M03E

Innovative and remarkable chemistries, conjugation methods, labeling and functionalisation (PH)

| Hydrazone chemistry Introduction | More info | Conjugation kit | Tagging | Immob.(beads) | Peptide/Oligos | Crosslinkers | Antibody labeling | Organic Synthesis | WB RapidDirect

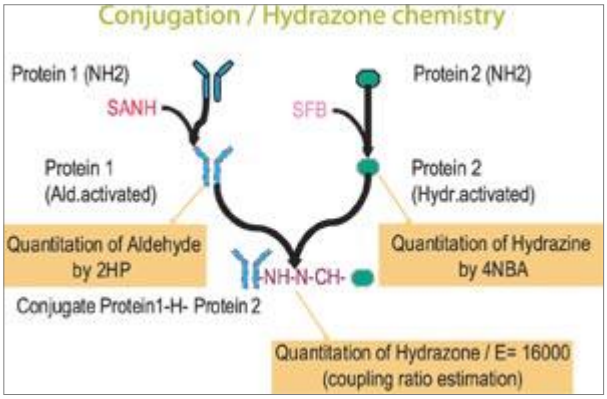
■ Hydrazone Chemistry (Hydralink, Controlled Amine) - Introduction

The new technology to conjugate simply and efficiently your biomolecules

Advantages : efficient and flexible!

- **easy** operating (modify each molecule, mix),
- activate the biomolecules in advance (stable for months),
- **better control** of the coupling ratio,
- **excellent yield** of conjugation,
- **no reduction or de-protection step**,
- **orientated** heteroconjugation **highly selective** (no homoconjuguates) ,
- conjugation keeping the bioactivity of the components, and very stable,
- **flexibility** for many applications / samples.

- label/conjugate proteins
- immobilisation on supports



This technique replaces advantageously the standard methods based on NHS/Maleimide, epoxides, glutaraldehyde, ... and is more flexible and satisfying for various applications, i.e. for conjugation in solution to solid phase synthesis. Its suits perfectly proteins and any biomolecule or support containing amines or derivatized by conventional biochemistry (proteins, peptides, oligonucleotides, cDNAs, carbohydrates, fluorophores, beads, glass, silica ...) .

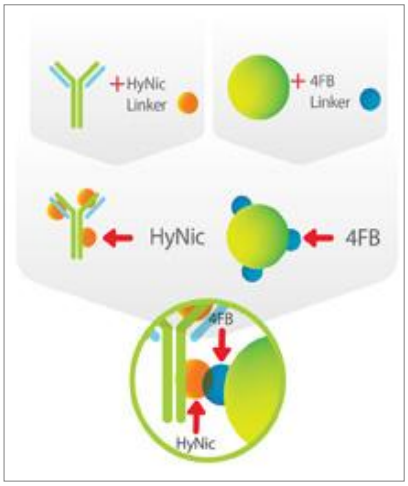
This technology is offered as stand-alone reagents for organic synthesis, peptides, antibodies, nucleic acids conjugation and labeling, but also in convenient conjugation or labeling kits:

All purpose controlled amine conjugation kit | Peptide-oligos conjugation kits | Antibody labeling kits (HRP, AP, RPE, APC)

Immobilisation/supports (agarose beads, magnetic beads)

■ Hydralink™ ControlledAmine Protein conjugation kits

General use linking kits for proteins, peptides, aminated oligos, beads or surfaces. Everything included, no experience needed.



The S-HyNic Conjugation Kit can be used to covalently crosslink any two amine-containing biomolecules. HydraLink™ conjugation chemistry is based on the reaction of a HyNic linker with a 4FB linker to form a stable hydrazone bond. The bond created is both stable and UV-traceable. This unique covalent bond is created when the HyNic linker, incorporated into one type of biomolecule reacts with the 4FB linker, incorporated into the second biomolecule. S-HyNic and S-4FB react only with each other and not with other protein functional groups (selective, oriented and bioorthogonal reaction).

All Purpose SANH Conjugation Kit (S-HyNic) BL150A, 1 kit
Contains SANH and S-4FB activators,and all needed reagents for conjugating to amine-containing molecules. Technical sheet
All Purpose SHTH Conjugation Kit BL152A, 1 kit
Contains SHNH and S-4FB activators,and all needed reagents for conjugating to amine-containing molecules.

Prices and technical sheets on-line

■ Hydralink™ Protein-Oligos conjugation kits

Innovative conjugation chemistry to prepare proteins-oligonucleotide conjugates efficiently and easily. .

- **Easy protocol**: activate, mix, clean-up
- **>95% conversion**
- **UV$_{354\ nm}$-traceable stable ligation**

Advantages
- **Simple and Easy to Use** – Requires only µpipette, µcentrifuge, and UV spectrophotometer
- **Automated Calculations** – Calculator with fully integrated input/output provided
- **High Yield** – 30 - 95% yield based on starting protein (depends on kit/protein)
- **Faster kinetics** for greater efficiency and yields thank to catalyzed conjugation
- **High Purity** – >95% purity without chromatographic purification (kit FV9100 only)
- **High Stability** – Conjugates are 10 times more stable than any other conjugation linker
- **Specificity** – Two-linker method avoids homoconjugate formation
- **Quantifiable** – Using a UV signature wavelength and simple UV scan

▪ The ***Antibody-Oligonucleotide All-in-One Conjugation Kit*** includes buffers, spin columns, and a calculator to determine MSR.
No column chromatography is required.
It generates **high-purity conjugates virtually free of residual antibody or oligonucleotide** (>95% conjugate).

