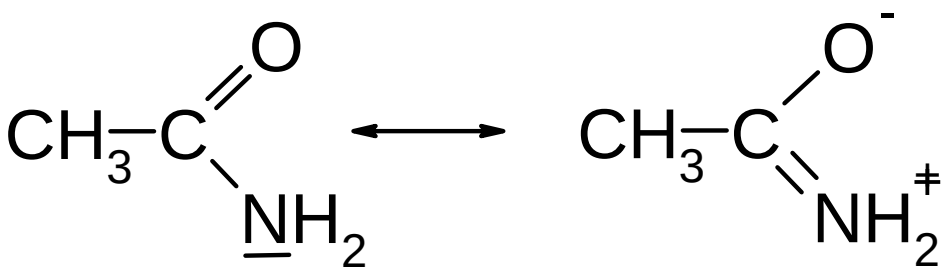
Peptides
Properties-Synthesis-Hydrolysis

Resonance



Properties:

Neutral (free electron pair of N: delocalized)

Planar unit (sp^2-hybridisation of C=N), C-N bond is shortened

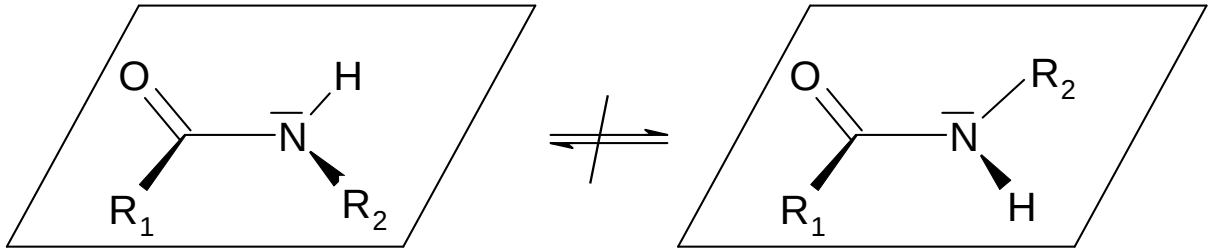
Restricted rotation – partial double bond character!

Stable (loss of positive character on carbonyl atom - nucleophilic addition less likely)

The amide linkage forms the basis of stability and three-dimensional structure of peptides and proteins

Hydrolysis of peptides: 6 M HCl, 105°

Peptides
Properties-Synthesis-Hydrolysis

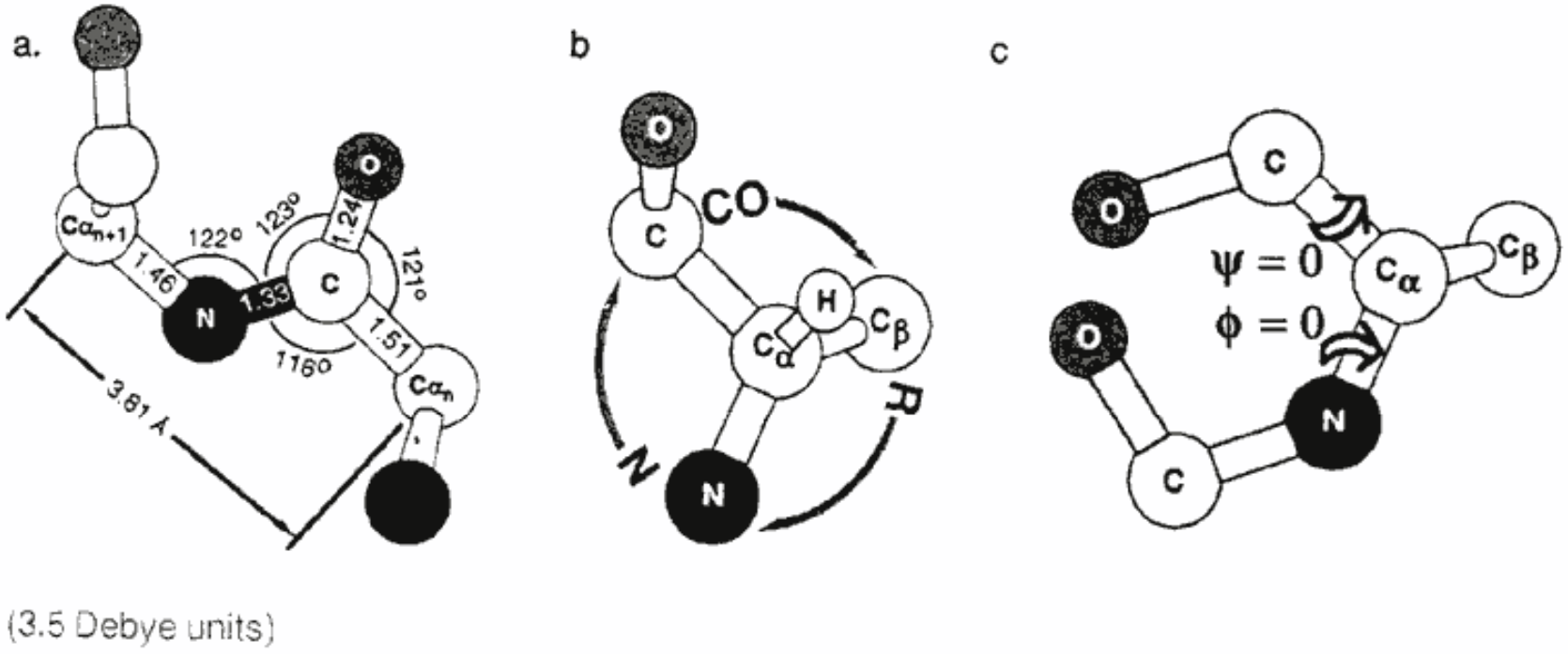


cis *trans*

Rotational barrier: 18.8 kcal/mol (41.8 kJ/mol)

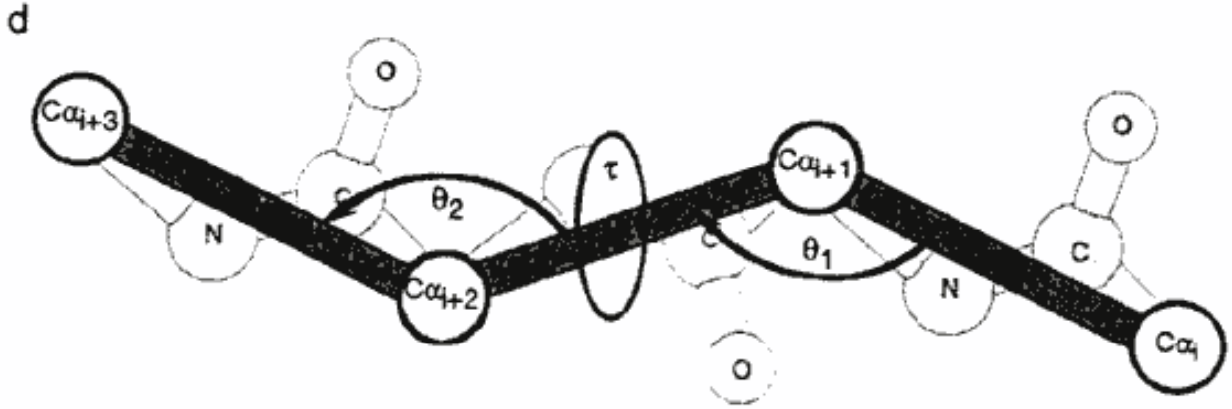
Rate of isomerisation at 40º : ~ 0.15 s^{-1}
Trans-form more stable (factor 10^3)
Exception: proline (factor 4)

