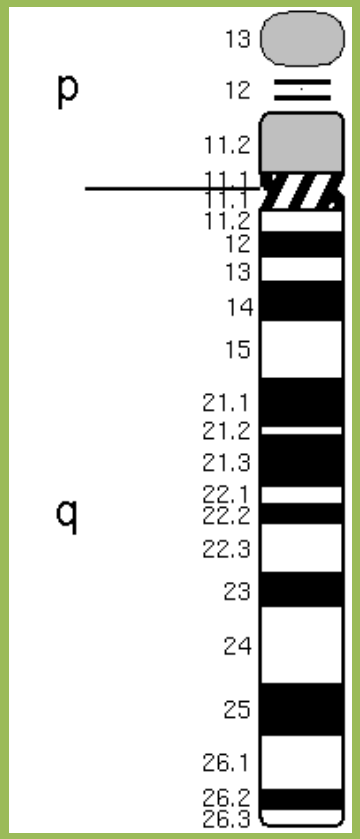
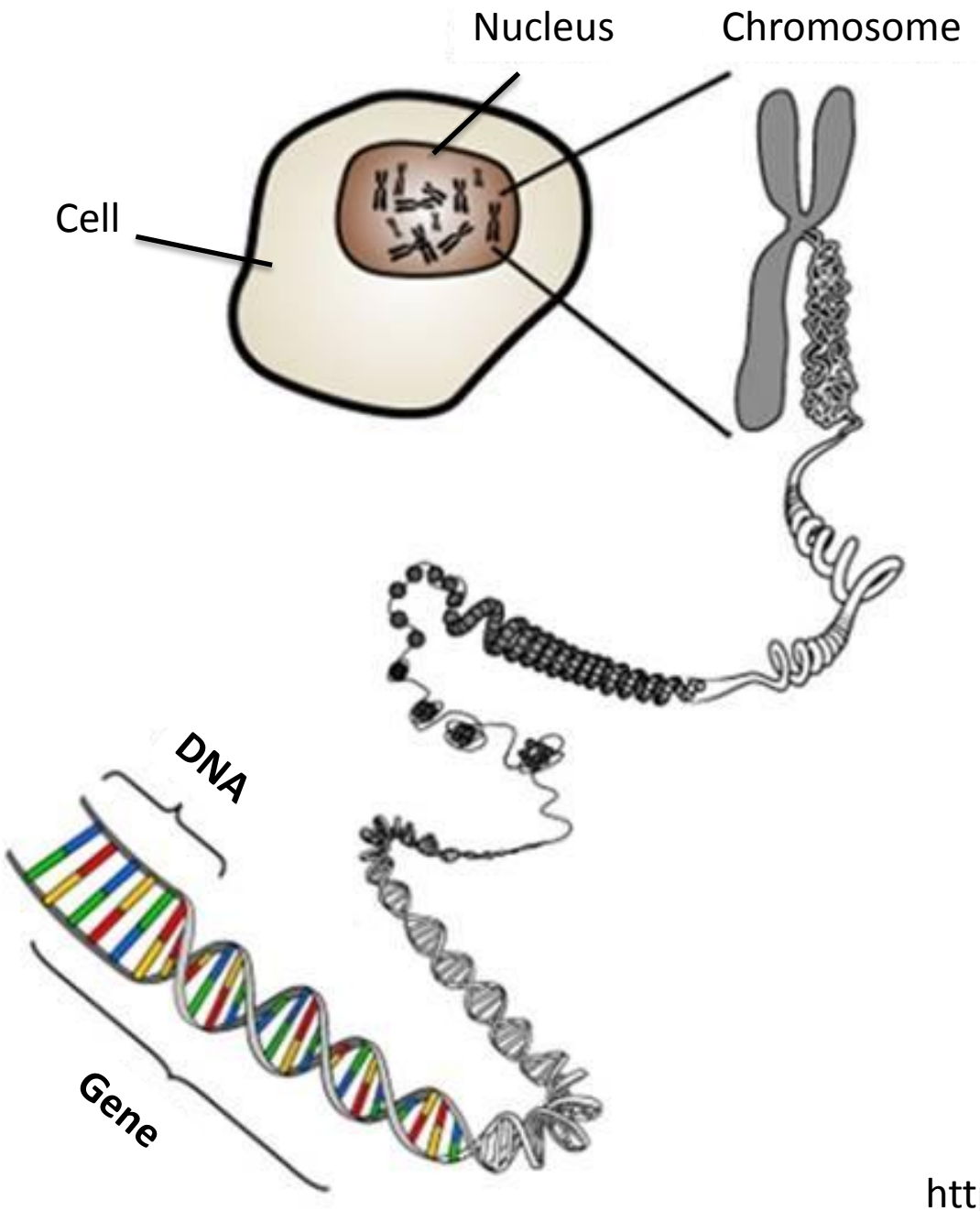
angelman SYNDROME FOUNDATION



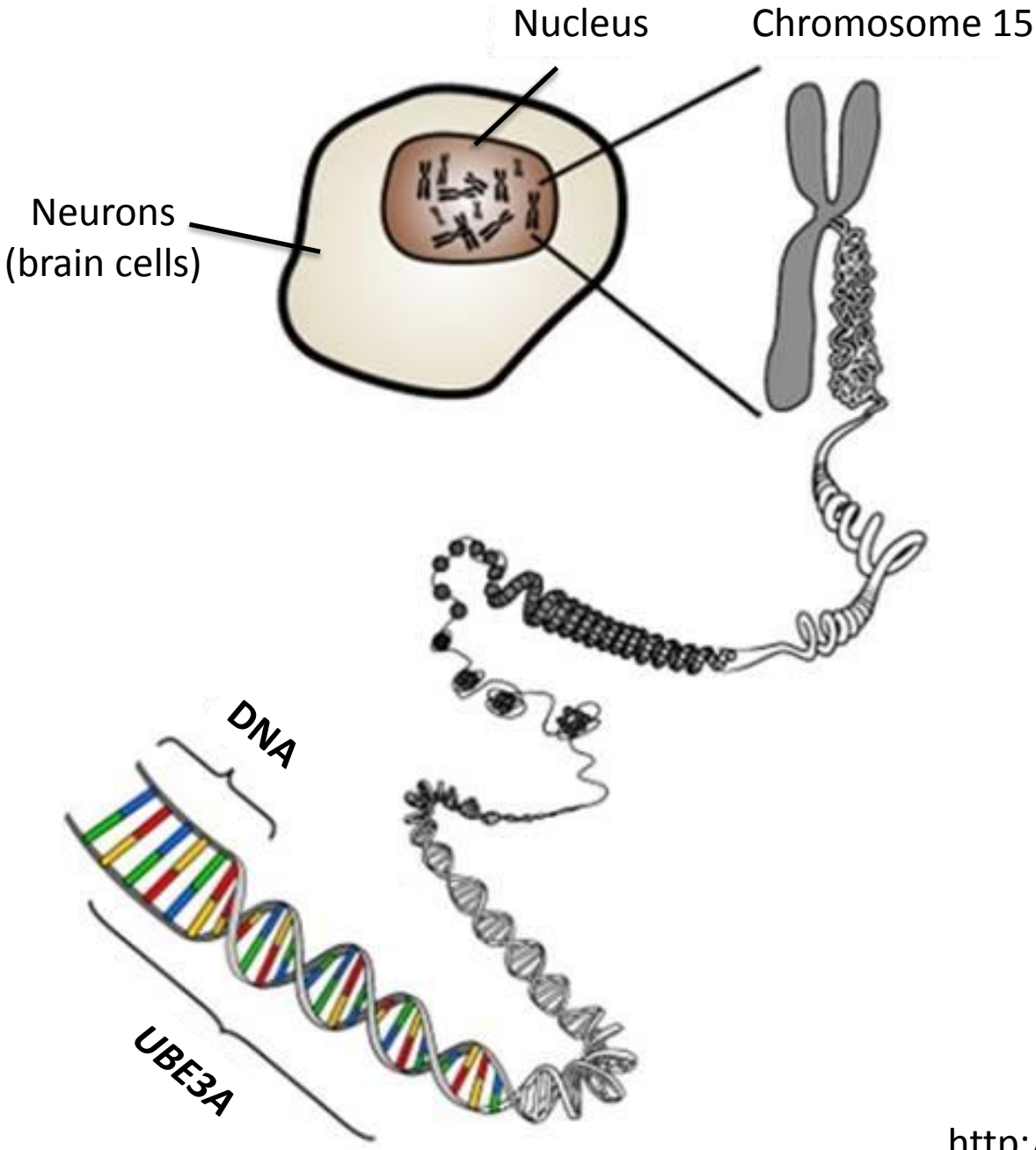
Genetics 101

Stormy J. Chamberlain
Department of Genetics and Genome Sciences,
University of Connecticut Health Center

