

HIS CAPTURE KIT 2

for **ligand strand 2** with green dye **Ga** - only for **Y-structure**

Dynamic Biosensors GmbH
HK-NTA-2 v1.0



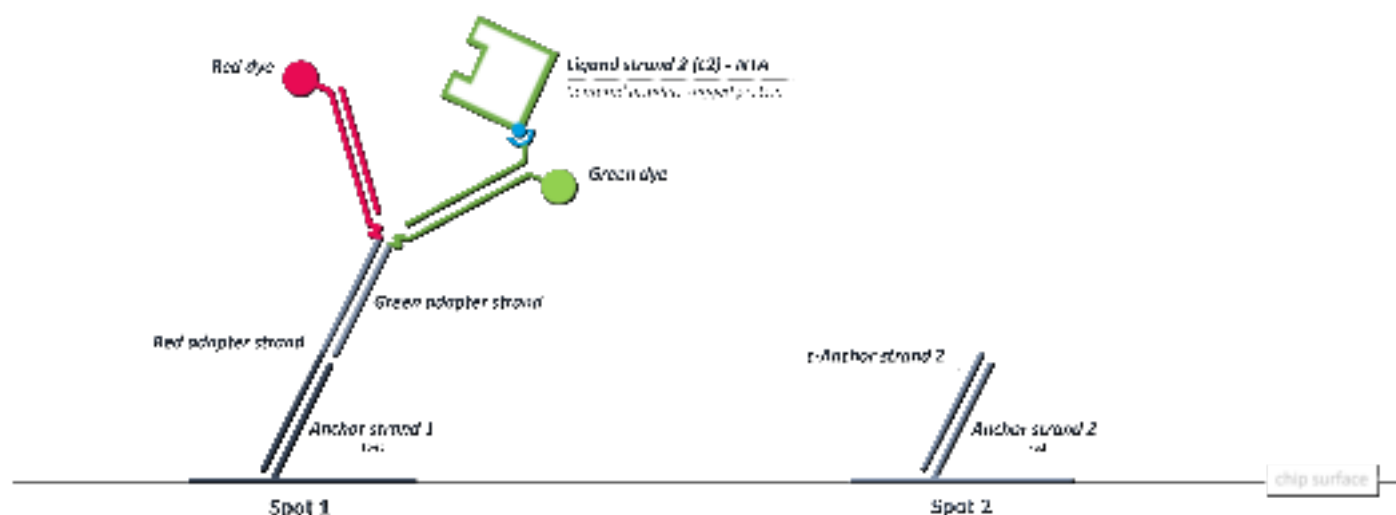
Key Features

- This kit is designed for capture of **histidine-tagged proteins** (His6 or His10) using **Tris-NTA**.
- Includes **Ligand strand 2** with Tris-NTA for **20 regenerations**.
- For functionalization of **ligand strand 2** (green arm) of the **Y-structure**.
- Used in combination with **HK-NTA-1** for the red arm of the **Y-structure**, it is ideal for dimerization projects.
- Homo-/hetero-proteins can be coupled easily to the arms via his-tag capture.
- Compatible with **heliX[®] Adapter Chip**, both **Spot 1** and **Spot 2**

