

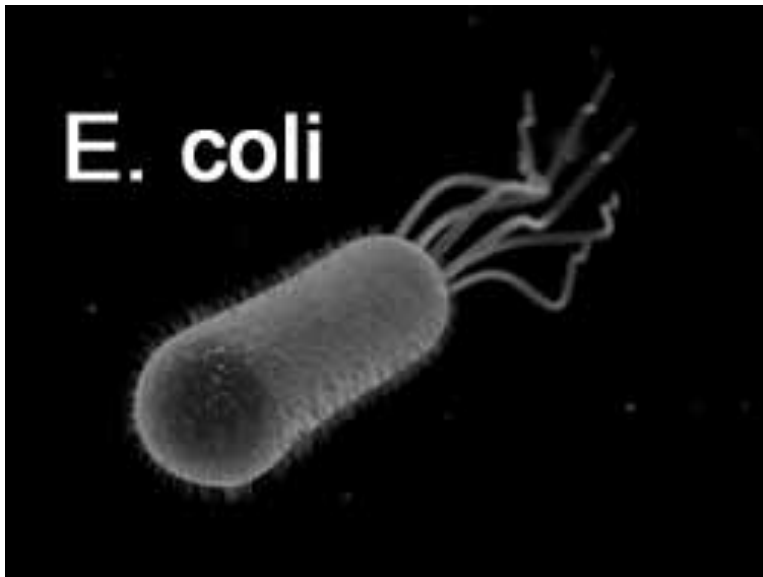
## SUMMER INSTITUTE - ONLINE MODALITY CALENDAR 2022

SUN	MON	TUE	WED	THU	FRI	SAT
June 26	27	28	29	30	31	July 02
	9:00-10:00am Virtual Program Orientation for Summer Institute Online Modality					
July 03	04	05	06	07	08	09
	Holiday (Independence Day)	8:30-10:00am -Welcome Reception and Buddy Meet & Greet Event	Free Day	Classes begin! 8:30-11am: BIOL4905 INTRODUCTION 8-10:20pm: Afternoon course	8:30-11am: BIOL4905 DNA PREPARATION 8-10:20pm: Afternoon course	
10	11	12	13	14	15	16
	8:30-11am:BIOL4905 PROTEOMICS I 8-10:20pm: Afternoon course	8:30-11am:BIOL4905 PROTEOMICS II 8-10:20pm: Afternoon course	8:30-11am:BIOL4905 PROTEOMICS III 8-10:20pm: Afternoon course	8:30-11am: BIOL4905 RNA PREPARATION 8-10:20pm: Afternoon course	Virtual Independence Day Activity	
17	18	19	20	21	22	23
	8:30-11am:BIOL4905 qPCR / ROBOTS 8-10:20pm: Afternoon course	8:30-11am:BIOL4905 DNA Sequence Analysis 8-10:20pm: Afternoon course	Midterm Break	8:30-11am:BIOL4905 Next Gen. Sequencing 8-10:20pm: Afternoon course	8:30-11am:BIOL4905 Automated Microscopy /AFM	
24	25	26	27	28	29	30
	8:30-11am:BIOL4905 Microarray I 8-10:20pm: Afternoon course	8:30-11am:BIOL4905 Microarray II 8-10:20pm: Afternoon course	8:30-11am:BIOL4905 Nanostring 8-10:20pm: Afternoon course	8:30-11am:BIOL4905 Flow Cytometry 8-10:20pm: Afternoon course	FINALS	
31	August 01	02	03			
	9:00-10:00am: Closing Reception		Grades available in PAWS			

Legend:

Orange: Courses

Blue: Activities



Bacteria thrive on many different types of food. But most yeasts can live only on sugars and starches. From these, they produce carbon dioxide gas and alcohol. Thus, they have been useful to man for centuries in the production of certain foods and beverages. They are responsible for the rising of bread dough and the fermentation of wines, whiskey, brandy and beer. They also play the initial role in the production of vinegar.

~ 1.5 - 3  $\mu\text{m}$

**Prokaryote**

**DNA is Circular**

**Cell membrane**



Yeasts are small, *single-celled plants*. They are members of the family *fungi* (singular, *fungus*), which also includes mushrooms. Fungi differ from other plants in that they have no chlorophyll.

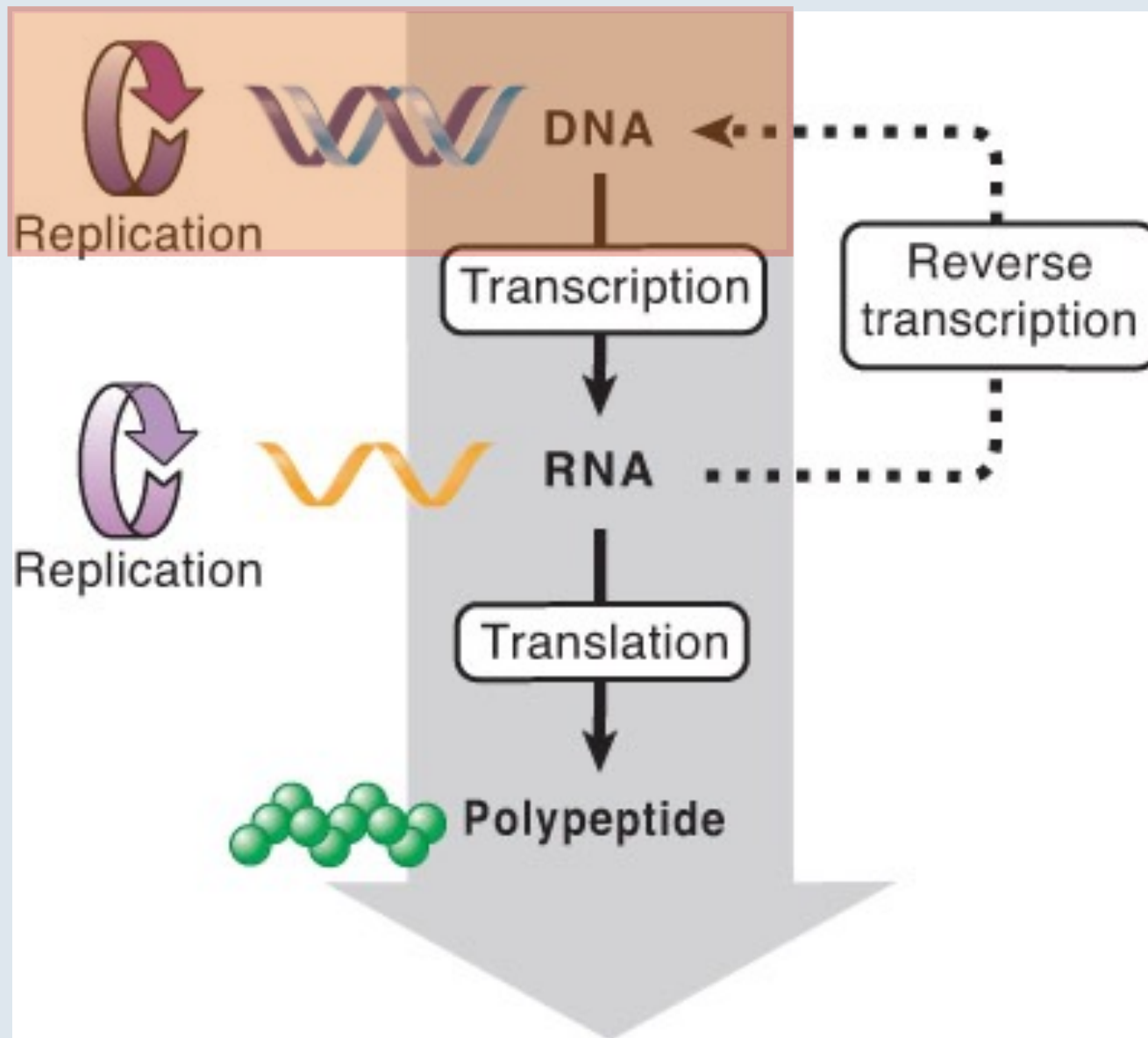
~ 50  $\mu\text{m}$

**Eukaryote**

**DNA is linear**

**Cell wall**

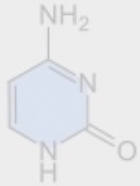
# DNA Preparation



Information in nucleic acid can be perpetuated or transferred, but the transfer of information into a polypeptide is irreversible.

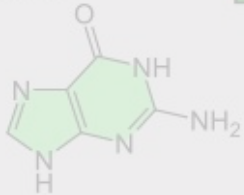


Cytosine



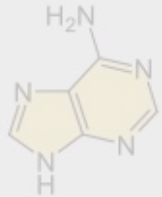
**C**

Guanine



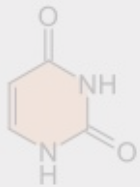
**G**

Adenine



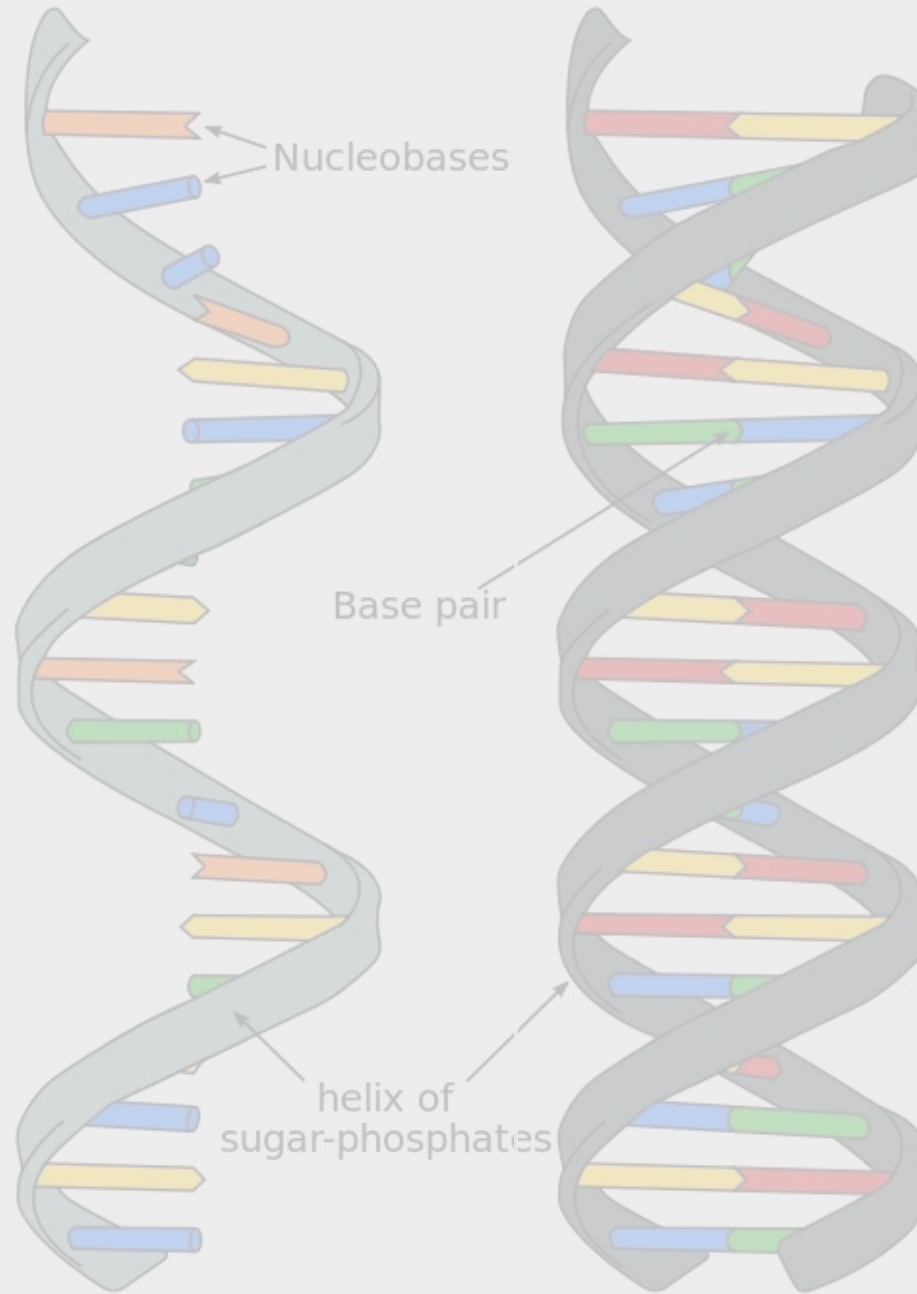
**A**

Uracil

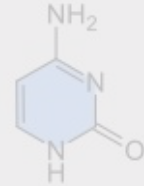


**U**

Nucleobases  
of RNA

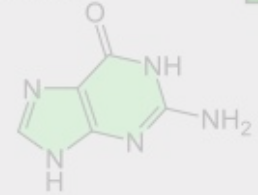


Cytosine



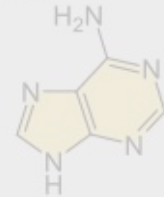
**C**

Guanine



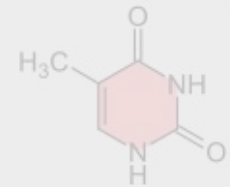
**G**

Adenine



**A**

Thymine



**T**

Nucleobases  
of DNA

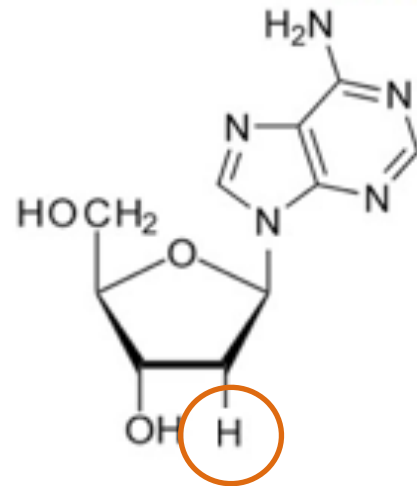
**RNA**

Ribonucleic acid

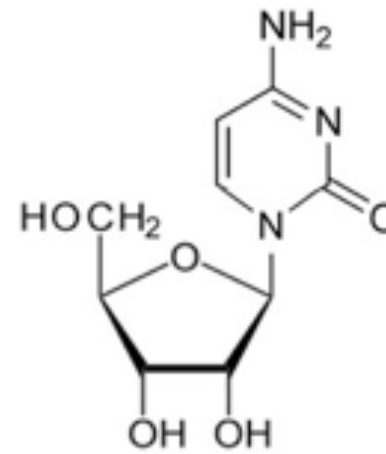
**DNA**

Deoxyribonucleic acid

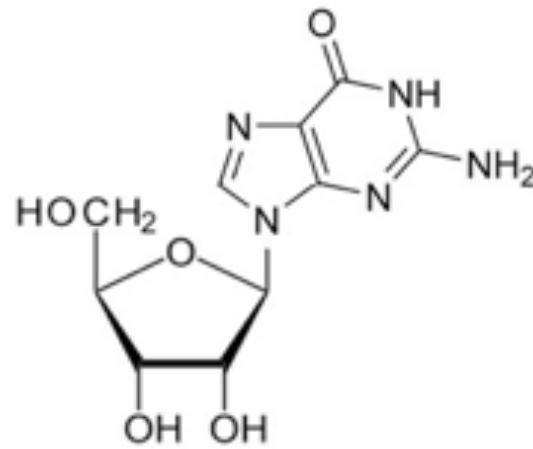
**deoxyAdenosine**



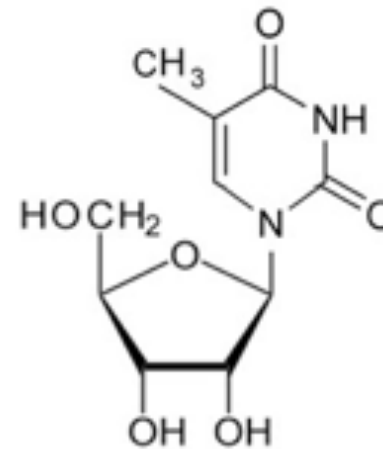
**Cytidine**

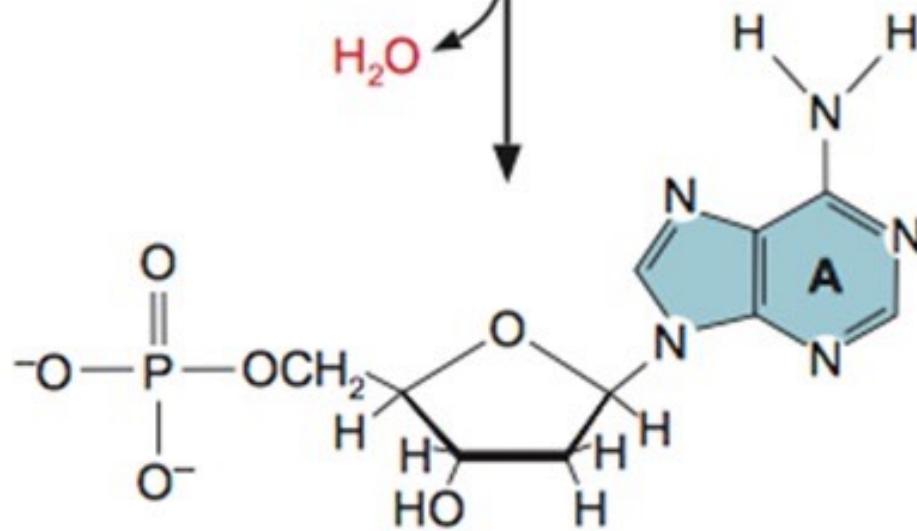
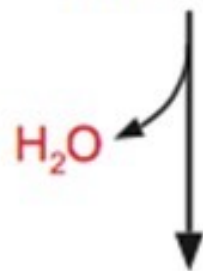
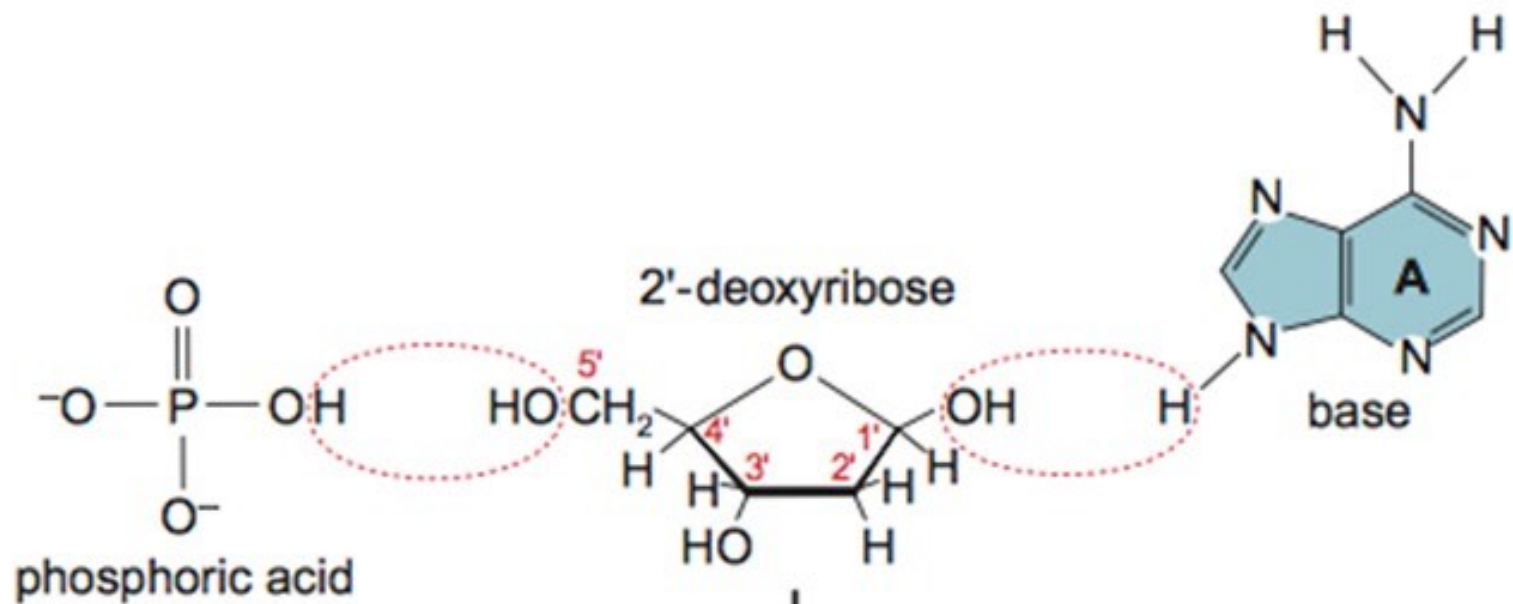


