Circumventing DNA depurination by micellar Broensted acid catalysis – synthesis of DNA-tagged tetrahydroquinolines

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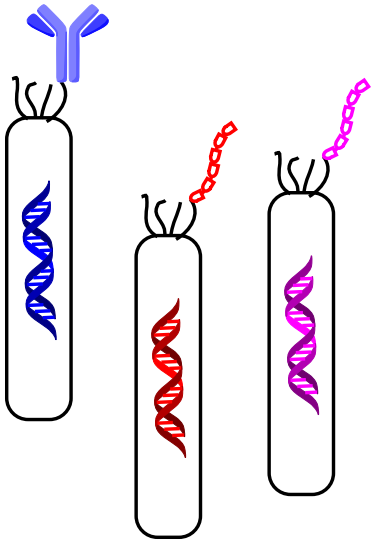
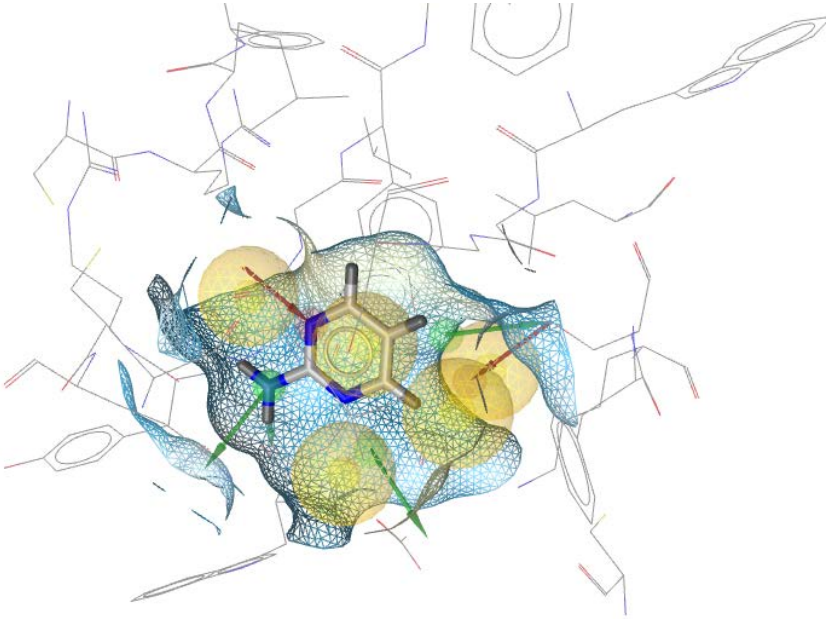
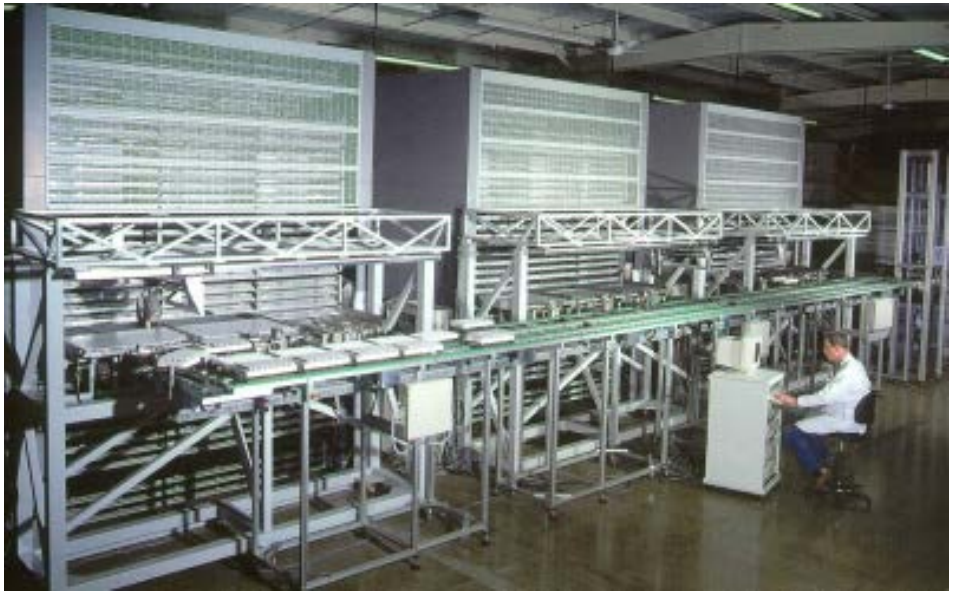


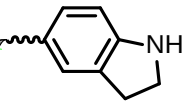
Identification of biomacromolecules

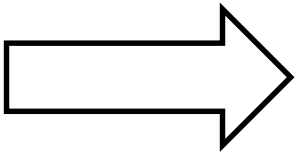
Source:https://www.nobelprize.org/prizes/chemistry/ Frances Arnold, Georg Smith, Sir Gregory Winter
Kunig, Biol. Chem., 2018

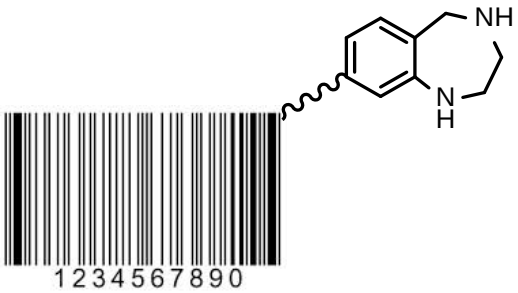


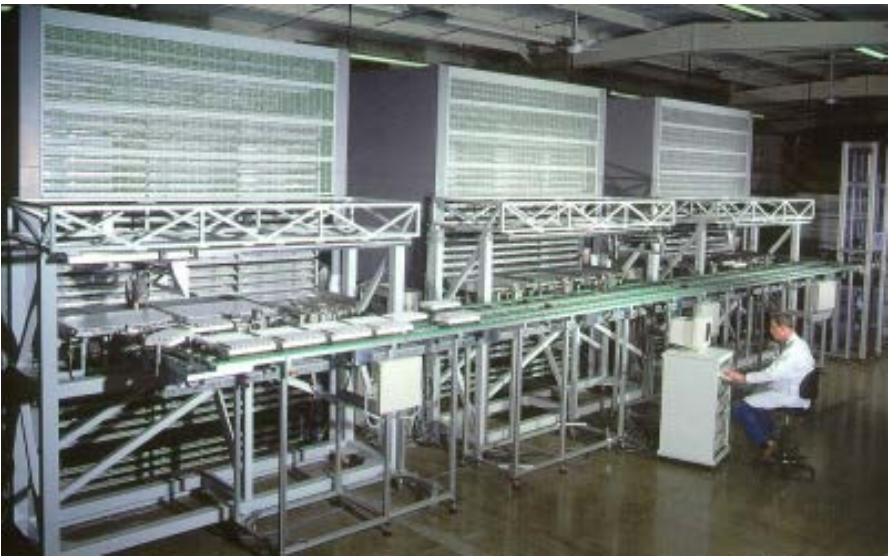


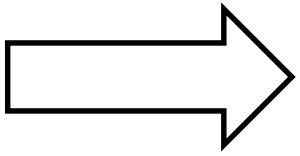
Identification of bioactive small molecules?

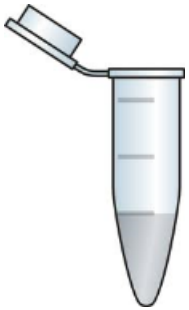




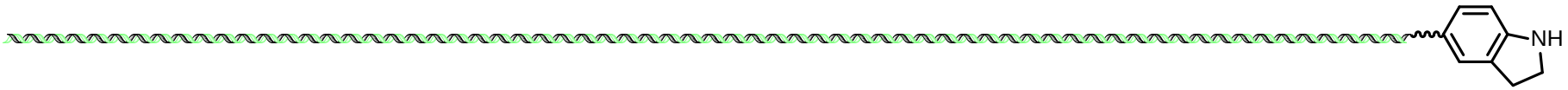








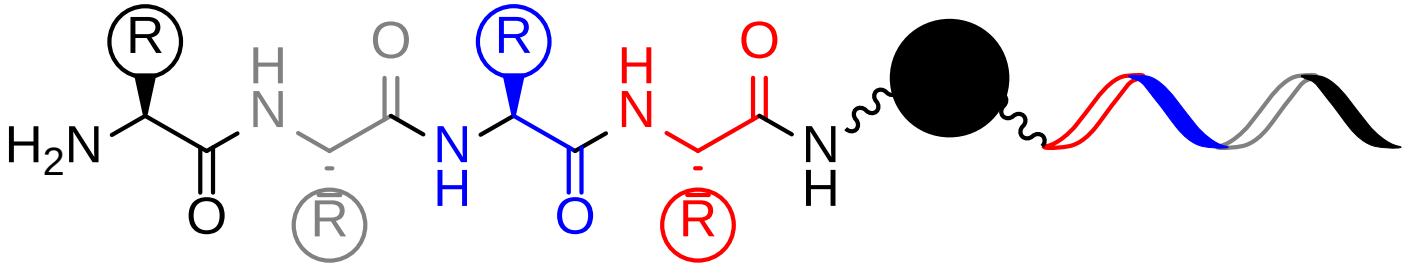
Small molecule screening libraries



Encoded combinatorial chemistry

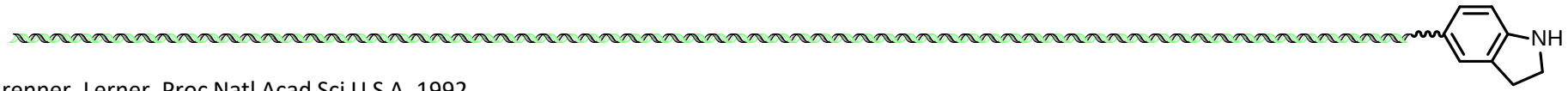
(chemical repertoire/encoded libraries/commaless code)

SYDNEY BRENNER AND RICHARD A. LERNER

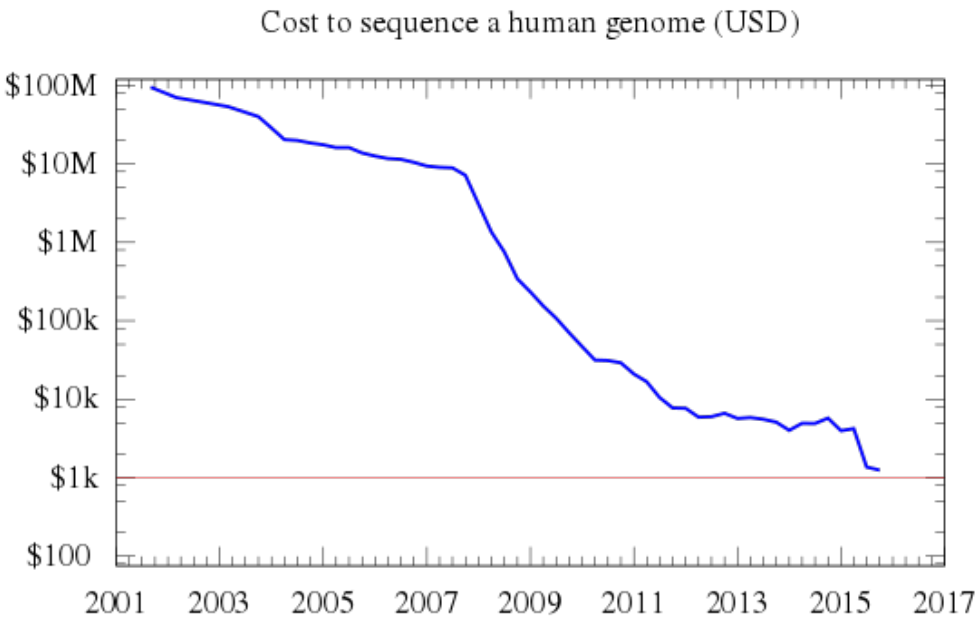


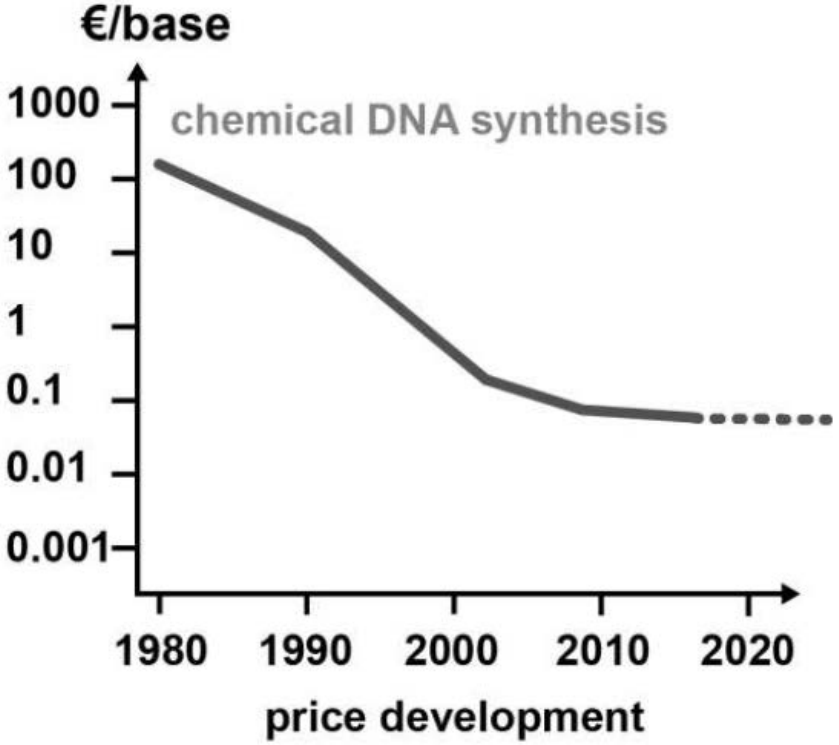
AGCTACTTCCCAAGGCACATGCACATGCACATGGGGCCCTATTCTTAG–LINK–Gly–Gly–Gly
AGCTACTTCCCAAGGCACATGCACATGACGGTAGGGCCCTATTCTTAG–LINK–Met–Gly–Gly
AGCTACTTCCCAAGGCACATGACGGTACACATGGGGCCCTATTCTTAG–LINK–Gly–Met–Gly
AGCTACTTCCCAAGGCACATGACGGTAACGGTAGGGCCCTATTCTTAG–LINK–Met–Met–Gly
AGCTACTTCCCAAGGACGGTACACATGCACATGGGGCCCTATTCTTAG–LINK–Gly–Gly–Met
AGCTACTTCCCAAGGACGGTACACATGACGGTAGGGCCCTATTCTTAG–LINK–Met–Gly–Met
AGCTACTTCCCAAGGACGGTAACGGTACACATGGGGCCCTATTCTTAG–LINK–Gly–Met–Met
AGCTACTTCCCAAGGACGGTAACGGTAACGGTAGGGCCCTATTCTTAG–LINK–Met–Met–Met