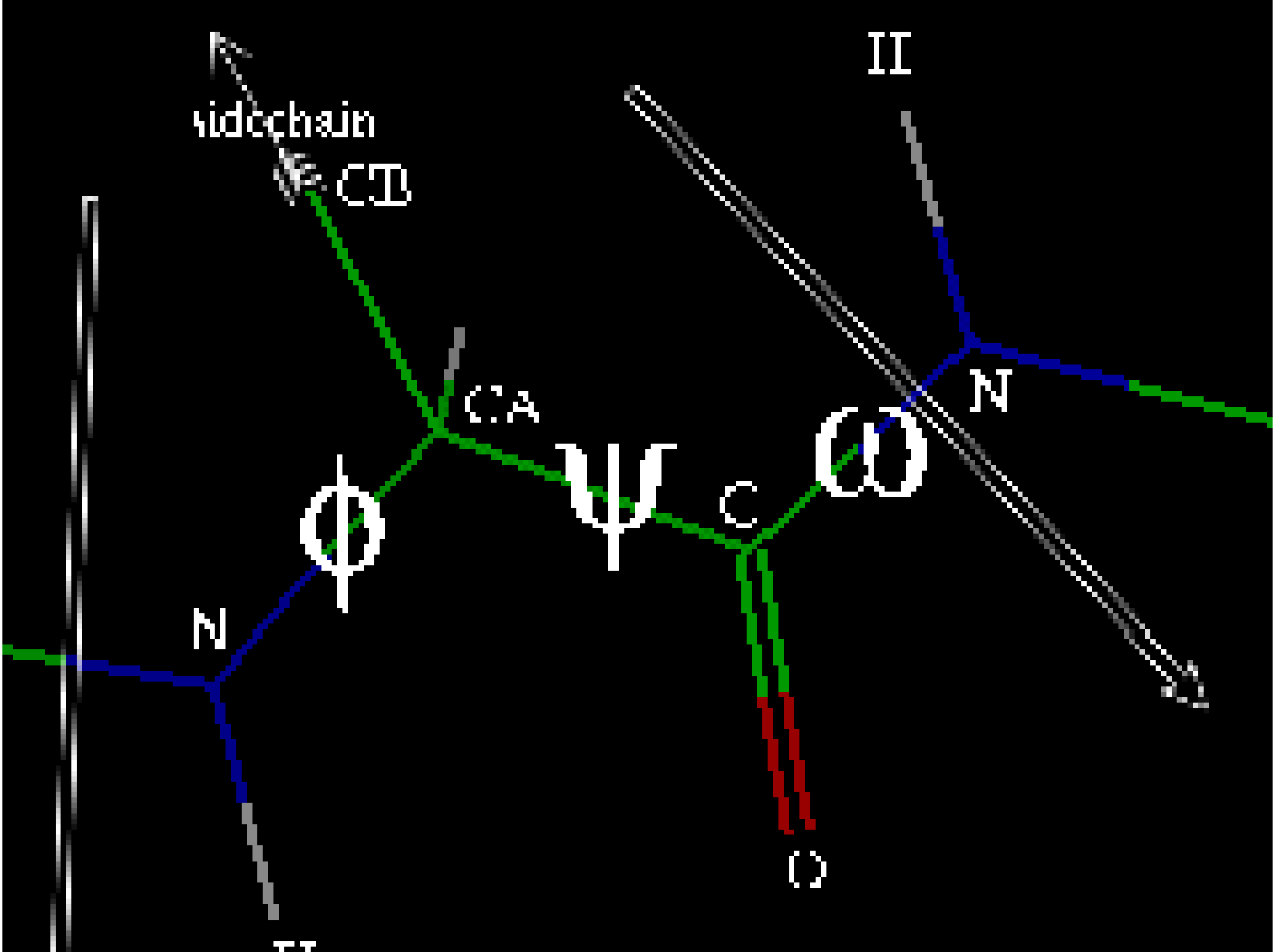
Geometry of peptide bond



(3.5 Debye units)

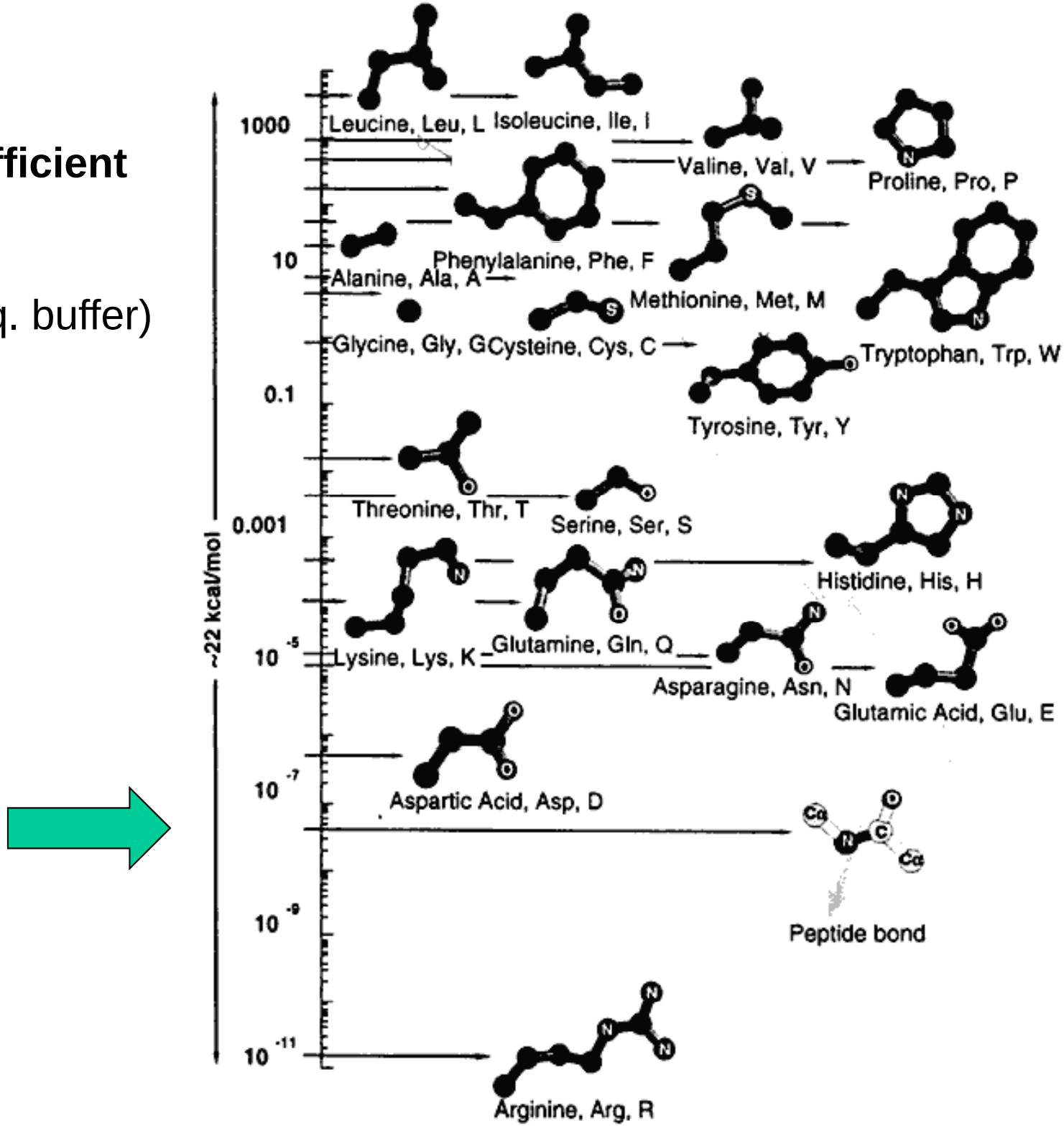
Distance between two amino acid residues: 0.381 nm




