Bio-orthogonal Chemistry

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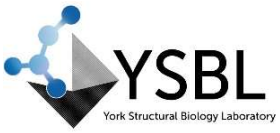
- B/K/256 (Biology site, K block, 2nd floor)
- https://github.com/Zarmstronglab/Courseware



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Supplementary Resources

- Click chemistry sees first use in humans, C&EN News, 2020, October 10
- Bioorthogonal Reactions for Labeling Proteins, Lang and Chin*, ACS Chem. Biol. 2014, 9, 16−20
- From Mechanism to Mouse: A Tale of Two Bioorthogonal Reactions, Sletten and Bertozzi, Accounts of Chemical Research 2011, 44, 9, 666–676
- The Future of Bioorthogonal Chemistry, Devaraj, ACS Cent. Sci. 2018, 4, 952−959
- Available at https://github.com/Zarmstronglab/Courseware

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Learning objectives

- Understand and discuss the difficulties of performing organic chemistry on proteins, in cells and organisms
- Describe the concept of bio-orthogonal chemistry
- Recognise the reactants and products of bio-orthogonal reactions
- Understand and discuss the benefits and drawbacks of different types of bio-orthogonal reactions
- Understand the mechanism of strained alkyne and alkene cycloadditions and roles in probing cell biology

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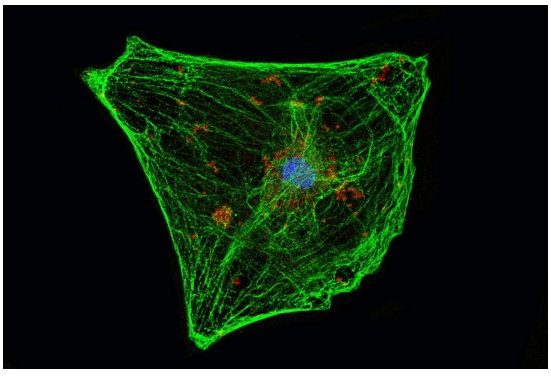
Why do chemistry in biological systems?

- Track biomolecules in their native environment
  - Especially Post-translational modifications, small molecules and metabolites
- Make small-molecule protein conjugates
  - Site specific drug Delivery
- Release pro-drugs are specific sites
  - Only activate drugs at the site of interest



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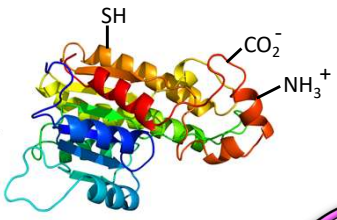
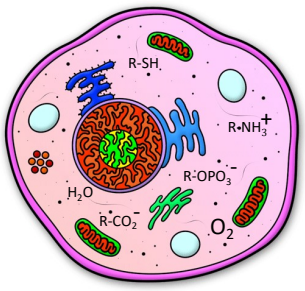
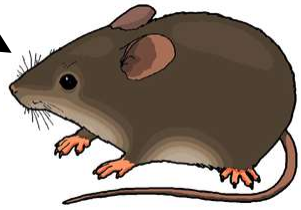
Chemistry in living systems is difficult







Neutral pH, water, body temperature

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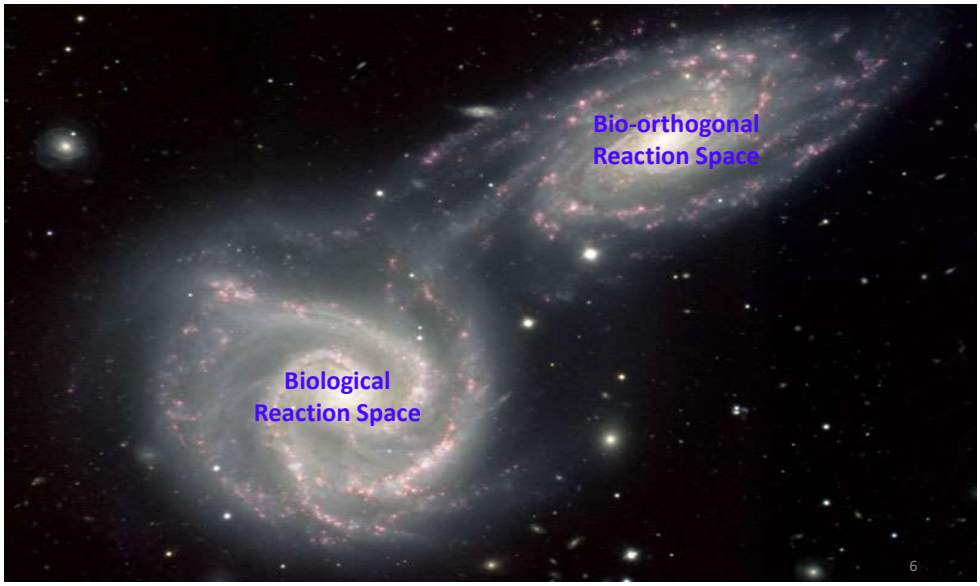
Bio-orthogonal chemistry

