

More than Meets the Eye: The Genetics of Eye Color

by

Annie Prud'homme-Généreux
Life Sciences
Quest University, Canada



Part I – High School Blues

Evan and Alexia had been happily married for seven years and had a delightful five-year-old son named Ryan. One day, while going through his old high school biology textbook, Evan stumbled on some troubling information. In the section on the genetics of eye color, he read that two blue-eyed parents cannot produce a brown-eyed child. This was disturbing to him because both he and Alexia had blue eyes, but Ryan had brown eyes.

He and Alexia were very much in love and Evan didn't believe his wife had been unfaithful. Puzzled, he questioned his wife, who confirmed she had been faithful to him. Evan had known Alexia long enough to recognize when she was lying and detected nothing but honesty in her response.

What is going on?

Questions

1. What protein does the “eye color gene” encode? Propose what the function of this protein might be, and how this protein might differ in people with brown and blue eyes.
2. Which is dominant and which is recessive: the blue eye allele or the brown eye allele?
3. If Evan has blue eyes, what genotype (which two alleles) is he likely to have?
4. If Alexia has blue eyes, what genotype (which two alleles) is she likely to have?
5. Draw a Punnett square showing the genotype of all the possible children this couple could have. Based solely on this information, what is the likelihood that Ryan is their child?
6. Assuming that Alexia has been unfaithful, what is (are) the possible genotype(s) of the man who is Ryan's biological father?
7. Assuming that Alexia has been faithful, suggest as many hypotheses as you can to explain Ryan's phenotype.

Part II – Eye Coloration

Puzzled, Alexia and Evan used the internet to research what gives the eye its color.

“Eye color” refers to the color of the iris of the eye. Melanin is a dark pigment produced by cells in the iris that gives the eye its color. What determines the color of the eye is a combination of the amount, location, and qualities (e.g., different types) of the melanin present in the iris (Sturm & Larsson, 2009).

The iris has a front layer and a back layer. The space in between them, called the stroma, is filled with various proteins, including white collagen fibers. For almost all eye colors, there is a lot of melanin on the back layer of the iris (Sturm & Larsson, 2009). Where people differ is in the melanin in the front layer of the iris.

A lot of melanin in the front of the iris makes the eye look brown because, as light hits the front of the iris, the pigments absorb the light.

Blue irises have less melanin in the front layer, so light can go through it. As light travels through the stroma, it encounters the collagen fibrils. This scatters the short blue wavelengths to the surface. In other words, when light hits the collagen fibrils, the light is refracted, or bent, and this makes the light appear blue or green. This effect is also experienced when looking at the sky. The sky is actually black. However, as light travels through the Earth’s atmosphere, it encounters particles that bend the light and cause the sky to appear blue. This effect is called Rayleigh Scattering (Southworth, 2007; Sturm & Larsson, 2009).

A lot of pigment in the front of the iris gives brown, less melanin gives green or hazel, and little pigment gives blue (Figure 1).

While blue irises have little melanin of any kind in the front of the iris, other eye colors vary in the relative amount of the different types of melanin (called eumelanin and pheomelanin), giving a spectrum of eye shades (Sturm & Larson, 2009).

Questions

- Based on what you now know, how many genes may be involved in determining eye color? Suggest what each gene does to affect this trait.
- What do you suspect that the blue/brown eye color gene studied in high school does in the cell? What type of protein might this gene encode? Offer several possibilities.

