

PlayMais 3-D DNA Model

*Instructions for the lecture "Data Management in Digital Health"
adapted from (Caine et al. 2015).*

Instructions

Part 1: Constructing the Nucleotides

The purpose of this step is to prepare the main units of the DNA molecule, namely the nucleotides (Figure 1A–C).

- Take a yellow flake (deoxyribose), a blue flake (phosphate group), and a colored flake (nucleobase). Using a damp sponge, wet one end of a blue flake. Stick the wet end of the blue flake onto one end of a yellow flake by gently pushing them together. Keep them in position with your hands for approximately 30 s.
- Once the blue and yellow flakes are stuck together, take the colored flake (i.e., red, orange, white, green) and wet one of the ends with the damp sponge. Stick the wet end of the colored flake onto the side of the yellow flake.
- Repeat these steps until all the DNA units that you will need, according to the reference sequence (see your group sheets), have been prepared. Note: The reference sequence provides information about only one of the two strands. In order to build both strands, it will be necessary to assemble the nucleotides of the reference sequence plus their complementary nucleotides (i.e., for each guanine nucleotide that you assemble, also assemble a cytosine nucleotide and vice versa. For each adenine nucleotide that you assemble, also assemble a thymine nucleotide and vice versa).

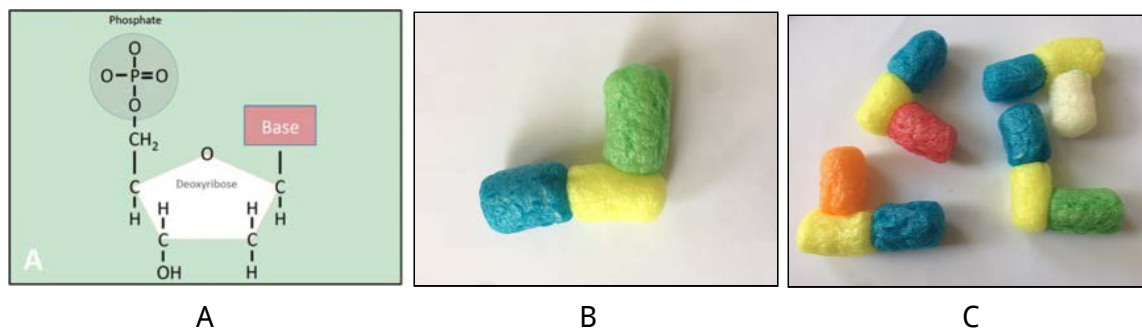


FIGURE 1 PlayMais nucleotides. (A) Chemical structure of a nucleotide. (B) PlayMais DNA units consisting of a phosphate (blue flake), a deoxyribose (yellow flake), and a base (colored flake). (C) PlayMais nucleotides.

Table 1 Flake Color Code

	Flake Color
Adenine base	Red
Cytosine base	Orange
Guanine base	White
Thymine base	Green
Deoxyribose	Yellow
Phosphate group	Blue

Part 2: Building the first Strand

First, one of the two DNA strands will be prepared. In this case, the first DNA strand to be built will be the one reported on the reference sequence. It is better not to make excessively long strands because you will need to twist the double helix while adding the second strand.

- Take the first and the second nucleotides according to the reference sequence (Figure 2A).
- Wet the end of the blue flake of the second nucleotide and stick it onto the yellow flake of the first nucleotide (Figure 2B). Note: In order to facilitate the rotation of the DNA double helix, when you add a nucleotide, this should be slightly rotated compared to the previous one (clockwise rotation) (Figure 2C). The standard conformation of the DNA double helix takes 10 nucleotide-pairs for one complete turn around its axis. In order to achieve this, stick the nucleotides of the first strand with its base rotated 45 compared to the base of the previous nucleotide (Figure 2C). In principle, 36 should be enough ($360 / 10$ nucleotides), but the elastic torsion of the double helix, when completed, will tend to reduce this angle. Therefore, we recommend a rotation of 45 between each nucleotide.
- Continue as described in the first steps for the remaining nucleotides of the sequence (Figure 2D).
- Mark all codons with numbers in a continuous fashion on the coding strand.



A

B

C



D

FIGURE 2 Assembly of the first PlayMais strand. (A) Two PlayMais nucleotides are selected according to the reference sequence. (B–C) The blue flake of one of the two PlayMais nucleotides is wet and stuck onto the yellow flake of the second one, respecting the nucleobase rotation of 45 around the backbone chain. (D) First strand overview.

Part 3: Building the Complementary Strand like a DNA Polymerase

During this step, you will act as if you were a DNA polymerase synthesizing the complementary strand. Using the remaining units, you will add them one by one to the previously built first strand. To do so, you will have to respect the law of base-pairing (Figure 3A).

