Old Dominion University/BIOL294-Genetics/Rinehart-Kim/Romanov Project

Internet Resources

You may use other Internet sources here, but please cite any sources that you use unless they are one of the following.

http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0004838 (You should be able to access the entire article. You may need to copy and paste the site address.)

http://www.ncbi.nlm.nih.gov/pubmed/20557352 (You won't be able to access the entire article, but the abstract will give you important information.)

http://www.nature.com.proxy.lib.odu.edu/ng/journal/v9/n1/pdf/ng0195-9.pdf (Please note that this is a PDF of the article and is provided via the ODU Library.)

Question Allocation:

Teresa Hooker: #1-13 + #42 Finished
Tanatswa Sambana: #14-23 +#42 Finished
Savannah Crew - #24-32 + #42 - Finished
Danielle Wolcott #33-41+#42 - Finished

History

1. Nicholas II was the last Romanov to hold power in Russia. What was his title?
   Tsar

2. How long had the Romanov family been in power in Russia?
   About 300 Years

3. Politically, what happened to Nicholas II?
   The Bolshevik Revolution

4. Who took control after Nicholas II abdicated the throne?
   The Bolsheviks

5. What happened to Nicholas II and his family after he abdicated the throne?
   They were executed

6. One of the reasons that the family of Nicholas II was executed (vs. just imprisoned) was because there was a fear that the White Russian Army would save them. Who was the White Russian Army?
   People who fought against the Bolsheviks in the Russian Civil War

Hemophilia

The pedigree chart found at the end of this assignment comes from the Module powerpoint lecture notes.

7. How was Nicholas II wife, Alix, ***genetically*** related to Queen Victoria of England?
   He was married to her Great-Grandaughter

Both Queen Victoria and Alix are designated as being carriers for hemophilia.

8. In a couple of sentences, describe the physiology of the disease hemophilia.
   With Hemophilia, the blood is unable to clot because a mutation in the gene that codes the clotting factor. This could mean the clotting factor could not work properly, or they could not have it altogether. (Yes, I know it is severe bleeding because the blood cannot clot. But WHY can't the blood clot? Be *very* specific.)

9. What does it mean to be a carrier for a disease?
   A carrier is someone who is able to pass on the genetic mutation for a disease, whether it is active in themselves or not.

10. What type of hemophilia (A or B) is (probably) represented in the pedigree chart?

Type B

11. On what chromosome is the gene that, when mutated, causes hemophilia?

The X chromosome

12. Describe the mutation (at the molecular level) that apparently caused hemophilia in Alix, (and probably all of the European families that had hemophilia). Be *very* specific.

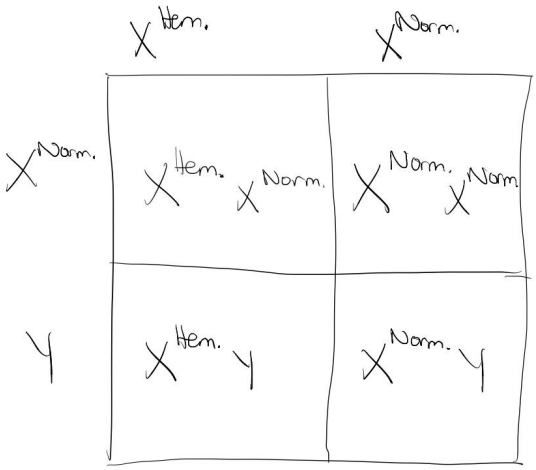
This Mutation is called a Substitution Mutation. TThis occurs when one base is swapped with another between parent chromosomes. In this case, the mutation for hemophilia is on exon 4, gene F9 on the X chromosome.

13. How could the mutation you described in #12 result in a faulty gene product. Be *very* specific in your description.

This could be a faulty gene product because the substitution of the mutated gene segment caused a change in the amino acid and protein expressed.

