CPPC 2021
First Canadian Peptide and Protein Community Virtual Symposium
27–28 MAY 2021 | ONLINE
Canadian Peptide Protein Community
biomedicines
MDPI

Genetically encoded fragment-based discovery from phage-displayed macrocyclic libraries with genetically encoded unnatural pharmacophores

Dr. Arunika Ekanayake
Derda Research Group
Department of Chemistry
University of Alberta

UNIVERSITY OF ALBERTA

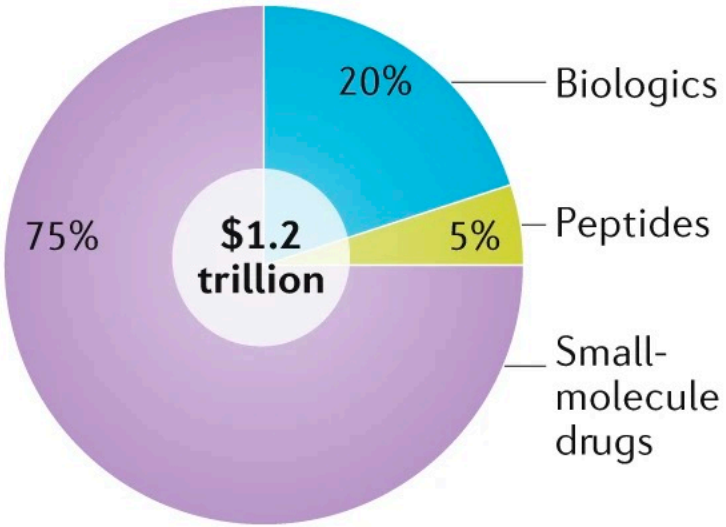
DERDA RESEARCH GROUP

Abstract

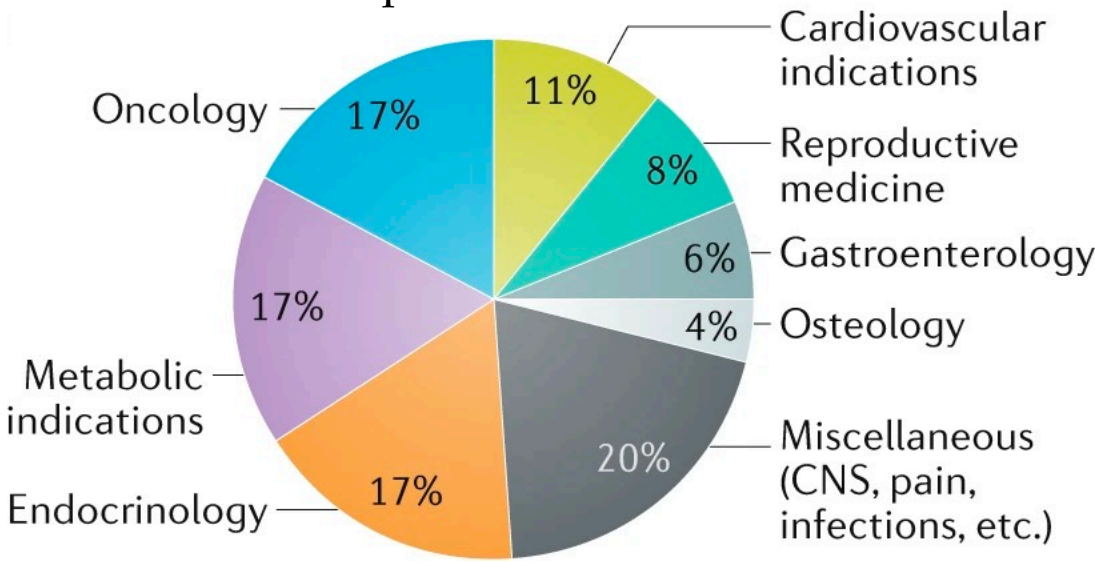
Peptide therapeutics are known to have high selectivity and strong interactions with their targets of interest. While there is increased interest in the therapeutic application of peptides, there is a continuous need to generate peptides with improved stability and cellular internalization properties. This requires the incorporation of pharmacophores or chemical fragments onto peptides. Generation of genetically encoded (GE) macrocyclic peptide libraries containing unnatural fragments can give rise to libraries of value-added ligands against a plethora of protein targets. Generally, these fragments are installed at the macrocyclization step, requiring optimization of different reactions depending on the chemical fragment. In Derda Research Group, we have employed the potent Knorr-pyrazole synthesis reaction to develop a robust two-step ligation reaction for late-stage functionalization of GE-peptide libraries. A readily available 1,5-dichloropentane-2,4-dione linchpin coverts peptide libraries displayed on phage to 1,3-diketone macrocyclic peptides that provide a handle to be functionalized by a variety of hydrazines. The phage libraries carry a silent-DNA barcode that encodes the amino acid sequence and the unnatural fragment displayed on the peptides. These libraries can be applied for genetically encoded-fragment-based discovery (GE-FBD) against various therapeutic targets.

