

WHEAT GERM COATED SURFACE

Wheat Germ Lectin, belonging to the lectins family, is a Hemagglutinin obtained from the common wheat germ *Triticum Vulgaris*. It is well known that lectins have been used extensively for the isolation of glyco-conjugates and glycoproteins with specific carbohydrate structures.

Wheat Germ Lectin shows specific affinity for molecules containing N-acetyl-D-glucosamine residue.

Wheat Germ Lectin coated surfaces offer a powerful and sensitive instrument for binding in specific way the carbohydrate fraction of glycoproteins, enzymes and cell membranes.

The optical properties of polystyrene remain unchanged, allowing to use the modified surface as powerful tool for diagnostic assays.

Example of applications:

- studies of surfaces of normal and transformed cells
- glycoprotein purification including membrane glycoproteins
- studies of cell surface changes during development and the cell cycle

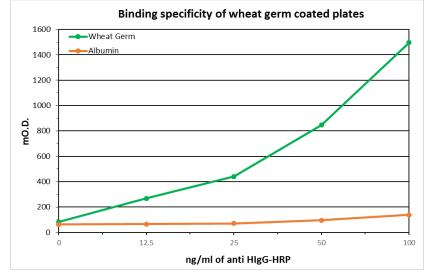
TECHNICAL NOTE N. 17

General procedure for binding a biomolecule to Wheat Germ coated surface

- 1. Dilute your biomolecule (sample) to 0.5- 5 μ g/ml in an appropriate neutral pH buffer (buffer should contain 1mM Ca⁺⁺ and 1mM Mn⁺⁺; in fact these ions promote the interaction between saccharide groups and Wheat Germ coated surface)
- 2. Proceed with incubation: conditions depend on biomolecule structure
- 3. Wash four times to remove the unbound material
- 4. Proceed with your specific test: to point out the bound biomolecule and/or to use the bound biomolecule to point out a specific counter molecule

Example of test: binding specificity of Wheat Germ coated plates

- Dilute AHIgG-Pod from 100 ng/ml to 12.5 ng/ml in pure distilled water containing 1 mM CaCl₂•2 H₂O + 1 mM MnCl₂•4 H₂O
- 2. Add 100µl of each solution to the wells of Wheat Germ coated plate and incubate 30' R.T.; add the same solutions to albumin coated plate as comparison for evaluate the specificity of binding
- 3. Leave blank wells as control
- 4. Empty the wells and wash with 0.1M PBS pH 7.2 + 0.05% Tween[®] 20 four times
- 5. Add 100 µl /well of TMB substrate solution and incubate 10 minutes at room temperature



6. Stop the substrate reaction by adding $100 \mu l$ of sulphuric acid 1 N and read the optical density at 450 nm