

## **NC DNA DAY - Pharmacogenomics Module Script**

**Pharmacogenomics Title Slide:** Today I am going to talk to you about the exciting topic of pharmacogenomics. Does anyone know or have a guess what this word means?

**What is DNA Day?** On DNA Day we commemorate some very important events in the history of our knowledge about DNA. In April 1953, scientists Watson and Crick described the double helix structure of DNA. Fifty years later, in 2003, scientists published the entire SEQUENCE of the human genome which has enabled many advances in our understanding of how our genes function, but also in how we can treat diseases.

**What is pharmacogenomics?:** Let's break up the word. Pharma sounds like pharmacy – another word you may be more familiar with that means drug or medicine. Genomics, which sounds similar to the word genetics, means the study of genes. So, the word pharmacogenomics means personalized medicine that is tailored to you the individual, based on the genes you have. Your genes will determine how you respond to a drug, is the drug toxic, is the dose too low, will the drug give you a headache, make you feel sick to your stomach, or make you feel better? All of these side effects and outcomes are dictated by your genes. If doctors understand which genes interact with which medicines or drug treatments, they can design therapies that fit you as an individual patient. This approach will improve disease treatment and hopefully outcome.

Let's look at an example case study to help you understand why pharmacogenomics is really exciting!

**Case Study – Breast Cancer Patients:** Here are 10 women who have recently been diagnosed with breast cancer. They have been having radiation and chemotherapy or drug treatments. Their doctor tells them about a clinical trial with a new drug called Tumoricide. This drug is supposed to kill the cancer cells in their bodies. All 10 women agree to take the drug hoping that it will help them get better more rapidly.

### **Case Study – Breast Cancer Patients: Several slides build these concepts**

One year later, the women are screened again to see if the breast cancer cells have been eliminated from their bodies. Three of the women are doing really well and the cancer seems to be gone (shown in dark pink), but seven of them still have tumors (shown in light pink). One woman (in the top right) has a weak heart. She blames the Tumoricide for her heart problems. This suggests that Tumoricide is only 30% effective; it only helped 3 out of 10 women.

**Helped/Hurt Breast Cancer Case Study (Why?):** The scientists running the clinical trial take a step back and look at the results, 7 women were not helped by Tumoricide, one might have

even been hurt, but three women seemed to be better. Their cancer might be gone. All 10 women had the same disease – breast cancer – and all 10 took the same drug – Tumoricide. Why did these women respond so differently? To help you answer this important question, *let's talk about how scientists make personalized medications...*

### **How do scientists make personalized medicine?**

I will be reviewing the basics of DNA to better understand how scientists think about personalized medicine. First, its important to realize that Personalized medicine is about understanding what is in YOUR DNA. Its understanding your genetic make-up, whats inside your cells and identifying the small differences or variations inside your genes.

In summary, its understanding what makes your genetic code unique.

### **Genetic Code: DNA – Several slides build these concepts**

So-lets review the Genetic Code. The Genetic Code refers to your DNA. DNA stands for Deoxyribonucleic Acid (D-N-A) and contains the information that is necessary to make a complete organism. Remember that DNA is composed of 4 nucleotides usually symbolized by A, T, C, G:

A is Adenine

T is Thymine

C is cytosine

G is Guanine

### **The Central Dogma: DNA to RNA to Protein (Several slides build these concepts)**

To understand the how DNA works, we usually discuss this in terms of the Central Dogma. The Central Dogma refers to the fact the DNA is transcribed into RNA and RNA is translated into Protein.

- 1) DNA is made from nucleotides that arrange as a long double-stranded molecule. Genes are found on the DNA strand
- 2) The genes on the DNA are made into RNA, which is essentially a single-stranded copy of the gene
- 3) The RNA is then made into a protein. Proteins are composed of amino acids. Amino acids are made from triplets of nucleotides called codons.

In this example of the RNA molecule, the first three nucleotides make up codon 1 and the second three nucleotides make up codon 2. Codon 1 will be translated into amino acid 1 and Codon 2 will be translated into amino acid 2. This combination of amino acids is then called a Protein.

In Summary: Genes are on DNA which is made into RNA, then RNA is made into Protein. Thus, the gene sequence is very important for making the correct protein.

### **A small change in a gene sequence can result in a very different protein – several slides build these concepts**

Although humans have the same genes in their DNA, there are small differences in our DNA sequence that can result in very different proteins. To better understand how small changes in a DNA sequence can change a protein—lets go over this example. This DNA sequence is broken into codons or triplets of nucleotides. DNA is made into RNA and each codon will encode for a specific amino acid. It's the string of amino acids that make up a protein.

