

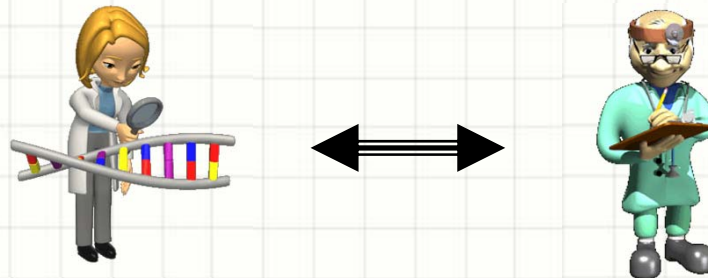


GENETIC TESTING: IN WHOM AND WHEN

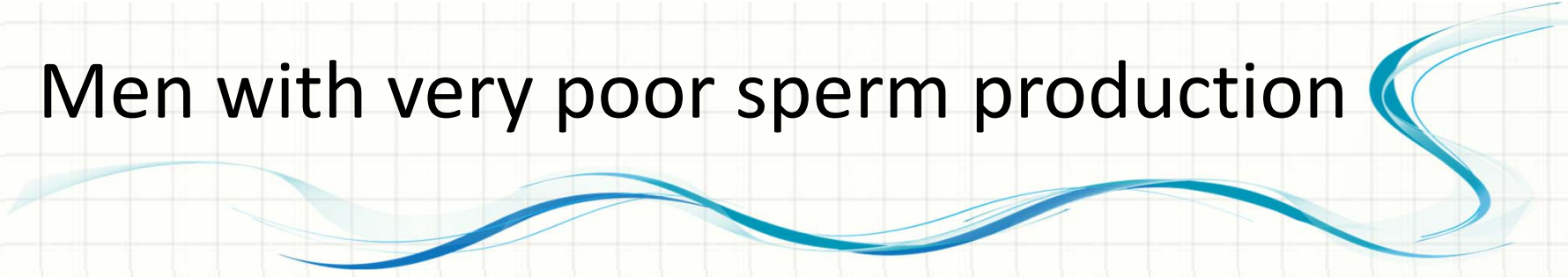
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My background in this field

- I was the first to link Cystic Fibrosis Mutations with Congenital Absence of the Vas Deferens
 - One of the world's leading experts in this area
 - See below
- I was one of the first clinical researchers in the field of the genetic basis of extremely poor sperm production
 - One of the world's leading experts in this area
 - See below



Men with very poor sperm production



- Some definitions

- Oligospermia

- Low sperm count

- Severe Oligospermia

- Extremely low sperm count – usually less than 3 million

- Azoospermia

- No sperm in the ejaculate (no sperm on the semen analysis)

- Non-obstructive azoospermia (NOA)

- No sperm due to a problem with sperm production


- Not a blockage of any type or anywhere in the system

- Obstructive azoospermia

- No sperm because there is a blockage somewhere in the system

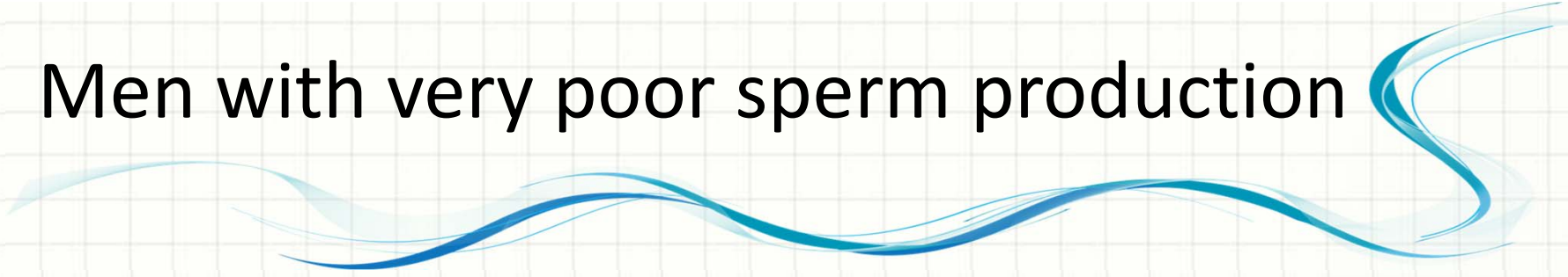
- Sperm production in the testicles is normal, it just can't get out