Angelman Syndrome Foundation Family Conference
Phoenix, AZ
July 13th, 2017



http://www.gencodys.eu/

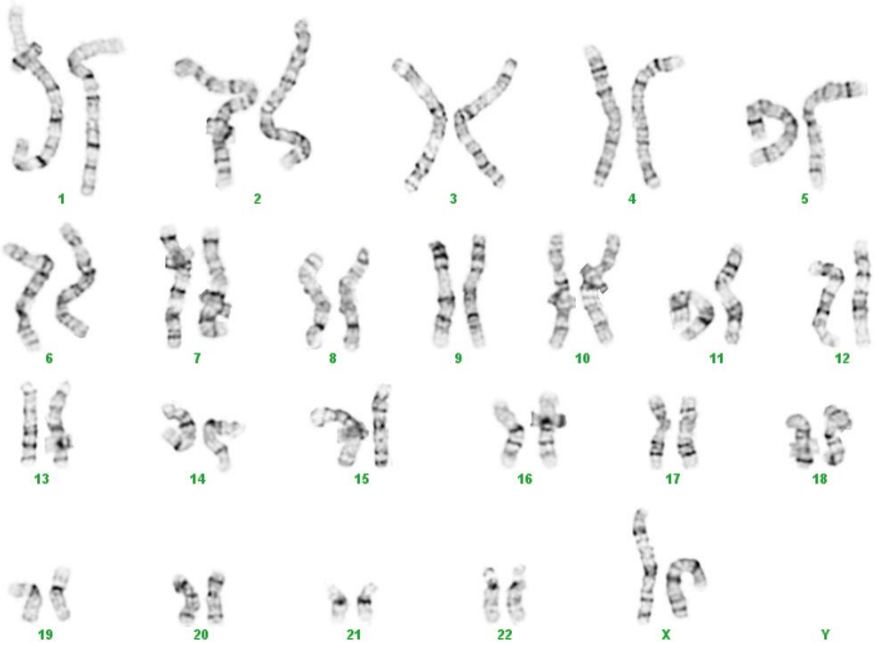


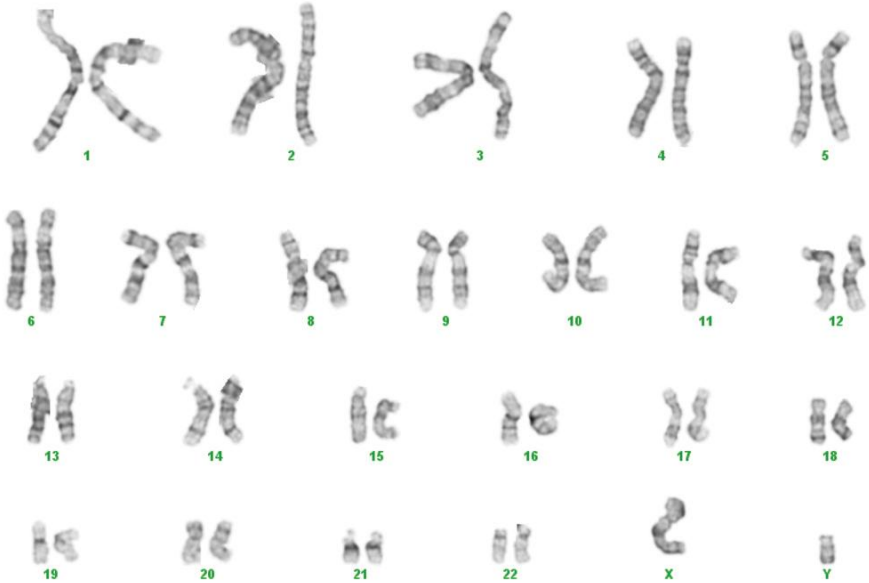
http://www.gencodys.eu/

Humans have 22 pairs of chromosomes plus 2 sex chromosomes

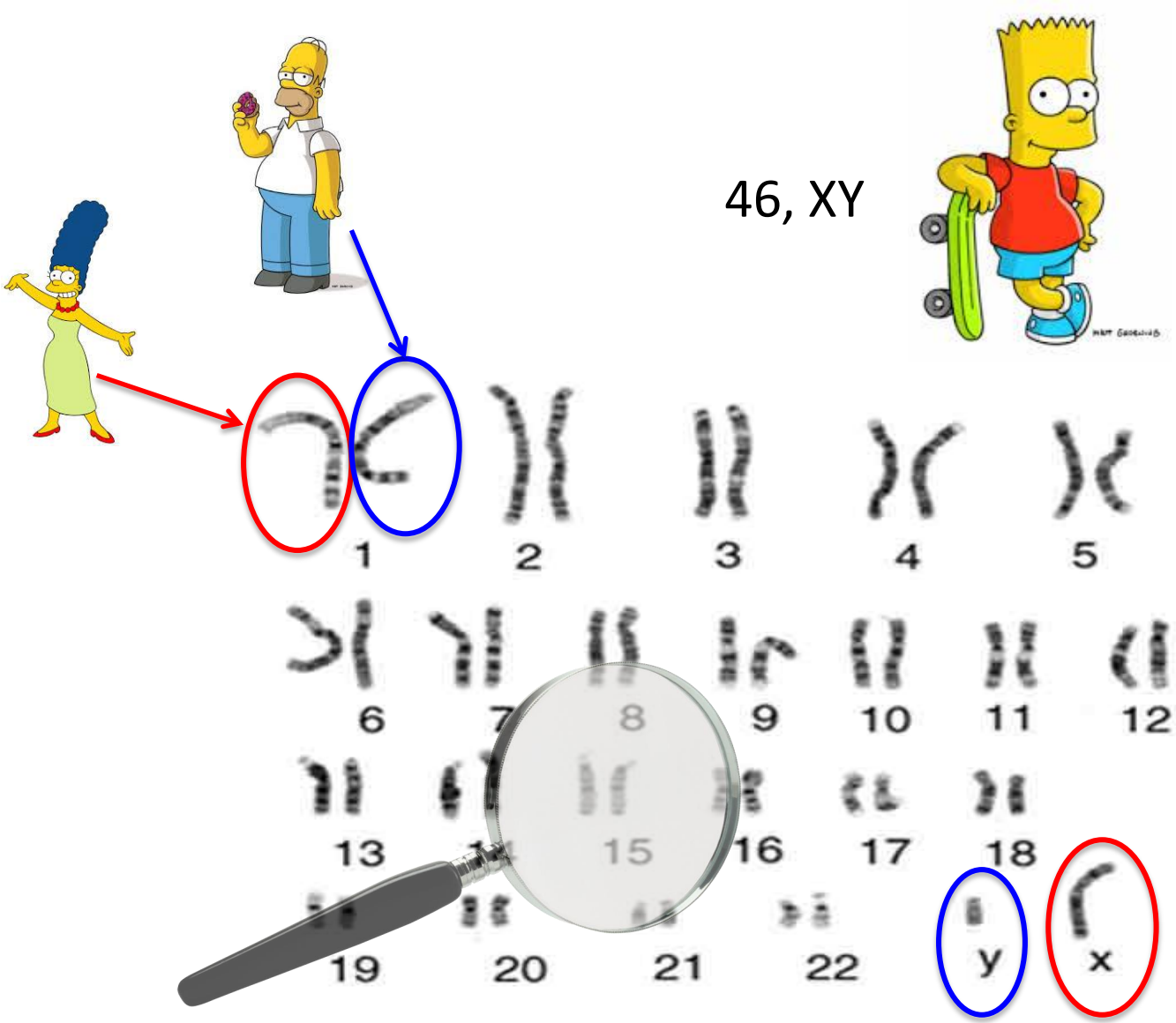
46, XX

46, XY

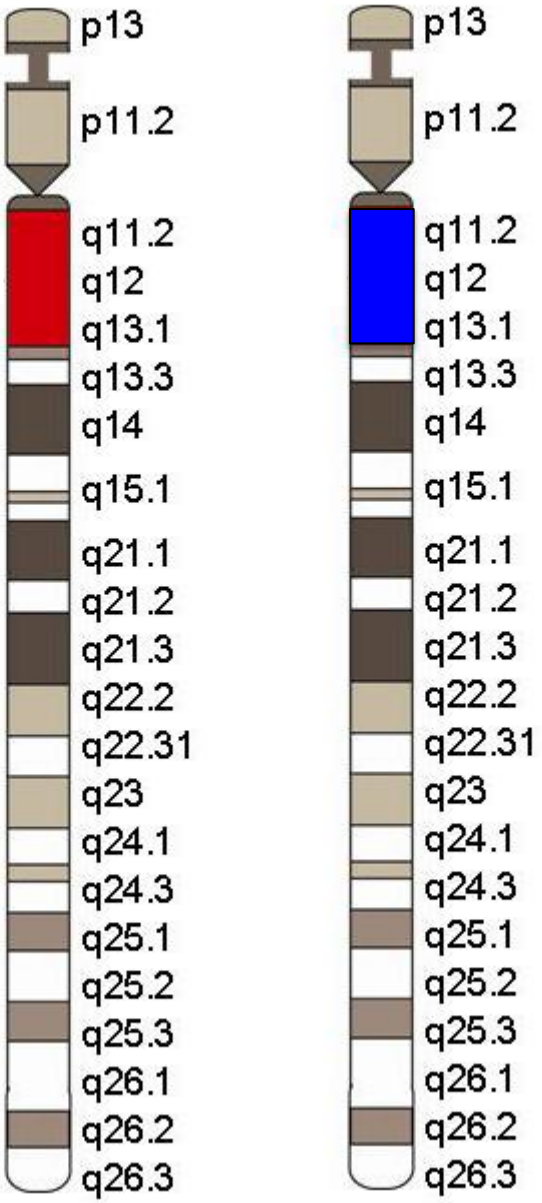




We get one copy of each chromosome from mom and one copy from dad



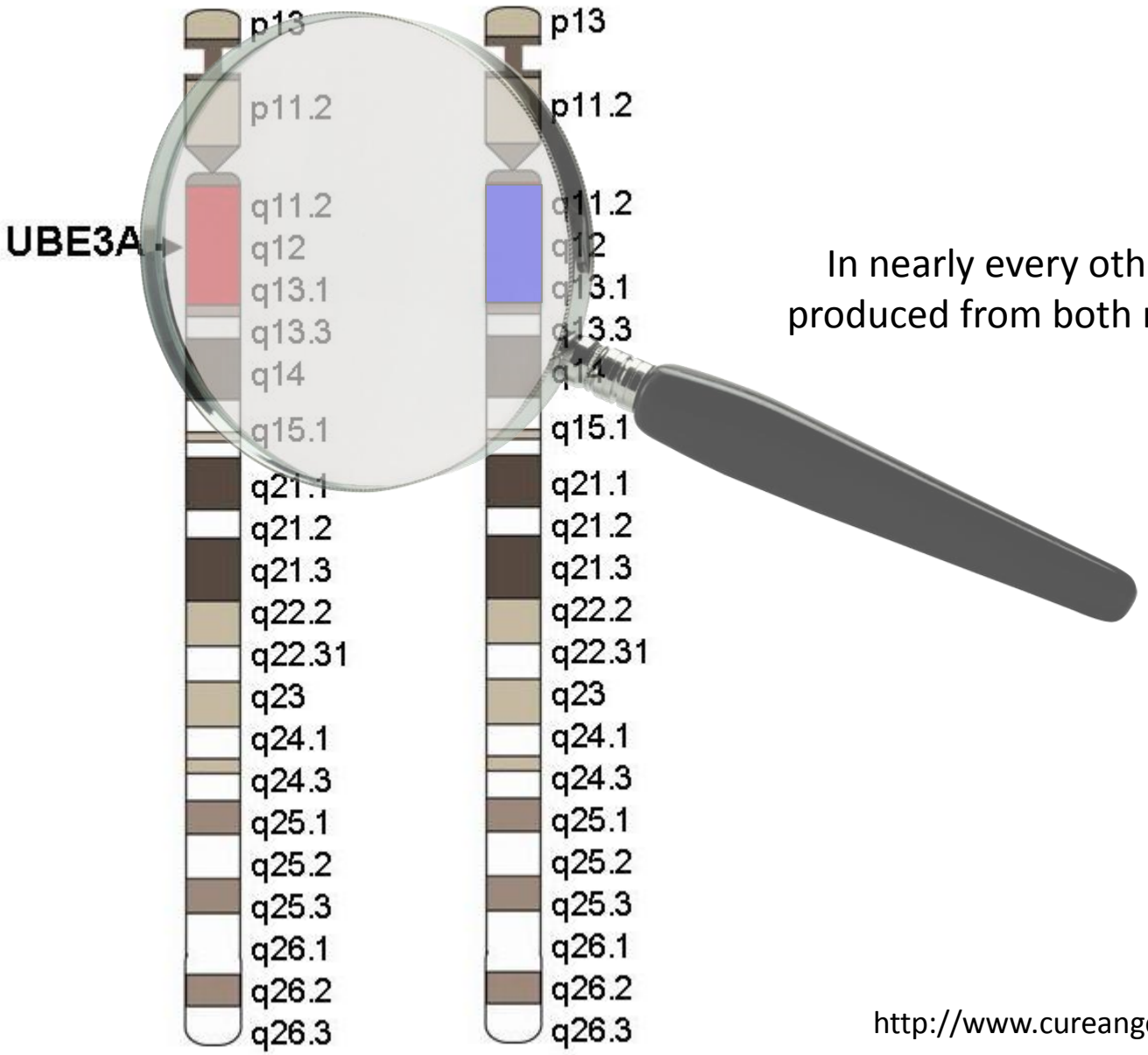
Unlike most chromosomes, mom's and dad's copies of chromosome 15 are different from one another



The chromosome 15q11-q13 region is where the *UBE3A* gene is located.

http://www.cureangelman.org/what-genetics101.html

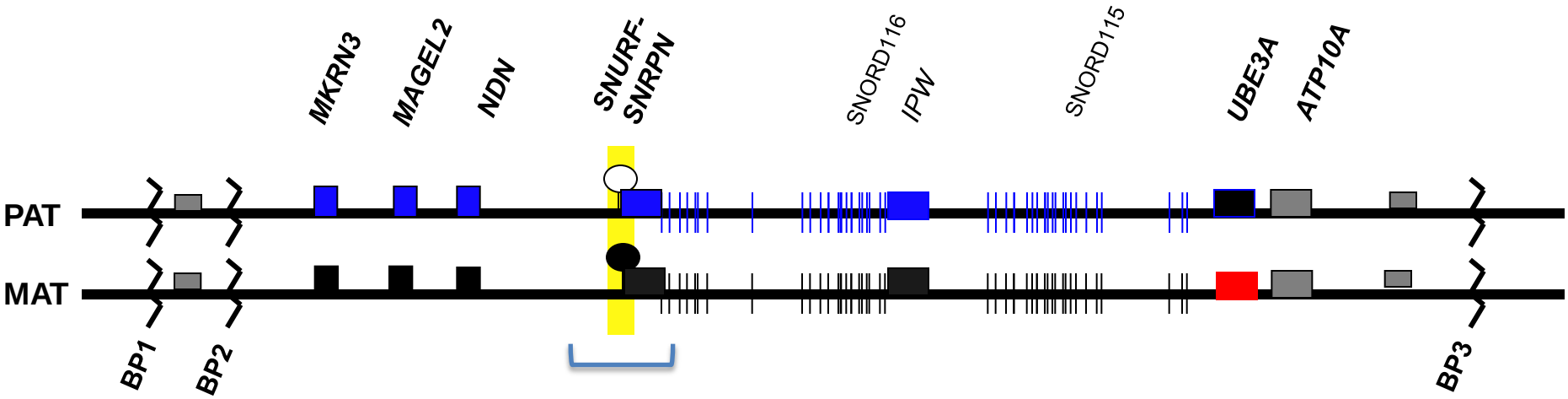
In neurons, UBE3A is produced from mom's copy, but not dad's



In nearly every other cell type, UBE3A is produced from both mom's and dad's copies

http://www.cureangelman.org/what-genetics101.html

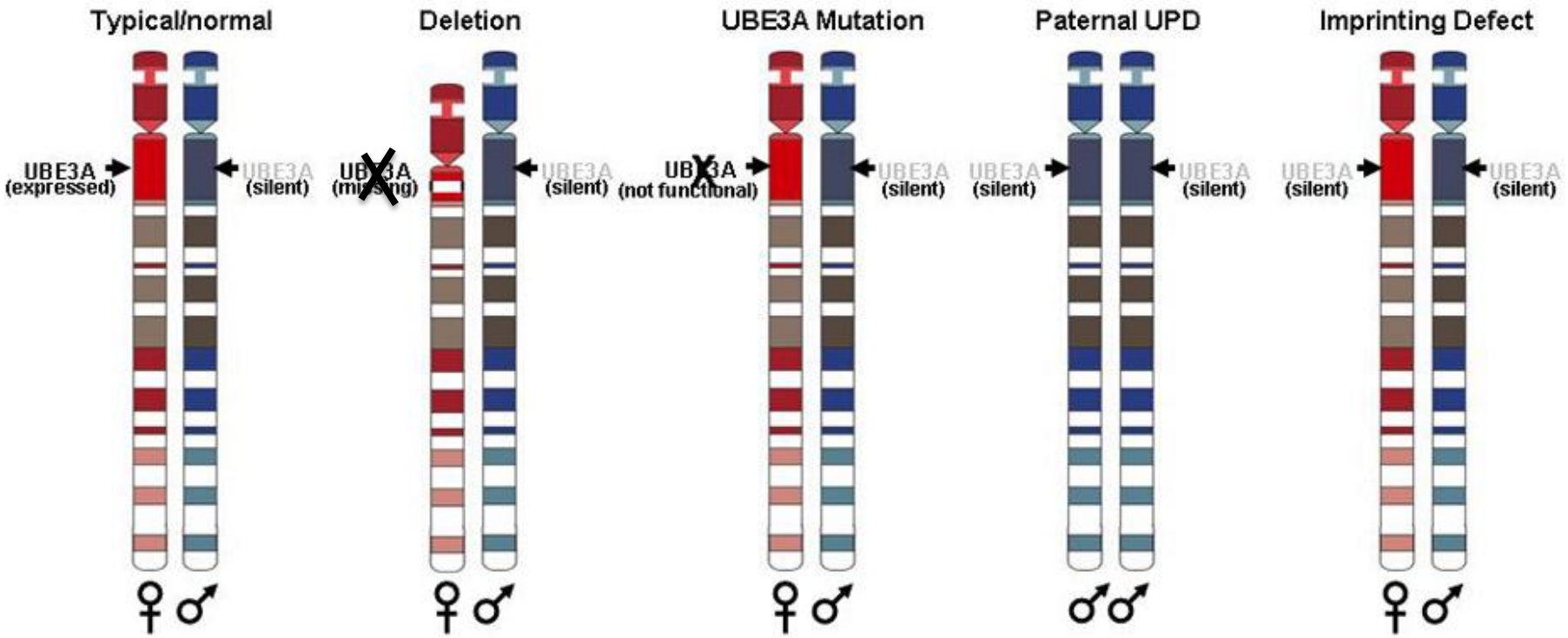
Genes in the chromosome 15q11-q13 region



Imprinting center –has a methylation imprint

* Methylation imprint ultimately determines the difference between mom's and dad's chromosomes 15.