Insulin to present: the story of peptide therapeutics

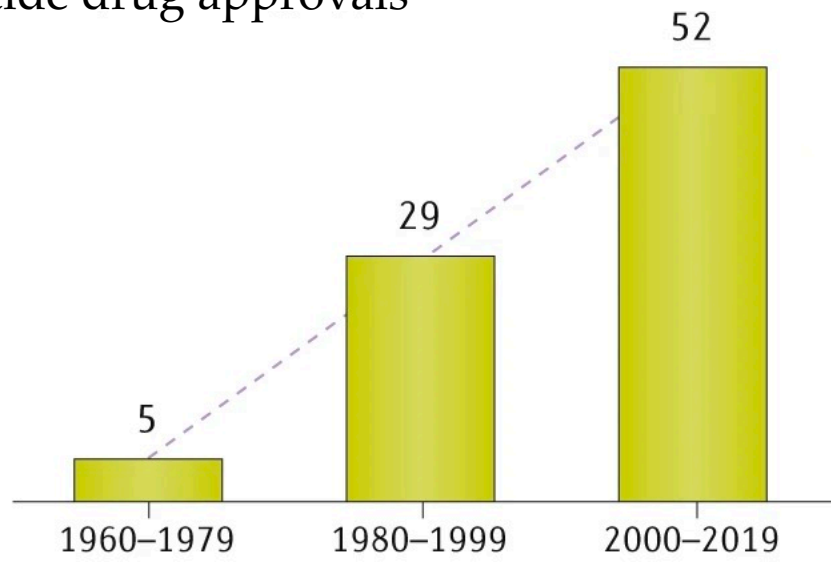
Global pharmaceutical market (2019)



Therapeutic indications



Peptide drug approvals



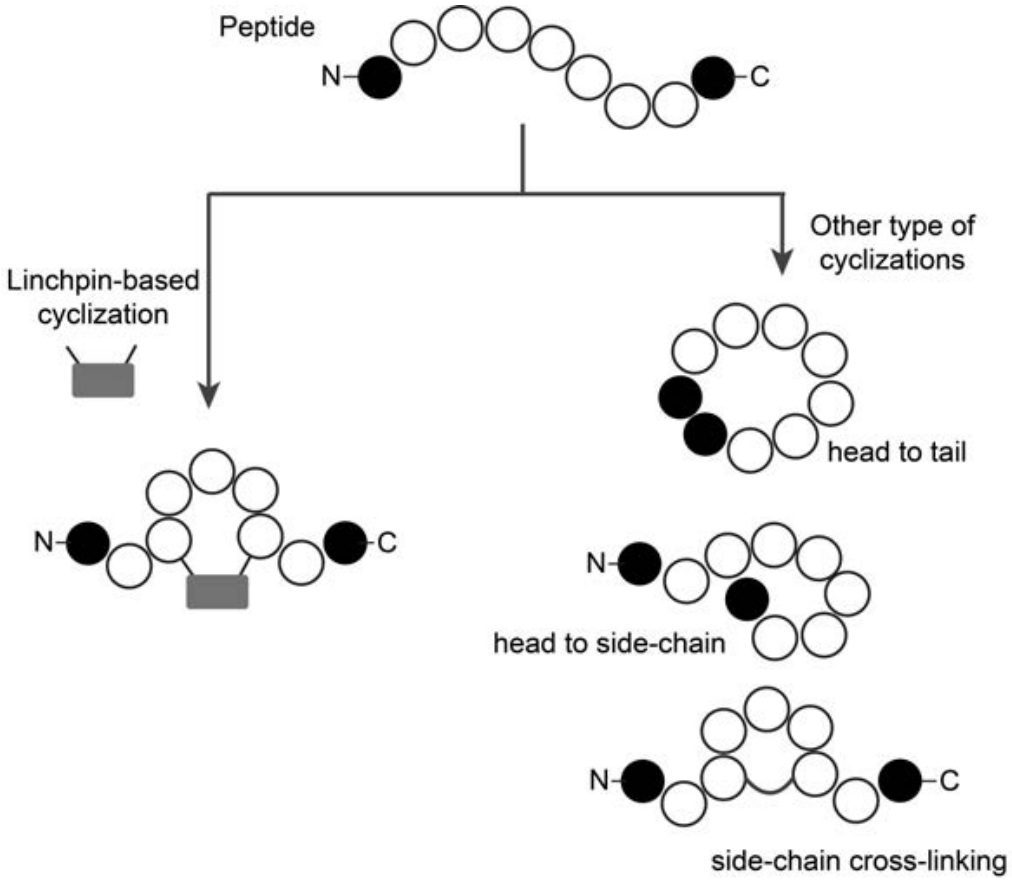
Nature Reviews Drug Discovery
20, 309-325 (**2021**)