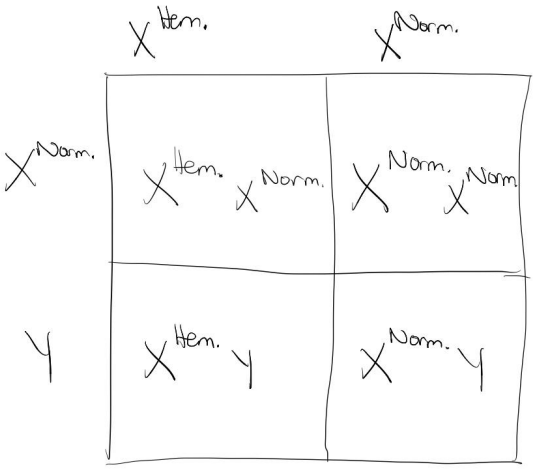
14. The Romanov's son, Alexis, had hemophilia. Describe how Alexis genetically acquired hemophilia. (Use a Punnett square. You can either draw a table or line up the genotypes.)

Nichols was a normal male who had no hemophilia, and Alix was a carrier of hemophilia. Nichols had a Y chromosome which he gave to Alexis which made him a male, and Alix gave the mutated X chromosome to Alexis. The XY combination is the reason why Alexis had hemophilia.



15. Using a Punnett square (again, draw a table or line up the genotypes), explain why only males in the pedigree chart have hemophilia. (Choose at least one of the males represented in the pedigree chart, and show his parents in the Punnett square.)

The X-linked recessive disease is called hemophilia. The only way for a female to have hemophilia is if her father suffers from the hemophilia disease, and the mother is a carrier or suffers from hemophilia. This recessive gene is seen in males more instead of females because the mother passese her X chromosome, and the father passes the Y chromosome. The punnett square below shows none of the women were suffering from hemophilia, but they carried the gene but did not express it, the men expressed the hemophilia gene.



16. Is it possible for a female to inherit hemophilia, and, if so, how?

Yes, it is possible for a female to inherit the hemophilia gene but it is a rare situation. The only way a female will inherit the gene is if her father suffers from hemophilia and her mother is a carrier of the gene, or also suffers from hemophilia.

17. Some historians speculate that Alexis' hemophilia condition could have led to the Russian Revolution. Explain.

You should look up the faith healer Rasputin and read about his relationship to the Romaov family.

Before his death, Alexis was the next heir to be the Czar of Russia. Once Alexis' condition of hemophilia was discovered he received help to heal him from Grigori Rasputin. Rasputin became close to the family. Unfortunately, Rasputin was killed which ended up causing the downfall of the royal family.

<u>Molecular Analysis of People in a Mass Grave</u>

18. Two "graves" were discovered near Yekaterinburg, Russia. Describe the number of bodies in each grave.

The first grave that was discovered was the largest grave which contained nine of the family members and staff. The second grave, which was the smaller grave, had the remains of two of the Romanov children.

19. When were these graves discovered?

In 1991, the first grave was discovered which was the largest grave. In 2007, the second smaller grave was discovered.

20. What type of testing was done to confirm <u>sex</u> and <u>familial relationships</u> among the remains found in the mass grave?

Nuclear DNA in cell nucleus and Mitochondrial DNA in cell mitochondria were the two types of test used. The purpose of these tests was to investigate the DNA markers.

21. Genetically, what does STR "stand" for? Be very specific in your answer.

Short Tandem Repeat, which is an analysis that uses quadruplex 3 and mtDNA analysis using the modification of a solid phase sequencing protocol.

22. Mitochondrial DNA testing was also done on both Nicholas II and Alix. Why was information from Alix's, but not Nicholas' mitochondrial DNA used to identify three females as belonging to Alix?

The main way to inherit Mitochondrial DNA is from the mother. An ovule which is found in females has about 1000 mitochondria. At fertilization there is no mitochondria found in fertilized eggs.

23. HRH Prince Philip, the Duke of Edinburgh, provided mitochondrial DNA used to identify Alix and her three daughters. HRH Prince Philip, the former Duke of Edinburgh, is married to Queen Elizabeth II of England. Wait, isn't Queen Elizabeth II related to Queen Victoria? So why was ***Prince Philip's*** mitochondrial DNA used?

How has Prince Philip been in the news in 2021?

Queen Victoria and Queen Elizabeth the second of England are related. Also Queen Elizabeth the second is also related to her husband Prince Philip. Therefore, at the end of the day it did not matter whose DNA was used because they have the same maternal mitochondrial DNA.

