

PractiChrom™ Histamine Assay Kit (PHIS-25)

Quantitative Histamine Determination Using PICOEXPLORER™

DESCRIPTION

HISTAMINE (C₅H₉N₃) is a biogenic amine naturally present in many foods and body cells in amounts without toxicological significance. It is also a contaminant that mostly found in the *Scombridae* family fishes such as tuna and mackerel or other seafood products when improperly handled or stored. The consumption of foods containing high level of histamine may lead to an allergy-like food poisoning known as scombroid poisoning.

BioAssay Systems' histamine assay kit is based on enzyme catalyzed oxidation of histamine in which the formed electron mediator reduces a formazan reagent. The intensity of product color is directly proportional to histamine concentration in the sample.

KEY FEATURES

Sensitive and accurate. Detection limit of 0.012 mM (1.3 ppm, 0.13 mg/dL) and linearity to 1 mM (111 ppm, 11.1 mg/dL) Histamine.

Convenient. Assay performed with portable PiCO Explorer device.

Cost efficient. No need for expensive plate readers.

APPLICATIONS

Direct Assays: histamine in food, beverage, and agricultural products.

KIT CONTENTS (25 TESTS)

Extraction Buffer: 8 mL **Standard:** 1.0 mL 20 mM Histamine

HDH Enzyme: 30 µL

Storage conditions: The kit is shipped at room temperature. Store the Standard and HDH Enzyme at -20°C upon receiving. The Extraction Buffer can be stored at room temperature. For long term storage (>30 days), we recommend storing all components at -20°C. Shelf life: 6 months after receipt.

Precautions: reagents are for research use only. Equilibrate all components to room temperature prior assay. Normal precautions for laboratory reagents should be exercised while using the reagents. Please refer to Material Safety Data Sheet for detailed information.

PROCEDURES

Sample Preparation:

Seafood. Weigh 500 mg of fish and place it in an Eppendorf tubes. Then add 500 µL Extraction buffer to the tube and homogenize. Centrifuge the tube for 10 min at full speed. Assay the supernatant. (Dilution factor: 2)

Fish Sauce & Soy sauce. Dilute 10× in dH₂O by adding 10 µL sample to 90 µL dH₂O. (Dilution factor: 10)

Other colored samples will require a dilution. For preparation protocols for other samples, please contact our technical support at info@bioassaysys.com

Procedure

1. Prepare 1 mM Histamine Standard by mixing 5 µL of the provided 20 mM Standard and 95 µL dH₂O in an Eppendorf tube.
2. In separate PCR tubes, add 10 µL dH₂O and 10 µL 1 mM Histamine Standard. Add 10 µL Sample to two separate tubes, one serving as the Sample tube and one as the Sample Blank tube.

Prepare sufficient Working Reagent for all dH₂O, Standard, and Sample tubes by mixing, for each tube: 43 µL Extraction Buffer plus 2 µL HDH Enzyme.

Then quickly add 40 µL Working Reagent to all dH₂O, Standard, and Sample tubes. To each Sample Blank tube, only add Extraction Buffer (No HDH enzyme). Close the tubes, briefly vortex or tap to mix. Tap tube on bench to settle liquid to the bottom of the tube. Incubate for 20 min at room temperature in the dark.

3. Please refer to the PICOEXPLORER™ User's Manual for detailed instructions for operating the device.

Download the PAS-110 application. Turn on Bluetooth.

Push the Power button on the device. Then, open the app and tap the Connection Setting button and connect the device.

Measuring a Standard Curve (See pg 17-19 in User's Manual)

Return to the main menu and tap the Standard Curve button. Set the following:

LED Output: 10%

Unit: mM

RBG Selection: G

Tap the first Known Concentration Data Input Area box and input 0.0. Then, tap on the second box and input 1.0 (this represents the 0 and 1 mM Histamine standards). Place the dH₂O tube into the measurement chamber of the photo absorbance sensor. Tap the Known Concentration Measurement Input Area (the box below 0.0), and click Measure. Remove the tube, then place the 1 mM Standard into the measurement chamber. Tap the box below 1.0 and click Measure. Click Graph to view the standard curve.

Measuring Sample Concentrations

Return to the main menu and tap the Measure button. Edit the LED output, Units, and RBG selection as done above for the standard curve.

Place each Sample and Sample Blank tube into the measurement chamber of the photo absorbance sensor and tap measure.

CALCULATION

The "concentration" will be displayed on the PICOEXPLORER™ for each Sample and Sample Blank. To calculate the Histamine concentration in the sample, subtract the Sample Blank concentration from the Sample concentration and multiply by the dilution factor used. (e.g. 2, 10, etc)

$$[\text{Histamine}] = ([\text{Sample}] - [\text{Sample Blank}]) \times n \quad (\text{mM})$$

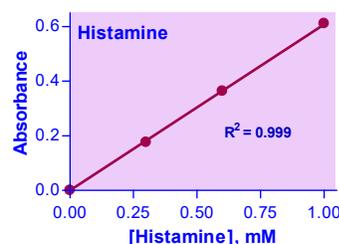
where [Sample] is the concentration of sample plus HDH, [Sample Blank] is the concentration of sample *without* HDH, and *n* is the dilution factor.

Note: if the sample concentration says "Out of range" the sample is not within the linear range of the assay. If the color of the tube is yellow like the dH₂O tube, then the sample has low levels of Histamine that cannot be detected by the assay. If the sample is very dark, dilute further in dH₂O and repeat the assay. Multiply the results by the dilution factor.

Conversions: 1 mM histamine equals 11.1 mg/dL, or 111 ppm.

MATERIALS REQUIRED, BUT NOT PROVIDED

Pipetting devices, PCR tubes (e.g. Watson 137-211c 0.2 mL; or Cat# PCR-50 from BioAssay Systems), Eppendorf tubes (e.g. Phenix Cat# MAX-715, or Cat # EPP-50 from BioAssay Systems), centrifuge, and PICOEXPLORER™ (Cat # PICO001).



Standard Curve in water measured with PICOEXPLORER™

RELATED PRODUCTS

PICOEXPLORER™ (Handheld Colorimeter): Cat # PICO001

PCR Tubes: Cat # PCR-50, Pack of 50 tubes

Eppendorf tubes: Cat # EPP-50, Pack of 50 tubes

Mini Centrifuge: Cat # CTFG, Small battery operated centrifuge for fish sample preparation.

Histamine Quick Test Strips: Cat # QQHIST10