Pharmacogenetics and pharmacogenomics

Vincent Bours

Genetics Center

CHU / University of Liège

Pharmacogenomics in the clinic

Mary V. Relling[1] and **William E. Evans**[1]

[1]Department of Pharmaceutical Sciences, St. Jude Children's Research Hospital, Memphis, TN

Pharmacogenetics

- **Study of genetic differences between individuals that influence the clinical response to a drug**

- **Genetic factors predictive of the response to a specific drug**

- **Choice of the best medicine for an individual patient**

Personnalized medicine
Precision medicine

Pharmacogenomics

- **Study the interactions between drugs and the genome**

- **Global approach / drug discovery / genomic markers**

Pharmacogenetics

- **Genetic factors predictive of the response to a specific drug**

  - **Probability of a clinical response**
  - **Risk of severe side effects**

- **Drug prescription: method by « try and error »**

- **USA: adverse reactions to prescription drugs:**
- **2 million people each year**
- **100 000 deaths/year**
- **7% of hospital admissions**
- **Cost: > 30 billion $**

- **Causes:**
  - **Environmental: drug interactions**
  - **Co-morbidity**
  - **Genetic factors**

- **Pharmacogenetics: Could genetic factors be anticipated?**

If it were not for the great variability among individuals, medicine might as well be a science and not an art.

Sir William Osler, 1892

REVIEW ARTICLE

GENOMIC MEDICINE

Alan E. Guttmacher, M.D., and Francis S. Collins, M.D., Ph.D., *Editors*

Inheritance and Drug Response

Richard Weinshilboum, M.D.

REVIEW ARTICLE

DRUG THERAPY

Alastair J.J. Wood, M.D., *Editor*

Pharmacogenomics — Drug Disposition, Drug Targets, and Side Effects

William E. Evans, Pharm.D., and Howard L. McLeod, Pharm.D.

With your genes? Take one of these, three times a day

HUMAN GENETICS SPECIAL

Truly 'personalized' medicine remains a distant goal. But researchers are now thinking about how to use genomic data to avoid prescribing drugs that may kill, or won't work.

Pharmacogenomics and Drug Toxicity

Yusuke Nakamura, M.D., Ph.D.

The NEW ENGLAND JOURNAL of MEDICINE
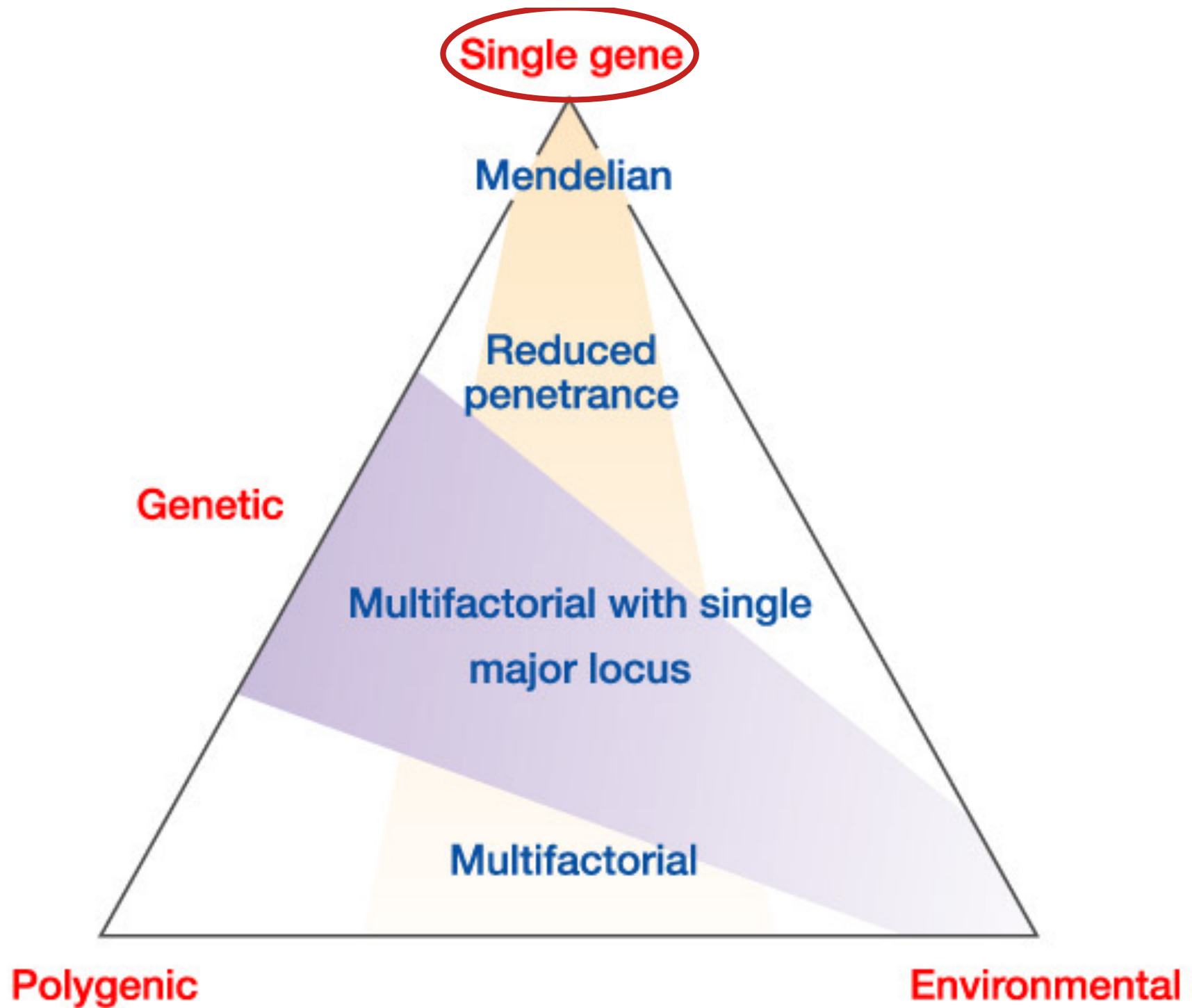
Estimation of the Warfarin Dose with Clinical and Pharmacogenetic Data

The International Warfarin Pharmacogenetics Consortium*
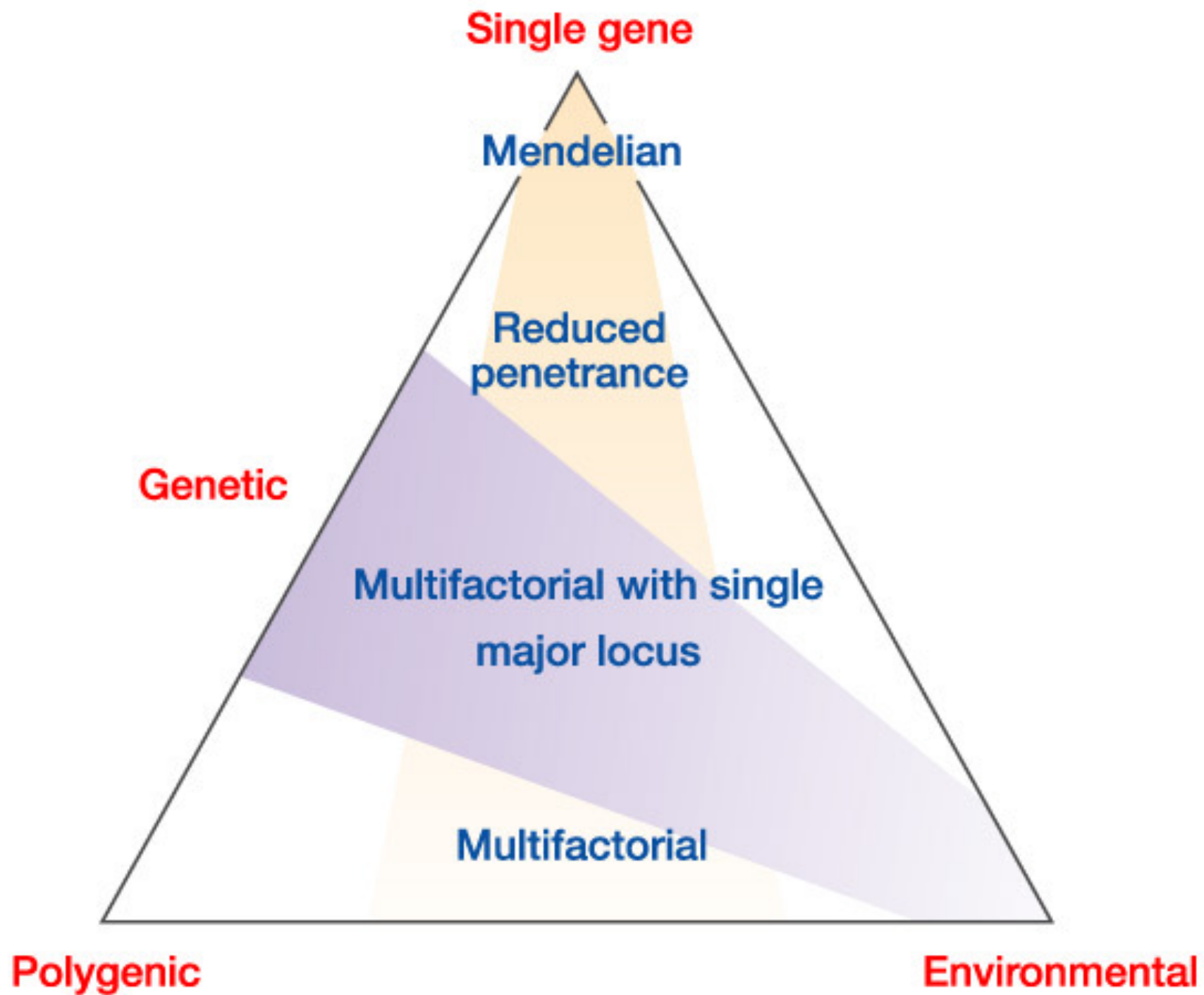
Alcohol

Several genes are involved in alcohol metabolism.