Who needs a genetic evaluation



- Men with sperm counts below 3 million
 - Not due to a blockage of some type
 - Not due to a pituitary or hypothalamic problem
 - Not due to anabolic steroid / testosterone use
 - Due to poor sperm production in the testicles ! !
- Men with Cystic Fibrosis or CBAVD
 - CBAVD = Congenital Bilateral Absence of the Vas Deferens
 - In these two conditions, the tubing that brings the sperm into the body is not present

Men with very poor sperm production



- We need to find out why the testes do not work well
 - The information may change what you do in terms of:
 - Is there a hope that I actually make sperm ?
 - Should I have an operation to try and find sperm ?
 - Is there a risk of using my sperm ?
 - Is there a problem with my Y chromosome that any son will inherit ?
 - Is there a problem with my chromosomes that we need to know about ?
 - Do I have Klinefelter Syndrome ?
 - Do I have a different and unusual syndrome that affects sperm also ?
 - It all starts with a good history and physical examination
 - The semen analysis then confirms any need for testing

What tests do we do for poor sperm production?

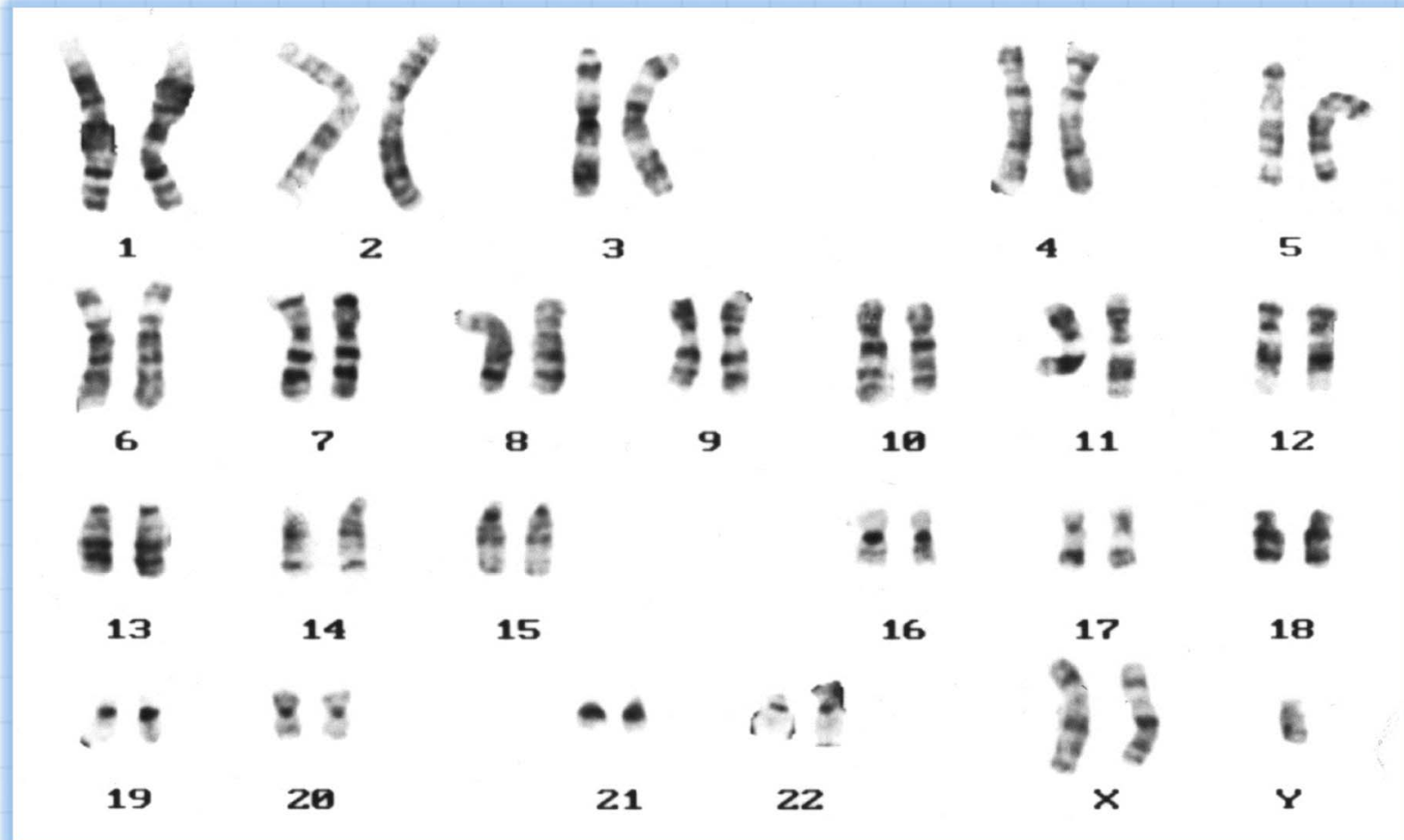
- 1. Karyotype

- Blood test
- Shows us the number and structures of the chromosomes
 - There are normally 46 chromosomes
 - The sex chromosomes in the male are X and Y
 - In the female the sex chromosomes are X and X
- There may be abnormalities in number, for example:
 - Klinefelter Syndrome – 47 chromosomes with an extra X
