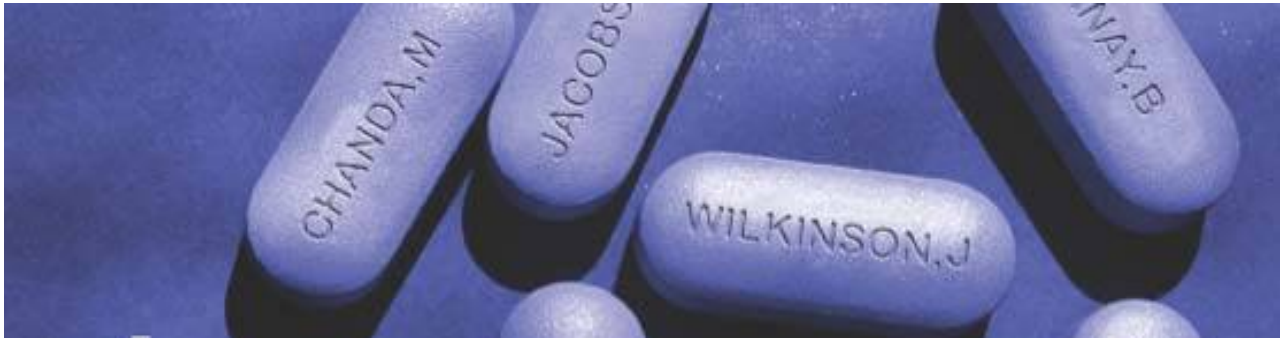
Pharmacogenetics
&
Pharmacogenomics



Personalized Medicine

- So, how many of you have tried ibuprofen (Motrin, Advil)? How many of you have tried naproxen (Aleve)?
- Who thinks ibuprofen works better? How about naproxen?
- Why do you think this is? These drugs work on the same target, cyclooxygenase.
- We all have small, inherited differences in our cyclooxygenase enzymes. Thus, we respond differently to these drugs. Due to pharmacogenetics, we are forced to respond in a different way.

Drugs and Genes

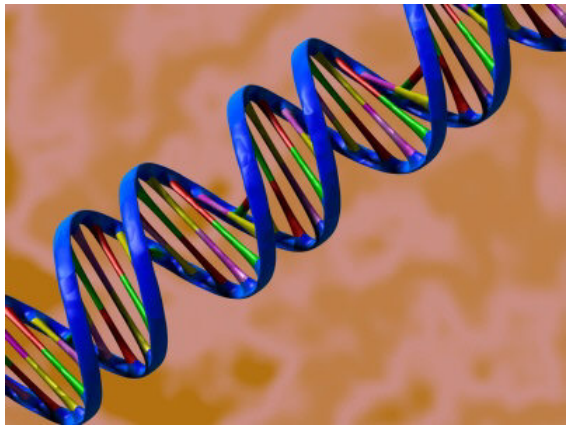
- Pharmacogenetics:
  - Study of the genetic factors that influence an organism's reaction to a drug

- Pharmacogenomics:
  - Development of drug therapies to compensate for genetic differences in patients

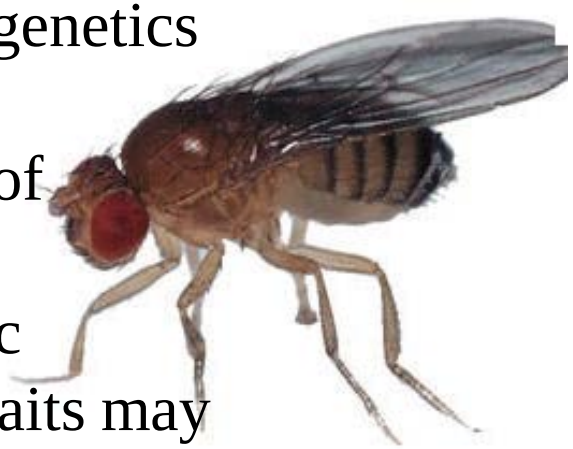
- You guys had a lot of definitions yesterday when learning about dose response, and you will have a bunch here too.
- Dose response and pharmacogenetics are related. How drugs behave in the body will depend on genes in some way.
- First, we'll define pharmacogenetics, whicalcohol. This is a classic example of pharmacogenetics. Some of you may respond with a flushed face, rapid heart rate, dizziness, while others may not. We'll elaborate some more on this concept in a few minutes. h is…
- I bet some of you in the room have had
- Pharmacogenomics is more on a population/clinical trial level and is defined as…

Review of Genes

- Genotype:
  - The genetic makeup of an organism
- Phenotype:
  - An organisms' observable traits



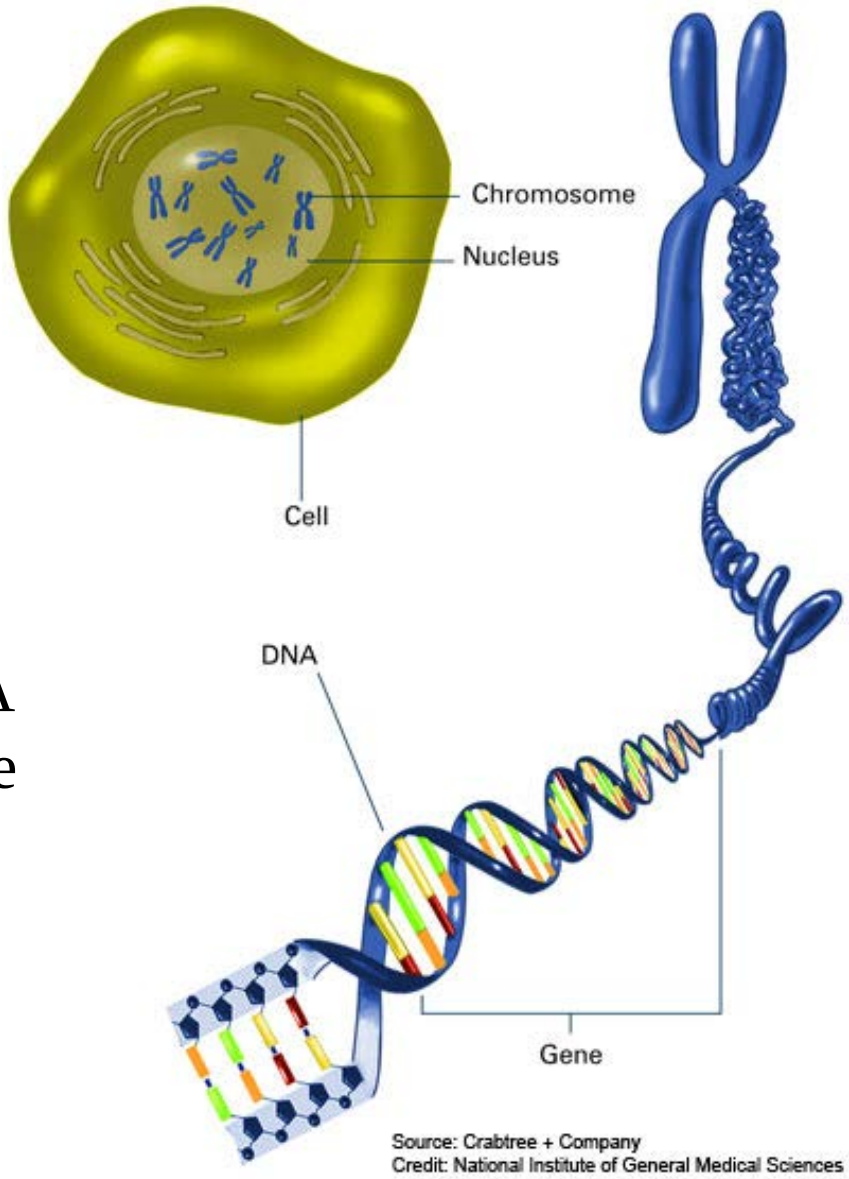
Here is a brief review of the central dogma of genetics
Genotype involves the DNA sequence, while phenotype is any measurable, observable trait of an organism that results form the genotype.
Gene expression results in patterns and specific interactions to give you what you see. These traits may include physical characteristics, behavior, biochemical measurements, etc.
We are going to talk today about how genotypes cause different phenotypes, including how we react to drugs.



Human Genome is Variable

DNA is wrapped into structures called chromosomes within a cell's nucleus, and DNA is made up of base pairs of different nucleotides.

Remember your A's, T's, C's, and G's?



Human Genome is Variable

- Contains $3x10^9$ base pairs of DNA
  - 3 billion!

- Between 2 people (except identical twins) the rate of genetic variation (individuality) is about 0.1%

  [0.1% of 3 billion = 3 million base pair differences]

DNA contains…

Every once in a while, there can be a different pair of nucleotides than what is found in the population. It is replaced with a different pair.

Between 2 people…

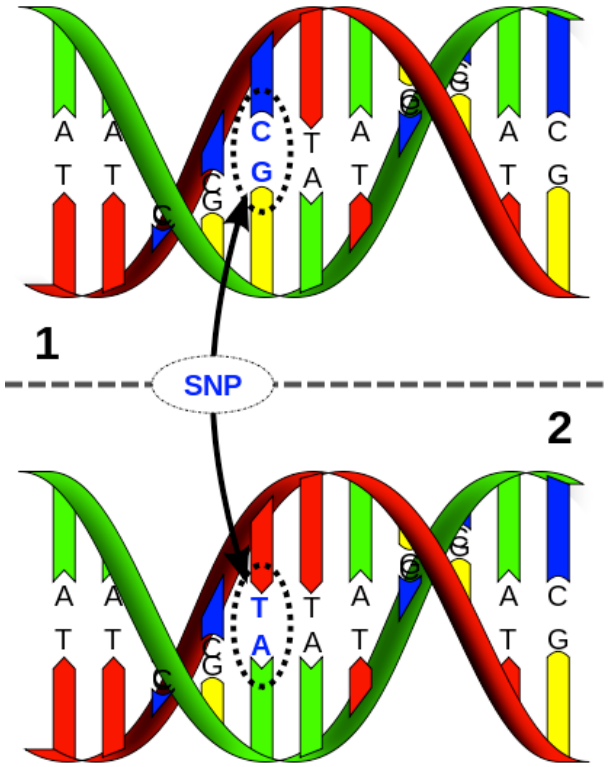
That is still a lot of differences (3 million base pairs are different). What can explain this variance?