Alcool $\xrightarrow{\text{ADH}}$ **Acétaldéhyde** $\xrightarrow{\text{ALDH}}$

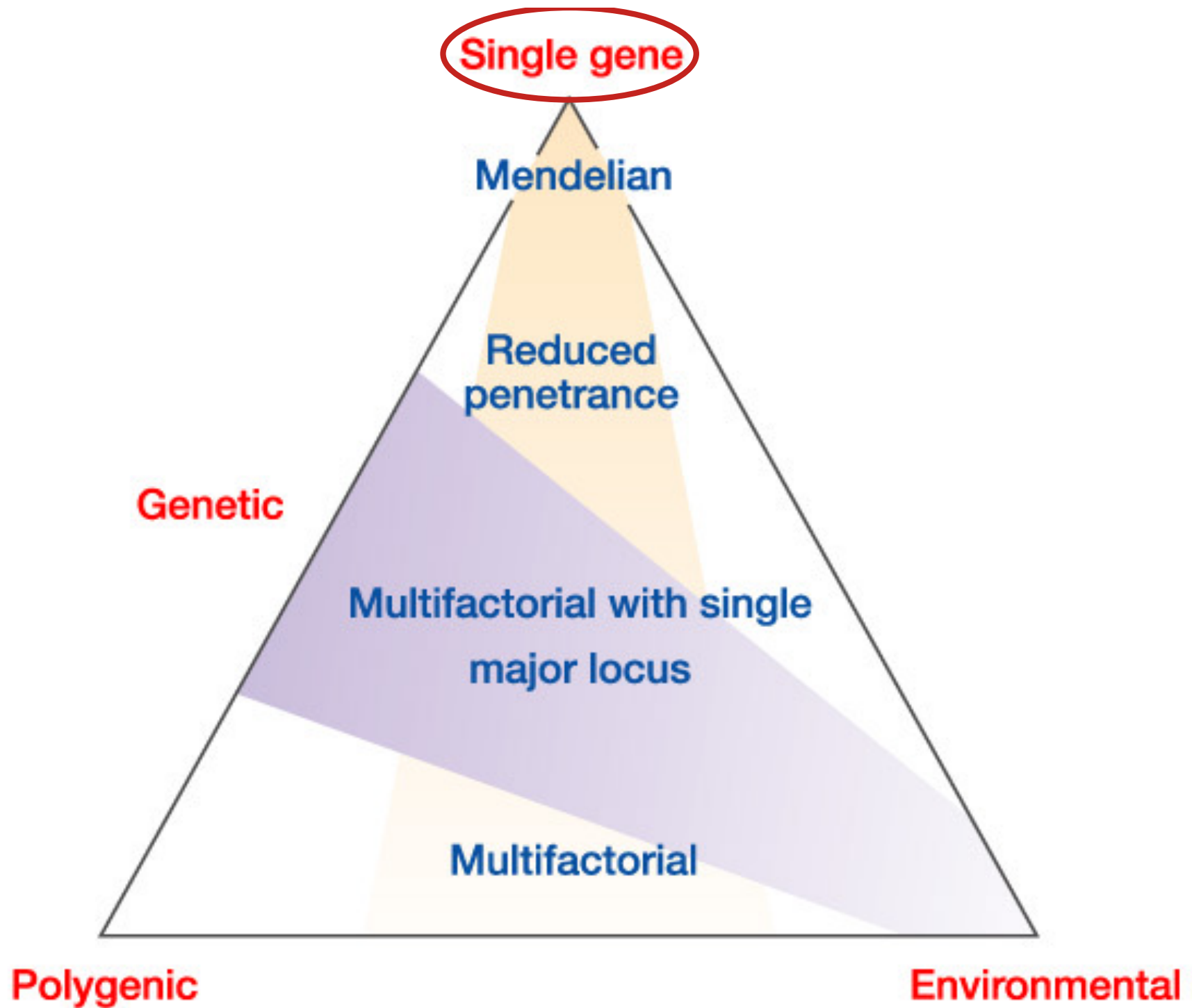# ALDH2-2

Moderate dose of alcohol.
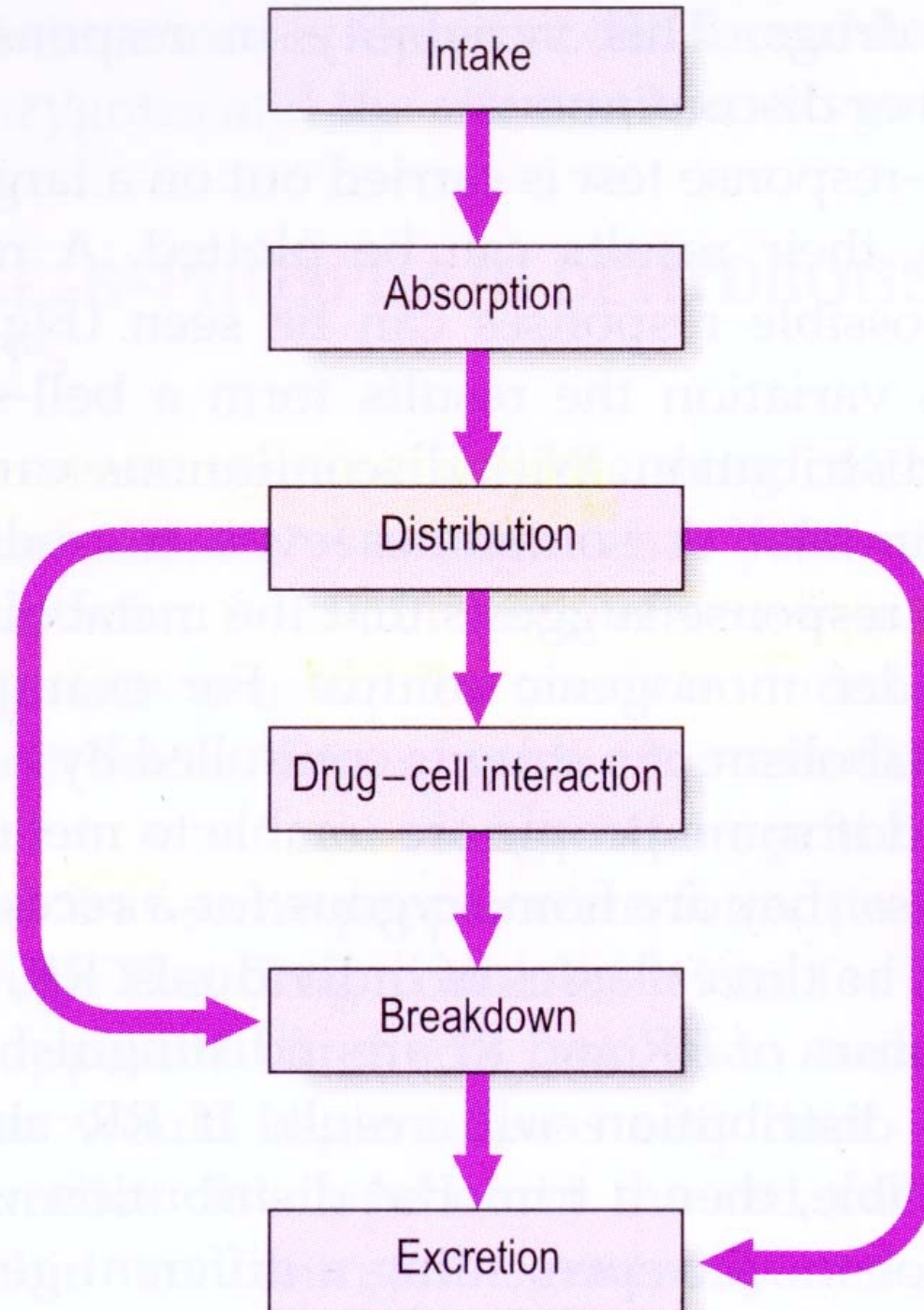
Good metabolism
OK

Bad metabolism
KO

Environment
Genetic factors
Ethnic differences

Table 11.1 Ethnic variations in some pharmacogenetic disorders

Disorder	Ethnic group	Frequency (%)
Slow acetylation	Europeans	50
	Orientals	10
Pseudocholinesterase variants	Europeans	<1
	Eskimos	1–2
G6PD deficiency	N. Europeans	<1
	S. Europeans	up to 25
	Afro-Caribbeans	10
Atypical ADH	Europeans	5
	Orientals	85

Single gene

Mendelian

Reduced penetrance

Genetic

Multifactorial with single major locus

Multifactorial

Polygenic

Environmental

ADME

Pharmacogenetics

1. Drug metabolism
- Phase I: CYP, ...
- Phase II: TPMT, NAT2, GST, ...

2. Transport
- MDR (ABC)

3. Targets
- Beta-adrenergic receptor

4. Unexpected side effects
- Long QT
- Deafness and aminoglycosids, ...

Pharmacogenetics

1. Drug metabolism

- Phase I: CYP, ...
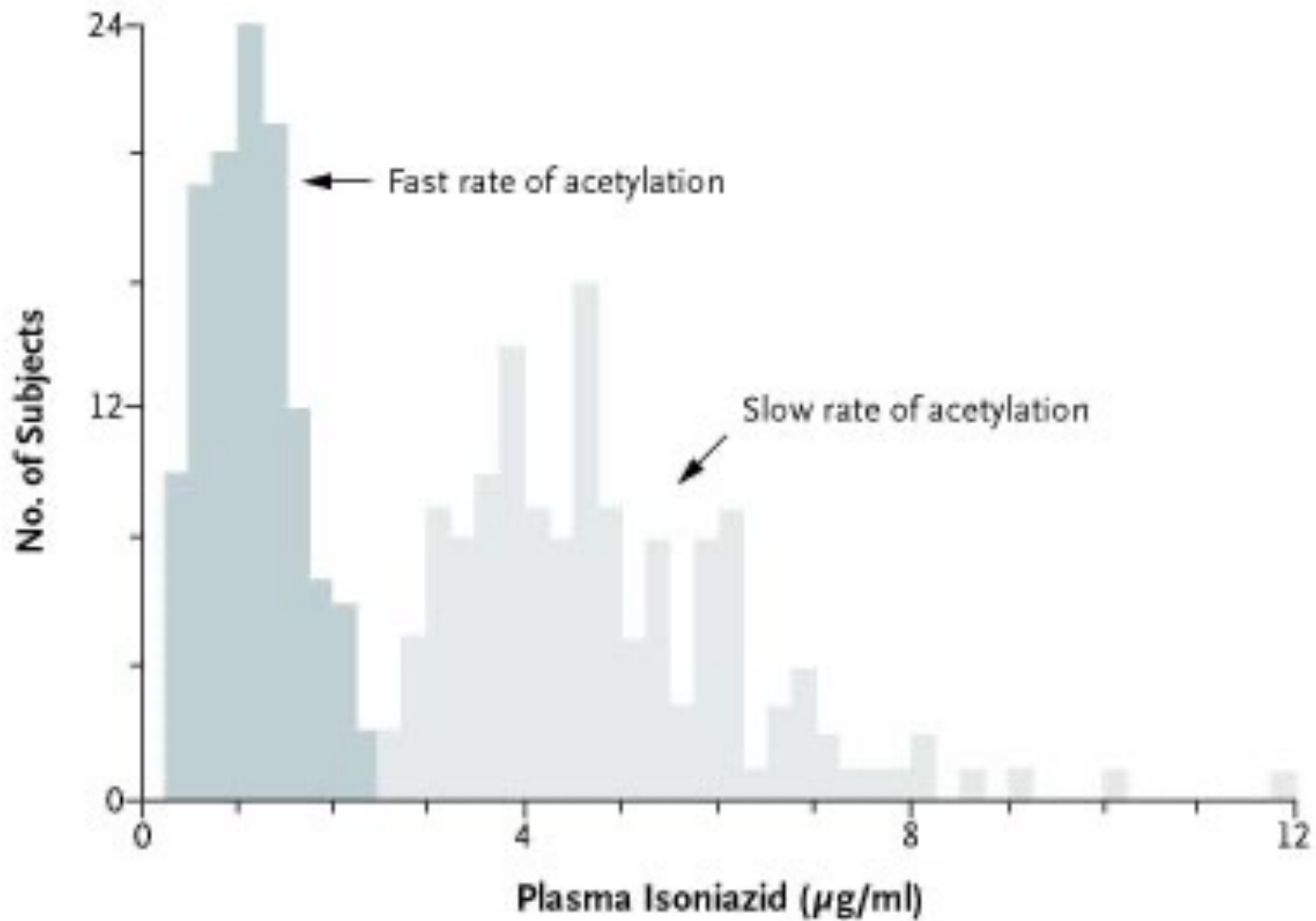- Phase II: TPMT, NAT2, GST, ...

