Inheritance of Hemophilia
About NHF
The National Hemophilia Foundation is dedicated to finding the cures for inherited bleeding disorders and to preventing and treating the complications of these disorders—through education, advocacy, and research.

Inheritance of Hemophilia by Connie Miller, PhD

Acknowledgments
We thank the many individuals who reviewed drafts of and provided guidance for this publication, including Carol Kasper, MD, and Robert Resta, MS. We are grateful to Sharon Cross, PhD, for her extensive contributions to the manuscript and to Marion Koerper, MD, for her insightful comments.

This publication was developed under the supervision of the Educational Materials Working Group of the National Hemophilia Foundation, Casey Hannan, MPH, Chair.

Managing Editor: ..................Steven Humes, MPH
Editorial Coordinator: ..............Maribel Johnson, RN
Graphic Designer: ....................Eliot Weinstein
Printing: ................................Marko Press

This publication was made possible by an educational grant from:
Centers for Disease Control and Prevention.

CDC

This material is provided for your general information only. NHF does not give medical advice or engage in the practice of medicine. NHF under no circumstances recommends particular treatments for specific individuals and in all cases recommends that you consult your physician or local treatment center before pursuing any course of treatment.

©1998 National Hemophilia Foundation. Material in this publication may not be reproduced without written permission from the National Hemophilia Foundation.
Hemophilia is a *hereditary disease*—one that can be passed down through families or *inherited*. This condition is caused by changes in the genes (units of inherited information) that make *proteins* needed for blood clotting. These genes, like all genes contained in cells, are passed from parents to children. Just as a parent may sometimes pass a physical trait such as a dimple to a child, so, too, a parent may pass the gene that results in hemophilia to a child. In this booklet, the phrase "hemophilia gene" will sometimes be used as a shorthand term for the changed clotting protein genes that can cause hemophilia.

Until recently, there was no way to know which women might carry the genes for hemophilia or which children might be born with the condition. In the last few years, scientists have found new ways to test for hemophilia genes. This publication discusses how hemophilia is inherited. It also describes tests used to find out if the hemophilia gene is present, particularly in women who may carry the gene (*carriers*) but show no signs of excessive bleeding. Reproductive choices for men and women with the hemophilia gene are also reviewed.

*Terms appearing in italics are defined in the glossary on page 14.*
The Inheritance of Hemophilia: A History

Hemophilia has been known for hundreds of years to "run in" families. Jewish law written 1500 years ago did not allow the circumcision of a baby boy if two older brothers had bled to death after the procedure. Even then, it was understood that a brother of a male child with a bleeding problem was at high risk for having a similar condition.

In more recent times, hemophilia became widely known through its presence among members of many European royal families. All of the affected male members of the European royal families were direct descendants of Queen Victoria of England, the first known hemophilia carrier in her family. Queen Victoria's family tree is shown in Figure 1.

In the United States, the transmission of hemophilia from mothers to sons was first described in the early 1800s. In the 1980s, hemophilia made headlines as one of the first human disorders for which the responsible genes had been identified and cloned (copied in the laboratory). The discovery of the genes for hemophilia has helped pave the way to better methods for detecting these genes in people who do not show signs of excessive bleeding.

The Causes of Hemophilia

There are at least ten proteins in the blood that must work in a precise order to make blood clot. These proteins are called clotting factors. A change in any of these factors can lead to abnormal bleeding. There are many types of inherited bleeding disorders, each caused by a defect in one of the factors needed to form clots. This publication describes only two of these bleeding disorders, hemophilia A and hemophilia B.

Hemophilia A, or classic hemophilia, is caused by a defect in a clotting protein called factor VIII (eight). Hemophilia B is caused by a defect in another protein necessary for clotting, factor IX (nine). Hemophilia B is sometimes called "Christmas disease"; Christmas was the last name of the first person described with this disorder.

Depending on the exact defect in the clotting factor gene, hemophilia can be mild, moderate, or severe. All affected individuals in a given family have the same change in their factor VIII or factor IX gene. Accordingly, the disease usually has the same severity in different members of the same family.

Hemophilia A and B have similar symptoms and are inherited the same way. In fact, they were only first recognized as separate disorders in 1952. As used throughout this booklet unless otherwise noted, "hemophilia" refers to both hemophilia A and hemophilia B.

---

Figure 1. Queen Victoria's family tree.
How Genetic Traits are Inherited

All the genetic information needed to form a new human being is contained in the sperm cell it receives from its father and the egg cell it receives from its mother. This information determines physical traits or characteristics such as hair color, eye color, blood type, and height as well as less apparent features such as the proteins needed for the body to function.

The information for inherited traits is contained in units called genes. Genes are made up of the chemical substance deoxyribonucleic acid (DNA), which contains a code that tells the cell how to make proteins. There are thousands of genes in every human cell. They are packaged into tiny threadlike structures called chromosomes. When examined under a microscope, each chromosome has a unique appearance or size. These differences allow chromosomes to be numbered 1 through 23; the 23rd chromosome is usually referred to as the "sex chromosome."

