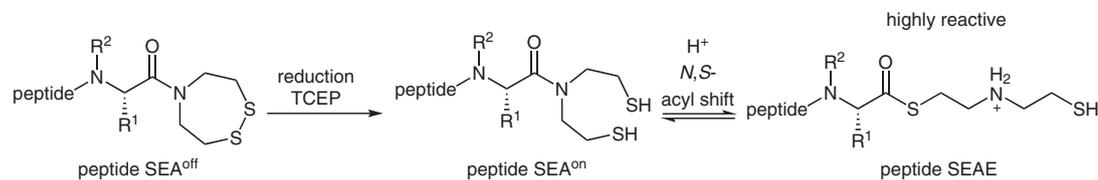


Native Chemical Ligation by Microflow Chemistry



Microfluidic system:

feed solution 1

pPeptide SEA^{on} (7 mM)
 TCEP (100 mM), pH 1
 CyslleLeuLysGluProValHisGlyVal-SEA^{on}
 CyslleLeuLysGluGlyValHisGlyVal-SEA^{on}
 CyslleLeuAsnGlyProValHisGlyVal-SEA^{on}
 CyslleLeuAspGlyProValHisGlyVal-SEA^{on}
 CyslleLeuAsnGlyProValHisGlyLeu-SEA^{on}
 CyslleLeuAspGlyProValHisGlyLeu-SEA^{on}
 CyslleLeuLysGluGlyValHisGlyThr-SEA^{on}
 CyslleLeuLysGluGlyValHisGlyIle-SEA^{on}
 CyslleLeuLysGluGlyValHisGlyPro-SEA^{on}

feed solution 2

decane

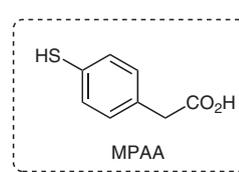
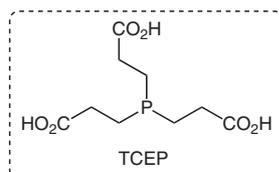
$t^1 = 60$ min

$T^1 = 90$ °C or 65 °C

peptide SEAE

cyclic peptides

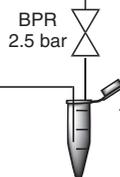
c-(CyslleLeuLysGluProValHisGlyVal) (46%)
 c-(CyslleLeuLysGluGlyValHisGlyVal) (46%)
 c-(CyslleLeuAsnGlyProValHisGlyVal) (66%)
 c-(CyslleLeuAspGlyProValHisGlyVal) (37%)
 c-(CyslleLeuAsnGlyProValHisGlyLeu) (80%)
 c-(CyslleLeuAspGlyProValHisGlyLeu) (64%)
 c-(CyslleLeuLysGluGlyValHisGlyThr) (49%)
 c-(CyslleLeuLysGluGlyValHisGlyIle) (51%)
 c-(CyslleLeuLysGluGlyValHisGlyPro) (38%)



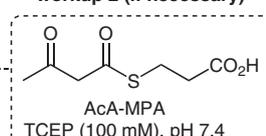
feed solution 3

MPAA (0.4 M)
 Na₂HPO₄ (0.2 M)
 pH = 7.8

pH = 7.3
 $t^2 = 4$ min
 $T^2 = 37$ °C



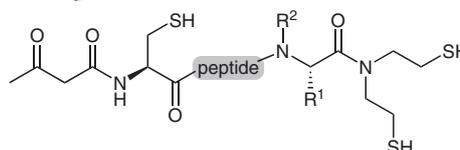
workup 2 (if necessary)



workup 1 (all)

acidification with 10% aq AcOH
 then extraction with Et₂O

HPLC



Significance: The authors reported a fast and highly efficient intramolecular cyclization of peptides with native chemical ligation under homogeneous microfluidic conditions, in which the formation of highly active S-[2-[(2-sulfanylethyl)amino]-ethyl] peptidyl thioesters is a key step.

Comment: This scale-independent microfluidic native chemical ligation proceeds rapidly, even for difficult junctions, and realizes an expedient preparation of bioactive macrocyclic peptides.