Ask for White Paper: Antibody-Oligonucleotide Conjugate Preparation

How it works?

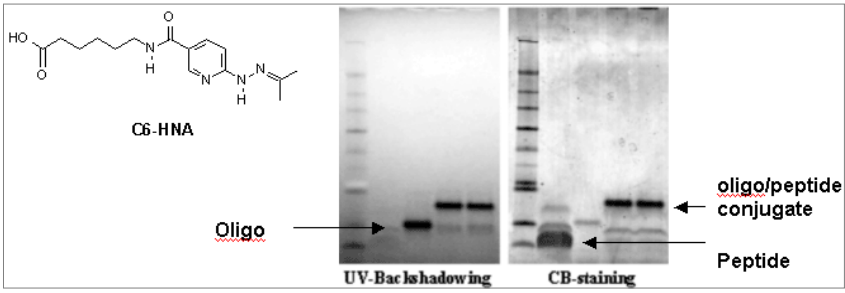
1. Amine-modified, 20 to 60-mer oligonucleotide is modified using an excess of the Sulfo-S-4FB linker2. Polyclonal or monoclonal antibody (100 µg) is modified using the S-HyNic linker.

3. The 4FB and HyNic modified biomolecules are mixed together in the presence of the TurboLink™ catalyst, leading to rapid and conjugation through formation of stable bis-arylhydrazone bonds, followed by magnetic-affinity, solid phase purification.

4. The antibody-oligonucleotide conjugate is ready for use in the most demanding and sensitive applications.

Application: Oligonucleotide-Protein conjugation

PAGE gel demonstrating a 5'-aldehyde modified oligonucleotide (1 equivalent) was reacted with a 15mer peptide that was modified by C6-HNA at N-terminus). Simple addition of the hydrazine-modified peptide to the aldehyde-modified oligonucleotide (lane 1) directly yielded the peptide/oligonucleotide conjugate without the requirement of reducing reagents.



▪ The ***Protein-oligos conjugation kit*** is similar, and contains the material to synthesize two conjugation reactions for any immunoPCR or hybridization-type assay in just under 4 hours each, yielding between 40–60% conjugate after purification. 2 features facilitate easy purification and identification of the conjugate from the excess oligo using size exclusion purification methods such as FPLC or diafiltration: 1/the ligation consist in a **stable bond** (bis-arylhydrazone) with measurable **absorbance at 354 nm**. 2/it yields **>95% conversion** of protein to conjugate when more than 4 molar equivalents of oligo are added.

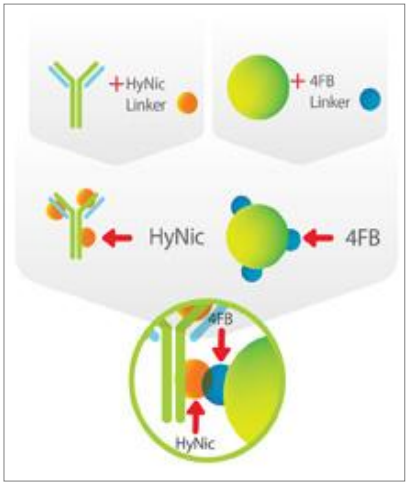
Antibody-Oligonucleotide All-in-One Conjugation Kit **FV9100, 1 kit***
*:the kit conjugates 100ug of antibody; Technical sheet
Protein-oligos All-in-One conjugation kit **IV2490, 1 kit**
*:the kit provide 2 conjugations reactions, Technical sheet

Prices and technical sheets on-line

interchim 211 bis Av. J.F. - BP 1140 03103 Montluçon Cedex - Tel. 33 (0) 4 70 03 88 55 - Fax 33 (0) 4 70 03 82 60
e-mail interchim@interchim.com - web www.interchim.com

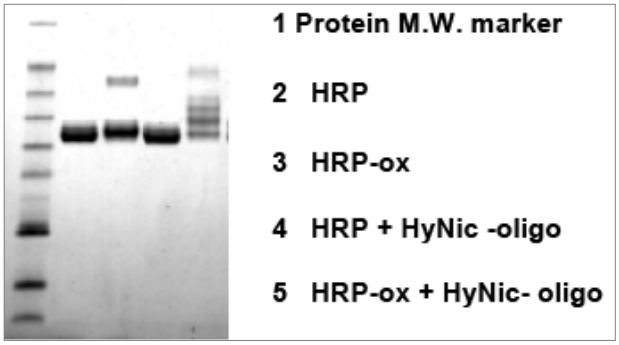
■ Antibody labeling kits

■ HydraLink BioConjugation & Labeling kits (HRP, Fluorescein, PE,...)

