



CYSTIC FIBROSIS
Genetic Counseling
Teacher instructions and answers

In this exercise students will play the role of genetic counselors. First they will construct a pedigree from a family who has a member of CF. Then they will calculate the odds that different family members will have CF or will carry CF. Based on this information they will decide whether to recommend genetic counseling to the couple. Then students will run gels of the DNA from the couple and analyze the results. They will then decide how to discuss their findings with the couple.

This exercise assumes the students are already familiar with basic Mendelian genetics and patterns of inheritance. It also assumes that students are familiar with terms such as gene, allele, and locus.

Students are first asked to construct a pedigree. An example pedigree is given in the exercise. If students need more help in how to construct a pedigree, an accompanying handout goes over this process in more detail.

Similarly, students are asked to calculate the odds that an individual will be a CF carrier given their family history. Again, if students need more help with this process, an accompanying handout goes over some examples in more detail.

The general instructions for the gel electrophoresis are given in an accompanying handout. This handout tells where to buy supplies as well as how to make and run the gels.

Below is a description of how to make the electrophoresis samples for this exercise. This information is not included in the gel electrophoresis handout.

Preparation of Samples for Gel for CF testing

Set 1 - Jeff and Susan are both carriers

- 1A. xylene cyanole only
- 1B. bromphenol blue only
- 1C. bromphenol blue and xylene cyanole
- 1D. bromphenol blue and xylene cyanole

Set 2 - Jeff is a carrier, Susan is not a carrier

- 2A. xylene cyanole only
- 2B. bromphenol blue only
- 2C. bromphenol blue and xylene cyanole
- 2D. xylene cyanole only

Summary

In this exercise, you will play the role of a genetic counselor. Susan and Jeff plan to have a child soon, but are concerned that a relative of Jeff's died of cystic fibrosis.

Goals

- Be able to construct a pedigree
- Review dominant and recessive inheritance patterns
- Given a pedigree, calculate the probability that a given individual will have a genetically inherited disease, or be a carrier for that disease.
- Understand how genetic testing works
- Be aware of what questions genetic testing can answer, and what questions it can't answer
- Be able to discuss different ethical issues surrounding genetic testing including
 - the values of genetic testing
 - how to use the results of genetic testing
 - the limitations and negative aspects of genetic testing

Background

You are a genetic counselor. Susan and Jeff have been referred to you because they want to have their first child soon. A relative of Jeff's died of cystic fibrosis (CF), and he is concerned about the risk of having a child with cystic fibrosis. In cystic fibrosis, the mucus lining the respiratory and gastrointestinal tracts is extremely thick, making it for individuals with this disease to fight infections or absorb nutrients. CF individuals used to die within the first years of life. Over time, as advances in diagnosis and treatments have been made, the life expectancy has lengthened. It is currently 31 years for males and 29 years for females.

Cystic fibrosis (CF) has an autosomal recessive pattern of inheritance. If you are unfamiliar with patterns of inheritance, review the information in the box on the next page. About 1 in 25 Caucasians carry one mutant cystic fibrosis gene. The rate is lower for Hispanics (1 in 40), Blacks (1 in 60), and Asians (1 in 90). About 1 in 2500 Caucasian babies born will have cystic fibrosis.

There are over 600 different CF-causing mutations that have been discovered to date in the CFTR gene (the gene responsible for CF). There is currently a genetic test that screens for the 70 most common mutations. Depending on ethnic background, this test can detect 30-97% of all CF carriers. This means that a positive result definitely indicates the person is a carrier. However, a negative result does not necessarily mean the individual is not a carrier. The test is most effective for Caucasians and Ashkenazi Jews since it detects the mutations most commonly found in these populations. Soon there will be a genetic test that screens for the 90 most common mutations.

Patterns of Inheritance

Autosomal dominant - The mutation is not located on the X or Y chromosome and is dominant over other alleles at this locus. This means that individuals with only one copy of the mutant gene will have the disease. With diseases that are autosomal dominant, 50% of an affected parent's offspring are affected, and males and females are affected with equal frequency.

Autosomal recessive - The mutation is not located on the X or Y chromosome and is recessive to other alleles at this locus. This means that individuals who carry one copy of the mutant gene are unaffected, although they are carriers, and individuals with two copies of the mutant gene will have the disease. For diseases that are autosomal recessive, 25% of the offspring of 2 carriers are affected, and males and females are affected with equal frequency.

