

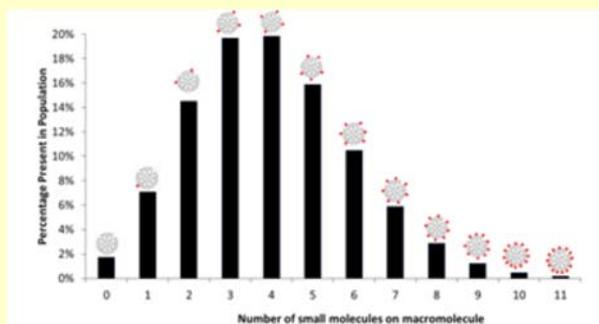
# FACULTY-UNDERGRADUATE RESEARCH

## PROFESSOR CASEY DOUGHERTY

### BIOCHEMISTRY RESEARCH

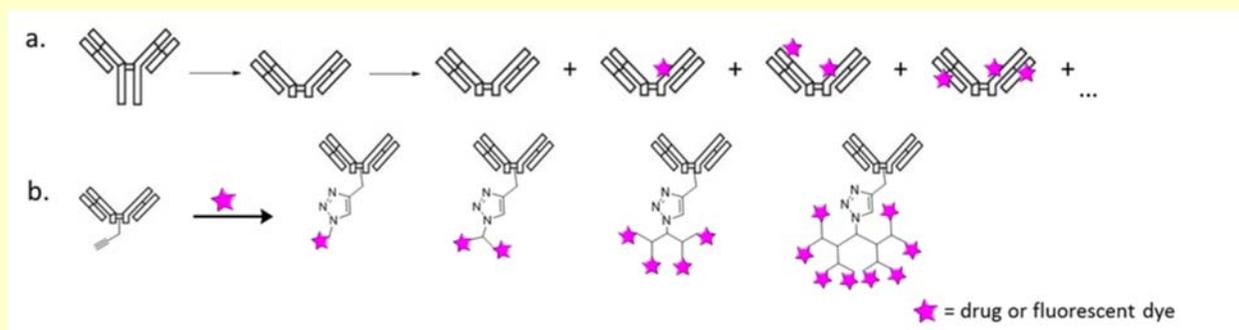
Macromolecules have become a material of interest in the development of novel imaging agents and therapeutics in medicine. Many macromolecules, such as synthetic multivalent polymers and biologically interesting antibodies, can have multiple copies of small molecules, such as fluorescent dyes and drugs, attached to them. This can provide a higher efficiency for delivering a drug or better detection of an imaging agent than using just small molecule treatment.

However, the inherent random nature of conjugating small molecules to a macromolecule leads to a large mixture of different ratios of small molecules to macromolecule (Figure 1). Since the small molecules used for imaging and therapeutics are typically hydrophobic and the macromolecule is typically hydrophilic, the different ratios of small molecule to macromolecule can lead to very different degrees of hydrophobicity of the molecule. The degree of hydrophobicity has the ability to alter a macromolecule's biological behavior, and controlling the ratio of hydrophobicity has been an obstacle in developing effective macromolecules for biomedical applications.



**Figure 1.** Example of a predicted distribution of a ratio of 4 small molecules conjugated to a macromolecule with 128 reaction sites.

Our work will focus on the synthesis and characterization of antibody fragments and antibody-polymer conjugates with controlled ratios of fluorophores/drugs. The research is a combination of biochemistry, organic synthesis, and analytical characterization. We will show the difference in fluorescence and absorption of the different materials based on a having a distribution (Figure 2a) versus a precise ratio of small molecules to macromolecule (Figure 2b). We will fragment antibodies and attach a ligand that allows further conjugation of a fluorescent dye, drug, or polymer with multiple copies of fluorescent dye or drug (Figure 2b). Biological controls will be conducted, and the materials could be applied in cell studies to see how their behavior (amount of uptake, where it goes in the cell, etc.) changes based on hydrophobic ratio.



**Figure 2.** a) reaction scheme of a typical antibody fragment conjugation with a small molecule that results in a distribution of ratios of small molecule to antibody. b) proposed synthesis to obtain precisely one, two, four, or eight small molecules on an antibody fragment.

#### Publications

1. van Dongen, M. A.; Dougherty, C. A.; Holl, M. M. B. Multivalent Polymers for Drug Delivery and Imaging: The Challenges of Conjugation. *Biomacromolecules* 2014, 15, 3215-3234.
2. Dougherty, C. A.; Furgal, J. C.; van Dongen, M. A.; Goodson, T.; Holl, M. M. B.; Manono, J.; DiMaggio, S. Isolation and Characterization of Precise Dye/Dendrimer Ratios. *Chem-Eur J* 2014, 20, 4638-4645.
3. Dougherty, C. A.; Vaidyanathan, S.; Orr, B. G.; Holl, M. M. B. Fluorophore: Dendrimer Ratio Impacts Cellular Uptake and Intracellular Fluorescence Lifetime. *Bioconjugate Chem* 2015, 26, 304-315.