Potential Connections Between Sickle Cell Disease and Iron Deficiency Anemia

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Abstract

Sickle Cell Disease (SCD) is a common genetic disease which is an inherited red blood cell (RBC) disorder. SCD affects a protein called Hemoglobin causing it to become abnormal. Many RBCs contain Hemoglobin which is responsible for transporting oxygen throughout the blood. Due to the abnormality of the Hemoglobin protein, the RBC loses its round shape and forms a C-shape causing the blood to become hard and sticky. This may lead to other health problems such as infection, acute chest syndrome, and stroke. According to the Centers for Disease Control (CDC), about 1 in 365 African Americans are born with SCD and 1 in 13 African Americans are born with the Sickle Cell Trait (SCT). SCT is when a child inherits a mutated hemoglobin gene from one parent and a normal gene from the other. Usually, no signs are detected, but in rare cases health problems may develop.

Anemia is also another very common red blood cell disorder that results from the lack/dysfunction of the RBC within the body. This disorder has a low amount of Hemoglobin which can lead to reduced oxygen flow to some of the body's organs, causing destruction and low production of the RBC, along with blood loss. The lack of healthy red blood cells causes Sickle Cell Anemia (SCA). Also, Iron Deficiency Anemia can develop through some blood loss, genetic, and/or heavy menstrual periods.

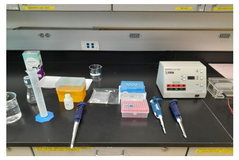
In this study, we will investigate the restriction enzyme, Dde I, which cleaves normal human DNA at the position corresponding to amino acid number 5 of the beta-globin chain. This will discriminate between HbA (normal) and HbS (disease) genes. We will collect and analyze samples from a hypothetical small family cohort (n= 3) to evaluate the presence or absence of the affected genes and subsequent disease. We aim to investigate, firsthand, Medelian genetics and molecular biology to gain skills in preparation for the molecular detection of disease, in space, as well as forensic science on Earth.

Objectives

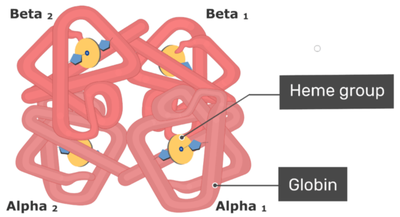
- Explain SCD
- Explain Iron Deficiency Anemia
- Conduct a molecular genetics experiment
- Analyze and interpret the results

MATERIALS AND METHODS

- Ready-to-Load QuickStrip™ DNA Samples
- UltraSpec-Agarose™,
- Electrophoresis Buffer (50X)
- Practice Gel Loading Solution
- FlashBlue™ DNA Stain.
- DNA Eletrophoresis
- Micropipettes
- White Light Box
- Microwave or Hot Plate



Hemoglobin Protein



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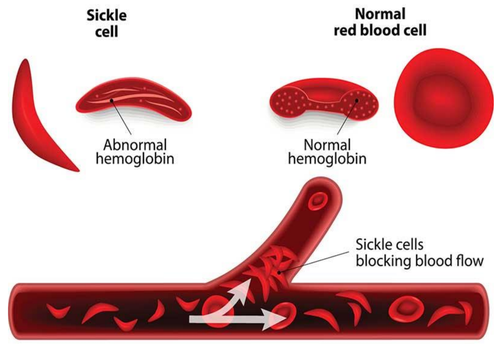
Effect of Hemoglobin Mutation on RBCs



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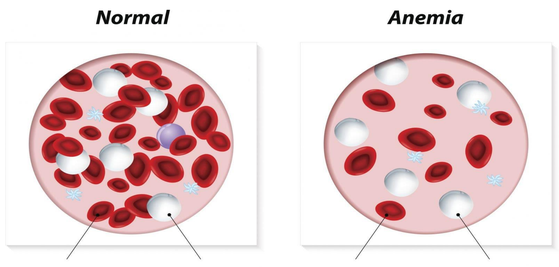
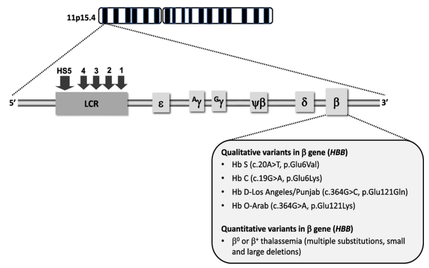
Anemia RBC



https://www.google.com/url?sa=i&url=https%3A%2F%2Fwww.drugs.com%2Fhealth-guide%2Fanemia.html&psig=AOvVaw335H-BzaPMKYde2MzDotNp&ust=1682107556453000&source=images&cd=vfe&ved=0CBAQjRxqFwoTCLj48Iyhuf4CFQAAAAAdAAAAABAE

Restriction enzyme that discriminates between HbA (normal) and HbS (disease) genes



https://www.researchgate.net/figure/Genetic-and-molecular-basis-of-sickle-cell-disease-SCD-is-caused-by-mutations-in-the-b_fig1_358990385

Restriction enzymes are endonucleases which catalyze the cleavage of phosphodiester bands with both DNA strands. During digestion, the DNA results are cut into 4 to 8 base pair in length, leaving different size DNA fragments for each individual test.

