Iris Biotech

# LINKEROLOGY®

Version: IB6_4

# Empowering Peptide Innovation

With this guiding theme in mind, Iris Biotech's mission is to support researchers by supplying

- innovative technologies,
- rare compounds,
- as well as a broad portfolio on standard consumables,

available in flexible quantities from small scale to bulk quantities. To fulfill our dedication "Empowering Peptide Innovation", we are attending various conferences, symposia, and exhibitions each year. This allows us to remain in direct contact with scientists all over the world, both from academia and industry, to exchange knowledge, and to gather new ideas to tackle your current challenges.

Guided by our dedication to provide

- competent service,
- as well as novel substances and
- latest technologies,

Iris Biotech is your trusted partner for the world of peptides, while having strong expertise in associated disciplines. Thus, our portfolio comprises reagents and tools for the synthesis and modification of peptides, e.g., amino acids, resins and solvents but also for related technologies such as drug delivery, linkerology® and life sciences.

Owed to the growing demand for tailor-made compounds, our portfolio is fine-tuned by our custom synthesis service at Iris Biotech Laboratories. Our skilled scientists offer profound expertise in

- *de novo* route development,
- upscaling towards larger scale production,
- as well as synthesis optimization for increased efficiency.

Examples are the synthesis of rare chiral building blocks, unnatural amino acid derivatives, sophisticated orthogonal protecting groups, heterocycles, building blocks for nucleotides, PEGs and PEG-analogs as well as specific linkers for controlled drug delivery and release.

***Amino Acids*** ***Building Blocks*** ***Life Sciences*** ***Drug Delivery*** ***Reagents*** ***Resins*** ***Linkerology®*** ***Click Chemistry***

# Portfolio Overview

## Peptide Synthesis and Modification

### (Protected) Amino Acids

Standards such as Fmoc-D/L-AAA and Boc-D/L-AAA, Smoc amino acids for peptide synthesis in water, variety of protecting groups (e.g., Pbf, Trt, $^{t}$Bu, Bzl, Acm, Mob, SIT, Phacm, Allocam, Mmt), unusual amino acids, fluorinated derivatives, substituted prolines, arginine analogs

### Building Blocks

Amino alcohols, amino aldehydes, diamines and hydrazines, (pseudoproline) dipeptides, polyamines and spermines, fatty acid derivatives, peptide nucleic acids (PNAs)

### Reagents

Coupling reagents, solvents and scavengers, protecting groups

### Resins

Preloaded resins (e.g., based on Trityl, TCP, TentaGel, Methoxybenzhydryl, Merrifield, PAM, Rink, Wang), scavenger resins, hydrazone resins, poly(acrylamide) resins, Cyclover

## Linkerology® and Drug Delivery

### Linkers for Solid Phase Peptide Synthesis

### Cleavable Linkers

Val-Ala-based, Val-Cit-based, disulfide-based, Dde-helping hands, pH-sensitive linkers

### Photo-Activatable Linkers

### Functionalized Linkers

Clickable linkers, trifunctional linkers, linkers with maleimide function, cross-linkers, selective N-term acylation and biotinylation, 5HP2O

### PROTACs

Ligands, linkers & modules

### Fullerenes, Poly(2-oxazolines), Dextrans & Plant-Derived Cholesterol

### Superparamagnetic Iron Oxide Nanoparticles

### Poly-Amino Acids

Poly-Arg, Poly-Glu, Poly-Lys, Poly-Orn, Poly-Sar

### PEGylation

Branched PEGylating reagents, (amino-)PEG-acids, PEG-amines & hydrazides & guanidines, reagents for Click-conjugation, Biotin-PEG-reagents, PEG-thiols, PEG-maleimides, other PEGylating reagents

## Life Sciences

### Biotinylation Reagents

### Carbohydrates

Galactose, Glucose, Mannose, Xylose and others

### Drug Metabolites

### Peptides

### Substrates & Inhibitors

E.g., protein kinase inhibitors, substrates for fusion (Halo/Snap/Clip)-tagged proteins

### Natural Products

### Dyes and Fluorescent Labels

E.g., ICG, AMC, DAPI

### Maillard & Amadori Reaction Products

Large portfolio of derivatives useful as standards for food, pharma and cosmetics industry

### Vitamins

# Custom Synthesis

Your project requires a compound not listed in our portfolio?
Get in contact and inquire about our custom synthesis capabilities.

**Our experienced scientists are excited to accept your synthetic challenge!**
In such cases, your request undergoes the following stages:

### Step-by-Step Analysis

- Customer's demands

### Process Evaluation

- Detailed literature review
- Synthetic possibilities

### Strategy Development

- Protocol development
- Method development and validation
- Customized synthesis

### Quality Consistency

- Identity confirmation
- Purity verification

# Our Service Promise

All our services are based on high standards, transparency & documentation, trust, honesty & confidentiality, as well as the required know-how.

### High Standards

- Values: sustainability & responsibility
- State-of-the-art equipment & latest technologies
- High quality standards
- Qualified suppliers & regular audits

### Transparency & Documentation

- Talk to our specialists – customer care
- Certificates of analysis & origin
- Impurity profiling
- Safety data sheets
- Analytical and process reports

### Trust, Honesty & Confidentiality

- Intergenerational business valuing partnerships
- Meeting the customer's expectations
- Integrity towards our customers

### Our Know-How

- One-step reactions & complex multi-step synthesis
- Scalability from mg to kg quantities
- Route scouting

# Table of Contents

# 1. The Concept of Antibody-Drug Conjugation (ADC)

## 1.1. Technical and Market Background

Conjugating highly potent small molecules to vastly target specific biomolecules, like antibodies, has become a modern and sophisticated approach, particularly in the field of cancer therapy. The list of ADCs in clinics continues to grow, bolstered by the success of two pioneers in this field:

***Adcetris®*** (Seattle Genetics) has been approved in 2011 for the treatment of Hodgkin's lymphoma and systemic anaplastic large cell lymphoma (ALCL) and reached $476.9 million sales per year in 2018. This drug is composed of a monoclonal antibody targeting CD30 conjugated to four molecules of monomethyl auristatin E *via* a self-immolative linkage (*Fig. 1*). Reduction of interchain disulfide bonds provides reactive cysteine residues, which are then conjugated with maleimide payload linker systems, yielding the final drug compound.

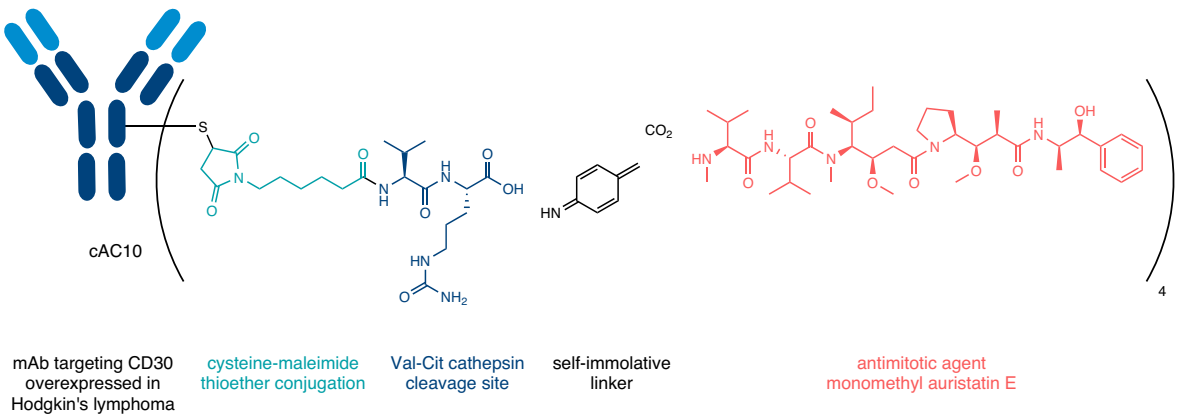


*Fig. 1: Composition of Adcetris®, one of the first FDA-approved ADCs.*

***Kadcyla®***, another pioneer in this field, has been approved in 2013 for the treatment of HER-2 positive metastatic breast cancer and reached $981 million sales per year in 2018. In this case, payloads are conjugated to surface accessible lysines resulting in a heterogeneous modification of the core antibody.

***Reference:***

→ *Antibody-drug conjugates in tumor therapy; B. Sammet, C. Steinkuhler, N. Sewald;* ***Pharm Pat Anal*** *2012;* ***1****: 65-73.* https://doi.org/10.4155/ppa.12.4

## ADCs – Mode of Action

The typical mode of action of ADCs is shown in *Fig. 2*. An ADC circulates in plasma until it reaches the target cell. The antibody portion of an ADC then nails to a cell-surface antigen that is ideally specific to a cancer cell. Upon binding, the ADC-antigen protein complex becomes internalized into the cancer cell. When the complex is degraded, it releases the cytotoxin which then binds to its target to cause cancer cell apoptosis. The linker between antibody and payload is typically either permanent or cleavable by hydrolases, such as the protease cathepsin B, by glucoronidases or through reductive conditions and the presence of glutathion.

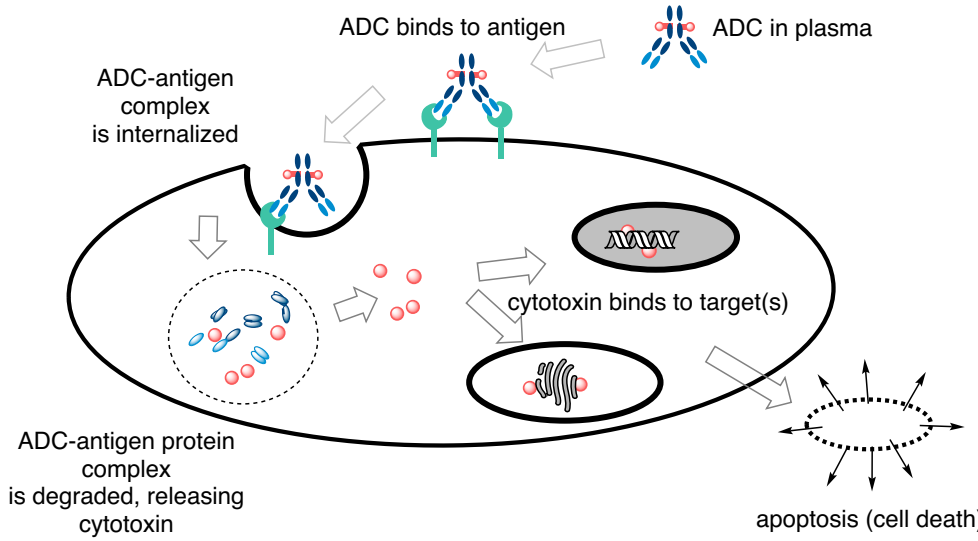


*Fig. 2: Mode of action of ADCs.*

This concept is a sophisticated approach combining the high specificity of antibodies with the high potency of (small) drug molecules. The disadvantages of antibodies, like low potency, as well as the drawbacks of small drug molecules, like low specificity accompanied by high toxicity through many side effects, are compensated by the advantages of the other counterpart. A smart synergistic combination of both elements significantly enlarges the narrow therapeutic window of a small drug molecule between minimum (efficacious) and maximum (toxic) dosage (*Fig. 3*). ADC drugs expand the therapeutic window, as they can increase efficacy and decrease toxicity in comparison to traditional chemotherapeutic cancer treatments. Targeted delivery to cancer cells increases the amount of dosed drug reachingthe tumor, thus lowering the minimum effective dose (MED). The MTD (maximum, highest tolerable dose without serious side effects) is increased, as less drug compound reaches healthy, non-target tissues. Historically, defining MTD was the primary objective of phase 1 oncology trials. More recently, especially for new targeted drugs (including ADCs), emphasis has been placed on determining the recommended phase 2 dose (RP2D), which better captures chronic toxicities emergent after multiple treatment cycles (e.g., edema, effusion, pneumonitis, ocular toxicities) and certain grade 2 side effects (e.g., diarrhea, mucositis, cytopenia, neuropathy, severe fatigue) that may become intolerable over time. However, interpreting small molecule and ADC doses in clinical trials requires specific interpretation and accurate conversion of the doses to a common unit and expansion of the therapeutic window needs to be discussed individually. An appreciation that ADCs do not significantly enhance the MTDs of their payloads may provide insight into several existing observations in this field, like ADCs that feature a common drug linker often encounter similar MTDs because of payload-associated platform toxicities, independent of the target antigen. This highlights that most off-target adverse events are antibody independent.

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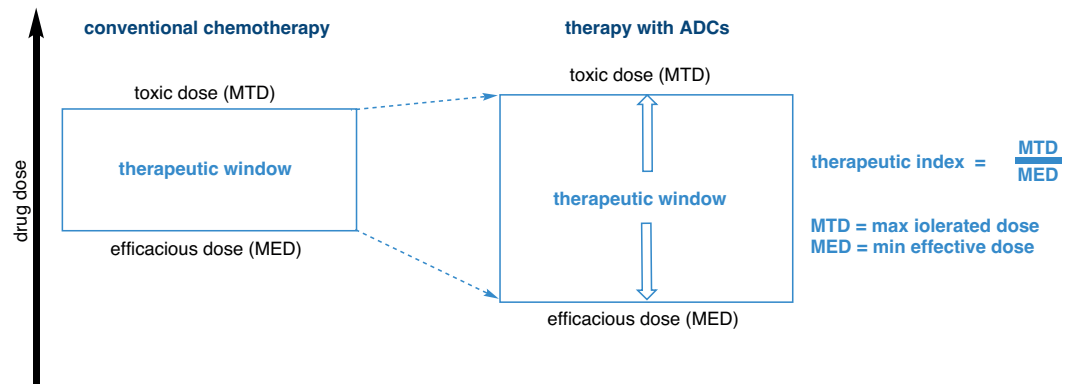


*Fig. 3: The therapeutic window is significantly enlarged using ADCs compared to small-molecule drugs.*

***References:***


- *L. Anthony, (2019). ADC Landscape Review 2019 [PowerPoint slides]. Retrieved from http://worldadc-usa.com*
- *Design and Synthesis of Tesirine, a Clinical Antibody-Drug Conjugate Pyrrolobenzodiazepine Dimer Payload; A. C. Tiberghien, J. N. Levy, L. A. Masterson, N. V. Patel, L. R. Adams, S. Corbett, D. G. Williams, J. A. Hartley, P. W. Howard;* ***ACS Med Chem Lett*** *2016;* ***7****: 983-987. https://doi.org/10.1021/acsmedchemlett.6b00062*
- *Recent advances of antibody drug conjugates for clinical applications; P. Zhao, Y. Zhang, W. Li, C. Jeanty, G. Xiang, Y. Dong;* ***Acta Pharmaceutica Sinica B*** *2020;* ***10(9)****: 1589-1600. https://doi.org/10.1016/j.apsb.2020.04.012*
- *The therapeutic window of antibody drug conjugates: A dogma in need of revision; R. Colombo, J. R. Rich;* ***Cancer Cell*** *2022; 40. https://doi.org/10.1016/j.ccell.2022.09.016*
- *Threapeutic index improvement of antibody-drug conjugates; H.-P. Gerber, S. Gangwar, A. Betts;* ***Mabs*** *2023;* ***15(1)****: 2230618. https://doi.org/10.1016/j.ccell.2022.09.016*


While initially only small molecules or short peptides have been used as payloads, the panel of conjugates has opened to chelators for radioactive nuclides and larger biomolecules, such as toxic enzymes. Additional variations have been introduced on the antibody side by utilizing antibody fragment combinations or diabodies (*Fig. 4*).

Points of conjugation are typically the thiol groups of cysteines, the amino functions of lysines or the N-terminus of a monoclonal antibody. Due to the inherent heterogeneity of conjugation to the multiple amines or cysteines found in mAbs, significant research efforts are directed toward the production of discrete, homogeneous ADC products *via* site-specific conjugation. This may involve genetic engineering of the mAb to introduce discrete, available cysteines or non-natural amino acids with an orthogonally reactive functional handle such as an aldehyde, ketone, azido, or alkynyl tag. These site-specific approaches increase the homogeneity of ADCs and enable novel bioorthogonal chemistries which utilize, reactive moieties rather than thiols or amines. This broad diversity of applicable linkers can then be utilized leading to improved design in future generations of ADCs.

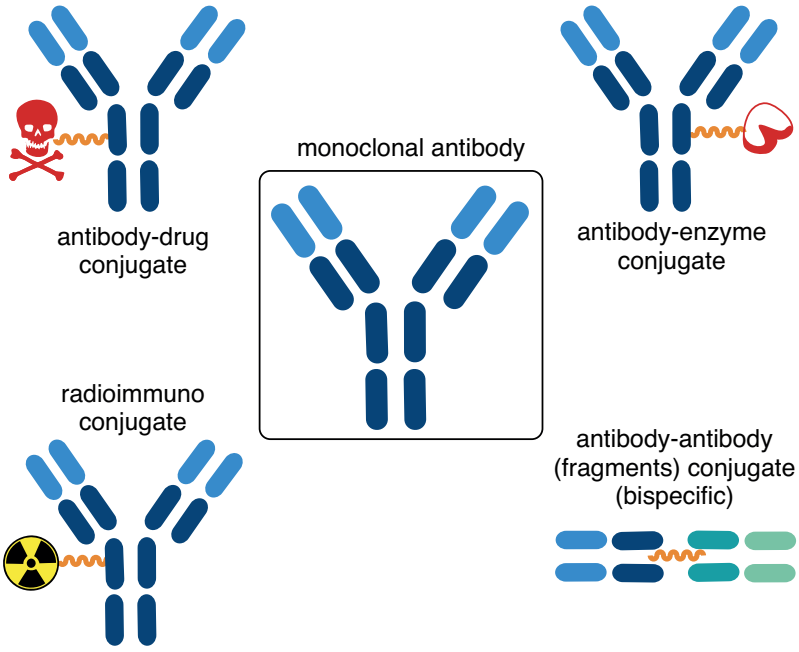


*Fig. 4: The concept of antibody-drug conjugation can be extended from conjugations of small cytotoxic molecules to conjugation with chelators for radionuclides, proteins or with antibody fragments.*

*Tab. 1: Global Approved ADCs (Jan. 2025).*

| Approval Year | (Trade) Name | Payload | Linker |
|---|---|---|---|
| 2011 | Adcetris | MMAE | enzyme cleavable (vc) |
| 2013 | Kadcyla | DM1 | non-cleavable thioether |
| 2017 | Besponsa | Calicheamicin | acid cleavable disulfide |
| 2017 | Mylotarg | Calicheamicin | acid cleavable disulfide |
| 2018 | Lumoxiti | Pseudomonas exotoxin A fragment (PE38) | fused peptide linkage |
| 2019 | Enhertu | Deruxtecan | enzyme cleavable (ggfg) |
| 2019 | Padcev | MMAE | enzyme cleavable (vc) |
| 2019 | Polivy | MMAE | enzyme cleavable (vc) |
| 2020 | Trodelvy | SN-38 | acid cleavable *p*-aminobenzyl carbonate |
| 2020 | Blenrep | MMAF | non-cleavable (vvi) |
| 2020 | Akalux | IR700 | non-cleavable |
| 2021 | Tivdak | MMAE | enzyme cleavable (vc) |
| 2021 | Zynlonta | PBD dimer | enzyme cleavable (va) |
| 2021 | Aidixi | MMAE | enzyme cleavable (vc) |
| 2022 | ELAHERE | DM4 | non-cleavable |
| expected for 2025 | *Datopotamab deruxtecan* | Deruxtecan | enzyme cleavable (ggfg) |
| expected for 2025 | *Patritumab deruxtecan* | Deruxtecan | enzyme cleavable (ggfg) |
| expected for 2025 | *Telisotuzumab vedotin* | MMAE | enzyme cleavable (vc) |

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***References:***

- *Site-specific antibody drug conjugates for cancer therapy; S. Panowski, S. Bhakta, H. Raab, P. Polakis, J. R. Junutula;* ***MAbs*** *2014;* ***6****: 34-45.* https://doi.org/10.4161/mabs.27022
- *Advances in Precision Oncology: Targeted Thorium-227 Conjugates As a New Modality in Targeted Alpha Therapy; U. B. Hagemann, K. Wickstroem, S. Hammer, R. M. Bjerke, S. Zitzmann-Kolbe, O. B. Ryan, J. Karlsson, A. Scholz, H. Hennekes, D. Mumberg , A. S. Cuthbertson;* ***Cancer Biother Radiopharm*** *2020;* ***35(7)****: 497-510.* https://doi.org/10.1089/cbr.2020.3568

### Background Information

| | | |
|---|---|---|
| **$IC_{50}$**<br>**Inhibitory Concentration**<br>Concentration causing 50% of maximal inhibition of the desired activity. | **$EC_{50}$**<br>**Effective Concentration**<br>Concentration causing 50% of maximal response of the desired effect. | **$ED_{50}$**<br>**Effective Dose**<br>Dose causing the desired effect in 50% of individuals. |
| **$GI_{50}$**<br>**Growth Inhibition**<br>Concentration causing 50% inhibition of cell proliferation/cell growth. | **$TC_{50}$**<br>**Toxic Concentration**<br>Concentration causing a defined toxic effect in 50% of individuals. | **$TD_{50}$**<br>**Toxic Dose**<br>Dose causing a defined toxic effect in 50% of individuals. |
| **$CC_{50}$**<br>**Cytotoxic Concentration**<br>Concentration killing 50% of cells. | **$LC_{50}$**<br>**Lethal Concentration**<br>Concentration killing 50% of individuals. | **$LD_{50}$**<br>**Lethal Dose**<br>Dose killing 50% of individuals. |

## 1.2. Linker Design, Connectivity, Degradability, and Drug-Antibody Ratio (DAR)

Antibody-drug conjugates (ADCs), which combine the specificity, favorable pharmacokinetics, and biodistribution of a monoclonal antibody (mAb) with the cytotoxic potency of a drug are promising new therapeutics for cancer. Along with the development of mAbs and cytotoxic drugs, the design of the linker is essential, as it impacts the efficacy and tolerability of ADCs. The linker needs to provide sufficient stability during systemic circulation while providing rapid and efficient release of the cytotoxic drug in its active state inside the tumor cells.

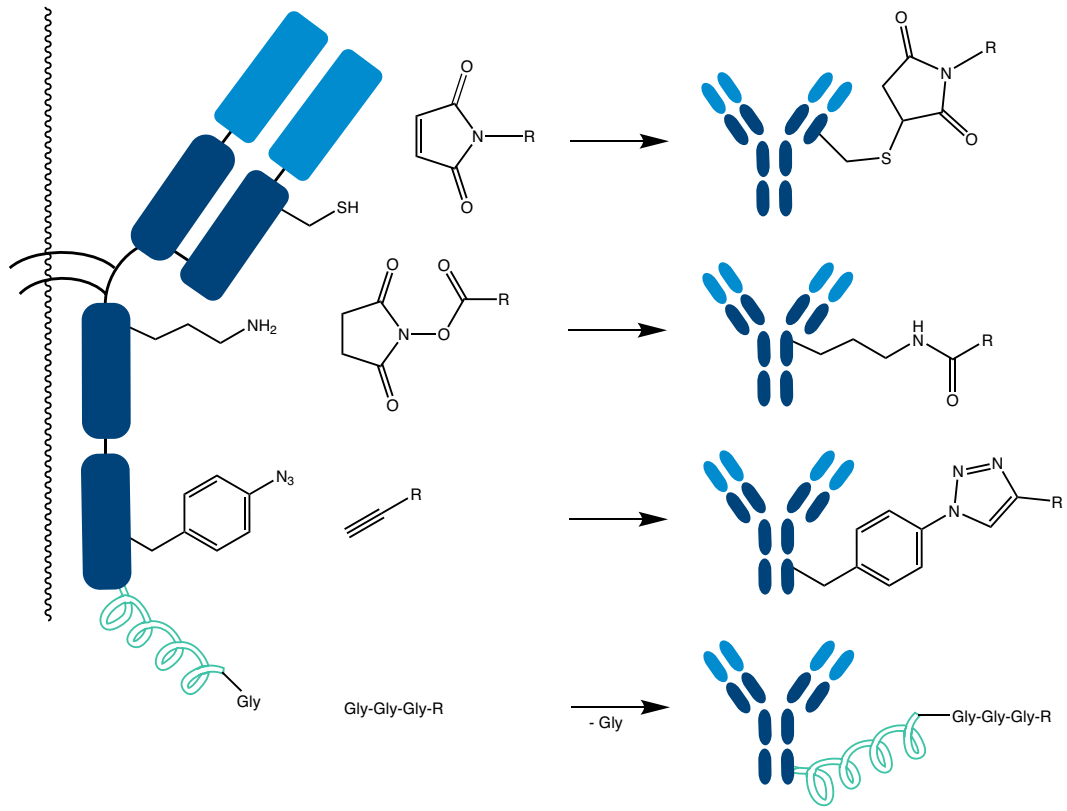


*Fig. 5: Thiols from cysteines, amines from lysines, azido functions from non-canonical amino acids and specific sequences accessible on the surface of antibodies can be addressed by different chemical or enzymatical conjugation methodologies.*

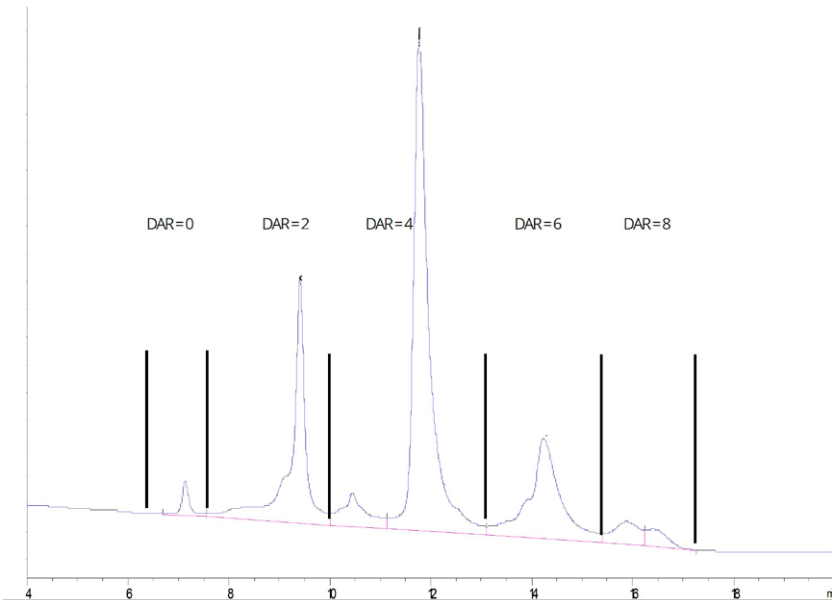


*Fig. 6: Drug-antibody ratio (DAR) is an important parameter of an ADC. Low DAR could reduce the antitumor efficacy, while high DAR may affect antibody structure, stability, and antigen binding etc. therefore causing loss of activity. DAR values are also important for the therapeutic index of ADCs. In most ADC drug candidates, the DAR values were maintained at about 2-4. Hence, controlling DAR during ADC preparation is a key procedure. Figure provided by Glycotope.*

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The type of linkage between payload and biomolecule can basically either be permanent or cleavable unnder certain well-defined circumstances (*Fig. 5*). As payloads typically are highly cytotoxic, it would be fatal if they were released from their carrier during circulation in plasma. Hence, the linker part should be stable to conditions such as pH, redox potential, presence of proteases in plasma, and all other parameters of plasma. However, after internalization it is favorable that the linker is fragmenting in order to release the drug molecule, ideally in a traceless manner. Conjugations with the antibody can rather easily be achieved using active esters forming amide bonds with lysines, which are usually accessible in a high number on the surface. The resulting conjugate, hence, is rather heterogenic with different numbers of payloads attached at different positions. A more and well-defined drug-antibody ratio (DAR) can be achieved by utilizing the disulfide bridges between heavy and light chains of the antibody.

After reductive cleavage of the disulfide bonds, conjugation chemistry can be performed by different kinds of reactions like conventional maleimides or disulfide bond formation. Heterogeneity can be observed if heavy and light antibody chains do not recombine in the original manner.

A highly accurate and specific DAR with well-defined connectivity can be achieved, if unnatural amino acids, e.g., *p*-azidophenylalanine, can be introduced recombinantly. Click chemistry or other Diels-Aldertype reactions can be used to introduce linkers and payloads. In a similar manner, certain peptide fragments can be added, which serve as substrates for ligases in order to conjugate to appropriate linker-payload conjugates.

***References:***


→ *Linker Technologies for Antibody–Drug Conjugates; B. Nolting;* ***Antibody-Drug Conjugates L. Ducry*** *2013;* ***1045****: 71-100.* https://doi.org/10.1007/978-1-62703-541-5_5

→ *In Vivo Applications of Bioorthogonal Reactions: Chemistry and Targeting Mechanisms; M. M. A. Mitry, F. Greco, H. M. I. Osborn;* ***Chemistry*** *2023; n/a: e202203942.* https://doi.org/10.1002/chem.202203942


## Cleavage Mechanisms

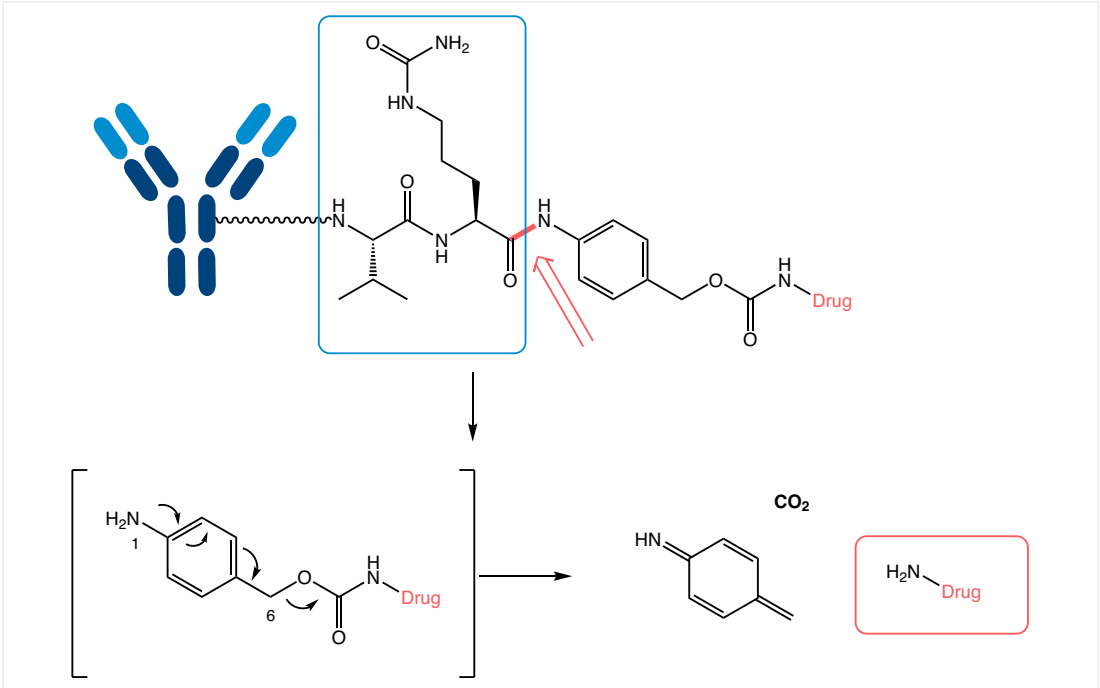


*Fig. 7: Valyl-citrullyl dipeptide fragment serves as substrate for cathepsin and suffers cleavage by hydrolysis leading to a 1,6-elimination with fragmentation and traceless release of the drug molecule.*

An ADC travels through plasma until it reaches the target cell. After internalization, the complex degrades and releases the payload even with a stable linker. However, release can be accelerated through implementation of moieties which fragmentize under certain conditions. One of the most commonly used spacers is the bifunctional *p*-aminobenzyl alcohol group, which is linked to the peptide through the amino group forming an amide bond, while amine containing cytotoxic drugs are attached through carbamate functionalities to the benzylic hydroxyl group of the linker. The resulting prodrugs are activated upon protease mediated hydrolysis and cleavage of the amide bond of citrulline to the *p*-aminobenzyl fragment, leading to a 1,6-elimination reaction releasing the unmodified drug, carbon dioxide, and remnants of the linker group (*Fig. 7*, *Fig. 8*).

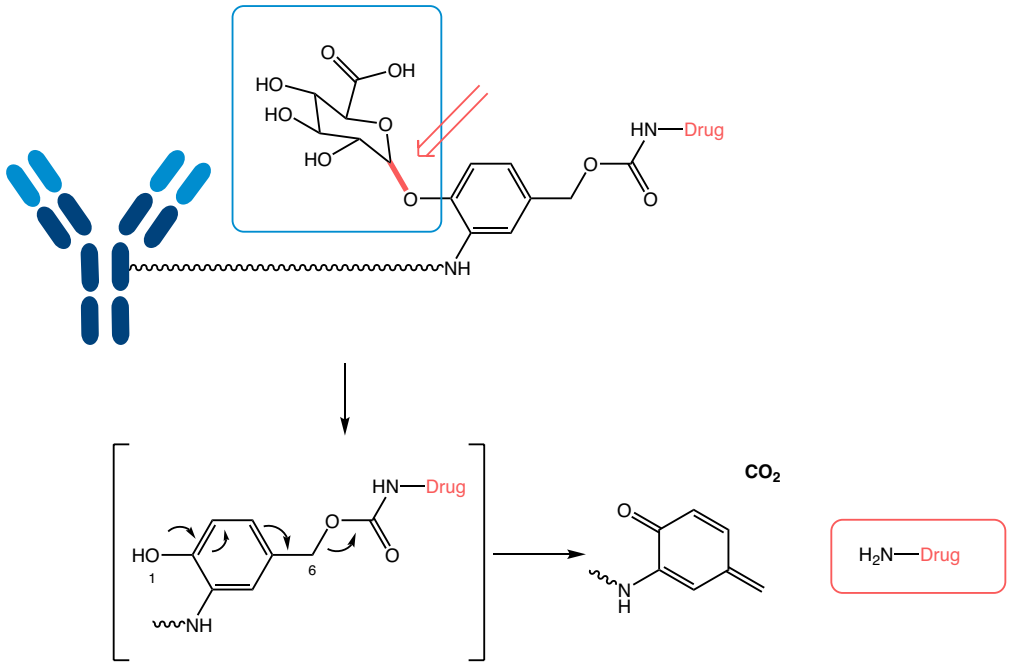


*Fig. 8: Glucuronic acid capped p-aminobenzyl will be cleaved by glucuronidases resulting in 1,6-elmination, fragmentation, and traceless release of the drug molecule.*

In an extension of the peptide-based linker strategies to provide high ADC stability, β-glucuronic acid-based linkers were developed. Facile release of the active drug is realized through cleavage of the β-glucuronide glycosidic bond by the lysosomal enzyme β-glucuronidase. This enzyme is abundantly present in lysosomes and overexpressed in some tumor types, while its activity outside cells is low. The linker is hydrophilic, stable against circulation, and provides ADCs that are highly active both *in vitro* and *in vivo*.

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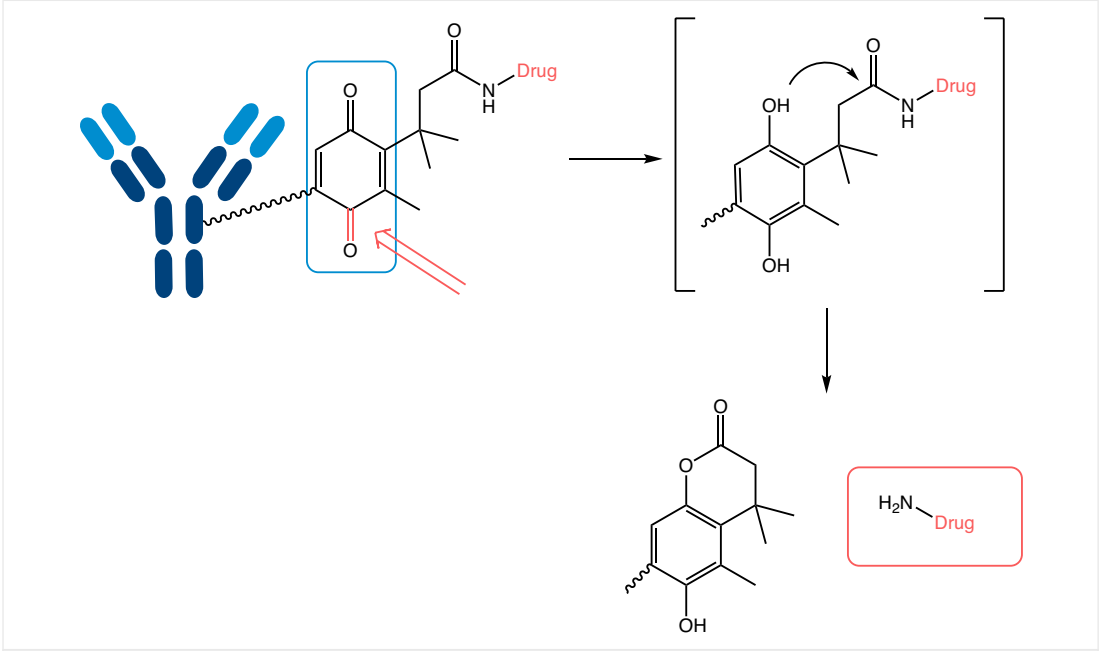



*Fig. 9: Chinoidic variations of trimethyl locks are reduced to the corresponding diphenol followed by traceless release of a drug molecule via lacton formation.*

Besides hydrolases, the presence of oxidoreductases in the lysosome is being utilized for the design of cleavable linkers. Cytochrome P450 oxidoreductase (CPR), nitroquinone oxidoreductase 1 (NQO1), and cellular reductants such as glutathione (GSH) transform reducible fragments like chinone or disulfide to self-immolative intermediates.

## Trimethyl Lock

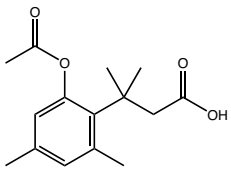
The sterical demand of three closely positioned methyl groups (*Fig. 9*) favors the cleavage of a carbonyl bond by lacton formation. The acidity of the phenol is sufficient to accelerate lactonization at neutral pH and any residue carrying a hydroxyl or amino function will be unlocked, i.e. tracelessly released. The hydroxy group of phenol can be protected and released by a variety of methodologies. This reaction usually requires no elevated temperature. Hence, it will work nicely at physiological conditions.

Product details

### RL-2960 Acetyl-Trimethyl-Lock

**3-(2-Acetoxy-4,6-dimethylphenyl)-3-methylbutyric acid**

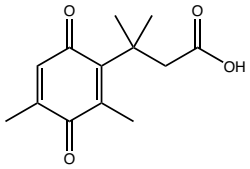
| | |
|---|---|
| CAS-No. | 134098-68-3 |
| Formula | $C_{15}H_{20}O_4$ |
| Mol. weight | 264,14 g/mol |



## RL-2950 Fourmethyl-Lock

3-(2,4-dimethyl-3,6-dioxocyclohexa-1,4-dienyl)-3-methylbutanoic acid

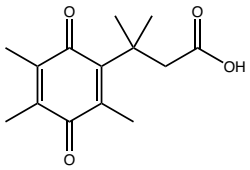
| | |
|---|---|
| CAS-No. | 133544-77-1 |
| Formula | $C_{13}H_{16}O_4$ |
| Mol. weight | 236,26 g/mol |



## RL-2940 Fivemethyl-Lock

3-methyl-3-(2,4,5-trimethyl-3,6-dioxocyclohexa-1,4-dienyl)butanoic acid

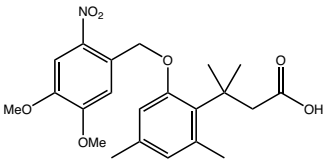
| | |
|---|---|
| CAS-No. | 40662-29-1 |
| Formula | $C_{14}H_{18}O_4$ |
| Mol. weight | 250,29 g/mol |



## RL-2970 Photo-Trimethyl-Lock

3-(2-Nitroveratryl-4,6-dimethylphenyl)-3-methylbutyric acid

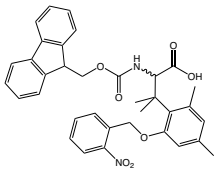
| | |
|---|---|
| CAS-No. | 2095134-25-9 |
| Formula | $C_{22}H_{27}NO_7$ |
| Mol. weight | 417,45 g/mol |



## FAA7190 Fmoc-Spr(oNB)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-beta,beta-dimethyl-(2,4-dimethyl-6-(2-nitrobenzyloxy)phenyl) alanine (rac.)

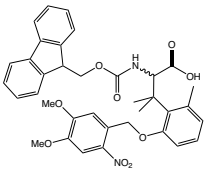
| | |
|---|---|
| CAS-No. | 1032400-98-8 |
| Formula | $C_{35}H_{34}N_2O_7$ |
| Mol. weight | 594,66 g/mol |



## FAA7200 Fmoc-Spr(oNv)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-beta,beta-dimethyl-(2-methyl-6-(2-nitroveratryl)phenyl)alanine (rac.)

| | |
|---|---|
| CAS-No. | 1228829-20-6 |
| Formula | $C_{36}H_{36}N_2O_9$ |
| Mol. weight | 640,68 g/mol |



***References:***


→ *Trimethyl lock: A trigger for molecular release in chemistry, biology, and pharmacology; M. N. Levine, R. T. Raines;* ***Chem. Sci.*** *2012;* ***3****: 2412-2420. https://doi.org/10.1039/C2SC20536J*

→ *Photo-triggered fluorescent labelling of recombinant proteins in live cells; D. Jung, K. Sato, K. Min, A. Shigenaga, J. Jung, A. Otaka, Y. Kwon;* ***Chem Commun*** *2015;* ***51****: 9670-3. https://doi.org/10.1039/C2SC20536J*

→ *Detection of DT-diaphorase Enzyme with a ParaCEST MRI Contrast Agent; I. Daryaei, K. M. Jones, M. D. Pagel;* ***Chemistry*** *2017;* ***23****: 6514-6517. https://doi.org/10.1002/chem.201700721*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- *Syntheses and kinetic studies of cyclisation-based self-immolative spacers; S. Huvelle, A. Alouane, T. Le Saux, L. Jullien, F. Schmidt;* ***Org Biomol Chem*** *2017;* ***15****: 3435-3443.* https://doi.org/10.1039/c7ob00121e
- *Invention of stimulus-responsive peptide-bond-cleaving residue (Spr) and its application to chemical biology tools; A. Shigenaga, J. Yamamoto, T. Kohiki, T. Inokuma, A. Otaka;* ***J Pept Sci*** *2017;* ***23****: 505-513.* https://doi.org/10.1002/psc.2961
- *Trimethyl Lock: A Multifunctional Molecular Tool for Drug Delivery, Cellular Imaging, and Stimuli-Responsive Materials; O. A. Okoh, P. Klahn;* ***ChemBioChem*** *2018;* ***19****: 1668-1694.* https://doi.org/10.1002/cbic.201800269


## Disulfide Linkers

Disulfide linkers (*Fig. 10*) are likely first degraded in the lysosome to generate a cysteine-disulfide catabolite followed by disulfide reduction in the cytosol by cellular reductants such as GSH. The kinetics of reduction can be tailored by neighboring one to four methyl groups next to both sulfurs.

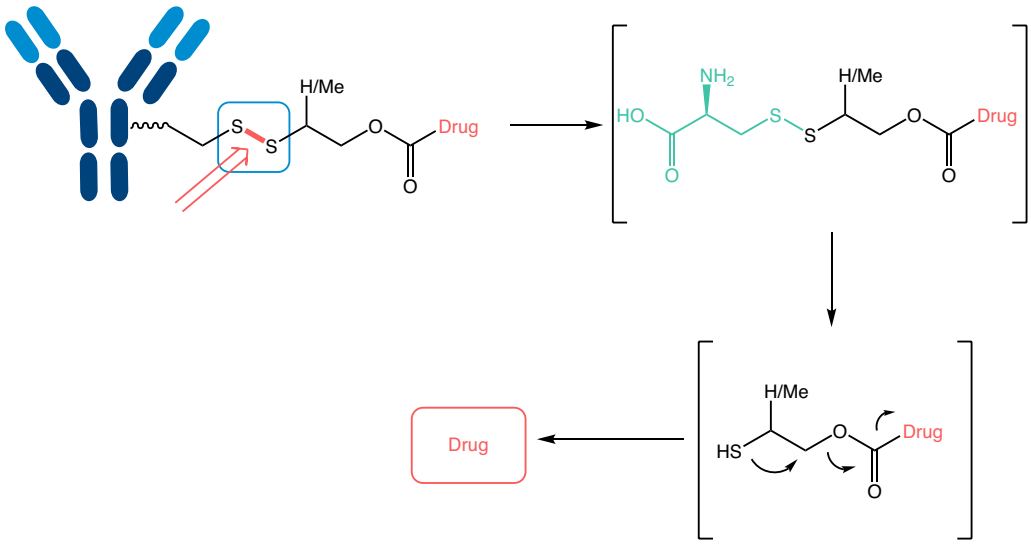


*Fig. 10: Mechanism of disulfide bond cleavage in lysosomal compartments.*

***References:***


- *Modulating Therapeutic Activity and Toxicity of Pyrrolobenzodiazepine Antibody-Drug Conjugates with Self-Immolative Disulfide Linkers; T. H. Pillow, M. Schutten, S. F. Yu, R. Ohri, J. Sadowsky, K. A. Poon, W. Solis, F. Zhong, G. Del Rosario, M. A. T. Go, J. Lau, S. Yee, J. He, L. Liu, C. Ng, K. Xu, D. D. Leipold, A. V. Kamath, D. Zhang, L. Masterson, S. J. Gregson, P. W. Howard, F. Fang, J. Chen, J. Gunzner-Toste, K. K. Kozak, S. Spencer, P. Polakis, A. G. Polson, J. A. Flygare, J. R. Junutula;* ***Mol. Cancer. Ther.*** *2017;* ***16****: 871-878.* https://doi.org/10.1158/1535-7163.MCT-16-0641
- *Mechanisms of drug release in nanotherapeutic delivery systems; P. T. Wong, S. K. Choi;* ***Chem Rev*** *2015;* ***115****: 3388-432.* https://doi.org/10.1021/cr5004634
- *Expanded Utility of the beta-Glucuronide Linker: ADCs That Deliver Phenolic Cytotoxic Agents; S. C. Jeffrey, J. De Brabander, J. Miyamoto, P. D. Senter;* ***ACS Med Chem Lett*** *2010;* ***1****: 277-80.* https://doi.org/10.1021/ml100039h

## Multiple Payloads with one self-immolative Linker

*p*-Hydroxy- and *p*-amino-benzyl fragments will release payloads by a 1,6-elimination cascade, resulting in chinoide intermediates. Under physiological conditions, they readily add water to reform the aromatic ring structure. In case appropriate carbamate substitutions are also placed on position 2 and 2', a fragmentation will occur in a similar manner as by a 1,4-elimination and release any molecules at these positions (*Fig. 11*).

One of the major challenges related to anticancer chemotherapy is resistance against anticancer drugs. A strategy to revert the resistance of tumor cells is the combined use of different anticancer drugs.

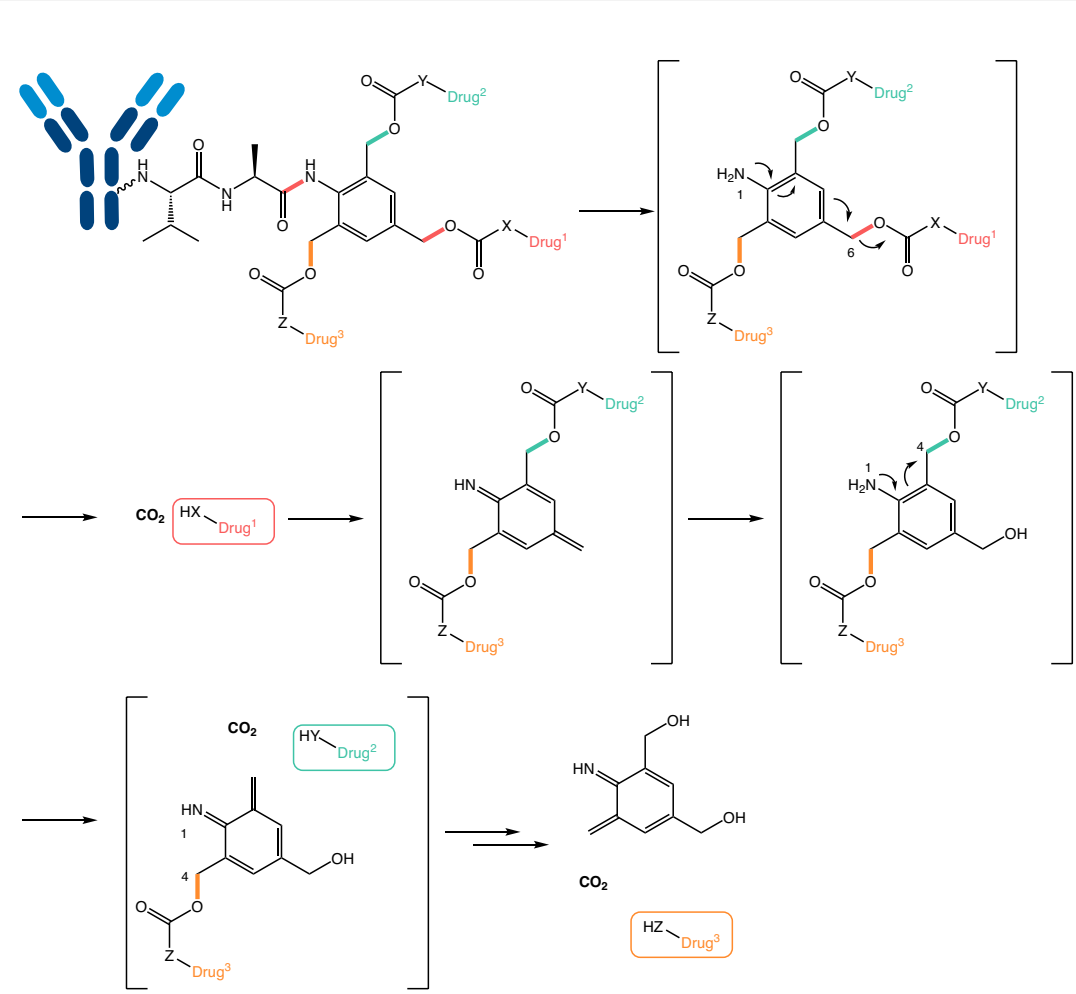


*Fig. 11: Mechanism of multiple traceless releas[e.*



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***References:***

- *A novel connector linkage applicable in prodrug design; P. L. Carl, P. K. Chakravarty, J. A. Katzenellenbogen; **J. Med. Chem.** 1981; **24**: 479-80. https://doi.org/10.1021/jm00137a001*
- *The azaquinone-methide elimination: comparison study of 1,6- and 1,4-eliminations under physiological conditions; R. Erez, D. Shabat; **Org Biomol Chem** 2008; **6**: 2669-72. https://doi.org/10.1039/b808198k*
- *Dendritic chain Dendritic chain reaction: responsive release of hydrogen peroxide upon generation and enzymatic oxidation of methanol; M. Avital- Shmilovici, D. Shabat; **Bioorg Med Chem** 2010; **18**: 3643-7. https://doi.org/10.1016/j.bmc.2010.02.038*
- *ABC transporters as multidrug resistance mechanisms and the development of chemosensitizers for their reversal; C. H. Choi; **Cancer Cell Int** 2005; **5**: 30. https://doi.org/10.1186/1475-2867-5-30*

It has been reported that payload release can be supported by introducing a *N,N'*-dimethylethane-1,2-diamine bridge between carbamate and payload. After release of carbon dioxide, it will cyclize and form 1,3-dimethylimidazolidin-2-one and liberate the payload from the linker construction (*Fig. 12*).

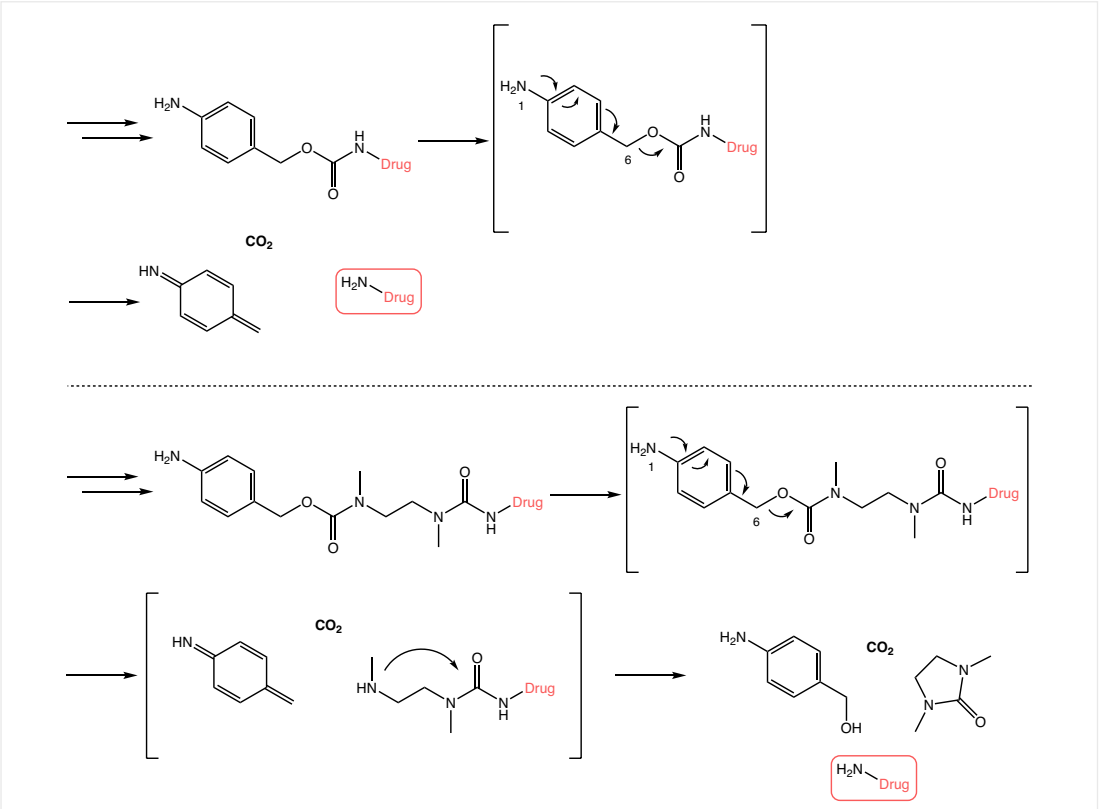


*Fig. 12: Mechanism of assisted traceless release by N,N'-dimethylethane-1,2-diamine.*

Besides the benzyl system, other moieties have been used for fragmentation reactions. In *Fig. 13* different methods are summarized, which have been studied and published. PG is the protecting group and LG the leaving group belonging to the payload to be released. X needs to be a strong electron-donating group, such as O, X or NH, in order to initiate the elimination cascade. While the 1,6-elimination of a benzyl system tends to be the most common system, *ortho*-benzyl undergoing a 1,4-elimination can alternatively be used, as well as styrene fragments (1,8-elimination). However, neither naphthalene rings nor biphenyl structures (1,10-elimination) work, even with a strongly donating amino group.

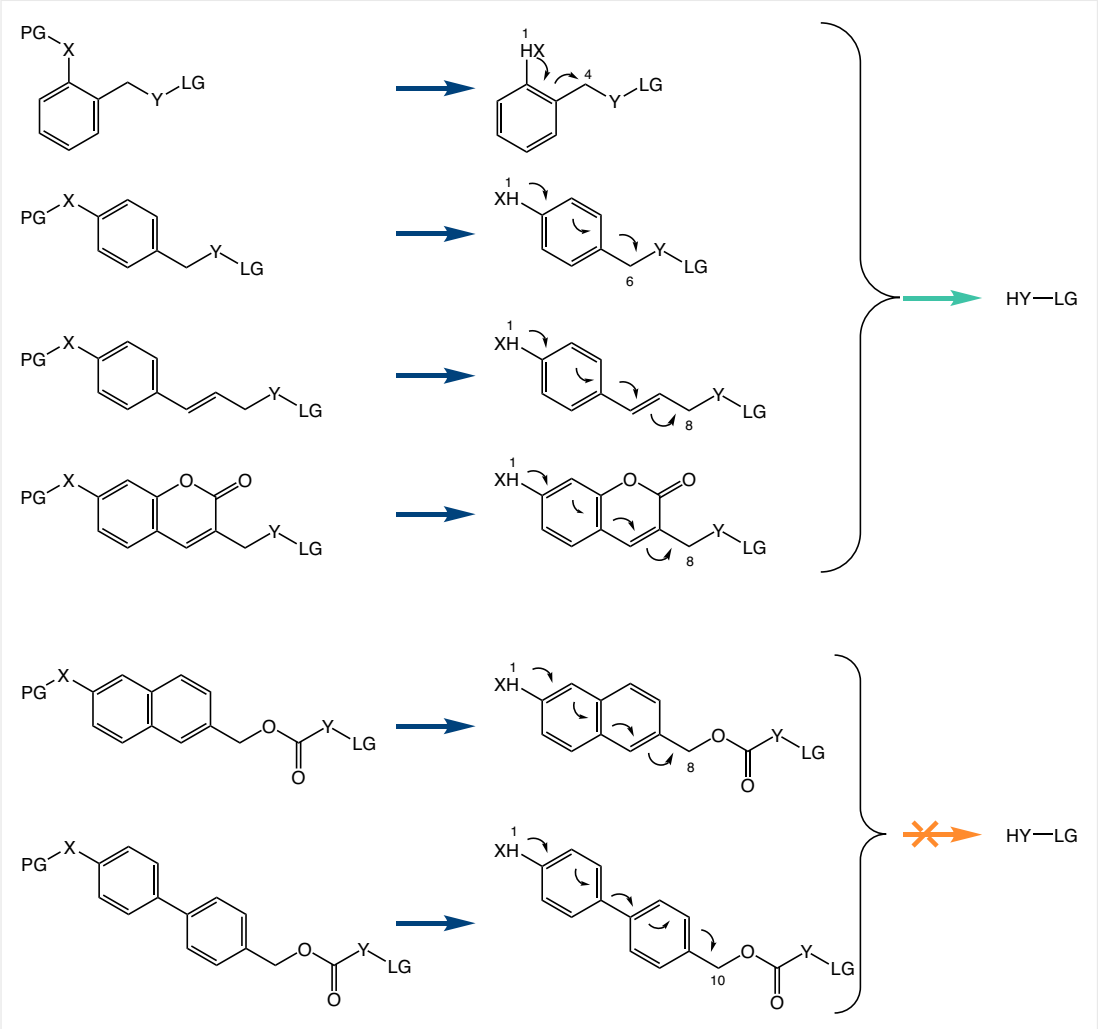


*Fig. 13: Possible fragments enabling traceless release of payloads.*

***References:***


→ *Self-immolative spacers: kinetic aspects, structure-property relationships, and applications; A. Alouane, R. Labruere, T. Le Saux, F. Schmidt, L. Jullien;* ***Angew Chem Int Ed*** *2015;* ***54****: 7492-509. https://doi.org/10.1002/anie.201500088*

→ *Cleavable linkers in chemical biology; G. Leriche, L. Chisholm, A. Wagner;* ***Bioorg Med Chem*** *2012;* ***20****: 571-82. https://doi.org/10.1016/j.bmc.2011.07.048*




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## Dioxoborolane Cross-Linker:

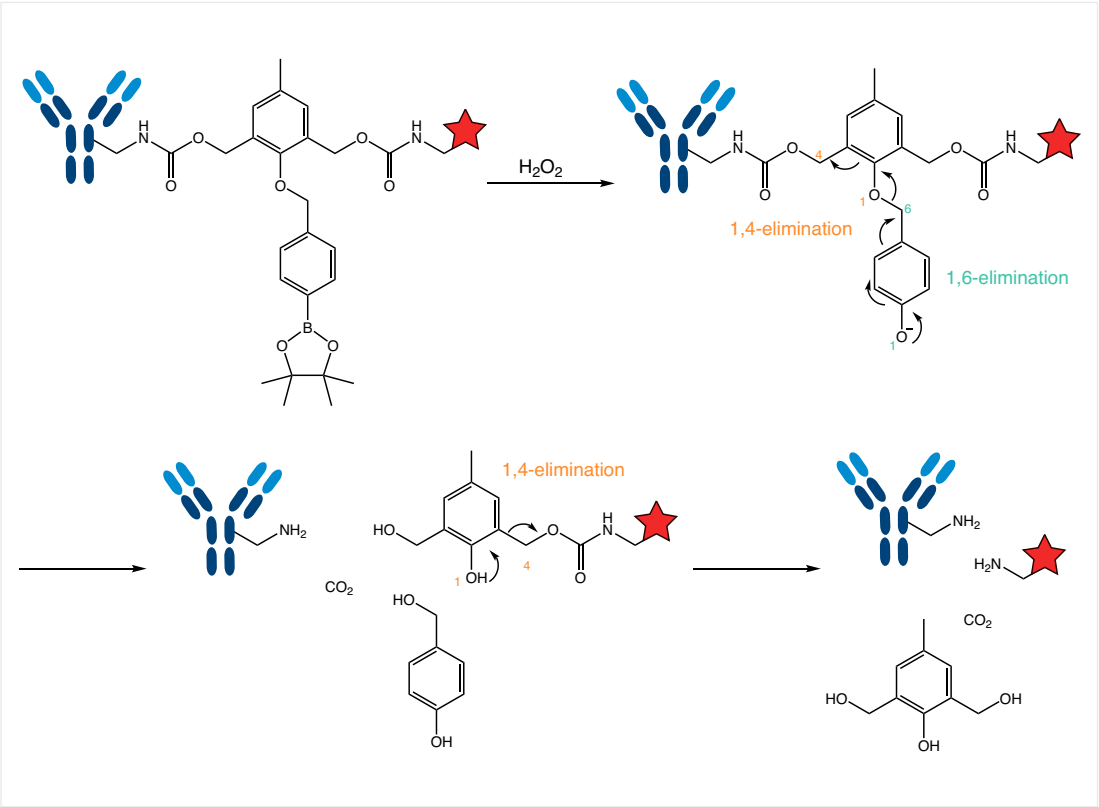


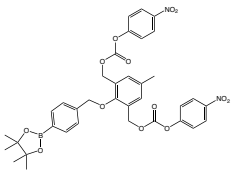

*Fig. 14: Tetramethyldioxoborolane cross-linker fragmentize in the presence of $H_2O_2$.*

Tetramethyldioxoborolane linkers are used as cross-linkers or masking protecting groups of alcohols or amines. They release their payloads under mild oxidative conditions, as it is in the presence of $H_2O_2$. Oxidation initially creates a free phenolate which triggers an initial 1,6-elimination. The second step in this cascade is a 1,4-elimination, followed by a fragmentation under release of the first conjugate. A second 1,4-elimination follows releasing the second conjugated compound. Such moieties are for example being used for the preparation of masked $H_2O_2$ probes releasing their active fluorophore upon trigger detection. Another application is the generation of liposomes, which disrupt upon $H_2O_2$ detection and then release the active cargo. Peptides bearing two lysines can be cyclized and masked inactive *via* such a borolane cross-linker. Activity of the bioactive peptide will be restored, if $H_2O_2$ is present in the tissue environment.

