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Romanov Assignment

History

1. Nicholas II was the last czar to hold power in Russia. How long had the Romanov family been in power in Russia?

The Romanov family ruled Russia for over 300 years.

2. Nicholas II abdicated the throne. Who took power then?

The Ural Soviets had power after Nicholas II abdicated the throne.

3. Describe what happened to Nicholas II and his family after he abdicated the throne?

Nicholas II and his family were held exile with four members of their staff and then were executed.

4. 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?

The White Russian Army was a group that were anti-Bolshevik and fought against them during the Civil War for control of Russia.

Hemophilia

One of the pedigree charts found at the end of this assignment comes from the Module PowerPoint lecture notes.

5. How was Alix, the wife of Nicholas II, related to Queen Victoria of England? (Look at the pedigree chart very carefully.)

Alix is the granddaughter of Queen Victoria of England.

6. On what chromosome is the gene that, when mutated, causes hemophilia, and how does this contribute to its inheritance pattern?

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

The chromosome that when mutated, causes hemophilia is the X chromosome. Since this is X-chromosome related, that means that males are more likely to have hemophilia because they only have one X chromosome. Since they only have one X chromosome, they will either have it, or won't and cannot just be a carrier of it.

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

Being a carrier means that the disease is recessive and that one of the chromosomes contains the mutation, while the other does not. In order to have a disease that is recessive, both the chromosomes must have the mutated disease, but if only one is infected, then you do not have the disease, but are carrying an infected chromosome.

8. Why aren't males considered *carriers* for hemophilia?

Males are not considered carriers because they only have one X chromosome, so if it contains the mutated disease then there is no other option than having the disease. Whereas a female has two X chromosomes and with only one infected, the noninfected chromosome protects enough to not acquire the disease.

9. 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 severe bleeding when people have low levels of factor eight or factor nine in their bodies. This occurs when someone has a mutation in one of the genes that instructs for blood clotting to occur. There are not enough blood clotting proteins due to the mutations in someone with hemophilia.

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

Hemophilia B is probably represented in the pedigree chart. Based on the abstract provided (Lannoy, N., Hermans, C. 2010), scientists were able to find the tomb of Alexis and then captured biological information from it and results showed that there was a substitution of exon 4 in the gene F9. That specific mutation results in hemophilia B.

11. 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.

The mutation that occurred in probably all European families that had hemophilia was a causal substitution. This substitution occurred in the splice acceptor site in the F9 gene of exon 4. A substitution is when one nucleotide is removed and swapped with a different nucleotide.

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

The mutation described in the previous question could result in a faulty gene if the substitution leads to a completely different amino acid. When substitution occurs in the first or second base, that can lead to a completely different amino acid, but not with the third base.

13. 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.)

Alexis's mother Alix was a carrier of hemophilia and this is how Alexis acquired the disease. Since Alexis is a boy, the only way he could acquire hemophilia is from the mother because males acquire the X chromosome from the mother and the Y chromosome from the father. With hemophilia being X chromosome related, a son can only get it from the mother. With Alix being a carrier, one of the X chromosomes was affected, while the other wasn't, and Alexis genetically acquired the infected X chromosome. The below chart shows the possibilities of the children of Alix and Nicholas II with two females and two males as children. Alix had a 50/50% chance of being infected with hemophilia or not. In this case, Alexis is the highlighted XY as being infected. Inf stands for infected and noninf stands for non-infected.

	Xnoninf	Y
Xinf	XinfXnoninf	XinfY
Xnoninf	XnoninfXnoninf	XnoninfY

14. 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.)

All of the females from the pedigree chart are only carriers and do not actually have hemophilia, therefore only the males can be infected because the males will get their only X chromosome from the mother. The only possible way that a female could get hemophilia with a mother who is a carrier is if the mother mated with an infected man. There are no cases in the pedigree chart where a carrier mother mated and had kids with an infected father, so therefore only the sons had a chance of having hemophilia. I have chosen to examine Leopold as the example of how he got

hemophilia and why the females did not. Since hemophilia is linked to the X chromosome and recessive, females can only get it if both X chromosomes are infected whereas males can get it with their one and only X chromosomes being infected. In this case, Leopold had a 50/50% chance of having hemophilia since his dad Prince Albert was not infected, but his mother Queen Victoria was a carrier, and he got it. His other brothers did not have hemophilia though, so they landed with the noninfected X chromosome from their mother.

	X^{noninf}	Y
X^{inf}	$X^{inf}X^{noninf}$	$X^{inf}Y$
X^{noninf}	$X^{noninf}X^{noninf}$	$X^{noninf}Y$

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

Yes, it is possible for a female to inherit hemophilia. The way a female can inherit hemophilia is if a mother who has hemophilia or is a carrier of hemophilia has children with a male who has hemophilia as well. This would mean that the infected X chromosome from the mother would come together with the infected X chromosome from the father. The Punnett square below shows the two different ways that a female could get hemophilia.