Molecular classes of AS



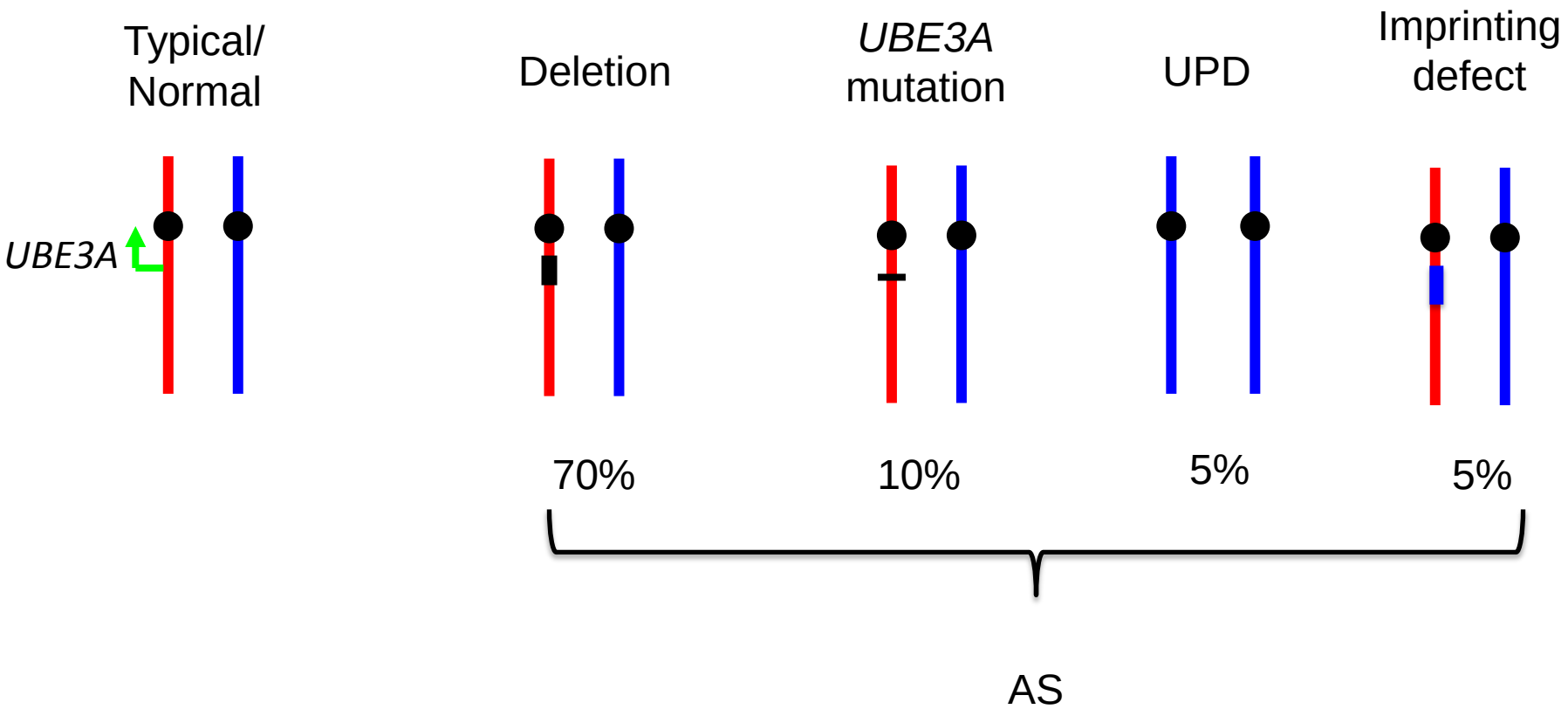
http://cureangelman.net/understanding-angelman/genetics101/

Different Molecular Classes of AS

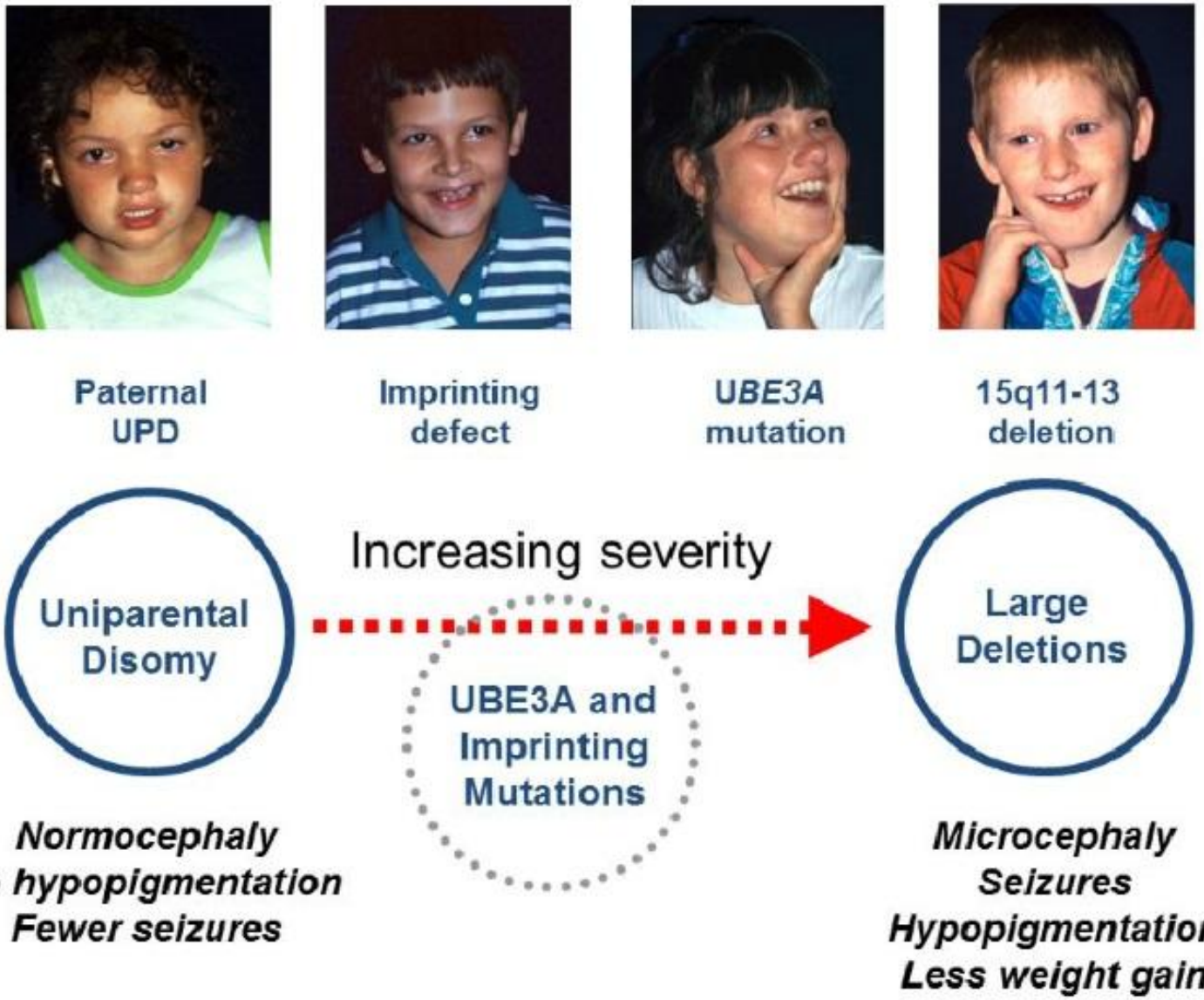
Chromosomal/Genetic Abnormality	% in AS
Deletion of maternal 15q11–q1	70%
Mutation in *UBE3A* gene	10
Paternal Uniparental Disomy (UPD)	5
Imprinting Defect (ID)	5
Unknown	10

Adapted from: Molecular epigenetics of Angelman syndrome (2007). M. Lalande and M. A. Calciano

Molecular Classes of AS

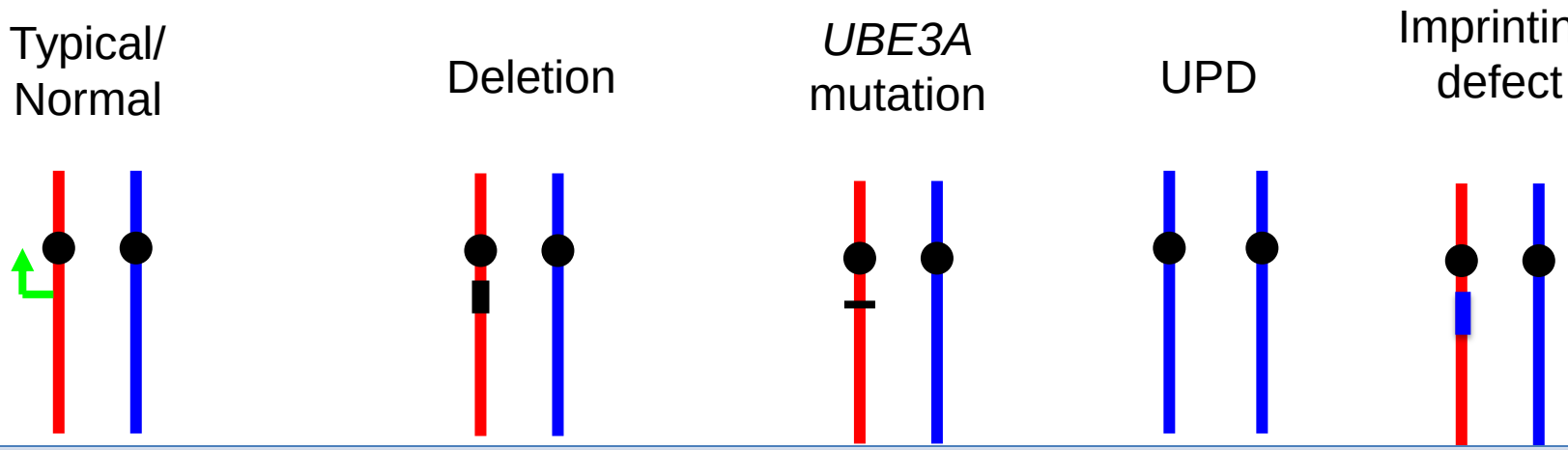


Why do we want to know what type of AS a child has?



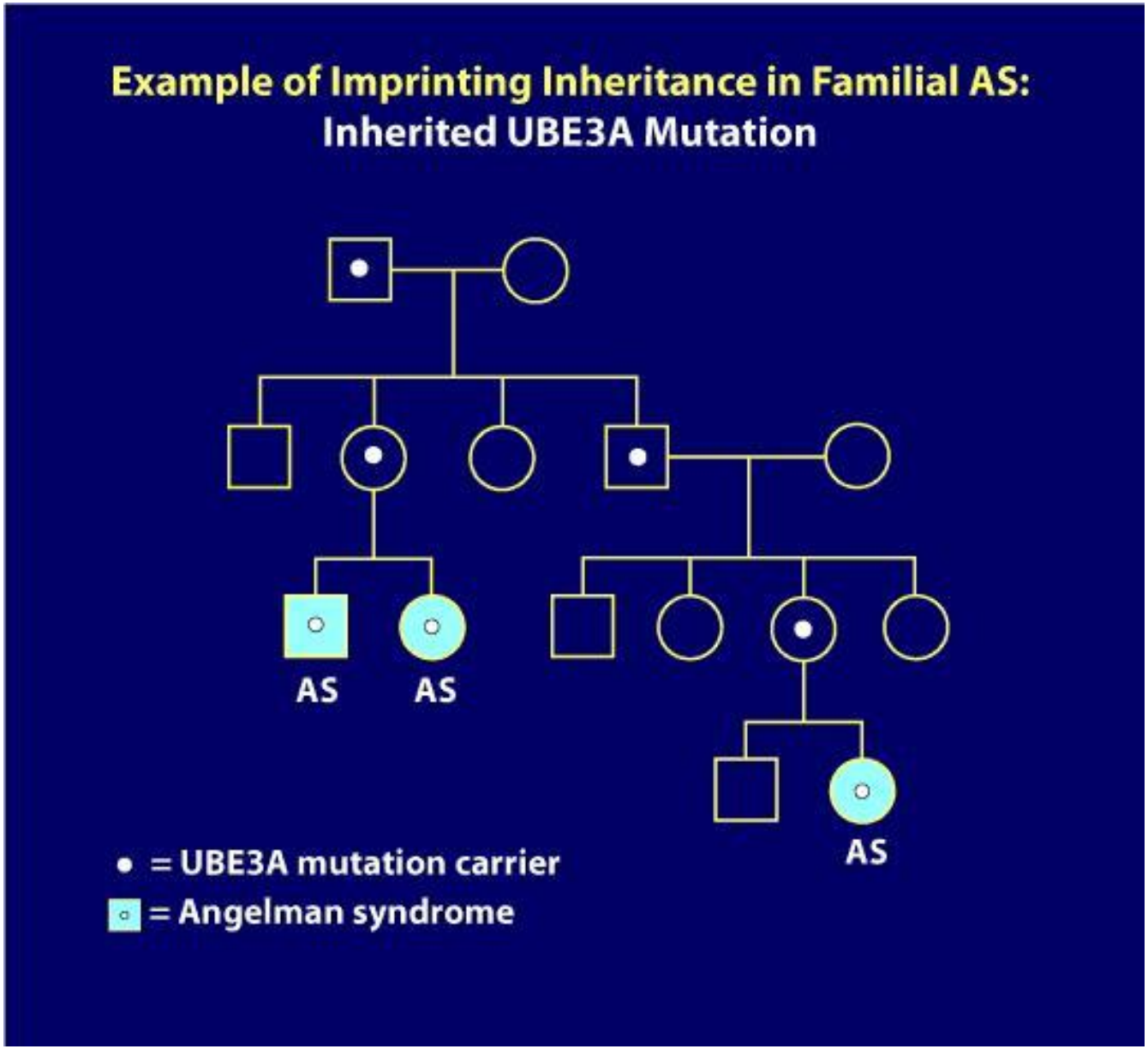
Credit: Dr. Charlie Williams

Risk of Recurrence—
What are the chances of having another child with AS?