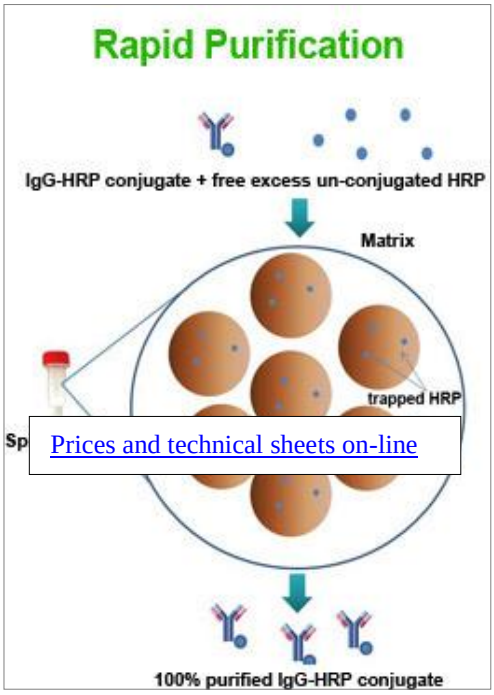


The S-HyNic Conjugation kits are designed to conjugate proteins with pre-activated high-activity markers such as HRP, PE, Biotin. Any suitably pure and sized molecule containing amines (antibody, aminoallyl-oligo can also be conjugated and purified in ~4 hours (30 minutes hands-on time).
All-in-One Conjugation kits contain the activated reagent for HyNic activation of 2 x 100 µg of any user-supplied antibody, the activated marker, and dual purification tool (affinity beads).
One-Shot Conjugation kits contains same reagents but purification tool is Gelfiltration column, while conjugation level can be assessed.

These kits use the unique HydraLink™ conjugation chemistry: the protein to label is activated as a HyNic group using SANH or SHTH reagents, while the marker is activated as an Aldehyde using SFB reagents. This allows more flexibility and control than conventional methods (glutaraldehyde, Mal/NHS, periodate...). Then HyNic and 4FB:
- react only with each other (**no interference** of chemical groups found in biomolecules) in **mild conditions** (gentle chemistry)
- create a unique covalent bond that is both stable and **UV-traceable** (@354nm).



Commassie-stained PAGE gel of a 5'-HyNic-modified oligonucleotide (22-mer) conjugated to periodate oxidized horseradish Peroxidase (Lane 5). Lanes 2 and 3 are HRP and ox-HRP, respectively. Lane 4 is a negative control reaction using non-oxidized HRP



Prices and technical sheets on-line

HRP All-in-One Antibody Labeling Kit **FK1640, Kit** for 2x100µg Ab
" **(Large scale kit)** **FK1641, Kit** for 5 mg Ab
Contains all reagents to conjugate antibody with 4FB pre-activated Horseradish Peroxidase*. Technical sheet
AP All-in-One Antibody Labeling Kit **FK8880, 1 kit** (for 2x100µg Ab)
Contains all reagents to conjugate 2x100µg of antibody with pre-activated Alkaline Phosphatase. Technical sheet
R-PE All-in-One Antibody Labeling Kit **FK8900, 1 kit** (for 2x100µg Ab)
Contains all reagents to conjugate 2x100µg of antibody with pre-activated R-Phycoerythrin*. Technical sheet
*: with affinity magnetic beads + spin filter (for AP Lab.Kit FK8880: 30K spin filter alone)r

R-PE Antibody Labeling Kit **JO2380, 1 kit** (for 2x500µg Ab)
Contains all reagents to conjugate 2x~0.5mg of antibody with pre-activated R-Phycoerythrin and 0.5ml gelfiltration columns. Technical sheet
APC Antibody Labeling Kit **FK8890, 1 kit** (for 2x500µg Ab)
Contains all reagents to conjugate 2x~0.5mg of antibody with pre-activated Allophycocyanin and 0.5ml gelfiltration columns. Technical sheet
Fluorescein One-Shot Antibody Labeling Kit **RJ3201, 1 kit** (for 2x100µg Ab)
Contains all reagents to conjugate 2x~100µg of antibody with NHS pre-activated Fluorescein and 0.5ml gelfiltration columns. Technical sheet

Associated reagents:

RapidDirect™ Primary Antibody polyHRP **1I6151** (for 2x100µg Ab)
Contains all reagents to conjugate 2x100µg of antibody with pre-activated HRP See technical sheet
Modify your primary antibody directly with polyHRP for superior sensitivity. No purification. No secondary antibodies needed, hence minimal background,, eliminate II Ab antibody incubation steps, thereby reduce the time required to complete the protocol, and eliminate any cross-species contamination.
Ideal for IP/WB applications, and for those who use the same antibody for multiple western blots with the additional benefit of time savings.

TurboLink Buffer **HT1820, 1.5ml**
Catalyze the HyNic/4FB reaction to yield stable hydrazonie link..

HRP - 4FB **Inquire**
4FB-modified Horse Radish Peroxidase - Conjugates retain their high enzymatic activity.

AP - 4FB **Inquire**
4FB-modified Alkaline Phosphatase - Conjugates retain their high enzymatic activity

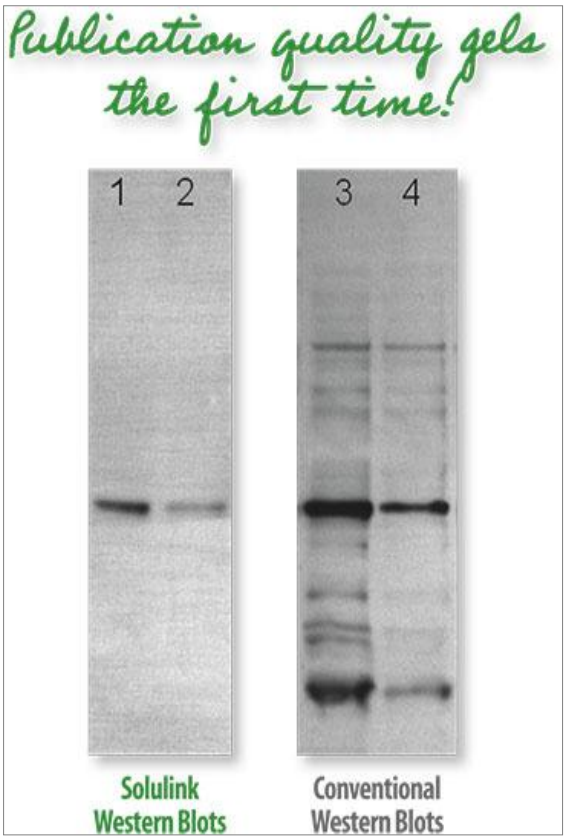
R-PE - 4FB **Inquire**
4FB-modified R-PhycoErythrin in 100mM Sodium Phosphate 150mM NaCL, pH6.0