Product details

### RL-4140 TetraMe-Dioxoborolane-(OpNC)2

(5-methyl-2-((4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)benzyl)oxy)-1,3-phenylene)bis(methylene) bis(4-nitrophenyl) bis(carbonate)

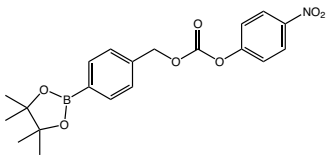


| | |
|---|---|
| CAS-No. | 1355342-68-5 |
| Formula | $C_{36}H_{35}BN_2O_{13}$ |
| Mol. weight | 714,49 g/mol |

## RL-4130 TetraMe-Dioxoborolane-OpNC

4-Nitrophenyl 4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)benzyl carbonate

| | |
|---|---|
| CAS-No. | 1254765-89-3 |
| Formula | $C_{20}H_{22}BNO_7$ |
| Mol. weight | 399,21 g/mol |



***References:***


- *Influence of Linkers within Stimuli-Responsive Prodrugs on Cancer Therapy: A Case of Five Doxorubicin Dimer-Based Nanoparticles; Q. Wang, C. Wang, S. Li, Y. Xiong, H. Wang, Z. Li, J. Wan, X. Yang, Z. Li;* ***Chem. Mater.*** *2022;* ***34(5)****: 2085-2097.* https://doi.org/10.1021/acs.chemmater.1c03346
- *Reactive Oxygen Species-Responsive Liposomes via Boronate-Caged Phosphatidylethanolamine; J. Lou, M. D. Best;* ***Bioconjugate Chem.*** *2020;* ***31(9)****: 2220-2230.* https://doi.org/10.1021/acs.bioconjchem.0c00397
- *Magnetic Resonance Imaging of PSMA-Positive Prostate Cancer by a Targeted and Activatable Gd(III) MR Contrast Agent; H. Li, D. Luo, C. Yuan, X. Wang, J. Wang, J. P. Basilion, T. J. Meade;* ***J. Am. Chem. Soc.*** *2021;* ***143(41)****: 17097–17108.* https://doi.org/10.1021/jacs.1c07377
- *Novel N-Methylated Cyclodepsipeptide Prodrugs for Targeted Cancer Therapy; C. Wu, Z. Cheng, D. Lu, K. Liu, Y. Cheng, P. Wang, Y. Zhou, M. Li, X. Shao, H. Li, W. Su, L. Fang;* ***J. Med. Chem.*** *2021;* ***64(2)****: 991-1000.* https://doi.org/10.1021/acs.jmedchem.0c01387
- *Structure-Based Identification of Potent Lysine-Specific Demethylase 1 Inhibitor Peptides and Temporary Cyclization to Enhance Proteolytic Stability and Cell Growth-Inhibitory Activity; H. Kitagawa, M. Kikuchi, S. Sato, H. Watanabe, N. Umezawa, M. Kato, Y. Hisamatsu, T. Umehara, T. Higuchi;* ***J. Med. Chem.*** *2021;* ***64(7)****: 3707-3719.* https://doi.org/10.1021/acs.jmedchem.0c01371
- *A mitochondrial-targetable dual functional near-infrared fluorescent probe to monitor pH and $H_2O_2$ in living cells and mice; X. Bi, Y. Wang, D. Wang, L. Liu, W. Zhu, J. Zhang, X. Zha;* ***RSC Adv.*** *2020;* ***10****: 26874-26879.* https://doi.org/10.1039/D0RA03905E
- *GSH Activated Biotin-tagged Near-Infrared Probe for Efficient Cancer Imaging; R. Guo, F. Huang, B. Zhang, Y. Yan, J. Che, Y. Jin, Y. Zhuang, R. Dong, Y. Li, B. Tan, R. Song, Y. Hu, X. Dong, X. Li, N. Lin;* ***Theranostics*** *2019;* ***9(12)****: 3515-3525.* https://doi.org/10.7150/thno.32742 https://www.thno.org/v09p3515.htm
- *Photoactivatable Organic Semiconducting Pro-nanoenzymes; J. Li, J. Huang, Y. Lyu, J. Huang, Y. Jiang, C. Xie, K. Pu;* ***J. Am. Chem. Soc.*** *2019;* ***141(9)****: 4073-4079.* https://doi.org/10.1021/jacs.8b13507
- *Versatile Histochemical Approach to Detection of Hydrogen Peroxide in Cells and Tissues Based on Puromycin Staining; C. Yik-Sham Chung, G. A. Timblin, K. Saijo, C. J. Chang;* ***J. Am. Chem. Soc.*** *2018;* ***140(19)****: 6109-6121.* https://doi.org/10.1021/jacs.8b02279
- *Facile Fabrication of 10-Hydroxycamptothecin-Backboned Amphiphilic Polyprodrug with Precisely Tailored Drug Loading Content for Controlled Release; X. Zhang, M. Zhang, M. Wang, H. Peng, Q. Hua, L. Ma, B. Wang, H. Wei;* ***Bioconjugate Chem.*** *2018;* ***29(7)****: 2239-2247.* https://doi.org/10.1021/acs.bioconjchem.8b00238
- *Self immolative dioxetane based chemiluminescent probe for H2O2 detection; O. Seven, F. Sozmen, I. S. Turan;* ***Sens. Actuators B*** *2017;* ***239****: 1318-1324.* https://doi.org/10.1016/j.snb.2016.09.120
- *Reactive Oxygen Species-Responsive Protein Modification and Its Intracellular Delivery for Targeted Cancer Therapy; M. Wang, S. Sun, C. I. Neufeld, B. Perez-Ramirez, Q. Xu;* ***Angew. Chem. Int. Ed.*** *2014;* ***53(49)****: 13444-13448.* https://doi.org/10.1002/anie.201407234
- *pH and hydrogen peroxide dual responsive supramolecular prodrug system for controlled release of bioactive molecules; Y. Wang, H. Wang, Y. Chen, X. Liu, Q. Jin, J. Ji;* ***Colloids and Surfaces B: Biointerfaces*** *2014;* ***121****: 189-195.* https://doi.org/10.1016/j.colsurfb.2014.06.024
- *Investigation of self-immolative linkers in the design of hydrogen peroxide activated metalloprotein inhibitors; J. L. Major Jourden, K. B. Daniel, S. M. Cohen;* ***Chem. Commun.*** *2011;* ***47****: 7968-7970.* https://doi.org/10.1039/C1CC12526E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# 2. Permanent Linkers

## 2.1. PEG-Based Spacer Molecules

This class of linkers is considered non-cleavable, meaning linker cleavage and payload release do not depend on the differential properties between plasma and cytoplasmic compartments. Instead, the release of the cytotoxic drug is postulated to occur after internalization of the ADC *via* antigen-mediated endocytosis and delivery to lysosomal compartments, where the antibody is degraded to the level of amino acids through intracellular proteolytic degradation. This process releases a drug derivative, formed by the cytotoxic drug, the linker, and the amino acid residue to which the linker was covalently attached.

The following section displays examples of hetero-bifunctional PEG-based spacer molecules. As payloads are quite often rather hydrophobic, PEG fragments help to solubilize the linker-payload conjugate, which is essential to perform successful conjugation to the antibody. It further helps to increase the solubility in physiological media and to improve the pharmacokinetic properties of the whole ADC construct.

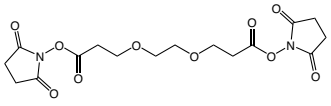
Two of the latest approved ADCs, Trodelvy and Zynlonta, were developed with PEG spacers as part of their linkers to improve solubility and stability *in vivo*.

Product details

### PEG4120 NHS-PEG(2)-NHS

**3,6-Dioxaoctandioic acid bissuccinimidyl ester**

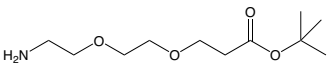
| | |
|---|---|
| CAS-No. | 65869-63-8 |
| Formula | $C_{16}H_{20}N_2O_{10}$ |
| Mol. weight | 400,34 g/mol |



### PEG1365 $H_2N$-PEG(2)-CO-O*t*Bu

**3-(2-(2-Aminoethoxy)ethoxy)propanoic acid *t*-butyl ester**

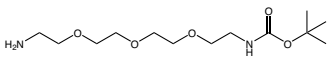
| | |
|---|---|
| CAS-No. | 756525-95-8 |
| Formula | $C_{11}H_{23}NO_4$ |
| Mol. weight | 233,3 g/mol |



### PEG6835 Boc-NH-PEG(3)-$NH_2$*HCl

***tert*-butyl (2-(2-(2-(2-aminoethoxy)ethoxy)ethoxy) ethyl)carbamate**

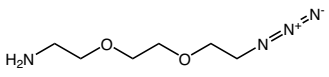
| | |
|---|---|
| CAS-No. | 101187-40-0 net |
| Formula | $C_{13}H_{28}N_2O_5$*HCl |
| Mol. weight | 292,38*36,46 g/mol |



## PEG4980 $H_2N$-PEG(2)-$N_3$*TosOH

2-[2-(2-Azidoethoxy)ethoxy]ethanaminium tosylat

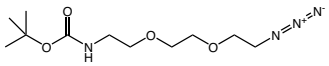
| | |
|---|---|
| CAS-No. | 166388-57-4 |
| Formula | $C_7H_{14}N_4O_2$*$C_7H_8O_3S$ |
| Mol. weight | 174,20*172,20 g/mol |



## PEG4960 Boc-NH-PEG(2)-$N_3$

1-(*t*-Butyloxycarbonyl-amino)-3,6-dioxa-8-octaneazide

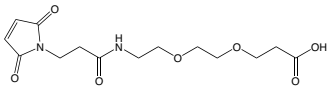
| | |
|---|---|
| CAS-No. | 950683-55-3 |
| Formula | $C_{11}H_{22}N_4O_4$ |
| Mol. weight | 274,32 g/mol |



## PEG1555 mal-PEG(2)-COOH

3-(2-(2-(3-Maleimidopropanamido)ethoxy)ethoxy) propanoic acid

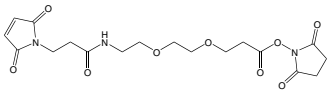
| | |
|---|---|
| CAS-No. | 756525-98-1 |
| Formula | $C_{14}H_{20}N_2O_7$ |
| Mol. weight | 328,32 g/mol |



## PEG1560 mal-PEG(2)-NHS

3-(2-(2-(3-Maleimidopropanamido)ethoxy)ethoxy) propanoic acid succinimidyl ester

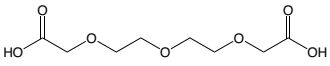
| | |
|---|---|
| CAS-No. | 955094-26-5 |
| Formula | $C_{18}H_{23}N_3O_9$ |
| Mol. weight | 425,39 g/mol |



## PEG2030 TUDA

3,6,9-Trioxaundecandioic acid

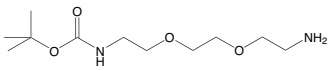
| | |
|---|---|
| CAS-No. | 13887-98-4 |
| Formula | $C_8H_{14}O_7$ |
| Mol. weight | 222,19 g/mol |



## BNN1016 Boc-DOOA

1-(*t*-Butyloxycarbonyl-amino)-3,6-dioxa-8-octaneamine, liq.

| | |
|---|---|
| CAS-No. | 153086-78-3 |
| Formula | $C_{11}H_{24}N_2O_4$ |
| Mol. weight | 248,32 g/mol |



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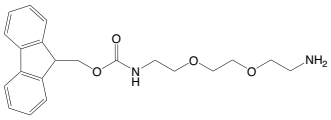
Product details

## FNN1007 Fmoc-DOOA*HCl

1-(9-Fluorenylmethyloxycarbonyl-amino)-3,6-dioxa-8-octaneamine hydrochloride

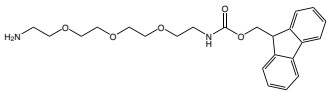
| | |
|---|---|
| CAS-No. | 868599-73-9 |
| Formula | $C_{21}H_{26}N_2O_4$*HCl |
| Mol. weight | 370,45*36,45 g/mol |



## RL-4390 Fmoc-NH-PEG(3)-$NH_2$*HCl

(9H-fluoren-9-yl)methyl (2-(2-(2-aminoethoxy)ethoxy)ethoxy)ethyl)carbamate hydrochloride

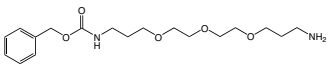
| | |
|---|---|
| CAS-No. | 906079-91-2 |
| Formula | $C_{23}H_{30}N_2O_5$*HCl |
| Mol. weight | 414,50*36,45 g/mol |



## ZNN1120 Z-TOTA

1-Benzyloxycarbonyl-4,7,10-trioxa-13-tridecaneamine

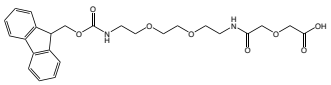
| | |
|---|---|
| CAS-No. | 220156-99-0 |
| Formula | $C_{18}H_{30}N_2O_5$ |
| Mol. weight | 354,45 g/mol |



## PEG5180 Fmoc-DOOA-DIG-OH

2-(2-(2-(2-((9-Fluorenylmethyloxycarbonyl)amino)ethoxy)ethoxy)ethylamino)-diglycolic acid

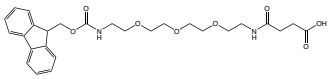
| | |
|---|---|
| CAS-No. | 669073-64-7 |
| Formula | $C_{25}H_{30}N_2O_8$ |
| Mol. weight | 486,51 g/mol |



## RL-4400 Fmoc-NH-PEG(3)-NH-Suc-OH

1-(9H-fluoren-9-yl)-3,17-dioxo-2,7,10,13-tetraoxa-4,16-diazaicosan-20-oic acid

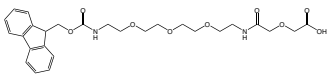
| | |
|---|---|
| CAS-No. | 1653992-32-5 |
| Formula | $C_{27}H_{34}N_2O_8$ |
| Mol. weight | 514,57 g/mol |



## RL-4410 Fmoc-NH-PEG(3)-DIG-OH

1-(9H-fluoren-9-yl)-3,17-dioxo-2,7,10,13,19-pentaoxa-4,16-diazahenicosan-21-oic acid

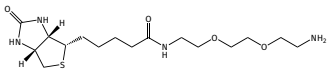
| | |
|---|---|
| CAS-No. | 489427-26-1 |
| Formula | $C_{27}H_{34}N_2O_9$ |
| Mol. weight | 530,57 g/mol |



## RL-4060 Biotin-DOOA

Biotinyl-1-amino-3,6-dioxa-8-octanamine

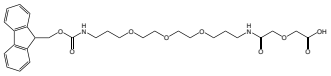
CAS-No. 138529-46-1
Formula $C_{16}H_{30}N_4O_4S$
Mol. weight 374,50 g/mol



## FAA5730 Fmoc-TTD-DIG-OH

[N1-(9-Fluorenylmethoxycarbonyl)-1,13-diamino-4,7,10-trioxatridecan-diglycolic acid

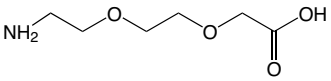
CAS-No. 75-09-2
Formula $C_{29}H_{38}N_2O_9$
Mol. weight 558,62 g/mol



## PEG2420 H-$O_2$Oc-OH

[2-(2-aminoethoxy)ethoxy]acetic acid

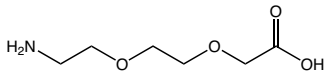
CAS-No. 134978-97-5
Formula $C_6H_{13}NO_4$
Mol. weight 163,17 g/mol



## PEG7940 H-$O_2$Oc-OH*HCl

8-amino-3,6-dioxaoctanoic acid hydrochloride

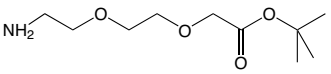
CAS-No. 134979-01-4
Formula $C_6H_{13}NO_4$*HCl
Mol. weight 163,17*36,45 g/mol



## PEG2430 H-$O_2$Oc-O*t*Bu*HCl

[2-(2-aminoethoxy)ethoxy]acetic acid *tert*-butyl ester*HCl

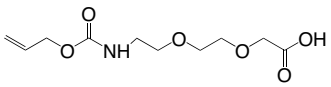
CAS-No. 2098500-69-5
Formula $C_{10}H_{21}NO_4$*HCl
Mol. weight 219,28*36,45 g/mol



## AAA1905 Aloc-$O_2$Oc-OH*DCHA

8-(Allyloxycarbonyl-amino)-3,6-dioxaoctanoic acid dicyclohexylamine

CAS-No. 560088-74-6
Formula $C_{10}H_{17}NO_6$*$C_{12}H_{23}N$
Mol. weight 247,11*181,32 g/mol



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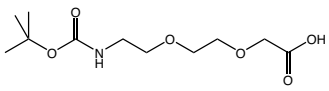
## PEG8080 Boc-$O_2$Oc-OH

(2-(2-(*t*-Butyloxycarbonylamino)ethoxy)ethoxy)acetic acid

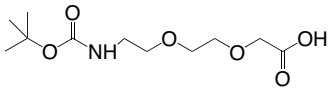
| | |
|---|---|
| CAS-No. | 108466-89-3 |
| Formula | $C_{11}H_{21}NO_6$ |
| Mol. weight | 263,29 g/mol |



## BAA1466 Boc-$O_2$Oc-OH*DCHA

8-(*t*-Butyloxycarbonyl-amino)-3,6-dioxaoctanoic acid dicyclohexylammonium salt

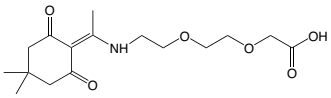
| | |
|---|---|
| CAS-No. | 560088-79-1 |
| Formula | $C_{11}H_{21}NO_6$*$C_{12}H_{23}N$ |
| Mol. weight | 263,29*181,32 g/mol |



## DAA1016 Dde-$O_2$Oc-OH

8-[(4,4-Dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl-amino]-3,6-dioxaoctanoic acid, {2-[2-(Dde-amino) ethoxy]ethoxy}acetic acid

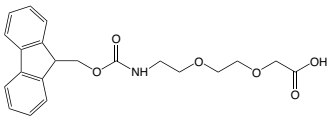
| | |
|---|---|
| CAS-No. | 1263045-93-7 |
| Formula | $C_{16}H_{25}NO_6$ |
| Mol. weight | 327,37 g/mol |



## FAA1435 Fmoc-$O_2$Oc-OH

8-(9-Fluorenylmethyloxycarbonyl-amino)-3,6-dioxaoctanoic acid

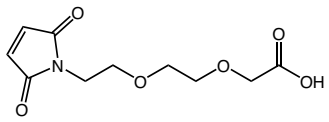
| | |
|---|---|
| CAS-No. | 166108-71-0 |
| Formula | $C_{21}H_{23}NO_6$ |
| Mol. weight | 385,42 g/mol |



## PEG4870 Mal-$O_2$Oc-OH

{2-[2-(2,5-Dioxo-2,5-dihydro-1H-pyrrol-1-yl)ethoxy] ethoxy}acetic acid

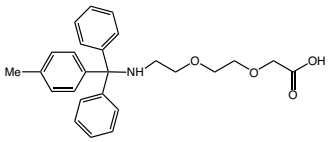
| | |
|---|---|
| CAS-No. | 173323-23-4 |
| Formula | $C_{10}H_{13}NO_6$ |
| Mol. weight | 243,21 g/mol |



## PEG4650 Mtt-$O_2$Oc-OH*DEA

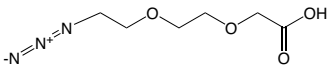
N-(4-Methyltrity)-8-amino-3,6-dioxaoctanoic acid diethylamine

| | |
|---|---|
| CAS-No. | 2098500-66-2 |
| Formula | $C_{26}H_{29}NO_4$*$C_4H_{11}N$ |
| Mol. weight | 419,51*73,14 g/mol |



## PEG2780 $N_3$-$O_2$Oc-OH*CHA

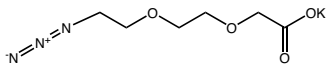
[2-(2-azidoethoxy)ethoxy]acetic acid cyclohexylamine salt



| | |
|---|---|
| CAS-No. | 2098500-94-6 |
| Formula | $C_6H_{11}N_3O_4$*$C_6H_{13}N$ |
| Mol. weight | 189,17*99,17 g/mol |

## PEG7950 $N_3$-AEEA-OK

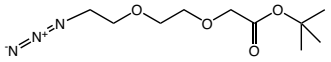
Potassium 8-azido-3,6-dioxaoctanoate



| | |
|---|---|
| CAS-No. | 882518-90-3 |
| Formula | $C_6H_{10}KN_3O_4$ |
| Mol. weight | 39,10*188,16 g/mol |

## PEG5390 $N_3$-$O_2$Oc-O*t*Bu

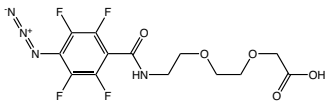
8-Azido-3,6-dioxaoctanoic acid *t*-butyl ester



| | |
|---|---|
| CAS-No. | 251564-45-1 |
| Formula | $C_{10}H_{19}N_3O_4$ |
| Mol. weight | 245,28 g/mol |

## PEG5000 $N_3$-TFBA-$O_2$Oc

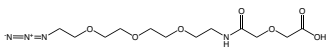
{2-[2-(4-Azido-2,3,5,6-tetrafluorobenzoyl-amino)ethoxy] ethoxy}acetic acid



| | |
|---|---|
| CAS-No. | 1993119-45-1 |
| Formula | $C_{13}H_{12}F_4N_4O_5$ |
| Mol. weight | 380,25 g/mol |

## RL-4370 $N_3$-PEG(3)-NH-DIG-OH

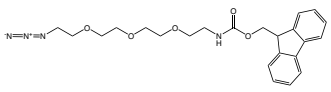
Diglycolic acid PEG3 azide



| | |
|---|---|
| CAS-No. | 239081-53-9 |
| Formula | $C_{12}H_{22}N_4O_7$ |
| Mol. weight | 334,33 g/mol |

## RL-4380 Fmoc-NH-PEG(3)-$N_3$

(9H-fluoren-9-yl)methyl (2-(2-(2-(2-azidoethoxy)ethoxy) ethoxy)ethyl)carbamate



| | |
|---|---|
| CAS-No. | 1172605-58-1 |
| Formula | $C_{23}H_{28}N_4O_5$ |
| Mol. weight | 440,50 g/mol |

The Concept of Antibody-Drug Conjugation (ADC)
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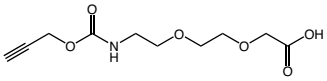
## PAA1050 Poc-$O_2$Oc-OH*DCHA

8-(Popargylyloxycarbonyl-amino)-3,6-dioxaoctanoic acid dicyclohexylamine

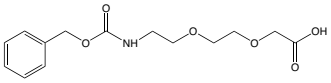
| | |
|---|---|
| CAS-No. | 2988660-57-5 |
| Formula | $C_{10}H_{15}NO_6$*$C_{12}H_{23}N$ |
| Mol. weight | 245,23*181,32 g/mol |



## ZAA1186 Z-$O_2$Oc-OH*DCHA

8-(Benzyloxycarbonyl-amino)-3,6-dioxaoctanoic acid dicyclohexylamine

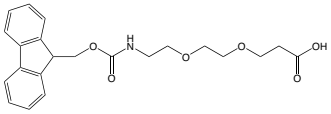
| | |
|---|---|
| CAS-No. | 560088-84-8 |
| Formula | $C_{14}H_{19}NO_6$*$C_{12}H_{23}N$ |
| Mol. weight | 297,31*181,32 g/mol |



## PEG1810 Fmoc-AEEP

3-(2-(2-(9-Fluorenylmethyloxycarbonyl)aminoethoxy) ethoxy)propanoic acid

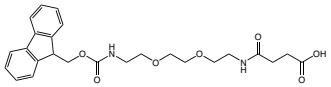
| | |
|---|---|
| CAS-No. | 872679-70-4 |
| Formula | $C_{22}H_{25}NO_6$ |
| Mol. weight | 399,44 g/mol |



## PEG4970 Fmoc-Ebes

N-[8-(9-Fluorenylmethyloxycarbonyl)amino-3,6-dioxaoctyl]succinamic acid

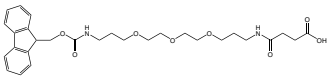
| | |
|---|---|
| CAS-No. | 613245-91-3 |
| Formula | $C_{25}H_{30}N_2O_7$ |
| Mol. weight | 470,51 g/mol |



## FAA1568 Fmoc-TTDS-OH

[N1-(9-Fluorenylmethoxycarbonyl)-1,13-diamino-4,7,10-trioxatridecan-succinamic acid

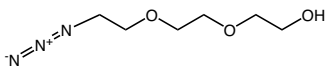
| | |
|---|---|
| CAS-No. | 172089-14-4 |
| Formula | $C_{29}H_{38}N_2O_8$ |
| Mol. weight | 542,63 g/mol |



## PEG4900 $N_3$-EEEt-OH

2-[2-(2-Azidoethoxy)ethoxy]ethanol

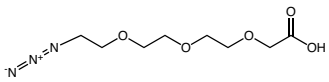
| | |
|---|---|
| CAS-No. | 86520-52-7 |
| Formula | $C_6H_{13}N_3O_3$ |
| Mol. weight | 175,19 g/mol |



## PEG5400 $N_3$-AEEEA*CHA

11-Azido-3,6,9-trioxaundecanoic acid cyclohexylamine

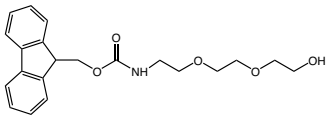
CAS-No. 172531-37-2
Formula $C_8H_{15}N_3O_5*C_6H_{13}N$
Mol. weight 233,22*99,17 g/mol



## PEG5370 Fmoc-AEEE

2-(2-(2-(9-Fluorenylmethyloxycarbonyl)aminoethoxy) ethoxy)ethanol

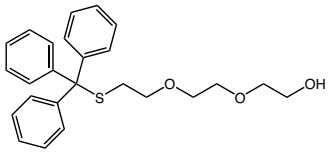
CAS-No. 560088-66-6
Formula $C_{21}H_{25}NO_5$
Mol. weight 371,43 g/mol



## PEG7010 Trt-S-EEE

S-Trityl-2-(2-(2-mercaptoethoxy)ethoxy)ethanol

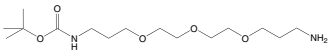
CAS-No. 728033-15-6
Formula $C_{25}H_{28}O_3S$
Mol. weight 408,55 g/mol



## BNN1028 Boc-TOTA

1-(*t*-Butyloxycarbonyl-amino)-4,7,10-trioxa-13-tridecanamine, liq.

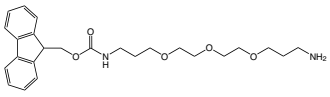
CAS-No. 194920-62-2
Formula $C_{15}H_{32}N_2O_5$
Mol. weight 320,43 g/mol



## FNN1011 Fmoc-TOTA*HCl

1-(9-Fluorenylmethyloxycarbonyl-amino)-4,7,10-trioxa-13-tridecanamine hydrochloride

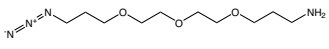
CAS-No. 868599-75-1
Formula $C_{25}H_{34}N_2O_5*HCl$
Mol. weight 442,56*36,45 g/mol



## BNN1150 $N_3$-TOTA

1-Azido-4,7,10-trioxa-13-tridecanamine

CAS-No. 1162336-72-2
Formula $C_{10}H_{22}N_4O_3$
Mol. weight 246,31 g/mol



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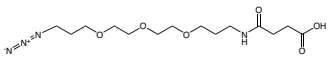
## PEG5170 $N_3$-TOTA-Suc

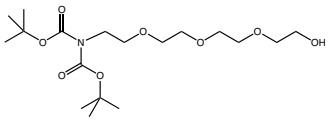
1-Azido-4,7,10-trioxa-13-tridecaneamine succinamic acid



| | |
|---|---|
| CAS-No. | 1993176-74-1 |
| Formula | $C_{14}H_{26}N_4O_6$ |
| Mol. weight | 346,38 g/mol |

## PEG7860 Boc2-AEEEE

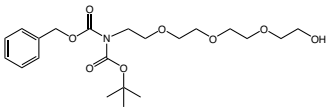
2-(2-(2-(2-(Di-(*t*-butylmethyloxycarbonyl))aminoethoxy) ethoxy)ethoxy)ethanol



| | |
|---|---|
| CAS-No. | 2389064-37-1 |
| Formula | $C_{18}H_{35}NO_8$ |
| Mol. weight | 393,47 g/mol |

## PEG5385 Boc,Z-AEEEE

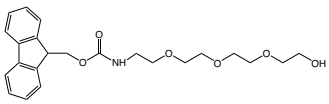
2-(2-(2-(2-(Benzyloxycarbonyl-*tert*-Butylmethyloxycarbonyl)aminoethoxy)ethoxy)ethoxy)ethanol



| | |
|---|---|
| CAS-No. | 2389064-46-2 |
| Formula | $C_{21}H_{33}NO_8$ |
| Mol. weight | 427,49 g/mol |

## PEG5380 Fmoc-AEEEE

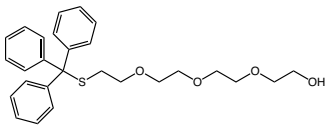
2-(2-(2-(2-(9-Fluorenylmethyloxycarbonyl)aminoethoxy)ethoxy)ethoxy)ethanol



| | |
|---|---|
| CAS-No. | 868594-41-6 |
| Formula | $C_{23}H_{29}NO_6$ |
| Mol. weight | 415,48 g/mol |

## PEG6730 Trt-S-EEEE

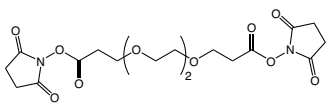
S-Trityl-2-(2-(2-(2-mercaptoethoxy)ethoxy)ethoxy) ethanol



| | |
|---|---|
| CAS-No. | 125607-10-5 |
| Formula | $C_{27}H_{32}O_4S$ |
| Mol. weight | 452,61 g/mol |

## PEG4130 NHS-PEG(3)-NHS

3,6,9-Trioxaundecandioic acid bissuccinimidyl ester

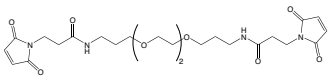


| | |
|---|---|
| CAS-No. | 1314378-16-9 |
| Formula | $C_{18}H_{24}N_2O_{11}$ |
| Mol. weight | 444,39 g/mol |

## PEG1485 mal-PEG(3)-mal

Bis-(1,13-(3-maleimidopropionyl)amido)-4,7,10-trioxatridecane

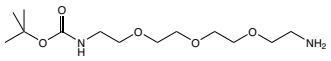
CAS-No. 756525-89-0
Formula $C_{24}H_{34}N_4O_9$
Mol. weight 522,55 g/mol



## PEG7870 Boc-NH-PEG(3)-$NH_2$

1-(*t*-Butyloxycarbonyl)amino-3,6,9-trioxa-undecan-11-amine

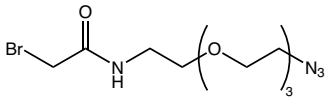
CAS-No. 101187-40-0
Formula $C_{13}H_{28}N_2O_5$
Mol. weight 292,37 g/mol



## PEG7190 Bromoacetamido-PEG(3)-$N_3$

Bromoacetamido-tri(ethylene glycol)-azide

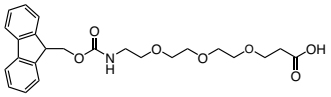
CAS-No. 940005-81-2
Formula $C_{10}H_{19}BrN_4O_4$
Mol. weight 339,19 g/mol



## PEG4370 Fmoc-NH-PEG(3)-COOH

12-(9-Fluorenylmethyloxycarbonylamino)-4,7,10-trioxa-dodecanoic acid

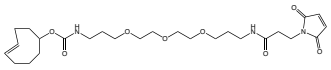
CAS-No. 867062-95-1
Formula $C_{24}H_{29}NO_7$
Mol. weight 443,49 g/mol



## TCO1050 TCO-PEG(3)-mal

*trans*-Cyclooctene-PEG(3)-maleimide

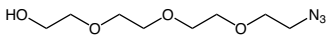
CAS-No. 1809356-72-6
Formula $C_{26}H_{41}N_3O_8$
Mol. weight 523,62 g/mol



## PEG3760 $N_3$-PEG(3)-OH

alpha-Azido-omega-hydroxy tetra(ethylene glycol)

CAS-No. 86770-67-4
Formula $C_8H_{17}N_3O_4$
Mol. weight 219,24 g/mol



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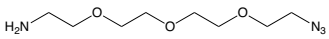
Product details

## PEG3060 $H_2N$-PEG(3)-$N_3$

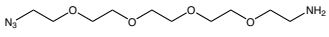
1-Amino-11-azido-3,6,9-trioxaundecane



| | |
|---|---|
| CAS-No. | 134179-38-7 |
| Formula | $C_8H_{18}N_4O_3$ |
| Mol. weight | 218,25 g/mol |

## PEG5320 $N_3$-PEG(4)-$NH_2$

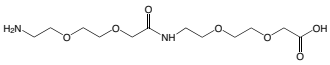
14-Azido-3,6,9,12-tetraoxatetradecan-1-amine



| | |
|---|---|
| CAS-No. | 951671-92-4 |
| Formula | $C_{10}H_{22}N_4O_4$ |
| Mol. weight | 262,31 g/mol |

## PEG1221 H-$O_2$Oc-$O_2$Oc-OH

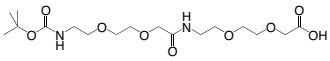
17-Amino-10-oxo-3,6,12,15-tetraoxa-9-azaheptadecan-1-oic acid



| | |
|---|---|
| CAS-No. | 1143516-05-5 |
| Formula | $C_{12}H_{24}N_2O_7$ |
| Mol. weight | 308,33 g/mol |

## BAA1485 Boc-$O_2$Oc-$O_2$Oc-OH

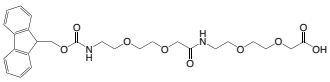
17-(*t*-Butyloxycarbonyl-amino)-9-aza-3,6,12,15-tetraoxa-10-on-heptadecanoic acid



| | |
|---|---|
| CAS-No. | 1069067-08-8 |
| Formula | $C_{17}H_{32}N_2O_9$ |
| Mol. weight | 408,45 g/mol |

## FAA1787 Fmoc-$O_2$Oc-$O_2$Oc-OH

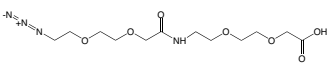
17-(9-Fluorenylmethyloxycarbonyl-amino)-9-aza-3,6,12,15-tetraoxa-10-on-heptadecanoic acid



| | |
|---|---|
| CAS-No. | 560088-89-3 |
| Formula | $C_{27}H_{34}N_2O_9$ |
| Mol. weight | 530,58 g/mol |

## PEG2790 $N_3$-$O_2$Oc-$O_2$Oc-OH

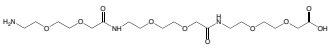
8-(8-Azido-3,6-dioxaoctanoylamido)-3,6-dioxaoctanoic acid



| | |
|---|---|
| CAS-No. | 1254054-60-8 |
| Formula | $C_{12}H_{22}N_4O_7$ |
| Mol. weight | 334,33 g/mol |

## PEG2770 H-$O_2$Oc-$O_2$Oc-$O_2$Oc-OH

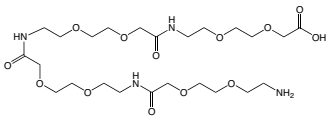
26-amino-10,19-dioxo-3,6,12,15,21,24-hexaoxa-9,18-diazahexacosan-1-oic acid



| | |
|---|---|
| CAS-No. | 2773558-06-6 |
| Formula | $C_{18}H_{35}N_3O_{10}$ |
| Mol. weight | 453,48 g/mol |

## PEG8060 H-$O_2$Oc-$O_2$Oc-$O_2$Oc-$O_2$Oc-OH

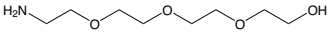
8-amino-3,6-dioxaoctanoic acid tetramer



| | |
|---|---|
| CAS-No. | 2773558-66-8 |
| Formula | $C_{24}H_{46}N_4O_{13}$ |
| Mol. weight | 598,64 g/mol |

## PEG1320 $H_2$N-PEG(4)-OH

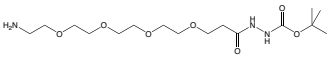
2-(2-(2-(2-Aminoethoxy)ethoxy)ethoxy)ethanol



| | |
|---|---|
| CAS-No. | 86770-74-3 |
| Formula | $C_8H_{19}NO_4$ |
| Mol. weight | 193,24 g/mol |

## PEG1335 $H_2$N-PEG(4)-NHNH-Boc

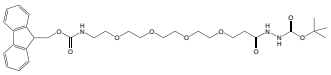
15-Amino-4,7,10,13-tetraoxa-pentadecanoyl-*N*'-(*t*-butyloxycarbonyl)-hydrazid



| | |
|---|---|
| CAS-No. | 1263047-17-1 |
| Formula | $C_{16}H_{33}N_3O_7$ |
| Mol. weight | 379,45 g/mol |

## PEG1805 Fmoc-NH-PEG(4)-NHNH-Boc

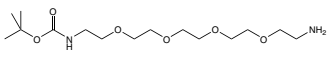
15-(9-Fluorenylmethyloxycarbonyl)amino-4,7,10,13-tetraoxa-pentadecanoyl-*N*'-(*t*-butyloxycarbonyl)hydrazid



| | |
|---|---|
| CAS-No. | 1263044-77-4 |
| Formula | $C_{31}H_{43}N_3O_9$ |
| Mol. weight | 601,69 g/mol |

## PEG7880 Boc-NH-PEG(4)-$NH_2$

1-(*t*-Butyloxycarbonyl)amino-3,6,9,12-tetraoxatetradecan-14-amine



| | |
|---|---|
| CAS-No. | 811442-84-9 |
| Formula | $C_{15}H_{32}N_2O_6$ |
| Mol. weight | 336,42 g/mol |

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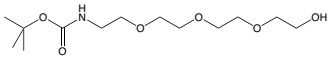
Product details

## PEG1915 Boc-NH-PEG(4)-OH

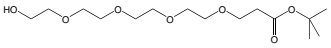
2-(2-(2-(2-(*t*-Butyloxycarbonylamino)ethoxy)ethoxy) ethoxy)ethanol



| | |
|---|---|
| CAS-No. | 106984-09-2 |
| Formula | $C_{13}H_{27}NO_6$ |
| Mol. weight | 293,36 g/mol |

## PEG1535 HO-PEG(4)-CO-OtBu

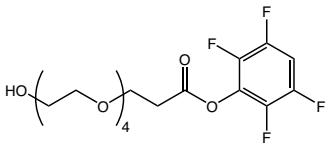
15-Hydroxy-4,7,10,13-tetraoxa-pentadecanoic acid *t*-butyl ester



| | |
|---|---|
| CAS-No. | 518044-32-1 |
| Formula | $C_{15}H_{30}O_7$ |
| Mol. weight | 322,39 g/mol |

## PEG7220 HO-PEG(4)-TFP

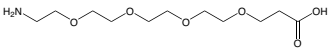
Hydroxy-tetra(ethylene glycol)-propionyl 2,3,5,6-tetrafluorophenyl ester



| | |
|---|---|
| CAS-No. | 2130036-53-0 |
| Formula | $C_{17}H_{22}F_4O_7$ |
| Mol. weight | 414,35 g/mol |

## PEG1370 $H_2N$-PEG(4)-COOH

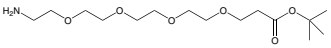
15-Amino-4,7,10,13-tetraoxa-pentadecanoic acid



| | |
|---|---|
| CAS-No. | 663921-15-1 |
| Formula | $C_{11}H_{23}NO_6$ |
| Mol. weight | 265,3 g/mol |

## PEG1375 $H_2N$-PEG(4)-CO-O*t*Bu

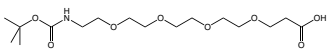
15-Amino-4,7,10,13-tetraoxa-pentadecanoic acid *t*-butyl ester



| | |
|---|---|
| CAS-No. | 581065-95-4 |
| Formula | $C_{15}H_{31}NO_6$ |
| Mol. weight | 321,41 g/mol |

## PEG1920 Boc-NH-PEG(4)-COOH

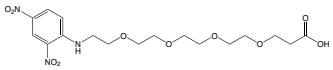
15-*t*-Butyloxycarbonylamino-4,7,10,13-tetraoxa-pentadecanoic acid



| | |
|---|---|
| CAS-No. | 756525-91-4 |
| Formula | $C_{16}H_{31}NO_8$ |
| Mol. weight | 365,42 g/mol |

## PEG2145 Dnp-NH-PEG(4)-COOH

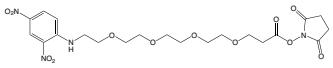
1-(2,4-Dinitrophenylamino)-3,6,9,12-tetraoxapentadecanoic acid



| | |
|---|---|
| CAS-No. | 858126-76-8 |
| Formula | $C_{17}H_{25}N_3O_{10}$ |
| Mol. weight | 431,39 g/mol |

## PEG2150 Dnp-NH-PEG(4)-NHS

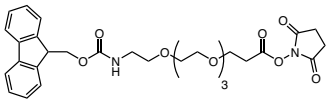
1-(2,4-Dinitrophenylamino)-3,6,9,12-tetraoxapentadecanoic acid succinimidyl ester



| | |
|---|---|
| CAS-No. | 858126-78-0 |
| Formula | $C_{21}H_{28}N_4O_{12}$ |
| Mol. weight | 528,47 g/mol |

## PEG4410 Fmoc-NH-PEG(4)-NHS

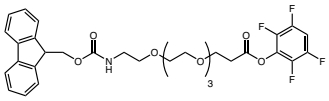
15-(9-Fluorenylmethyloxycarbonyl)amino-4,7,10,13-tetraoxa-pentadecanoic acid succinimidyl ester



| | |
|---|---|
| CAS-No. | 1314378-14-7 |
| Formula | $C_{30}H_{36}N_2O_{10}$ |
| Mol. weight | 584,24 g/mol |

## PEG7810 Fmoc-NH-PEG(4)-TFP

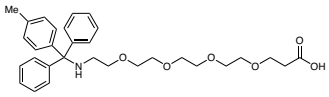
15-(9-Fluorenylmethyloxycarbonyl)amino-4,7,10,13-tetraoxa-pentadecanoic acid (2,3,5,6-tetrafluorophenyl) ester



| | |
|---|---|
| CAS-No. | 2247993-77-5 |
| Formula | $C_{32}H_{33}F_4NO_8$ |
| Mol. weight | 635,6 g/mol |

## PEG2161 Mtt-NH-PEG(4)-COOH*TEA

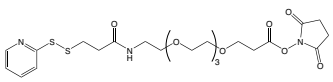
1-(*p*-Methytritylamino)-3,6,9,12-tetraoxapentadecanoic acid triethylamionium salt



| | |
|---|---|
| CAS-No. | 1310680-33-1 (net) |
| Formula | $C_{31}H_{39}NO_6*C_6H_{15}N$ |
| Mol. weight | $C_{31}H_{39}NO_6*C_6H_{15}N$ g/mol |

## PEG2230 OPSS-PEG(4)-NHS

N-[3-(*o*-Pyridyldisulfido)propanoyl]-15-amino-4,7,10,13-tetraoxa-pentadecanoyl succinimidyl ester



| | |
|---|---|
| CAS-No. | 1334177-95-5 |
| Formula | $C_{23}H_{33}N_3O_9S_2$ |
| Mol. weight | 559,65 g/mol |

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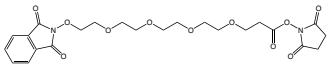
## PEG5080 Phth-NO-PEG(4)-NHS

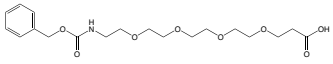
1-Phthalimidoxy-3,6,9,12-tetraoxapentadecan-15-oic acid succinimdyl ester



| | |
|---|---|
| CAS-No. | 1415328-95-8 |
| Formula | $C_{23}H_{28}N_2O_{11}$ |
| Mol. weight | 508,48 g/mol |

## PEG1495 Z-NH-PEG(4)-COOH

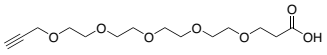
15-Benzyloxycarbonylamino-4,7,10,13-tetraoxa-pentadecanoic acid



| | |
|---|---|
| CAS-No. | 756526-00-8 |
| Formula | $C_{19}H_{29}NO_8$ |
| Mol. weight | 399,44 g/mol |

## PEG8170 Propargyl-PEG(5)-COOH

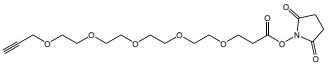
4,7,10,13,16-pentaoxanonadec-18-ynoic acid



| | |
|---|---|
| CAS-No. | 1245823-51-1 |
| Formula | $C_{14}H_{24}O_7$ |
| Mol. weight | 304,34 g/mol |

## PEG5410 Alkyne-PEG(4)-NHS

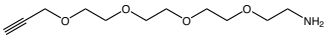
Alkyne-PEG(4)-succinimidyl ester



| | |
|---|---|
| CAS-No. | 1393330-40-9 |
| Formula | $C_{18}H_{27}NO_9$ |
| Mol. weight | 401,41 g/mol |

## PEG5430 Alkyne-PEG(4)-$NH_2$

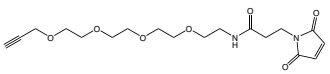
Alkyne-PEG(4)-amine



| | |
|---|---|
| CAS-No. | 1013921-36-2 |
| Formula | $C_{11}H_{21}NO_4$ |
| Mol. weight | 231,29 g/mol |

## PEG5440 Alkyne-PEG(4)-mal

Alkyne-PEG(4)-maleimide

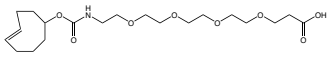


| | |
|---|---|
| CAS-No. | 1609651-90-2 |
| Formula | $C_{18}H_{26}N_2O_7$ |
| Mol. weight | 382,41 g/mol |

## TCO1040 TCO-PEG(4)-COOH

*trans*-Cyclooctene-PEG(4)-Acid

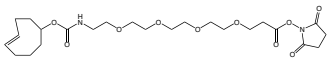
CAS-No. 1802913-21-8
Formula $C_{20}H_{35}NO_8$
Mol. weight 417,49 g/mol



## TCO1010 TCO-PEG(4)-NHS

*trans*-Cyclooctene-PEG(4)-carboxy succinimidyl ester

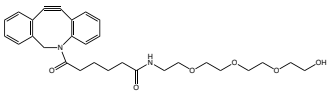
CAS-No. 1621096-79-4
Formula $C_{24}H_{38}N_2O_{10}$
Mol. weight 514,57 g/mol



## RL-2510 DBCO-PEG(4)-OH

Dibenzoazacyclooctyne-tetra(ethylene glycol)

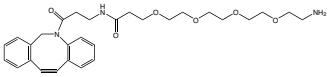
CAS-No. 1416711-60-8
Formula $C_{29}H_{36}N_2O_6$
Mol. weight 508,61 g/mol



## RL-2420 DBCO-PEG(4)-$NH_2$*TFA

Dibenzoazacyclooctyne-tetra(ethylene glycol)-amine trifluoro acetic acid salt

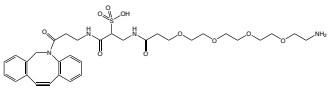
CAS-No. 1255942-08-5
Formula $C_{29}H_{37}N_3O_6*C_2F_3HO_2$
Mol. weight 523,62*114,02 g/mol



## RL-2421 DBCO-Sulfo-PEG(4)-$NH_2$

Dibenzoazacyclooctyne-tetra(ethylene glycol)amine

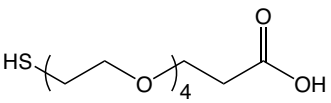
CAS-No. 2055198-05-3
Formula $C_{32}H_{42}N_4O_{10}S$
Mol. weight 674,76 g/mol



## PEG1970 HS-PEG(4)-COOH

15-Mercapto-4,7,10,13-tertaoxa-pentadecanoic acid

CAS-No. 749247-06-1
Formula $C_{11}H_{22}O_6S$
Mol. weight 282,35 g/mol

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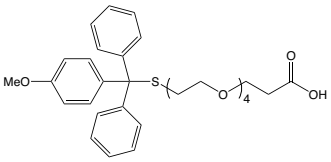
## PEG1740 Mmt-S-PEG(4)-COOH

15-(4-Methoxytrityl)thio-4,7,10,13-tertaoxa-pentadecanoic acid

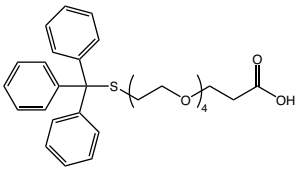
| | |
|---|---|
| CAS-No. | 1263047-31-9 |
| Formula | $C_{31}H_{38}O_7S$ |
| Mol. weight | 554,69 g/mol |



## PEG6710 Trt-S-PEG(4)-COOH*H2O

15-Tritylmercapto-4,7,10,13-tetraoxapentadecanoic acid monohydrate

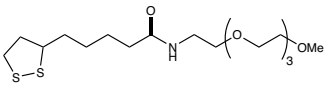
| | |
|---|---|
| CAS-No. | 882847-05-4 net |
| Formula | $C_{30}H_{36}O_6S*H_2O$ |
| Mol. weight | 524,67*18,01 g/mol |



## PEG3590 Lipoamide-PEG(4)-OMe

alpha-Lipoamide-omega-methoxy tetra(ethylene glycol)

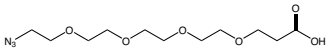
| | |
|---|---|
| CAS-No. | 1334172-66-5 |
| Formula | $C_{17}H_{33}NO_5S_2$ |
| Mol. weight | 395,58 g/mol |



## PEG2345 $N_3$-PEG(4)-COOH

15-Azido-4,7,10,13-tetraoxa-pentadecanoic acid

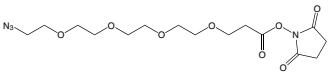
| | |
|---|---|
| CAS-No. | 1257063-35-6 |
| Formula | $C_{11}H_{21}N_3O_6$ |
| Mol. weight | 291,3 g/mol |



## PEG1400 $N_3$-PEG(4)-NHS

15-Azido-4,7,10,13-tetraoxa-pentadecanoic acid succinimidyl ester

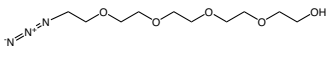
| | |
|---|---|
| CAS-No. | 944251-24-5 |
| Formula | $C_{15}H_{24}N_4O_8$ |
| Mol. weight | 388,37 g/mol |



## PEG5300 $N_3$-PEG(4)-OH

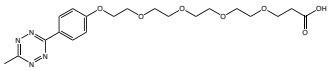
2-(2-(2-(2-(2-Azidoethoxy)ethoxy)ethoxy)ethoxy) ethanol

| | |
|---|---|
| CAS-No. | 86770-68-5 |
| Formula | $C_{10}H_{21}N_3O_5$ |
| Mol. weight | 263,29 g/mol |



## RL-2310 MeTz-PEG(4)-COOH

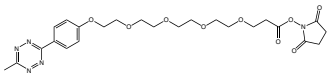
**Methyltetrazine-PEG(4)-acid**



CAS-No. 1802907-91-0
Formula $C_{20}H_{28}N_4O_7$
Mol. weight 436,56 g/mol

## RL-2330 MeTz-PEG(4)-NHS

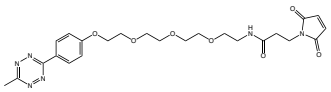
**Methyltetrazine-PEG(4)-propanoyl succinimidyl ester**



CAS-No. 1802907-92-1
Formula $C_{24}H_{31}N_5O_9$
Mol. weight 533,53 g/mol

## RL-2340 MeTz-PEG(4)-mal

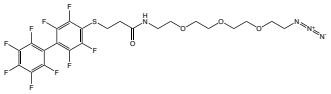
**Methyltetrazine-PEG(4)-maleimide**



CAS-No. 1802908-02-6
Formula $C_{24}H_{30}N_6O_7$
Mol. weight 514,53 g/mol

## RL-4030 PFB-mercaptopropionyl-PEG3-$N_3$

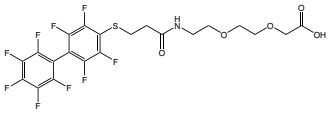
**Perfluorobiphenyl-mercaptopropionyl-PEG(3)-$N_3$**



Formula $C_{23}H_{21}F_9N_4O_4S$
Mol. weight 620,49 g/mol

## RL-4040 PFB-mercaptopropionyl-AEEA

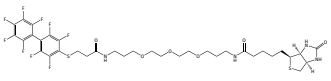
**Perfluorobiphenyl-mercaptopropionyl-AEEA**



Formula $C_{21}H_{16}F_9NO_5S$
Mol. weight 565,41 g/mol

## RL-4050 PFB-mercaptopropionyl-TOTA-Biotin

**Perfluorobiphenyl-mercaptopropionyl-TOTA-Biotin**



Formula $C_{35}H_{41}F_9N_4O_6S_2$
Mol. weight 848,84 g/mol

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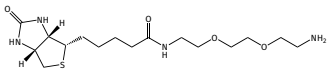
### RL-4060 Biotin-DOOA

Biotinyl-1-amino-3,6-dioxa-8-octanamine

CAS-No. 138529-46-1
Formula $C_{16}H_{30}N_4O_4S$
Mol. weight 374,50 g/mol



## 2.2. Hydrophobic Spacer Molecules

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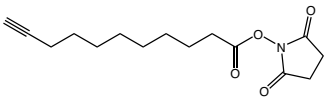
### RL-3460 10-Undecynoyl-OSu

10-Undecynoic acid N-hydroxysuccinimide ester

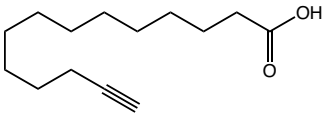
CAS-No. 1006592-57-9
Formula $C_{15}H_{21}NO_4$
Mol. weight 279,34 g/mol



### RL-2055 Alkyne-myristic acid

13-Tetradecynoic acid

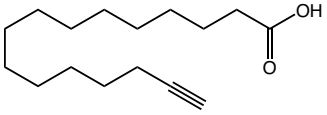
CAS-No. 82909-47-5
Formula $C_{14}H_{24}O_2$
Mol. weight 224,34 g/mol



### RL-2060 Alkyne-palmitic acid

15-Hexadecynoic acid

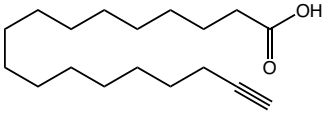
CAS-No. 99208-90-9
Formula $C_{16}H_{28}O_2$
Mol. weight 252,39 g/mol



### RL-2065 Alkyne-stearic acid

17-Octadecynoic acid

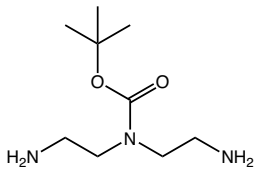
CAS-No. 34450-18-5
Formula $C_{18}H_{32}O_2$
Mol. weight 280,45 g/mol



## BNN1330 DETA(HBH)*2HCl

*tert*-butyl bis(2-aminoethyl)carbamate

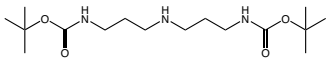
| | |
|---|---|
| CAS-No. | 1914917-65-9 |
| Formula | $C_9H_{21}N_3O_2$*2HCl |
| Mol. weight | 203,29*72,92 g/mol |



## BNN1340 DPTA(BHB)*HCl

di-*tert*-butyl (azanediylbis(propane-3,1-diyl))dicarbamate

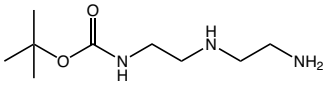
| | |
|---|---|
| CAS-No. | 82409-03-8 |
| Formula | $C_{16}H_{33}N_3O_4$*HCl |
| Mol. weight | 331,46*36,46 g/mol |



## BNN1350 DETA(BHH*2HCl)

*tert*-butyl (2-((2-aminoethyl)amino)ethyl)carbamate dihydrochloride

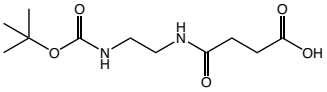
| | |
|---|---|
| CAS-No. | 162279-67-6 |
| Formula | $C_9H_{21}N_3O_2$*2HCl |
| Mol. weight | 203,29*72,92 g/mol |



## BNN1380 Boc-EDA-Suc-OH

Boc,Succinoyl-ethylenediamine

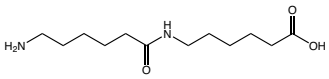
| | |
|---|---|
| CAS-No. | 891781-87-6 |
| Formula | $C_{11}H_{20}N_2O_5$ |
| Mol. weight | 260,29 g/mol |



## HAA9300 H-Aca-Aca-OH

6-(6-Aminohexanamido)hexanoic acid

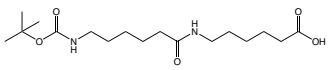
| | |
|---|---|
| CAS-No. | 2014-58-6 |
| Formula | $C_{12}H_{24}N_2O_3$ |
| Mol. weight | 244,34 g/mol |



## BAA4870 Boc-Aca-Aca-OH

N-Boc-6-(6-Aminohexanamido)hexanoic acid

| | |
|---|---|
| CAS-No. | 14254-45-6 |
| Formula | $C_{17}H_{32}N_2O_5$ |
| Mol. weight | 344,45 g/mol |



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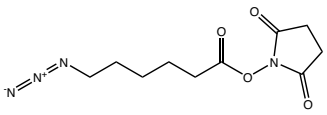
## RL-2980 $N_3$-Aca-OSu

6-Azidocaproic acid N-hydroxysuccinimidyl ester

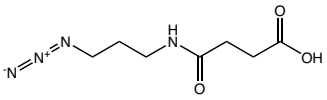
| | |
|---|---|
| CAS-No. | 866363-70-4 |
| Formula | $C_{10}H_{14}N_4O_4$ |
| Mol. weight | 254,24 g/mol |



## RL-4350 $N_3$-DAPr-Suc-OH

Azido-propylenediamine-succinoyl-OH

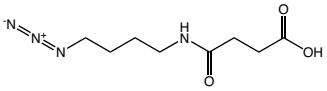
| | |
|---|---|
| CAS-No. | 929894-58-6 |
| Formula | $C_7H_{12}N_4O_3$ |
| Mol. weight | 200,20 g/mol |



## RL-4360 $N_3$-DABu-Suc-OH

Azido-butylenediamine-succinoyl-OH

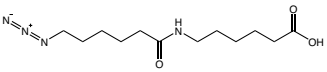
| | |
|---|---|
| CAS-No. | 2226183-50-0 |
| Formula | $C_8H_{14}N_4O_3$ |
| Mol. weight | 214,23 g/mol |



## HAA6990 $N_3$-Aca-Aca-OH

6-(6-azidohexanamido)hexanoic acid

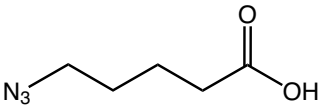
| | |
|---|---|
| CAS-No. | 866363-71-5 |
| Formula | $C_{12}H_{22}N_4O_3$ |
| Mol. weight | 270,33 g/mol |



## AAA1970 $N_3$-Pen-OH

5-Azido-pentanoic acid

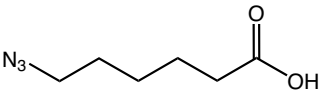
| | |
|---|---|
| CAS-No. | 79583-98-5 |
| Formula | $C_5H_9N_3O_2$ |
| Mol. weight | 143,14 g/mol |



## AAA1960 $N_3$-Hx-OH

6-Azido-hexanoic acid

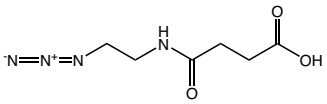
| | |
|---|---|
| CAS-No. | 79598-53-1 |
| Formula | $C_6H_{11}N_3O_2$ |
| Mol. weight | 157,17 g/mol |



## BNN1370 $N_3$-EDA-Suc-OH

Azido-ethylenediamine-succinoyl-OH

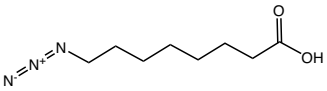
| | |
|---|---|
| CAS-No. | 2225891-73-4 |
| Formula | $C_6H_{10}N_4O_3$ |
| Mol. weight | 186,17 g/mol |



## RL-4430 $N_3$-C7H14-COOH

8-azidooctanoic acid

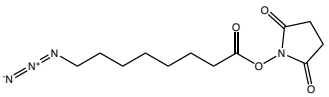
| | |
|---|---|
| CAS-No. | 217180-76-2 |
| Formula | $C_8H_{15}N_3O_2$ |
| Mol. weight | 185,23 g/mol |



## RL-3480 8-Azido-octanoyl-OSu

8-Azidodooctanoic acid N-hydroxysuccinimide ester

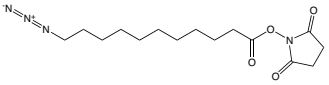
| | |
|---|---|
| CAS-No. | 2576471-56-0 |
| Formula | $C_{12}H_{18}N_4O_4$ |
| Mol. weight | 282,30 g/mol |



## RL-3170 11-Azido-undecanoyl-OSu

11-Azidoundecanoic acid N-hydroxysuccinimide ester

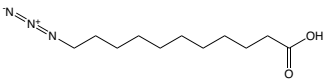
| | |
|---|---|
| CAS-No. | 850080-13-6 |
| Formula | $C_{15}H_{24}N_4O_4$ |
| Mol. weight | 324,38 g/mol |



## RL-3200 11-Azidoundecanoic acid

11-Azido-undecanoic acid

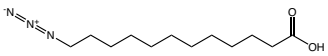
| | |
|---|---|
| CAS-No. | 118162-45-1 |
| Formula | $C_{11}H_{21}N_3O_2$ |
| Mol. weight | 227,30 g/mol |



## RL-3210 12-Azidododecanoic acid

12-Azido-dodecanoic acid

| | |
|---|---|
| CAS-No. | 80667-36-3 |
| Formula | $C_{12}H_{23}N_3O_2$ |
| Mol. weight | 241,33 g/mol |



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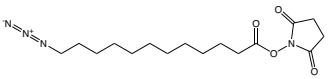
## RL-3220 12-Azido-dodecanoyl-OSu

12-Azidododecanoic acid N-hydroxysuccinimide ester

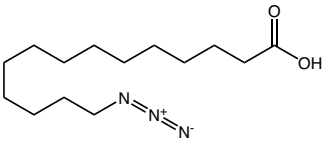
CAS-No. 2489524-00-5
Formula $C_{16}H_{26}N_4O_4$
Mol. weight 338,40 g/mol



## RL-3230 14-Azido-myristic acid

14-azidotetradecanoic acid

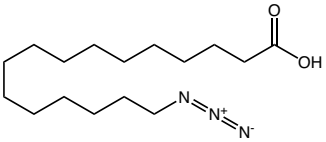
CAS-No. 176108-61-5
Formula $C_{14}H_{27}N_3O_2$
Mol. weight 269,38 g/mol



## RL-3240 16-Azido-palmitic acid

16-azidohexadecanoic acid

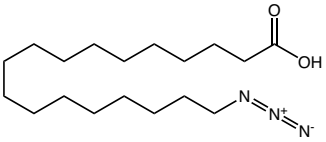
CAS-No. 112668-54-9
Formula $C_{16}H_{31}N_3O_2$
Mol. weight 297,44 g/mol



## RL-3250 18-Azido-stearic acid

18-azidooctadecanoic acid

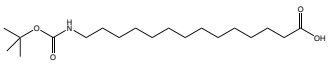
CAS-No. 1529763-58-3
Formula $C_{18}H_{35}N_3O_2$
Mol. weight 325,49 g/mol



## BAA4240 14-(Boc-amino)-myristic acid

14-((*t*-Butyloxycarbonyl)amino)tetradecanoic acid

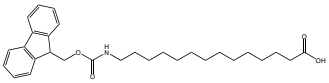
CAS-No. 2307778-46-5
Formula $C_{19}H_{37}NO_4$
Mol. weight 343,51 g/mol



## FAA8160 14-(Fmoc-amino)-myristic acid

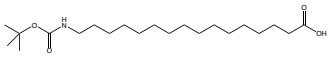
14-((9-Fluorenylmethyloxycarbonyl)amino)tetradecanoic acid

CAS-No. 1931109-55-5
Formula $C_{29}H_{39}NO_4$
Mol. weight 465,63 g/mol



## BAA3900 16-(Boc-amino)-palmitic acid

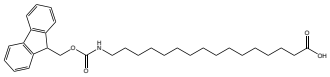
16-((*t*-Butyloxycarbonyl)amino)hexadecanoic acid



| | |
|---|---|
| CAS-No. | 135747-73-8 |
| Formula | $C_{21}H_{41}NO_4$ |
| Mol. weight | 371,55 g/mol |

## FAA7460 16-(Fmoc-amino)-palmitic acid

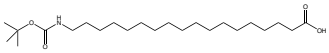
16-((9-Fluorenylmethyloxycarbonyl)amino)hexadecanoic acid



| | |
|---|---|
| CAS-No. | 1356220-22-8 |
| Formula | $C_{31}H_{43}NO_4$ |
| Mol. weight | 493,68 g/mol |

## BAA3910 18-(Boc-amino)-stearic acid

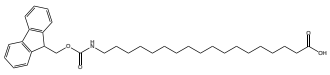
18-((*t*-Butyloxycarbonyl)amino)octadecanoic acid



| | |
|---|---|
| CAS-No. | 2389064-45-1 |
| Formula | $C_{23}H_{45}NO_4$ |
| Mol. weight | 399,61 g/mol |

## FAA7450 18-(Fmoc-amino)-stearic acid

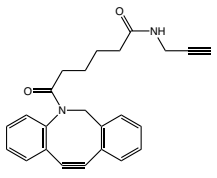
18-((9-Fluorenylmethyloxycarbonyl)amino)octadecanoic acid



| | |
|---|---|
| CAS-No. | 1199580-37-4 |
| Formula | $C_{33}H_{47}NO_4$ |
| Mol. weight | 521,73 g/mol |

## RL-4020 DBCO-C6-Alkyne

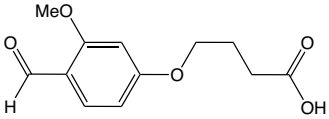
N-(propargylamidoadipoyl)-dibenzoazacyclooctyne




| | |
|---|---|
| Formula | $C_{24}H_{22}N_2O_2$ |
| Mol. weight | 370,45 g/mol |

## RL-1002 FMPB-Linker

4-(4'-Formyl-3'-methoxyphenoxy) butanoic acid




| | |
|---|---|
| CAS-No. | 309964-23-6 |
| Formula | $C_{12}H_{14}O_5$ |
| Mol. weight | 238,24 g/mol |

The Concept of Antibody-Drug Conjugation (ADC)
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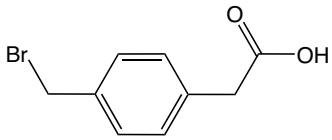
## RL-1008 Br-PAM-Linker

4-Bromomethylphenyl-acetic acid

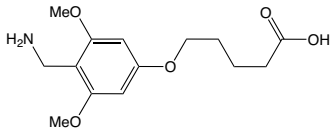
| | |
|---|---|
| CAS-No. | 13737-36-5 |
| Formula | $C_9H_9BrO_2$ |
| Mol. weight | 229,1 g/mol |




## RL-1050 H-PAL-Linker

5-[4-Aminomethyl-3,5-dimethoxyphenoxy]-pentanoic acid

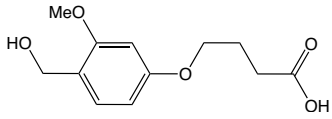
| | |
|---|---|
| CAS-No. | 125666-66-2 |
| Formula | $C_{14}H_{21}NO_5$ |
| Mol. weight | 283,31 g/mol |




## RL-1114 HMPB-Linker

4-(4-Hydroxymethyl-3-methoxyphenoxy)-butyric acid

| | |
|---|---|
| CAS-No. | 136849-75-7 |
| Formula | $C_{12}H_{16}O_5$ |
| Mol. weight | 240,24 g/mol |

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## 2.3. Permanent Linkers with Maleimide Function

Michael addition of a thiol to a maleimide is commonly used for numerous bioconjugations. Several commercial constructs like Brentuximab vedotin, Trastuzumab emtansine, and Cimzia contain a thiol-maleimide adduct. However, this reaction is reversible. During the journey of an appropriate thioether containing drug through physiological media, this bond can break, and fragments are released which might contribute to certain unwanted or even toxic reactions.