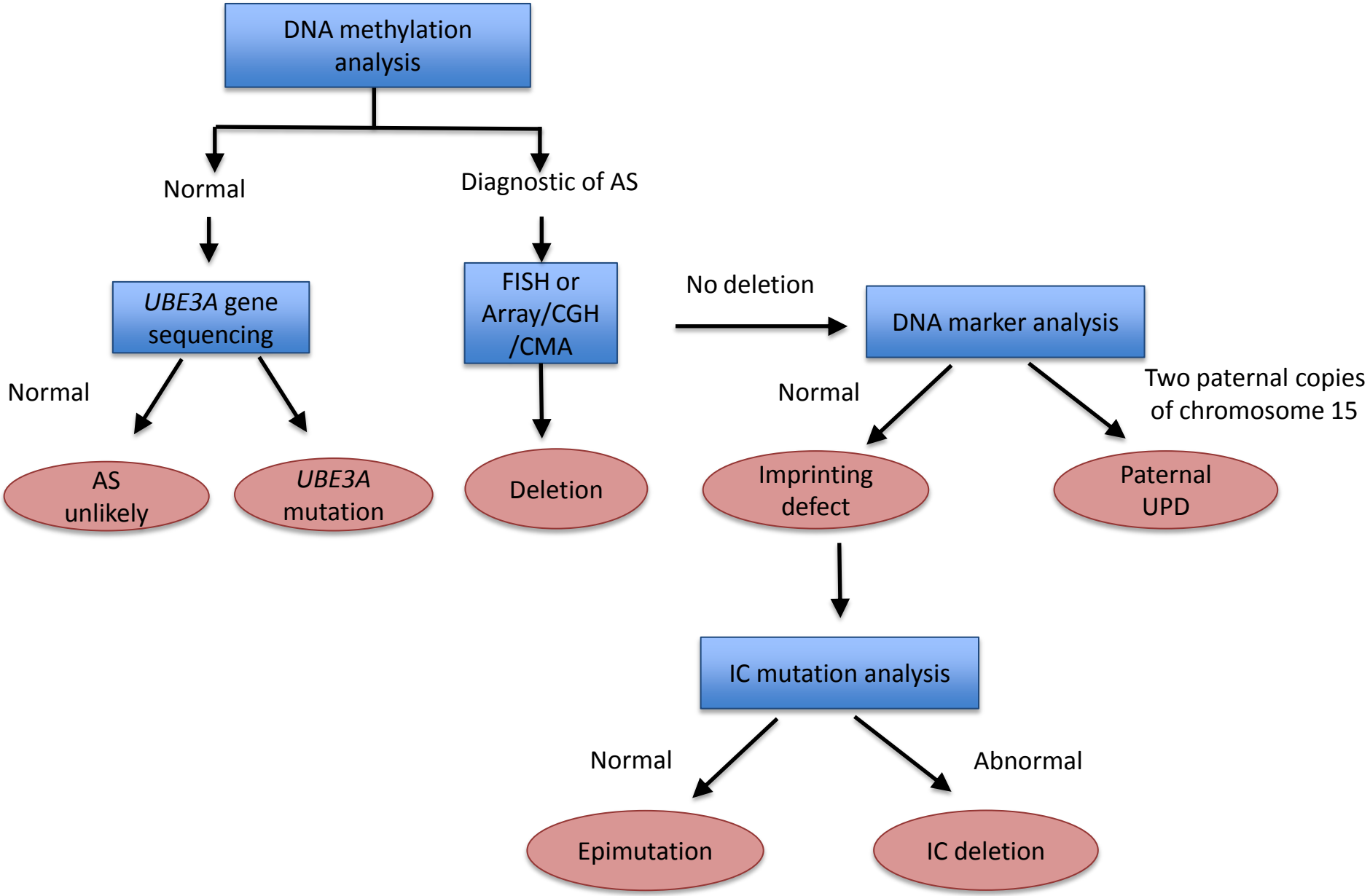
	Typical/ Normal	Deletion	*UBE3A* mutation	UPD	Imprinting defect
Prevalence in AS:		70%	10%	5%	5%
Risk of recurrence		<<1%*	50%* if inherited <1%* if spontaneous	<<1%*	<1% for most 50% if deletion

* exceptions—translocation, germline mosaicism

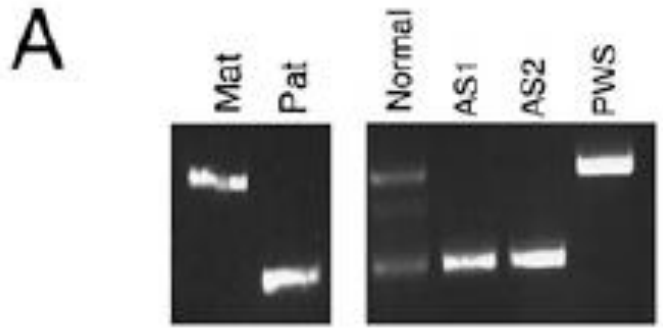


http://www.angelman.org/understanding-as/medical-info/genetic-counseling-in-angelman-syndrome/

Diagnostic Testing for AS

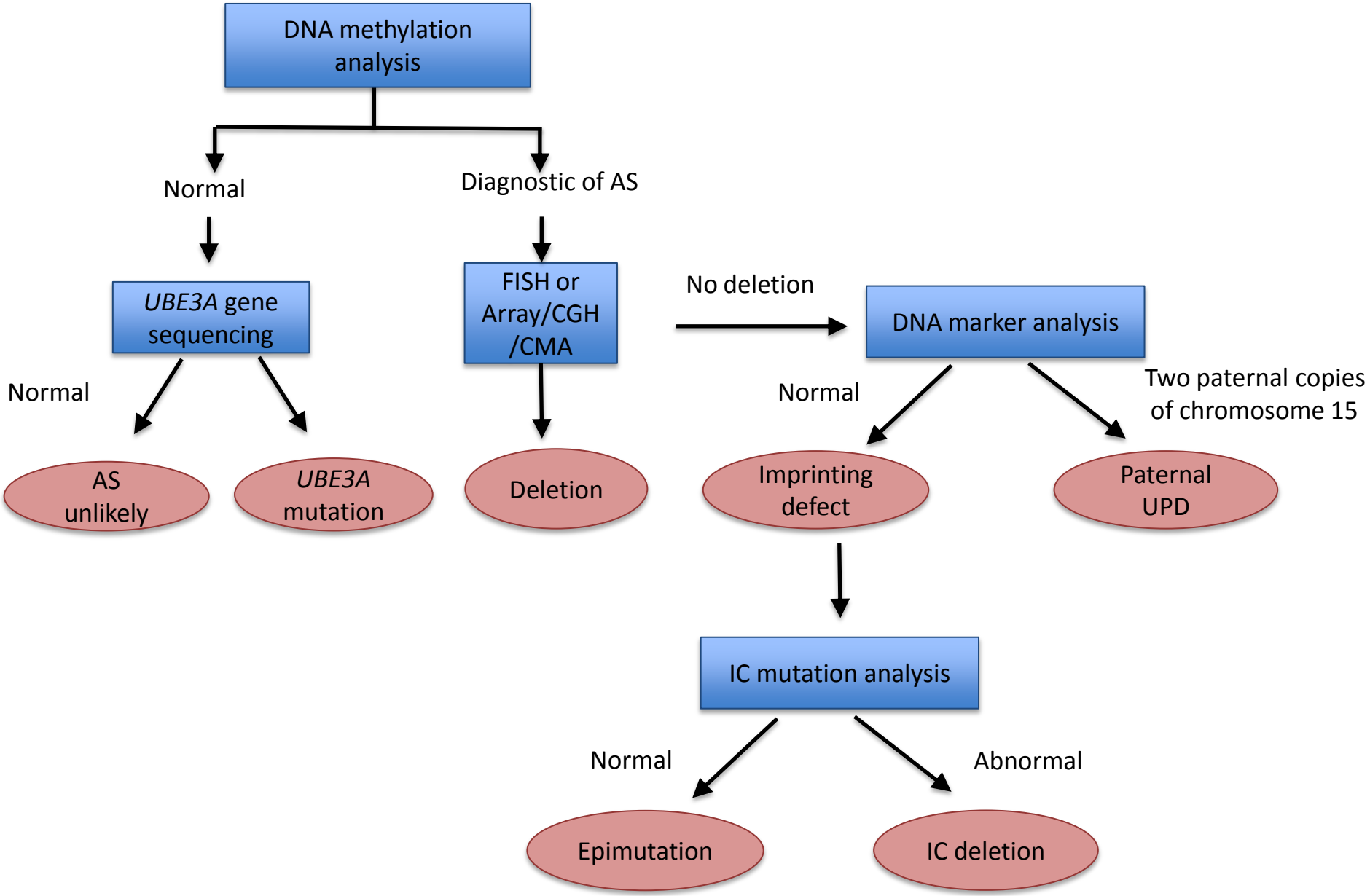


DNA methylation testing

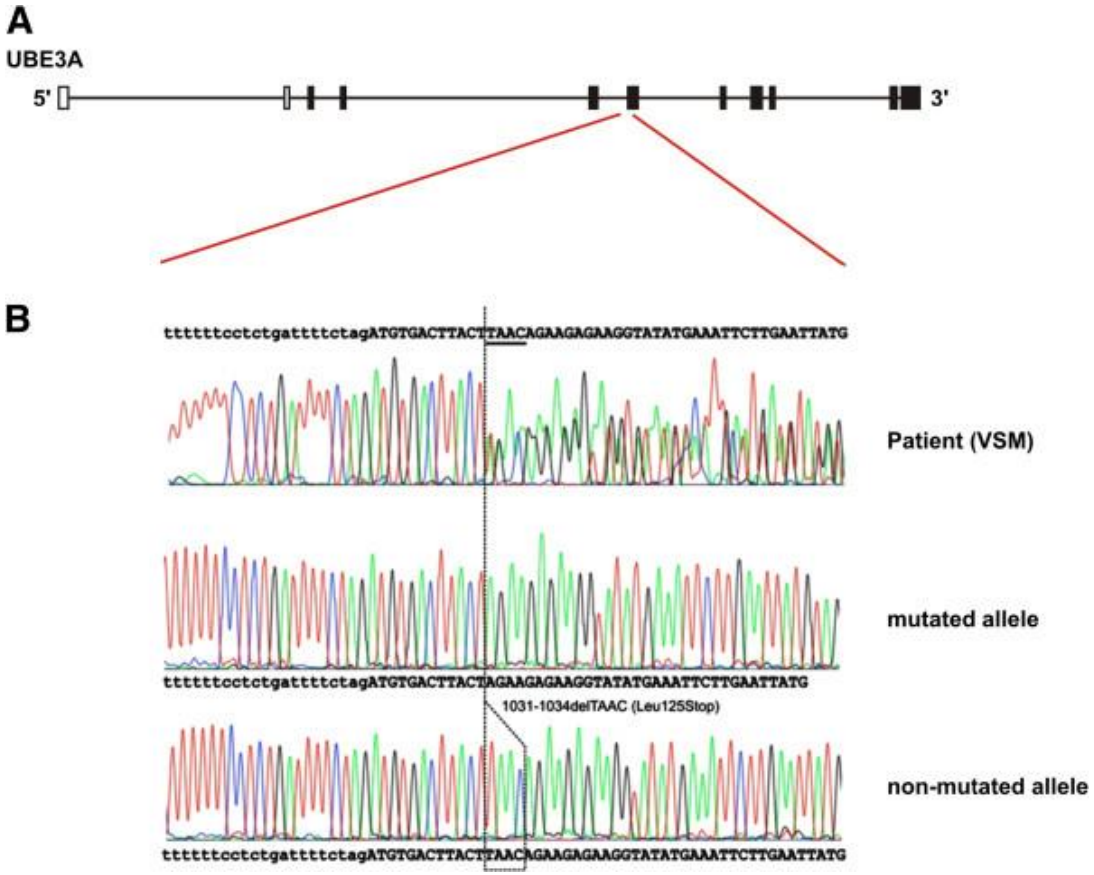


* methylation-specific PCR

Diagnostic Testing for AS

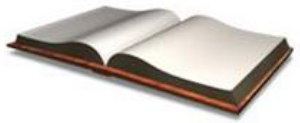


UBE3A gene sequencing



DeMolfetta et al., BMC Med Genet. 2012 Dec 20;13:124. doi: 10.1186/1471-2350-13-124

What do we learn from UBE3A gene sequencing



ACC GUG CUU ACG GGG UAC AUA
T V L T G Y I

The UBE3A gene encodes a protein called a ubiquitin ligase.

1. A polymorphism is a change that doesn't change the meaning of the sequence.

ACC GUG CUU ACC GGG UAC AUA
T V L T G Y I

The UBE3A gene encodes a protein known as a ubiquitin ligase.

2. Some mutations may change the meaning of the sequence impacting the protein.

ACC GUG CCU ACC GGG UAC AUA
T V P T G Y I

The UBE3A gene explodes a protein called a ubiquitin ligase.

3. A "stop" mutation may alter the sequence and truncate the protein so it isn't functional.

ACC GUG CUU ACC GGG UAA AUA
T V L T G *

The UBE3A gene encodes a protein called.