CPPC 2021

Chemistry as a tool to design value added ligands

Improving pharmacokinetics by macrocyclization

- Proteolytic stability
- Targeting active sites
- Internalization

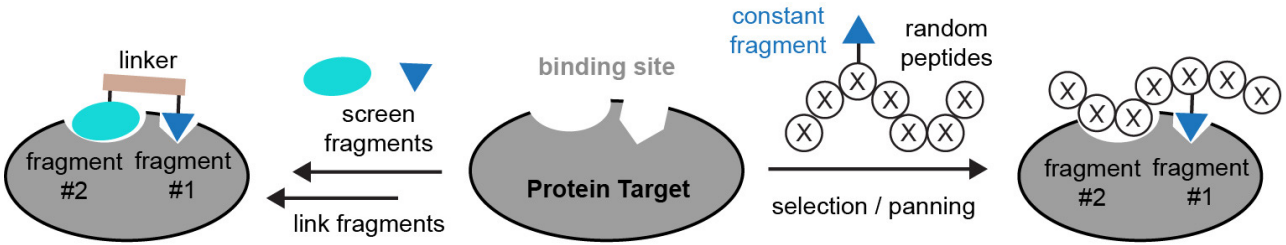


Protein & Peptide Letters, 25, 1-25 (**2018**)