Sex-linked - The mutation is located on the X chromosome. Since males have only one copy of the X chromosome, sex-linked diseases are carried by females and almost always affect males.

Step 1 - Take a family history

You ask both Jeff and Susan to list the relatives they know of, and whether each of these relatives had cystic fibrosis or was determined to be a carrier through genetic testing.

Jeff's family

Ethnicity - Caucasian

His Aunt Beth (his mother's sister) died of CF.

None of his other relatives have cystic fibrosis (listed below)

Joe - Father (there is no family history of CF in Joe's family)

Helen - Mother

Paul and Brian - 2 brothers

1 sister, Cindy was tested to see whether she was a CF carrier. She does not carry the CF mutation.

4 grandparents

Uncle Tom (mother's brother, known to carry CF, his wife does not carry CF)

Uncle Tom's 3 sons (John, Eric, and Steve)

Susan's family

Ethnicity - Hispanic

There are no relatives in Susan's family with cystic fibrosis.

The rate of carrying the CF gene is shown below for several ethnic groups

Caucasians - 1 in 25

Hispanics - 1 in 40

Blacks - 1 in 60

Asians - 1 in 90

Step 2 - Construct a pedigree

First, read the example below and note how the pedigree is constructed. Then, using the information about Jeff's family, construct a pedigree including all family members.

Example of a pedigree

A few weeks after Tania was born, she was diagnosed with cystic fibrosis. Her parents, Helen and Dave, had two additional children, Christy and Mark, and had each tested in vitro. Neither has CF. Mark is a carrier, Christy is not. When Dave's sister Josephine found out that Tania had CF, she immediately had a sweat test done on her son Joe. Joe had a negative sweat test. She then had herself tested genetically to see if she carried CF. She tested negative for CF genes, as did her husband Steve. Josephine and Steve later had a daughter, Sue. Dave's other sister Mary and Helen's brother Bob do not have CF and have not had themselves tested.

Key to symbols

square = male

circle = female

open symbol = does not have or carry CF

filled symbol = has CF (2 copies of mutant gene)

symbol with a line through it = carries CF (1 copy of mutant gene)

