Knowing the Risks

The Genetics of Breast and Ovarian Cancer

In 2013, actor and director Angelina Jolie underwent a preventive double mastectomy after testing positive for a mutation in the tumor-suppressing BRCA1 gene. That spring, she announced the news in a widely read New York Times opinion piece.

In the op-ed, Jolie discussed how her mother died of cancer at the age of 56 and how the mutation Jolie inherited increased the risks she faced for developing ovarian and breast cancer. She described the procedures that removed and reconstructed her breasts. And she shared her desire to reassure her children that their mother was doing everything she could to stay with them as long as possible.

She also urged women — particularly those with a family history of breast or ovarian cancer — to seek out professional medical advice and make an informed decision on possible genetic testing and preventive treatment. Mutations on the BRCA1 gene and the similarly tumor-suppressing BRCA2 gene also carry increased lifetime risk for cancers of the pancreas and prostate.

“Cancer is still a word that strikes fear into people’s hearts, producing a deep sense of powerlessness,” Jolie wrote. “But today it is possible to find out through a blood test whether you are... What’s in a Name? Continued on page 3...
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Knowing the Risks (Continued from front cover)

highly susceptible to breast and ovarian cancer, and then take action.”

About 12 percent of women in the country will develop breast cancer at some point in their lives. For women with a BRCA2 mutation, that number jumps to 69 percent. A BRCA1 mutation leads to breast cancer in 72 percent of women.

Of note, however, surgical interventions can carry significant known risks in addition to psychological and economic burdens. All without the certainty of clear findings that point toward the wisdom of a particular treatment.

Currently, genetic laboratories can look for well-established disease-causing mutations in specific populations, such as Ashkenazi Jews, who are more likely to pass on any of two clearly defined mutations of the BRCA1 gene or one particular mutation of the BRCA2 gene. People of Ashkenazi Jewish decent with a parent carrying one of these mutations have a 50 percent chance of inheriting the mutated gene from that parent.

However, even five years after Jolie’s announcement, it remains unclear what causes the 95% of total breast cancer cases unrelated to BRCA1/2 mutations.

And not all mutations are alike. In fact, there are thousands of mostly benign variants in BRCA1/2 genes, more accurately called either benign polymorphisms or variants of uncertain significance (or VUS). Some VUS are harmless. Some can be disease-causing mutations. There is no easy way to tell which is which. Treatment decisions following genetic testing classified as a BRCA1 or BRCA2 variant of uncertain significance are based on probabilistic models, not specific risk associated with a patient’s particular variant or variants.

But one of the world’s experts on this subject has been constructing a model to uncover how changes like these in genes lead to disease.

“We’re examining BRCA1 and BRCA2 to understand the progression of cancer at the most fundamental level,” said Dr. Ryan Jensen, Associate Professor of Therapeutic Radiology at Yale Medical School. “We need to recognize what is happening to the genes to create this elevated cancer risk, and then we can better address ways to treat and prevent these diseases.”

Jensen’s work, spurred by a WHRY-funded grant in 2013, continues progress toward a rapid laboratory test that will clear up the ambiguity and help guide patients and doctors toward decisions that can produce the best health outcomes.

“There are thousands of women in the United States with thousands of different alterations to these important genes,” Jensen said. “We need to learn everything we can to help them.”

Uncorrected Errors

In 1990, Dr. Mary-Claire King at the University of California, Berkeley, discovered a gene shared by families susceptible to breast cancer, sparking great interest in biomedical science to link genetics to disease. She called her discovery BRCA1 (breast cancer susceptibility gene 1) for its breast cancer connection. A group of scientists led by Michael Stratton at the

**BRCA2 MUTATIONS**

Dr. Ryan Jensen’s lab is purifying the most common BRCA2 variants, labeled here in an illustration of the gene's crystal structure, to determine if the alterations are benign or cause disease.
University of Cambridge identified the BRCA2 (breast cancer susceptibility gene 2) gene in 1994.

The BRCA1/2 genes work to correct potential tumor-causing mistakes made during the replication of DNA, the material carrying the body’s genetic code that directs the production of proteins and passes traits from parents to offspring. Proteins form the building blocks of cells and trigger biochemical reactions.