Each egg cell and each sperm cell contains a single copy of each of the 23 chromosomes. When an egg cell and a sperm cell join together, they form a fertilized egg that contains 23 chromosomes from the mother and 23 chromosomes from the father. As a result, each of the 23 chromosomes now has a mate, for a total of 46 chromosomes. From a genetic viewpoint, this system has a lot of advantages. If one chromosome has a gene that does not work correctly, the other chromosome can often supply a working copy of the gene. The working copy makes up for the information missing in the damaged gene, and thus no harm is done.

Except for egg cells and sperm cells, all cells in the human body contain 23 pairs of chromosomes. Figure 2a shows the chromosomes in a cell photographed, cut out, and arranged in matching pairs. Figure 2b shows a similar pattern except for the final pair of chromosomes. In Figure 2a, the chromosomes in this pair look the same. Both are X chromosomes, and the individual with these chromosomes is thus a female (XX). In Figure 2b, the chromosomes in the last pair are different. One is an X chromosome, and the other, smaller chromosome is a Y chromosome. This individual is a male (XY).

All of a female's egg cells contain an X chromosome for the sex chromosome. In contrast, half of a male's sperm cells contain an X chromosome and half contain a Y. As shown in Figure 3, if an X-bearing sperm fertilizes an egg, the result will be a girl (XX). If a Y-bearing sperm fertilizes an
egg, the XY product will be a boy. The sperm, therefore, determines the sex of the child. At each conception, there is an equal chance of producing a girl or a boy.

How Hemophilia is Inherited

The genes involved in factor VIII and factor IX production are both located on the X chromosome; for this reason, they are sometimes referred to as X-linked or sex-linked. A change (mutation) in one of these genes can cause the clotting factor to be made incorrectly. The result is abnormal blood clotting. Such a genetic mutation results in hemophilia.

Hemophilia occurs almost exclusively in males. This is because males, as noted above, have only one X chromosome. The Y chromosome found in males primarily determines gender; it does not contain genes that make clotting factors. When a male inherits an X chromosome with an altered clotting factor gene from his mother, he will be born with hemophilia because his Y chromosome has no genetic instructions to make up for the clotting factor information that is missing on the X chromosome passed from the mother. In contrast, females have two X chromosomes. An X chromosome with a working copy of the gene that makes clotting factor can mask the gene on the other X chromosome that does not make the clotting factor. Only in rare cases do females have hemophilia as severe as that occurring in males.

Mutations are changes that occur by chance; once they are present in a gene, they can be passed on to offspring. In most affected families, the hemophilia gene has been present for many generations. Occasionally, however, a new mutation arises, and hemophilia appears for the first time with the birth of a baby. About one out of three persons with hemophilia has no family history of the disorder. However, in most cases the hemophilia gene was carried by one or more previous generations. In some families, the abnormal gene may have only been passed on to females and so remained undetected. In other cases, affected males may have gone undiagnosed, either because the disorder was a mild form or because they died as infants and the cause of death was never known.

Transmission of Hemophilia by Family Members

The gene that causes hemophilia can be passed down by the mother (maternal transmission) or by the father (paternal transmission). In rare situations, both parents may have altered genes. The pattern of inheritance expected from each of these situations is described and illustrated in Figures 4-6. The following discussion is intended as an overview of how hemophilia is inherited. Genetic counselors can provide additional information and answer questions on this topic.

Women and Bleeding Disorders

Although hemophilia is rare in females, it can occur if the woman or girl has an altered clotting factor gene on both X chromosomes. This may happen if: (1) a girl is born to a man with hemophilia and a woman who is a carrier; (2) one X chromosome with an altered hemophilia gene is inherited from a man with hemophilia or a woman who is a carrier and the other X chromosome acquires a new mutation in the hemophilia gene; or (3) both X chromosomes acquire new mutations in the hemophilia gene. In some cases, girls are born with only one X chromosome. If this X chromosome carries the hemophilia gene, the child will have hemophilia.

Another cause of bleeding in women who carry the hemophilia gene is nonrandom X inactivation. In all female cells, one X chromosome is always "turned off" while the other works normally to make proteins; this is called lyonization. Usually, one X chromosome is turned off in about half the cells, and the other one is turned off in the other half. Sometimes, however, the same X chromosome is turned off in almost all the cells. This is referred to as nonrandom X inactivation. If the X chromosome with a normal clotting factor gene is turned off in most of the cells of a woman who carries the hemophilia gene on the other X chromosome, then the woman will have clotting factor levels similar to hemophilic men in the family. In less severe cases, women who carry the hemophilia gene may make more factor VIII or IX than the hemophilic men in the family, but still less than normal. These women may be at risk for excess bleeding following trauma, surgery, or tooth extractions. Any carrier of hemophilia who has less than 50% factor VIII or IX activity is considered to have mild hemophilia. She can have abnormal bleeding and will require the same treatment as men with similar clotting factor levels.