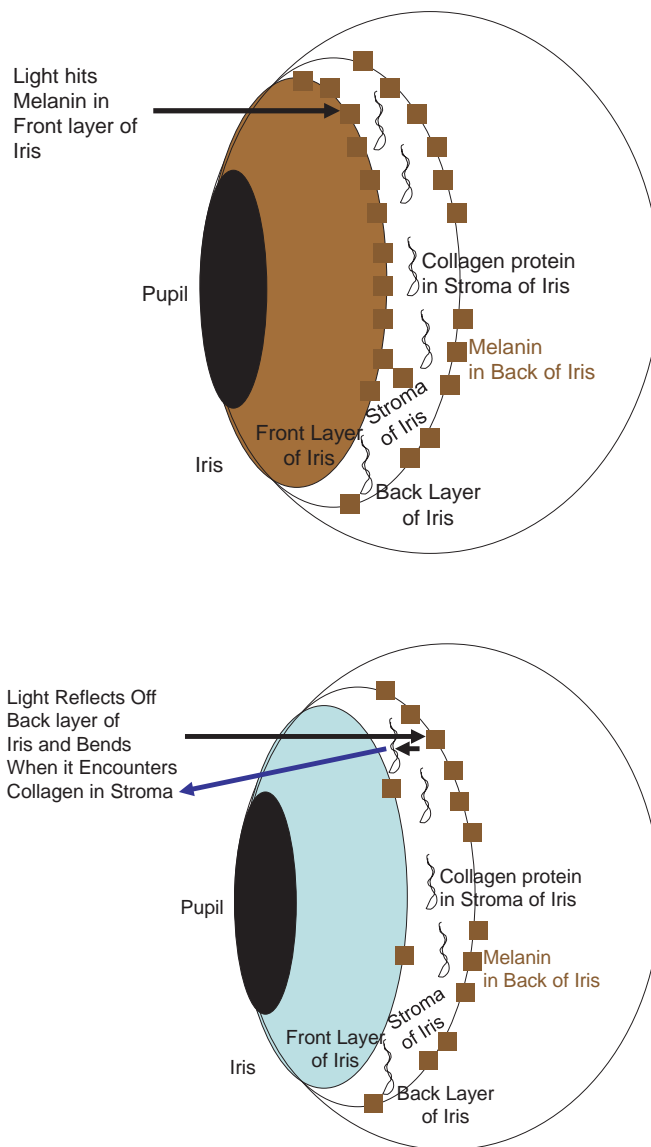


Figure 1: Top figure illustrates a brown eye, and bottom figure shows a blue eye. The small brown squares in the front and back of the iris represent melanin molecules. The relative number of brown squares represents the relative density of melanin in various regions of the iris in eyes of different color.

10. Based on your previous answer, how might the blue and brown alleles differ (how might they differ in function, in sequence, in the resulting protein, in structure, etc.)?
11. Eyes can be brown, blue, or green/hazel. How could these three differences be encoded genetically? Suggest several ways to achieve these three phenotypes.
12. Does this information suggest ways in which two blue-eyed individuals could have a brown-eyed child?

References

- Southworth, L. (2007). Ask a Geneticist: Are gray eyes the same as blue in terms of genetics? *Understanding Genetics*. The Tech Museum, Stanford University School of Medicine. Retrieved 2 April 2010 from <http://www.thetech.org/genetics/ask.php?id=232>.
- Sturm, R.A., and Larsson, M. (2009). Genetics of human iris colour and patterns. *Pigment Cell and Melanoma Research* 22: 544–562.

Part III – Genetics of Eye Color

Their curiosity further piqued, Alexia and Evan contacted Dr. Rick Sturm¹ at the University of Queensland, a leading expert on eye color.

“It used to be thought that eye colour was what we call a simple Mendelian recessive trait—in other words, brown eye colour was dominant over blue, so a person with two brown versions of the gene or a brown and a blue would have brown eyes, and only two blues with no brown could produce blue eyes. But the model of eye colour inheritance using a single gene is insufficient to explain the range of eye colours that appear in humans. We believe instead that there are two major genes—one that controls for brown or blue, and one that controls for green or hazel—and others that modify this trait. The mechanism that determines whether an eye is brown or blue is like switching on a light, whereas an eye becoming green or hazel is more like someone unscrewing the light bulb and putting in a different one,” said Dr. Sturm (Sturm, quoted from: “Eyes Have it on Multiple Gene Question,” n.d).

To answer Alexia and Evan’s question, he added: “So contrary to what used to be thought, it is possible for two blue-eyed parents to have a brown-eyed child, although this is not common.” (Sturm, quoted from “Eyes Have it on Multiple Gene Question,” n.d.).

Hearing these words, Evan sighed in relief. Even though he trusted his wife, it was reassuring to hear one of the world’s leading experts on the matter say that Ryan could be their son.

