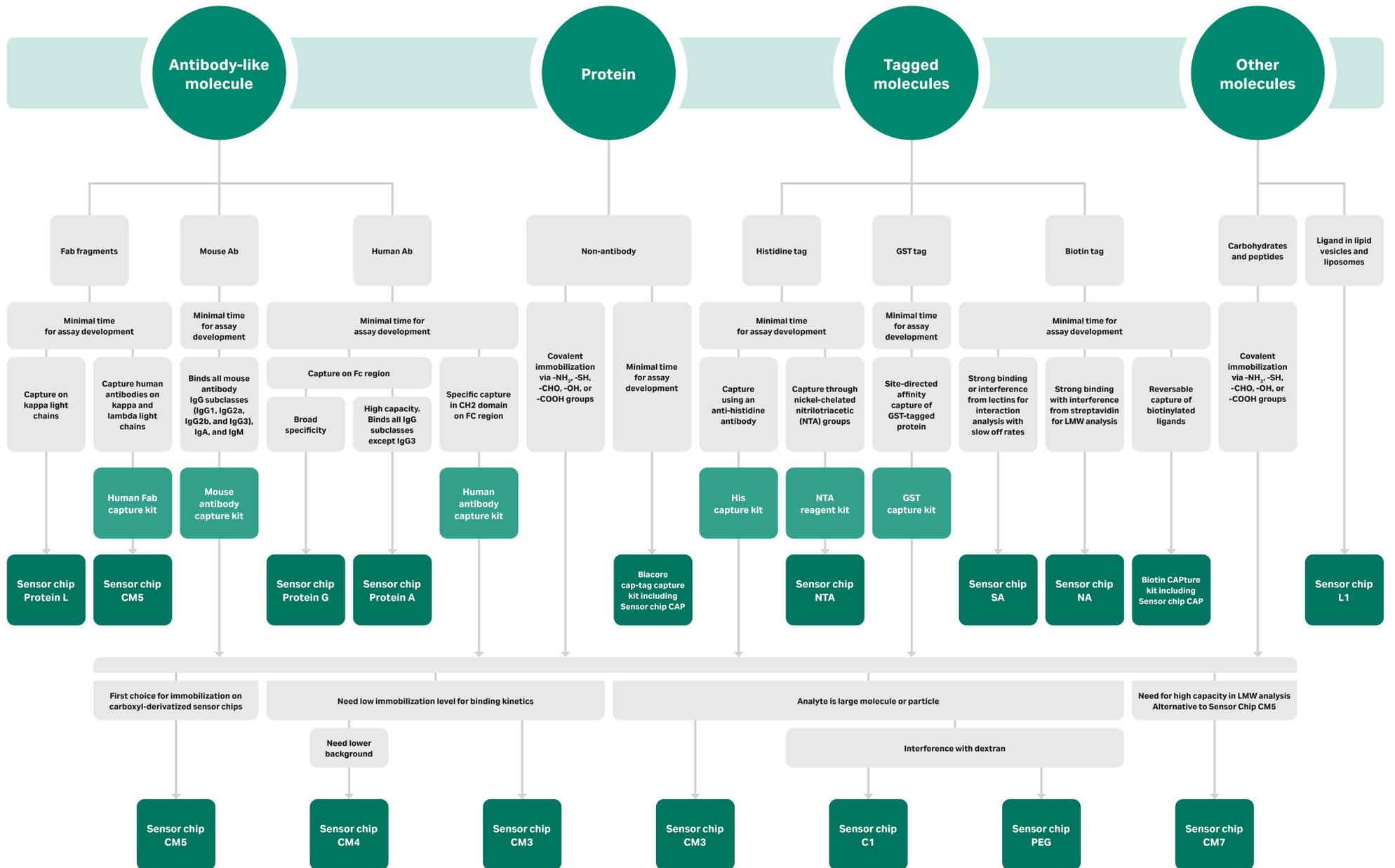


How to choose a suitable attachment approach for your ligand

Save time and effort for your Biacore™ SPR interaction analysis study



Terminology

- Ligand:** interaction partner attached to the surface.
- Analyte:** interaction partner in solution passed over the attached ligand.
- Capture molecule:** binding partner attached to the surface for easy attachment of ligand of interest.
- Prefunctionalized surface:** binding partner immobilized to the surface for easy assessment of analyte binding.

Sensor chip formats

- Sensor chip classic format are designed for Biacore X100, Biacore 3000, and Biacore C SPR systems.
- Series S sensor chip are for use with Biacore 8 series, Biacore 1 series, Biacore S200, Biacore T200, and Biacore 4000 SPR systems.



Which molecule should I attach to the surface?

- Take advantage of affinity tags** that are present on your molecule.
- Valency:** attach the interactant with most binding sites to avoid avidity problems.
- Molecule amount:** attach the interaction partner with lowest amount to save reagents.
- Molecule purity:**
 - Prefunctionalized or capture surfaces: no specific purity consideration needed.
 - Direct coupling: attach the purest interactant to ensure specific binding interactions.
- Molecular weight:** avoid attachment of very large (relative molecular mass, $M_r > 1\,000\,000$) or small ($M_r < 500$) molecules. If significant difference in molecular weight of interacting proteins, attach the smaller protein molecule.

Note: cross-reactivity examination or consideration is always needed in interaction analysis experiments

Direct immobilization or capture approach?

Covalent direct attachment advantages

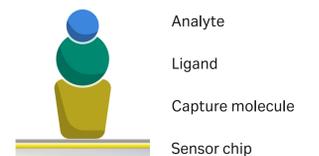
- Consumes less ligand
- Higher binding capacity
- Very stable immobilization with no leakage
- Shorter cycle times

Examples: Amine coupling, ligand-thiol coupling, surface-thiol coupling, maleimide coupling, aldehyde coupling



Capture-based attachment advantages

- Orientation-specific attachment
- Selective capture of ligand from crude samples
- Simpler assay development (regeneration is independent of the captured molecule¹)
- Interactant on the surface is easily changed¹
- For ligands that are difficult to immobilize or regenerate
- For unstable ligands



Examples: see selection guide above.
¹ Except for streptavidin or biotin due to the extremely high affinity