D-Xylose Assay Kit

Catalog No.: abx298936

Size: 96 tests

Detection Range: 0.007 mmol/L – 4 mmol/L

Sensitivity: 0.007 mmol/L

Storage: Store all components at 4°C in the dark for up to 12 months.

Application: For detection and quantification of D-Xylose concentration in serum, plasma, and urine samples.

Abbexa's D-Xylose Assay Kit is a quick, convenient, and sensitive method for measuring and calculating D-Xylose activity. The absorbance should be measured at 554 nm. The intensity of the color is proportional to the concentration of D-Xylose, which can then be calculated.

Kit components

1. Phloroglucinol: 6 × 60 ml
2. Standard (13.3 mmol/L): 1 ml
3. Standard diluent: 10 ml

Materials Required But Not Provided

1. Spectrophotometer (554 nm)
2. Double distilled water
3. Normal saline (0.9% NaCl) or PBS (0.01 M, pH 7.4)
4. Pipette and pipette tips
5. Glass vials/tubes
6. 100°C water bath
7. Centrifuge
8. Vortex mixer
9. Timer

Protocol

A. Preparation of samples and reagents

1. Samples

The following sample preparation methods are intended as a guide and may be adjusted as required depending on the specific samples used.

- **Serum:** Samples should be collected into a serum separator tube. Coagulate the serum by leaving the tube undisturbed in a vertical position overnight at 4°C or at room temperature for up to 1 hr. Centrifuge at approximately 2000 × g for 15 mins at 4°C. If a precipitate appears, centrifuge again. Take the supernatant, keep on ice and assay immediately, or aliquot and store at -80°C for up to 1 month.
- **Plasma:** Collect plasma using heparin as the anticoagulant. Centrifuge for 10 mins at 1000-2000 × g at 4°C, within 30 mins of collection. If precipitate appears, centrifuge again. Avoid hemolytic samples. Take the supernatant (avoid taking the middle layer containing white blood cells and platelets), keep on ice and assay immediately, or aliquot and store at -80°C for up to 1 month.
- **Urine:** Collect urine and centrifuge at 10,000 × g for 15 minutes at 4°C. Take the supernatant, keep on ice and assay immediately, or aliquot and store at -80°C for up to 1 month.

We recommend carrying out a preliminary experiment to determine the optimal dilution factor of samples before carrying out the formal experiment.

Note:

- Fresh samples or recently obtained samples are recommended to prevent degradation and denaturalization that may lead to erroneous results.
- Lysis buffers may interfere with the kit. It is therefore recommended to use mechanical lysis methods for cell lysates and tissue homogenates.
- Some reagents in this kit are irritants, the procedure should be carried out in a fume hood.
- **Test samples should be pre-treated with D-Xylose before assay, and control samples should not be pre-treated with D-Xylose before assay.**

2. Reagents

- **1.33 mmol/L standard solution:** Dilute the standard with standard diluent solution 10-fold (for example, 9 ml of standard diluent should be added to 1 ml of 13.3 mmol/L standard solution). This produces a 1.33 mmol/L standard solution. Prepare immediately before carrying out the assay and mix fully. After dilution, the 1.33 mmol/L standard solution can be stored at 4°C in the dark for up to 3 months.

B. Assay Procedure

1. Set blank, sample control, standard and sample glass tubes. *Each sample requires a sample control tube.*
2. **Serum and plasma samples:** Add 30 µl of treated sample to the sample tube. Add 30 µl of untreated sample to the sample control tube. Add 30 µl of 1.33 mmol/L standard to the standard tube. Add 30 µl of double distilled water to the blank tube.
3. **Urine samples:** Add 50 µl of treated sample to the sample tube. Add 50 µl of untreated sample to the sample control tube. Add 50 µl of 1.33 mmol/L standard to the standard tube. Add 50 µl of double distilled water to the blank tube.

4. Add 3 ml of Phloroglucinol to all tubes and mix fully.
5. Incubate all tubes at 100°C in the water bath, and begin the timer. After 4 minutes, remove the tubes and cool immediately with running cold water.
6. Calibrate the spectrophotometer to zero using double distilled water.
7. Measure the OD values of each tube at 554 nm with a 1 cm optical path cuvette.

C. Calculation of Results

$$\text{D Xylose (mmol/L)} = \frac{\text{OD}_{\text{Sample}} - \text{OD}_{\text{Sample Control}}}{\text{OD}_{\text{Standard}} - \text{OD}_{\text{Blank}}} \times c \times f$$

where:

$\text{OD}_{\text{Sample}}$	absorbance of the sample
$\text{OD}_{\text{Sample Control}}$	absorbance of the sample control
$\text{OD}_{\text{Stamdard}}$	absorbance of the 1.33 mmol/L standard
OD_{Blank}	absorbance of the blank
c	concentration of the standard (1.33 mmol/L)
f	dilution factor of the sample before carrying out the assay