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Plant Total Phenol (TP)Assay Kit

Note: Take two or three different samples for prediction before test.

Operation Equipment: Spectrophotometer/ microplate reader

Cat No: AK0449

Size: 100T/48S

Components:

Extract solution: 60% alcohol, self-provided reagent.

Reagent I: Liquid 5 mL×1, store at 4℃ .

Reagent II: Liquid 8 mL×1, store at 4℃ .

Standard: Powder×1, store at 4℃ . 5 mg of gallic acid. Before use, add 1mL of distilled water, heat it at 50℃ and dissolve it to prepare 5mg/mL standard solution.

Description:

Plant phenols have the function of scavenging free radicals, anti-oxidation and anti-aging. It is widely used in cosmetics, food, medicine and other fields because of its high nutritional value and health care function. In alkaline conditions, phenolic substance reduce tungsten-molybdic acid to form blue compounds which has a absorption peak at 760 nm. The total phenol content of the sample is obtained by measuring the absorbance at 760 nm.

Required but not provided:

Balance, oven, crusher, sieve, ultrasonic breaker, centrifuge, 60% alcohol, spectrophotometer/microplate reader, micro glass cuvette/96 well plate, distilled water.

Procedure:

I. Total phenol extraction:

Dry the sample to constant weight, smash. After screening with the 40 mesh sieve, add 2.5 mL of Extract solution to 0.1 g of tissue and extract by ultrasonic breaker, (power 300W, crush 5s, interval 8s, 60℃ for 30 min). centrifuge at 12000 rpm for 10 min at 25℃. Take supernatant and make the liquid to a volume of 2.5 mL with the Extract solution.

II. Preparation of standard.

The standard solution of 5 mg/mL standard solution is diluted to 0.3125, 0. 15625, 0.078125, 0.039, 0.02, 0.01, 0.005, 0.0024 mg/mL for test.

III. Determination procedure.

1. Preheat spectrophotometer or microplate reader for 30 min, adjust wavelength to 760 nm, set zero with distilled water.

2. Add reagents according to the following table.

Reagent Name (μL)	Control tube (A_C)	Test tube (A_T)	Standard tube (A_S)	Blank tube (A_B)

Sample	10	10		-
Standard			10	
Distilled water				10
Reagent I		50	50	50
Mix thoroughly, incubate at room temperature for 2 min.				
Reagent II	50	50	50	50
Distilled water	140	90	90	90
Mix thoroughly, incubate at room temperature for 10 min. Detect the absorbance of 760 nm in micro glass cuvette or 96 well flat-bottom plate.				

Note: Blank tube just test once or twice.

IV. Calculation.

1. Draw the standard curve.

With the concentration of different standard solution as x-axis, $\Delta A(A_S-A_B)$ as y-axis, draw standard curve y=kx+b. Bring $\Delta A=A_T-A_C$ to standard curve, calculate x(mg/mL).

2. Calculation of plant total phenol

a. Sample weight

Total phenol (mg/g) = $x\times V_E\div W=2.5x\div W$

b. Protein concentration

Total phenol (mg/mg prot) = $x\times V_E\div(Cpr\times V_E) = x\div Cpr$

V_E: Extract solution volume；2.5 mL；

W: Sample weight, g；

Cpr: Protein concentration, mg/mL.

Note:

1. If OD>1, determine after diluting, multiply dilution multiple in equation.
2. Reagent I have a certain irritation to the skin, please take precautions during operation.

Examples:

1. Add 0. 1g treated purple flower to 2.5mL extract solution, after treating sample follow the determination procedure to operate, with 96-well flat-bottom plates to calculate: ΔA =A(T)-A(B)=0.506-0.041=0.465, standard curve: y=3.083x+0.01, calculate x=0. 1476, according with mass of sample to calculate: Total phenol (mg/mg mass) =2.5x÷W=2.5×0. 1476÷0. 1=3.69 mg/g mass.

Recent Product citations：

[1] Wang Y, Gao S, He X, et al. Response of total phenols, flavonoids, minerals, and amino acids of four edible fern species to four shading treatments[J]. PeerJ, 2020, 8: e8354.

Related Products:

AK0254/AK0253	Ceruloplasmin (CP) Assay Kit
AK0456/AK0455	Total antioxidant capacity (T-AOC) Assay Kit
AK0444/AK0443	Total Sulfhydryl Assay Kit