**Guanosine**



**Thymidine**





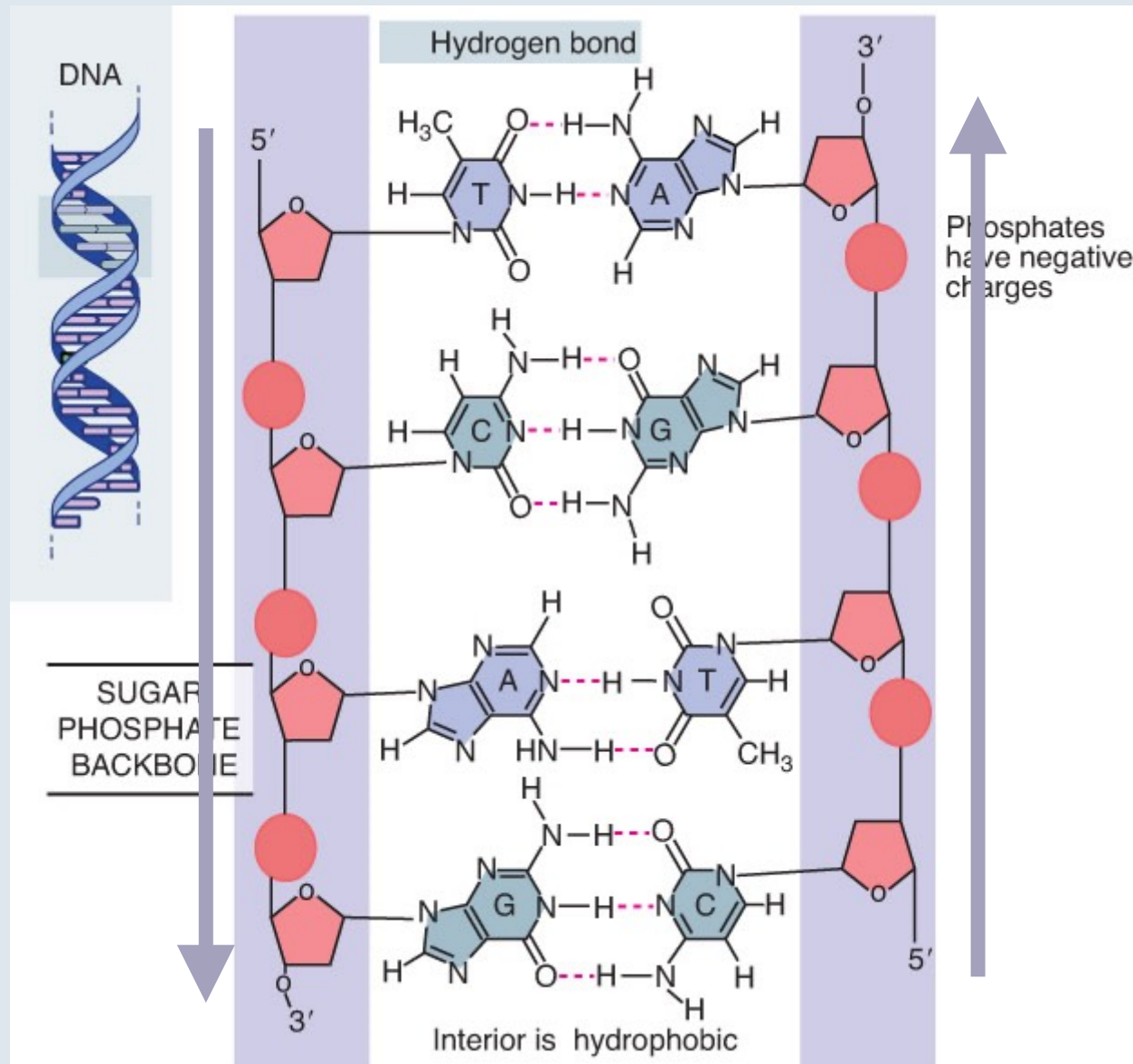
nucleotide (dAMP)



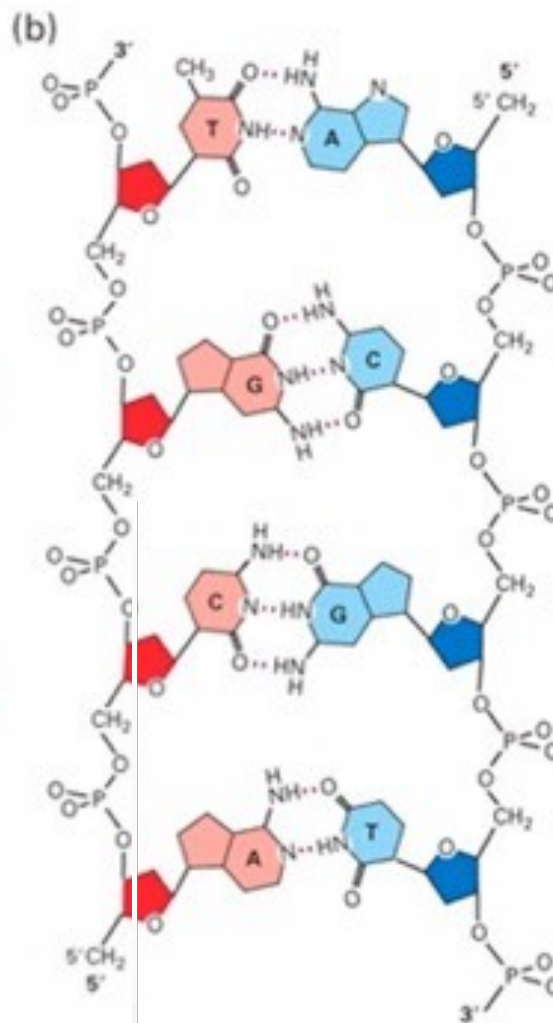
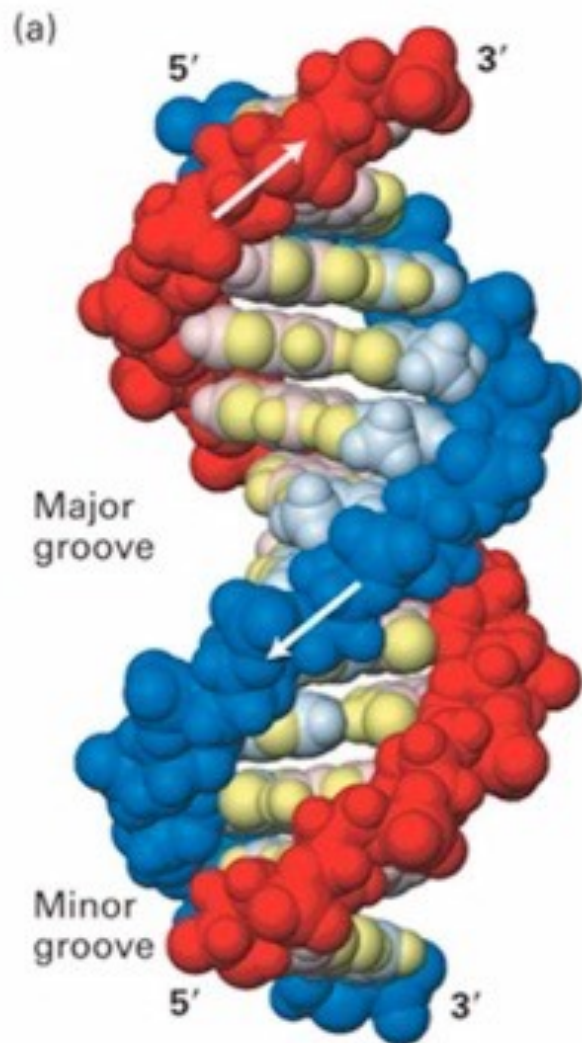








The double helix maintains a constant width because purines always face pyrimidines in the complementary A-T and G-C base pairs.



## B form DNA

2.0 nM dia (20 Å)

0.36 nM (3.6 Å)

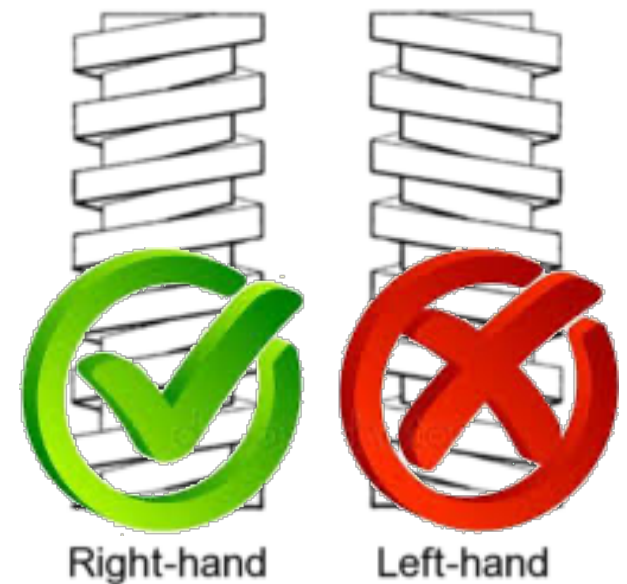
between bases

~10 bases per turn

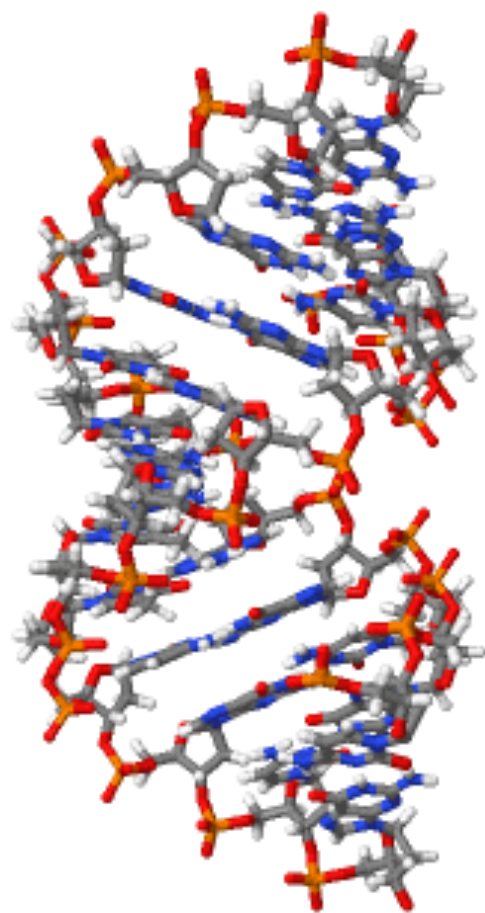
antiparallel strands

bases perpendicular

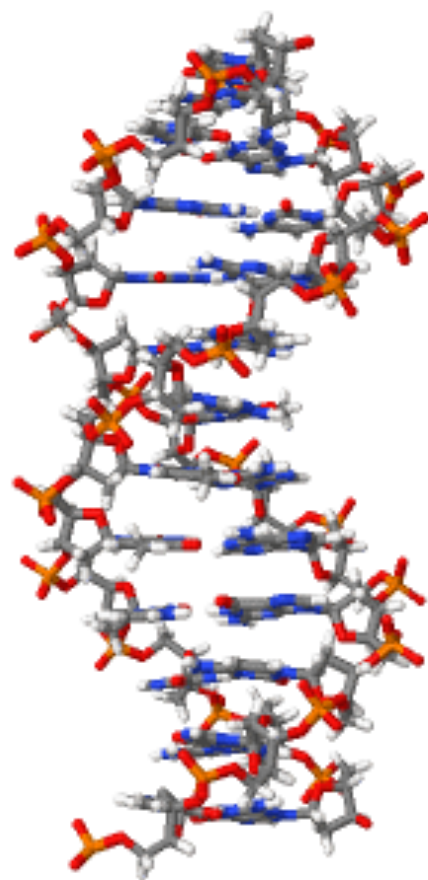
to axis



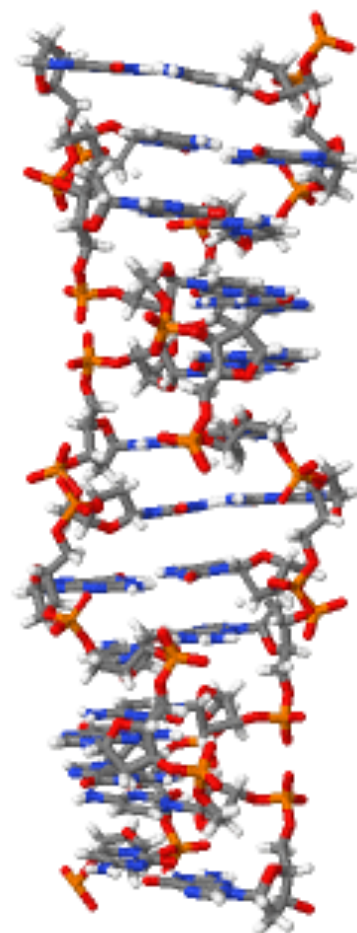




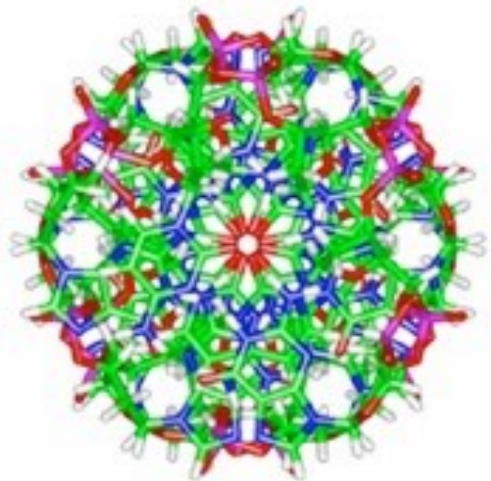
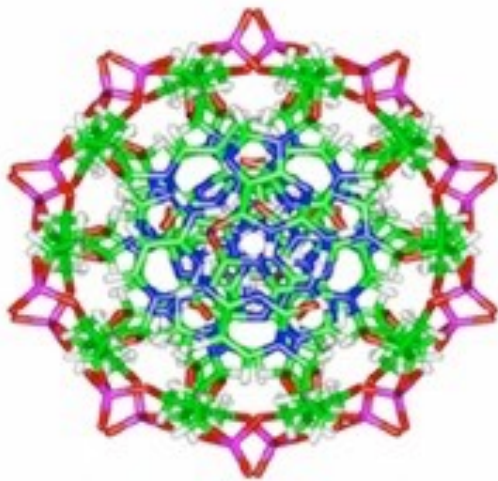
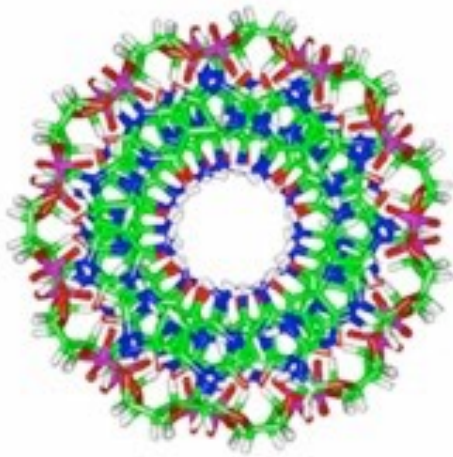
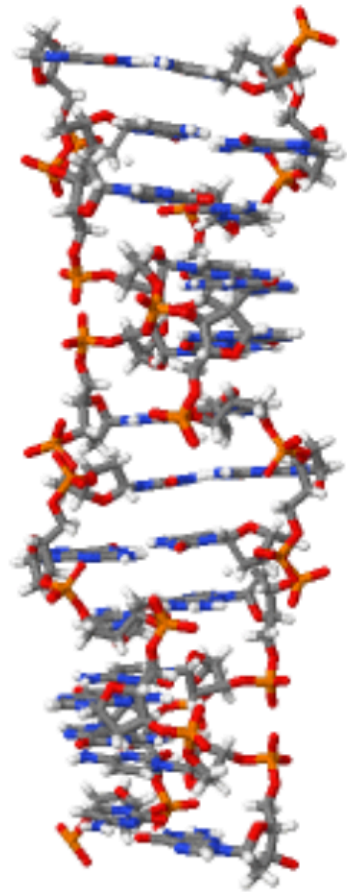
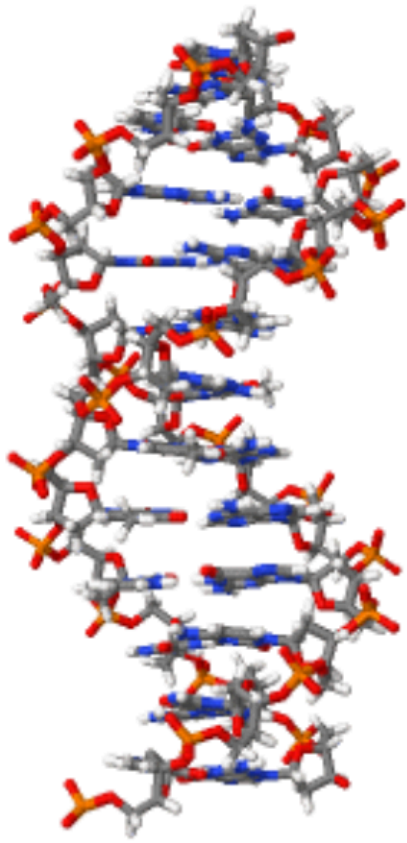
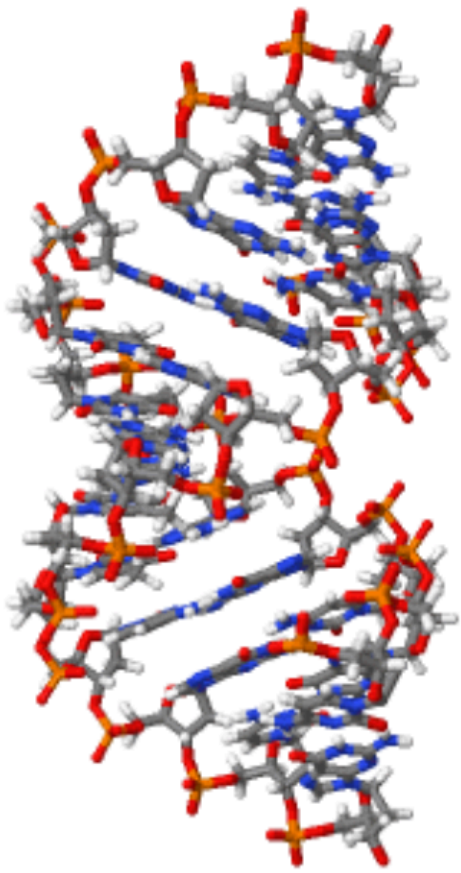
**A DNA**



**B DNA**



**Z DNA**



Review

> [Front Biosci.](#) 2007 May 1;12:4424-38. doi: 10.2741/2399.

## Z-DNA, an active element in the genome

Guliang Wang<sup>1</sup>, Karen M Vasquez

Affiliations + expand

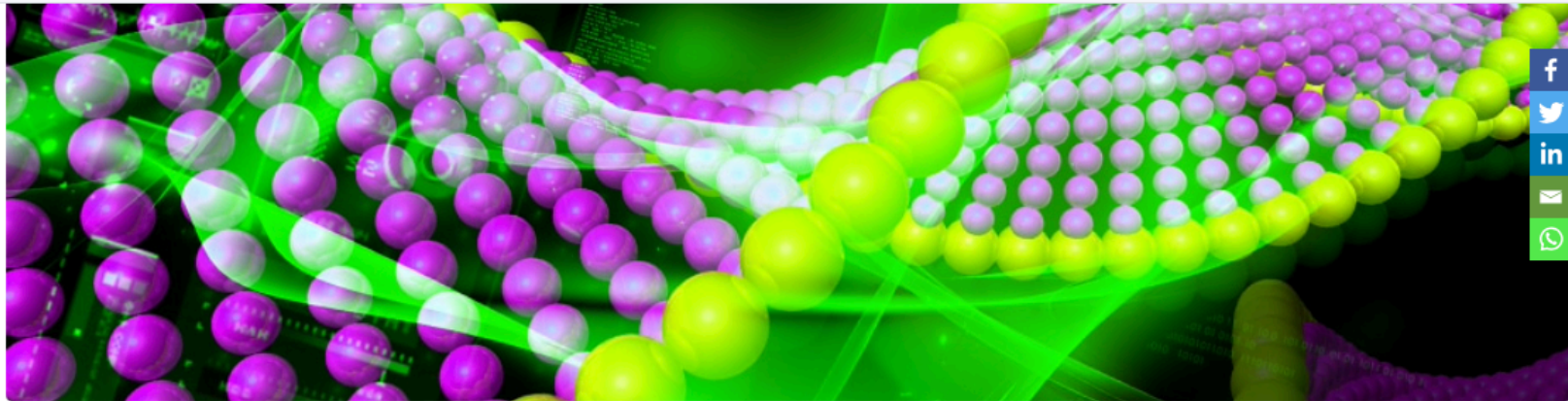
PMID: 17485386 DOI: [10.2741/2399](#)

**Free article**

### Abstract

Z-DNA is a left-handed helical form of DNA in which the double helix winds to the left in a zigzag pattern. DNA containing alternating purine and pyrimidine repeat tracts have the potential to adopt this non-B structure in vivo under physiological conditions, particularly in actively transcribed regions of the genome. Z-DNA is thought to play a role in the regulation of gene expression; Z-DNA is also thought to be involved in DNA processing events and/or genetic instability. For example, Z-DNA-forming sequences have the potential to enhance the frequencies of recombination, deletion, and translocation events in cellular systems. Although the biological function(s) of Z-DNA and related Z-DNA-binding proteins are not fully understood, accumulating experimental and clinical evidence support the idea that this non-B DNA conformation is involved in several important biological processes and may provide a target for the prevention and treatment of some human diseases. In this review, we discuss the properties of Z-DNA, proteins that are known to bind specifically to Z-DNA, and potential biological functions of this non-canonical DNA structure.





## Z-DNA the new biology: The third dimension of cancer therapeutics

DNA comes in many different shapes and sizes. Z-DNA, also known as left-handed DNA, is different from the more familiar right-handed B-DNA. Until recently, the role of Z-DNA in humans was a mystery. In a scientific breakthrough, Dr Alan Herbert of InsideOutBio Inc., Charlestown, Massachusetts, has identified the purpose of unusual DNA sequences called “flipons”. Flipons get their name from their ability to “flip” their conformation, from right-handed to left-handed DNA. Flipons change the way that genes are read out, altering the programming of cells and their response to the environment. Flipons can turn-off the immune response, an ability sometimes hijacked by cancer cells to avoid rejection by the body’s immune system. These discoveries hold the promise of new treatments for diseases like cancer in the future.

The unique structure of the DNA double helix is one of the most recognisable images in biology. Most of the DNA in our cells takes the form of B-DNA. The familiar B-DNA is also known as right-handed DNA because the DNA strands wind to the right. The B-DNA double helix structure was discovered by Watson and Crick in 1953. Less well-known, however, is a different type of DNA: the left-handed Z-DNA. Z-DNA was discovered by chance and, until recently, its role – or even whether it had any purpose at all – remained a mystery. Z-DNA is not a mirror image of B-DNA, but has its own unique shape, a feature nature makes use of.