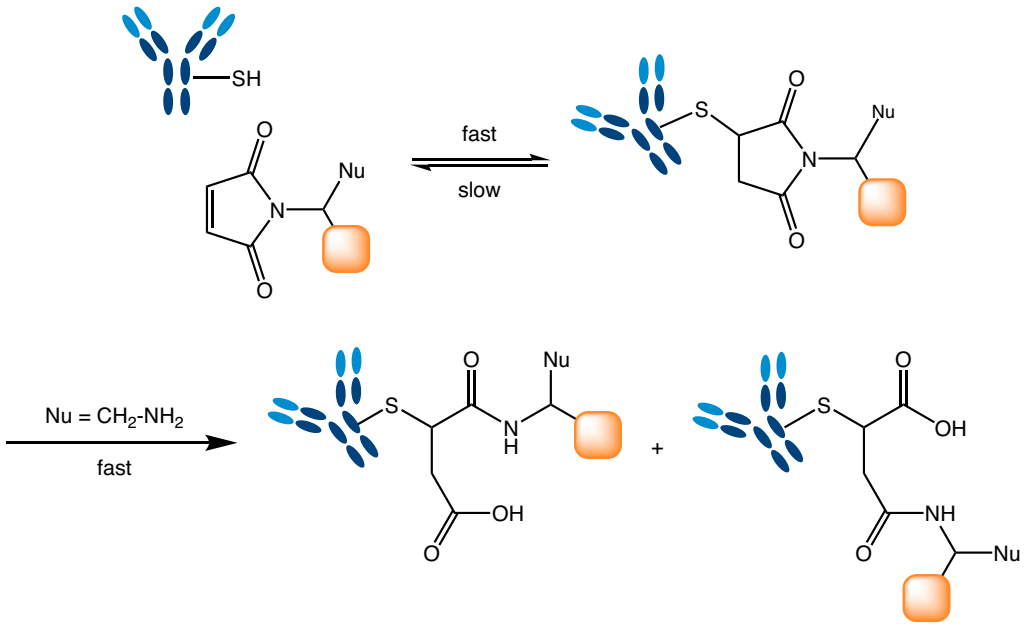


*Fig. 15: Maleimides bind reversibly to thiols resulting in thioethers. This linkage turns stable once the maleimide ring is opened through hydrolysis.*

However, if the succinimide moiety of a maleimide-thiol conjugate is hydrolyzed, the ring-opened product is fully stabilized towards cleavage (*Fig. 15*). The rates of ring-opening hydrolysis are greatly accelerated by electron withdrawing N-substituents and good nucleophiles in the proximity of the carbonyl functions. Thus, conjugates made with nucleophilic side-chains and electron-withdrawing maleimides may be purposefully hydrolyzed to their ring-opened counterparts and ensure good *in vivo* stability.

***References:***


→ *Covalent Modification of Biomolecules through Maleimide-Based Labeling Strategies; K. Renault, J. W. Fredy, P. Y. Renard, C. Sabot;* ***Bioconjug Chem*** *2018;* ***29****: 2497-2513.* https://doi.org/10.1021/acs.bioconjchem.8b00252

→ *Optimisation of the dibromomaleimide (DBM) platform for native antibody conjugation by accelerated post-conjugation hydrolysis; M. Morais, J. P. M. Nunes, K. Karu, N. Forte, I. Benni, M. E. B. Smith, S. Caddick, V. Chudasama, J. R. Baker;* ***Org Biomol Chem*** *2017;* ***15****: 2947-2952.* https://doi.org/10.1039/c7ob00220c

→ *Use of a next generation maleimide in combination with THIOMAB™ antibody technology delivers a highly stable, potent and near homogeneous THIOMAB™ antibody-drug conjugate (TDC); J. P. M. Nunes, V. Vassileva, E. Robinson, M. Morais, M. E. B. Smith, R. B. Pedley, S. Caddick, J. R. Baker, V. Chudasama;* ***RSC Advances*** *2017;* ***7****: 24828-24832.* https://doi.org/10.1039/c7ra04606e


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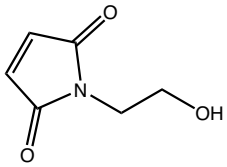
- *Long-term stabilization of maleimide-thiol conjugates; S. D. Fontaine, R. Reid, L. Robinson, G. W. Ashley, D. V. Santi;* ***Bioconjug Chem*** *2015;* ***26****: 145-52.* https://doi.org/10.1021/bc5005262
- *Self-hydrolyzing maleimides improve the stability and pharmacological properties of antibody-drug conjugates; R. P. Lyon, J. R. Setter, T. D. Bovee, S. O. Doronina, J. H. Hunter, M. E. Anderson, C. L. Balasubramanian, S. M. Duniho, C. I. Leiske, F. Li, P. D. Senter;* ***Nat Biotechnol*** *2014;* ***32****: 1059-62.* https://doi.org/10.1038/nbt.2968
- *Mild method for succinimide hydrolysis on ADCs: impact on ADC potency, stability, exposure, and efficacy; L. N. Tumey, M. Charati, T. He, E. Sousa, D. Ma, X. Han, T. Clark, J. Casavant, F. Loganzo, F. Barletta, J. Lucas, E. I. Graziani;* ***Bioconjug Chem*** *2014;* ***25****: 1871-80.* https://doi.org/10.1021/bc500357n


Product details

## RL-3000 Mal-Et-OH

N-(2-Hydroxyethyl)maleimide

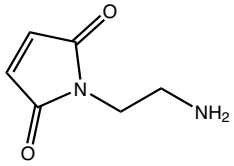
| | |
|---|---|
| CAS-No. | 1585-90-6 |
| Formula | $C_6H_7NO_3$ |
| Mol. weight | 141,12 g/mol |



## RL-2780 Mal-$NH_2$*HCl

2-Maleimidoethylamine hydrochloride

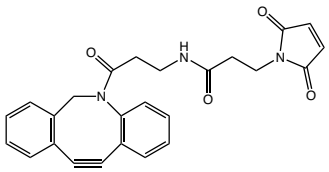
| | |
|---|---|
| CAS-No. | 134272-64-3 |
| Formula | $C_6H_8N_2O_2$*HCl |
| Mol. weight | 140,14*36,45 g/mol |



## RL-2490 DBCO-mal

Dibenzoazacyclooctyne-maleimide

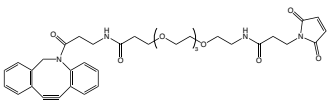
| | |
|---|---|
| CAS-No. | 1395786-30-7 |
| Formula | $C_{25}H_{21}N_3O_4$ |
| Mol. weight | 427,45 g/mol |



## RL-2500 DBCO-PEG(4)-mal

Dibenzoazacyclooctyne-tetra(ethylene glycol)-maleimide

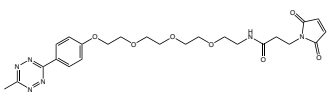
| | |
|---|---|
| CAS-No. | 1480516-75-3 |
| Formula | $C_{36}H_{42}N_4O_9$ |
| Mol. weight | 674,74 g/mol |



## RL-2340 MeTz-PEG(4)-mal

Methyltetrazine-PEG(4)-maleimide

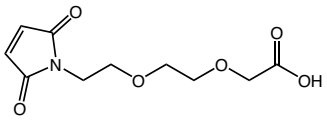
| | |
|---|---|
| CAS-No. | 1802908-02-6 |
| Formula | $C_{24}H_{30}N_6O_7$ |
| Mol. weight | 514,53 g/mol |



## PEG4870 Mal-$O_2$Oc-OH

{2-[2-(2,5-Dioxo-2,5-dihydro-1H-pyrrol-1-yl)ethoxy] ethoxy}acetic acid

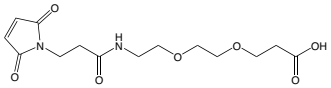
| | |
|---|---|
| CAS-No. | 173323-23-4 |
| Formula | $C_{10}H_{13}NO_6$ |
| Mol. weight | 243,21 g/mol |



## PEG1555 mal-PEG(2)-COOH

3-(2-(2-(3-Maleimidopropanamido)ethoxy)ethoxy) propanoic acid

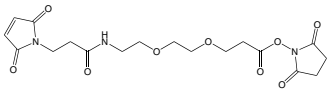
| | |
|---|---|
| CAS-No. | 756525-98-1 |
| Formula | $C_{14}H_{20}N_2O_7$ |
| Mol. weight | 328,32 g/mol |



## PEG1560 mal-PEG(2)-NHS

3-(2-(2-(3-Maleimidopropanamido)ethoxy)ethoxy) propanoic acid succinimidyl ester

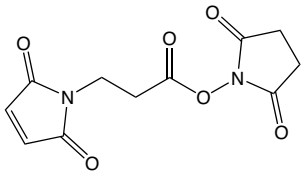
| | |
|---|---|
| CAS-No. | 955094-26-5 |
| Formula | $C_{18}H_{23}N_3O_9$ |
| Mol. weight | 425,39 g/mol |



## MAA1020 Mal-beta-Ala-OSu

3-(Maleimido)propionic acid N-succinimidyl ester

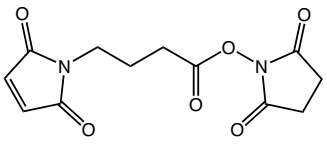
| | |
|---|---|
| CAS-No. | 55750-62-4 |
| Formula | $C_{11}H_{10}N_2O_6$ |
| Mol. weight | 266,21 g/mol |



## RL-2640 Mal-Bu-NHS

4-Maleimidobutyric acid-NHS ester

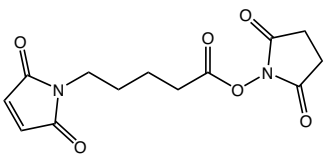
| | |
|---|---|
| CAS-No. | 80307-12-6 |
| Formula | $C_{12}H_{12}N_2O_6$ |
| Mol. weight | 280,23 g/mol |



## RL-2670 Mal-Pen-NHS

5-Maleimidopentanoic acid-NHS ester

| | |
|---|---|
| CAS-No. | 103750-03-4 |
| Formula | $C_{13}H_{14}N_2O_6$ |
| Mol. weight | 294,26 g/mol |



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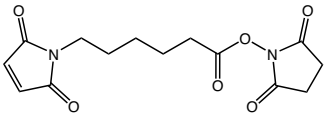
## RL-2660 Mal-Hx-NHS

6-Maleimidohexanoic acid-NHS ester

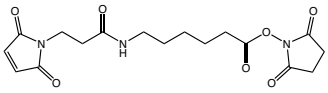
CAS-No. 55750-63-5
Formula $C_{14}H_{16}N_2O_6$
Mol. weight 308,29 g/mol



## RL-2690 Mal-PrHx-NHS

6-(3-Maleimidopropionylamino)-hexanoic acid-NHS ester

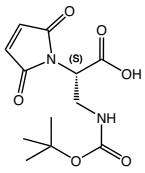
CAS-No. 367927-39-7
Formula $C_{17}H_{21}N_3O_7$
Mol. weight 379,36 g/mol



## MAA1040 Mal-L-Dap(Boc)-OH*DCHA

N-alpha-MaleimidoN-beta-*t*-butyloxycarbonyl-L-2,3-diaminopropionic acid dicyclohexylamine

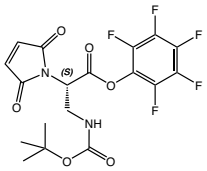
CAS-No. 2004724-16-5
Formula $C_{12}H_{16}N_2O_6*C_{12}H_{23}N$
Mol. weight 284,27*181,32 g/mol



## MAA1080 Mal-L-Dap(Boc)-OPfp

N-alpha-MaleimidoN-beta-*t*-butyloxycarbonyl-L-2,3-diaminopropionic acid pentafluorophenolate

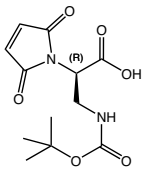
CAS-No. 1887132-90-2
Formula $C_{18}H_{15}F_5N_2O_6$
Mol. weight 450,31 g/mol



## MAA1060 Mal-D-Dap(Boc)-OH*DCHA

N-alpha-MaleimidoN-beta-*t*-butyloxycarbonyl-D-2,3-diaminopropionic acid dicyclohexylamine

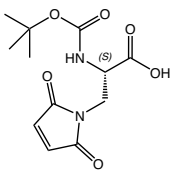
CAS-No. 2382651-11-6 net
Formula $C_{12}H_{16}N_2O_6*C_{12}H_{23}N$
Mol. weight 284,27*181,32 g/mol



## BAA6475 Boc-L-Dap(Mal)-OH*DCHA

(2S)-2-((*tert*-butoxycarbonyl)amino)-3-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)propanoic acid dicyclohexylamine

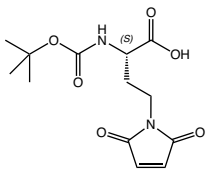
Formula $C_{12}H_{16}N_2O_6*C_{12}H_{23}N$
Mol. weight 284,27*181,32 g/mol



## BAA6480 Boc-L-Dab(Mal)-OH

(2S)-2-((*tert*-butoxycarbonyl)amino)-4-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)butanoic acid

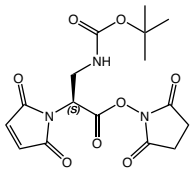
CAS-No. 135631-02-6
Formula $C_{13}H_{18}N_2O_6$
Mol. weight 298,30 g/mol



## MAA1120 Mal-L-Dap(Boc)-OSu

N-alpha-Maleimido-N-beta-Boc-L-2,3-diaminopropionic acid NHS ester

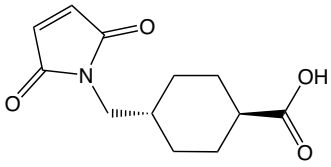
CAS-No. 1703778-79-3
Formula $C_{16}H_{19}N_3O_8$
Mol. weight 381,34 g/mol



## MAA5400 Mal-AMCHC-OH

*trans*-4-(maleimidomethyl)cyclohexane-1-carboxylic acid

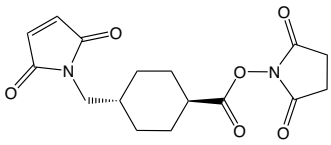
CAS-No. 69907-67-1
Formula $C_{12}H_{15}NO_4$
Mol. weight 237,25 g/mol



## MAA1000 Mal-AMCHC-OSu

*trans*-N-Succinimidyl 4-(maleimidomethyl)cyclohexane-1-carboxylate

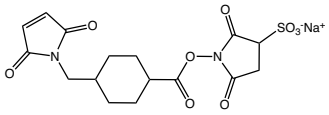
CAS-No. 71875-81-5
Formula $C_{16}H_{18}N_2O_6$
Mol. weight 334,33 g/mol



## MAA1050 Sulfo-SMCC

4-(N-Maleimidomethyl)cyclohexane-1-carboxylic acid 3-sulfo-N-hydroxysuccinimide ester sodium salt (*cis*/*trans* mixture)

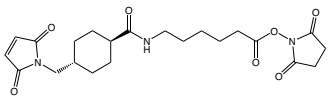
CAS-No. 92921-24-9
Formula $C_{16}H_{17}N_2NaO_9S$
Mol. weight 436,37 g/mol



## RL-2650 Mal-cHxHx-NHS

6-[*trans*-4-(Maleimidomethyl)-cyclohexanoylamino]-hexanoic acid-NHS ester

CAS-No. 125559-00-4
Formula $C_{22}H_{29}N_3O_7$
Mol. weight 447,48 g/mol



The Concept of Antibody-Drug Conjugation (ADC)
Permanent Linkers
Cleavable Linkers
Trifunctional Linkers
Cross-Linkers for other Bio Applications
Preparing Carriers for Conjugation
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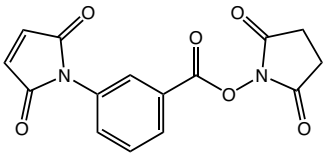
## RL-2600 3-Mal-Bz-NHS

3-Maleimidobenzoic acid-NHS ester

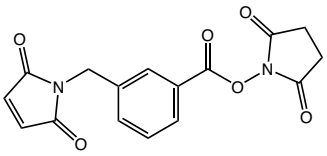
CAS-No. 58626-38-3
Formula $C_{15}H_{10}N_2O_6$
Mol. weight 314,25 g/mol



## RL-2610 3-Mal-MBz-NHS

3-(Maleimidomethyl)-benzoic acid-NHS ester

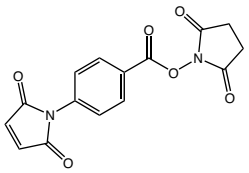
CAS-No. 91574-36-6
Formula $C_{16}H_{12}N_2O_6$
Mol. weight 328,28 g/mol



## RL-2620 4-Mal-Bz-NHS

4-Maleimidobenzoic acid-NHS ester

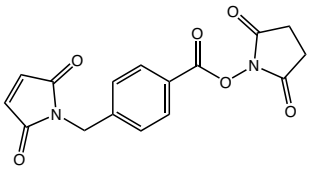
CAS-No. 64191-06-6
Formula $C_{15}H_{10}N_2O_6$
Mol. weight 314,25 g/mol



## RL-2630 4-Mal-MBz-NHS

4-(Maleimidomethyl)-benzoic acid-NHS ester

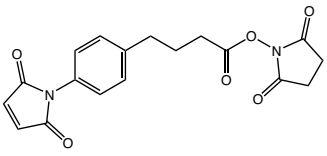
CAS-No. 64987-84-4
Formula $C_{16}H_{12}N_2O_6$
Mol. weight 328,28 g/mol



## RL-2680 Mal-PhBu-NHS

4-(4-Maleimidophenyl)-butyric acid-NHS ester

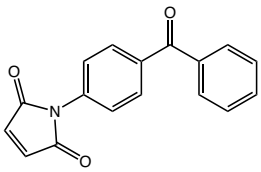
CAS-No. 79886-55-8
Formula $C_{18}H_{16}N_2O_6$
Mol. weight 356,33 g/mol



## LS-3350 4-(N-Maleimido)benzophenone

1-(4-Benzoylphenyl)-1H-pyrrole-2,5-dione

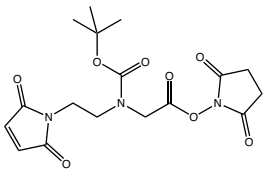
CAS-No. 92944-71-3
Formula $C_{17}H_{11}NO_3$
Mol. weight 277,28 g/mol



## RL-3430 Mal-N-Boc-Aeg-NHS

N-(*t*-butoxycarbonyl)-N-(2-(maleinimido)ethyl)glycine N-Hydroxysuccinimidyl ester

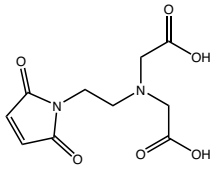
CAS-No. 2576471-29-7
Formula $C_{17}H_{21}N_3O_8$
Mol. weight 395,37 g/mol



## RL-3450 Mal-$CH_2CH_2$-N-(CH2-COOH)$_2$

2,2'-((2-(maleinimido)ethyl)azanediyl)diacetic acid

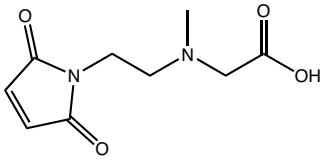
CAS-No. 207612-92-8
Formula $C_{10}H_{12}N_2O_6$
Mol. weight 256,21 g/mol



## RL-3400 Mal-$CH_2CH_2$-N(Me)-CH2-COOH

N-(2-(maleinimido)ethyl)-N-methylglycine

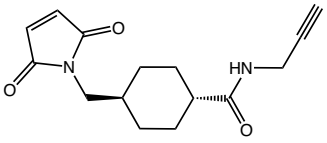
CAS-No. 2576471-52-6
Formula $C_9H_{12}N_2O_4$
Mol. weight 212,21 g/mol



## MAA1100 Mal-AMCHC-N-Propargylamide

*trans*-4-[(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)methyl]-N-(prop-2-yn-1-yl)cyclohexane-1-carboxamide

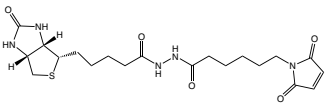
CAS-No. 2027476-42-0
Formula $C_{15}H_{18}N_2O_3$
Mol. weight 274,32 g/mol



## RL-8420 Biotin-NH-NH-Mal

6-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)-*N'*-(5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanoyl)hexanehydrazide

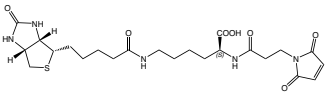
CAS-No. 116919-18-7
Formula $C_{20}H_{29}N_5O_5S$
Mol. weight 451,54 g/mol



## RL-8425 Biocytin-Mal

N2-(3-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)propanoyl)-N6-(5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanoyl)-L-lysine

CAS-No. 102849-12-7
Formula $C_{23}H_{33}N_5O_7S$
Mol. weight 523,61 g/mol



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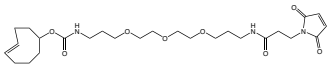
Product details

**TCO1050** **TCO-PEG(3)-mal**

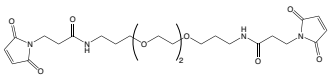
*trans*-Cyclooctene-PEG(3)-maleimide



| | |
|---|---|
| CAS-No. | 1809356-72-6 |
| Formula | $C_{26}H_{41}N_3O_8$ |
| Mol. weight | 523,62 g/mol |

**PEG1485** **mal-PEG(3)-mal**

Bis-(1,13-(3-maleimidopropionyl)amido)-4,7,10-trioxatridecane



| | |
|---|---|
| CAS-No. | 756525-89-0 |
| Formula | $C_{24}H_{34}N_4O_9$ |
| Mol. weight | 522,55 g/mol |

## 2.4. 5HP2O as Maleimide Alternative

Despite various possibilities for the modification of and conjugation to side-chain functional groups of histidine, lysine, methionine, tryptophane, and tyrosine, respectively, cysteine remains the most attractive target due to its rare occurrence in natural proteins typically allowing for its site-selective modification upon introduction at a specific position. In terms of cysteine modification, maleimides are the reactive moieties of choice acting *via* a Michael addition reaction. However, maleimides are susceptible to hydrolysis, retro-Michael reactions and/or thiol exchange reactions leading to protein conjugates with overall poor stability.

In this context, starting from furan, the research group around Prof. Annemieke Madder developed 5-hydroxy-1,5-dihydro-2H-pyrrol-2-ones (5HP2Os) – representatives of the 5-hydroxy-pyrrolone family – as maleimide replacement technology avoiding all the above-mentioned maleimide-drawbacks. The substituents R1 and R2 – as shown in the scheme below – allow for further derivatization and can be chosen depending on the intended subsequent use.

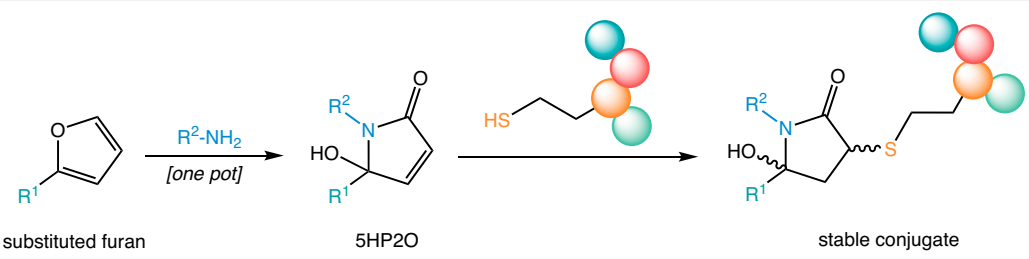



*Fig. 16: Singlet oxygen-induced formation of 5HP2O-derivatives in one pot starting from a substituted furan and a substituted-amine in the presence of methylene blue, oxygen, and dimethyl sulfide in methanol upon visible light irradiation. The 5HP2O reacts with a peptide containing a sulfhydryl group (typically from a cysteine). This reaction usually completes within 1 hour with 0.3 equivalents Na4EDTA in MeOH or with aqueous MeCN (3/2) at pH 8.0 as solvent.*

## Interested in more details about 5HP2O as maleimide replacement technology?

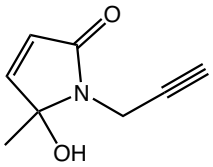
### Watch the recording of our online workshop!

Product details

### RL-8670 5HP2O-alkyne

5-hydroxy-5-methyl-1-(prop-2-yn-1-yl)-1,5-dihydro-2H-pyrrol-2-one

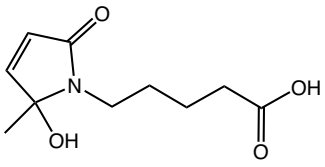
| | |
|---|---|
| CAS-No. | 2484704-61-0 |
| Formula | $C_8H_9NO_2$ |
| Mol. weight | 151,16 g/mol |



### RL-8675 5HP2O-(CH2)4-COOH

5-(2-hydroxy-2-methyl-5-oxo-2,5-dihydro-1H-pyrrol-1-yl)pentanoic acid

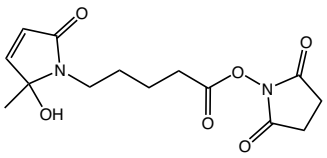
| | |
|---|---|
| Formula | $C_{10}H_{15}NO_4$ |
| Mol. weight | 213,23 g/mol |



### RL-8680 5HP2O-(CH2)4-NHS

2,5-dioxopyrrolidin-1-yl 5-(2-hydroxy-2-methyl-5-oxo-2,5-dihydro-1H-pyrrol-1-yl)pentanoate

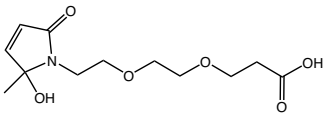
| | |
|---|---|
| Formula | $C_{14}H_{18}N_2O_6$ |
| Mol. weight | 310,31 g/mol |



### RL-8685 5HP2O-PEG(2)-COOH

3-(2-(2-(2-hydroxy-2-methyl-5-oxo-2,5-dihydro-1H-pyrrol-1-yl)ethoxy)ethoxy)propanoic acid

| | |
|---|---|
| Formula | $C_{12}H_{19}NO_6$ |
| Mol. weight | 273,28 g/mol |



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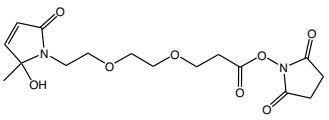
### RL-8690 5HP2O-PEG(2)-NHS

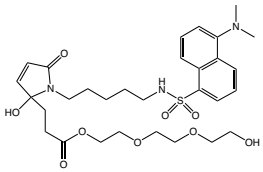
2,5-dioxopyrrolidin-1-yl 3-(2-(2-(2-hydroxy-2-methyl-5-oxo-2,5-dihydro-1H-pyrrol-1-yl)ethoxy)ethoxy) propanoate



Formula $C_{16}H_{22}N_2O_8$
Mol. weight 370,36 g/mol

### LS-4670 5HP2O((PEG)2-OH)-(CH2)5-Dansyl

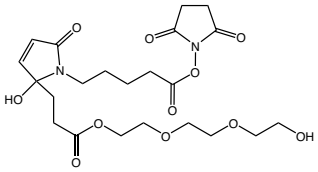
2-(2-(2-hydroxyethoxy)ethoxy)ethyl 3-(1-(5-((5-(dimethylamino)naphthalene)-1-sulfonamido)pentyl)-2-hydroxy-5-oxo-2,5-dihydro-1H-pyrrol-2-yl)propanoate



Formula $C_{30}H_{43}N_3O_9S$
Mol. weight 621,75 g/mol

### RL-8695 5HP2O((PEG)2-OH)-(CH2)4-NHS

2,5-dioxopyrrolidin-1-yl 5-(2-hydroxy-2-(3-(2-(2-(2-hydroxyethoxy)ethoxy)ethoxy)-3-oxopropyl)-5-oxo-2,5-dihydro-1H-pyrrol-1-yl)pentanoate



Formula $C_{22}H_{32}N_2O_{11}$
Mol. weight 500,50 g/mol

***References:***


- *Bioconjugation Reagent and Methods. A. Madder, E. De Geyter, E. Antonatou, S. Smolen, D. Kalaitzakis, D. Vassilikogiannakis; 2020. WO2020/174086A2*
- *2-Hydroxypyrrolidin-5-ones for bioconjugation and methods for their production. A. Madder, E. De Geyter, E. Antonatou, S. Smolen, D. Kalaitzakis, D. Vassilikogiannakis; 2020. WO2020/174086A3*
- *5-Hydroxy-pyrrolone based building blocks as maleimide alternatives for protein bioconjugation and single-site multi-functionalization; E. De Geyter, E. Antonatou, D. Kalaitzakis, S. Smolen, A. Iyer, L. Tack, E. Ongenae, G. Vassilikogiannakis, A. Madder; **Chem. Sci.** 2021; **12**: 5246-5252. https://doi.org/10.1039/d0sc05881e*


## 2.5. Photoactivatable Linkers

Irradiation of diazirines with UV light (ca. 350-360 nm) yields a highly reactive carbene species that can undergo insertions into C–C, C–H, O–H, and X–H (X = heteroatom) bonds of neighboring molecules to irreversibly form a covalent bond (*Fig. 17*). The diazirine moiety is the smallest of all photoreactive groups, so introduction of a diazirine-bearing amino acid into a peptide or protein usually does not impair its biological activity. Further, advantages of diazirine crosslinkers are their stability at room temperature and their relative stability against nucleophiles as well as towards both acidic and basic conditions.

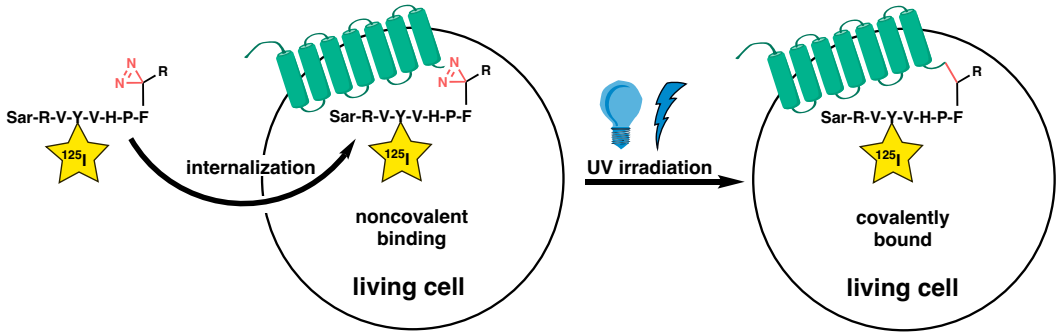



*Fig. 17: Use of photo-phenylalanine for the identification of angiotensin-II receptor binding sites; $^{125}I$ is used as a radiotracer.*

***References:***


- *Protein-polymer conjugation via ligand affinity and photoactivation of glutathione S-transferase; E. W. Lin, N. Boehnke, H. D. Maynard;* ***Bioconjug Chem*** *2014;* ***25****: 1902-9.* https://doi.org/10.1021/bc500380r
- *Cell-based proteome profiling of potential dasatinib targets by use of affinity-based probes; H. Shi, C. J. Zhang, G. Y. Chen, S. Q. Yao;* ***J Am Chem Soc*** *2012;* ***134****: 3001-14.* https://doi.org/10.1021/ja208518u
- *Probing protein-protein interactions with a genetically encoded photo-crosslinking amino acid; H. W. Ai, W. Shen, A. Sagi, P. R. Chen, P. G. Schultz;* ***ChemBioChem*** *2011;* ***12****: 1854-7.* https://doi.org/10.1002/cbic.201100194
- *Proteome profiling reveals potential cellular targets of staurosporine using a clickable cell-permeable probe; H. Shi, X. Cheng, S. K. Sze, S. Q. Yao;* ***Chem Commun*** *2011;* ***47****: 11306-8.* https://doi.org/10.1039/c1cc14824a
- *Direct interaction between an allosteric agonist pepducin and the chemokine receptor CXCR4; J. M. Janz, Y. Ren, R. Looby, M. A. Kazmi, P. Sachdev, A. Grunbeck, L. Haggis, D. Chinnapen, A. Y. Lin, C. Seibert, T. McMurry, K. E. Carlson, T. W. Muir, S. Hunt, 3rd, T. P. Sakmar;* ***J Am Chem Soc*** *2011;* ***133****: 15878-81.* https://doi.org/10.1021/ja206661w
- *Aliphatic diazirines as photoaffinity probes for proteins: recent developments; J. Das;* ***Chem Rev*** *2011;* ***111****: 4405-17.* https://doi.org/10.1021/cr1002722
- *Synthesis and application of photoproline - a photoactivatable derivative of proline; B. VanderMeijden, J. A. Robinson;* ***Arkivoc*** *2011;* ***2011****: 130-136.* https://doi.org/10.3998/ark.5550190.0012.611
- *Photo-crosslinking of proteins in intact cells reveals a dimeric structure of cyclooxygenase-2 and an inhibitor-sensitive oligomeric structure of microsomal prostaglandin E2 synthase-1; P. O. Hetu, M. Ouellet, J. P. Falgueyret, C. Ramachandran, J. Robichaud, R. Zamboni, D. Riendeau;* ***Arch Biochem Biophys*** *2008;* ***477****: 155-62.* https://doi.org/10.1016/j.abb.2008.04.038
- *Covalent capture of phospho-dependent protein oligomerization by site-specific incorporation of a diazirine photo-cross-linker; M. Vila-Perello, M. R. Pratt, F. Tulin, T. W. Muir;* ***J Am Chem Soc*** *2007;* ***129****: 8068-9.* https://doi.org/10.1021/ja072013j
- *Photo-leucine incorporation reveals the target of a cyclodepsipeptide inhibitor of cotranslational translocation; A. L. MacKinnon, J. L. Garrison, R. S. Hegde, J. Taunton;* ***J Am Chem Soc*** *2007;* ***129****: 14560-1.* https://doi.org/10.1021/ja076250y
- *Synthesis of photoactive analogues of a cystine knot trypsin inhibitor protein; T. Durek, J. Zhang, C. He, S. B. Kent;* ***Org Lett*** *2007;* ***9****: 5497-500.* https://doi.org/10.1021/ol702461z
- *Photo-leucine and photo-methionine allow identification of protein-protein interactions in living cells; M. Suchanek, A. Radzikowska, C. Thiele;* ***Nat Methods*** *2005;* ***2****: 261-7.* https://doi.org/10.1038/nmeth752



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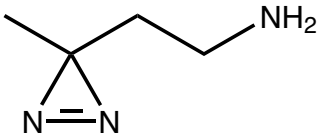
Product details

## RL-2910 Photo-Butylamine

2-(3-methyl-3H-diazirin-3-yl)ethan-1-amine hydrochloride

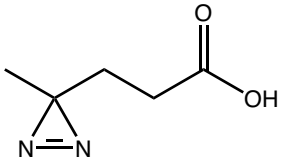
| | |
|---|---|
| CAS-No. | 25055-95-2 |
| Formula | $C_4H_9N_3$*HCl |
| Mol. weight | 99,13*36,45 g/mol |




## RL-2890 Photo-Pentanoic acid

3-(3-methyl-3H-diazirin-3-yl)propanoic acid

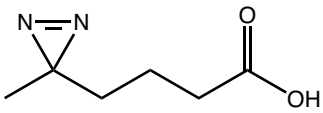
| | |
|---|---|
| CAS-No. | 25055-86-1 |
| Formula | $C_5H_8N_2O_2$ |
| Mol. weight | 128,13 g/mol |




## RL-2900 Photo-Hexanoic acid

4-(3-methyl-3H-diazirin-3-yl)butanoic acid

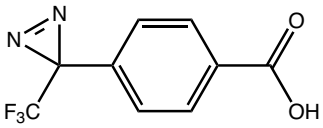
| | |
|---|---|
| CAS-No. | 16297-97-5 |
| Formula | $C_6H_{10}N_2O_2$ |
| Mol. weight | 142,16 g/mol |




## RL-2920 Photo-Benzoic acid

4-[3-(Trifluoromethyl)-3H-diazirin-3-yl]benzoic acid

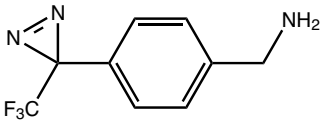
| | |
|---|---|
| CAS-No. | 85559-46-2 |
| Formula | $C_9H_5F_3N_2O_2$ |
| Mol. weight | 230,14 g/mol |




## RL-2930 Photo-Benzylamine*HCl

4-[3-(Trifluoromethyl)-3H-diazirin-3-yl]benzylamine hydrochloride

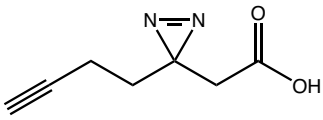
| | |
|---|---|
| CAS-No. | 1258874-29-1 |
| Formula | $C_9H_8N_3F_3$*HCl |
| Mol. weight | 215,18*36,45 g/mol |




## RL-3410 Photo-Click-Heptanoic acid

2-(3-(but-3-ynyl)-3H-diazirin-3-yl)acetic acid

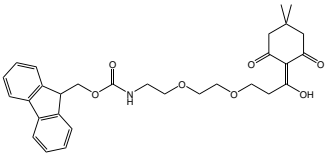
| | |
|---|---|
| CAS-No. | 2049109-24-0 |
| Formula | $C_7H_8N_2O_2$ |
| Mol. weight | 152,15 g/mol |



## RL-3270 Fmoc-AEEP-DIM

3-(2-(2-(9-Fluorenylmethyl)oxycarbonylaminoethoxy) ethoxy)-1-(4,4-dimethyl-2,6-dioxocyclohexylide-ne)-propan-1-ol

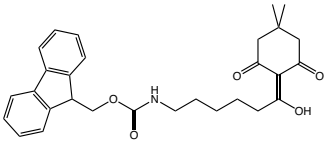
| | |
|---|---|
| CAS-No. | 1988771-96-5 |
| Formula | $C_{30}H_{35}NO_7$ |
| Mol. weight | 521,60 g/mol |



## RL-3260 Fmoc-Aca-DIM

6-((9-Fluorenylmethyl)oxycarbonylamino)-1-(4,4-di-methyl-2,6-dioxocyclohexylidene)-hexan-1-ol

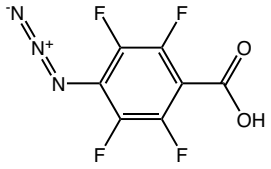
| | |
|---|---|
| CAS-No. | 2379561-08-5 |
| Formula | $C_{29}H_{33}NO_5$ |
| Mol. weight | 475,58 g/mol |



## RL-2035 ATFB

4-Azido-2,3,5,6-tetrafluorobenzoic acid

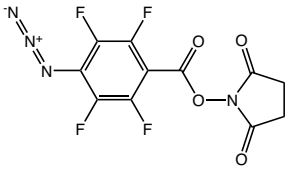
| | |
|---|---|
| CAS-No. | 122590-77-6 |
| Formula | $C_7HF_4N_3O_2$ |
| Mol. weight | 235,1 g/mol |



## RL-2045 ATFB-NHS

N-Succinimidyl 4-azido-2,3,5,6-tetrafluorobenzoate

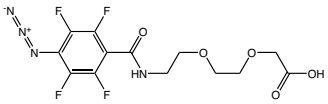
| | |
|---|---|
| CAS-No. | 126695-58-7 |
| Formula | $C_{11}H_4F_4N_4O_4$ |
| Mol. weight | 332,17 g/mol |



## PEG5000 $N_3$-TFBA-$O_2$Oc

{2-[2-(4-Azido-2,3,5,6-tetrafluorobenzoyl-amino)ethoxy] ethoxy}acetic acid

| | |
|---|---|
| CAS-No. | 1993119-45-1 |
| Formula | $C_{13}H_{12}F_4N_4O_5$ |
| Mol. weight | 380,25 g/mol |



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***References:***


- *Tri- and Tetravalent Photoactivable Cross-Linking Agents; A. Welle, F. Billard, J. Marchand-Brynaert;* ***Synthesis*** *2012;* ***44****: 2249-2254.* https://doi.org/10.1055/s-0031-1290444
- *Perfluorophenyl azides: new applications in surface functionalization and nanomaterial synthesis; L. H. Liu, M. Yan;* ***Acc Chem Res*** *2010;* ***43****: 1434-43.* https://doi.org/10.1021/ar100066t
- *Chemistry of Bifunctional Photoprobes; R. S. Pandurangi, P. Lusiak, S. Desai, R. R. Kuntz;* ***Bioorganic Chemistry*** *1998;* ***26****: 201-212.* https://doi.org/10.1006/bioo.1998.1098
- *Recent Trends in the Evaluation of Photochemical Insertion Characteristics of Heterobifunctional Perfluoroaryl Azide Chelating Agents: Biochemical Implications in Nuclear Medicine; R. S. Pandurangi, S. R. Karra, R. R. Kuntz, W. A. Volkert;* ***Photochemistry and Photobiology*** *1997;* ***65****: 208-221.* https://doi.org/10.1111/j.1751-1097.1997.tb08547.x
- *Chemistry of Bifunctional Photoprobes. 1. Perfluoroaryl Azido Functionalized Phosphorus Hydrazides as Novel Photoreactive Heterobifunctional Chelating Agents: High Efficiency Nitrene Insertion on Model Solvents and Proteins; R. S. Pandurangi, S. R. Karra, K. V. Katti, R. R. Kuntz, W. A. Volkert;* ***J Org Chem*** *1997;* ***62****: 2798-2807.* https://doi.org/10.1021/jo961867b


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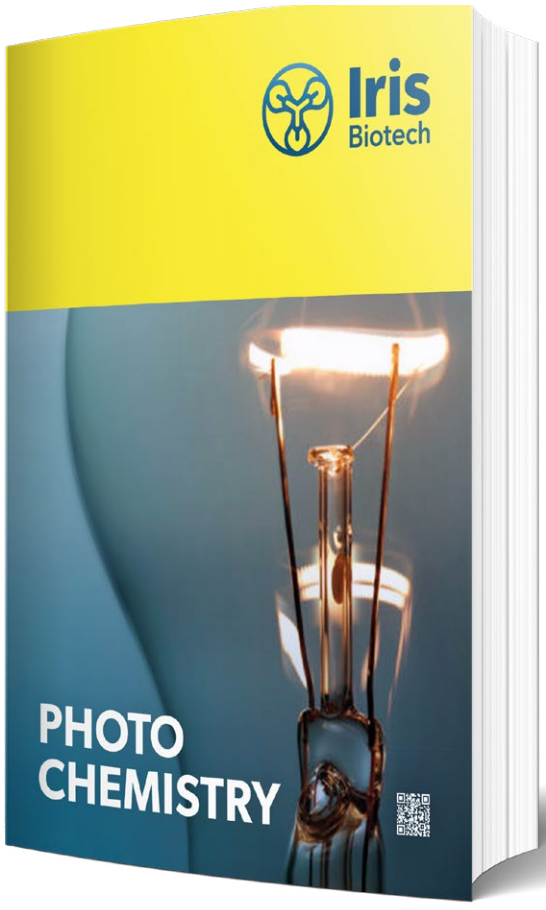

# 3. Cleavable Linkers

Peptidic bonds are expected to have a high serum stability, as lysosomal proteolytic enzymes show reduced activities in blood due to endogenous inhibitors and the unfavorably high pH value of blood compared to lysosomes. This was confirmed by preclinical *in vivo* studies, which revealed half-lives of seven to ten days for peptide linkers. Release of a drug conjugated *via* a peptidyl linker to monoclonal antibodies (mAb) occurs specifically due to the action of lysosomal proteases (e.g., cathepsin and plasmin). These proteases may be present at elevated levels in certain tumor tissues. Therefore, peptide linkers combine greater systemic stability with rapid enzymatic release of the drug in the target cell. Besides Val-Ala, Val-Cit and Phe-Lys, other sequences have been reported as lysosomally cleavable peptides, like Gly-Phe-Leu-Gly and Ala-Leu-Ala-Leu.

***References:***


- *Star structure of antibody-targeted HPMA copolymer-bound doxorubicin: a novel type of polymeric conjugate for targeted drug delivery with potent antitumor effect; M. Kovar, J. Strohalm, T. Etrych, K. Ulbrich, B. Rihova;* ***Bioconjug Chem*** *2002;* ***13****: 206-15.* https://doi.org/10.1021/bc010063m
- *Synthesis of a lipophilic daunorubicin derivative and its incorporation into lipidic carriers developed for LDL receptor-mediated tumor therapy; A. J. Versluis, E. T. Rump, P. C. Rensen, T. J. Van Berkel, M. K. Bijsterbosch;* ***Pharm Res*** *1998;* ***15****: 531-7.* https://doi.org/10.1023/a:1011917508056
- *Influence of a peptide linker on biodistribution and metabolism of antibody-conjugated benzyl-EDTA. Comparison of enzymatic digestion in vitro and in vivo; M. Studer, L. A. Kroger, S. J. DeNardo, D. L. Kukis and C. F. Meares;* ***Bioconjug Chem*** *1992;* ***3****: 424-9.* https://doi.org/10.1021/bc00017a012


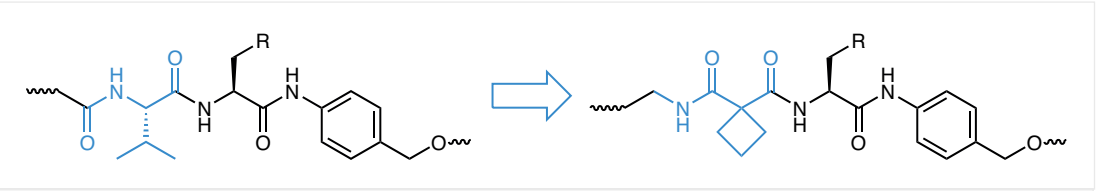

*Fig. 18: Cyclobutane-1,1-dicarboxamide can replace valine in dipeptide linker systems, resulting in improved ADC selectivity.*

Peptide-based ADC linkers, such as Val-Cit or Val-Ala, that are cleaved by lysosomal proteases have shown sufficient stability in serum and effective payload-release in targeted cells. However, the use of peptide-based linkers limits the ability to modulate protease specificity. Furthermore, if the linker can preferentially be hydrolyzed by tumor-specific proteases only, safety margin may improve. In this context, a cyclobutane-1,1-dicarboxamide-containing linker (*Fig. 18*) replacing valine in other sequences has been invented which is hydrolyzed predominantly by cathepsin B, while the typical valine-citrulline dipeptide linker is rather less. ADCs bearing the nonpeptidic linker are as efficacious and stable *in vivo* as those with the dipeptide linker. Hence, the application of the peptidomimetic linker presents new opportunities for improving the selectivity of ADCs.