Single Nucleotide Polymorphism

- The most common cause of genetic variation
- SNPs occur on average every 1000 bases
- Understanding SNPs has shown promise for improving disease detection and treatments

Polymorphism
"Poly" *Many*
"Morphe" *Form*

General Population 94%



Single Nucleotide Polymorphism 6%

- Well, the variations in the DNA have a name, they are called single…
- The term polymorphism can be broken down into the root words for “many” and “form.”
- SNPs occur…

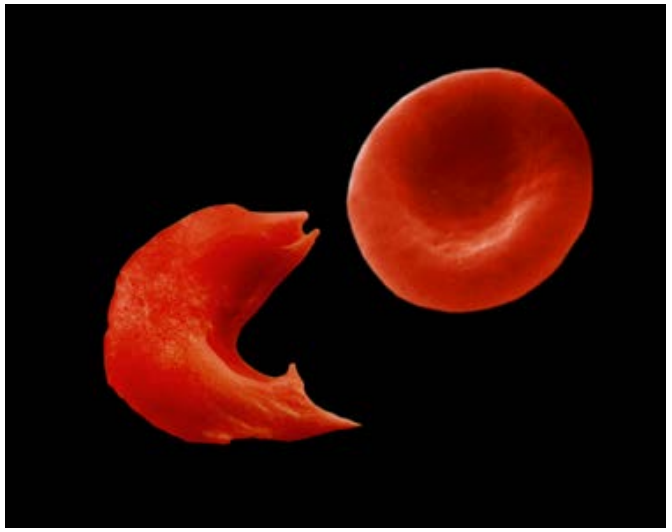
In the diagram in the corner, the general population may have a G at a certain point in the DNA sequence, but 6% of the population has a SNP at this location. In order for a SNP to be called a SNP must have occurred in ≥1% of population. Therefore, 6% indicates the SNP is authentic.

- We can accumulate mutations over the course of our lives.
- Understanding…
- Can you think of any diseases in which certain groups are at a higher risk for the disease than other groups?

Examples of SNPs and Disease

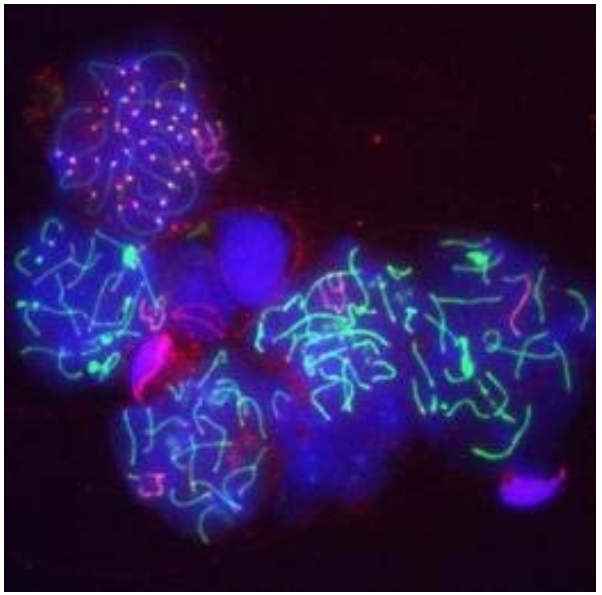
SNPs in Beta-globin

Sickle Cell Anemia



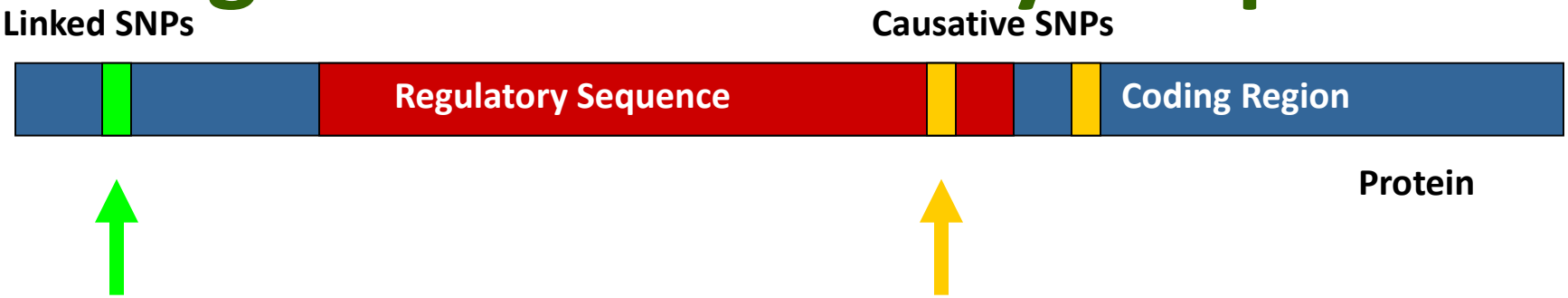
SNPs in BRCA1

Predisposed for breast cancer



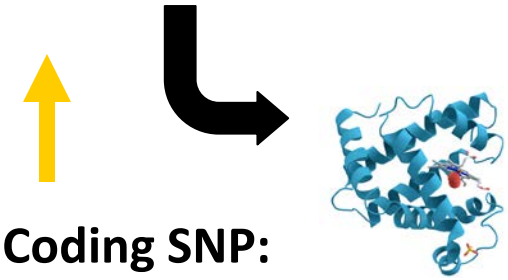
Here are some examples of how SNPs affect us in terms of disease.
In sickle cell anemia, a SNP in B-globin causes the cell to be unable to carry oxygen properly.
Having a SNP in BRCA1 doesn’t cause cancer. However, the SNP causes a loss of the ability for the cell to be protected form environmental risks that may contribute to breast cancer.
We know more in terms of SNPs and how they affect disease states.
We want to use this same information to see how SNPs may affect response to drugs.
Let’s look at SNPs in a graphical way. Where do SNPs hang out?

Single Nucleotide Polymorphisms



Outside gene

Non-coding SNP: changes amount of protein



Coding SNP: changes amino acid sequence (function)

Does everybody understand this so far?
Changes can occur anywhere in the DNA sequence.
Let's take three minutes to do a think, pair, share.
We have these changes in protein where we can either get a different flavor of protein or different amounts. You also learned that you can have different subtypes of proteins that may bind better to a drug.
There can be changes outside the working gene (these SNPs help scientists find or "map" where the business end of a molecule may be changed), but let's focus on the two SNPs in the working part of the gene.
What do you think happens to protein production when there is a SNP in the regulatory sequence (where instructions for proteins are located)? What happens when there is a change in the coding region?

The region in the red has instructions as to how and how much protein should be produced.
The blue coding region involves the specific amino acid sequence.
Will you be getting the correct protein? One mistake can lead to a big change in proteins.

Here are some examples of how SNPs affect us in terms of disease.
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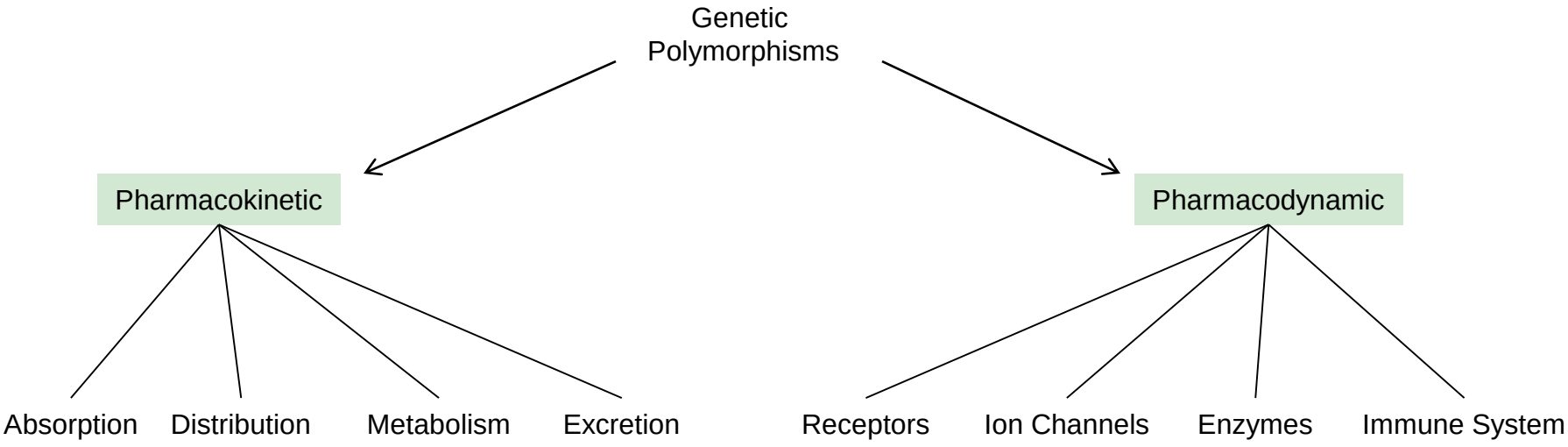
SNPs Can Change Drug Response

- Pharmacokinetic: changes in drug metabolism
- Pharmacodynamic: changes in drug targets
  - enzymes, receptors, or transporters



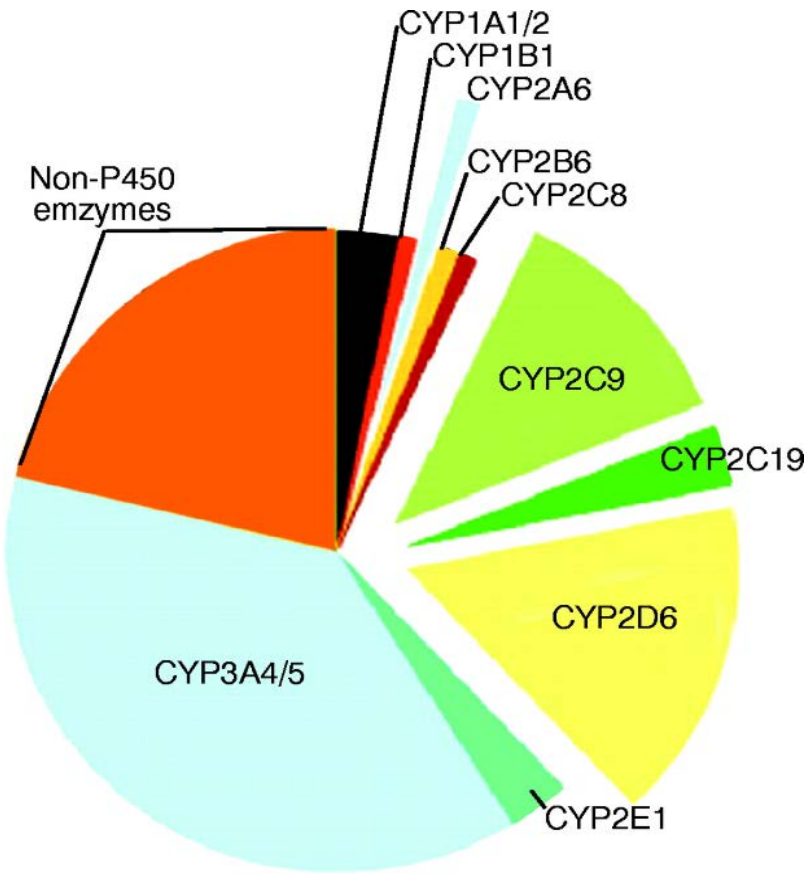
This slide is the heart of this lecture.