- Not interacting or interfering with a biological system

Carolyn Bertozzi



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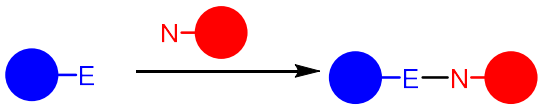
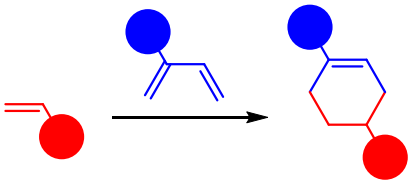
Types of Bio-orthogonal chemistry

- Nucleophile – electrophile
  - Oxime Ligation
  - Staudinger Ligation



- Cycloadditions
  - Azide Alkye Cycloadditions
  - Tetrazine ligations



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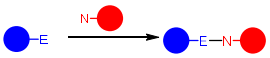
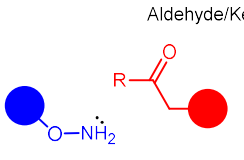
Nucleophile – Electrophile



Oxime Ligation



- Amnioxy groups a small and relatively rare in living systems
- Aldehydes can be incorporated into proteins
- No catalyst other than acid needed
- Somewhat slow – may take days to completely label
- Aminoxy groups can be hydrolysed under acidic conditions

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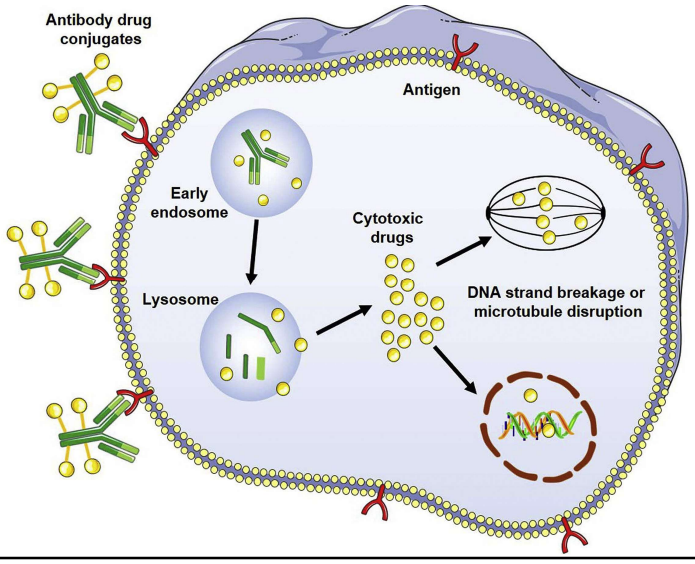
Antibody Drug Conjugates



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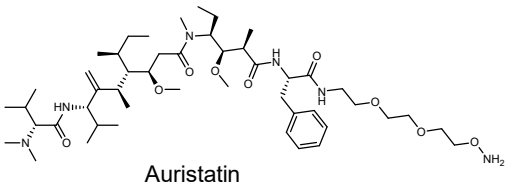
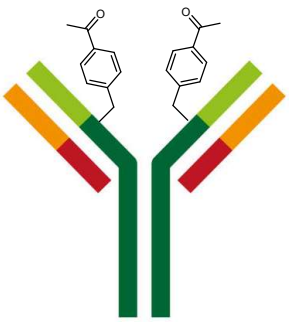
Antibody Drug Conjugates