24. Who was missing from the mass grave (the one with the most skeletons)?

Tsarevich Alexei and one of his sisters of the Romanov family were missing from the mass grave and were later found in a separate smaller grave.

Molecular Analysis of People in a Mass Grave, cont.

25. The Duke of Fife and Princess Xenia provided mitochondrial DNA used to identify Nicholas. One of these is a female and another is a male. Does that matter? What general statement can you make about their genetic relationship to Nicholas and Alexandra? Are these people still living?

When using mitochondrial DNA comparison sex does matter. Since children recieve mitochondrial DNA from their mothers, a female's mitochondrial DNA would have to be used to determine a maternal relationship. This means that the mother of Nicholas passed her mtDNA to him and to her other female children which continued to pass down from female offspring until reaching Xenia. The Duke and Princess are still living (at least until after their DNA was collected to be analyzed).

26. What was discovered in the mitochondrial DNA of Nicholas that was not identified in either the Duke of Fife or Princess Xenia?

A point of heteroplasmy was found in the mtDNA sequence of Nicholas which was not found in the mtDNA of the Duke of Fife or Princess Xenia.

27. What is the term given to the existence of two (or more) genetically different mitochondria in the cell?

Heteroplasmy

28. What three types of DNA were used to test the remains found in a second grave?

The three types of DNA used in testing these remains were mitochondrial DNA, autosomal STR DNA, and Y- STR DNA.

29. Of the three types of DNA you listed in #28, which one would have been used specifically to identify Alexei?

The Y- STR DNA test was used to identify Alexei.

30. What was the source of the DNA used to identify Alexei?

The DNA from living relatives of the Romanov family were used to compare to the remains in the grave.

31. Was Anastasia in the grave in which Alexei was found?

It could not be determined because they did not have any DNA specific to Anastasia or Maria to compare to the remains to prove one way or the other.

Who Wants to Be Anastasia?

Apparently, about 200 people have wanted to be Anastasia and have claimed to be her! One of the most famous imposters was a woman named Anna Anderson (Manahan).

32. Give a brief history (2-3 sentences) of Anna Anderson-both her claims and what is thought to be true.

Anna Anderson Manahan came forward and claimed to be the Duchess Anastasia from the Romanov family and that she had survived and escaped her attempted murder via firing squad. Since it could not be determined that the body in the second grave was or was not Anastasia, many people believed her claim. Upon extensive investigation, nobody could indefinitely determine that she was or was not Anastasia at the time.

33. Where in the US did Anna Anderson eventually settle?

Anna Anderson eventually settled down in Charlottesville, Virginia ("Anna Anderson: The Great Imposter" 2014).

34. Whom did she eventually marry?

Anna Anderson married John Manahan ("Anna Anderson: The Great Imposter" 2014).

35. What were the sources of Anna Andersons's nuclear DNA?

A small sample of tissues were taken from her small bowels, removed during a surgery.

36. What were the sources of Nicholas' and Alix's nuclear DNA?

Blood samples of living relatives of the Russian Royal Family, as well as recovered bone fragments from Nicholas and his family.

37. What type of analysis was done on DNA from Anna Anderson, Nicholas, and Alix?

DNA testing was done using mitochondrial DNA, autosomal STR, and Y-STR testing.

38. Anna Anderson's mitochondrial DNA was compared to the mitochondrial DNA of what two people?

Anna Anderson's mitochondrial DNA was compared to the mitochondrial DNA of HRH the Duke of Edinburgh, a direct maternal descendant of the Tsarina, Alexandria. Anderson's mitochondrial DNA was also compared to the mitochondrial DNA of Carl Maucher, the great nephew of Schanzkowska, a direct relative through the maternal line.

39. A hypervariable region of the mitochondrial DNA was analyzed. Define a hypervariable region?

A hypervariable region is an area in mitochondrial DNA of nucleotide repetition and substitution.

40. What were the conclusions from the mitochondrial DNA comparisons?

Anderson's samples indicated that there was no association with a maternal relative of the Tsarina or of HRH Prince Philip. A match was indicated suggesting that Carl Maucher was a maternal relative of Anna Anderson, with the possibilities that they may be maternally related or they are unrelated. However, the findings did support the hypothesis that Anna Anderson was actually Franzisca Schankowska.