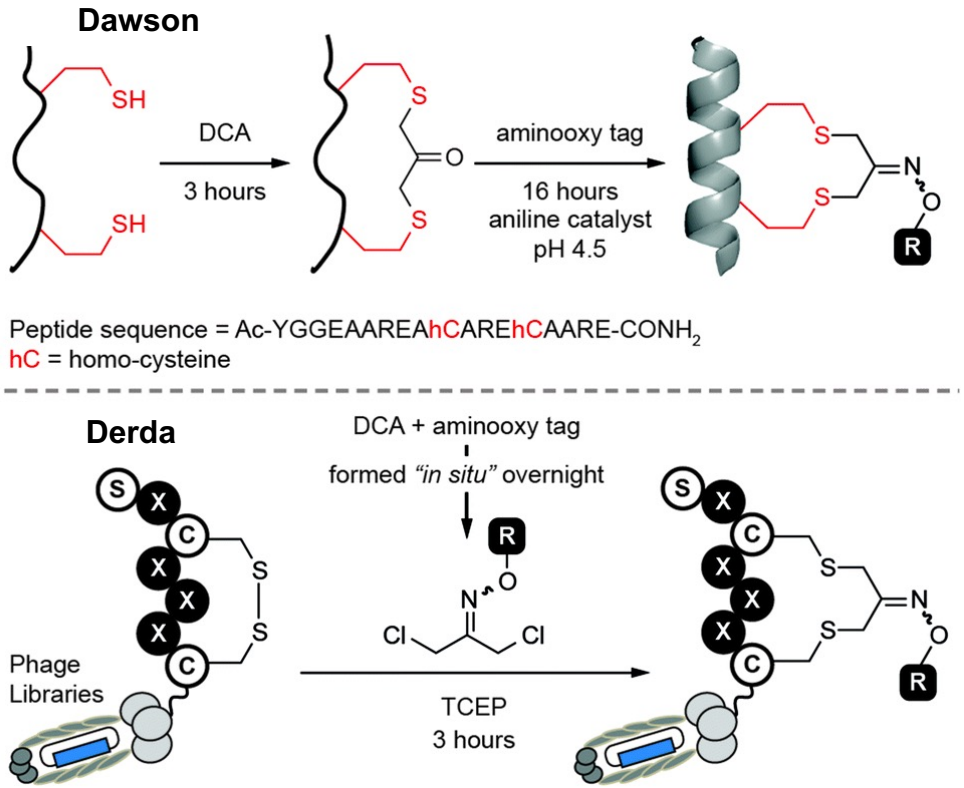
CPPC 2021

Providing a platform to introduce phamacophores

Canonical fragment-based drug design (FBDD)



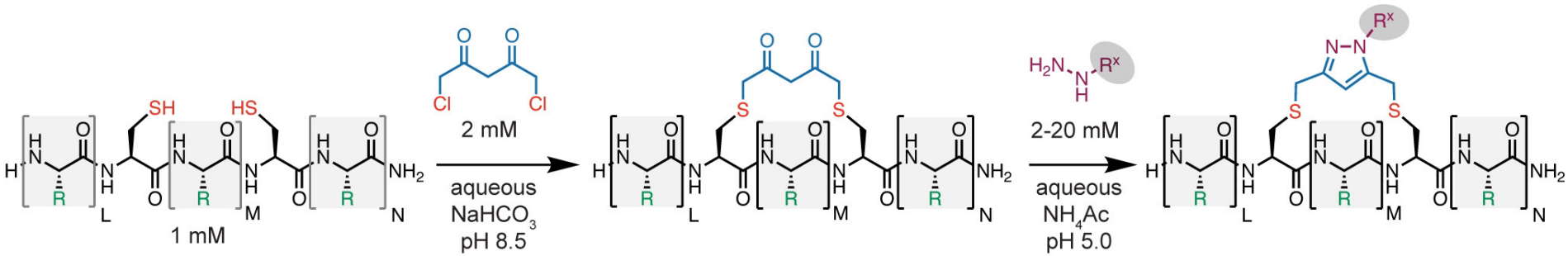
Genetically-encoded fragment - based discovery (GE-FBD)



Current opinion in chem. Bio,50, 128-137 (**2019**)
Org. Biomol. Chem., 14, 5539-5545 (**2016**)

CPPC 2021

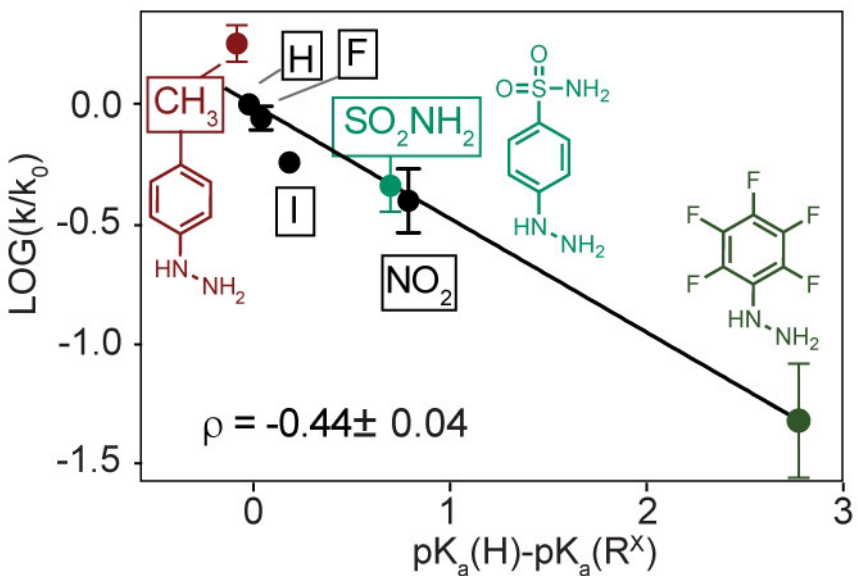
Late-stage functionalization of 1,3-diketone macrocycles