- If we have SNPs and take a drug, we can have different things happen.
- Let's look at the pharmacokinetic side of the story.
- Pharmacokinetics involves changes in drug metabolism, or what the body does to process a drug.
- If you had SNPs in metabolizing enzymes, what would it do to the drug? We talked a lot about morphine yesterday. Do you guys remember the main metabolite?
- What would happen if you had a SNP in the enzyme that makes morphine sulfate?
- You can have more or less of the active drug circulating which could lead to too either heightened side effects such as increased sedation or not enough pain relief, respectively.
- Pharmacodynamics involves changes in drug targets, or what the drug does to the body. What would happen if you had a SNP in the receptor for morphine?
- You could have better or worse binding to the drug. Other drugs may be more appropriate for pain relief.

SNPs and CYPs

- Cytochrome P450 enzymes are commonly found with SNPs

As a brief review, CYPs (or Cytochrome P450 genes) produce enzymes involved in the metabolism of molecules, including drugs, and are subdivided into groups and subfamilies (CYP3A4, CYP2C9, etc). These enzymes are mostly found in the liver, but they are also found throughout the body (brain, intestine, etc).
This diagram shows the proportion of drugs metabolized by P450 enzymes. Most drugs are metabolized by CYP3A4. We worry about polypharmacy or taking multiple drugs metabolized by CYP3A4 due to drug-drug interactions.



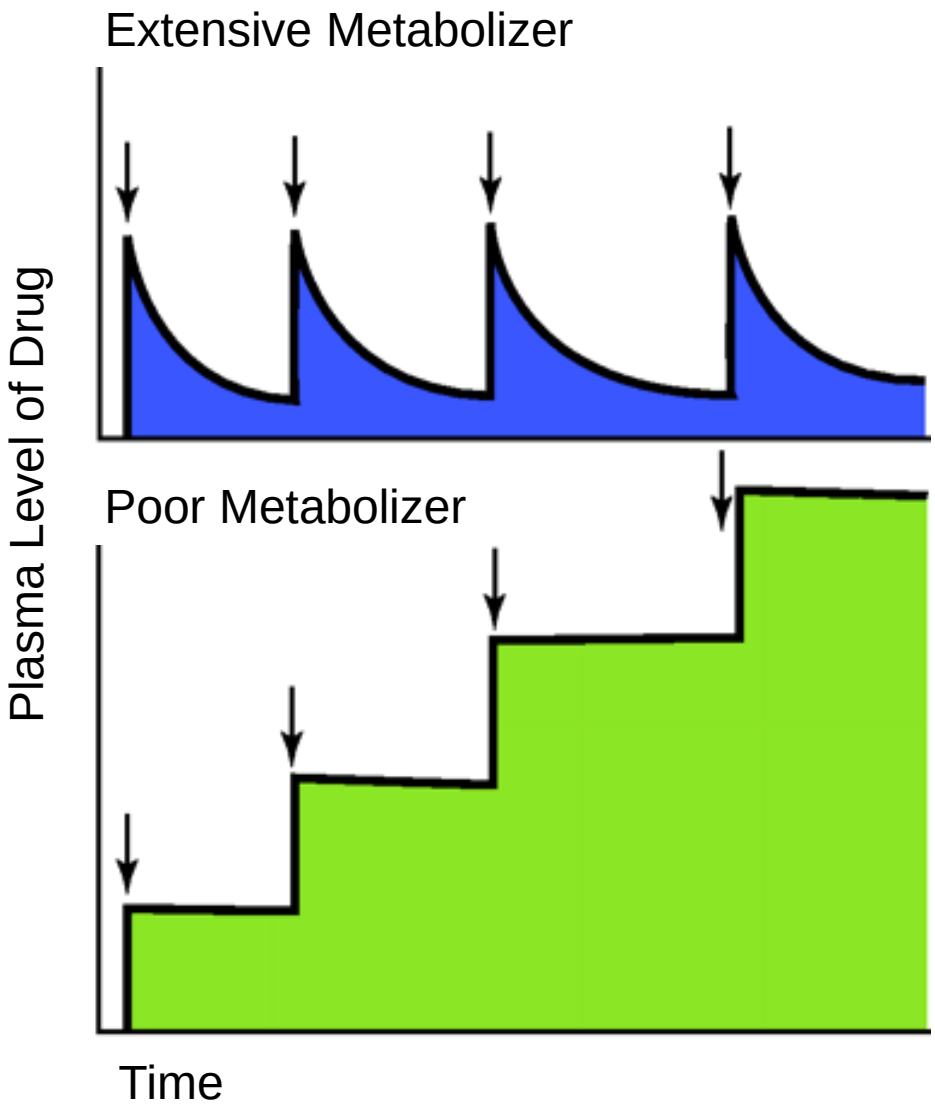
Drug Metabolism

Polymorphism	Substrate
CYP2B6	doxorubicin, nicotine
CYP2C9	warfarin, doxorubicin
CYP2C19	diazepam, proton pump inhibitors
CYP2D6	beta-blockers, anti-depressants, codeine, tamoxifen
CYP3A4	erythromycin, HIV protease inhibitors

- Here is just a little SNP-it of CYPs with important polymorphisms that affect drug metabolism. Whenever we have a SNP in a CYP, there may be a difference in the way drugs work.
- For example, CYP2C9 has more than 50 different SNPs in the regulatory and coding regions. CYP2C9 metabolizes over 100 therapeutic drugs.
- CYP2D6 may metabolize up to 25% of drugs in the clinic.

The idea is not to memorize this list. Drugs have many different substrates, and many different drugs are metabolized by CYPs.

Effects of SNP in CYP2D6



1: Normal enzyme activity

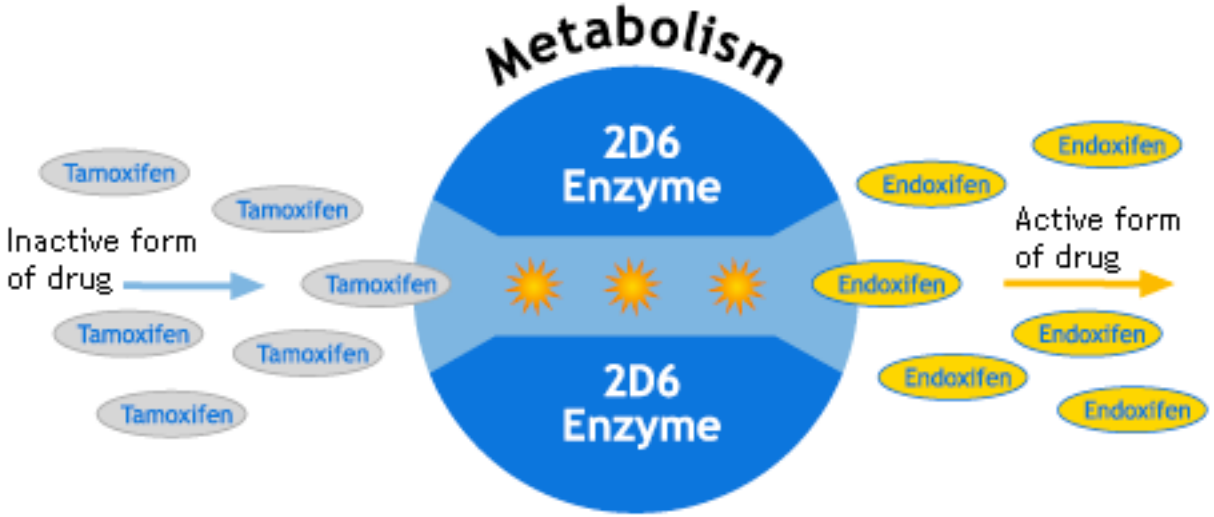
2: Decreased enzyme activity

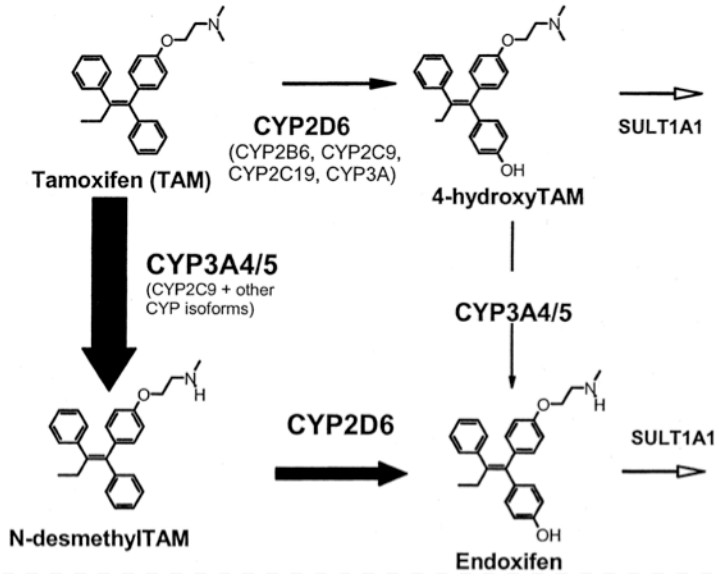
- Let’s take another couple of minutes for a think, pair, share activity.

- Patient 1 has normal enzyme activity. The y-axis is the plasma level of the drug, and the x-axis is time. Each arrow represents a dose of medication. There is an increase in the plasma level of the drug at each dose which gradually decreases over time.

- Patient 2 has decreased activity of their CYP2D6 enzyme. If you were this patient’s physician, how might you adjust their drug treatment? How might a graph of the plasma level of the drug over time look over a few doses?