Chemically synthesized DNA, encoded beads



Brenner, Lerner, Proc Natl Acad Sci U S A. 1992





National Human Genome Research Institute, price development: courtesy Dr. Thorsten-Lars Schmidt, Kent State University

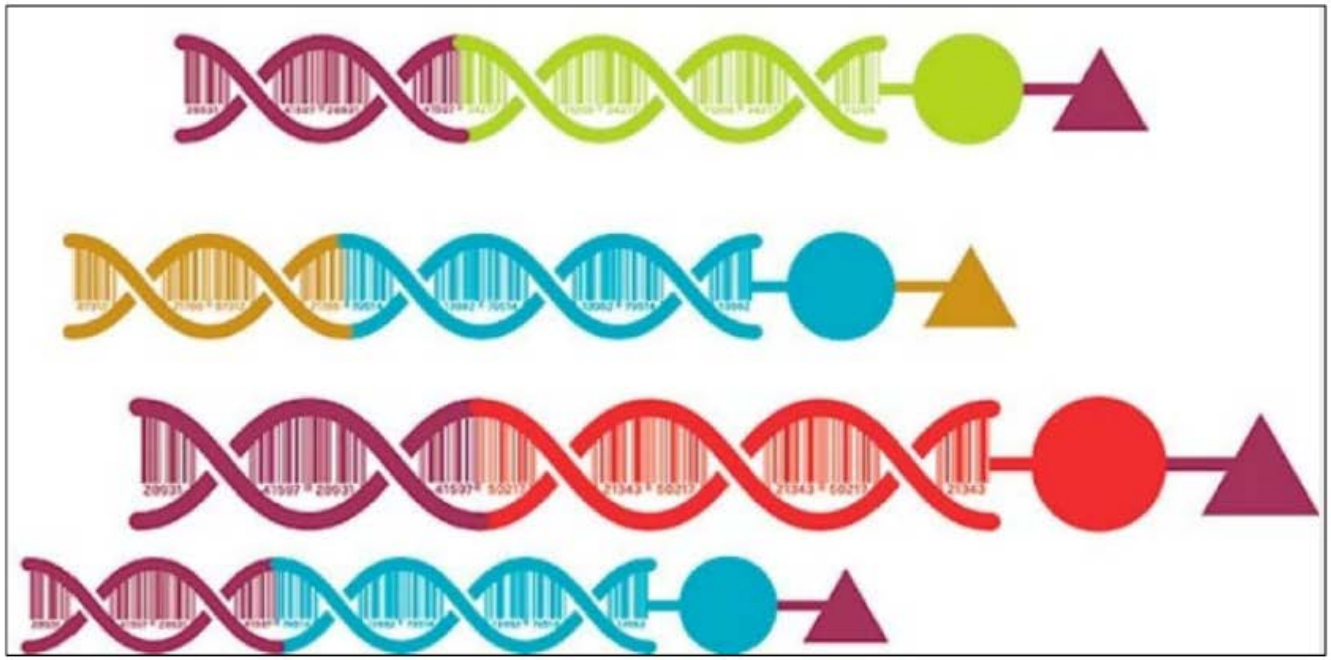
c&en
CHEMICAL & ENGINEERING NEWS

COVER STORY

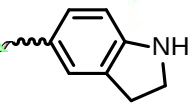
How DNA-encoded libraries are revolutionizing drug discovery

Issue Date: June 19, 2017

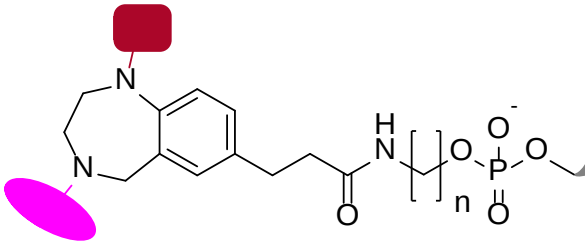
With the bar-coding technology, drugmakers leverage the chemistry of large numbers



Available in: http://cen.acs.org/articles/95/i25/DNA-encoded-libraries-revolutionizing-drug.html.

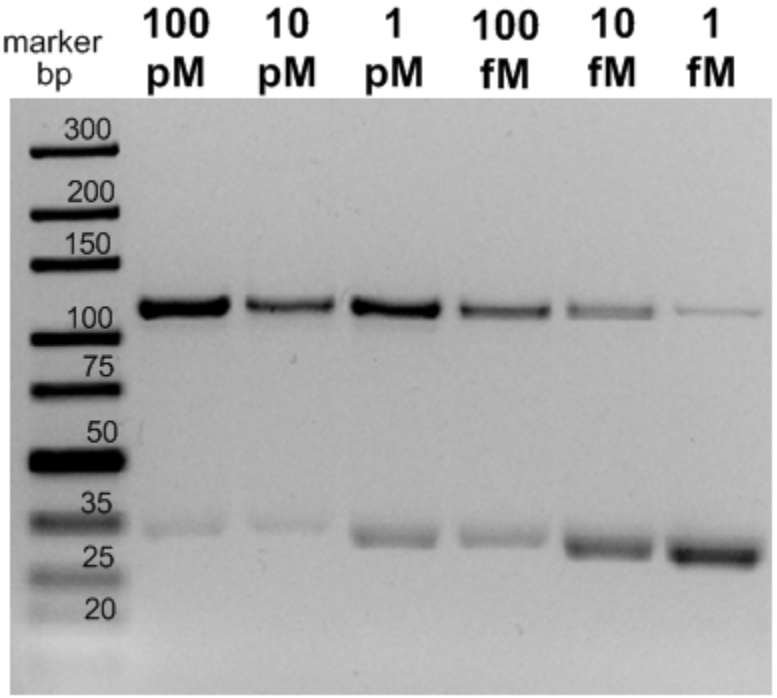


Detection of DNA-small molecule conjugates

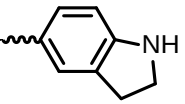


primer primer

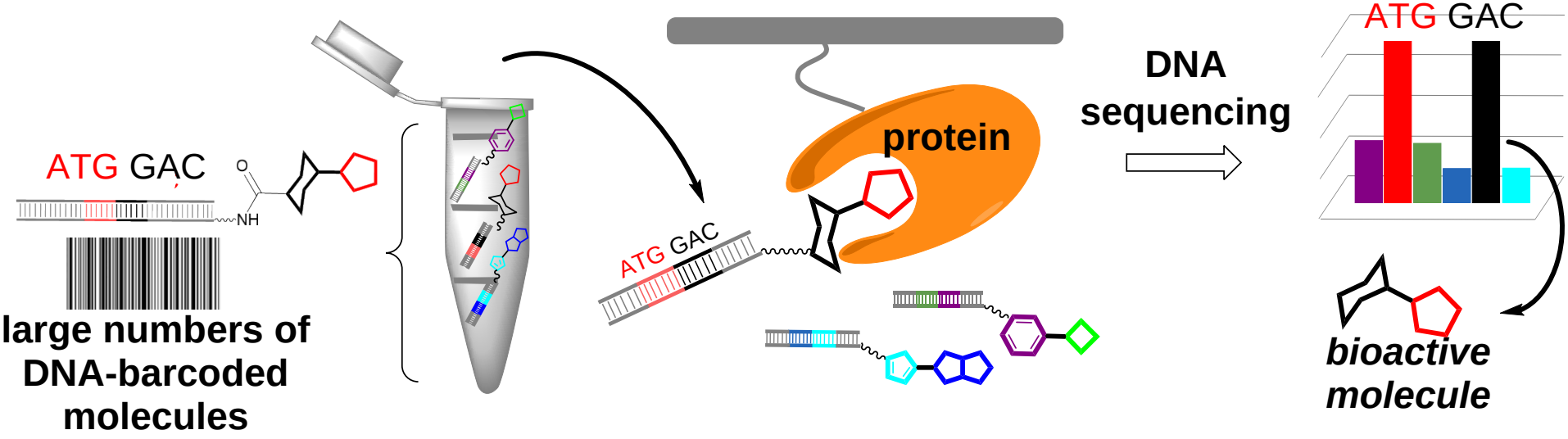
„phenotype“ **„genotype“**



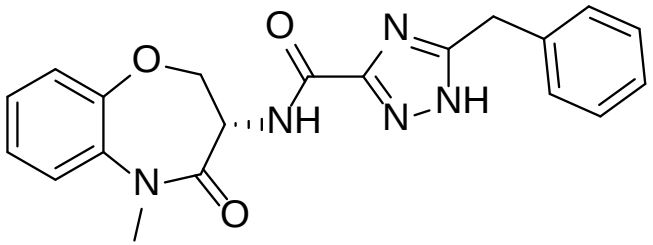
PCR:
40 µL volume
25 cycles
Taq-polymerase



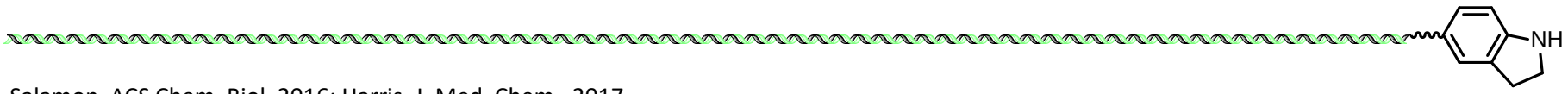
Finding protein binders with encoded libraries



- **efficient**
- **generic**
- **simple infrastructure**
- **validated**

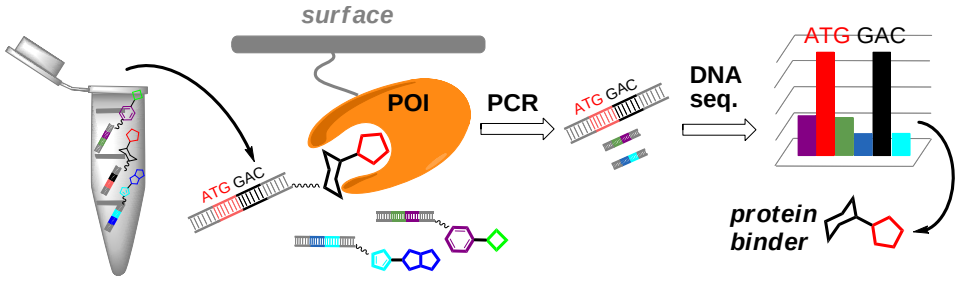


RIP1 kinase inhibitor

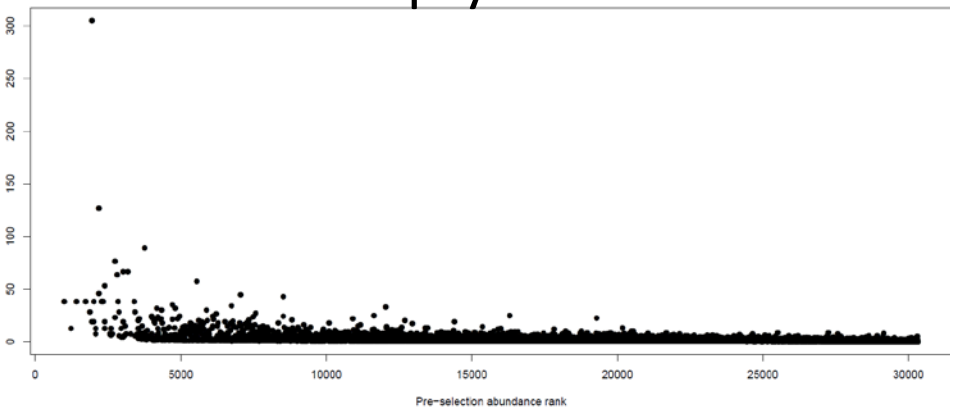


H. Salamon, ACS Chem. Biol. 2016; Harris, J. Med. Chem. 2017

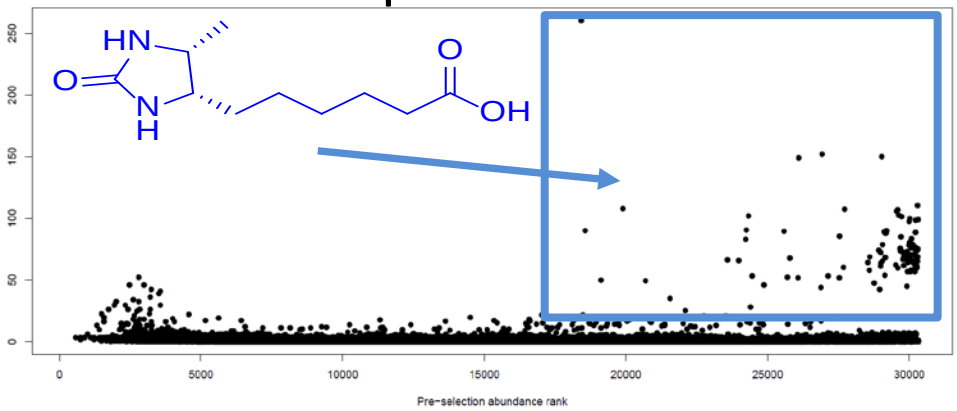
Proof of concept selection with streptavidin