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***Reference:***

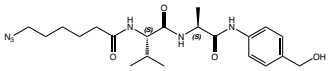
→ *Discovery of Peptidomimetic Antibody-Drug Conjugate Linkers with Enhanced Protease Specificity; B. Wei, J. Gunzner-Toste, H. Yao, T. Wang, J. Wang, Z. Xu, J. Chen, J. Wai, J. Nonomiya, S. P. Tsai, J. Chuh, K. R. Kozak, Y. Liu, S. F. Yu, J. Lau, G. Li, G. D. Phillips, D. Leipold, A. Kamath, D. Su, K. Xu, C. Eigenbrot, S. Steinbacher, R. Ohri, H. Raab, L. R. Staben, G. Zhao, J. A. Flygare, T. H. Pillow, V. Verma, L. A. Masterson, P. W. Howard, B. Safina;* ***J. Med. Chem.*** *2018;* ***61****: 989-1000.* https://doi.org/10.1021/acs.jmedchem.7b01430

## 3.1. Valine-Alanine-Based Enzymatically Cleavable Linkers

Product details

### ADC1290 6-Azidohexanoyl-Val-Ala-PAB

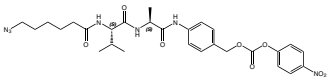
6-azidohexanoyl-valyl-alanyl-(4-aminobenzyl alcohol)



| | |
|---|---|
| CAS-No. | 2706564-30-7 |
| Formula | $C_{21}H_{32}N_6O_4$ |
| Mol. weight | 432,52 g/mol |

### ADC1300 6-Azidohexanoyl-Val-Ala-PAB-PNP

6-azidohexanoyl-valyl-alanyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| Formula | $C_{28}H_{35}N_7O_8$ |
| Mol. weight | 597,62 g/mol |

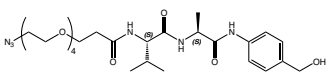
***Reference:***

→ *NKT cell-dependent glycolipid-peptide vaccines with potent anti-tumour activity; R. J. Anderson, B. J. Compton, C. W. Tang, A. Authier-Hall, C. M. Hayman, G. W. Swinerd, R. Kowalczyk, P. Harris, M. A. Brimble, D. S. Larsen, O. Gasser, R. Weinkove, I. F. Hermans, G. F. Painter;* ***Chem. Sci.*** *2015;* ***6****: 5120-5127.* https://doi.org/10.1039/c4sc03599b

Product details

### ADC1330 Azido-PEG(4)-Val-Ala-PAB

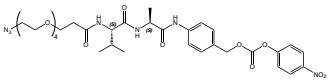
azido-tetraethyleneglycol-propanoyl-valyl-alanyl-(4-aminobenzyl alcohol)



| | |
|---|---|
| Formula | $C_{26}H_{42}N_6O_8$ |
| Mol. weight | 566,65 g/mol |

## ADC1340 Azido-PEG(4)-Val-Ala-PAB-PNP

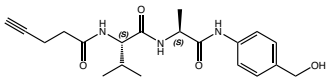
azido-tetraethyleneglycol-propanoyl-valyl-alanyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| Formula | $C_{33}H_{45}N_7O_{12}$ |
| Mol. weight | 731,75 g/mol |

## ADC1310 4-Pentynoyl-Val-Ala-PAB

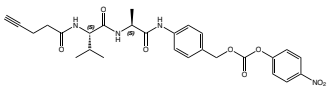
4-pentynoyl-valyl-alanyl-(4-aminobenzyl alcohol)



| | |
|---|---|
| CAS-No. | 1956294-75-9 |
| Formula | $C_{20}H_{27}N_3O_4$ |
| Mol. weight | 373,45 g/mol |

## ADC1320 4-Pentynoyl-Val-Ala-PAB-PNP

4-pentynoyl-valyl-alanyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| CAS-No. | 1956294-76-0 |
| Formula | $C_{27}H_{30}N_4O_8$ |
| Mol. weight | 538,55 g/mol |

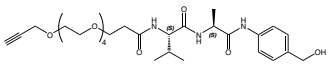
***Reference:***

→ *Integrin-Targeting Knottin Peptide-Drug Conjugates Are Potent Inhibitors of Tumor Cell Proliferation; N. Cox, J. R. Kintzing, M. Smith, G. A. Grant, J. R. Cochran;* ***Angew Chem Int Ed*** *2016;* ***55****: 9894-7.*
*https://doi.org/10.1002/anie.201603488*

Product details

## ADC1350 Alkyne-PEG(4)-Val-Ala-PAB

propargyl-tetraethyleneglycol-propanoyl-valyl-alanyl-(4-aminobenzyl alcohol)



| | |
|---|---|
| CAS-No. | 2348405-90-1 |
| Formula | $C_{29}H_{45}N_3O_9$ |
| Mol. weight | 579,68 g/mol |

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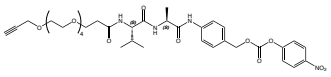
Product details

## ADC1360 Alkyne-PEG(4)-Val-Ala-PAB-PNP

propargyl-tetraethyleneglycol-propanoyl-valyl-alanyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| CAS-No. | 2348405-91-2 |
| Formula | $C_{36}H_{48}N_4O_{13}$ |
| Mol. weight | 744,79 g/mol |

***References:***

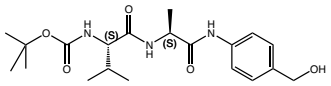

→ *Exploration of the carmaphycins as payloads in antibody drug conjugate anticancer agents; J. Almaliti, B. Miller, H. Pietraszkiewicz, E. Glukhov, C. B. Naman, T. Kline, J. Hanson, X. Li, S. Zhou, F. A. Valeriote, W. H. Gerwick;* ***Eur J Med Chem*** *2019;* ***161****: 416-432.* https://doi.org/10.1016/j.ejmech.2018.10.024

→ *Design and synthesis of novel dual-cyclic RGD peptides for alphavbeta3 integrin targeting; J. Liu, X. Cheng, X. Tian, D. Guan, J. Ao, Z. Wu, W. Huang, Z. Le;* ***Bioorg Med Chem Lett*** *2019;* ***29****: 896-900.* https://doi.org/10.1016/j.bmcl.2019.01.043


Product details

## ADC1040 Boc-Val-Ala-PAB

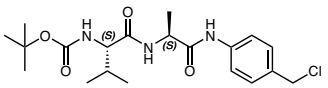
*t*-Butyloxycarbonyl-valyl-alanyl-4-aminobenzylalcohol



| | |
|---|---|
| CAS-No. | 1884577-99-4 |
| Formula | $C_{20}H_{31}N_3O_5$ |
| Mol. weight | 393,48 g/mol |

## ADC1660 Boc-Val-Ala-PAB-Cl

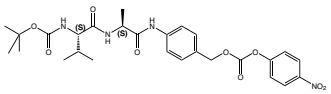
*tert*-butyloxycarbonyl-valyl-alanyl-4-aminobenzylchloride



| | |
|---|---|
| Formula | $C_{20}H_{30}ClN_3O_4$ |
| Mol. weight | 411,93 g/mol |

## ADC1050 Boc-Val-Ala-PAB-PNP

*t*-Butyloxycarbonyl-valyl-alanyl-(4-aminobenzyl)-(4-nitrophenyl)carbonate

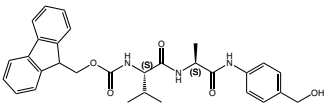


| | |
|---|---|
| CAS-No. | 1884578-00-0 |
| Formula | $C_{27}H_{34}N_4O_9$ |
| Mol. weight | 558,58 g/mol |

## ADC1060 Fmoc-Val-Ala-PAB

9-Fluorenylmethyloxycarbonyl-valyl-alanyl-4-aminobenzylalcohol

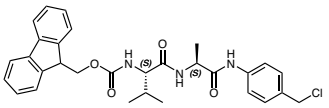
| | |
|---|---|
| CAS-No. | 1394238-91-5 |
| Formula | $C_{30}H_{33}N_3O_5$ |
| Mol. weight | 515,61 g/mol |



## ADC1670 Fmoc-Val-Ala-PAB-Cl

9-Fluorenylmethyloxycarbonyl-valyl-alanyl-4-aminobenzylchloride

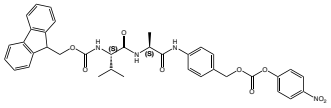
| | |
|---|---|
| CAS-No. | 1491136-17-4 |
| Formula | $C_{30}H_{32}ClN_3O_4$ |
| Mol. weight | 534,05 g/mol |



## ADC1070 Fmoc-Val-Ala-PAB-PNP

9-Fluorenylmethyloxycarbonyl-valyl-alanyl-(4-aminobenzyl)-(4-nitrophenyl)carbonate

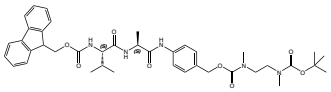
| | |
|---|---|
| CAS-No. | 1394238-92-6 |
| Formula | $C_{37}H_{36}N_4O_9$ |
| Mol. weight | 680,71 g/mol |



## ADC1410 Fmoc-Val-Ala-PAB-NMeCH$_2$CH$_2$NMe-Boc

9-Fluorenylmethyloxycarbonyl-valyl-alanyl-4-aminobenzyloxycarbonyl-((*t*-buytl methyl(2-methylamino)ethyl)carbamate)

| | |
|---|---|
| CAS-No. | 1691196-82-3 |
| Formula | $C_{40}H_{51}N_5O_8$ |
| Mol. weight | 729,86 g/mol |



***References:***


- *Multivalency Increases the Binding Strength of RGD Peptidomimetic-Paclitaxel Conjugates to Integrin alphaV beta3; A. Raposo Moreira Dias, A. Pina, A. Dal Corso, D. Arosio, L. Belvisi, L. Pignataro, M. Caruso and C. Gennari;* ***Chemistry*** *2017;* ***23****: 14410-14415.* https://doi.org/10.1002/chem.201703093
- *Synthesis and biological evaluation of RGD peptidomimetic-paclitaxel conjugates bearing lysosomally cleavable linkers; A. Dal Corso, M. Caruso, L. Belvisi, D. Arosio, U. Piarulli, C. Albanese, F. Gasparri, A. Marsiglio, F. Sola, S. Troiani, B. Valsasina, L. Pignataro, D. Donati, C. Gennari;* ***Chemistry*** *2015;* ***21****: 6921-9.* https://doi.org/10.1002/chem.201500158
- *Elongated multiple electronic cascade and cyclization spacer systems in activatible anticancer prodrugs for enhanced drug release; F. M. de Groot, W. J. Loos, R. Koekkoek, L. W. van Berkom, G. F. Busscher, A. E. Seelen, C. Albrecht, P. de Bruijn, H. W. Scheeren;* ***J Org Chem*** *2001;* ***66****: 8815-30.* https://doi.org/10.1021/jo0158884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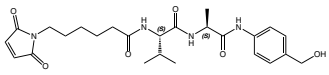
Product details

## ADC1270 MC-Val-Ala-PAB

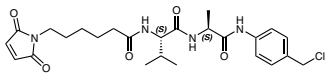
6-maleimidohexanoyl-valyl-alanyl-(4-aminobenzyl alcohol)



| | |
|---|---|
| CAS-No. | 1870916-87-2 |
| Formula | $C_{25}H_{34}N_4O_6$ |
| Mol. weight | 486,56 g/mol |

## ADC1700 MC-Val-Ala-PAB-Cl

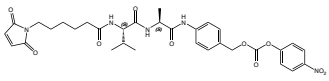
6-Maleimidohexanoyl-valyl-alanyl-(4-aminobenzyl chloride)



| | |
|---|---|
| CAS-No. | 2983182-03-0 |
| Formula | $C_{25}H_{33}ClN_4O_5$ |
| Mol. weight | 521,01 g/mol |

## ADC1280 MC-Val-Ala-PAB-PNP

6-maleimidohexanoyl-valyl-alanyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



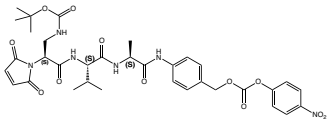
| | |
|---|---|
| CAS-No. | 1639939-40-4 |
| Formula | $C_{32}H_{37}N_5O_{10}$ |
| Mol. weight | 651,66 g/mol |

***References:***


- *Improved Methodology for the Synthesis of a Cathepsin B Cleavable Dipeptide Linker, Widely Used in Antibody-Drug Conjugate Research; D. Mondal, J. Ford, K. G. Pinney;* ***Tetrahedron Lett*** *2018;* ***59****: 3594-3599. https://doi.org/10.1016/j.tetlet.2018.08.021*
- *Next generation maleimides enable the controlled assembly of antibody-drug conjugates via native disulfide bond bridging; F. F. Schumacher, J. P. Nunes, A. Maruani, V. Chudasama, M. E. Smith, K. A. Chester, J. R. Baker, S. Caddick;* ***Org Biomol Chem*** *2014;* ***12****: 7261-9. https://doi.org/10.1039/c4ob01550a*
- *Site-Specific Conjugation of Auristatins onto Engineered scFv Using Second Generation Maleimide to Target HER2-positive Breast Cancer in Vitro; N. Aubrey, E. Allard-Vannier, C. Martin, F. Bryden, S. Letast, C. Colas, Z. Lakhrif, N. Collinet, I. Dimier-Poisson, I. Chourpa, M. C. Viaud-Massuard, N. Joubert;* ***Bioconjug Chem*** *2018;* ***29****: 3516-3521. https://doi.org/10.1021/acs.bioconjchem.8b00668*
- *Impact of cathepsin B-sensitive triggers and hydrophilic linkers on in vitro efficacy of novel site-specific antibody-drug conjugates; F. Bryden, C. Martin, S. Letast, E. Lles, I. Vieitez-Villemin, A. Rousseau, C. Colas, M. Brachet-Botineau, E. Allard-Vannier, C. Larbouret, M. C. Viaud-Massuard, N. Joubert;* ***Org Biomol Chem*** *2018;* ***16****: 1882-1889. https://doi.org/10.1039/c7ob02780j*

## ADC1080 Mal-Dap(Boc)-Val-Ala-PAB-PNP

N-alpha-Maleimido-N-beta-*t*-butyloxycarbonyl-L-2,3-diaminopropionyl-valyl-alanyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| Formula | $C_{34}H_{40}N_6O_{12}$ |
| Mol. weight | 721,71 g/mol |

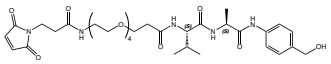
***References:***


- *Linker Technologies for Antibody–Drug Conjugates; B. Nolting;* ***Antibody-Drug Conjugates L. Ducry*** *2013;* ***1045****: 71-100. https://doi.org/10.1007/978-1-62703-541-5_5*
- *Self-hydrolyzing maleimides improve the stability and pharmacological properties of antibody-drug conjugates; R. P. Lyon, J. R. Setter, T. D. Bovee, S. O. Doronina, J. H. Hunter, M. E. Anderson,C. L. Balasubramanian, S. M. Duniho, C. I. Leiske, F. Li, P. D. Senter;* ***Nat Biotechnol*** *2014;* ***32****: 1059-62. https://doi.org/10.1038/nbt.2968*
- *Self-Stabilizing Linker Conjugate; Lyon R., Doronina S., Bovee T.; Seattle Genetics, Inc.; U.S. Patent No. 9,504,756, 2013*
- *In Vivo Antitumor Activity of a Novel Acetazolamide-Cryptophycin Conjugate for the Treatment of Renal Cell Carcinomas; S. Cazzamalli, E. Figueras, L. Petho, A. Borbely, C. Steinkuhler, D. Neri, N. Sewald;* ***ACS Omega*** *2018;* ***3****: 14726-14731. https://doi.org/10.1021/acsomega.8b02350*


Product details

## ADC1390 Mal-beta-Ala-PEG(4)-Val-Ala-PAB

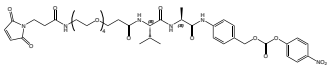
maleimido-beta-alanyl-tetraethyleneglycol-propanoyl-valyl-alanyl-(4-aminobenzyl alcohol)



| | |
|---|---|
| CAS-No. | 2417003-93-9 |
| Formula | $C_{33}H_{49}N_5O_{11}$ |
| Mol. weight | 691,77 g/mol |

## ADC1400 Mal-beta-Ala-PEG(4)-Val-Ala-PAB-PNP

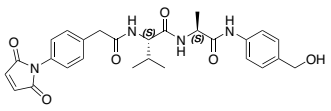
maleimido-beta-alanyl-tetraethyleneglycol-propanoyl-valyl-alanyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| CAS-No. | 2417003-94-0 |
| Formula | $C_{40}H_{52}N_6O_{15}$ |
| Mol. weight | 856,87 g/mol |

## ADC1730 Mal-PhAc-Val-Ala-PAB

(S)-2-(2-(4-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl) phenyl)acetamido)-N-((S)-1-((4-(hydroxymethyl)phenyl) amino)-1-oxopropan-2-yl)-3-methylbutanamide



| | |
|---|---|
| Formula | $C_{27}H_{30}N_4O_6$ |
| Mol. weight | 506,56 g/mol |

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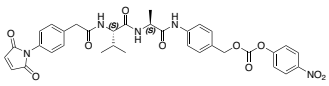
Product details

## ADC1740 Mal-PhAc-Val-Ala-PAB-PNP

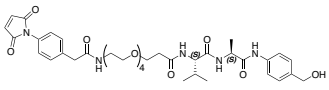
4-((S)-2-((S)-2-(2-(4-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)phenyl)acetamido)-3-methylbutanamido)propanamido)benzyl (4-nitrophenyl) carbonate



Formula $C_{34}H_{33}N_5O_{10}$
Mol. weight 671,66 g/mol

## ADC1770 Mal-PhAc-PEG(4)-Val-Ala-PAB

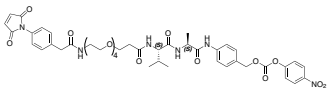
(S)-2-(3-(2-(2-(4-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)phenyl)acetamido)ethoxy)propanamido)-N-((S)-1-((4-(hydroxymethyl)phenyl)amino)-1-oxopropan-2-yl)-3-methylbutanamide



Formula $C_{32}H_{39}N_5O_8$
Mol. weight 621,69 g/mol

## ADC1780 Mal-PhAc-PEG(4)-Val-Ala-PAB-PNP

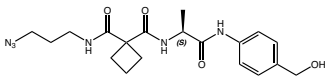
4-((2S,5S)-15-(4-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)phenyl)-5-isopropyl-2-methyl-4,7,14-trioxo-10-oxa-3,6,13-triazapentadecanamido)benzyl (4-nitrophenyl) carbonate



Formula $C_{39}H_{42}N_6O_{12}$
Mol. weight 786,80 g/mol

## ADC1580 Azido-cyclobutane-1,1-dicarboxamide-Ala-PAB

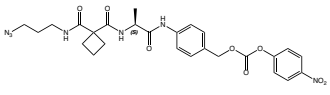
3-azidopropyl-cyclobutane-1,1-dicarboxamide-alanyl-(4-aminobenzyl alcohol)



CAS-No. 2576471-45-7
Formula $C_{19}H_{26}N_6O_4$
Mol. weight 402,45 g/mol

## ADC1590 Azido-cyclobutane-1,1-dicarboxamide-Ala-PAB-PNP

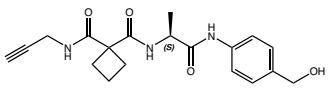
3-azidopropyl-cyclobutane-1,1-dicarboxamide-alanyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



CAS-No. 2576471-36-6
Formula $C_{26}H_{29}N_7O_8$
Mol. weight 567,55 g/mol

## ADC1600 Propargyl-cyclobutane-1,1-dicarboxamide-Ala-PAB

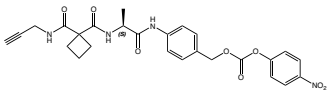
propargyl-cyclobutane-1,1-dicarboxamide-alanyl-(4-aminobenzyl alcohol)



CAS-No. 2576471-28-6
Formula $C_{19}H_{23}N_3O_4$
Mol. weight 357,40 g/mol

## ADC1610 Propargyl-cyclobutane-1,1-dicarboxamide-Ala-PAB-PNP

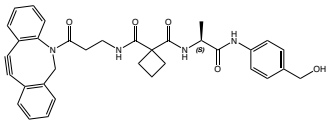
propargyl-cyclobutane-1,1-dicarboxamide-alanyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| CAS-No. | 2576471-42-4 |
| Formula | $C_{26}H_{26}N_4O_8$ |
| Mol. weight | 522,51 g/mol |

## ADC1620 DBCO-cyclobutane-1,1-dicarboxamide-Ala-PAB

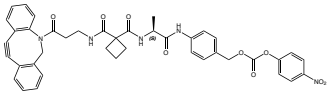
dibenzoazacyclooctyne-cyclobutane-1,1-dicarboxamide-alanyl-(4-aminobenzyl alcohol)



| | |
|---|---|
| CAS-No. | 2576471-46-8 |
| Formula | $C_{34}H_{34}N_4O_5$ |
| Mol. weight | 578,66 g/mol |

## ADC1630 DBCO-cyclobutane-1,1-dicarboxamide-Ala-PAB-PNP

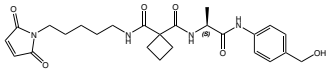
dibenzoazacyclooctyne-cyclobutane-1,1-dicarboxamide-alanyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| CAS-No. | 2576471-43-5 |
| Formula | $C_{41}H_{37}N_5O_9$ |
| Mol. weight | 743,76 g/mol |

## ADC1560 Mal-cyclobutane-1,1-dicarboxamide-Ala-PAB

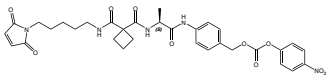
5-maleimidopentyl-cyclobutane-1,1-dicarboxamide-alanyl-(4-aminobenzyl alcohol)



| | |
|---|---|
| CAS-No. | 2576471-41-3 |
| Formula | $C_{25}H_{32}N_4O_6$ |
| Mol. weight | 484,54 g/mol |

## ADC1570 Mal-cyclobutane-1,1-dicarboxamide-Ala-PAB-PNP

5-maleimidopentyl-cyclobutane-1,1-dicarboxamide-alanyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| CAS-No. | 2576471-35-5 |
| Formula | $C_{32}H_{35}N_5O_{10}$ |
| Mol. weight | 649,65 g/mol |

***Reference:***

→ *Discovery of Peptidomimetic Antibody-Drug Conjugate Linkers with Enhanced Protease Specificity; B. Wei, J. Gunzner-Toste, H. Yao, T. Wang, J. Wang, Z. Xu, J. Chen, J. Wai, J. Nonomiya, S. P. Tsai, J. Chuh, K. R. Kozak, Y. Liu, S. F. Yu, J. Lau, G. Li, G. D. Phillips, D. Leipold, A. Kamath, D. Su, K. Xu, C. Eigenbrot, S. Steinbacher, R. Ohri, H. Raab, L. R. Staben, G. Zhao, J. A. Flygare, T. H. Pillow, V. Verma, L. A. Masterson, P. W. Howard, B. Safina;* ***J. Med. Chem.*** *2018;* ***61****: 989-1000.* https://doi.org/10.1021/acs.jmedchem.7b01430

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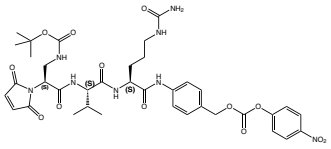
## 3.2. Valine-Citrulline-Based Enzymatically Cleavable Linkers

Product details

### ADC1090 Mal-Dap(Boc)-Val-Cit-PAB-PNP

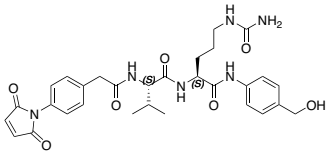
N-alpha-Maleimido-N-beta-*t*-butyloxycarbonyl-L-2,3-diaminopropionyl-valyl-citrullyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| CAS-No. | 2281797-57-5 |
| Formula | $C_{37}H_{46}N_8O_{13}$ |
| Mol. weight | 810,81 g/mol |

### ADC1750 Mal-PhAc-Val-Cit-PAB

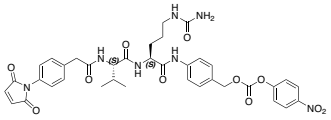
(S)-2-((S)-2-(2-(4-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)phenyl)acetamido)-3-methylbutanamido)-N-(4-(hydroxymethyl)phenyl)-5-ureidopentanamide



| | |
|---|---|
| Formula | $C_{30}H_{36}N_6O_7$ |
| Mol. weight | 592,65 g/mol |

### ADC1760 Mal-PhAc-Val-Cit-PAB-PNP

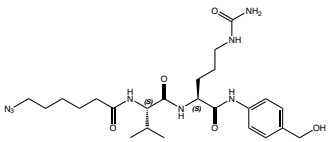
4-((S)-2-((S)-2-(2-(4-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)phenyl)acetamido)-3-methylbutanamido)-5-ureidopentanamido)benzyl (4-nitrophenyl) carbonate



| | |
|---|---|
| CAS-No. | 3034752-26-3 |
| Formula | $C_{37}H_{39}N_7O_{11}$ |
| Mol. weight | 757,76 g/mol |

### ADC1120 6-Azidohexanoyl-Val-Cit-PAB

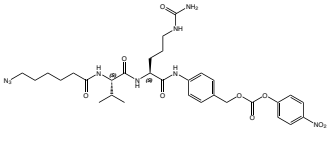
6-azidohexanoyl-valyl-citrullyl-(4-aminobenzyl alcohol)



| | |
|---|---|
| CAS-No. | 1613321-02-0 |
| Formula | $C_{24}H_{38}N_8O_5$ |
| Mol. weight | 518,61 g/mol |

### ADC1130 6-Azidohexanoyl-Val-Cit-PAB-PNP

6-azidohexanoyl-valyl-citrullyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| CAS-No. | 1613321-01-9 |
| Formula | $C_{31}H_{41}N_9O_9$ |
| Mol. weight | 683,71 g/mol |

***Reference:***

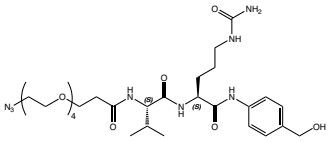
→ *NKT cell-dependent glycolipid-peptide vaccines with potent anti-tumour activity; R. J. Anderson, B. J. Compton, C. W. Tang, A. Authier-Hall, C. M. Hayman, G. W. Swinerd, R. Kowalczyk, P. Harris, M. A. Brimble, D. S. Larsen, O. Gasser, R. Weinkove, I. F. Hermans, G. F. Painter;* ***Chem. Sci.*** *2015;* ***6****: 5120-5127.* https://doi.org/10.1039/c4sc03599b

Product details

## ADC1160 Azido-PEG(4)-Val-Cit-PAB

azido-tetraethyleneglycol-propanoyl-valyl-citrullyl-(4-aminobenzyl alcohol)

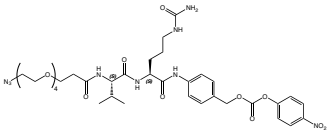
| | |
|---|---|
| CAS-No. | 2055024-64-9 |
| Formula | $C_{29}H_{48}N_8O_9$ |
| Mol. weight | 652,74 g/mol |



## ADC1170 Azido-PEG(4)-Val-Cit-PAB-PNP

azido-tetraethyleneglycol-propanoyl-valyl-citrullyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate

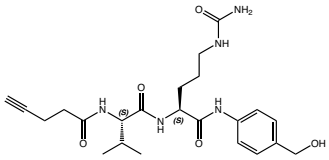
| | |
|---|---|
| CAS-No. | 1869126-60-2 |
| Formula | $C_{36}H_{51}N_9O_{13}$ |
| Mol. weight | 817,84 g/mol |



## ADC1140 4-Pentynoyl-Val-Cit-PAB

4-pentynoyl-valyl-citrullyl-(4-aminobenzyl alcohol)

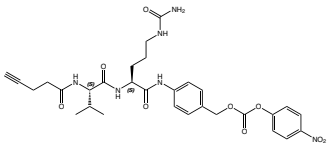
| | |
|---|---|
| CAS-No. | 2708150-97-2 |
| Formula | $C_{23}H_{33}N_5O_5$ |
| Mol. weight | 459,54 g/mol |



## ADC1150 4-Pentynoyl-Val-Cit-PAB-PNP

4-pentynoyl-valyl-citrullyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate

| | |
|---|---|
| Formula | $C_{30}H_{36}N_6O_9$ |
| Mol. weight | 624,64 g/mol |



***Reference:***

→ *Integrin-Targeting Knottin Peptide–Drug Conjugates Are Potent Inhibitors of Tumor Cell Proliferation. N. Cox, J. R. Kintzing, M. Smith, G. A. Grant, J. R. Cochran;* ***Angew. Chem. Int. Ed.*** *2016;* ***55(34)****: 9894-9897.* https://doi.org/10.1002/anie.201603488

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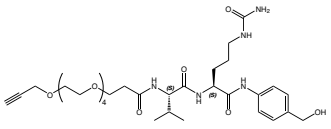
Product details

## ADC1180 Alkyne-PEG(5)-Val-Cit-PAB

propargyl-tetraethyleneglycol-propanoyl-valyl-citrullyl-(4-aminobenzyl alcohol)

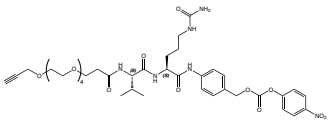
Formula $C_{32}H_{51}N_5O_{10}$
Mol. weight 665,77 g/mol



## ADC1190 Alkyne-PEG(5)-Val-Cit-PAB-PNP

propargyl-tetraethyleneglycol-propanoyl-valyl-citrullyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate

Formula $C_{39}H_{54}N_6O_{14}$
Mol. weight 830,88 g/mol



***References:***


→ *Exploration of the carmaphycins as payloads in antibody drug conjugate anticancer agents. J. Almaliti, B. Miller, H. Pietraszkiewicz, E. Glukhov, C. B. Naman, T. Kline, J. Hanson, X. Li, S. Zhou, F. A. Valeriote, W. H. Gerwick;* ***Eur J Med Chem.*** *2019;* ***161****: 416-432.* https://doi.org/10.1016/j.ejmech.2018.10.024

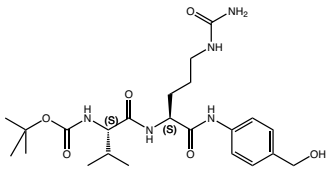
→ *Design and synthesis of novel dual-cyclic RGD peptides for avβ3 integrin targeting. J. Liu, X. Cheng, X. Tian, D. Guan, J. Ao, Z. Wu, W. Huang, Z. Le;* ***Bioorg Med Chem Lett.*** *2019;* ***29(7)****: 896-900.* https://doi.org/10.1016/j.bmcl.2019.01.043


Product details

## ADC1020 Boc-Val-Cit-PAB

*t*-Butyloxycarbonyl-valyl-citrullyl-4-aminobenzylalcohol

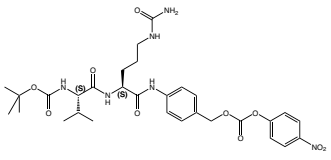
CAS-No. 870487-09-5
Formula $C_{23}H_{37}N_5O_6$
Mol. weight 479,59 g/mol



## ADC1010 Boc-Val-Cit-PAB-PNP

*t*-Butyloxycarbonyl-valyl-citrullyl-(4-aminobenzyl)-(4-nitrophenyl)carbonate

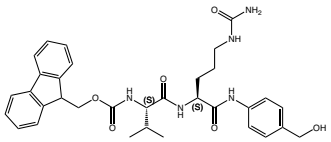
CAS-No. 870487-10-8
Formula $C_{30}H_{40}N_6O_{10}$
Mol. weight 644,67 g/mol



## ADC1030 Fmoc-Val-Cit-PAB

9-Fluorenylmethyloxycarbonyl-valyl-citrullyl-4-aminobenzylalcohol

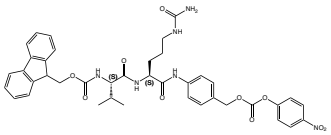
| | |
|---|---|
| CAS-No. | 159858-22-7 |
| Formula | $C_{33}H_{39}N_5O_6$ |
| Mol. weight | 601,29 g/mol |



## ADC1000 Fmoc-Val-Cit-PAB-PNP

9-Fluorenylmethyloxycarbonyl-valyl-citrullyl-(4-aminobenzyl)-(4-nitrophenyl)carbonate

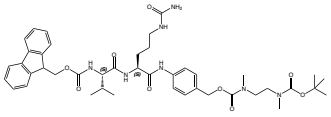
| | |
|---|---|
| CAS-No. | 863971-53-3 |
| Formula | $C_{40}H_{42}N_6O_{10}$ |
| Mol. weight | 766,80 g/mol |



## ADC1240 Fmoc-Val-Cit-PAB-NMeCH$_2$CH$_2$NMe-Boc

9-Fluorenylmethyloxycarbonyl-valyl-citrullyl-4-aminobenzyloxycarbonyl-((*t*-buytl methyl(2-methylamino)ethyl)carbamate)

| | |
|---|---|
| CAS-No. | 1802297-96-6 |
| Formula | $C_{43}H_{57}N_7O_9$ |
| Mol. weight | 815,95 g/mol |



***References:***


→ *Multivalency Increases the Binding Strength of RGD Peptidomimetic-Paclitaxel Conjugates to Integrin αVβ3. A. R. M. Dias, A. Pina, A. Dal Corso, D. Arosio, L. Belvisi, L. Pignataro, M. Caruso, C. Gennari;* ***Chem. Eur. J.*** *2017;* ***23(58)****: 14410-14415.* https://doi.org/10.1002/chem.201703093

→ *Synthesis and Biological Evaluation of RGD Peptidomimetic–Paclitaxel Conjugates Bearing Lysosomally Cleavable Linkers. A. D. Corso, M. Caruso, L. Belvisi, D. Arosio, U. Piarulli, C. Albanese, F. Gasparri, A. Marsiglio, F. Sola, S. Troiani, B. Valsasina, L. Pignataro, D. Donati, C. Gennari;* ***Chem. Eur. J.*** *2015;* ***21(18)****: 6921-6929.* https://doi.org/10.1002/chem.201500158

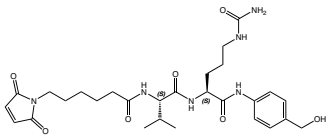
→ *Elongated Multiple Electronic Cascade and Cyclization Spacer Systems in Activatible Anticancer Prodrugs for Enhanced Drug Release. F. M. H. de Groot, W. J. Loos, R. Koekkoek, L. W. A. van Berkom, G. F. Busscher, A. E. Seelen, C. Albrecht, P. Bruijn, H. W. Scheeren;* ***J. Org. Chem.*** *2001;* ***66(26)****: 8815-8830.* https://doi.org/10.1021/jo0158884




## ADC1100 MC-Val-Cit-PAB

6-maleimidohexanoyl-valyl-citrullyl-(4-aminobenzyl alcohol)

| | |
|---|---|
| CAS-No. | 159857-80-4 |
| Formula | $C_{28}H_{40}N_6O_7$ |
| Mol. weight | 572,65 g/mol |



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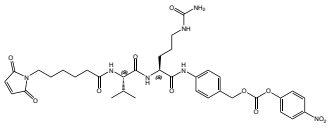
Product details

## ADC1110 MC-Val-Cit-PAB-PNP

6-maleimidohexanoyl-valyl-citrullyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| CAS-No. | 159857-81-5 |
| Formula | $C_{35}H_{43}N_7O_{11}$ |
| Mol. weight | 737,76 g/mol |

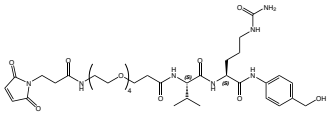
***Reference:***

→ *Improved Methodology for the Synthesis of a Cathepsin B Cleavable Dipeptide Linker, Widely Used in Antibody-Drug Conjugate Research. D. Mondal, J. Ford, K. G. Pinney;* ***Tetrahedron Lett.*** *2018;* ***59(40)****: 3594-3599. https://doi.org/10.1016/j.tetlet.2018.08.021*

Product details

## ADC1220 Mal-beta-Ala-PEG(4)-Val-Cit-PAB

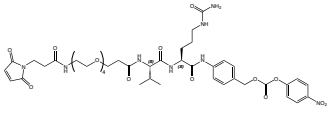
maleimido-beta-alanyl-tetraethyleneglycol-propanoyl-valyl-citrullyl-(4-aminobenzyl alcohol)



| | |
|---|---|
| CAS-No. | 1949793-41-2 |
| Formula | $C_{36}H_{55}N_7O_{12}$ |
| Mol. weight | 777,86 g/mol |

## ADC1230 Mal-beta-Ala-PEG(4)-Val-Cit-PAB-PNP

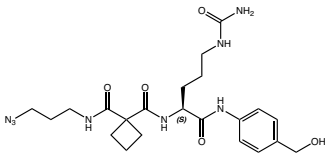
maleimido-beta-alanyl-tetraethyleneglycol-propanoyl-valyl-citrullyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| CAS-No. | 2003260-12-4 |
| Formula | $C_{43}H_{58}N_8O_{16}$ |
| Mol. weight | 942,96 g/mol |

## ADC1480 Azido-cyclobutane-1,1-dicarboxamide-Cit-PAB

3-azidopropyl-cyclobutane-1,1-dicarboxamide-citrullyl-(4-aminobenzyl alcohol)

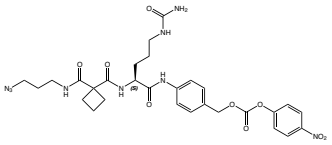


| | |
|---|---|
| CAS-No. | 2576471-33-3 |
| Formula | $C_{22}H_{32}N_8O_5$ |
| Mol. weight | 488,54 g/mol |

## ADC1490 Azido-cyclobutane-1,1-dicarboxamide-Cit-PAB-PNP

3-azidopropyl-cyclobutane-1,1-dicarboxamide-citrul-lyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate

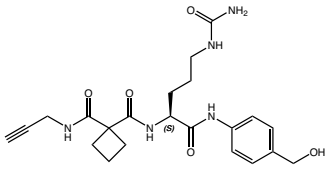
| | |
|---|---|
| CAS-No. | 2576471-44-6 |
| Formula | $C_{29}H_{35}N_9O_9$ |
| Mol. weight | 653,64 g/mol |



## ADC1500 Propargyl-cyclobutane-1,1-dicarboxamide-Cit-PAB

propargyl-cyclobutane-1,1-dicarboxamide-citrul-lyl-(4-aminobenzyl alcohol)

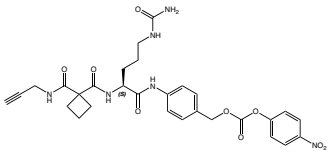
| | |
|---|---|
| CAS-No. | 2576471-27-5 |
| Formula | $C_{22}H_{29}N_5O_5$ |
| Mol. weight | 443,50 g/mol |



## ADC1510 Propargyl-cyclobutane-1,1-dicarboxamide-Cit-PAB-PNP

propargyl-cyclobutane-1,1-dicarboxamide-citrul-lyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate

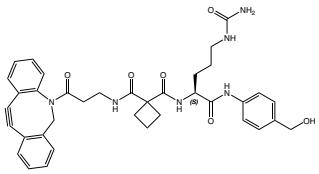
| | |
|---|---|
| CAS-No. | 2576471-40-2 |
| Formula | $C_{29}H_{32}N_6O_9$ |
| Mol. weight | 608,60 g/mol |



## ADC1520 DBCO-cyclobutane-1,1-dicarboxamide-Cit-PAB

dibenzoazacyclooctyne-cyclobutane-1,1-dicarboxami-de-citrullyl-(4-aminobenzyl alcohol)

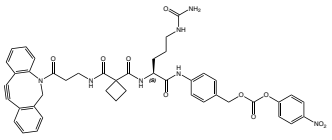
| | |
|---|---|
| CAS-No. | 2576471-51-5 |
| Formula | $C_{37}H_{40}N_6O_6$ |
| Mol. weight | 664,75 g/mol |



## ADC1530 DBCO-cyclobutane-1,1-dicarboxamide-Cit-PAB-PNP

dibenzoazacyclooctyne-cyclobutane-1,1-dicarboxami-de-citrullyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate

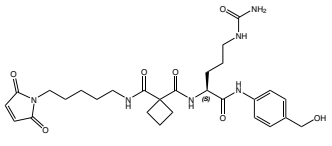
| | |
|---|---|
| CAS-No. | 2576471-34-4 |
| Formula | $C_{44}H_{43}N_7O_{10}$ |
| Mol. weight | 829,85 g/mol |



## ADC1460 Mal-cyclobutane-1,1-dicarboxamide-Cit-PAB

5-maleimidopentyl-cyclobutane-1,1-dicarboxamide-ci-trullyl-(4-aminobenzyl alcohol)

| | |
|---|---|
| CAS-No. | 1799663-03-8 |
| Formula | $C_{28}H_{38}N_6O_7$ |
| Mol. weight | 570,64 g/mol |



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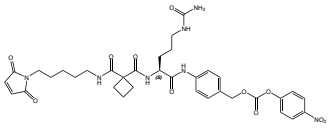
Product details

ADC1470 Mal-cyclobutane-1,1-dicarboxamide-Cit-PAB-PNP

5-maleimidopentyl-cyclobutane-1,1-dicarboxamide-citrullyl-(4-aminobenzyl)-(4-nitrophenyl)-carbonate



| | |
|---|---|
| CAS-No. | 2204228-34-0 |
| Formula | $C_{35}H_{41}N_7O_{11}$ |
| Mol. weight | 735,74 g/mol |

***Reference:***

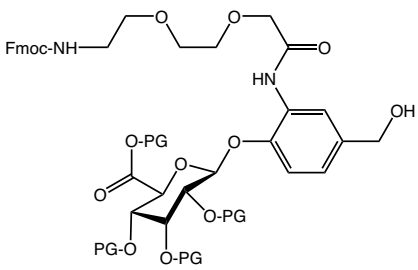
→ *Discovery of Peptidomimetic Antibody–Drug Conjugate Linkers with Enhanced Protease Specificity. B. Wei, J. Gunzner-Toste, H. Yao, T. Wang, J. Wang, Z. Xu, J. Chen, J. Wai, J. Nonomiya, S. Ping Tsai, J. Chuh, K. R. Kozak, Y. Liu, S. Yu, J. Lau, G. Li, G. D. Phillips, D. Leipold, A. Kamath, D. Su, K. Xu, C. Eigenbrot, S. Steinbacher, R. Ohri, H. Raab, L. R. Staben, G. Zhao, J. A. Flygare, T. H. Pillow, V. Verma, L. A. Masterson, P. W. Howard, B. Safina;* ***J. Med. Chem.*** *2018;* ***61(3)****: 989-1000.* https://doi.org/10.1021/acs.jmedchem.7b01430

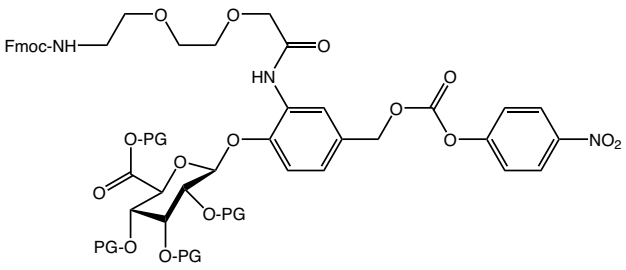
## 3.3. β-Glucuronide Enzymatically Cleavable Linkers

As an extension of the linkerology® toolbox, the design of linkers with improved stability during systemic circulation is highly desired. As the drug-releasing lysosomal enzyme β-glucuronidase is abundantly present within lysosomes and overexpressed in some tumor types but low outside cells, β-glucuronic acid-based linkers provide the potential for high ADC stability in the systemic circulation and selective intracellular drug release. Especially for ADCs based on highly hydrophobic drugs, the incorporation of the highly hydrophilic β-glucuronides may circumvent the tendency of aggregation. For example, a drug-linker consisting of a β-glucuronide linked to auristatin MMAF was prepared. Rat plasma stability analysis revealed an extrapolated half-life of 81 days, compared with about six days for the corresponding valine-citrulline dipeptide-linked MMAF.

### Interested in β-Glucuronide Enzymatically Cleavable Linkers?

**Please contact our Custom Synthesis for more details!**



***References:***


- *Expanded Utility of the β-Glucuronide Linker: ADCs That Deliver Phenolic Cytotoxic Agents; S. C. Jeffrey, J. De Brabander, J. Miyamoto, P. D. Senter;* ***ACS Med. Chem. Lett.*** *2010;* ***1****: 277-280.* https://doi.org/10.1021/ml100039h
- *Development and Properties of β-Glucuronide Linkers for Monoclonal Antibody-Drug Conjugates; S. C. Jeffrey, J. B. Andreyka, S. X. Bernhardt, K. M. Kissler, T. Kline, J. S. Lenox, R. F. Moser, M. T. Nguyen, N. M. Okeley, I. J. Stone, X. Zhang, P. D. Senter;* ***Bioconjugate Chem.*** *2006;* ***17****: 831-840.* https://doi.org/10.1021/bc0600214
- *Linker Technologies for Antibody–Drug Conjugates; B. Nolting;* ***Antibody-Drug Conjugates L. Ducry*** *2013;* ***1045****: 71-100.* https://doi.org/10.1007/978-1-62703-541-5_5


## 3.4. PH-Responsive Linkers

Various linker types for conjugation are available on the market, responding to and being – ideally tracelessly – cleaved by virtue of a certain trigger, e.g., peptidic linkers are cleaved in the presence of specific enzymes.

Thus, conditions solely present in the affected tissue need to be selected to avoid premature drug release. Compared to healthy cells, abnormal metabolism and proliferation in tumor cells lead to a lowered intracellular pH of around 5.0-6.0, while a lower pH (4.0-5.0) is reported for the lysosomes.

At Iris, we offer a pH-sensitive linker based on an alkyne-functionalized, *para*-nitrophenyl (PNP)-modified 5-(hydroxymethyl)-pyrogallol-orthoester (HMPO).

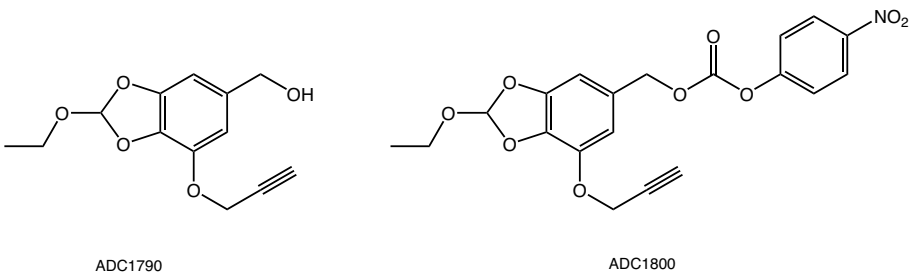



*Fig. 19: Chemical structures of the pH-sensitive self-immolative alkyne-HMPO-PNP linker (ADC1800 on page 74) and its precursor alkyne-HMPO-OH (ADC1790 on page 74).*

This construct is stable during circulation in plasma (tested for 24 hours at pH values of 7.4 and 6.6.) while being cleaved tracelessly *via* 1,6-elimination at pH 5.5. Payloads can be easily coupled: The HMPO-PNP linker ADC1800 reacts with base-activated amides while releasing the nitrophenyl residue.

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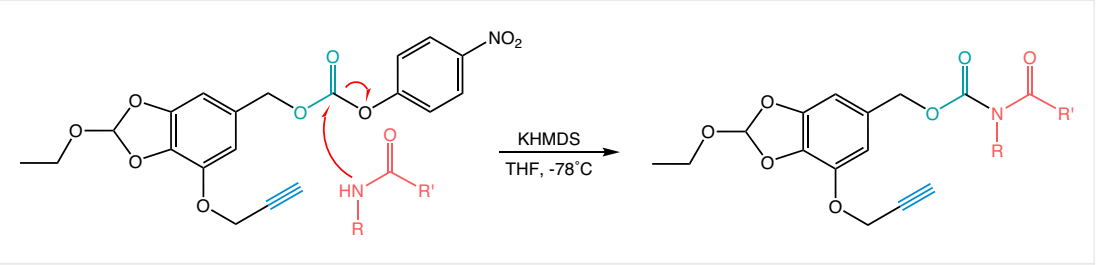



*Fig. 20: Coupling of payload and linker. Reaction conditions: The drug (depicted in red) is dissolved in THF at -78°C. The strong base potassium hexamethyldisilyzide (KHMDS) and then ADC1800 on page 74 are added. For the workup, the temperature is allowed to rise to -5 °C over 4 hours. Ammonium chloride is added to quench excess linker, then the product is extracted and purified by chromatography.*

While the payload is attached as carbamate, the connection to the carrier protein (usually a tumor-specific antibody) is realized *via* the propargyl residue through biorthogonal CuAAC click reaction, forming a 1,2,3-triazole. For this, the antibody must carry azido groups, which may be introduced *via* recombinant biosynthesis, utilizing pyrrolysyl-tRNA synthetase and a suitably modified lysine (e.g., *https://www.iris-biotech.de/HAA2080),* or with an amino-reactive azido linker like, e.g., *PEG1400 on page 33* or *RL-2980 on page 37*.

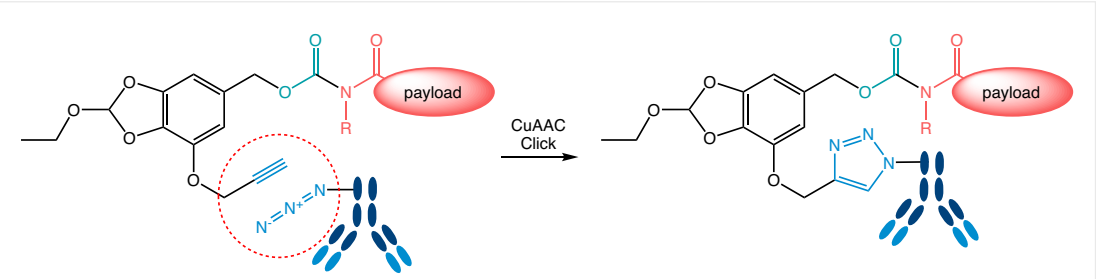



*Fig. 21: Combination of antibody and drug-linker adduct to generate the antibody-drug-conjugate (ADC).*

At acidic pH, the linker gets protonated inducing a 1,6-elimination reaction. The oxocarbonyl moiety is released as carbon dioxide, leading to the irreversible and traceless linker cleavage and payload release.

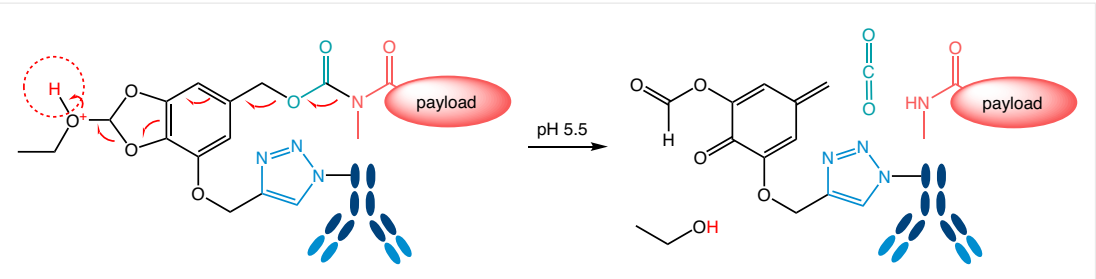


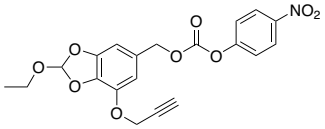

*Fig. 22: Payload release at acidic pH in the tumor microenvironment.*

While *ADC1800 on page 74* as the activated form of the linker reacts directly with the amide-containing drug molecule, we also offer the precursor *ADC1790 on page 74* (Alkyne-HMPO-OH) which can be further customized with your linker of choice.

## ADC1800 Alkyne-HMPO-PNP

(2-ethoxy-7-(prop-2-yn-1-yloxy)benzo[d][1,3]dioxol-5-yl) methyl (4-nitrophenyl) carbonate

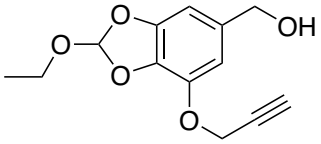
| | |
|---|---|
| CAS-No. | 3028213-53-5 |
| Formula | $C_{20}H_{17}NO_9$ |
| Mol. weight | 415,35 g/mol |



## ADC1790 Alkyne-HMPO-OH

(2-ethoxy-7-(prop-2-yn-1-yloxy)benzo[d][1,3]dioxol-5-yl) methanol

| | |
|---|---|
| CAS-No. | 3028213-63-7 |
| Formula | $C_{13}H_{14}O_5$ |
| Mol. weight | 250,25 g/mol |



***References:***


- *A pH-responsive crosslinker platform for antibody-drug conjugate (ADC) targeting delivery; F. Migliorini, E. Cini, E. Dreassi, F. Finetti, G. Ievoli, G. Macri, E. Petricci, E. Rango, L. Trabalzini, M. Taddei;* ***Chem. Commun.*** *2022;* ***58(75)****: 10532-10535.* https://doi.org/10.1039/D2CC03052G
- *A Self-Immolative Linker for the pH-Responsive Release of Amides; A. Petrini, G. Ievoli, F. Migliorini, M. Taddei, S. Siciliano;* ***Molecules*** *2023;* ***28(6)****: 2445.* https://doi.org/10.3390/molecules28062445
- *Conjugates of PSMA-binding moieties with cytotoxic agents; WO2024028258*
- *Compositions and methods for treatment of sexual dysfunction and related diseases, disorders, and conditions; WO2022251699*


## 3.5. Disulfide-Based (Self-Immolative) Linkers

Another chemically labile linkage extensively exploited in the development of antibody-drug conjugates are disulfides. They are stable at physiological pH and are designed to release the drug upon internalization inside cells. The cytosol provides a significantly more reducing environment compared to the extracellular milieu and the presence of cytoplasmic thiol cofactor, such as reduced glutathione (GSH). Additionally, the intracellular enzyme protein disulfide isomerase, or similar enzymes capable of cleaving disulfide bonds, may also contribute to the preferential cleavage of disulfide bonds inside cells. GSH is reported to be present in cells in the concentration range of 0.5-10 mM, compared with a significantly lower concentration of GSH or cysteine in plasma at approximately 5 µM. This is especially true for tumor cells, where irregular blood flow leads to a hypoxic state, resulting in enhanced activity of reductive enzymes and therefore in even higher glutathione concentrations.

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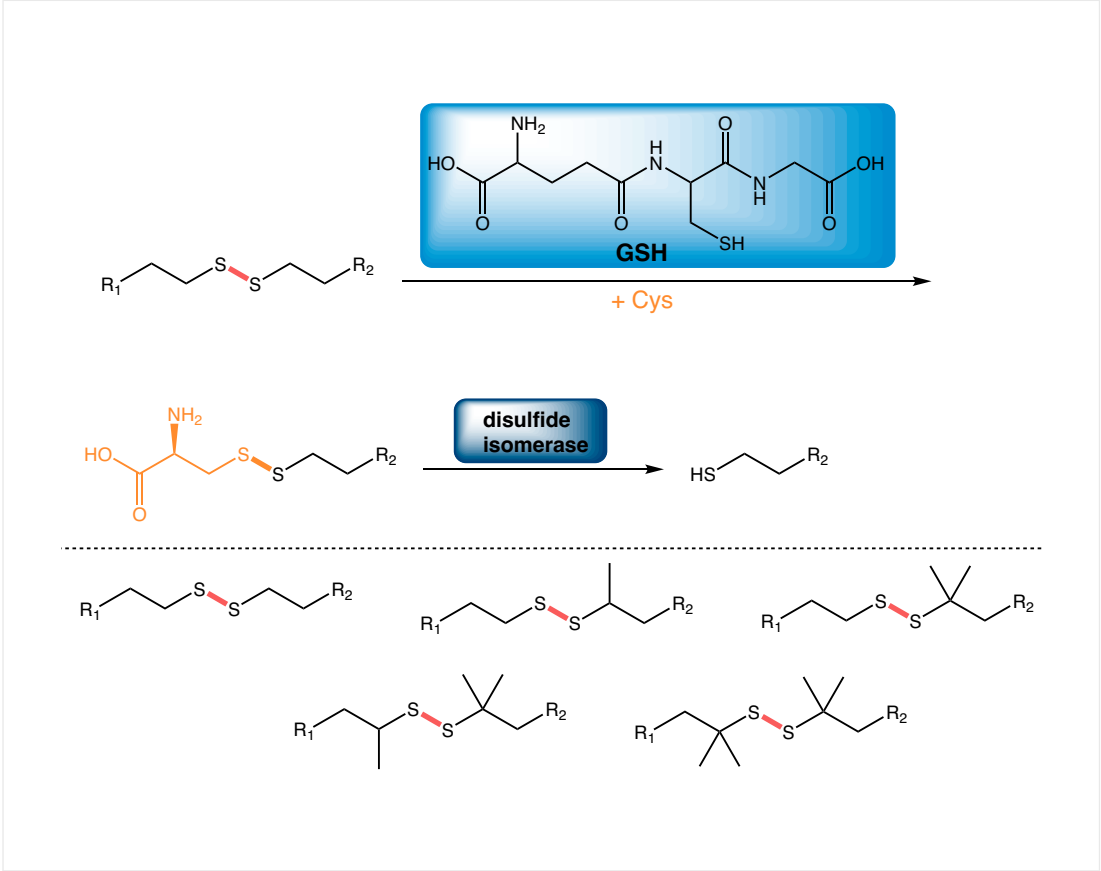



*Fig. 23: The stability of disulfide linkers can be finetuned by neighboring methylation.*

The stability of disulfide bridges can be fine-tuned by adjacent residues (*Fig. 23*). Methyl groups are bulky enough to have a significant influence on the thermodynamic stability of the disulfide bridge. While one additional methyl group already enhances the stability drastically, two methyl groups make the disulfide bond practically stable towards reductive cleavage. A methylation number of three or four will comple tely lock the disulfide bridge towards further modifications. As the direct conjugation of cleavable triggers to bioactive agents through disulfide bridges suffers from ineffective cleavage in case of bulky moieties and resulting steric hindrance as well as restricted possibilities for trigger-drug combinations, disulfide based self-immolative linkers (DSILs) provide a robust strategy for selective activation upon disulfide cleavage in the reductive cytoplasmic milieu.

Disulfide-based self-immolative linkers benefit of the reversibility of disulfide-bond formation. Upon oxidation, free thiols form less nucleophilic disulfide bonds, preventing self-immolative fragmentation. However, this process can be reversed in the presence of reducing agents, such as GSH. Those specifi-cations allow for sufficient stability in the extracellular milieu but spontaneous self-immolative reaction within the cytosol upon GSH-mediated disulfide cleavage. Variations in the linker's chemical composition (disulfide ethoxycarbonyl (SSE) vs. disulfide benzyloxycarbonyl (SSB)) result in chemically tunable kinetics of the self-immolative cleavage due to different response rates towards GSH, showing higher rates for SSB-based DSILs compared to SSE-based ones (*Fig. 24*). Thus, the choice of the linker allows for fine-tun-ing of the cleavage speed and payload release.

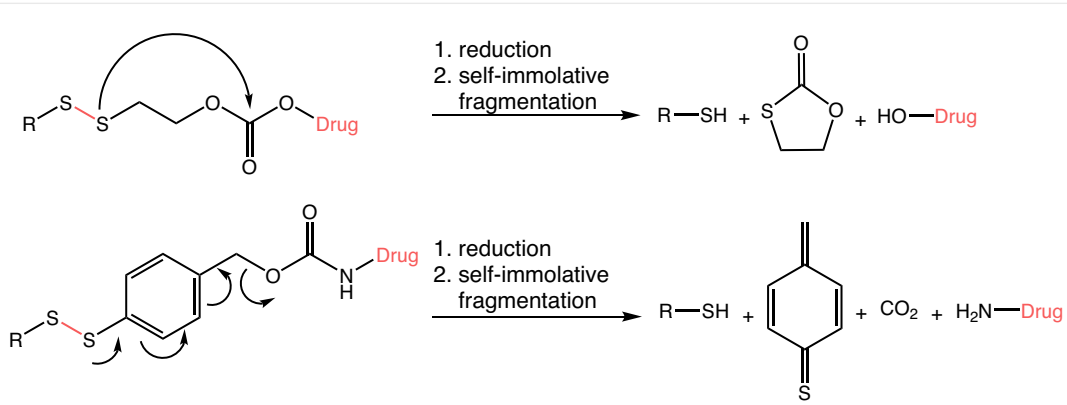



*Fig. 24: Disulfide-based self-immolative linkers either based on an intramolecular cyclization cascade mechanism or on a 1,6-elimination.*

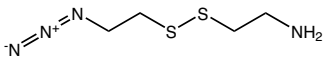
For ease of synthesis, Iris Biotech offers pyridyl disulfides as building blocks for the preparation of disulfide-based self-immolative linkers. Pyridyl disulfides undergo a disulfide exchange reaction with sulfhydryl groups to form disulfide bonds over a broad pH range also suitable for physiological pH. During the reaction, a disulfide exchange occurs between the biomolecule's thiol group and the reagent's 2-pyridyldithiol group. As a result, pyridine-2-thione is released, which can be followed spectrophotometrically ($\lambda_{max}$ = 343 nm) to monitor the progress of the reaction.

