

### **BTS** Provides the Best tools for Protein Purification!

## AFFINITY HIS-TAG PURIFICATION

# CHELATING AGAROSE BEADS GENERAL DESCRIPTION

Purification of proteins is a vital part of modern research. Impure extracts generally contain a wide range of proteins with diverse biological functions and different chemistry, which need to be separated.

Affinity Chromatography (IMAC) is the purification technique which is widely used. It is based on the interaction between certain superficial protein residues (histidines, cysteines and to a lesser extent tryptophans), with transition metal cations, forming chelates.

These transition metals are bound through a chemical reaction to the agarose beads, giving the agarose an activation degree.

ABT manufactures two types of chelating beads utilizing standard crosslinked beads and the new highly crosslinked Rapid Run™ beads, both in 6% concentrations. The binding between the metal and the agarose bead is by means of the IDA.

IDA cross-linked Agarose resin consists of iminodiacetic acid groups ligated by stable ether linkages via a spacer arm. IDA is a tridentate chelating agent, covalently coupled to cross-linked agarose beads. This resin is loaded with a divalent metal (Ni<sup>2+</sup>, Cu<sup>2+</sup>, Zn<sup>2+</sup> or Co<sup>2+</sup>). The resulting resin ("ready to use") is ideal for rapid purifications of His-tagged proteins.

In comparison with other chelating resins such as NTA-agarose, the IDA has three sites available for the interaction with metal ions, instead of the four with NTA. IDA resins are usually more easily regenerated allowing a better elution of the fused proteins bound with smaller concentrations of imidazole.

The product range covers 4 different types of metal and two different densities of groups on the beads.

The choice of resin depends on the objectives / priorities for each purification (binding capacity / selectivity) and the type of protein to be purified (easy or difficult to separate).

The easiest strategy is to screen with the correct Test kit, and decide on options based on both the protein and the purpose and objectives of the purification.

The user can optimize the best recovery system by using the test kit approach.

### DEPENDING ON THE OBJECTIVES /PRIORITIES

PRIOR OBJECTIVE	RECOMMENDED TEST KIT	DESCRIPTION	REMARKS
High Binding Capacity	HIGH Density CHELATE KIT	Includes: 2 ml HIGH Density METAL FREE 2 ml HIGH Density NICKEL 2 ml HIGH Density ZINC 2 ml HIGH Density COBALT 40 mini-columns (optional)	In general a High Density of Chelate groups offers a greater number of opportunities for the binding of target-protein, but also increases the possibilities that other proteins will be adsorbed.
High Selectivity	LOW Density CHELATE KIT	Includes: 2 ml LOW Density METAL FREE 2 ml LOW Density NICKEL 2 ml LOW Density ZINC 2 ml LOW Density COPPER 40 mini-columns (optional)	On the other hand, a Low Density of Chelate groups reduces the opportunities for binding for target-protein as well as other proteins present in the extract.

<sup>\*</sup> For laboratory use only. Not for use in diagnostic or therapeutic procedures.



#### • DEPENDING ON THE TYPE OF PROTEIN TO BE PURIFIED

TYPE OF PROTEIN	RECOMMENDED TEST KIT	DESCRIPTION	REMARKS
Proteins easy to separate	NICKEL CHELATE KIT	Includes: 2 ml HIGH Density NICKEL 2 ml LOW Density NICKEL 20 empty mini-columns (optional)	Nickel and Cobalt are the most widely used cations for this type of Chromatography because they provide effective bonding and selectivity for a wide range of proteins.
	NICKEL & COBALT CHELATE KIT	Includes: 2 ml HIGH Density NICKEL 2 ml LOW Density NICKEL 2 ml HIGH Density COBALT 30 empty mini-columns (optional)	In this case, both metals generally provide better "One Step" Purifications.
Proteins difficult to separate	ZINC CHELATE KIT	Includes: 2 ml HIGH Density ZINC 2 ml LOW Density ZINC 20 empty mini-columns (optional)	Although Nickel and Cobalt have excellent properties, sometimes other chelates are more appropriate for particular purifications.  ABT also offers resins charged with Copper and Zinc.
	ZINC & COPPER CHELATE KIT	Includes: 2 ml HIGH Density ZINC 2 ml LOW Density ZINC 2 ml LOW Density COPPER 30 empty mini-columns (optional)	These products allow purifications where Nickel or Cobalt may cause inhibition, or irreversible adsorption of the target-protein.

Once the user has chosen the best option, ABT offers further different product formats:

Spin Columns

Bulk Resins

Pre-Packed Columns

Nickel & Cobalt are the most commonly used metal ion for IMAC purifications. Nickel/ Cobalt Rapid Run™ beads combine the advantages of the metal with the high flow rates of the Rapid Run™ resin. This product is excellent for large scale His-tagged protein purifications.

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