

Pectin Microplate Assay Kit

Catalog # AS0176

Detection and Quantification of Pectin Content in Tissue extracts, Other biological fluids Samples.

This instruction must be read in its entirety before using this product.

For research use only, Not for use in diagnostic procedures.

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I. INTRODUCTION

Pectin, any of a group of water-soluble carbohydrate substances that are found in the cell wallsand intercellular tissues of certain plants. In the fruits of plants, pectin helps keep the walls of adjacent cells joined together. Immature fruits contain the precursor substance protopectin, which is converted to pectin and becomes more water-soluble as ripening proceeds. At this stage the pectin helps ripening fruits to remain firm and retain their shape. As a fruit becomes overripe, the pectin in it is broken down to simple sugars that are completely water-soluble. As a result, the overripe fruit becomes soft and begins to lose its shape.

Pectic substances consist of an associated group of polysaccharides that are extractable with hot water or with aqueous solutions of dilute acids. The chief sources of commercial pectin are the peels of citrus fruits, and to a lesser extent apple pomace (residue from cider presses). Very small amounts of pectin suffice in the presence of fruit acids and sugar to form a jelly.

Pectin also has several health benefits in humans. Included among these are its ability to reduce low-density lipoprotein (LDL) levels, thereby lowering cholesterol levels, and its ability to slow the passage of food through the intestine, relieving diarrhea. Pectins can also activate cell death pathways in cancer cells, indicating that pectins may play an important role in preventing certain types of cancer. Pectin Microplate Assay Kit provides a very sensitive and convenient means to measure pectincontent in a variety of samples. In the assay, pectinase reacts with pectin, resulting in the formation of galacturonic acid, which react with dye reagent and determined at 540nm, is directly proportional to the pectincontent in the sample.



II.KIT COMPONENTS

Component	Volume	Storage
96-Well Microplate	1 plate	
Assay Buffer	30 ml x 6	RT
Reaction Buffer	20 ml x 1	4 °C
Dye Reagent	10 ml x 1	4 °C, keep in dark
Enzyme	Powderx 1	-20 °C
Standard	Powderx 1	4 °C
Plate Adhesive Strips	3 Strips	
Technical Manual	1 Manual	

Note:

Enzyme:add1 mlReaction Bufferto dissolve before use.

Standard: add 1 ml Distilled waterto dissolve before use; then add 0.3 ml into 0.7 ml

Distilled water, mix, the concentration will be 3 mmol/L.

III. MATERIALS REQUIRED BUT NOT PROVIDED

- 1. Microplate reader to read absorbance at 540 nm
- 2. Distilled water
- 3. Pipettor
- 4. Pipette tips
- 5. Mortar
- 6. Centrifuge
- 7. Timer
- 8. Convection oven



IV. SAMPLE PREPARATION

1.For tissue samples

Weigh out 0.1 g tissue, homogenize with 1 ml Assay Buffer in the mortar; then transfer all the sample into the microcentrifuge tube; put it into the water bath of 80 °C for 30 minutes; centrifuge at 8,000g for 10minutes, discard the supernatant; then add 0.5ml Assay Buffer again, mix, put it in water bath of 80 °C for 30 minutes, centrifuge at 8,000g for 10minutes, discard the supernatant; add 150 µlReaction Buffer into the microcentrifuge tube, mixfor detection.

2. For liquid samples

Add 10 μl sample and 150 μl Reaction Buffer into the microcentrifuge tube, mix for detection.



V. ASSAY PROCEDURE

Add following reagents into the sample microcentrifuge tube:

Reagent	Sample	Standard	Blank		
Enzyme	40 μl				
Mix, put it intothe water bath, 50 °C for 60 minutes.Centrifuge at 8,000g for					
10minutes, add the supernatant into the microplate.					
Sample	100 μl				
Standard		100 μl			
Distilled water			100 µl		
Dye Reagent	100 μl	100 μl	100 µl		
Mix, put it into the convection oven, 90 °Cfor 10 minutes, record absorbance					
measured at 540nm.					



VI. CALCULATION

1. According to the weight of sample

Pectin(µmol/g) = (C_{Standard}×V_{Standard}) ×(OD_{Sample} - OD_{Blank}) / (OD_{Standard} - OD_{Blank})/ (V_{Sample}×W/ V_{Total})

= $0.6 \times (OD_{Sample} - OD_{Blank}) / (OD_{Standard} - OD_{Blank}) / W$

2. According to the volume of sample

Pectin (µmol/ml) = (C_{standard}×V_{standard}) ×(OD_{sample} - OD_{Blank}) / (OD_{standard} -OD_{Blank})/ (V_{sample}×V/ V_{Total})

= 0.6× (OD_{Sample} - OD_{Blank}) / (OD_{Standard} - OD_{Blank}) / V

V_{Sample}: the volume of sample, 0.1 ml;

V_{Standard}: the volume of standard, 0.1 ml;

V_{Total}: the volume of all sample, reaction buffer and enzyme, 0.2 ml;

 $C_{Standard}$: the standard concentration, $3mmol/L = 3\mu mol/ml$;

C_{Protein}: the protein concentration, mg/ml;

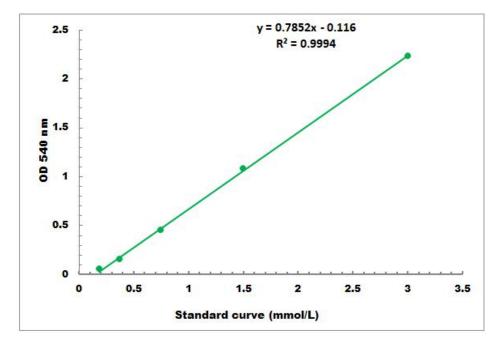
W: the weight of sample, g;

V: the volume of sample, ml.



VII. TYPICAL DATA

The standard curve is for demonstration only. A standard curve must be run with each assay.



Detection Range: 0.3mmol/L -3mmol/L

VIII. TECHNICAL SUPPORT

For troubleshooting, information or assistance, please go online to www.sabbiotech.cn or contact us at techcn@signalwayantibody.com

IX. NOTES