II
CB
CA
N
N
C
O
O

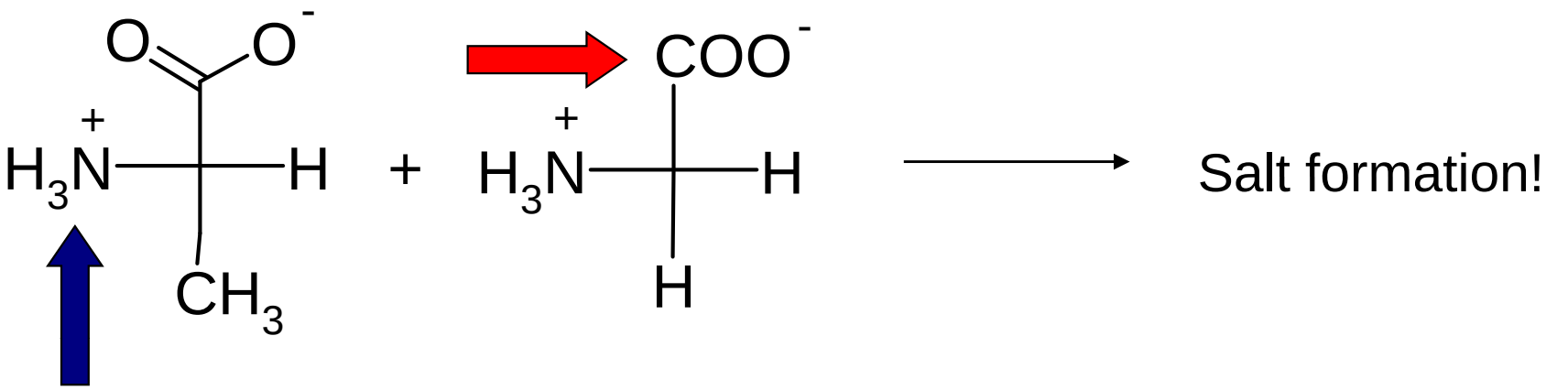
Distribution coefficient of amino acids

(Cyclohexane / aq. buffer)

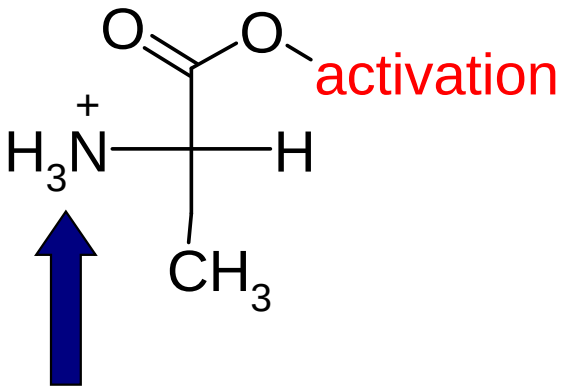


Principles of peptide synthesis

1. Reaction of the α-amino group with an activated carboxylic group



2. Protection needed for amino and carboxylic groups



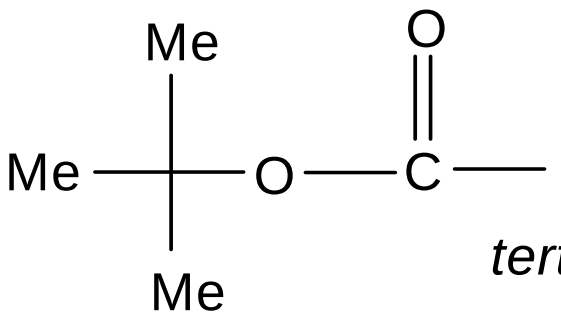
4 potential products from 2 amino acids A,B

AA
AB, BA
BB

3. Protecting groups for side chain residues (-SH, -OH, -COOH, -NH_2)

Protecting groups

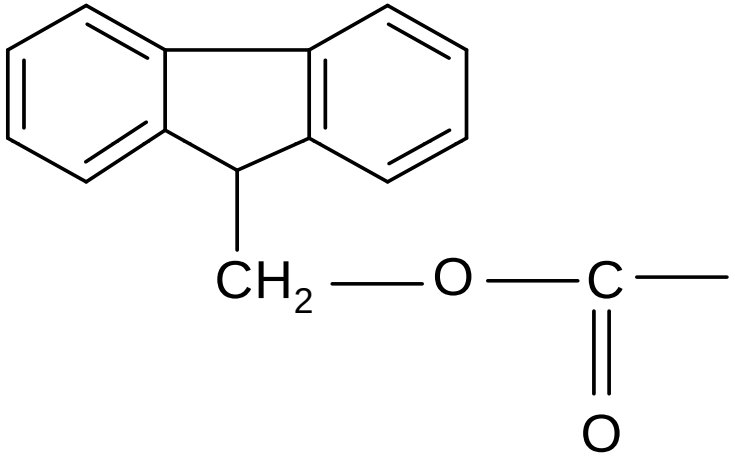
Amino functions:



tert-Butoxycarbonyl (Boc)

Cleavage (quantitative, without racemate formation)

H^+ (TFA)



9-Fluorenylmethoxycarbonyl (Fmoc)

Weak bases (piperidine, morpholine)

Carboxyl groups:

Benzyl ester

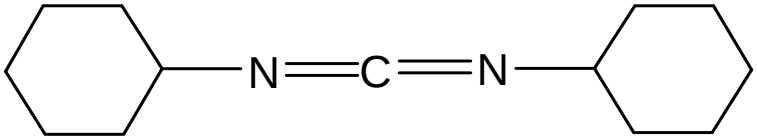
Hydrogenation

Boc-ester

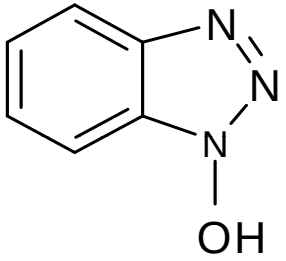
H^+

Activation

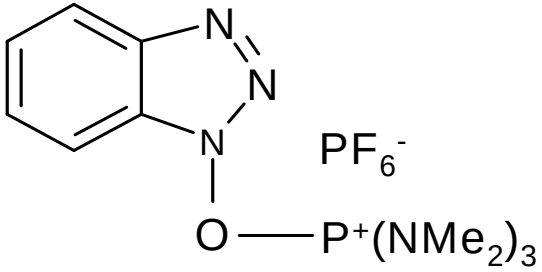
Coupling: via water elimination or by using active esters



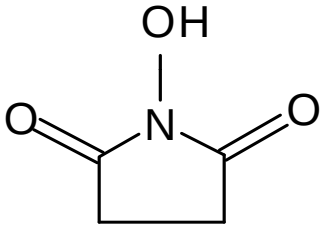
Dicyclohexylcarbodiimid (DCC)



1H-Hydroxybenzotriazol (HOBt)



Benzotriazol-1-yl-oxy-tris-(dimethylamino)phosphonium hexafluorophosphate (BOP)



