DNA & DNA : Protein Interactions

BIBC 100
Sequence = Information

• Alphabet = language
  – L, I, F, E    LIFE

• DNA = DNA code
  – A, T, C, G    CAC=Histidine
                  CAG=Glutamine
                  GGG=Glycine

• Protein = Protein code
  – 20 a.a.      LIVE ≠ EVIL
                  L, I, F, E    N → C  N → C
DNA basics

- A DNA chain has polarity
- Just as for proteins we have \( \text{N} \rightarrow \text{C} \),
  
  Amino-end \quad Carboxy-end

  for DNA the base sequence is \( 5' \rightarrow 3' \)

  5' OH unlinked \quad 3' OH unlinked

- Both proteins and DNA are polymers with sequence specificity and directionality
  - Protein: Polypeptide
  - DNA: Polynucleotide
What is a nucleotide?

• Nucleotide:
  – Base  +  Sugar  +  Phosphate
  A,G,C,T  ribose, deoxyribose  3 phosphates

Nucleoside:
  – Base  +  Sugar
Four kinds of bases joined to a sugar-phosphate backbone

- **A** = Adenosine (purine) \( \rightarrow \) 2H bonds
- **T** = Thymine (pyrimidine) \( \rightarrow \) Length of H bond < 3Å
- **G** = Guanine (purine) \( \rightarrow \) 3H bonds
- **C** = Cytosine (pyrimidine)
Double stranded DNA

- Packed – right handed helical staircase (double helix)
- Rails: antiparallel sugar-phosphate chains
- Steps: Purine-Pyrimididine base pairs H-bonded
- Polarity: sequence 5’→3’ direction
  - ACG ≠ GCA
Types of DNA

• **B-DNA**
  – Most common
  – Right handed helices
  – 10 bp/turn
  – 1 full turn = 34 Å
  – Width of dsDNA 20 Å
  – Helical twist of \( \sim 36^\circ \)

• **A-DNA**
  – Right handed helices
  – Dehydrated dsDNA

• **Z-DNA**
  – Left handed helices
  – Alternating G-C content
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dsDNA has Major and Minor Groove

- B-DNA has distorted helices
- Bases within the grooves exposed to solution
- Protein binds DNA within these grooves
  - binding of α helix of protein to DNA can bend DNA structure
  - Major groove binding most common
How Proteins recognize Genes

- Most genes are silent, unless specifically turned on

- **DNA → RNA → Protein**
  - Transcription
  - Translation

- **Switch Protein: Transcription Factor**
  - Activator
  - Repressor
  - Can be regulated by small molecule
Gene Activators and Repressors

• Main Controls of
  – Growth
  – Differentiation
  – Oncogenesis

• Recognition
  – Affinity
  – Specificity
(a) **Negative regulation**
Molecular signal causes dissociation of repressor from DNA, inducing transcription.

Repessor → DNA → Promoter → OFF

(b) **Negative regulation**
Molecular signal causes binding of repressor to DNA, inhibiting transcription.

Operator → ON → mRNAr

(c) **Positive regulation**
Molecular signal causes dissociation of activator from DNA, inhibiting transcription.

Activator → RNA polymerase → ON → mRNA

(d) **Positive regulation**
Molecular signal causes binding of activator to DNA, inducing transcription.

Activator binding site → OFF → mRNA

**Figure 28-4**
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Negative regulation
Molecular signal causes dissociation of repressor from DNA, inducing transcription.

Figure 28-4a
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Negative regulation
Molecular signal causes binding of repressor to DNA, inhibiting transcription.
Positive regulation
Molecular signal causes dissociation of activator from DNA, inhibiting transcription.

Figure 28-4c
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Positive regulation
Molecular signal causes binding of activator to DNA, inducing transcription.
# Activators and Repressors

<table>
<thead>
<tr>
<th>LOCK</th>
<th>KEY</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nucleotide Base sequence</td>
<td>Protein Motif</td>
</tr>
<tr>
<td><strong>DNA</strong></td>
<td></td>
</tr>
<tr>
<td>Edges at major and minor groove</td>
<td></td>
</tr>
<tr>
<td><strong>• Helx-Turn-Helix</strong></td>
<td></td>
</tr>
<tr>
<td>• Prokaryotes acts as dimer :HTH</td>
<td></td>
</tr>
<tr>
<td>• Eukaryotes acts as monomer:Homeodomain</td>
<td></td>
</tr>
<tr>
<td>• Metal Free</td>
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<tr>
<td><strong>• Zinc-Stabilized</strong></td>
<td></td>
</tr>
<tr>
<td>• Helix-turn- β-sheet</td>
<td></td>
</tr>
<tr>
<td>• Leucine-Zipper</td>
<td></td>
</tr>
</tbody>
</table>
DNA: Where?

- Only the edges of the nucleotides are accessible to the solvent or to protein, primarily in the major groove of the DNA double helix

Protein: How?

- Protruding groups from protein surface to contact nucleotides at base of groove
- Motifs
Helix turn Helix Motif

- 2 α helices connected by loop
- 2 motifs for functional binding
- 1 α helix from each motif interacts with DNA ~ 10 bp
  - Sequential interaction with back bone and base pairs dependent upon specificity
  - Interaction of Dimer bends the DNA
For Bacterial HTH

- DNA is distorted
- Protein is dimeric
- H-bonds between sugar-phosphate backbone and protein
  - anchor DNA to protein
- Sequence-specific recognition between DNA bases and \(\alpha\)-helix
- Recognition helices in dimer are separated 34\(\AA\) apart, i.e. one turn of B-DNA
- If one helix binds to major groove, the second binds (34\(\AA\)) to major groove one turn away
- DNA bends and can interact with other regions of protein
Allosteric Effects on Binding

Small Molecule that acts far from protein binding site with DNA causing a conformational change in the Recognition Helix.
Cap Binding Protein

- Cap: catabolite activating protein
- Activated by cAMP binding
- Dimer, binds major groove
- Both CBP & DNA are distorted upon binding
Zn Finger Motif

- 2 β strands + 1 α helix coordinated by Zinc ion
- 2 Cys + 2 His coordinate metal
- Highly conserved sequence esp. around coordination sites
- Functional interactions
  - 3 fingers wrapping around DNA along major groove
  - Dimer aligning motifs 34Å apart and interacting at major groove
Zn Finger interactions with DNA

1. Recognition helix positioning:
   DNA backbone to side chain of the loop
2. Base pair specific interactions with the Recognition helix side chains
Leucine Zipper

- 2 amphipathic $\alpha$ helices form coiled coil to stabilize dimer
- Distal, basic region on each helix interacts with major groove of DNA
Heptad repeats in Leucine Zipper

• 7 amino acid repeat stabilizes dimer interactions within the coiled coil
Figure 28-14

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