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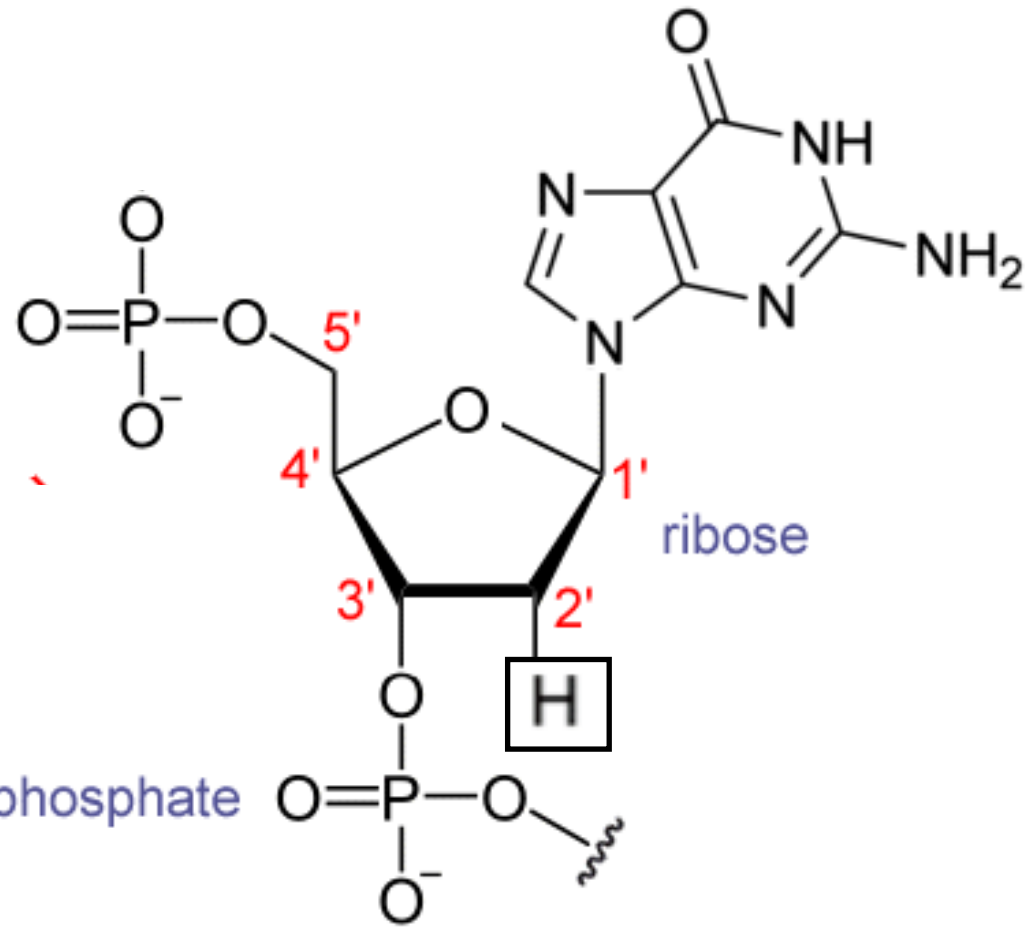
### Article References

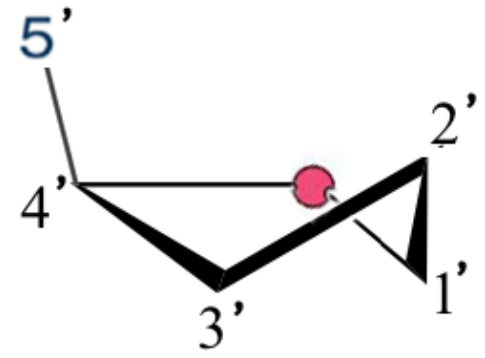
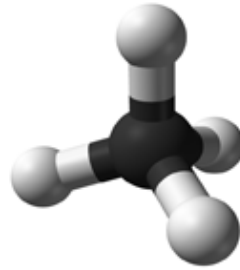
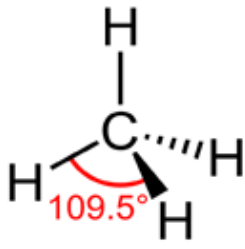
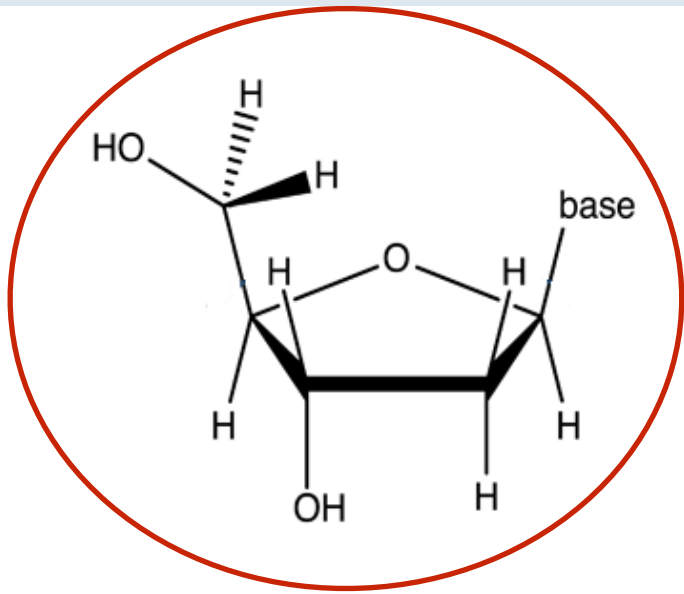
Herbert, A. (2019). Z-DNA and Z-RNA in human disease. *Commun Biol.*, [online] 2(7).  
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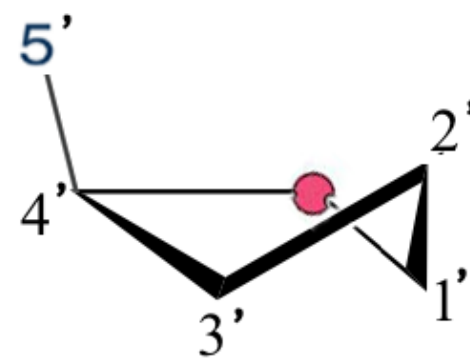
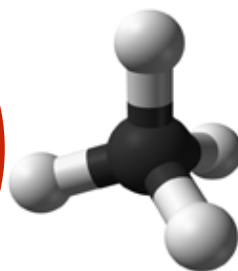
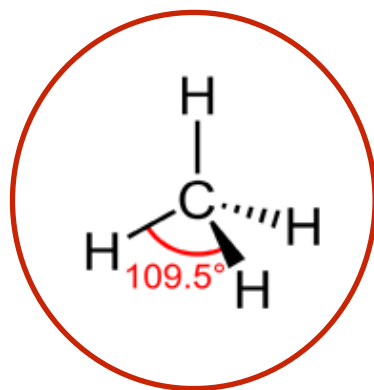
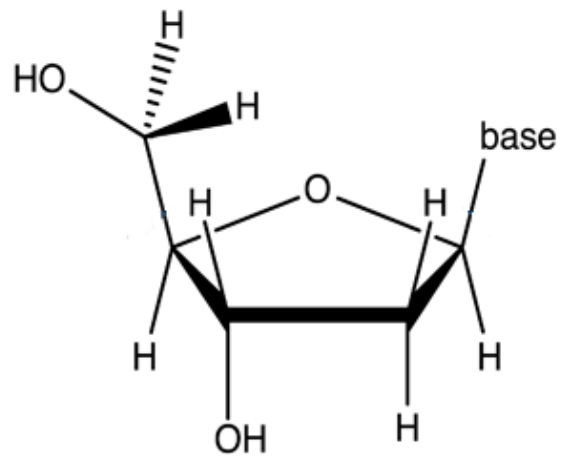
Herbert, A. (2019). Mendelian disease caused by variants affecting recognition of Z-DNA and Z-RNA by the Zα domain of the double-stranded RNA editing enzyme ADAR. *European Journal of Human Genetics*, [online]  
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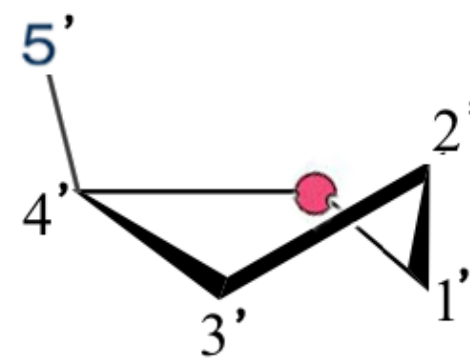
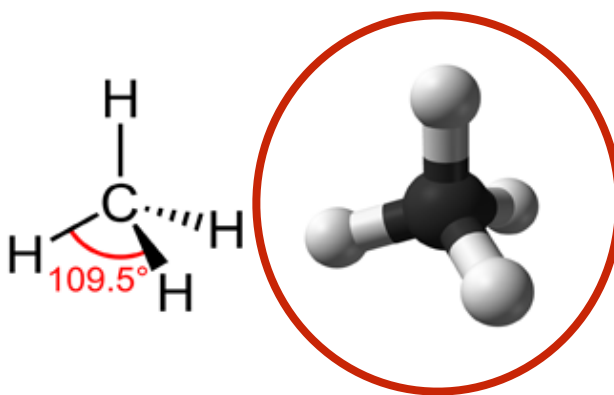
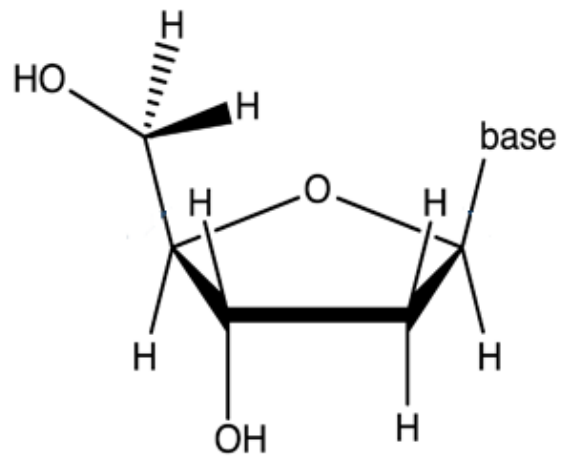


base  
(here: guanine)

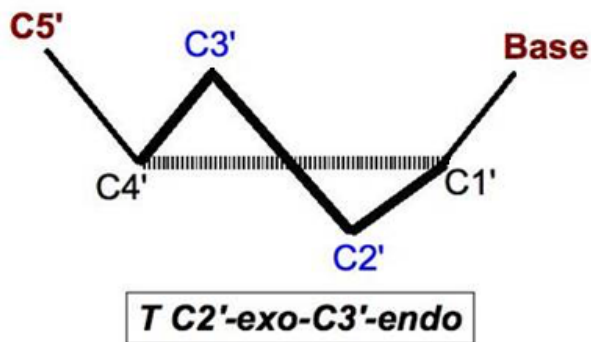
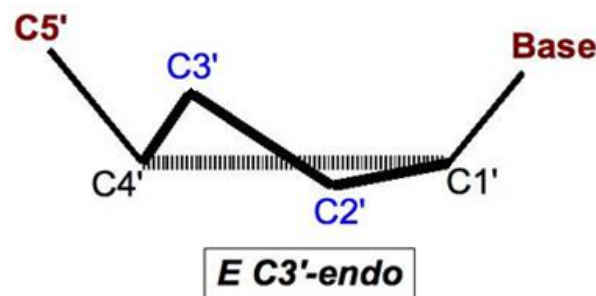
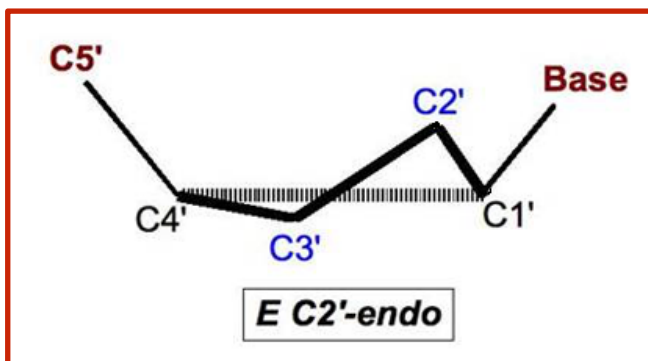
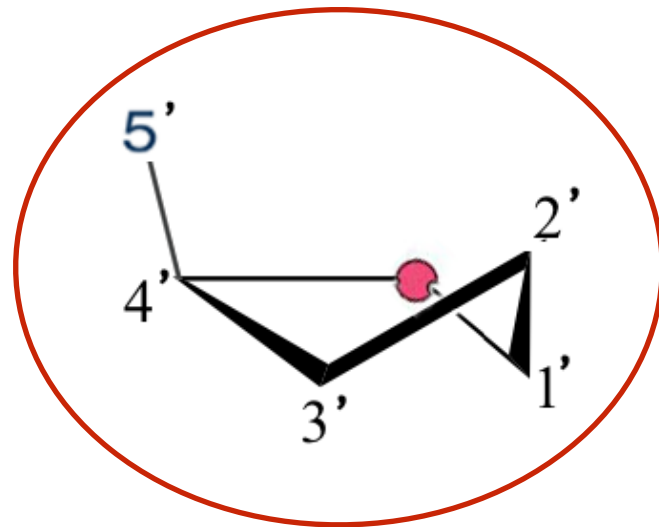
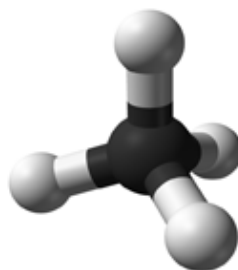
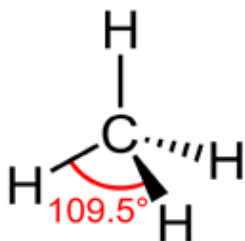
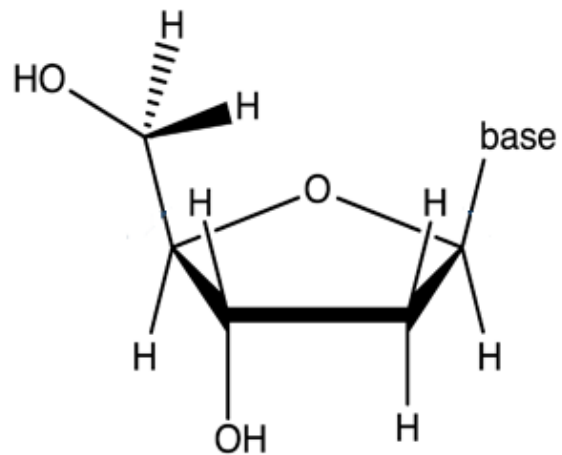


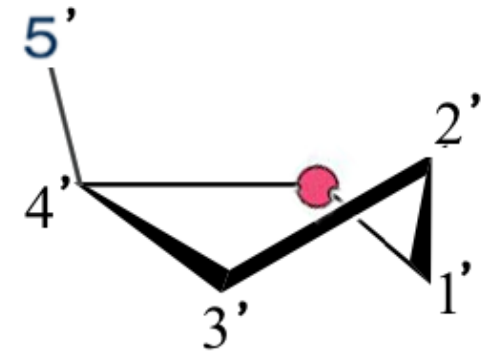
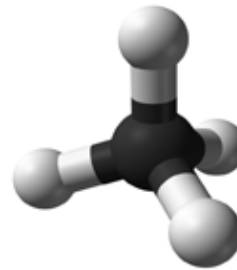
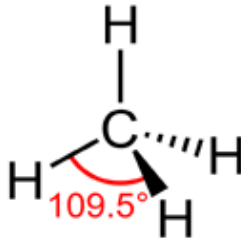
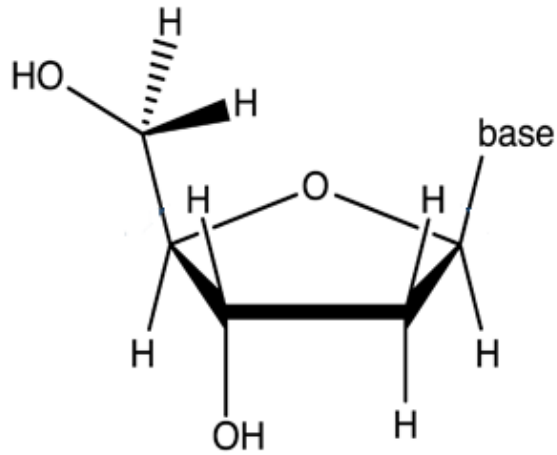






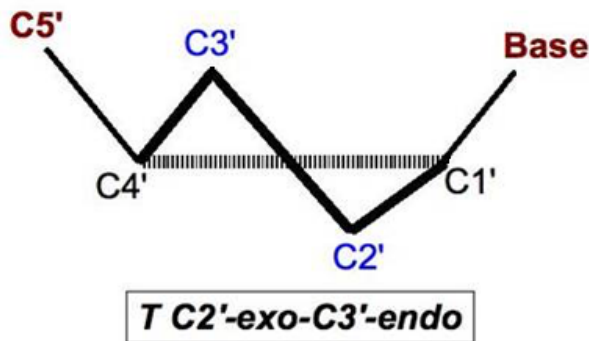
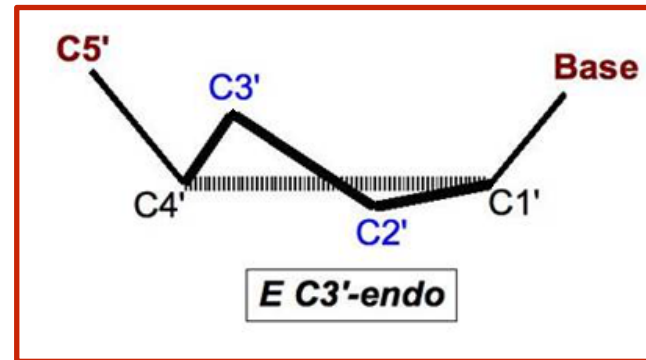
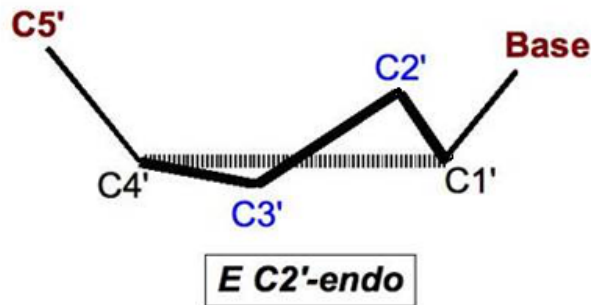