- The building of the new strand starts from the last nucleotide of the old strand (i.e., the last one that has been added). In this way, the synthesis “direction” of the DNA polymerase will be respected. Moreover, as described in the background section, the two strands comprising the double helix possess an anti- parallel orientation. To respect such a design, the strand’s backbone will start with a blue flake and end with a yellow flake (as done with the old strand).
- Choose a nucleotide that is complementary to the one of the old strand where it will be stuck. Wet the free surface of its colored flakes and stick it onto the colored flake of the corresponding nucleotide (Figure 3A).
- Take the second nucleotide that you want to add and wet both its colored flake and its blue flake. First, stick the blue flake onto the yellow flake of the previous nucleotide, maintaining a right-handed angle of 45 between the two nucleobases (Figure 3B). Second, stick the two colored flakes of the complementary nucleotides (old and new strand) together (Figure 3C). Perform this second step gently, since the elasticity of the torsion will start pulling the two strands apart.
- Continue adding nucleotides until the double helix is completed (Figure 3D). While adding the nucleotides, remember to respect the right-handed rotation (i.e., circa 45) of the newly added nucleotides.

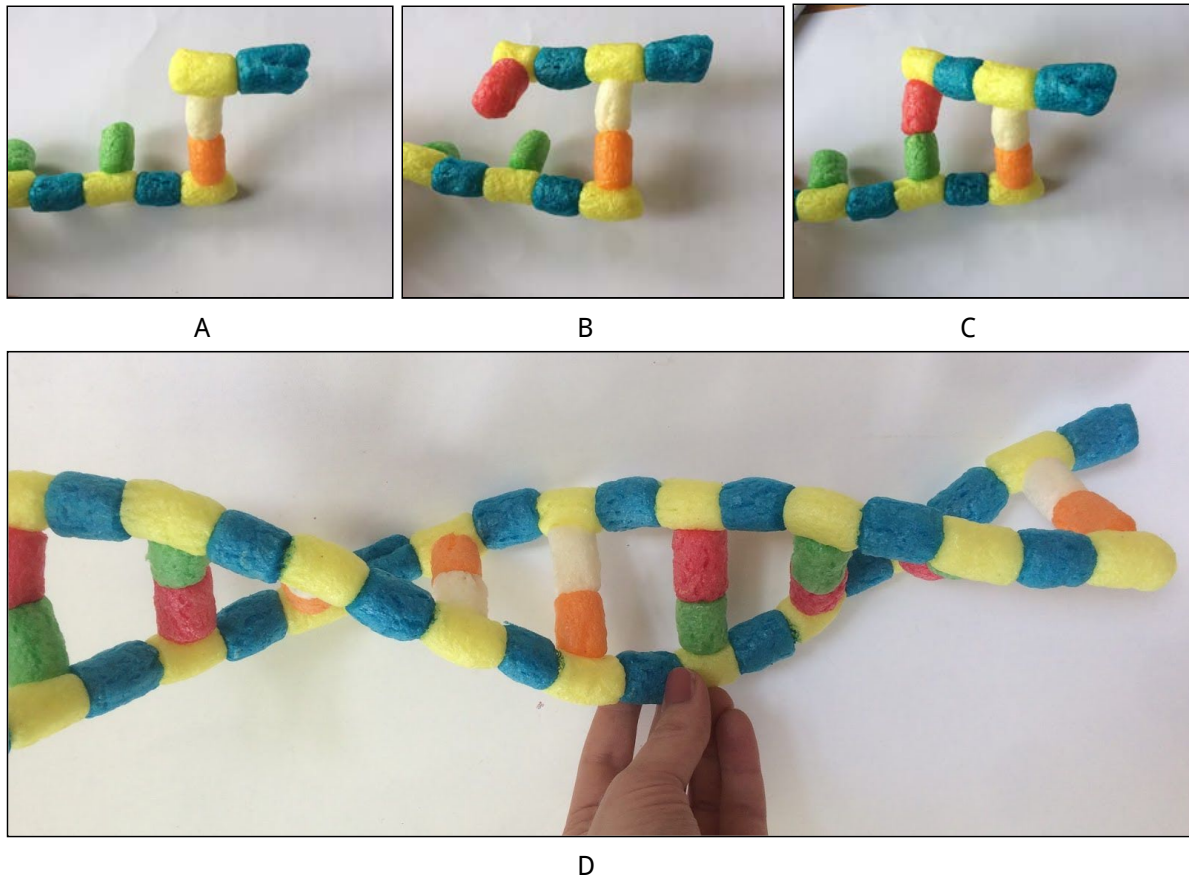


FIGURE 3 Building of the second strand. (A) The first PlayMais nucleotide added is complementary to the PlayMais nucleotide of the old strand. (B-C) The second PlayMais nucleotide is added to the first one respecting the 45 right-handed rotation. (D) Completed first part of the double strand.