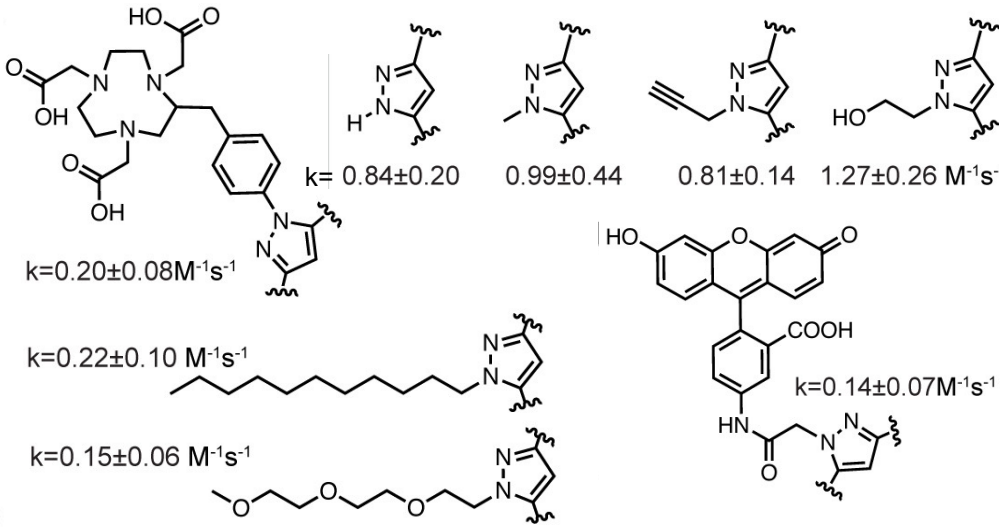


1

2

Knorr pyrazole synthesis

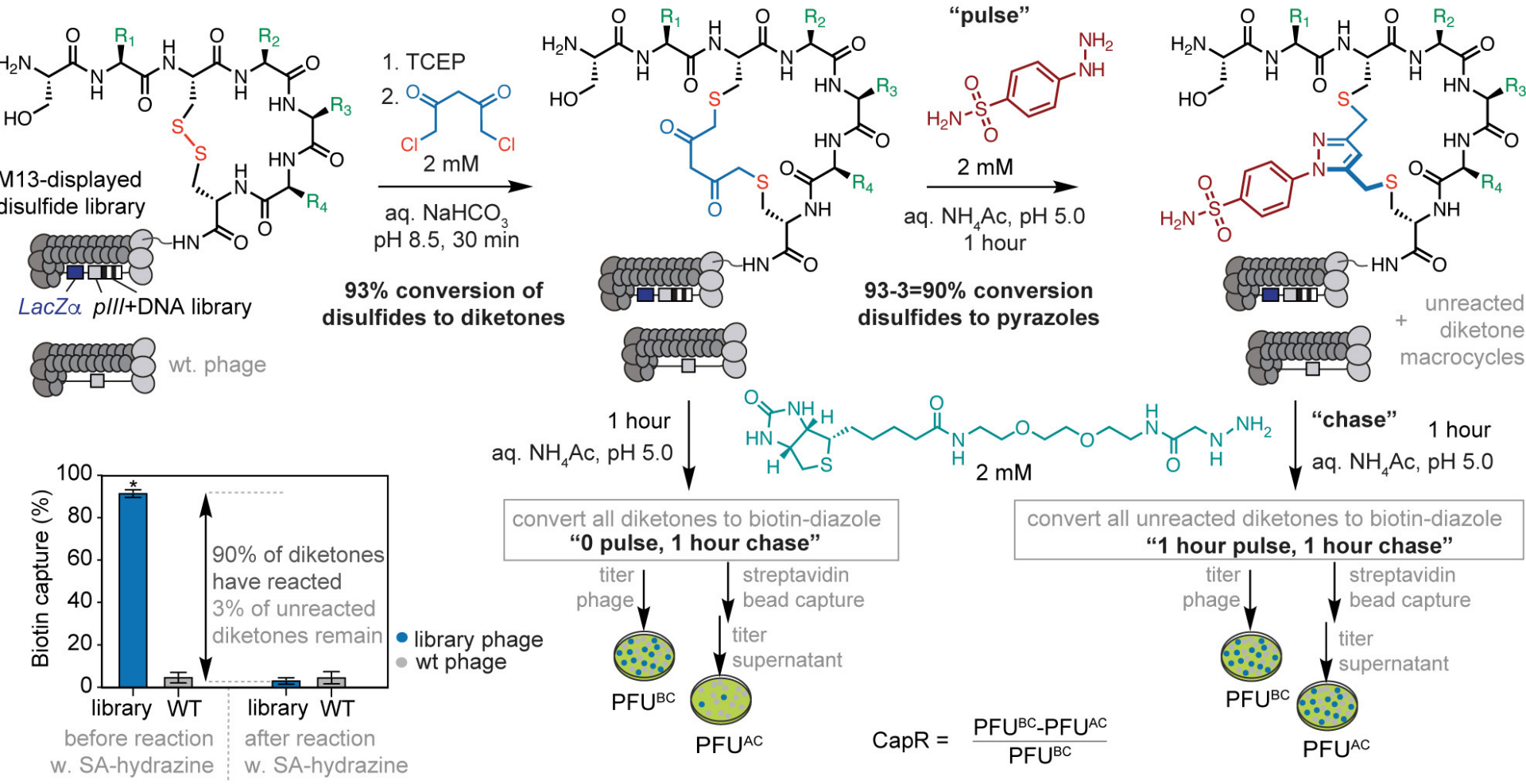