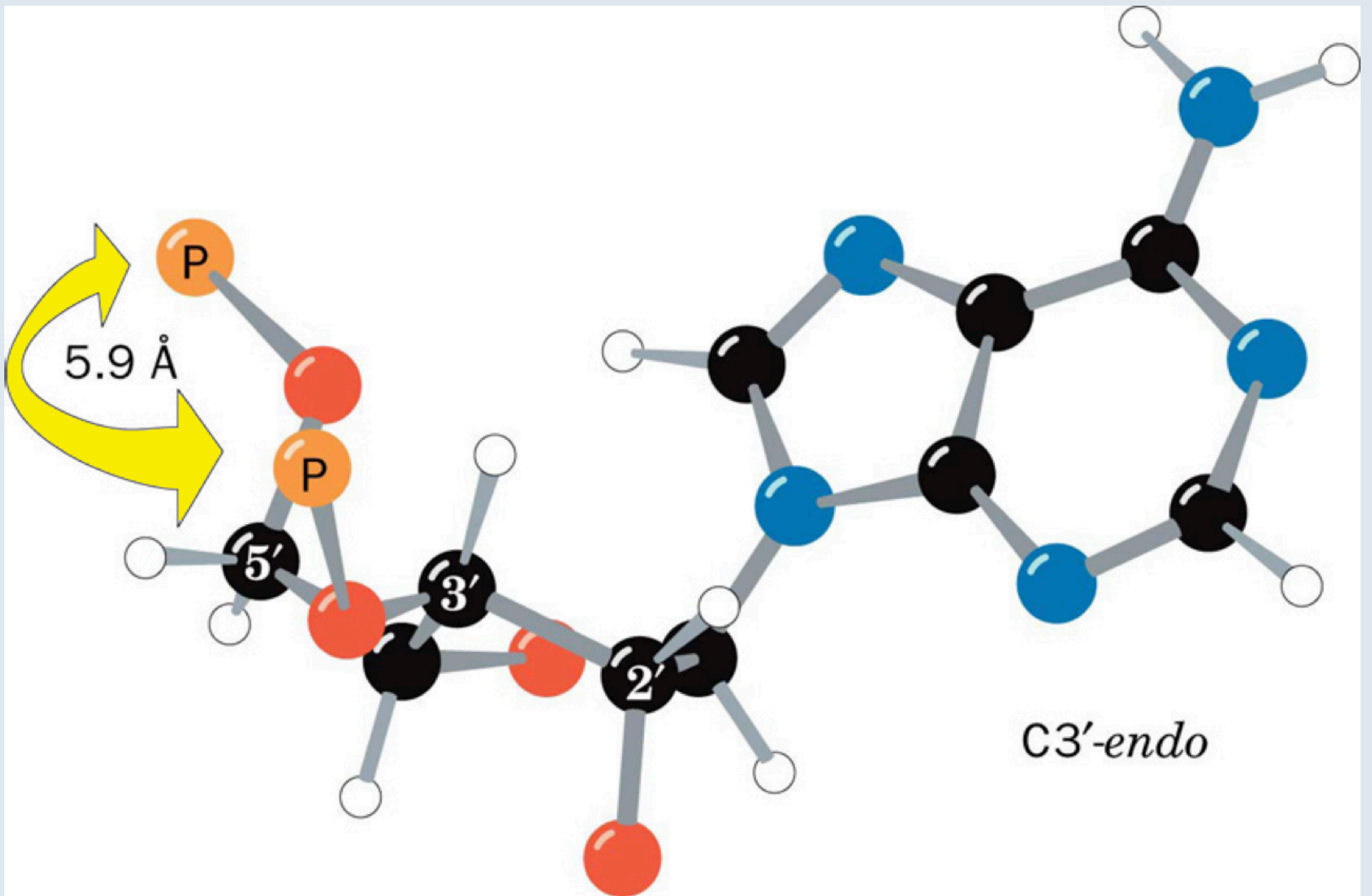


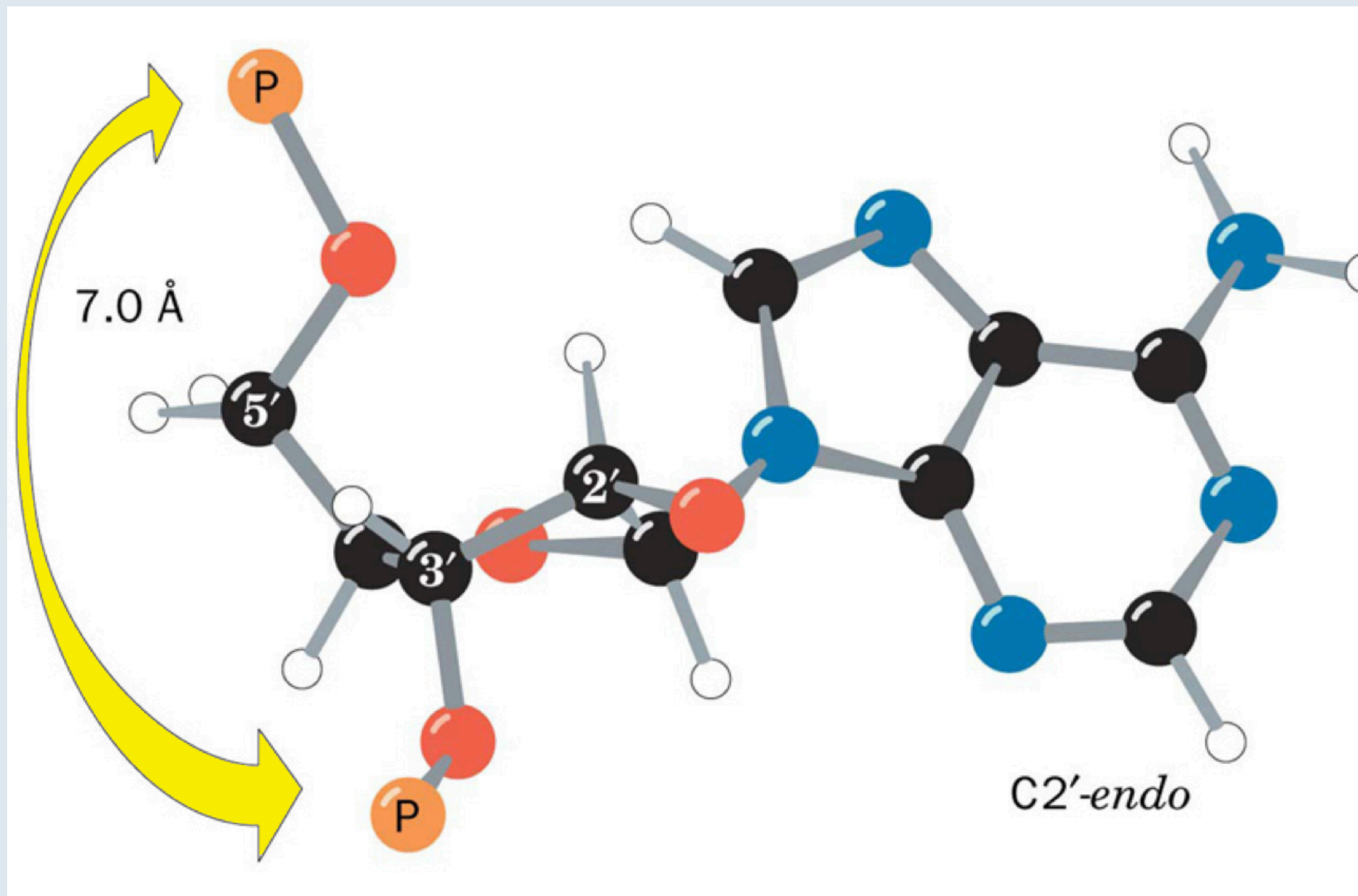


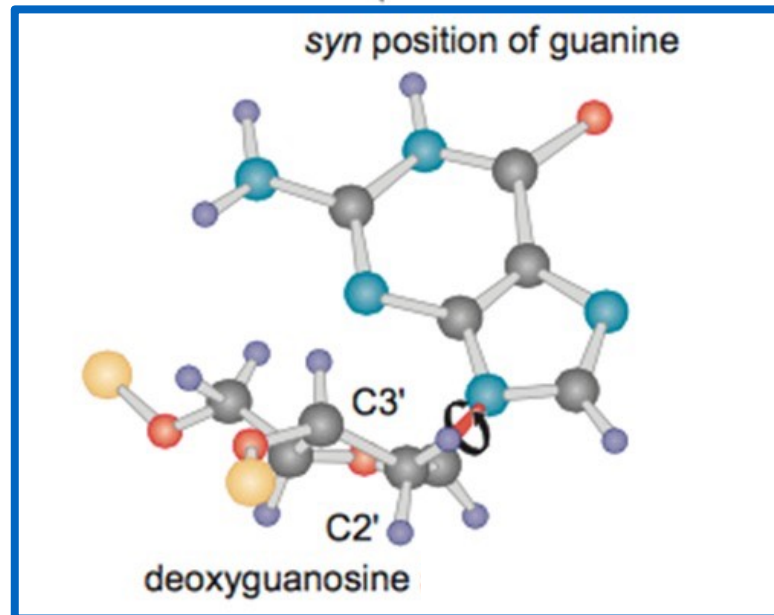
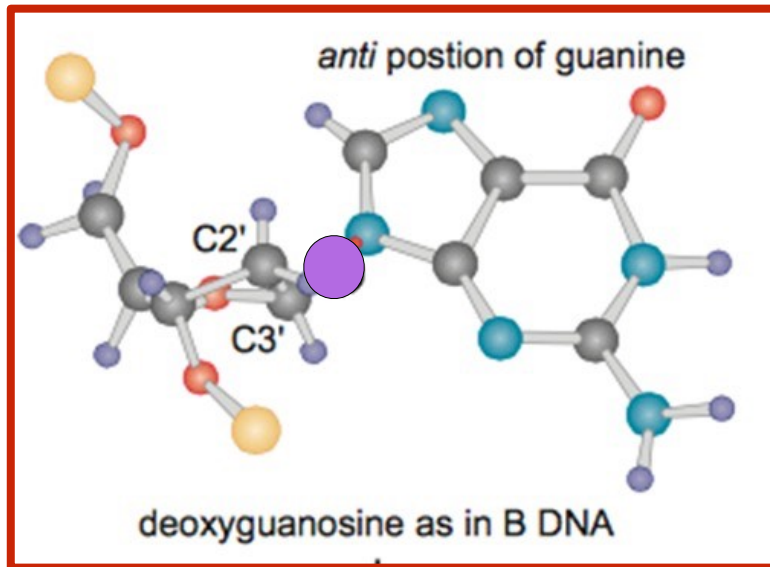
## B DNA

## A DNA (RNA or DNA/RNA hybrid)



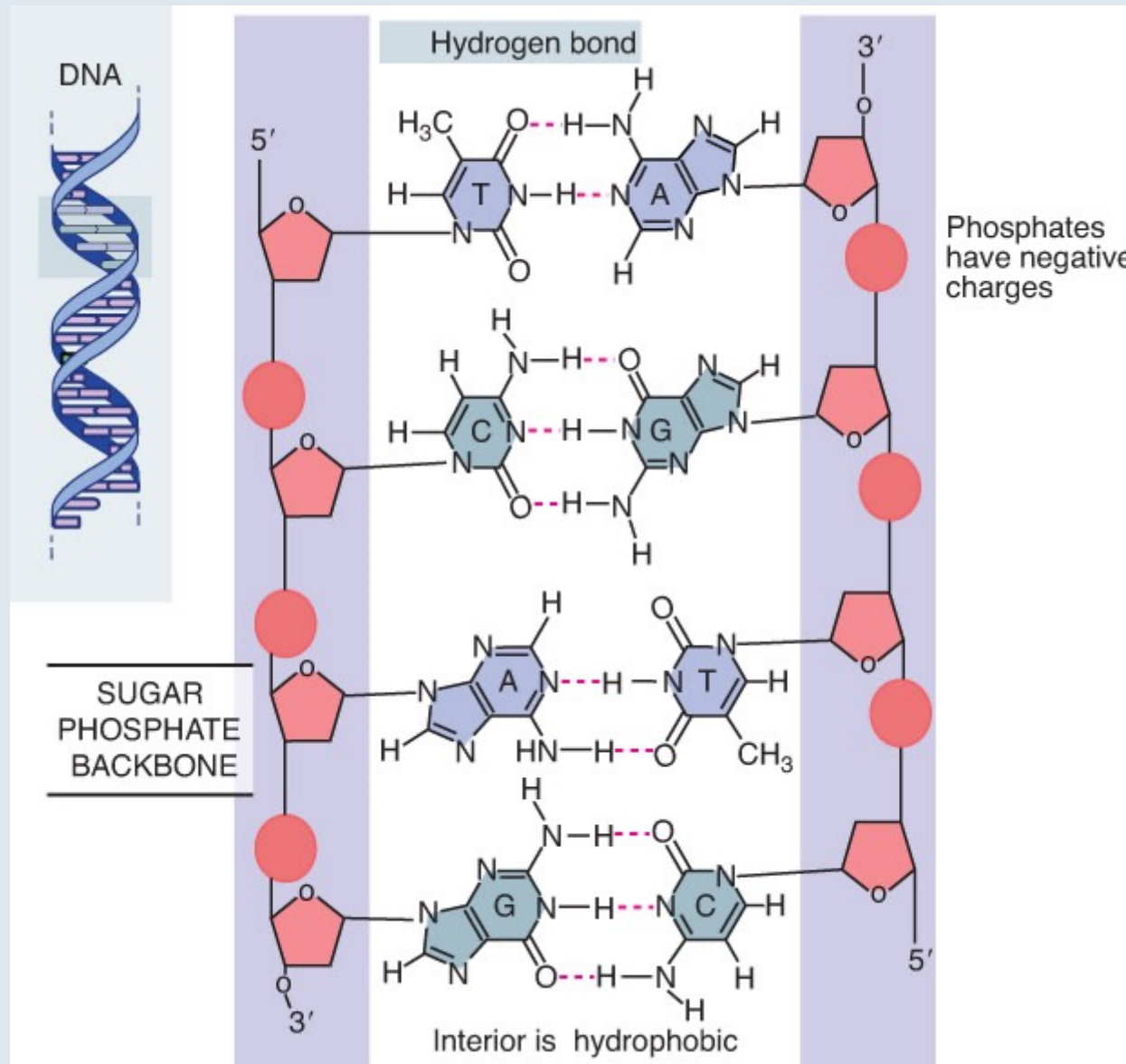




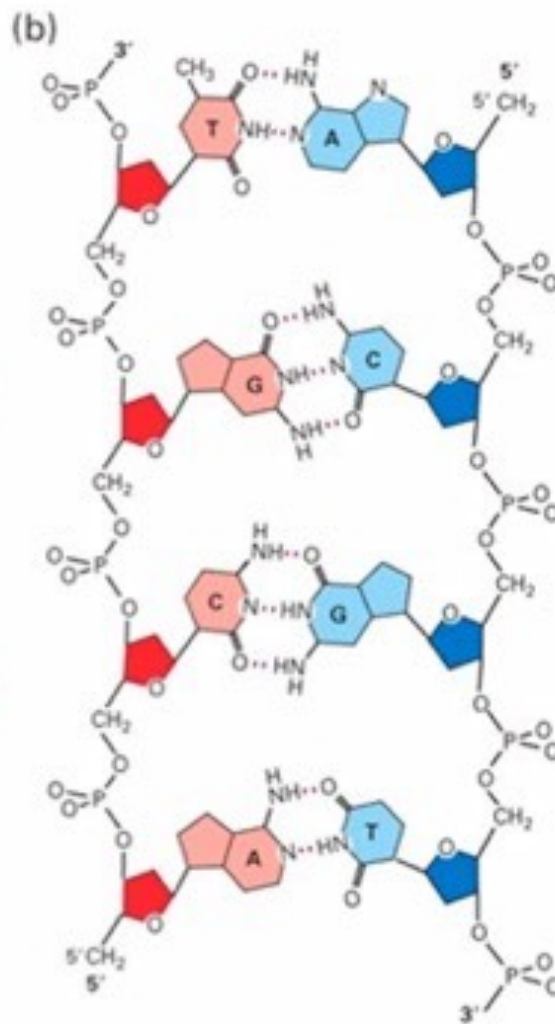
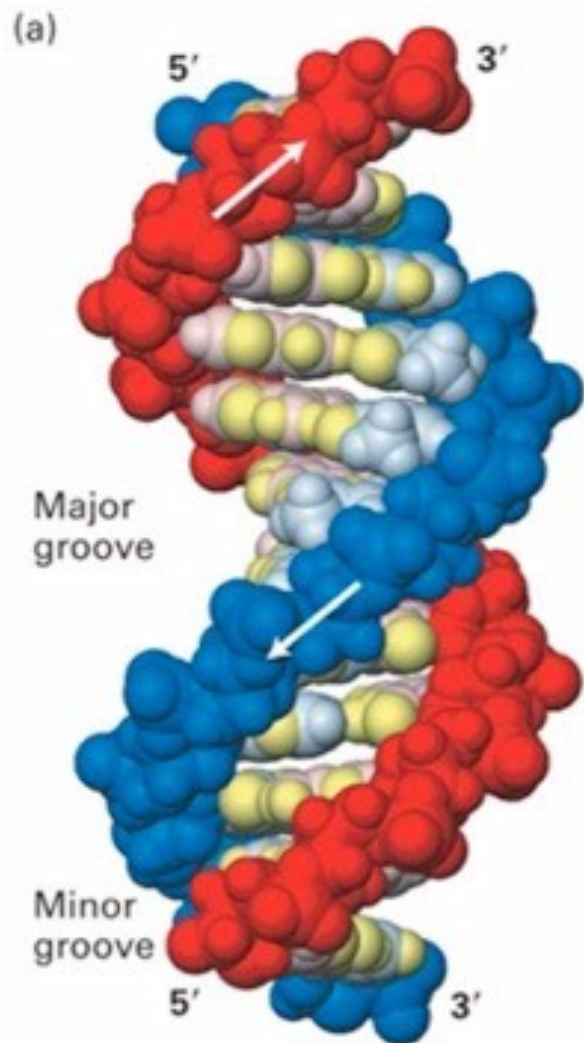




<b>Geometry attribute</b>	<b>A-form</b>	<b>B-form</b>	<b>Z-form</b>
Helix sense	right-handed	right-handed	left-handed
Repeating unit	1 bp	1 bp	2 bp
Rotation/bp	32.7°	35.9°	60°/2
bp/turn	11	10.5	12
Inclination of bp to axis	+19°	-1.2°	-9°
Rise/bp along axis	2.3 Å (0.23 nm)	3.32 Å (0.332 nm)	3.8 Å (0.38 nm)
Pitch/turn of helix	28.2 Å (2.82 nm)	33.2 Å (3.32 nm)	45.6 Å (4.56 nm)
Mean propeller twist	+18°	+16°	0°
Glycosyl angle	anti	anti	C: anti, G: syn
Sugar pucker	C3'-endo	C2'-endo	C: C2'-endo, G: C3'-endo
Diameter	23 Å (2.3 nm)	20 Å (2.0 nm)	18 Å (1.8 nm)



The double helix maintains a constant width because purines always face pyrimidines in the complementary A-T and G-C base pairs.



## B form DNA

2.0 nM dia (20 Å)

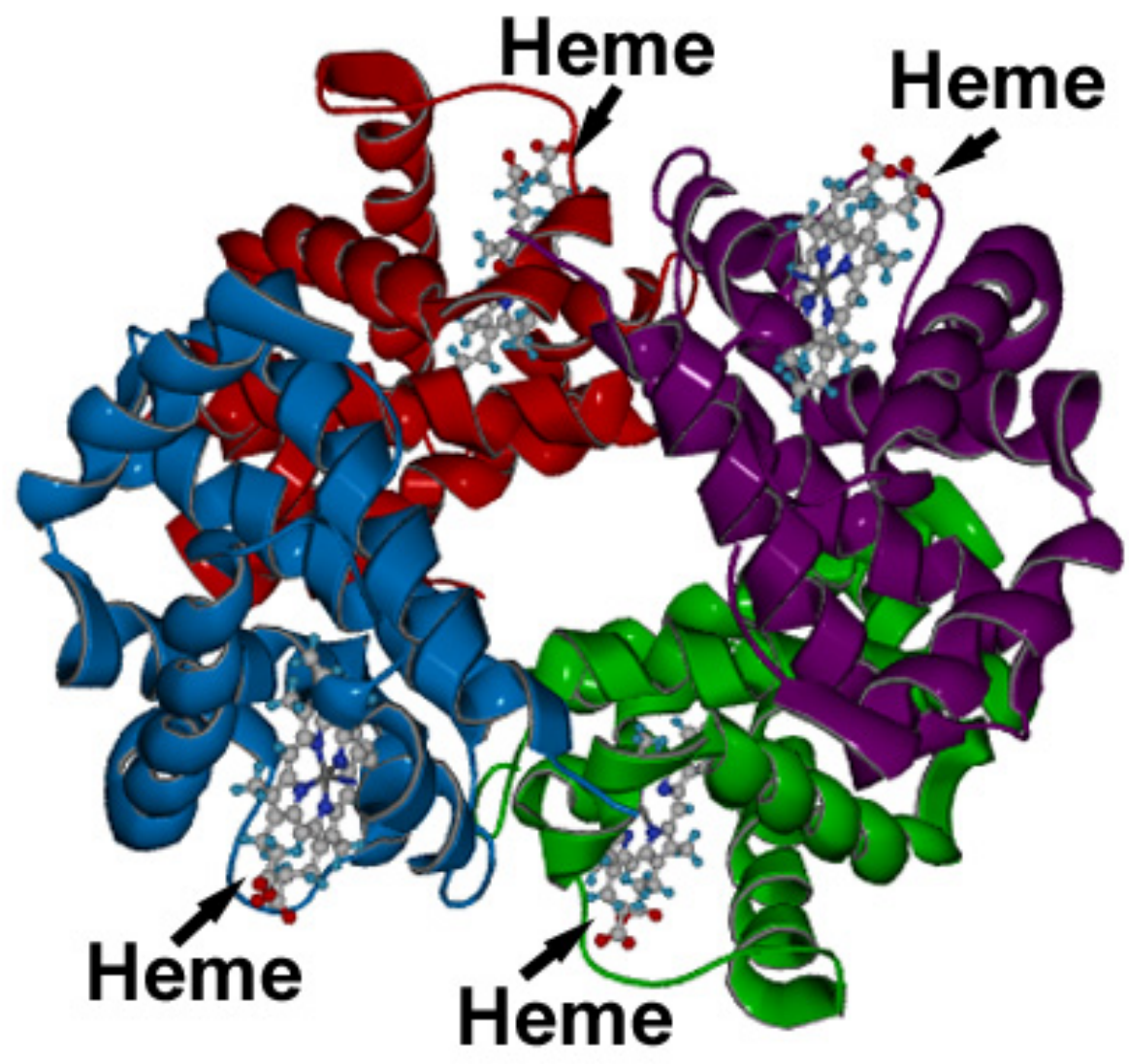
0.36 nM (3.6 Å)