Some women who are called hemophiliac actually have von Willebrand disease, another bleeding disorder that can closely mimic hemophilia. Some women diagnosed as having von Willebrand disease are actually carriers of hemophilia. Confusion between the two conditions may arise easily. In von Willebrand disease, excess bleeding is a result of a decrease or defect in von Willebrand factor (vWF), a protein that carries factor VIII in the blood and helps factor VIII work properly. When the levels of vWF in the blood are low, there is not enough protein to carry factor VIII and the factor VIII levels also become low. For this reason, von Willebrand disease is sometimes confused with hemophilia A. Usually, though, these disorders can be distinguished by testing for the levels of vWF. People with mild hemophilia and those who are hemophilia carriers usually have normal levels of vWF, whereas people with von Willebrand disease usually have low levels of this protein. However, one form of von Willebrand disease results in normal vWF levels and activity and is difficult to distinguish from hemophilia. Unlike hemophilia, von Willebrand disease affects males and females equally.
Transmission through the Mother

Even though women only rarely have hemophilia, unaffected women can still pass on the gene for hemophilia to their children. Women who carry the gene that causes hemophilia are called "carriers." Like hemophilia, carrier status can be passed down from previous generations or it can be the result of a new mutation. If a new mutation occurs, the presence of the altered gene may only become apparent after the birth of a son with hemophilia.

Women who are genetic carriers of hemophilia have one X chromosome with an altered gene and one X chromosome with a normal gene. As shown in Figure 4, transmission of the chromosome with the affected gene results in a son with hemophilia or a daughter who is a carrier. Transmission of the chromosome with the normal gene results in children who are unaffected by hemophilia: the son will not have the disorder, and the daughter will not be a carrier. There is a 50:50 chance that each son of a carrier will be affected and a 50:50 chance that each daughter of a carrier will be a carrier. These odds are the same for each pregnancy. Having a son with hemophilia or a carrier daughter does not change the risk of having another child with the hemophilia gene.

In some cases, a new mutation may arise during development of the mother's egg cells. Depending on exactly when the mutation occurs, some eggs may have it and others may not. This is called germline mosaicism. In these cases, the mother does not have the mutation anywhere but in her eggs, so tests on the genes in her other cells will not show an altered factor VIII or IX gene. Germline mosaicism can make it difficult to calculate the risk of inheriting the altered gene. Usually, however, not all of the mother's eggs are affected, so the risk of her passing on the condition is less than when the altered gene is carried in all of her cells.

Transmission through the Father

Males have one X chromosome and one Y chromosome. A man who has an X chromosome with the altered gene has hemophilia. Such men always pass the affected X chromosome on to their daughters. If they pass the Y chromosome, the baby will be a male, and he will not have hemophilia. Men who do not have hemophilia themselves cannot transmit it, even if other family members do have the disorder.

Figure 5 shows the results of paternal transmission of an X chromosome carrying the hemophilia gene. Boys born to a father with hemophilia and a mother who is not a carrier will not have the condition because the Y chromosome of the father does not transmit hemophilia. However, because fathers have only one X chromosome, all female offspring born to fathers with hemophilia will inherit the altered gene on the X chromosome and will thus become obligate carriers of hemophilia.

Table 1.

<table>
<thead>
<tr>
<th>A woman is definitely a carrier if she is:</th>
</tr>
</thead>
<tbody>
<tr>
<td>the biological daughter of a man with hemophilia</td>
</tr>
<tr>
<td>the biological mother of more than one son with hemophilia</td>
</tr>
<tr>
<td>the biological mother of one hemophilic son and has at least one other blood relative with hemophilia</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>A woman may or may not be a carrier if she is:</th>
</tr>
</thead>
<tbody>
<tr>
<td>the biological mother of one son with hemophilia</td>
</tr>
<tr>
<td>the sister of a male with hemophilia</td>
</tr>
<tr>
<td>an aunt, cousin, or niece of an affected male related through maternal ties</td>
</tr>
<tr>
<td>the biological grandmother of one grandson with hemophilia</td>
</tr>
</tbody>
</table>

Transmission from Two Affected Parents

Figure 6 shows the possible outcomes of a union between a man with hemophilia and a woman who is a carrier. Sons born to a father with hemophilia and a carrier mother have a 50:50 chance of having hemophilia. Girls also have a 50:50 chance of having hemophilia, because both parents have an X chromosome with the hemophilia gene. The girls who do not have hemophilia will be carriers.

Tests for Determining Carrier Status

There are two important reasons to test females who might carry the hemophilia gene to see if they are carriers: (1) to find out if special medical...
measures might be needed should the woman be injured or require surgery or dental work, and (2) to provide information that may help in making reproductive choices.

Many doctors who treat hemophilia patients recommend that possible carriers have their clotting factor levels tested early in life. Carriers may have lower clotting factor levels than normal and need special treatment during any situation involving bleeding, such as an accident or even dental work. If it has not already been performed, possible carriers should have their clotting factor levels tested before a planned surgery. Possible carriers, including those with normal clotting factor levels, should seriously consider other tests to determine carrier status before beginning to have sexual relations or becoming pregnant. Counseling and education may help the woman, or her family if the female is a child, make decisions affecting her health and reproductive options.