Now, lets use this same DNA sequence, except the in the last codon there is single nucleotide that is different. So instead of CCT the codon reads ACT. ACT will encode for a different amino acids, shown in red. Because the amino acids are different, the function of the protein may be different too.

To make this concept clearer...Let's associate amino acids with words and a protein as a sentence. Thus, the first protein sequence might make a sentence like ---TOM AND SAM ARE BAD. However, the second sentence might read like TOM AND SAM ARE SAD. Notice how the meaning of those sentences changes, with just a single letter difference between BAD and SAD.

These changes in DNA are called variations or mutations. Importantly, variations in the DNA or (genotype) can result in observable changes (known as a phenotype) in individuals.

If you are having a hard time remembering the difference between genotype and phenotype— here is how I first remembered them. Gene sounds like GENE—so a genotype refers to changes in your genes. Pheno means displaying- so a phenotype is when you display or show a certain characteristic that is governed by your gene

### **Variations in our DNA makes us Unique!**

Importantly, these variations are what make us unique and what makes us genetically diverse! This genetic diversity is very important for understanding Personalized Medicine because it is these small differences in our genes that make us react differently to medicine.

## Case Study

Remember our case study...why did some patients have such a positive reaction to Tumorcide and some patients did not.

Next, I will help you understand how proteins have a role in understanding how our bodies react to drugs.

### What are the reasons a person would react differently to drugs?

- So Now that you know all this great stuff about DNA, what does it have to do with medicine?
- Well we are going to look at a few genetic reasons why different people may have different reactions to the same drug, like the Tumorcide story Sarah told you all about.
- The first way people react differently is differences in the receptor proteins they have to recognize drugs.

### Drugs and Receptors – Several animations build this concept

- Our cells have all the DNA in them to make all the different kinds of proteins they need.
- This includes receptor proteins. Receptors are proteins that sit on the outside of our cells and receive signals from the external environment.
- Sometimes, these signals are drugs!
- Molecules that bind to proteins are also called ligands.
- Drugs are ligands that bind to a specific type of receptor protein.
- When the cells receives the signal, this gives a therapeutic effect by blocking other molecules from binding to the receptor or telling the cell to do something different that makes us feel better.

### Your DNA and Drugs – Several animations build this concept

- But, still, why would your DNA change your body's reaction to a drug?
- Since DNA gives the code for proteins, changes in your genes can cause changes to your receptors.
- For example mutations in DNA can cause the cells to make more receptors than average, so the ligand has more receptors to bind to.
- Or a mutation may cause the cell to make less receptors, so the drug doesn't have as many to bind to.
- And sometimes mutations cause a change in the receptor itself, which makes it unable to bind with the drug.
- These variations in our genes can cause variations in how we respond to drugs

- With too many receptors the cell may receive more signals and amplify the drug effect. Overreaction to a drug is called hypersensitivity.
- With too few receptors and the drug may not find enough to bind to and the drug's effect will be small. Under reaction to a drug is called hyposensitivity.
- Or a Mutated receptor may not be able to bind the drug at all, so the cell sees no effect. When a patient sees no reaction to a drug it is called insensitivity.

### **Where Drugs “Fit” In**

- But how does a drug know where to bind? Drugs are designed to fit into specific receptors on our cells, just like a key is designed to fit into a specific lock.

### **Let's do a class case study! Several animations**

- Now that we have learned a bit about our bodies reactions to drugs, lets do an in-class “personalized medicine” case study to test our genetic variation.
- You all have PTC strips, pick it up and place it on your tongue for five seconds to taste it. Don't just lick it, but leave it on there for a bit to see if you can taste anything. Don't worry- PTC is a non-toxic chemical, so tasting it won't hurt you!
- What do you taste? Write down what you tasted and compare it with your classmates. PAUSE POINT.
- Did it taste yucky to some of you and just like plain paper to others?
- Why do you think that is? Do you think it could have anything to do with your genetics?
- Discuss your hypothesis with your classmates. PAUSE POINT.

### **Why can some people taste PTC and others can't? Several animations**

- It does have to do with genetics and receptors!
- A key must fit into the lock to open a door just like PTC must be able to bind the receptor to cause an effect
- One genetic variant of the PTC receptor binds PTC well - PTC tastes bitter
- Another genetic variant of the PTC receptor cannot bind PTC- so the strip doesn't taste like anything. This key does not fit into the lock!
- How does this happen?

### **Where does tasting PTC come from? Several animations**

- Remember how I told you that we get one copy of each gene from each of our parents? Well, lets review.
- But genes can be dominant or recessive.
- When a dominant gene is present it's trait will overcome the recessive gene.