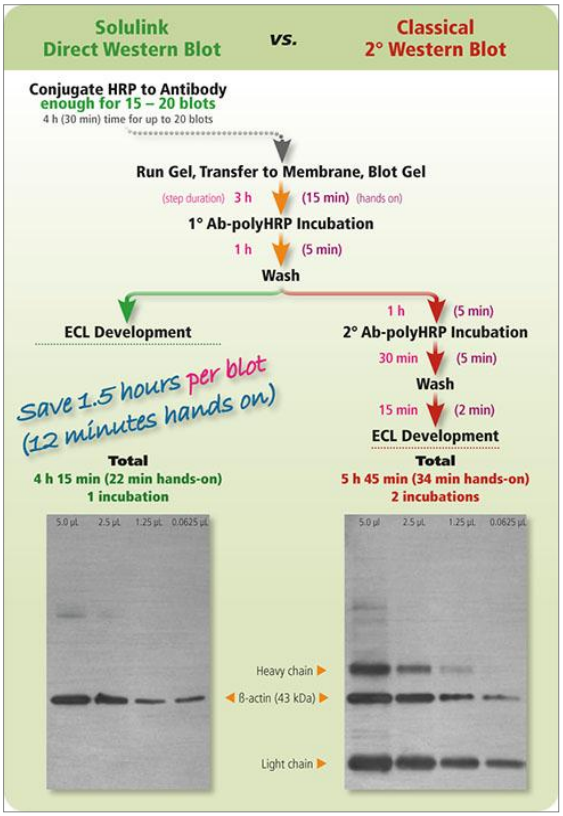
APC - 4FB **Inquire**
4FB-modified AlloPhycoCyanine in 100mM Sodium Phosphate 150mM NaCL, pH6.0.

Fluorescein-PEG$_3$-4FB **Inquire**
Conjugates retain their high quantum yield (~0.98) and are easy to purify using gel filtration methods.

■ RapidDirect™ Western Blot Kits

The NEW RapidDirect™ Western Blot Products, using patented linking technology, attaches multiple HRP proteins to any user-supplied primary antibody, offering several advantages over the use of secondary antibodies for western blot detection.





- **Publication quality** gels the first time
- **Far less background** — does not detect any antibody contaminants on blot, such as light and heavy chains
- **Sensitivity better or equal** to the classical secondary antibody methods in ALL immunoassay formats
- Significant **time savings of 1.5 hours** per blot or IHC tissue staining

Select from Two Versions of Western Blot Kits

■ RapidDirect™ Primary Antibody polyHRP Western Blot Kit

For standard western blots and other immunoassay applications. Modify your primary antibody directly with HRP. No purification. No secondary antibodies needed for downstream applications. Ideal for those who use the same antibody for multiple western blot or immunoassay applications with the additional benefit of time savings.

■ RapidDirect Primary Antibody polyHRP IP/Western Blot Kits

IP/western techniques are typically used to detect low-copy antigens. The primary antibody-HRP/antigen complex is immobilized by α-species IgG immobilized on NanoLink™ magnetic beads (pull-dwon assay), then eluted wit hantigen and analyzed by WB.

Ask for more information about these kits

Prices and technical sheets on-line

RapidDirect™ Primary Antibody polyHRP **1I6151** (for 2x100µg Ab)

Contains all reagents to conjugate 2x100µg of antibody with pre-activated HRP See technical sheet

Modify your primary antibody directly with polyHRP for superior sensitivity. No purification. No secondary antibodies needed, hence minimal background,, eliminate II Ab antibody incubation steps, thereby reduce the time required to complete the protocol, and eliminate any cross-species contamination.

Ideal for IP/WB applications, and for those who use the same antibody for multiple western blots with the additional benefit of time savings.

RapidDirect Primary Antibody polyHRP IP/Western Blot Kits / pull-down assay

with goat anti-mouse	**JO1990** (inquire)
with goat anti-rabbit	**JO2000** (inquire)
with rabbit anti-goat	**JO2010** (inquire)

