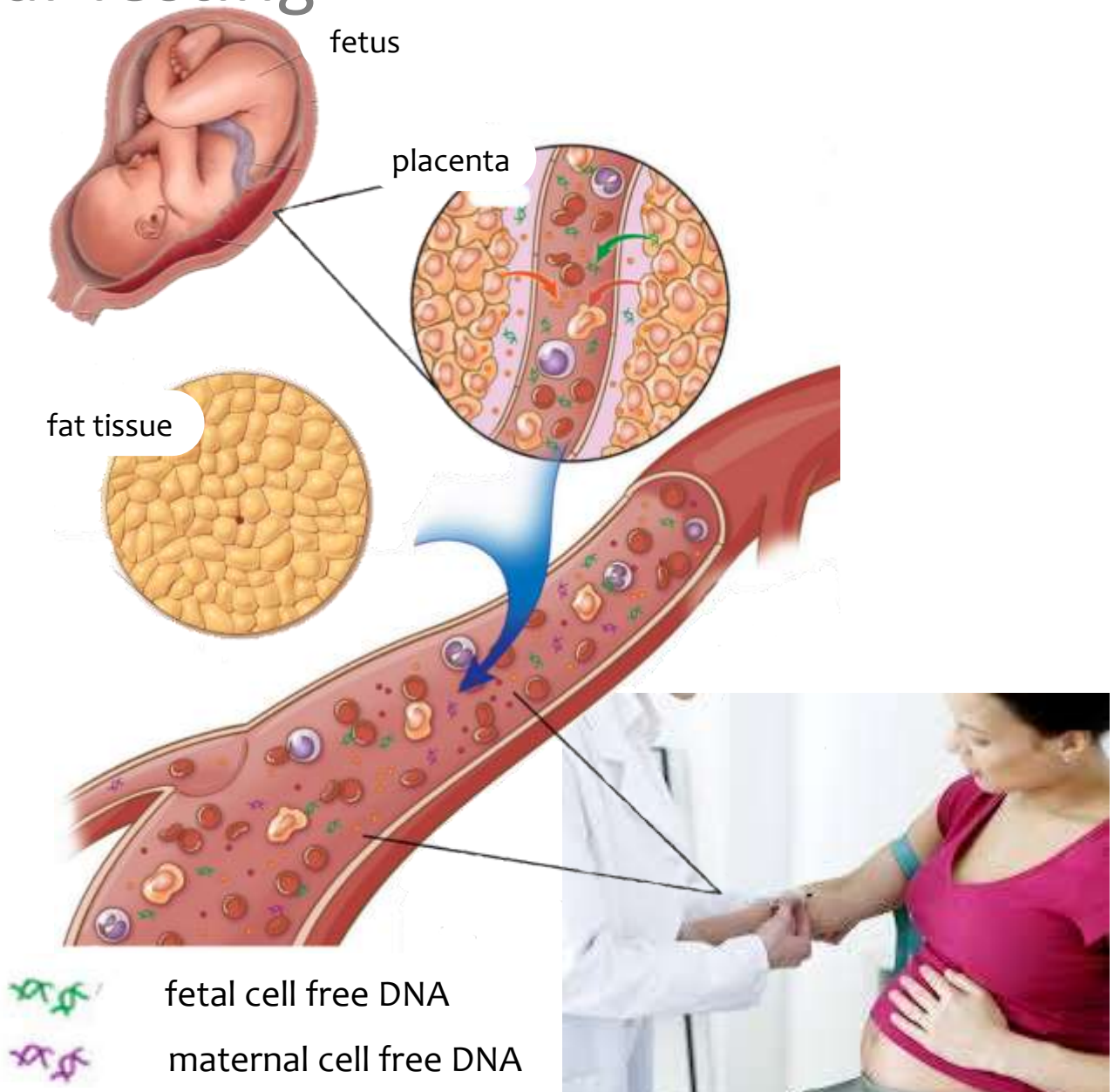
New developments in Prenatal Genetic Testing: towards comprehensive genome screening?