- This determines which trait is seen, which is a person's phenotype. So, Tasting PTC comes from our genes!

### **Tasting PTC is dominant (T) over inability taste PTC which is recessive (t); Several animations**

- We each have two copies of the PTC receptor gene- one from our Mom, and one from our Dad
- The gene for tasting PTC is dominant over the gene for not tasting PTC which is recessive.
- Lets look at an example of three people with different genotypes. How do you think each of these people would respond to tasting PTC?
- The person on the left has two copies of the dominant gene for tasting PTC, so all of their receptors will be good at binding it.
- The person in the middle has one dominant PTC tasting gene and one recessive non-tasting gene, so some of their receptors will be good at binding PTC while others wont.
- The double recessive genotype, little T little T, gives a NON-TASTER phenotype that never tastes anything on the strip.
- Which person were you? Where you a supertaster that tasted the bitterness right away? Did it taste a little bitter after a while? Or did you just taste paper? Write down your phenotype, we are going to use it later in our data analysis.

### **Drug receptor summary**

- But how is our experiment related to personalized medicine?
- As we have learned, the Ability to taste PTC has a very strong **genetic component**.
- PTC is a chemical and Drugs are also chemicals.
- You have just performed a genetic test for your reaction to a chemical!
- And the differences in how your class tasted PTC is very similar to the difference in patients reactions to drugs
- How did scientists find that patients react differently to Tumoricide?

**Helped/Hurt Breast Cancer Case Study (Home slide):** Remember the case study from earlier with the 10 patients, 7 who were not helped by the drug Tumoricide and 3 who benefitted? Why do you think these women responded differently to this drug? What do you think scientists found? Do you think it has anything to do with a receptor?

**Tumoricide is a personalized medication:** Scientists found that there are two different kinds of breast cancer cells. One kind have receptors called Her2 receptors, while the others do not. When the Her2 receptors are present, Tumoricide can bind and helps the patients by killing the cancer cells. Just like PTC binds to the PTC receptor allowing you to taste it. However, when

the cells do not have Her2 receptors, Tumoricide doesn't work and may actually do harm to the patient.

**Helped/Hurt Breast Cancer Case Study (Home slide) Her2+/-:** In this case study, only the three women who were helped by Tumoricide had the Her2 receptor on their breast cancer cells. The other women were Her2 negative or lacked the receptor. By identifying patients with Her2+ breast cancer cells, doctors are able to prescribe Tumoricide to the right patients!

**Screening for Her2+ Cells:** One way patients are screened for the Her2 receptor is through staining a tissue biopsy from their tumor. The membranes of the cells stain brown when the receptor is present in high amounts on the cells. The image on the left shows tissue from a Her2 negative tumor, there is no brown staining. On the right is an image from a Her2 positive tumor, see the brown membranes? This staining technique is a way to detect Her2 receptors in breast tumors, just as the PTC strip test was a way to detect PTC receptors on your tongue.

**Screening for Her2+ Cells (Would you test this patient?):** Do you think a patient with this staining result should be administered Tumoricide?

**Screening for Her2+ Cells (Would you test this patient?) YES:** Yes, this patient is positive for Her2 and would likely respond well to Tumoricide!

**Breast Cancer Treatment Timeline:** If the three women in this case study had been diagnosed with breast cancer in 1990 they would have had chemotherapy and radiation, but not Tumoricide treatment. Now with the discovery of Tumoricide and the ability to test for the receptor doctors can do a better job of giving Tumoricide to those women who will benefit from the drug and not cause risk of heart problems in women who will not be helped because they don't have the receptor.

*Now, let's talk about other physiological traits that can influence an individual's response to drugs.*

### **Transition from Tumoricide back to PTC**

So we just discussed how the HER-2 receptor influences a patient's response to Tumoricide..

...and we saw in class how the PTC receptor influences the ability to taste PTC.

### **Where are the PTC receptors? Several Animations**

To start address the questions how other physiological factors may influence your ability to respond to a medicine, let's revisit the PTC story. Can anyone think about where the PTC receptors are located in our bodies? Since we know that we sense PTC by tasting, where would the cells be that contain the PTC receptors...

If you were thinking about the cells on your tongue—then you are right! The PTC receptors are found on your taste buds—which are on your tongue. We often think of taste buds as helping us enjoy a yummy piece of cake, like shown in this picture of a little boy who has just enjoyed some cake himself.