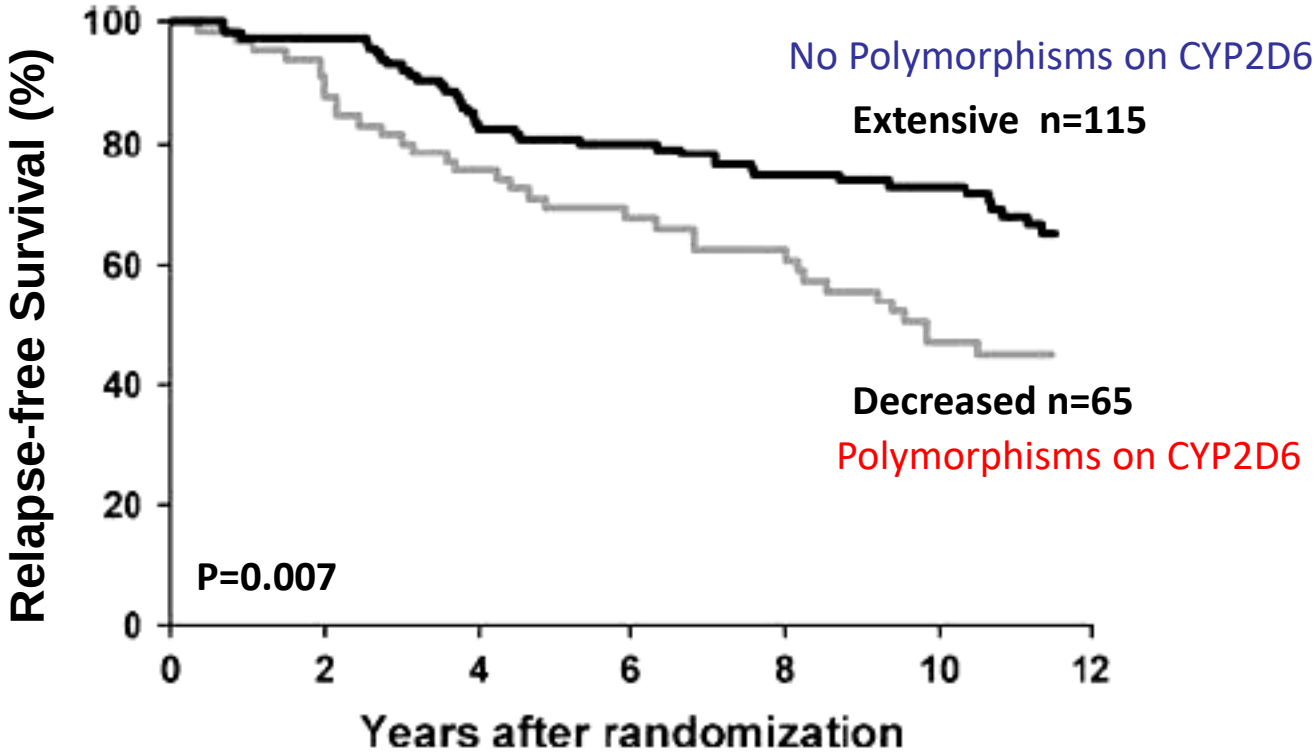
Pharmacokinetics of Tamoxifen





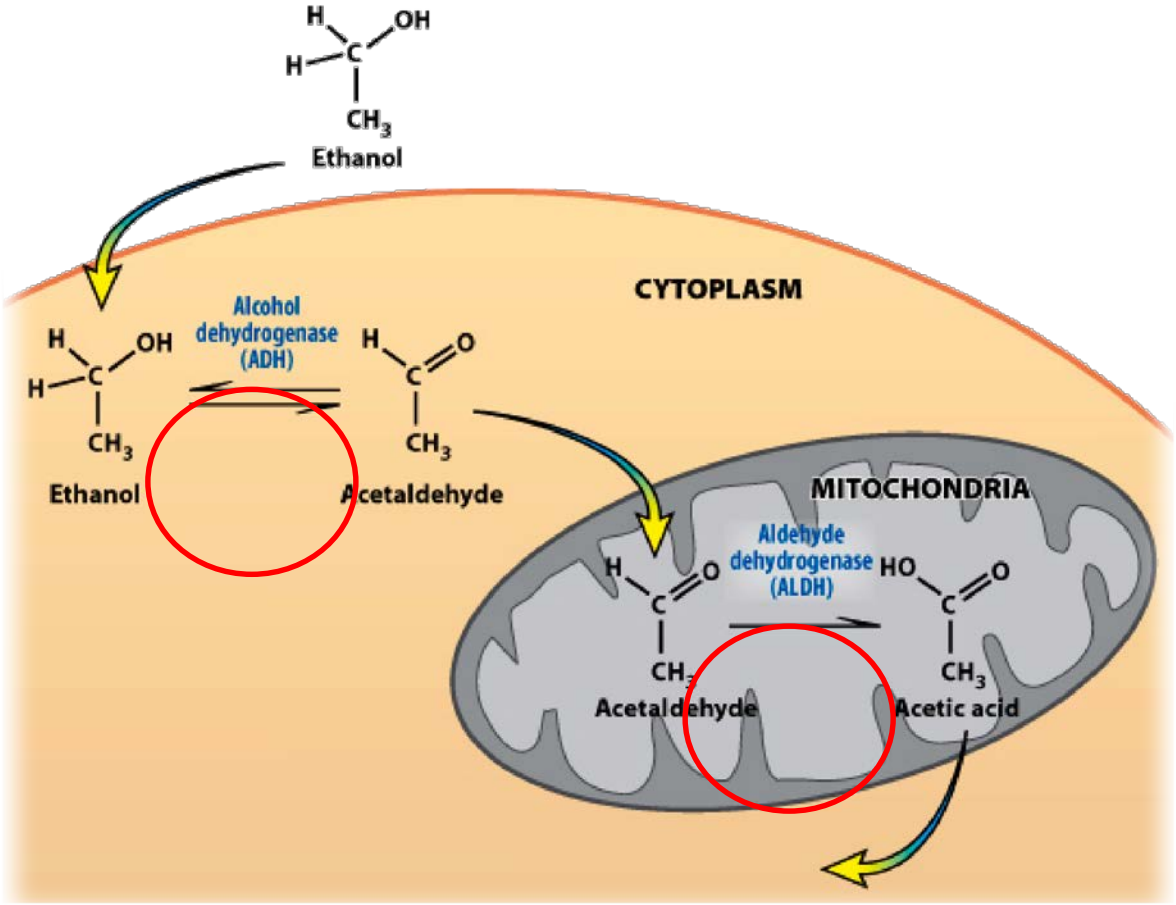
- We talked about this drug the other day during the metabolism lecture.
- Tamoxifen is a drug used for the treatment of breast cancer and is metabolized by the CYP2D6 enzyme. This is a special case where the metabolite is active. Do you remember what it is called? It is a prodrug. We see testing for the CYP2D6 enzyme in clinical practice.
- What happens when you have SNPs in the CYP2D6 enzyme? You can have reduced active amounts of the drug, which may result in therapeutic failure. It is important to know a patient's CYP2D6 status. They may be on the drug for 5 years after surgery and/or chemotherapy to prevent cancer reoccurrence.

Mutations Impact Treatment Outcome



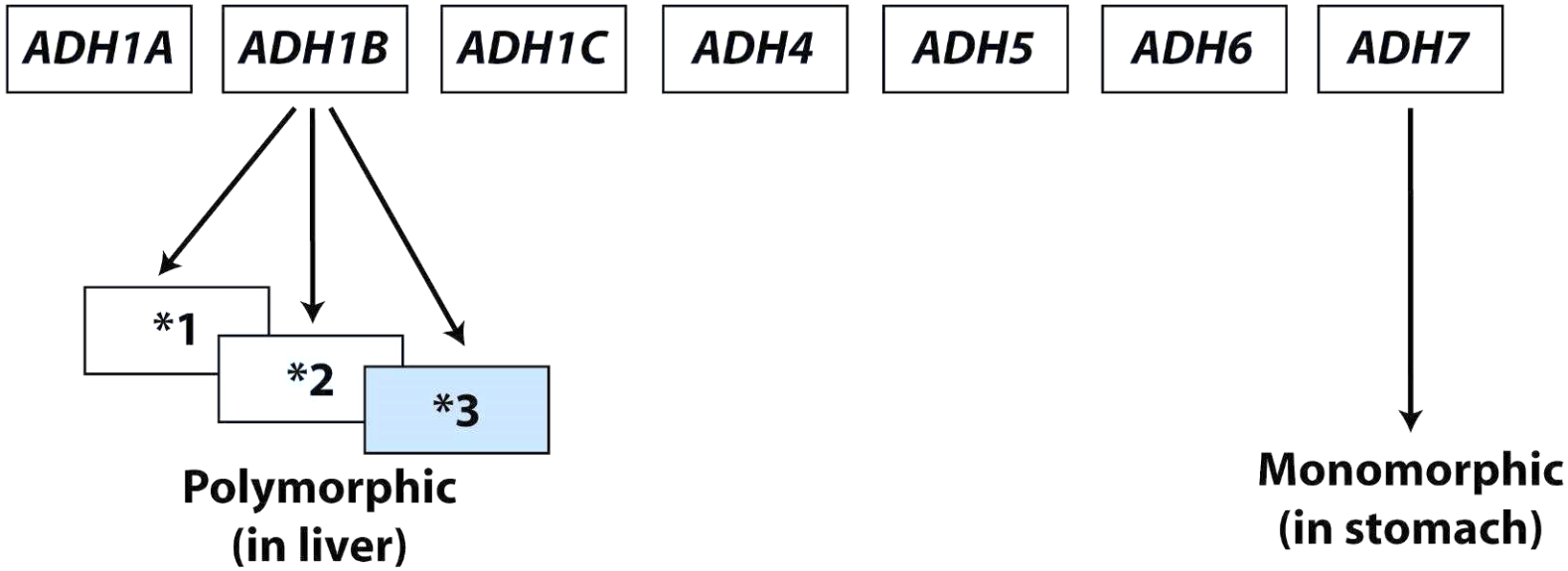
Clin Pharmacol Ther. 2008 January; 83(1): 160–166.

Metabolism of Alcohol… 2 Steps



There are two steps to alcohol metabolism. Similar to how the intermediate of acetaminophen is toxic, the intermediate of acetaldehyde is also toxic.

Genetic Polymorphisms and Differences in Alcohol Metabolism



There are 7 different kinds of the first enzyme, alcohol dehydrogenase, in the metabolic pathway of alcohol.

In people, there are polymorphisms, particularly in the ADH1B (an asterisk denotes a polymorphism). I bet that a lot of you in this room have had different reactions to alcohol. We are going to do an activity this afternoon that deals with how alcohol is metabolized.

Genetic Polymorphisms and Differences in Alcohol Metabolism

ADH Polymorphism	Metabolism Rate	Populations with High Prevalence
*ADH1B*1*	Slow	Most populations, including most Caucasians, Mexican Americans, and Native Americans Heavy drinkers
*ADH1B*2*	Fast	Most Asians, 25% of Jews, and some Hispanics
*ADH1B*3*	Fast	African-Americans and 6% of Mission Indians (S. West California)

Here is a table showing that different groups of polymorphisms tend to aggregate with certain populations.
If you are a fast metabolizer, you will have more acetaldehyde. Acetaldehyde is toxic. If you have an accumulation of acetaldehyde, then you will not like to drink. What about that second enzyme in the pathway that metabolizes acetaldehyde? Half of all Han Chinese have a polymorphism in the second enzyme so that they cannot effectively metabolize acetaldehyde.