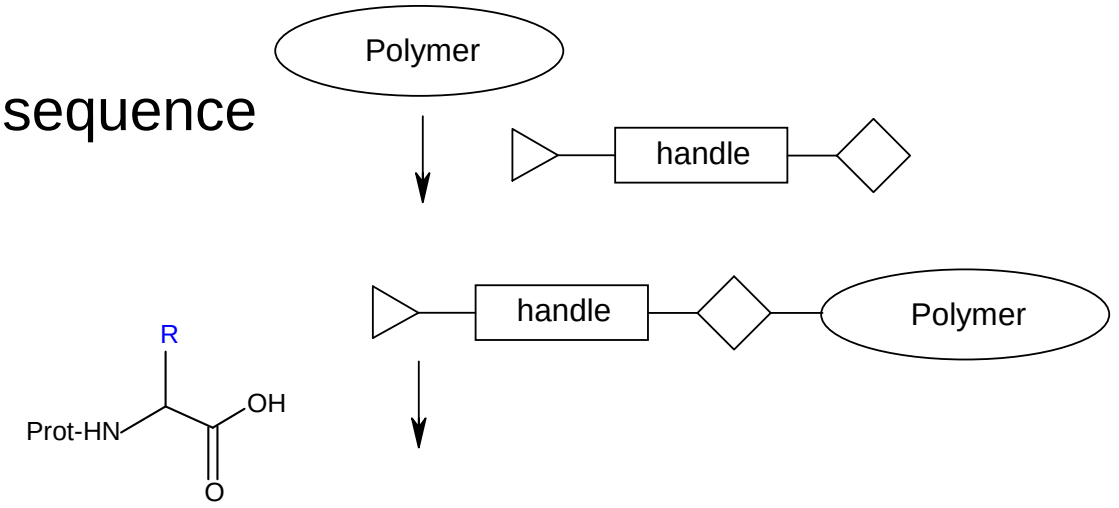
N-Hydroxysuccinimid

Solid phase synthesis

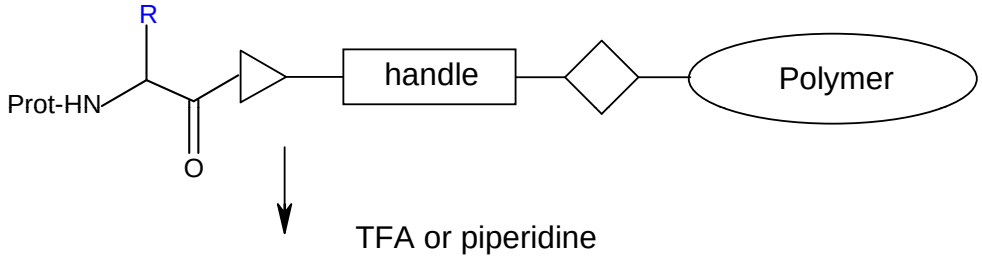
(Merrifield)

Synthesis sequence

1. Binding of a bifunctional linker to the polymer

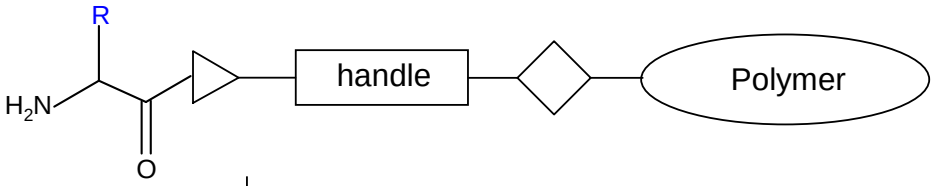


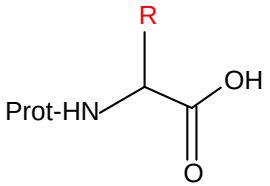
2. Binding of first amino acid to linker