CENTRUM MEDISCHE GENETICA GENT

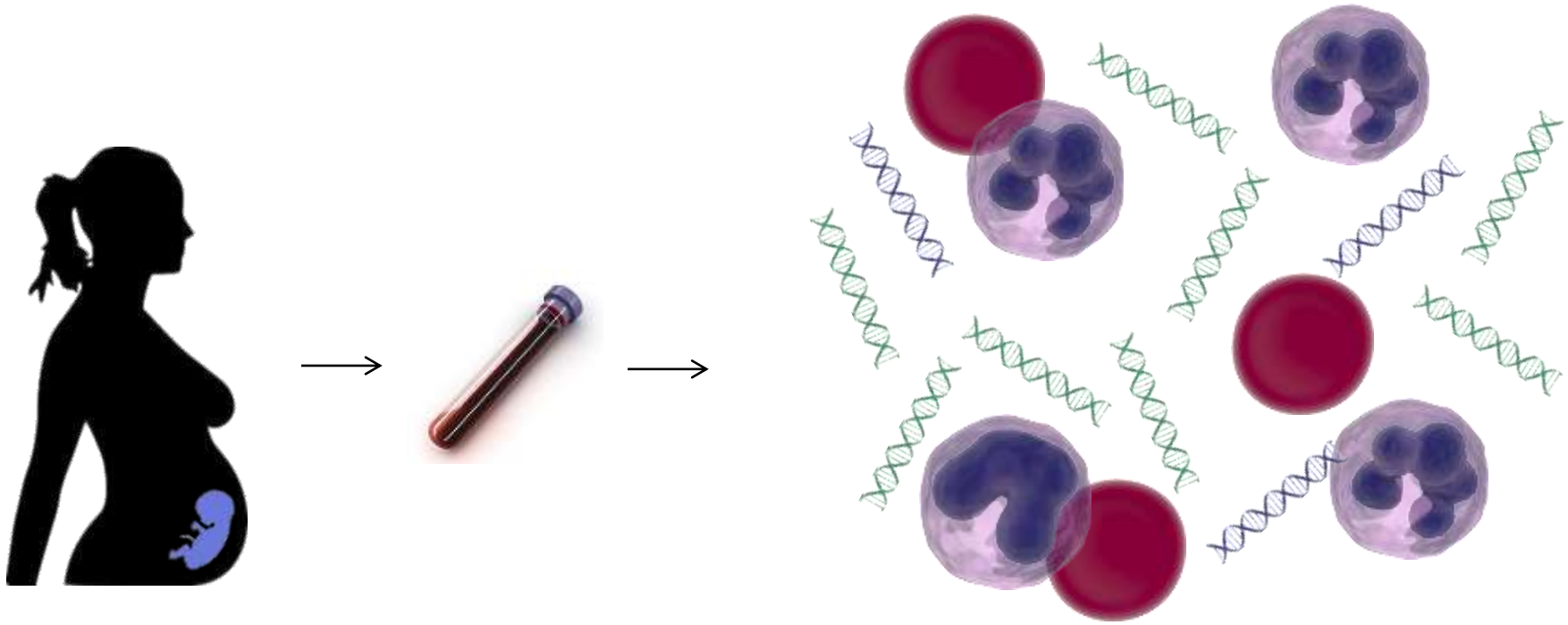
Björn Menten

Non Invasive Prenatal Testing

Next Generation Sequencing

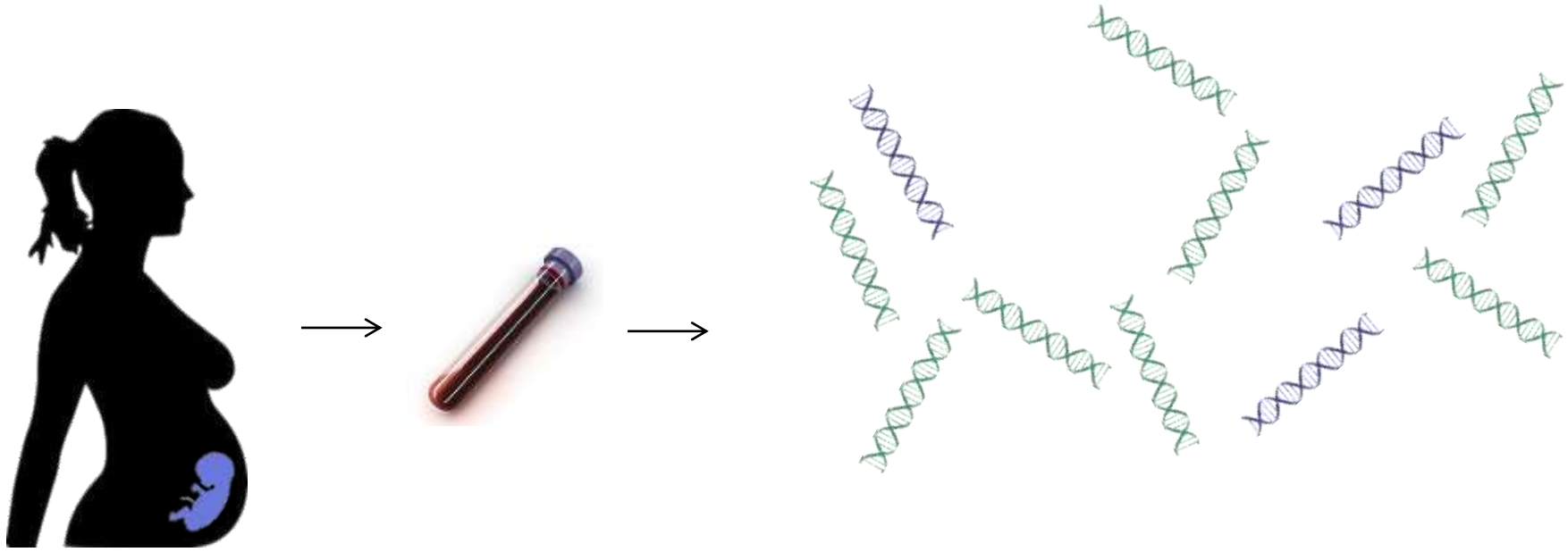