JACS ,143, 5497-5507 (**2021**)

CPPC 2021

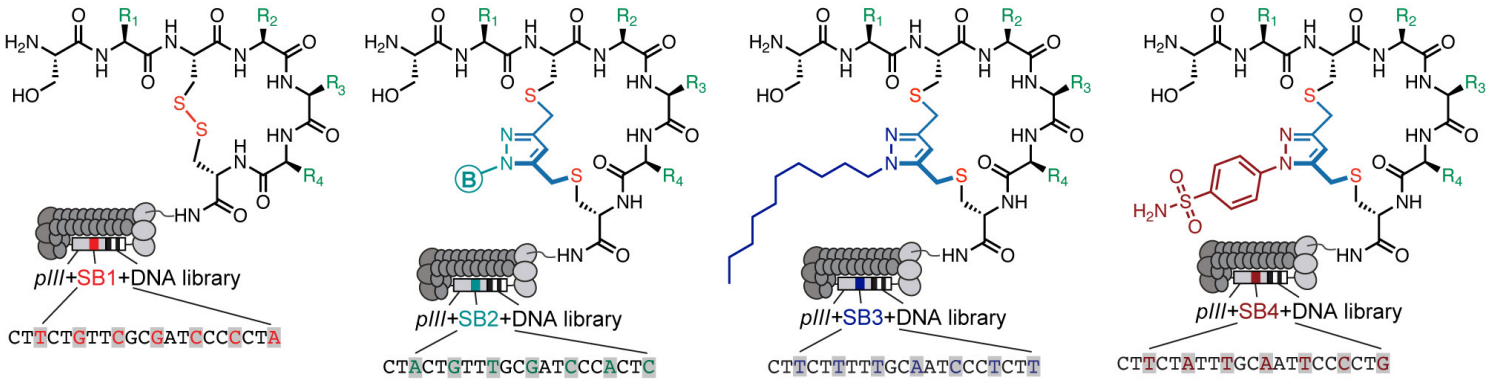
Macrocyclization of genetically encoded peptides