There are several ways to find out if a female is a carrier, these will be discussed in more detail in the following sections. In a few cases, a careful family history may determine if an individual is a carrier. If not, then DNA tests that look either directly or indirectly for altered factor VIII or factor IX genes can often tell if a girl or woman is a carrier. These tests may require a blood sample not only from the woman but also from an affected male member of the family (see below). If DNA methods are not informative, laboratory tests that measure clotting factor levels in the blood may be useful although not as precise. With the help of some or all of these approaches, most females can find out whether they carry the hemophilia gene. In some cases, unfortunately, none of these tests can fully determine whether a female is a carrier. For instance, if germline mosaicism has occurred and the mutation is only found in some of the woman’s eggs, then no useful information will be gained from studies of DNA from the woman’s blood cells.

**Family History**

A detailed family history is an important first step in finding out whether a female is a hemophilia carrier. As shown in Table 1, family history information can indicate whether it is definite or possible that the woman is a carrier. Women whose family history indicate that they definitely carry the hemophilia gene are sometimes referred to as obligate carriers. Women who have a relative or one son with hemophilia may or may not carry the hemophilia gene. All likely or possible carriers can benefit from carrier testing.

**DNA Testing**

The most accurate way to tell whether a woman is a carrier is to examine her DNA for changes that cause hemophilia. DNA-based methods for detecting hemophilia carriers improve every year,

![Diagram](image)

**Figure 4. Maternal transmission of genes that result in hemophilia.** The chromosome with the hemophilia gene is designated X. If a woman carries the hemophilia gene, each time she is pregnant there is the possibility she will produce one of the following: a girl who is a carrier, a girl who does not carry the hemophilia gene, a boy with hemophilia, or a boy without hemophilia.

![Diagram](image)

**Figure 5. Paternal transmission of genes that result in hemophilia.** The chromosome with the hemophilia gene is designated X. If a man has hemophilia, all of his daughters will be carriers, and none of his sons will have hemophilia.

![Diagram](image)

**Figure 6. Transmission of the gene that results in hemophilia from a hemophilic father and a mother who is a carrier.** The chromosome with the hemophilia gene is designated X. If both parents carry the hemophilia gene, there are four possible outcomes: girls who have hemophilia, girls who are carriers, boys who have hemophilia, or boys without hemophilia. Figures 4-6 are adapted with permission from Hematol (June 1994).
so only the general principles involved in these techniques will be discussed here. Questions concerning a certain method or laboratory test should be referred to the provider who recommends the test or to a genetic counselor.

Two types of DNA analysis are available for finding mutations that cause hemophilia: (1) direct detection of the gene mutation, and (2) indirect detection using easy-to-find DNA markers that are located close to the gene mutation. These two approaches are described below.

Direct Detection of Gene Changes

The most straightforward method of finding changes in a gene is to look directly at the gene to see if a mutation has occurred. This is often done by using chemical methods to find out the order of the gene's base pairs (the smallest piece of information in a gene). This order, or gene sequence, is then compared to that of a normal gene. Differences in the gene sequence may point to a mutation that causes hemophilia.

This method works best if the person being tested for carrier status has a living affected male in her family. In these cases, a specific gene change can sometimes be found in the male, and then the female's genes can be examined for the same change. If that mutation is found in the female's DNA, she is a carrier. If it is not there, then in most cases she is not a carrier. (The exception to this rule is germline mosaicism, in which the mutation that causes hemophilia is present only in the woman's eggs and not in her blood cells.) Even in families with no living affected males, direct detection methods may provide helpful information. Sometimes a change is found that is known to occur in other individuals with hemophilia. Such a finding suggests that the woman being tested carries the gene for hemophilia.

Direct detection of mutations through gene sequencing techniques has been used with the most success in persons with hemophilia B.13 If an affected male is available, the specific change can almost always be found. However, if no affected male is available for testing, it can be hard to tell whether differences found in the DNA sequence of a female will cause hemophilia or whether they are simply changes that have nothing to do with hemophilia.

The factor VIII gene is too large to be routinely sequenced by current methods. However, about 50% of individuals with severe hemophilia A have a large (about 10,000 base pairs) portion of the gene that has been flipped around (Figure 7).14,15 This type of change is called an inversion. Inversions disrupt the gene because the cellular machinery can "read" a gene in only one direction. When an inversion occurs, part of the gene's

Finding Mutations

The goal of all genetic tests is to examine the gene in question for signs of changes that can cause disease. The smaller the gene, the easier this is to do. For instance, the gene involved in sickle cell anemia consists of 1600 bits of information (DNA base pairs). By genetic standards, this is fairly small. The genes involved in hemophilia are much larger. The factor IX gene is made up of approximately 34,000 base pairs, and the factor VIII gene consists of about 186,000 base pairs. The factor VIII gene is one of the largest genes analyzed to date.