between bases

~10 bases per turn

antiparallel strands

bases perpendicular  
to axis





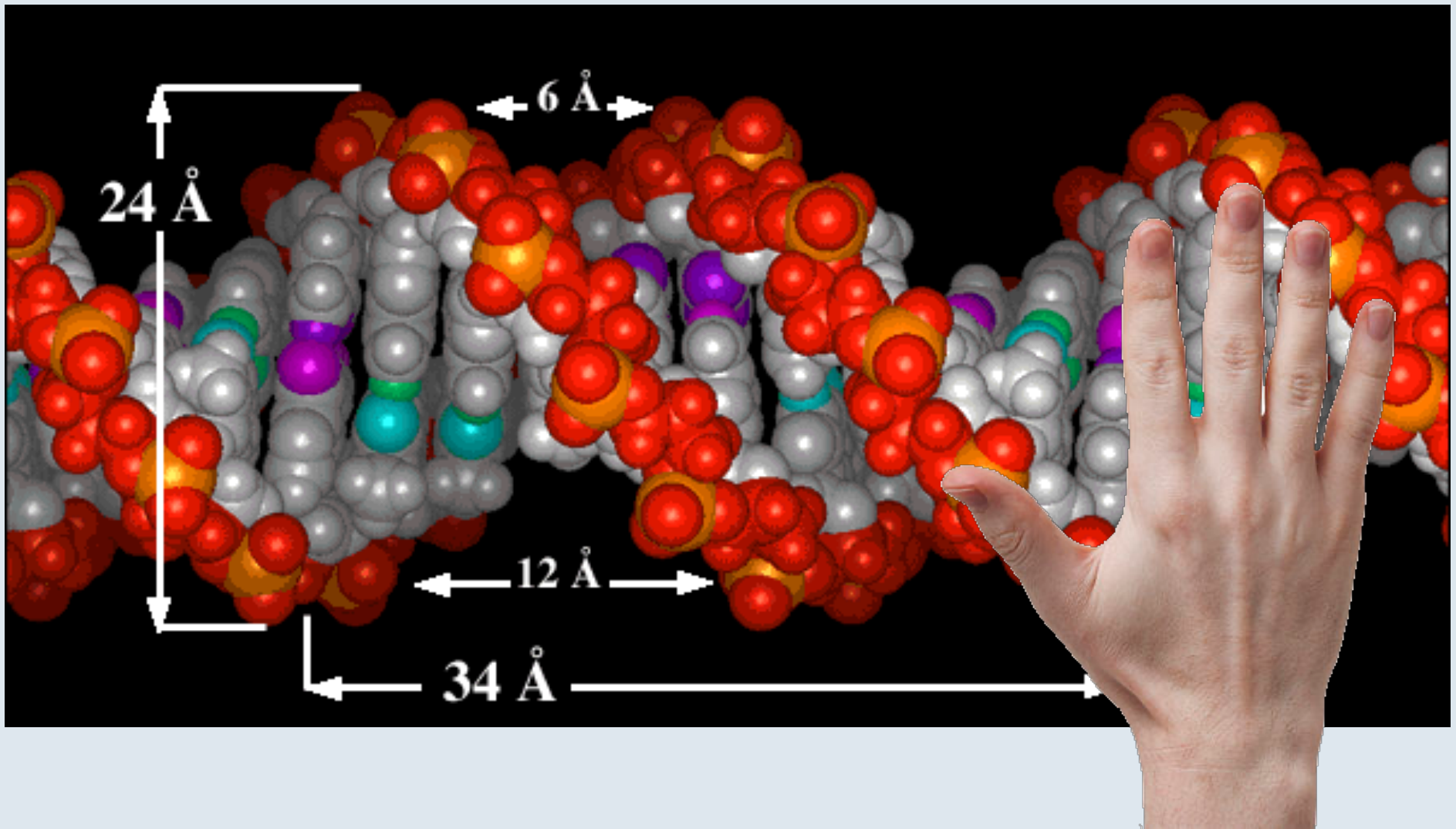
Rubin's Vase illusion

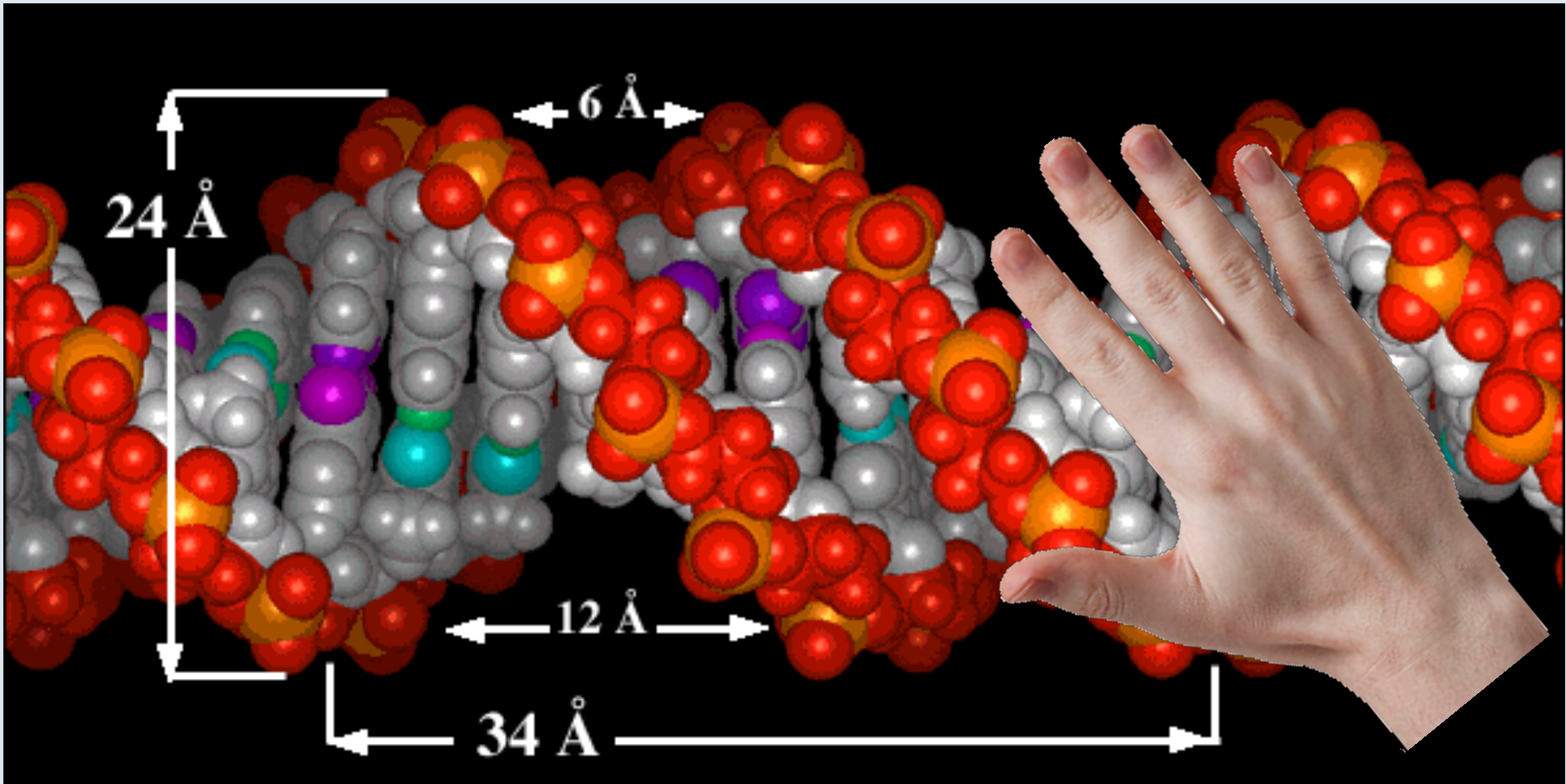




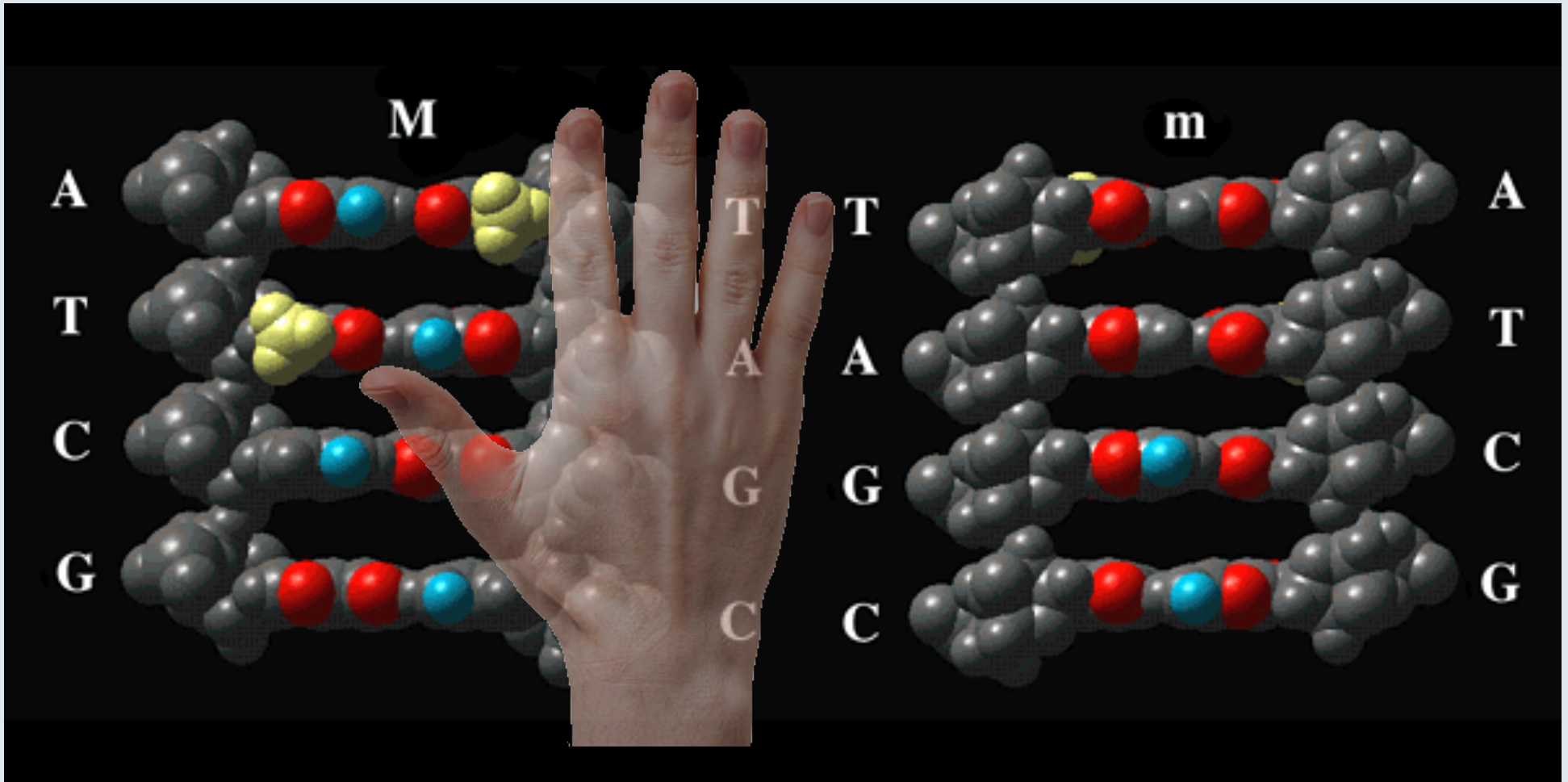
Handwritten signature or mark at the bottom of the image.



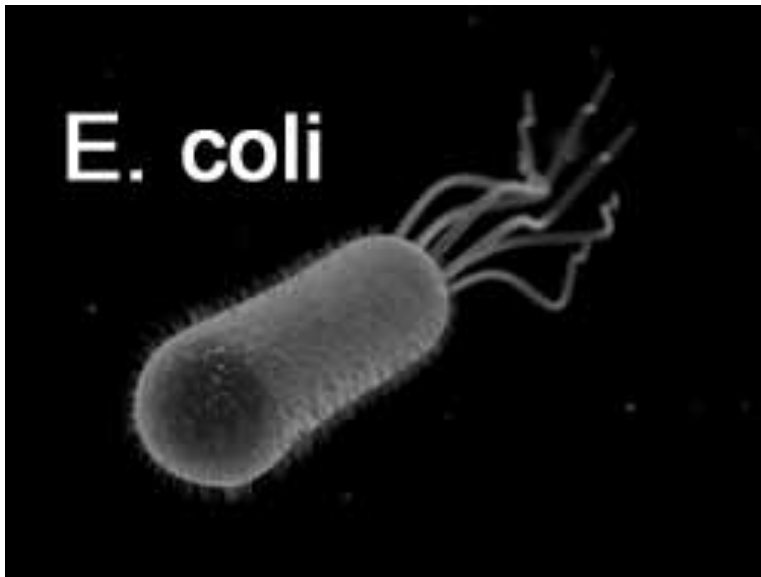












Bacteria thrive on many different types of food. But most yeasts can live only on sugars and starches. From these, they produce carbon dioxide gas and alcohol. Thus, they have been useful to man for centuries in the production of certain foods and beverages. They are responsible for the rising of bread dough and the fermentation of wines, whiskey, brandy and beer. They also play the initial role in the production of vinegar.

**~ 1.5 - 3  $\mu\text{m}$**

**Prokaryote**

**DNA is Circular**

**Cell membrane**



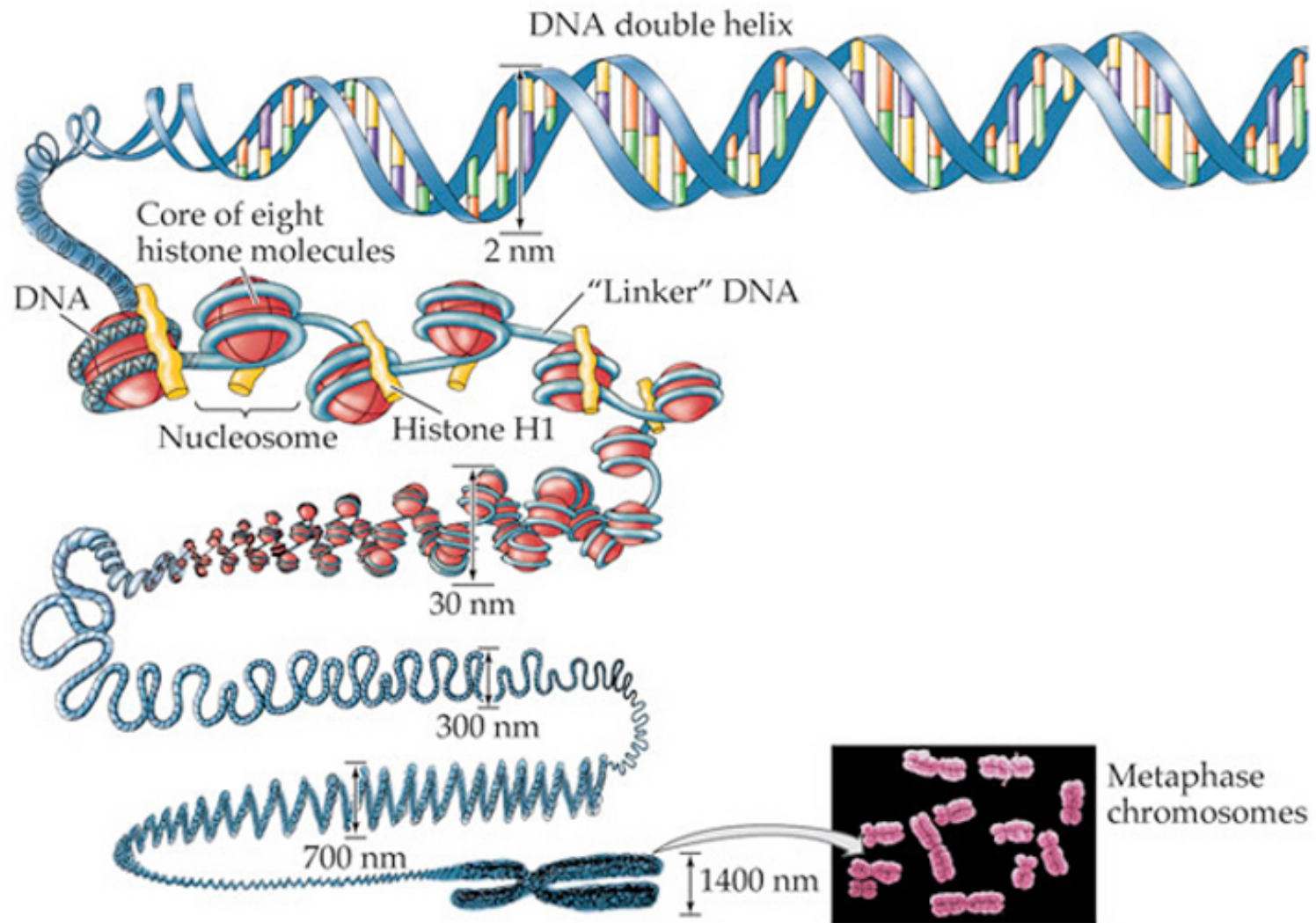
Yeasts are small, *single-celled plants*. They are members of the family *fungi* (singular, *fungus*), which also includes mushrooms. Fungi differ from other plants in that they have no chlorophyll.

**~ 50  $\mu\text{m}$**

**Eukaryote**

**DNA is linear**

**Cell wall**



LIFE: THE SCIENCE OF BIOLOGY, Seventh Edition, Figure 9.8 DNA Packs into a Mitotic Chromosome  
 © 2004 Sinauer Associates, Inc. and W. H. Freeman & Co.

- There are effectively five classes of histones.

There are effectively five classes of histones,

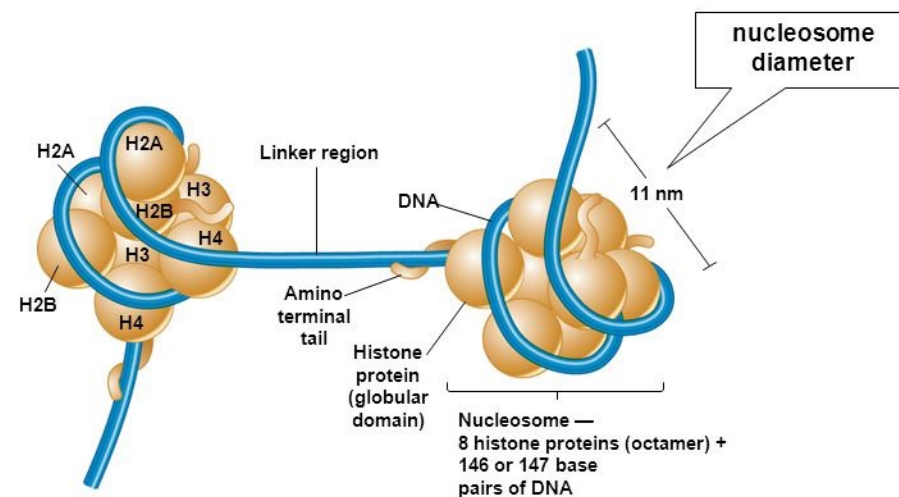
The core of a nucleosome contains eight histone molecules, two each from four of the histone classes.

There are **146 - 160 base pairs** of DNA wrapped around the core, or  $>1.65$  turns of DNA.

One molecule from the remaining histone class, histone H1, clamps the DNA to the core, and helps form the next level of packaging.

During **mitosis** and **meiosis**, the **chromatin** becomes even more heavily coiled and condensed.

Nucleosomes shorten DNA ~seven-fold



(a) Nucleosomes showing core histone proteins

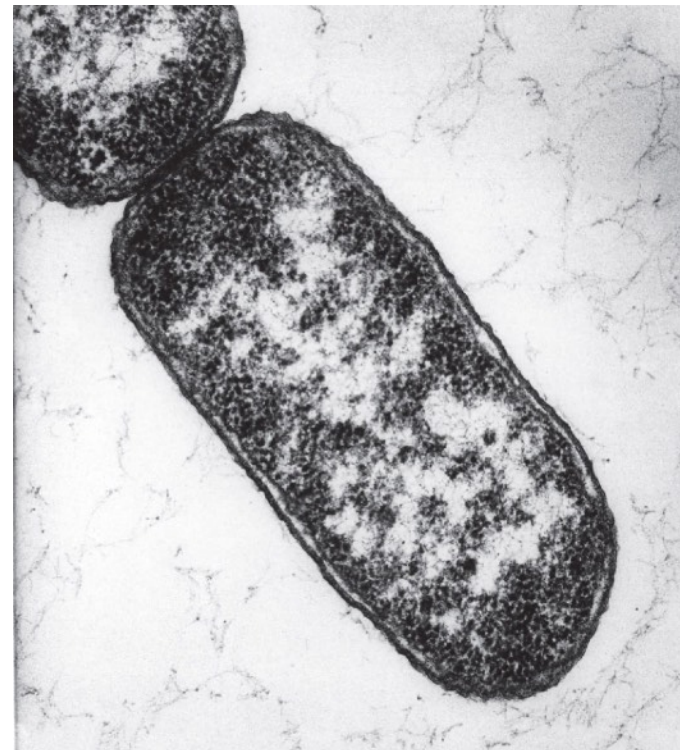
Brooker, Fig 12.10a

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# The Bacterial Genome Is a Nucleoid

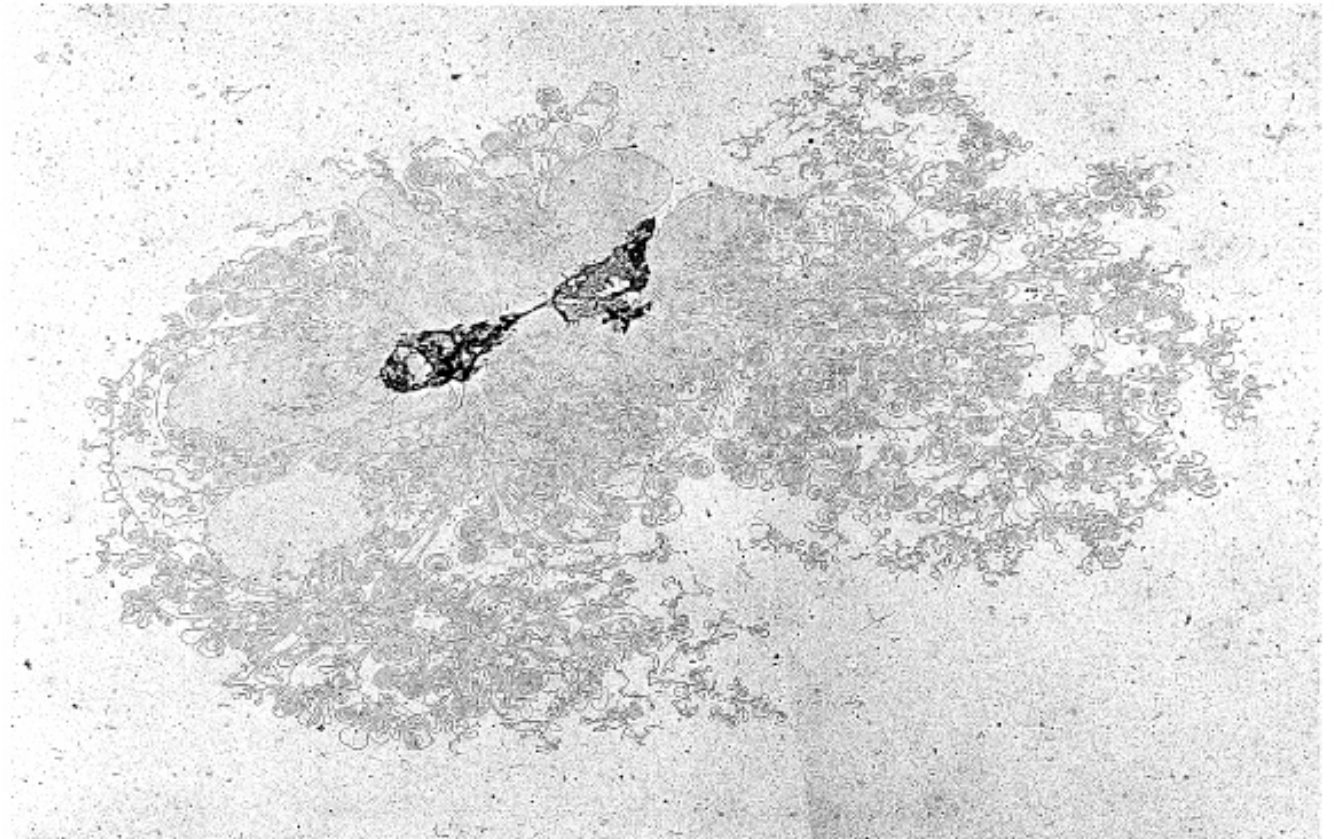
- The bacterial nucleoid is ~80% DNA by mass and can be unfolded by agents that act on RNA or protein.
- The proteins that are responsible for condensing the DNA have not been identified.