1950s

Inherited traits: plasma or urine drug concentrations

Response to succinylcholine

- Pseudocholinesterase
- 1/3500 white subjects
- Missense mutation

Pharmacokinetics of isoniazid

- N-acetyltransferase

Ethnic variations !

Pharmacokinetics
....

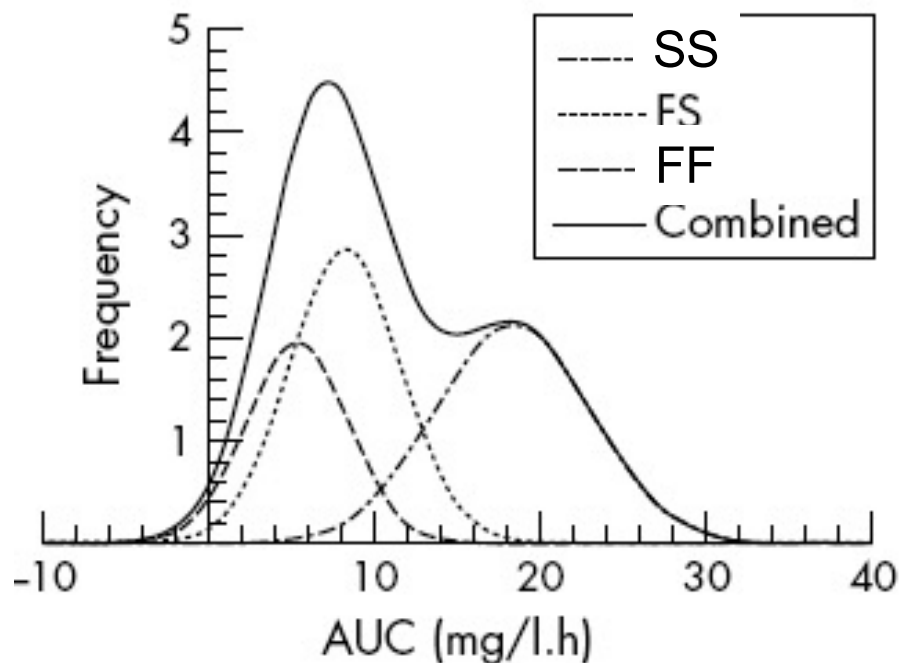# Isoniazid treatment of children: can genetics help guide treatment?

N Cranswick, K Mulholland

Isoniazid pharmacokinetics in children treated for respiratory tuberculosis

H S Schaaf, D P Parkin, H I Seifart, C J Werely, P B Hesseling, P D van Helden, J S Maritz, P R Donald
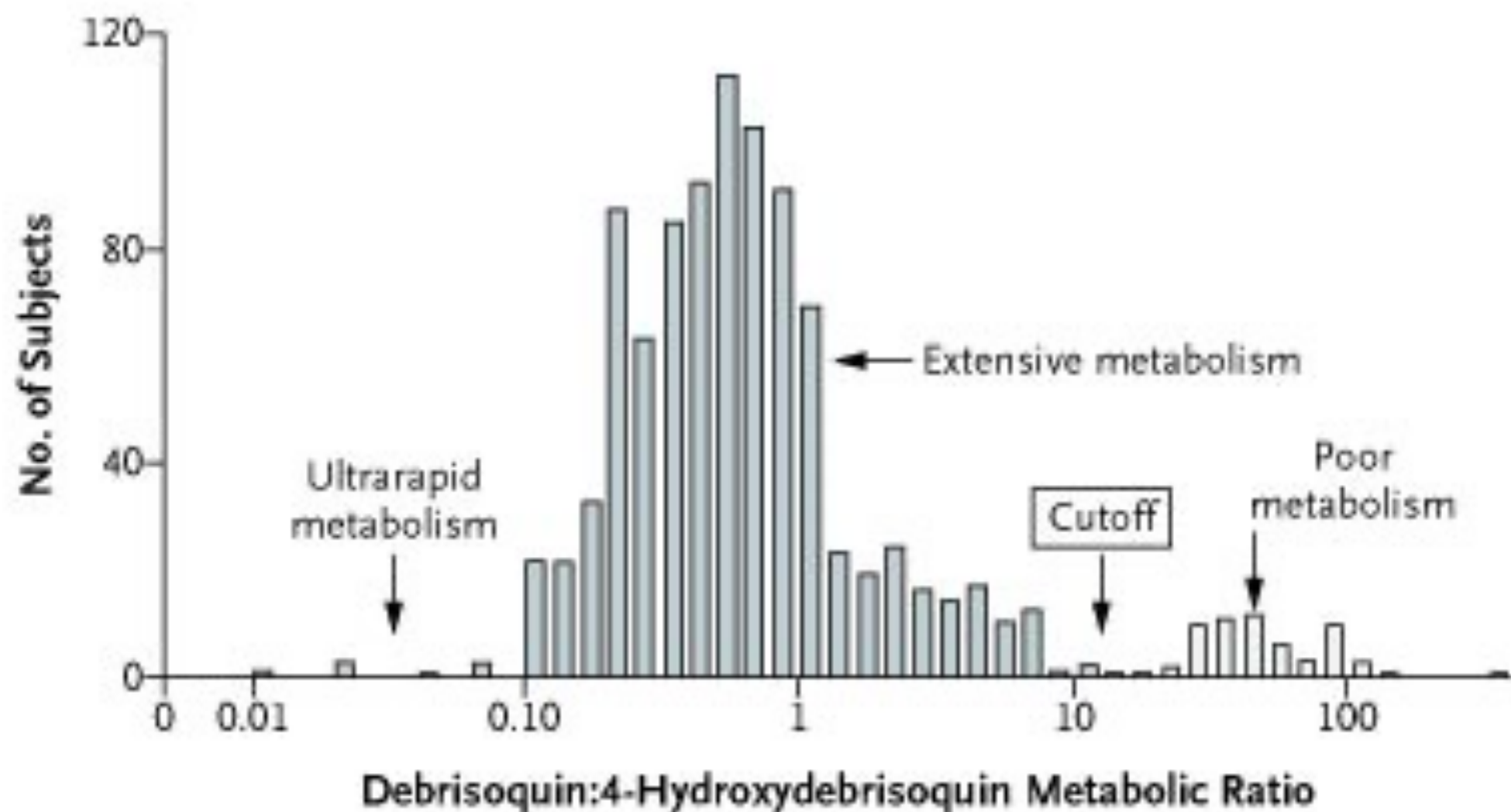

10 mg/kg

Important proportion of FF patients are under recommended concentrations

Recommendation for FF detection and dose adaptation.

Emerging countries?

Cytochrome P450 2D6 (CYP2D6)

Codeine, nortryptyline, ...

No. of Subjects

120

80

40

0

Ultrarapid metabolism

Extensive metabolism

Poor metabolism

Cutoff

0   0.01   0.10   1   10   100

Debrisoquin:4-Hydroxydebrisoquin Metabolic Ratio
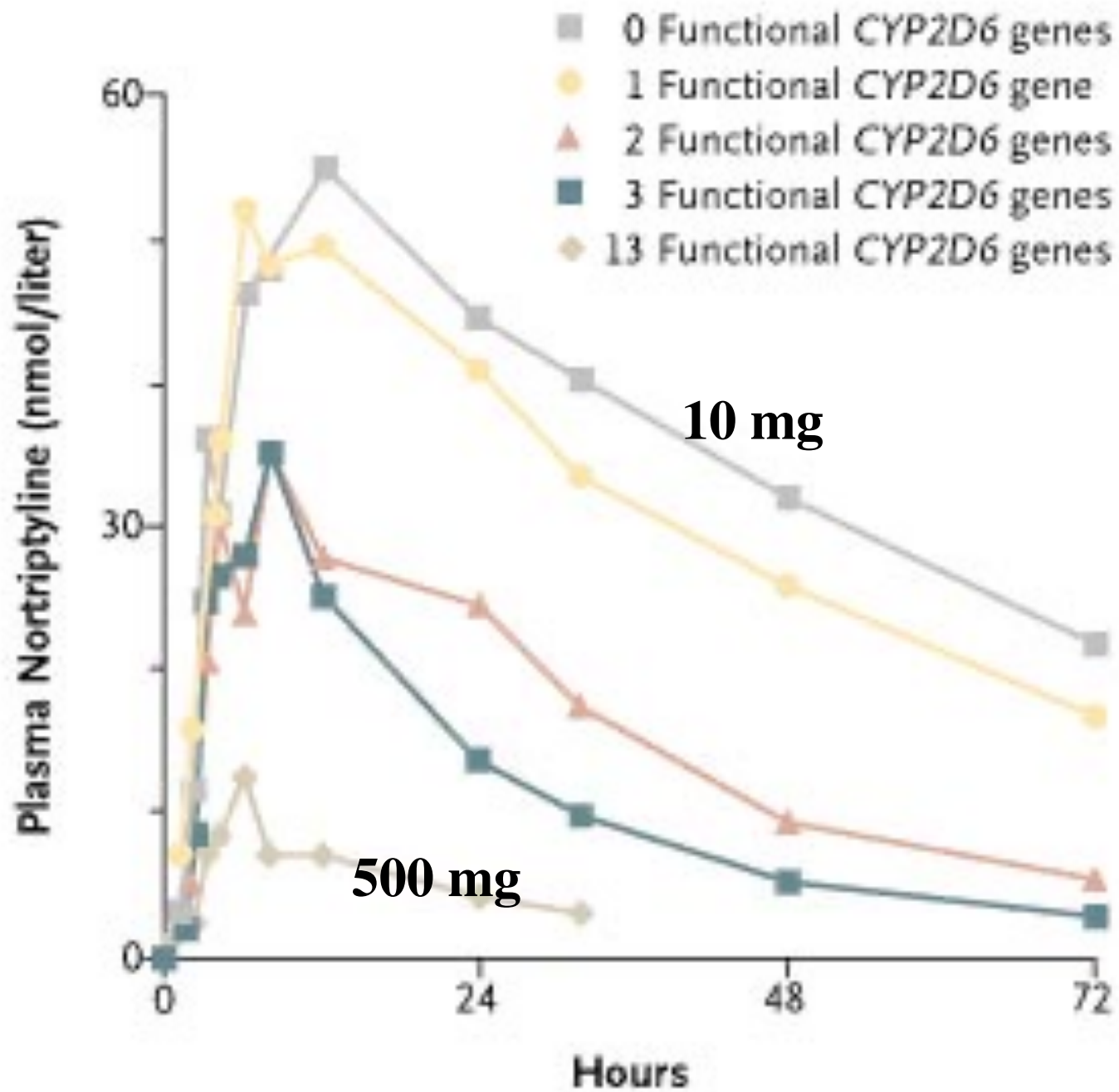
Cytochrome P450 2D6 (CYP2D6)
Codeine, nortryptyline, ...