Non Invasive Prenatal Testing: cell free (fetal) DNA in maternal plasma



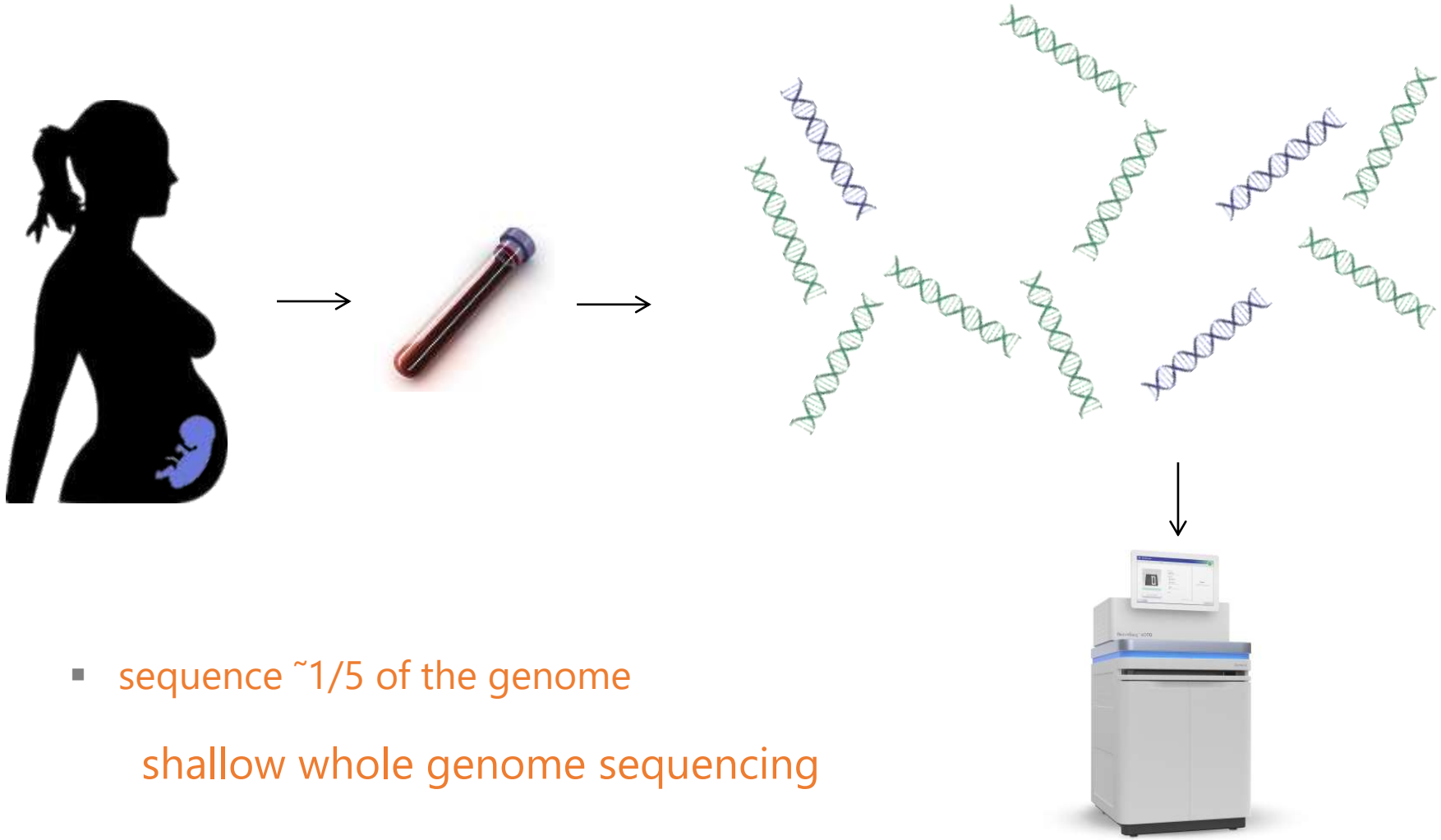
- from **syncytiotrophoblasts**
- cffDNA: 0-25% of total cell free DNA
- short sequences (~169bp)