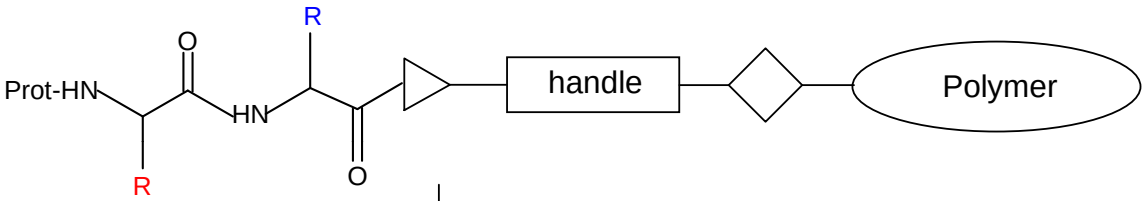
TFA or piperidine

3. Deprotection of amino group





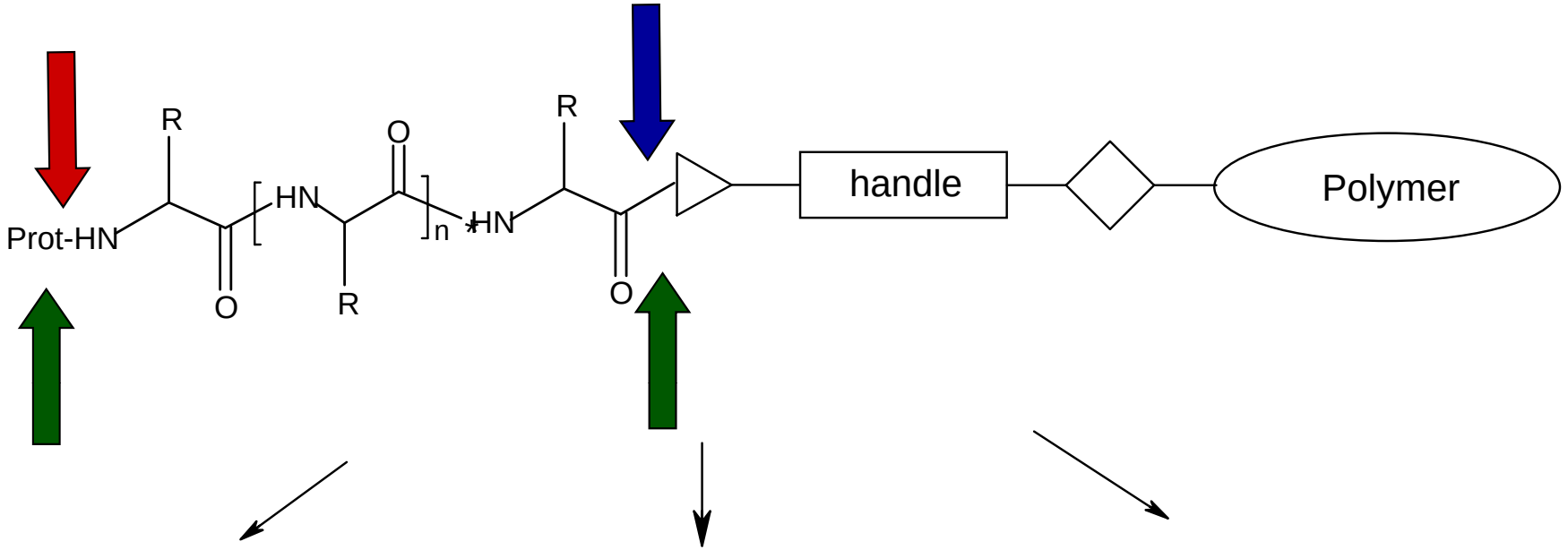
4. Coupling of second amino acid



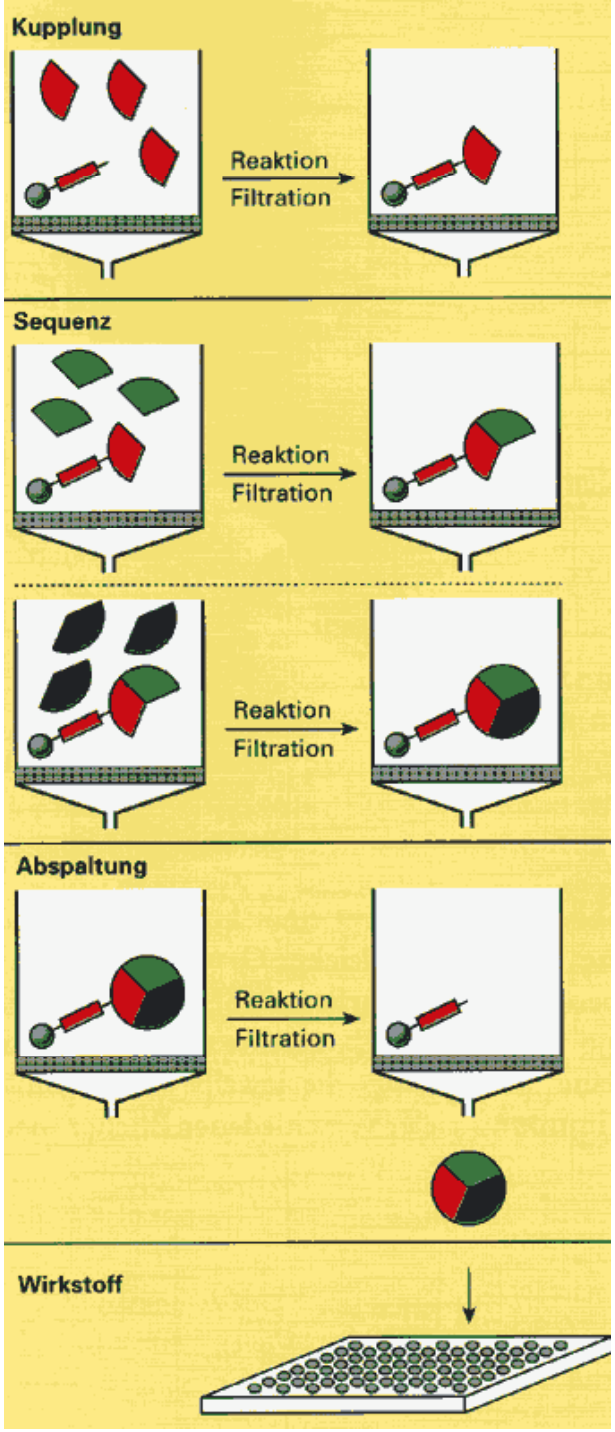
5. Repetition of cycles

Synthesis sequence

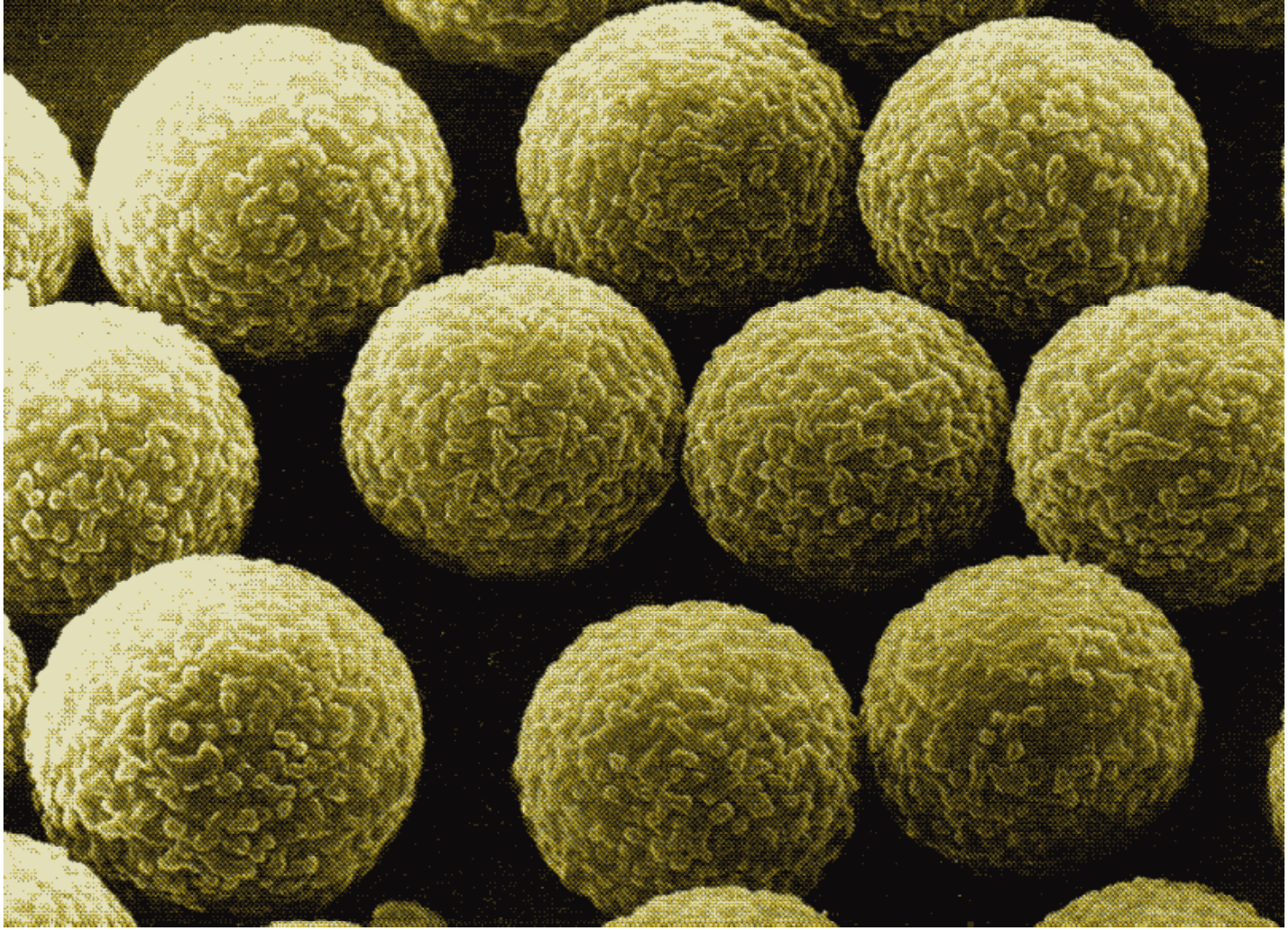
6. Removal of protecting groups and cleavage from linker



protected peptide **deblocked peptide-resin** **deprotected peptide**



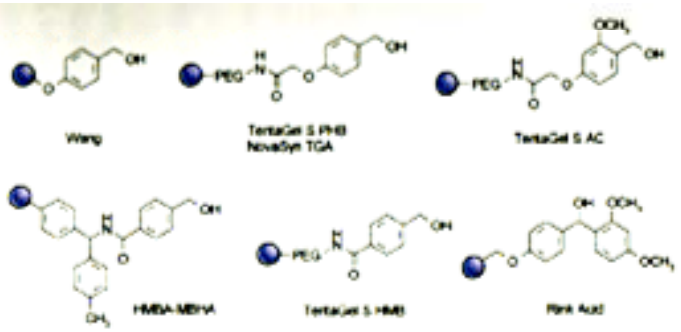
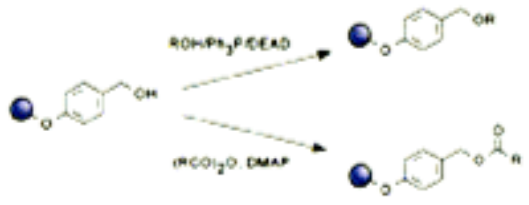
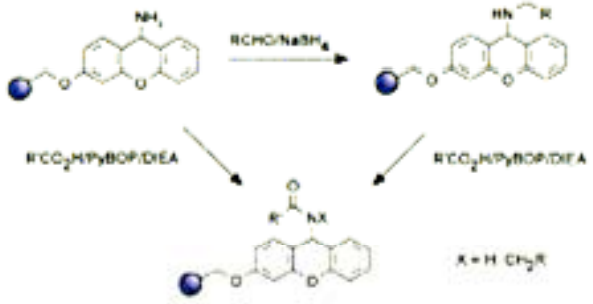
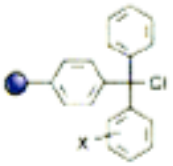
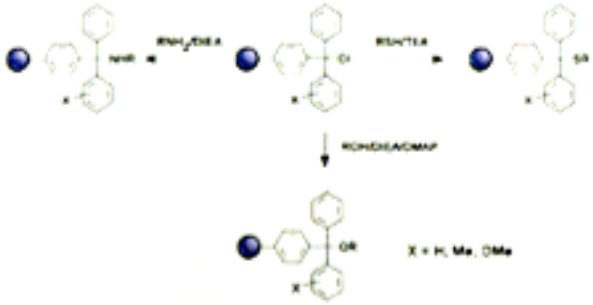
Scheme for solid phase synthesis

