

Name: _____

The Mystery of the Crooked Cell

Investigating the Heredity of a Blood Disorder

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Dr. James Herrick of Chicago has just received a patient who he thinks may have a blood disorder. This patient feels well the majority of the time though has reported specific recurring health problems since his childhood. In one instance, the patient reported feeling so tired after a short swim that he could hardly move. He was unusually short of breath, had extreme pain in his joints and muscles, a yellowish tint in the whites of his eyes, and tenderness in his abdomen. Consequently, the patient had to spend two weeks on bed rest.

A look at his immediate family history reveals that neither of his parents have had similar symptoms. However, upon further investigation, it is revealed that the patient's paternal uncle and grandmother shared the same symptoms under the same circumstances. Based on this medical history, Dr. Herrick believes the patient may have sickle cell anemia.

Sickle cell anemia is a hereditary disease that is passed down to an individual from their parents. The disease causes red blood cells to form into an abnormal crescent shape which prevents the cells from efficiently carrying oxygen to the body's tissues. This crescent, or sickle, shape is due to a mutation in the gene that codes for a protein in red blood cells called hemoglobin, the component of red blood cells which binds to oxygen. In particular, the mutation leads to a change in one of the amino acids in the hemoglobin protein.



Dr. Herrick has asked for your help in confirming his diagnosis. To do so, you will be testing the patient's hemoglobin protein using gel electrophoresis. Based on your results, you can then confirm the patient's diagnosis.

What is heredity?

MATERIALS

Patient's hemoglobin	Micropipette and tips
Normal hemoglobin	Electrophoresis equipment
Sickle hemoglobin	Microcentrifuge

PART I - Agarose gel preparation for protein electrophoresis

- 1. Locate the clear electrophoresis tray and white comb on the laboratory bench.
- 2. Insert the white comb into the tray to form the wells. There are two different slots in the tray for the comb. Be sure to use the slot closest to the end of the tray.
- 3. Remove the cap from the agarose gel tube, and pour its entire contents into the tray. Allow the gel 15-20 minutes to solidify.

PART II - Sample preparation & protein electrophoresis

- 4. Locate the following samples in your colored tube rack, and, along with the instructor, fill in the following chart:

Sample	What is the label on the tube?	Well number
Normal Hemoglobin		
Sickle Hemoglobin		
Patient Hemoglobin		

- 5. Centrifuge each sample for two (2) seconds to collect the contents at the bottom of the tube. Make sure you balance your centrifuge.

QUICK CHECK: Compare the hemoglobin samples to each other. Can you tell a difference between them?

- 6. Load 15 μ L of each sample into the well that it was assigned to in the chart above.
- 7. Locate the electrophoresis box, and place your sample into one of the six gel slots. The gel should be completely covered by the electrophoresis buffer. Place the lid on the box, ensuring that the black wire is on top.
- 8. Run the gel at 220 volts for at least 10 minutes.

- 9. Hemoglobin is a protein that has a red color. You will be able to see how far your hemoglobin samples have migrated through the agarose gel. Fill in the example below with your MdBioLab instructor.

Write which sample is in which well

Wells →

— Write what the charge is closest to the wells.

Draw an arrow on this line to show what direction the hemoglobin protein will travel in the agarose gel.

— Write what the charge is closest to the bottom of the agarose gel.

Why does normal hemoglobin move at a different speed than sickle hemoglobin?

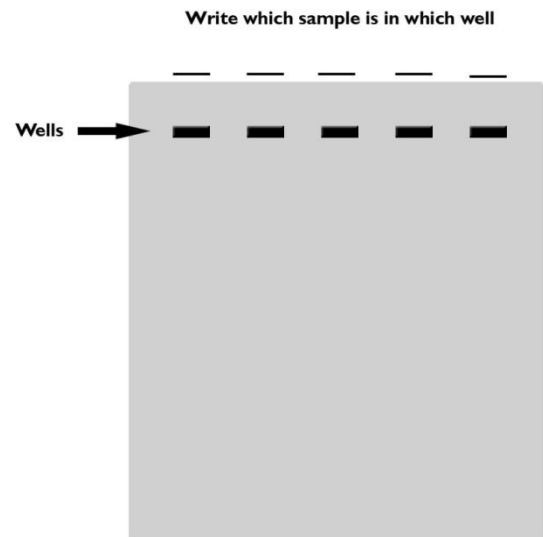
How will you use your patient's results to determine if they have sickle cell disease?

Since neither of the patient's parents have sickle cell disease, is it possible that the patient could have it?

Support your theory with a Punnett square which depicts the patient's possible genotypes:

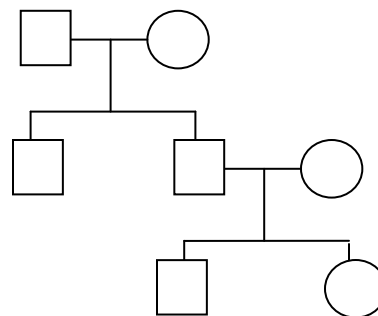
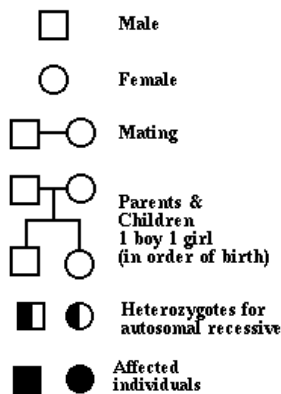
- 10. Draw your results in the diagram, then fill in the patient's genotype and phenotype in the chart below.

Genotype	Phenotype



PART III – Data analysis

Genetic counselors use pedigree charts to show the occurrence and appearance of genotypes and phenotypes within a family. Recall the patient's family history as described on page 1 and his genotype as determined from the analysis of his hemoglobin protein; using the pedigree below, attempt to trace the occurrence of sickle cell disease within his family.



Patient

PART IV - Conclusion

It's known that the patient's sister does not have sickle cell anemia, though her genotype is unknown. Considering that the patient's sister is a serious athlete, do you think she should be tested for the sickle-cell trait?