5-10% of caucasians:
deficit of Cyt P450 2D6

> 75 alleles
Slow metabolisers: nortryptiline side effects, no response to codeine

Very-fast metabolisers
Multiples gene copies (0-13)
East Africa: 29%

Legend:
- 0 Functional *CYP2D6* genes
- 1 Functional *CYP2D6* gene
- 2 Functional *CYP2D6* genes
- 3 Functional *CYP2D6* genes
- 13 Functional *CYP2D6* genes

X-axis: Hours (0, 24, 48, 72)
Y-axis: Plasma Nortriptyline (nmol/liter) (0, 30, 60)

10 mg

500 mg

Cytochrome P450 2D6 (CYP2D6)
Codeine

Activated in morphine

Slow metabolisers: no response
Fast metabolisers: morphine overdose

FDA: No Codeine After Tonsillectomy for Children.

A review of cases reported to the FDA's Adverse Event Reporting System between 1969 and May 2012 identified 10 deaths and 3 overdoses in children who had been treated with codeine; 7 of the cases were also reported in the medical literature. Of the 13 cases, 8 occurred in children after adenotonsillectomy

Some of the affected children may have been rapid metabolizers of codeine, according to the FDA's warning. All humans convert codeine into morphine, but individuals who have certain genetic variants encoding the enzyme cytochrome P450 2D6 do so more rapidly

FDA Warns of Rare Morphine Overdose in Breastfed Babies

Nursing mothers with a genetic predisposition for rapidly metabolizing codeine in pain medication can seriously overdose their babies with morphine.

Safety of codeine during breastfeeding

Fatal morphine poisoning in the breastfed neonate of a mother prescribed codeine

Parvaz Madadi, Gideon Koren, MD, FRCPC, [...], and Katarina Aleksa,

Abstract

QUESTION Recently a newborn died from morphine poisoning when his mother used codeine while breastfeeding. Many patients receive codeine for postlabour pain. Is it safe to prescribe codeine for nursing mothers?

ANSWER When a mother is an ultrarapid metabolizer of cytochrome P450 2D6, she produces much more morphine when taking codeine than most people do. In this situation, newborns might be exposed to toxic levels of morphine when breastfeeding.

Other Cytochrome P450 isoforms

2C9

2C19

3A5

...

Estimation of the Warfarin Dose with Clinical and Pharmacogenetic Data

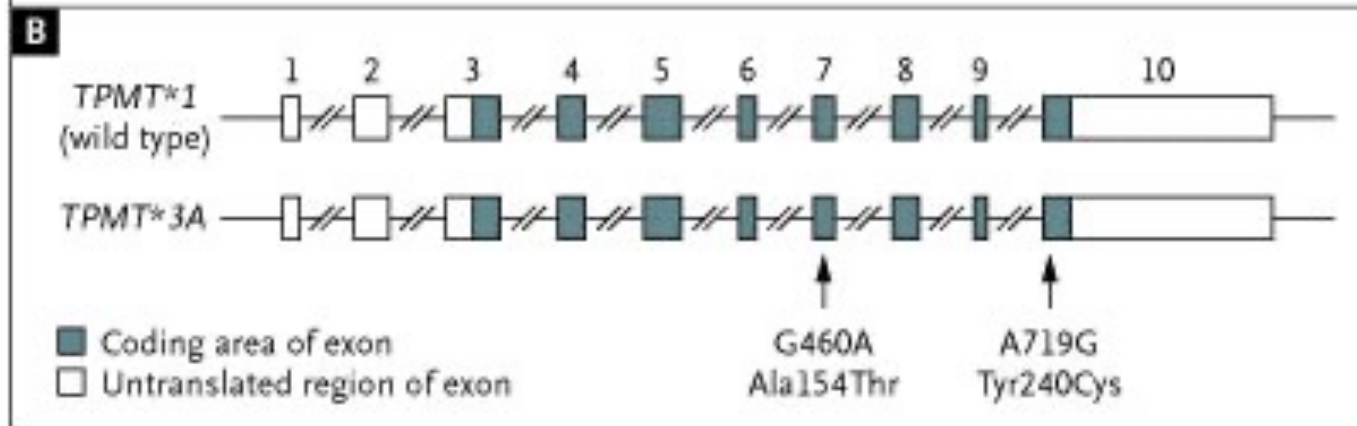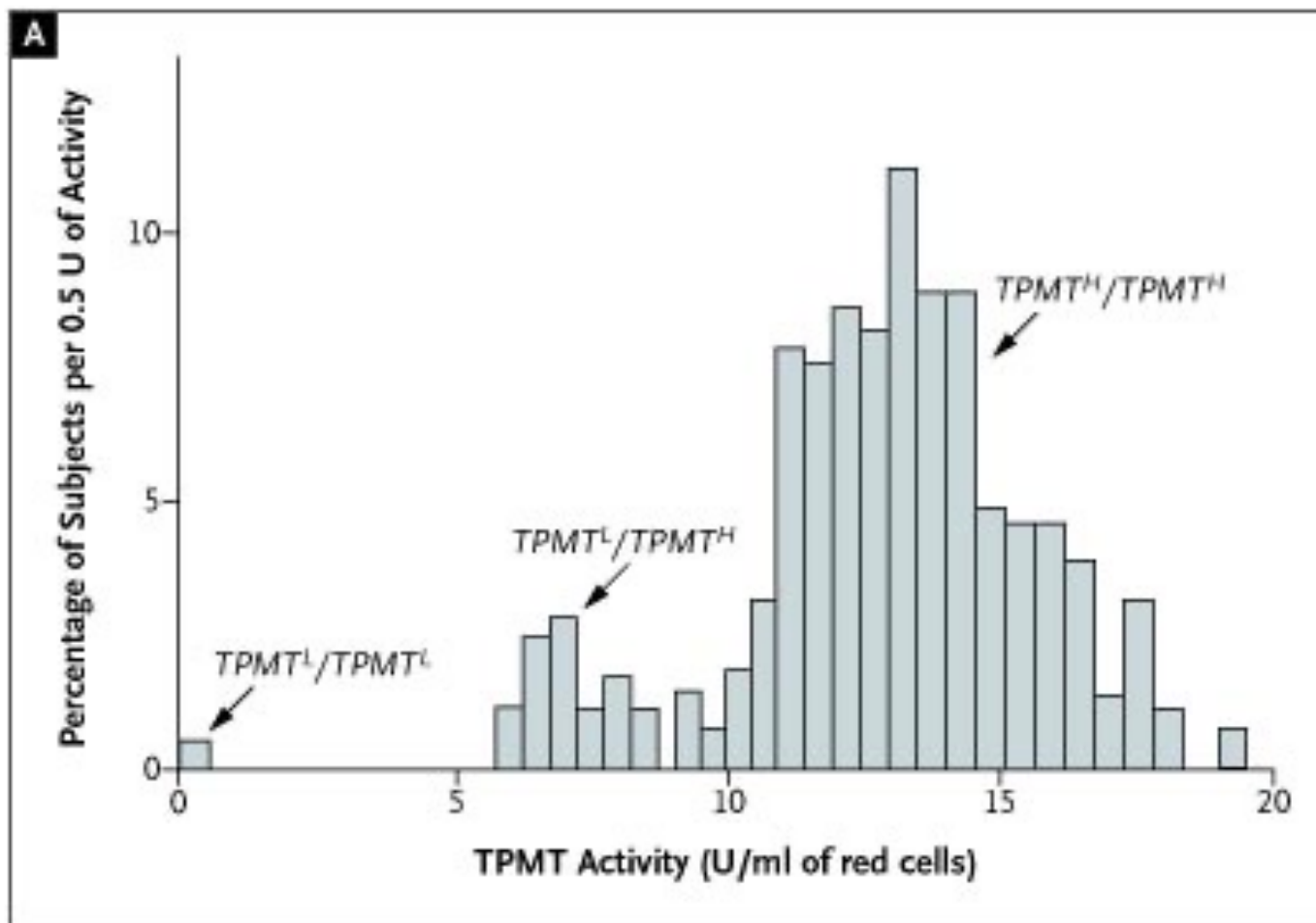The International Warfarin Pharmacogenetics Consortium*

Génotyping of the cyt P450 2C9 (3 alleles) and the VKORC1 gene (1 SNP) allow a good prediction of the appropriate warfarin dose (>4000 pts)

The FDA recommended (2007) to include information on pharmacogenetics in the drug sheet and encouraged to use pharmacogenetics to define the treatment

Clinical utility ? Reduction of side effects?

Thiopurine S-methyltransferase (TPMT)
Mercaptopurine, Azathioprine

A

Percentage of Subjects per 0.5 U of Activity

TPMT^L/TPMT^L

TPMT^L/TPMT^H

TPMT^H/TPMT^H

TPMT Activity (U/ml of red cells)

B

TPMT*1 (wild type)

1 2 3 4 5 6 7 8 9 10

TPMT*3A

G460A
Ala154Thr

A719G
Tyr240Cys

■ Coding area of exon
□ Untranslated region of exon

Thiopurine S-methyltransferase (TPMT)
Mercaptopurine, Azathioprine

Weak metabolisers: myelosuppression after regular doses

Clinically relevant test

Ethnic differences

Table 1. Pharmacogenetics of Phase I Drug Metabolism.*

Drug-Metabolizing Enzyme	Frequency of Variant Poor-Metabolism Phenotype	Representative Drugs Metabolized	Effect of Polymorphism
Cytochrome P-450 2D6 (CYP2D6)	6.8% in Sweden 1% in China[17]	Debrisoquin[15] Sparteine[16] Nortriptyline[23] Codeine[27,28]	Enhanced drug effect Enhanced drug effect Enhanced drug effect Decreased drug effect
Cytochrome P-450 2C9 (CYP2C9)	Approximately 3% in England[29] (those homozygous for the *2 and *3 alleles)	Warfarin[29,30] Phenytoin[31,32]	Enhanced drug effect[29-32]
Cytochrome P-450 2C19 (CYP2C19)	2.7% among white Americans[33] 3.3% in Sweden 14.6% in China[17] 18% in Japan[33]	Omeprazole[34,35]	Enhanced drug effect[36,37]
Dihydropyrimidine dehydrogenase	Approximately 1% of population is heterozygous[38]	Fluorouracil[39,40]	Enhanced drug effect[39,40]
Butyrylcholinesterase (pseudocholinesterase)	Approximately 1 in 3500 Europeans[41]	Succinylcholine[9,41]	Enhanced drug effect[9,41]

* Examples of genetically polymorphic phase I enzymes are listed that catalyze drug metabolism, including selected examples of drugs that have clinically relevant variations in their effects.

