

AIM: To study the bioassay of Histamine using guinea pig ileum by matching method.

APPARATUS:

- Reservoir, tubing, hemostatic forceps, isolated organ bath, aeration tube, isotonic frontal writing lever and recording drum.

EXPERIMENTAL CONDITION:

- Physiological Salt solution (PSS) : Tyrode
- Temperature : 37 (+ or -) 10C
- Aeration : Carbogen (95% O₂ and 5% CO₂)
- Basal tension on the tissue : 500 gm
- Magnification of the response : 10 times
- Drug : Histamine (1, 10 or 100 µg/mL)

PRINCIPLE:

Histamine produces a dose dependent contraction of guinea pig ileum smooth muscle. Two responses of the standard histamine are taken. The doses are adjusted such that one is giving response of approximately 20% and other 70% of the maximum. The response of unknown that lies between two responses of standard histamine dose is taken. The panel is repeated by increasing or decreasing the doses of standard till all three equal responses are obtained. The dose of test sample is kept constant. In the end, a response of the double dose of the standard and test that match each other are taken and the concentration of unknown is determined by the formula.

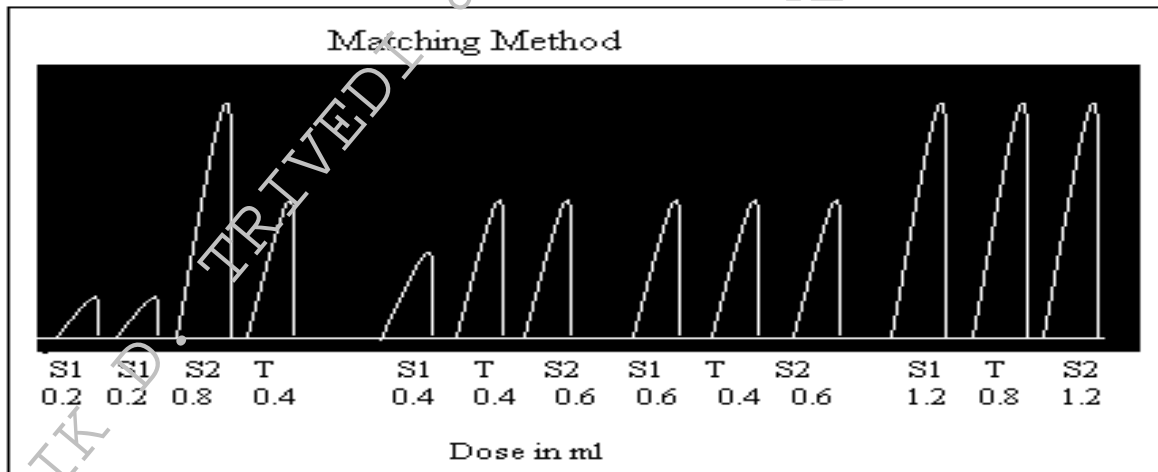
PROCEDURE:

1. The assembly is set up and arrangements are made for experimental conditions mentioned above.
2. A guinea pig which is fasted overnight is sacrificed as per CPCSEA recommended guidelines. The abdominal cavity is quickly opened & a piece of ileum is isolated. It placed in a Petri dish containing Tyrode solution maintained at 37°C.
3. The mesentery of ileum is removed & the lumen of ileum is cleaned by passing warm Tyrode solution through it from a pipette held at an angle of about 20-30 degrees.

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4. The tissue is mounted in mammalian organ bath and connected to isotonic frontal writing lever. The tissue is allowed to stabilize for 30 min.
5. The responses of standard Histamine solution are taken till the maximum effect is obtained.
6. Two responses of standard acetylcholine are taken in such a way that it produces approximately 20% and 70% respectively.
7. The dose of unknown sample (t) should be arranged in such a way that its response (T) should be between S1 & S2.
8. Then start to decrease dose of S2 & increase dose of S1 till all three equal responses are obtained. The dose of test sample is kept constant.
9. At the end, a response of the double dose of the standard and test which match each other are taken. These should give equal responses.

GRAPH:



DRAW GRAPH:

CALCULATION

$$\text{Concentration of unknown} = \frac{\text{Dose of std.}}{\text{Dose of test}} \times \text{conc. of std.} \times \text{dil. factor}$$

=

RESULT: The concentration of given unknown sample is _____ μg/ml.

QUESTIONS:

1. Give the principle of matching method.
2. Explain mechanism of histamine.

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