empty beads



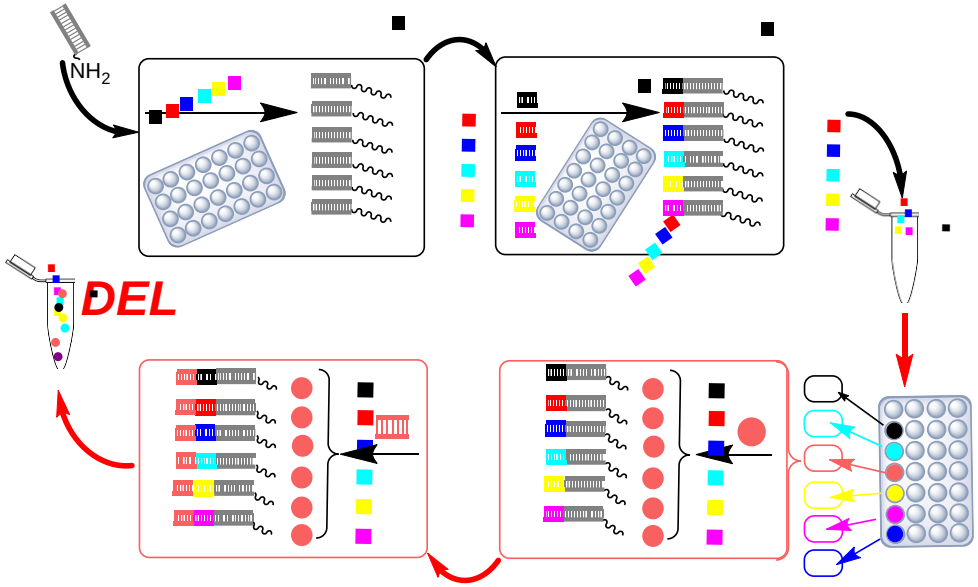
streptavidin beads

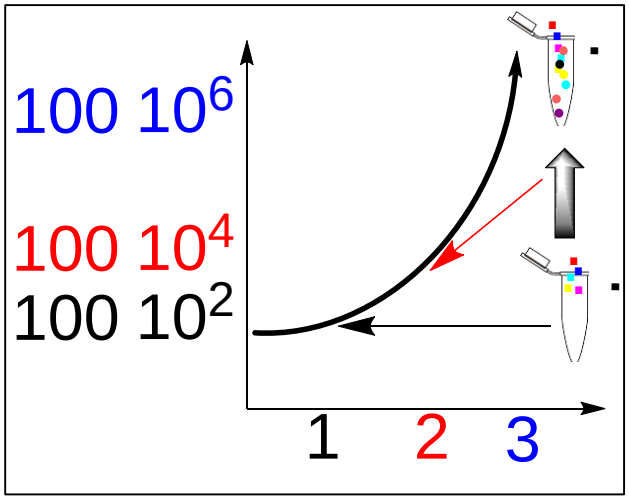


- process validated
- algorithm for sequencing data analysis
- evaluation of sequencing platforms
- barcoding strategies

DNA-recorded chemistry

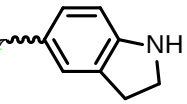
„headpiece“





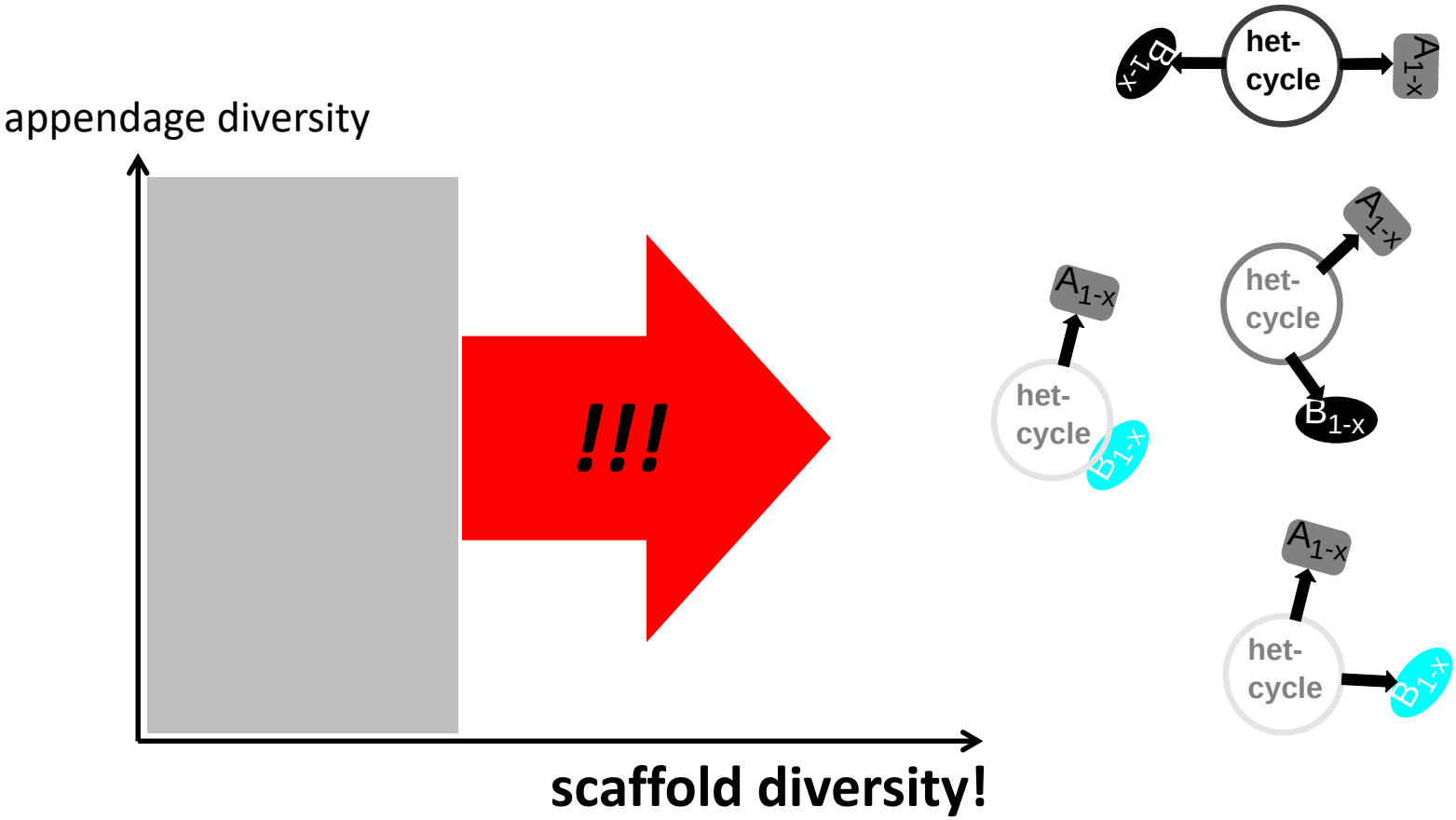
- split-and-pool synthesis
- exponential growth of libraries
- numerically large libraries

- DNA-compatible reactions
- water-tolerant reactions
- restricted synthesis toolbox



Clark, Nat. Chem. Biol., 2007
alternative formats: Liu et al. „DNA-templated chemistry; Harbury et al. „DNA-routing“; Neri et al. fragment selection

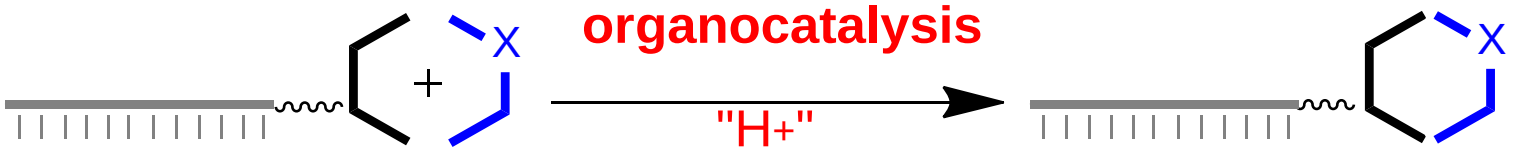
Our vision for DNA-encoded chemistry



- ✓ **Expand the chemical space**
- ✓ **Catalysis is a source of diversity**

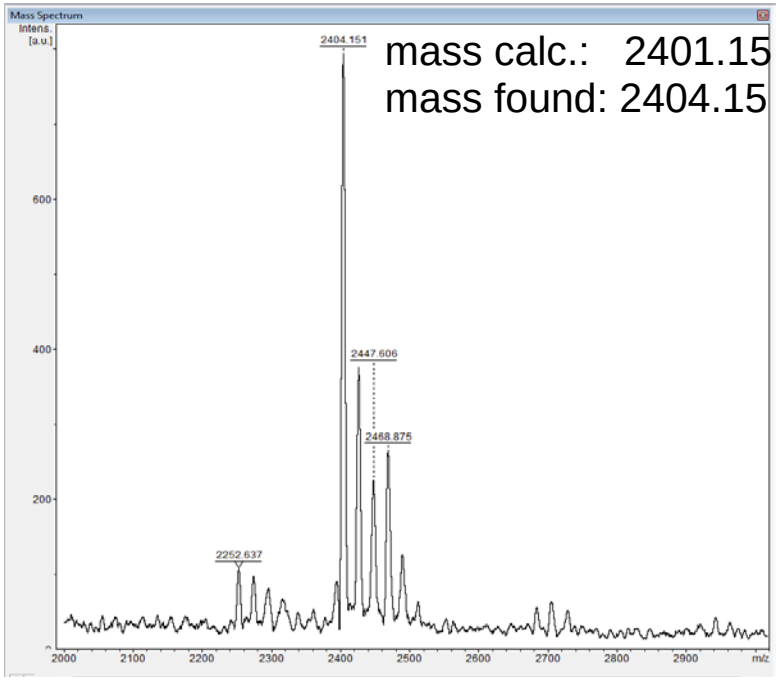
a) T. Kodadek, Chem Commun. 2011; b) A. L. Satz, Bioconjug Chem. 2015;
c) J. Wang, Proc Nat Acad Sci. 2018; d) D. C. Blakemore, Nat Chem. 2018.

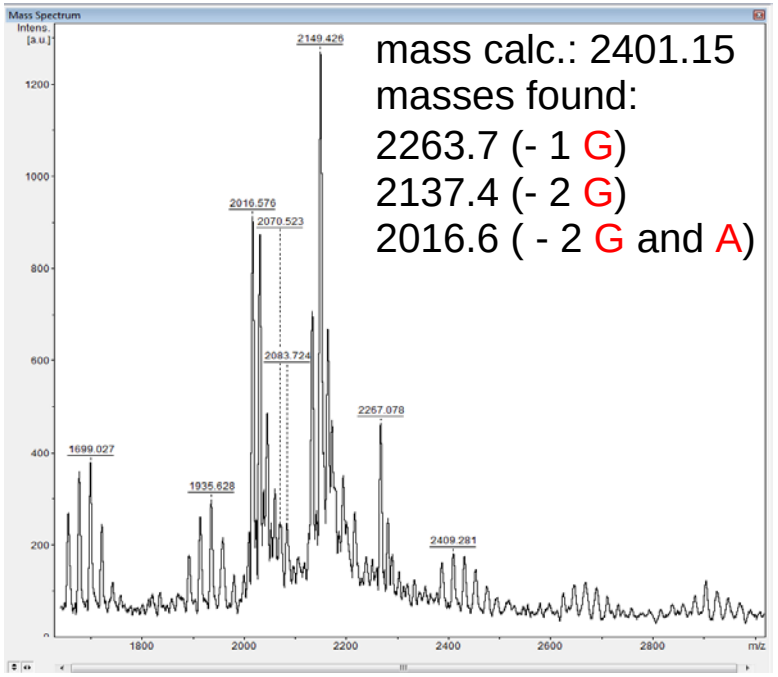
Chemical instability of DNA



3'—G-C-T-G-A-C—5'

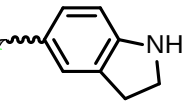
3'—~~G~~-C-T-~~G~~-~~A~~-C—5'