Non Invasive Prenatal Testing: cell free (fetal) DNA in maternal plasma



- 1-100ng total cfDNA/ml plasma

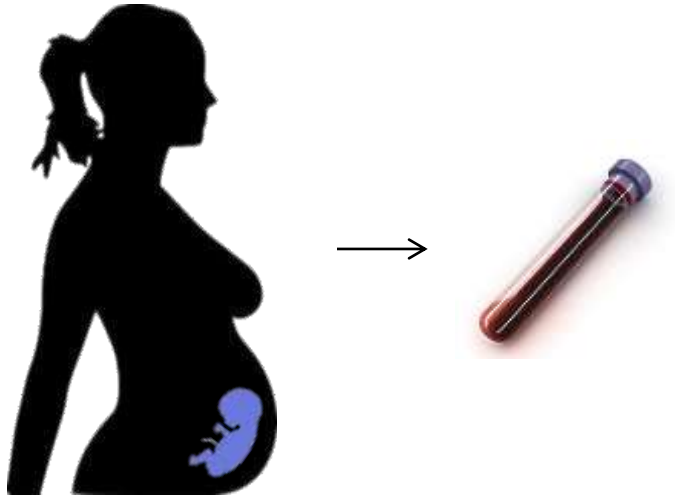
Non Invasive Prenatal Testing by massive parallel sequencing