***Contains all reagents for 10-40 Immunoprecipitations and Western-blots analysis**

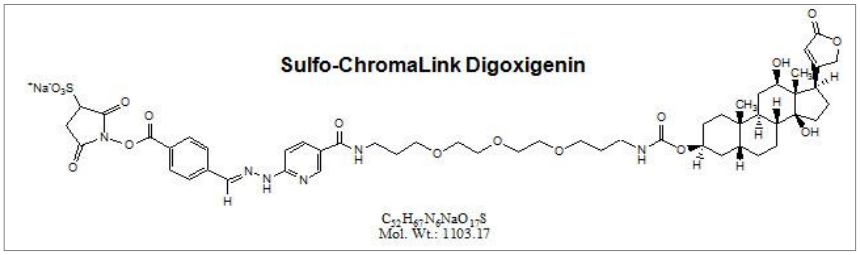
■ Hydralink tagging

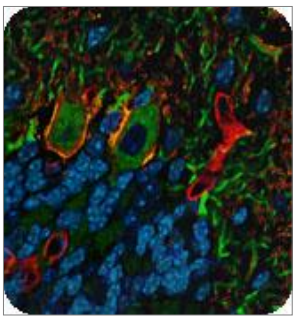
Hydralink technology* allows for tagging chemically any purified biomolecule, extract or supports to yield easy and complete labeling. Affinity tags such as c-Myc or TAT binding moieties are available activated by the hydralink technology.
Bioactive peptides are also available on inquire at interbiotech@interchim.com.
* See the Hydrazone chemistry principle (HyNic and SFB reactions).

■Digoxigenin ChromaLink Labeling

DIG label your Antibody with greater confidence, consistency and reproducibility

UV-Traceable Digoxigenin Linker allows simple UV measurement for fool-proof incorporation of Digoxigenin into any 1' or 2' antibody from any source (Rabbit, mouse, human, IgY)





Goat Primary Antibodies

ChromaLink Digoxigenin OneShot Ab Labeling Kit **FK1630, 1Kit** (labels 100µg Ab)
Complete kit to label 100%g of antibody in just 2 hours. Technical sheet

ChromaLink Digoxigenin reagent **Inquire**

Application: Multi-Color Immunofluorescence Technique using Primary Antibodies raised in the **Same** Host Species

Triple-labeling immunofluorescence in IHC was achieved using 3 different primary antibodies derived from a single host source:
-one primary antibody with biotin (ChromaLink Biotin #BT3614)
-a second primary antibody with DIG (ChromaLink Digoxigenin #DW133A).

■Hydralink tagging

Tags such as c-Myc or TAT binding moieties are available activated by the hydralink technology*, to ensure easy and complete chemical labeling when the genetic expression tagging (cell systems) is not possible or suitable. They can be used for labeling peptides or other biomolecules to be detected using anti tag antibodies, for controls on cells expressing the tag, for preparing tag-affinity supports...
* See the Hydrazone chemistry principle (HyNic and SFB reactions).

- Penetrating peptides

HyNic-TAT **FK5491, 0.5 mg** **FK5492, 1 mg**
HyNic-$(Arg)_8$ **FK5481, 0.5 mg** **FK5482, 1 mg**

- Protein expression reporter peptides:

HyNic-$(His)_6$ Tag **FK5471, 0.5 mg** **FK5472, 1 mg**
HyNic-S-Tag **FK5501, 0.5 mg** **FK5502, 1 mg**
HyNic c-Myc Tag **FK5511, 0.5 mg** **FK5512, 1 mg** **HyNic-Flag Tag** **FK5531, 0.5 mg** **FK5532, 1 mg**

- Bioactive peptides HyNic or 4FB conjugated : (Ghrelin, Obestatin,…)

Custom PepLink Peptides **Inquire at interbiotech@interchim.com**

- Lipids

4FB-DSPE **FK5671, 5x20 mg**
4FB/disteroyl-phosphatidylethanolamide

Related products: other tags

■**Biotin tag**: see section 'biotin labeling', i.e.:
Biotin-PEO_4-NHS **R2027A, 50 mg**
Reactive with amino groups
AB-NTA free acid **BE8210, 100mg**
Reactive with aldehydes and NHS groups

■**Chelate / poly-His tag**: see section 'chelate labeling', i.e.:
Maleimido-C_3-NTA **T3212A, 10 mg**
Reactive with thiol groups.

■**Other tags:** see the section 'protein expression' for transfection methods
■**Anti tags:** see the section 'Secondary antibodies' for labeled anti tag antibodies.
■**tag detection reagents**: see in proteins analysis section 'Fusion tags detection'.

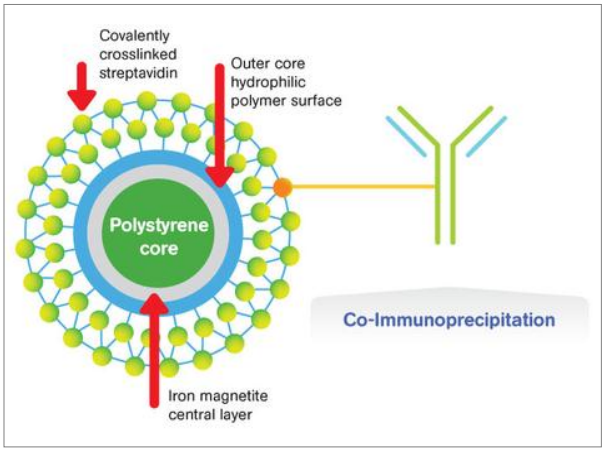
Hydralink supports: resins and beads

Hydralink fuunctionnalized magnetic beads

NanoLink™ Magnetic Beads are polymer encapsulated, 800 nanometer-sized, super-paramagnetic particles.
MagnaLink™ Magnetic Beads are polymer encapsulated, uniform, 2.8µm-sized, super-paramagnetic particles.

