AB308323 – Aldehyde Assay Kit (Colorimetric)

Measurement of aldehyde in various samples.
For research use only - not intended for diagnostic use.

For overview, typical data and additional information please visit: abcam website

Storage and Stability

Store the kit at -20 °C, protected from light. Briefly centrifuge all small vials prior to opening. Read the entire protocol before performing the assay. Use within six months after opening.

Materials Supplied

Item	Quantity	Storage Condition
Aldehyde Assay Buffer	25 ml	4 °C or -20 °C
Aldehyde Indicator	5 vials	-
Aldehyde Developer	1.2 m	-20°C
Aldehyde Standard	15 µl	-20°C

Materials Required, Not Supplied

These materials are not included in the kit, but will be required to successfully utilize this assay:

- 1N NaOH
- 96-well clear plate with flat bottom
- Multi-well spectrophotometer (plate reader)
- Dounce Tissue Homogenizer
- 10 kD Spin Column

Reagent Preparation

Aldehyde Assay Buffer: Store at 4 °C or -20 °C. Bring to room temperature (RT) before use.

Aldehyde Indicator: Reconstitute each vial in 220 µl of 1N NaOH and mix well. Keep on ice, while in use. Discard within 4 hrs.
Δ Note: *The mixture is not stable.*

Aldehyde Developer: Ready to use. Thaw at RT. Keep on ice, while in use. Store at -20 °C.

Aldehyde Standard: Ready to use. Thaw at RT. Keep on ice, while in use. Store at -20 °C.

Aldehyde Assay Protocol

Sample Preparation

For tissues or cells: Homogenize pelleted cells (~5 x 105 cells) with 100 µl ice-cold Aldehyde Assay Buffer using Dounce Tissue Homogenizer and keep on ice for 10-15 min. Centrifuge samples at 12,000 x g and 4 °C for 15 min and collect the supernatant for the assay. Keep the sample on ice, while in use.
For biological fluids: Centrifuge samples at 12,000 x g and 4 °C for 15 min and collect the supernatant. Keep the sample on ice, while in use. **To minimize the matrix effect, filter all samples through a 10 kDa spin column. Centrifuge at 10,000 x g and 4 °C for 10 min and collect the filtrate. Repeat the process, if needed. Add 2-20 µl of the filtrate into each Sample well. Adjust the volume of each Sample well to 75 µl/well with Aldehyde Assay Buffer. Prepare a Reagent Background Control well by adding 75 µl of Aldehyde Assay Buffer.

Δ Note: *For unknown samples, we recommend testing several doses of the samples to ensure that the readings are within the Standard Curve range.*

Standard Curve Preparation:

Prepare a 5 mM Aldehyde Standard by adding 5 µl of Aldehyde Standard stock to 95 µl Aldehyde Assay Buffer. Add 0, 2, 4, 6, 8, 10 µl of 5 mM Aldehyde Standard into a series of wells to generate 0, 10, 20, 30, 40, 50 nmol/well of Aldehyde Standard respectively. Adjust the volume to 75 µl/well with Aldehyde Assay Buffer.

Sample Reaction: Add 13 µl of Aldehyde Indicator into each well containing Sample(s), Reagent Background Control and Aldehyde Standards and mix well. Incubate at RT for 30 min for the reaction to finish.

Δ Note: *Do not re-use the reconstituted Aldehyde Indicator.*

Sample Development:

Add 12 µl of Aldehyde Developer to each well containing Sample(s), Reagent Background Control and Aldehyde Standards and mix well. Incubate at RT for 30 min.

Measurement:

Measure the absorbance of all wells at RT at OD 530 nm in end-point mode.

Δ Note: *The absorbance and color of the sample solution might vary according to the type of aldehyde.*

Calculation:

Subtract 0 Standard reading from all Standard readings. Plot the Aldehyde Standard Curve. Subtract the Reagent Background Control reading from all Sample readings to get the corrected Sample readings (ΔOD). Apply the corrected Sample readings (ΔOD) to the Aldehyde Standard Curve to obtain the corresponding amount of aldehyde (B in nmol). Calculate the amount of Aldehyde in Sample as:

Aldehyde in Sample = B x D / (V x P) = nmol / µg protein

Where: **B** = Amount of Aldehyde from the Aldehyde Standard Curve (nmol)
V = Sample volume added into the reaction well (µl)
P = Initial Sample concentration (µg/µl)
D = Sample dilution factor (D = 1 for undiluted samples)

Technical Support

For all technical or commercial enquiries please go to:
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