A thin section shows the bacterial nucleoid as a compact mass in the center of the cell





# The Bacterial Genome Is Supercoiled



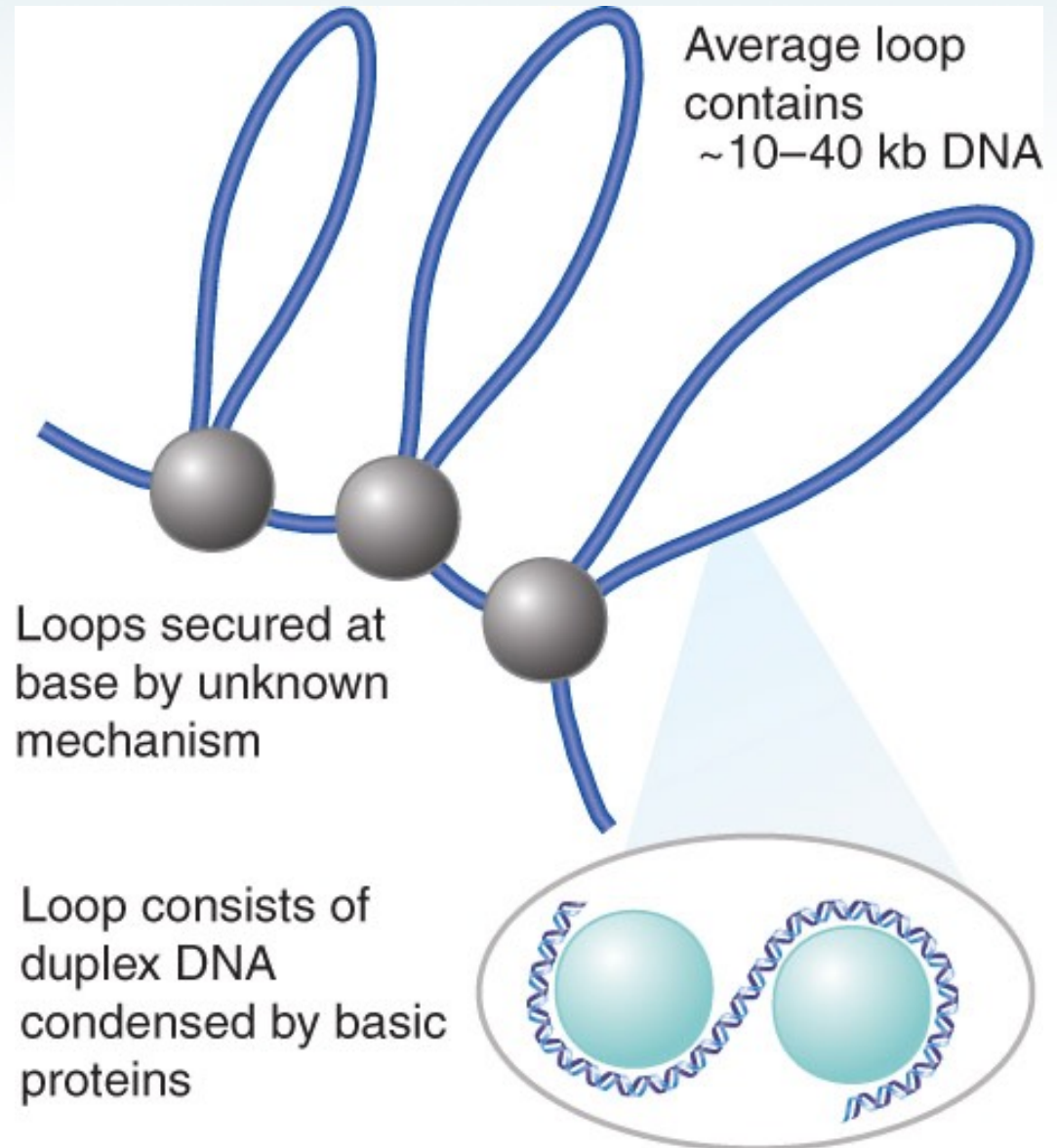
2  $\mu\text{m}$



# The Bacterial Genome Is Supercoiled

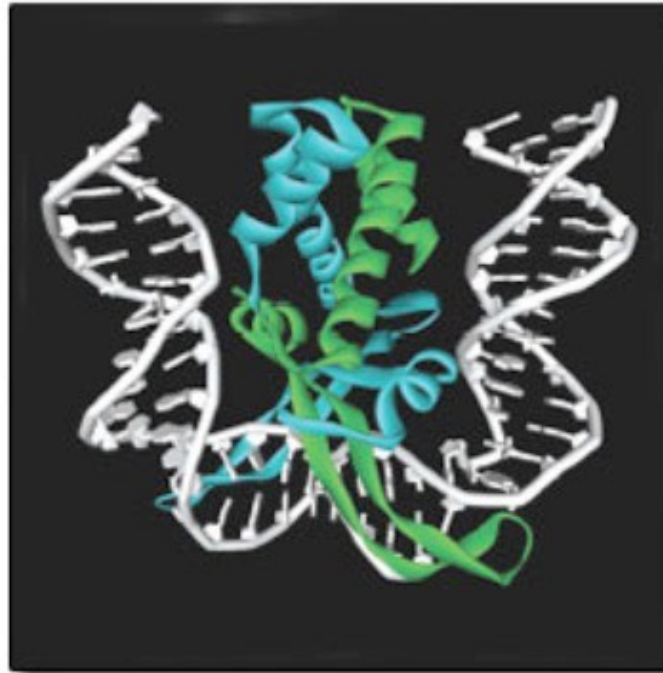
- The nucleoid has ~400 independent negatively supercoiled **domains**.
- The average density of supercoiling is... ~1 turn / 100bp.

The bacterial genome consists of a large number of loops of duplex DNA

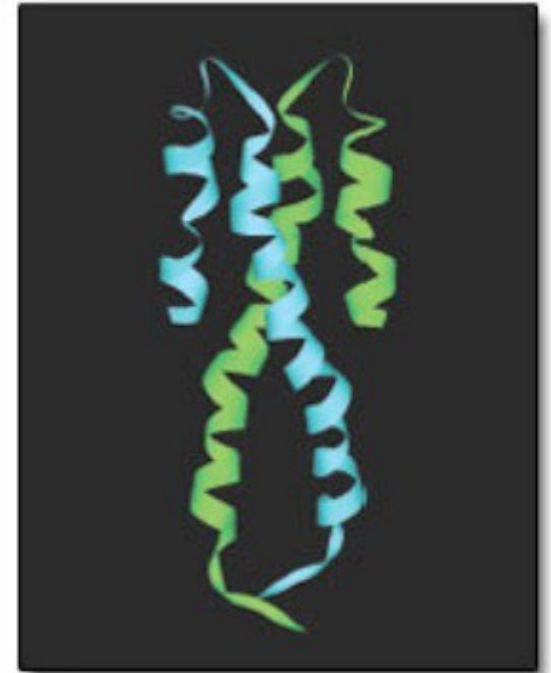




(a)



(b)

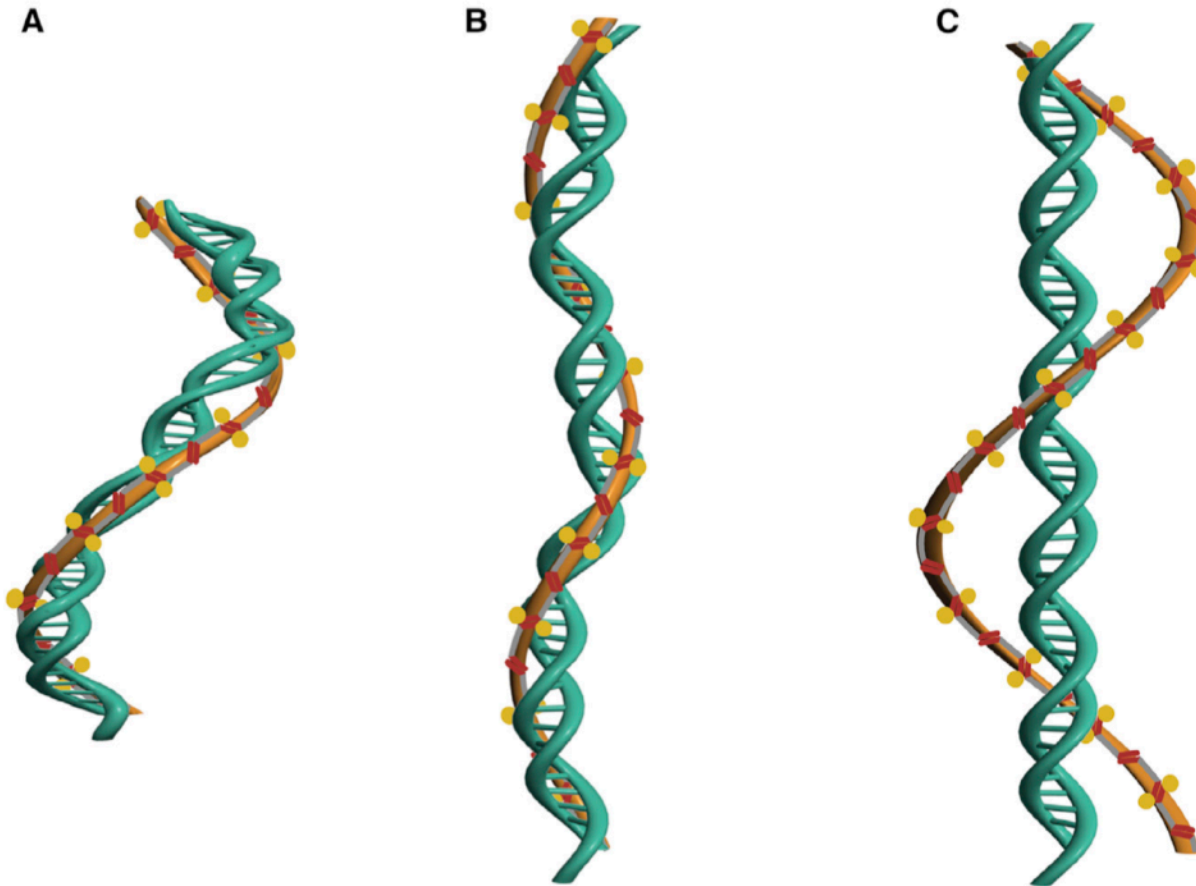
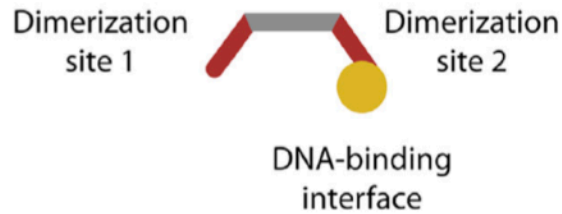


(c)

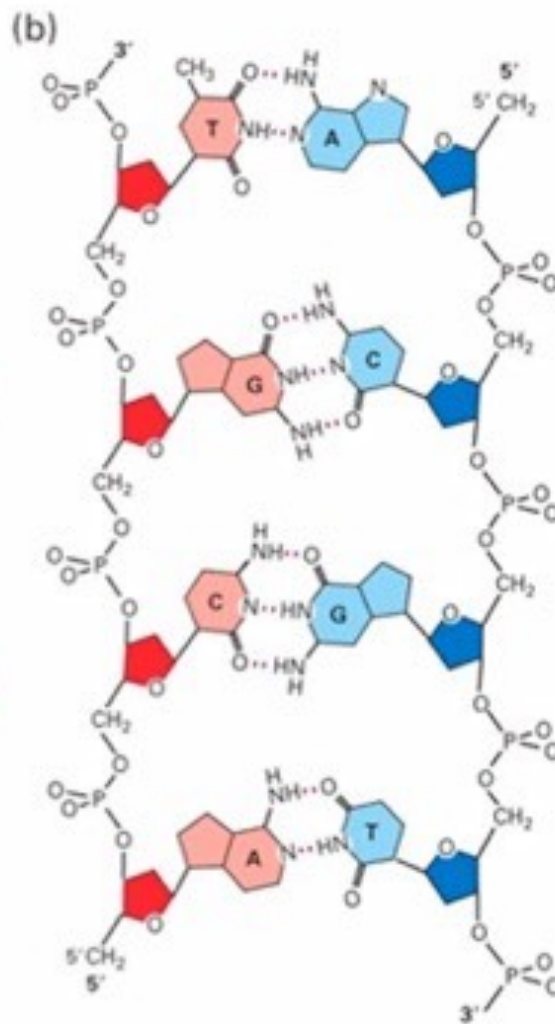
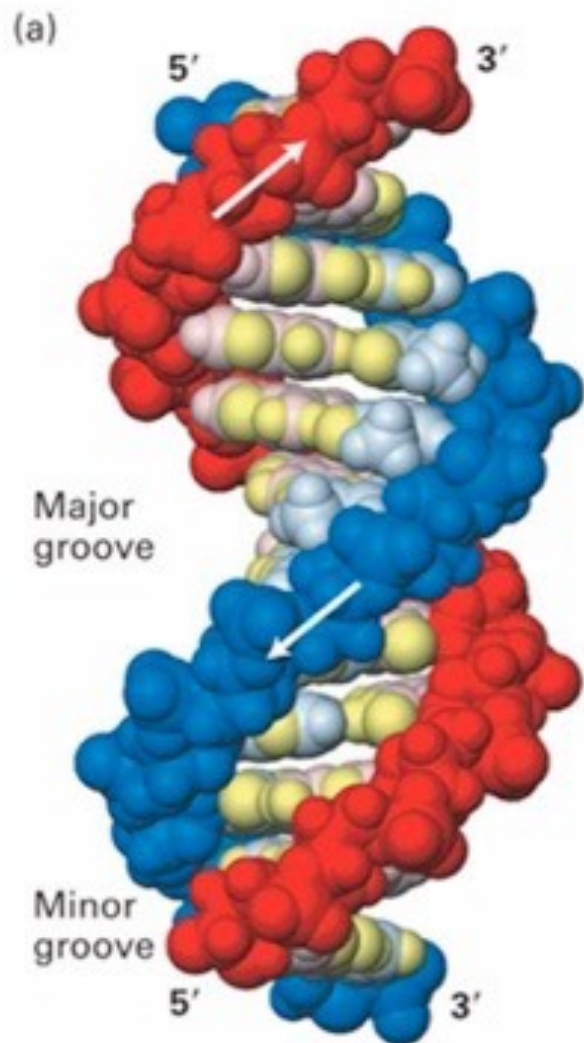
- (a) The HU protein dimer complexed with DNA
- (b) Binding of an *E. coli* IHF dimer to DNA induces a 180° Turn
- (c) Structure of the N-terminal domain of *E. coli* H-NS dimer.

**All structures show protein secondary structures and tubular DNA**

## H-NS



**FIGURE 3** H-NS forms a right-handed helical nucleoprotein filament. An H-NS molecule has two dimerization domains (Fig. 1), which enable H-NS to form a chain of linked H-NS molecules on DNA. Three H-NS nucleoprotein filament structures are proposed. (A) DNA wraps around an H-NS helix, reducing the contour length to ~50%. (B) A flexible H-NS nucleoprotein filament allows deformation into a thinner and longer structure. (C) H-NS wraps around the DNA in a helix, forming thick nucleoprotein filaments. The DNA is drawn to scale to highlight the change in contour length. Yellow circles (free DNA-binding domains) that are directed toward the outside of these helical filaments may allow interaction with naked DNA, hence forming filament-mediated DNA bridges under certain conditions.



## B form DNA

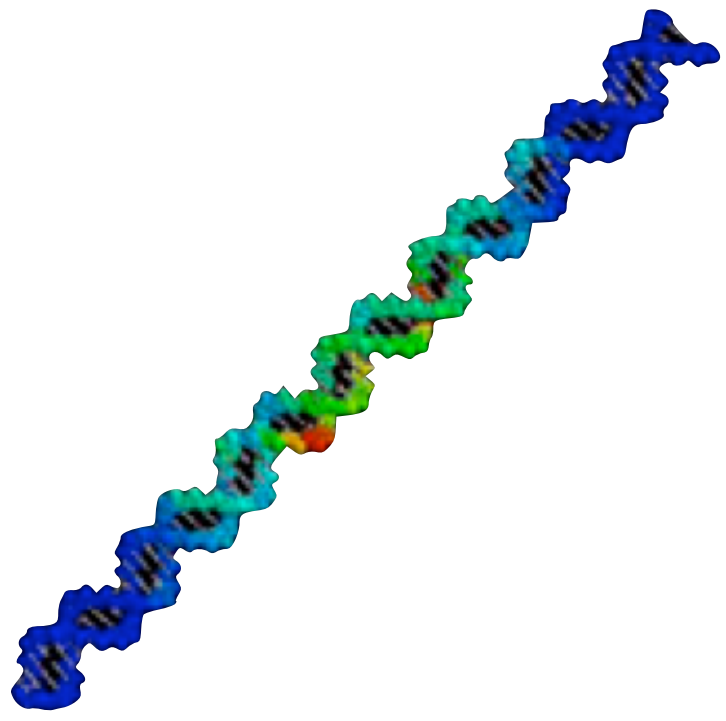
2.0 nM dia (20 Å)

0.36 nM (3.6 Å)

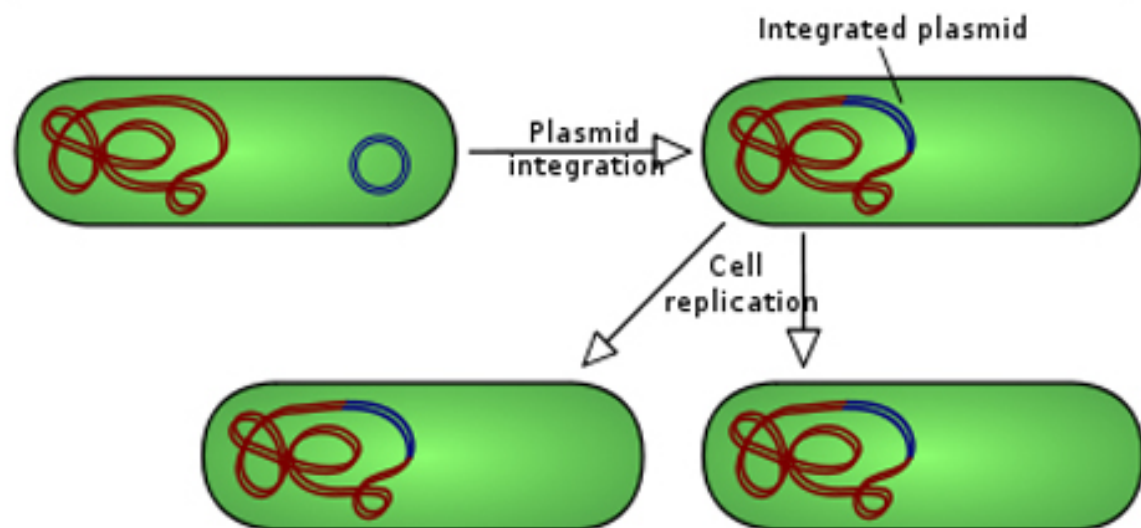
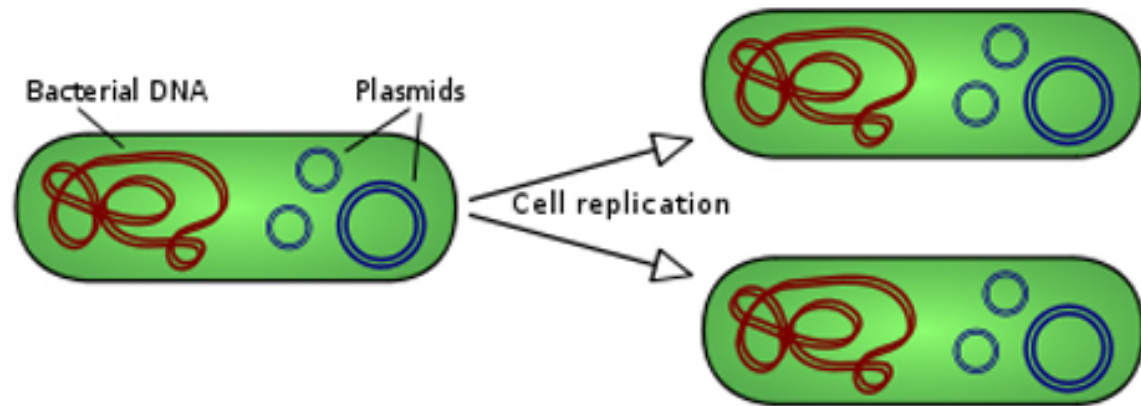
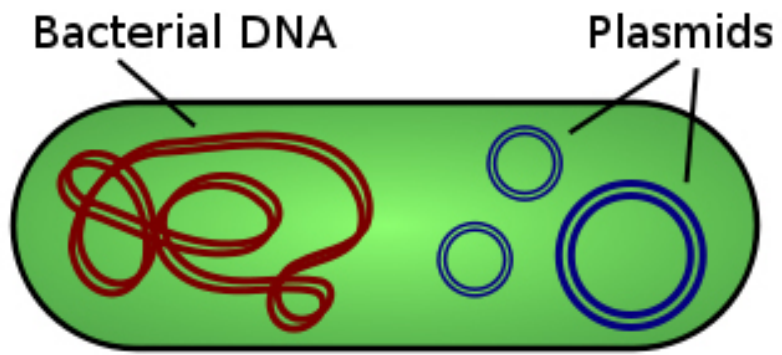
between bases