NanoLink and MagnaLink beads have the most consistent, ultra-high binding of any particle on the market (>12 nmol/mg), thanks to the proprietary coupling technology.
The high surface area and lower non-specific binding of both Beads are ideal for immobilizations and Co-IP applications.
These microspheres are particularly suited for high throughput robotic applications where high biotin loads must be removed or immobilized without the presence of an iron leachage.

Applications include capture and immobilization of biotinylated biomolecules, such as DNA, RNA, peptides, proteins and antibodies.

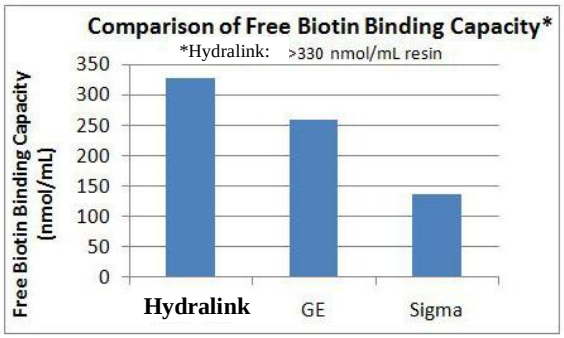


NanoLink 4FB Magnetic Beads 0.8µm, at 10mg/mL	**IO6430, 1mL**		
NanoLink Amino Magnetic Beads 0.8µm, at 10mg/mL	**IO6440, 1mL**		
NanoLink Streptavidin Magnetic Beads 0.8µm, at 10mg/mL	**CP5530, 1mL**	**CP5531, 5ml**	**CP5532, 10ml**
MagnaLink 4FB Magnetic Beads 2.8µm, at 10mg/mL	**IO3320, 1mL**		
MagnaLink Amino Magnetic Beads 2.8µm, at 10mg/mL	**IO6450, 1mL**		
MagnaLink Streptavidin Magnetic Beads 2.8µm, at 10mg/mL	**AWJ460, 1mL**	**AWJ461; 5ml**	**AWJ462, 10ml**
Magnetic Stand (holds 1.5 mL tube for separation of magnetic particles)	**L77191, 1u**		
HyLink™ Glass Slides, HyNic-activated	**DZ1381, 10 Slides**		

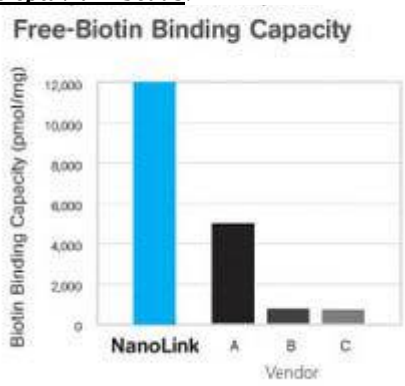
Hydralink immobilization on magnetic beads

NanoLink BeadLink Kit	**IO6470, 1mL**
MagnaLink BeadLink Kit	**IO6470, 1mL**

Streptavidin Agarose beads:



MagnaLink™and NanoLink™ streptavidin beads:



- ●Highest biotin-binding capacity on the market
- ●Binding capacity can be "dialed in"
- ●Lower background noise
- ●Available versions:
  - Streptavidin
  - Amino
  - 4FB

Hydralink immobilization on Agarose beads

Agarose Gel activated by the hydralink technology are available for easy immobilisation of HyNic derivatised ligands, for further capture, depletion or affinity purifications.

Agarose – 4 FB **IV2510, 1ml**
High binding of HyNic derivatised molecules

Agarose – Streptavidin **IO6460, 2mL** **IO6461, 5mL** **IO6462, 10mL**
High biotin-binding streptavidin agarose(>330nmol biotin/ml gel)

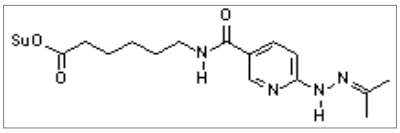
More: see the section 'Purification by affinity'
see the section 'Immobilization of biomolecules'

■ Hydrazone Crosslinkers – for biochemistry

(see principle and benefits of this chemistry).

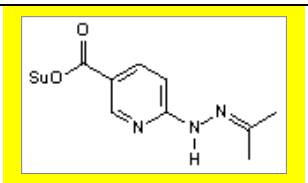
All these reagents are packaged under dry argon, and to store at –20°C for long term. See the Technical sheet

SANH **BL9270 10 mg** **BL9271, 25 mg**
Succinimidyl 4-Hydrazinonicotinate Acetone Hydrazone; MW: 290.2



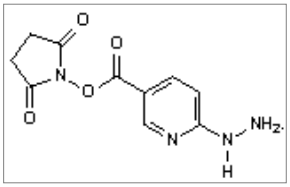
Used to convert primary amines to hydrazinopyridine moieties where protection of the hydrazine is required. The protecting group leaves during formation of the hydrazone conjugate

C6-SANH **BL9330 10 mg** **BL9331, 25 mg**
C6-Succinimidyl 4-Hydrazinonicotinate Acetone Hydrazone. MW: 403.4



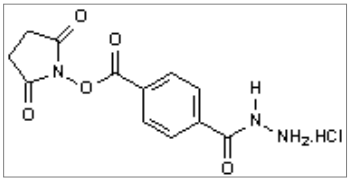
Used to convert primary amines to hydrazinopyridine moieties with an extended six carbon linker where protection of the hydrazine is required. The protecting group leaves during formation of the hydrazoneconjugate.