Phase 1 reactions: oxidation, reduction, hydrolysis ...

Table 2. Pharmacogenetics of Phase II Drug Metabolism.[*]

Drug-Metabolizing Enzyme	Frequency of Variant Poor-Metabolism Phenotype	Representative Drugs Metabolized	Effect of Polymorphism
N-Acetyltransferase 2	52% among white Americans[10] 17% of Japanese[58]	Isoniazid[10] Hydralazine[11] Procainamide[12]	Enhanced drug effect[13]
Uridine diphosphate–glucuronosyltransferase 1A1 (TATA-box polymorphism)	10.9% among whites[59] 4% of Chinese[60] 1% of Japanese[60]	Irinotecan[61] Bilirubin[62]	Enhanced drug effect[63] Gilbert's syndrome[62]
Thiopurine S-methyltransferase	Approximately 1 in 300 whites[50,57] Approximately 1 in 2500 Asians[57]	Mercaptopurine[51] Azathioprine	Enhanced drug effect (toxicity)[51-53]
Catechol O-methyltransferase	Approximately 25% of whites[51,64]	Levodopa[51,65]	Enhanced drug effect[51,65]

[*] Examples of genetically polymorphic phase II (conjugating) enzymes are listed that catalyze drug metabolism, including selected examples of drugs that have clinically relevant variations in their effects.

Phase 2 reactions: conjugation (acetylation, methylation, glucoronidation, ...

Pharmacogenetics

1. Drug metabolism

- Phase I: CYP, ...
- Phase II: TPMT, NAT2, GST, ...
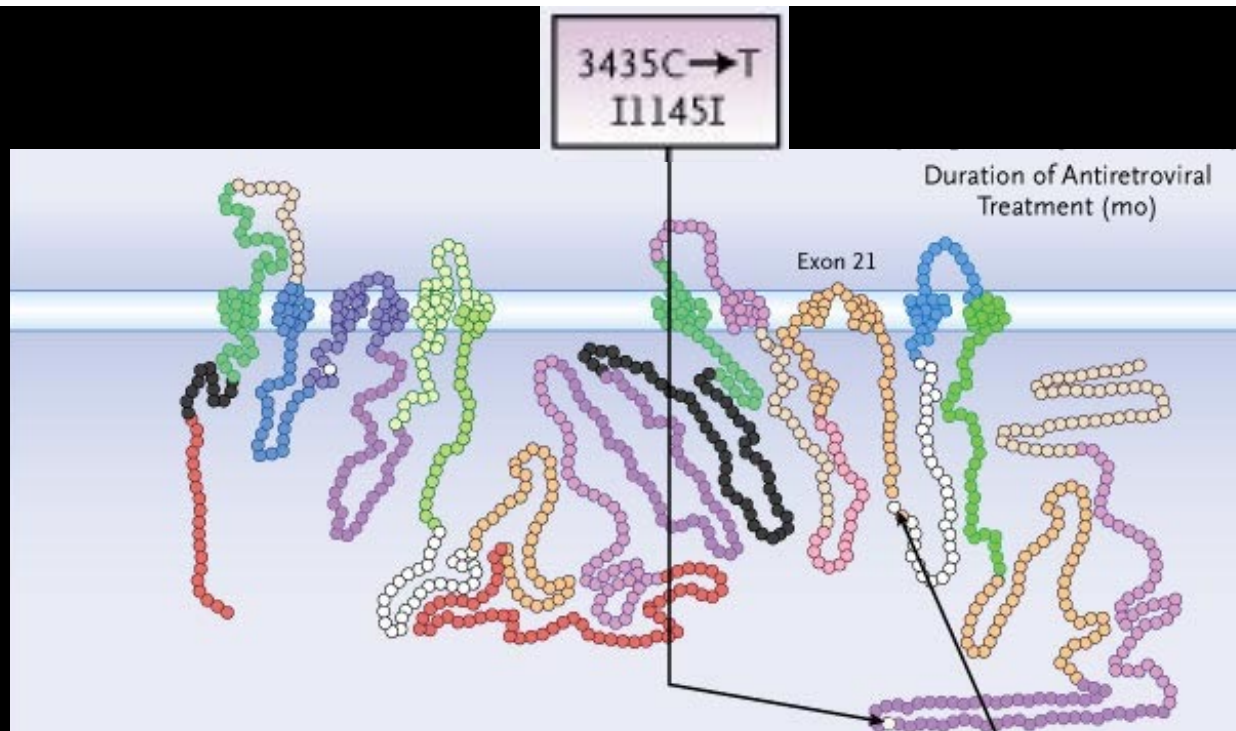
2. Transport

- MDR (ABC)

Drug Transporters

ATP-binding cassette
P-glycoprotéine MDR1 ABCB1

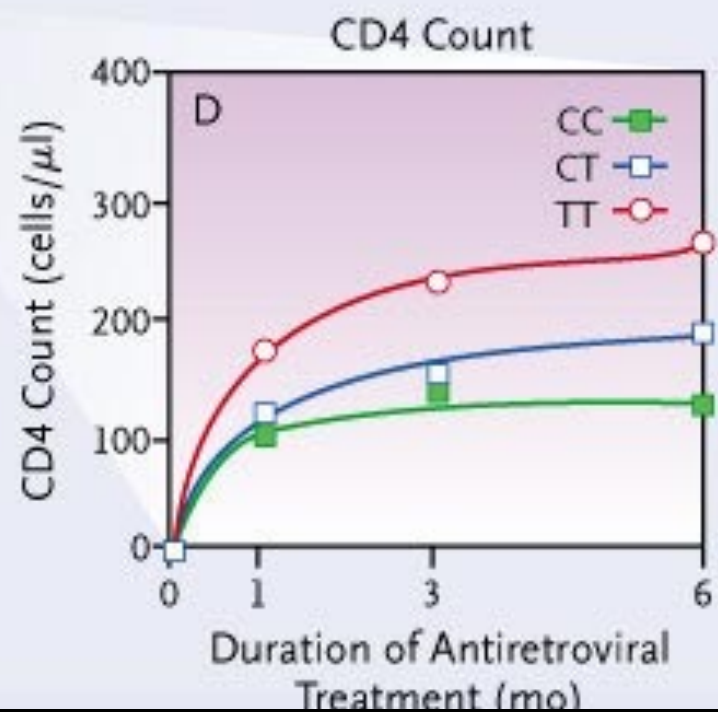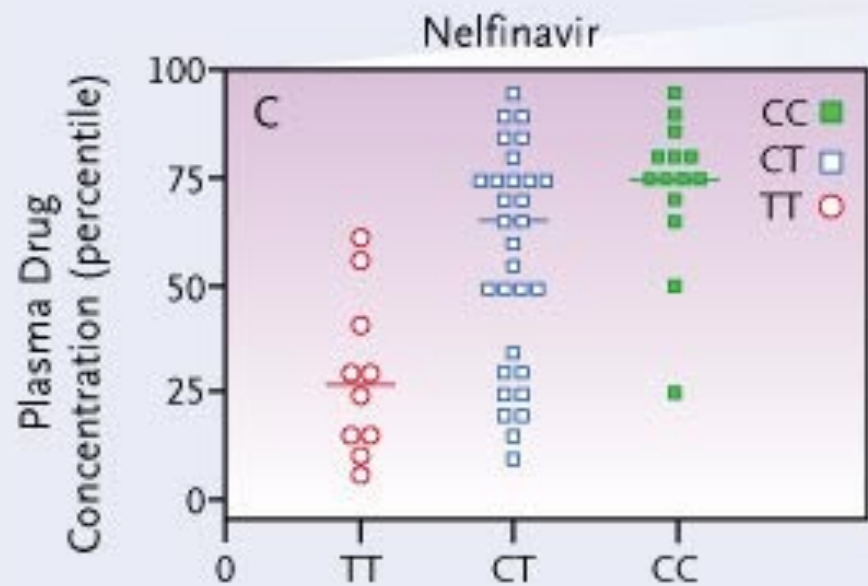**Drug efflux, blood-brain barrier, urine or bile excretion of xenobiotics**

Variable expression of P-glycoprotein

TT lower expression
CC higher expression, lower cell drug retention

Linked with another polymorphism (missense)

3435C→T
I1145I

C — Nelfinavir

Plasma Drug Concentration (percentile)

CC ■
CT □
TT ○

TT   CT   CC

D — CD4 Count

CD4 Count (cells/μl)

CC ■
CT □
TT ○

Duration of Antiretroviral Treatment (mo)

Pharmacogenetics

1. Drug metabolism

- Phase I: CYP, ...
- Phase II: TPMT, NAT2, GST, ...
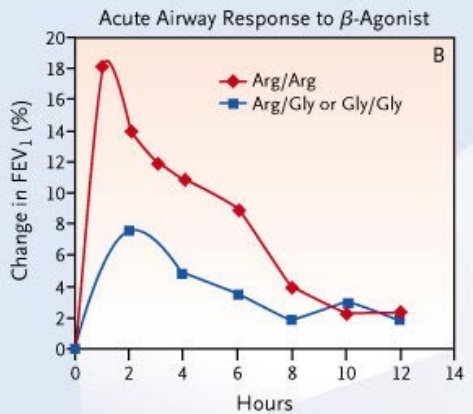
2. Transport

- MDR (ABC)

3. Targets

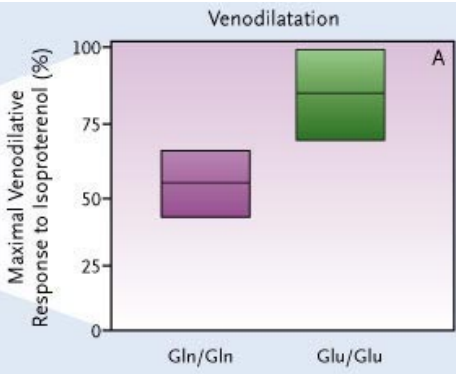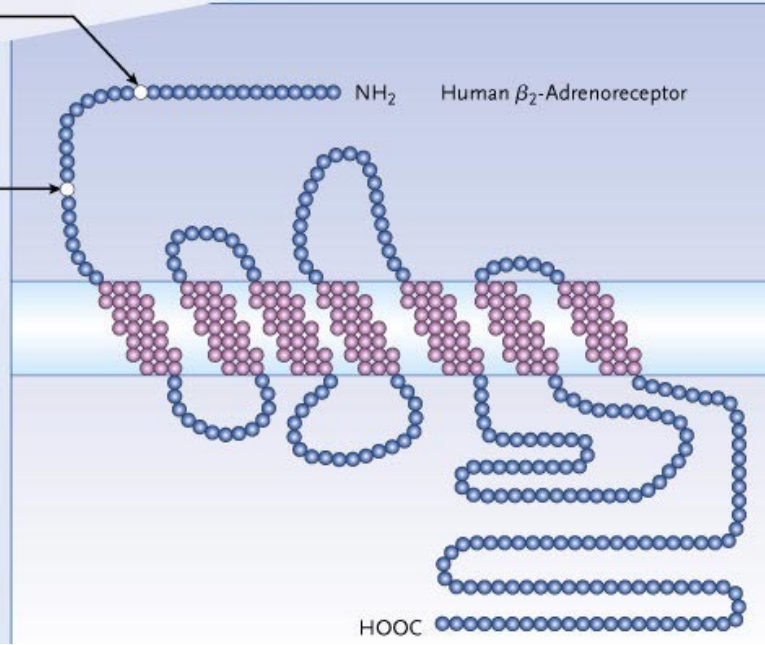- Beta-adrenergic receptor

