Regeneration Protocol for 100 INDIGO Ni-Agarose

(03/17/2025)

Protocols for washing and regenerating 100 INDIGO Ni-Agarose by alkaline buffers

CV= column volume. I.e. for 1 ml column bed volume use 10 CV = 10 ml of buffer

Wash and Regenerate (recommended after each run, latest after five runs)

Please take care, that all solutions have a temperature of 4 °C or ambient temperature! Hot sodium hydroxide can harm resin and chromatography material!

1. 10 CV H_2O
2. 10 CV 500 mM NaOH
3. 10 CV H_2O
4. 10 CV Neutralization buffer (150 mM sodium chloride; 200 mM Na_2HPO_4, pH 7.0)
5. 10 CV H_2O
6. 10 CV 20% (v/v) Ethanol, 10 mM sodium acetate, pH 6.50*

*Note: 20% (v/v) Ethanol, 10 mM sodium acetate, pH 6.50, is the recommended storage buffer for the column and resin.

Note: Sodium hydroxide (Danger! Causes eye and skin burns, causes digestive and respiratory tract burns, hygroscopic, absorbs moisture from the air) and ethanol (HIGHLY FLAMMABLE (R16 S43), FLASH POINT: (R11) 9°C, IGNITION TEMPERATURE: 425°C, EXPLOSION LIMITS: LOWER: 3.5%v/v, UPPER: 19%v/v) are hazardous chemicals!

Carefully read the SDS, and use protective action, as recommended! (lab coat, protective goggled and gloves, use a hood, ventilate well, avoid breathing vapours, Keep away from oxidizers, heat and flames, take measures to prevent electrostatic charges...)

Carefully read and follow the SDS for Na_2HPO_4, sodium chloride, and sodium acetate.