SHNH **BL9360 10 mg** **BL9361, 25 mg**
Succinimidyl Hydraziumnicotinate Hydrochloride; MW: 286.7



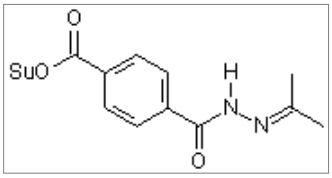
Used to convert primary amines to hydrazinopyridine moieties. Also chelates99mTc.

SHTH **BL9370 10 mg** **BL9371, 25 mg**
Succinimidyl 4-Hydrazidoterephthalate.Hydrochloride; MW: 313.7



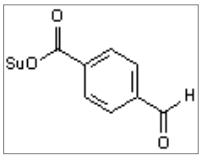
Used to convert primary amines to aromatic hydrazide moieties.

SATH **BL9390 25 mg**
Succinimidyl 4-hydrazidoterephthate acetone hydrazone; MW: 317.4



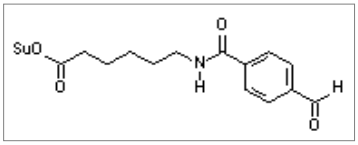
Used to incorporate 4-hydrazidoterephthalamide moieties on proteins or other amine-containing moieties.

SFB **M11771 100mg**
Succinimidyl 4-formylbenzoate; MW: = 247.1



Used to convert primary amines to benzaldehyde moieties.

C6-SFB **BL9410, 25mg** C6-Succinimidyl 4 –formylbenzoate; MW: 360.4



Used to convert primary amines to benzaldeyhde moieties with an extended six carbon linker.

SFB **BI1311 25mg**
SulfoSuccinimidyl 4-formylbenzoate; MW: = 327.3

The sulfo derivative of SFB, for improved water solubility. Especially useful for for conversion of amino surfaces such as beads and plates.

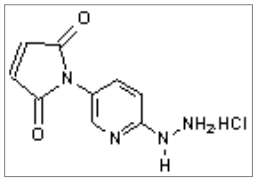
PEO4-SFB (PEG4-SFB) **CP3981 10mg**
SulfoSuccinimidyl 4-formylbenzoate; MW: = 563.5

This derivative of SFB contains an extended and hydrophilic PEO spacer, taht alleviates steric hindrance and increase the conjugate hydrophilicity and stability.

SS-SFB **CP3991 10mg**
Succinimidyl-4-formylbezoate; MW 410.5

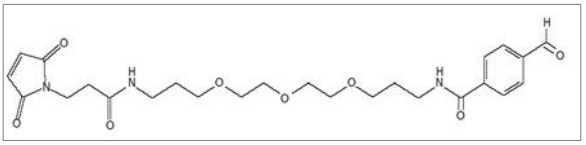
This derivative of SFB contains a S-S link in the spacer allowing thiol mediated cleavage.

MHPH **BL9400 10 mg** **BL9401 25 mg**
5-Maleimido-2-hydraziniumpyridine hydrochloride; MW: 204.2



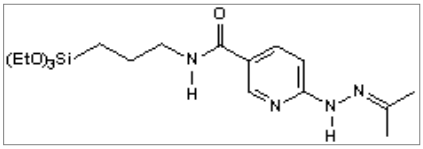
Used to convert thiol moieties to hydrazinopyridine moieties. Its advantages include thiol-reaction specificity, UV-traceability. 2-hydrazinopyridine-modified proteins then react to form stable conjugates in the presence of other (aromatic) aldehyde-modified proteins or biomolecules.

MTFB **BZ0770 10mg** **BZ0774 5x1mg**
MW: 503.5



Used to convert thiol moieties to 4FB (4-formylbenzamide) moieties. Possess a PEG3 linker for increased solubility of modified biomolecule.

Hydrazine-silane **BL9420, 25 mg**
MW: 396.4



Used to incorporate hydrazinopyridine moieties on silica or glass surfaces

Also available: Hydrazine Silane Coated Glass Slides #DZ1381, 10u

2SBA (2-Sulfobenzaldehyde) **A42050, 100mg**
MW: 208.2
Used to quench or cap hydrazone conjugation reactions.

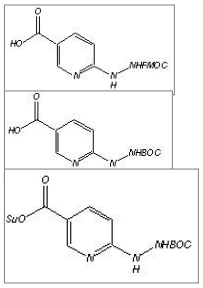
TurboLink Catalist Buffer **HT1820**
Speeds up de reaction between 4FB and HyNic.

Hydrazone linkers for organic synthesis

The following reagents are designed for modifying biomolecules and oriented conjugation according hydrazone chemistry. (see principle and benefits of this chemistry). See the Technical sheet

6-FMOC-HNA **BL9740, 100mg BL9741, 500mg**
6-FMOC-hydrazinonicotinic acid; MW: 375.2
Used to incorporate hydrazine moieties during solid or solution phase peptide synthesis.

6-FMOC-HNA-Osu **BL9760, 100mg BL9761, 500mg**
Succinimidyl 6-FMOC-hydrazinonicotinate; MW: 472.2
Used to incorporate hydrazine moieties during solid or solution phase peptide synthesis.

