

## 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. 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 ( $\text{Ni}^{2+}$ ,  $\text{Cu}^{2+}$ ,  $\text{Zn}^{2+}$  or  $\text{Co}^{2+}$ ). 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:<br>2 ml HIGH Density METAL FREE<br>2 ml HIGH Density NICKEL<br>2 ml HIGH Density ZINC<br>2 ml HIGH Density COBALT<br>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:<br>2 ml LOW Density METAL FREE<br>2 ml LOW Density NICKEL<br>2 ml LOW Density ZINC<br>2 ml LOW Density COPPER<br>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.                                      |

# AFFINITY HIS-TAG PURIFICATION CHELATING AGAROSE BEADS GENERAL DESCRIPTION



- 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:<br>2 ml HIGH Density NICKEL<br>2 ml LOW Density NICKEL<br>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:<br>2 ml HIGH Density NICKEL<br>2 ml LOW Density NICKEL<br>2 ml HIGH Density COBALT<br>30 empty mini-columns (optional) | In this case, both metals generally provide better "One Step" Purifications.                                                                                                                           |
| Proteins difficult to separate | ZINC CHELATE KIT            | Includes:<br>2 ml HIGH Density ZINC<br>2 ml LOW Density ZINC<br>20 empty mini-columns (optional)                                 | Although Nickel and Cobalt have excellent properties, sometimes other chelates are more appropriate for particular purifications. <a href="#">ABT</a> also offers resins charged with Copper and Zinc. |
|                                | ZINC & COPPER CHELATE KIT   | Includes:<br>2 ml HIGH Density ZINC<br>2 ml LOW Density ZINC<br>2 ml LOW Density COPPER<br>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:

- Bulk Resins
- Pre-Packed Columns
- Nickel Cartridges

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.