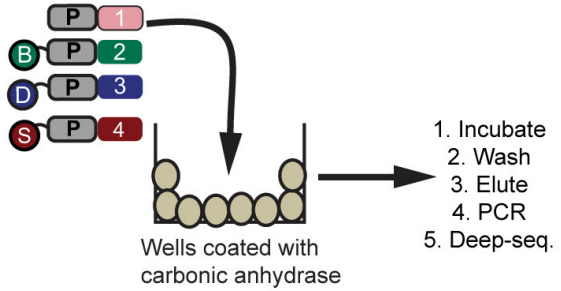


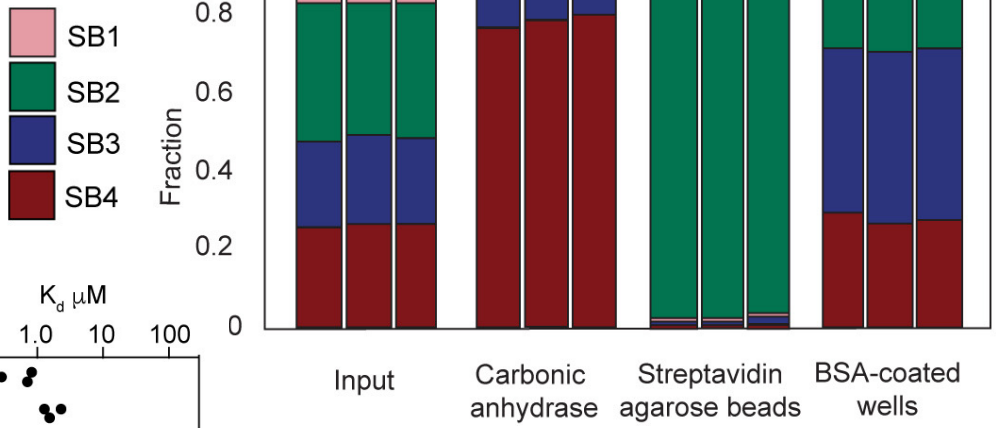
JACS ,143, 5497-5507 (**2021**)

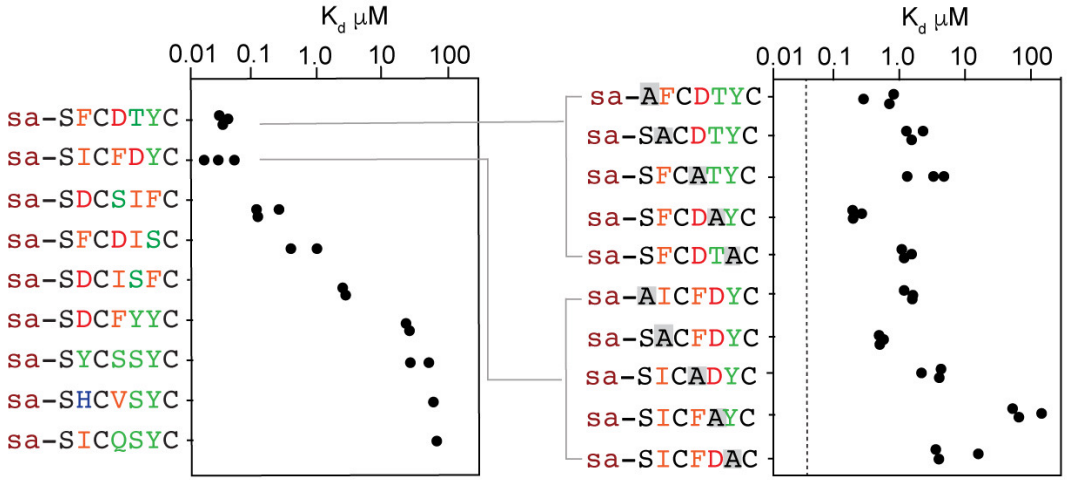
CPPC 2021

Selection of ligands from functionalized macrocyclic libraries





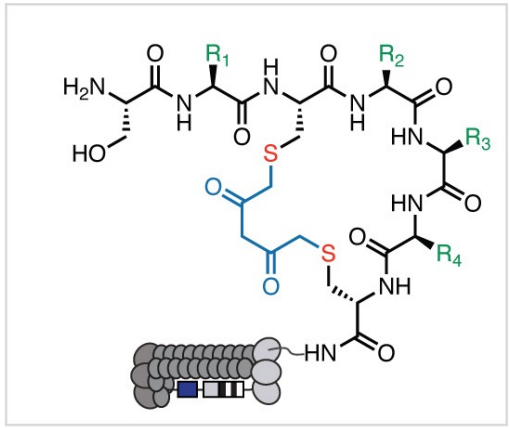


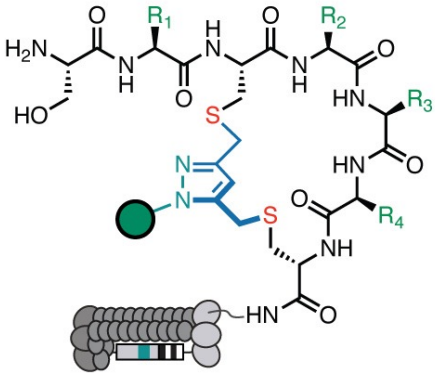


JACS ,143, 5497-5507 (**2021**)

