

- INTRODUCTION

To determine pD₂ value of histamine using guinea pig ileum.

- To record the dose response curve for histamine on an isolated tissue of guinea pig ileum.
- To plot the molar concentration vs percent response curves for histamine.
- To determine the half maximal effective concentration (EC₅₀) of histamine for guinea pig ileum
- To determine the pD₂ value of histamine on guinea pig ileum.

- EQUIPMENT REQUIRED

Animal :-	Guinea Pig
Drug:-	Histamine stock solution (10 µg /ml)
Instrument:-	Student Organ Bath, kymograph.
Physiological salt solution:-	Thyroid's solution

- PRINCIPLE

The principle for determining the pD₂ value of histamine using the guinea pig ileum involves measuring the contraction response of the isolated ileum to varying concentrations of histamine. The procedure follows these steps:

1. **Tissue Preparation:** An isolated segment of guinea pig ileum is suspended in an organ bath containing a physiological solution.
2. **Stimulation:** Histamine is administered in incremental concentrations to the tissue.
3. **Response Measurement:** The tension or contraction of the ileum is recorded for each concentration of histamine.
4. **Analysis:** The concentration-response curve is plotted, and the pD₂ value is calculated, which is the negative logarithm of the molar concentration of histamine required to produce 50% of the maximal response (EC₅₀).
5. **EC₅₀:** Effective Concentration 50% refers to the concentration of a drug that produces 50% of the maximum response. It is critical parameter for evaluating the potency of a substance: the lower the EC₅₀ value the more potent the substance as the concentration is needed to achieve half of its maximum effect.

- PROCEDURE:

1. The guinea pig is sacrificed by a blow on the head.
2. Cut open the abdomen and elevate the caecum to locate the ileocecal junction. Cut and remove a few centimetres of the ileal section, then insert it in the heated watch glass with Tyrode solution.

3. Trim the mesentery and gently clean the contents of the ileum by inserting the Tyrode solution into its lumen. To avoid causing harm to the stomach muscle, extreme caution should be used.
4. Cut the ileum into 2-3 cm-long segments.
5. Tie a thread to the top and bottom ends of 2-3cm of ileum tissue without closing the lumen. Place the tissue in an organ bath with Tyrode solution at 32-33°C and O₂ bubbles.
6. A tension of 0.5g is used, and the tissue is allowed to equilibrate for 30 minutes before being added to the organ bath.
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7. Using a frontal lever to record concentration-dependent responses to histamine.
8. Contact time is 30 seconds, and the time cycle is 3 minutes, with a wash period of 60 seconds twice. are preserved to ensure correct recording of responses.
9. Record the pD₂ value of histamine

- **CONCLUSION**

The conclusion for determining the pD₂ value of histamine using the guinea pig ileum is that the experiment successfully quantifies the potency of histamine by analyzing the concentration-response curve. The pD₂ value, derived from the EC₅₀, provides a quantitative measure of histamine's effectiveness in inducing contraction in smooth muscle. A higher pD₂ value indicates greater potency, while a lower pD₂ value suggests lower potency. This method is valuable for studying the pharmacological effects of histamine and comparing its potency to other compounds that may affect smooth muscle activity.

- **PAGE 4: IDEAL OBSERVATION**

Sr. No.	Conc. Of histamine (µg/mL)	Amount Added in Organ bath		Conc. In µg/mL histamine (In organ bath contains 20 ml solution)	Response (in mm)	% Response	Conc. Of histamine (µg/L)	Conc. Of histamine (µmol/mL)	Log Conc.
		In mL	In µg						
1.	10	0.1	1	0.05	7	16.66	0.449	4.49 x 10 ⁻⁴	-3.34775
2.	10	0.1	1	0.05	7	16.66	0.449	4.49 x 10 ⁻⁴	-3.34775
3.	10	0.2	2	0.1	16	38.09	0.899	8.99 x 10 ⁻⁴	-3.04624
4.	10	0.4	4	0.2	22	52.38	1.799	17.99 x 10 ⁻⁴	-2.74497
5.	10	0.8	8	0.4	34	80.95	3.589	35.98 x 10 ⁻⁴	-2.44394
6.	10	1.6	16	0.8	42	100.00	7.197	71.97 x 10 ⁻⁴	-2.14285
7.	10	3.2	32	1.6	42	100.00	14.394	143.94 x 10 ⁻⁴	-1.84182

RESULT:

The result of the experiment to determine the pD₂ value of histamine using the guinea pig ileum showed a concentration-response curve, where the contraction response increased with higher concentrations of histamine. The EC₅₀ value, representing the concentration of histamine that produces 50% of the maximum contraction, was used to calculate the pD₂ value. For example, if the EC₅₀ was 1×10^{-7} M, the pD₂ value would be 7, indicating histamine's potency in inducing ileum contraction.

DISCUSSION:

The pD₂ value of histamine reflects its potency in inducing contraction of the guinea pig ileum. A higher pD₂ indicates greater potency. The result is consistent with known histamine receptor activity, primarily through H₁ receptors. Experimental factors like tissue preparation and environmental conditions can influence results. Comparing the pD₂ value with literature helps confirm the experiment's accuracy. The findings also provide insight into histamine's pharmacological effects, with potential applications for studying antihistamines and other related drugs.