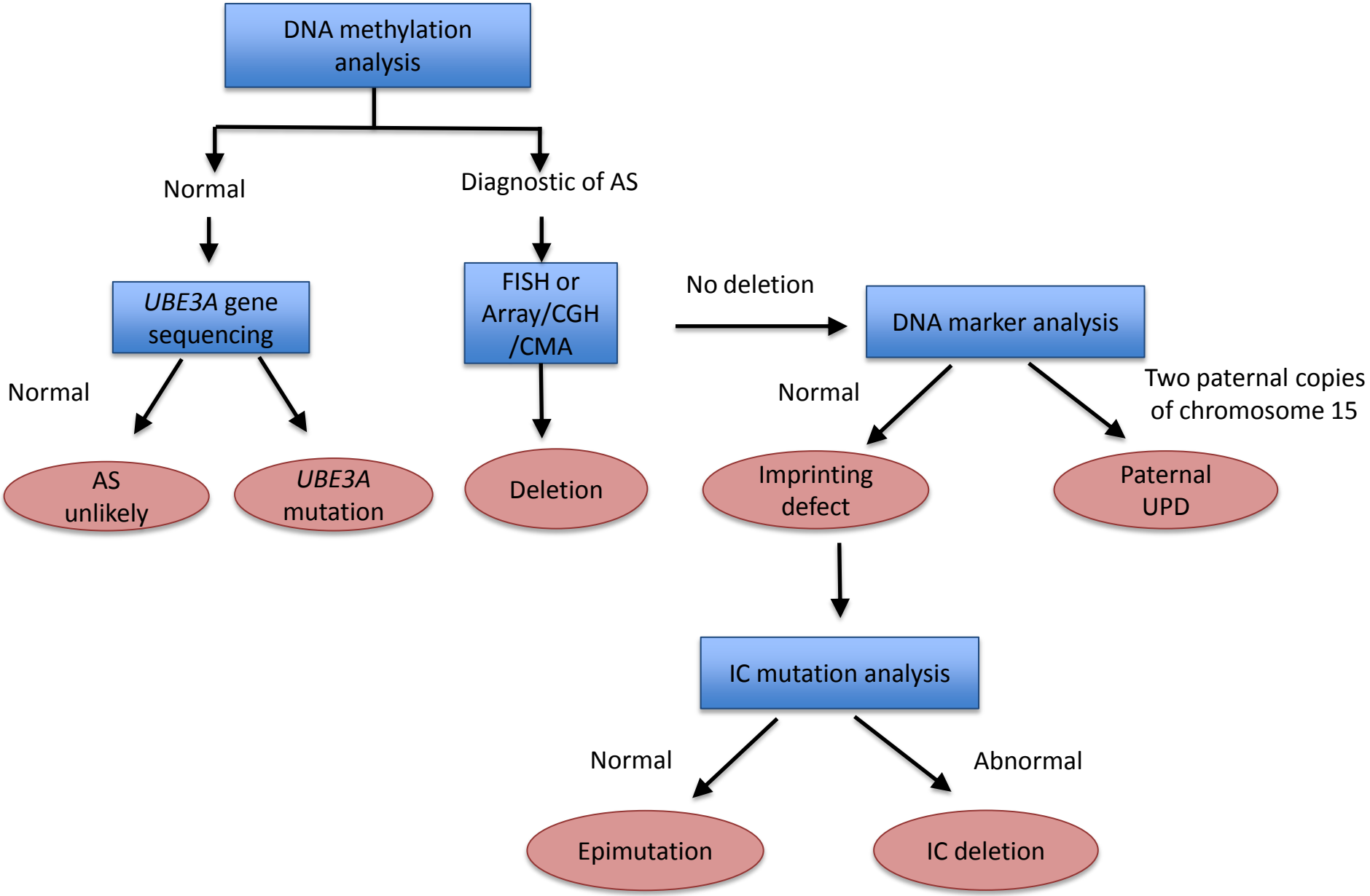
4. Mutations that add or delete sequence will change how the sequence is read impacting the protein.

ACC GUG CUU *AAC GGG GUA CAU A*
T V L N G V H

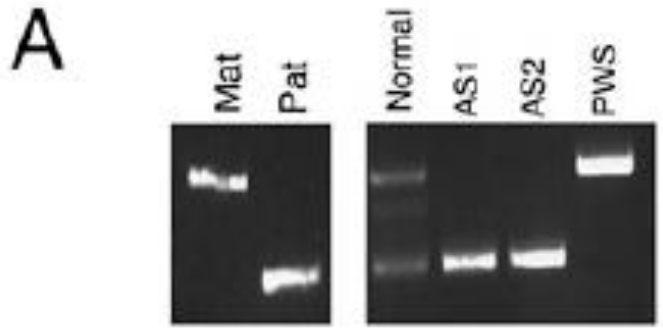
The UBE3A gene encod esapro teinca llediquil igase.