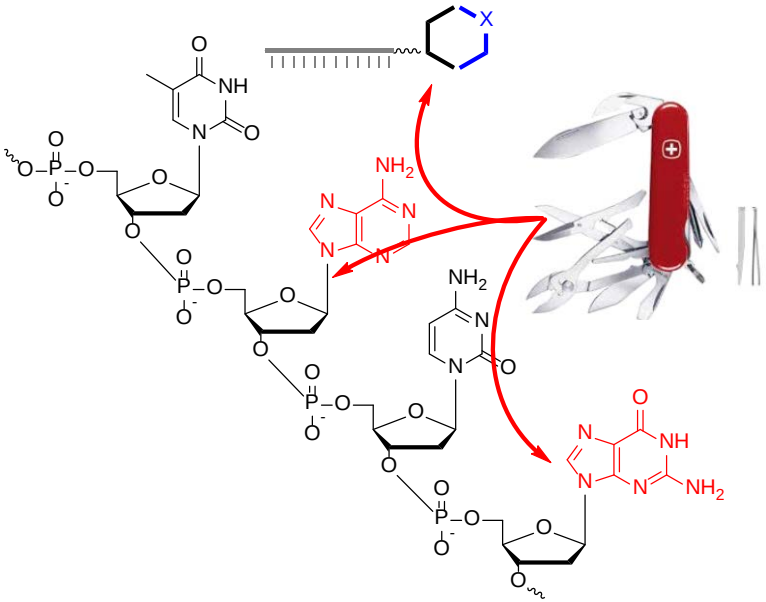


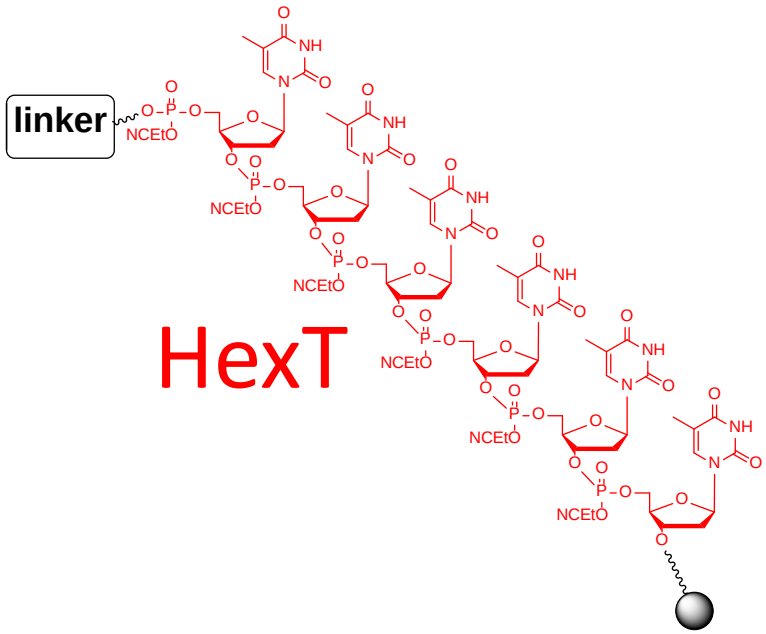


MS of a DNA prior treatment with 1% TFA

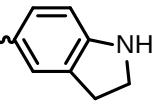
… and the day after



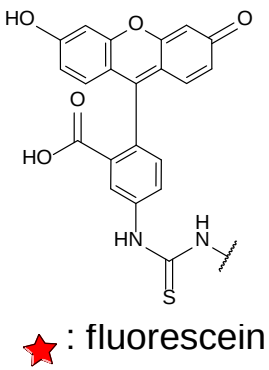




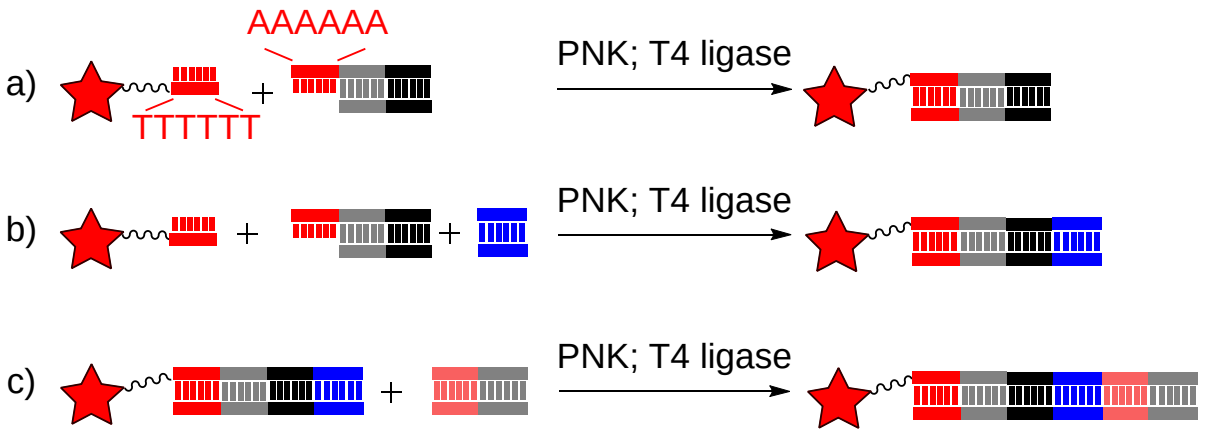
Adapt the encoding strategy to the chemistry

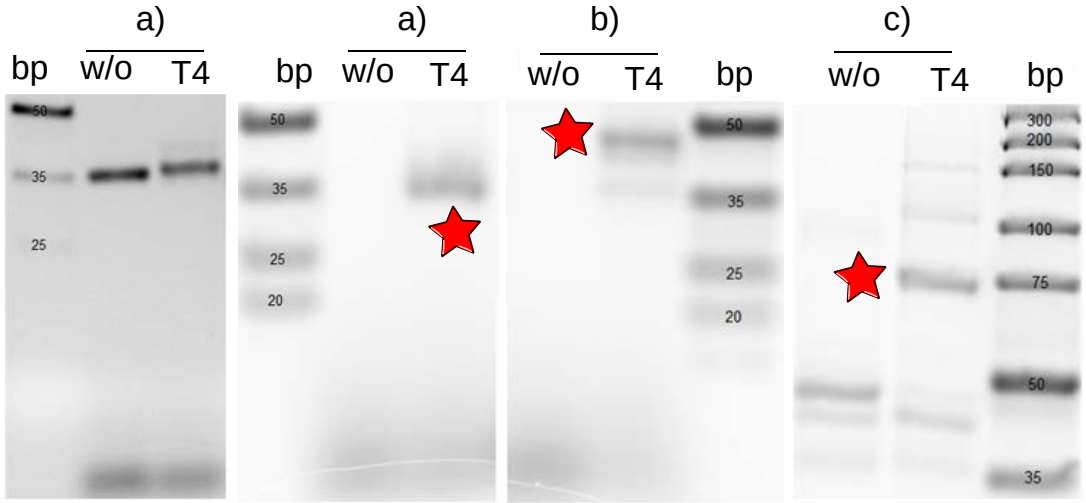


Ligation of hexT to dsDNA



: fluorescein

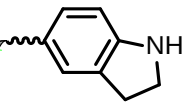




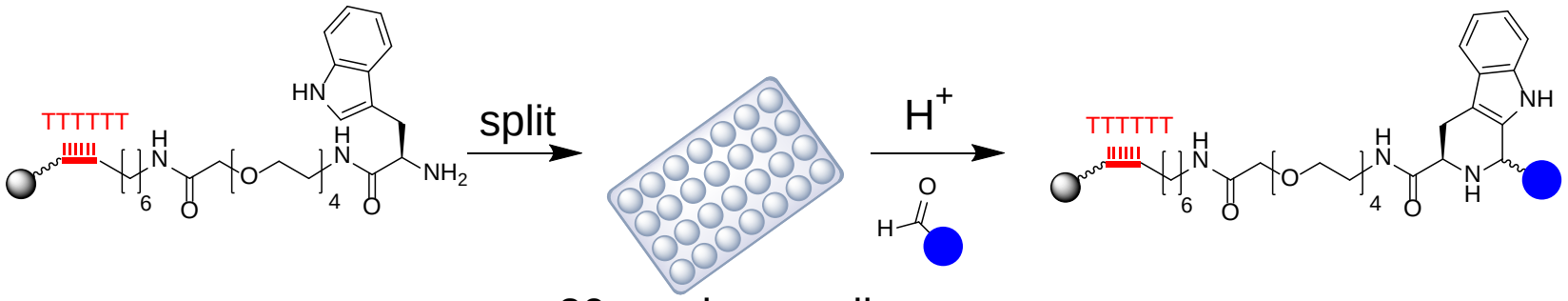
DNA stain

fluorescence scan

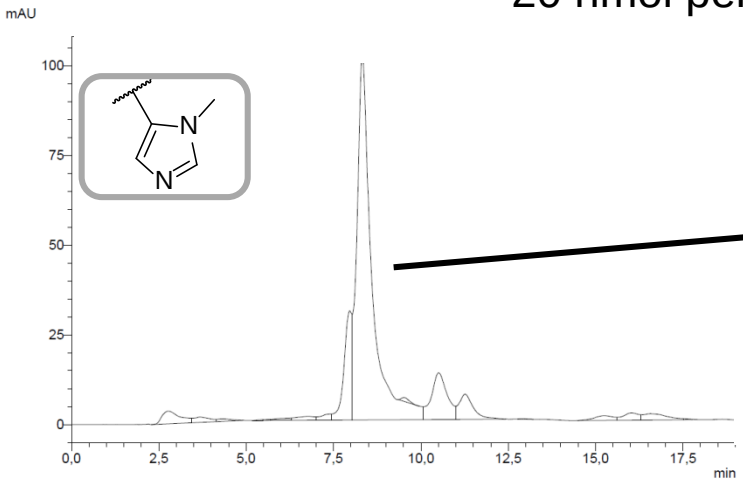
5,5% agarose gel; 75 min; 150V;
gel stain: Midori Green
grey: primer sequence

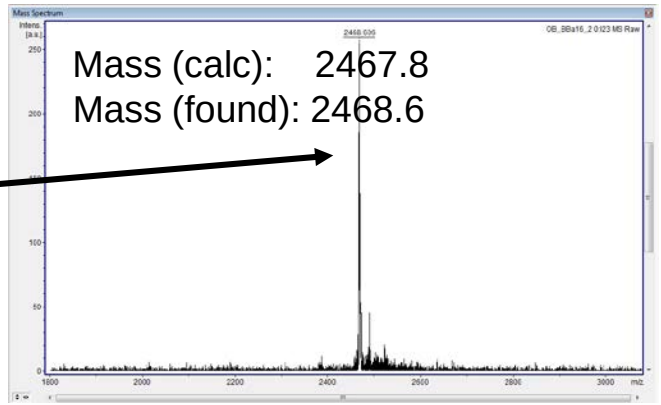


Catalysis



20 nmol per well



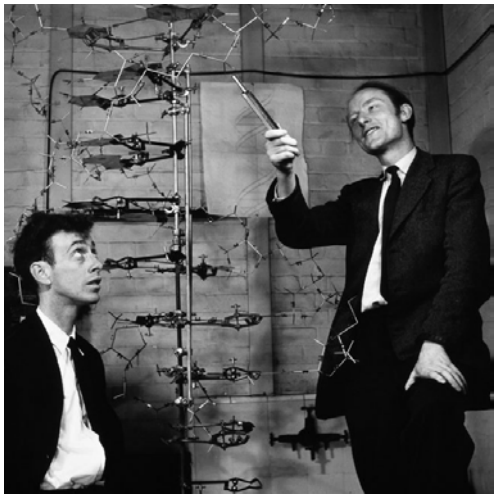


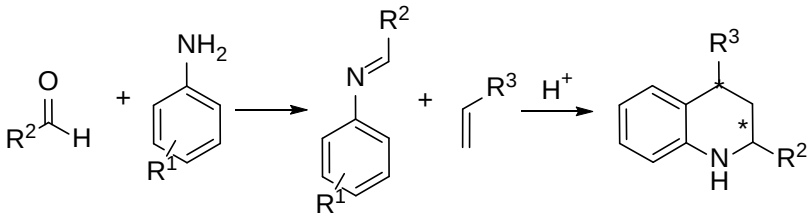
hexT tolerates H^+, and Lewis acids, e.g. Au(I), Cu(I/II), Ag(I), Ru(III), Pd(II)

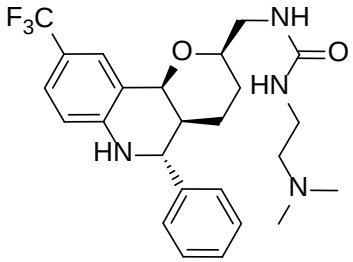
➡ larger scope of methods to initiate library synthesis

Klika Škopić, Org Biomol Chem., 2017
Klika Škopić, Chem Sci., 2017

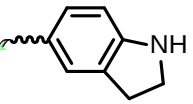
Gene-compatible catalysis?





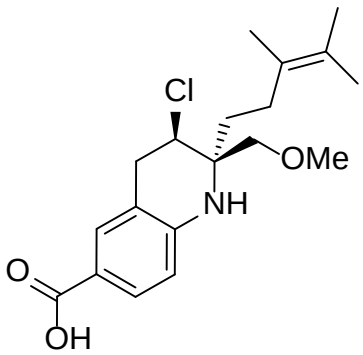


EMD 534085

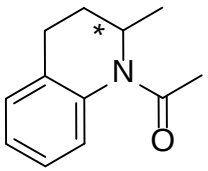


Schiemann, Bioorg Med Chem Lett. 2010

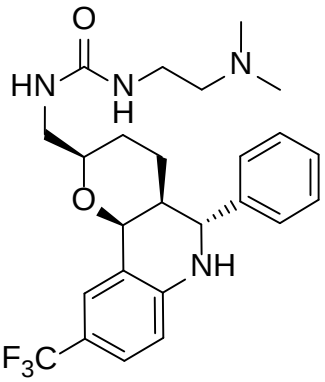
Target molecules



(-)-virantmycin

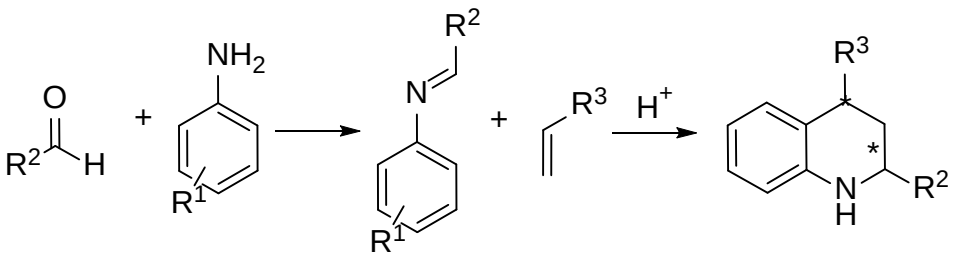


BET-targeting fragment



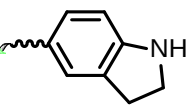
EMD 534085

tetrahydroquinoline – a scaffold found in many bioactive compounds

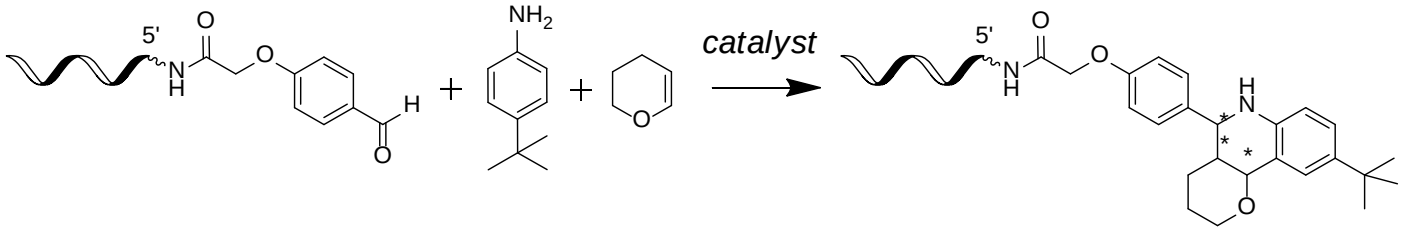


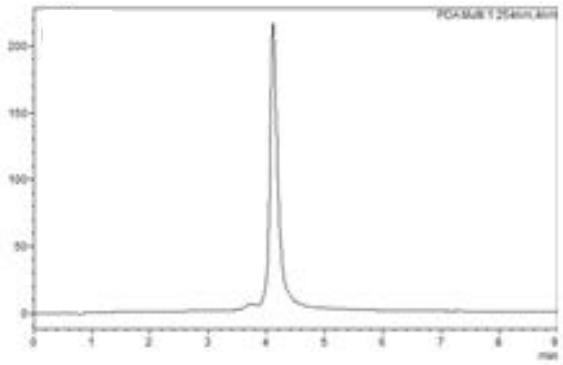
Povarov reaction gives access to densely substituted THQs

a) L. S. Povarov, Izv. Akad. Nauk SSR, Ser. Khim. 1963; b) T. Akiyama, J. Am. Chem. Soc. 2006;
c) H. Liu, J. Am. Chem. Soc. 2009; d) G. Dagousset, J. Am. Chem. Soc. 2011.

