NATIONAL CENTER FOR CASE STUDY TEACHING IN SCIENCE

A Family in Need: In-Class Case Study on Cancer Genetics

by
Janet A. De Souza-Hart
School of Arts & Sciences
Massachusetts College of Pharmacy & Health Sciences, Boston, MA

Instructions: Using the information that you have studied in class and in your textbook, complete the following questions.

Part I – Genetic Testing

You are a second-year medical student in an innovative medical school that allows you to get in-depth clinical experience early in your medical education. You are currently assigned to work with Dr. Aikenhed, a pediatric oncologist. She has done a biopsy on a tumor-like growth in the adrenal gland of her 17-year-old male patient, Lee F. You write down the following notes while Dr. Aikenhed takes a detailed family history from the patient's parents:

- Lee has a sister, Leah (age 10), and a brother, Luke (age 6). Both are healthy.
- Lee's mother, Grace, was diagnosed with bilateral breast cancer last year at age 35. Lee's father, Brian, and Lee's paternal grandparents have no history of cancer.
- Grace is the youngest of four children. Her eldest brother, Greg (age 42), and sister, Greta (age 40), have never had any signs or symptoms of cancer. She had a brother, Geoff, who died of leukemia at age 8. He was the third child of Grace's parents.
- Greg's two fraternal twin daughters have no signs or symptoms of cancer.
- Grace's father, Roger, died of a soft tissue sarcoma at age 35. Grace's mother, Renee, is still living and in excellent health.
- Roger's mother died of a brain tumor at age 30. Roger was her only child.

Questions

1. Draw the pedigree for this family in the space provided below. Make sure to indicate Lee, the proband, with an arrow and use the proper symbol for individuals who have died from cancer. Remember to also label each generation with roman numerals.

It is clear from the pedigree that there is a pattern. You strongly suspect that the high incidence of cancer in this family is caused by an inherited genetic defect. Due to the severity of the phenotype, the defective allele is probably rare in the population.

2. Complete the following table to help organize your thoughts. Indicate in the table if a mode of inheritance is definitely possible, possible but unlikely, or not possible based on the pedigree. Also briefly explain your rationale behind these choices in the space provided.

Table 1 – Determining Mode of Inheritance

Mode of Inheritance	Possible	Possible but Unlikely	Not possible	Brief Rationale
Autosomal recessive				
Autosomal dominant				
X-linked recessive				
X-linked dominant				
Y-linked				
Mitochondrial				

3. What mode of inheritance pattern *best* fits this pedigree in your opinion?

In order to impress Dr. Aikenhed, you begin to research genetic disorders that can cause adrenal cancer. A quick search of the Genetics Home Reference (http://ghr.nlm.nih.gov/) using the key words "autosomal dominant adrenal cancer" gives seven results. You decide to rule out two immediately since they do not usually cause malignant tumors. Here is your summary of the clinical symptoms for the other five, including the OMIM (Online Mendelian Inheritance in Man database) number:

Table 2 – Clinical & Genetic Characteristics of Selected Syndromes Associated with Adrenal Cancer

Genetic Disorder	OMIM #	Gene(s) involved	Clinical manifestations
Multiple endocrine neoplasia	131100 & others	*MEN, RET* or *CDKN1B*	Tumors of the parathyroid/pituitary glands and pancreas; kidney stones, hypertension, fatigue, vomiting, nausea.
Carney complex type I	160980	*PRKAR1A*	Signs/symptoms commonly begin in teens/early adulthood. Changes in skin pigmentation (brown spots), heart tumors, tumors in endocrine tissues (thyroid, testes, ovaries)
Li Fraumeni syndrome	151623	*TP53*	Breast, bone and soft tissue cancers common. Cancers of blood-forming tissues and adrenocortical carcinomas are possible among others.
Neuroblastoma	256700	*KIF1B* & others	Most often affects children under age 5. Tumor originates in adrenal gland but can also form in nerve tissue of abdomen, chest, and pelvis. Can metastasize to bone, liver, skin. Fatigue, pain, loss of appetite, and more possible.
Von-Hippel-Lindau syndrome	193300	*VHL*	Tumors (both benign and malignant) and cysts in various locations (kidneys, pancreas, male genital tract, inner ear), non-cancerous blood vessel tumors.

4. What kind of information can you look up using the OMIM number for a genetic disorder?

5. Based on the information from *both your patient and his family,* what genetic disorder should you most suspect?

6. What is the OMIM number for this disorder?

7. What gene(s) is/are most often defective in this genetic disorder?

8. Dr. Aikenhed shares your suspicion. She then shares with you her concerns about Leah and Luke, despite the fact that they do not appear to have cancer. Why is she concerned?

9. With respect to this genetic disorder, would it be possible for a person with cancer to have two parents who do *not* have cancer? Explain.