http://www.cureangelman.org/what-testing101.html

Diagnostic Testing for AS

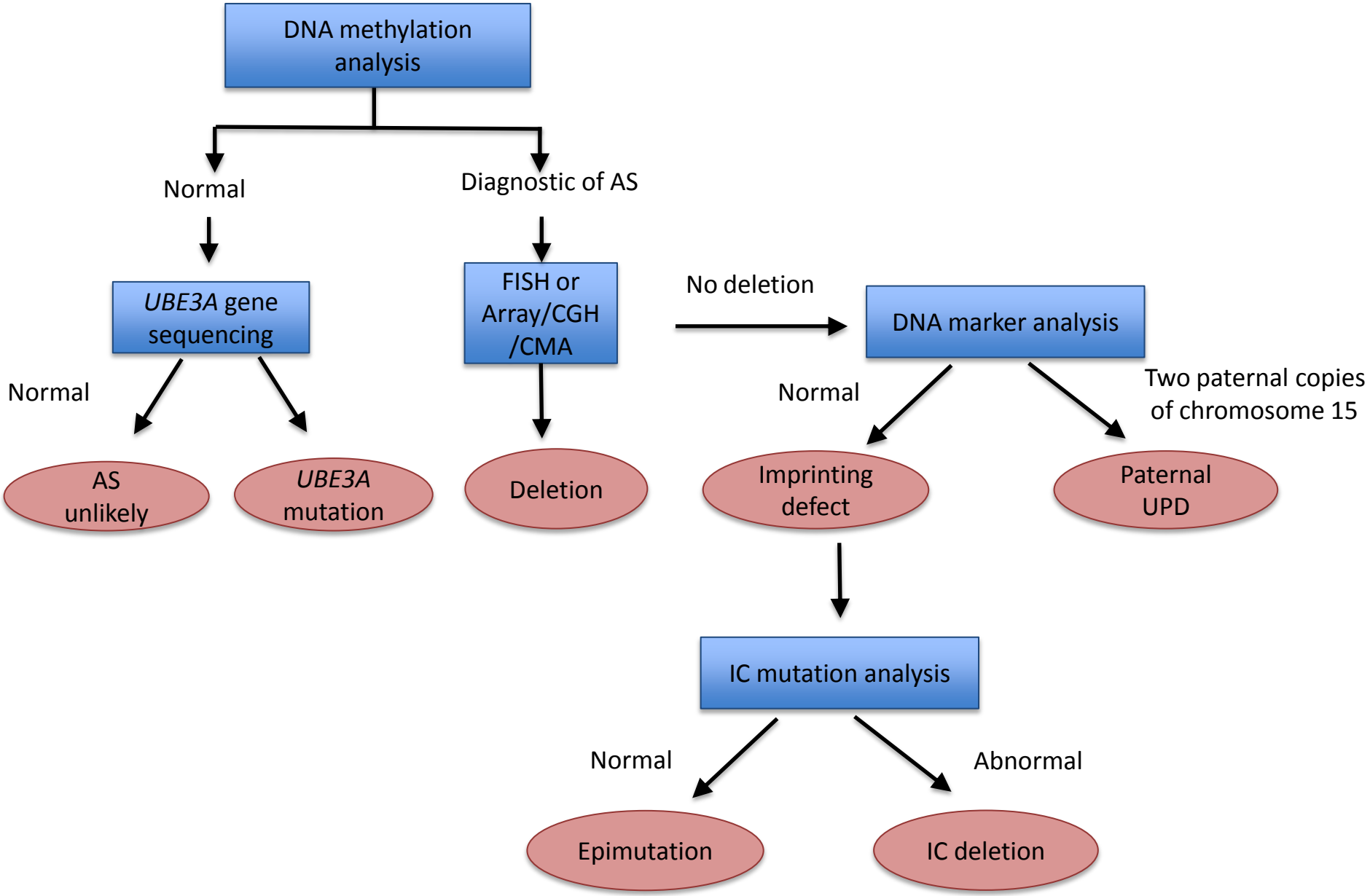


DNA methylation testing

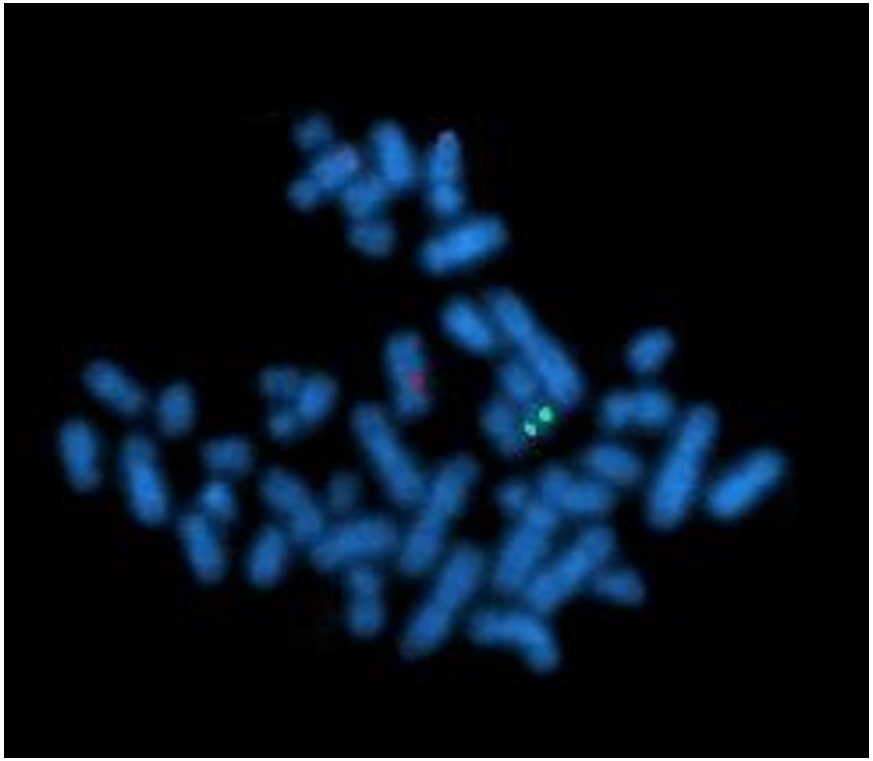


* methylation-specific PCR

Diagnostic Testing for AS



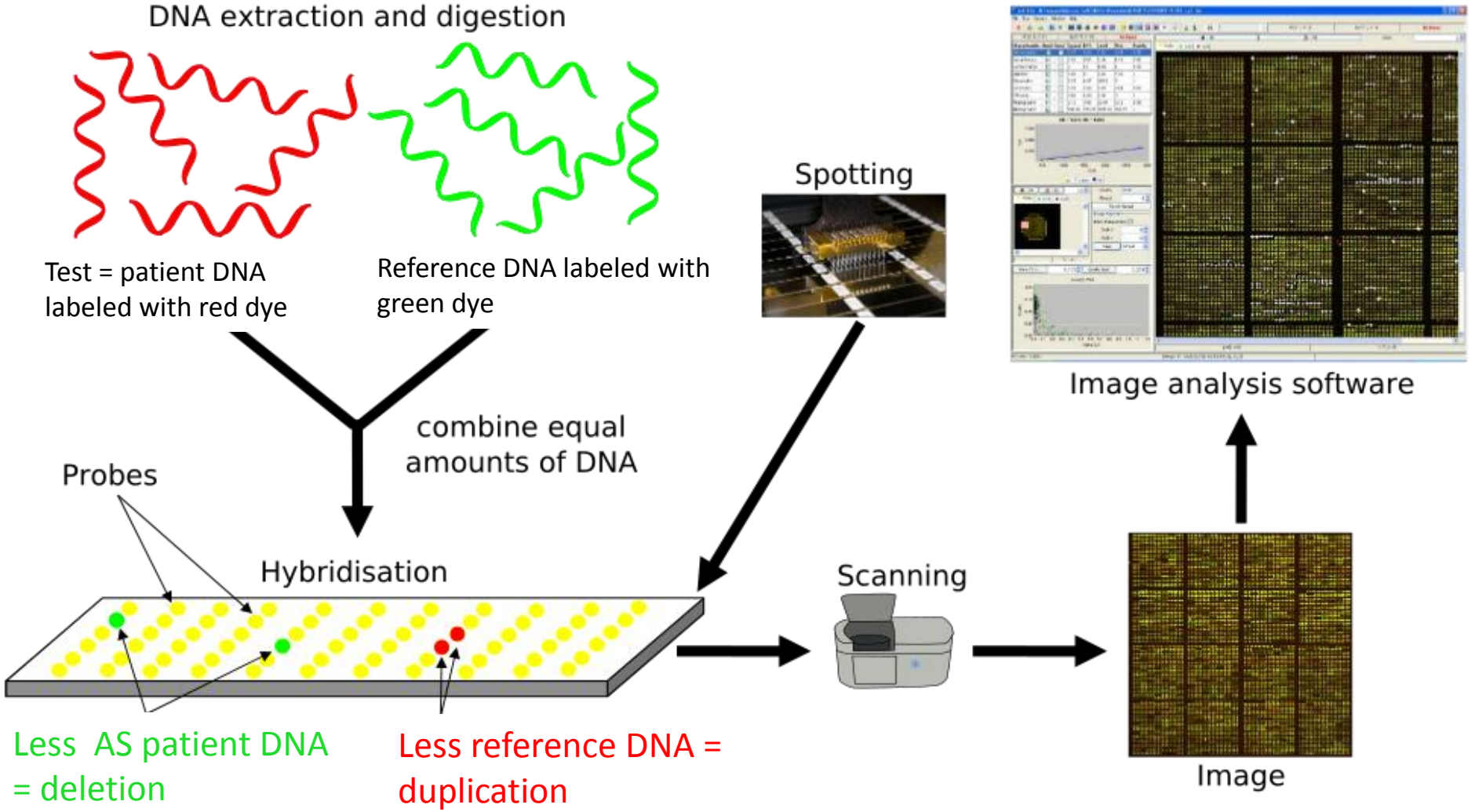
FISH—fluorescent in situ hybridization



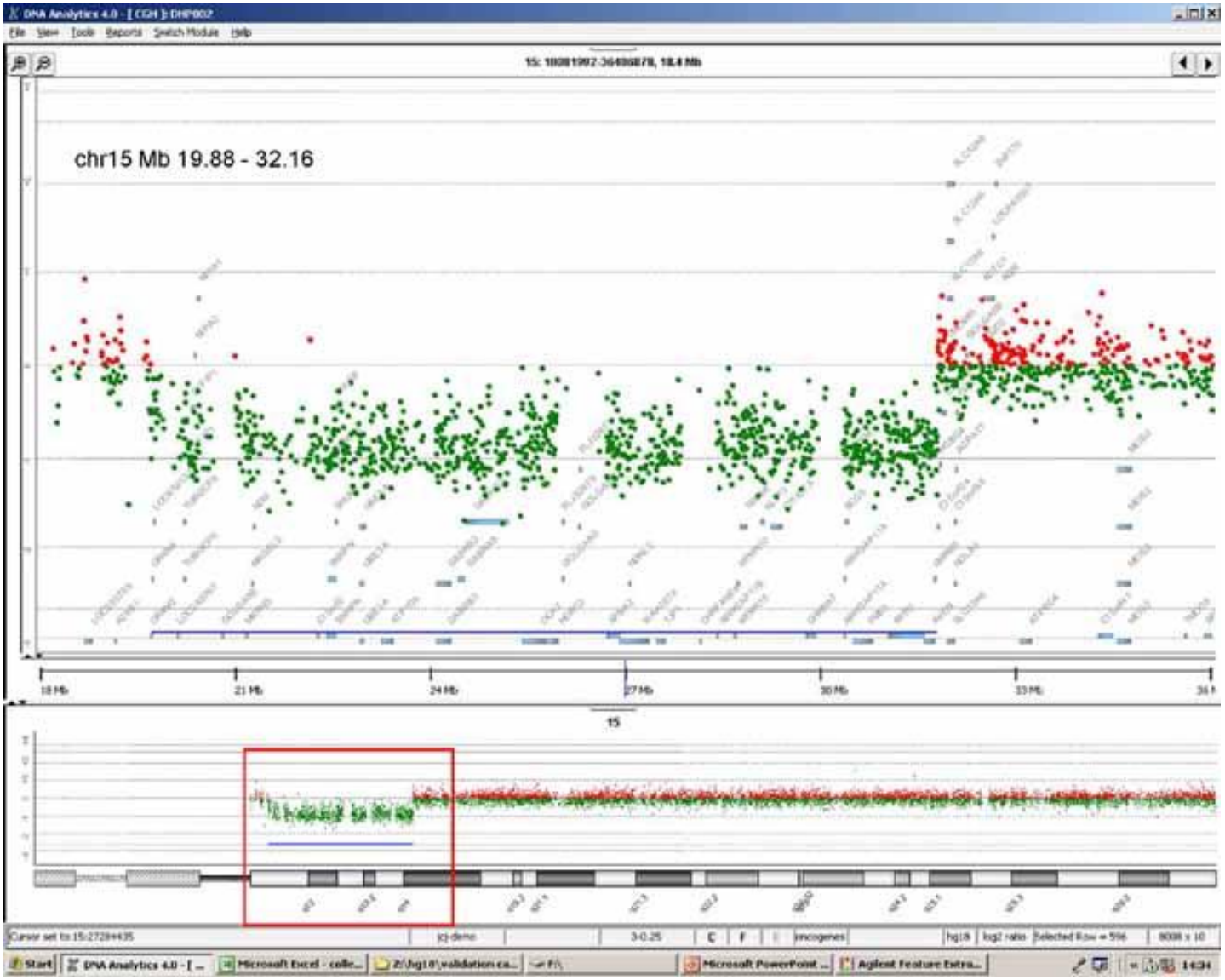
Green = the tip of chromosome 15, which should be present in everyone
Red = UBE3A gene. If missing, then the individual is deletion-positive.

http://www.cureangelman.org/what-testing101.html

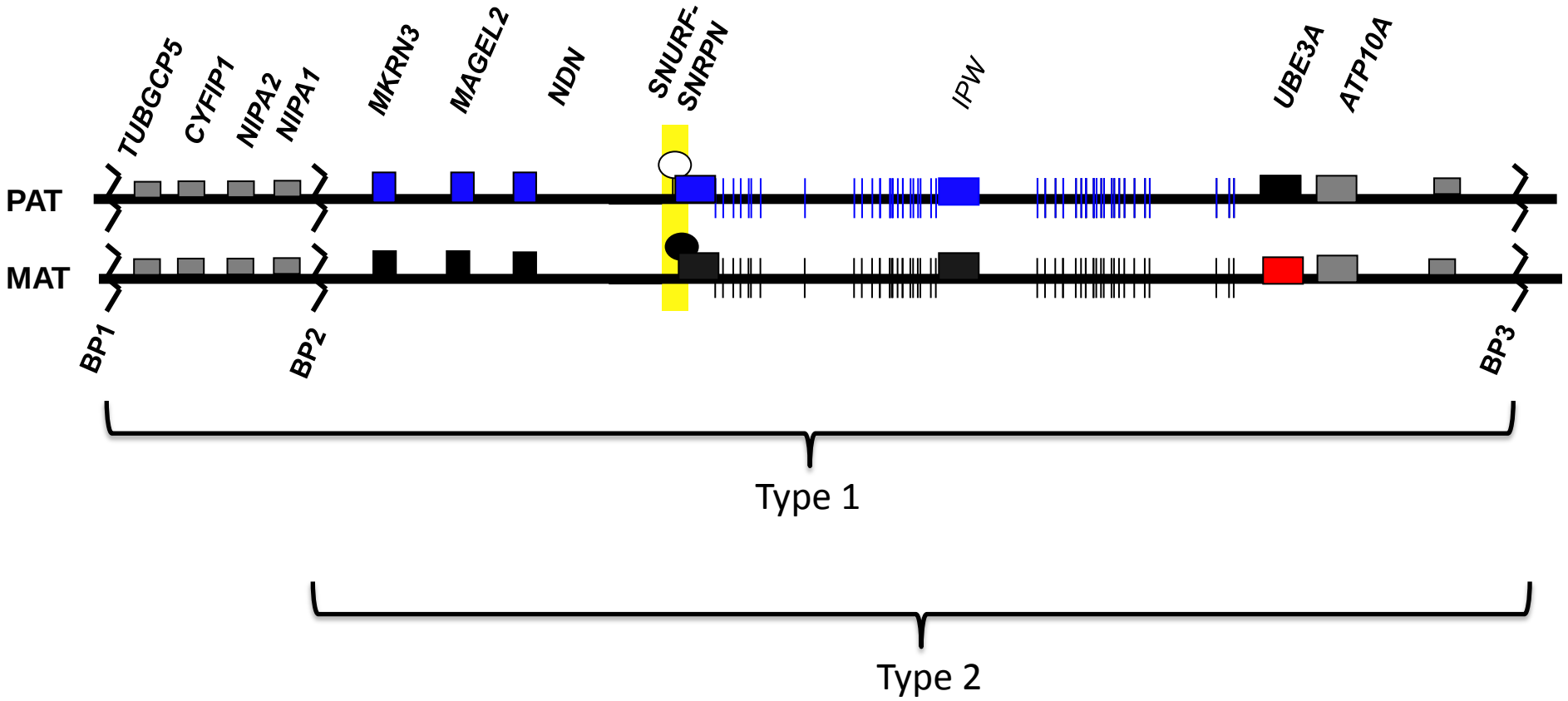
Microarray/Chromosomal Microarray/aCGH



Microarray/Chromosomal Microarray/aCGH

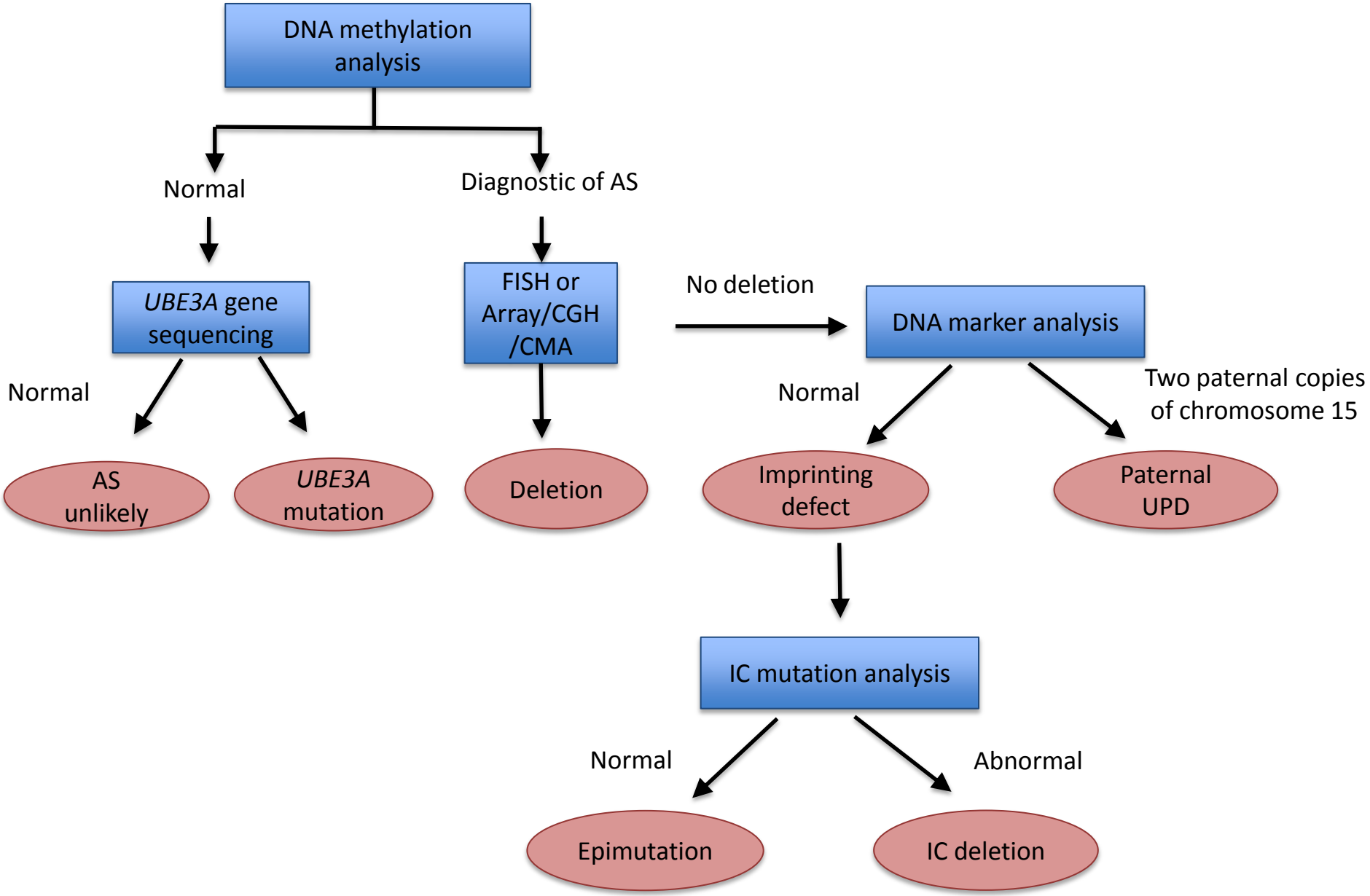


Type 1 vs Type 2



- There are additional breakpoints (BPs) downstream of BP3 that are used less frequently
- The BPs are nearly identical DNA sequences that are repeated. Repeats = possible deletions

