

HIGH SCHOOL GENETICS CURRICULAR MATERIAL

This science curricular material was developed by Tanya Manternach as part of the NSF-funded RET summer internship with the Iowa Turtle Army in Dr. Nicole Valenzuela's Lab at Iowa State University, and is intended for use by other peer teachers with proper acknowledgement. Funding was provided in part by NSF grant IOS 0743284 and IOS-0924290 to Nicole Valenzuela.

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Fig. 4 RNA. Image courtesy of Ophardt 2003.

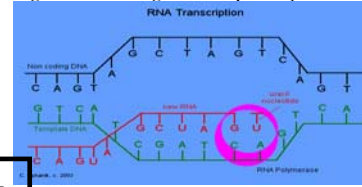
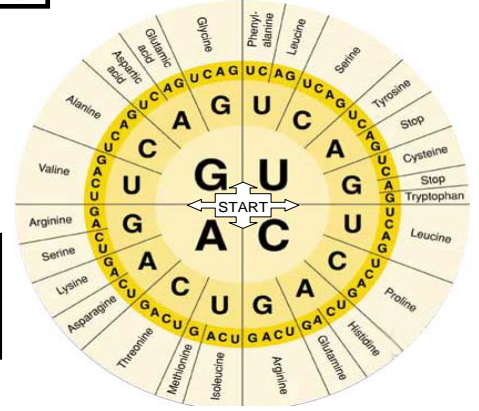


Fig. 3 Amino Acid - Codon Key. (Image modified from Biology. Glencoe/McGraw-Hill, 2006.)



7. Cut the hydrogen bonds that hold the nitrogen bases of your classmate's DNA strands together and copy the DNA codons from strand one below. Transcribe a complimentary strand of messenger RNA (mRNA) as shown in fig. 4. Note that RNA substitutes the nitrogen base, Thymine (T) with Uracil (U). Record the DNA and mRNA code below.

DNA code: (from strand 1)	---	---	---	---	---	---	---
	codon 1	codon 2	codon 3	codon 4	codon 5	codon 6	codon 7
mRNA: compliment	---	---	---	---	---	---	---
	codon 1	codon 2	codon 3	codon 4	codon 5	codon 6	codon 7

8. Copy the DNA code and mRNA you transcribed into part two on the attached form and exchange it with a different classmate.
9. Translate your classmate's code into amino acids using the key in fig. 3 to form a protein. Record the amino acid chain you translated:

mRNA:	---	---	---	---	---	---
	codon 1	codon 2	codon 3	codon 4	codon 5	codon 6
amino acids:	a.acid 1	a.acid 2	a.acid 3	a.acid 4	a.acid 5	a.acid 6

10. Cut out the protein (chain of amino acids) from question 9 and color the amino acids of the protein according to the chart below. Use the starting letter of the amino acid's name to determine which color to use.

First letter of the A. Acid	A	C	G	H	I	L	M	P	R	all other
Color	Red	Green	Blue	Yellow	White	Orange	Purple	Pink	Black	Brown

11. Proteins perform different functions in your body. Their function is determined by their shape and their shape or conformation is determined by the different amino acids. Fold your protein into its correct conformation by following the rules below:

Color	Shape formation
Red	Bond two adjacent red boxes together with glue or tape
Orange	Fold the box like an accordion
Blue	Fold the box up to make a 90 degree angle
Green	Fold the box down to make a 90 degree angle
Yellow	Twist the box
Pink	Fold in half
All other colors	Leave unchanged

12. Return the protein to the original creator of the message. Once you receive the protein made from your original message check to see if it was assembled correctly by reviewing the steps. Errors in the transcription or translation of the message could have generated variations in the conformation of the protein. However, some errors may not have generated different variations in the protein conformation. Explain.
13. As mentioned earlier, the conformational shape of the protein allows the protein to perform different jobs in the body. If the shape is faulty, the protein cannot work properly. This may lead to genetic disorders. For example, if hemoglobin proteins are not shaped properly they will not be able to carry oxygen or may alter the shape of the red blood cells that carry these proteins causing sickle-cell anemia.