23andMe

Why do your genetics matter?

Personalize your medication with your doctor.

Knowledge about variations in your DNA can help your doctor determine if you need more or less of a medication, or whether you might be at increased risk for certain side effects.

Your DNA can impact how effective drugs will be.

Clopidogrel (Plavix®) helps prevent heart attacks by keeping blood cells from sticking together. But a genetic variation that interferes with the drug's metabolism prevents some people from getting the full effect.

Genetics can determine if you'll have side effects.

Two genetic variations analyzed by 23andMe can increase the chances of experiencing severe muscular pain and weakness when taking high doses of cholesterol-lowering statins.

»See our health reports on 248 diseases and conditions.

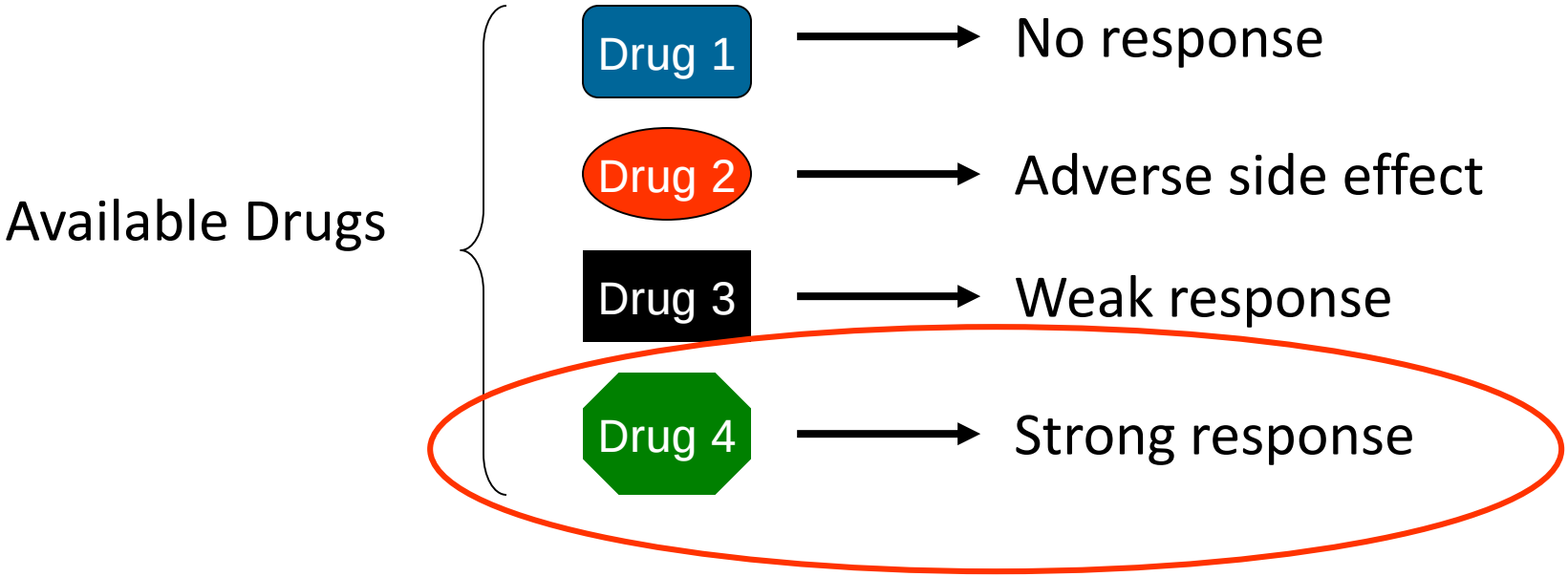
You don’t have to wait until you get a certain disease to find out if you have polymorphisms in your drug metabolizing enzymes.
23andMe is a commercially available Personal Genome Service. For $99 (recently reduced from $299, and before that nearly $1000), you can spit in a tube, ship it off, and, in 6-8 weeks, obtain information on your ancestry (relative finder, global origins, and ancestral lineages) and health (carrier status, disease risk, and drug response).
I sent my kit off about 3 weeks ago and am eagerly waiting for the results.

Better Drug Discovery with Pharmacogenetics & Pharmacogenomics

- Increase drug efficacy
- Eliminate adverse drug reactions
- Ensure success for drug approval in clinical trials
- Increase efficiency of clinical trials

Transitioning from the individual level, let's see how we can apply genomics on a clinical trial level. How much do you think it costs to bring a drug to market? It costs well over $1 billion to currently bring one drug to market from molecular discovery.

Drug Treatment

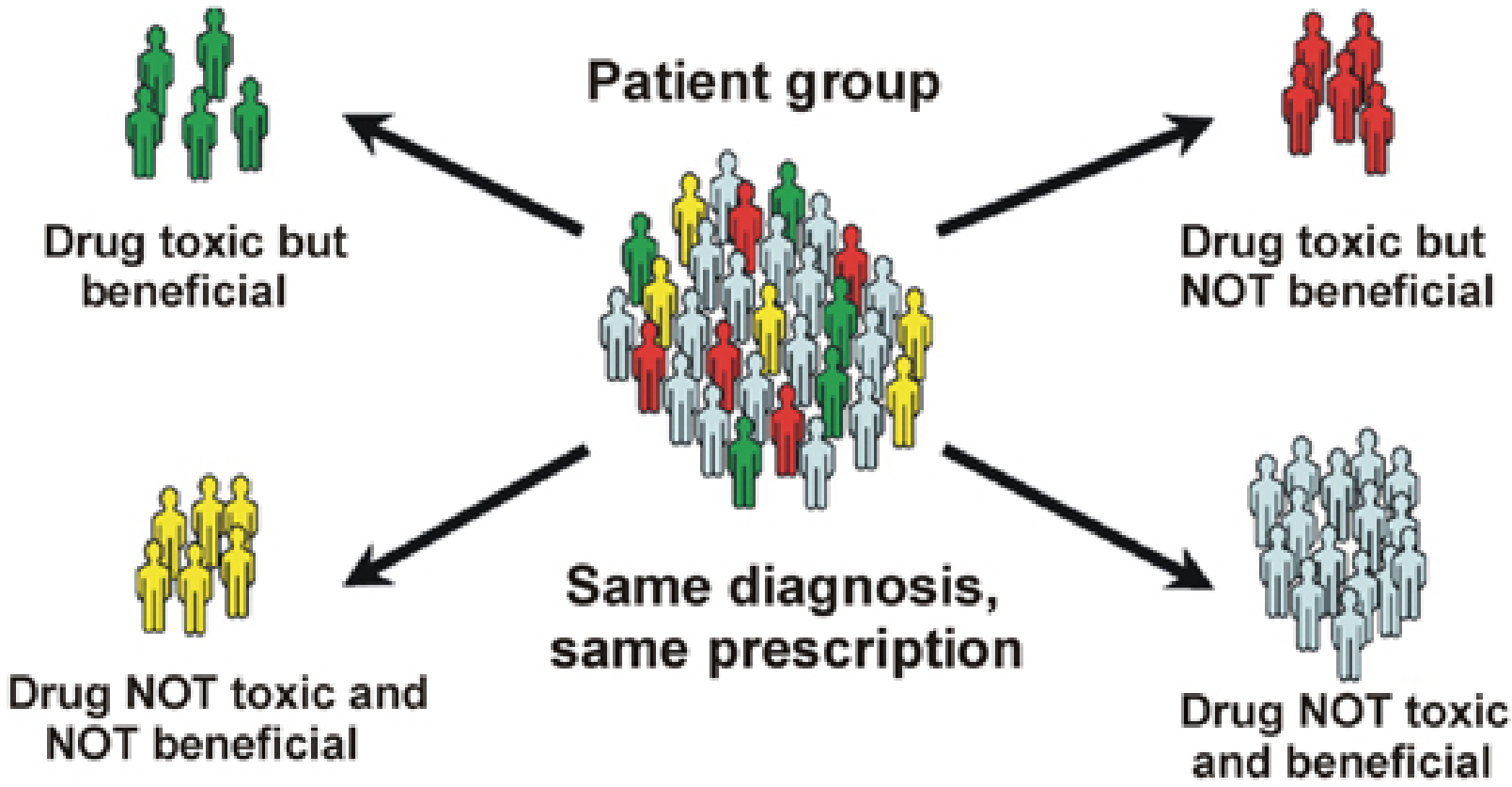


Currently best treatment determined by trial and error.

Future genomics may enable us to pick the right drug the first time.