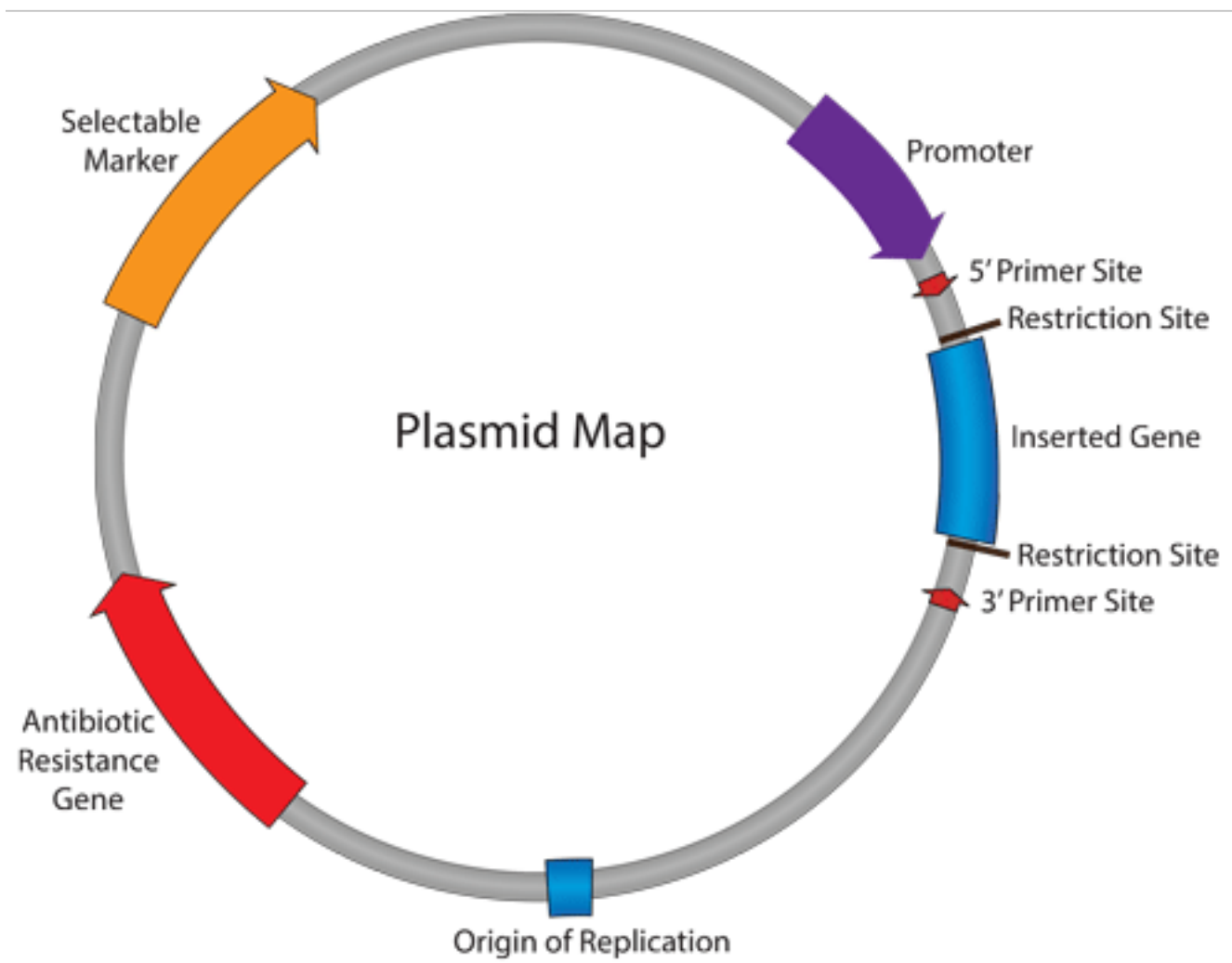
~10 bases per turn

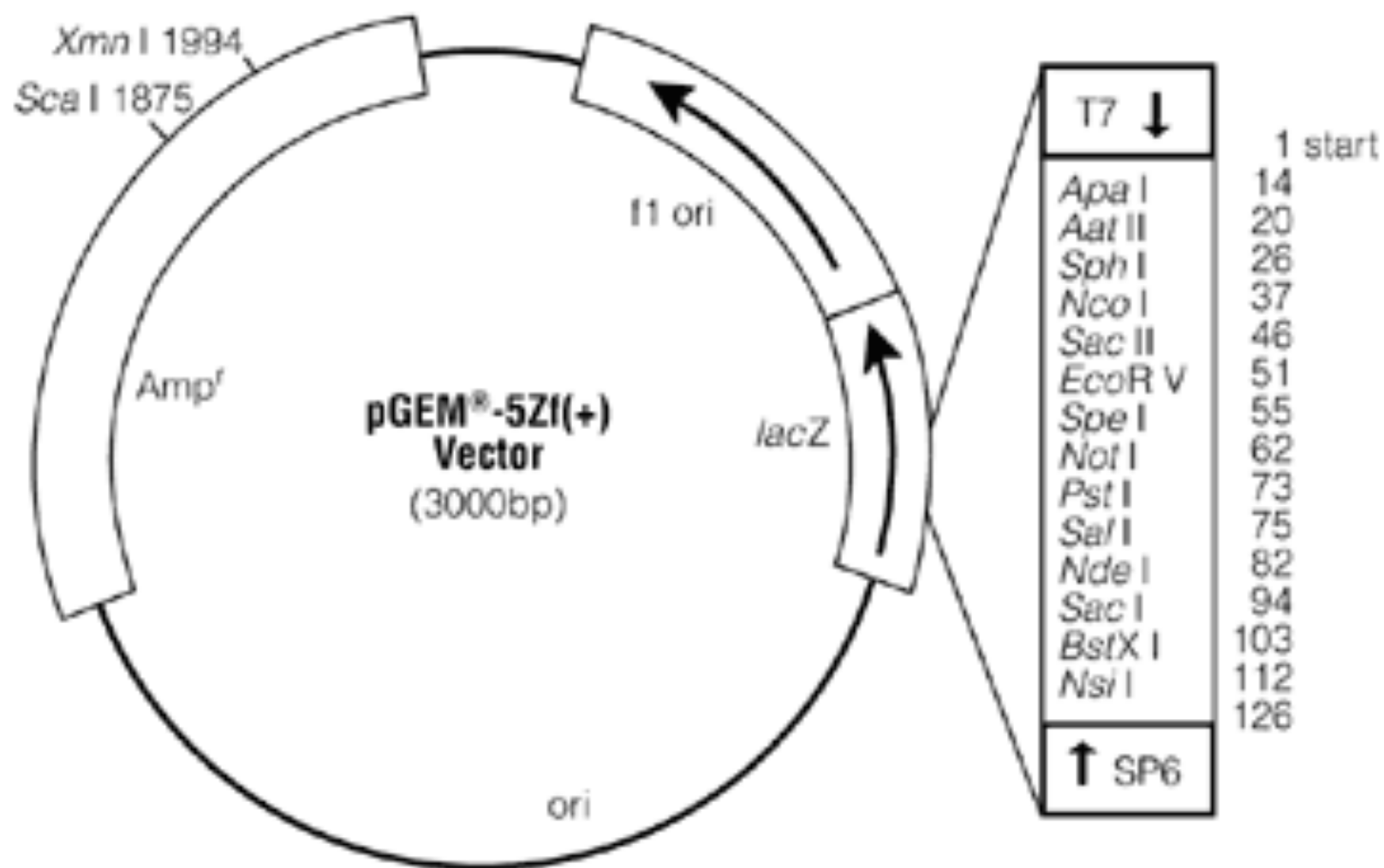
antiparallel strands  
bases perpendicular  
to axis

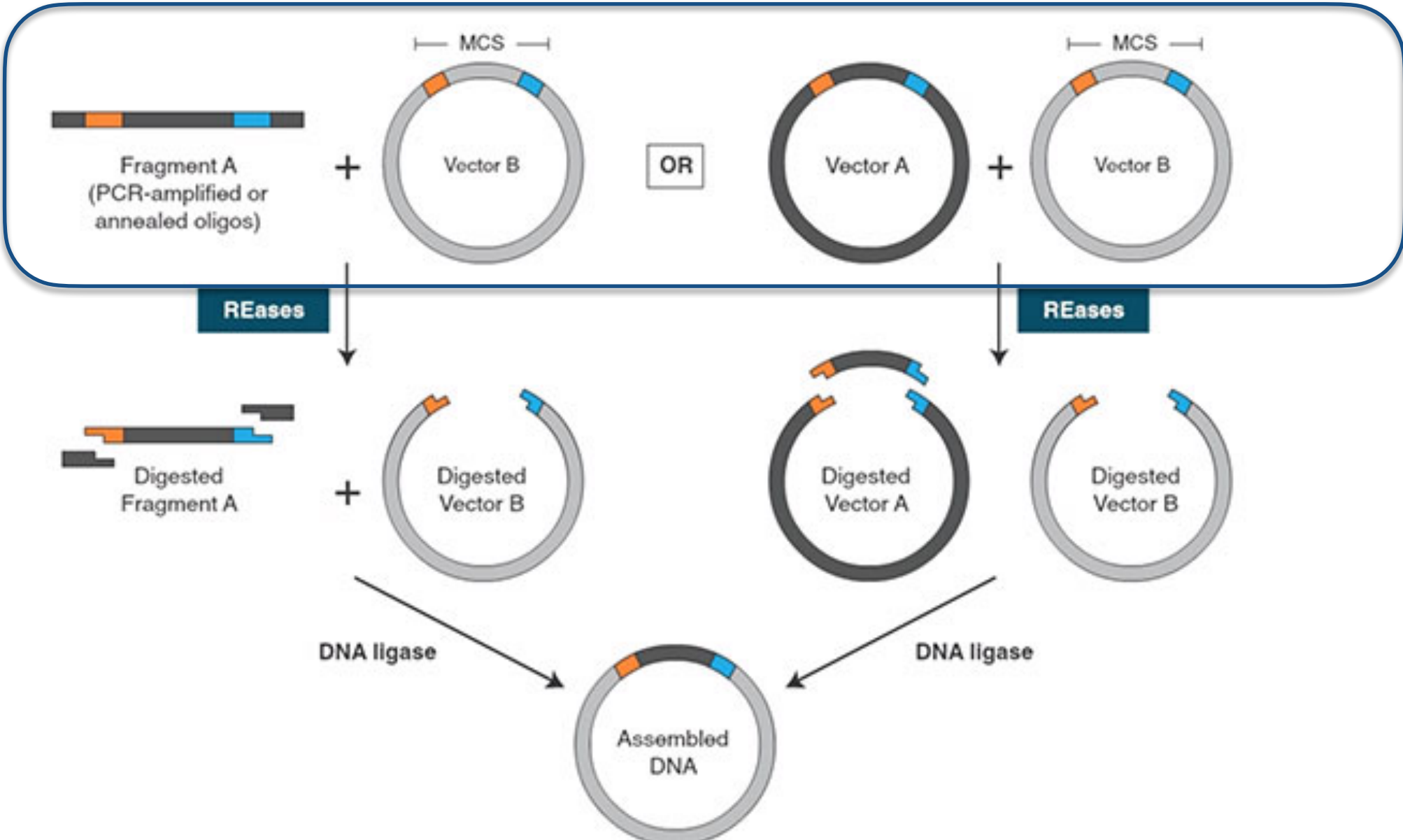


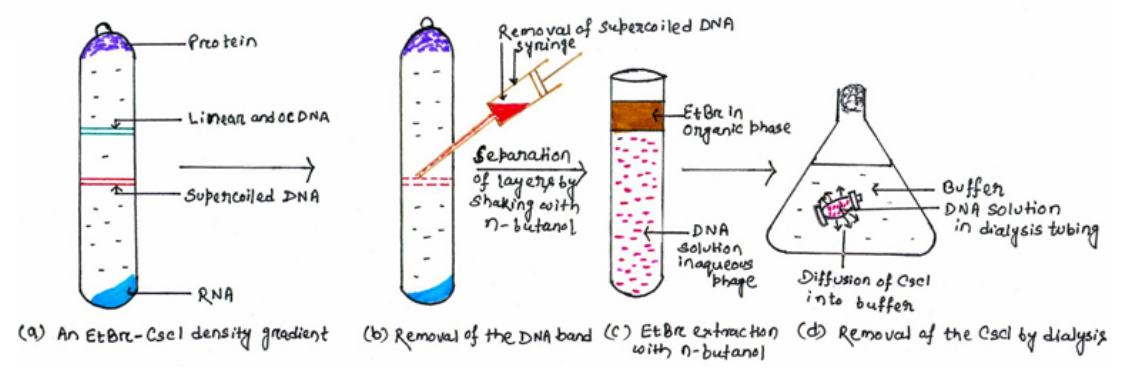
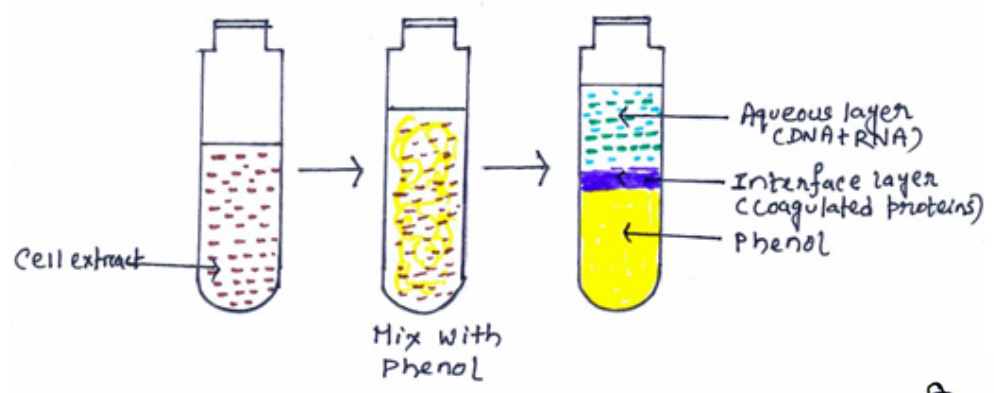
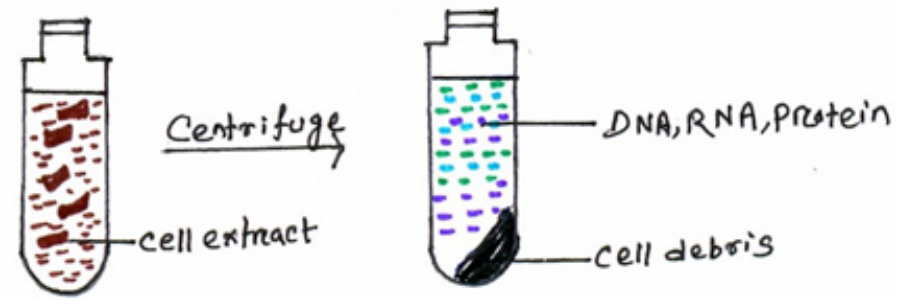












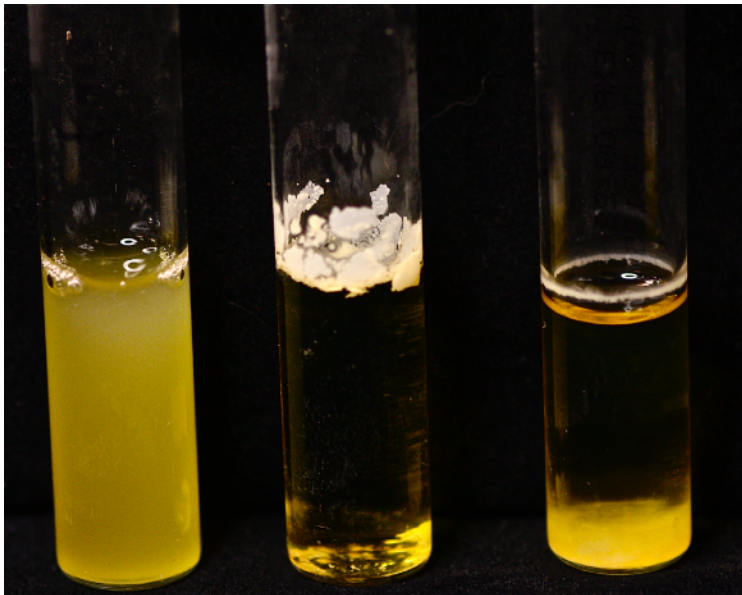
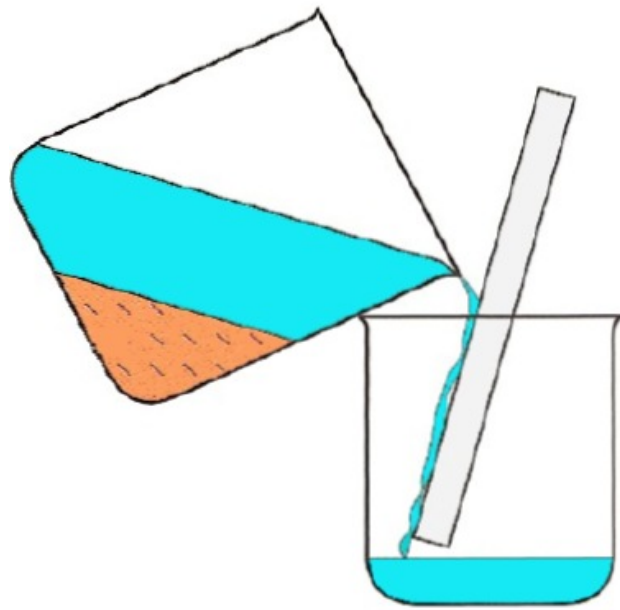




## QIAprep® Spin Miniprep Kit | Protocol for DNA Preparation

1. Pellet 1 ml of bacterial “overnight” culture by centrifugation at  $> 8000$  rpm ( $6,800 \times g$ ) for 3 min at room temperature  
(15 - 25) and discard the supernatant. Then add another 1 ml of overnight culture to the same tube and repeat this centrifugation step.
2. Resuspend pelleted bacterial cells in **250  $\mu$ l Buffer P1**. There should be no cell clumps visible after the resuspension of the pellet in the Buffer P1. **USE “FLICKING” technique to help resuspend cells.**
3. Add **250  $\mu$ l Buffer P2** and mix thoroughly by **GENTLY** inverting the tube 4-6 times until the solution becomes clear. Continue mixing until there is a homogenously colored blue mixture; there should be no localized regions of different shades of blue or brown sections. Let the mixture sit for 4-5 minutes.  
**Do not allow the lysis reaction to proceed for more than 5 min.**
4. Add **350  $\mu$ l Buffer N3** and mix **immediately and thoroughly** by inverting the tube 4-6 times. The solution will be a homogenously colorless solution when properly mixed.
5. Centrifuge for 10 min at 13,000 rpm ( $\sim 17,900 \times g$ ) in a table-top microcentrifuge.
6. Apply the **supernatant** from **step 5** to the **QIAprep spin column** by decanting or pipetting. Centrifuge for 30 - 60 s and discard the “flow-through”.
7. Wash the **QIAprep spin column** by adding **500  $\mu$ l Buffer PB**. Centrifuge for **30 - 60 seconds** and **discard the flow-through**. **Note:** This step is only required when using endA<sup>+</sup> strains or other bacterial strains with high nuclease activity or carbohydrate content.
8. **Wash the QIAprep spin column by adding 750  $\mu$ l Buffer PE**. Centrifuge for **30 - 60 seconds** and **discard the flow-through**.
9. **Repeat Step 8.**
10. **Centrifuge for 1 min** to remove residual wash buffer.
11. Place the **QIAprep column** into a **clean 1.5 ml microcentrifuge tube**.
12. To elute DNA, **add 50  $\mu$ l Buffer EB** (10 mM Tris·Cl, pH 8.5) or water to the center of the QIAprep spin column.
13. Let it stand for 1 min, and centrifuge again for 1 min.

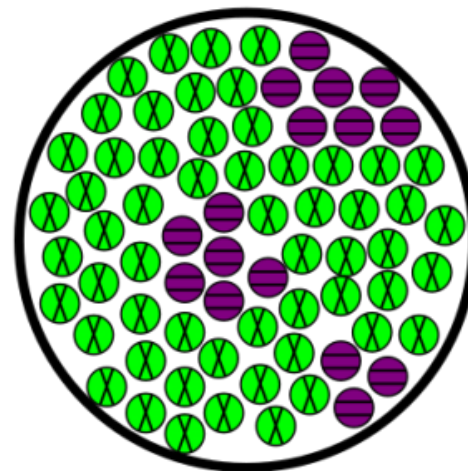
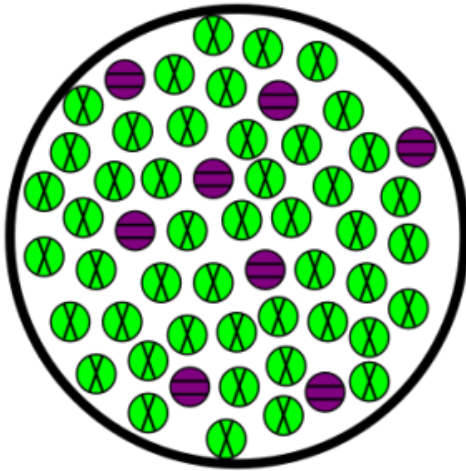
“Decant”



“Flocculant”

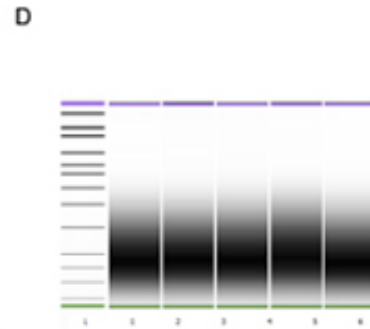
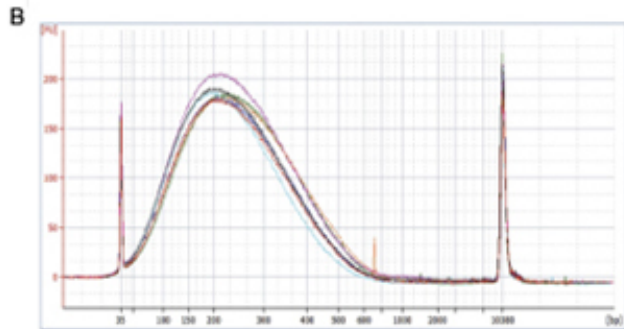
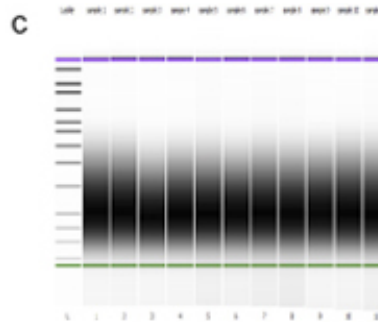
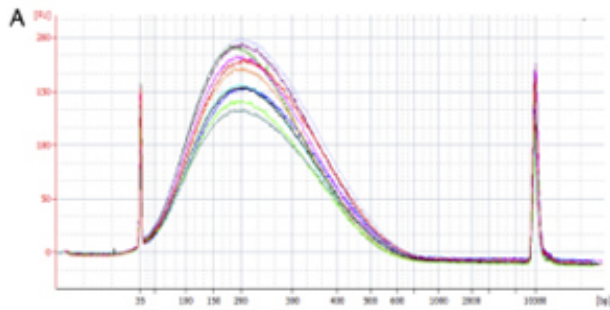
**“Homogeneous” -well mixed suspension...**

Mixtures



Homogeneous Heterogeneous

“Vortex”



“Shear”



## Gel Electrophoresis Preparation

1. Prepare 200 ml 1x TBE buffer from 10x stock (20 ml 10x TBE + 180 ml diH<sub>2</sub>O)
2. Prepare 60 ml of 1% **agarose** with 1x TBE buffer (0.5g agarose + 50 ml 1x TBE). Microwave for 40 seconds two times (not 80 seconds).