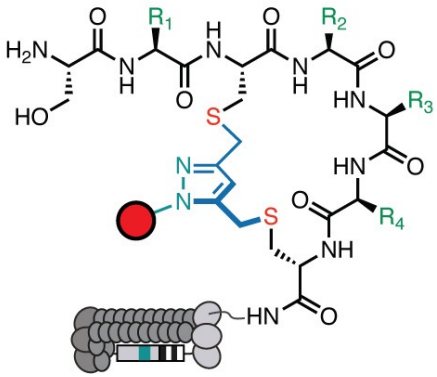
CPPC 2021

Applications of GE-FBD

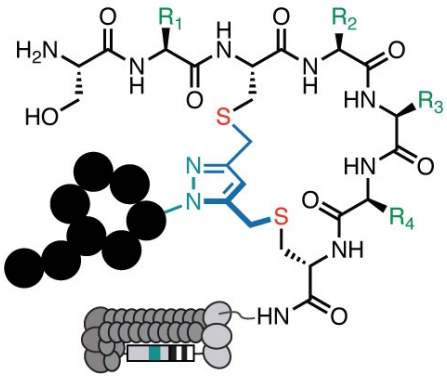




Electrophilic fragments reactive towards active site cysteines



Electrophilic fragments reactive towards allosteric site cysteine



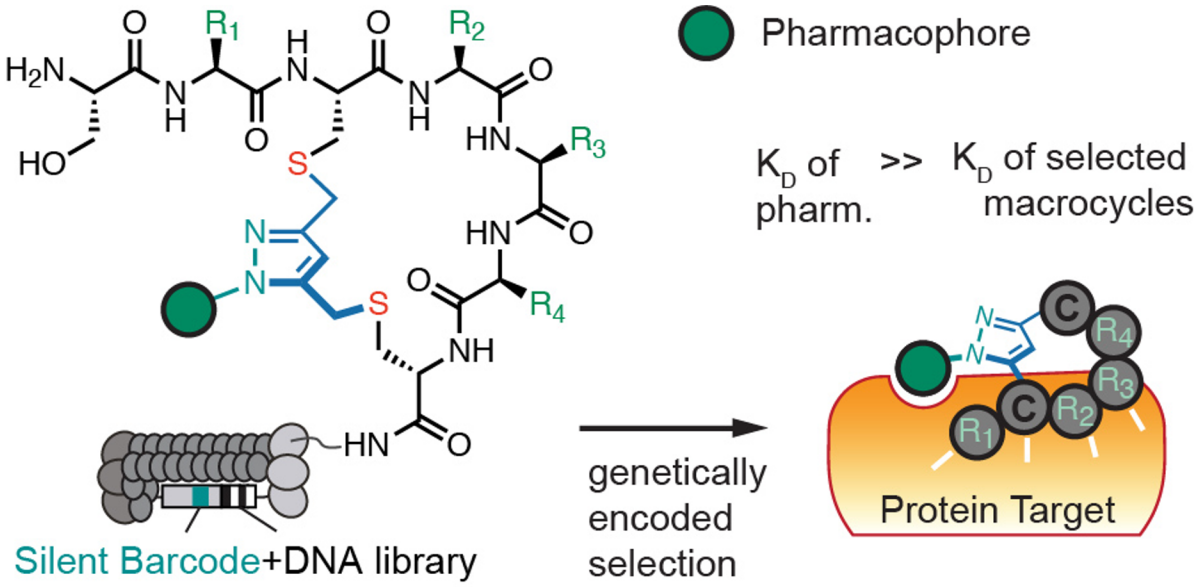
CPP for intracellular targets

(*Ongoing work*)

CPPC 2021

Conclusions and Perspective

- Broad substrate scope of Knorr Pyrazole Synthesis
- Attractive strategy for diversification of macrocycles with built in 1,3-diketones using a large range of hydrazines
- Robust and straightforward macrocyclization with late-stage functionalization
- Ability to generate large number of ligands already containing post validation modifications
- Shelf stable linchpin allows storage of intermediate libraries for immediate modification and application



JACS ,143, 5497-5507 (**2021**)

CPPC 2021

Acknowledgments

Dr. Ratmir Derda
Lena Sobze
Dr. Raja Mukherjee
Jihea Youk
Dr. Nichols J. Bennett

Collaborators
Dr. Lela Vukovich
Payam Kelich
Dr. Frank Wuest
Dr. Atul Bhardwaj

NSERC CRSNG

CIHR IRSC
Canadian Institutes of Health Research
Instituts de recherche en santé du Canada

UNIVERSITY OF ALBERTA

DERDA RESEARCH GROUP

CPPC 2021