### **What are taste buds? Animations build these concepts**

But what exactly are taste buds. Taste buds are found on the papillae of your tongue—these are the bumpy things on your tongue. Taste buds are actually clusters of cells that are found on the papilla and the PTC receptors are found on your taste buds. The PTC receptor is just one type of receptor that is found on tastebuds—there are many different kinds of receptors that allow you to taste different types of foods.

Okay—so the PTC receptors are found on your tongue, but how does this result in us “tasting “ the PTC chemical. When the PTC chemical binds to the PTC receptor on the taste bud, it excites a nerve cell that is connected to the papilla. The nerve cell then transmits a signal to the brain. When your brain receives this signal..you respond by thinking –WOW! That’s really bitter.

### **Question**

So—lets ask ourselves a scientific question—Are there other traits that can allow a person to more strongly taste PTC?

Can you think of any reasons or hypotheses as to why some people could taste the PTC chemical more strongly than others—knowing that the PTC receptors are found on your tastebuds?

One hypothesis could be stated like this: If a person has more tastebuds, then he/she may be able to taste the PTC chemical more strongly.

### **Let’s test our hypothesis and count our taste buds!**

- 1) Lollipop time! Lick your lollipop such that the blue gets all over your tongue...especially the tip of your tongue.
- 2) Once your tongue is really blue, place one hole reinforcer on the tip of your tongue—so it looks like the picture on the bottom of this slide.
- 3) Have your partner count the bumps or papillae on your tongue—these will not stain blue.

**\*\*Remember that your taste buds are on your papillae. Therefore, the number of papillae correlates to the amount of taste buds on your tongue\*\***



## **Counting the number of papillae**

Here are some examples of what you may see.

Come to the front of the class to report your PTC phenotype (taster, super-taster and non-taster) and the number of papillae on your tongue.

## **Ideal Graph – Animations build these concepts**

Here is an ideal graph representing the number of tongue papillae related to the ability to taste PTC. As you can see, these results support our hypothesis that the super-taster has more papillae!

Now, let's think about the non-taster. The number of papillae in the non-taster is variable—as shown here by the large error bars. Why would the number of papillae in a non-taster be variable—or to put it another way—why could non-tasters sometimes have a small number of papillae and a large number of papillae?

The answer to this question is that even if a non-taster has lots of papillae, they don't have the receptor; therefore, PTC chemical can not bind to the cell - therefore, it would not matter how many papillae are on a non-taster's tongue, because without the correct receptor these people are unable to taste the PTC regardless of the number of papillae.

## **What it takes to be a PTC taster?**

So in summary—What does it take to be a PTC taster?

There are two traits that are important for determining PTC taste sensitivity

- 1) The first one we learned about is your PTC receptor genotype—Do you have the receptors that enable you to taste PTC.
- 2) The second trait refers to how the density of papillae on your tongue correlates to the sensitivity of tasting PTC.

Thinking about how these two traits both are important for PTC taste sensitivity is really important when we are trying to understand personalized medicine. We need to consider many different aspects of how we might respond to medicine..

Another factor in this puzzle is understanding how your body processes drugs after receiving them.

## **What are the reasons a person would react differently to drugs?**

So far we've talked about two reasons a person's reaction to a drug may differ based on their genetics –finally we will talk about how the body processes drugs.

### **A Drug's Life**

Scientists have names for the four basic stages of a medicine's life in the body: absorption, distribution, metabolism, and excretion or ADME.

Medicines can be absorbed into the body in many different ways.

*Can anyone give me an example of how a drug enters the body?*

A few of the most common ways to administer drugs are oral like swallowing a pill, inhalation like using an asthma inhaler, intramuscular like getting a flu shot in an arm

After that the drug is absorbed into the blood stream it is distributed throughout the body to the targeted organ.

Then it is broken down, or metabolized. This usually occurs in the Liver, the body's chemical processing and detoxifying plant where chemicals are cut apart, stuck together, or transformed, making the substance easier to get rid of or excreted.

There are many proteins involved in each of these processes. You can imagine if a person had a genetic variation in a gene that affects one of these proteins then there may be a roadblock in the ADME process and a drug might not work the way it was intended.

### **Metabolic enzymes**

For example, there are several genetic variations in a group of proteins in the liver that are involved in breaking down and deactivating drugs. These specific metabolic proteins are called enzymes.

A genetic variation in liver enzymes can affect a person's ability to process certain drugs. Less active or inactive forms of these enzymes are unable to break down and efficiently eliminate drugs from the body thus causing a drug overdose

While more active forms of these enzymes can cause a patient to excrete the drug before it has a chance to work.