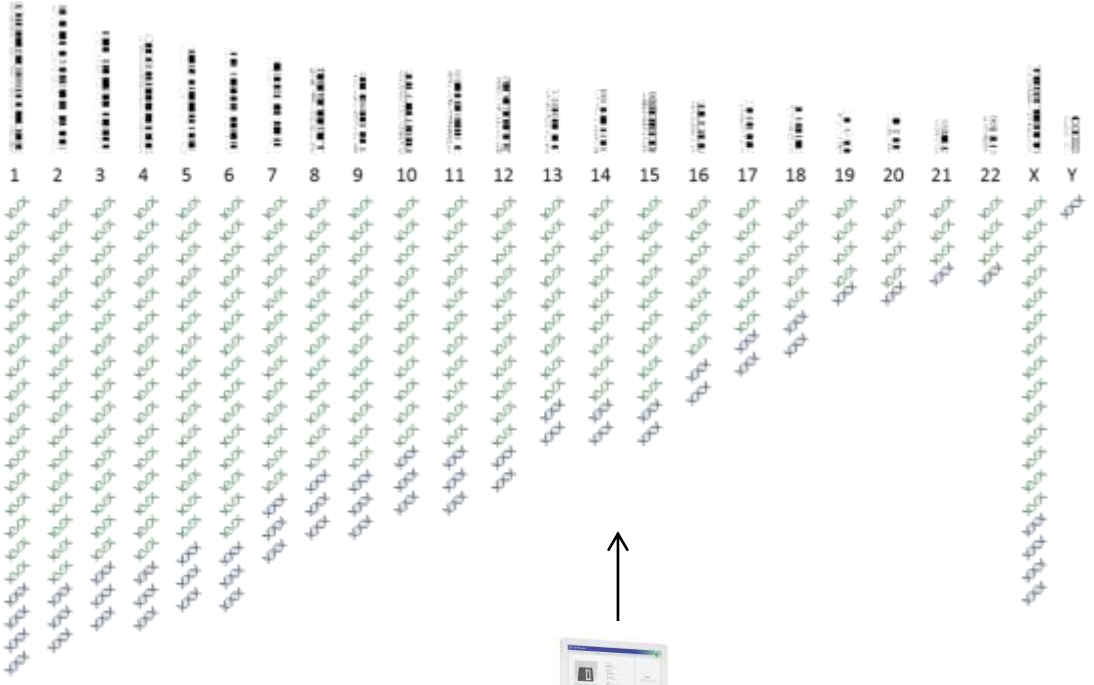


- sequence ~1/5 of the genome

  shallow whole genome sequencing

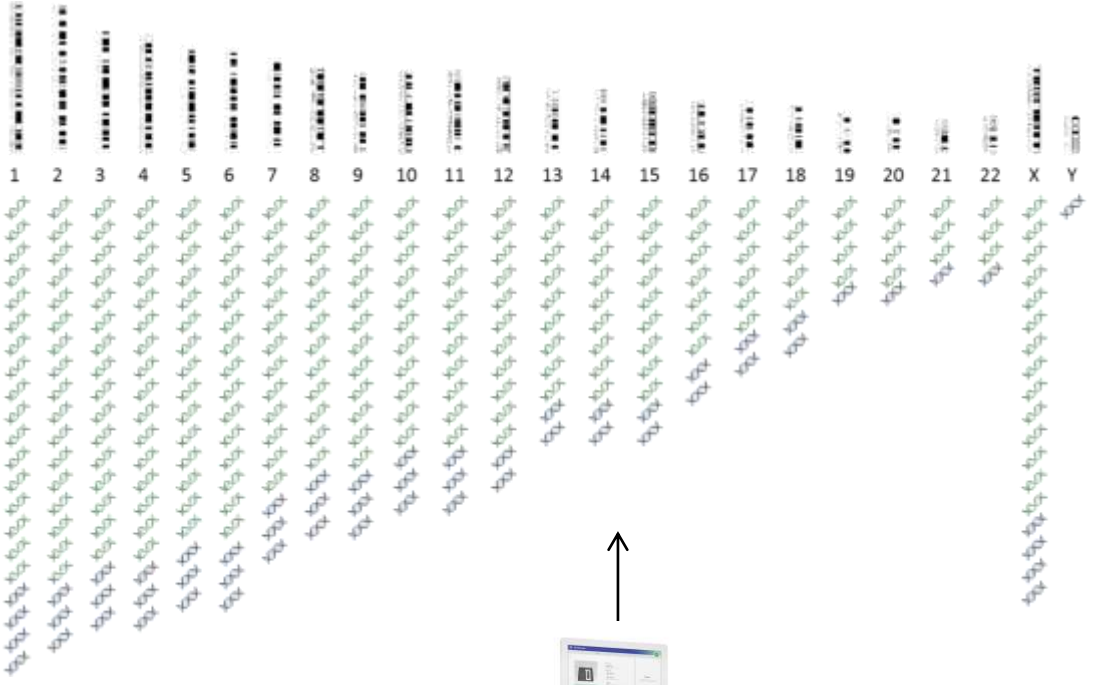
Non Invasive Prenatal Testing by massive parallel sequencing