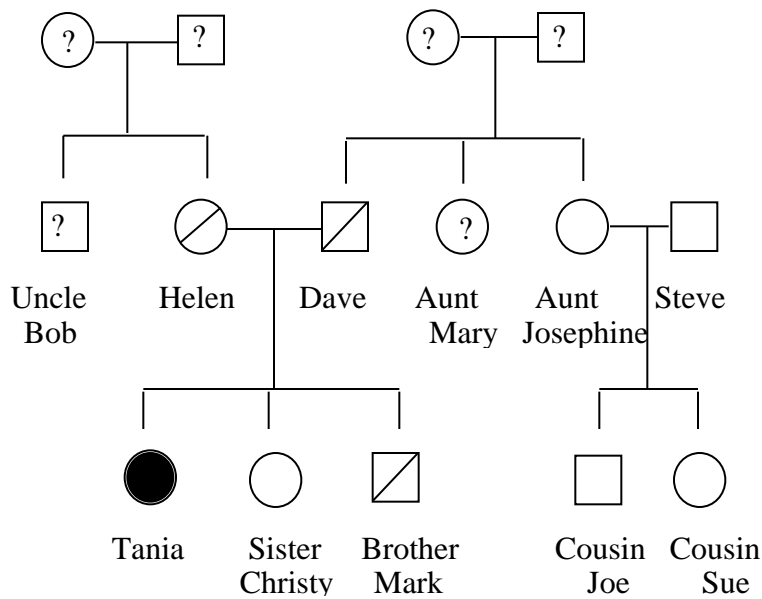
+ = normal CF gene

- = mutant CF gene

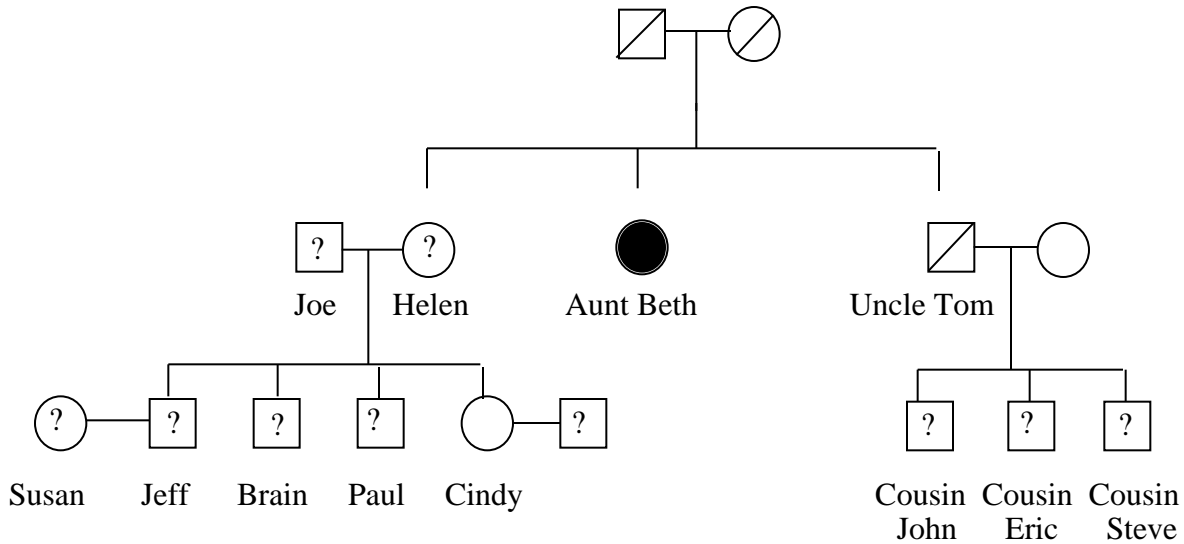
? in symbol = genotype of individual unknown

horizontal lines connecting two symbols indicate couples

symbols connected by a line above them are siblings and have the same parents



Now, using the information about Jeff's family, construct a pedigree including all his family members. If you need additional help, consult the handout, constructing a pedigree.



Step 3 - Calculate the probability that different members of Jeff's family are carriers

If you need additional help calculating the probabilities, consult the accompanying handout "Inherited Diseases: Calculating Probabilities"

What is the probability that Jeff's father is a carrier?

1/25 or 4% - the rate of carrying CF in the Caucasian population (you could choose to change this for other ethnic backgrounds)

What is the probability that Jeff's mother is a carrier? Note that Helen herself does not have CF.

2/3 or 67%
 (Note that the answer is not 1/2 as you would get if you solved this mathematically. This is because we know that Jeff's mother does not have cystic fibrosis so that possibility is eliminated.)

If both of Jeff's parents are carriers, calculate the probability that Jeff himself is a carrier. Note that Jeff himself does not have CF.

2/3 or 67%
 (Note that the answer is not 1/2 as you would get if you solved this mathematically. This is because we know that Jeff does not have cystic fibrosis so that possibility is eliminated.)

What is the probability that Susan is a carrier?

1/40 - the rate of carrying CF in the Hispanic population

Step 4 - Discuss the test with Jeff and Susan

Will you recommend genetic testing for Jeff? for Susan? Whom would you test first and why? Would you recommend they be tested for other genetically inherited diseases such as sickle cell anemia or Tay Sach's, at the same time? What issues would you discuss with them in helping them decide whether to be tested for CF or for additional diseases?

From my visit to a genetic counselor

- *They will offer to test Jeff and Susan to see whether they carry a CF gene.*
- *They will not test for other genetically inherited diseases at the same time unless there is reason to suspect that someone is at particularly high risk due to the high cost of these tests.*
- *In discussing testing for additional diseases it would be important to ask about ethnicity and family history of other genetically inherited diseases. If only one partner is from a ethnic group with a high rate of carrying a disease, testing is not usually performed. However, if both partners are from a high-risk ethnic group, testing might be considered.*
- *In discussing CF testing it would be important to point out that the test does not detect all mutations leading to CF. Thus, a positive result for a CF gene could identify someone as a carrier, however, a negative result for a CF gene does not guarantee someone does not carry a CF mutation.*
- *A genetic counselor would discuss the disease, its consequences, and the type of life an individual with CF could have with the parents. This will allow the parents to make an informed decision as to whether to have the test and what they would like to do with test results.*