### **Adverse drug reactions (ADR)**

When we think of taking a medication we usually think of them treating or preventing a problem. However, most medications, even when given at the normal dose, have the potential of harming us. Doctors call these unwanted side-effects an adverse drug reaction or ADR.

*Can you name some examples of an ADR?*

They range from the less severe like rash or headache to life-threatening like stroke or heart attack.

There are multiple causes for ADRs. We know that both genetics and environment play a role in how people react to medications. Some environmental causes may be someone's diet, taking other drugs, age, and amount of physical activity.

An example of a genetic cause could be a genetic variation in a metabolic enzyme in the liver. Someone who is a poor metabolizer - the enzymes in their liver are not as efficient- can experience an ADR at even a normal drug dose.

### **Case study: Nortriptyline metabolism**

Let's take for example three women all the same height, weight, age, and racial background. These women are depressed and go to the doctor. After the doctor examines them she prescribes an antidepressant, Nortriptyline at a dose of 100mg.

However, each of the women react differently to the drug.

Person **A** has an adverse reaction. Person **B** nothing happens – she still is feeling depressed, and person **C** feels better.

### **Case study: Nortriptyline metabolism (2)**

*Why do you think this happened?*

#### **ADME of Nortriptyline**

Well, a doctor trying to figure out what was going on in these women decided to examine the ADME process by measuring the amount of active Nortriptyline in their blood.

*Can you guess which patient will have the most and which patient will have the least amount of active drug in their blood?*

#### **ADME of Nortriptyline (2)**

As you can see the doctors saw that patient A had 95mg, patient B 5mg, and patient C 50mg.

#### **DNA variations influence drug metabolism**

Patient A is considered a poor metabolizer – there is still a lot of drug left in their blood causing unwanted side effects because the enzyme in her liver is not breaking it down fast enough,

## **DNA variations influence drug metabolism (2)**

Patient B is an ultrarapid metabolizer – the active drug is excreted out of her system before it has a chance to work because her liver enzyme is so efficient,

## **DNA variations influence drug metabolism (3)**

and Patient C is an intermediate metabolizer – the drug is metabolized at a moderate rate which left enough drug in her body to treat her symptoms but did not cause side effects.

Scientists recently determined that the differences in people's metabolism for Nortriptyline is due to a genetic variation in a specific enzyme in their liver.

We should also note that drug metabolism is drug specific, so one person may metabolize one drug quicker than another drug and there are environmental aspects that can affect metabolism.

## **2011 – What do doctors do?**

Doctors today would re-examine the patients and change the drug or the dose for each patient who does not respond or responds with an adverse drug reaction after they have tried out the “normal” dose.

For example, patients like person A who is a poor metabolizer would need less drug

And patient B who is an ultra-rapid metabolizer would need a much higher dose to achieve the same effect.

## **Today One-Size-Fits-All drugs**

Currently drugs are developed for the average patient, and as we saw from the example of Nortriptyline, right now there is no simple way to determine who will respond well and who will respond poorly without a guess and test method.

We know that each of us are different genetically and one size does not fit all! Drugs need to be personalized.

So what is the solution?

## **Today One-Size-Fits-All drugs**

Pharmacogenomics or personalized medicine

By using the information found in your DNA, pharmacogenomics will take the guesswork out of finding the right drug, it will speed recovery time and increase safety as the likelihood of adverse reactions is decreased. This may ultimately decrease the cost of healthcare.

## **APRIL, 2050**

Pharmacogenomics offers a very appealing alternative to the one-size fits all. Imagine a day when you go into your doctor's office and, after a simple and rapid test of your DNA, your doctor changes their mind about which drug they will offer you.

Researchers predict that the medicines of the future may not only look and work differently than those you take today, but tomorrow's medicines will be tailored to your genes boosting their effectiveness and minimizing possible side effects.

### **Summary**

Today we learned that variation can lead to phenotypic differences and therefore there are differences in how we all process drugs -

Like the variation in people's receptors that allow them to taste PTC. Or the variation in receptors on a breast cancer tumor that makes it more or less susceptible to the drug Tumoricide.

We learned that there are other physiological traits that enable you to respond to a drug

Like the number of taste buds on your tongue

We also learned how drugs undergo absorption, distribution, metabolism, and excretion and about enzymes in the liver where genetic variation can lead to people metabolizing drugs differently.

### **Pharmacogenomics**

We learned that most of today's medicines are one-size-fits all.

Pharmacogenomics is a developing research field that is still in its infancy.

We need a new generation of research scientists, and doctors to work together to find the best ways to develop new therapies.

We will one day give patients the right drug at the right dose at the right time based on the information that is within their DNA.

---