Dr. Sturm went on to explain that, contrary to what is taught in high school genetics classes, there are many genes involved in the determination of eye color. Two of them are most important in determining eye color.

The first gene, called *OCA2* (or *bey2* or *EYCL3*), is on chromosome 15. Although the exact function of the *OCA2* protein in the cell is not known, it seems to be involved in the production of melanin (“*OCA2*”, 2009). *OCA2* exists in two allelic forms: brown or blue. The brown (B) allele stimulates the production of high levels of melanin. The blue allele (b) does not produce the *OCA2* protein, leading to the loss of melanin production. The brown allele is dominant over the blue allele. The mutation that produces the blue allele is a single point mutation (from T to C) in the intron of a gene upstream from *OCA2* called *HERC2* (Eiberg et al., 2008; Sturm et al., 2008). This point mutation occurs in a regulatory element for *OCA2*. The mutation prevents the binding of a helicase, which makes the DNA accessible to transcription factors for *OCA2* transcription.

The second gene, called *gye*, is on chromosome 19 and comes in two forms: blue or green. Green (G) codes for a protein that results in the production of some melanin, and is dominant over blue (g), which codes for a protein that results in no production of the pigment.

In effect, there is a hierarchy: the *OCA2* gene is epistatic to the *gye* gene (in other words, the product of the *OCA2* gene masks the product of the *gye* gene). If one allele at the *OCA2* gene site encodes the brown eye trait, the alleles at the *gye* sites are irrelevant and the eyes are brown. However, if the alleles at the *OCA2* site are bb, then the combination of alleles at the *gye* site determines whether the eyes are blue or green (Starr, 2005).

Questions

13. Which of the two genes is likely to have been the one referred to in high school genetics problems of eye color?
14. Determine all the possible genotypes (at the *OCA2* and *gye* loci) for Alexia and Evan.
15. Are the *OCA2* and *gye* genes genetically linked? Why or why not?
16. Make a Punnett square showing the genotypes of all possible children produced by this couple (show the *OCA2* and *gye* gene loci in your Punnett Square). Determine the eye color of all possible children.
17. Can the *OCA2* and *gye* genes explain why Ryan has brown eyes?

¹ Dr. Rick Sturm is a leading expert in the field of the genetics of eye color. While the comments attributed to him are taken from interviews and press releases (as indicated in the references to this part), this case is a work of fiction. It is not based on Dr. Sturm’s actions or comments in response to this particular situation.

References

- Eiberg, H., Troelsen, J., Nielsen, M., Mikkelsen, A., Mengel-From, J., Kjaer, K.W., and Hanse, L. (2008). Blue eye color in humans may be caused by a perfectly associated founder mutation in a regulatory element located within the HERC2 gene inhibiting OCA2 expression. *Human Genetics* 123: 177–187.
- “OCA2” (2009). Genetics Home Reference: Your Guide to Understanding Genetic Conditions. U.S. National Library of Medicine. Retrieved 5 May 2010 from <http://ghr.nlm.nih.gov/gene=oca2>
- Starr, B. (2005). Ask a geneticist: Has any progress been made in explaining eye colors other than brown, blue, and green? Understanding Genetics, The Tech Museum, Stanford University School of Medicine. Retrieved 2 April 2010 from <http://www.thetech.org/genetics/ask.php?id=126>
- Sturm, R.A., Duffy, D.L., Zhen Zhaw. Z., Leite, F.P.N., Stark, M.S., Hayward, N.K., Martin, N.G., and Montgomery, G.W. (2008). A single SNP in an evolutionary conserved region within intron 86 of the HERC2 gene determines human blue-brown eye color. *American Journal of Human Genetics* 82: 424–431.
- “The eyes have it on multiple gene question” (n.d.). The University of Queensland Australia, Institute for Molecular Bioscience. Retrieved 2 April 2010 from <http://www.imb.uq.edu.au/index.html?page=57400&pid=11690>

Part IV – What Else Could Be Going On?

Alexia and Evan were baffled. The two genes were not sufficient to explain how a blue-eyed couple could have a brown-eyed child.