Product details

## HNN1090 $N_3$-Cystamine*HCl

**Azido-cystamine hydrochloride**

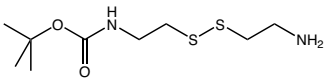
| | |
|---|---|
| CAS-No. | 1807512-40-8 net |
| Formula | $C_4H_{10}N_4S_2$*HCl |
| Mol. weight | 178,28*36,45 g/mol |



## BNN1170 Boc-Cystamine

**2-(*t*-Butyloxycarbonylamino)ethyldithio-2'-ethylamine**

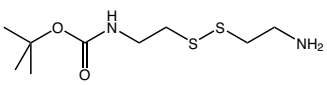
| | |
|---|---|
| CAS-No. | 485800-26-8 |
| Formula | $C_9H_{20}N_2O_2S_2$ |
| Mol. weight | 252,40 g/mol |



## BNN1063 Boc-Cystamine*HCl

**2-(*t*-Butyloxycarbonylamino)ethyldithio-2'-ethylamine hydrochloride**

| | |
|---|---|
| CAS-No. | 93790-49-9 |
| Formula | $C_9H_{20}N_2O_2S_2$*HCl |
| Mol. weight | 252,40*36,45 g/mol |



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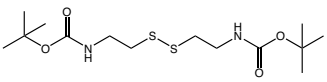
Product details

## BNN1360 Di-Boc-Cystamine

*N,N'*-Bis-*tert*-butoxycarbonyl-cystamine

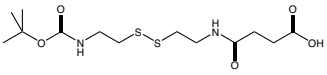
CAS-No. 67385-10-8
Formula $C_{14}H_{28}N_2O_4S_2$
Mol. weight 352,51 g/mol



## BAA2180 Boc-Cystamine-Suc-OH

4-(2-((2-*t*-Butyloxycarbonylaminoethyl)disulfanyl) ethylamino)-4-oxobutanoic acid

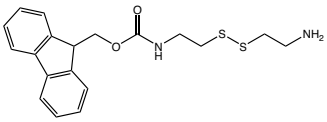
CAS-No. 946849-79-2
Formula $C_{13}H_{24}N_2O_5S_2$
Mol. weight 352,47 g/mol



## RL-3370 Fmoc-Cystamine*HCl

2-((9-Fluorenylmethyloxycarbonylamino)ethyl)disulfanyl-(2-aminoethane) hydrochloride

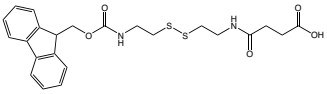
CAS-No. 2893917-85-4
Formula $C_{19}H_{22}N_2O_2S_2$*HCl
Mol. weight 374,52*36,45 g/mol



## RL-3310 Fmoc-Cystamine-Suc

4-(2-((2-(9-Fluorenylmethyloxycarbonylamino)ethyl) disulfanyl)ethylamino)-4-oxobutanoic acid

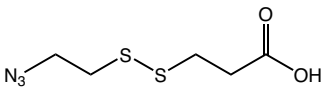
CAS-No. 946849-80-5
Formula $C_{23}H_{26}N_2O_5S_2$
Mol. weight 474,59 g/mol



## RL-4100 Azido-SS-COOH

3-((2-azidoethyl)disulfanyl)propanoic acid

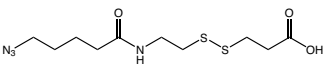
CAS-No. 2228857-32-5
Formula $C_5H_9N_3O_2S_2$
Mol. weight 207,27 g/mol



## RL-3320 Azido-Pen-SS-COOH

3-((2-(5-azidopentanamido)ethyl)disulfanyl)propanoic acid

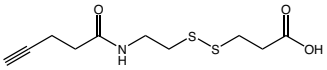
CAS-No. 2576471-47-9
Formula $C_{10}H_{18}N_4O_3S_2$
Mol. weight 306,40 g/mol



## RL-3330 Alkyne-SS-COOH

3-((2-pent-4-ynamidoethyl)disulfanyl)propanoic acid

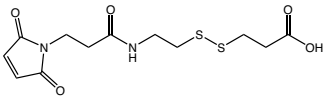
CAS-No. 2279938-29-1
Formula $C_{10}H_{15}NO_3S_2$
Mol. weight 261,36 g/mol



## RL-4090 Mal-SS-COOH

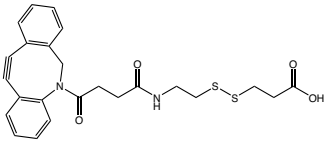
3-((2-(3-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)propanamido)ethyl)disulfanyl)propanoic acid

CAS-No. 2128735-24-8
Formula $C_{12}H_{16}N_2O_5S_2$
Mol. weight 332,39 g/mol



## RL-4110 DBCO-Suc-SS-COOH

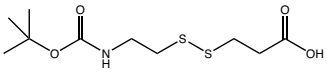
CAS-No. 2749426-25-1
Formula $C_{24}H_{24}N_2O_4S_2$
Mol. weight 468,59 g/mol



## RL-2190 Boc-SS-COOH

3-((2-(*tert*-butoxycarbonylamino)ethyl)disulfanyl) propanoic acid

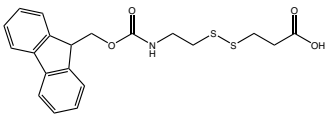
CAS-No. 485800-27-9
Formula $C_{10}H_{19}NO_4S_2$
Mol. weight 281,39 g/mol



## RL-2200 Fmoc-SS-COOH

3-((2-(((9H-fluoren-9-yl)methoxy)carbonylamino)ethyl) disulfanyl)propanoic acid

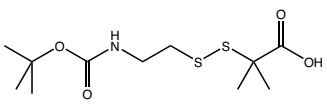
CAS-No. 864235-83-6
Formula $C_{20}H_{21}NO_4S_2$
Mol. weight 403,52 g/mol



## RL-2810 Boc-AEDI-OH

2-((2-(*t*-Butyloxycarbonylamino)ethyl)disulfanyl)-2-methylpropanoic acid

CAS-No. 144700-78-7
Formula $C_{11}H_{21}NO_4S_2$
Mol. weight 295,42 g/mol



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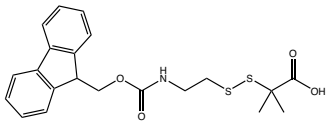
Product details

## RL-2800 Fmoc-AEDI-OH

2-((2-((9-Fluorenylmethyloxycarbonyl)amino)ethyl) disulfanyl)-2-methylpropanoic acid

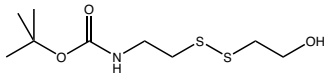
| | |
|---|---|
| CAS-No. | 1823244-38-7 |
| Formula | $C_{21}H_{23}NO_4S_2$ |
| Mol. weight | 417,54 g/mol |



## RL-3510 Boc-NH-SS-OH

2-((2-(*t*-Butyloxycarbonylamino)ethyl)disulfaneyl) ethan-1-ol

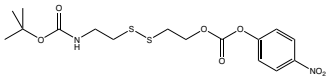
| | |
|---|---|
| CAS-No. | 877864-07-8 |
| Formula | $C_9H_{19}NO_3S_2$ |
| Mol. weight | 253,38 g/mol |



## RL-3520 Boc-NH-SS-OpNC

2-((2-(*t*-Butyloxycarbonylamino)ethyl)disulfaneyl) ethan-1-yl *p*-nitrophenylcarbonate

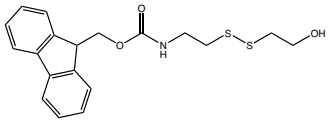
| | |
|---|---|
| CAS-No. | 2040301-00-4 |
| Formula | $C_{16}H_{22}N_2O_7S_2$ |
| Mol. weight | 418,48 g/mol |



## RL-3530 Fmoc-NH-SS-OH

2-((2-((9-Fluorenylmethyloxycarbonyl)amino)ethyl) disulfaneyl)ethan-1-ol

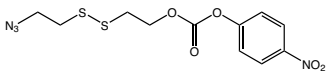
| | |
|---|---|
| CAS-No. | 2576471-39-9 |
| Formula | $C_{19}H_{21}NO_3S_2$ |
| Mol. weight | 375,50 g/mol |



## RL-4150 Azido-SS-OpNC

2-((2-azidoethyl)disulfanyl)ethyl (4-nitrophenyl) carbonate

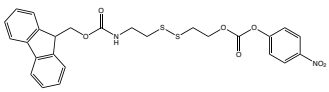
| | |
|---|---|
| CAS-No. | 2766027-28-3 |
| Formula | $C_{11}H_{12}N_4O_5S_2$ |
| Mol. weight | 344,36 g/mol |



## RL-3540 Fmoc-NH-SS-OpNC

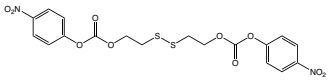
2-((2-((9-Fluorenylmethyloxycarbonyl)amino)ethyl) disulfaneyl)ethan-1-yl *p*-nitrophenylcarbonate

| | |
|---|---|
| CAS-No. | 2576471-53-7 |
| Formula | $C_{26}H_{24}N_2O_7S_2$ |
| Mol. weight | 540,61 g/mol |



## RL-4160 pNCO-SS-OpNC

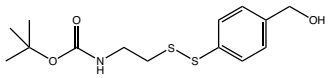
disulfanediylbis(ethane-2,1-diyl) bis(4-nitrophenyl) bis(carbonate)



| | |
|---|---|
| CAS-No. | 1435972-52-3 |
| Formula | $C_{18}H_{16}N_2O_{10}S_2$ |
| Mol. weight | 484,45 g/mol |

## RL-3560 Boc-NH-SS-Bzl-OH

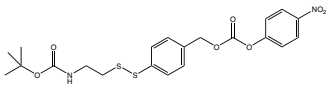
4-((2-(*t*-Butyloxycarbonylamino)ethyl)disulfaneyl) benzylalcohol



| | |
|---|---|
| CAS-No. | 2576471-54-8 |
| Formula | $C_{14}H_{21}NO_3S_2$ |
| Mol. weight | 315,45 g/mol |

## RL-3570 Boc-NH-SS-Bzl-OpNC

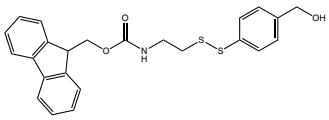
4-((2-(*t*-Butyloxycarbonylamino)ethyl)disulfaneyl) benzyl *p*-nitrophenylcarbonate



| | |
|---|---|
| CAS-No. | 2576471-38-8 |
| Formula | $C_{21}H_{24}N_2O_7S_2$ |
| Mol. weight | 480,55 g/mol |

## RL-3580 Fmoc-NH-SS-Bzl-OH

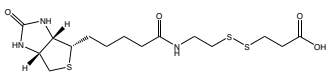
4-((2-((9-Fluorenylmethyloxycarbonyl)amino)ethyl) disulfaneyl)benzylalcohol



| | |
|---|---|
| CAS-No. | 2064282-26-2 |
| Formula | $C_{24}H_{23}NO_3S_2$ |
| Mol. weight | 437,57 g/mol |

## RL-3300 Biotin-SS-COOH

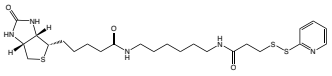
3-((2-(5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d] imidazol-4-yl)pentanamido)ethyl)disulfanyl)propanoic acid



| | |
|---|---|
| CAS-No. | 104582-29-8 |
| Formula | $C_{15}H_{25}N_3O_4S_3$ |
| Mol. weight | 407,57 g/mol |

## RL-8415 Biotin-Hx-SS-Py

5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imida-zol-4-yl)-N-(6-(3-(pyridin-2-yldisulfanyl)propanamido) hexyl)pentanamide



| | |
|---|---|
| CAS-No. | 129179-83-5 |
| Formula | $C_{24}H_{37}N_5O_3S_3$ |
| Mol. weight | 539,77 g/mol |

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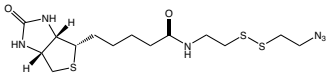
## RL-4120 Biotin-SS-$N_3$

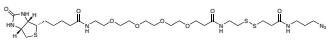
N-(2-((2-azidoethyl)disulfanyl)ethyl)-5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanamide



| | |
|---|---|
| CAS-No. | 1620523-64-9 |
| Formula | $C_{14}H_{24}N_6O_2S_3$ |
| Mol. weight | 404,57 g/mol |

## PEG8100 Biotin-PEG(4)-SS-Azide

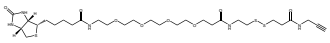
N-(2-((3-((3-azidopropyl)amino)-3-oxopropyl)disulfaneyl)ethyl)-1-(5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanamido)-3,6,9,12-tetraoxapentadecan-15-amide



| | |
|---|---|
| CAS-No. | 1260247-52-6 |
| Formula | $C_{29}H_{52}N_8O_8S_3$ |
| Mol. weight | 736,96 g/mol |

## PEG8110 Biotin-PEG(4)-SS-Alkyne

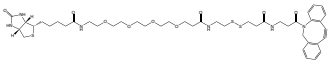
N-(2-((3-oxo-3-(prop-2-ynylamino)propyl)disulfanyl)ethyl)-1-(5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanamido)-3,6,9,12-tetraoxapentadecan-15-amide



| | |
|---|---|
| CAS-No. | 1260247-54-8 |
| Formula | $C_{29}H_{49}N_5O_8S_3$ |
| Mol. weight | 691,92 g/mol |

## PEG8120 Biotin-PEG(4)-SS-DBCO

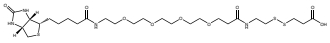
N-(2-((3-(3-(azadibenzocyclooctyn-1-yl)-3-oxopropylamino)-3-oxopropyl)disulfanyl)ethyl)-1-(5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanamido)-3,6,9,12-tetraoxapentadecan-15-am ide



| | |
|---|---|
| Formula | $C_{44}H_{60}N_6O_9S_3$ |
| Mol. weight | 913,18 g/mol |

## PEG8090 Biotin-PEG(4)-SS-COOH

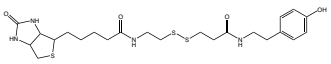
9,25-dioxo-29-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)-12,15,18,21-tetraoxa-4,5-dithia-8,24-diazanonacosan-1-oic acid



| | |
|---|---|
| CAS-No. | 1380166-80-2 |
| Formula | $C_{26}H_{46}N_4O_9S_3$ |
| Mol. weight | 654,86 g/mol |

## LS-3570 Biotin-SS-Tyramide

N-(2-((3-(4-hydroxyphenethylamino)-3-oxopropyl) disulfanyl)ethyl)-5-(2-oxohexahydro-1H-thieno[3,4-d] imidazol-4-yl)pentanamide



| | |
|---|---|
| CAS-No. | 678975-20-7 |
| Formula | $C_{23}H_{34}N_4O_4S_3$ |
| Mol. weight | 526,74 g/mol |

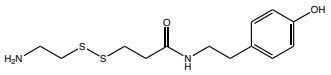
## LS-3930 Biotin-PEG(4)-SS-Tyramide

N-(2-((3-(4-hydroxyphenethylamino)-3-oxopropyl)di-sulfanyl)ethyl)-1-(5-(2-oxohexahydro-1H-thieno[3,4-d] imidazol-4-yl)pentanamido)-3,6,9,12-tetraoxapenta-decan-15-amide

| | |
|---|---|
| Formula | $C_{34}H_{55}N_5O_9S_3$ |
| Mol. weight | 774,02 g/mol |

## LS-3960 Tyramide-SS-amine*HCl

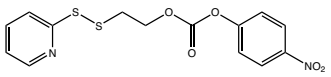
3-((2-aminoethyl)disulfanyl)-N-(4-hydroxyphenethyl) propanamide hydrochloride



| | |
|---|---|
| CAS-No. | 2576471-37-7 net |
| Formula | $C_{13}H_{20}N_2O_2S_2$*HCl |
| Mol. weight | 300,44*36,45 g/mol |

## RL-3500 OPSS-OpNC

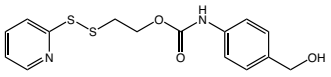
2-(2-Pyridithio)ethyl-*p*-nitrophenylcarbonate



| | |
|---|---|
| CAS-No. | 874302-76-8 |
| Formula | $C_{14}H_{12}N_2O_5S_2$ |
| Mol. weight | 352,38 g/mol |

## RL-3890 OPSS-PAB

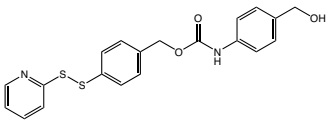
2-(pyridin-2-yldisulfaneyl)ethyl (4-(hydroxymethyl) phenyl)carbamate



| | |
|---|---|
| CAS-No. | 2362536-42-1 |
| Formula | $C_{15}H_{16}N_2O_3S_2$ |
| Mol. weight | 336,42 g/mol |

## RL-3920 OPSS-BzI-PAB

4-(pyridin-2-yldisulfaneyl)benzyl (4-(hydroxymethyl) phenyl)carbamate



| | |
|---|---|
| Formula | $C_{20}H_{18}N_2O_3S_2$ |
| Mol. weight | 398,50 g/mol |

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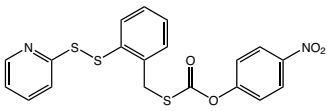
Product details

## RL-4170 2-OPSS-Bzl-OpNC

O-(4-nitrophenyl) S-(2-(pyridin-2-yldisulfanyl)benzyl) carbonothioate

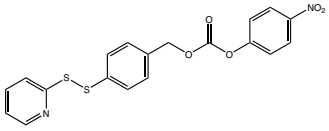
| | |
|---|---|
| CAS-No. | 1384425-52-8 |
| Formula | $C_{19}H_{24}N_2O_4S_3$ |
| Mol. weight | 430,51 g/mol |



## RL-3550 OPSS-Bzl-OpNC

(4-(pyridin-2-yldisulfaneyl)benzyl) *p*-nitrophenylcarbonate

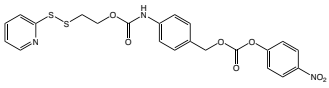
| | |
|---|---|
| CAS-No. | 1151989-04-6 |
| Formula | $C_{19}H_{14}N_2O_5S_2$ |
| Mol. weight | 414,45 g/mol |



## RL-3820 OPSS-PAB-OpNC

2-(pyridin-2-yldisulfaneyl)ethyl (4-((((4-nitrophenoxy) carbonyl)oxy)methyl)phenyl)carbamate

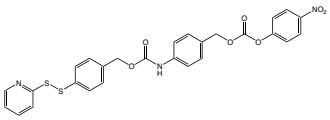
| | |
|---|---|
| CAS-No. | 2362536-70-5 |
| Formula | $C_{22}H_{19}N_3O_7S_2$ |
| Mol. weight | 501,53 g/mol |



## RL-3850 OPSS-Bzl-PAB-OpNC

4-(pyridin-2-yldisulfaneyl)benzyl (4-((((4-nitrophenoxy) carbonyl)oxy)methyl)phenyl)carbamate

| | |
|---|---|
| Formula | $C_{27}H_{21}N_3O_7S_2$ |
| Mol. weight | 563,60 g/mol |



***References:***


- *Labeling Carboxyl Groups of Surface-Exposed Proteins Provides an Orthogonal Approach for Cell Surface Isolation; N. E. Ozkan Kucuk, E. Sanal, E. Tan, T. Mitchison, N. Ozlu;* ***J Proteome Res*** *2018;* ***17****: 1784-1793. https://doi.org/10.1021/acs.jproteome.7b00825*
- *Development of trifunctional probes for glycoproteomic analysis; C. S. Tsai, P. Y. Liu, H. Y. Yen, T. L. Hsu, C. H. Wong;* ***Chem Commun*** *2010;* ***46****: 5575-7. https://doi.org/10.1039/c0cc00345j*
- *Fluorometric assay for tissue transglutaminase-mediated transamidation activity; C. Gnaccarini, W. Ben-Tahar, W. D. Lubell, J. N. Pelletier, J. W. Keillor;* ***Bioorg Med Chem*** *2009;* ***17****: 6354-9. https://doi.org/10.1016/j.bmc.2009.07.031*
- *Convenient synthesis of photoaffinity probes and evaluation of their labeling abilities; T. Kan, Y. Kita, Y. Morohashi, Y. Tominari, S. Hosoda, T. Tomita, H. Natsugari, T. Iwatsubo, T. Fukuyama;* ***Org Lett*** *2007;* ***9****: 2055-8. https://doi.org/10.1021/ol070376i*
- *Facile synthesis toward the construction of an activity probe library for glycosidases; T. H. Shie, Y. L. Chiang, J. J. Lin, Y. K. Li, L. C. Lo;* ***Carbohydr Res*** *2006;* ***341****: 443-56. https://doi.org/10.1016/j.carres.2005.12.005*
- *A simple photo-affinity labeling protocol; H.-y. Li, Y. Liu, K. Fang, K. Nakanishi;* ***Chemical Communications*** *1999; 365-366. https://doi.org/10.1039/a809507h*

→ *Linker Technologies for Antibody–Drug Conjugates; B. Nolting;* ***Antibody-Drug Conjugates L. Ducry*** *2013;* ***1045****: 71-100. https://doi.org/10.1007/978-1-62703-541-5_5*
→ *Disulfide-Based Self-Immolative Linkers and Functional Bioconjugates for Biological Applications; Z. Deng, J. Hu, S. Liu;* ***Macromol Rapid Commun*** *2020;* ***41****: e1900531. https://doi.org/10.1002/marc.201900531.*
→ *Reduction-Triggered Transformation of Disulfide-Containing Micelles at Chemically Tunable Rates; Z. Deng, S. Yuan, R. X. Xu, H. Liang, S. Liu;* ***Angew. Chem. Int. Ed.*** *2018;* ***57****: 8896-8900. https://doi.org/10.1002/anie.201802909*
→ *Modulated Fragmentation of Proapoptotic Peptide Nanoparticles Regulates Cytotoxicity; T. Suma, J. Cui, M. Müllner, S. Fu, J. Tran, K. F. Noi, Y. Ju, F. Caruso;* ***J. Am. Chem. Soc.*** *2017;* ***139****: 4009-4018. https://doi.org/10.1021/jacs.6b11302*
→ *Engineering Intracelular Delivery Nanocarriers and Nanoreactors from Oxidation-Responsive Polymersomes via Synchronized Bilayer Cross-Linking and Permeabilizing Inside Live Cells; Z. Deng, Y. Qian, Y. Yu, G. Liu, J. Hu, G. Zhang, S. Liu;* ***J. Am. Chem. Soc.*** *2016;* ***138****: 10452-10466. https://doi.org/10.1021/jacs.6b04115*

## 3.6. Dde-Based Linkers

The Dde [N-1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)-3-ethyl] protecting group is commonly utilized to protect the side-chain amine groups of lysine, ornithine, 2,4-diaminobutyric acid, and 2,3-diaminopropionic acid. Dde shows orthogonal cleavage conditions to Fmoc (piperidine or DBU) and *t*Bu (TFA) deprotecting protocols and is stable to denaturing washing conditions, while allowing for a mild and selective removal in the presence of other protecting groups using a buffered aqueous solution of hydrazine or hydroxylamine, thus representing a versatile tool for the site-specific modification of peptides. Advantageously, the cleavage can be followed spectrophotometrically since the reaction product of Dde with hydrazine is a chromophoric derivative.

Placing Dde as one terminal group of a linker and a functional group prone for conjugation as the other, or using Dde as the central connective portion of a linker, allows for the creation of new bifunctional linkers that can be selectively and temporarily attached to:

- Appropriately modified biomolecules for binding to streptavidin (with terminal biotin) (*Fig. 25 (A)*), or conjugation to any solid supports, e.g., *via* Click reaction (*Fig. 25 (B)*).
- Solubilizing tags, e.g., hexa-lysine (“helping-hand linkers”, *Fig. 25 (C)*), oligo-arginine, PEGs (*Fig. 25 (D)*) or other hydrophilic groups improving solubility of hydrophobic peptides or other compounds when being attached to either the N-terminus or any lysine side-chain within a peptide sequence.
- Dyes and any other conjugate for monitoring, diagnostics, targeting or other purposes.



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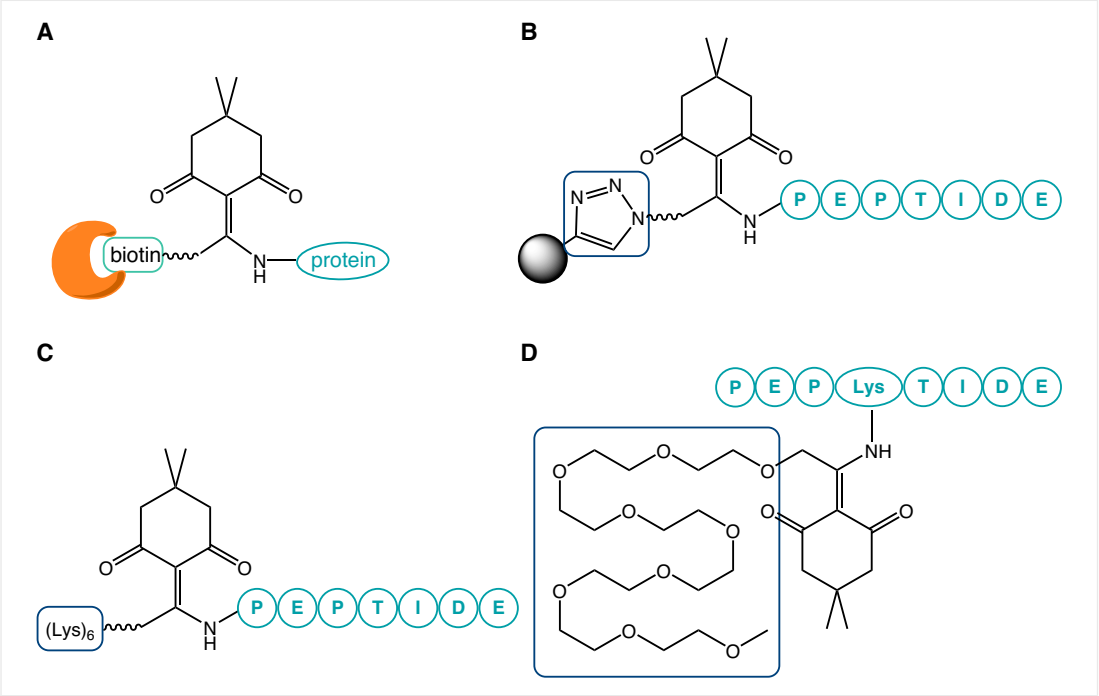


*Fig. 25: Dde-based linkers can be utilized for various applications: Attachment of a cleavable biotin tag to proteins for catch-and-release affinity purification over Streptavidin beads (A), reversible labeling or conjugation to other biomolecules, or reversible immobilization on solid supports via Click chemistry (B), temporary attachment of solubilizing tags like oligo-lysine (C) or PEGs (D).*

**Dde/ivDde linkers are implemented in simple and nearly quantitative steps:**

1. Orthogonal deprotection of lysine residues in a peptide or N-terminus or any other amino function of a hydrophobic compound.
2. On-resin incorporation of the linker.
3. Fmoc-SPPS elongation.
4. Cleavage of the peptide from the resin and removal of all side-chain protecting groups.
5. The tagged peptide can be separated from truncated sequences.
6. In-solution cleavage using mild aqueous hydrazine to cleave the Dde linker after purification, streptavidin attachment, NCL-based assembly or another reaction step. The cleavage can be monitored spectroscopically as the resulting pyrazole shows a strong absorption at 290 nm.

Dde/ivDde becomes particularly useful for handling and purification of insoluble and aggregationprone peptides, as any appropriate solubilizing promoting group can be attached to create so-called “helping-hand” linkers that can be removed in a traceless manner (*Fig. 26*).

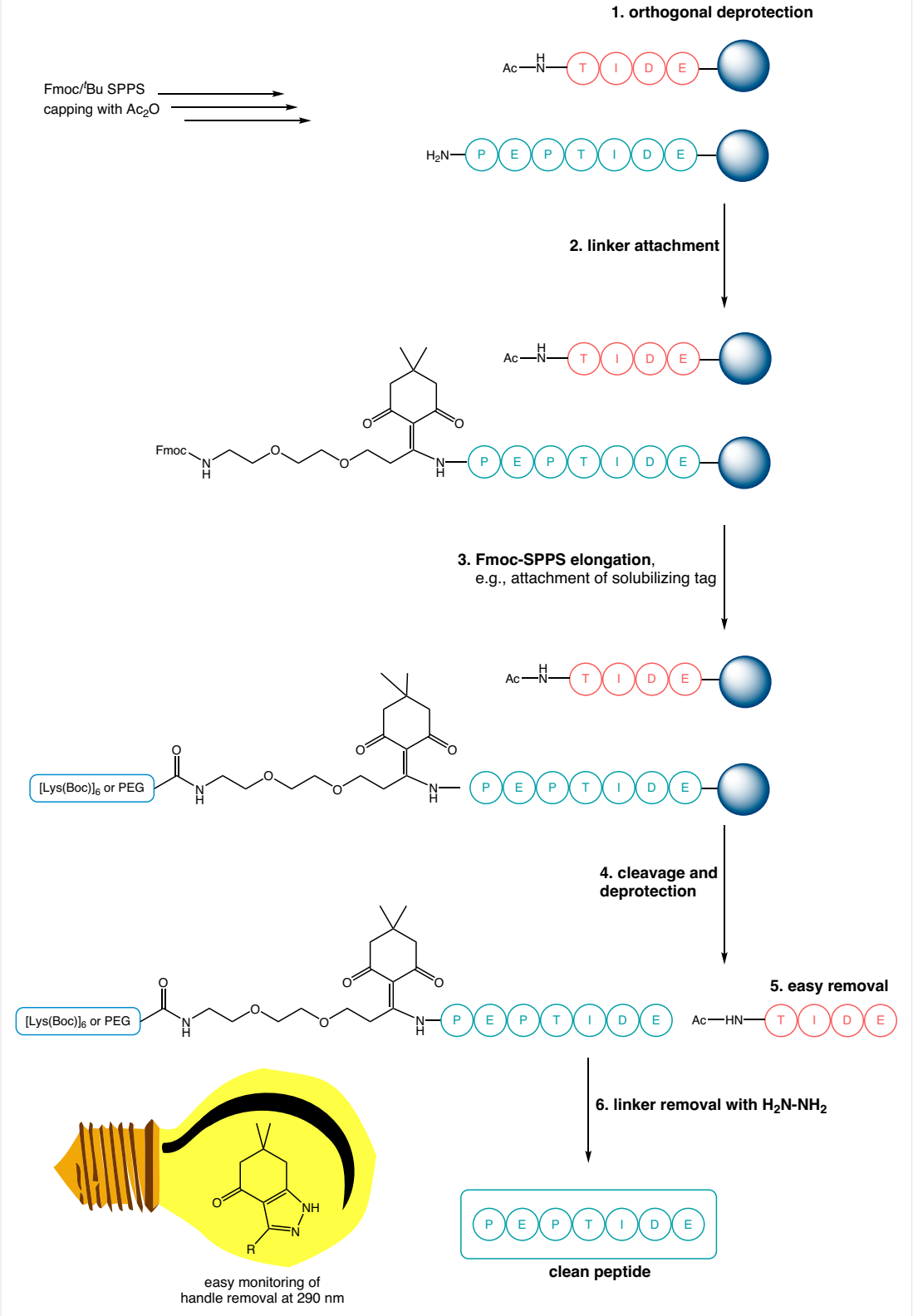


*Fig. 26: Dde-based "helping-hand" linkers improve the solubility and allow for the purification of hydrophobic peptides. Removal of the handle can be easily monitored spectroscopically at 290 nm.*

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***Procedure for Removing Helping-Hands from Peptides*** (adapted from Jacobsen *et al.*, *J Am Chem Soc* 2016):

10 mL of 2 M hydrazine stock solution (pH 7.5) is being prepared as follows:

1. Weigh 5.7 g Guanidinium chloride and 75 mg DTT into 15 mL Falcon tube.
2. Add 1 mL of 1 M $NaH_2PO_4$.
3. Add 2 mL of 10 M hydrazine in water.
4. Add 0.5 mL of 12 M HCl.
5. Dissolve solution by thorough vortexing.
6. Adjust pH to 7.5 by adding concentrated HCl.
7. Fill to a final volume of 10 mL with water.
8. Filter solution using 0.2 µm syringe filter.

Cleavage of the helping-hand can be triggered by equivolume addition of 2 M hydrazine stock solution into the solution of the peptide. After adding the hydrazine solution, subtle adjustment may be necessary to achieve a final solution pH of 7.5. The reaction is normally completed within minutes. Deprotection can be monitored spectrophotometrically at 290 nm.

Despite its widespread use, the biotinylation of proteins for subsequent purification *via* Streptavidin beads bears certain hurdles, e.g., concerning the removal of the proteins from the beads due to the strong binding. One possible improvement is represented by the use of appropriately derivatized Dde-linkers. The connection of such a bifunctional linker with a biotin moiety on the one end, and a clickable group (alkyne, e.g., DBCO) or tyramide on the other, allows for the selective attachment to appropriately modified biomolecules, as well as the mild release of captured proteins from the beads after purification (*Fig. 27*).

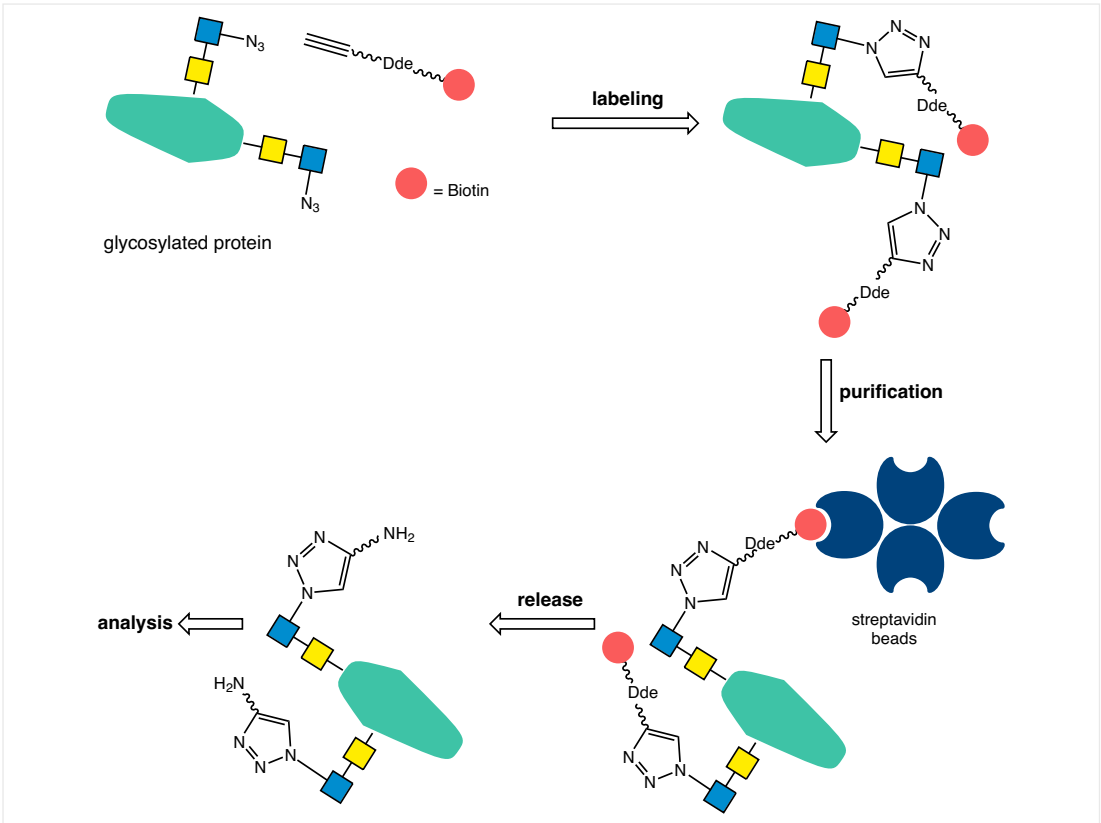


*Fig. 27: Biotinylation of an azide-bearing glycoprotein using Click chemistry, followed by purification of the labelled protein over Streptavidin beads, release of the protein by hydrazinolysis of the Dde-group, and analysis of the isolated glycoprotein (adapted from Griffin et al. Mol. Biosys. 2016).*

Aside from the commonly used cleavage solution for Dde consisting of 2% hydrazine monohydrate in $H_2O$, the following procedure may be used in order to ensure full orthogonality between Dde and Fmoc.

**Selective Removal of Dde/ivDde using hydroxylamine** (adapted from Díaz-Mochón *et al.*, *Org. Lett.* 2004):

1.25 g (1.80 mmol) of $NH_2OH{\cdot}HCl$ and 0.918 g (1.35 mmol) of Imidazole were suspended in 5 mL NMP, and the mixture sonicated until complete dissolution. This solution can be stirred for at least 2 weeks at -20 °C. Just before reaction, five volumes of this solution were diluted with one volume of alternatively DCM or DMF.

Dde is easy to cleave, but not very robust. Thus, during Fmoc cleavage, Dde might migrate to free lysine ε-amino groups ("scrambling") or, in rare cases, even to the peptide's free amino terminus. Especially during the synthesis of longer peptide sequences, a certain extent of Dde is removed during Fmoc cleavage with piperidine.

The sterically more demanding protecting group ivDde is more stable towards piperidine and does usually not migrate to free lysine amino groups. However, for some sequences, total removal of the robust ivDde is hardly possible - especially near the C-terminus or in aggregating sequences.

A newer group for the orthogonal protecting of amino groups reported in literature is MeDmb (methyl dimethylbarbituric acid). At Iris Biotech, we are also offering the sterically more demanding ivDmb. There is no "one-fits-all" when you are to decide on the protecting group selection for orthogonal lysine side-chain protection. When your peptide shows a low tendency for scrambling, Lys(Dde) may be fine. Otherwise, you can switch to more robust ones such as Lys(ivDde) or Lys(ivDmb). If you do not succeed in completely removing the ivDde protecting group, the brand-new ivDmb should be your choice. For more details on the ivDmb protecting group, see our blog post:
https://www.iris-biotech.de/blog/potm-next-generation-lysine-side-chain-protecting-groups/

***References:***


- *A novel lysine-protecting procedure for continuous flow solid phase synthesis of branched peptides; B. W. Bycroft, W. C. Chan, S. R. Chhabra, N. D. Hone;* ***Journal of the Chemical Society, Chemical Communications*** *1993: 778-779.* https://doi.org/10.1039/c39930000778
- *An appraisal of new variants of Dde amine protecting group for solid phase peptide synthesis; S. R. Chhabra, B. Hothi, D. J. Evans, P. D. White, B. W. Bycroft, W. C. Chan;* ***Tetrahedron Letters*** *1998;* ***39****: 1603-1606.* https://doi.org/10.1016/s0040-4039(97)10828-0
- *Investigation on the stability of the Dde protecting group used in peptide synthesis: migration to an unprotected lysine; K. Augustyns, W. Kraas, G. Jung;* ***J Pept Res*** *1998;* ***51****: 127-33.* https://doi.org/10.1111/j.1399-3011.1998.tb00630.x
- *Evaluation of ivDde as a quasi-orthogonal protecting group for Fmoc solid-phase peptide synthesis; R. R. Wilhelm, A. Srinivasan, M. A. Schmidt; Peptides for the New Millennium: Proceedings of the 16th American Peptide Symposium June 26–July 1, 1999, Minneapolis, Minnesota, USA 2000: 58-59.* https://doi.org/10.1007/0-306-46881-6_19
- *Synthesis of a chlorothalonil peptide conjugate mimicking protein-bound pesticide residues; H. Hrenn, W. Schwack, W. Seilmeier, H. Wieser;* ***Tetrahedron Lett*** *2003;* ***44****: 1911-1913.* https://doi.org/10.1016/S0040-4039(03)00121-7
- *Reaction of 1,3-dimethyl-5-acetyl-barbituric acid (DAB) with primary amines. Access to intermediates for selectively protected spermidines; E. T. da Silva, E. L. S. Lima;* ***Tetrahedron Lett*** *2003;* ***44****: 3621-3624.* https://doi.org/10.1016/S0040-4039(03)00709-3
- *Scope and Limitations of Barbituric and Thiobarbituric Amino Acid Derivatives as Protecting Groups for Solid-Phase Peptide Synthesis: Towards a Green Protecting Group; S. Ramkisson, H. H. Al-Rasheed, K. A. Dahlous, B. G. De La Torre, A. El-Faham, F. Albericio;* ***ChemistrySelect*** *2021;* ***6****: 6626-6630.* https://doi.org/10.1002/slct.202101539


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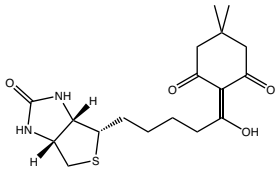
Product details

## LS-4020 Biotin-Dde

2-(1-hydroxy-5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentylidene)-5,5-dimethylcyclohexane-1,3-dione

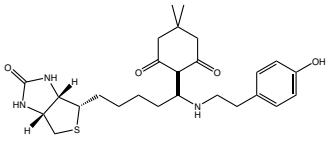
CAS-No. 194038-08-9
Formula $C_{18}H_{26}N_2O_4S$
Mol. weight 366,48 g/mol



## LS-4000 Biotin-Dde-Tyramide

2-(1-(4-hydroxyphenethylamino)-5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentylidene)-5,5-dimethylcyclohexane-1,3-dione

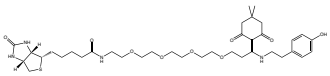
CAS-No. 2819732-80-2
Formula $C_{26}H_{35}N_3O_4S$
Mol. weight 485,64 g/mol



## PEG8130 Biotin-PEG(4)-Dde-Tyramide

N-(15-(4,4-dimethyl-2,6-dioxocyclohexylidene)-18-(4-hydroxyphenyl)-3,6,9,12-tetraoxa-16-azaoctadecyl)-5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanamide

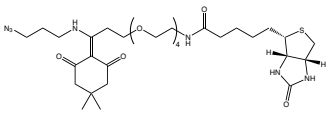
CAS-No. 2814457-48-0
Formula $C_{34}H_{56}N_4O_9S$
Mol. weight 732,93 g/mol



## PEG7960 Biotin-PEG(4)-Dde-$N_3$

N-(19-azido-15-(4,4-dimethyl-2,6-dioxocyclohexylidene)-3,6,9,12-tetraoxa-16-azanonadecyl)-5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanamide

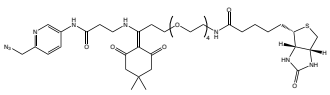
CAS-No. 1802907-93-2
Formula $C_{32}H_{53}N_7O_8S$
Mol. weight 695,87 g/mol



## PEG7970 Biotin-PEG(4)-Dde-Picolyl-$N_3$

N-(6-(azidomethyl)pyridin-3-yl)-15-(4,4-dimethyl-2,6-dioxocyclohexylidene)-1-(5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanamido)-3,6,9,12-tetraoxa-16-azanonadecan-19-amide

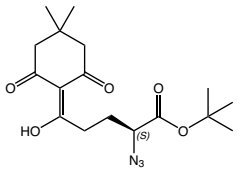
CAS-No. 2055048-42-3
Formula $C_{38}H_{57}N_9O_9S$
Mol. weight 815,98 g/mol



## RL-8700 $N_3$-L-Glu(Dde)-O*t*Bu

*tert*-butyl (S)-2-azido-5-(4,4-dimethyl-2,6-dioxocyclohexylidene)-5-hydroxypentanoate

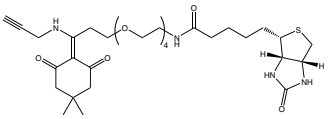
| | |
|---|---|
| Formula | $C_{17}H_{25}N_3O_5$ |
| Mol. weight | 351,40 g/mol |



## PEG7980 Biotin-PEG(4)-Dde-Alkyne

N-(15-(4,4-dimethyl-2,6-dioxocyclohexylidene)-3,6,9,12-tetraoxa-16-azanonadec-18-ynyl)-5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanamide

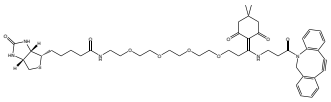
| | |
|---|---|
| CAS-No. | 1802908-00-4 |
| Formula | $C_{32}H_{50}N_4O_8S$ |
| Mol. weight | 650,83 g/mol |



## PEG8140 Biotin-PEG(4)-Dde-DBCO

N-(15-(4,4-dimethyl-2,6-dioxocyclohexylidene)-19-oxo-19-(azadibenzocyclooctyn-1-yl)-3,6,9,12-tetraoxa-16-azanonadecyl)-5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d]imidazol-4-yl)pentanamide

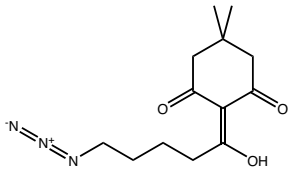
| | |
|---|---|
| CAS-No. | 1807512-43-1 |
| Formula | $C_{47}H_{61}N_5O_9S$ |
| Mol. weight | 872,08 g/mol |



## RL-3280 $N_3$-Pen-Dde

2-(5-azido-1-hydroxypentylidene)-5,5-dimethylcyclohexane-1,3-dione

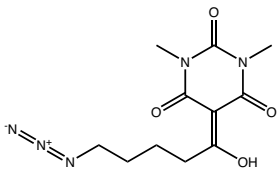
| | |
|---|---|
| CAS-No. | 1867129-38-1 |
| Formula | $C_{13}H_{19}N_3O_3$ |
| Mol. weight | 265,31 g/mol |



## RL-3290 $N_3$-Pen-Dtpp

5-(5-azido-1-hydroxypentylidene)-1,3-dimethylpyrimidine-2,4,6(1H,3H,5H)-trione

| | |
|---|---|
| CAS-No. | 1867129-42-7 |
| Formula | $C_{11}H_{15}N_5O_4$ |
| Mol. weight | 281,27 g/mol |



The Dde derived linker might cleave under mildly acidic and even neutral conditions in the one or the other case. The DTPM derived linker is totally stable under acidic conditions as well as to a wide range of chemical treatments, including particularly harsh sodium methoxide-based deacetylation of chemically introduced glycans.

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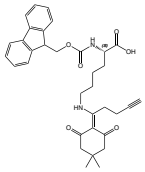
***Reference:***

→ *Combining triazole ligation and enzymatic glycosylation on solid phase simplifies the synthesis of very long glycoprotein analogues; M. Galibert, V. Piller, F. Piller, V. Aucagne, A. F. Delmas;* ***Chem. Sci.*** *2015;* ***6****: 3617-3623.*
*https://doi.org/10.1039/c5sc00773a*

Product details

## FAA8115 Fmoc-L-Lys(Pentynoyl-DIM)-OH

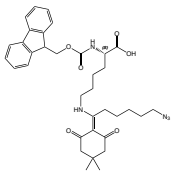
N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-epsilon-[1-(4,4-dimethyl-2,6- dioxocyclohexylidene)pent-4-yn-1-yl]-L-lysine



| | |
|---|---|
| CAS-No. | 2408993-33-7 |
| Formula | $C_{34}H_{38}N_2O_6$ |
| Mol. weight | 570,69 g/mol |

## FAA8145 Fmoc-L-Lys($N_3$-Aca-DIM)-OH

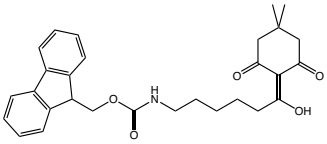
N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-epsilon-[6-azido-1-(4,4-dimethyl-2,6- dioxocyclohexylidene)hexyl]-L-lysine



| | |
|---|---|
| CAS-No. | 2408993-39-3 |
| Formula | $C_{35}H_{43}N_5O_6$ |
| Mol. weight | 629,76 g/mol |

## RL-3260 Fmoc-Aca-DIM

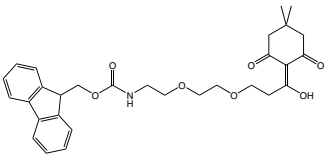
6-((9-Fluorenylmethyl)oxycarbonylamino)-1-(4,4-dimethyl-2,6-dioxocyclohexylidene)-hexan-1-ol



| | |
|---|---|
| CAS-No. | 2379561-08-5 |
| Formula | $C_{29}H_{33}NO_5$ |
| Mol. weight | 475,58 g/mol |

## RL-3270 Fmoc-AEEP-DIM

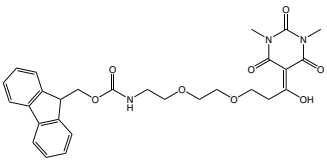
3-(2-(2-(9-Fluorenylmethyl)oxycarbonylaminoethoxy) ethoxy)-1-(4,4-dimethyl-2,6-dioxocyclohexylidene)-propan-1-ol



| | |
|---|---|
| CAS-No. | 1988771-96-5 |
| Formula | $C_{30}H_{35}NO_7$ |
| Mol. weight | 521,60 g/mol |

## RL-3470 Fmoc-AEEP-DMB

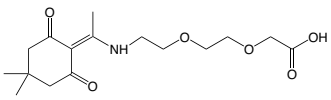
(9-Fluorenylmethyloxycarbonyl)amino-PEG(2)-Dtpp



| | |
|---|---|
| Formula | $C_{28}H_{31}N_3O_8$ |
| Mol. weight | 537,57 g/mol |

## DAA1016 Dde-$O_2$Oc-OH

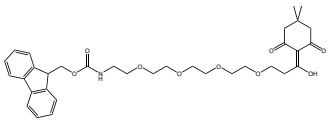
8-[(4,4-Dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl-amino]-3,6-dioxaoctanoic acid, {2-[2-(Dde-amino) ethoxy]ethoxy}acetic acid



| | |
|---|---|
| CAS-No. | 1263045-93-7 |
| Formula | $C_{16}H_{25}NO_6$ |
| Mol. weight | 327,37 g/mol |

## PEG8150 Fmoc-PEG(4)-Dde

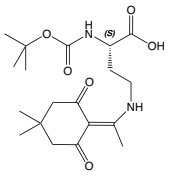
1-(9H-Fluorenylmethyloxycarbonylamino)-15-(4,4-dimethyl-2,6-dioxocyclohexylidene)-3,6,9,12-tetraoxapentadecyl-15-ol



| | |
|---|---|
| CAS-No. | 2093409-87-9 |
| Formula | $C_{34}H_{43}NO_9$ |
| Mol. weight | 609,71 g/mol |

## BAA1191 Boc-L-Dab(Dde)-OH

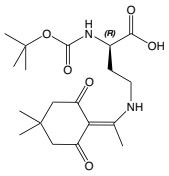
N-alpha-*t*-Butyloxycarbonyl-N-gamma-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)ethyl]-L-2,4-diaminobutyric acid



| | |
|---|---|
| CAS-No. | 1263045-50-6 |
| Formula | $C_{19}H_{30}N_2O_6$ |
| Mol. weight | 382,46 g/mol |

## BAA1171 Boc-D-Dab(Dde)-OH

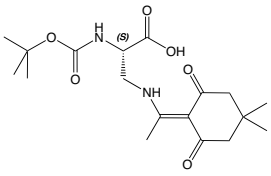
N-alpha-*t*-Butyloxycarbonyl-N-gamma-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)ethyl]-D-2,4-diaminobutyric acid



| | |
|---|---|
| CAS-No. | 1263046-41-8 |
| Formula | $C_{19}H_{30}N_2O_6$ |
| Mol. weight | 382,46 g/mol |

## BAA1193 Boc-L-Dap(Dde)-OH

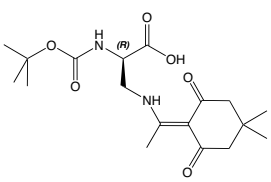
N-alpha-*t*-Butyloxycarbonyl-N-beta-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)ethyl]-L-2,3-diaminopropionic acid



| | |
|---|---|
| CAS-No. | 1263045-09-5 |
| Formula | $C_{18}H_{28}N_2O_6$ |
| Mol. weight | 368,43 g/mol |

## BAA1176 Boc-D-Dap(Dde)-OH

N-alpha-*t*-Butyloxycarbonyl-N-beta-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)ethyl]-D-2,3-diaminopropionic acid



| | |
|---|---|
| CAS-No. | 1263047-33-1 |
| Formula | $C_{18}H_{28}N_2O_6$ |
| Mol. weight | 368,43 g/mol |

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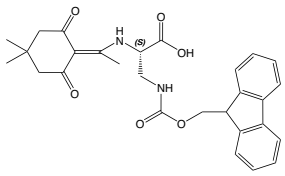
Product details

## DAA1012 Dde-L-Dap(Fmoc)-OH

N-alpha-(4-4-Dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl-N-beta-(9-fluorenylmethyloxycarbonyl)-L-2,3-diaminopropionic acid

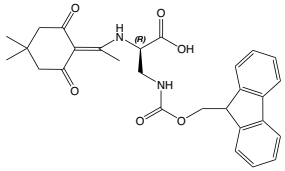
CAS-No. 1263046-98-5
Formula $C_{28}H_{30}N_2O_6$
Mol. weight 490,56 g/mol



## DAA1006 Dde-D-Dap(Fmoc)-OH

N-alpha-(4-4-Dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl-N-beta-(9-fluorenylmethyloxycarbonyl)-D-2,3-diaminopropionic acid

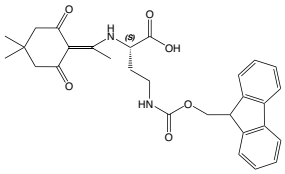
CAS-No. 1263046-87-2
Formula $C_{28}H_{30}N_2O_6$
Mol. weight 490,56 g/mol



## DAA1010 Dde-L-Dab(Fmoc)-OH

N-alpha-(4-4-Dimethyl-2,6-dioxocyclohex-1-ylidene)ethyl-N-gamma-(9-fluorenylmethyloxycarbonyl)-L-2,4-diaminobutyric acid

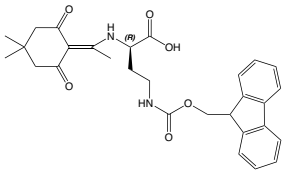
CAS-No. 1263045-85-7
Formula $C_{29}H_{32}N_2O_6$
Mol. weight 504,59 g/mol



## DAA1004 Dde-D-Dab(Fmoc)-OH

N-alpha-(4-4-Dimethyl-2,6-dioxocyclohex-1-ylidene)ethyl-N-gamma-(9-fluorenylmethyloxycarbonyl)-D-2,4-diaminobutyric acid

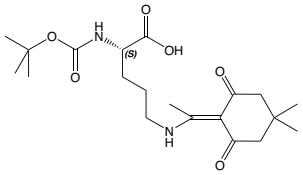
CAS-No. 1263046-84-9
Formula $C_{29}H_{32}N_2O_6$
Mol. weight 504,59 g/mol



## BAA1197 Boc-L-Orn(Dde)-OH

N-alpha-*t*-Butyloxycarbonyl-N-delta-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)ethyl]-L-ornithine

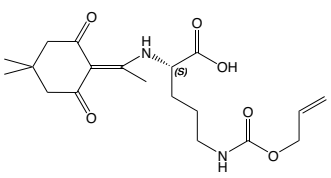
CAS-No. 1272755-14-2
Formula $C_{20}H_{32}N_2O_6$
Mol. weight 396,49 g/mol



## DAA1001 Dde-L-Orn(Aloc)-OH

N-alpha-(4-4-Dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl-N-delta-allyloxycarbonyl-L-ornithine

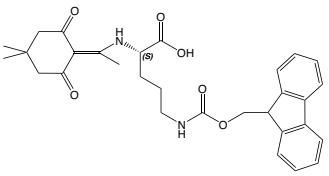
CAS-No. 1423017-98-4
Formula $C_{19}H_{28}N_2O_6$
Mol. weight 380,44 g/mol



## DAA1002 Dde-L-Orn(Fmoc)-OH

N-alpha-(4-4-Dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl-N-delta-(9-fluorenylmethyloxycarbonyl)-L-ornithine

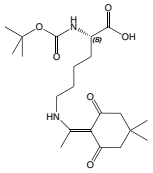
CAS-No. 1423017-87-1
Formula $C_{30}H_{34}N_2O_6$
Mol. weight 518,62 g/mol



## BAA1286 Boc-L-Lys(Dde)-OH*DCHA

N-alpha-*t*-Butyloxycarbonyl-N-epsilon-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)ethyl-L-lysine dicyclohexylamine

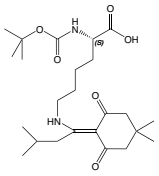
CAS-No. 444795-66-8
Formula $C_{21}H_{34}N_2O_6*C_{12}H_{23}N$
Mol. weight 410,51*181,32 g/mol



## BAA1287 Boc-L-Lys(ivDde)-OH

N-alpha-*t*-Butyloxycarbonyl-N-epsilon-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)-3-methylbutyl]-L-lysine

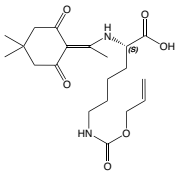
CAS-No. 862847-44-7
Formula $C_{24}H_{40}N_2O_6$
Mol. weight 452,6 g/mol



## DAA1013 Dde-L-Lys(Aloc)-OH*DCHA

N-alpha-(4-4-Dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl-N-epsilon-allyloxycarbonyl-L-lysine dicyclohexylamine

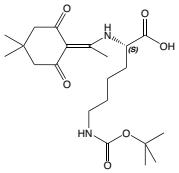
CAS-No. 264230-73-1 net
Formula $C_{20}H_{30}N_2O_6*C_{12}H_{23}N$
Mol. weight 394,47*181,32 g/mol



## DAA1014 Dde-L-Lys(Boc)-OH

N-alpha-(4-4-Dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl-N-epsilon-*t*-butyloxycarbonyl-L-lysine

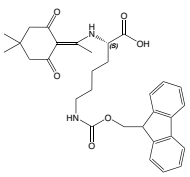
CAS-No. 1189586-14-8
Formula $C_{21}H_{34}N_2O_6$
Mol. weight 410,51 g/mol



## DAA1015 Dde-L-Lys(Fmoc)-OH

N-alpha-(4-4-Dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl-N-epsilon-(9-fluorenylmethyloxycarbonyl)-L-lysine

CAS-No. 156648-40-7
Formula $C_{31}H_{36}N_2O_6$
Mol. weight 532,64 g/mol



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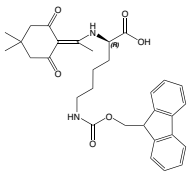
Product details

## DAA1017 Dde-D-Lys(Fmoc)-OH

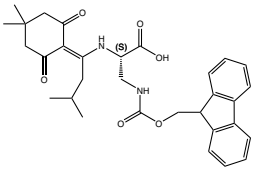
N-alpha-(4-4-Dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl-N-epsilon-(9-fluorenylmethyloxycarbonyl)-D-lysine



CAS-No. 1301706-71-7
Formula $C_{31}H_{36}N_2O_6$
Mol. weight 532,64 g/mol

## DAA1018 ivDde-L-Dap(Fmoc)-OH

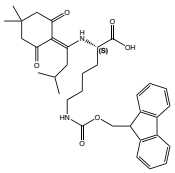
N-alpha-[(4,4-Dimethyl-2,6-dioxocyclohex-1-ylidene)-3-methylbutyl]-N-beta-(9-fluorenylmethyloxycarbonyl)-L-2,3-diaminopropionic acid



CAS-No. 2389078-71-9
Formula $C_{31}H_{36}N_2O_6$
Mol. weight 532,63 g/mol

## DAA1019 ivDde-L-Lys(Fmoc)-OH

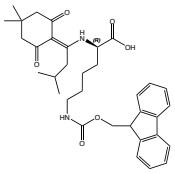
N-alpha-[(4,4-Dimethyl-2,6-dioxocyclohex-1-ylidene)-3-methylbutyl]-N-epsilon-(9-fluorenylmethyloxycarbonyl)-L-lysine



CAS-No. 1446752-60-8
Formula $C_{34}H_{42}N_2O_6$
Mol. weight 574,71 g/mol

## DAA1030 ivDde-D-Lys(Fmoc)-OH

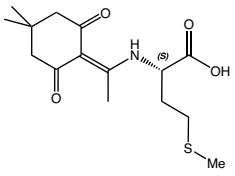
N-alpha-[(4,4-Dimethyl-2,6-dioxocyclohex-1-ylidene)-3-methylbutyl]-N-epsilon-(9-fluorenylmethyloxycarbonyl)-D-lysine



CAS-No. 2308529-94-2
Formula $C_{34}H_{42}N_2O_6$
Mol. weight 574,71 g/mol

## DAA1020 Dde-L-Met-OH

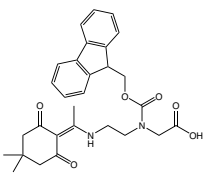
N-alpha-[1-(4,4-dimethyl-2,6-dioxocyclohexylidene) ethyl]-L-methionine



CAS-No. 1435266-87-7
Formula $C_{15}H_{23}NO_4S$
Mol. weight 313,13 g/mol

## FAA8690 Fmoc-Aeg(Dde)-OH

(9-Fluorenylmethyloxycarbonyl)-N-(2-((1-(4,4-dimethyl-2,6-dioxocyclohexylidene)ethyl)amino)ethyl) glycine

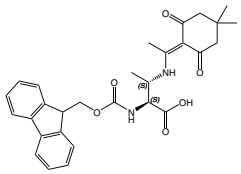


CAS-No. 2988301-08-0
Formula $C_{29}H_{32}N_2O_6$
Mol. weight 504,58 g/mol

## FAA8815 Fmoc-L-Abu(3-Dde-amino)-OH (2S,3S)

2-(Fmoc-amino)-3-(Dde-amino)butanoic acid (2S,3S)

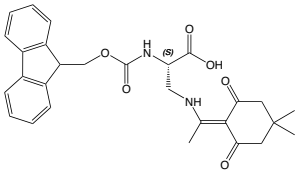
Formula $C_{29}H_{32}N_2O_6$
Mol. weight 504,58 g/mol



## FAA1462 Fmoc-L-Dap(Dde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-beta-[(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl]-2,3-diaminopropionic acid

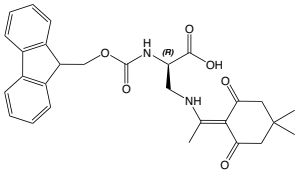
CAS-No. 247127-51-1
Formula $C_{28}H_{30}N_2O_6$
Mol. weight 490,56 g/mol



## FAA1476 Fmoc-D-Dap(Dde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-beta-[(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl]-D-2,3-diaminopropionic acid

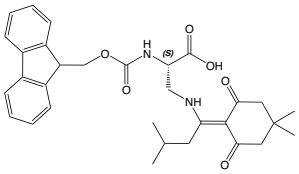
CAS-No. 210830-03-8
Formula $C_{28}H_{30}N_2O_6$
Mol. weight 490,56 g/mol



## FAA1464 Fmoc-L-Dap(ivDde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-beta-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)-3-methylbutyl]-L-2,3-diaminopropionic acid

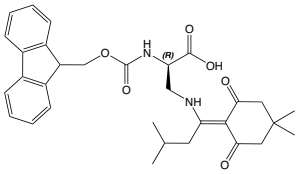
CAS-No. 607366-20-1
Formula $C_{31}H_{36}N_2O_6$
Mol. weight 532,64 g/mol



## FAA1478 Fmoc-D-Dap(ivDde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-beta-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)-3-methylbutyl]-D-2,3-diaminopropionic acid

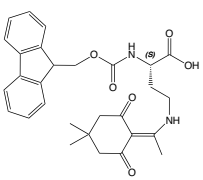
CAS-No. 1228900-15-9
Formula $C_{31}H_{36}N_2O_6$
Mol. weight 532,64 g/mol



## FAA1365 Fmoc-L-Dab(Dde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-gamma-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl-L-2,4-diaminobutyric acid

CAS-No. 235788-61-1
Formula $C_{29}H_{32}N_2O_6$
Mol. weight 504,59 g/mol



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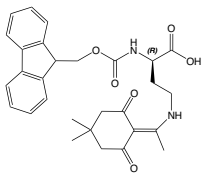
Product details

## FAA1318 Fmoc-D-Dab(Dde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-gamma-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl-D-2,4-diaminobutyric acid

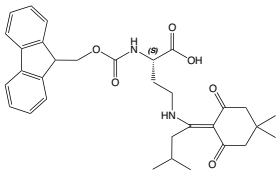
| | |
|---|---|
| CAS-No. | 596797-14-7 |
| Formula | $C_{29}H_{32}N_2O_6$ |
| Mol. weight | 504,59 g/mol |



## FAA1458 Fmoc-L-Dab(ivDde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-gamma-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)-3-methylbutyl]-L-2,4-diaminobutyric acid

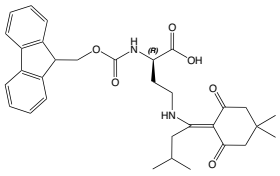
| | |
|---|---|
| CAS-No. | 607366-21-2 |
| Formula | $C_{32}H_{38}N_2O_6$ |
| Mol. weight | 546,67 g/mol |



## FAA1473 Fmoc-D-Dab(ivDde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-gamma-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)-3-methylbutyl]-D-2,4-diaminobutyric acid

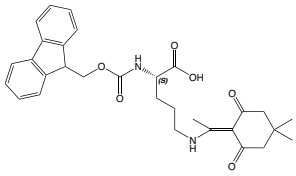
| | |
|---|---|
| CAS-No. | 872169-32-9 |
| Formula | $C_{32}H_{38}N_2O_6$ |
| Mol. weight | 546,67 g/mol |



## FAA1502 Fmoc-L-Orn(Dde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-delta-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl]-L-ornithine

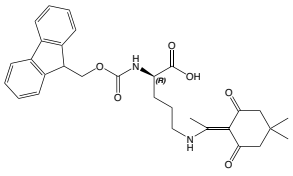
| | |
|---|---|
| CAS-No. | 269062-80-8 |
| Formula | $C_{30}H_{34}N_2O_6$ |
| Mol. weight | 518,62 g/mol |



## FAA2090 Fmoc-D-Orn(Dde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-delta-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl]-D-ornithine

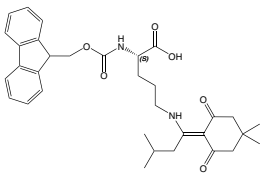
| | |
|---|---|
| CAS-No. | 1419640-31-5 |
| Formula | $C_{30}H_{34}N_2O_6$ |
| Mol. weight | 518,62 g/mol |



## FAA1503 Fmoc-L-Orn(ivDde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-delta[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)-3-methylbutyl]-L-ornithine solvate

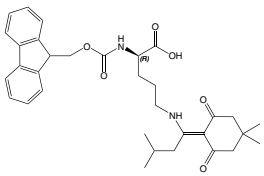
| | |
|---|---|
| CAS-No. | 1198321-33-3 |
| Formula | $C_{33}H_{40}N_2O_6$ |
| Mol. weight | 560,7 g/mol |



## FAA1493 Fmoc-D-Orn(ivDde)-OH.solv.

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-delta-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)-3-methylbutyl]-D-ornithine.solv.