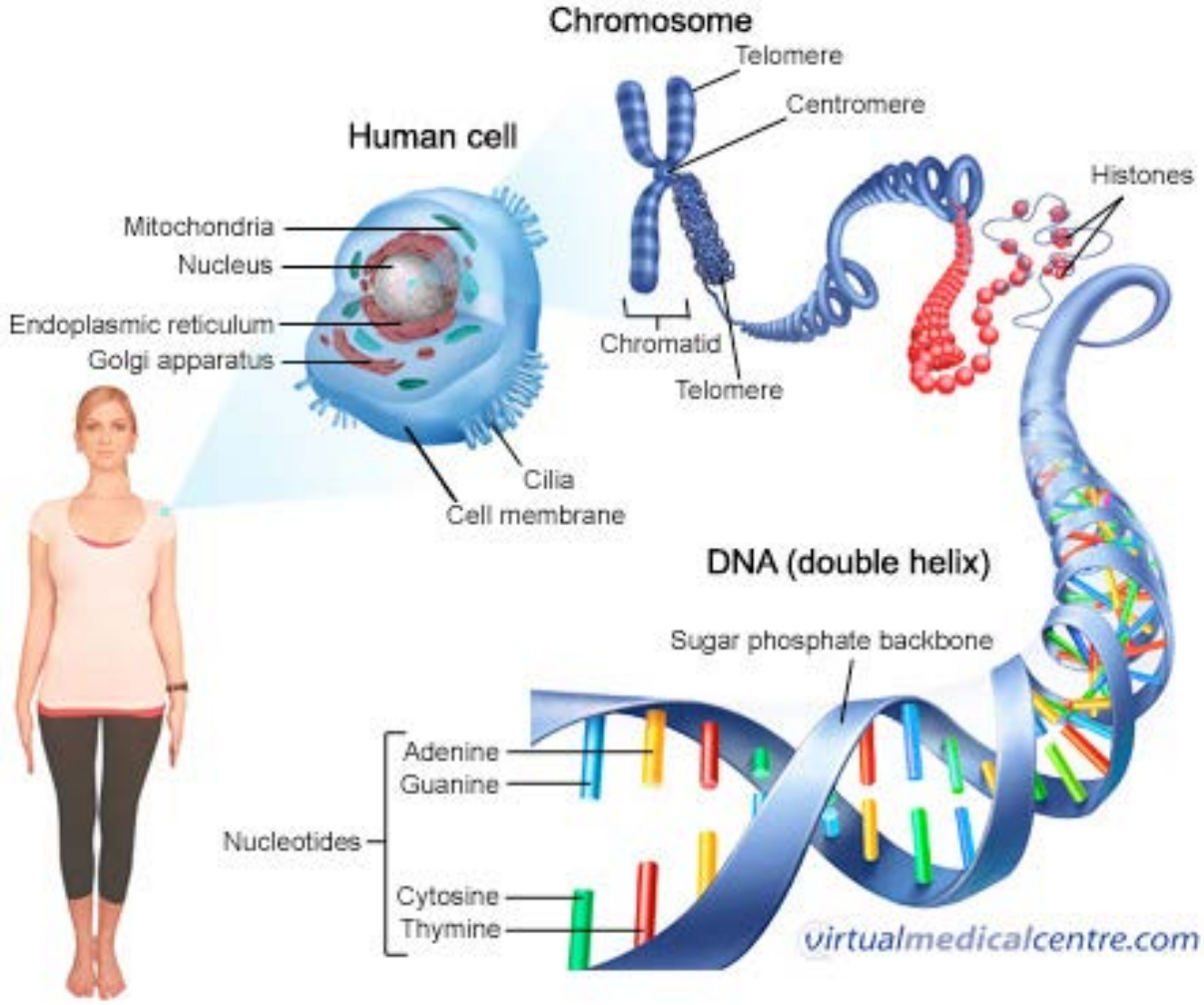
What issues do we have with drugs that we currently have on the market available to us? Currently, the best treatment is determined by trial and error. In the case of antidepressants (we have multiple kinds of antidepressants, may ask if your relatives responded to a certain medication before prescribing), the healthcare team try multiple drugs until one works. However, the study of pharmacogenetics/-genomics can help us pick the correct ones the first time.

Yesterday's Medicine



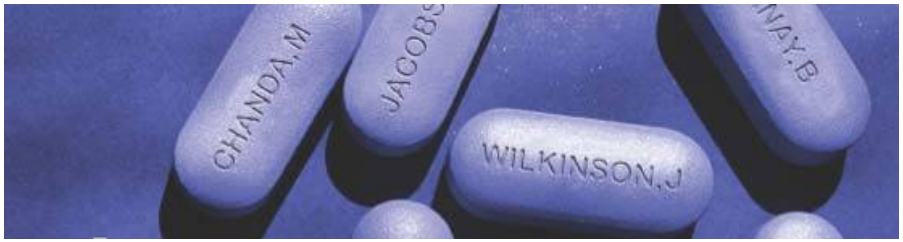
Many of today's drugs are only effective in about 40-60% of the population, but as long as there is no adverse affect in the remaining patients, physicians are able to prescribe them on a trial-and-error basis.

Tomorrow's Medicine



Tomorrow's vision is to utilize the genome to individualize therapy, build huge databases, and predict the patient's response to the drug before he or she even takes the drug.

Personalized Medicine



This individualization is called…

The right dose of

the right drug for

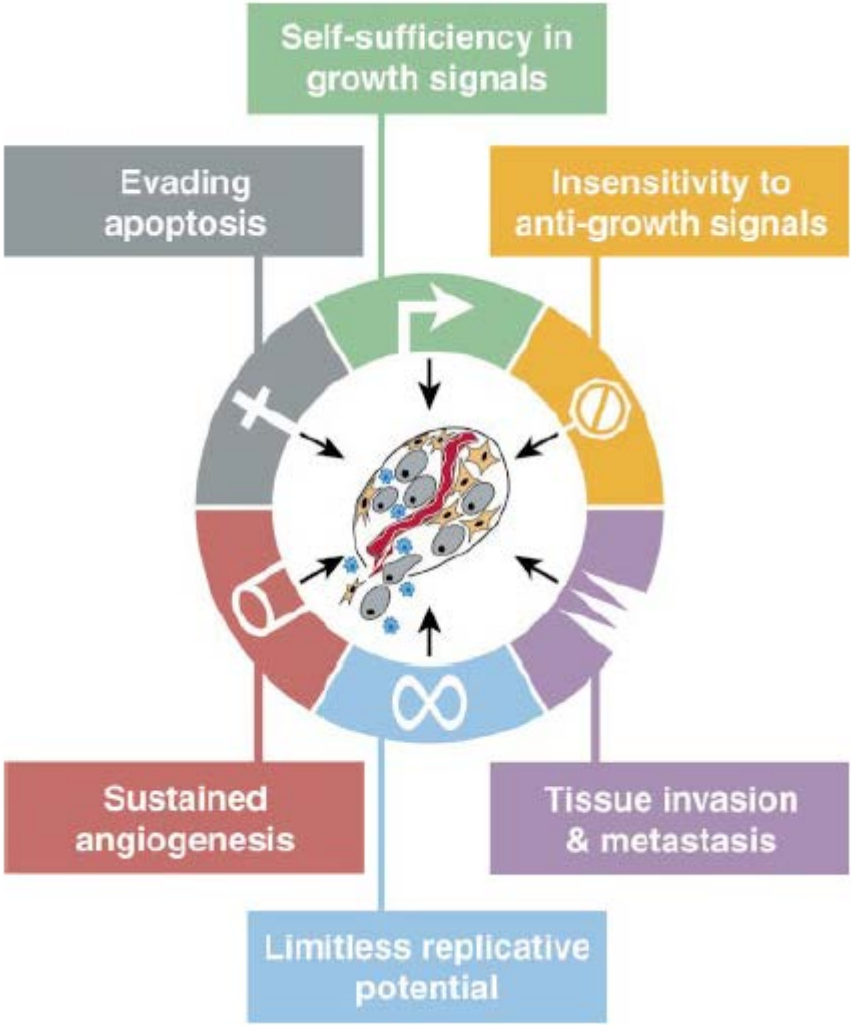
the right indication for

the right patient at

the right time.

Personalized Medicine: Cancer

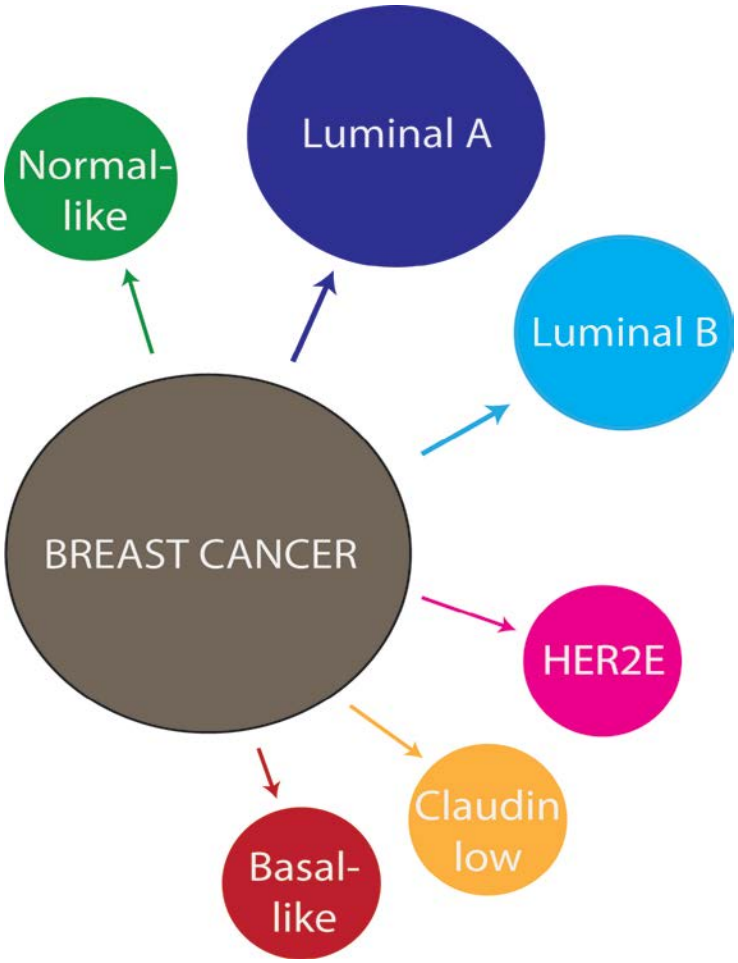
Many factors contribute to tumor formation.



Let's talk about a disease where personalized medicine is perhaps being most utilized. Many things contribute to tumor formation, and we'll talk about it next week in more depth. Let's focus on a certain type of cancer for this discussion.

