

A 35-year romance with
a seafaring microbe *p. 1006*

Social ties and policy reforms in
China's S&T system *pp. 1019 & 1022*

Designing zeolites
to react *pp. 1028 & 1051*

Science

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AAAS

SYNTHETIC CHROMOSOMES

Remodeling the yeast genome
piece by piece *p. 1038*



LECTURE 4 BUILDING DNA

LECTURE

DNA nomenclature
single strand
double strand
bases
base pairs
linear
circular
blunt end
sticky end

DNA products
oligos
fragments
parts
plasmids/integrations

Denovo synthesis
solid phase chemistry
<300bases

PCR
DNA replication
components (buffer, dNTPs, polymerase, template)
protocol
first 3 cycles
thermostable polymerases
DNA modification
Overhangs
primer design
validation
agarose gel electrophoresis
sanger sequencing (GCTA, KBRG)

DNA assembly
Restriction (EcoRI, GAATTC)
components (buffer, RE, DNA)
Ligation
components (buffer, ligase, DNA)
Restriction, ligation
Time frame

Golden gate assembly
Type II's restriction enzyme (BsaI, GGTCTCN1)
Restriction ligation
Cyclical protocol

EXERCISE

Design primers to amplify GFP orf
Execute virtual PCR
Design primers to amplify GFP orf with BsaI OH
Design primers to amplify vector with OH
Assemble a gene with 3XGFP

LECTURE 3

POLYPEPTIDE SYNTAX

Amino acid structures
C1, Calpha carbons
hydrophobic - AGILPV
aromatic - FWY
neg charge - DE
pos charge - RHK
hydroxylic - ST
sulfur containing - CM
polar - NQ

protein structure
peptide bond
dihedral angles
phi (N-Ca)
psi (Ca-C1)
omega (C1-N) = 180deg unless proline (0deg)
protein folding
bond forces
stretching, bending, rotating
short range forces
Pauli repulsion, van der Waals' interactions
Electrostatic forces
Coulomb's law
interaction with solvent
Motif's
alpha helix
3.6 residues per turn
each residue 100deg, 1.5A
beta sheet
planar
isoelectric point
molecular weight

protein domains
binding domain - leucine zipper, TALEN
PTM domain
phosphorylation
methylation
ubiquitination
enzymatic domains
protease
kinase
targeting domains
NLS
signal peptide
anchoring sequence

EXERCISE
make a DNA file of GFP
create a translation
annotate the protein secondary structure
compute the molecular weight of GFP
compute the isoelectric point
visualize GFP in chimera

