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.0004*](http://journals.plos.org/plosone/article?id=10.1371/journal.pone.0004)*838 (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*](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.)

Blakemore, E. Why Czar Nicholas II and the Romanovs Were Murdered. History; <https://www.history.com/news/romanov-family-murder-execution-reasons> (2019)

Llewellyn, J. Thompson, S. The White Armies. Alpha History; <https://alphahistory.com/russianrevolution/white-armies/#:~:text=The%20White%20armies%20> (2019)

U.S. National Library of Medicine. Hemophilia; MedlinePlus. <https://medlineplus.gov/genetics/condition/hemophilia/#inheritance> (2020)

Britannica, The Editors of Encyclopaedia. Grigori Rasputin. Britannica; <https://www.britannica.com/biography/Grigory-Yefimovich-Rasputin> (2021)

González-Candelas, F. López-Labrador F. Hypervariable Region. Brenner’s Encyclopedia of Genetics <https://www.sciencedirect.com/topics/biochemistry-genetics-and-molecular-biology/hypervariable-region> (2013)

**History**

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

The last ruling Russian Czar

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

For over 300 years

3. Politically, what happened to Nicholas II?

Nicholas II promised representative government and civil liberties in Russia with a manifesto. He went back on this and was forced to abdicate his throne by the revolution going on in Russia led by the Bolsheviks.

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

Nicholas II tried to give his throne to his brother Grand Duke Michael Alexandrovich but his brother declined. Russia’s provisional government went into control just to get overthrown by the Bolsheviks six months later.

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

After Nicholas II abdicated his throne his whole family and four loyal members of the staff were held in exile in Yekaterinburg, Russia. Then, 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?

They were loyalists who obeyed the crown and opposed the Bolsheviks.

**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, related to Queen Victoria of England?

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

Alix was her granddaughter.

8. In a couple of sentences, describe the physiology of the disease hemophilia. (Yes, I know it is severe bleeding because the blood cannot clot. But WHY can’t the blood clot? Be *very* specific.)

Hemophilia is a disorder in which an individual’s blood lacks certain blood clotting proteins. There are different types of hemophilia because there are different blood clotting proteins that can be absent in an individual's blood due to a flaw in the gene. The blood can not clot because the coagulation platelets can not link to each other to form an effective seal over the wound opening.

9. What does it mean to be a carrier for a disease?

To be a carrier an individual must have the inactivated mutation in their genome. (Or the mutation has minimal affect) They must also possess the ability to pass on the disease.

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

Hemophilia type B

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

The single X chromosome that is passed into males with a Y chromosome.

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

It was substitution of Alix’s DNA with the splice acceptor site of exon 4 on the F9 gene.

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

The mutation is a substitution which means one nitrogenous base pair is replaced by a different base pair. Since the mutation causes a physiological change it is either a missense or nonsense mutation because the mutation causes protein malfunction. The mutation either codes for the wrong amino acid or the mutation causes the codon to become a stop codon.

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.)

|  |  |  |
| --- | --- | --- |
|  | XH | Xh |
| XH | XHXH | XHXh |
| Y | XHY | XhY |

Since Alexis’ genotype can be XHY or XhY he has a 1:1 ratio on whether he would be affected or not.

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.)

Being that the disease is a X-linked recessive disorder, only one copy would be needed for males to have the disease opposed to women needing two copies of the gene. Since no affected male mated with a carrier female there were no affected females.

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

Yes, while rare, females can get hemophilia if both X chromosomes are affected by the mutation. This can occur if both parents are affected or if the parents are an affected male and carrier female, with the likelihood of having an affected daughter being 100% and 50% respectively.

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 Romanov family.***

Rasputin was a supposed healer of ailments via alternative techniques, the Romanov family trusted him and promoted him to their people. When it was clear he was not healing Alexis via his methods, the people got angry and thought the royal family was deceiving them. Rasputin already had controversy from other important families who called him fraudulent and not to be trusted, which worsened the Romanov’s public image. It was also rumored that Rasputin was sleeping with the tzar’s wife which led to more massive civil unrest of the people. It is theorized that the Romanov family trusted Rasputin so much as he was actually able to “heal” Alexei. However, the healing was simply making doctors stop giving alexei ibuprofen as it is a blood thinner.

**Molecular Analysis of People in a Mass Grave**

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

The mass grave, or the one with nine bodies, had five members of the royal family and the four loyal members of their staff. The second grave contained the two missing royal children.

19. When were these graves discovered?

The mass grave was first discovered in the late 1970s but official testing was not done on the site until 1991. The second grave was found in the summer of 2007.

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

DNA testing of the STR markers for familial relations and Y-STR testing for sex.

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

Short Tandem repeat. It is used in biology and forensic to match DNA samples.

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?

Being that mitochondrial DNA is only passed from the mother to child it is only possible to compare children mitochondrial DNA to their mother.

23. HRH Prince Philip, the Duke of Edinburgh, provided mitochondrial DNA used to identify Alix and her three daughters. HRH Prince Philip, the 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?

Prince Philip is the husband of Queen Elizabeth II with their grandchildren being Prince William and Prince Harry. Prince Philip’s mitochondrial DNA was used because of his lineage to the Tsarina.

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

Russians say that Maria was missing but Americans claim Anastasia was the one who was missing. Alexis, the son with hemophilia, was also missing.

**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?

It does not matter. They would just have to be of maternal lineage for Nicholas’s mitochondrial DNA to show the relationship.

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

Nicholas had a fixed hetroplasmy point at 16169 and the Duke and Princess did not; they were just T.

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

Chimera Cells.

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

Autosomal STR, Y-STR, and mitochondrial DNA.

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

Y-STR testing could be performed to identify Alexis.

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

They used Prince Andrew Andreevich Romanov as the DNA source.

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

It is unknown.

**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’s birth name according to the findings in Nature is Franzisca Schanzkowska. Schanzkowska was born in 1896 in Germany. Then, during world war 1 she was badly injured by an explosion while working in a munitions factory. She disappeared in 1920.

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

Here, in Charlottesville.

34. Whom did she eventually marry?

John Manahan

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

Samples from her stomach

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

From the bones of the czar and his wife.

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

Mitochondrial DNA testing was done on the DNA of all three people.

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 the Duke of Edinburgh, who is the great nephew of the tsarina, and Carl Maucher, who is the great nephew of Franzisca Schanzkowska.

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

Hypervariable regions are portions of the DNA in a particular species that has much higher levels of variation than similar portions of DNA. A hypervariable region on mitochondrial DNA is a location on the mitochondrial DNA where nitrogenous base pairs have substitutions. These regions can be tested to show lineage because of the high variability among a certain species.

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

The conclusions from the mitochondrial DNA testing of Anna Anderson compared to the mitochondrial DNA testing of Carl Maucher and the Duke of Edinburgh showed a lineage between Anna Anderson and Carl Maucher.

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?

In the summer of 2007 the second grave was found with the two bodies. The scientists knew at some point during their studies that the two bodies were the missing Romanov Children. However, the results were not published until 2009.

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

Stephen Blanchard. I learned how mitochondrial DNA testing works. I learned about the History of Nicholas II. I also learned how hemophilia works and was able to review pedigree charts and x-linked recessive disorders.

Evan Howell. I learned about the process of blood clotting and how hemophilia is passed down. I also learned of the supposed magic healer who caused civil unrest amongst the royal family.

Taylor Barr. I gained a better understanding of how sex-linked traits work, how they are tracked and passed down.

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/>

**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.**

­­Taylor Barr

Nathan Cook

Stephen Blanchard

Evan Howell

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