41. The article which describes the analysis of Anna Anderson's DNA was published in 1995. When were all of Nicholas' and Alix's children finally accounted for?

Nicholas' and Alix's children were all finally accounted for in late 2007. The Russian government brought in scientists to conduct DNA testing of the remains of a second grave site, ending the mystery of the missing Romanov children.

42. What did you learn from doing this assignment? (Each person in a group should answer this question. It is not a group answer.)

Savannah Crew - From doing this assignment, not only did I learn about the history of the Romanov family but I learned a great deal about genetic testing. Before, I had no idea that there were so many different ways to test DNA and different ways to determine lineage of individuals. The assignment particularly solidified my knowledge of mitochondrial DNA and how it is passed down lineages through solely the mother.

Danielle Wolcott - While working on this assignment, I learned that there are multiple ways and methods of genetic testing. In the case of the Romanov family there were multiple methods of genetic testing used to identify the family members and to compare DNA to the others. This assignment expanded my knowledge and taught me how genetic testing is done based on the source of DNA and how multiple methods may have to be used to come to an overall conclusion.

Tanatswa Sambana - While working on this project I learned that hemophilia is a x-linked disease that is recessive. The disease is commonly found in males, but is very rare in females. I learned that hemophilia is a disease that was mostly common in the European families because of how they would marry or breed with people of royal blood, which led to marriages within families.

Teresa Hooker- While working on this assignment, I was fascinated to learn about the history of the Romanov family and their medical history with carrying the mutation for Hemophilia from generation to generation. In

addition to the history of it, I learned much more about the Hemophilia Disease and its' mutation on the X-chromosome as a whole.

Are you still interested in the life of the last Tsar of Russia and his relationship to British royalty? The headline for the following article showed up on my Internet browser earlier this year. While I can't vouch for it as it did not appear in a peer-reviewed journal, it might be interesting reading for you.
https://www.townandcountrymag.com/society/tradition/a31028924/windsors-romanovs-relationship-last-gathering-true-story/

Citation(s):

33&34- Anna Anderson: The Great Imposter. *Legacy.com*;
https://www.legacy.com/news/anna-anderson-the-great-imposter/

WHILE THIS IS A GROUP PROJECT, EACH STUDENT SHOULD UNDERSTAND EACH QUESTION AND ANSWER GIVEN. BY PLACING YOUR NAME BELOW (AND INCLUDING A DATE), YOU HAVE REVIEWED THE ANSWERS ON THIS ASSIGNMENT, AND AGREE TO THEM.

Teresa Hooker (06/24/2021)
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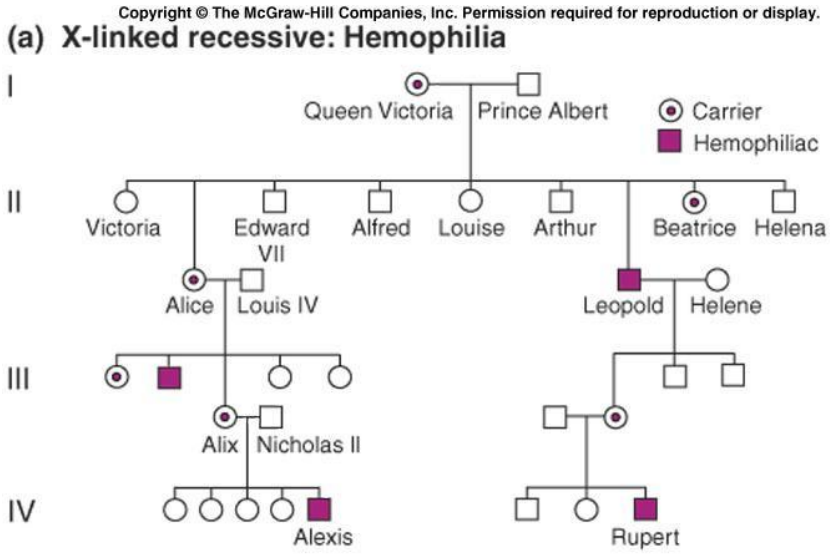