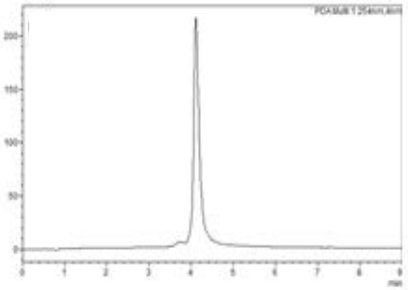


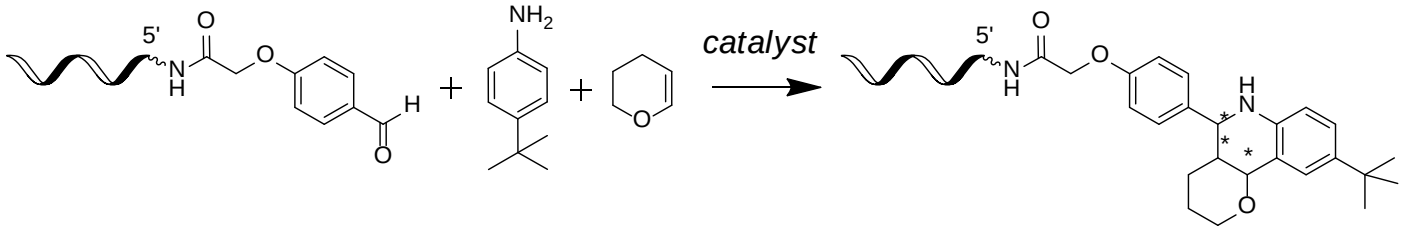
Homogeneous catalysis

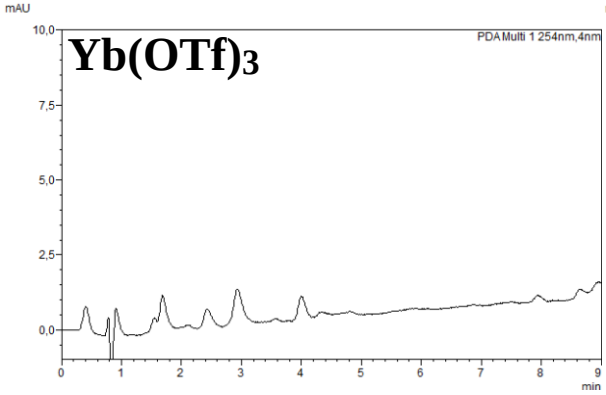


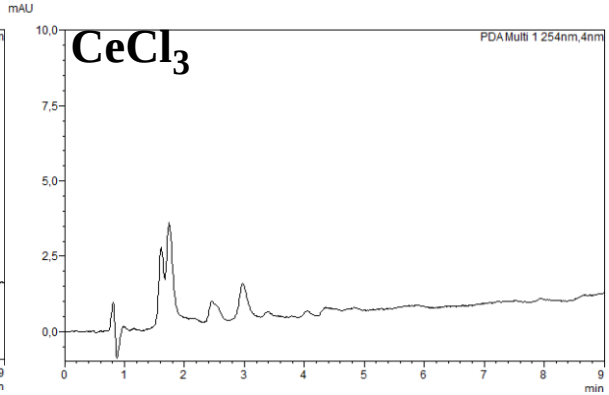


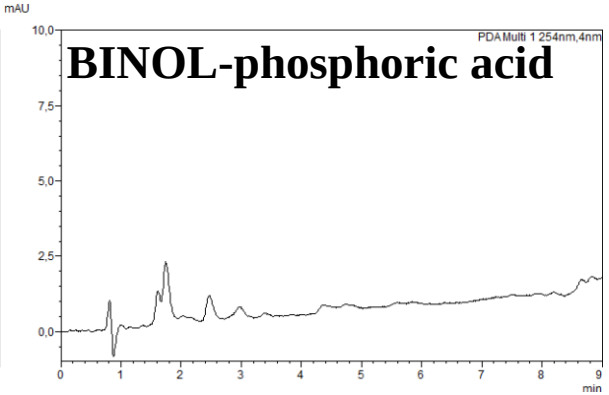
Homogeneous catalysis

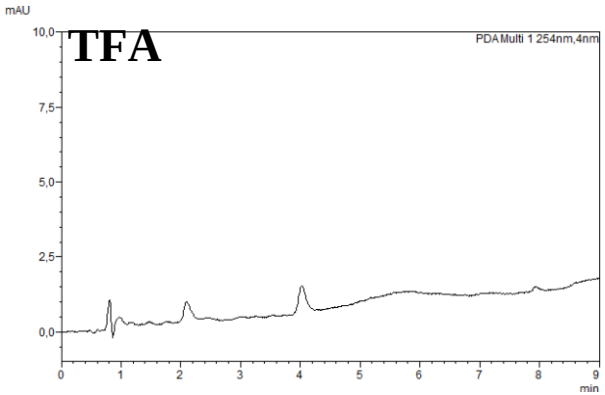


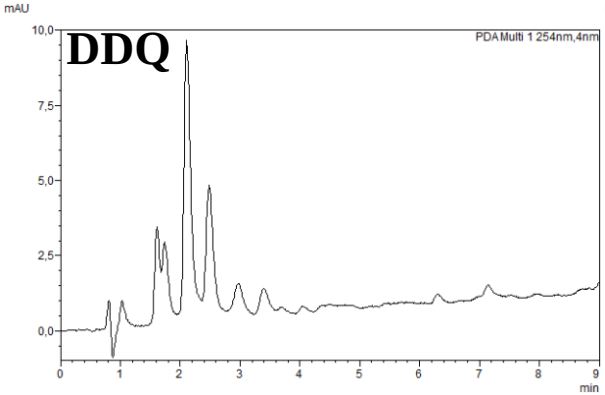


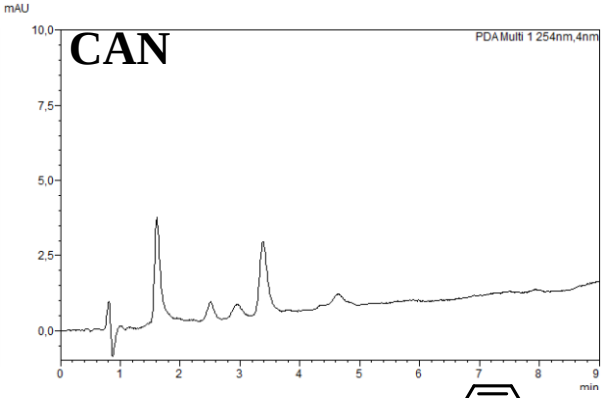


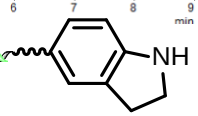




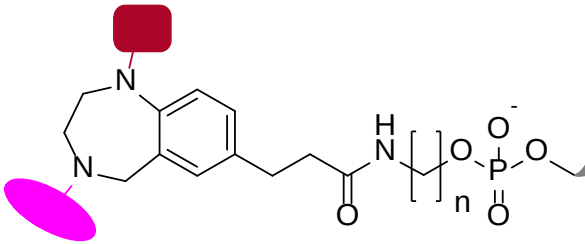




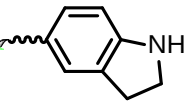




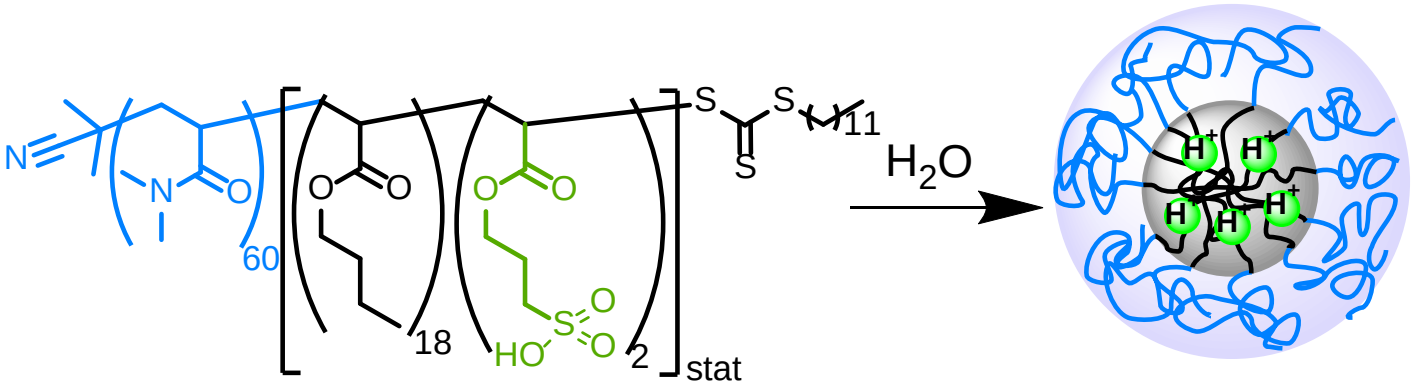
DNA conjugates – amphiphilic structures



„phenotype“ **„genotype“**



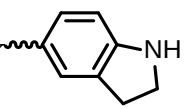
Amphiphilic polymers



- amphiphilic polymer with low CMC (ca. 1 µM)
- formation of spherical nanoreactors with steep polarity gradient
- compartmentation of reaction medium
- sulfonic acid immobilized in the core

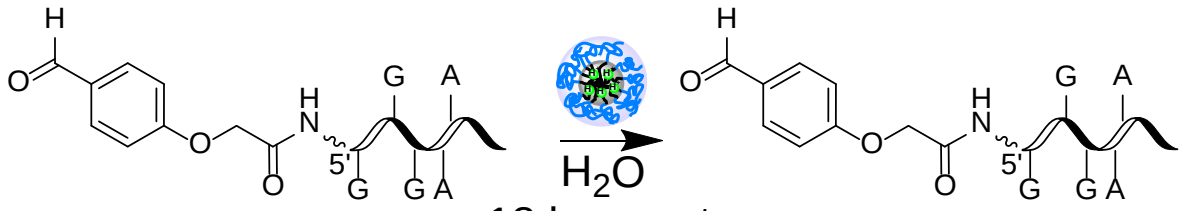
Selective reaction of DNA-conjugated starting materials?

with Prof. R. Weberskirch (TU Dortmund)

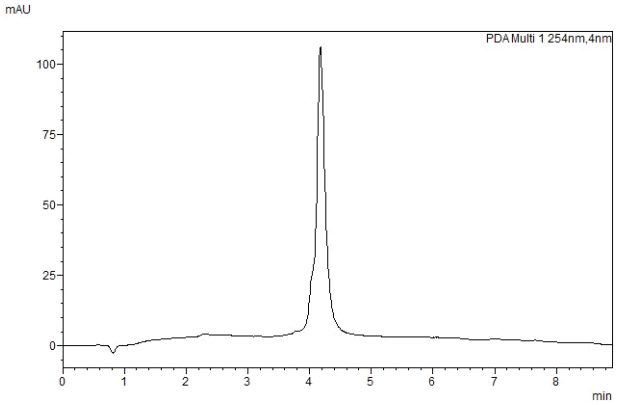


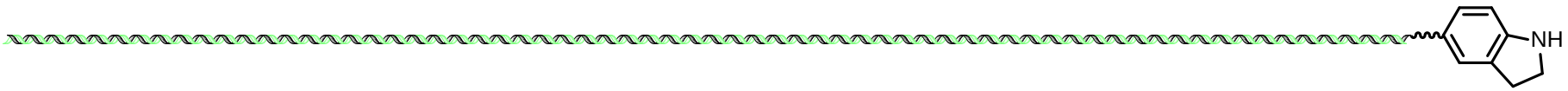
a) I. V. Berezin, Russ. Chem. Rev. 1973; b) T. Dwars, Angew. Chem. Int. Ed. 2005;
c) B. H. Lipshutz, Aldrichimica Acta. 2012; d) A. Lu, Curr. Opin. Biotechnol. 2013.