However, they remembered Dr. Sturm's words: *"So contrary to what used to be thought, it is possible for two blue-eyed parents to have a brown-eyed child, although this is not common."*

The couple pooled their knowledge of genetics to think of ways in which two blue-eyed parents might have a brown-eyed child.

Questions

18. Re-examine your answer to Question 11 and the information learned in the previous section. How similar are the DNA at the associated regions of blue-eyed and brown-eyed people likely to be? Based on this, propose a mechanism that could account for Ryan's eye color. Be as specific as you can in explaining what might have happened.
19. Many genes are known to affect eye color. More than two are known to have an effect. You have already reviewed the effects of OCA2, HERC2, and *gcy*, and your answer to Question 8 may have identified a few more possibilities. These genes work together to affect the quantity, distribution, and quality of the melanin found in the front of the iris. Each must work well for the formation of brown irises. Non-brown eyes result when there is a mutation *in any one* of these gene products. Do blue-eyed parents always share the same mutation? What if they didn't? How might this account for Ryan's eyes?
20. Do organisms that have identical DNA always share a common phenotype (think of identical twins)? What can account for observed differences in phenotype? How might this play out at the molecular level? How might this apply in Ryan's case?

Part V – Three Hypotheses

Following their reflections, Alexia and Evan contacted Dr. Barry Starr² at Stanford University to confirm their three hypotheses.

Hypothesis 1: A Mutation

“Genetics is complicated by the fact that genes don’t always stay the same.[...] Our DNA copying machinery is nearly perfect but it still will make an occasional mistake. If that mistake happens in sperm or egg cells, it will get passed on. And if the change is in the right place in the blue eye gene, blue-eyed parents can now have a brown-eyed child. [...] Genes for things like blue and brown eyes are very, very similar. In fact, they are really just different versions of the same gene. [...] So, to turn a blue eye gene into a brown eye gene, you may only need to change a single letter” (Starr, n.d.)

Hypothesis 2: Epigenetic Effects

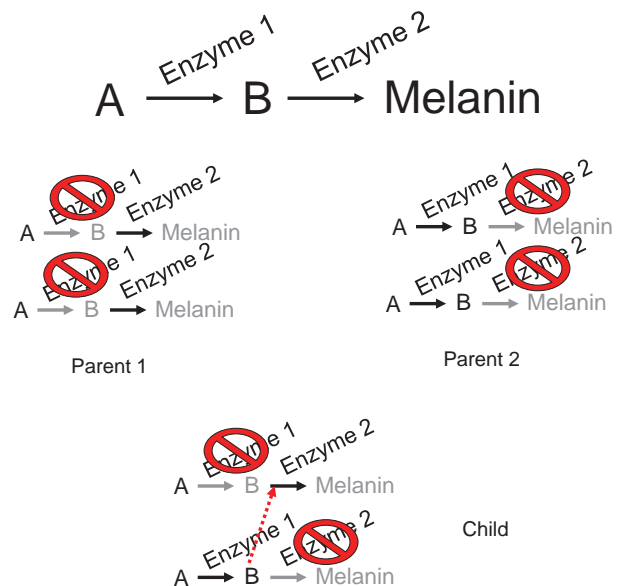
Dr. Starr also offered an alternate hypothesis.

“Sometimes a gene can be read in one person but is unreadable in another. What happens if a gene is unreadable in a parent but a child’s cells can read it? That’s right, a blue-eyed parent can have a brown-eyed child. Believe it or not, sometimes what your mom eats while she is pregnant can affect your hair color. Well, if you’re a mouse, anyway... Scientists did an experiment where they fed a mouse one food and her pups were black. A different food resulted in [yellow] pups. And all of the A, G, C, or T’s were the same between the pups. What happened? The food ended up attaching little chemical groups called methyls to the DNA. These methyls made the gene unreadable. So even though genetics would predict the same color pups, the environment changed the outcome” (Starr, n.d.)

Hypothesis 3: Genetic Complementation

Alexia and Evan also contacted another biologist, Ky Sha,³ while they were at Stanford University.