You are worried about sharing your suspicions with Lee and Grace, but for different reasons. Grace, who is still recovering from breast cancer surgery and chemotherapy, seems very fragile as well as distraught that her son has a tumor-like growth. If Lee did, in fact, inherit a faulty gene from her that will predispose him to cancer, you are worried about how Grace will handle the news. She seems at great risk for deep feelings of guilt and serious depression. Lee, on the other hand, is a very upbeat young man with a great sense of humor. He seems mostly concerned about improving his mother's spirits. You would dread being the one to tell him if it turns out that he has a genetic condition that will predispose him to various types of cancers for his entire life. In order to best communicate the pros and cons of genetic testing to them, you review the different types available.

10. Here are four general categories of tests along with four specific examples. Give the letter corresponding to the correct example after each category:

    Pre-implantation Genetic Diagnosis ____

    Carrier Testing ____

    Predictive (pre-symptomatic) Testing ____

    Prenatal Testing ____

    Examples:

    A) Two parents are interested in learning their chances of having a child with Tay-Sachs disease.

    B) A 30-year-old man with a family history of Huntington disease wants to know if he carries the defective allele.

    C) Amniocentesis is performed in order to determine if a fetus has a major chromosomal disorder.

    D) A couple with achondroplasia want to ensure that they have a child who is unaffected by this condition.

11. What types of issues come up when determining whether to *test* someone for a genetic illness?

12. If Lee has inherited a gene from his mother that predisposes him to cancer, would you recommend that the family test Leah and Luke at this point? Why or why not?

Part II – Applications of DNA Technology

While you are waiting for Lee's biopsy results, you decide to do some concentrated studying for the USMLE Step 1, the exam that all second-year medical students must take to progress in medical school. Since you want to find a clear and straightforward way of explaining to Lee how genetic testing works, you review material related to genetic diagnosis, prevention, and treatment when you get home.

Questions

1. In order to test Lee for a genetic disorder, Dr. Aikenhed will probably take a sample of cells containing his genomic DNA by swabbing his cheek or taking a blood sample. Why is it important to use non-cancerous cells to test Lee for a genetic disorder instead of the biopsy tissue?

2. Describe the main technique for amplifying a segment of DNA (like the one you suspect is involved in Lee's cancer) from a complex mixture of genomic DNA. Remember that the entire human genome sequence is known. (*Hint:* This is a technique that is commonly used by laboratories that do genetic testing and various other applications of molecular biology.)

3. If Dr. Aikenhed wanted to see if there was mutation within the protein-coding sequence of the gene implicated in this disorder (as opposed to mutations affecting regulatory elements), what technique *involving dideoxynucleotides* could be used? Briefly describe this technique.

Lee and his family have inspired in you a desire to find out more about how to prevent and treat this disorder. In your medical genetics textbook, you read that some genetic disorders have specific treatments and/or prevention strategies. The symptoms and progression of phenylketonuria (PKU), for example, can be mitigated with a strict diet that limits phenylalanine intake. Cystic fibrosis is treated with palpitations (tapping) of the chest to loosen mucus and with antibiotics to help prevent infection, among other strategies. Most genetic disorders, however, do not have such clear cut recommendations.

4. Frequent screening/check-ups and avoidance of risk factors (e.g., smoking) may be advised in cases involving inherited genes which predispose a person to cancer. Do you think that this advice will make a difference in morbidity or mortality related to this particular disorder? Why or why not?

5. What is the name of the genetic technique that would help a person who inherited this genetic disorder to have a child without the defective allele?

Part III – Gene Expression and Disease

Unfortunately, the results from Lee's biopsy are in and it appears that he has adrenocortical carcinoma (ACC). Fortunately, Dr. Aikenhed (who, in addition to her clinical practice in the area of oncology also conducts research) is on the cutting edge of developing treatments for this condition. She is part of a consortium of researchers that are trying to catalog all of the different genes that may be involved in adrenal cancer. This will help physicians and scientists to discover drugs to target specific cancer-contributing gene products.

Cancer genes can be categorized in many different ways. *Proto-oncogenes* produce proteins that are involved in promoting the cell cycle. Mutations in these genes tend to cause either an over-production of protein (over-expression) or a protein to be constitutively active (pushing the cell cycle forward even in the absence of a signal to divide (e.g. growth factor)).

Tumor suppressor genes, on the other hand, put the brakes on the cell cycle, usually at key checkpoints. Mutations in tumor suppressor genes that contribute to carcinogenesis can cause either abnormally low levels of protein or no functional protein (under-expression) at all. A mutation like this could allow a damaged or rogue cell to divide when it shouldn't.

Genome-maintenance genes can also play a role in cancer and they include the telomerase gene (which helps extend the life of a chromosome and, consequently, the number of cell division cycles that it can undergo) and DNA repair genes (which keep the level of mutations in all genes low).

