Phenols Assay Kit

ARG83415 Phenols Assay Kit can be used to measure Phenols in urine, serum, plasma, tissue extracts, cell lysate, cell culture media and other biological fluids.

Catalog number: ARG83415

Package: 96 wells

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INTRODUCTION

In organic chemistry, phenols, sometimes called phenolics, are a class of chemical compounds consisting of one or more hydroxyl groups (−OH) bonded directly to an aromatic hydrocarbon group. The simplest is phenol, C6H5OH. Phenolic compounds are classified as simple phenols or polyphenols based on the number of phenol units in the molecule.

PRINCIPLE OF THE ASSAY

The Phenols Assay Kit can measure Phenols in urine, serum, plasma, tissue extracts, cell lysate, cell culture media and other biological fluids. Phenols can react with phosphomolybdic acid. The product increase in absorbance at 760 nm is directly proportional to the Phenols content.

MATERIALS PROVIDED & STORAGE INFORMATION

Component	Quantity	Storage
Microplate	1 X 96-well plate	
Standard	1 vial (lyophilized)	4°C
Assay Buffer	4 X 30 ml	4°C
Reaction Buffer	3 ml (ready to use)	4°C
Reagent Dye	1 ml (ready to use)	4°C

MATERIALS REQUIRED BUT NOT PROVIDED

- Microplate reader capable of measuring absorbance at 760 nm
- Pipettes and pipette tips
- Deionized or distilled water

TECHNICAL HINTS AND PRECAUTIONS

- Wear protective gloves, clothing, eye, and face protection especially while handling blood or body fluid samples.
- All component store at 4°C.
- Briefly spin down the reagents before use.
- It is highly recommended that the standards and samples be assayed in at least duplicates.
- Change pipette tips between the addition of different reagent or samples.

SAMPLE COLLECTION & STORAGE INFORMATION

The sample collection and storage conditions listed below are intended as general guidelines. Sample stability has not been evaluated.

Tissue - Weigh out 0.1 g tissue, homogenize with 1 ml Assay Buffer, put it in water bath of 60 °C for 2 hours with shaking, centrifuged at 10,000g for 10 minutes, take the supernatant into a new centrifuge tube.

*Note: liquid samples can detect directly.

REAGENT PREPARATION

- Standard: Add 1 ml of Assay Buffer to dissolve standard; then add 0.2 ml into 0.8 ml Assay Buffer. The concentration will be 4 µmol/ml. Perform 2-fold serial dilution of the top standards to make the standard curve.

ASSAY PROCEDURE

Standards and samples should be assayed in at least duplicates.

1. Sample wells: Add **10 µl Sample** into Sample wells.
2. Standard wells: Add **10 µl Standard** into Standard wells.
3. Add **120 µl Distilled water** to each wells (**130 µl** for Blank wells).
4. Add **60 µl Reaction Buffer** to each wells.
5. Mix well. Incubate at **RT** for **5 min**
6. Add **10 µl Reagent Dye** to each wells.
7. Mix well. Incubate at **RT** for **10 min**. Read the OD at **760nm**

Reagent	Sample	Standard	Blank
Sample	10 µl	-	-
Standard	-	10 µl	-
Distilled water	120 µl	120 µl	130 µl
Reagent Buffer	60 µl	60 µl	60 µl
Mix well. Incubate at **RT** for **5 min**			
Reagent Dye	10 µl	10 µl	10 µl
Mix well. Incubate at **RT** for **10 min**. Read the OD at **760nm**			

CALCULATION OF RESULTS

1. Calculate the average absorbance values for each set of samples, standard and blank.

2. Calculation:

A. Definition:

$C_{Standard}$: the standard concentration, 4 µmol /ml;

W: the weight of sample, g;

V_{Sample}: the volume of reaction sample, 10 µl = 0.01 ml;

$V_{standard}$: the volume of standard, 10 µl = 0.01 ml;

V_{assay}: the volume of Assay Buffer, 1000 µl = 1 ml.

B. Formula:

a). According to the weight of sample

Phenols (µmol/g) =

$[(C_{Standard} \times V_{standard}) \times (OD_{Sample} - OD_{blank})] / [(OD_{Standard} - OD_{Blank}) \times (W \times V_{Sample} / V_{assay})]$

$=4 \times (OD_{Sample} - OD_{blank}) / [(OD_{Standard} - OD_{Blank}) \times W]$

b). According to the volume of sample

Phenols (µmol/ml) =

$[(C_{Standard} \times V_{standard}) \times (OD_{Sample} - OD_{blank})] / [(OD_{Standard} - OD_{Blank}) \times V_{Sample})]$

$=4 \times (OD_{Sample} - OD_{blank}) / (OD_{Standard} - OD_{Blank})$

3. Detection range:

The detection range is from 0.04 mg/ml - 4 mg/ml.

4. If the samples have been diluted, the calculated activity must be further converted by the appropriate dilution factor according to the sample preparation procedure as described above.

EXAMPLE OF TYPICAL RESULT

The following data is for demonstration only and cannot be used in place of data generations at the time of assay. Please note this data is for demonstration only and serial diluted standards are not necessary for this kit.