But even as people might consider their genetic code to be a permanent part of themselves, DNA is not static. Every time a cell divides, all 3.2 billion base pairs that form a DNA molecule’s coded double-helix structure have to be copied. Sometimes there are mistakes.

“The BRCA1 and 2 genes function like an auto correct function in text messaging,” Jensen said. “When the genes that code for BRCA1 and 2 proteins are mutated and fail to correct DNA damage, genetic mistakes accumulate, eventually leading a cell down a path to becoming cancerous.”

But in order to better understand how these mutations function and what work goes unaccomplished when the gene is not intact, someone first needed to isolate a pure sample of the proteins that the unmutated genes create. This presented a difficult task because of the large size of the proteins, their instability, the complicating presence of other proteins that attach to them, and the low levels at which they can be found in human cells.

In 2010, after six years working in a 4 degree Celsius cold room, Jensen was the lead author on a study culminating a total of 15 years of research and published in the journal Nature to announce the successful purification of the BRCA2 gene’s protein. In 2017, he collaborated with Dr. Patrick Sung, Yale Professor of Molecular Biophysics and Biochemistry, Therapeutic Radiology, and Epidemiology, to successfully purify the BRCA1 protein.

“If that was the hard part, next came the equally hard part,” Jensen said.

So-called truncating mutations in BRCA2, like those found among Ashkenazi Jews, delete an easy-to-spot large section of the protein strand needed to bind to the DNA. If the protein can’t bind to the DNA, it can’t repair the DNA, which leads to the development and spread of cancer.

**DEFINITIONS**

**DNA**
Deoxyribonucleic acid, a self-replicating material that carries the genetic code of almost all living things and directs the production of proteins, the building blocks of cells.

**Gene**
A part of the DNA molecule that forms the basis of heredity, passing traits from parents to offspring.

**Variant**
A permanent change in the sequence of DNA created through replication error or unrepaired damage. Some variants, known as benign polymorphisms, do not affect the gene’s ability to produce a working protein and therefore do not cause disease.

**Variant of Uncertain Significance (VUS)**
A change to a gene’s code for which there is limited or conflicting evidence regarding its ability to cause cancer.

**Mutation**
A gene variant with sufficient evidence to classify it as capable of causing disease because the change is known to affect the gene’s ability to produce a working protein.

**Truncating mutation**
A gene variant with a high risk of producing cancer that deletes an easy-to-spot large section of the protein strand needed to bind to the DNA. If the protein can’t bind to the DNA, it can’t repair the DNA, which leads to the development and spread of cancer.

**Amino Acid**
An organic compound that forms proteins, which are the building blocks of cells and the triggers for biochemical reactions.

**Missense**
A gene variant that results in the change of a single amino acid, either having no effect on a protein’s ability to correct DNA errors or leaving that protein dysfunctional, thus likely to cause cancer.

**Cancer**
A group of diseases that occur when abnormal cells, often altered through mutation, begin dividing without stopping and then spread to surrounding tissue.

**BRCA1 and BRCA2**
Human genes responsible for producing proteins that repair DNA replication errors responsible for development of cancer cells. Harmful mutations of these genes are linked to increased lifetime risk of breast, ovarian, prostate, and pancreatic cancer.

**Gene purification**
Isolating an extract of a protein from all other bio-molecules, such as RNA and other proteins, to better understand its structure and function.

**Genome**
The complete set of genes or genetic material present in a cell or organism.
strand needed to bind to the DNA and are associated with very high cancer risk. If the protein can’t bind to the DNA, it can’t repair the DNA, which leads to the development and spread of cancer.

A more subtle variant is called missense, which involves changing a single amino acid, the organic substance that forms proteins. That’s one altered amino acid in a chain of 3,418 amino acids that make up the DNA-correcting protein. Such missense mutations are commonly labeled as variants of uncertain significance because of their relative rareness and difficulty to link to disease.

“How does one amino acid affect a protein and the risk for cancer?” Jensen said. “Answering that question has been keeping us busy.”

Looking Closer

In 2013, Women’s Health Research at Yale funded a two-year study in which Jensen’s team began the painstaking effort of purifying the BRCA2 gene when it contains 10 of the most common variants of uncertain significance and conducting experiments to determine if the alterations in the DNA code affect the function of the intact BRCA2 protein.