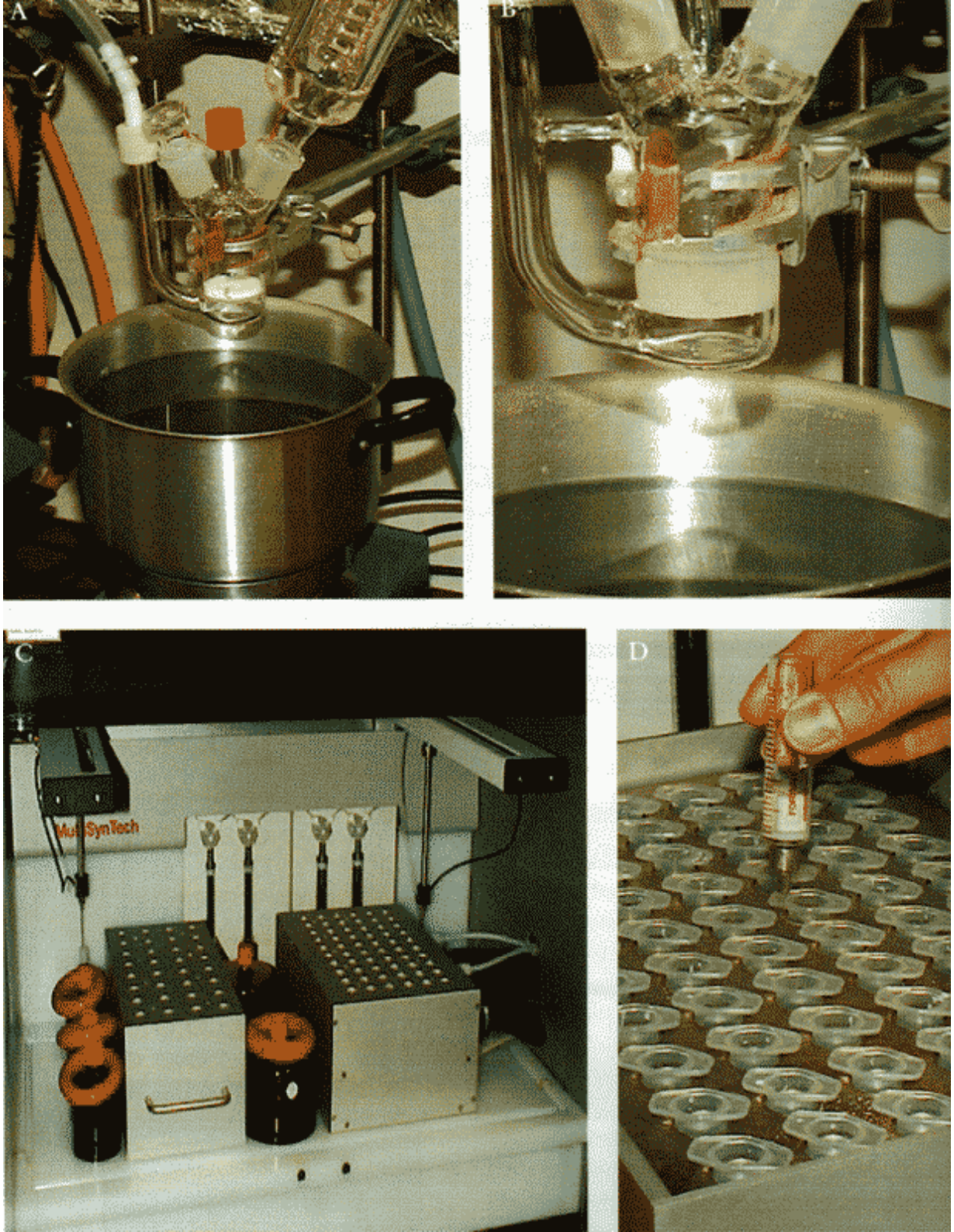


TentaGel beads (crosslinked polystyrene, 130 μm grafted with polyethyleneglycol)

Capacity: 0.2-0.8 mmol / g
15 mg product (M: 500) ~ 40-150 mg resin

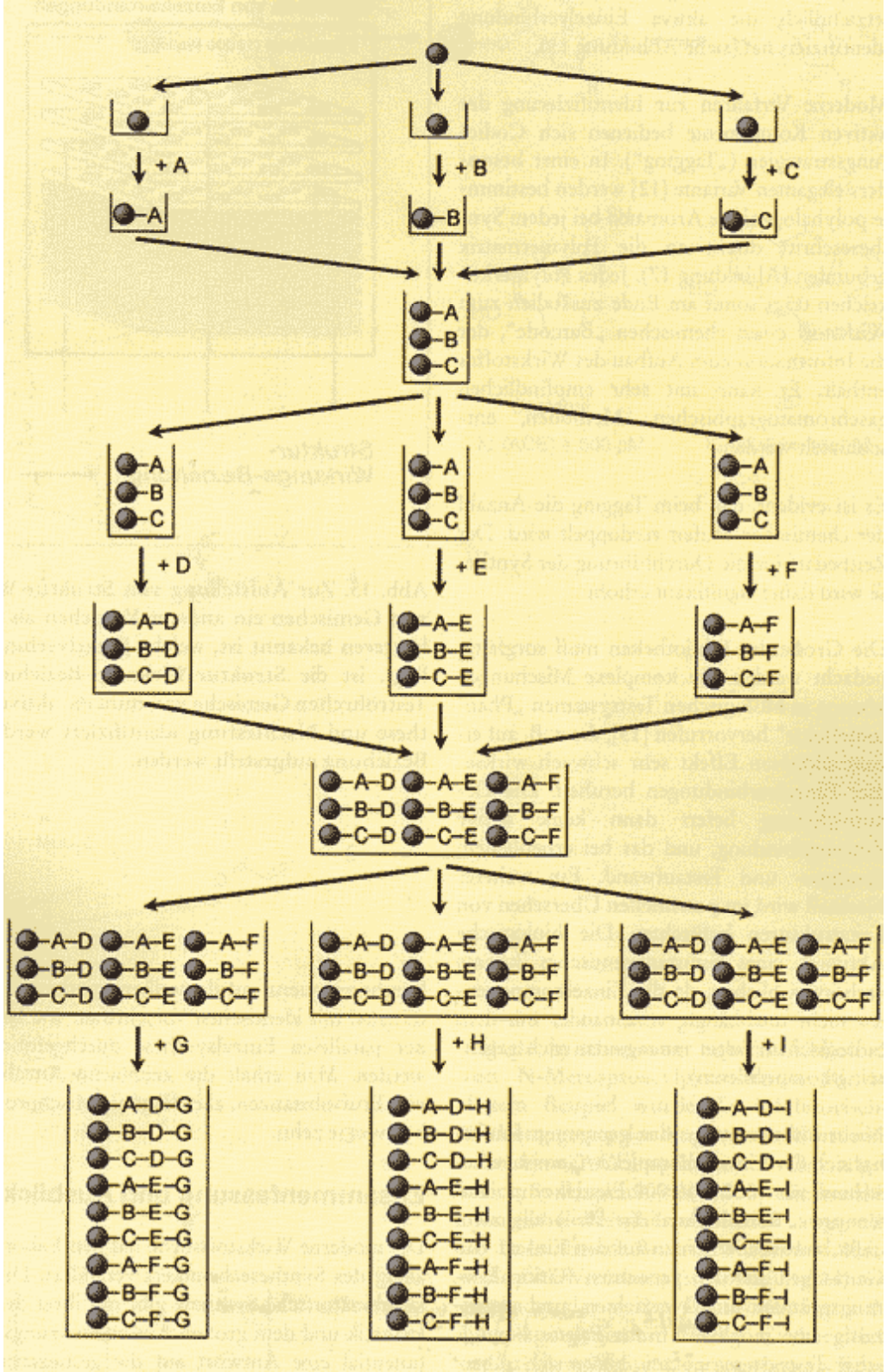
Chiuz 1996 (30) 270-285.