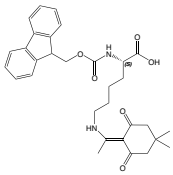
| | |
|---|---|
| CAS-No. | 1272754-86-5 |
| Formula | $C_{33}H_{40}N_2O_6$ |
| Mol. weight | 560,7 g/mol |



## FAA1390 Fmoc-L-Lys(Dde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-epsilon-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl-L-lysine

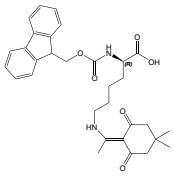
| | |
|---|---|
| CAS-No. | 150629-67-7 |
| Formula | $C_{31}H_{36}N_2O_6$ |
| Mol. weight | 532,64 g/mol |



## FAA1486 Fmoc-D-Lys(Dde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-epsilon-[(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene) ethyl]-D-lysine

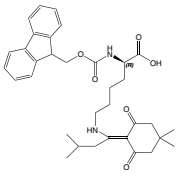
| | |
|---|---|
| CAS-No. | 333973-51-6 |
| Formula | $C_{31}H_{36}N_2O_6$ |
| Mol. weight | 532,64 g/mol |



## FAA1488 Fmoc-D-Lys(ivDde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-epsilon-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)-3-methylbutyl]-D-lysine

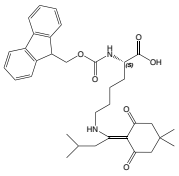
| | |
|---|---|
| CAS-No. | 1272755-33-5 |
| Formula | $C_{34}H_{42}N_2O_6$ |
| Mol. weight | 574,72 g/mol |



## FAA1500 Fmoc-L-Lys(ivDde)-OH

N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-epsilon-[1-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)-3-methylbutyl]-L-lysine

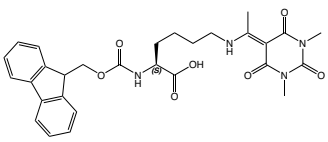
| | |
|---|---|
| CAS-No. | 204777-78-6 |
| Formula | $C_{34}H_{42}N_2O_6$ |
| Mol. weight | 574,72 g/mol |



## FAA8840 Fmoc-L-Lys(MeDmb)-OH

(2S)-6-{[1-(1,3-dimethyl-2,4,6-trioxo-1,3-diazinan-5-ylidene)ethyl]amino}-2-({[(9H-fluoren-9-yl)methoxy]carbonyl}amino)hexanoic acid

| | |
|---|---|
| Formula | $C_{29}H_{32}N_4O_7$ |
| Mol. weight | 548,60 g/mol |



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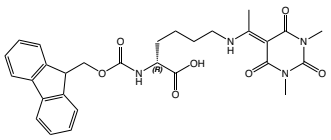
Product details

## FAA8845 Fmoc-D-Lys(MeDmb)-OH

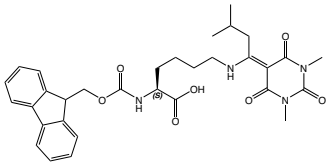
(2R)-6-{[1-(1,3-dimethyl-2,4,6-trioxo-1,3-diazinan-5-ylidene)ethyl]amino}-2-({[(9H-fluoren-9-yl)methoxy]carbonyl}amino)hexanoic acid



| | |
|---|---|
| Formula | $C_{29}H_{32}N_4O_7$ |
| Mol. weight | 548,60 g/mol |

## FAA7975 Fmoc-L-Lys(ivDmb)-OH

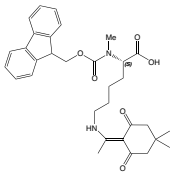
N2-(((9H-fluoren-9-yl)methoxy)carbonyl)-N6-(1-(1,3-dimethyl-2,4,6-trioxotetrahydropyrimidin-5(2H)-ylidene)-3-methylbutyl)-L-lysine



| | |
|---|---|
| Formula | $C_{32}H_{38}N_4O_7$ |
| Mol. weight | 590,68 g/mol |

## FAA1401 Fmoc-L-MeLys(Dde)-OH

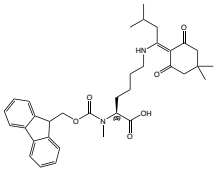
N-alpha-(9-Fluorenylmethyloxycarbonyl)-N-alpha-methyl-N-epsilon-(4,4-dimethyl-2,6-dioxocyclohex-1-ylidene)ethyl-L-lysine



| | |
|---|---|
| CAS-No. | 1428229-84-8 |
| Formula | $C_{32}H_{38}N_2O_6$ |
| Mol. weight | 546,67 g/mol |

## FAA7935 Fmoc-L-MeLys(ivDde)-OH

N2-(((9H-fluoren-9-yl)methoxy)carbonyl)-N6-(1-(4,4-dimethyl-2,6-dioxocyclohexylidene)-3-methylbutyl)-N2-methyl-L-lysine



| | |
|---|---|
| CAS-No. | 1173996-67-2 |
| Formula | $C_{35}H_{44}N_2O_6$ |
| Mol. weight | 588,75 g/mol |

***References:***


- *Synthesis of hydrophobic insulin-based peptides using a helping hand strategy; M. M. Disotuar, M. E. Petersen, J. M. Nogueira, M. S. Kay, D. H. Chou;* ***Org Biomol Chem*** *2019;* ***17****: 1703-1708.* https://doi.org/10.1039/c8ob01212a
- *Chemical synthesis of Shiga toxin subunit B using a next-generation traceless "helping hand" solubilizing tag; J. M. Fulcher, M. E. Petersen,R. J. Giesler, Z. S. Cruz, D. M. Eckert, J. N. Francis, E. M. Kawamoto, M. T. Jacobsen, M. S. Kay;* ***Org Biomol Chem*** *2019;* ***17****: 10237-10244.* https://doi.org/10.1039/c9ob02012h
- *Mapping the Binding Site of BMS-708163 on gamma-Secretase with Cleavable Photoprobes; N. Gertsik, C. W. Am Ende, K. F. Geoghegan, C. Nguyen, P. Mukherjee, S. Mente, U. Seneviratne, D. S. Johnson Y. M. Li;* ***Cell Chem Biol*** *2017;* ***24****: 3-8.* https://doi.org/10.1016/j.chembiol.2016.12.006
- *A Helping Hand to Overcome Solubility Challenges in Chemical Protein Synthesis; M. T. Jacobsen, M. E. Petersen, X. Ye, M. Galibert, G. H. Lorimer, V. Aucagne, M. S. Kay;* ***J Am Chem Soc*** *2016;* ***138****: 11775-82.* https://doi.org/10.1021/jacs.6b05719
- *Compounds and methods for purifying peptides produced by solid phase peptide synthesis; Aucagne V., Delmas A.; CNRS; U.S. Patent No. 9,073,969, 2015*
- *Cleavable trifunctional biotin reagents for protein labelling, capture and release; Y. Yang, S. H. Verhelst;* ***Chem Commun*** *2013;* ***49****: 5366-8.* https://doi.org/10.1039/c3cc42076k
- *Investigation on the stability of the Dde protecting group used in peptide synthesis: migration to an unprotected lysine; K. Augustyns, W. Kraas, G. Jung;* ***J Pept Res*** *1998;* ***51****: 127-33.* https://doi.org/10.1111/j.1399-3011.1998.tb00630.x
- *Full Orthogonality between Dde and Fmoc: The Direct Synthesis of PNA-Peptide Conjugates; J. J. Díaz-Mochón, L. Bialy, M. Bradley;* ***Org. Lett.*** *2004;* ***7****: 1127-1129.* https://doi.org/10.1021/ol049905y
- *Scope and Limitations of Barbituric and Thiobarbituric Amino Acid Derivatives as Protecting Groups for Solid-Phase Peptide Synthesis: Towards a Green Protecting Group; S. Ramkisson, H. H. Al-Rasheed, K. A. Dahlous, B. G. De La Torre, A. El-Faham, F. Albericio;* ***Chem. Select*** *2021;* ***6(26)****: 6626-6630.* https://doi.org/10.1002/slct.202101539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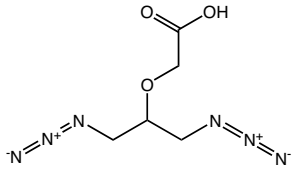
# 4. Trifunctional Linkers

Product details

## AAA2190 DAPOA*DCHA

2-(1,3-diazidopropan-2-yloxy)acetic acid dicyclohexylamine

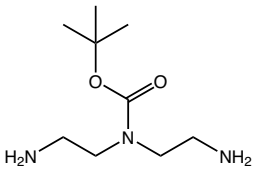
| | |
|---|---|
| CAS-No. | 2389064-43-9 |
| Formula | $C_5H_8N_6O_3$*$C_{12}H_{23}N$ |
| Mol. weight | 200,16*181,32 g/mol |



## BNN1330 DETA(HBH)*2HCl

*tert*-butyl bis(2-aminoethyl)carbamate

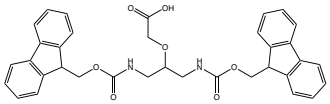
| | |
|---|---|
| CAS-No. | 1914917-65-9 |
| Formula | $C_9H_{21}N_3O_2$*2HCl |
| Mol. weight | 203,29*72,92 g/mol |



## FAA7570 Fmoc2-DAPOA

2-((1,3-bis((9-fluorenylmethyloxycarbonyl)amino)propan-2-yl)oxy)acetic acid

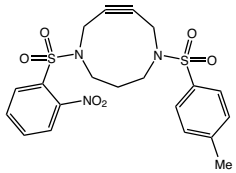
| | |
|---|---|
| CAS-No. | 688350-01-8 |
| Formula | $C_{35}H_{32}N_2O_7$ |
| Mol. weight | 592,64 g/mol |



## RL-2710 DACN(Tos,Ns)

N-(*o*-nitrobenzenesulfonyl)-*N'*-(*p*-toluenesulfonyl)-4,8-diazacyclononyne

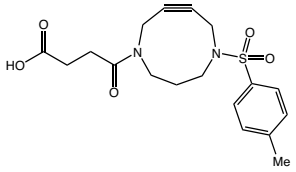
| | |
|---|---|
| CAS-No. | 1797508-58-7 |
| Formula | $C_{20}H_{21}N_3O_6S_2$ |
| Mol. weight | 463,53 g/mol |



## RL-2720 DACN(Tos,Suc-OH)

N-succinoyl-*N'*-(*p*-toluenesulfonyl)-4,8-diazacyclononyne

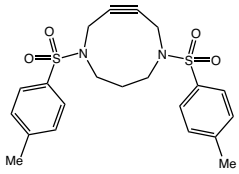
| | |
|---|---|
| CAS-No. | 2109751-68-8 |
| Formula | $C_{18}H_{22}N_2O_5S$ |
| Mol. weight | 378,44 g/mol |



## RL-2730 DACN(Tos$_2$)

*N,N'*-bis(*p*-toluenesulfonyl)-4,8-diazacyclononyne

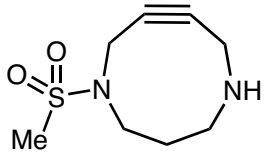
CAS-No. 1797508-57-6
Formula $C_{21}H_{24}N_2O_4S_2$
Mol. weight 432,56 g/mol



## RL-3600 DACN(Ms)*HCl

N-(Mesyl)-4,8-diazacyclononyne hydrochloride

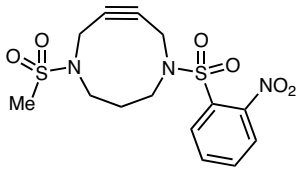
CAS-No. 2331322-16-6
Formula $C_8H_{14}N_2O_2S$*HCl
Mol. weight 202,27*36,46 g/mol



## RL-3610 DACN(Ms,Ns)

N-(Mesyl)-*N'*-(2-nosyl)-4,8-diazacyclononyne

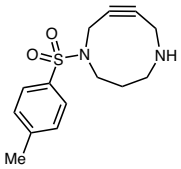
CAS-No. 2411082-25-0
Formula $C_{14}H_{17}N_3O_6S_2$
Mol. weight 387,43 g/mol



## RL-2735 DACN(Tos)*HCl

N-(*p*-toluenesulfonyl)-4,8-diazacyclononyne hydrochloride

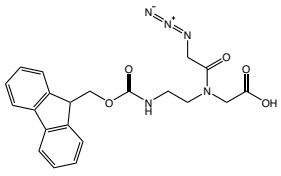
CAS-No. 2331322-18-8
Formula $C_{14}H_{18}N_2O_2S$*HCl
Mol. weight 278,37*36,46 g/mol



## HAA9330 $N_3$-Gly-Aeg(Fmoc)-OH

N-(2-((((9H-fluoren-9-yl)methoxy)carbonyl)amino) ethyl)-N-(2-azidoacetyl)glycine

CAS-No. 2606227-07-8
Formula $C_{21}H_{21}N_5O_5$
Mol. weight 423,43 g/mol



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# 5. Cross-Linkers for other Bio Applications

## 5.1. Substrates for Fusion (Halo/Snap/Clip)-Tagged Proteins

Site-specific protein labeling is a versatile tool for studying protein function and interaction in living cells. Various peptide-based protein tags have been developed. Herein, we are focusing on HaloTag®, SNAP-Tag® and CLIP-Tag™, as well as corresponding substrates offered by Iris Biotech. Those labeling systems enable the specific, covalent attachment of in principle any molecule of choice to a protein of interest.

The HaloTag® (*Fig. 28*) is a 33 kDa self-labeling protein tag derived from the haloalkane dehalogenase DhaA from *Rhodococcus rhodochrous*. Its active site reacts in a nucleophilic attack with chloroalkane linker substrates to form an irreversible bond in the case of the mutated enzyme. The chloroalkane linker can easily be functionalized with a label of choice, e.g., with a fluorophore or biotin. For the wild-type enzyme, this intermediate would be hydrolyzed, leading to the regeneration of the enzyme.

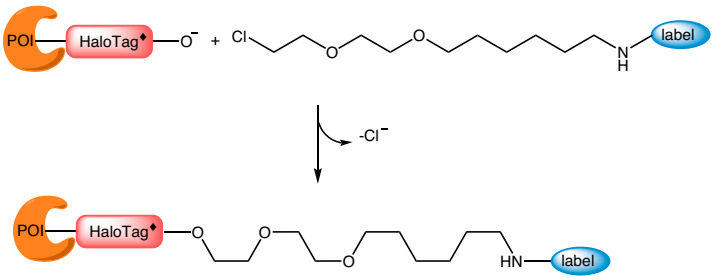


*Fig. 28: Schematic illustration of the HaloTag® reaction.*

The SNAP-tag® (*Fig. 29*) is a 20 kDa self-labeling protein tag based on a modified form of the human O6-alkyl-guanine-DNA-alkyltransferase (hAGT), a DNA repair enzyme. A cysteine residue within the SNAP-tag® undergoes an irreversible reaction with synthetic O6-benzylguanine (BG) derivatives, resulting in a covalent thioether bond. The BG moiety can easily be further functionalized with a label of choice, e.g., fluorophore, biotin, generally without affecting the reaction of the substrate with the SNAP-tag®.

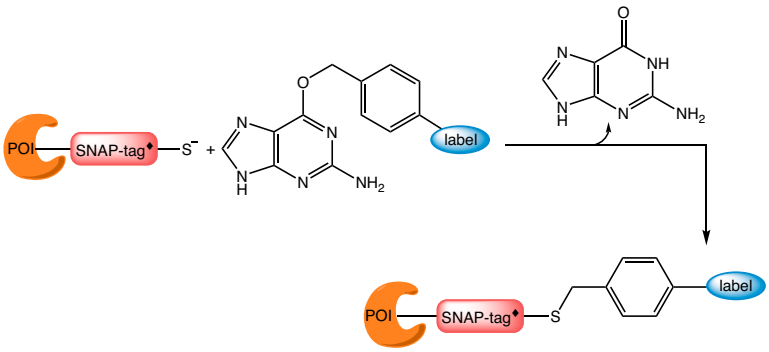


*Fig. 29: Schematic illustration of the SNAP-tag® reaction.*

The CLIP-tag™ (*Fig. 30*) (20 kDa) is a modified version of the SNAP-tag, engineered to react with benzylcytosine (BC) instead of benzylguanine (BG). Thus, properties are similar. CLIP-tag™- and SNAP-tag®-fused proteins can be labeled simultaneously in the same cells.

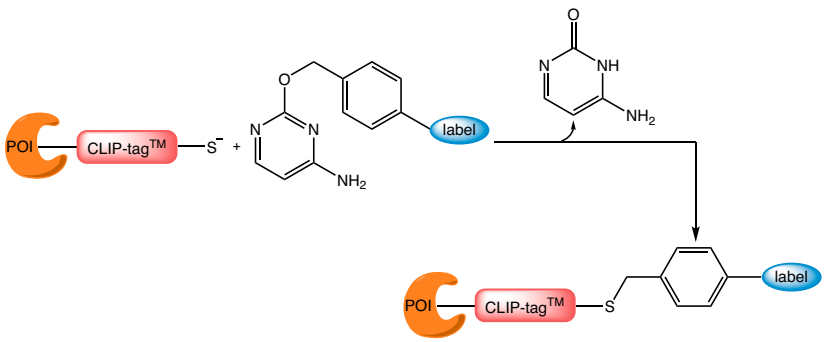


*Fig. 30: Schematic illustration of the CLIP-tag™ reaction.*

*Tab. 2: Table summarizing main properties of HaloTag®, SNAP-tag® and CLIP-tag™.*

| | HaloTag | SNAP-tag | CLIP-tag |
|---|---|---|---|
| Origin | Haloalkane dehalogenase *(Rhodococcus rhodochrous)* | Human O6-alkylguanine-DNA-alkyltransferase | Human O6-alkylguanine-DNA-alkyltransferase |
| Reactivity | Chloroalkane derivatives | O6-benzylguanine derivatives | Benzylcytosine derivatives |
| Length | 297 amino acids | 182 amino acids | 182 amino acids |
| Molecular Weight | 33.6 kDa | 19.4 kDa | 19.4 kDa |

Iris Biotech offers a biotin- (*RL-3860 on page 108*) as well as an ICG-functionalized (*RL-3830 on page 108*) SNAP-tag® substrate, as well as the corresponding CLIP-tag™ suitable (*RL-3840 on page 106*, *RL-3870 on page 106*) derivatives. Biotinylated proteins can for example be selectively isolated based on the high affinity towards avidin representing a useful tool for purification, immobilization, and labeling. Indocyanine green (ICG) is a near-infrared fluorescence imaging dye (absorption maximum 800 nm + slight absorption in the visible range; emission maximum 810 nm) approved by the FDA.

As substrates for the HaloTag® various products are offered, e.g., suitable for further functionalization *via* Chlick chemistry.





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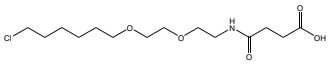
Product details

## RL-3180 Halo-PEG(2)-Suc

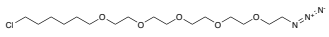
4-((2-(2-((6-chlorohexyl)oxy)ethoxy)ethyl) amino)-4-oxobutanoic acid



| | |
|---|---|
| CAS-No. | 1488363-39-8 |
| Formula | $C_{14}H_{26}ClNO_5$ |
| Mol. weight | 323,81 g/mol |

## RL-3640 Halo-PEG(5)-azide

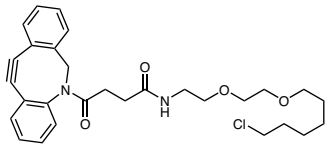
1-azido-21-chloro-3,6,9,12,15-pentaoxahenicosane



| | |
|---|---|
| CAS-No. | 1261238-21-4 |
| Formula | $C_{16}H_{32}ClN_3O_5$ |
| Mol. weight | 381,90 g/mol |

## RL-3670 Halo-DBCO

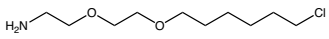
N-[2-[2-[(6-chlorohexyl)oxy]ethoxy]ethyl]-gamma-oxo-dibenz[b,f]azocine-5(6H)-butanamide



| | |
|---|---|
| CAS-No. | 1808119-16-5 |
| Formula | $C_{29}H_{35}ClN_2O_4$ |
| Mol. weight | 511,06 g/mol |

## RL-3680 Halo-PEG(2)-$NH_2$*HCl

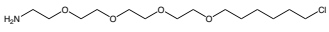
12-Chloro-3,6-dioxa-dodecan-1-amine hydrochloride



| | |
|---|---|
| CAS-No. | 1035373-85-3 |
| Formula | $C_{10}H_{22}ClNO_2$*HCl |
| Mol. weight | 223,74*36,46 g/mol |

## RL-3690 Halo-PEG(4)-$NH_2$*HCl

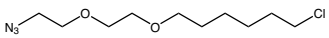
18-Chloro-3,6,9,12-tetraoxa-octadecan-1-amine hydrochloride



| | |
|---|---|
| CAS-No. | 1261238-20-3 |
| Formula | $C_{14}H_{30}ClNO_4$*HCl |
| Mol. weight | 311,85*36,46 g/mol |

## RL-3700 Halo-PEG(2)-Azide

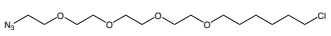
1-Azido-12-chloro-3,6-dioxadodecane



| | |
|---|---|
| CAS-No. | 2568146-55-2 |
| Formula | $C_{10}H_{20}ClN_3O_2$ |
| Mol. weight | 249,74 g/mol |

## RL-3710 Halo-PEG(4)-Azide

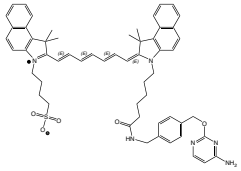
1-Azido-18-chloro-3,6,9,12-tetraoxaoctadecane



CAS-No. 2989398-83-4
Formula $C_{14}H_{28}ClN_3O_4$
Mol. weight 337,85 g/mol

## RL-3840 ICG-CLIP

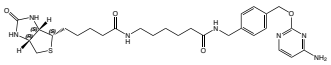
4-(2-((1E,3E,5E,7E)-7-(3-(6-((4-(((4-aminopyrimidin-2-yl) oxy)methyl)benzyl)amino)-6-oxohexyl)-1,1-dimethyl-1,3-dihydro-2H-benzo[e]indol-2-ylidene)hepta-1,3,5-trien-1-yl)-1,1-dimethyl-1H-benzo[e]indo l-3-ium-3-yl) butane-1-sulfonate



Formula $C_{57}H_{62}N_6O_5S$
Mol. weight 943,22 g/mol

## RL-3870 Biotin-Clip

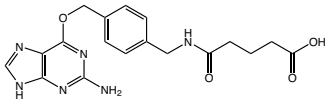
N-(4-(((4-aminopyrimidin-2-yl)oxy)methyl)benzyl)-6-(5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d] imidazol-4-yl)pentanamido)hexanamide



CAS-No. 1004524-73-5
Formula $C_{28}H_{39}N_7O_4S$
Mol. weight 569,73 g/mol

## RL-3835 SNAP-acid

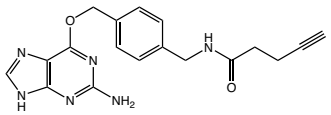
5-((4-(((2-amino-9H-purin-6-yl)oxy)methyl)benzyl) amino)-5-oxopentanoic acid



CAS-No. 881663-34-9
Formula $C_{18}H_{20}N_6O_4$
Mol. weight 384,40 g/mol

## RL-3930 Alkyne-SNAP

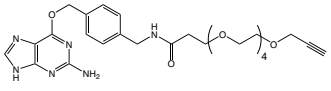
N-(4-(((2-amino-9H-purin-6-yl)oxy)methyl)benzyl) pent-4-ynamide



CAS-No. 1104822-07-2
Formula $C_{18}H_{18}N_6O_2$
Mol. weight 350,38 g/mol

## RL-3940 Alkyne-PEG(5)-SNAP

N-(4-(((2-amino-9H-purin-6-yl)oxy)methyl)benzy-l)-4,7,10,13,16-pentaoxanonadec-18-ynamide



Formula $C_{27}H_{36}N_6O_7$
Mol. weight 556,62 g/mol

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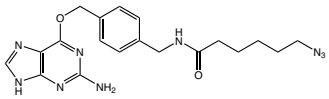
Product details

## RL-3950 Azide-SNAP

N-(4-(((2-amino-9H-purin-6-yl)oxy)methyl)benzyl)-6-azidohexanamide

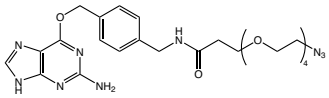
Formula $C_{19}H_{23}N_9O_2$
Mol. weight 409,45 g/mol



## RL-3960 Azide-PEG(4)-SNAP

N-(4-(((2-amino-9H-purin-6-yl)oxy)methyl)benzyl)-1-azido-3,5,7,9-tetraoxadodecan-12-amide

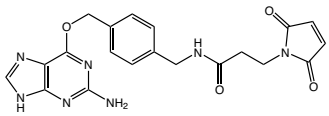
Formula $C_{24}H_{33}N_9O_6$
Mol. weight 543,59 g/mol



## RL-3970 Mal-SNAP

N-(4-(((2-amino-9H-purin-6-yl)oxy)methyl)benzyl)-3-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)propanamide

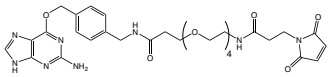
Formula $C_{20}H_{19}N_7O_4$
Mol. weight 421,42 g/mol



## RL-3980 Mal-PEG(4)-SNAP

N-(4-(((2-amino-9H-purin-6-yl)oxy)methyl)benzyl)-1-(3-(2,5-dioxo-2,5-dihydro-1H-pyrrol-1-yl)propanamido)-3,6,9,12-tetraoxapentadecan-15-amide

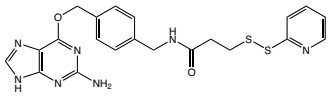
CAS-No. 1151762-31-0
Formula $C_{31}H_{40}N_8O_9$
Mol. weight 668,71 g/mol



## RL-3990 OPSS-SNAP

N-(4-(((2-amino-9H-purin-6-yl)oxy)methyl)benzyl)-3-(pyridin-2-yldisulfaneyl)propanamide

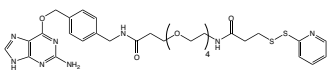
Formula $C_{21}H_{21}N_7O_2S_2$
Mol. weight 467,57 g/mol



## RL-4000 OPSS-PEG(4)-SNAP

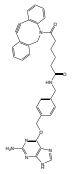
N-(4-(((2-amino-9H-purin-6-yl)oxy)methyl)benzyl)-1-(3-(pyridin-2-yldisulfaneyl)propanamido)-3,6,9,12-tetraoxapentadecan-15-amide

CAS-No. 2278175-10-1
Formula $C_{32}H_{42}N_8O_7S_2$
Mol. weight 714,86 g/mol



## RL-4010 DBCO-SNAP

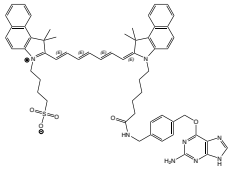
Formula $C_{34}H_{31}N_7O_3$
Mol. weight 585,67 g/mol



## RL-3830 ICG-SNAP

4-(2-((1E,3E,5E,7E)-7-(3-(6-((4-(((2-amino-9H-purin-6-yl) oxy)methyl)benzyl)amino)-6-oxohexyl)-1,1-dimethyl-1,3-dihydro-2H-benzo[e]indol-2-ylidene)hepta-1,3,5-trien-1-yl)-1,1-dimethyl-1H-benzo[e]indo l-3-ium-3-yl) butane-1-sulfonate

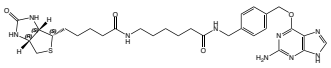
Formula $C_{58}H_{62}N_8O_5S$
Mol. weight 983,25 g/mol



## RL-3860 Biotin-SNAP

N-(4-(((2-amino-9H-purin-6-yl)oxy)methyl)benzyl)-6-(5-((3aS,4S,6aR)-2-oxohexahydro-1H-thieno[3,4-d] imidazol-4-yl)pentanamido)hexanamide

CAS-No. 471918-16-8
Formula $C_{29}H_{39}N_9O_4S$
Mol. weight 609,75 g/mol



***References:***


- *Site-specific protein labeling with SNAP-Tags; N. B. Cole;* ***Curr Protoc Protein Sci.*** *2013;* ***73(30)****: 1-30. https://doi.org/10.1002/0471140864.ps3001s73*
- *A general method for the covalent labeling of fusion proteins with small molecules in vivo; A. Keppler, S. Gendrezig, T. Gronemeyer, H. Pick, H. Vogel, K. Johnsson;* ***Nat. Biotechnol.*** *2003;* ***21****: 86-89. https://doi.org/10.1038/nbt765*
- *HaloTag: A Novel Protein Labeling Technology for Cell Imaging and Protein Analysis; g: v: Los; L. P. Encell, M. G. McDougall, D. D. Hartzell, N. Karassina, C. Zimprich, M. G. Wood, R. Learish, R. F. Ohana, M. Urh, D. Simpson, J. Mendez, K. Zimmerman, P. Otto, G. Vidugiris, J. Zhu, A. Darzins, D. H. Klaubert, R. F. Bulleit, K. V. Wood;* ***ACS Chem. Biol.*** *2008;* ***3(6)****: 373-382. https://doi.org/10.1021/cb800025k*
- *Directed evolution of O6-alkylguanine-DNA alkyltransferase for efficient labeling of fusion proteins with small molecules in vivo; A. Juillerat, T. Gronemeyer, A. Keppler, S. Gendreizig, H. Pick, H. Vogel, K. Johnsson;* ***Chem. Biol.*** *2003;* ***10(4)****: 313-317. https://doi.org/10.1016/s1074-5521(03)00068-1*
- *Site-specific, Covalent Labeling of Recombinant Antibody Fragments via Fusion to an Engineered Version of 6-O-Alkylguanine DNA Alkyltransferase; F. Kampmeier, M. Ribbert, T. Nachreiner, S. Dembski, F. Beaufils, A. Brecht, S. Barth;* ***Bioconjugate Chem.*** *2009;* ***20(5)****: 1010-1015. https://doi.org/10.1021/bc9000257*
- *SNAP-Tag Technology: A Useful Tool to Determine Affinity Constants and Other Functional Parameters of Novel Antibody Fragments; J. Niesen, M. Sack, M. Seidel, R. Fendel, S. Barth, R. Fischer, C. Stein;* ***Bioconjugate Chem.*** *2016;* ***27(8)****: 1931-1941. https://doi.org/10.1021/acs.bioconjchem.6b00315*
- *The clinical use of indocyanine green as a near-infrared fluorescent contrast agent for image-guided oncologic surgery. Boudewijn E. Schaafsma MD, J. Sven D. Mieog MD, Merlijn Hutteman MSc, Joost R. van der Vorst MD, Peter J.K. Kuppen PhD, Clemens W.G.M. Lwik PhD, John V. Frangioni MD, PhD, Cornelis J.H. van de Velde MD, PhD, Alexander L. Vahrmeijer MD PhD;* ***Surg. Oncol.*** *2011;* ***104****: 323-332. https://doi.org/10.1002/jso.21943*
- *Degradation kinetics of indocyanine green in aqueous solution. Vishal Saxena, Mostafa Sadoqi, Jun Shao;* ***J. Pharm. Sci.*** *2003;* ***92****: 2090-2097. https://doi.org/10.1002/jps.10470*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- *Stability assessment of indocyanine green within dextran-coated mesocapsules by absor- bance spectroscopy. Mohammad Abbas Yaseen; Jie Yu; Michael S. Wong; Bahman Anvari;* ***Journal of Biomedical Optics*** *2007;* ***12(6)****: 064031.* https://doi.org/10.1117/1.2821423
- *Imaging proteins inside cells with fluorescent tags; G. Crivat, J. W. Taraska;* ***Trends Biotechnol*** *2012;* ***30(1)****: 8-16.* https://doi.org/10.1016/j.tibtech.2011.08.002
- *Visualizing Biochemical Activities in Living Cells through Chemistry; L. Reymond, G. Lukinavicius, K. Umezawa, D. Maurel, M. A. Brun, A. Masharina, K. Bojkowska, B. Mollwitz, A. Schena, R. Griss, K. Johnsson;* ***CHIMICA Int. J. Chem.*** *2011;* ***65(11)****: 868-871.* https://doi.org/10.2533/chimia.2011.868.
- *Cell Penetration Profiling Using the Chloroalkane Penetration Assay; L. Peraro, K. L. Deprey, M. K. Moser, Z. Zou, H. L. Ball, B. Levine, J. A. Kritzer;* ***J. Am. Chem. Soc.*** *2018;* ***140(36)****: 11360-11369.* https://doi.org/10.1021/jacs.8b06144
- *HaloTag technology: a versatile platform for biomedical applications; C. G. England, H. Luo, W. Cai;* ***Bioconjug Chem*** *2015;* ***26****: 975-86.* https://doi.org/10.1021/acs.bioconjchem.5b00191
- *HaloTag: a novel protein labeling technology for cell imaging and protein analysis; G. V. Los, L. P. Encell, M. G. McDougall, D. D. Hartzell, N. Karassina, C. Zimprich, M. G. Wood, R. Learish, R. F. Ohana, M. Urh, D. Simpson, J. Mendez, K. Zimmerman, P. Otto, G. Vidugiris, J. Zhu, A. Darzins, D. H. Klaubert, R. F. Bulleit, K. V. Wood;* ***ACS Chem Biol*** *2008;* ***3****: 373-382.* https://doi.org/10.1021/cb800025k
- *Self-labelling enzymes as universal tags for fluorescence microscopy, super-resolution microscopy and electron microscopy; V. Liss, B. Barlag, M. Nietschke, M. Hensel;* ***Scientific Reports*** *2016; 5.* https://doi.org/10.1038/srep17740
- *Snap-, CLIP- and Halo-Tag Labelling of Budding Yeast Cells; F. Stagge, G. Y. Mitronova, V. N. Belov, C. A. Wurm, S. Jakobs;* ***PLoS ONE 8(10):*** *e78745.* https://doi.org/10.1371/journal.pone.0078745


## 5.2. Specific His Tag Acylation

Gly-His tag for selective α-amine acylation:


Lys-His tag for selective ε-amine acylation:

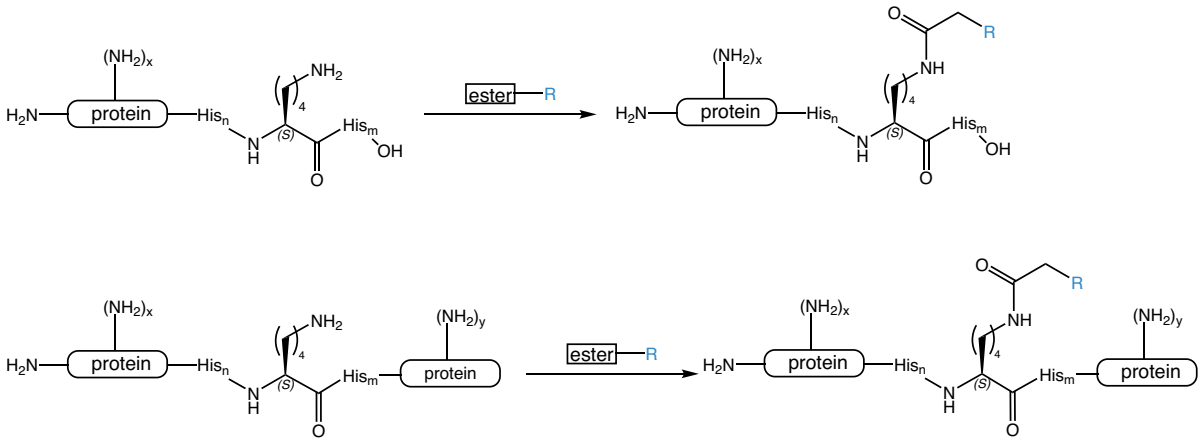


*Fig. 31: Schematic illustration of Gly-His tag acylation and Lys-His tag acylation. Functionalized esters can be used for the introduction of the desired moieties, e.g., azides, biotin.*

A frequently used tag for protein modification is the so-called His tag. His tags, or polyhistidine tags, comprise a consecutive series of six to ten histidine residues. His tags are widely used for protein purification by immobilized-metal ion affinity chromatography, allowing to extract a protein of interest from thousands of other proteins present in a cell lysate. However, as an inadvertent side-reaction, it is reported that His tagged proteins can undergo N-terminal acylation with gluconolactone (GDL). The discovery of this reaction by Geoghegan and coworkers triggered the inspiration for the development of His tag-based peptide segments for selective acylations. Thus, Jensen *et al*. developed two methods that use poly-histidine sequences to direct the highly selective acylation of proteins, either at the N-terminus or at a specific lysine residue.

The highly selective and efficient N-terminal acylation of proteins is based on a Gly-$His_6$ segment (Gly-His tag) complemented by the use of 4-methoxy phenyl esters as finely tuned acylating agents, resulting in stable conjugates. Other acylating agents, e.g., lactons, thioesters, N-hydroxysuccinimide ester were also tested but gave only limited N-terminal acylation or low selectivity.

**General Conjugation Protocol:**

A 35 µM solution of GH6-protein in 200 mM HEPES buffer at pH 7.5 is incubated with 40 equiv. of azido-acetyl 4-methoxyphenylester for 24 h at 4 °C. The formation of the mono-functionalized product can be observed by ESI-MS and can reach 70% to 90% conversion. A higher conversion rate can be achieved by the addition of two aliquots of 10 equiv. of the acylating agent in the course of the next 48 h.

For the acylation of lysine, Jensen *et al*. developed the peptide sequence $His_n$-Lys-$His_m$ (Lys-His tag) that directs the acylation of the designated lysine N-ε amine under mild conditions and with high selectivity over native lysine residues.

Both methods provide highly selective acylation, yield stable conjugates, and do not require the use of metal ions. In detail, referring to literature, yields for the protein modification with Gly-His are typically 60–80% mono-acylation with 1–5% over-acylation, while for Lys-His it is typically 50–70% monoacylation with 1–8% over-acylation. The Gly-His tag as well as the Lys-His tag maintain the capacity for immobilized metal ion affinity chromatography and have been shown robust for the attachment of azides, fluorophores, and biotin to different proteins, including antibodies.

Mechanistic studies indicate that the very high selectivity of the His tag acylation is based on specific base catalysis, in which a histidine side-chain assists deprotonation during the direct acylation of the glycine α-amine (*Fig. 32*). The ester preferentially reacts with assistance from histidine side-chain imidazoles since they are not protonated ($pK_a$ ~ 6.0) at the pH of the reaction, in contrast to the N-terminal α-amine ($pK_a$ ~ 7.6-8.0) and lysine side-chains ($pK_a$ ~ 10.5). The presence of the additional five His residues in the His tag may serve to modulate the basicity of the imidazole nitrogen of the catalytic residue. A recent study has shown that the $pK_a$ values of individual histidine side-chains in a $His_6$-tag span a range from 4.8-7.5.

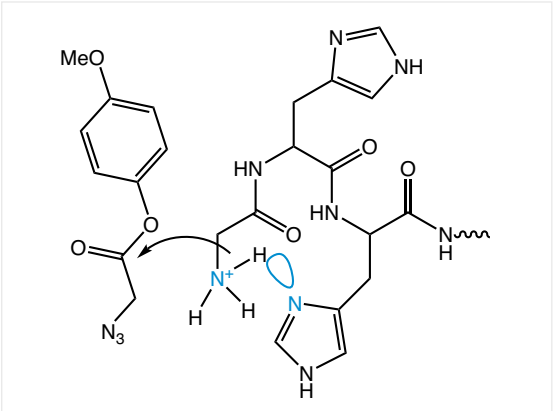


*Fig. 32: Imidazole rings of neighboring histidines in a His tag catalyze the acylation of a glycyl N-terminus via a base catalyzed mechanism.*

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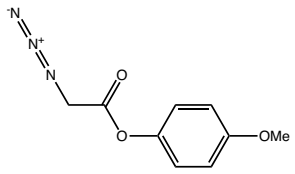
Product details

## RL-3010 $N_3$Ac-OPhOMe

4-Methoxyphenyl 2-azidoacetate

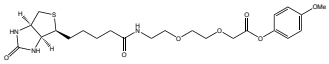
| | |
|---|---|
| CAS-No. | 2546513-31-7 |
| Formula | $C_9H_9N_3O_3$ |
| Mol. weight | 207,19 g/mol |




## RL-3100 Biotin-AEEA-OPhOMe

2-(2-(2-(Biotinamido)ethoxy)ethoxy)acetic acid 4-methoxyphenyl ester

| | |
|---|---|
| CAS-No. | 2546513-67-9 |
| Formula | $C_{23}H_{33}N_3O_7S$ |
| Mol. weight | 495,59 g/mol |



***References:***


- *Selective Acylation of Proteins at Gly and Lys in His tags; K. J. Jensen, M. B. Thygesen, K. K. Sørensen, S. Wu, T. Treiberg, S. Schoffelen;* ***ChemBioChem*** *2022.* https://doi.org/10.1002/cbic.202200359
- *Site-specific covalent labeling of His tag fused proteins with N-acyl-N-alkyl sulfonamide reagent; V. Thimaradka, J. H. Oh, C. Heroven, A. R. Aricescu, M. Yuzaki, T. Tamura, I. Hamachi;* ***Bioorg. Med. Chem.*** *2021;* ***15(30)****: 115947.* https://doi.org/10.1016/j.bmc.2020.115947
- *Selective N-terminal acylation of peptides and proteins with a Gly-His tag sequence; M. C. Martos-Maldonado, C. T. Hjuler, K. K. Sorensen, M. B. Thygesen, J. E. Rasmussen, K. Villadsen, S. R. Midtgaard, S. Kol, S. Schoffelen, K. J. Jensen;* ***Nat Commun*** *2018;* ***9****: 3307.* https://doi.org/10.1038/s41467-018-05695-3
- *Spontaneous alpha-N-6-phosphogluconoylation of a "His tag" in Escherichia coli: the cause of extra mass of 258 or 178 Da in fusion proteins; K. F. Geohegan, H. B. Dixon, P. J. Rosner, L. R. Hoth, A. J. Lanzetti,K. A. Borzilleri, E. S. Marr, L. H. Pezzullo, L. B. Martin, P. K. LeMotte, A. S. McCol, A. V. Kamath, J. G. Stroh;* ***Anal Biochem.*** *1999;* ***267(1)****: 169-84.* https://doi.org/10.1006/abio.1998.2990.


**You are interested in autocatalytic His tags for the chemical modification of proteins?**

**Watch the recording of our workshop!**

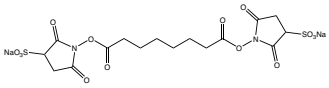
## 5.3. Bifunctional Protein Cross-Linkage

Product details

**RL-2770** **BSSS**

**Bis(sulfosuccinimidyl) suberate sodium salt**

| | |
|---|---|
| CAS-No. | 127634-19-9 |
| Formula | $C_{16}H_{18}N_2Na_2O_{14}S_2$ |
| Mol. weight | 572,43 g/mol |



This molecule (*Fig. 33*) carries amino reactive sulfo-NHS esters on both ends and is a water-soluble, homo-bi functionalprotein cross-linker (spacer length: 11.4 Å). Due to its water solubility, conjugation reactions can conveniently take place at physiological conditions. This 8-atom spacer is non-cleavable and the molecule is not cell membrane permeable. It can be used to prepare antibody-protein conjugates, for crosslinking cell surface proteins, and for covalently binding an antibody to an immobilized Protein A or Protein G resin.

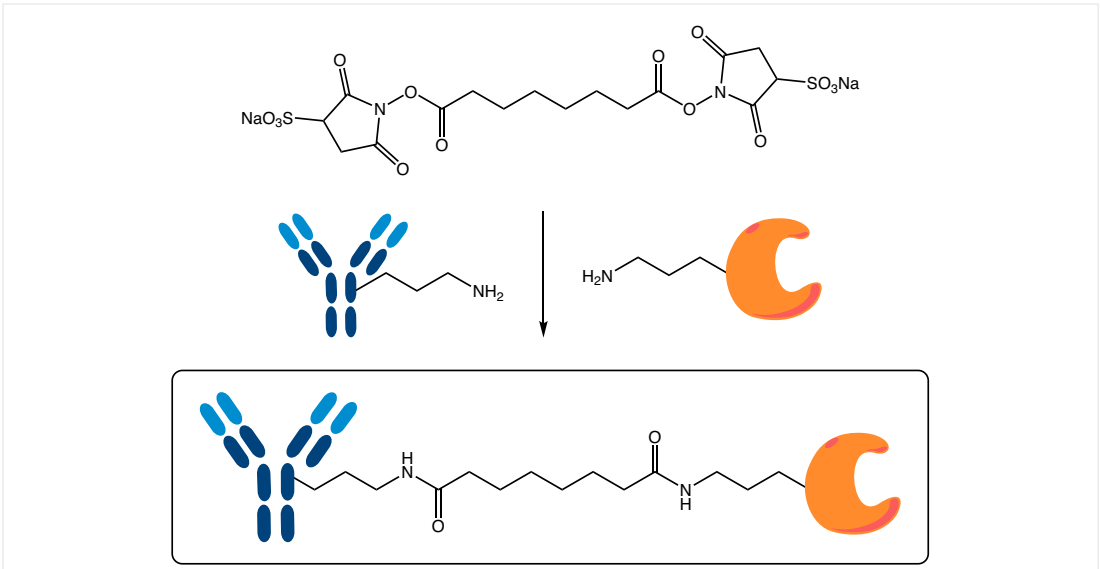



*Fig. 33: BSSS can be used for cross-linkage of different biomolecules.*

The Concept of Antibody-Drug Conjugation (ADC)
Permanent Linkers
Cleavable Linkers
Trifunctional Linkers
Cross-Linkers for other Bio Applications
Preparing Carriers for Conjugation
Index

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**General BSSS Cross-Linking Protocol:**

1. Allow vial of BSSS to fully equilibrate to ambient temperature before opening to prevent condensation inside the vial (BSSS is moisture-sensitive).
2. Immediately before use, prepare a 50 mM solution of BSSS, by dissolving 10 mg BSSS in 350 mL of 25 mM sodium phosphate, pH 7.4 (do not use amine containing buffers for the conjugation reaction).
3. Add BSSS solution (20-fold excess cross-linker to protein) to the protein sample so that the final concentration is between 0.5 to 5 mM.
4. Allow the sample to react at room temperature for 45 minutes to 1 hour. Allow slightly longer if reaction must be done on ice (the reaction rate is only slightly slower at low temperatures).
5. Quench any unreacted BSSS with 25 mM to 60 mM Tris and allow to react for 10-15 minutes at room temperature.
6. Desalt sample to remove unreacted BSSS, i.e., by gel filtration, dialysis, etc.

***References:***


→ *Isotope-tagged cross-linking reagents. A new tool in mass spectrometric protein interaction analysis; D. R. Muller, P. Schindler, H. Towbin, U. Wirth, H. Voshol, S. Hoving, M. O. Steinmetz;* ***Anal Chem*** *2001;* ***73****: 1927-34. https://doi.org/10.1021/ac001379a*

→ *The Golgi-associated hook3 protein is a member of a novel family of microtubule-binding proteins; J. H. Walenta, A. J. Didier, X. Liu, H. Kramer;* ***J Cell Biol*** *2001;* ***152****: 923-34. https://doi.org/10.1083/jcb.152.5.923*

→ *Rapamycin potentiates transforming growth factor beta-induced growth arrest in nontransformed, oncogene-transformed, and human cancer cells; B. K. Law, A. Chytil, N. Dumont, E. G. Hamilton, M. E. Waltner-Law, M. E. Aakre, C. Covington, H. L. Moses;* ***Mol Cell Biol*** *2002;* ***22****: 8184-98. https://doi.org/10.1128/mcb.22.23.8184-8198.2002*

→ *Chemical cross-linking and mass spectrometry for mapping three-dimensional structures of proteins and protein complexes; A. Sinz;* ***J Mass Spectrom*** *2003;* ***38****: 1225-37. https://doi.org/10.1002/jms.559*

→ *Mapping low-resolution three-dimensional protein structures using chemical cross-linking and Fourier transform ion-cyclotron resonance mass spectrometry; G. H. Dihazi, A. Sinz;* ***Rapid Commun Mass Spectrom*** *2003;* ***17****: 2005-14. https://doi.org/10.1002/rcm.1144*

→ *Mapping the topology and determination of a low-resolution three-dimensional structure of the calmodulin-melittin complex by chemical cross-linking and high-resolution FTICRMS: direct demonstrationof multiple binding modes; D. M. Schulz, C. Ihling, G. M. Clore, A. Sinz;* ***Biochemistry*** *2004;* ***43****: 4703-15. https://doi.org/10.1021/bi036149f*

→ *Selective inactivation of adrenomedullin over calcitonin gene-related peptide receptor function by the deletion of amino acids 14-20 of the mouse calcitonin-like receptor; D. Koller, L. M. Ittner, R. Muff, K. Husmann, J. A. Fischer, W. Born;* ***J Biol Chem*** *2004;* ***279****: 20387-91. https://doi.org/10.1074/jbc.M313058200*

→ *Mactinin, a fragment of cytoskeletal alpha-actinin, is a novel inducer of heat shock protein (Hsp)-90 mediated monocyte activation; S. D. Luikart, A. Panoskaltsis-Mortari, T. Hinkel, R. T. Perri, K. Gupta, T. R. Oegema, P. Gupta;* ***BMC Cell Biol*** *2009;* ***10****: 60. https://doi.org/10.1186/1471-2121-10-60*

→ *The program for processing newly synthesized histones H3.1 and H4; E. I. Campos, J. Fillingham, G. Li, H. Zheng, P. Voigt, W. H. Kuo, H. Seepany, Z. Gao, L. A. Day, J. F. Greenblatt, D. Reinberg;* ***Nat Struct Mol Biol*** *2010;* ***17****: 1343-51. https://doi.org/10.1038/nsmb.1911*

→ *Sar1 assembly regulates membrane constriction and ER export; K. R. Long, Y. Yamamoto, A. L. Baker, S. C. Watkins, C. B. Coyne, J. F. Conway, M. Aridor;* ***J Cell Biol*** *2010;* ***190****: 115-28. https://doi.org/10.1083/jcb.201004132*

→ *Mps1 directs the assembly of Cdc20 inhibitory complexes during interphase and mitosis to control M phase timing and spindle checkpoint signaling; J. Maciejowski, K. A. George, M. E. Terret, C. Zhang, K. M. Shokat, P. V. Jallepalli;* ***J Cell Biol*** *2010;* ***190****: 89-100. https://doi.org/10.1083/jcb.201001050*

→ *Protection against protein aggregation by alpha-crystallin as a mechanism of preconditioning; J. E. Ferns, C. S. Theisen, E. E. Fibuch, N. W. Seidler;* ***Neurochem Res*** *2012;* ***37****: 244-52. https://doi.org/10.1007/s11064-011-0601-4*

→ *Identification of IGPR-1 as a novel adhesion molecule involved in angiogenesis; N. Rahimi, K. Rezazadeh, J. E. Mahoney, E. Hartsough, R. D. Meyer;* ***Mol Biol Cell*** *2012;* ***23****: 1646-56. https://doi.org/10.1091/mbc.E11-11-0934*

→ *TWEAK-independent Fn14 self-association and NF-kappaB activation is mediated by the C-terminal region of the Fn14 cytoplasmic domain; S. A. Brown, E. Cheng, M. S. Williams, J. A. Winkles;* ***PLoS One*** *2013;* ***8****: e65248. https://doi.org/10.1371/journal.pone.0065248*

- *ZRF1 controls the retinoic acid pathway and regulates leukemogenic potential in acute myeloid leukemia; S. Demajo, I. Uribesalgo, A. Gutierrez, C. Ballare, S. Capdevila, M. Roth, J. Zuber, J. Martin-Caballero, L. Di Croce;* ***Oncogene*** *2014;* ***33****: 5501-10.* https://doi.org/10.1038/onc.2013.501
- *Ubiquitin Associates with the N-Terminal Domain of Nerve Growth Factor: The Role of Copper(II) Ions; V. Lanza, A. Travaglia, G. Malgieri, R. Fattorusso, G. Grasso, G. Di Natale, V. Zito, G. Arena, D. Milardi, E. Rizzarelli;* ***Chemistry*** *2016;* ***22****: 17767-17775.* https://doi.org/10.1002/chem.201603650
- *Potentiation of Surface Stability of AMPA Receptors by Sulfhydryl Compounds: A Redox-Independent Effect by Disrupting Palmitoylation; J. Han, H. Zhang, S. Wang, J. Zhou, Y. Luo, L. H. Long, Z. L. Hu, F. Wang, J. G. Chen, P. F. Wu;* ***Neurochem Res*** *2016;* ***41****: 2890-2903.* https://doi.org/10.1007/s11064-016-2006-x
- *Dramatic Domain Rearrangements of the Cyanobacterial Orange Carotenoid Protein upon Photoactivation; H. Liu, H. Zhang, G. S. Orf, Y. Lu, J. Jiang, J. D. King, N. R. Wolf, M. L. Gross, R. E. Blankenship;* ***Biochemistry*** *2016;* ***55****: 1003-9.* https://doi.org/10.1021/acs.biochem.6b00013
- *Nuclear Speckle-related Protein 70 Binds to Serine/Arginine-rich Splicing Factors 1 and 2 via an Arginine/Serine-like Region and Counteracts Their Alternative Splicing Activity; C. H. Kim, Y. D. Kim, E. K. Choi, H. R. Kim, B. R. Na, S. H. Im, C. D. Jun;* ***J Biol Chem*** *2016;* ***291****: 6169-81.* https://doi.org/10.1074/jbc.M115.689414
- *PGL germ granule assembly protein is a base-specific, single-stranded RNase; S. T. Aoki, A. M. Kershner, C. A. Bingman, M. Wickens, J. Kimble;* ***Proc Natl Acad Sci U S A*** *2016;* ***113****: 1279-84.* https://doi.org/10.1073/pnas.1524400113


## 5.4. Triazolecarbaldehydes for Selective *N*-Terminal Protein Modification

The selective modification of the N-terminus (i.e., an α-amino group) is frequently required when, e.g., payloads are attached to antibodies to generate ADCs, or proteins need to be precisely labeled, e.g., with fluorescent dyes or biotin. Besides, the N-terminus is an attractive site for modification as it represents a unique position in proteins and is typically not involved in folding. Classical reagents for the conjugation of molecules to amines are NHS and TFP esters, iso(thio)cyanates and the reductive amination with aldehydes. However, all of them react rather unspecific.

Herein, we introduce a novel reagent for the specific N-terminal labeling of proteins and peptides: 1-(4-nitrophenyl)-1H-1,2,3-triazole-4-carbaldehyde (1-NP-triazole-4-CHO) (*RL-8650 on page 116*).

This nitrophenol-functionalized triazole aldehyde reacts with primary amines under mild conditions and high conversion rates with reported yields of 90% and more. This allows to introduce labels, e.g., for imaging. The coupling reaction may be performed in aqueous solution buffered with 10 mM potassium phosphate or MOPS at pH 7.5, by adding the aldehyde dissolved in DMF and incubating for 4-16 hours. Work-up may be performed by chromatography.



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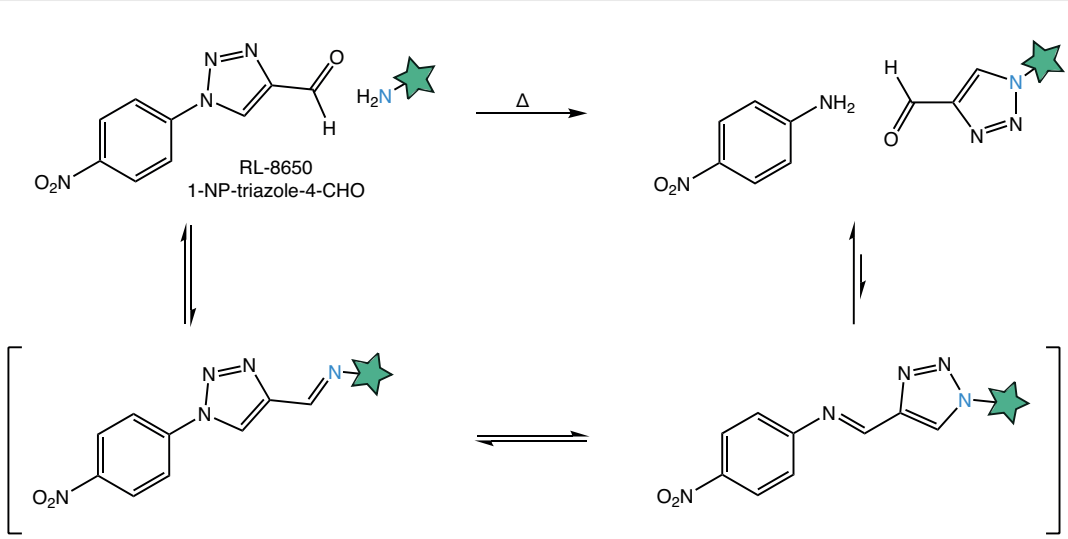



*Fig. 34: Activation of a primary amine (e.g., carrying a label = green star) with 1-NP-triazole-4-carbaldehyde. The amine binds to the aldehyde, forming an imine, which undergoes a rearrangement thus integrating the amine nitrogen as part of the triazole. Then, p-nitrophenylaniline is eliminated and an activated aldehyde triazole is formed which then can be coupled to the N-terminus of a peptide.*

With the success of Chlick chemistry the triazole moiety gained more and more presence in drug design and discovery. Several triazole derivatives with interesting biological activities are already reported in literature making it an attractive structural motive for peptide modification.

The activated triazole aldehyde – bearing the desired label (illustrated as green star in the scheme below) – can be specifically conjugated to the N-terminus of a peptide forming a N-terminal 4-imidazolinone. The reaction may be performed overnight, in an aqueous buffer at physiological pH, e.g., 10 mM phosphate buffer, at 37 °C.

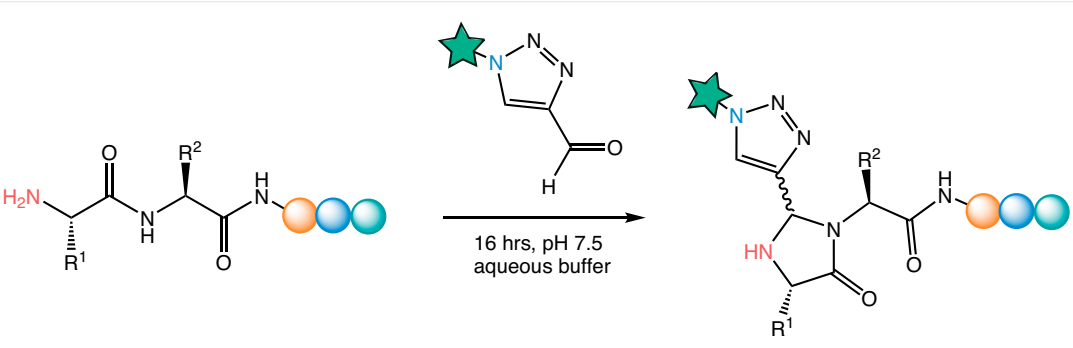
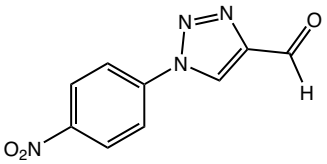



*Fig. 35: Coupling of the aldehyde-functionalized triazole reagent to the N-terminus of a peptide forming an imidazolinone.*

### RL-8650 1-NP-triazole-4-CHO

1-(4-nitrophenyl)triazole-4-carbaldehyde

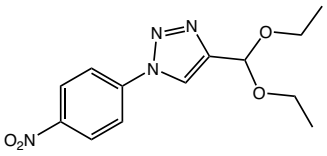
| | |
|---|---|
| CAS-No. | 113934-26-2 |
| Formula | $C_9H_6N_4O_3$ |
| Mol. weight | 218,17 g/mol |



### RL-8655 4-(Diethoxymethyl)-1-NP-triazole

4-(Diethoxymethyl)-1-(4-nitrophenyl)-1H-1,2,3-triazole

| | |
|---|---|
| CAS-No. | 1012081-58-1 |
| Formula | $C_{13}H_{16}N_4O_4$ |
| Mol. weight | 292,29 g/mol |



***References:***


- *Tandem synthesis of 1-formyl-1,2,3-triazoles; J. T. Fletcher, J. A. Christensen, E. M. Villa;* ***Tetrahedron Lett.*** *2017;* ***58(47)****: 4450-4454. https://doi.org/10.1016/j.tetlet.2017.10.023*
- *Synthesis and antibiotic activity of a small molecules library of 1,2,3-triazole derivatives; M. Aufort, J. Herscovici, P. Bouhours, N. Moreau, C. Girard;* ***Bioorg Med Chem Lett.*** *2008;* ***18(3)****: 1195-1198. https://doi.org/10.1016/j.bmcl.2007.11.111*
- *Triazolecarbaldehyde reagents for one-step N-terminal protein modification, A. Onoda, N. Inouse, E. Sumiyoshi, T. Hayashi;* ***ChemBioChem.*** *2019;* ***21(9)****: 1274-1278. https://doi.org/10.1002/cbic.201900692*


## 5.5. Proteolysis Targeting Chimeras (PROTACs®)

Targeted protein degradation (*Fig. 36*) *via* proteolysis-targeting chimeras (PROTACs) is an emerging attempt to cure diseases caused by the irregular expression of certain disease-causing proteins. Such protein degraders act as bifunctional linkers and allow to feed the protein of interest (POI) to the cell's Ubiquitin-Proteasome system, thus, to eliminate the malexpressed proteins. These PROTACs consist of three components: one ligand with high affinity for E3 ubiquitin ligase, another one with high affinity for the protein of interest (POI) and an appropriate cross-linker joining both ligands. This linker can also be used to increase the solubility, if needed, e.g., by incorporation of PEGs. The resulting proximity of both, the recruited POI and the E3 ligase, allows the polyubiquitination of the POI by the E3 associated E2 enzyme. This leads to a labeling of the POI for degradation through the proteasome.



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**Mode of action:**

1. A cross-linker unites the POI ligand and E3 ligase ligand = PROTAC.
2. The three-component PROTAC recruits the POI and the E2-associated E3 ligase *via* the respective ligands = Ternary complex.
3. Several Ubiquitins are added to lysine residues of the POI = Polyubiquitination.
4. The ubiquitinated POI is degraded by the proteasome.

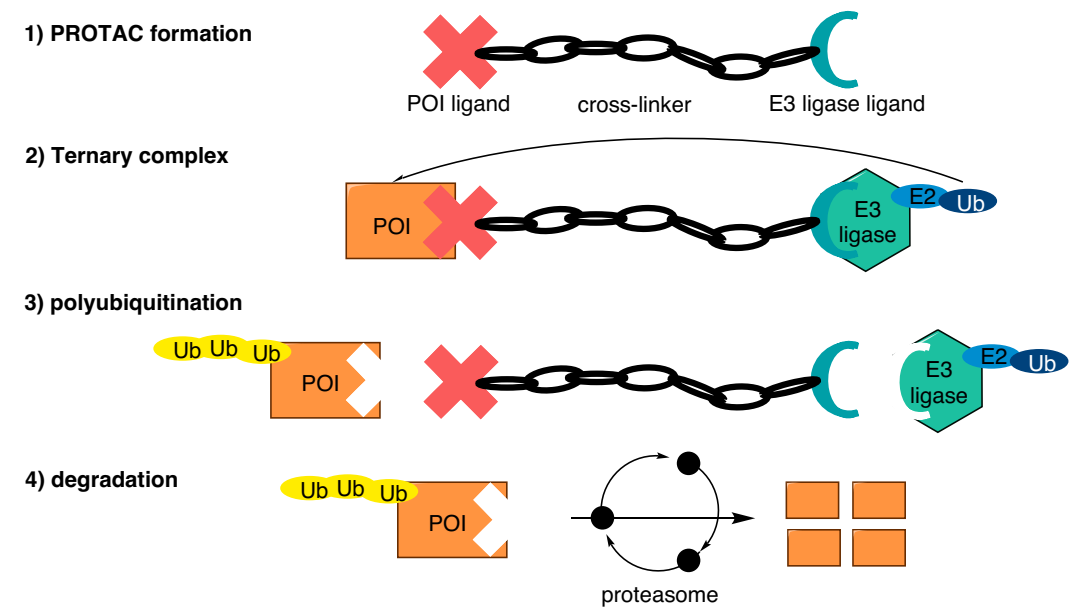



*Fig. 36: Targeted protein degradation via proteolysis-targeting chimeras.*

To construct a suitable PROTAC, we provide a variety of E3 ubiquitin ligase ligands in combination with linkers of various length and an elective amino-, carboxyl-, click- or thiol-reactive end (“Partial PROTACs”).