For some genetic diseases, such as sickle cell anemia, all persons with the disease have the same mutation. This is not true for hemophilia. In one study of 1142 individuals with hemophilia B, 476 different, unique mutations were found scattered throughout the gene.16 Similarly, among 543 persons with hemophilia A, 296 unique mutations were detected.17 This variability in the site of gene changes, together with the large sizes of these genes, can make finding the mutation responsible for hemophilia a bit like "looking for a needle in a haystack." Nevertheless, scientists have had remarkable success in developing methods to look for mutations that result in hemophilia.
because there is a chance that the marker will become separated from the hemophilia gene. In rare cases, pieces of the X chromosome outside the gene can switch places with a piece of the other X chromosome. As a result, the 'marker' ends up on the normal X chromosome. Finally, in families where there is only one person with hemophilia, an indirect marker seldom tells whether the genetic change occurred in the child with hemophilia or was already present in his mother. In these cases, the woman still would not know whether she is a carrier.

**Indirect detection of gene changes**

Indirect detection of gene changes relies on genetic markers-DNA pieces that are easy to identify but that themselves have nothing to do with hemophilia—to track the hemophilia gene through the family. In this type of testing, the mother of an affected male is the key person. Her X chromosomes are checked for known markers that have one pattern on one X chromosome and a different pattern on the other X chromosome. Once such a marker is found, the affected male is checked to see which pattern his X chromosome has; this tells which of his mother's X chromosomes carries the hemophilia gene. His sisters can then be tested to see if they received the same X chromosome that he did. The biologic father should also be tested for the marker pattern on his X chromosome. This knowledge is needed so that the X chromosome the sister got from her mother can be distinguished from the one she got from her father. Because samples from the father help tell whether the daughter has a chromosome with an altered gene, it is very important that the identity of the biologic father is certain. Otherwise, a woman may be told that she is not a carrier, when in fact her DNA pattern is being compared to someone to whom she is not related. Figure 8 shows an example of the use of indirect DNA testing methods to determine carrier status.

Indirect markers can be very powerful tools in following the inheritance of an altered gene as long as both of the carrier's parents and an affected male member are available. If not, then the answer is not as certain. Other problems can also produce unclear results. In some women, all the markers checked are the same on both X chromosomes. In these cases, the chromosome carrying the hemophilia gene cannot be identified. In other women, the only helpful marker is outside the factor VIII gene. This makes the test less accurate.
Counselling

Clotting Factors

If DNA tests cannot tell if a woman is a carrier, clotting factor levels may be helpful. The accuracy of clotting factor tests in determining carrier status is approximately 70% to 90%. On average, carriers have lower factor VIII or IX levels than women who are not carriers. Because carriers have only one normal X chromosome, one might expect their clotting factor levels to be half of the normal level. However, there is such a wide range of "normal" for clotting factor levels that there is a large overlap between the levels of carriers and noncarriers (Figure 9). In addition, clotting factor levels can be affected by age, health, pregnancy, and medications, including birth control pills.

Figure 9. Distribution curve of factor VIII activity among carrier and noncarrier women. Although factor VIII activity levels are generally higher in noncarriers than in carriers, many women from both groups fall into a "middle" activity level range. For this reason, it may be difficult to tell whether a particular woman is a noncarrier with a relatively low factor VIII activity level or a carrier with a relatively high factor VIII activity level.

Information on Carrier Status.

When to Test

Many physicians feel that women who might be carriers should have their clotting factor levels tested early in life. About 50% of female carriers have clotting factor levels that are less than 50% of normal levels, and 10% may have clotting factor levels below 30%. These clotting factor levels are low enough to result in dangerous bleeding if the woman is in an accident or has surgery. Knowing that a woman's clotting factor level is low can help physicians provide the best care in situations involving bleeding. In addition, if potential carriers are tested at a young age, family members whose blood is needed for the DNA tests described earlier are more likely to be available. If surgery is planned, factor level testing should definitely occur before the operation.

For a woman who has normal clotting factor levels, a family history analysis or DNA tests will be required to tell whether she is a carrier. This type of carrier testing is not an emergency and should be planned for a time when appropriate counseling and education can be provided. Some families may prefer to delay DNA testing until individuals are old enough to request it themselves and can understand the results and the implications.

It is very important, however, that a woman who wants to know whether she is a carrier be tested before she becomes pregnant. This knowledge will help her and her doctor plan and monitor her pregnancy. In addition, pregnancy can cause factor VIII levels to rise; this makes it difficult to accurately measure factor levels.

Parents who have just learned that their child has hemophilia may want to know if the mother is a carrier. Because of the changes in factor VIII levels that occur during pregnancy, the mother's factor levels cannot be accurately determined until about three months after giving birth. However, DNA tests to determine if the mother is a carrier can be performed whenever the couple wishes. Carrier testing should definitely be performed before a new pregnancy occurs.

Genetic Counseling

Genetic counseling is available to all members of a family, male and female, in which hemophilia is present. The counseling process may involve a study of the family tree, laboratory testing, and one or more discussions with a genetic counselor or other members of the hemophilia team. Counseling can take place at a hemophilia treatment center or at a genetics clinic (this may require a referral from the family's healthcare provider).

The goals of genetic counseling are: (1) to provide information on transmission of the gene for hemophilia, and (2) to explain tests used to determine whether family members carry the gene. Genetic counselors also provide emotional support to families as they deal with the information and begin to think about their reproductive options.