Linker type	Attachment	Cleavage		
		Resin	Reagent	Prod.
Hydroxy-Resin			Wang, TGA, TentaGel	TFA TFA	RCOOH ROH
			Rink Acid	10% HOAc/DCM 5% TFA/DCM	RCOOH ROH
			HMBA-MBHA	NaOH aq. NH_3/MeOH $NaBH_4$/EtOH MeOH/TFA NH_2NH_2/DMF	RCOOH $RCONH_2$ RCH_2OH RCOOMe $RCONHNH_2$
Amino-Resin			Rink Amide, Rink Amide MBHA, TGR, TentaGel	TFA	RCONHX
			Sieber	1% TFA/DCM	RCONHX
Trityl-Resin				TFA/DCM	RCOOH ROH RSH RNH



Combinatorial synthesis

(Compound libraries)



Combinatorial synthesis

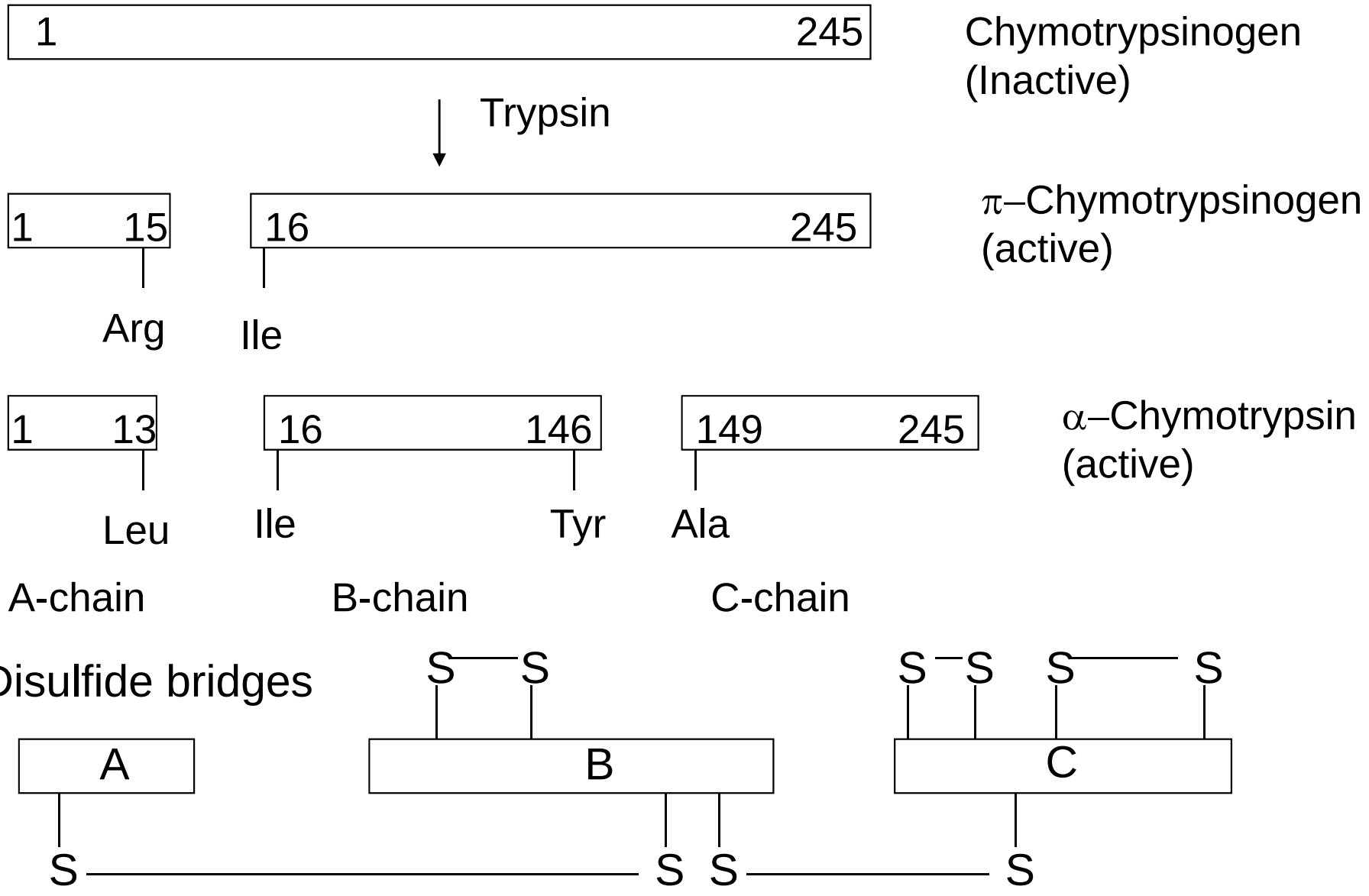
(Compound libraries)

„Mix and Split“

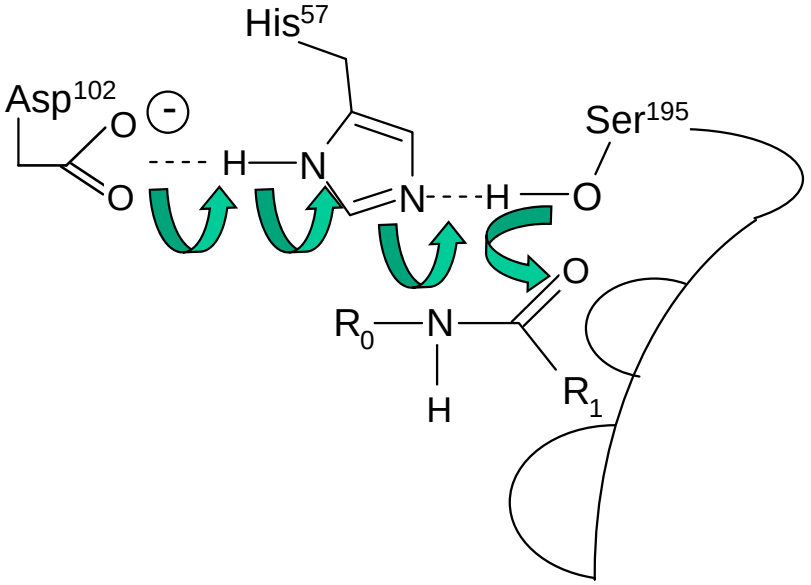
9 Reactions yield 27 compounds
(3 groups of 9 components)