Part 4: Connect all individual Parts and Decipher the Amino Acid Sequence Resulting from Transcription and Translation

- To do so, wet the blue flakes of each fragment and stick them onto the corresponding yellow flakes from the other fragment.
- Identify the Start-Codon in sequence 1.
- Transcribe and translate the sequence according to the translation process as described in the background information and using the RNA codons as given in Figure 6. Please use the one-letter amino acid code.

Part 5: Introduce Mutations/DNA modifications

- Introduce the mutation/DNA modification as given on the team sheets at an appropriate position within the region you created
- One person of each group explains the given mutation, how and where it was introduced, and the effects on the corresponding amino acid sequence to the rest of the class

Background Information

DNA is the biological medium that carries the genetic information in all living organisms. It can be seen as a very long right-handed spiral ladder made up of two biopolymers (complementary DNA strands) that together form a double helix structure. The chemical structure of the two complementary strands displays opposite direction such that they are antiparallel to each other. As a result, at one end of the DNA molecule, one strand will expose a chemical structure (i.e., a phosphate group), whereas the other strand will not. Each DNA strand consists of repeating fundamental units called nucleotides. Nucleotides are composed of a phosphate group, a deoxyribose (monosaccharide sugar), and a nucleobase (simply called base). Sugars and phosphates form the backbone of each strand, whereas four different bases - adenine (A), guanine (G), cytosine (C), and thymine (T)—hold the two DNA strands together. To do so, each base specifically forms hydrogen bonds with a base from the other strand. In detail, an adenine will always pair with a thymine, whereas a guanine will always pair with a cytosine. This complementarity is essential for the double helix arrangement of the DNA molecule in space and is the basis of faithful DNA replication prior to cell division (Watson and Crick 1953).

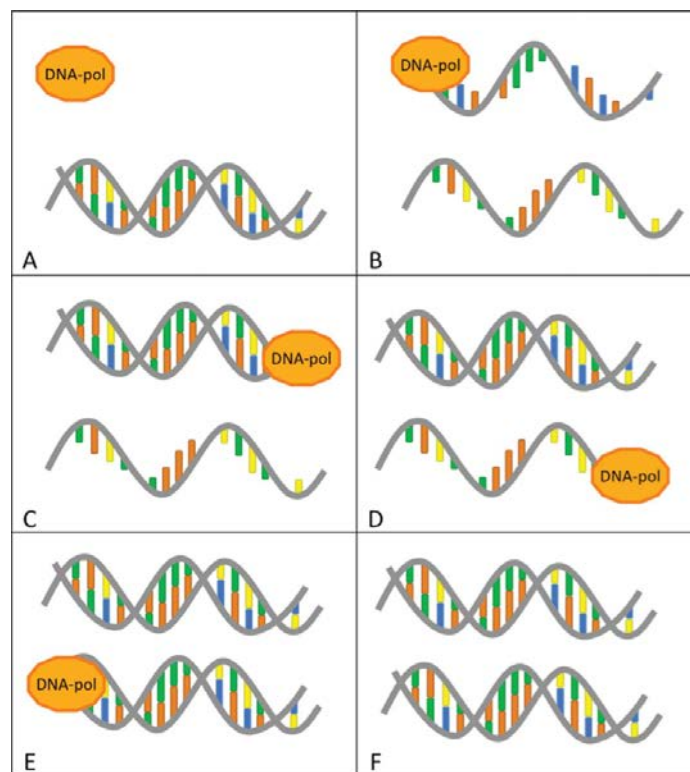


FIGURE 4 DNA replication. (A–F) A schematic representation of the DNA replication process. Phosphate-sugar backbones are represented as gray strands. Nucleotides are depicted using the following color code: adenine-green, thymine-orange, guanine-yellow, cytosine-blue. (B–C) The first of the two parental strands comprising the original DNA double helix is used as a template to synthesize the complementary strand. (D–E) The second parental strand is then used to synthesize a second complementary strand. (F) At the end of the process, two identical DNA double helix molecules are produced.

The order of nucleotides comprising a DNA sequence is not random. Specific sequences of nucleotides, called genes, encode fundamental information for cell function. Moreover, within a single cell, thousands of genes act coordinately towards a common purpose: to support the organism's life. Therefore, it is crucial that these instructions are transmitted from one generation to the next when cells divide. DNA-polymerase is the cellular machinery that fulfills this function (Figure 4A). It replicates the DNA molecule using each strand of the parental molecule as a template to build a new complementary strand (Figure 4B–E). In this way, the newly synthesized strand will respect the nucleotide sequence of the one it is replacing, and two new DNA molecules will be formed at each replication round (Figure 4F). Due to this old/new strand double helix synthesis, the process is called semiconservative replication. The replication process ensures that instructions are preserved and transmitted to subsequent generations. These DNA-encoded instructions allow the cell to build the main components of its cellular environment: proteins. To do so, the first step consists in a process called transcription (Figure 5A - D). The RNA-polymerase transcribes the gene sequence into a messenger molecule called ribonucleic acid, RNA (Figure 5C–D). Like DNA, RNA is made up of nucleotides and is synthesized using as a template the sequence of the gene from which it is transcribed (Figure 5D). The instructions contained within the RNA molecule are then used for protein synthesis. This second step performed by macromolecular machines (i.e., ribosomes) is called translation (Figure 5E–I).