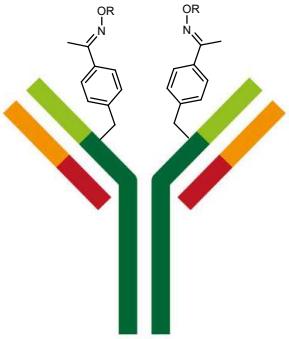
Auristatin



p-Acetylphenylalanine containing Antibody

Oxime Ligation

pH 4.5
37 °C
1 - 4 days



Antibody Drug Conjugate

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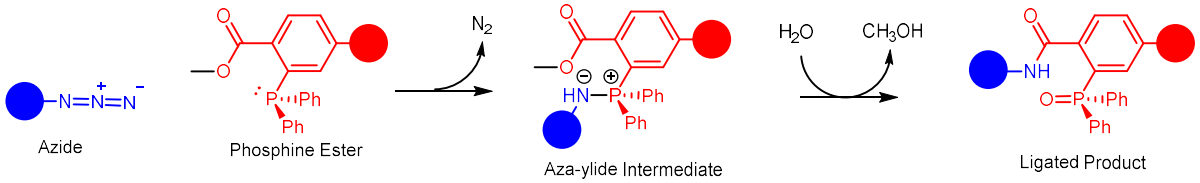
Staudinger ligation



Hermann Staudinger

- Alkyl Azides are small, absent in living systems and easy to incorporate into small molecules
- R-N_3 are a mild electrophile, but unreactive to biological nucleophiles
- Triarylphosphines are ideal partners
- Somewhat slow reaction, excess of phosphine needed
- Phosphines can be oxidized in vivo

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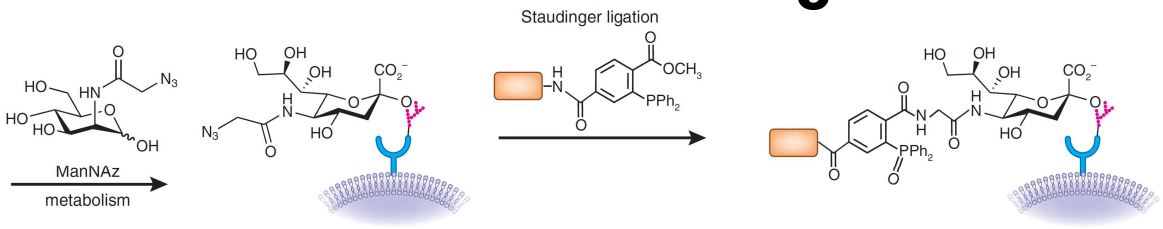
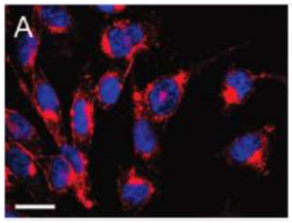
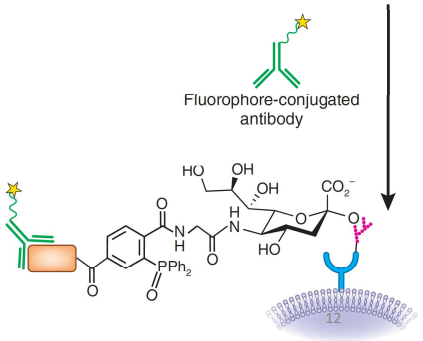
Metabolic cell surface labeling



Carolyn Bertozzi



Labeled Cells



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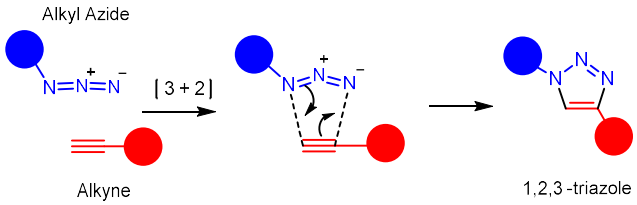
3+2 Cycloadditions



- Uncatalyzed Reaction needs heat - incompatible with biosystems
- Can also be performed with a Cu(I) catalyst – classic 'click-chemistry'
  - Copper catalysed Azide Alkyne Cycloaddition (CuAAC)
- Cu(I) is toxic so it must be performed on the outside of cells

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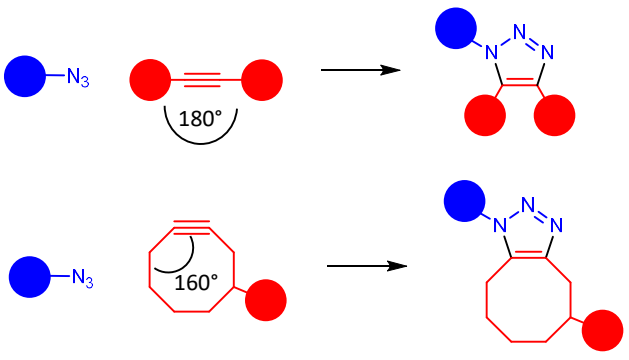
Strain Promoted Azide Alkyne Cycloaddition (SPAAC)



