Histamine Three-point Bioassay

Experiment 11 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Teaching mode: use approved simulation, prepared demonstration records or supplied experimental data. No live-animal dosing, surgery or challenge is prescribed in these notes.

Aim

To estimate histamine activity from a supplied two-standard/one-test dataset.

Principle

A test response is compared with reference responses under a common valid assay design. The calibration relationship and response range must be appropriate.

Requirements

- Instructor-provided labelled diagrams, traces or data for this activity.
- Calculator, graph paper or spreadsheet, and the activity record.

Procedure

1. Read the assay design

Identify standard concentrations, test dilution and replicates. For the three-point exercise, identify low-standard, high-standard and test responses in the supplied balanced design.

2. Check the bracket

Confirm that the test response is within the applicable standard-response range and that the assigned assay validity criteria are satisfied.

3. Estimate

Use the fitted standard curve or the prescribed local log-concentration relation to calculate the test equivalent.

4. Correct and report

Apply the recorded dilution factor and state the assay units, uncertainty and validity limitation.

Observations and Record

Record item	Entry
Activity / material identifier	_____
Measurement or visible feature	_____
Comparison / calculated outcome	_____
Interpretation and limitation	_____

Worked Example

The numbers below are illustrative; enter your own observations in the record.

- For a local linear response versus log10 concentration: log10(Cu) = log10(C1) + (Ru − R1)/(R2 − R1) × log10(C2/C1). This illustrative interpolation does not replace the prescribed full statistical assay analysis.
- Example: C1 = 1, C2 = 10 units/mL, R1 = 10, R2 = 30 and Ru = 20 give Cu = 10^0.5 = 3.16 units/mL before dilution correction.

Result

The recorded finding is ____; the supporting observation or calculation is ____. The main limitation is ____.

Precautions

- Use the supplied teaching material; distinguish a measured observation from an assumed value.
- Keep units, axes, labels and measurement conditions consistent.
- Record limitations and seek instructor review before applying a conclusion to a live case.

Viva Questions and Answers

1. Why bracket the test response?

Extrapolation outside the validated standard range can give unreliable activity estimates.

2. Does an interpolated number prove assay validity?

No. Replicate variation, response model and assigned statistical validity checks must also be assessed.

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