Ky had a different idea to explain their situation. Because eye color is determined by many genes, it is possible that each gene product collaborates to synthesize melanin as though they were stations in an assembly line (in other words, the gene products work in series; this is called a biochemical pathway). If this is true, different enzymes work one after the other on intermediates in the assembly line. If an enzyme working on the melanin pigment ahead of them “breaks down,” then all the enzymes downstream on the assembly line cannot do their job. The assembly line stalls. Imagine that Parent 1 has a mutation in Enzyme 1 (on both chromosomes) that prevents the formation of melanin. Parent 2 has a different mutation (also on both chromosomes) that affects a station more downstream in the assembly line, but Parent 2 is also not able to put together melanin. Both parents will have blue eyes because they lack the final product: melanin. However, when the parents combine their genes to produce a child, the child inherits one production line that breaks down at Enzyme 1,



² Dr. Barry Starr is a biologist at Stanford University. While the comments attributed to him are taken from an article he has written (as indicated in the references to this part), this case is a work of fiction. It is not based on Dr. Starr’s actions or comments in response to this particular situation.

³ Ky Sha is a biologist at Stanford University. While the comments attributed to him are taken from an article he has written (as indicated in the references to this part), this case is a work of fiction. It is not based on Mr. Sha’s actions or comments in response to this particular situation.

and one production line that breaks down at Enzyme 2. Since the child has some functional Enzyme 1 and Enzyme 2 floating in his or her cells, a complete and working production line can be assembled. As a result, some melanin can be formed, giving the child brown eyes. This is called “genetic complementation” (Sha, 2004).

Questions

21. It was recently discovered that almost all people who have blue eyes most likely share a common ancestor that lived 6,000 to 10,000 years ago. This was determined independently by three research groups who came to similar conclusions about the single locus responsible for blue eye color (Eiberg et al., 2008; “*Blue-eyed humans*,” 2008; Sturm et al., 2008; Kayser et al., 2008). Given this information, which of the three hypotheses to explain Ryan’s eye color are you likely to dismiss?
22. Eiberg et al. (2008) discovered that all blue-eyed humans likely share a common ancestor and mutation; however, the authors of the study and the other similar studies leave open the possibility that there are blue-eyed people that do not have the proposed causative allele in HERC2. For instance, one of the other studies that came out at the same time states, “Individuals carrying the C/C genotype had only a 1% probability of having brown eyes. By contrast, T/T carriers had an 80% probability of being brown eyed” (Sturm et al., 2008, p.425). Apparently, 1% of the people studied in Sturm’s study who were homozygous for the proposed causative mutation for blue eyes had brown eyes. What does this suggest about the genetics of eye color and your conclusions about Ryan’s eye color?
23. Given the information that you have been provided about the nature of the mutation that gave rise to the blue iris allele, determine the likelihood of a backward mutation occurring in a genome to create a brown eye allele starting from a blue eye one. What sort of mutation would need to happen? Is this difficult to achieve?
24. A recent survey of published work from 1930 to 1990 showed that the non-paternity rate is currently about 2–3% in western society (not the 10% commonly reported by popular and scientific literature) (Voracek et al., 2008). Meanwhile, the estimated rate of mutations in humans is 2.5×10^{-8} mutations per nucleotide site or 175 mutations per diploid genome per generation (Nachman & Crowell, 2000). Weigh this evidence in determining whether you believe that Ryan is Evan’s son.
25. While most eye color researchers accept that two blue-eyed parents can have a brown-eyed child, most of the original reports in the scientific literature date back to the early 1900s. Here is an example: “*Blue x blue as a rule gives only blue. However the rule is not without exceptions. In one case a man with brown eyes was born of blue-eyed parents. Another instance was communicated by a gentleman who stated that both his parents had light blue eyes. Of their seven children all had blue eyes except one sister whose eyes were described as brown or dark hazel. Pearson gives one instance in which one member of a family of six children born of blue-eyed parents had dark brown eyes like those of the father’s maternal grandfather*” (Holmes & Loomis, 1909). What’s missing from this type of evidence to be convincing? How would this type of phenomenon be investigated today?
26. According to a 2002 Loyola University study in Chicago (Grant & Lauderdale, 2002), almost 60% of people born in North America at the turn of the last century had blue eyes. By mid century, that number had dropped to a third. Today, it is 1 in 6 Americans (Belkin, 2006; Starr, 2010). Propose a hypothesis to explain this phenomenon.
27. Assuming that the mutation that causes people to have blue eye arose 6,000 years ago (Eiberg et al., 2008; “*Blue-eyed humans*,” 2008), and given that at the turn of the last century more than half of Americans had blue eyes, it can be assumed that this mutation is extremely successful. It must confer an advantage. Hypothesize why evolution might have favored the selection of blue eye color. What advantage might it confer? Think of the different mechanisms of evolution that could result in this observation. For explanations that rely on natural selection, specify whether it may be a case of sexual selection.
28. How could this information about the genetics of eye color be used by forensics experts and archeologists? What are some of the challenges of using this technique in these fields?