“Studying the top 10 can direct us to understand other nearby mutations or similar functions of the protein,” Jensen said. “We are using it as a model to guide us and assess all of these variants that may or may not cause disease.”

Individuals in the general population are not screened for specific mutations. Instead, labs analyze the entirety of their genes to find deviations from a complete set of intact genes assembled by scientists in a database to use as a reference. Such comparisons show a lot of normal variation from individual to individual. These are changes, but not necessarily disease-causing mutations. What Jensen and others are trying to determine is which of the variants are most likely to lead to disease.

Jensen’s goal is to develop a practical test that can quickly and accurately determine whether an individual’s variant is harmful or not. His WHRY-funded study proved the principle of such a test and its effectiveness in distinguishing harmful from benign BRCA2 variants by comparing those known to be harmful and others known to be harmless as confirmed by studies of patients with the genetic mutations and the presence or absence of cancer diagnoses.

Jensen’s WHRY-funded proof-of-principle study allowed him to successfully apply for a large grant from the National Institutes of Health to continue this work toward determining which BRCA2 variants prove harmful so that patients and their health care providers can make

ABOUT THE INVESTIGATOR

Dr. Ryan Jensen earned his Ph.D. from Yale University and his B.A. from the University of California at Berkeley. He is an Associate Professor of Therapeutic Radiology and of Pathology.

His research focuses on DNA repair and how repairs to this basic hereditary material go awry, leading to the initiation of tumors and cancer. A major focus of his work is to understand the role harmful BRCA1 and BRCA2 mutations play in increasing the risk of breast, ovarian, and other types of cancer.

Dr. Ryan Jensen’s lab is now uncovering why there is such a high prevalence of ovarian cancer in BRCA2 mutation carriers to generate the needed guidance for women to treat or prevent this deadly disease.
informed decisions about potential treatments.

And the study advanced his lab’s larger goal of establishing a model that can accurately describe and predict the mechanisms that turn a healthy, functioning cell into a tumor that not only survives without an intact BRCA1 or BRCA2 gene, but thrives and spreads.

Toward this end, he will purify more BRCA2 protein samples and examine their structure in crystal form or by using a special electron microscope technique called cryo-electron microscopy (cryoEM) in which samples are cooled to cryogenic temperatures (below -180 degrees Celsius). If successful, he can use these images to create a 3-D picture of the BRCA2 protein to further our understanding of how the protein works and why certain mutations interfere with its normal functions.

“Once we get a handle on what BRCA2 looks like, we can learn so much... we can look at the protein and make predictions.”

Enabling Early Detection and Treatment of Ovarian Cancer

In response to the success of Jensen’s earlier work, WHRY has granted him this year’s Wendy U. and Thomas C. Naratil Pioneer Award to uncover why there is such a high prevalence of ovarian cancer in BRCA2 mutation carriers and offer needed guidance for women to treat or prevent this deadly disease.

About 22,500 women in the country will receive a diagnosis of ovarian cancer this year. And 14,000 will die of the disease. The best chance for survival relies on early detection, but current methods remain inadequate.

“Ovarian cancer is of particular importance for women’s health as it is often diagnosed too late for effective therapeutic intervention,” Jensen said. “By identifying the molecular circuitry disrupted by loss of the tumor-suppressing protein produced by BRCA2, we have the opportunity to expose vulnerabilities that can be selectively targeted or lead to novel biological clues to aid detection and vastly improve the standard of care for this unique population of women.”

In this new study, Jensen will manipulate fallopian tube cells and cells from the surface of ovaries that have been identified as the potential origin of ovarian tumors to determine how the loss of BRCA2 proteins can influence this process.

Jensen has already created fallopian and ovarian surface cells in which he can turn the BRCA2 protein “on” and “off” like a switch. This experimental system will provide a powerful model to test his hypothesis that cells without the BRCA2 protein experience profound genomic instability, a cellular state in which mutations begin to accumulate throughout a person’s genome at an alarming rate.

“Our 20TH YEAR — WHRY’S LONG FIGHT AGAINST CANCER

In one of WHRY’s earliest breast cancer studies, Dr. Bruce Haffty, currently Chief of Staff at Rutgers Robert Wood Johnson Medical School in New Jersey, demonstrated that BRCA1 and BRCA2 mutations predict a vulnerability to breast cancer recurrence in either the treated breast or the untreated breast.