DNA stability

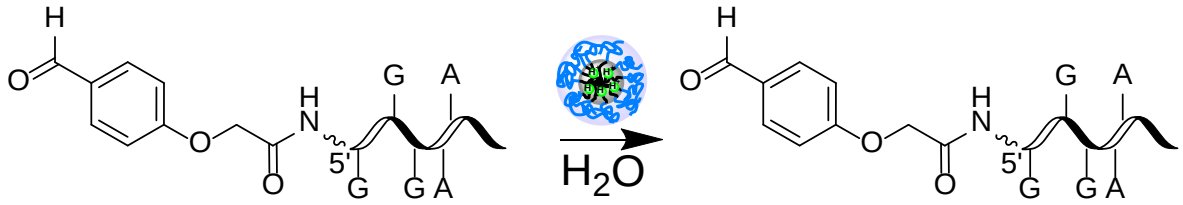


5'-GTC TTG CCG AAT TC





DNA stability



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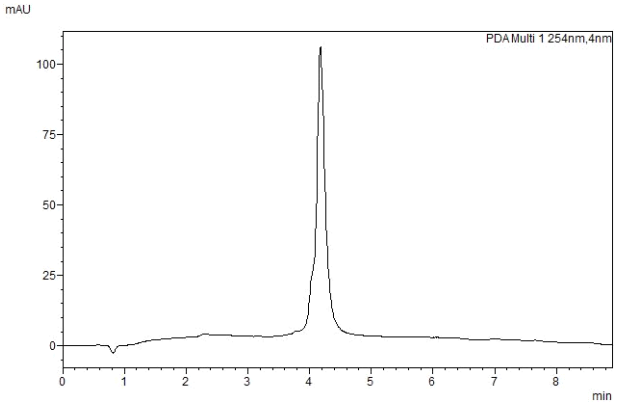
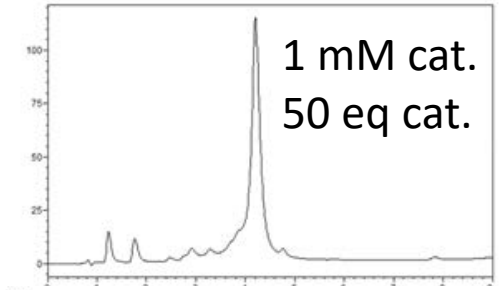
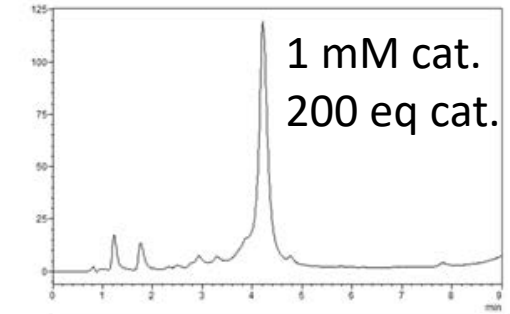
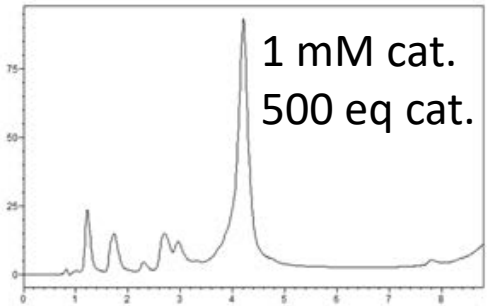
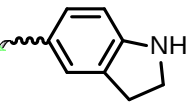


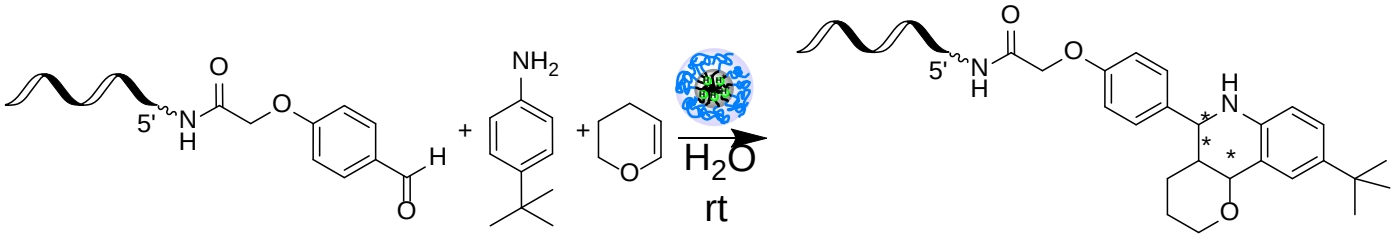


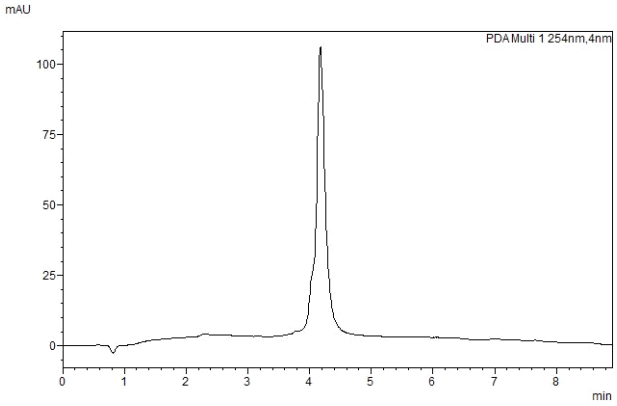


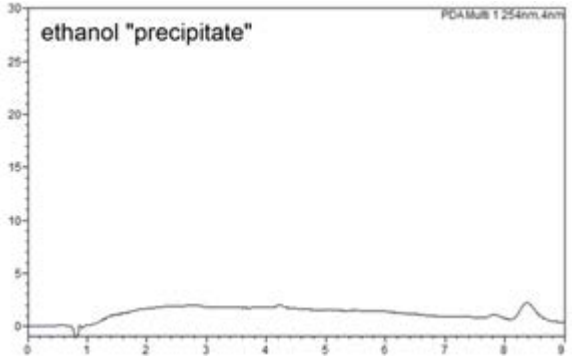


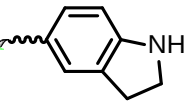


Micellar catalysis

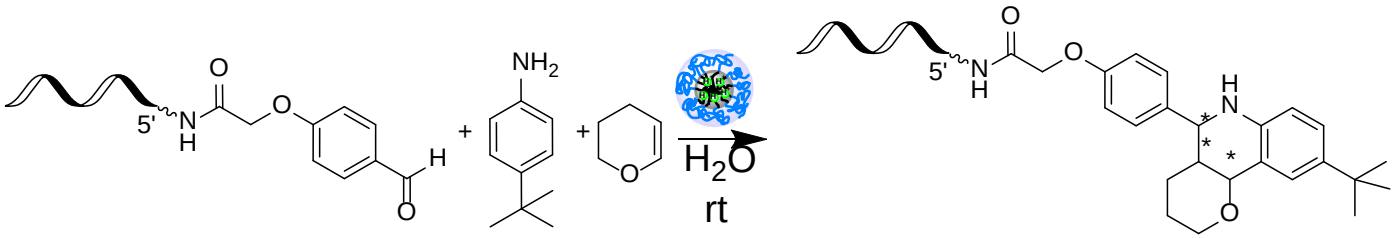


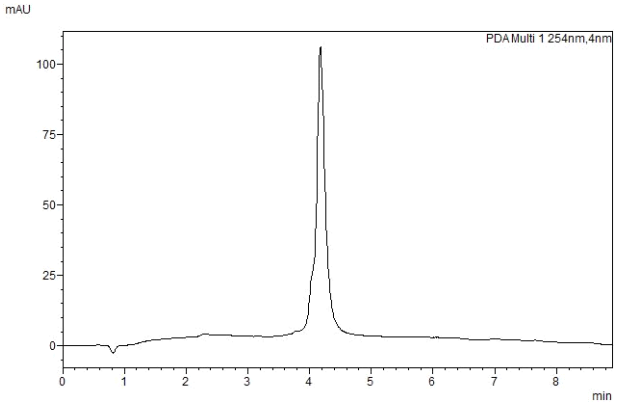


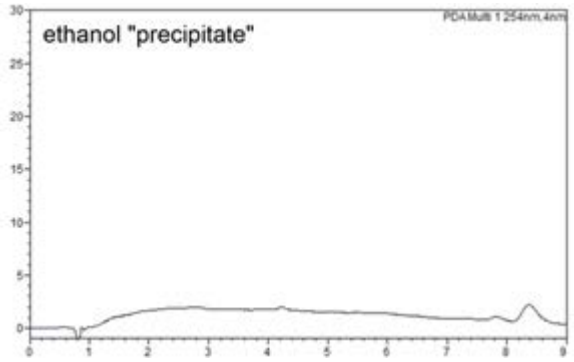


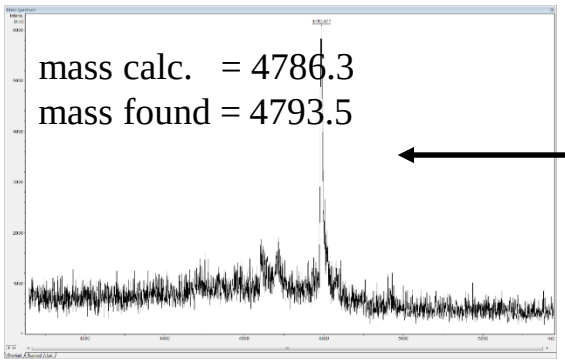


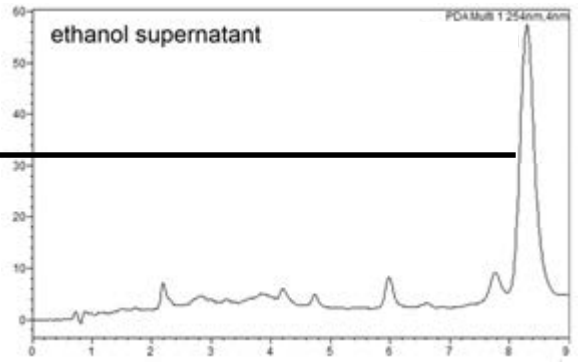
Micellar catalysis





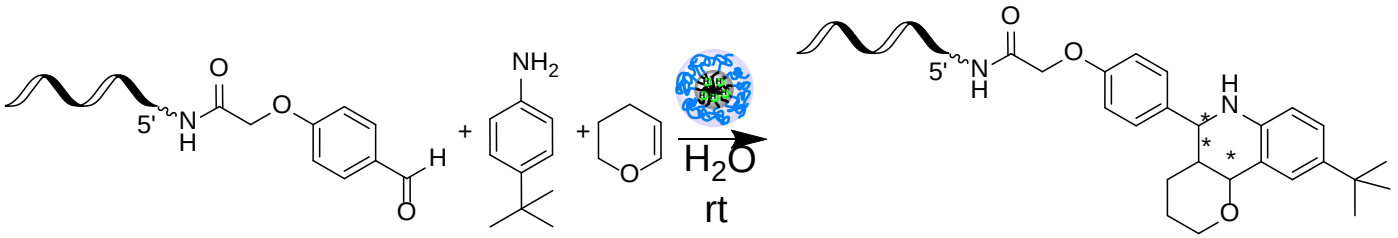


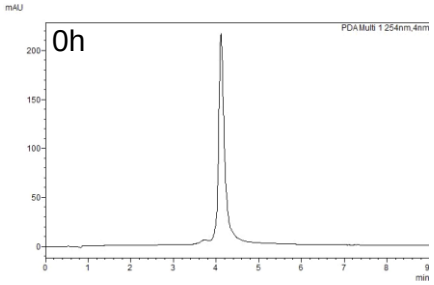


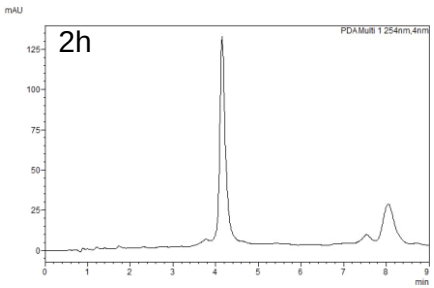


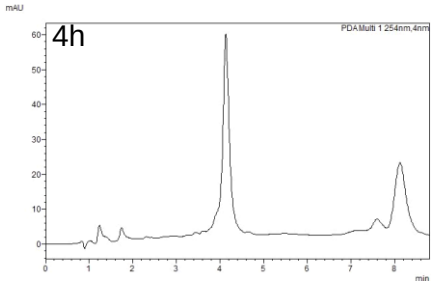
Thorough extraction of the reaction mixture depletes the polymer.