Récepteurs

Récepteur β2-Adrénergique

Frequent SNPs affecting signal transduction

Venodilatation

Acute Airway Response to β-Agonist

Desensitization

Gln or Glu at codon 27
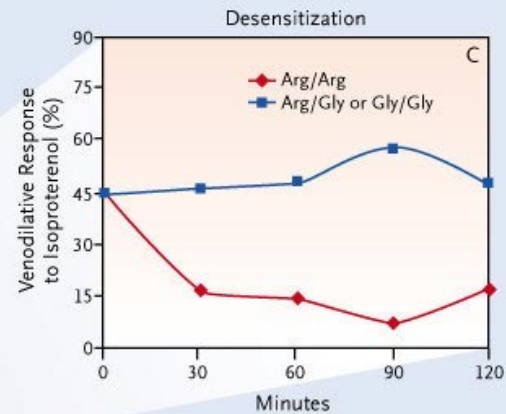
Arg or Gly at codon 16

Arg/Arg at codon 16
Loss of activity after repeated administrations

β2-Adrenergic Receptor

At least 13 SNPs

12 haplotypes

Correlation clinical response/haplotype

Table 1. Genetic Polymorphisms in Drug Target Genes That Can Influence Drug Response.*

Gene or Gene Product	Medication	Drug Effect Associated with Polymorphism
ACE	ACE inhibitors (e.g., enalapril)	Renoprotective effects, blood-pressure reduction, reduction in left ventricular mass, endothelial function[32-40]
	Fluvastatin	Lipid changes (e.g., reductions in low-density lipoprotein cholesterol and apolipoprotein B); progression or regression of coronary atherosclerosis[41]
Arachidonate 5-lipoxygenase	Leukotriene inhibitors	Improvement in FEV_1[42]
β_2-Adrenergic receptor	β_2-Agonists (e.g., albuterol)	Bronchodilatation, susceptibility to agonist-induced desensitization, cardiovascular effects[43-50]
Bradykinin B2 receptor	ACE inhibitors	ACE-inhibitor–induced cough[51]
Dopamine receptors (D2, D3, D4)	Antipsychotics (e.g. haloperidol, clozapine)	Antipsychotic response (D2, D3, D4), antipsychotic-induced tardive dyskinesia (D3), antipsychotic-induced acute akathisia (D3)[52-56]
Estrogen receptor-α	Conjugated estrogens	Increase in bone mineral density[57]
	Hormone-replacement therapy	Increase in high-density lipoprotein cholesterol[58]
Glycoprotein IIIa subunit of glycoprotein IIb/IIIa	Aspirin or glycoprotein IIb/IIIa inhibitors	Antiplatelet effect[59]
Serotonin (5-hydroxytryptamine) transporter	Antidepressants (e.g., clomipramine, fluoxetine, paroxetine)	5-Hydroxytryptamine neurotransmission, antidepressant response[60-62]

Pharmacogenetics

1. Drug metabolism
- Phase I: CYP, ...
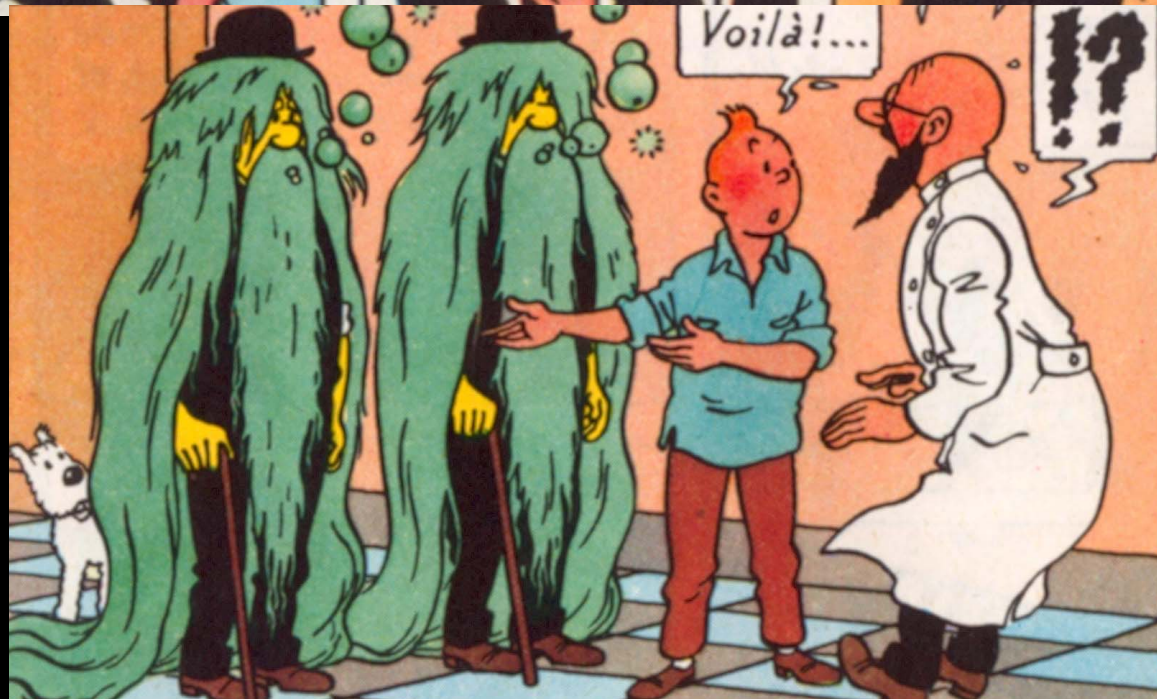- Phase II: TPMT, NAT2, GST, ...

2. Transport
- MDR (ABC)

3. Targets
- Beta-adrenergic receptor

4. Unexpected side effects
- Long QT
- Deafness and aminoglycosids, ...

Deafness induced by aminoglycosids

Polymorphisms in the mitochondrial 12S rRNA

Targeted by the antibiotics

Hypersensitivity reactions

Carbamazepin:
- HLA B*15:02: Asia, standard practice in Taiwan before carbamazepin prescription
-HLA A*31:01: Europe

Abacavir: HLA-B*5701

Table 2. Genetic Polymorphisms in Disease-Modifying or Treatment-Modifying Genes That Can Influence Drug Response.*

Gene or Gene Product	Disease or Response Association	Medication	Influence of Polymorphism on Drug Effect or Toxicity
Adducin	Hypertension	Diuretics	Myocardial infarction or strokes[69]
Apolipoprotein E (APOE)	Progression of atherosclerosis, is-chemic cardiovascular events	Statins (e.g., simvastatin)	Enhanced survival[70,71]
Apolipoprotein E (APOE)	Alzheimer's disease	Tacrine	Clinical improvement[72]
HLA	Toxicity	Abacavir	Hypersensitivity reaction[73,74]
Cholesterol ester transfer protein (CETP)	Progression of atherosclerosis	Statins (e.g., pravastatin)	Slowing of progression of atherosclerosis by pravastatin[75]
Ion channels (HERG, KvLQT1, Mink, MiRP1)	Congenital long-QT syndrome	Erythromycin, terfenadine, cisa-pride, clarithromycin, quinidine	Increased risk of drug-induced torsade de pointes[76-78]
Methylguanine methyl-transferase (MGMT)	Glioma	Carmustine	Response of glioma to carmustine[63]
Parkin	Parkinson's disease	Levodopa	Clinical improvement and levodopa-induced dyskinesias[79]
Prothrombin and factor V	Deep-vein thrombosis and cerebral-vein thrombosis	Oral contraceptives	Increased risk of deep-vein and cerebral-vein thrombosis with oral contraceptives[80]
Stromelysin-1	Atherosclerosis progression	Statins (e.g., pravastatin)	Reduction in cardiovascular events by prava-statin (death, myocardial infarction, stroke, angina, and others); reduction in risk of repeated angioplasty[81]

Recommended tests

DPYD (5-Fu)
TPMT
UGT1A1 (irinotecan)

HLA-B*57:01 (abacavir)

...

Website Stanford University

http://www.pharmgkb.org

Reviews the clinical utility of pharmogenetic tests

http://www.pharmgkb.org/search/clinicalAnnotationList.action?levelOfEvidence=top

CPIC

Clinical pharmacogenetics Implementation Consortium

https://cpicpgx.org

Recommended tests

DPYD (5-Fu)
TPMT
UGT1A1 (irinotecan)

HLA-B*57:01 (abacavir)

…

DPYD (5-Fu)

French recommendations (2018)

Based on the levels of evidence from the literature data and considering current French practices, the Group of Clinical Pharmacology in Oncology (GPCO)-UNICANCER and the French Network of Pharmacogenetics (RNPGx) recommend the following: (1) to screen DPD deficiency before initiating any chemotherapy containing 5-FU or capecitabine; (2) to perform DPD phenotyping by measuring plasma uracil (U) concentrations (possibly associated with dihydrouracil/U ratio), and DPYD genotyping (variants *2A, *13, p.D949V, HapB3); (3) to reduce the initial FU dose (first cycle) according to DPD status, if needed, and further, to consider increasing the dose at subsequent cycles according to treatment tolerance.