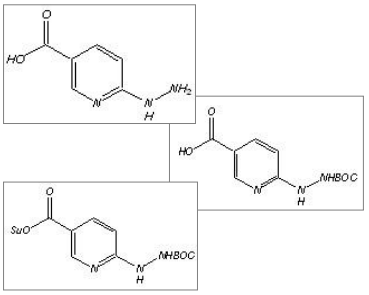


6-BOC-HNA **BL9750, 100mg BL9751, 500mg**
6-BOC-hydrazinonicotinic acid; MW: 253.1
Used to incorporate hydrazine moieties during solid or solution phase peptide synthesis.

6-BOC-HNA-OSu **BL9770, 100mg BL9771, 500mg**
Succinimidyl 6-BOC-hydrazinonicotinate; MW: = 350.3
Used to incorporate hydrazine moieties during solid or solution phase peptide synthesis.

C6-HNAA **BL9780, 100mg BL9781, 500mg**
6-hydrazinonicotinic acid acetone hydrazone; MW: 306.4
Used to incorporate protected hydrazine moieties with extended six carbon linker during peptide synthesis

HNA **BL9790, 100mg**
6-Hydrazinonicotinic Acid; MW: 153.1
Precursor molecule.



BOC-HTA **BL9810, 100mg BL9811, 500mg**
(4-BOC-hydrazido) terephthalic acid; MW: 280.3
Precursor molecule.

BOC-HTA-OSu **BL9820, 100mg BL9821, 500mg**
Succinimidyl 4-BOC-hydrazido) terephthalate; MW: 377.4
Precursor molecule.

4FB-Amidite **FV7031, 250mg**
Building block for nucleic acids synthesis / modification.

More about Hydrazone chemistry

Principle:

The method includes one (or 2 separate) activation step(s) of amine, thiols, or silanol to aromatic functionalities, aldehyde (4SFB) and hydrazine (HyNic) able to form an hydrazone bond. The level of activation is fully controllable using chemical groups quantitation reagents (4NBA #BL9650 for hydrazines, 2HP #019022 for aldehydes). Then the modified molecules are simply mixed to yield a stable conjugate. The hydrazone bound formed is fully stable, in contrast to the hydrazone formed by more commonly accessible hydrazides (unstable acyl hydrazones). It is the only known example of a stable Schiff base, which requires no additional steps (reduction) to stabilize the bond.

Applications:

Hydrazone chemistry provides additional benefits for surface modifications, compared to conventional methods such a glutaraldehyde (that is not oriented chemistry) or other aldehyde (4hours procedure), SMCC (Mal-NHS) (that has poor yield), CNBr activation (that yields a charged bond): it is specific and flexible, controllable activation steps, UV-traceability, and quick (2 hours).

HydraLink conjugation system is a privileged new method to conjugate and immobilize a variety of biomolecules, including peptides, peptides, carbohydrates and nucleic acids. It is covered by US patent 5.206.370. 5.420.285. 5.753.520 and 5.769.778. and EU Patent 0.384.769). The involved reaction is highly selective and mild, derivatized molecules are stable and non-susceptible to non-specific binding. Hydrazone link is kinetically and metabolically stable analog of a cysteine bridge.
It is a superior alternative to step-wise methods, in which difficulties come from the need to separate the modified or conjugates molecules from excess modifiers, unavailable reactive groups requiring tedious activation steps or labor-intensive site specific engineering methods, from intramolecular undesired crosslinking, and from heterogeneous conjugation at the molecular level.

Comparison of conjugation methods:

	Hydrazine/carbonyl	Avidin/Biotin	Maleimide/Thiol
Stability of Activated biomolecules	+++	++	-
High selectivity	+++	+++	+
No reticulation	+++	+	-
No undesirable covalent modification	+++	+++	-
No non-specific binding of conjugate	++	+	++
No need of reductant	+++	+++	-
Covalent (stable) linkage	++	(non cov)	++
Fast reaction kinetics	+++	+++	++
Suitable to a variety of biomolecules&support	+++	+	+
Amenable to solid phase synthesis	+++	++	+
Reproducible/adjustable coupled ratio	+++	++	+
Scalable	+++	-	++
pH range (optimum)	3-(4.7)-7, 5-11	4-7.5	

The technology is ideal for:
-protein to protein conjugation (see Conjugation Kit #BL150A)
-peptide to oligonucleotides conjugation (see Conjugation Kit #IV2490)
-immobilization of peptides and nucleic acids onto microarrays, microplates. (see here)
-organic synthesis of peptides and nucleic acids (see here)
-other biomolecule conjugation (see application below)
-labeling biomolecules by enzymes, fluorophores (see All-In-One kits) or tags (see here).

Related products lines

Interbiotec - BioSciences innovation – proposes a complete range of products for protein biochemistry.

Innovative and remarkable chemistries, conjugation methods, labeling and functionalisation (PH)

- **Standard Click Chemistry reagents** (PH)
- **Copper-free Click Chemistry reagents** (DBCO reagents) (PH)
- **Staudinger reaction** (effective conjugations/chemical modification) (PH)

- **PEGylation reagents** (conjugation reagents, linkers and building blocks) (PH)
- **SDA reagents** (effective photo reactions) (PH)
- **STELLA labeling** (azocycloaddition reactions) (PH)

- **SAM reagents** (Self-Assembled Monolayers for surface modification) (PH)

- **Gold nano-particules and materials**
- **Carbone nanotubes**
- **ITO slides** (PH)

- FluoProbes labeling agents

Desalting tools – CelluSep tubings, SpectraPor tubings, GebaFlex, FloatALyser, SlideALyser,...

Products HighLights Overview

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