- There may be abnormalities in structure, for example:
 - Translocations: a piece of one chromosome is flipped with another
- Necessary prior to attempting to find testis sperm in NOA !

Example of a Klinefelter karyotype:

*22 pairs and the sex chromosomes

* note the extra X chromosome (so 47 total and 2 X's)



What tests do we do for poor sperm production?

- 2. Y Chromosomal Microdeletion Assay (YCMD)
 - Blood test
 - Looks at the Y chromosome to see if there are missing areas
 - There are genes on the Y chromosome important for sperm production
 - If the area where one of the genes lives is missing
 - The testicle may make a very small number of sperm
 - The testicle may not be able to make any sperm at all
 - **Necessary prior to attempting to find testis sperm in NOA !**
 - If a specific area is missing and there is no sperm production
 - An operation (TESE) to look for sperm will NOT be helpful
 - If a specific area is missing and there might be a little bit of sperm
 - Each male offspring will be infertile or sterile

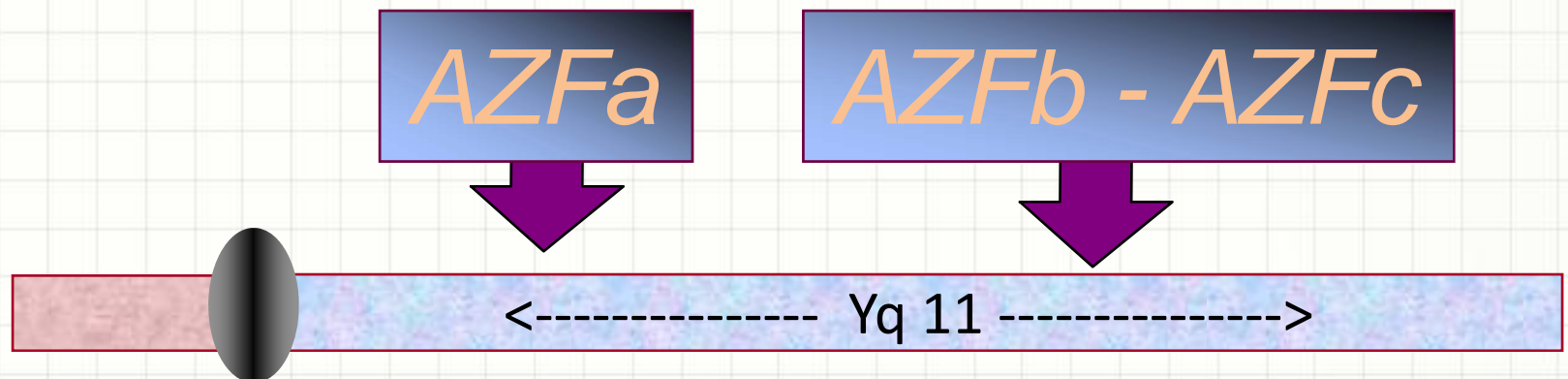
The three regions on the Y chromosome that we look at:

*AZFa, AZFb, and AZFc

If you have non-obstructive azoospermia (NOA) and are:

- Missing AZFa – no sperm will be found in the testicles
- Missing AZFb – no sperm will be found in the testicles
- Missing AZFb and AZFc – no sperm will be found in the testicles
- Missing AZFc – sperm might be found in the semen or in the testicles