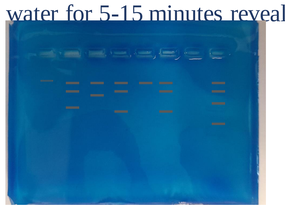
Background

Sickle Cell Disease (SCD) is a common genetically inherited red blood cell (RBC) disorder. SCD affects a protein, Hemoglobin, causing it to become abnormal. Hemoglobin proteins are within Red Blood Cells (RBC) which travel throughout the body. The circulatory system is designed to help deliver oxygen and nutrients to our cells and dispose of waste, which is carried out, in part, by RBCs. SCD affects the Hemoglobin S (HbS) protein by mutations of the β globin gene that can be located within chromosome 11. HbS results from the replacement of glutamic acid by valine in the sixth position of the β-globin chain of hemoglobin. (Brandow, A.M &Leim, R.I., 2022). This causes the RBC to lose its normal morphology and forms a C-shape, resembling a sickle, also causing the RBC to become hard and sticky. This effect may take up to 5 months after birth for symptoms to occur which can cause acute or chronic pain and occlude blood flow. The pain can be localized in areas such as the legs, arms, and back, due to infarction. Ultimately, SCD can lead to major health problems such as infection, acute chest syndrome, stroke, and other end-organ damage.

Iron Deficiency, which is a lack of iron, but in some cases may also be also caused by SCA due to the lack of Hemoglobin which helps with the transportation of iron within the RBC. Iron deficiency anemia (IDA) is when the hemoglobin containing iron decreases in the blood. More than 2 billion individuals worldwide suffer from IDA, and it is still the leading cause of anemia. (Alzahrani et al. BMC). Iron Deficiency Anemia can also be developed through blood loss, genetic, and/or heavy menstrual periods.

Experiment

For our experiment, we carefully located the Beta-globin gene that is found on chromosome 11 and detect the allele that interfere with the body's ability to create hemoglobin. Using the micropipette, 15 µl of each sample was loaded into Agarose gel and placed into electrophoresis buffer within the electrophoresis chamber. After running the samples for 30 to 35 mins at 150V, the gel was then stained in a Flashblue stain solution for 2-3 minutes and rinsed in warm water for 5-15 minutes revealing results below.



Lane 1: Control Sample w/ SCD
Lane 2: Control Sample w/ SCT (Heterozygous)
Lane 3: Control Sample w/ normal gene (Homozygous)
Lane 4: Mother DNA Sample
Lane 5: Child DNA Sample
Lane 6: Father DNA Sample

Results

Lane 1: Pattern has one band due to no restriction enzyme digestion of either band.

Lane 2: Pattern has three bands due to containment of restriction enzyme to one of the pair of genes, (the normal gene), that has been cut to produce small pieces of DNA. The second copy of the gene show signs of mutation and could not be cut into two pieces.

Lane 3: Both genes shown restriction enzyme and are digested into smaller DNA band.

Lane 4: Mother is heterozygous and has Sickle cell trait(SCT). Pattern has three bands due to containment of restriction enzyme to one of the pair of genes, (the normal gene), that has been cut to produce small pieces of DNA. The second copy of the gene show signs of mutation and could not be cut into two pieces.

Lane 5: Child shown to have SCT and Sickle cell anemia (SCA) due to Pattern has one band due to no restriction enzyme digestion of either band.

Lane 6: Father is heterozygous and has SCT. Pattern has three bands due to containment of restriction enzyme to one of the pair of genes, (the normal gene), that has been cut to produce small pieces of DNA. The second copy of the gene show signs of mutation and could not be cut into two pieces.

Conclusion

In conclusion, even though 5 million individuals are currently affected with SCD worldwide, the only potential connections we were able to link IDA and SCD is loss of Hemoglobin within SCA. Also, the results of our experiment further explained how Heterozygous individuals with the dominant normal gene can mask the presence of the disease that cause the mutation, leaving the offspring positive for SCA and SCT.

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