29. What are some potential implications of the popular state of knowledge about the genetic determinants of eye color?

References

- Belkin, D. (2006). Blue eyes are increasingly rare in America. *The New York Times*. Retrieved 2 April 2010 from http://www.nytimes.com/2006/10/18/world/americas/18iht-web.1018eyes.3199975.html?_r=1
- “Blue-eyed humans have a single common ancestor” (2008). Science Daily Website. Retrieved 2 April 2010 from <http://www.sciencedaily.com/releases/2008/01/080130170343.htm>
- Eiberg, H., Troelsen, J., Nielsen, M., Mikkelsen, A., Mengel-From, J., Kjaer, K.W., and Hanse, L. (2008). Blue eye color in humans may be caused by a perfectly associated founder mutation in a regulatory element located within the HERC2 gene inhibiting OCA2 expression. *Human Genetics* 123: 177–187.
- Grant, M.D., and Lauderdale, D.S. (2002). Cohort effects in a genetically determined trait: Eye colour among US whites. *Annals of Human Biology* 29 (6): 657–666.
- Holmes S.J., and Loomis, H.M. (1909). The heredity of eye color and hair color in man. *Biological Bulletin* 18(1): 50–65. Retrieved 4 April 2010 from <http://www.jstor.org/stable/1535989>
- Kayser, M., Liu, F., Janssens, A.C., Rivadeneira, F., Lao, O., van Duijn, K., et al. (2008). Three genome-wide association studies and a linkage analysis identify HERC2 as a human iris color gene. *American Journal of Human Genetics* 82(2):411–23.
- Nachman, M.W., and Crowell, S.L. (2000). Estimates of the mutation rate per nucleotide in humans. *Genetics* 156: 297–304.
- Sha, K. (2004). Ask a geneticist: How did I get green eyes? Understanding Genetics, The Tech Museum, Stanford University School of Medicine. Retrieved 2 April 2010 from <http://www.thetech.org/genetics/ask.php?id=29>
- Starr, B. (n.d.) Ask a geneticist: Can blue-eyed parents have a baby with brown eyes? Understanding Genetics, The Tech Museum, Stanford University School of Medicine. Retrieved 2 April 2010 from <http://www.thetech.org/genetics/ask.php?id=101>
- Starr, B. (2010). Ask a Geneticist: Why are blue eyes so uncommon anymore? Understanding Genetics, The Tech Museum, Stanford University School of Medicine. Retrieved 4 May 2010 from <http://www.thetech.org/genetics/ask.php?id=355>
- Sturm, R.A., Duffy, D.L., Zhen Zhaw, Z., Leite, F.P.N., Stark, M.S., Hayward, N.K., Martin, N.G., and Montgomery, G.W. (2008). A single SNP in an evolutionary conserved region within intron 86 of the HERC2 gene determines human blue-brown eye color. *American Journal of Human Genetics* 82: 424–431.
- Voracek, M., Haubner, T., and Fisher, M.L. (2008). Recent decline in nonpaternity rates: a cross-temporal meta-analysis. *Psychological Reports* 103(3): 799–811.



Image credits: Photo in title block © Alexshalamov | Dreamstime.com. Illustrations on pages 2 and 7 by the author.

Case copyright held by the **National Center for Case Study Teaching in Science**, University at Buffalo, State University of New York. Originally published January 13, 2011. Please see our **usage guidelines**, which outline our policy concerning permissible reproduction of this work.