Serin Proteases

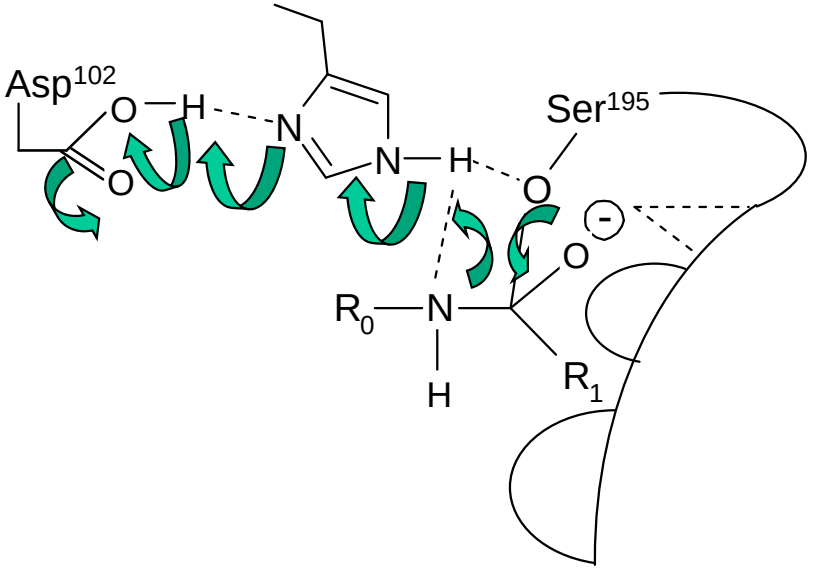
Examples:
Chymotrypsin, Trypsin, Elastase, Subtilisin



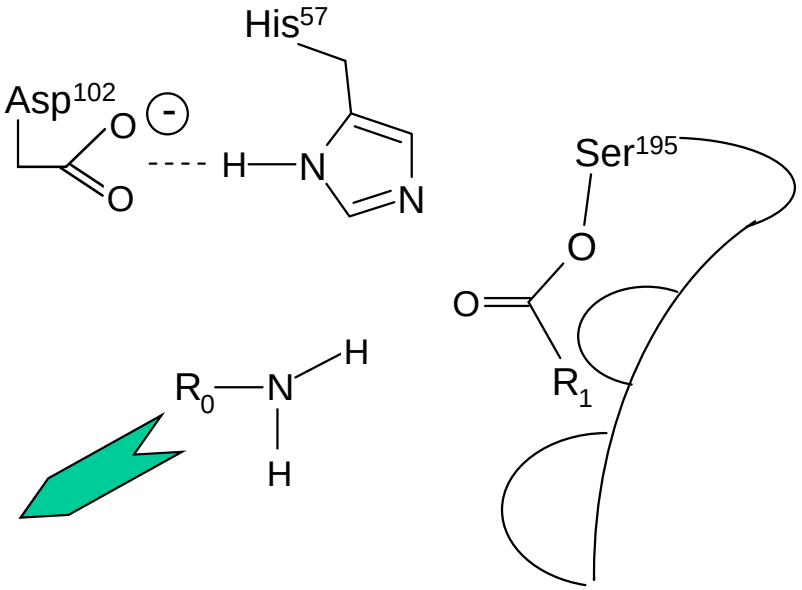
Mechanism of Serin Protease-hydrolysis of peptides



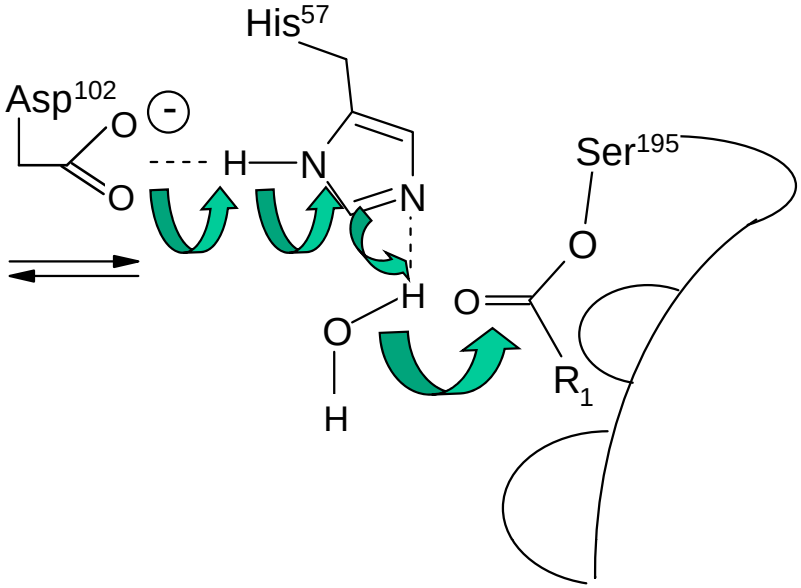
1. Enzyme-substrate complex



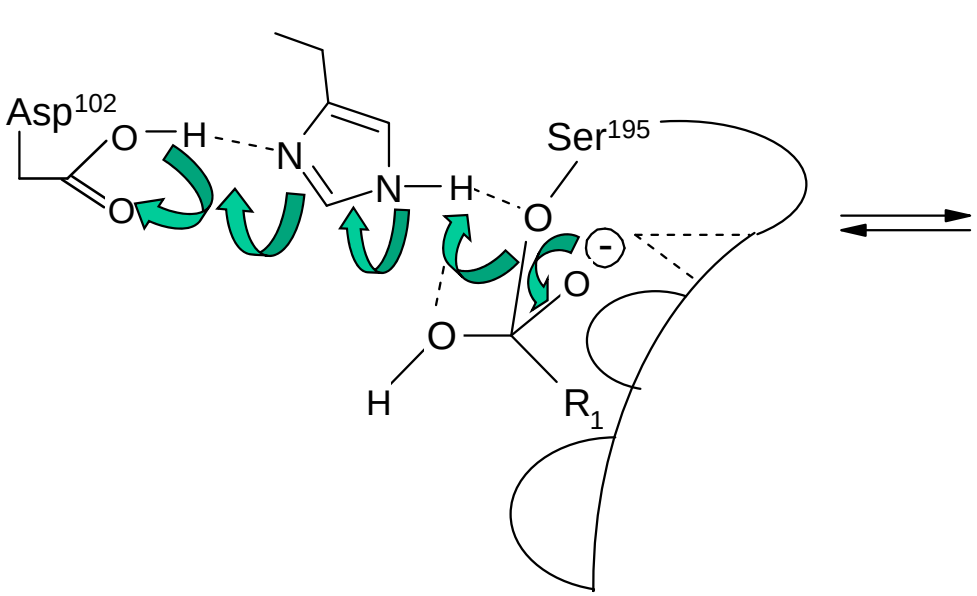
2. Tetrahedral intermediate

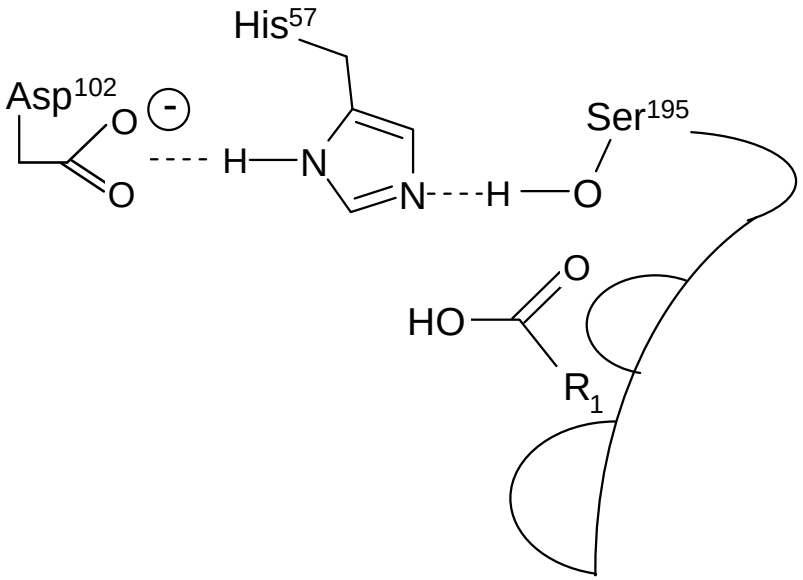


3. Acyl enzyme-intermediate



4. Hydrolysis of acyl enzyme





5. Tetrahedral intermediate

6. Enzyme-product complex

Mechanism
catalytic triade, proton shuttle