18 Kcal/mol strain

~ 6 Kcal/mol strain

https://youtu.be/kwvzjedDteE

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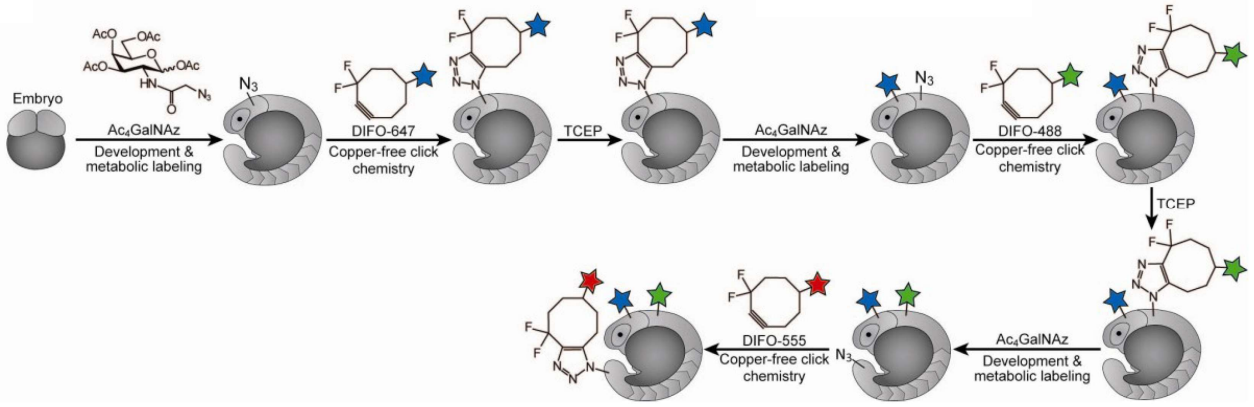
Imaging Glycans



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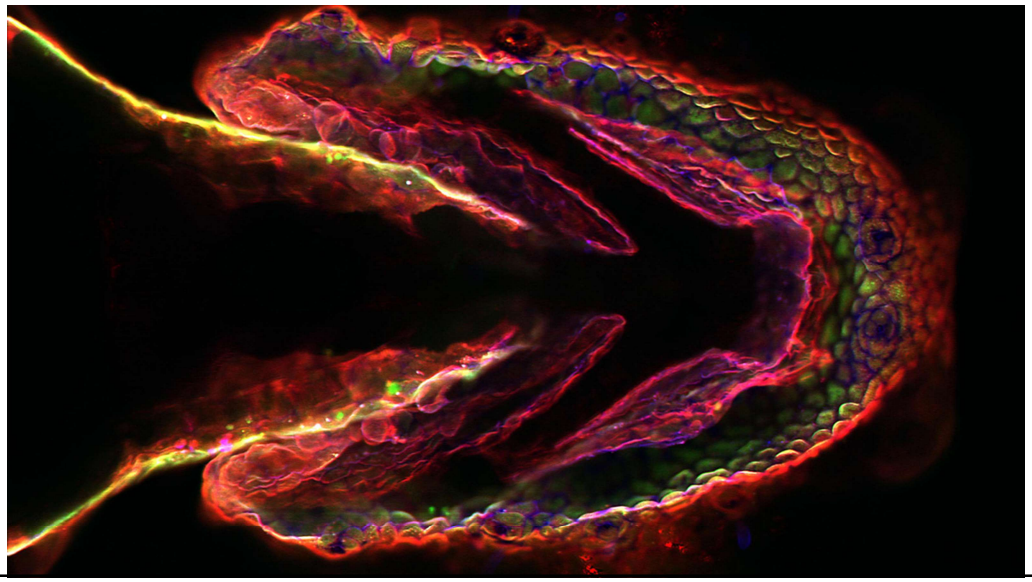
Time Course of Glycans in Zebrafish