DPYD (5-Fu)

Common mutations with enzyme low activity
Do not identify all patients with DPYD deficiency

Dosage of the ratio dihydrouracil/uracil in the plasma : better sensitivity

DPYD (5-Fu)

EMA recommendations (March 2020)

https://www.esmo.org/oncology-news/ema-provides-new-testing-and-treatment-recommendations-for-fluorouracil-capecitabine-and-tegafur

DPYD (5-Fu)

EMA recommendations (March 2020)

EMA PROVIDES NEW TESTING AND TREATMENT RECOMMENDATIONS FOR FLUOROURACIL CAPECITABINE AND TEGAFUR

Patients should be tested for DPD deficiency before starting treatment

Date:20 Mar 2020
Topics:Anticancer agents & Biologic therapy

DPYD (5-Fu)

Joint Belgian Recommendation on Screening for DPD-deficiency in patients treated with 5-FU, capecitabine (and tegafur)

we recommend phenotype or targeted genotype testing for DPD deficiency before starting 5-FU, capecitabine or tegafur. We strongly suggest a stepwise approach using phenotype testing upfront because of the higher sensitivity and the lower cost for the society.
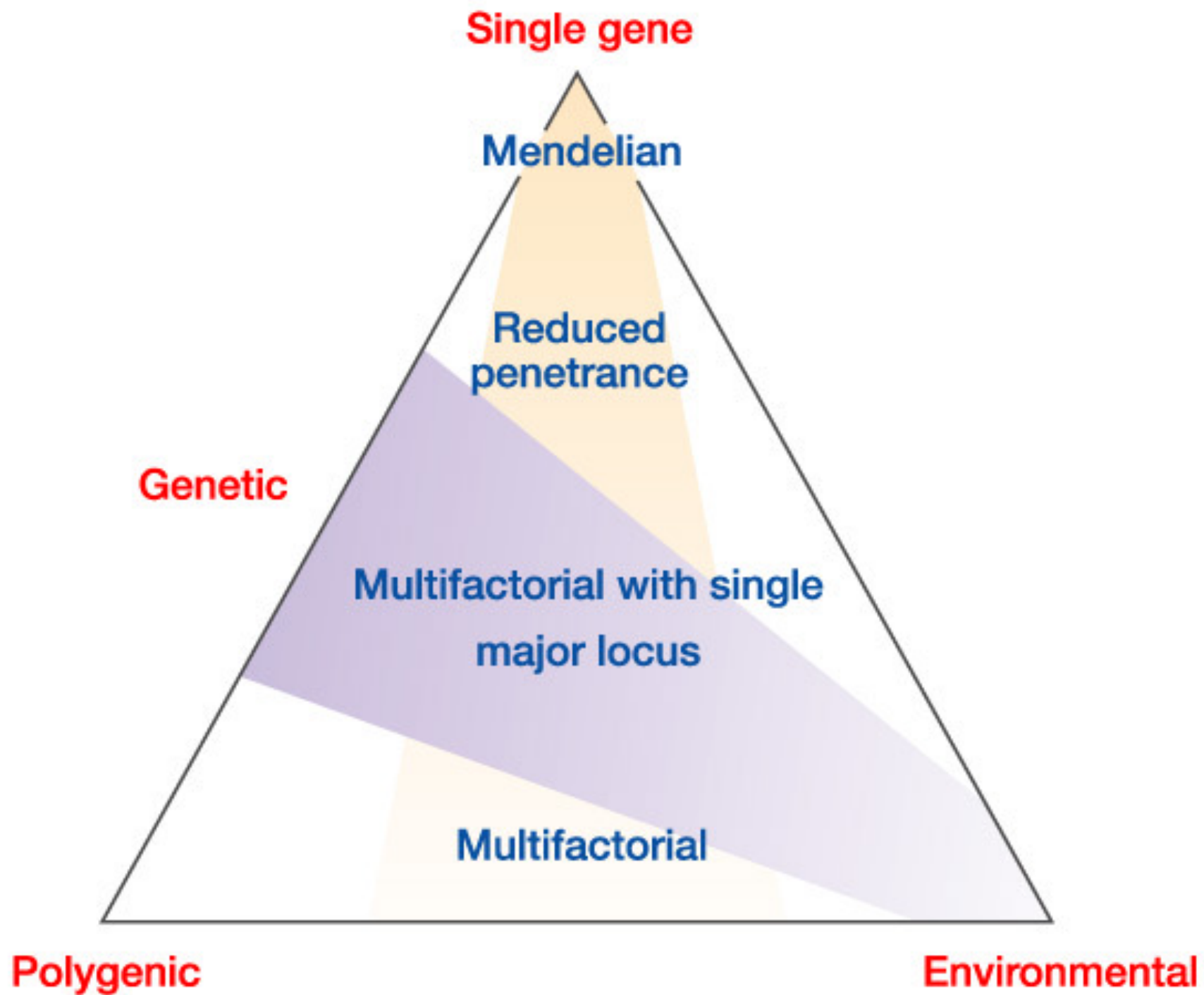
CANCER

1. « Host » pharmacogenetics

- TPMT and mercaptopurine.
- Dihydropyrimidine dehydrogenase and 5-fluoruracile
- UGT1A1

2. « Tumour » pharmacogenetics / theranostics

- HER2 amplification and Herceptin
- Ras mutation and resistance to anti-EGFR antibodies
- Bcr-Abl fusion gene and response to glivec
- MGMT methylation and response to alkylating agents , ….

= target identification or acquired resistance

CANCER

1. « Host » pharmacogenetics

+

2. « Tumour » pharmacogenetics

BRCA mutations linked with clinical response to platine agents and PARP inhibitors

Clinical relevance

Goal: to predict a clinical response, to prevent side effects, to adapt the doses. Tailored treatment.

BUT very few pharmacogenetic tests have shown a real clinical relevance (TPMT, warfarin).

Often, multiple genes are involved.

Political/economic relevance

Goal: to save money (avoiding useless treatments or important side effects).

August 2006

« Genomics and personalized medicine »

Access to genetic tests in order to allow a personnalized medicine for all the Americans.

To better target medical cares.

Barack Obama

Companies

Goal: to save money

Defining the target population
Reducing the size of clinical trials
Reducing and predicting side effects

Current research and future directions

Include pharmacogenetic/pharmacogenomic tests in clinical trials.

Consider ethnical differences.

Perspectives for a large screening of PG variants in every individual ??

Pharmacogenetics in emerging countries

Impact of pharmacogenomics on neglected diseases in the developing world
T Pang, Am J Pharmacogenomics, 2003

Important impact on treatment of tuberculosis, malaria and HIV

Response rates different than in Occident

Development of therapeutics more adapted to African patients??

Pharmacogenetics in emerging countries

Pharmacogenetics and rational drug use around the world.
Roederer et al., Pharmacogenomics, 2011

Pharmacogenetics in emerging countries

http://apps.who.int/iris/bitstream/handle/10665/43669/9789241595469_eng.pdf?sequence=1&isAllowed=y

Pharmacogenetics: new developments

Genotype-based treatment of genetic diseases

Cystic fibrosis

CFTR modulators
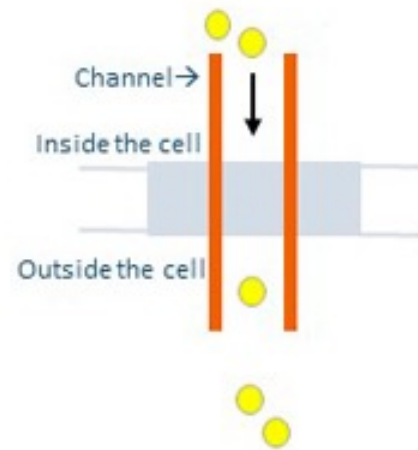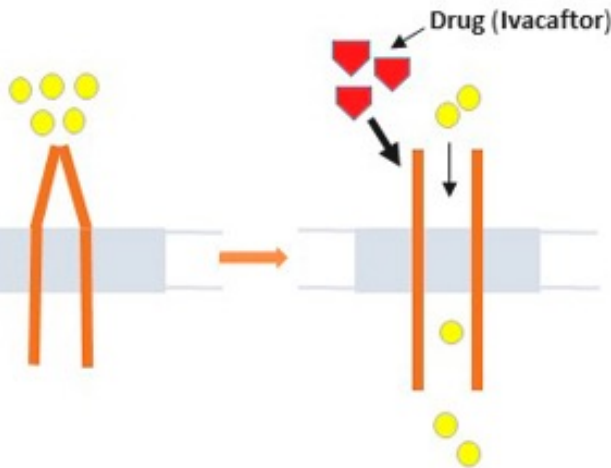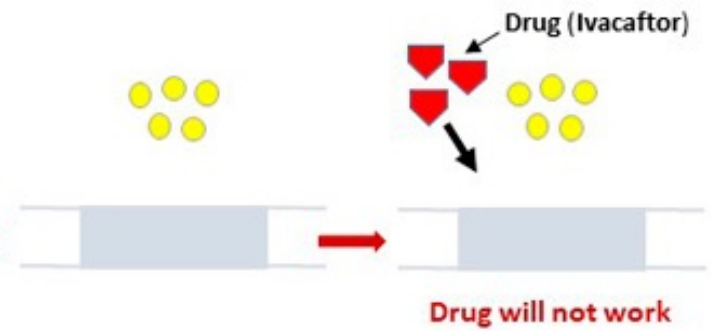
CF: genotype – treatment correlation

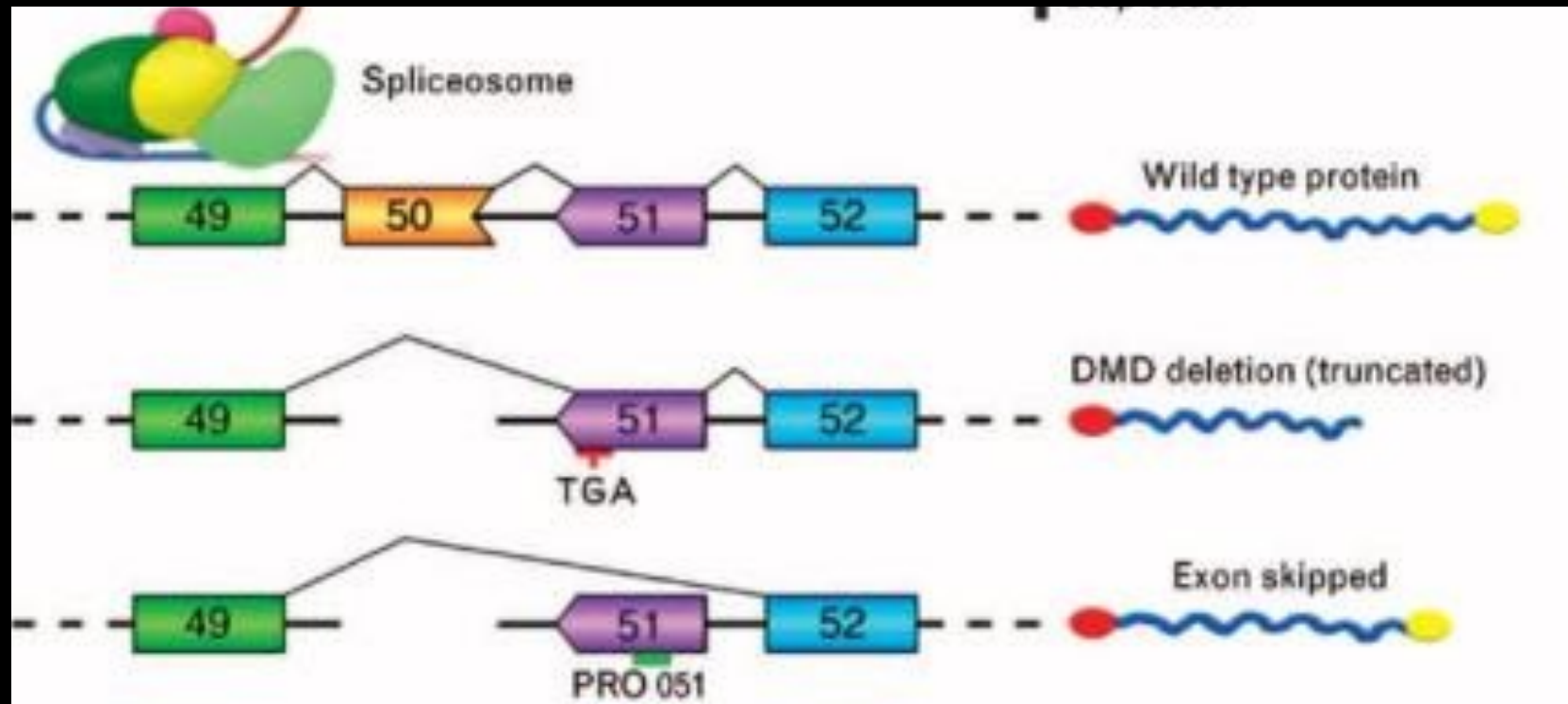| Mutation class	Defect	Phenotype	Example	Treatment strategy
I	Reduced CFTR protein expression	No protein	Gly542X Trp1282X	Production correctors (ataluren)
II	Misfolded CFTR protein not transported to the cell surface	No traffic	Phe508del (ΔF508) Asn1303Lys Ala561Glu	Corrector + potentiator (lumacaftor + ivacaftor, VX-661+ ivacaftor)
III	Reduced/lack of CFTR channel opening	Impaired gating	Gly551Asp Ser549Arg Gly1349Asp	Potentiator (ivacaftor)
IV	Misshaped CFTR pore restricts Cl⁻ movement	Decreased conductance	Arg117His Arg334Trp Ala455Glu	Potentiator (ivacaftor)
V	Reduced CFTR protein production	Less protein	3849+10 kb C→T Ala455Glu 3272-26A → G	No data available
VI	High CFTR protein turnover at the cell surface	Less stable	120del23 rPhe508del	No data available
VII	No transcription due to large deletions on *CFTR* gene	No mRNA	dele2,3 (21kb) 1717-1G →A	Unrescuable (By pass therapies?)