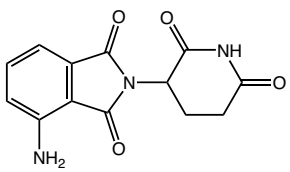
## E3-Ligase Ligands & Negative Controls

Product details

### PTC1000 Pomalidomide

1,3-dioxo-2-(2,6-dioxopiperidin-3-yl)-4-aminoisoindoline,

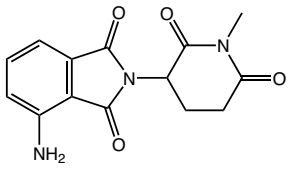
| | |
|---|---|
| CAS-No. | 19171-19-8 |
| Formula | $C_{13}H_{11}N_3O_4$ |
| Mol. weight | 273,24 g/mol |



## PTC1010 N-Methylated pomalidomide

4-Amino-2-(1-methyl-2,6-dioxopiperidin-3-yl)isoindoline-1,3-dione

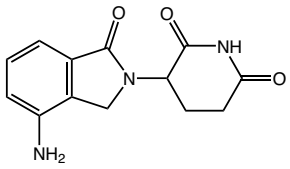
CAS-No. 1352827-50-9
Formula $C_{14}H_{13}N_3O_4$
Mol. weight 287,27 g/mol



## PTC1020 Lenalidomide

1-Oxo-4-amino-2-(2,6-dioxopiperidin-3-yl)isoindole

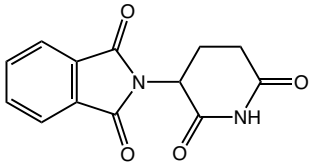
CAS-No. 191732-72-6
Formula $C_{13}H_{13}N_3O_3$
Mol. weight 259,26 g/mol



## PTC1030 (±)-Thalidomide

(±)-2-(2,6-Dioxo-3-piperidinyl)-1H-isoindole-1,3(2H)-dione

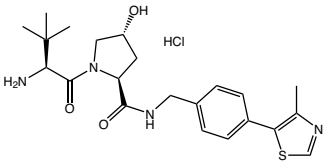
CAS-No. 50-35-1
Formula $C_{13}H_{10}N_2O_4$
Mol. weight 258,23 g/mol



## PTC1040 (S,R,S)-AHPC hydrochloride

(2S,4R)-1-((S)-2-Amino-3,3-dimethylbutanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide hydrochloride

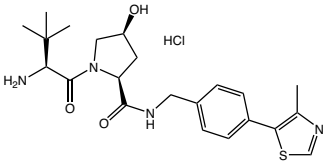
CAS-No. 1448189-80-7
Formula $C_{22}H_{30}N_4O_3S*xHCl$
Mol. weight 430,56 (free base) g/mol



## PTC1050 (S,S,S)-AHPC hydrochloride

(2S,4S)-1-((S)-2-Amino-3,3-dimethylbutanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide hydrochloride

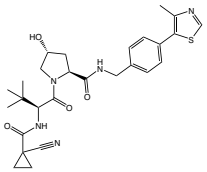
CAS-No. 2115897-23-7
Formula $C_{22}H_{30}N_4O_3S*xHCl$
Mol. weight 430,56 (free base) g/mol



## PTC1060 VH298

(2S,4R)-1-((S)-2-(1-Cyanocyclopropanecarboxamido)-3,3-dimethylbutanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide

CAS-No. 2097381-85-4
Formula $C_{27}H_{33}N_5O_4S$
Mol. weight 523,65 g/mol



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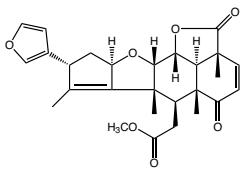
## PTC1070 Nimbolide

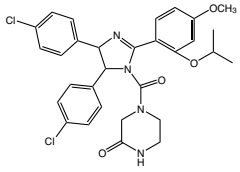
CAS-No. 25990-37-8
Formula $C_{27}H_{30}O_7$
Mol. weight 466,52 g/mol



## PTC1080 Nutlin-3

(±)-4-[4,5-Bis(4-chlorophenyl)-2-(2-isopropoxy-4-methoxy-phenyl)-4,5-dihydro-imidazole-1-carbonyl]-piperazin-2-one

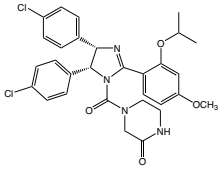
CAS-No. 548472-68-0
Formula $C_{30}H_{30}Cl_2N_4O_4$
Mol. weight 581,49 g/mol



## PTC1090 Nutlin-3a

(-)-4-(4,5-Bis(4-chlorophenyl)-2-(2-isopropoxy-4-methoxyphenyl)-4,5-dihydro-1H-imidazole-1-carbonyl) piperazin-2-one

CAS-No. 675576-98-4
Formula $C_{30}H_{30}Cl_2N_4O_4$
Mol. weight 581,49 g/mol



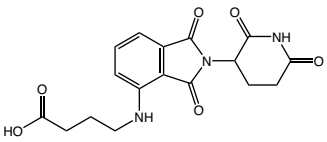
## Amino-Reactive Partial PROTACs

Product details

## PTC1100 Pomalidomide-C3-COOH

4-((2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl) amino)butanoic acid

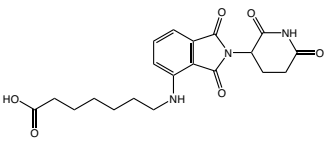
CAS-No. 2225940-47-4
Formula $C_{17}H_{17}N_3O_6$
Mol. weight 359,33 g/mol



## PTC1110 Pomalidomide-C6-COOH

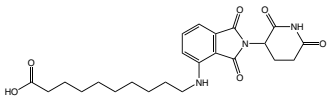
7-((2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl) amino)heptanoic acid

CAS-No. 2225940-50-9
Formula $C_{20}H_{23}N_3O_6$
Mol. weight 401,41 g/mol



## PTC1120 Pomalidomide-C9-COOH

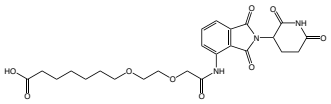
10-((2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)amino)decanoic acid



| | |
|---|---|
| CAS-No. | 2243000-24-8 |
| Formula | $C_{23}H_{29}N_3O_6$ |
| Mol. weight | 443,5 g/mol |

## PTC1130 Pomalidomide-PEG2-butyl COOH

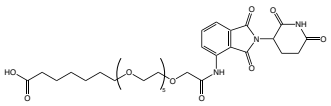
7-(2-(2-((2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)amino)-2-oxoethoxy)ethoxy)heptanoic acid



| | |
|---|---|
| CAS-No. | 2421216-99-9 |
| Formula | $C_{24}H_{29}N_3O_9$ |
| Mol. weight | 503,5 g/mol |

## PTC1140 Pomalidomide-PEG6-butyl COOH

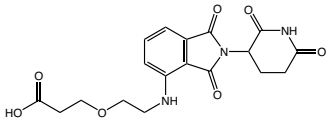
1-((2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)amino)-1-oxo-3,6,9,12,15,18-hexaoxapentacosan-25-oic acid



| | |
|---|---|
| Formula | $C_{32}H_{45}N_3O_{13}$ |
| Mol. weight | 679,71 g/mol |

## PTC1150 Pomalidomide-PEG1-COOH

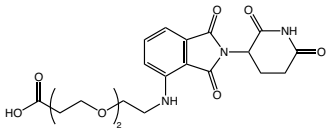
3-(2-((2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)amino)ethoxy)propanoic acid



| | |
|---|---|
| CAS-No. | 2139348-60-8 |
| Formula | $C_{18}H_{19}N_3O_7$ |
| Mol. weight | 389,36 g/mol |

## PTC1160 Pomalidomide-PEG2-COOH

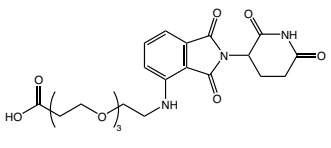
3-(2-(2-((2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)amino)ethoxy)ethoxy)propanoic acid



| | |
|---|---|
| CAS-No. | 2140807-17-4 |
| Formula | $C_{20}H_{23}N_3O_8$ |
| Mol. weight | 433,42 g/mol |

## PTC1170 Pomalidomide-PEG3-COOH

3-(2-(2-(2-((2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)amino)ethoxy)ethoxy)ethoxy)propanoic acid



| | |
|---|---|
| CAS-No. | 2138440-82-9 |
| Formula | $C_{22}H_{27}N_3O_9$ |
| Mol. weight | 477,46 g/mol |

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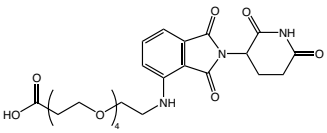
## PTC1180 Pomalidomide-PEG4-COOH

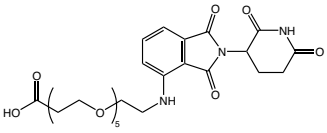
1-((2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl) amino)-3,6,9,12-tetraoxapentadecan-15-oic acid



| | |
|---|---|
| CAS-No. | 2138440-81-8 |
| Formula | $C_{24}H_{31}N_3O_{10}$ |
| Mol. weight | 521,52 g/mol |

## PTC1190 Pomalidomide-PEG5-COOH

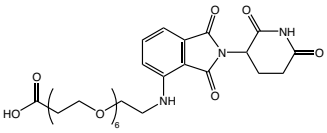
1-((2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl) amino)-3,6,9,12,15-pentaoxaoctadecan-18-oic acid



| | |
|---|---|
| CAS-No. | 2139348-63-1 |
| Formula | $C_{26}H_{35}N_3O_{11}$ |
| Mol. weight | 565,57 g/mol |

## PTC1200 Pomalidomide-PEG6-COOH

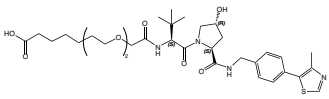
1-((2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl) amino)-3,6,9,12,15,18-hexaoxahenicosan-21-oic acid



| | |
|---|---|
| CAS-No. | 2225148-49-0 |
| Formula | $C_{28}H_{39}N_3O_{12}$ |
| Mol. weight | 609,62 g/mol |

## PTC1220 (S,R,S)-AHPC-PEG2-butyl COOH

(S,R,S)-AHPC-2-2-6-acid,7-(2-(2-(((S)-1-((2S,4R)-4-Hydroxy-2-((4-(4-methylthiazol-5-yl)benzyl)carba-moyl)pyrrolidin-1-yl)-3,3-dimethyl-1-oxobutan-2-yl) amino)-2-oxoethoxy)ethoxy)heptanoic acid



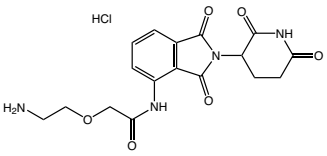
| | |
|---|---|
| CAS-No. | 2421187-89-3 |
| Formula | $C_{33}H_{48}N_4O_8S$ |
| Mol. weight | 660,82 g/mol |

## Carboxy-Reactive Partial PROTACs

Product details

## PTC1230 Pomalidomide-PEG1-$NH_2$ hydrochloride

2-(2-Aminoethoxy)-N-(2-(2,6-dioxopiperi-din-3-yl)-1,3-dioxoisoindolin-4-yl)acetamide hydroch-loride

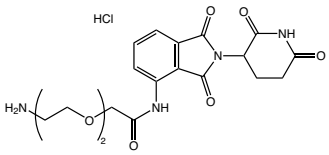


| | |
|---|---|
| CAS-No. | 2380273-67-4 |
| Formula | $C_{17}H_{18}N_4O_6$*xHCl |
| Mol. weight | 374,35 (free base) g/mol |

## PTC1240 Pomalidomide-PEG2-$NH_2$ hydrochloride

2-(2-(2-Aminoethoxy)ethoxy)-N-(2-(2,6-dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)acetamide hydrochloride

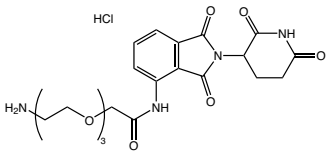
| | |
|---|---|
| CAS-No. | 2380273-73-2 |
| Formula | $C_{19}H_{22}N_4O_7$*xHCl |
| Mol. weight | 418,40 (free base) g/mol |



## PTC1250 Pomalidomide-PEG3-$NH_2$ hydrochloride

2-(2-(2-(2-Aminoethoxy)ethoxy)ethoxy)-N-(2-(2,6-dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)acetamide hydrochloride

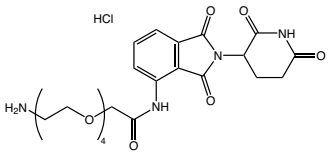
| | |
|---|---|
| CAS-No. | 2380273-75-4 |
| Formula | $C_{21}H_{26}N_4O_8$*xHCl |
| Mol. weight | 462,45 (free base) g/mol |



## PTC1260 Pomalidomide-PEG4-$NH_2$ hydrochloride

14-Amino-N-(2-(2,6-dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)-3,6,9,12-tetraoxatetradecanamide hydrochloride

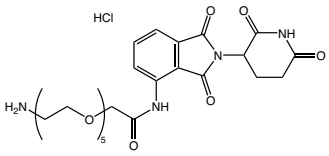
| | |
|---|---|
| CAS-No. | 2331259-45-9 |
| Formula | $C_{23}H_{30}N_4O_9$*xHCl |
| Mol. weight | 506,41 (free base) g/mol |



## PTC1270 Pomalidomide-PEG5-$NH_2$ hydrochloride

17-Amino-N-(2-(2,6-dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)-3,6,9,12,15-pentaoxaheptadecanamide hydrochloride

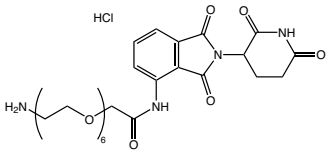
| | |
|---|---|
| CAS-No. | 2421217-05-0 |
| Formula | $C_{25}H_{34}N_4O_{10}$*xHCl |
| Mol. weight | 550,56 (free base) g/mol |



## PTC1280 Pomalidomide-PEG6-$NH_2$ hydrochloride

20-Amino-N-(2-(2,6-dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)-3,6,9,12,15,18-hexaoxaicosanamide hydrochloride

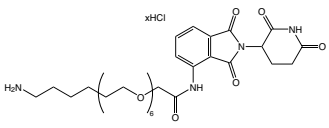
| | |
|---|---|
| Formula | $C_{27}H_{38}N_4O_{11}$*xHCl |
| Mol. weight | 594,61 (free base) g/mol |



## PTC1300 Pomalidomide-PEG6-butyl-$NH_2$ hydrochloride

4-Amino-N-(2-(2,6-dioxopiperidin-3-yl)-1,3-dioxoisoindolin

| | |
|---|---|
| Formula | $C_{31}H_{46}N_4O_{11}$*xHCl |
| Mol. weight | 650,72 (free base) g/mol |



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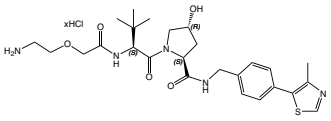
## PTC1310 (S,R,S)-AHPC-PEG1-$NH_2$ hydrochloride

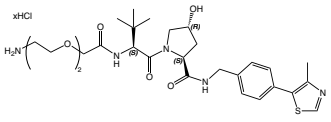
(2S,4R)-1-((S)-2-(2-(2-Aminoethoxy)acetamido)-3,3-dimethylbutanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide hydrochloride



| | |
|---|---|
| CAS-No. | 2711076-33-2 |
| Formula | $C_{26}H_{37}N_5O_5S$*xHCl |
| Mol. weight | 531,67 (free base) g/mol |

## PTC1320 (S,R,S)-AHPC-PEG2-$NH_2$ hydrochloride

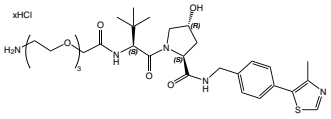
(2S,4R)-1-((S)-2-(2-(2-(2-Aminoethoxy)ethoxy)acetamido)-3,3-dimethylbutanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide hydrochloride



| | |
|---|---|
| CAS-No. | 2097973-72-1 |
| Formula | $C_{28}H_{41}N_5O_6S$*xHCl |
| Mol. weight | 575,72 (free base) g/mol |

## PTC1330 (S,R,S)-AHPC-PEG3-$NH_2$ hydrochloride

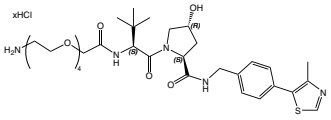
(2S,4R)-1-((S)-14-Amino-2-(*tert*-butyl)-4-oxo-6,9,12-trioxa-3-azatetradecanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide hydrochloride



| | |
|---|---|
| CAS-No. | 2097971-11-2 |
| Formula | $C_{30}H_{45}N_5O_7S$*xHCl |
| Mol. weight | 619,77 (free base) g/mol |

## PTC1340 (S,R,S)-AHPC-PEG4-$NH_2$ hydrochloride

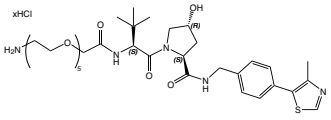
(2S,4R)-1-((S)-17-Amino-2-(*tert*-butyl)-4-oxo-6,9,12,15-tetraoxa-3-azaheptadecanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide hydrochloride



| | |
|---|---|
| CAS-No. | 2010159-57-4 |
| Formula | $C_{32}H_{49}N_5O_8S$*xHCl |
| Mol. weight | 663,83 (free base) g/mol |

## PTC1350 (S,R,S)-AHPC-PEG5-$NH_2$ hydrochloride

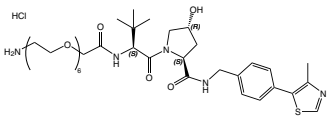
(2S,4R)-1-((S)-20-Amino-2-(*tert*-butyl)-4-oxo-6,9,12,15,18-pentaoxa-3-azaicosanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide hydrochloride



| | |
|---|---|
| CAS-No. | 2377275-23-3 |
| Formula | $C_{34}H_{53}N_5O_9S$*xHCl |
| Mol. weight | 707,88 (free base) g/mol |

## PTC1360 (S,R,S)-AHPC-PEG6-$NH_2$ hydrochloride

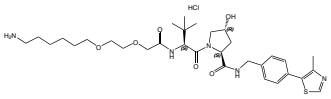
(2S,4R)-1-((S)-23-Amino-2-(*tert*-butyl)-4-oxo-6,9,12,15,18,21-hexaoxa-3-azatricosanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide hydrochloride



| | |
|---|---|
| CAS-No. | 2924759-90-8 |
| Formula | $C_{36}H_{57}N_5O_{10}S$*xHCl |
| Mol. weight | 751,93 (free base) g/mol |

## PTC1370 (S,R,S)-AHPC-PEG2-butyl-$NH_2$ hydrochloride

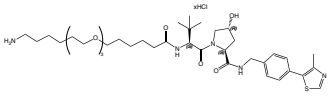
(2S,4R)-1-((S)-2-(2-(2-((6-Aminohexyl)oxy)ethoxy) acetamido)-3,3-dimethylbutanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide



| | |
|---|---|
| CAS-No. | 2421187-85-9 net |
| Formula | $C_{32}H_{49}N_5O_6S$*xHCl |
| Mol. weight | 631,83 (free base) g/mol |

## PTC1380 (S,R,S)-AHPC-C6-PEG3-butyl-$NH_2$ hydrochloride

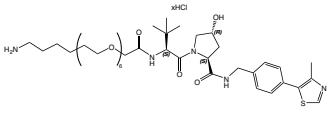
(2S,4R)-1-((S)-22-Amino-2-(*tert*-butyl)-4-oxo-10,13,16-trioxa-3-azadocosanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide



| | |
|---|---|
| CAS-No. | 2421187-84-8 net |
| Formula | $C_{38}H_{61}N_5O_7S$*xHCl |
| Mol. weight | 731,99 (free base) g/mol |

## PTC1390 (S,R,S)-AHPC-PEG6-butyl-$NH_2$ hydrochloride

(2S,4R)-1-((S)-27-Amino-2-(*tert*-butyl)-4-oxo-6,9,12,15,18,21-hexaoxa-3-azaheptacosanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide



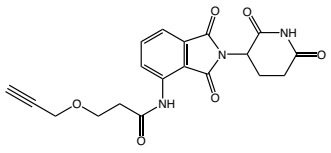
| | |
|---|---|
| Formula | $C_{40}H_{65}N_5O_{10}S$*xHCl |
| Mol. weight | 808,04 (free base) g/mol |

# Click-Reactive Partial PROTACs

Product details

## PTC1400 Pomalidomide-PEG1-Alkyne

N-(2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)-3-(prop-2-yn-1-yloxy)propanamide



| | |
|---|---|
| CAS-No. | 2236109-19-4 |
| Formula | $C_{19}H_{17}N_3O_6$ |
| Mol. weight | 383,35 g/mol |

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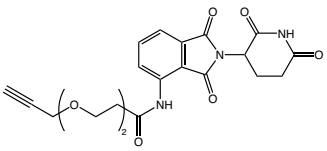
Product details

## PTC1410 Pomalidomide-PEG2-Alkyne

N-(2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)-3-(2-(prop-2-yn-1-yloxy)ethoxy)propanamide

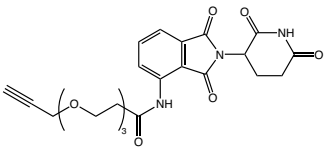
CAS-No. 2243000-25-9
Formula $C_{21}H_{21}N_3O_7$
Mol. weight 427,41 g/mol



## PTC1420 Pomalidomide-PEG3-Alkyne

N-(2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)-3-(2-(2-(prop-2-yn-1-yloxy)ethoxy)ethoxy)propanamide

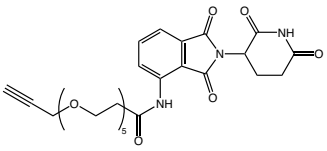
CAS-No. 2236109-20-7
Formula $C_{23}H_{25}N_3O_8$
Mol. weight 471,46 g/mol



## PTC1440 Pomalidomide-PEG5-Alkyne

N-(2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)-4,7,10,13,16-pentaoxanonadec-18-ynamide

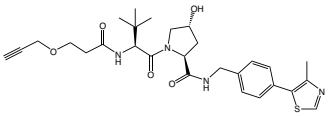
Formula $C_{27}H_{33}N_3O_{10}$
Mol. weight 559,57 g/mol



## PTC1460 (S,R,S)-AHPC-PEG1-Alkyne

(2S,4R)-1-((S)-3,3-Dimethyl-2-(3-(prop-2-yn-1-yloxy)propanamido)butanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide

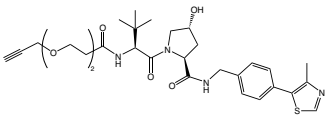
CAS-No. 2242965-71-3
Formula $C_{28}H_{36}N_4O_5S$
Mol. weight 540,67 g/mol



## PTC1470 (S,R,S)-AHPC-PEG2-Alkyne

(2S,4R)-1-((S)-3,3-Dimethyl-2-(3-(2-(prop-2-yn-1-yloxy)ethoxy)propanamido)butanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide

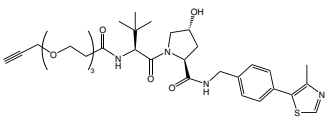
CAS-No. 2242965-72-4
Formula $C_{30}H_{40}N_4O_6S$
Mol. weight 584,73 g/mol



## PTC1480 (S,R,S)-AHPC-PEG3-Alkyne

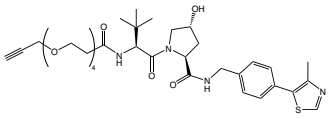
(2S,4R)-1-((S)-2-(*tert*-Butyl)-4-oxo-7,10,13-trioxa-3-azahexadec-15-ynoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide

CAS-No. 2374122-30-0
Formula $C_{32}H_{44}N_4O_7S$
Mol. weight 628,78 g/mol



## PTC1490 (S,R,S)-AHPC-PEG4-Alkyne

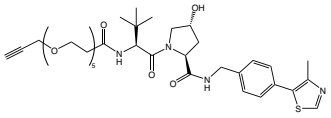
(2S,4R)-1-((S)-2-(*tert*-Butyl)-4-oxo-7,10,13,16-tetraoxa-3-azanonadec-18-ynoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide



| | |
|---|---|
| CAS-No. | 2267282-21-1 |
| Formula | $C_{34}H_{48}N_4O_8S$ |
| Mol. weight | 672,83 g/mol |

## PTC1500 (S,R,S)-AHPC-PEG5-Alkyne

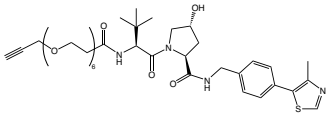
(2S,4R)-1-((S)-2-(*tert*-Butyl)-4-oxo-7,10,13,16,19-pentaoxa-3-azadocos-21-ynoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide



| | |
|---|---|
| CAS-No. | 2817805-63-1 |
| Formula | $C_{36}H_{52}N_4O_9S$ |
| Mol. weight | 716,88 g/mol |

## PTC1510 (S,R,S)-AHPC-PEG6-Alkyne

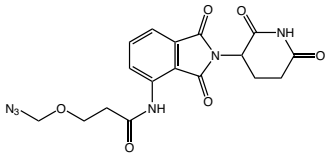
(2S,4R)-1-((S)-2-(*tert*-Butyl)-4-oxo-7,10,13,16,19,22-hexaoxa-3-azapentacos-24-ynoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide



| | |
|---|---|
| Formula | $C_{38}H_{56}N_4O_{10}S$ |
| Mol. weight | 760,94 g/mol |

## PTC1520 Pomalidomid- PEG1-$N_3$

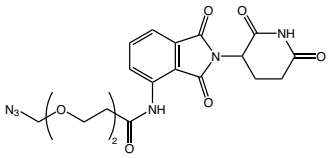
2-(2-Azidoethoxy)-N-(2-(2,6-dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)acetamide



| | |
|---|---|
| CAS-No. | 2133360-04-8 |
| Formula | $C_{17}H_{16}N_6O_6$ |
| Mol. weight | 400,35 g/mol |

## PTC1530 Pomalidomid- PEG2-$N_3$

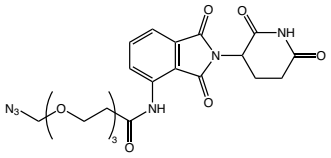
2-(2-(2-Azidoethoxy)ethoxy)-N-(2-(2,6-dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)acetamide



| | |
|---|---|
| CAS-No. | 2267306-14-7 |
| Formula | $C_{19}H_{20}N_6O_7$ |
| Mol. weight | 444,4 g/mol |

## PTC1540 Pomalidomid- PEG3-$N_3$

2-(2-(2-(2-azidoethoxy)ethoxy)ethoxy)-N-(2-(2,6-dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)acetamide



| | |
|---|---|
| CAS-No. | 2267306-15-8 |
| Formula | $C_{21}H_{24}N_6O_8$ |
| Mol. weight | 488,45 g/mol |

The Concept of Antibody-Drug Conjugation (ADC)
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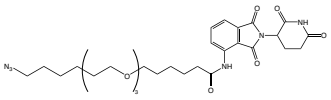
Product details

## PTC1560 Pomalidomid-C6-PEG3-butyl-$N_3$

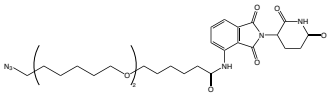
6-(2-(2-((6-Azidohexyl)oxy)ethoxy)ethoxy)-N-(2-(2,6-dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)hexanamide



| | |
|---|---|
| CAS-No. | 2300178-66-7 |
| Formula | $C_{29}H_{40}N_6O_8$ |
| Mol. weight | 600,66 g/mol |

## PTC1570 Pomalidomid-C6-PEG1-C3-PEG1-butyl-$N_3$

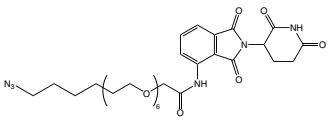
6-((5-((6-Azidohexyl)oxy)pentyl)oxy)-N-(2-(2,6-dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)hexanamide



| | |
|---|---|
| CAS-No. | 2300178-65-6 |
| Formula | $C_{30}H_{42}N_6O_7$ |
| Mol. weight | 598,69 g/mol |

## PTC1580 Pomalidomid-PEG6-butyl-$N_3$

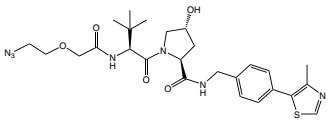
4-azido-N-(2-(2,6-dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)-3,6,9,12,15,18-hexaoxatetracosanamide



| | |
|---|---|
| Formula | $C_{31}H_{44}N_6O_{11}$ |
| Mol. weight | 676,71 g/mol |

## PTC1590 (S,R,S)-AHPC-PEG1-$N_3$

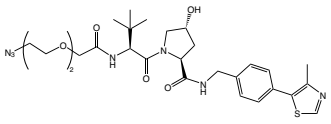
(2S,4R)-1-((S)-2-(2-(2-Azidoethoxy)acetamido)-3,3-dimethylbutanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide



| | |
|---|---|
| CAS-No. | 2101200-09-1 |
| Formula | $C_{26}H_{35}N_7O_5S$ |
| Mol. weight | 557,67 g/mol |

## PTC1600 (S,R,S)-AHPC-PEG2-$N_3$

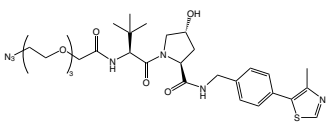
(2S,4R)-1-((S)-2-(2-(2-(2-Azidoethoxy)ethoxy)acetamido)-3,3-dimethylbutanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide



| | |
|---|---|
| CAS-No. | 2010159-45-0 |
| Formula | $C_{28}H_{39}N_7O_6S$ |
| Mol. weight | 601,72 g/mol |

## PTC1610 (S,R,S)-AHPC-PEG3-$N_3$

(2S,4R)-1-((S)-14-azido-2-(*tert*-butyl)-4-oxo-6,9,12-trioxa-3-azatetradecanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide

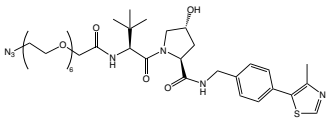


| | |
|---|---|
| CAS-No. | 1797406-80-4 |
| Formula | $C_{30}H_{43}N_7O_7S$ |
| Mol. weight | 645,77 g/mol |

Product details

## PTC1640 (S,R,S)-AHPC-PEG6-$N_3$

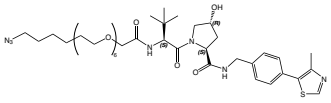
(2S,4R)-1-((S)-23-Azido-2-(*tert*-butyl)-4-oxo-6,9,12,15,18,21-hexaoxa-3-azatricosanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide



| | |
|---|---|
| CAS-No. | 2086298-71-5 |
| Formula | $C_{36}H_{55}N_7O_{10}S$ |
| Mol. weight | 777,93 g/mol |

## PTC1680 (S,R,S)-AHPC-PEG6-butyl-$N_3$

(2S,4R)-1-((S)-27-Azido-2-(*tert*-butyl)-4-oxo-6,9,12,15,18,21-hexaoxa-3-azaheptacosanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide



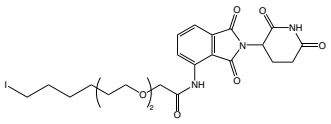
| | |
|---|---|
| Formula | $C_{40}H_{63}N_7O_{10}S$ |
| Mol. weight | 834,03 g/mol |

# Thiol-Reactive Partial PROTACs

Product details

## PTC1690 Pomalidomid-PEG2-butyl-I

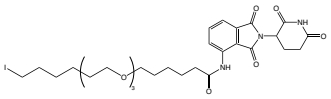
N-(2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)-2-(2-((6-iodohexyl)oxy)ethoxy)acetamide



| | |
|---|---|
| CAS-No. | 1835705-72-0 |
| Formula | $C_{23}H_{28}IN_3O_7$ |
| Mol. weight | 585,39 g/mol |

## PTC1700 Pomalidomid-C6-PEG3-butyl-I

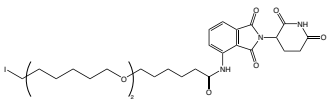
N-(2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)-6-(2-(2-((6-iodohexyl)oxy)ethoxy)ethoxy)hexanamide



| | |
|---|---|
| CAS-No. | 1835705-70-8 |
| Formula | $C_{29}H_{40}IN_3O_8$ |
| Mol. weight | 685,55 g/mol |

## PTC1710 Pomalidomid-C6-PEG1-C3-PEG1-butyl-I

N-(2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindolin-4-yl)-6-((5-((6-iodohexyl)oxy)pentyl)oxy)hexanamide



| | |
|---|---|
| CAS-No. | 1835705-76-4 |
| Formula | $C_{30}H_{42}IN_3O_7$ |
| Mol. weight | 683,57 g/mol |

The Concept of Antibody-Drug Conjugation (ADC)
Permanent Linkers
Cleavable Linkers
Trifunctional Linkers
Cross-Linkers for other Bio Applications
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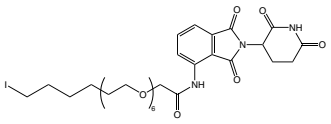
Product details

## PTC1720 Pomalidomid-PEG6-butyl-I

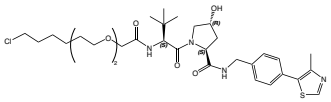
N-(2-(2,6-Dioxopiperidin-3-yl)-1,3-dioxoisoindo-lin-4-yl)-24-iodo-3,6,9,12,15,18-hexaoxatetracosanamide



| | |
|---|---|
| CAS-No. | 1835705-74-2 |
| Formula | $C_{31}H_{44}IN_3O_{11}$ |
| Mol. weight | 761,6 g/mol |

## PTC1730 (S,R,S)-AHPC-PEG2-butyl-Cl

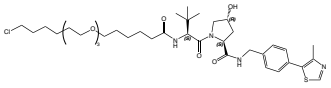
(2S,4R)-1-((S)-2-(2-(2-((6-Chlorohexyl)oxy)ethoxy) acetamido)-3,3-dimethylbutanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide



| | |
|---|---|
| CAS-No. | 1835705-57-1 |
| Formula | $C_{32}H_{47}ClN_4O_6S$ |
| Mol. weight | 651,26 g/mol |

## PTC1750 (S,R,S)-AHPC-C6-PEG3-butyl-Cl

(2S,4R)-1-((S)-2-(*tert*-Butyl)-22-chloro-4-oxo-10,13,16-trioxa-3-azadocosanoyl)-4-hydroxy-N-(4-(4-methylthiazol-5-yl)benzyl)pyrrolidine-2-carboxamide



| | |
|---|---|
| CAS-No. | 1835705-55-9 |
| Formula | $C_{38}H_{59}ClN_4O_7S$ |
| Mol. weight | 751,42 g/mol |

***References:***


- *Bifunctional Molecules beyond PROTACs; S. J. Conway;* ***J. Med. Chem.*** *2020;* ***63****: 2802-2806. https://doi.org/10.1021/acs.jmedchem.0c00293*
- *Targeted protein degradation by PROTACs; T. K. Neklesa, J. D. Winkler, C. M. Crews;* ***Pharmacol Ther*** *2017;* ***174****: 138-144. https://doi.org/10.1016/j.pharmthera.2017.02.027*
- *Targeted Protein Degradation: from Chemical Biology to Drug Discovery; P. M. Cromm, C. M. Crews;* ***Cell Chem Biol*** *2017;* ***24****: 1181-1190. https://doi.org/10.1016/j.chembiol.2017.05.024*
- *Targeted Protein Degradation by Small Molecules; D. P. Bondeson, C. M. Crews;* ***Annu Rev Pharmacol Toxicol*** *2017;* ***57****: 107-123. https://doi.org/10.1146/annurev-pharmtox-010715-103507*
- *Small-Molecule PROTACS: New Approaches to Protein Degradation; M. Toure, C. M. Crews;* ***Angew Chem Int Ed*** *2016;* ***55****: 1966-73. https://doi.org/10.1002/anie.201507978*
- *Impact of linker length on the activity of PROTACs; K. Cyrus, M. Wehenkel, E. Y. Choi, H. J. Han, H. Lee, H. Swanson, K. B. Kim;* ***Mol Biosyst*** *2011;* ***7****: 359-64. https://doi.org/10.1039/c0mb00074d*
- *Development of potent monoclonal antibody auristatin conjugates for cancer therapy; S. O. Doronina, B. E. Toki, M. Y. Torgov, B. A. Mendelsohn, C. G. Cerveny, D. F. Chace, R. L. DeBlanc, R. P. Gearing, T. D. Bovee, C. B. Siegall, J. A. Francisco, A. F. Wahl, D. L. Meyer, P. D. Senter;* ***Nat Biotechnol*** *2003;* ***21****: 778-84. https://doi.org/10.1038/nbt832*
- *Gemtuzumab ozogamicin, a potent and selective anti-CD33 antibody-calicheamicin conjugate for treatment of acute myeloid leukemia; P. R. Hamann, L. M. Hinman, I. Hollander, C. F. Beyer, D. Lindh, R. Holcomb, W. Hallett, H. R. Tsou, J. Upeslacis, D. Shochat, A. Mountain, D. A. Flowers, I. Bernstein;* ***Bioconjug Chem*** *2002;* ***13****: 47-58. https://doi.org/10.1021/bc010021y*

In addition to these pre-designed building blocks, we offer custom synthesis of your required ligand-linker combination (*Fig. 37*) or "complete PROTAC". This allows to design a library of slightly different PROTACs in order to find the best combination for your application, as even small changes in ligands and cross-linkers might affect the efficiency of the formation of the ternary complex.

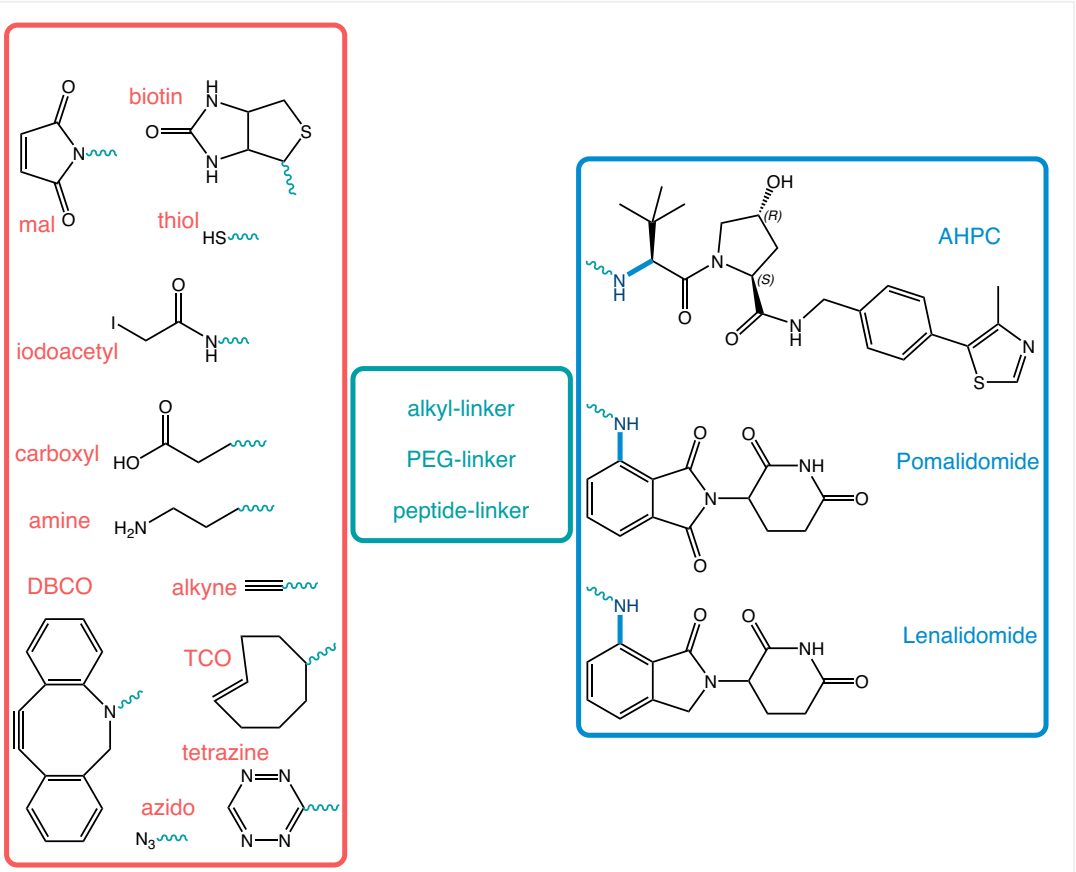



*Fig. 37: Possibilities of PROTAC design. Above displayed options for linker constructs can be conjugated to substrates of the protein of interest, in order to create the desired PROTAC®.*

**Please contact us for Custom Synthesis of the PROTAC® linker fragment of your choice or complete functional PROTAC®.**



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## 5.6. Site-Selective π-Clamp Mediated Cysteine Arylation

For the modification of proteins, cysteine is a suitable choice for bioconjugation because of the high nucleophilicity of its thiol group as well as the low abundance of cysteine residues in the majority of naturally occurring proteins (ca. 1.7%). However, conventional chemical cysteine-based conjugation techniques are not site-specific resulting in a multiple labelling mixture at random positions.

In nature, selective reactions in proteins are triggered by the formation of certain microenvironments by three-dimensional secondary structures of the surrounding polypeptide architecture. Inspired by nature, Pentelute *et al.* envisioned a new strategy for site-selective chemistry, the so called "π-clamp" arylation. cysteine embedded in the four-amino-acid sequence (Phe-Cys-Pro-Phe) tunes up the thiol reactivity for site-selective conjugation with perfluoroaromatic reagents. Thus, the π-clamp allows the selective modification of one cysteine site in a protein containing multiple other endogenous cysteine residues.

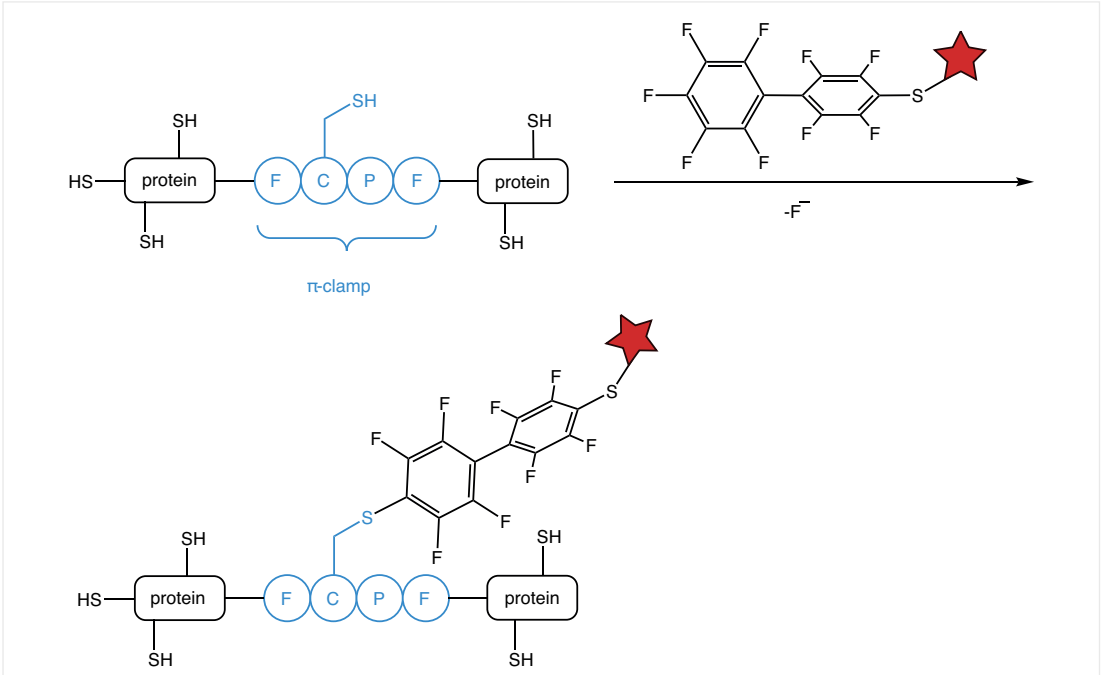



*Fig. 38: A cysteine residue inside the π-clamp selectively reacts with perfluorobiphenyl probes in thepresence of other competing cysteine residues or thiols.*

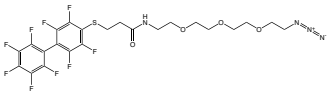
The reported reaction is site-specific, operational under physiological conditions, enzyme-free, and as efficient as the commonly used azide-alkyne click chemistry. Furthermore, the π-clamp works as part of the N-terminus, the C-terminus, as well as in the middle of a polypeptide chain. Besides, its small size hardly perturbs the target protein's native structure.

In addition to the possibility for site-selective cysteine labeling in any linear peptide, the π-clamp approach should allow for macrocyclization between cysteine thiolates as a last, high-yielding synthetic step either *via* an exogenously added perfluoroaryl-based linker, or by incorporating non-crosslinked perfluoroaryl-based moieties first, followed by their macrocyclization with dithiol reagents.

## RL-4030 PFB-mercaptopropionyl-PEG3-$N_3$

Perfluorobiphenyl-mercaptopropionyl-PEG(3)-$N_3$

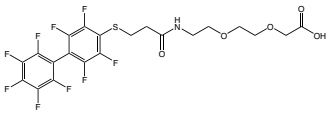
Formula $C_{23}H_{21}F_9N_4O_4S$
Mol. weight 620,49 g/mol



## RL-4040 PFB-mercaptopropionyl-AEEA

Perfluorobiphenyl-mercaptopropionyl-AEEA

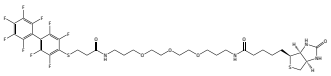
Formula $C_{21}H_{16}F_9NO_5S$
Mol. weight 565,41 g/mol



## RL-4050 PFB-mercaptopropionyl-TOTA-Biotin

Perfluorobiphenyl-mercaptopropionyl-TOTA-Biotin

Formula $C_{35}H_{41}F_9N_4O_6S_2$
Mol. weight 848,84 g/mol



***References:***


- *Site-Specific Small Molecule Labeling of an Internal Loop in JC Polyomavirus Pentamers Using the π-Clamp-Mediated Cysteine Conjugation; J. A. Baccile, P. J. Voorhees, A. J. Chillo, M. Berry, R. Morgenstern, T. J. Schwertfeger, F. M. Rossi, C. D. S. Nelson;* ***Chembiochem*** *2021;* ***22(21)****: 3037-3041. https://doi.org/10.1002/cbic.202100188*
- *Convergent diversity-oriented side-chain macrocyclization scan for unprotected polypeptides; Y. Zou, A. M. Spokoyny, C. Zhang, M. D. Simon, H. Yu, Y.-S. Lin, B. L. Pentelute;* ***Org. Biomol. Chem.*** *2014;* ***12****: 566-573. https://doi.org/10.1039/C3OB42168F*
- *Enzymatic „click" ligation: selective cysteine modification in polypeptides enabled by promiscuous glutathione S-transferase; C. Zhang, A. M. Spokoyny, Y. Zou, M. D. Simon, B. L. Pentelute;* ***Angew Chem Int Ed Engl.*** *2013;* ***52(52)****: 14001-5. https://doi.org/10.1002/anie.201306430*
- *π-Clamp-mediated cysteine conjugation; C. Zhang, M. Welborn, T. Zhu, N. J. Yang, M. S. Santos, T. Van Voorhis, B. L. Pentelute;* ***Nat Chem.*** *2016;* ***8****: 120-128. https://doi.org/10.1038/nchem.2413*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# 6. Preparing Carriers for Conjugation

*Tab. 3: Conceptual overview of conjugation technologies.*

| Carrier | Conjugation Chemistry | Cleavage Mechanism | Fragmentation for Traceless Release | Cargo Functionality |
|---|---|---|---|---|
| **Biopolymers:**<br>• Peptides<br>• Proteins<br>• Antibodies<br>• Single Chain<br>• Nanobodies<br>• Camelides<br>• Oligonucleotides<br>• Aptamers | Thioether formation with maleimide<br>Disulfide bond formation<br>Acylation of amines<br>His tag acylation<br>Click conjugation (CuAAC, SPAAC, IEDDA)<br>Enzyme supported conjugation:<br>• HaloTag®<br>• CLIP-Tag™<br>• SNAP-Tag®<br>• Sequence dependent conjugation (Sortase, Ligase) | **Enzymatic hydrolysis:**<br>• Val-Ala<br>• Val-Cit<br>• Phe-Lys<br>• Gly-Phe-Leu-Gly<br>• Ala-Leu-Ala-Leu<br>• Cyclobutyl-Ala<br>• Cyclobutyl-Cit<br>• Glucuronic acid | *p*-Aminobenzyl<br>*p*-Hydroxybenzyl<br>*p*-Mercaptobenzyl | Primary & secondary amines<br>Tertiary amines<br>Alcohols/Phenols<br>Carboxylic acids |
| **Carbon:**<br>• Nanotubes<br>• Fullerenes | Nitrene addition *via* photoactivation of perfluoroarylazides | **Reduction:** | Oxathiolone | |
| **Metals:**<br>• Gold<br>• Silver | Affinity of sulfur to gold and silver | | | |
| **Metal oxide** | Chelate formation | **Oxidation:**<br>Tetramethyl-dioxoborolane | Dimethylimidazolidinone | |
| **Plastic polymers:**<br>• Teflon<br>• Polyethylene<br>• Polystyrene<br>• Latex | Introduction of reactive functional groups by plasma treatment (e.g., ammonia or acrylic acid) | **pH:**<br>5-(hydroxymethyl)pyrogallol orthoester (HMPO) | | |
| **Silicates** | Affinity between silicon and oxygen | | | |

## 6.1. Antibodies, Antibody Formats and Proteins by (Cell-free) Recombinant Methodologies

Cell-free protein synthesis, often referred to as *in vitro* translation, is a fast and viable technique which, in comparison to *in vivo* protein synthesis, leads to the production of a target protein in a considerably less laborious way. Cell-free systems are based on lysates derived from *E. coli* or eukaryotic sources and they allow for the synthesis of a broad spectrum of structurally diverse and modified proteins. The path from a DNA template to the protein of interest is reduced to only a few hours of time and additionally, no genetically modified organisms (GMOs) are needed. A variety of proteins, such as certain membrane proteins (e.g., GPCRs), toxins or transcriptional and translational factors, whose synthesis in conventional *in vivo* systems is often associated with difficulties, can be synthesized *in vitro*. Only the target protein is synthesized in cell-free systems, since endogenous mRNA templates are removed during the process of lysate preparation. The open and flexible character of cell-free systems allows the well-tuned adjustment of the reaction environment by supplementing the reaction mixture with co-factors, chaperones, detergents, rare tRNAs and buffers of varying ion composition, depending on the demands of the individual protein. The reaction conditions during protein synthesis significantly impact protein folding, protein activity and functionality.

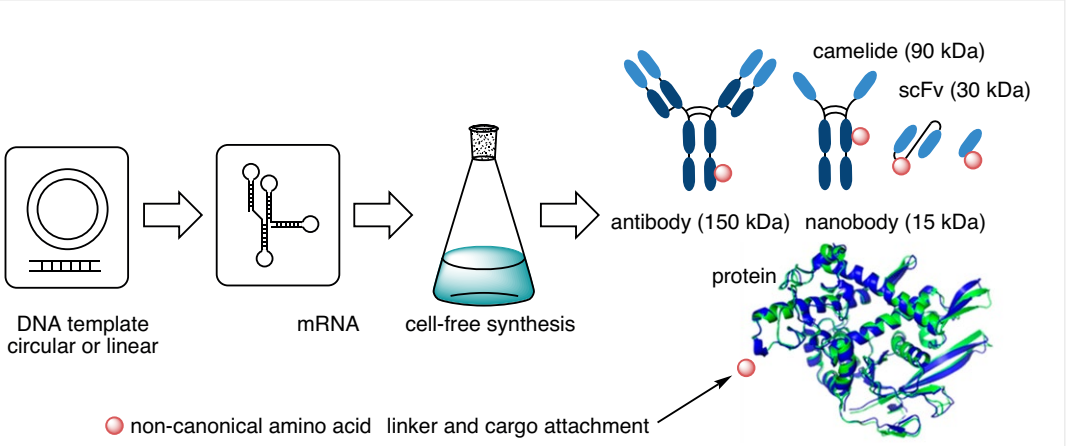


*Fig. 39: General principle of cell-free synthesis of biomolecules with position specific introduction of non-canonical amino acids as conjugation point for subsequent Linkerology®.*

Cell-free protein synthesis provides a fast access to proteins and antibody formats including introduction of mutations or non-canonical amino acids at defined positions for subsequent conjugation with permanent or self-immolative linkers and payloads. This includes otherwise difficult to synthesize toxic proteins, membrane proteins, labelled proteins, protein-conjugates, and antibody formats.

**You need more details about cell-free protein synthesis?**

**Watch the recording of our workshop!**

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*Do you have a certain bioconjugate in mind?*

- *Membrane protein carrying a conjugation function on a specific position.*
- *Antibody, single chain or nanobody decorated with azido or alkyne function for initial linker attachment.*
- *Biomolecule-linker conjugate, ready to load your payload.*
- *Or the final biomolecule-drug-conjugate.*
- *Adaption of your current protein ready for linker attachment.*

## Consult with us, we will work out the best solution for you!

***References:***


- *Enriched cell-free and cell-based native membrane derived vesicles (nMV) enabling rapid in-vitro electrophysiological analysis of the voltage-gated sodium channel 1.5; Y. Pandey, S. K. Dondapati, S. Kubick; **Biochim Biophys Acta Biomembr** 2023; **1865**: 184144. https://doi.org/10.1016/j.bbamem.2023.184144*
- *Rapid One-Step Capturing of Native, Cell-Free Synthesized and Membrane-Embedded GLP-1R; L. Haueis, M. Stech, E. Schneider, T. Lanz, N. Hebel, A. Zemella, S. Kubick; **Int J Mol Sci** 2023; **24**: 2808. https://doi.org/10.3390/ijms24032808*
- *Evaluation of the Ion Channel Assembly in a Eukaryotic Cell-Free System Focusing on Two-Pore Domain Potassium Channels K2P; J. Ullrich, C. Ohlhoff, S. K. Dondapati, A. Zemella, S. Kubick; **International Journal of Molecular Sciences** 2023; **24**: 6299. https://doi.org/10.3390/ijms24076299*
- *Cell-Free Systems Enable the Production of AB(5) Toxins for Diagnostic Applications; F. Ramm, L. Jack, D. Kaser, J. L. Schlosshauer, A. Zemella, S. Kubick; **Toxins (Basel)** 2022; **14**: 233. https://doi.org/10.3390/toxins14040233*
- *The Potential of Eukaryotic Cell-Free Systems as a Rapid Response to Novel Zoonotic Pathogens: Analysis of SARS-CoV-2 Viral Proteins; F. Ramm, S. K. Dondapati, H. A. Trinh, D. Wenzel, R. M. Walter, A. Zemella, S. Kubick; **Front Bioeng Biotechnol** 2022; **10**: 896751. https://doi.org/10.3389/fbioe.2022.896751*
- *Synthesis of an Anti-CD7 Recombinant Immunotoxin Based on PE24 in CHO and E. coli Cell-Free Systems; S. K. Krebs, M. Stech, F. Jorde, N. Rakotoarinoro, F. Ramm, S. Marinoff, S. Bahrke, A. Danielczyk, D. A. Wustenhagen, S. Kubick; **Int J Mol Sci** 2022; **23**: 13697. https://doi.org/10.3390/ijms232213697*
- *A Cell-free Expression Pipeline for the Generation and Functional Characterization of Nanobodies; L. Haueis, M. Stech, S. Kubick; **Front Bioeng Biotechnol** 2022; **10**: 896763. https://doi.org/10.3389/fbioe.2022.896763*
- *Synthesis of Fluorescently Labeled Antibodies Using Non-Canonical Amino Acids in Eukaryotic Cell-Free Systems; M. Stech, N. Rakotoarinoro, T. Teichmann, A. Zemella, L. Thoring, S. Kubick; Structural Proteomics: High-Throughput Methods R. J. Owens 2021: 175-190. https://doi.org/10.1007/978-1-0716-1406-8_9*
- *Cell-Free Protein Synthesis: A Promising Option for Future Drug Development; S. K. Dondapati, M. Stech, A. Zemella, S. Kubick; **BioDrugs** 2020; **34**: 327-348. https://doi.org/10.1007/s40259-020-00417-y*
- *Accelerating the Production of Druggable Targets: Eukaryotic Cell-Free Systems Come into Focus; L. Thoring, A. Zemella, D. Wüstenhagen, S. Kubick; **Methods and Protocols** 2019; **2**: 30. https://doi.org/10.3390/mps2020030*


## 6.2. Aptamers and other Oligonucleotides

Terminal azide bearing nucleotides closely resemble natural nucleotides and are the basis of chemoenzymatic oligonucleotide labeling. T7 RNA polymerase, poly(A) polymerase and the terminal deoxynucleotidyl transferase (Tdt) tolerate azide and alkyne decorated nucleotides and incorporate them at a defined position. Any mRNA can be site-specifically labeled without special requirements or altered production protocols. By click labeling at the 3'-end the half-life of the mRNA could be modulated, which might be an option for increased mRNA stability.

Feeding living cells with 5-ethynyl uridine (EU) will lead to EU labelled RNA, which further can be modified *via* Click conjugation, e.g., with fluorescent dyes or linkers carrying small molecules, peptides or proteins. Incorporation into DNA can also occur after some incubation time. This is most likely *via* conversion of the EU ribonucleoside into EdU aided by intracellular ribonucleotide reductases.

With 5-ethynyl-dA-CEP and 5-ethynyl-dU-CEP alkyne moieties can be used under standard conditions for solid phase synthesis of oligonucleotides followed by subsequence Click conjugation.

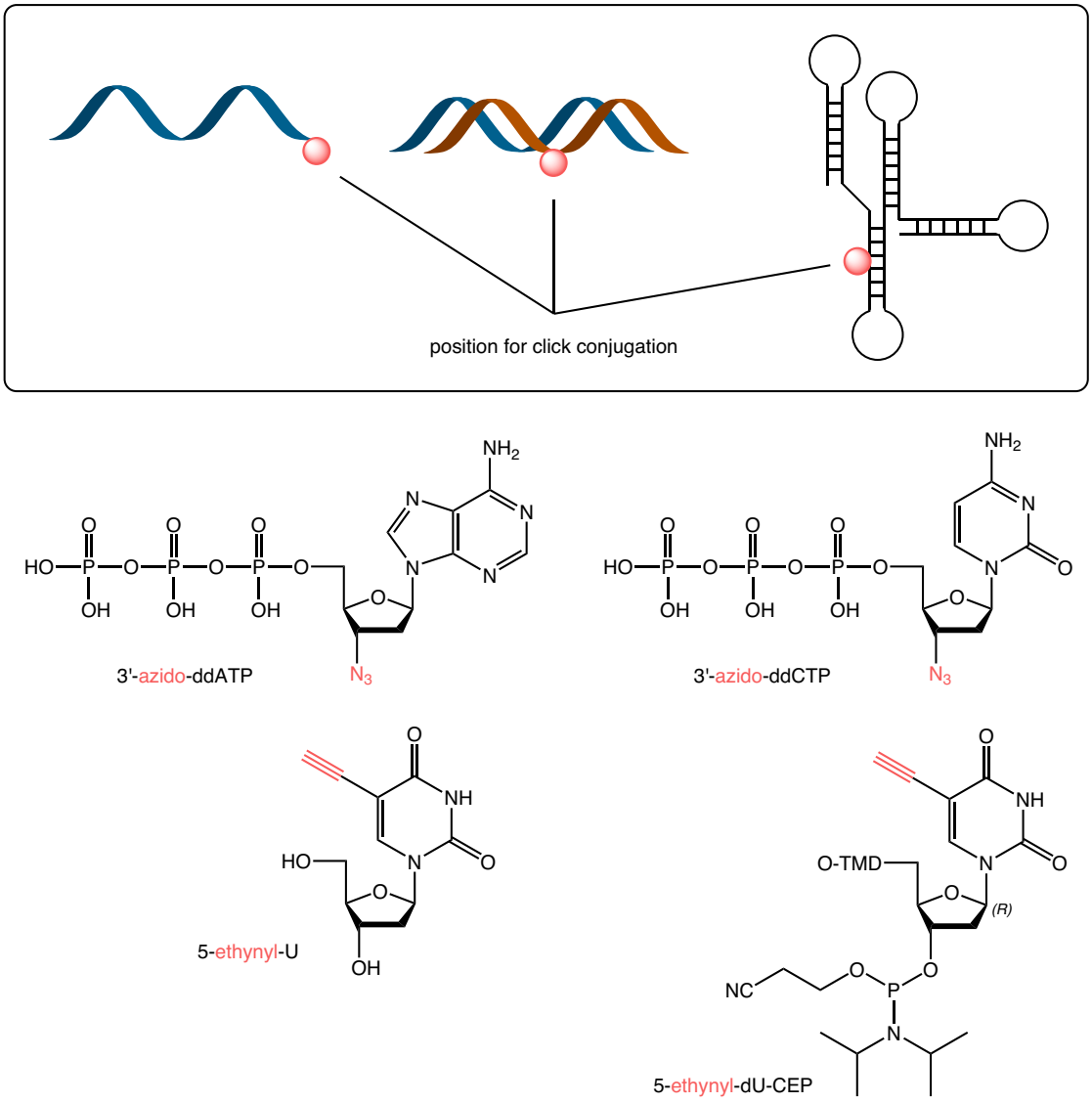


*Fig. 40: Alkyne and azido functions can be implemented at specific positions of oligonucleotide sequences by various building blocks.*

**With partners we have the possibility to provide modified oligonucleotide sequences ready for conjugation with suitable (self-immolative) linkers and payloads.**

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***References:***

- *Molecular Epitope Determination of Aptamer Complexes of the Multidomain Protein C-Met by Proteolytic Affinity-Mass Spectrometry; L. Lupu, P. Wiegand, N. Huttmann, S. Rawer, W. Kleinekofort, I. Shugureva, A. S. Kichkailo, F. N. Tomilin, A. Lazarev, M. V. Berezovski, M. Przybylski;* ***ChemMedChem*** *2020;* ***15****: 363-369. https://doi.org/10.1002/cmdc.201900489*
- *Chemoenzymatic Preparation of Functional Click-Labeled Messenger RNA; S. Croce, S. Serdjukow, T. Carell, T. Frischmuth;* ***Chembiochem : a European journal of chemical biology*** *2020;* ***21****: 1641-1646. https://doi.org/10.1002/cbic.201900718*
- *Exploring RNA transcription and turnover in vivo by using click chemistry; C. Y. Jao, A. Salic;* ***Proc Natl Acad Sci U S A*** *2008;* ***105****: 15779-84. https://doi.org/10.1073/pnas.0808480105*
- *Click Reaction on Solid Phase Enables High Fidelity Synthesis of Nucleobase-Modified DNA; F. Tolle, M. Rosenthal, F. Pfeiffer, G. Mayer;* ***Bioconjug Chem*** *2016;* ***27****: 500-3. https://doi.org/10.1021/acs.bioconjchem.5b00668*
- *Ethynyl side-chain hydration during synthesis and workup of „clickable" oligonucleotides: bypassing acetyl group formation by triisopropylsilyl protection; S. A. Ingale, H. Mei, P. Leonard, F. Seela;* ***J Org Chem*** *2013;* ***78****: 11271-82. https://doi.org/10.1021/jo401780u*
- *Synthesis of highly modified DNA by a combination of PCR with alkyne-bearing triphosphates and click chemistry; J. Gierlich, K. Gutsmiedl, P. M. Gramlich, A. Schmidt, G. A. Burley, T. Carell;* ***Chemistry*** *2007;* ***13****: 9486-94. https://doi.org/10.1002/chem.200700502*
- *Directed DNA metallization; G. A. Burley, J. Gierlich, M. R. Mofid, H. Nir, S. Tal, Y. Eichen, T. Carell;* ***J Am Chem Soc*** *2006;* ***128****: 1398-9. https://doi.org/10.1021/ja055517v*
- *A versatile modification of on-column oligodeoxynucleotides using a copper-catalyzed oxidative acetylenic coupling reaction; N. Minakawa, Y. Ono, A. Matsuda;* ***J Am Chem Soc*** *2003;* ***125****: 11545-52. https://doi.org/10.1021/ja036055t*
- *DNA duplexes stabilized by modified monomer residues: synthesis and stability; D. Graham, J. A. Parkinson, T. Brown; Journal of the Chemical Society,* ***Perkin Transactions 1*** *1998: 1131-1138. https://doi.org/10.1039/a707031d*

## 6.3. Carbon Compounds

Fullerenes and carbon nanotubes are subject of ongoing research as they possess unique geometrical shapes, as well as appealing photochemical, electrochemical, and physical properties. In addition, they act as efficient radical scavenger and antioxidant, as well as nano carrier for gene and drug delivery. Thus, a wide variety of operations can be considered. There are different options for conjugating linkers and payloads to such type of carriers.

a) Fullerenes carrying phenylbutyric methyl ester (PBM) can be activated by saponification followed by ester or amide formation with appropriate substitutions.

b) Fullerene derivatives with malonic acid moieties can react readily with nucleophiles, e.g., the amino functions of amino acids, amino-PEGs or other linkers and payloads enabling multiple payload decoration on one single fullerene.

c) Perhydroxylated fullerenols are a unique class of water-soluble fullerenes. Their alcohol functions can further be derivatized by ester or ether formation, e.g., *via* Mitsunobo reaction.

d) Fullerenes and carbon nanotubes carrying no functional group can be prepared for conjugation by photoaffinity labeling reagents such as aryl azides. Upon photolysis (258 nm), $N_2$ is liberated and a stabilized singlet perfluorophenylnitrene is being formed *in situ*, which reacts with neighboring molecules by insertion and addition reactions in moderate to good yields.