Azoospermia Factor - AZF

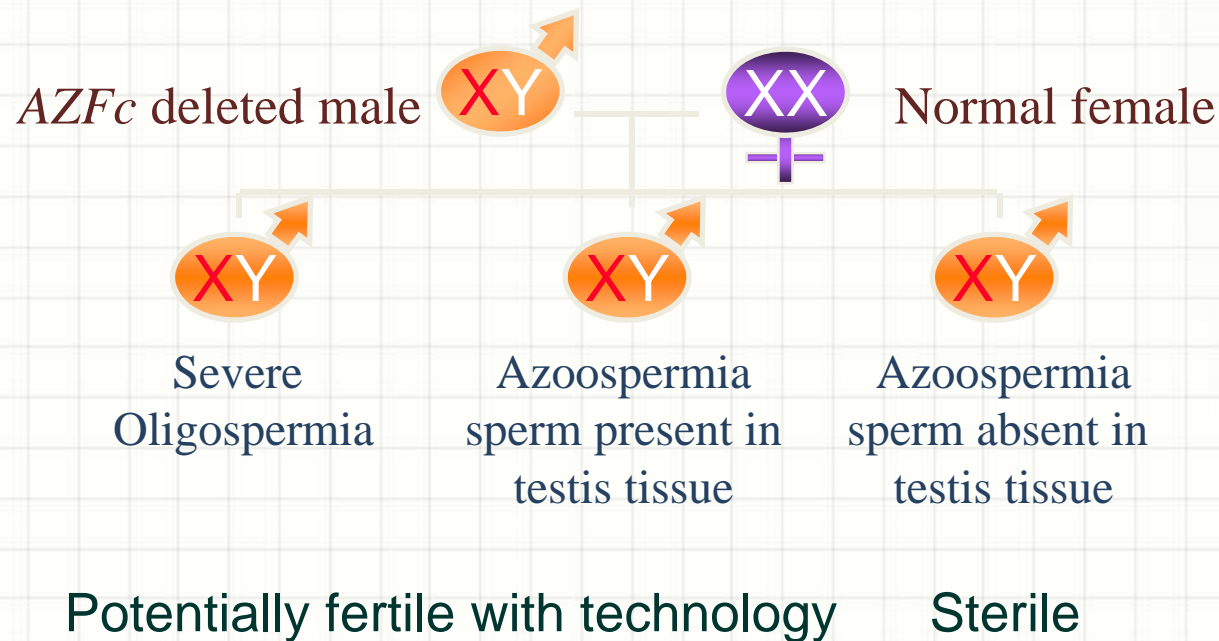


If you are missing the AZFc region:

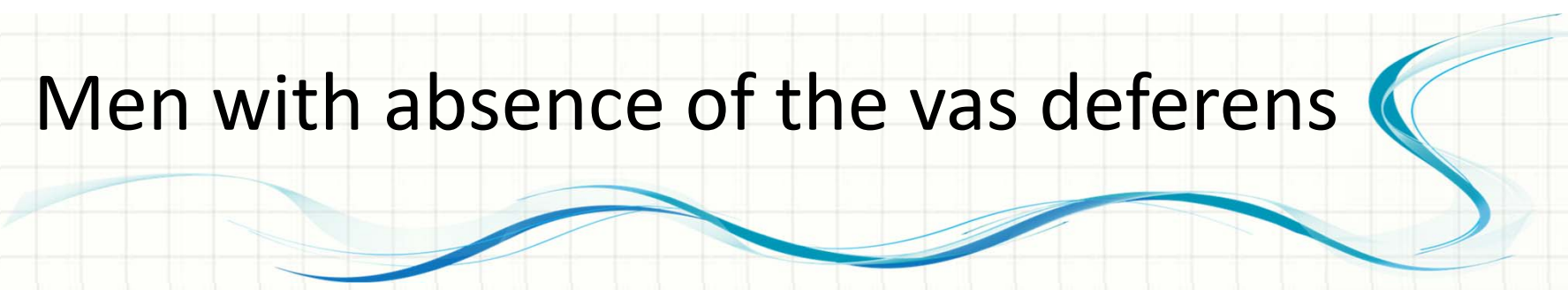
Any male offspring will be predicted to:

- a. have very low sperm production, if any
- b. have no sperm in the ejaculate
- c. possibly no sperm even in the testis tissue