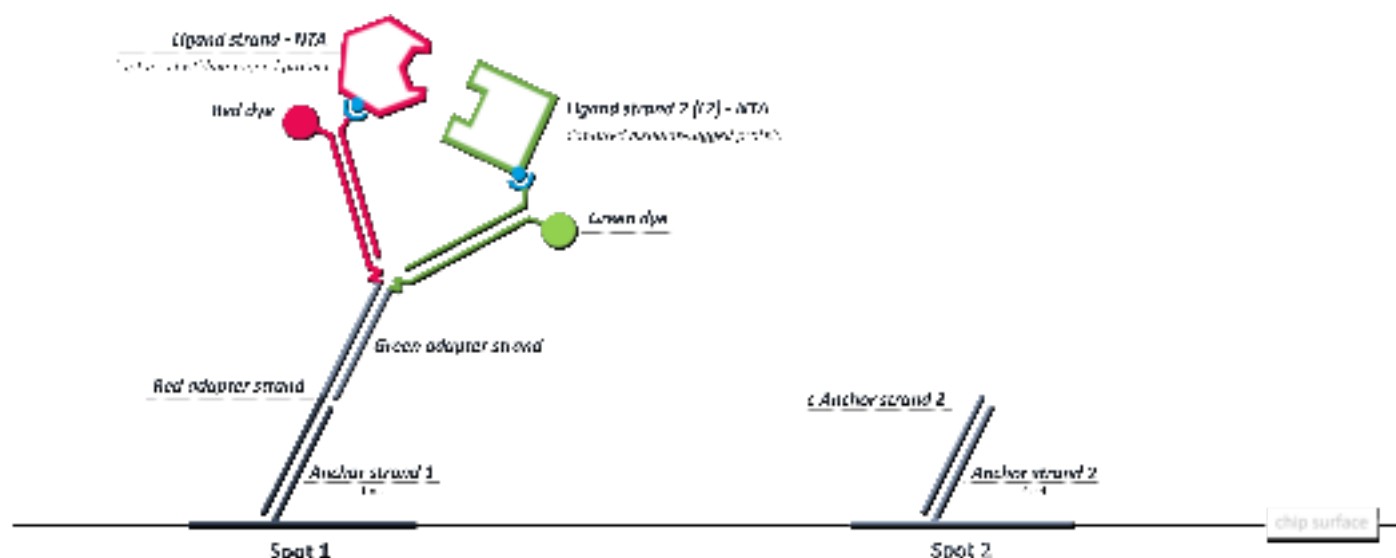
heliX[®] Adapter Chip Overview

2 spots with 2 different anchor sequences for DNA-encoded addressing. Spot 1 is functionalized with the capture molecule while Spot 2 is used as real-time reference.

Single Capture



Double Capture



Product Description

Order Number: **HK-NTA-2**

Table 1. Contents and Storage Information

Material	Cap	Concentration	Amount	Buffer	Storage
NTA - Ligand strand 2	Purple	500 nM	2 x 100 µL	TE40 ^[1]	-20°C
Loading Solution (NiCl₂)	Transparent	10 mM	5 x 1500 µL	TE40 ^[1]	-20°C
Imidazole Solution	Transparent	250 mM	10 x 2000 µL	TE140 ^[2]	-20°C

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This product has a limited shelf life, please see expiry date on label.
After preparation of ready to use solution the expiry date is **6 months**.

Preparation

Step 1

For surface functionalization, the **Y-structure Red Adapter strand** harboring the red dye **Ra** and the **Y-structure Green Adapter strand** harboring the green dye **Ga** need to be pre-hybridized with the **NTA - Ligand strand** and **NTA - Ligand strand 2** in order to build the **Y-structure**. In solution hybridization of **Y-structure** strands:

- Mix **Y-structure Red Adapter strand with Ra** (400 nM), **Y-structure Green Adapter strand with Ga** (400 nM), **NTA - Ligand strand** (500 nM) and **NTA - Ligand strand 2** (500 nM) at a 1:1 ratio (v/v).
- Incubate the solution of step i) at RT for at least **2 hours** to ensure complete hybridization. Overnight incubation at 4°C is also possible.

Step 2

Mix solution of step ii) and **cAnchor strand 2** (100 nM) at 1:1 ratio (v/v).

Step 3

Solution is ready to use for **heliX[®] Adapter Chip** functionalization.

Example

Required volume for one functionalization for double capture on **Y-structure** (Spot 1): **35 µL** with a final concentration of **50 nM**.

Vial 1				Vial 2
Red Adapter strand with Ra (400 nM)	Green Adapter strand with Ga (400 nM)	NTA - Ligand strand (500 nM)	NTA - Ligand strand 2 (500 nM)	cAnchor strand 2 (100 nM)
4.5 µL	4.5 µL	4.5 µL	4.5 µL	18 µL

Assay Setup in heliOS

This specific kit requires a customized method consisting in **His-tag capture** plus **Y-Structure FRET Kinetics**, which is currently not provided among the verified assay. It can be easily created by an advanced **heliOS** user by applying the default parameters already existing in the two different and separate workflows; however, for any help on creating the new method, please contact the support team at support@dynamic-biosensors.com.

TIP

As the stability of his capture is affected by the protein, in case of long dissociations, consider using the classic conjugation approach.

Useful Order Numbers

Table 2. Order Numbers

Product Name	Comment	Order No
heliX[®] Adapter Chip	Chip with 2 detection spots	ADP-48-2-0
Y-Structure Kit 1: for proximity binding assay on Spot 1	For studying ternary complex formation	HK-YS-1
Y-Structure Kit 2: for proximity binding assay on Spot 2	For studying ternary complex formation	HK-YS-2
His Capture Kit: with red dye (Ra)	For capturing of histidine-tagged proteins using Tris-NTA on the red arm of the Y-structure .	HK-NTA-1

Contact

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Instruments and chips are engineered and manufactured in Germany.

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[1] TE40: 10 mM Tris, 40 mM NaCl, 0.05 % Tween20, 50 µM EDTA, 50 µM EGTA

[2] TE140: 10 mM Tris, 40 mM NaCl, 0.05 % Tween20, 50 µM EDTA, 50 µM EGTA