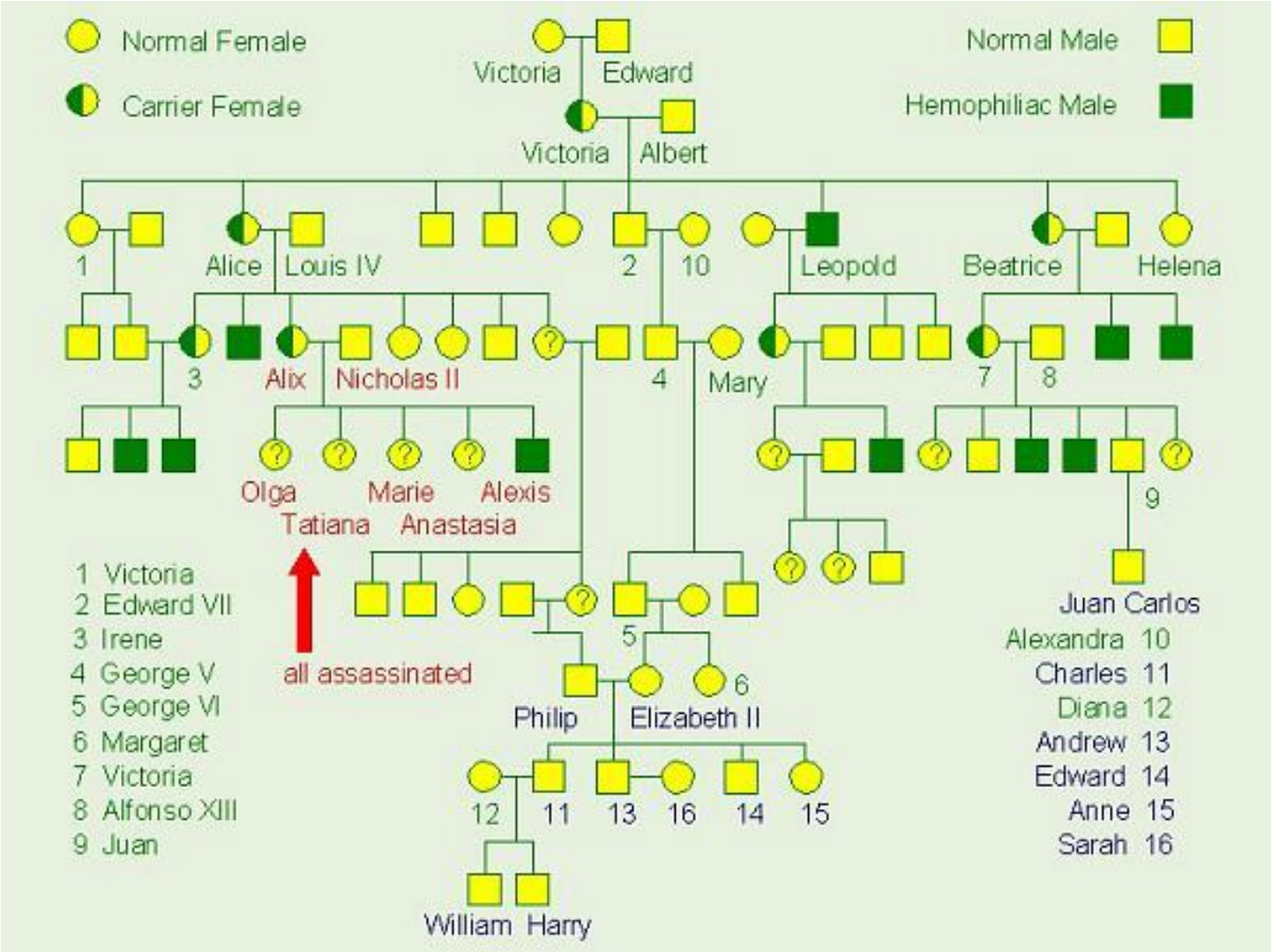

Source for the pedigree chart above: Janet Stein Carter, Biology Instr uctor at Clermont College, University of Cincinnati