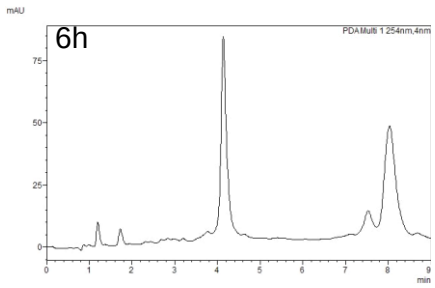
Micellar catalysis

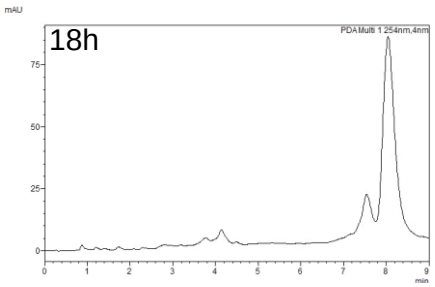


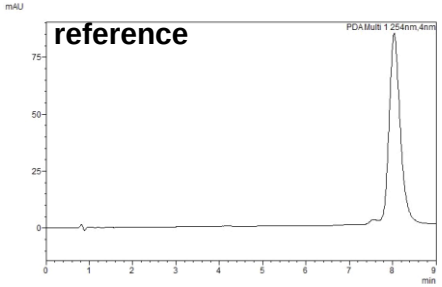


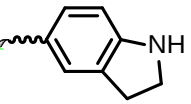




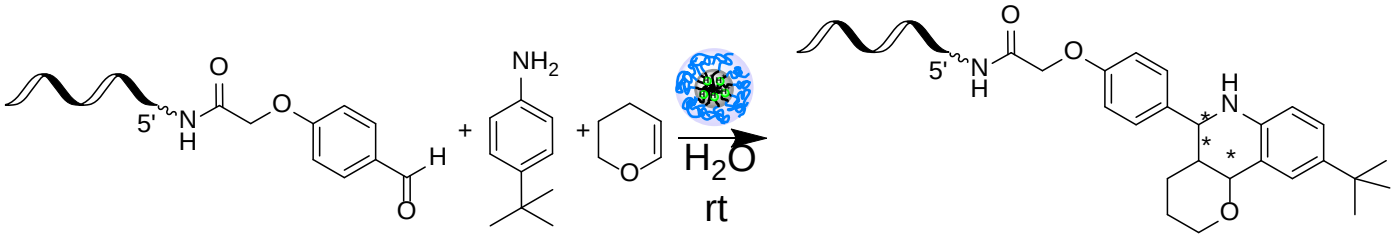


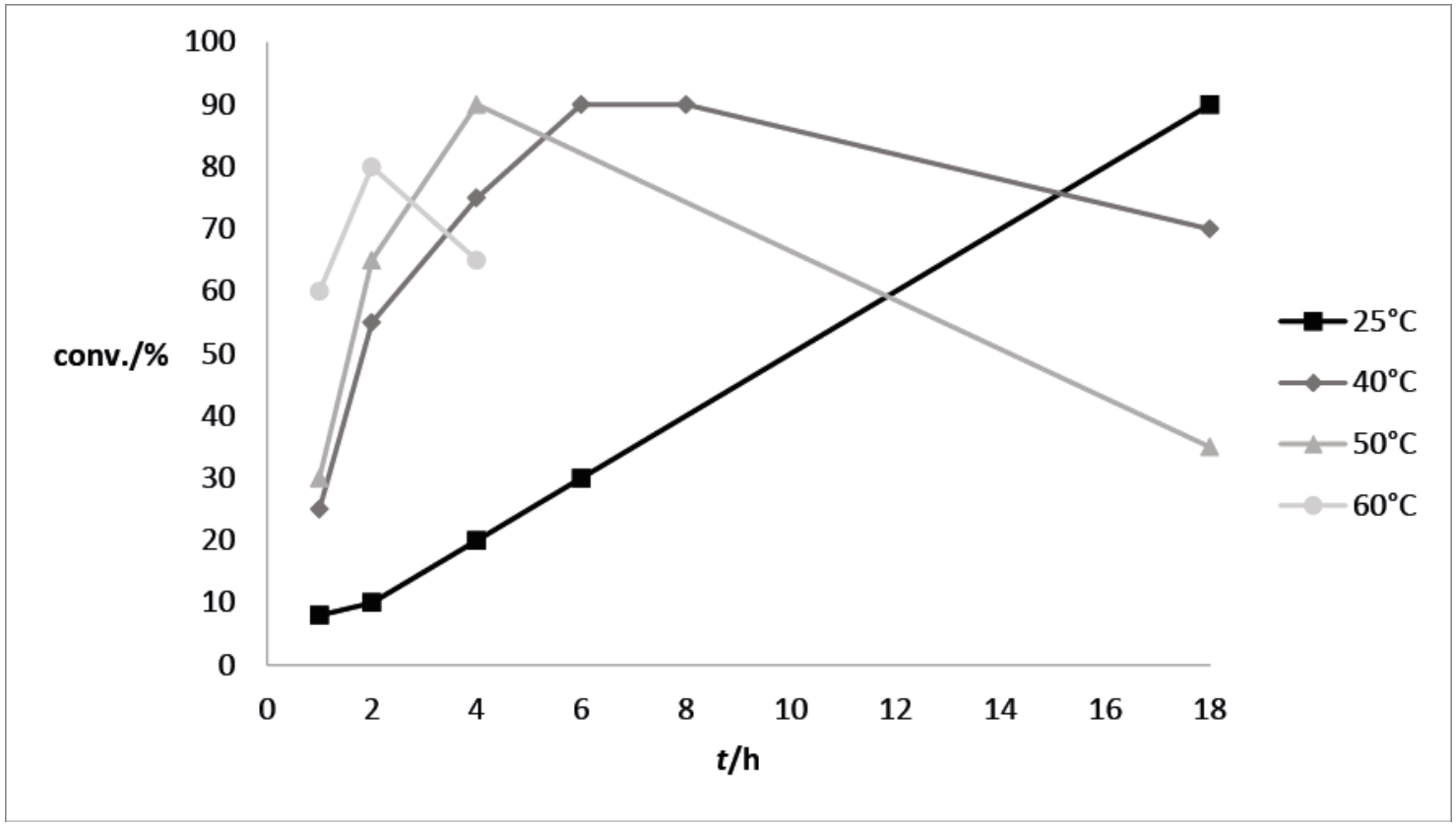


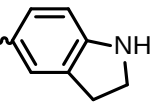




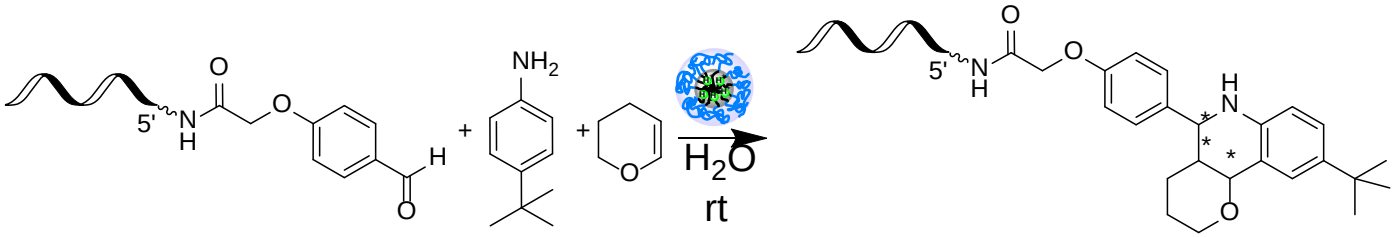
Micellar catalysis: kinetics

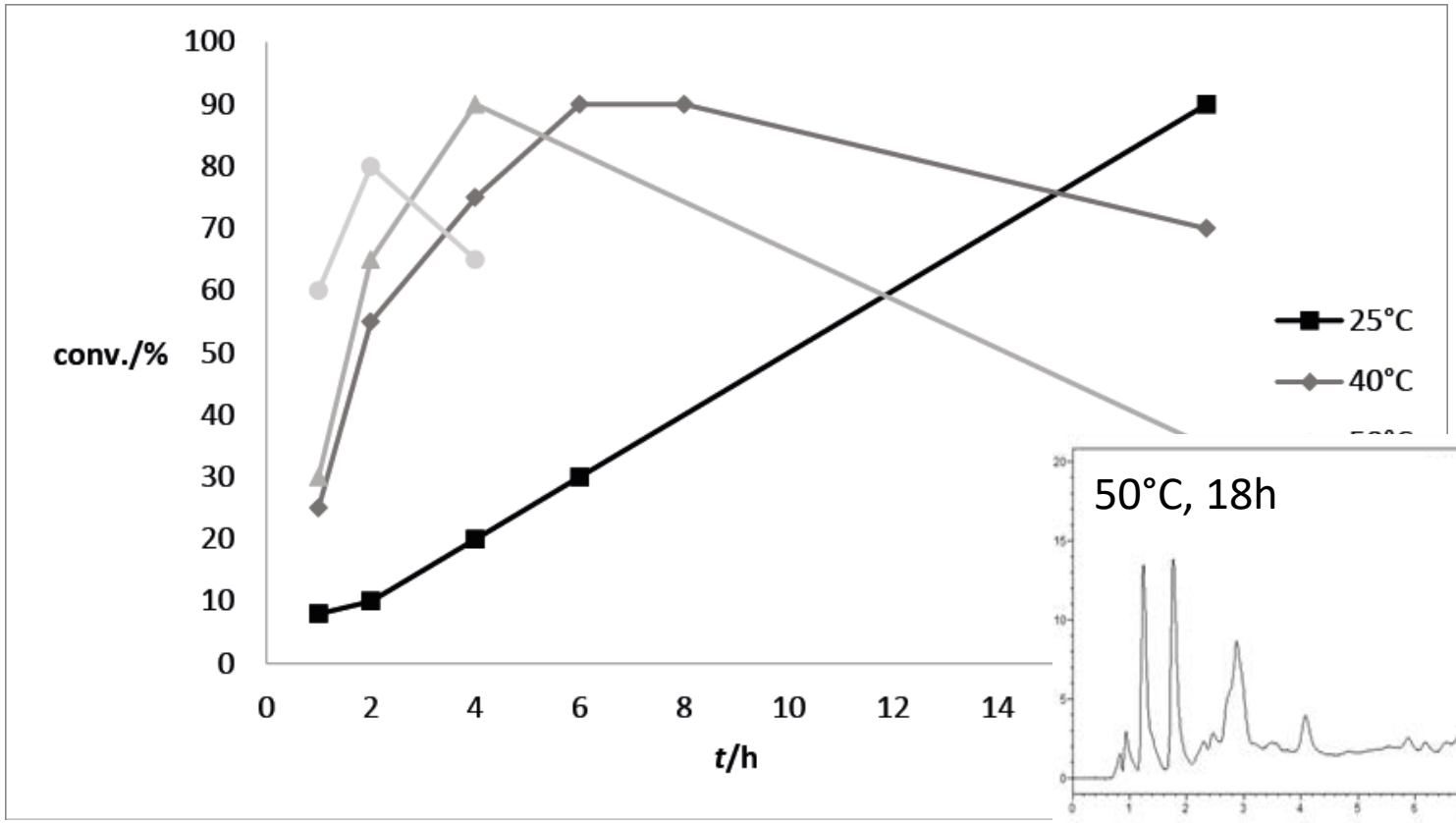




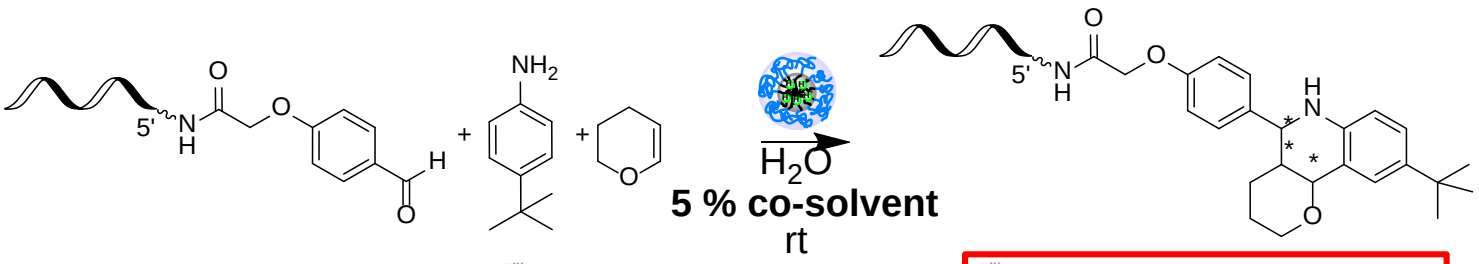


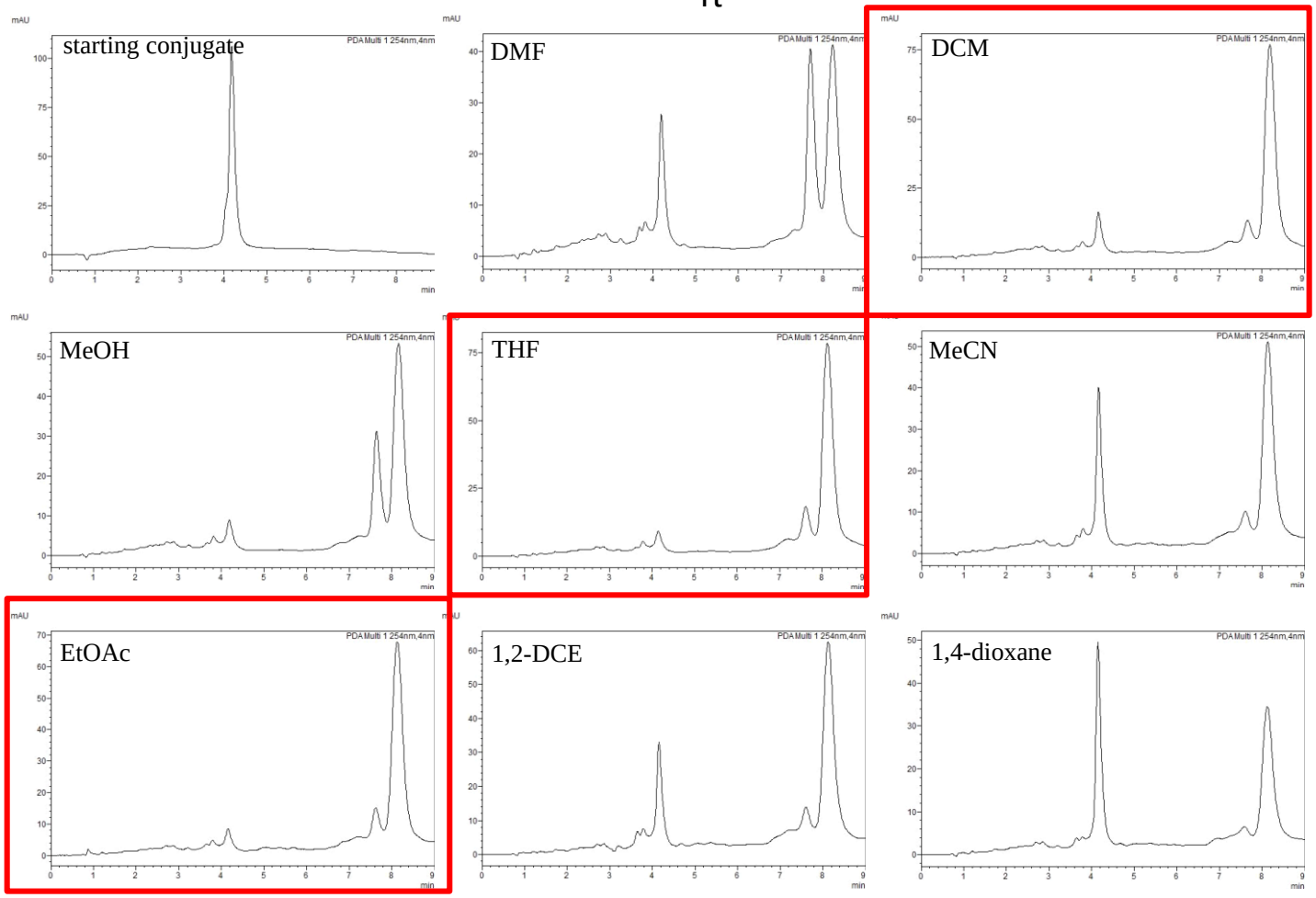
Micellar catalysis: kinetics



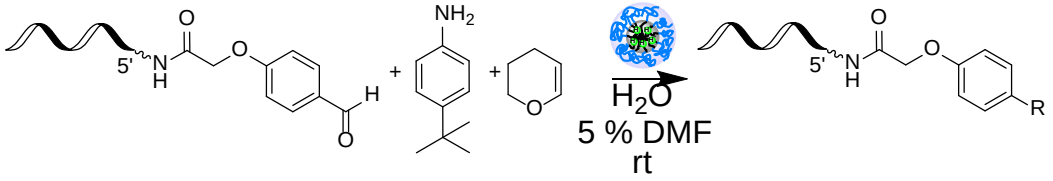


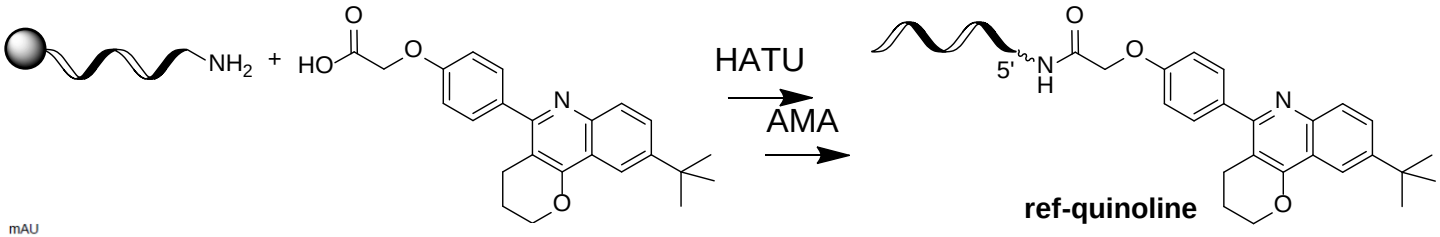
Micellar catalysis: co-solvents

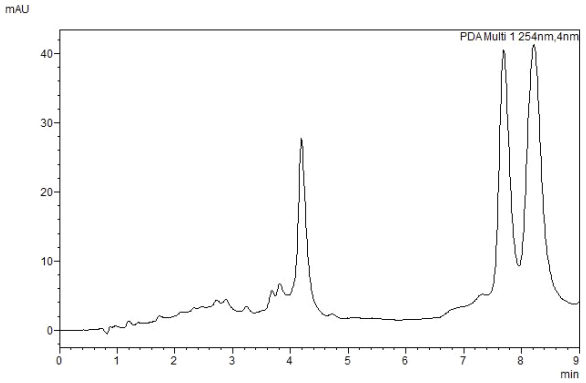