Variations in the protein conformation generated by error are known as mutations. Some mutations may not produce any noticeable affects while others could be deadly. However, sometimes the mutation may improve organism's fitness and become a helpful, survival adaptation that may lead to evolution.

Scientists, such as Dr. Nicole Valenzuela and her team of Ph. D and post doctoral students are studying the genes found in different populations of turtles within the Ecology, Evolution and Organismal Biology Department at Iowa State University to try to determine how DNA transcription and translation are regulated in order to determine the amount and type of proteins produced to control different physical and chemical processes within organisms. Understanding how these genes are regulated can help scientists understand how and why organisms develop differently. Studying the genes within or among species may provide clues to understanding evolutionary adaptations.

Her research team samples the amount of mRNA transcribed during different development stages of turtle embryos in order to see which genes are responsible for different developmental processes at that time, such as determining the sex of the turtle. She identified differences in gene expression between turtle species whose sex is predetermined by their genes (GSD) and turtles whose sex is determined by environmental temperature (TSD). She compares the amount of mRNA being expressed from the six major sex determining genes: Wt1, Sf1, Sox9, Dax 1, Dmrt1, and Aromatase during the sex determining stage of embryonic development for both GSD and TSD turtles to identify differences generated by gene expression and induced by incubation temperature. Genes important to determine sex in a TSD turtle should have high expression at a male producing temperature but low expression at a female producing temperature (or viceversa), but there should be no difference in expression by temperature in the GSD turtle since its sex is not influenced by the environment. To do this she collects the fragile mRNA being generated by the embryo during the sex determining stage and used it to produce a stable DNA copy (cDNA) which can be measured using a technique, called Quantitative Polymerase Chain Reaction (QPCR). The number of PCR cycles needed to bring the sample above a threshold visible to fluorescent microscopy is measured to determine the amount of gene expression. Genes that were most active (produced more mRNA) during this stage of development were thought to activate the sex determining stage of TSD turtles and could be compared to the amount and type of genes being expressed in GSD turtles of the same embryonic stage. Study fig. 4. Which genes do you think took less PCR cycles to reach threshold?

14. Which genes are thought to activate the temperature sensitive period in TSD turtles because they show the greatest expression leading up to the temperature sensitive period of embryonic development for sex determination in TSD turtles according to fig. 4?

15. These genes were not differentially active in GSD turtles. What could this suggest?

16. Dr. Valenzuela hypothesizes that Wt1 acts as a master switch gene used to activate the temperature sensitive period in TSD turtles by turning on the Sf1 gene prior to the onset of the sex determining stage. She suspects that Wt1 is responsible for TSD and is also present in GSD turtles, but the mechanism it uses to stimulate Sf1 is inactive since SF1 cannot activate the TSP in GSD turtles, which allows their genotype to determine the sex. Does this suggest that GSD evolved after the TSD in turtles if they both have the gene, but that it is vestigial (no longer activates the Sf1 gene) and cannot activate the temperature sensitive sex determination in GSD turtles allowing their genes to determine their sex instead of temperature?

17. Studying how regulatory gene networks differ among TSD and GSD turtles helps scientists understand how the genome of turtle populations can be affected by ecology. The potential selection for sex phenotypic plasticity or flexibility acting at the individual and population level favors the evolution of a regulatory system of sexual development that enables the embryo to assess its environment and differentiate into the sex that will attain the maximum potential fitness given the external conditions [Charnov and Bull, 1977; Bull, 1984; Valenzuela, 2004]. TSD may allow eggs to develop female at female optimal temperatures and male at male optimal temperatures. However, Valenzuela's research indicated that some level of temperature-sensitivity exists and is vestigial in GSD turtles. What does it tell us about gender plasticity (not having a fixed gender assignment predetermined by genetic makeup) of reptiles if it is vestigial in soft shell turtles demonstrating GSD?

Fig. 4. Genes expressed in TSD turtles. Courtesy of Valenzuela 2009.