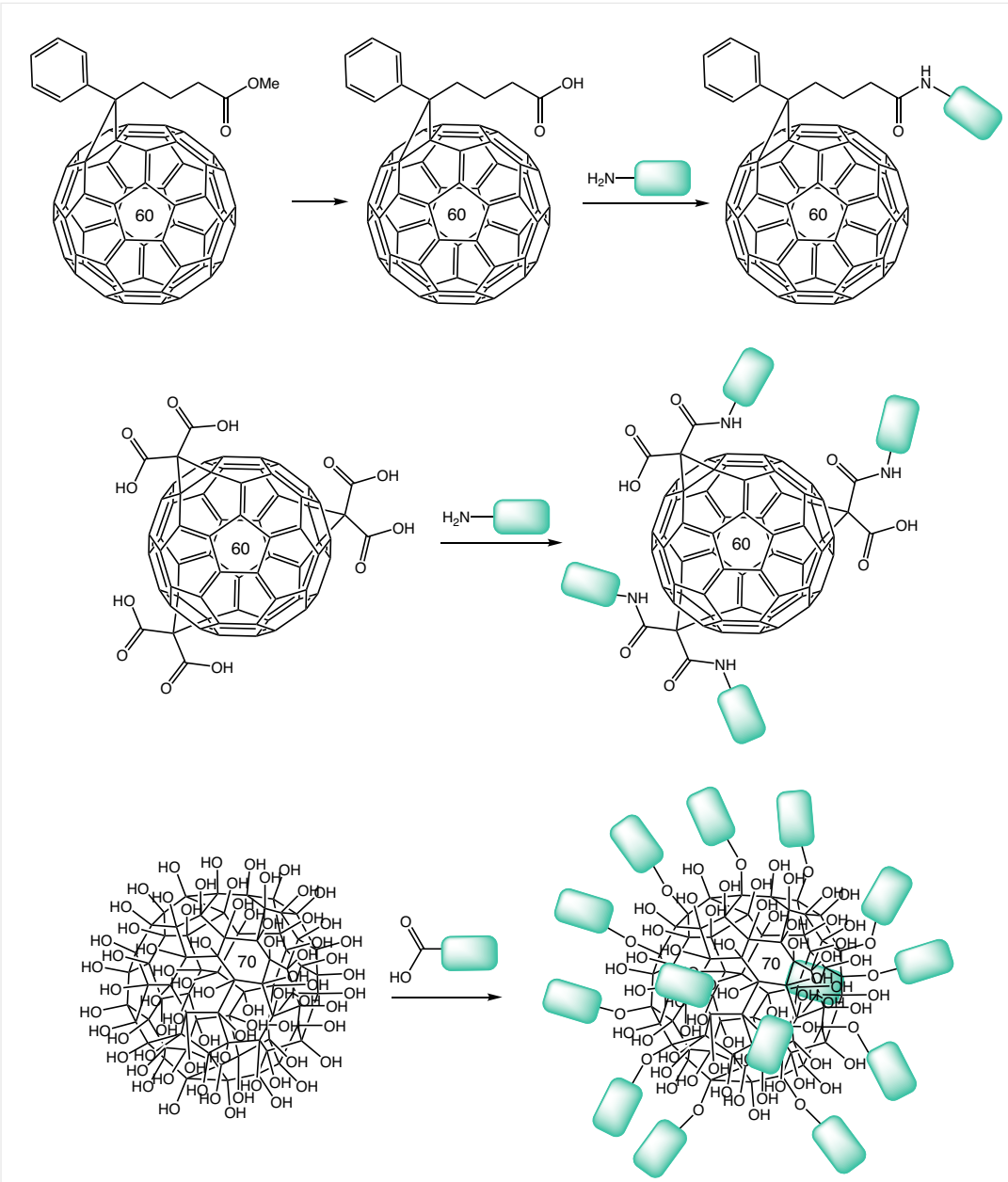


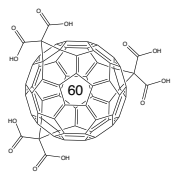

*Fig. 41: Pre-derivatized fullerenes allow direct surface conjugation.*

Product details

## FLL1070 Fullerene $C_{60}$ (malonic acid)n

**Buckminsterfullerene-n-(malonic acid)**

| | |
|---|---|
| Formula | $C_{60}(C_3H_2O_4)n$ |
| Mol. weight | 720,66+(102,05)n g/mol |

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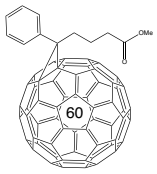
Product details

## FLL1020 Fullerene $C_{60}$ (PBM)

Fulleren-phenyl-(4-phenylbutyric acid methyl ester)

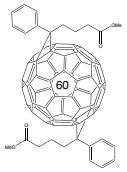
| | |
|---|---|
| CAS-No. | 160848-22-6 |
| Formula | $C_{72}H_{14}O_2$ |
| Mol. weight | 910,9 g/mol |



## FLL1010 Fullerene $C_{60}$ (PBM)2

Fulleren-diphenyl-bis(4-phenylbutyric acid methyl ester)

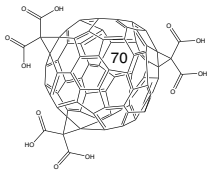
| | |
|---|---|
| CAS-No. | 1048679-01-1 |
| Formula | $C_{84}H_{28}O_4$ |
| Mol. weight | 1104,14 g/mol |



## FLL1080 Fullerene $C_{70}$ (malonic acid)n

Buckminsterfullerene-n-(malonic acid)

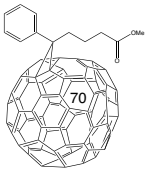
| | |
|---|---|
| Formula | $C_{70}(C_3H_2O_4)n$ |
| Mol. weight | 840,77+(102,05)n g/mol |



## FLL1060 Fullerene $C_{70}$ (PBM)

Fulleren-phenyl-(4-phenylbutyric acid methyl ester)

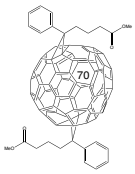
| | |
|---|---|
| CAS-No. | 609771-63-3 |
| Formula | $C_{82}H_{14}O_2$ |
| Mol. weight | 1031,01 g/mol |



## FLL1050 Fullerene $C_{70}$ (PBM)2

Fulleren-diphenyl-bis(4-phenylbutyric acid methyl ester)

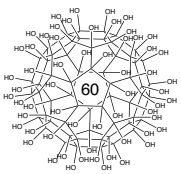
| | |
|---|---|
| Formula | $C_{94}H_{28}O_4$ |
| Mol. weight | 1221,25 g/mol |



## FLL1030 Fullerenol C60

Polyhydroxylated Fullerene

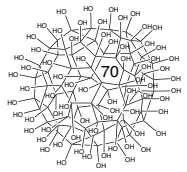
| | |
|---|---|
| Formula | $C_{60}(OH)n$ |
| Mol. weight | 720,66+(17,01)n g/mol |



## FLL1090 Fullerenol C70

Polyhydroxylated Fullerene

| | |
|---|---|
| Formula | $C_{70}(OH)n$ |
| Mol. weight | 840,77+(17,01)n g/mol |



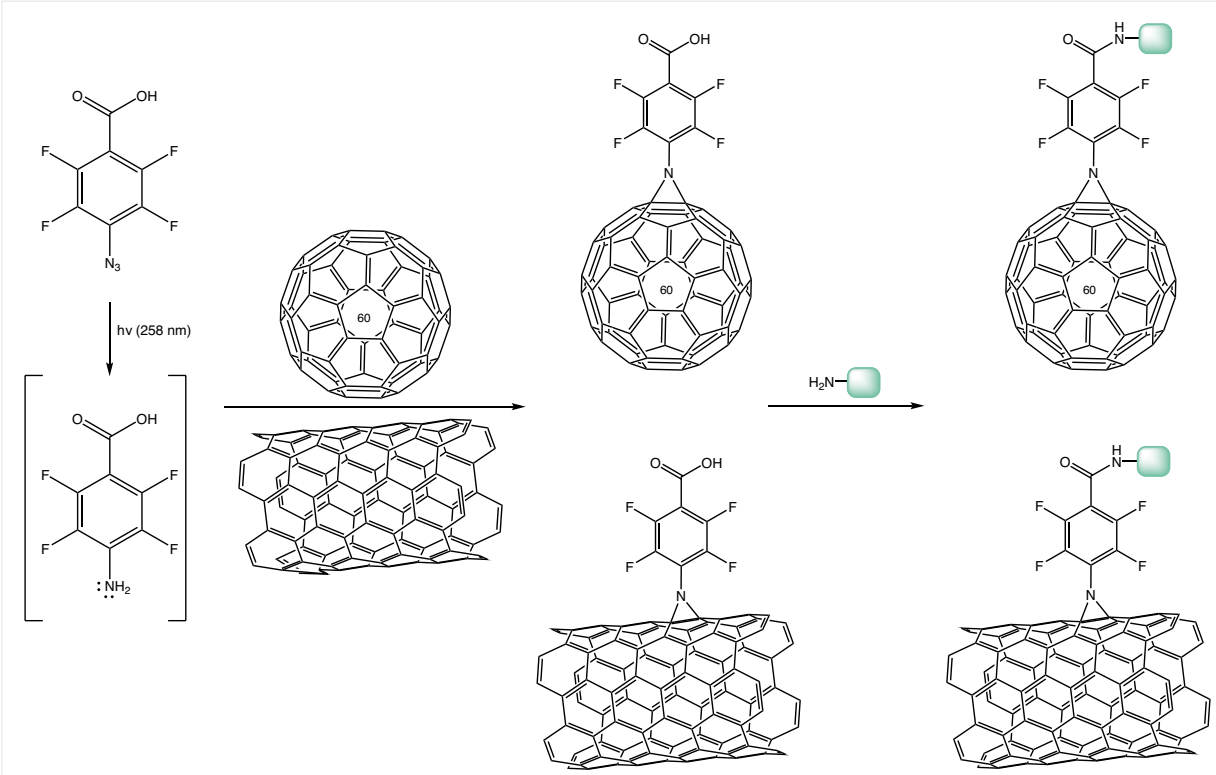



*Fig. 42: Carbon nanotubes and fullerenes can be surface decorated with carboxylic acid moieties for subsequent derivatization and linker attachment via photolysis of perfluoroarylazides which form in situ stable and reactive nitrenes.*

***References:***


- → *Fullerene derivatives with amino acids, peptides and proteins: From synthesis to biomedical application; E. I. Pochkaeva, N. E. Podolsky, D. N. Zakusilo, A. V. Petrov, N. A. Charykov, T. D. Vlasov, A. V. Penkova, L. V. Vasina, I. V. Murin, V. V. Sharoyko, K. N. Semenov;* ***Prog. Solid. State Ch.*** *2020;* ***57****: 100255.*
  *https://doi.org/10.1016/j.progsolidstchem.2019.100255*
- → *Fullerene-based delivery systems; H. Kazemzadeh, M. Mozafari;* ***Drug Discov Today*** *2019;* ***24****: 898-905.*
  *https://doi.org/10.1016/j.drudis.2019.01.013*
- → *Fullerenes in biology and medicine; E. Castro, A. H. Garcia, G. Zavala,, L. Echegoyen;* ***J. Mater. Chem. B.*** *2017;*
  *https://doi.org/10.1039/c7tb00855d*
- → *Fullerene C60 with cytoprotective and cytotoxic potential: prospects as a novel treatment agent in Dermatology? A. Rondags, W. Yan Yuen, M. F. Jonkman, B. Horváth;* ***Exp. Dermatol.*** *2017; 26 (3): 220-224.*
  *https://doi.org/10.1111/exd.13172*
- → *Water-soluble fullerenes for medical applications; I. Raovi;* ***Mater. Sci. Technol.*** *2016;* ***33****: 777-794.*
  *https://doi.org/10.1080/02670836.2016.1198114*


↑ back to content

- *Functionalized Fullerenes in Photodynamic Therapy; Y.-Y. Huang, S. K. Sharma, R. Yin, T. Agrawal, L. Y. Chiang, M. R. Hamblin;* ***J. Biomed. Nanotechnol.*** *2014;* ***10****: 1918-1936. https://doi.org/10.1166/jbn.2014.1963*
- *Fullerene-biomolecule conjugates and their biomedicinal applications; X. Yang, A. Ebrahimi, J. Li, Q. Cui;* ***Int. J. Nanomed.*** *2014;* ***19****: 77-92. http://dx.doi.org/10.2147/IJN.S52829*
- *Fullerenol Nanoparticles: Toxicity and Antioxidant Activity; R. Injac, M. Prijatelj, B. Strukelj; Oxidative Stress and Nanotechnology: Methods and Protocols. D. Armstrong, D. J. Bharali 2013: 75-100. https://doi.org/10.1007/978-1-62703-475-3_5*
- *Medicinal chemistry and pharmacological potential of fullerenes and carbon nanotubes; F. Cataldo, T. Da Ros; Springer Science & Business Media; 2008; 1.*
- *Medicinal applications of fullerenes; R. Bakry, R. M. Vallant, M. Najam-ul-Haq, M. Rainer, Z. Szabo, C. W. Huck, G. K. Bonn;* ***Int. J. Nanomedicine*** *2007;* ***2****: 639-649.*
- *Chapter 7 – Functionalization and application of [60]fullerene; A. Mateo-Alonso, D. Bonifazi, M. Prato;* ***Carbon Nanotechnology*** *2006; 155-189. https://doi.org/10.1016/B978-044451855-2/50010-3*
- *Fullerene derivatives: an attractive fool for biological applications; S. Bosi, T. Da Ros, G. Spalluto, M. Prato;* ***Eur. J. Med. Chem.*** *2003;* ***38****: 913-923. https://doi.org/10.1016/j.ejmech.2003.09.005*
- *Biological Applications of Fullerenes; A. W. Jensen, S. R. Wilson, D. I. Schuster;* ***Bioorg. Med. Chem.*** *1996;* ***4****: 767-779. https://doi.org/10.1016/0968.0896(96)00081-8*

## 6.4. Metals

Nanotechnology and nanobiotechnology using gold or silver particles are broadly diverse, rapidly expanding areas of study in medical diagnostics and therapeutics, sensorics and chemistry. Gold nanoparticles (AuNPs), particularly, have found a wide range of biomedical and environmental monitoring applications (drug delivery, diagnostics, biosensing, bio-imaging, theranostics, and hazardous chemical sensing) due to their excellent optoelectronic and enhanced physico-chemical properties.

Metal particles, however, are not water soluble without further modification. Due to the soft character of gold and sulfur, thiols readily form strong dative bonds to gold and silver surfaces creating a self-assembled monolayer (SAMs), which modifies surfaces for subsequent coupling of proteins, PEGs and other molecules. The formed nanoparticles show excellent stability and can be stored for years.

The bond between gold or silver surface atoms and monothiols is sensitive to reducing agents such as DTT (Cleland's Reagent), while the disulfide lipoic acid moiety (also known as thioctic acid) binds far stronger to metal surfaces and is much more resistant towards removal from the metal surface by DTT, TCEP and similar reagents than monothiols.

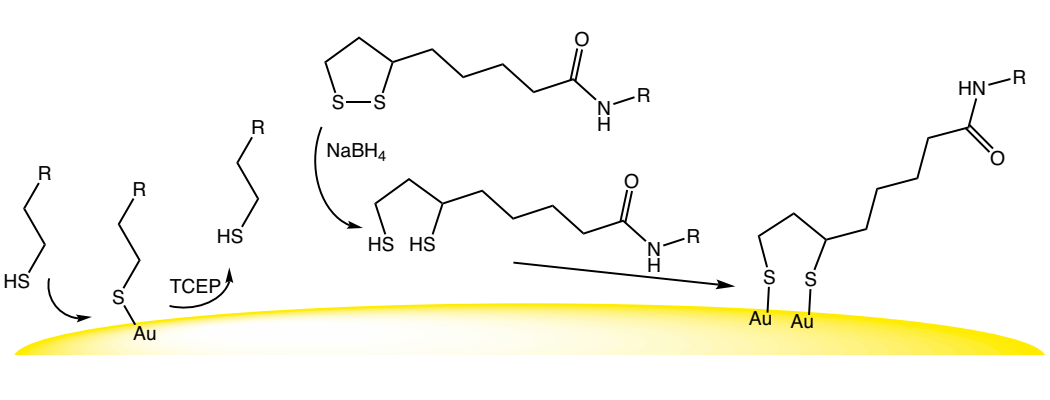



*Fig. 43: Thiols react instantly with gold and silver surfaces forming a strong metal-sulfur bond. Lipoic acid requires activation via reduction to the free dithiol and then binds much stronger to the surface as monothiols, which are sensitive to reducing agents.*

A variety of protocols exist in the literature for reducing lipoic acid to dihydrolipoic acid (DHLA), which binds instantly to the surface. Typically, tris(2-carboxyethyl)phosphine (TCEP) or sodium borohydride ($NaBH_4$) are being used as reducing agents. In general, TCEP reduction is carried out in water or aqueous buffer (excluding phosphate buffer, in which TCEP is unstable), in three times or greater molar excess to the lipoic acid derivative, using an incubation temperature of 25 °C to 50 °C, for about 1-2 hours. Each reduction procedure must be optimized for the particular lipoic acid derivative being reduced to the corresponding DHLA derivative.

Lipoamido-PEG-acids and lipoamido-PEG-alcohols can be used as intermediates for further derivatization after attachment to the surface. The density of functional groups on the surface can be tuned by co-coating the bifunctional mercapto- or lipoic-PEG with methoxy-PEG-lipoamides.

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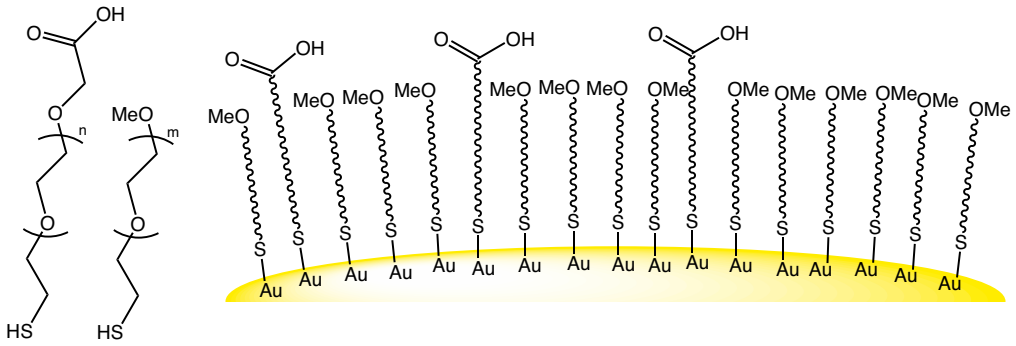


*Fig. 44: The density of functional groups can be tuned by co-coating of a PEG-thiol bearing a reactive group such as carboxylic acid with a non-reactive methoxy terminated PEG-thiol.*

***References:***


- *Gold nanoparticle surface engineering strategies and their applications in biomedicine and diagnostics; K. Mahato, S. Nagpal, M. A. Shah, A. Srivastava, P. K. Maurya, S. Roy, A. Jaiswal, R. Singh, P. Chandra;* ***3 Biotech*** *2019;* ***9****: 57.* https://doi.org/10.1007/s13205-019-1577-z
- *Recent advances in separation and detection methods for thiol compounds in biological samples; T. Toyo'oka;* ***J Chromatogr B Analyt Technol Biomed Life Sci*** *2009;* ***877****: 3318-30.* https://doi.org/10.1016/j.jchromb.2009.03.034
- *Polyethylene glycol-based bidentate ligands to enhance quantum dot and gold nanoparticle stability in biological media; B. C. Mei, K. Susumu, I. L. Medintz, H. Mattoussi;* ***Nat Protoc*** *2009;* ***4****: 412-23.* https://doi.org/10.1038/nprot.2008.243
- *Influence of anchoring ligands and particle size on the colloidal stability and in vivo biodistribution of polyethylene glycol-coated gold nanoparticles in tumor-xenografted mice; G. Zhang, Z. Yang, W. Lu, R. Zhang, Q. Huang, M. Tian, L. Li, D. Liang, C. Li;* ***Biomaterials*** *2009;* ***30****: 1928-36.* https://doi.org/10.1016/j.biomaterials.2008.12.038
- *Bioconjugate Techniques (Third Edition); G. T. Hermanson; 2013: 1146.* https://doi.org/10.1016/C2009-0-64240-9
- *Toward reliable gold nanoparticle patterning on self-assembled DNA nanoscaffold; J. Sharma, R. Chhabra, C. S. Andersen, K. V. Gothelf, H. Yan, Y. Liu;* ***J Am Chem Soc*** *2008;* ***130****: 7820-1.* https://doi.org/10.1021/ja802853r
- *Modular poly(ethylene glycol) ligands for biocompatible semiconductor and gold nanocrystals with extended pH and ionic stability; B. C. Mei, K. Susumu, I. L. Medintz, J. B. Delehanty, T. J. Mountziaris, H. Mattoussi;* ***Journal of Materials Chemistry*** *2008;* ***18****: 4949-4958.* https://doi.org/10.1039/b810488c
- *Oriented immobilization of antibodies with GST-fused multiple Fc-specific B-domains on a gold surface; T. H. Ha, S. O. Jung, J. M. Lee, K. Y. Lee, Y. Lee, J. S. Park, B. H. Chung;* ***Anal Chem*** *2007;* ***79****: 546-56.* https://doi.org/10.1021/ac061639+
- *Design of biotin-functionalized luminescent quantum dots; K. Susumu, H. T. Uyeda, I. L. Medintz, H. Mattoussi;* ***J Biomed Biotechnol*** *2007;* ***2007****: 90651.* https://doi.org/10.1155/2007/90651
- *Simultaneous determination of alpha-lipoic acid and its reduced form by high-performance liquid chromatography with fluorescence detection; S. Satoh, T. Toyo'oka, T. Fukushima, S. Inagaki;* ***J Chromatogr B Analyt Technol Biomed Life Sci*** *2007;* ***854****: 109-15.* https://doi.org/10.1016/j.jchromb.2007.04.003
- *Enhanced oligonucleotide-nanoparticle conjugate stability using thioctic acid modified oligonucleotides; J. A. Dougan, C. Karlsson, W. E. Smith, D. Graham;* ***Nucleic Acids Res*** *2007;* ***35****: 3668-75.* https://doi.org/10.1093/nar/gkm237
- *Biosensing with Luminescent Semiconductor Quantum Dots; K. Sapsford, T. Pons, I. Medintz, H. Mattoussi;* ***Sensors*** *2006;* ***6****: 925-953.*
- *Synthesis and reactions of functionalised gold nanoparticles; M. Brust, J. Fink, D. Bethell, D. J. Schiffrin, C. Kiely;* ***Journal of the Chemical Society, Chemical Communications*** *1995: 1655-1656.* https://doi.org/10.1039/c39950001655
- *Self-assembled organic monolayers: model systems for studying adsorption of proteins at surfaces; K. L. Prime, G. M. Whitesides;* ***Science*** *1991;* ***252****: 1164-7.* https://doi.org/10.1126/science.252.5009.1164

## 6.5. Metal Oxides

Nanotechnology and nanobiotechnology using quantum dots, magnetic particles, or metal oxides are broadly diverse, rapidly expanding areas of study in medical diagnostics and therapeutics, sensoric and chemistry. Many metal oxides are not water soluble without further modification.

Compounds containing functional groups which enable noncovalent binding or chelates to oxide surfaces and which carry residues like poly(ethylene glycol) (PEG) or other polymers equip metal oxide nanoparticles with colloidal stability and stealthiness.

Such functional groups are phosphonic acid compounds R–$H_2PO_3$ which display a strong affinity to metal ionic centers and are characterized by their multidentate binding ability. With appropriate residues the self-assembled monolayer (SAM) and multilayers can be designed. Examples have been published of cerium ($CeO_2$), iron ($\gamma$-$Fe_2O_3$), aluminum ($Al_2O_3$), and titanium ($TiO_2$) oxides of different sizes and morphologies.

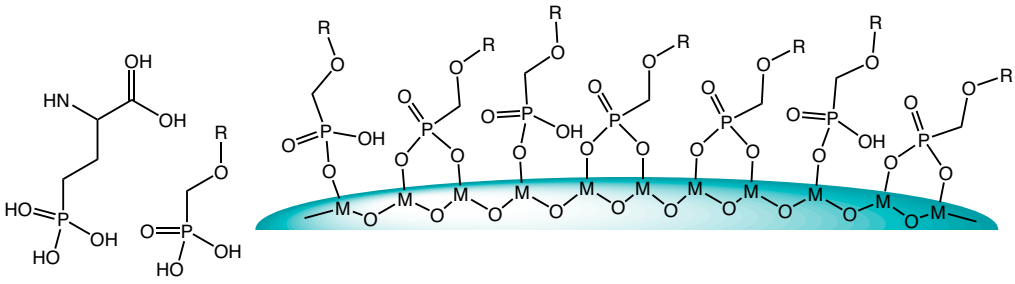

*Fig. 45: Metal oxide surfaces form stable bonds with phosphates and phosphonates by multidental, chelating interaction.*

***References:***


→ *Versatile Coating Platform for Metal Oxide Nanoparticles: Applications to Materials and Biological Science; J. F. Berret, A. Graillot;* ***Langmuir*** *2022;* ***38****: 5323-5338.* https://doi.org/10.1021/acs.langmuir.2c00338

→ *Efficient modification of metal oxide surfaces with phosphonic acids by spray coating; A. Bulusu, S. A. Paniagua, B. A. MacLeod, A. K. Sigdel, J. J. Berry, D. C. Olson, S. R. Marder, S. Graham;* ***Langmuir*** *2013;* ***29****: 3935-42.* https://doi.org/10.1021/la303354t


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## 6.6. Polymeric Surfaces by Plasma Treatment

Plastic polymers, like polyethylene, polystyrene, PTFE, or co-polymers thereof are materials used in many facets of daily life, including tools and devices with biological, human, and pharmacologic applications. A major property of such materials is that they are rather chemically inert to most environmental conditions.

Low pressure plasma treatment offers a new method to decorate such polymers with specific functional groups, such as amines or carboxylic acids, which offer the opportunity to further modify the surface with specific molecules. PEGs, for example, can turn such usually quite hydrophobic surfaces very hydrophilic. Attachments of fluorescent dyes will stain the particles accordingly. Attachment of peptides, proteins, like streptavidin, or antibodies opens the door to numerous biological and pharmacological applications.

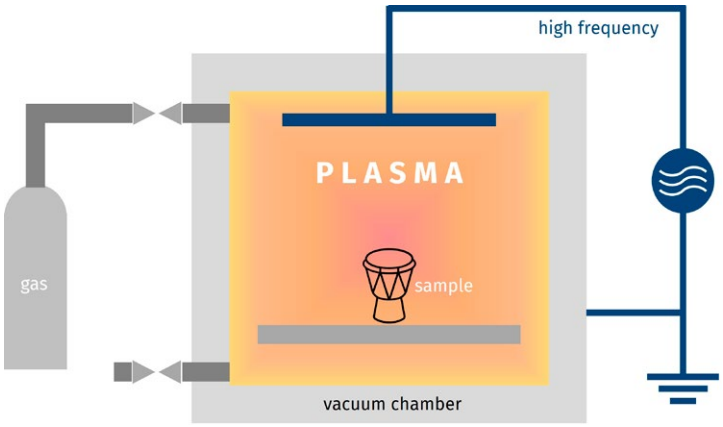


*Fig. 46: Plasma chamber for surface coating of samples with functional groups provided by the selected gas.*

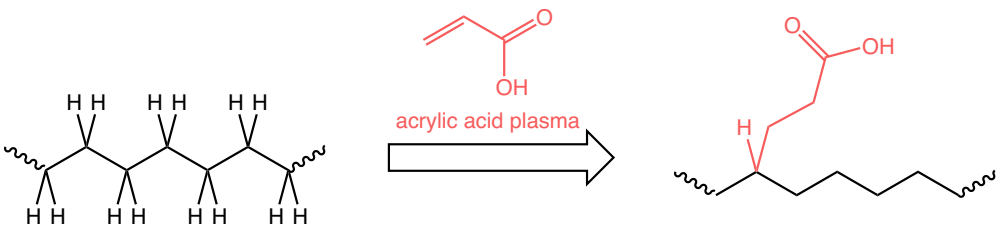


*Fig. 47: Introduction of carboxylic acid functions on polyethylene surfaces via acrylic acid plasma treatment.*

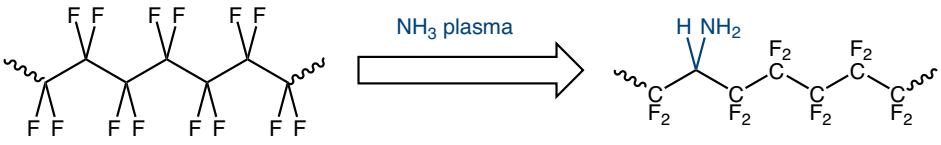


*Fig. 48: Introduction of amino functions on PTFE surfaces via ammonia plasma treatment.*

One major drawback of working on surfaces is the limitation of analytical methods, as conventional technologies, like mass spectroscopy, chromatic purification technologies, and other methodologies widely established for small molecules and biologics are not applicable in this case. For surface analytics X-ray photoelectron spectroscopy (XPS) is the method of choice. It is also known as ESCA (Electron Spectroscopy for Chemical Analysis) and is an established method for the analysis of chemical compositions of surfaces. It enables a highly sensitive, quantitative detection of all elements except hydrogen and helium, as well as for the identification of binding and oxidation states on solid surfaces. The method is very sensitive to surfaces, so that even very thin layers can be studied (2 nm to 10 nm depth of information).

The basic principle of XPS is based on the irradiation of a sample surface in vacuum with soft X-rays and analyzing the energy of the emitted photoelectrons. This energy is different from element to element and also depends on the oxidative level of the element. Therefore, analyzing a carbon signal a quantitative determination e.g., between carbon-hydrogen, carbon-nitrogen and carbonyl carbon can be measured. The XPS spectrum results from plotting the number of detected electrons per energy interval against their kinetic energy.

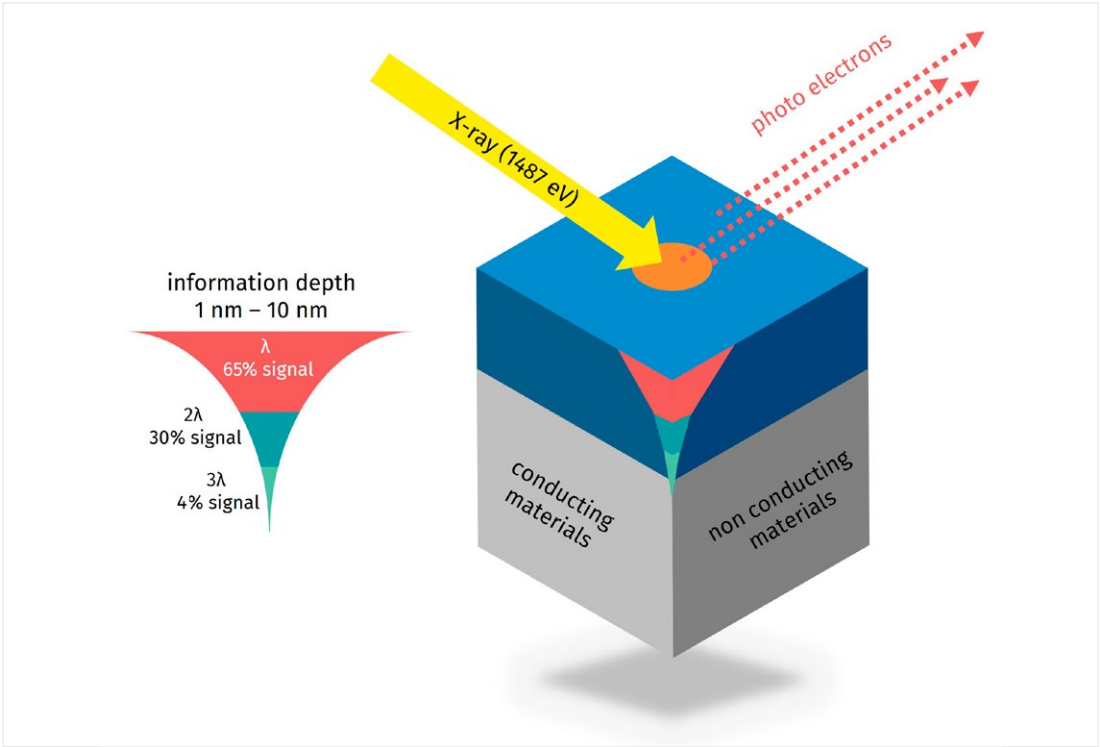


*Fig. 49: Principle of X-ray photoelectron spectroscopy and information retrieved by this technique.*

Information gained with X-ray photoelectron spectroscopy:

- Detection limit: 0.01 - 1 at%, sub-monolayer
- Detectable elements: Li – U
- Chemical bonding information
- Quantitative information
- Information depth: 1 nm to 10 nm
- Lateral resolution: ca. 30 µm
- Depth profiling
- Imaging/mapping

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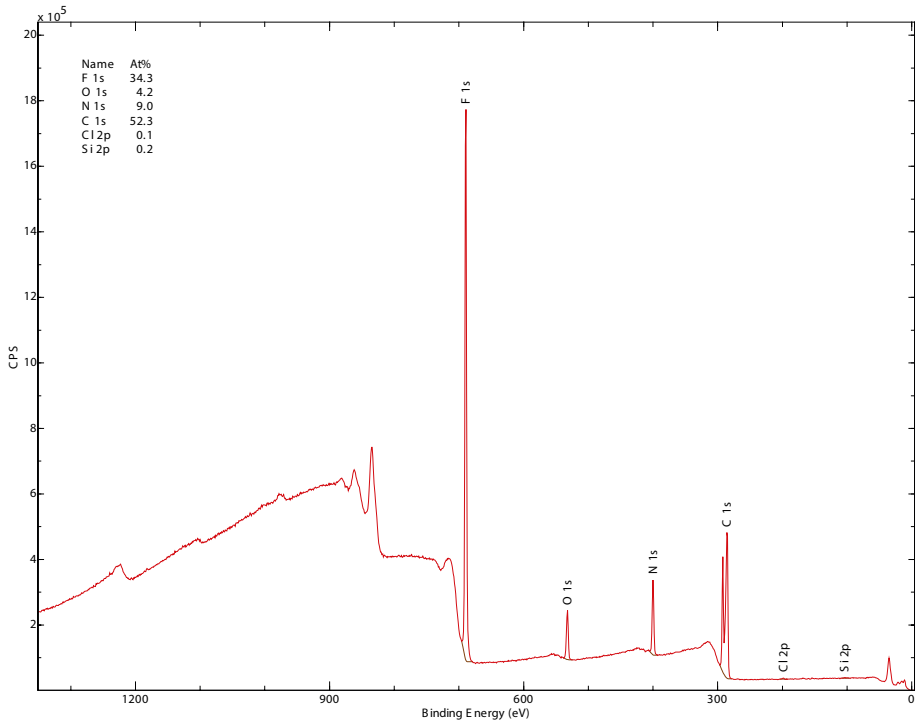



*Fig. 50: XPS representing signals of the elements sodium, fluorine, oxygen, nitrogen, calcium, carbon, chlorine, and silicon. The peak area allows quantitative determination of each element.*

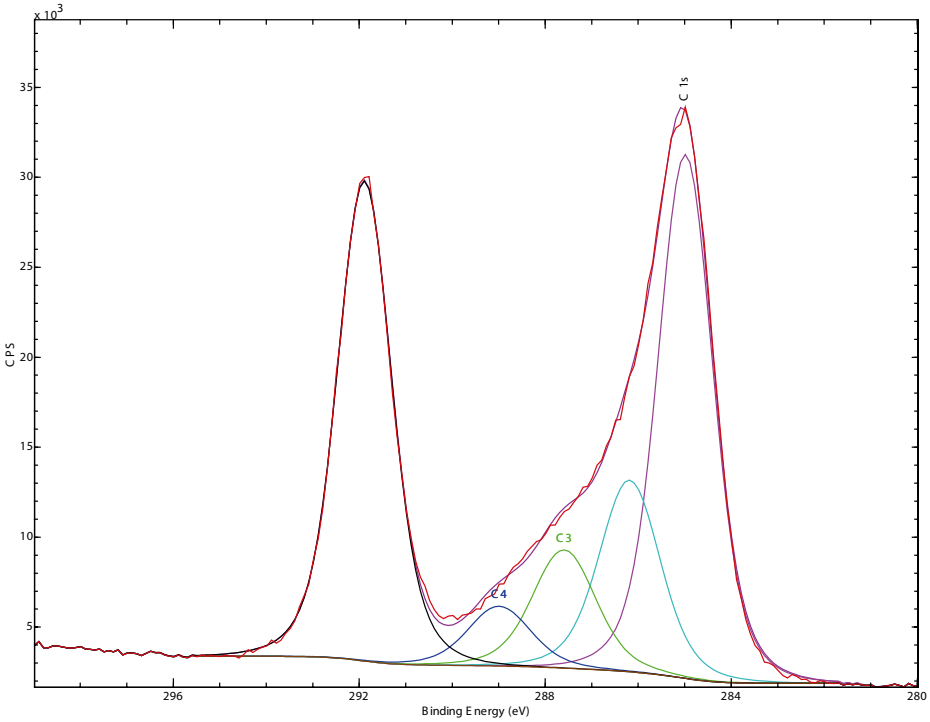



*Fig. 51: Expanded XPS spectrum of a carbon signal showing the ratio between C1 (aliphatic carbon), C2 (carbon with single bond to a hetero atom N or O), C3 (carbonyl, amide carbon), C4 (carboxyl, carbonate carbon), and C5 (fluorinated carbon).*

*Services available from us:*

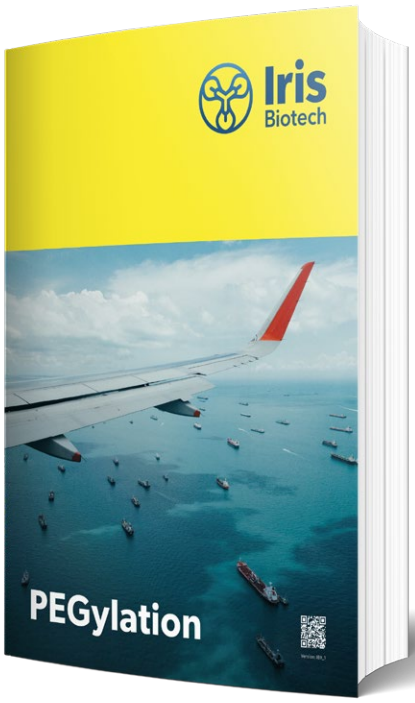
- *Beads of polyethylene, polystyrene, teflon or co-polymers thereof should be surface decorated with permanent or cleavable linkers and loaded with small or biomolecules.*
- *Turning polymer-based surface of devices hydrophilic, biocompatible or non-immunogenic.*
- *Equipping the surface of membranes, devices or other parts made of a certain material with an alternative or orthogonal property.*

## Consult with us, we will be happy to carry your application through to market.

***References:***


- *Characterisation of PEGylated PLGA nanoparticles comparing the nanoparticle bulk to the particle surface using UV/vis spectroscopy, SEC, 1H NMR spectroscopy, and X-ray photoelectron spectroscopy; S. Spek, M. Haeuser, M. M. Schaefer, K. Langer;* ***Applied Surface Science*** *2015;* ***347****: 378-385. https://doi.org/10.1016/j.apsusc.2015.04.071*
- *Reaction of human macrophages on protein corona covered TiO(2) nanoparticles; C. F. Borgognoni, M. Mormann, Y. Qu, M. Schafer, K. Langer, C. Ozturk, S. Wagner, C. Chen, Y. Zhao, H. Fuchs, K. Riehemann;* ***Nanomedicine*** *2015;* ***11****: 275-82. https://doi.org/10.1016/j.nano.2014.10.001*
- *Engineering Biomaterials Surfaces Using Micropatterning; L. Gagne, G. Laroche;* ***Advanced Materials Research*** *2006;* ***15-17****: 77-82. https://doi.org/10.4028/www.scientific.net/AMR.15-17.77*
- *Quantification of cation-exchanged zeolites by XPS and EDS: A comparative study; S. Fibikar, M. T. Rinke, A. Schäfer, L. D. Cola;* ***Microporous and Mesoporous Materials*** *2010;* ***132****: 296-299. https://doi.org/10.1016/j.micromeso.2010.02.016*
- *Comparison of Atmospheric-Pressure Plasma versus Low-Pressure RF Plasma for Surface Functionalization of PTFE for Biomedical Applications; C. Sarra-Bournet, S. Turgeon, D. Mantovani, G. Laroche;* ***Plasma Processes and Polymers*** *2006;* ***3****: 506-515. https://doi.org/10.1002/ppap.200600012*
- *Fast element mapping of titanium wear around implants of different surface structures; U. Meyer, M. Buhner, A. Buchter, B. Kruse-Losler, T. Stamm, H. P. Wiesmann;* ***Clin Oral Implants Res*** *2006;* ***17****: 206-11. https://doi.org/10.1111/j.1600-0501.2005.01184.x*


## Please find a variety of trimethylsilyl-PEG derivatives in our PEGylation Brochure.

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## 6.7. Silicates

Glass, quartz and silicates presenting silanol groups at their surface, which can further react with different silane reagents, such as chlorosilanes or alkoxysilanes. It can help to etch the surface prior to coating with acids to remove an outer non-reactive layer and expose silanol groups making them available to form silicon-oxygen-silicon structures and attaching a coating molecule on the surface.

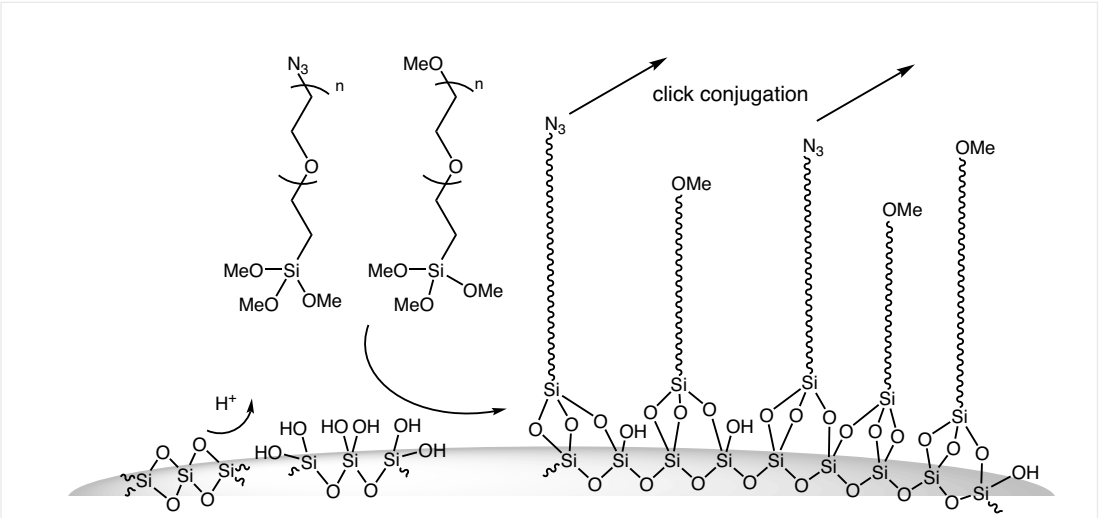


*Fig. 52: Silicate-type surfaces eventually require activation by acidic etching prior to reaction with silanizing reagents. Then trimethylsilyl-PEG-azides can conjugate to the surface. The terminal azido functions can then further coupled to Click reactive compounds. The density of functional groups can be diluted and fine-tuned by co-coating with non-reactive methoxy terminated trimethylsilyl-PEG compounds.*

***References:***


→ *Low cost and scalable method for modifying surfaces of hollow particles from hydrophilic to hydrophobic; J. Sharma, G. Polizos, D. Hun, K. Nawaz, R. Sahore;* ***RSC Adv*** *2020;* ***10****: 31065-31069.*
*https://doi.org/10.1039/d0ra06114j*

→ *Silica and Silane based polymer composite coating on glass slide by dip-Coating Method; S. Sriram, R. K. Singh, A. Kumar;* ***Surfaces and Interfaces*** *2020;* ***19****: 100472.*
*https://doi.org/10.1016/j.surfin.2020.100472*

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| PEG1495 | Z-NH-PEG(4)-COOH | 31 |
| ZAA1186 | Z-$O_2$Oc-OH*DCHA | 23 |
| ZNN1120 | Z-TOTA | 19 |

# Notes

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# Notes

# Code of Conduct

As business activity of Iris Biotech GmbH impacts people's lives and health, it must be operated in ethical and correct manner and act with integrity and responsibility. To ensure high ethical standards and fair business practices, Iris Biotech GmbH applies an integrated policy known as its Code of Conduct.

In 2001 Iris Biotech GmbH was founded just at the beginning of the Biotech movement and the first remarkable breakthrough of biotech pharma products. Although the biotech field is rather young compared to other industries we believe on long-term business, a good partnership between our business partners and Iris Biotech GmbH and a good reputation. It is our duty as well as our responsibility to maintain and to extend this over the next generations – based on the principles of an honourable and prudent tradesman which based upon the concept of honourable entrepreneurship.

This Code of Conduct has been developed following the "Voluntary Guidelines for Manufacturers of Fine Chemical Intermediates and Active Ingredients" issued by AIME (Agrochemical & Intermediates Manufacturers in Europe) and the requirements of some of our business associates.

Iris Biotech GmbH commits to hold this Code of Conduct and to include and apply its principles in the management system and the company policies.

## Ethics

Iris Biotech GmbH undertakes business in an ethical manner and acts with integrity. All corruption, extortion and embezzlement are prohibited. We do not pay or accept bribes or participate in other illegal inducements in business or government relationships. We conduct our business in compliance with all applicable anti-trust laws. Employees are encouraged to report concerns or illegal activities in the workplace, without threat of reprisal, intimidation or harassment.

## Labour

Iris Biotech GmbH is committed to uphold the human rights of workers and to treat them with dignity and respect. Child labour, workplace harassment, discrimination, and harsh and inhumane treatment are prohibited. Iris Biotech GmbH respects the rights of the employees to associate freely, join or not join labour unions, seek representation and join workers' councils. Employees are paid and their working timetable is established according to applicable wage and labour laws. Employees are able to communicate openly with management regarding working conditions without threat of reprisal, intimidation or harassment.

## General Policies

Contracts and Secrecy Agreements are binding and the confidential information received is only used for intended purposes. Clear management and organizational structures exist to provide efficient normal working and to address problems quickly. Know-how is protected and intellectual property isrespected.

## Health and Safety

Iris Biotech GmbH provides a safe and healthy working environment to the employees and protects them from overexposure to chemical and physical hazards. Products are produced, stored and shipped under the guidelines of the relevant chemical and safety legislation. Risks and emergency scenarios are identified and evaluated, and their possible impact is minimized by implementing emergency plans and written procedures. Safety information regarding hazardous materials is available to educate, train and protect workers from hazards. Preventive equipment and facilities maintenance is performed at suitable periods to reduce potential hazards. Employees are regularly trained in health and safety matters and are informed about product properties and risk classification when it is required.

## Environment

Iris Biotech GmbH operates in an environmentally responsible and efficient manner, minimizing adverse impacts on the environment. Waste streams are managed to ensure a safe handling, movement, storage, recycling and reuse, before and after being generated. Systems to prevent and mitigate accidental spills and releases to the environment are in place. All required environmental permits and licenses are obtained and their operational and reporting requirements are complied with.

## Production and Quality Management

A quality management system following the Good Distribution Practices (GDP rules) of Active Pharmaceutical Ingredients is established covering all the aspects of the worldwide distribution of products. Regular audits are performed to evaluate the efficiency and fulfilling of the quality system. Process controls to provide reproducible product quality are established. There are preventive maintenance procedures to ensure plant reliability and the lowest risk of failure. Staff is trained periodically about GMP and GDP rules. Procedures are established and installations are designed to avoid cross contamination. Batch and analytical records are kept for inspection and audit purposes for suitable periods according guidelines.

## Research and Development

Research and development staff education is appropriate to their functional activity and they are trained to develop, optimize and scale-up the processes. Intellectual property is respected and know-how protected. Development of manufacturing processes reflects the principles of the Green Chemistry according to the American Chemical Society Green Chemistry Institute. Animal testing is not used unless alternatives are not scientifically valid or accepted by regulators. If animal testing is carried out, animals are treated so that pain and stress are minimized.

# Terms and Conditions of Sales

All orders placed by a buyer are accepted and all contracts are made subject to the terms which shall prevail and be effective notwithstanding any variations or additions contained in any order or other document submitted by the buyer. No modification of these terms shall be binding upon Iris Biotech GmbH unless made in writing by an authorised representative of Iris Biotech GmbH.

## Placing of Orders

Every order made by the buyer shall be deemed an offer by the buyer to purchase products from Iris Biotech GmbH and will not be binding on Iris Biotech GmbH until a duly authorised representative of Iris Biotech GmbH has accepted the offer made by the buyer. Iris Biotech GmbH may accept orders from commercial, educational or government organisations, but not from private individuals and Iris Biotech GmbH reserves the right to insist on a written order and/or references from the buyer before proceeding.

There is no minimum order value. At the time of acceptance of an order Iris Biotech GmbH will either arrange prompt despatch from stock or the manufacture/acquisition of material to satisfy the order. In the event of the latter Iris Biotech GmbH will indicate an estimated delivery date. In addition to all its other rights Iris Biotech GmbH reserves the right to refuse the subsequent cancellation of the order if Iris Biotech GmbH expects to deliver theproduct on or prior to the estimated delivery date. Time shall not be of the essence in respect of delivery of the products. If Iris Biotech GmbH is unable to deliver any products by reason of any circumstances beyond its reasonable control („Force Majeure“) then the period for delivery shall be extended by the time lost due to such Force Majeure. Details of Force Majeure will be forwarded by Iris Biotech GmbH to the buyer as soon as reasonably practicable.

## Prices, Quotations and Payments

Prices are subject to change. For the avoidance of doubt, the price advised by Iris Biotech GmbH at the time of the buyer placing the order shall supersede any previous price indications. The buyer must contact the local office of Iris Biotech GmbH before ordering if further information is required. Unless otherwise agreed by the buyer and Iris Biotech GmbH, the price shall be for delivery ex-works. In the event that the buyer requires delivery of the products otherwise than ex-works the buyer should contact the local office of Iris Biotech GmbH in order to detail its requirements. Iris Biotech GmbH shall, at its discretion, arrange the buyer‘s delivery requirements including, without limitation, transit insurance, the mode of transit (Iris Biotech GmbH reserves the right to vary the mode of transit if any regulations or other relevant considerations so require) and any special packaging requirements (including cylinders). For the avoidance of doubt all costs of delivery and packaging in accordance with the buyer‘s requests over and above that of delivery in standard packaging ex-works shall be for the buyer‘s account unless otherwise agreed by both parties. Incoterms 2020 shall apply. Any tax, duty or charge imposed by governmental authority or otherwise and any other applicable taxes, duties or charges shall be for the buyer‘s account. Iris Biotech GmbH may, on request and where possible, provide quotations for multiple packs or bulk quantities, and non-listed items. Irrespective of the type of request or means of response all quotations must be accepted by the buyer without condition and in writing before an order will be accepted by Iris Biotech GmbH. Unless agreed in writing on different terms, quotations are valid for 30 days from the date thereof. Payment terms are net 30 days from invoice date unless otherwise agreed in writing. Iris Biotech GmbH reserves the right to request advance payment at its discretion. For overseas transactions the buyer shall pay all the banking charges of Iris Biotech GmbH. The buyer shall not

be entitled to withhold or set-off payment for the products for any reason whatsoever. Government/ Corporate Visa and MasterCard (and other such credit cards) may be accepted on approved accounts for payment of the products. Personal credit cards are not acceptable. Failure to comply with the terms of payment of Iris Biotech GmbH shall constitute default without reminder. In these circumstances Iris Biotech GmbH may (without prejudice to any other of its rights under these terms) charge interest to accrue on a daily basis at the rate of 2% per month from the date upon which payment falls due to the actual date of payment (such interest shall be paid monthly). If the buyer shall fail to fulfil the payment terms in respect of any invoice of Iris Biotech GmbH Iris Biotech GmbH may demand payment of all outstanding balances from the buyer whether due or not and/or cancel all outstanding orders and/or decline to make further deliveries or provision of services except upon receipt of cash or satisfactory securities. Until payment by the buyer in full of the price and any other monies due to Iris Biotech GmbH in respect of all other products or services supplied or agreed to be supplied by Iris Biotech GmbH to the buyer (including but without limitation any costs of delivery) the property in the products shall remain vested in Iris Biotech GmbH.

## Shipping, Packaging and Returns

The buyer shall inspect goods immediately on receipt and inform Iris Biotech GmbH of any shortage or damage within five days. Quality problems must be notified within ten days of receipt. Goods must not be returned without prior written authorisation of Iris Biotech GmbH. Iris Biotech GmbH shall at its sole discretion replace the defective products (or parts thereof) free of charge or refund the price (or proportionate price) to buyer. Opened or damaged containers cannot be returned by the buyer without the written prior agreement of Iris Biotech GmbH. In the case of agreed damaged containers which cannot be so returned, the buyer assumes responsibility for the safe disposal of such containers in accordance with all applicable laws.

## Product Quality, Specifications and Technical Information

Products are analysed in the Quality Control laboratories of Iris Biotech GmbH's production partners by methods and procedures which Iris Biotech GmbH considers appropriate. In the event of any dispute concerning reported discrepancies arising from the buyer's analytical results, determined by the buyer's own analytical procedures, Iris Biotech GmbH reserves the right to rely on the results of own analytical methods of Iris Biotech GmbH. Certificates of Analysis or Certificates of Conformity are available at the discretion of Iris Biotech GmbH for bulk orders but not normally for prepack orders. Iris Biotech GmbH reserves the right to make a charge for such certification. Specifications may change and reasonable variation from any value listed should not form the basis of a dispute. Any supply by Iris Biotech GmbH of bespoke or custom product for a buyer shall be to a specification agreed by both parties in writing. Technical information, provided orally, in writing, or by electronic means by or on behalf of Iris Biotech GmbH, including any descriptions, references, illustrations or diagrams in any catalogue or brochure, is provided for guidance purposes only and is subject to change.

## Safety

All chemicals should be handled only by competent, suitably trained persons, familiar with laboratory procedures and potential chemical hazards. The burden of safe use of the products of Iris Biotech GmbH vests in the buyer. The buyer assumes all responsibility for warning his employees, and any persons who might reasonably be expected to come into contact with the products, of all risks to person and property in any way connected with the products and for instructing them in their safe handling and use. The buyer also assumes the responsibility for the safe disposal of all products in accordance with all applicable laws.

## Uses, Warranties and Liabilities

All products of Iris Biotech GmbH are intended for laboratory research purposes and unless otherwise stated on product labels, in the catalogue and product information sheet of Iris Biotech GmbH or in other literature furnished to the buyer, are not to be used for any other purposes, including but not limited to use as or as components in drugs for human or animal use, medical devices, cosmetics, food additives, household chemicals, agricultural or horticultural products or pesticides. Iris Biotech GmbH offers no warranty regarding the fitness of any product for a particular purpose and shall not be responsible for any loss or damage whatsoever arising there from. No warranty or representation is given by Iris Biotech GmbH that the products do not infringe any letters patent, trademarks, registered designs or other industrial rights. The buyer further warrants to Iris Biotech GmbH that any use of the products in the United States of America shall not result in the products becoming adulterated or misbranded within the meaning of the Federal Food, Drug and Cosmetic Act (or such equivalent legislation in force in the buyer's jurisdiction) and shall not be materials which may not, under sections 404, 505 or 512 of the Act, be introduced into interstate commerce. The buyer acknowledges that, since the products of Iris Biotech GmbH are intended for research purposes, they may not be on the Toxic Substances Control Act 1976 („TSCA") inventory. The buyer warrants that it shall ensure that the products are approved for use under the TSCA (or such other equivalent legislation in force in the buyer's jurisdiction), if applicable. The buyer shall be responsible for complying with any legislation or regulations governing the use of the products and their importation into the country of destination (for the avoidance of doubt to include, without limitation, the TSCA and all its amendments, all EINECS, ELINCS and NONS regulations). If any licence or consent of any government or other authority shall be required for the acquisition, carriage or use of the products by the buyer the buyer shall obtain the same at its own expense and if necessary produce evidence of the same to Iris Biotech GmbH on demand. Failure to do so shall not entitle the buyer to withhold or delay payment. Any additional expenses or charges incurred by Iris Biotech GmbH resulting from such failure shall be for the buyer's account. Save for death or personal injury caused by negligence of Iris Biotech GmbH, sole obligation of Iris Biotech GmbH and buyer's exclusive remedy with respect to the products proved to the satisfaction of Iris Biotech GmbH to be defective or products incorrectly supplied shall be to accept the return of said products to Iris Biotech GmbH for refund of the actual purchase price paid by the buyer (or proportionate part thereof), or replacement of the defective product (or part thereof) with alternative product. Iris Biotech GmbH shall have no liability to the buyer under or arising directly or indirectly out of or otherwise in connection with the supply of products by Iris Biotech GmbH to the buyer and/or their re-sale or use by the buyer or for any product, process or services of the buyer which in any way comprises the product in contract tort (including negligence or breach of statutory duty) or otherwise for pure economic loss, loss of profit, business, reputation, depletion of brand, contracts, revenues or anticipated savings or for any special indirect or consequential damage or loss of any nature except as may otherwise be expressly provided for in these terms. All implied warranties, terms and representations in respect of the products (whether implied by statute or otherwise) are excluded to the fullest extent permitted by law. The buyer shall indemnify Iris Biotech GmbH for and against any and all losses, damages and expenses, including legal fees and other costs of defending any action, that Iris Biotech GmbH may sustain or incur as a result of any act or omission by the buyer, its officers, agents or employees, its successors or assignees, its customers or all other third parties, whether direct or indirect, in connection with the use of any product. For the avoidance of doubt and in the event that Iris Biotech GmbH supplies bespoke or custom product to the buyer's design or specification, this indemnity shall extend to include any claim by a third party that the manufacture of the product for the buyer or the use of the product by the buyer infringes the intellectual property rights of any third party.

## General

Iris Biotech GmbH shall be entitled to assign or sub-contract all or any of its rights and obligations hereunder. The buyer shall not be entitled to assign, transfer, sub-contract or otherwise delegate any of its rights or obligations hereunder. Any delay or forbearance by Iris Biotech GmbH in exercising any right or remedy under these terms shall not constitute a waiver of such right or remedy. If any provision of these terms is held by any competent authority to be invalid or unenforceable in whole or in part the validity of the other provisions of these terms and the remainder of the provision in question shall not be affected. These terms shall be governed by German Law and the German Courts shall have exclusive jurisdiction for the hearing of any dispute between the parties save in relation to enforcement where the jurisdiction of the German Courts shall be non-exclusive.

# Get in Contact

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# Distribution Partners

The list contains the current distributors of Iris Biotech in different regions of the world. The latest list of distribution partners and contact details is available at: *www.iris-biotech.de/distribution-partner*

## China:

**Chengdu Yoo Technology Co., Ltd.**

## Japan:

**BizCom Japan, Inc.**

**Shigematsu & Co., Ltd**

**Cosmo Bio Co., Ltd.**

## USA & Canada:

**Peptide Solutions LLC**

## India, Bangladesh, Oman, Sri Lanka, United Arab Emirates:

**Sumit Biosciences Pvt Ltd.**

# Empowering Peptide Innovation