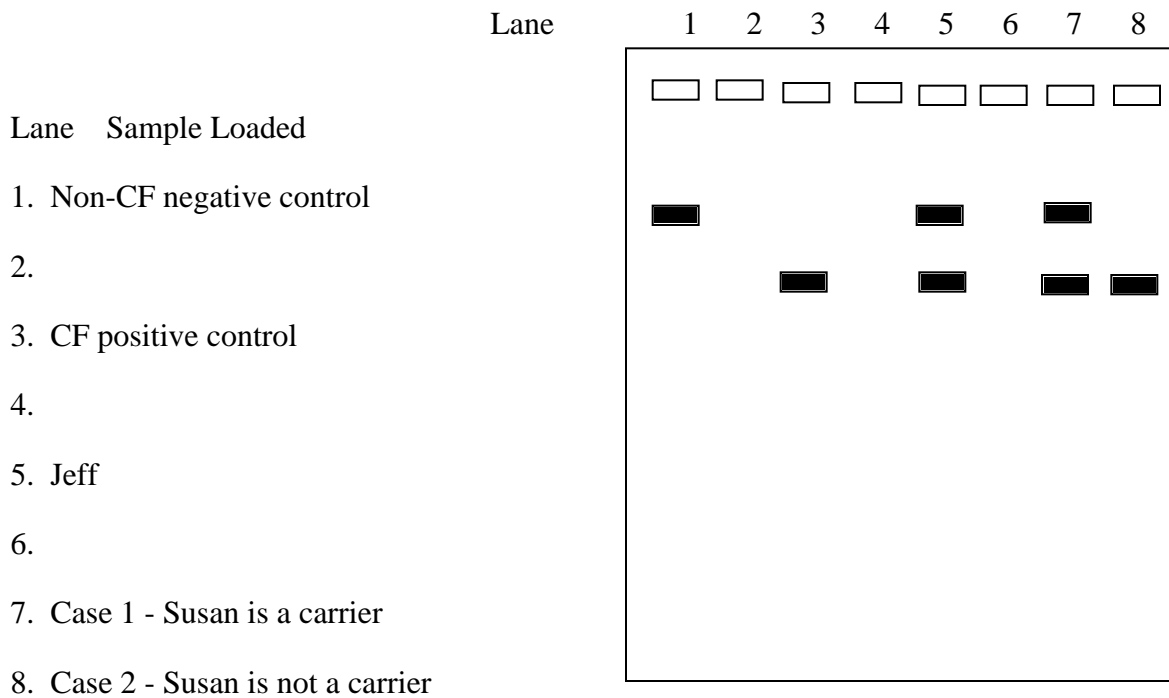
Step 5 - Perform the genetic test

Jeff and Susan have decided to go ahead with genetic testing. You take a sample of cheek cells from each of them. You send the cheek cell samples off to a lab where they isolate the DNA and run the reaction necessary to show the CFTR gene. You then receive 4 samples to run on a gel in order to analyze the results. The key for the labeling is shown below.

- A. Negative Control - DNA known to contain the normal allele of the CFTR gene
- B. Positive Control - DNA known to contain a mutant form of the CFTR gene
- C. Jeff's DNA
- D. Susan's DNA

Load the samples into separate wells of the gel. Be sure to note which sample you put in which well in the space provided on the next page. Run the gel until you can see a clear separation of bands. Then, sketch your gel in the box provided on the next page.

Results



Step 6 - Interpret the results by answering the following questions

Which allele of the CFTR gene (normal or CF mutant) is smaller (a shorter piece of DNA)? Why?

The mutant allele is shorter because it migrates further in the gel

Why is it important to run a negative control when doing genetic testing?

To be sure you know the position to which the normal allele will migrate

Why is it important to run a positive control when doing genetic testing?

To be sure you know the position to which the mutant allele will migrate

Which allele(s) does Jeff have (use + for the normal gene, - for the mutant gene)?

+/- or one normal/one mutant

Is Jeff a CF carrier?

Yes

Which allele(s) does Susan have?

Case 1 - +/- or one normal/ one mutant

Case 2 - +/+ two normal alleles

Is Susan a CF carrier?

Case 1 - yes

Case 2 - no

What is the probability their first child will be a CF child? a CF carrier?

Case 1 - CF child, 1 in 4 or 25% ($1/2 \times 1/2$) CF carrier, 1 in 2 or 50%

*Case 2 - CF child, close to 0 but not 0 since the genetic test can not find all possible CF mutations
CF carrier, 1/2*

If their first child does not have CF and is not a carrier, what is the probability that their second child will have CF? be a CF carrier?

The same as above. The state of the first child does not influence the probabilities for later children.

Step 7 - Discuss the results with Jeff and Susan

Make an outline of what you would discuss with Jeff and Susan?

Include the results of this test, and what it means with respect to the chances that Jeff and Susan will have a CF child.

Be prepared to present your results to the class.