Use hot gloves!

3. Once the side of the beaker is able to be touched, the TA will add 6 $\mu$ L of **GelStar**. Once the GelStar is added to the hot agarose, pour the agarose into casting tray and insert comb.
4. While the gel solidifies move onto DNA preparation, but don't forget to come back to the gel once it is solidified and insert the gel tray into the gel running tank. Fill the tank with 1x TBE until the gel is slightly covered.

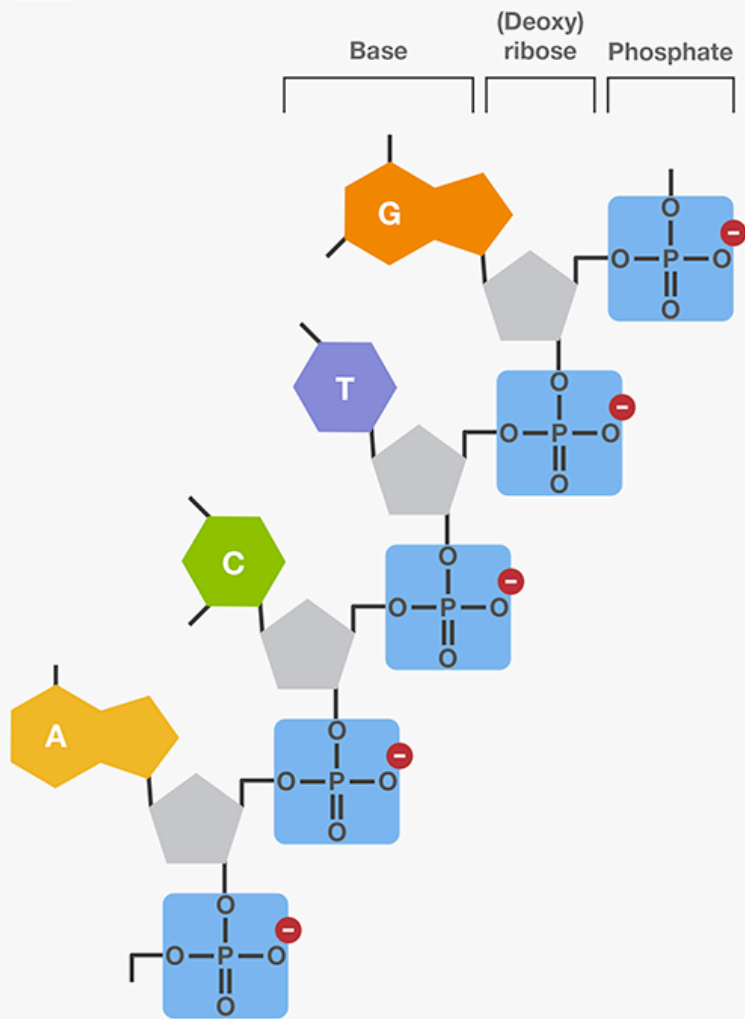
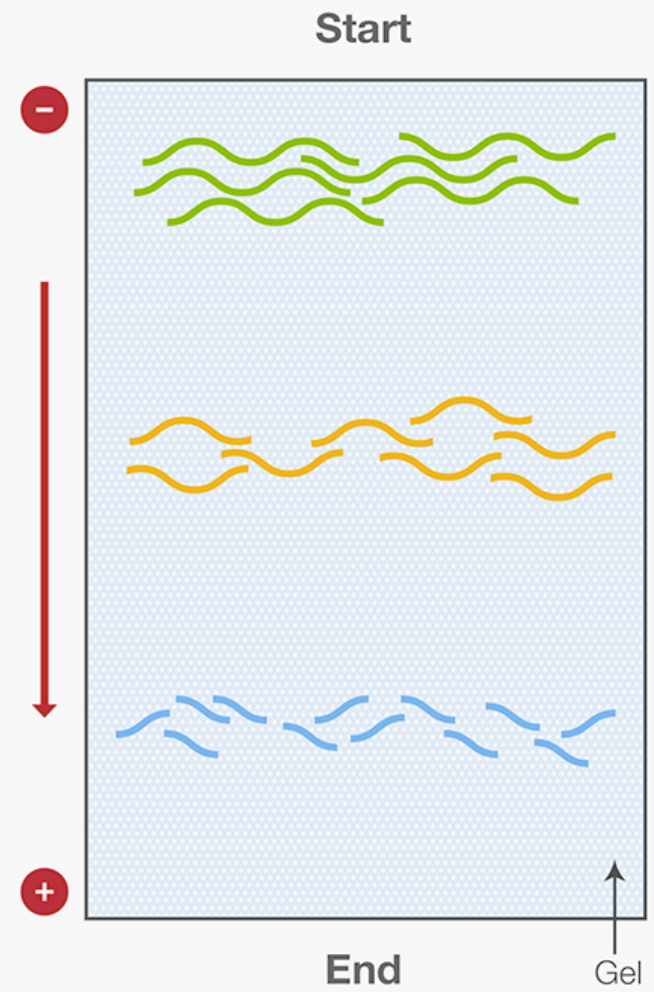
When placing the tray, ensure the comb side is aligned with the negative electrode side (black).

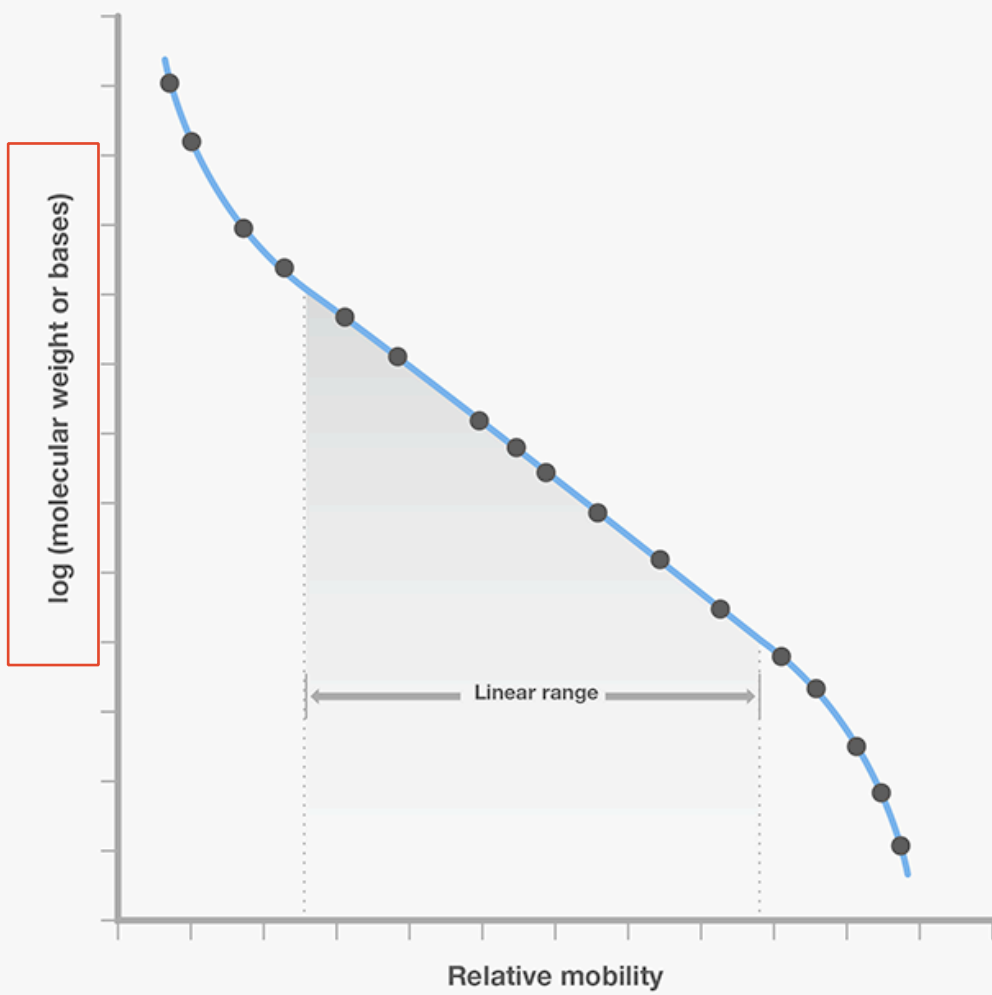
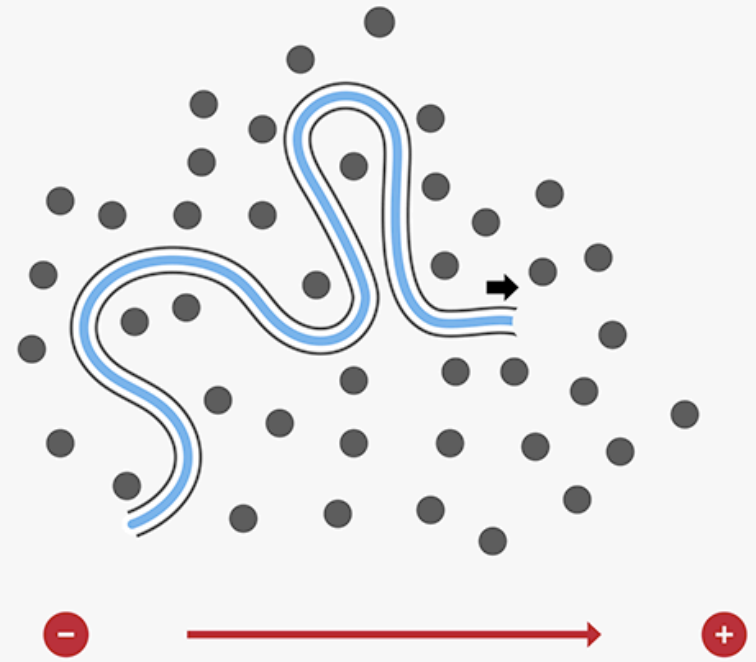
## Gel Electrophoresis

1. Take 5  $\mu$ l of your **DNA sample** and add it to a new microcentrifuge tube. Add 1  $\mu$ l of 6x DNA loading dye buffer.
2. Load 5  $\mu$ l of **DNA ladder** to the first lane and then add your DNA sample +dye to the lane beside it.
3. Start electrophoresis at **80V for 30 minutes** and then **power up to 100V**. DNA will migrate from the negative electrode to the positive electrode. **STOP when the front running dye reaches 1/4" from the "positive" end.**

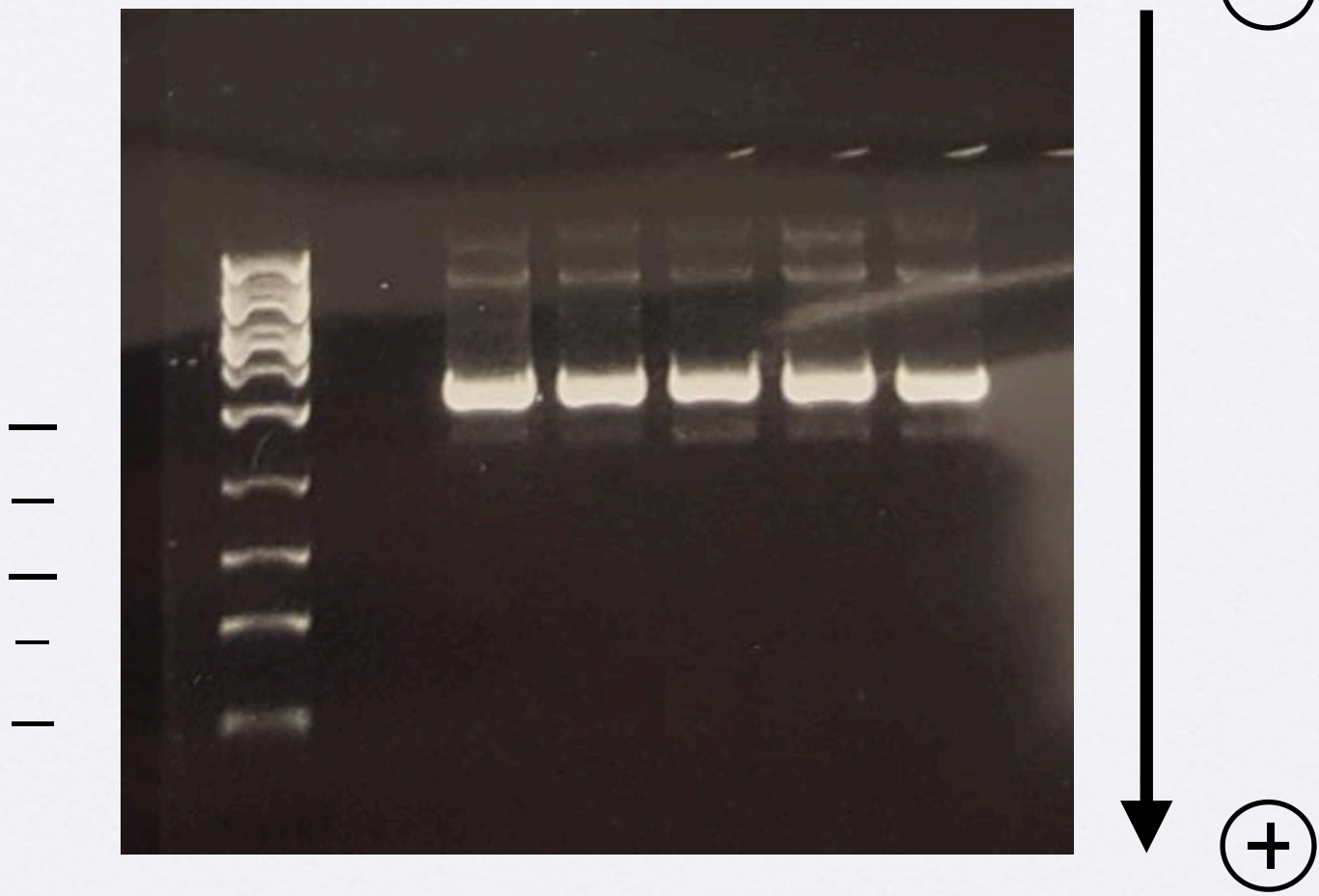
## DNA Concentration Determination

While your gel is running, check your DNA concentration using the NanoDrop spectrophotometer with the TA.

**A****B**

**A****B**

Control 6 7 8 9 10



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INSTRUMENTATION  
CELL PROTEIN and DNA

Personnel / Staff

Lab Technology /  
Training & Access

FAQs

Forms

Operation  
Manuals

Safety Information

"How to" GUIDES



Other, Related  
ABC Core Facilities

## CoreLab Technology Training & Access

Cell, Protein and DNA Core Facilities

### Introduction

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All researchers who would like to undertake research in the Natural Sciences at Georgia State University, and who wish to avail themselves of the research facilities and instrumentation within the **Natural Science Centre** and / or the **Petit Science Centre** must go through an initial series of training sessions to familiarize themselves with fundamental aspects of Laboratory Safety and Security, as well as what equipment is available and how to use it. These training sessions are **PRIMARILY** on-line, but one on training is available **UPON REQUEST** by a member of the **ABC Core facilities**.

Technology Training & Access **COVID-19 CHANGES for SPRING '21'**

### Registration

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## ABCORE FACILITIES

Training Videos -some with, some without audio

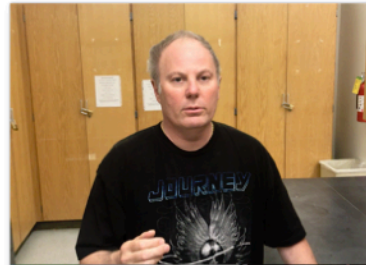
**BASIC TRAINING:** Videos in this section relate to Equipment that are available for "general use" in most Molecular Biology laboratories



### AUTOCLAVES PSC

A brief training video on how to use the AUTOCLAVES in PSC 545/645. For further, more detailed information, please contact -[Chip Foster](#)

[View Video](#)



### AUTOCLAVES NSC 3RD FLOOR

A brief training video on how to use the AUTOCLAVES in NSC 336. For further, more detailed information, please contact -[Chip Foster](#)

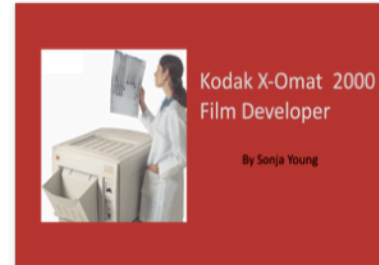
[View Video](#)



### AUTOCLAVES NSC 4TH FLOOR

A brief training video on how to use the AUTOCLAVES in NSC 484. For further, more detailed information, please contact -[Chip Foster](#)

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### FILM DEVELOPER

A brief guide on how to use the FILM DEVELOPER in the ABCore facilities. For further, more detailed, information please contact -[Sonja Young](#)

[View Video](#)

### Centrifuges

- High Speed, floor standing Centrifuge

by [Hyuk-Kyu Seoh](#)



### HIGH SPEED CENTRIFUGES

A brief guide on how to use the High Speed Centrifuges in the ABCore facilities. For further, more detailed information please contact -[Hyuk-Kyu Seoh](#)

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### Centrifuges

- Ultra High-speed Floor-standing Centrifuge

by [Hyuk-Kyu Seoh](#)



### ULTRACENTRIFUGES (FLOOR)

A brief guide on how to use the Ultracentrifuges floor models in the ABCore facilities. For further, more detailed, information please contact -[Hyuk-Kyu Seoh](#)

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### Centrifuges

- Ultra High-speed Floor-standing Centrifuge
- Ultra High-speed Table-top Centrifuge

by [Hyuk-Kyu Seoh](#)



### ULTRACENTRIFUGES (TABLE TOP)

A brief guide on how to use the Ultracentrifuges Table-Top models in the ABCore facilities. For further, more detailed, information please contact -[Hyuk-Kyu Seoh](#)

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### Lyophilizer (Freeze Dryer)



### LYOPHILIZERS

A brief guide on how to use the Lyophilizers in the ABCore facilities. For further, more detailed, information please contact -[Hyuk-Kyu Seoh](#)

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## ABCORE FACILITIES

Training Videos -some with, some without audio

**BASIC TRAINING:** Videos in this section relate to Equipment that are available for "general use" in most Molecular Biology laboratories



### PCR Thermocycler

Sandy Hsieh

#### PCR THERMOCYCLERS

A brief training video on how to use the PCR Thermocyclers in the ABCore facilities. For further, more detailed, information please contact -[Sandy Hsieh](#)

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### GSU ABCore Spectrophotometers

Nanodrop



#### NANODROP SPECTROPHOTOMETER

A brief guide on how to use the Nanodrop in the ABCore facilities. For further, more detailed, information please contact -[Jesse Gardner](#)

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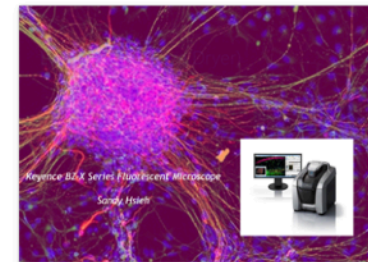
### Cellometer

Sandy Hsieh

#### CELLOMETER

A brief guide on how to use the Cellometer in the ABCore facilities. For further, more detailed, information please contact -[Sandy Hsieh](#)

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Keyence BZ-X Series Fluorescent Microscope

Sandy Hsieh

#### KEYENCE FLUORESCENT MICROSCOPE

A brief guide on how to use the Keyence BZ-X series microscope in the ABCore facilities. For further, more detailed, information please contact -[Sandy Hsieh](#)

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### GSU ABCore Plate Readers

Spectramax iD5 Multi Mode Microplate Reader



#### MD ID5 PLATE READER

A brief guide on how to use the MD iD5 Plate Reader in the ABCore facilities. For further, more detailed, information please contact -[Jesse Gardner](#)

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### Shaker Training

Sandy Hsieh

#### GENERAL SHAKER INCUBATORS

A brief guide on how to use the various flask/tube shaking incubators within the ABCore facilities. For further, more detailed, information please contact -[Sandy Hsieh](#)

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### "How to use" Video Guides



#### VIDEO 8

A brief guide on how to use the " " in the ABCore facilities. For further, more detailed, information please -[John Houghton](#)

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