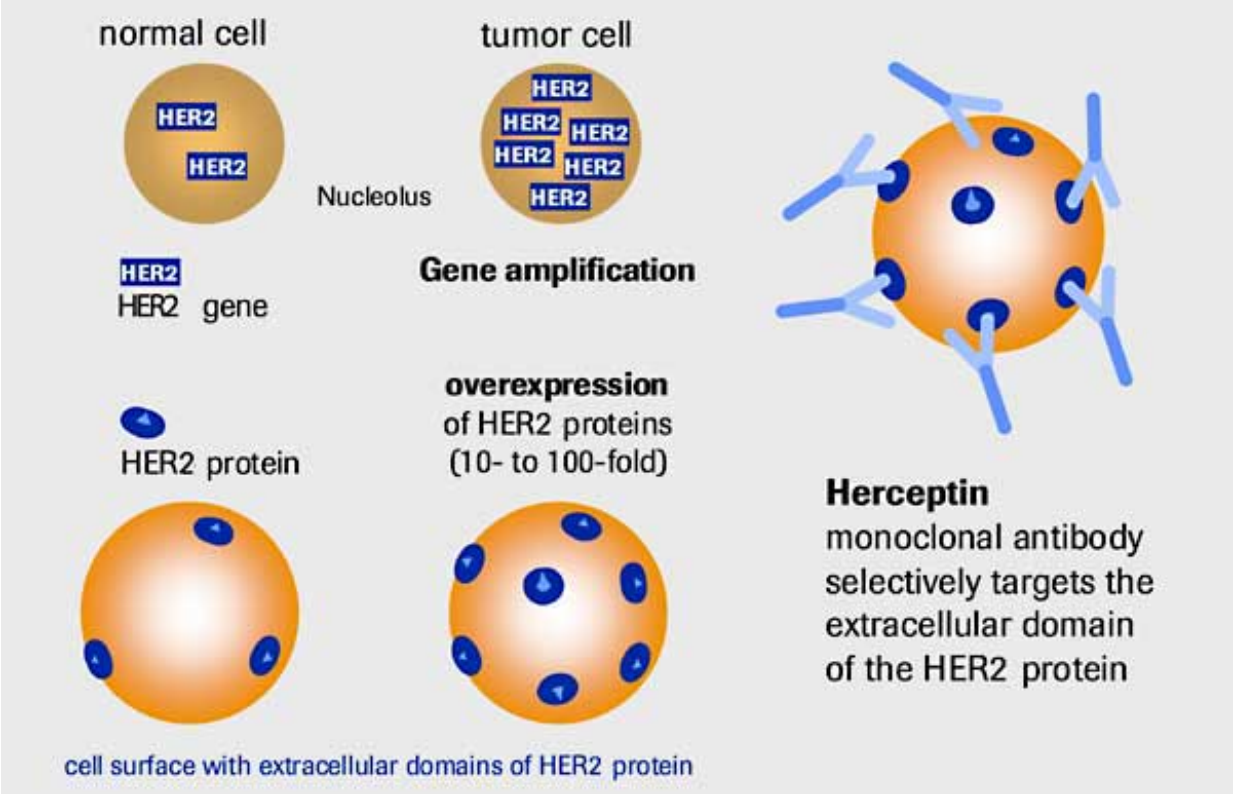
Breast Cancer is a Perfect Model

Breast cancer comes in many different subtypes, and we'll talk about one of them today. This subtype is conveniently named HER, which stands for Human Epidermal Growth Factor.



Pharmacogenomics: Gene Candidate to Drug Treatment

Human epidermal growth factor receptor-2 (HER2) in breast cancer cells



HER2 is a protein normally found in cells. It is a growth factor you need for normal cell function. In tumor cells, HER2 is found in larger than normal amounts. Polymorphisms lead to overamplification of the gene and overexpression of the HER2 proteins. We can create a drug to deactivate HER2 in tumor. Researchers developed an antibody, Herceptin or trastuzumab, to target HER2.

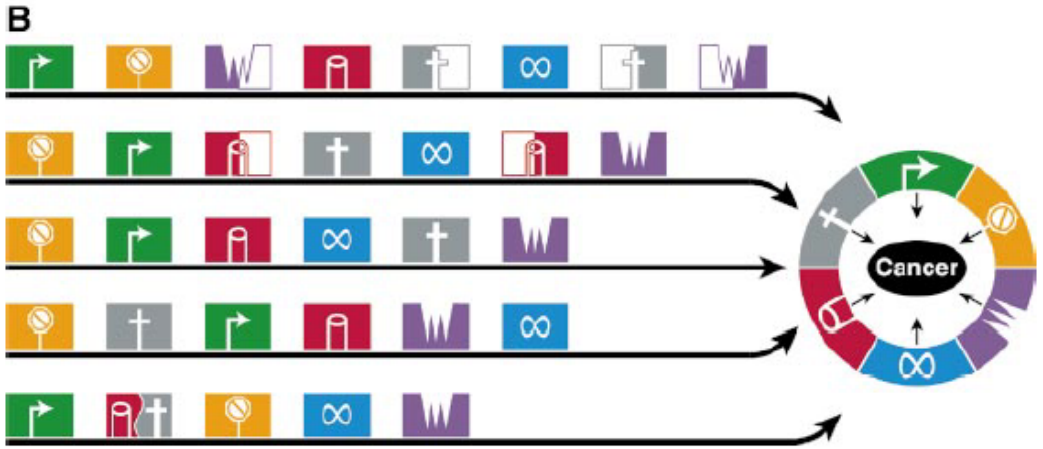
Herceptin in Clinical Trials

Trial Design	With HER2	Without
# of patients	470	2200
Response rate	50%	10%

So if we were to use information like this in clinical trials, we can really find if the drug has better efficacy without excessive trial and error. Look at how much better the response was in patients with HER22 versus patients negative for HER2.

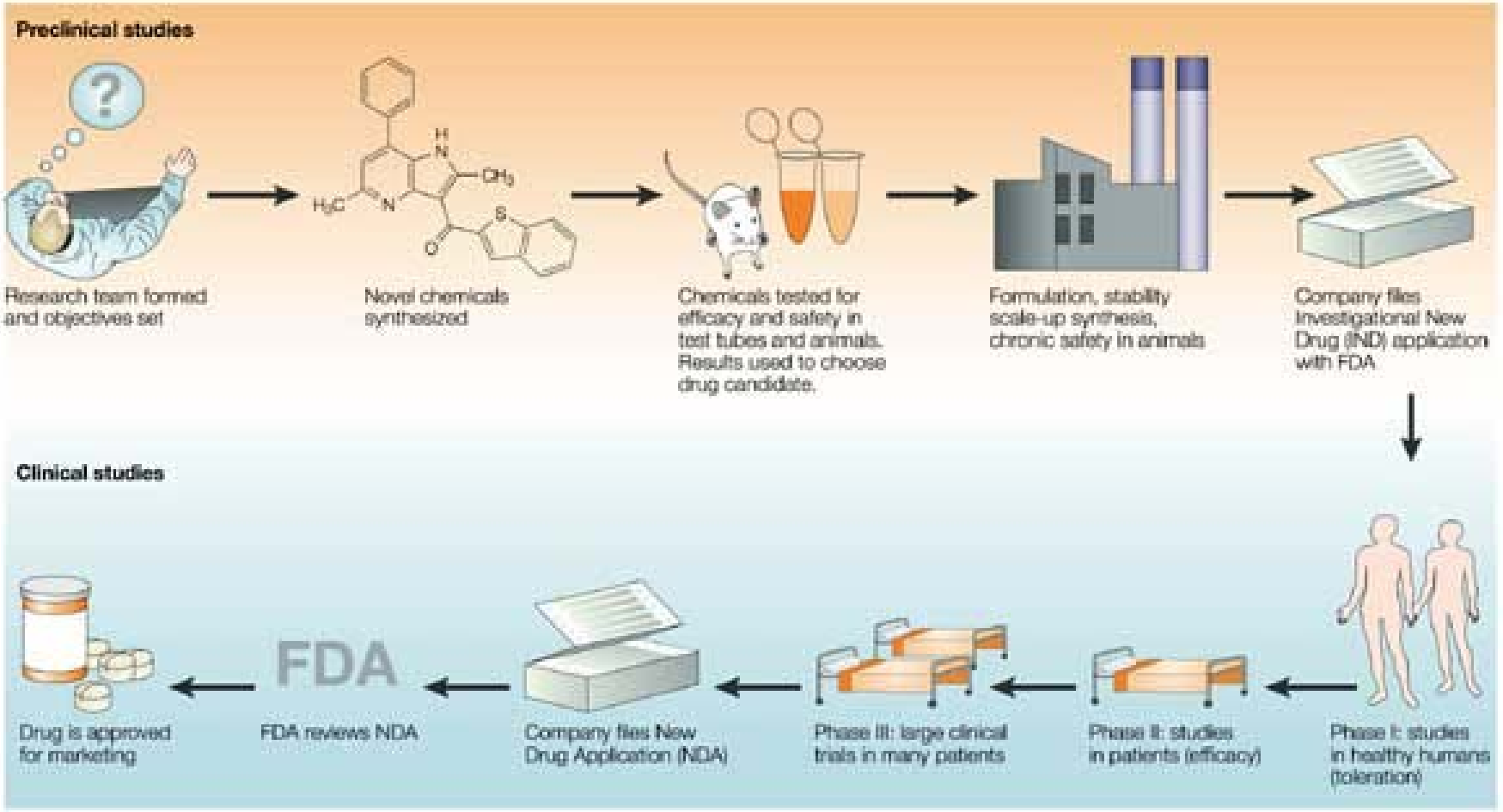
Cancer Results from a Combination of Genomic Changes

Component	Acquired Capability	Example of Mechanism
	Self-sufficiency in growth signals	Activate H-Ras oncogene
	Insensitivity to anti-growth signals	Lose retinoblastoma suppressor
	Evading apoptosis	Produce IGF survival factors
	Limitless replicative potential	Turn on telomerase
	Sustained angiogenesis	Produce VEGF inducer
	Tissue invasion & metastasis	Inactivate E-cadherin



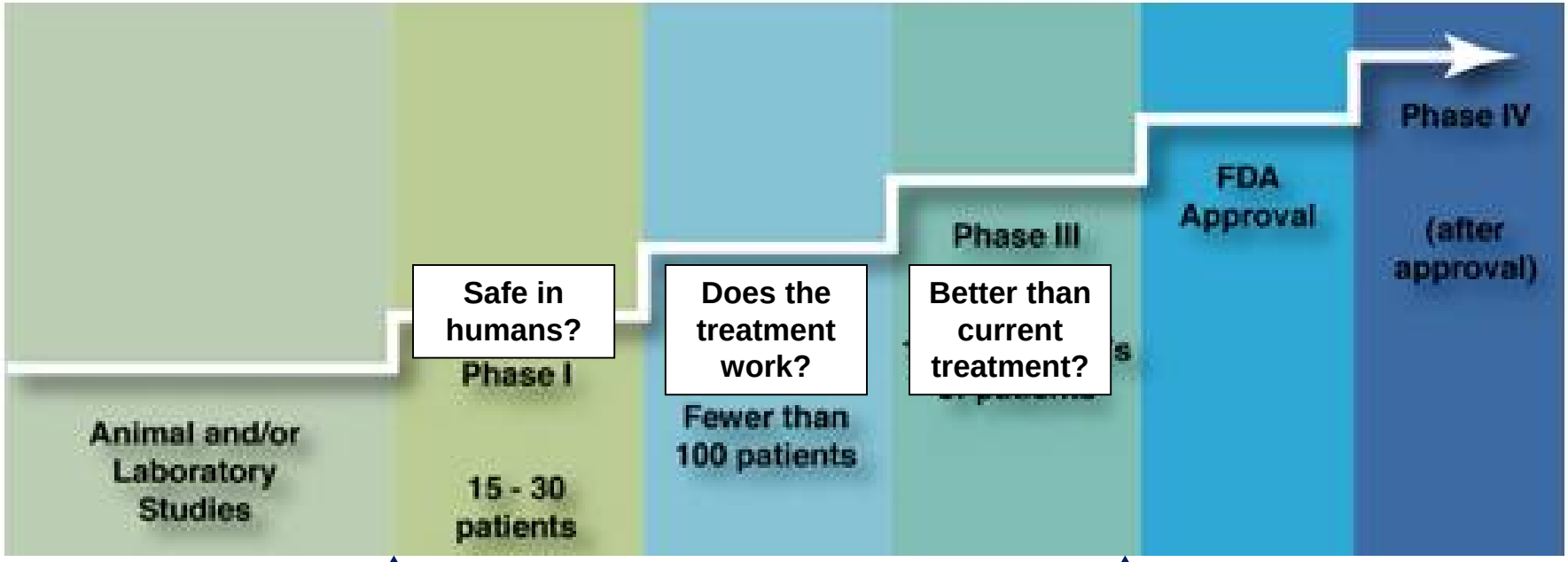
There is so much that we can target in cancer therapy. Cancers can also adapt to drug therapies, and we can predict the series of changes that may occur.