Diagnostic Testing for AS

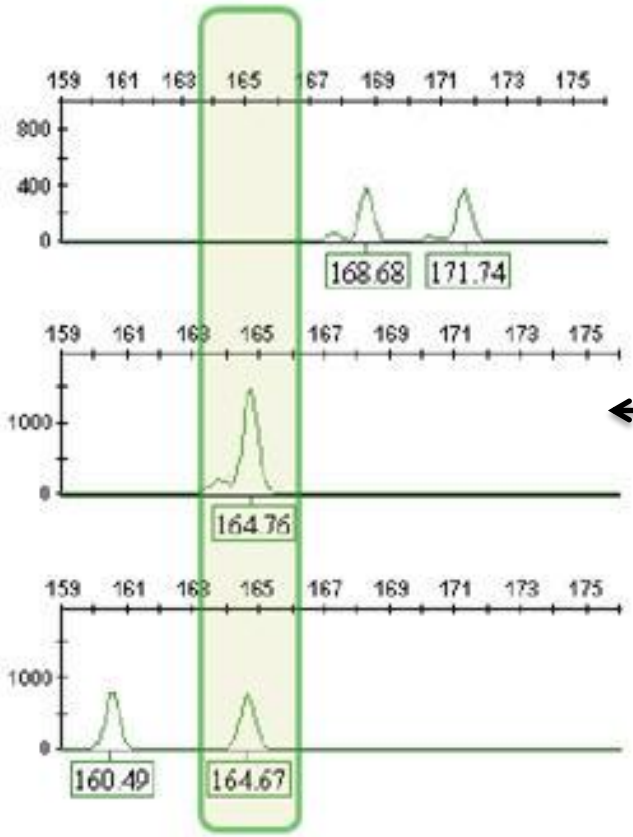


DNA marker analysis

mother

child w/AS

father

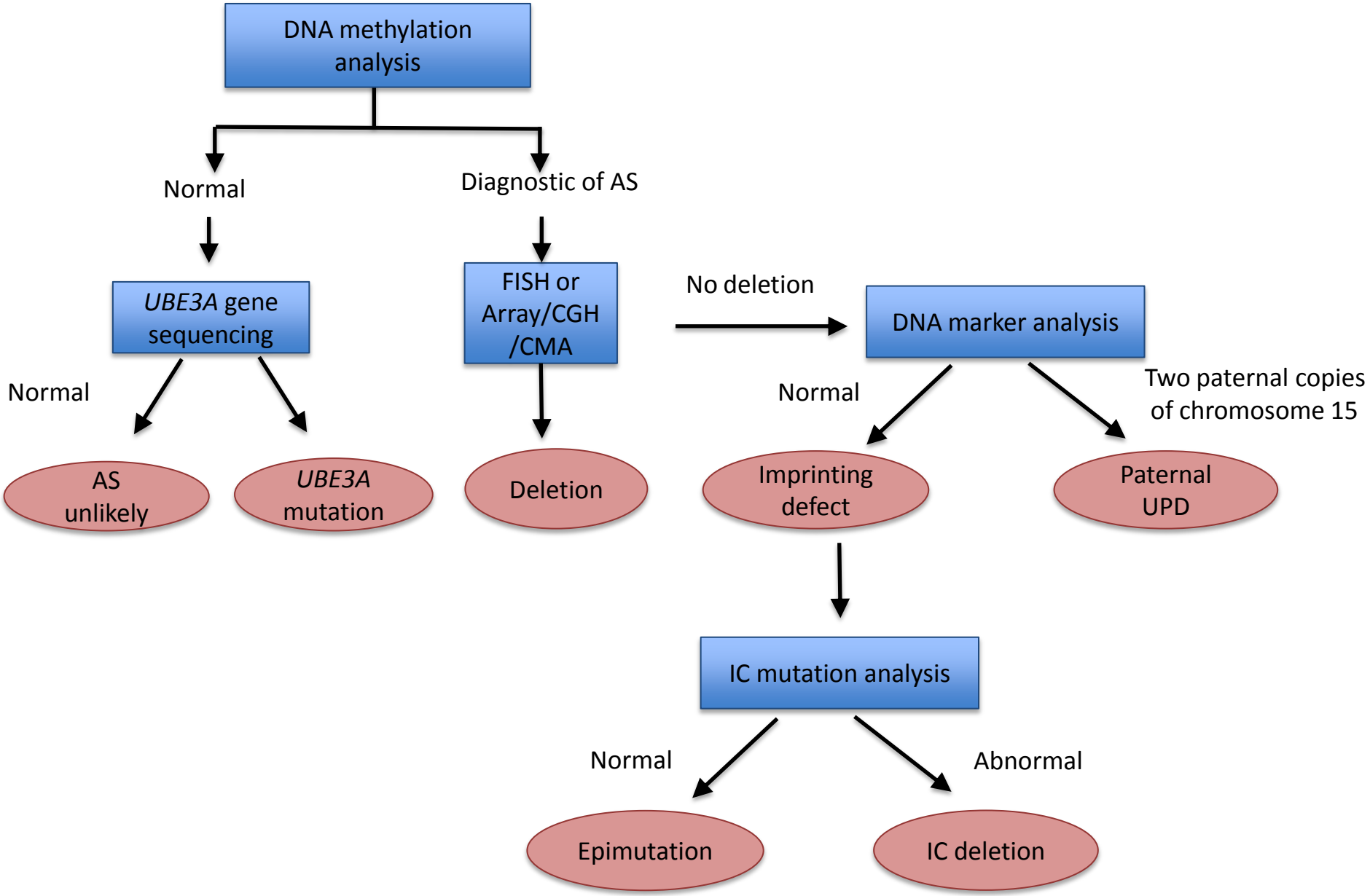


Maternal (upper), proband (middle) and paternal (lower) samples analysed together using Devyser UPD-15. The highlighted allele is inherited from the father. No allele is inherited from the mother