Whom to Tell

A woman who knows that she is a hemophilia carrier is advised to discuss it with the man who will be the father of her children. Ideally, they should be counseled together before she becomes pregnant. She should also inform all of her doctors about her carrier status. Whether to tell friends and family members about the carrier test results is a personal decision that should be made by the woman together with her partner and/or others she trusts.
How to Arrange Testing

Carrier testing is not available in all laboratories in the United States. In addition, the methods and accuracy of testing vary from laboratory to laboratory. For information about the laboratory nearest you that provides hemophilia carrier testing, consult the National Hemophilia Foundation, your hemophilia treatment center, your local hemophilia chapter, or a genetics clinic in your area.

Carrier testing always requires a sample of blood. Blood samples for DNA testing can be safely shipped to large, highly specialized laboratories. Costs vary depending upon the type of testing and the number of family members tested but range from several hundred dollars to higher per person tested. Insurance coverage for the testing will also vary depending on the policy. The effect on future insurance coverage may be another issue. Many families proceed with testing even though they are unsure what impact the identification of a new carrier might have on their insurance coverage. There is some concern that a carrier may be considered to have a "preexisting condition" and thus have her insurance denied or limited.

Reproductive Choices

How a person deals with the possibility of passing hemophilia on to a child depends on many things. Decisions must be made about parenthood, childbirth, whether to continue with a pregnancy, and raising a child with a chronic health condition. Ideally, the decision-making process involves both the man and the woman considering conceiving a child. Couples often discuss the personal or emotional aspects of these decisions with trusted family members, close friends, religious advisors, and/or counseling professionals. Health issues, financial matters, and the practical aspects of having a child with hemophilia are often discussed with their physician, nurse, or psychosocial professional at the hemophilia treatment center and with other parents of children with the condition. It may also be helpful to discuss these issues with a genetic counselor.

Options for Carriers

A woman who is a hemophilia carrier has a 25% chance of having a child with hemophilia each time she becomes pregnant. Several alternatives are available for a woman who carries the gene for hemophilia and wants to be a parent.

- She may choose to take the 25% risk of having a child with hemophilia and, with no testing, carry a pregnancy to term.
- During each pregnancy, she may choose to have a test to determine if the fetus carries the gene for hemophilia and then consider whether to
continue the pregnancy. Methods of prenatal diagnosis will be discussed below.

- She may choose to become pregnant by in vitro (test tube) fertilization using an egg donated by a noncarrier woman. Usually, the father’s sperm is used to fertilize the egg. The resulting embryo is placed into the carrier’s uterus so that she can carry the pregnancy to term. In this case, the baby has the father’s genes, and the maternal genes come from the noncarrier donor.

- She may choose to become pregnant by in vitro fertilization using her own fertilized eggs but with the embryo being tested before it is placed in her uterus. Because it is possible to determine the sex of the embryo, the parents may choose to implant only female embryos. If this choice is made, there is a 50% chance that the female will be a carrier. It may also be possible to detect whether the embryo carries the hemophilia gene so that an unaffected male or female embryo may be chosen. Diagnosis of embryos is very new and experimental and is not available at all locations.

- She may choose not to have her own biological children and consider adoption, foster parenting, or other nonbiological options instead. Effective contraception or permanent sterilization may be chosen to prevent conception.

Options for Affected Males
A man with hemophilia cannot transmit the disease to his sons. However, all of his daughters will be carriers and his daughters’ children may be affected. A man who has hemophilia has several choices regarding childbearing:

- He may choose to have children, knowing that none of them will have hemophilia. However, all of his daughters will be carriers.

- He may choose not to reproduce and consider adoption or other nonbiological alternatives. Either effective contraception or permanent sterilization may be used to prevent reproduction.

- He may choose to have a child who has the mother’s genes but not the father’s. Using a procedure called artificial insemination, a doctor injects sperm from an unaffected donor into the woman’s uterus in order to fertilize her egg. If the mother is not a hemophilia carrier, then the child born of this procedure will not have the hemophilia gene.

- He may choose to have prenatal diagnosis performed on each pregnancy with the aim of terminating pregnancies in which the fetus is female. Hemophilia would not be passed on to future generations.

- He may choose to use in vitro fertilization and only select male embryos for implantation.

Experimental techniques that may someday provide more options for carriers and affected males continue to be developed. For instance, sperm separation techniques, in which sperm carrying an X chromosome are separated from those carrying a Y chromosome, are being tested in some locations. Together with artificial insemination, such a method could be used to help the partner of a man with hemophilia conceive only male (unaffected) children or to help a carrier woman have only daughters rather than a possibly hemophilic son. At the present time (1998), sperm separation techniques have not been very successful. For this reason, many healthcare providers discourage their use.

Prenatal Diagnosis
Prenatal diagnosis can be used to determine the sex of the fetus and, in many cases, to find out if the fetus carries the hemophilia gene. These procedures may be important to hemophilic fathers and carrier mothers who are concerned about passing on the gene for hemophilia. To determine if the fetus carries the hemophilia gene, special tests are performed during pregnancy. The information from these tests gives the parents the opportunity to prepare for the birth of a hemophilic child and, with their physician, to plan a safe delivery. This prenatal testing also allows the couple to decide whether to continue with the pregnancy.