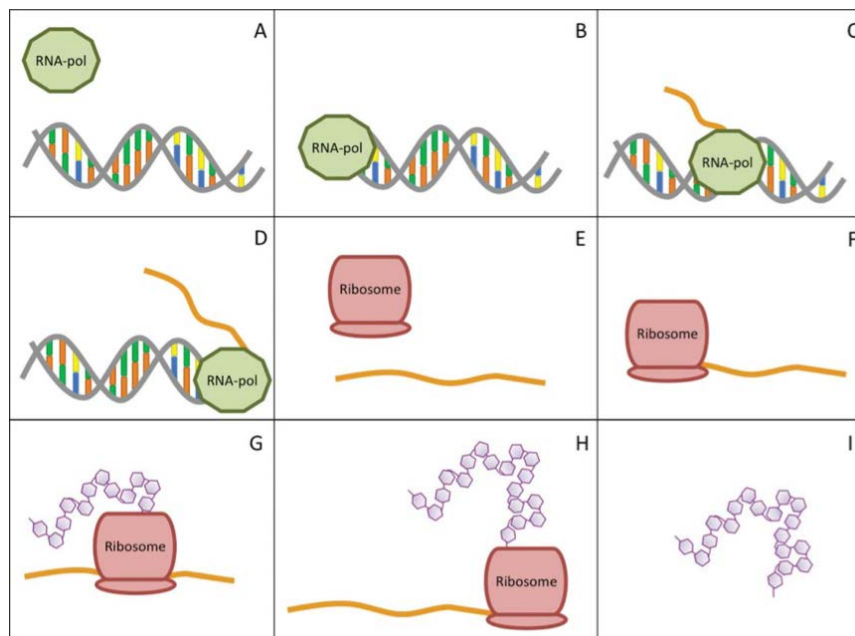


FIGURE 5 Transcription and translation. (A–D) The RNA-polymerase locates the gene within the DNA double helix and transcribes the sequence into a messenger RNA molecule (orange strand). (E–I) The ribosome translates the information contained within the RNA molecule into the corresponding protein (hexagonal sequence strand).

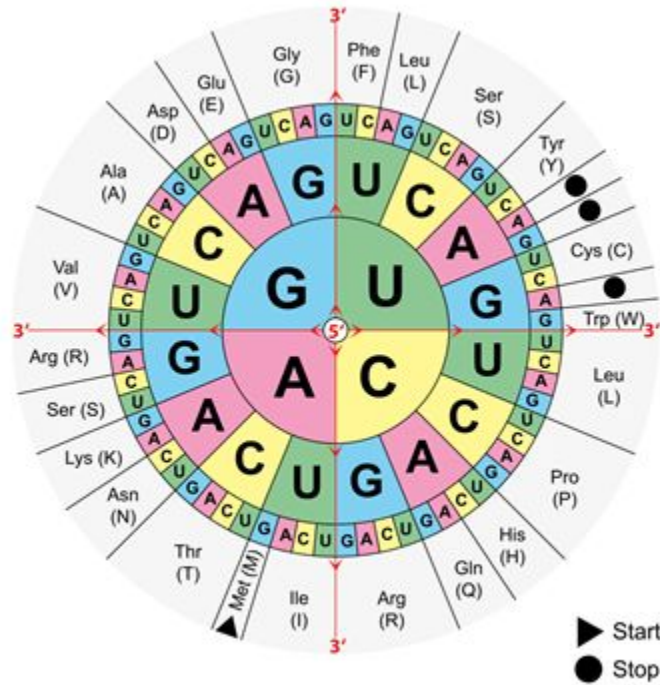


FIGURE 6 RNA Codons and how they translate into amino acids. One letter code of amino acids is given in brackets.

Works Cited

Caine, Massimo, Ninon Horié, Sandrine Zuchuat, Aurélie Weber, Verena Ducret, Patrick Linder, and Karl Perron. "A 3D-DNA Molecule Made of PlayMais." *Science Activities: Classroom Projects and Curriculum Ideas* 52.2 (2015): 31-44. Print.

Watson, J. D., and F. H. C. Crick. "Molecular Structure of Nucleic Acids: A Structure for Deoxyribose Nucleic Acid." *Nature* 171.4356 (1953): 737-38. Print.