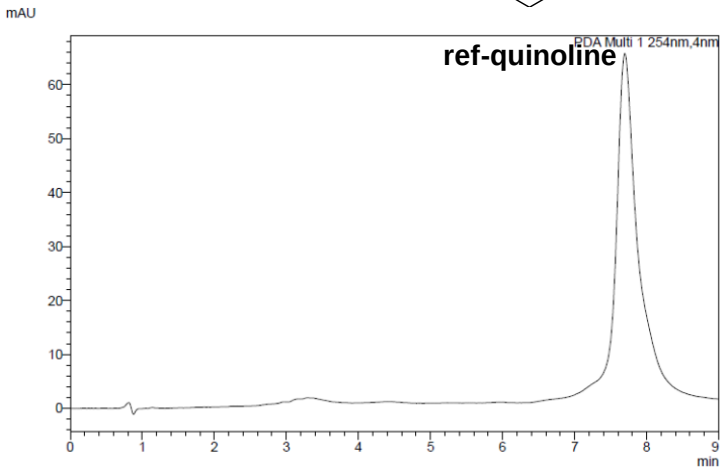


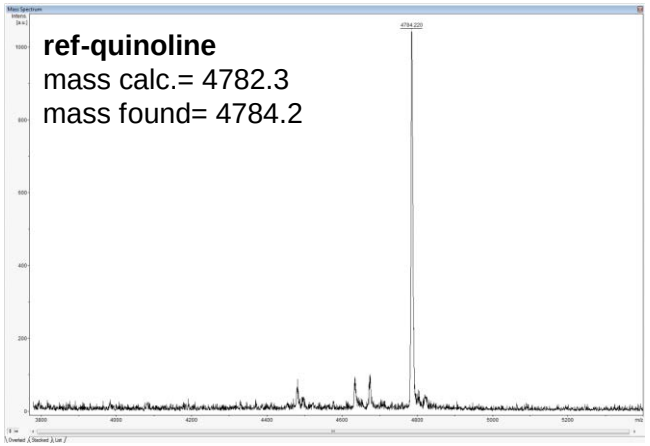
Micellar catalysis: side-product formation

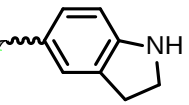




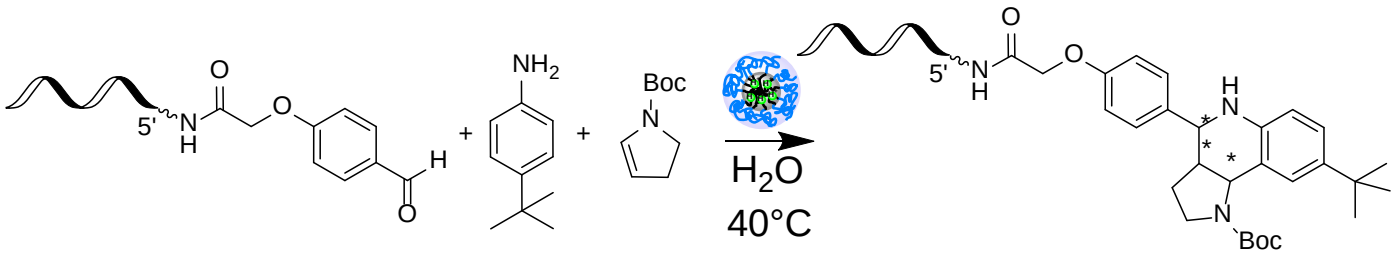


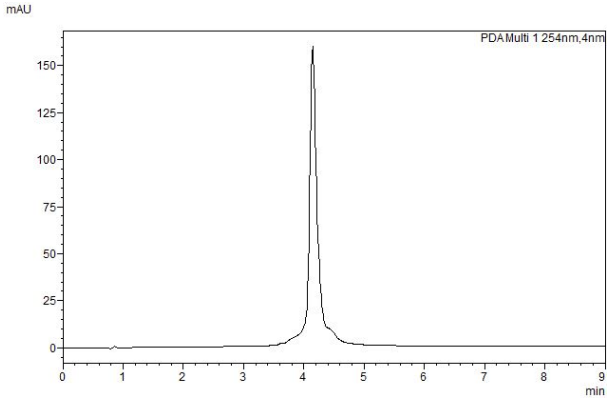


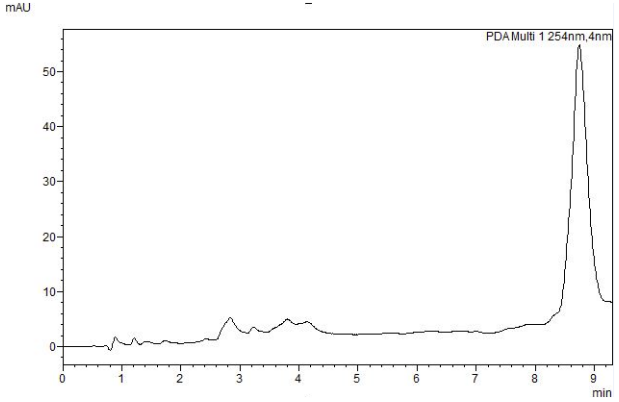


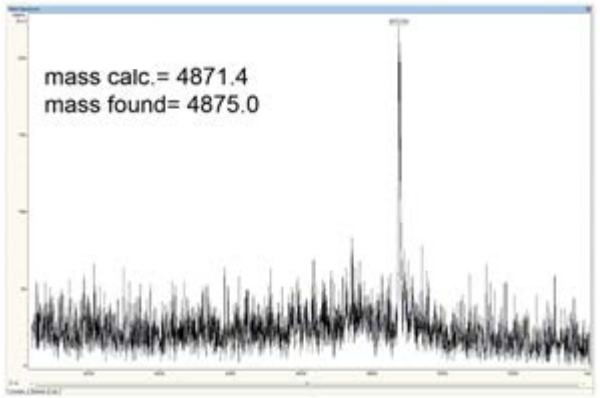


Micellar catalysis: scope

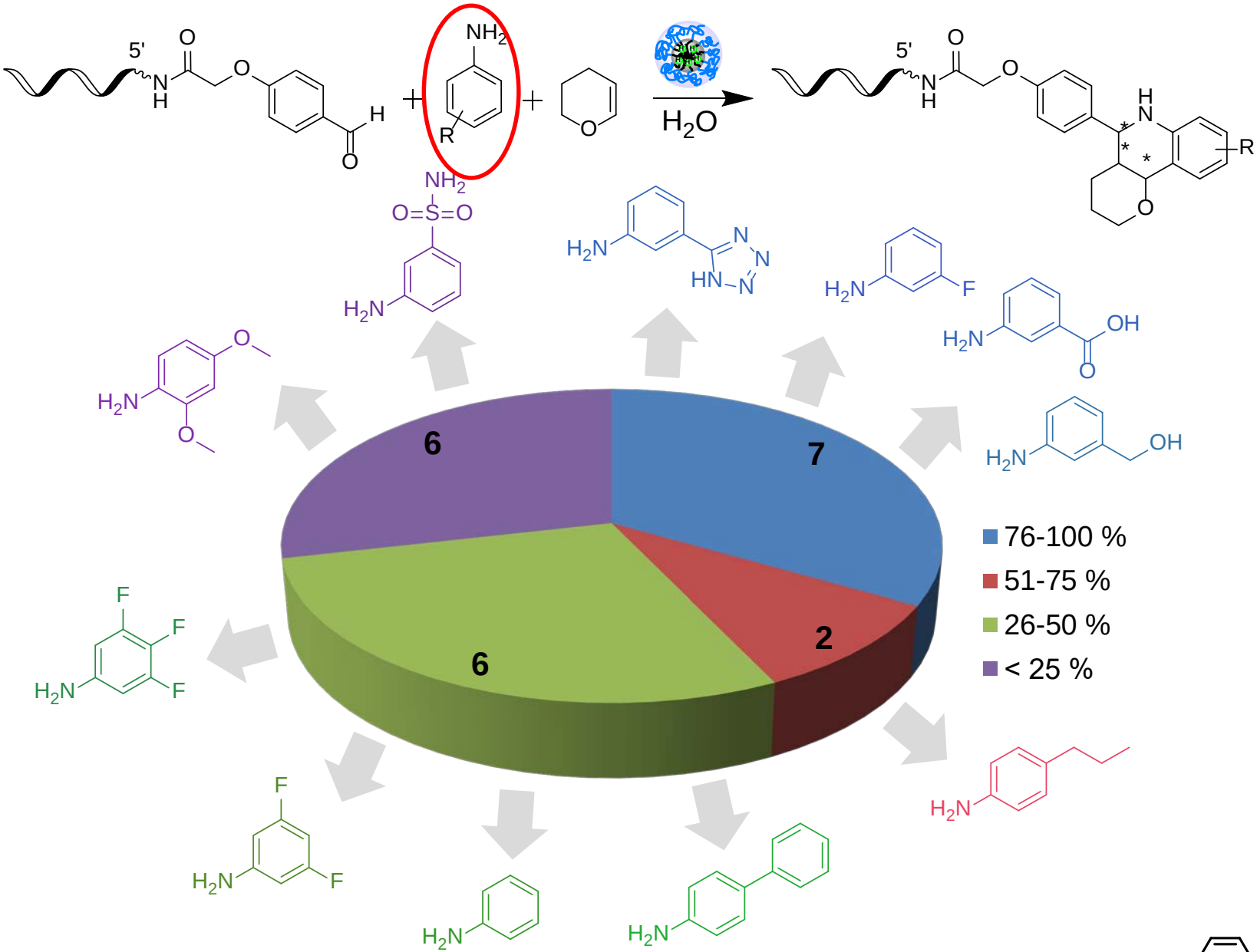






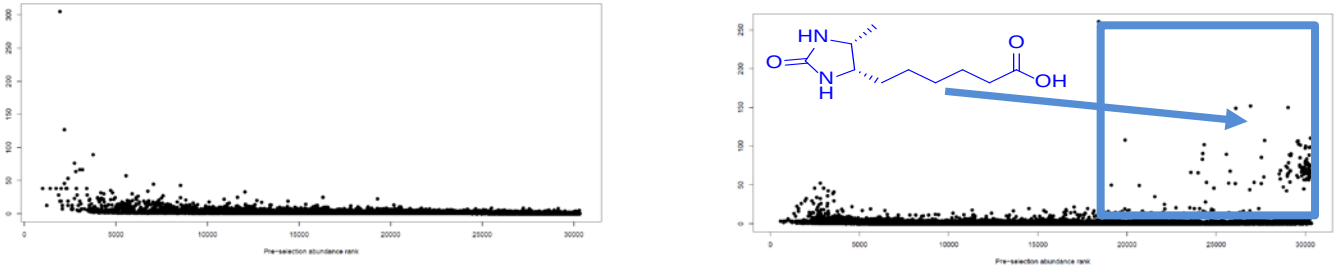


Micellar catalysis: scope

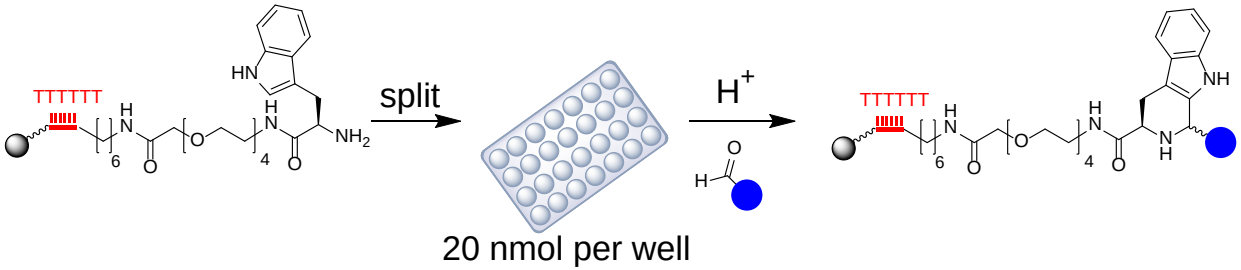
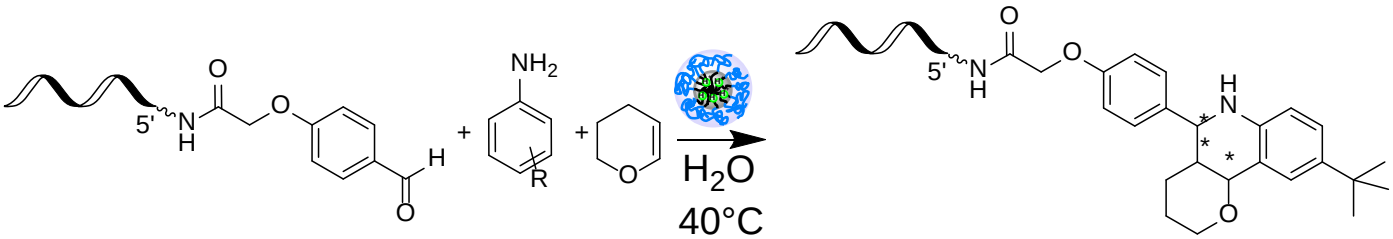


conclusions

- algorithm development for sequencing data analysis



- development of encoding strategies



- reaction systems for DEL synthesis