The prenatal diagnostic tests themselves can be complicated and may occasionally harm the fetus. The woman and her partner should be sure to discuss the tests and their risks with a genetic counselor. If possible, this meeting should occur before the woman becomes pregnant. A family study may be required if DNA testing is to be used for prenatal diagnosis (see previous section on "DNA Testing").

The first step in the prenatal diagnosis of hemophilia is usually determination of the sex of the fetus. This can be done by (1) ultrasound analysis, (2) chorionic villus sampling (CVS), or (3) amniocentesis. The second step is testing for the presence of the hemophilia gene.

Ultrasound is a noninvasive procedure; it does not involve puncturing or cutting the skin. Ultrasound often shows the sex of the fetus, but it cannot reveal whether the fetus carries the hemophilia gene. In contrast, CVS and amniocentesis are invasive procedures in which tubes or needles enter the woman’s body to collect fetal cells. CVS and amniocentesis may determine both the sex of the fetus and the presence of the hemophilia gene. Both CVS and amniocentesis also give information about Down syndrome, a genetic condition that causes retardation. Many physicians recommend that if the mother is over 35 years of age, either
CVS or amniocentesis be performed to see if the child has Down syndrome.

If these methods cannot be used or do not provide an answer, percutaneous umbilical blood sampling (PUBS) can be used to obtain a blood sample from the fetus. This blood sample can then be directly tested for factor VIII or IX levels or can be used to provide DNA for genetic tests. Like CVS and amniocentesis, PUBS is an invasive procedure.

**Prenatal Diagnostic Methods**

Ultrasound analysis is performed in the 14th to 16th week of pregnancy. In this procedure, sound waves directed at the woman's uterus produce a picture of the fetus. If the fetus is positioned correctly and is male, a skilled operator may be able to see its penis. Ultrasound causes no pain and carries no risk of miscarriage or harm to the fetus.

CVS is performed between the 10th and 12th week of pregnancy. A long thin tube called a catheter is inserted through the woman's vagina and cervix into her uterus. The tube is then positioned so that it can obtain a tissue sample from the chorionic villi, finger-like pieces of tissue that are part of the fetal membrane of the placenta, a structure that nourishes the fetus. The fetal cells are then grown in the lab to provide enough cells for DNA tests. A chromosomal picture of the fetus (like those in Figure 2) can be made from the fetal cells. This picture will reveal the sex of the fetus. DNA from the fetal cells can also be used to test for the hemophilia gene.

Amniocentesis is usually performed in the 15th to 18th week of pregnancy. In this procedure, a special needle is inserted through the skin of the woman's stomach into the uterus. This needle is then used to remove a sample of amniotic fluid, the fluid that surrounds the fetus and contains cells that have washed off the fetus. The cells are analyzed in the same way as those removed during CVS, but the results are not available until later in the pregnancy.

In some cases, DNA testing is not informative or is not possible in a particular family, or the woman comes in for testing too late in the pregnancy for CVS or amniocentesis. In these cases, fetal blood obtained through PUBS can be tested for factor VIII or factor IX levels or used for DNA testing. PUBS is usually performed from the 18th to 20th week of pregnancy. This procedure involves inserting a needle through the woman's skin into the uterus and then directly into a blood vessel in the umbilical cord, the cord that connects the mother and the fetus.

PUBS does not always provide helpful information. PUBS is performed later in pregnancy than CVS or amniocentesis, so DNA test results may not become available until after it is too late to terminate the pregnancy. Although the fetal blood
obtained by PUBS can be tested for factor level, only the most severe forms of hemophilia (factor level less than 1% of normal) can be detected by PUBS. Because of these problems, PUBS is seldom performed.

**Risks Associated with Prenatal Diagnostic Techniques**

Prenatal diagnosis of genetic changes may provide helpful information. However, it is not without difficulties. Sometimes technical problems prevent any information from being produced. Furthermore, CVS, amniocentesis, and PUBS present some risk to the fetus (Table 2). All three tests are associated with a low risk of miscarriage. For CVS, the risk is about 1 miscarriage out of every 100 to 200 tests. For amniocentesis, the risk is approximately 1 miscarriage out of every 200 to 400 tests. In addition, some studies suggest that approximately 1 out of every 3,000 to 10,000 fetuses tested with CVS are born with birth defects affecting the limbs, fingers, or toes if CVS is done before the 10th week of pregnancy. The risk is less when CVS is performed in the 11th to 12th week of pregnancy. For PUBS, there is a risk of causing fetal death due to bleeding, especially if the fetus has hemophilia. The risk associated with this procedure is about 1 fetal death out of every 100 tests. Risks and possible complications should be discussed with a genetic counselor or physician before having CVS, amniocentesis, or PUBS. CVS may also result in some risk to the mother. About 7 out of every 100 women who have CVS develop bleeding complications. In women who have mild hemophilia (factor VIII or IX level of less than 50% of normal), the risk of bleeding after CVS is greater and may cause the woman to miscarry. The risk of bleeding is much less following amniocentesis. This procedure may be safely done even in women whose factor VIII or factor IX level is below 50%. Because of the risk of bleeding, all women who are carriers should have their factor VIII or IX level tested during the week before CVS is performed. If the level is below 50%, CVS should be canceled and amniocentesis performed instead. If the woman prefers to have CVS, she can be treated with recombinant factor VIII or factor IX. Another treatment sometimes used to treat mild hemophilia, DDAVP (desmopressin acetate), should not be used in a pregnant woman because it may harm the fetus.

