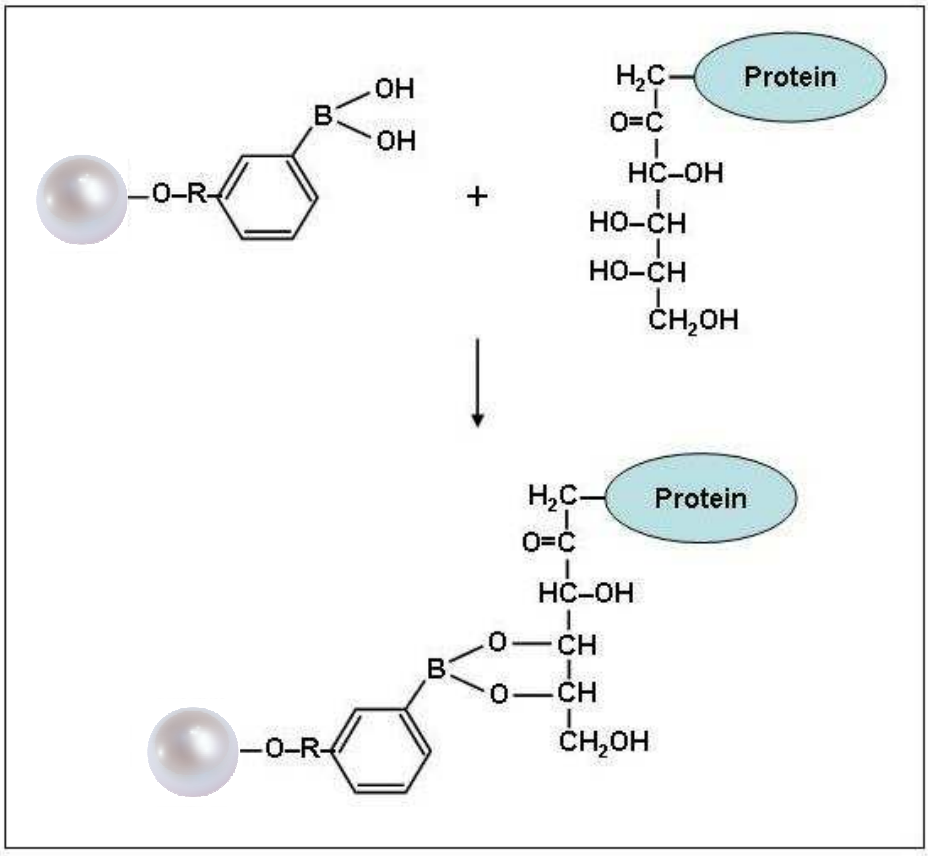
Protocol for isolation of vicinal cis-diol containing sugar compounds with beadBALL-Boronic acid

Introduction:

beadBALL-Boronic acid microspheres represent an easy-to-use affinity support for binding of glycoproteins, sugars, ribonucleotides and other small molecular weight compounds which contain vicinal cis-diol groups.

The immobilized boronic acid bind to vicinal cis-diol groups on sugars, such as mannose, galactose, glucose or ribose. After the binding and washing step the sugar containing sample can be eluted from the particles by lowering the pH or by addition of sorbitol.



Equipment and reagents:

- **beadBALL-Boronic acid**
  (w/v: 10 mg/ml in ddH_2O, 0.05 % sodium azide)

- **Wash & Binding buffer (W & B buffer):**
  0.25 M ammonium acetate, 50 mM $MgCl_2$, pH 9.5.

- **Elution buffer:**
  50 mM Tris, 0.1 M sorbitol, pH 8.5.

- **Vortex, Tube Rotator**

- **Microcentrifuge**

Protocol:

The following general protocol describes the binding of vicinal cis-diol containing molecules with beadBALL-Boronic acid.

1. Add 100 µl **beadBALL-Boronic acid** microspheres in a 2 ml microcentrifuge tube, add 1.0 ml **W & B buffer** and centrifuge for 1 minute at 500 x g. Remove the supernatant and repeat this step twice. Completely resuspend the microspheres in 0.5 ml **W & B buffer**.

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2. Add the sample to the microspheres and fill up with **W & B buffer** to a maximal volume of 1 ml.

3. Incubate at room temperature for 15 - 30 minutes by gentle mixing (tube rotator).

4. Spin down, remove and discard the supernatant.

5. Add 1.5 ml **W & B buffer** resuspend thoroughly, spin down, remove and discard the supernatant and repeat the washing step two times.

6. Add 100 µl **Elution buffer**, vortex and incubate for 15 minutes at room temperature in a thermo-mixer or shake the tube intermediately.

7. Spin down and transfer the supernatant in a new tube.