This landmark study, published in 2002 in The Lancet medical journal, continues to inform clinical decisions about treatments to prevent recurrence.
Advancing Our Common Purpose

Let’s agree on something basic: research should inform health care.

In 2018, that means science must take into account what makes each of us unique.

And, individual distinction begins with sex and gender.

When evaluating the development of diseases and the effectiveness of their treatments, it makes sense to begin with the biology that determines our sex classifications and the combination of biology, environment, and experience that forms our gendered self-representations.

And nowhere has this principle found a more successful champion than at Women’s Health Research at Yale.

Now entering its third decade, this self-supporting center continues to evolve and meet the challenges presented by the diseases and conditions that affect so many. WHRY does this by ensuring that science and clinical practice fully explore sex and gender and apply lessons learned in order to deliver their practical benefits.

Nowhere else will you find experts more committed to initiating and funding new studies on women’s health and exploring sex and gender differences. And, WHRY’s position within Yale School of Medicine offers unique opportunities for building interdisciplinary research partnerships and for training the next generation of researchers committed to advancing women’s health.

Further, WHRY shares its findings in order that people throughout the communities we serve can make informed decisions about their health and health care. In another dimension of our public purpose, WHRY works with policymakers to ensure public health initiatives reflect the best information available.

Women’s Health Research at Yale continues to accomplish these vital undertakings because of your generous and thoughtful support. I am tremendously grateful for our friends and their commitment to women’s health. Isn’t it good to know that there is a way we can act to benefit all people?

With thanks to everyone who has helped make WHRY a singular success,

Barbara M. Riley
Philanthropy Chair

Women’s Health Research at Yale thanks our generous partners who are helping to change the landscape of medical research and practice.

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In 1916, Louise Whitman Farnam wanted to study medicine, and one of the reasons she was told that women could not attend Yale School of Medicine was that there were no bathrooms for women.

She eventually attended and graduated four years later, thanks to her talent and drive, but also as a result of a gift to the school from her father, Henry Farnum.

“Many of us who have benefited from the opportunity of attending Yale School Medicine can thank the courageous women who came before us,” said Dr. Susan J. Baserga, Professor of Molecular Biophysics and Biochemistry, Genetics, and Therapeutic Radiology at YSM. “And we must also remember their fathers and mothers, who believed that their daughters’ education — particularly at this time — was just as important as their sons’. And also in the case of Henry Farnum, we must be grateful for his strategic donation of plumbing facilities so that the specious argument about why women can’t be doctors could be put to rest here at Yale.”

Baserga and other outstanding women who are or were faculty at YSM spoke in June at a symposium celebrating 100 years of women at YSM, showcasing the history, science, and clinical accomplishments of women and highlighting current issues women face in the field of medicine.

More than 700 people registered for the event, sponsored by the Committee on the Status of Women in Medicine (SWIM), the Minority Organization for Retention & Expansion (MORE), and the Dean’s Office.

Women’s Health Research at Yale Director Carolyn M. Mazure, Ph.D., presented the history of U.S. public policy on women’s health research and how WHRY has been changing the landscape of medical research and practice for 20 years.

“If we want to see improvement in health and health care, I think most of us would agree, that it depends upon developing new knowledge that we can incorporate into our personal practice and our professional practice,” Mazure said.

But early on in her education Mazure found that the prevailing scientific tradition in the United States and around the world was one in which women were not generally included as participants in clinical trials, a problem that persisted until the mid-1990s.

“If we want to see improvement in health and health care, that depends upon developing new knowledge.”

“And in the cases where women were included, we were taught growing up in science that we really didn’t have to analyze the data by gender outcome,” Mazure
said. “And as a consequence of that, it has really left us with this incredible gap in knowledge about women’s health.”

Mazure summarized why women should be studied. For starters, she said, women are more likely to suffer from chronic diseases and disability, have acute and chronic pain, die following a heart attack, develop depression and anxiety, have autoimmune diseases, develop Alzheimer’s disease, and have unique reproductive health requirements. She described how WHRY began funding pilot studies because those wishing to study women’s health did not have the necessary feasibility data to obtain external grants. Over 20 years, the center has funded $5 million in seed grants that have gone on to produce feasibility data for external grants of $102 million for the researchers to continue and extend their work.