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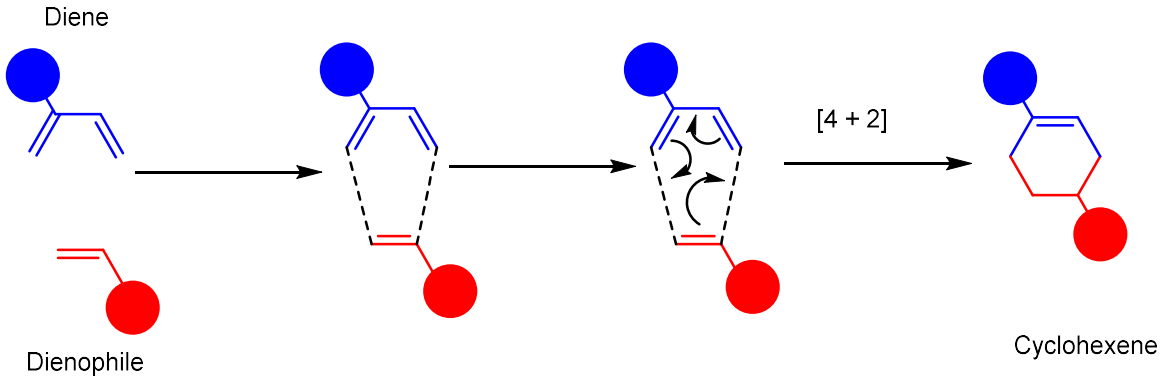
Diels-Alder like Cycloadditions [4+2]



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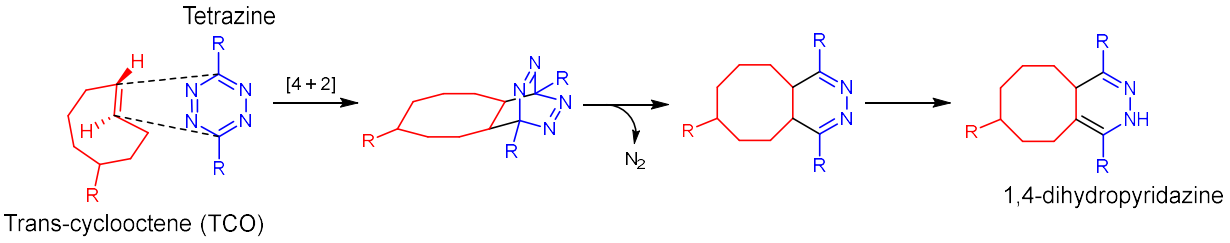
Tetrazine Ligation



- Tetrazine is absent in nature, TCO is very rare
- Super fast up to 10^6 times faster than the fastest SPAAC
  - 5 min to completion (15 µM + 30 µM substrates), in water/cell lyaste at 25 °C
- TCO is pre-distorted towards the transition state structure
- Drawback is the size of TCO and Tetrazine

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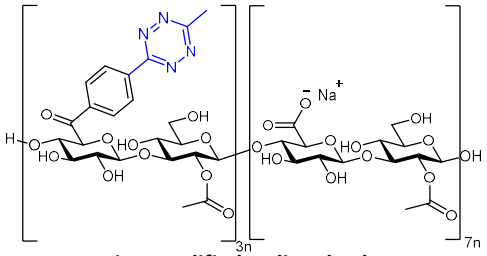
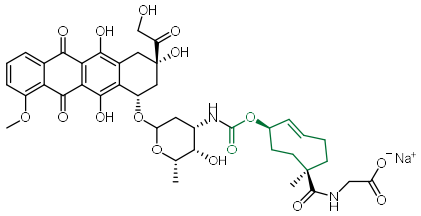
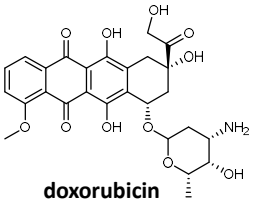
Cancer Therapeutics



Tetrazine-modified sodium hyaluronate



TCO-modified doxorubicin



doxorubicin

C&EN CHEMICAL & ENGINEERING NEWS

October 2020

'…biopolymer is injected into a cancer patient's tumor. That patient then receives five daily infusions of doxorubicin-TCO, which circulates through the body until it meets the tetrazine-modified biopolymer. At that point, the click reaction brings the tetrazine and TCO together, triggering a rearrangement that frees doxorubicin right next to the tumor cells.'

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Supplementary Resources

- Click chemistry sees first use in humans, C&EN News, 2020, October 10
- Bioorthogonal Reactions for Labeling Proteins, Lang and Chin*, ACS Chem. Biol. 2014, 9, 16−20
- From Mechanism to Mouse: A Tale of Two Bioorthogonal Reactions, Sletten and Bertozzi, Accounts of Chemical Research 2011, 44, 9, 666–676
- The Future of Bioorthogonal Chemistry, Devaraj, ACS Cent. Sci. 2018, 4, 952−959
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SCAN ME

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