Drug Discovery



To enhance drug discovery with pharmacogenomic information, we need to collect patient samples and can screen libraries of compounds against protein variants caused by SNPs.
Next, hits must be evaluated for chemical properties and potency and those worth pursing are synthesized, evaluated, and modified for drug qualities (lead optimization). The drug is now tested in a pre-clinical setting to determine toxicology, drug kinetics, and drug metabolism.

The Clinical Trial Process



File IND

File NDA

More info on the approved treatment

Normally, it takes 12 years to get to drug on market. Is there a way to reduce this timeframe?

Phase I trials determine the safety of a new treatment.

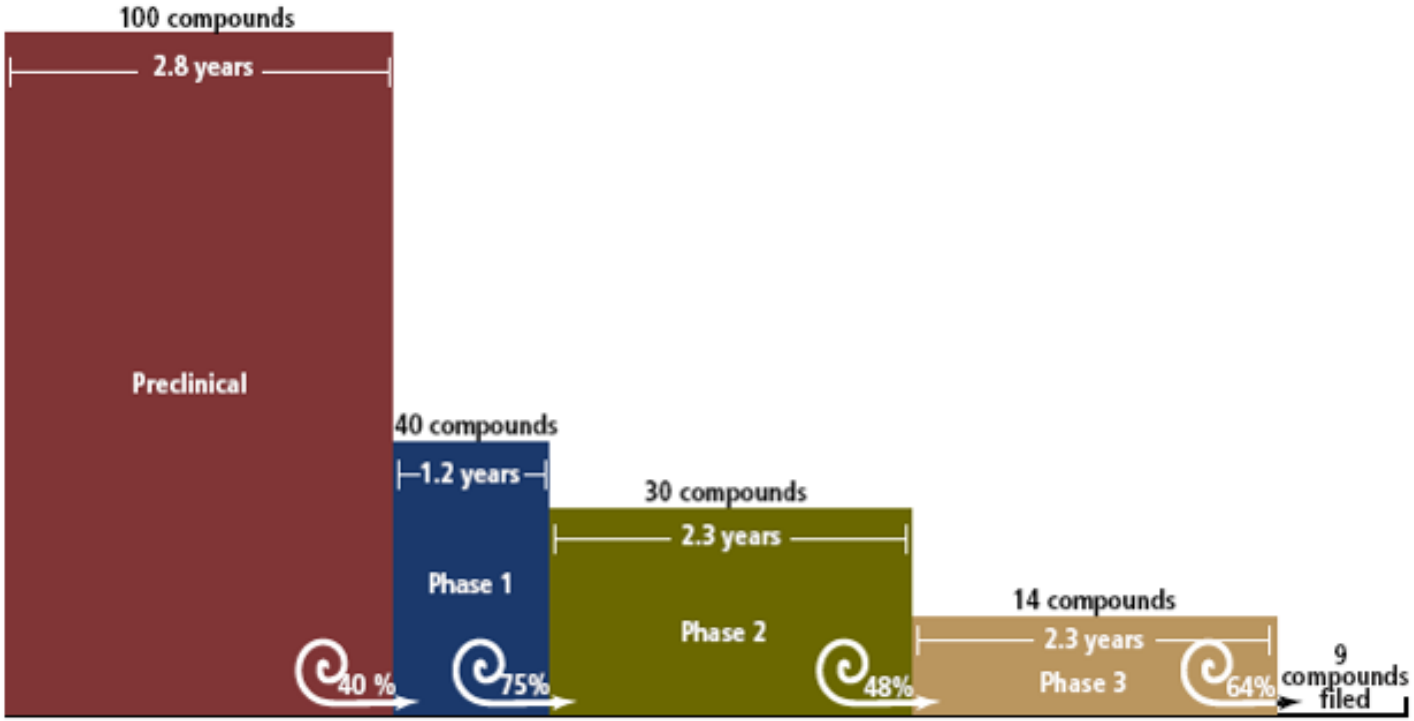
Phase II trials determine whether a certain disease responds to a new treatment.

Phase III trials study whether a new treatment is better than standard treatment.

Phase IV trials find more information about a new treatment that has been already approved for use in patients.

Where does pharmacogenomics currently play a role? In what phase is the pharmacogenomics of a drug usually examined?

Less than 1 in 10 Drugs in Clinical Trials Obtain a NDA



Source: Celera Genomics, 2004

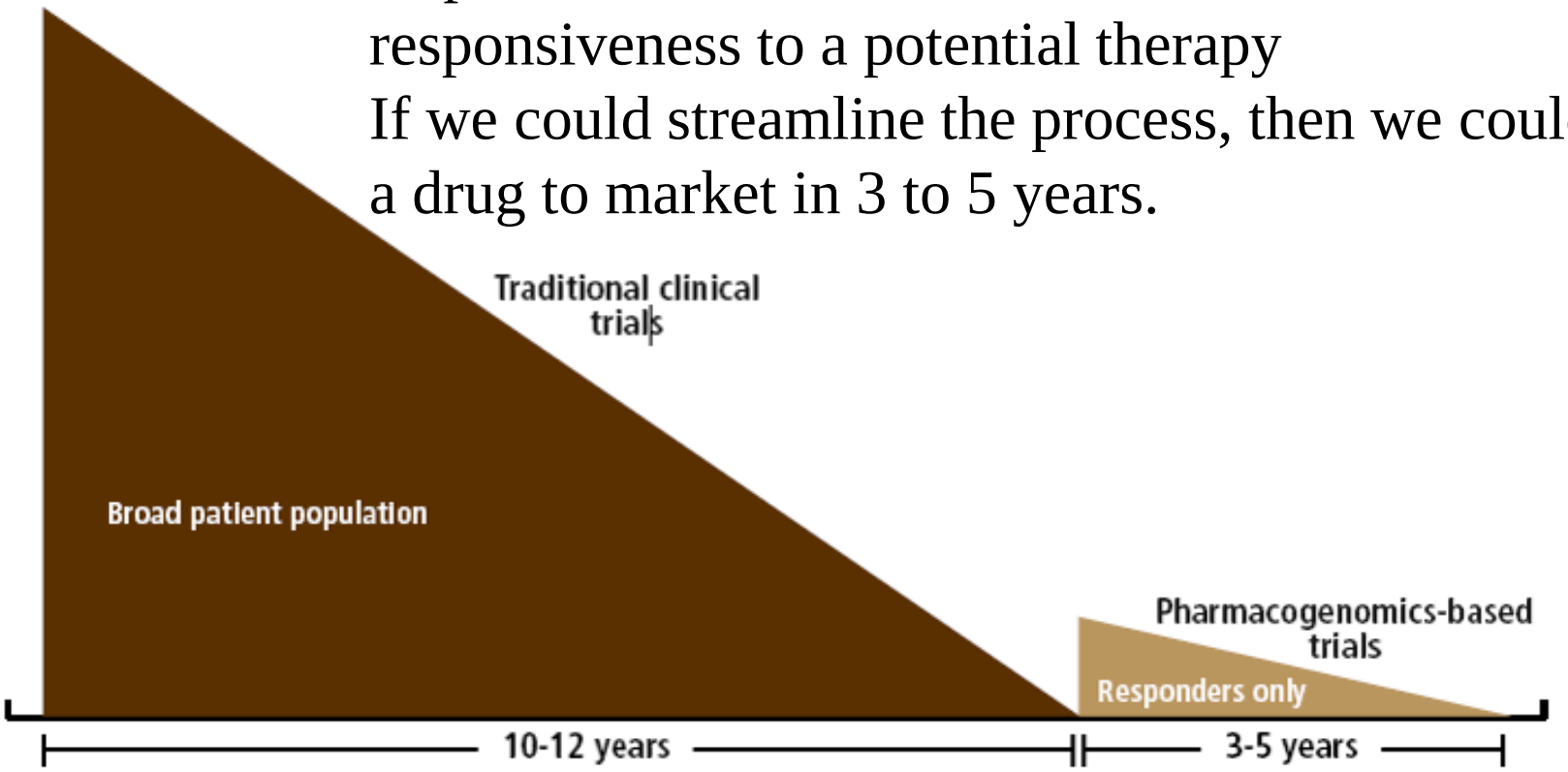
In a study of 14 clinical trials, 20-75% of subjects got no benefit

Less than 1 in 10 drugs make it to the NDA or New Drug Application during drug development from the preclinical phase. The average duration of each phase in the drug development process is shown in this bar graph. Also indicated is the likelihood, in percentages, of a compound making it to the next stage. Ultimately, only 9% if new drug candidates reach an NDA filing due in large part to safety and efficacy problems identified during clinical trials, and only 20 to 75% of subjects benefit in clinical trials, possibly because we are unaware of genetic factors that may elucidate how they may respond to drugs. For instance, drug induced liver injury (DILI) is a large issue. If we could genetically test for patients who are more susceptible to liver injury, we could bring more drugs to the market.

Streamlining of Clinical Trials is the Future

Pharmacogenomics-based clinical trials have the potential to be significantly less lengthy and costly than conventional trials because participants would be pre-screened for biomarkers that indicate their responsiveness to a potential therapy

If we could streamline the process, then we could get a drug to market in 3 to 5 years.



Source: Quintiles Transnational, 2004

Pharmacogenetics and Pharmacogenomics Summary

1. SNPs in coding and non-coding regions can alter the function or expression of an enzyme that metabolizes drugs
2. Pharmacogenetics impacts the kinetics and dynamics of drugs.
3. Personalized medicine will help treat patients according to their genetic profile, increase drug efficacy, and reduce side effects
4. Designing clinical trials based on SNP profiles can reduce FDA approval time for drugs

Concept Map

Pharmacogenomics

Pharmacodynamics

Pharmacokinetics

Gene

SNPs

Haplotype

Phenotype

Genotype

CYPs

Drug