The data now show that sex and gender differences contribute to the prevalence, risk factors, presentation, and course of diseases such as osteoporosis and stroke. We know that the response to a treatment can vary by sex and gender, such as with anesthesia and smoking. And prevention strategies for viruses such as HIV or conditions such as alcohol abuse often need to be sex-and-gender specific to be effective.

In addition, Mazure stressed how WHRY embraces the importance of communicating the latest findings to the widest audience possible so that people can make more informed decisions about their health. And she talked about how the center works to train the next generation of researchers and clinicians, while informing non-partisan public action.

“We really are just at the beginning of learning about women’s health,” Mazure said. “And most importantly, we need to seriously begin examining both the differences between and the differences among women and men.”

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... What’s Next?

**WHRY’s Undergraduate Fellows Prepare to Drive Progress**

Currently heading into its fourth year, WHRY’s Undergraduate Fellowship pairs students with faculty mentors and offers a “scientific home” to buttress their coursework. This active participation program equips Yale students with the knowledge and skills they will need to carry this vital work forward, influencing others throughout their promising careers.

“If more scientists appreciate the importance of studying the influence of sex and gender in health, they can help create the momentum for change,” said Haleigh Larson, a Class of ’18 graduate who spent two years as a WHRY fellow. “In the future, when I’m doing my own research, I want to conduct studies that are thoughtfully designed to consider sex and gender specificity and communicate results in a way that is directly relevant to women and men.”

In her time with WHRY, Larson helped produce and test the efficacy of a series of public health literacy videos designed to address the unique vulnerability of girls and young women to the negative effects of sexually transmitted infections, alcohol abuse, and stress. Her work ranged from researching topics, assisting with script writing, performing voiceover and on camera, and organizing, administering, and analyzing data from in-person survey sessions.

After graduation, Larson began a two-year position with The Brotman-Baty Institute in Seattle researching cancer-
causing gene mutations before applying to medical school and pursuing a career as a clinical geneticist.

**Rose Davis,** Class of ’18, worked with Dr. Lisa Freed, Director of Yale New Haven Hospital’s Women’s Heart and Vascular Program and a collaborator with WHRY on integrating health research into clinical practice.

For two years, Davis shadowed Dr. Freed at her clinic and participated in research studying women and their adherence to cholesterol-reducing medication guidelines established by the American Heart Association.

Talking to patients, administering questionnaires, and examining medical records for risk factors led Davis to a better understanding of how patients think and feel about taking medication.

“I feel I have a much more well-rounded understanding of what it means to be a patient and a physician,” Davis said. “I’m really grateful for that. Dr. Freed’s patients love her because she understands their challenges and, as a consequence, they are more likely to work with her to help themselves.”

Davis worked this summer in London at an internship with a new wellness company that runs retreats in Switzerland focusing on mental health, sleep, nutrition, and fitness. Next year, she plans to apply to medical school. And spread her WHRY knowledge wherever she goes.

“WHRY gave me an unparalleled education,” she said. “I found it extremely interesting. Every time I sat down with Dr. Mazure, I felt I took away something new.”

**Kanan Shah,** Class of ’18, was mentored by Dr. Kimberly Yonkers, Director of Yale’s Center for Wellbeing of Women and Mothers, working on projects to help pregnant mothers with substance abuse problems.

While crafting effective surveys, developing educational materials, and interacting with patients, Shah learned the value of gaining firsthand research experience as an undergraduate.

“I think this would fall into my most meaningful experiences at Yale,” Shah said. “I feel like before joining the WHRY fellowship and Dr. Yonkers’ lab, clinical research was just this term thrown around everywhere, but I didn’t know what it entailed.”

Shah is now taking a year off before applying to medical school, and she feels confident that she can transfer her skills to her current job at the New York University Department of Population Health as part of a health care innovation delivery team.

“I think this is something I want to keep going in my career,” Shah said. “I really like talking to people. Everybody is so different and unique. I don’t see how a provider can adequately treat patients without knowing about their life and not just that one condition they are treating them for.”