	X^{inf}	Y
X^{inf}	$X^{inf}X^{inf}$	$X^{inf}Y$
X^{noninf}	$X^{noninf}X^{inf}$	$X^{noninf}Y$

	X^{inf}	Y
X^{inf}	$X^{inf}X^{inf}$	$X^{inf}Y$
X^{inf}	$X^{inf}X^{inf}$	$X^{inf}Y$

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

Since Alexis was infected with hemophilia, his health conditions were very poor and there were no ways to cure or help him back then. The family reached for help from Grigory Rasputin who claimed to have mystic healing powers, which apparently helped some but ultimately Nicholas II resigned himself and the family from the throne which then led to them being executed and the Russian Revolution occurred as well.

Molecular Analysis of People in a Mass Grave

17. Two "graves" were discovered near Yekaterinburg, Russia. When were these graves discovered, and how many bodies were found in each grave?

The first grave was discovered in the late 1970's and had nine bodies. The second grave was discovered in 2007 and had two bodies.

18. What type of testing was done to confirm sex and familial relationships among the remains found in the mass grave? If you use an abbreviation, write it out and define what it is.

DNA testing was conducted in 1991 on the remains that were found in the mass grave. The familial relationships and sex of the skeletons were confirmed by nuclear DNA testing of five short tandem repeat markers.

19. 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? (To help you answer this question, look at the second pedigree chart.)
Prince Philip is also related to Alix based off of the pedigree chart through generations of marriages and children through the maternal side. Prince Philip is the great nephew of Alix and Queen Elizabeth II is the third cousin of Alix. Prince Philip is related to her through a direct sister of Alix rather than Queen Elizabeth II who is related to her through an uncle.
20. Who was missing from the mass grave (the one with the most skeletons)?
Two children of Tsar Nicholas II were missing from the mass grave and that was Alexis and one of his sisters.

Molecular Analysis of People in a Mass Grave, cont.
21. 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?
It essentially does not matter that one of them is a female and one of them is a male. Both of them are genetically related to Nicholas as maternal relatives.
22. What was discovered in the mitochondrial DNA of Nicholas that was not identified in either the Duke of Fife or Princess Xenia?
According to (Coble, M. et al., 2009), Nicholas II mtDNA portrayed a single point heteroplasmy at position 16169 (C/T="Y") but both the Duke of Fife and Princess Xenia were fixed for 16169 T.
23. What is the term given to the existence of two (or more) genetically different mitochondria in the cell?
The term given to the existence of two or more genetically different mitochondria in the cell is heteroplasmy.
24. What three types of DNA were used to test the remains found in a second grave? Again, if you use an abbreviation, write it out and define what it is.
The three types of DNA used to test the remains found in the second grave was mitochondrial DNA, autosomal short tandem repeat, and Y chromosome short tandem repeat.
25. Of the three types of DNA you listed in the previous answer, which one would have been used specifically to identify Alexis and why?
Y chromosome short tandem repeat would have been used specifically to identify Alexis because he is a male and only males have Y chromosomes.
26. Was Anastasia in the grave in which Alexis was found?
Alexis was found in the second grave and it cannot be determined if Anastasia was found in this second grave as well or if it was Maria based on the DNA tests that were ran and the results received.

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).
27. Give a brief history (2-3 sentences) of Anna Anderson-both her claims and what is thought to be true.
Anna Anderson had claimed to be Anastasia, the daughter of Nicholas II around 1920. Others believed that Anna Anderson was actually Franzisca Schanzkowska who was born in 1896 and was injured in an explosion at work in Germany and then disappeared after being admitted to

mental hospitals. It turned out that Anna was not Anastasia and was in fact Franzisca Schanzkowska based on different DNA testing.

28. Where in the US did Anna Anderson eventually settle and why?

Anna Anderson settled in Charlottesville, Virginia because she married an American history professor.

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

The sources of Anna Anderson's nuclear DNA were small bowel samples and some hairs from Anna Anderson.

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

The sources of Nicholas and Alix's nuclear DNA was from the bones of them and the DNA profiles that were built from the analysis of the bones.

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

Five short tandem repeats were performed and compared between the paraffin wax blocks of Anna Anderson to the bone DNA profiles of Nicholas and Alix. Only one of the five short tandem repeats lined up with each other.

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

The two other people whose mitochondrial DNA was compared to Anna Anderson's mitochondrial DNA was the Duke of Edinburgh and Carl Maucher.

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

A hypervariable region is a part of the genome that contains many different levels of variation compared to other areas that are similar to it. Variation can occur each generation or through mutations, rearrangement, and more.

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

It was concluded that Anna was not maternally related to the Duke of Edinburgh, but that Carl Maucher was a possible maternal relative of hers. To further confirm this, scientists compared the DNA of Anna and Carl to over 300 other sequences of Caucasians. The results of this confirmed that Carl Maucher and Anna Ander were related and that Anna was in fact Franzisca Schanzkowska.

35. 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?

All of Nicholas II and Alix's children were finally accounted for in 2007 when scientists further examined the remains found from the second grave.

36. What was the most surprising thing that you learned from doing this assignment?

The most surprising thing that I learned doing this assignment was how many people have tried to claim being Anastasia and the extent that Anna went, to try and prove that she was Anastasia. It's amazing how power can drive people to lie and Anna tried to take advantage of that. All of the DNA testing's that were performed are pretty incredible as well and how they have been able to rule people out and confirm that Anastasia died along with the rest of the family.

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