Dr. Aikenhed and her colleagues use microarrays to analyze gene expression (both over- or under-expressed genes) in cancer samples compared to normal tissue. You decide to review how microarrays work using a great tutorial from the Howard Hughes Medical Institute (http://www.hhmi.org/biointeractive/how-analyze-dna-microarray-data). It never ceases to amaze you how they can put thousands of tiny pieces of single-stranded DNA on these microarray chips in an organized way, creating all sorts of different sequences for samples to hybridize (bind) to.

Several microarrays were made with the most common genes implicated in ACC. DNA samples from six patients with ACC were analyzed. Lee F. is Patient #3. The last row represents a control using normal adrenal tissue from a healthy individual.

Table 3 – Different Genes* Implicated in ACC

	IGF2	TP53	RAS	BCLXL	CHRB	APC	DDB1	MRPL48	EGRF1	CTNNB1
Patient 1	R	Y	Y	Y	G	R	Y	Y	Y	Y
Patient 2	R	Y	Y	G	Y	R	Y	Y	Y	Y
Patient 3	Y	G	Y	Y	Y	Y	Y	Y	Y	Y
Patient 4	Y	G	Y	Y	Y	R	Y	Y	Y	R
Patient 5	Y	Y	R	Y	Y	Y	Y	Y	G	Y
Patient 6	Y	G	Y	Y	G	R	Y	Y	Y	Y
Control	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y

*gene symbols are consistent with HGNC & OMIM
R=red (high expression in diseased compared with normal cells)
G=green (low expression in diseased compared with normal cells)
Y=yellow (the same expression in both cell types)

Questions

1. What can you conclude about gene expression in your patient's cells? How is gene expression in your patient different than the other 5 patients?

2. The most common mutation for the genetic disorder affecting this family (occurring in ~72% of cases) involves the substitution of one amino acid for another in the protein. This type of change to the protein coding sequence is called a _____ mutation.

3. There are two other types of common *point* mutations that affect the coding sequence of various proteins. Briefly describe all three types of mutations in Table 4 and include whether you predict that the level of mRNA and protein might be affected (increased or decreased).

Table 4 – How Different Mutations Might Affect Gene Expression

Name of mutation	How does mutation alter mRNA sequence?	Could this mutation affect the level (increase or decrease) of functional *protein* in the cell? Why?
	substitution of one amino acid for another in the protein	

Sometimes mutations involve the deletion of large regions of genetic information. Other mutations affect gene expression by altering the DNA sequence of regulatory elements like promoter regions. Further analysis reveals that your patient appears to have a mutation that affects mRNA splicing (removal of introns and splicing together of various exons), which occurs approximately 8% of the time in this condition. This mutation reduces the level of mature mRNA in his cells. This is why the level of the affected gene shows a lower expression level than controls in your patient, Lee.

Part IV – Gene Therapy

Despite trying to maintain a professional level of emotional distance, you have become attached to Lee and his good natured family. You spend evenings searching the medical literature for potential treatments, including clinical trials, to help them. You remember learning about gene therapy trials for SCID (Severe Combined ImmunoDeficiency) and LCA (Leber's Congenital Amaurosis—a type of hereditary blindness) in the *New England Journal of Medicine*.

Questions

1. How would you describe the main goal of gene therapy to this family?

2. Several different types of viruses, including adenoviruses, have been used as vectors for gene therapy in humans. Why are viruses used for gene therapy?

3. The two basic types of gene therapy are _____ and _____. What is the main difference between them?

4. Which type would best help this patient and his family? Why?

5. What types of risks/problems are currently associated with gene therapy?

You discover that there have been gene therapy trials involving *TP53*. For a variety of reasons, however, the treatment has not been approved by the FDA (U.S. Food & Drug Administration) and the trials stalled. The main problem appeared to be that the company developing the treatment did not provide enough evidence to the FDA that the treatment was effective.

6. If there were a current *TP53* gene therapy clinical trial, would you recommend that Lee enroll in it? Why or why not?

7. Write a brief, encouraging letter addressed to Lee and his family that highlights the hope of a future gene therapy cure for the disorder affecting them. Make sure you explain to them what gene therapy is and what your opinion is on the potential for it to cure LFS.

For more information on gene therapy:

Aiuti, A., et al. 2009. Gene therapy for immunodeficiency due to adenosine deaminase deficiency. *New England Journal of Medicine* 360: 447–458.

Bainbridge, J.W.B., et al. 2008. Effect of gene therapy on visual function in Leber's congenital amaurosis. *New England Journal of Medicine* 358: 2231–2239.

Coune, P.G., Schneider, B.L., and Aebischer, P. 2012. Parkinson's disease: Gene therapies. *Cold Spring Harbor Perspectives in Medicine* 2(4):a009431.

Ortiz R. et al. 2012. New gene therapy strategies for cancer treatment: A review of recent patents. *Recent Patents on Anti-Cancer Drug Discovery* 7(3): 297–312.