**Lauren McNeel,** Class of ’18, was mentored by Dr. Kelly Cosgrove at the Yale PET Center, using brain scans to examine the causes and consequences of addiction and to determine what role sex and gender might play in the neurochemistry of addiction.

“It definitely added to my understanding of how nuanced research can be and how important it is to look for specific differences,” McNeel said. “And how investigations like these are really affecting the future of women’s health research.”

McNeel is taking the year off before applying to graduate programs. She wants to study women’s public health policy and law. And she knows she will

**Rose Davis, Class of ’18 (right) shadows Dr. Lisa Freed, Director of Yale New Haven Hospital’s Women’s Heart and Vascular Program (center) as part of her WHRY Undergraduate Fellowship.**
always apply lessons learned through biomedical research.

She has already shared the knowledge she gained from her work with WHRY and Dr. Cosgrove with friends and classmates, adding insight on historical inequities in health care research when discussing the dynamics of gender on markets and political power.

“It’s been eye-opening for me and also the people around me,” she said.

To learn about the rest of last year’s fellows and meet the new class joining us this fall, visit our website: yalewhr.org

What’s in a Name?

By Rick Harrison

When people ask me what I do for a living, I tell them I am the Communications Officer for Women’s Health Research at Yale. But it’s not that simple.

Because WHRY is unique, it is not easy to summarize all that the center does in a sentence or two. The name itself offers an accurate description, but not the entire story.

So what exactly is Women’s Health Research at Yale? It is a self-supporting center within Yale School of Medicine that for 20 years has been changing the landscape of medical research and practice by ensuring the study of women and examining health differences between women and men to improve the lives of everyone.

But even that not-so-brief description can lead to a lot of questions among people unfamiliar with our work.

Why, for example, does the landscape of medical research and practice need changing?

Most people are not aware that it wasn’t until the mid-1990s that women were required to be included as participants in clinical trials seeking grants from the National Institutes of Health, the world’s single largest funder of biomedical research. And most people are not aware that it wasn’t until 2016 that the NIH began to require the use of female animals, tissues, and cells in laboratory studies that form the necessary basis for later human trials.

In addition, people might wonder why a center at Yale needs to ensure that researchers examine health differences between sexes and genders. One reason is that even when females are included as research subjects in adequate numbers, the data are not always analyzed by sex or gender to determine if any significant differences exist. This leaves us in the dark about any potential divergences in the prevalence, development, and treatment of diseases and conditions.

So, Women’s Health Research at Yale most definitely benefits women, for whom a gap in health knowledge had grown for decades and whose specific health needs continue to require attention. But WHRY’s additional focus on sex and gender differences also allows men to benefit by illuminating aspects of men’s health that might otherwise get muddled in an unexamined mix of sex and gender data.

And Women’s Health Research at Yale does more than conduct research through our trailblazing Pilot Project Program and interdisciplinary research partnerships. The center also communicates the latest findings to the public to better inform health decisions and lead to better outcomes. Furthermore, we train the next generation of women’s health researchers and clinicians to carry on and spread this important work — both through hands-on mentorship and efforts to integrate sex and gender into medical school curriculums so that physicians and scientists investigate and account for important sex and gender differences.

Finally, it’s important to note that the Y in WHRY stands for Yale, a global institution of higher learning, innovation, and leadership that inspires the minds that inspire the world. WHRY has tremendous resources at our disposal to leverage the generous contributions our supporters make and achieve the greatest possible practical results to improve and even save lives.

In summary, when people ask me what I do, it’s never a short conversation. But it’s an important one. And one I’m eager to have with anyone interested in changing medical research and practice so we can better care for everyone.
Women’s Health Research at Yale

is changing the landscape of medical research and practice by ensuring the study of women and examining health differences between women and men to improve the lives of everyone.

Happy 20th anniversary, WHRY!

“Attending a WHRY community workshop nearly 20 years ago, I learned of the center’s dedication to ensuring that all people are included in and benefit from medical research. Just a couple of years ago, I had the opportunity to give a gift that was very important to me because it honored my mother. I chose WHRY. Your work sets the standard for a practical approach to getting the job done.”

— Ann Baker Pepe,
Member of WHRY’s Society of Friends

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