In addition to physical side effects, the psychological and emotional impact of testing should also be considered. Both amniocentesis and PUBS are done in the second trimester of pregnancy. By this point, the woman is showing her pregnancy and feeling fetal movements. It may be psychologically difficult for a woman to terminate a pregnancy at such a late date.

Complication rates vary from center to center, so women should ask for specific information about problems associated with prenatal diagnostic techniques at their location.

<table>
<thead>
<tr>
<th>Procedure</th>
<th>When performed (week of pregnancy)</th>
<th>Accuracy of diagnosis</th>
<th>Risk of miscarriage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Noninvasive</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ultrasound</td>
<td>14 to 16 weeks or later</td>
<td>95% to 100% for sex determination, 0% for hemophilia</td>
<td>0</td>
</tr>
<tr>
<td>Invasive</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CVS</td>
<td>10 to 12 weeks</td>
<td>95% to 100%</td>
<td>0.3% to 1.0%†</td>
</tr>
<tr>
<td>Amniocentesis</td>
<td>15 to 18 weeks</td>
<td>95% to 100%†</td>
<td>0.25% to 0.5%†</td>
</tr>
<tr>
<td>PUBS</td>
<td>18 to 20 weeks</td>
<td>95% to 100% for DNA tests; low accuracy for factor tests except for fetuses with severe hemophilia</td>
<td>1.0% to 2.0%†</td>
</tr>
</tbody>
</table>

*If a genetic marker has been determined by prior testing of affected hemophiliac in the family.
†Overall risk of miscarriage reported in reference 16. Risk of miscarriage may vary somewhat from center to center.
‡Risk of miscarriage determined at the Reproductive Genetics Center, University of California, San Francisco.

Courtesy of Marion Kooper, MD, Hemophilia Treatment Center, University of California, San Francisco.
Glossary

Artificial insemination: a method in which donor sperm are injected into the woman's uterus in order to fertilize her egg

Base pairs: the smallest piece of information in a gene

Carriers: individuals who carry the gene for a condition but do not have the condition themselves

Cloned: genes that have been copied by chemical methods in a laboratory

Clotting factors: proteins needed to make blood clot

Chromosomes: threadlike structures inside human cells that contain thousands of genes and that are passed down through families

Deoxyribonucleic acid (DNA): the chemical substance that makes up genes

Gene sequence: the order of base pairs in a gene

Gene therapy: methods to correct a gene mutation by adding an intact (normal) one or changing one that is already present

Genes: units of DNA that contain all the hereditary information needed to make a product, such as a protein

Genetic markers: DNA pieces that are easy to identify but that themselves have nothing to do with the gene being examined, such as the gene for hemophilia

Germline mosaicism: a situation that occurs when mutations are found in some, but not all, of a woman's eggs. These mutations occur during egg cell development and are not found in the woman's other cells, such as blood cells

Germline therapy: gene therapy directed at embryos; this method replaces all of the altered genes that cause disease so that the gene is not passed on to future generations

Heredity disease: a disease that can be passed down through families

Inherited: passed down through families

Invasive procedure: a procedure that requires cutting into the body

Inversion: a part of a gene that is flipped around so that it can no longer be "read" in the correct direction

In vitro fertilization: a technique in which sperm and eggs are mixed together outside the body (usually in a glass laboratory plate) with the goal of creating a fertilized egg

Lyonization: the process of shutting down one of the X chromosomes in each female cell. Usually, half of the cells shut down one of the X chromosomes and the other half shut down the other one

Maternal transmission: when a gene is passed from the mother

Mutation: a defect or change in a gene

Noninvasive procedure: a procedure that does not require cutting into the body

Nonrandom X inactivation: a phenomenon in which all the cells in a woman's body shut down the same X chromosome

Obligate carrier: a woman who, on the basis of family history, definitely carries the gene for hemophilia. Obligate carriers can be (1) the daughter of a biological father with hemophilia; (2) the mother of more than one son with hemophilia; or (3) the mother of a son with hemophilia who has one other blood relative with hemophilia

Paternal transmission: when a gene is passed from the father

Prenatal diagnosis: determining the medical condition of a child before it is born

Protein: any of a large class of substances consisting of amino acids. Proteins occur in all animal and vegetable matter and are necessary for growth and repair

Restriction enzymes: proteins that cut DNA at specific sites

von Willebrand factor: a protein that carries factor VIII in the blood and helps factor VIII work properly

X-linked: genes that are present on the X chromosome
References


Inheritance of Hemophilia