Vertical Transmission of AZFc Deletion



Men with absence of the vas deferens

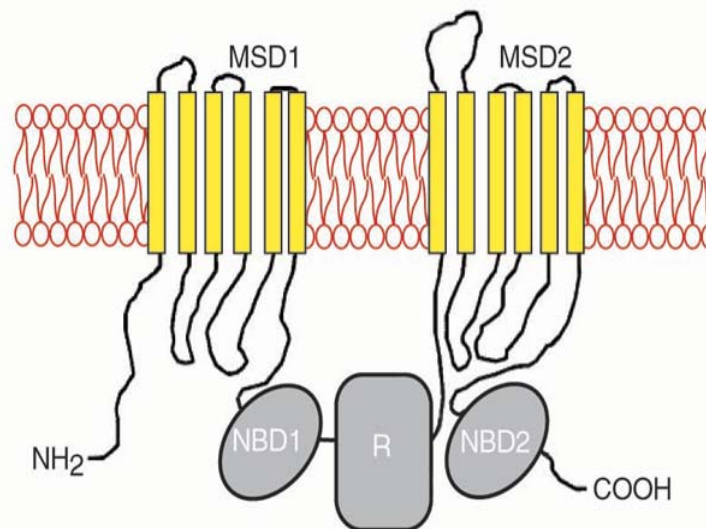


- The vas deferens transports the sperm up from the testicles
 - In men with Cystic Fibrosis, the vas is missing on both sides
 - In men with CBAVD, the vas is missing on both sides
 - I make the diagnosis of CBAVD in a minute on physical examination
 - There is no need for scrotal ultrasound
 - There is no need to “explore” the scrotum with an operation
 - The ejaculate volume is typically just a drop or two
 - There is no sperm in the semen analysis (no way for it to get there)
 - In both of these conditions, the testicles make sperm normally

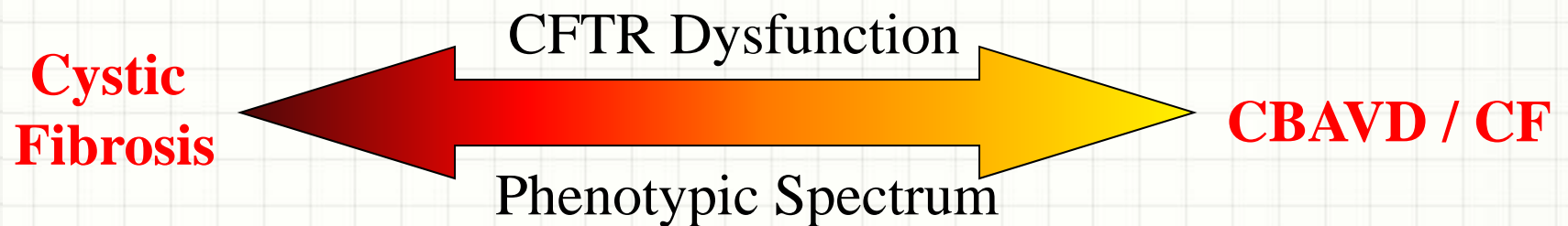
Both of these conditions are due to mutations in the “Cystic Fibrosis” genes

The Cystic Fibrosis gene is located on chromosome 7

1. Cystic fibrosis transmembrane conductance regulator (CFTR)
 - maintains proper fluidity of secretions respiratory and pancreatic systems
2. If dysfunctional, secretions are thick and tenacious
 - leads to pulmonary disease
 - leads to exocrine pancreatic failure
 - leads to absence of the vas



CFTR Dysfunction : A spectrum of disease



Depending on the CF mutation inherited from Mom and the CF mutation inherited from Dad, the presentation will be different – all the way from terrible Cystic Fibrosis on one end to CBAVD on the other end where the lungs and pancreas are just fine but the vas is absent

Since most CBAVD is caused by mutations in the “Cystic Fibrosis” Genes

- Prior to MESA, both partners need to have CF Mutation analysis
This should not be an option, but a necessity
- If the female partner is a carrier of a CF mutation, then any child conceived could have Cystic Fibrosis – unless we know about it beforehand and we can prevent this from happening
- The next picture is a Punnett Square showing the possibilities if the female partner is a carrier

Transmission of CFTR mutations if partner is a carrier

| | | <u>Male Genotype</u> | |
|------------------------|---------------|---|------------------------------------|
| | | $\Delta F508$ | 5T allele |
| <u>Female Genotype</u> | $\Delta F508$ | $\Delta F508 / \Delta F508$ <u>Cystic Fibrosis</u> | $\Delta F508 / 5T$ <u>CBAVD</u> |
| | + | $\Delta F508 / +$ <u>Carrier</u> | 5T / + <u>Carrier</u> |

This shows why it is so important to make the proper diagnosis and, if need be, perform CF mutation analysis on both partners – not just the male

In summary:

1

- The genetic basis of male infertility should always be looked for

2

- Before any intervention such as in- vitro fertilization or TESE

3

- It is a complex subject that you deserve to know about and understand

I am happy to see you in the office and determine what tests, if any, you really need and will benefit from