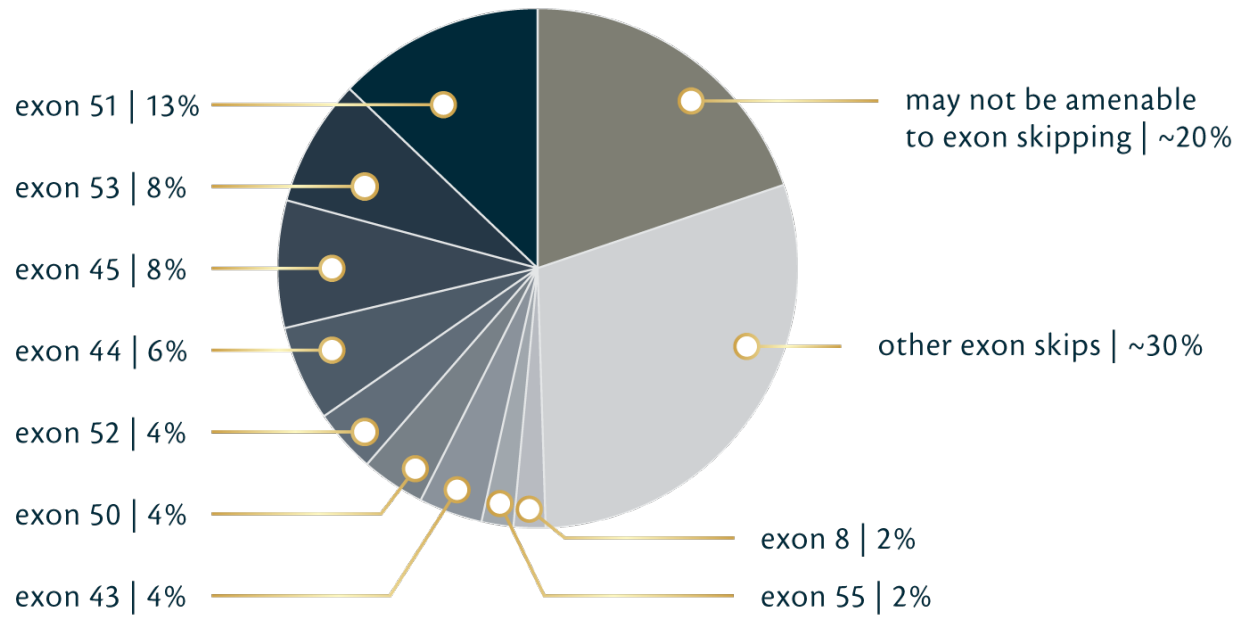Kb: Kilobases, CFTR: Cystic fibrosis transmembrane conductance regulator

Duchenne muscular dystrophy

Exon skipping

Correct splicing removes 3 introns by pairwise recognition of the junctions

exon 51 | 13%

exon 53 | 8%

exon 45 | 8%

exon 44 | 6%

exon 52 | 4%

exon 50 | 4%

exon 43 | 4%

may not be amenable
to exon skipping | ~20%
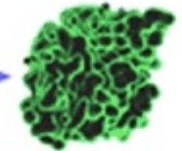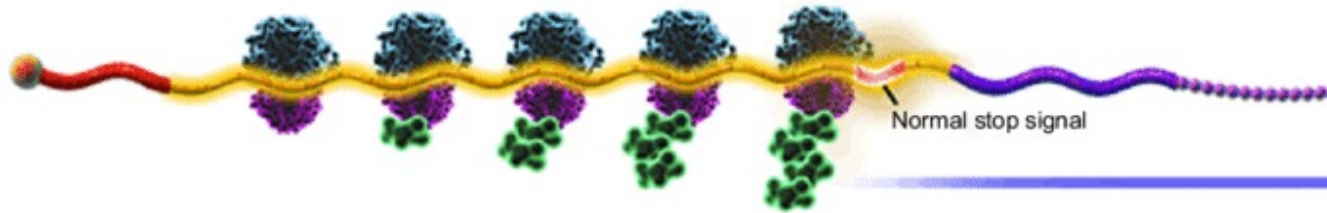
other exon skips | ~30%

exon 8 | 2%

exon 55 | 2%
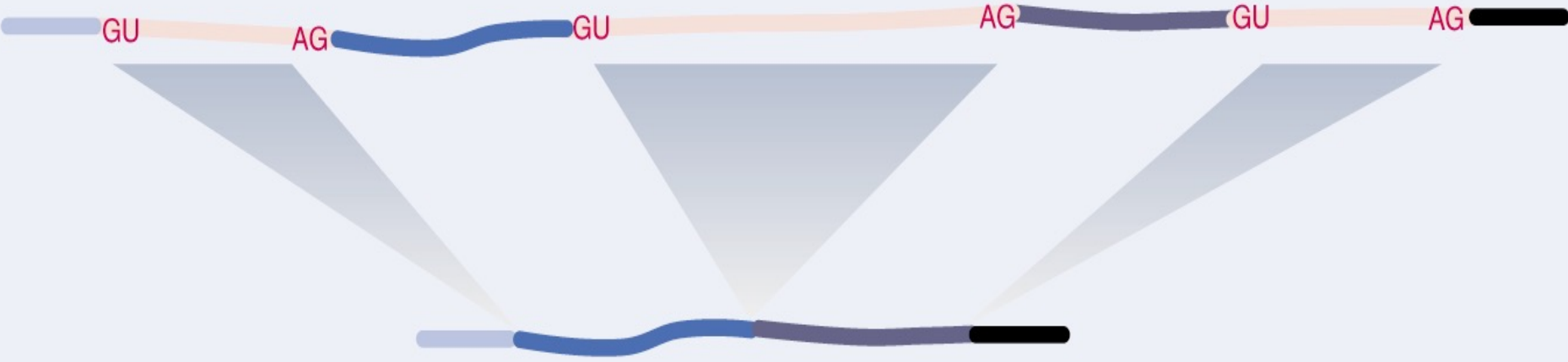
Approximately 13% of DMD
patients may be amenable
to exon 51 skipping.

Available data suggest up
to 80% of DMD patients
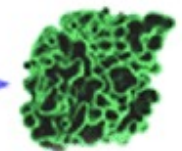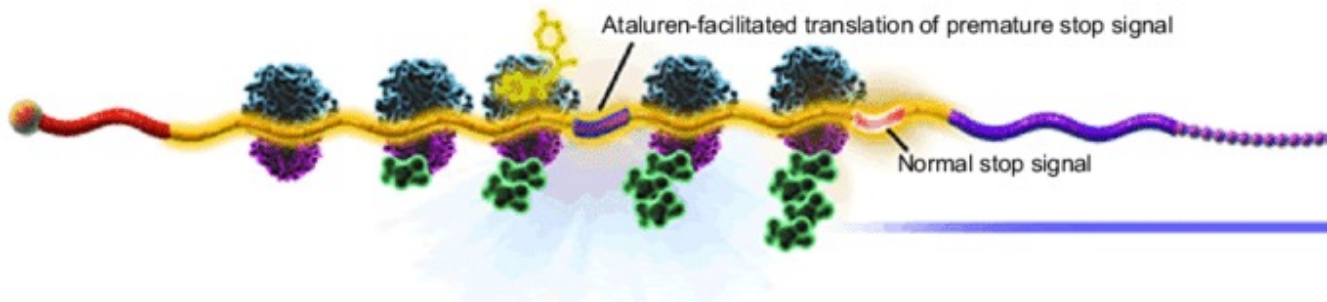have genotypes amenable
to exon skipping.

Normal control

Normal stop signal

Functional dystrophin

DMD with stop codon mutation

Premature stop signal   Normal stop signal
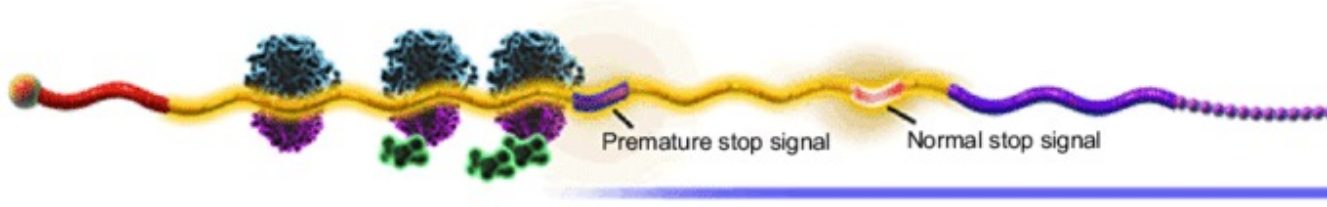
Truncated dystrophin

Ataluren-translated dystrophin

Ataluren-facilitated translation of premature stop signal

Normal stop signal

Functional truncated dystrophin

Pharmacogenetics

A few clinically actionable tests

To be done « on demand » or »prospectively »?

Ethnic variations in allele frequencies

Pharmacogenetics

Novel mutation-specific treatments for genetic diseases