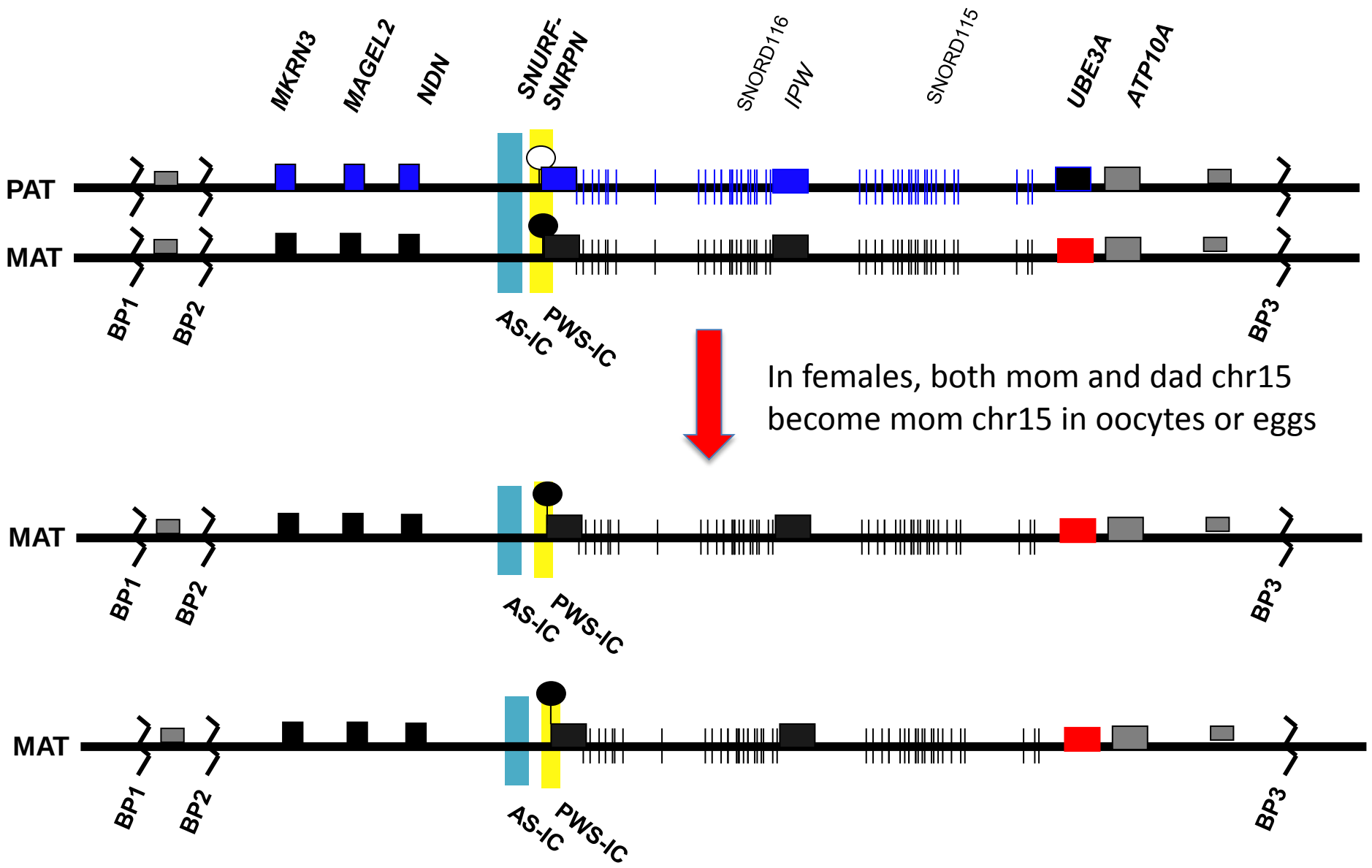
AS caused by paternal UPD

http://www.devyser.com/genetic-testing/devyser-upd-15

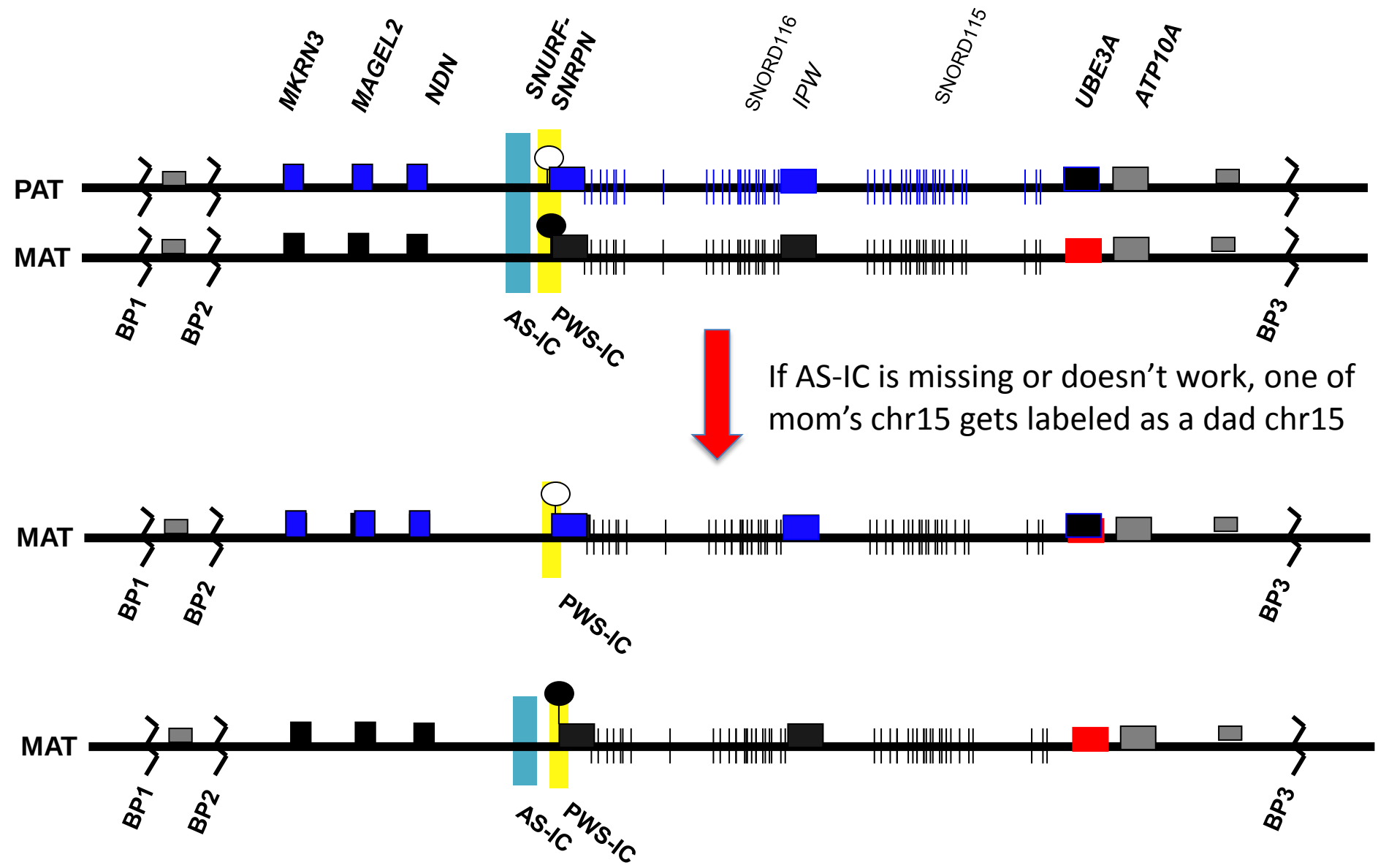
Diagnostic Testing for AS



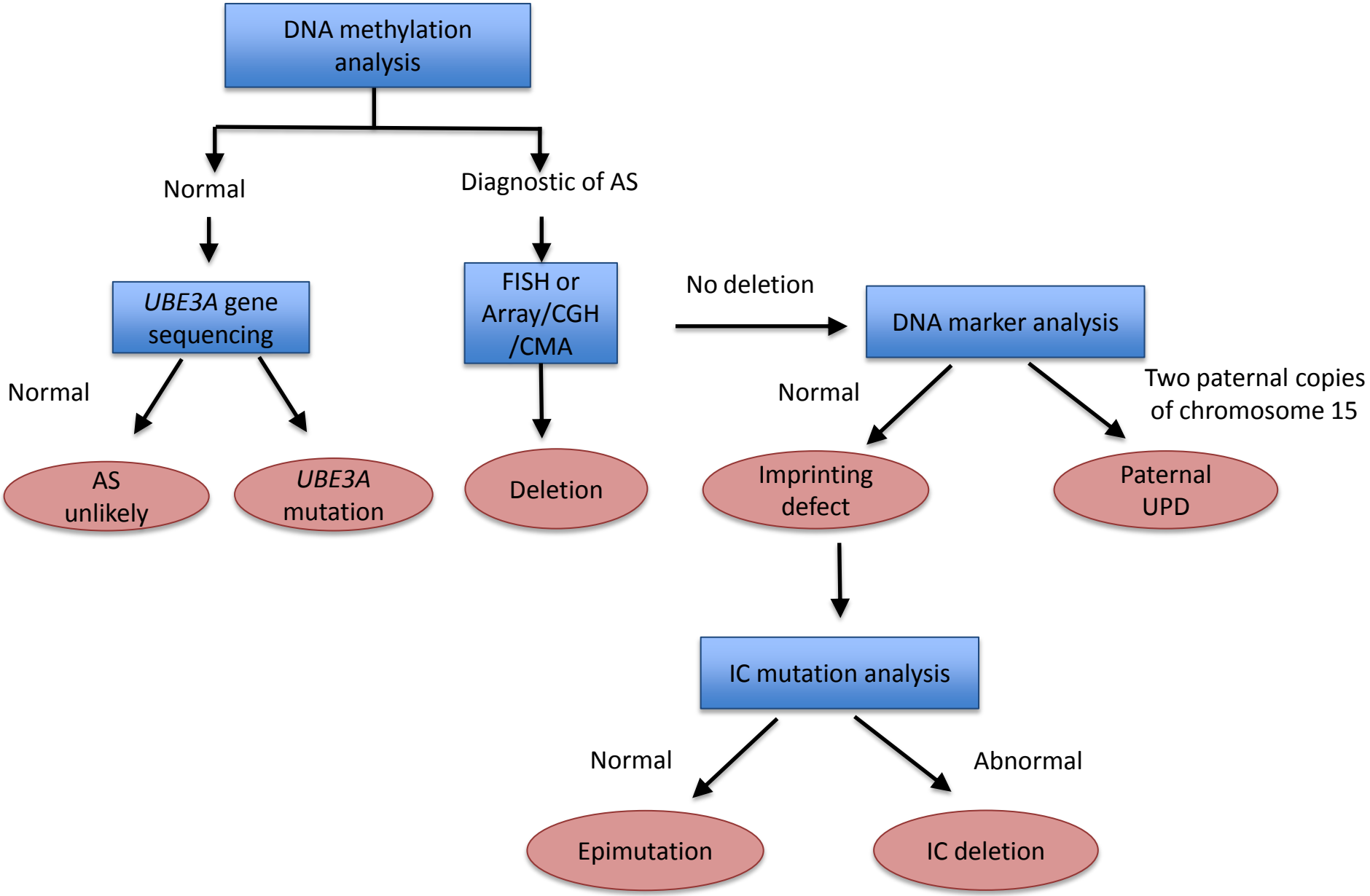
Imprinting defects



Imprinting defects

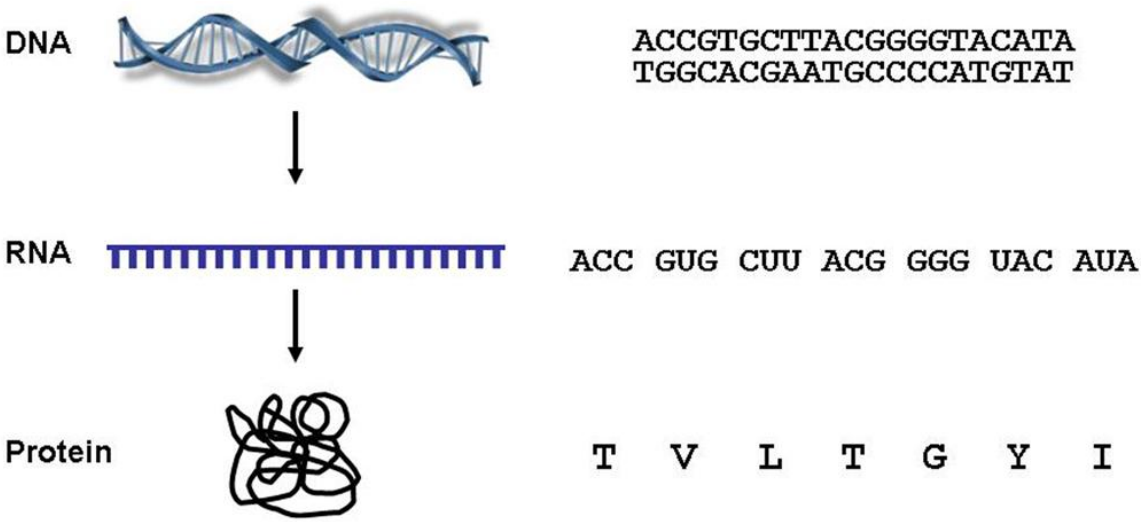


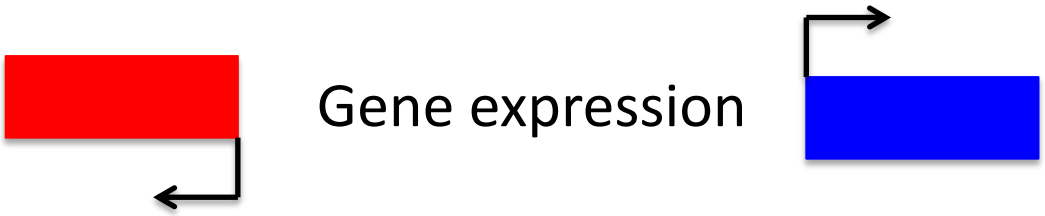
Diagnostic Testing for AS



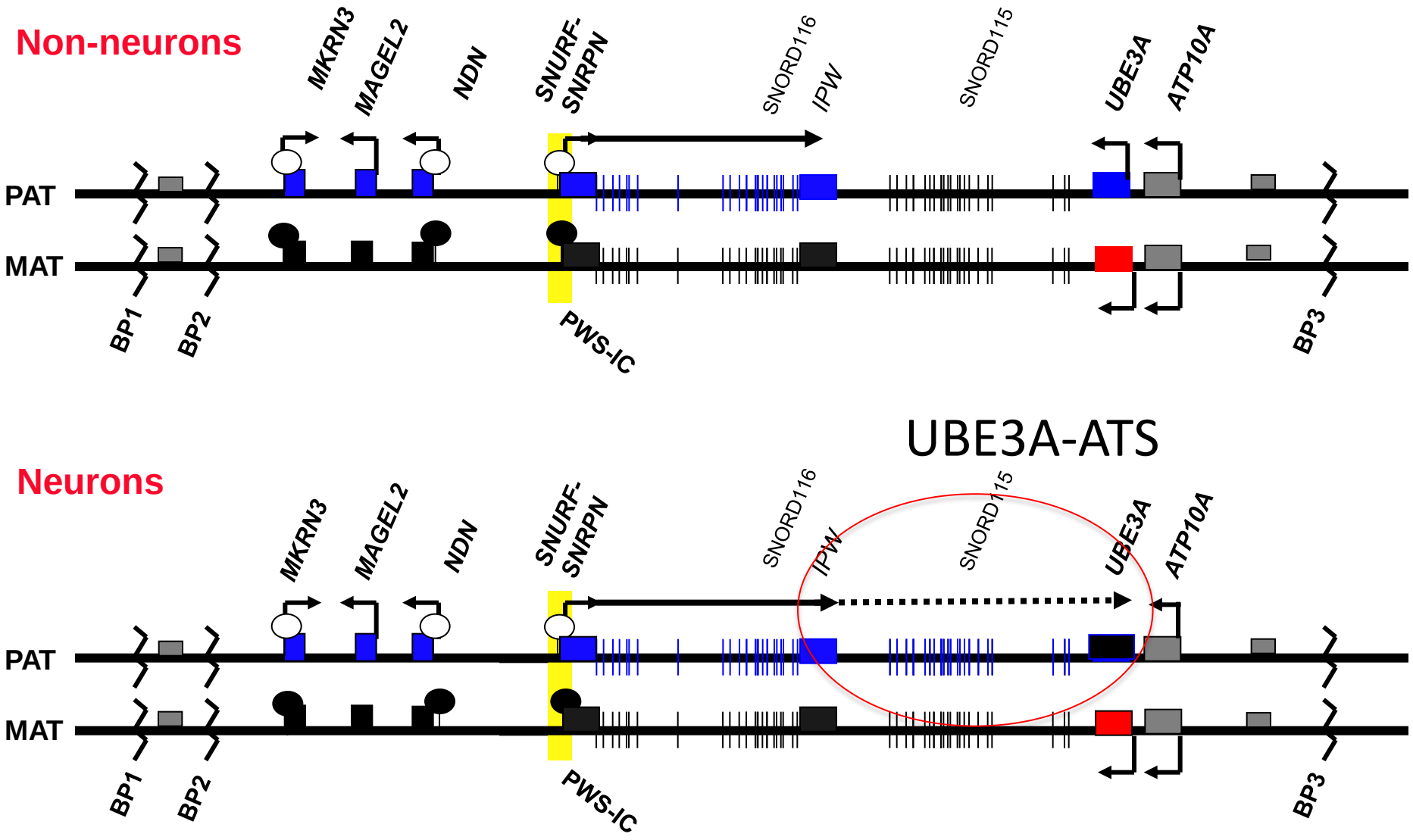
Advanced Genetics

DNA usually encodes RNA, which then encodes a protein



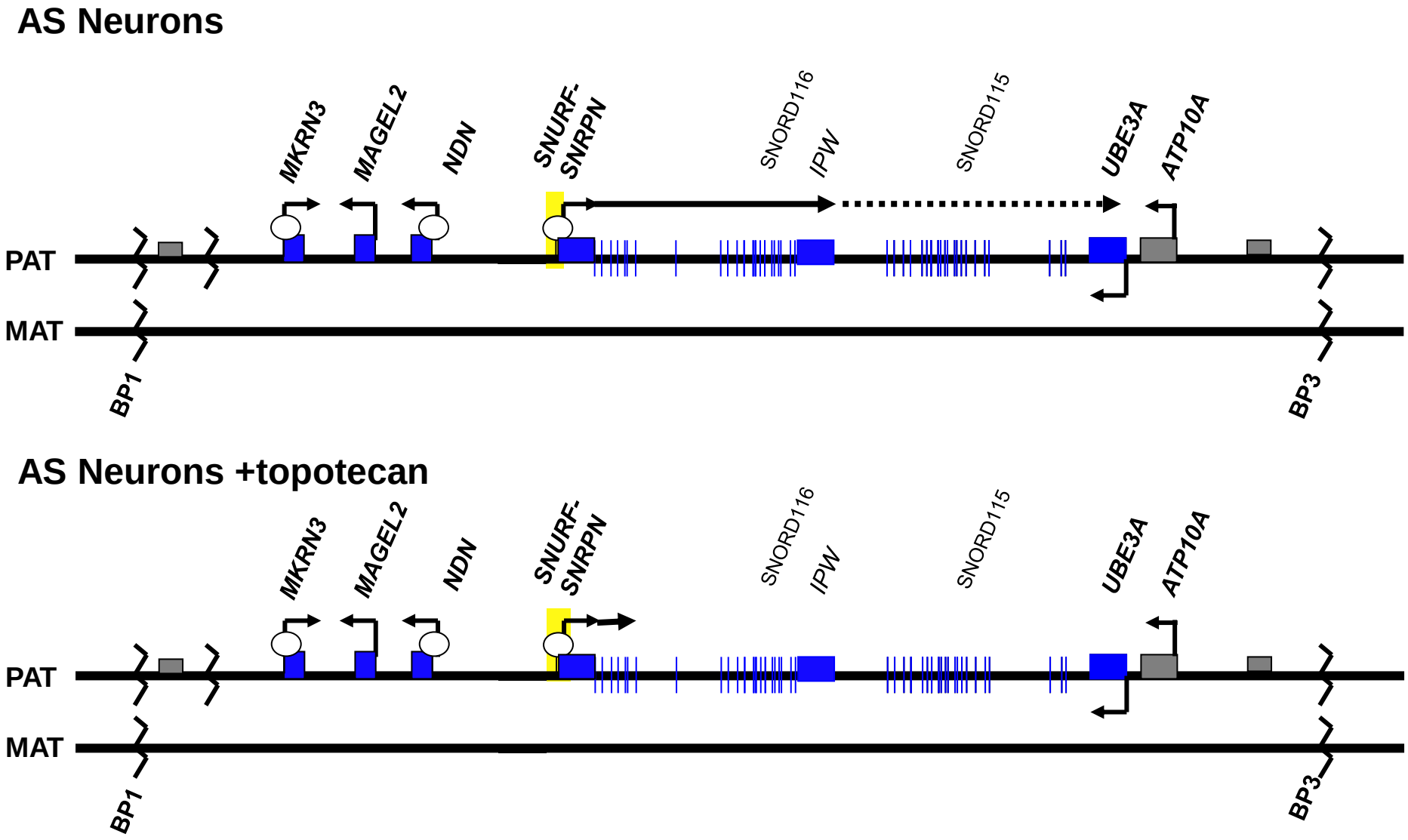


Gene expression across the chromosome 15q11-q13 region



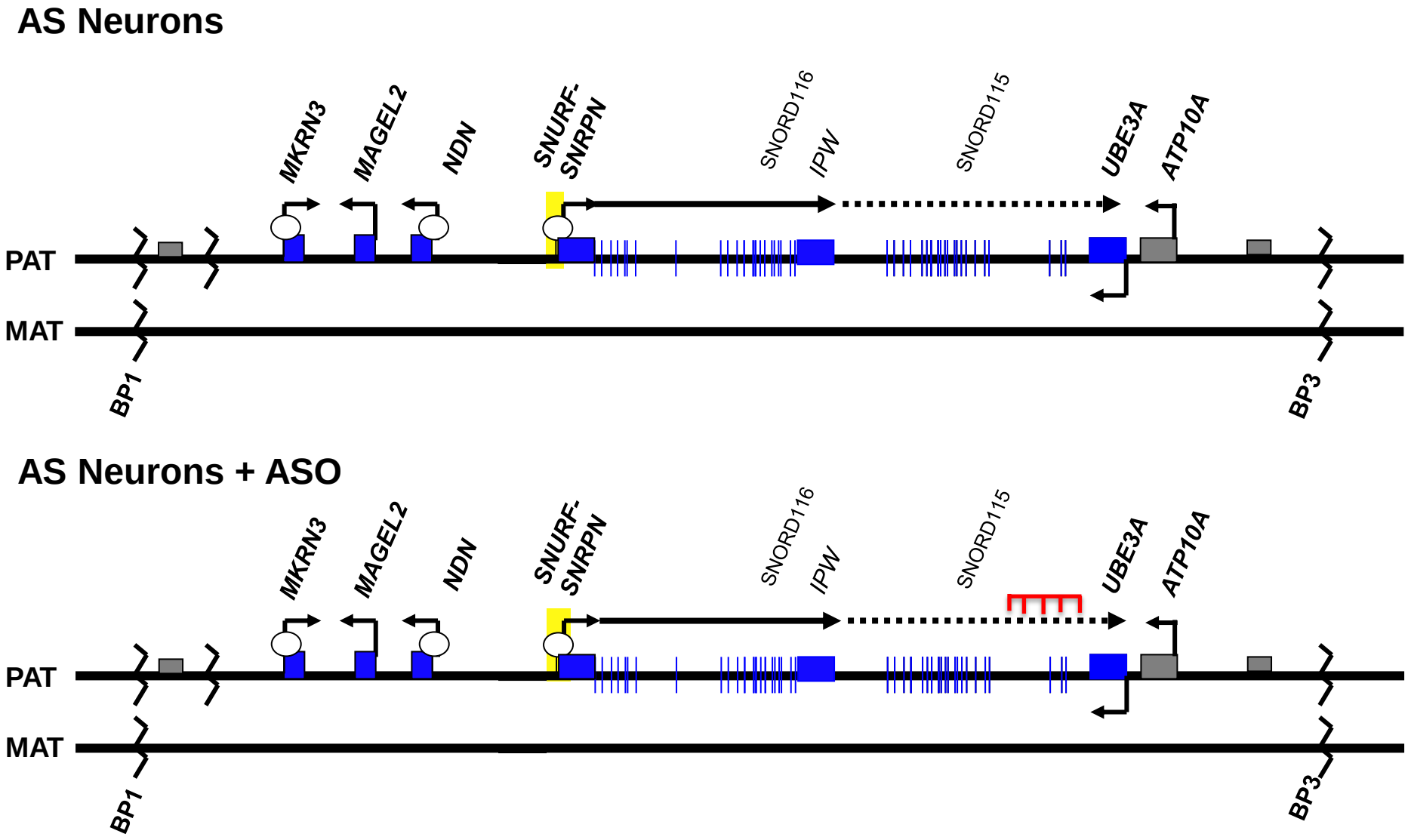
UBE3A-ATS silences paternal UBE3A

Topotecan reduces UBE3A-ATS



Topotecan reduces UBE3A-ATS

Antisense oligonucleotides (ASOs) reduce UBE3A-ATS



Antisense oligonucleotides (ASOs) reduce UBE3A-ATS

*

Acknowledgments

UCONN HEALTH

Families who have donated samples

Funding

Raymond and Beverly Sackler Foundation

NIH *Eunice Kennedy Shriver* National Institute of Child Health and Human Development

DPH Keeping Connecticut Healthy

Fighting Angels FOUNDATION *Fighting to talk...Fighting to walk...Fighting for a cure.*

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