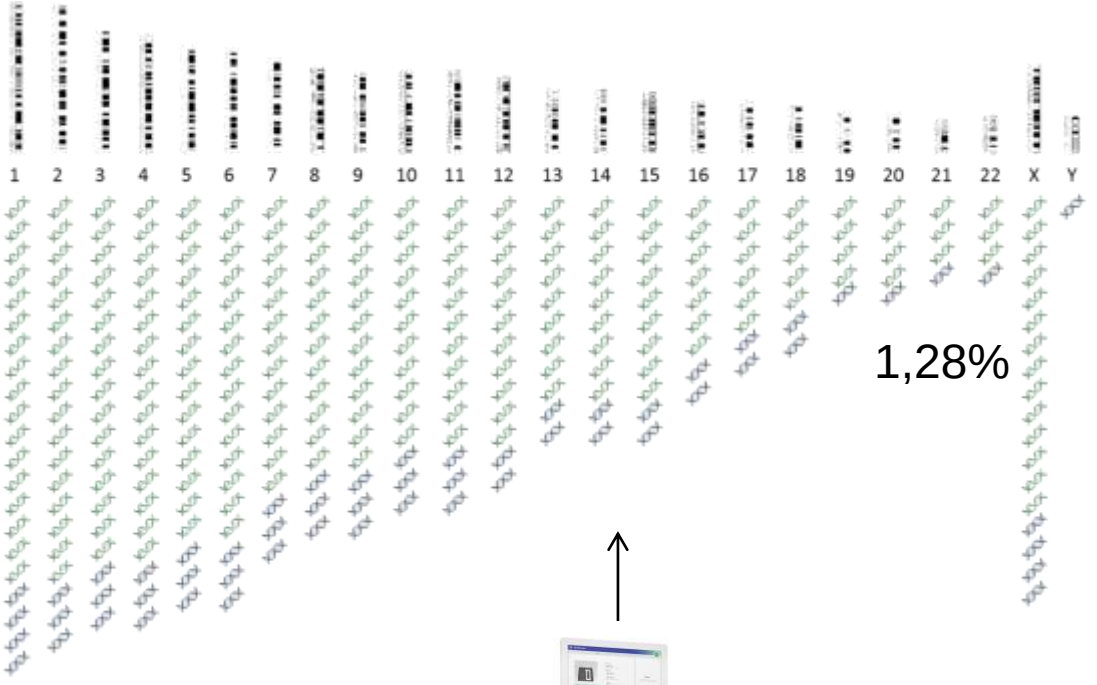
- map the sequences back to the genome

Non Invasive Prenatal Testing: counting statistics

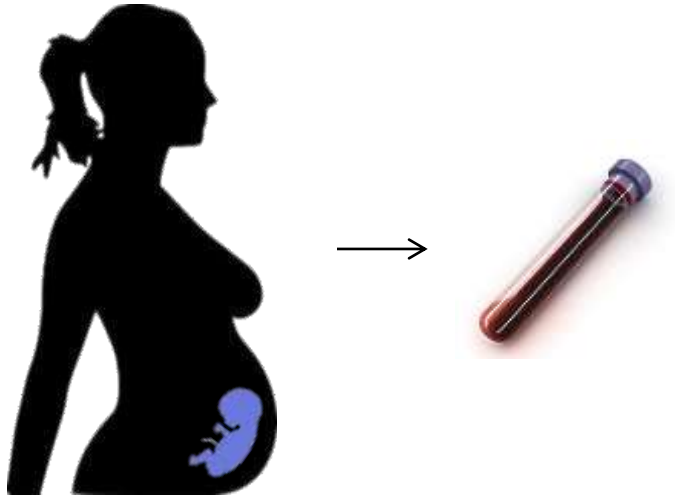


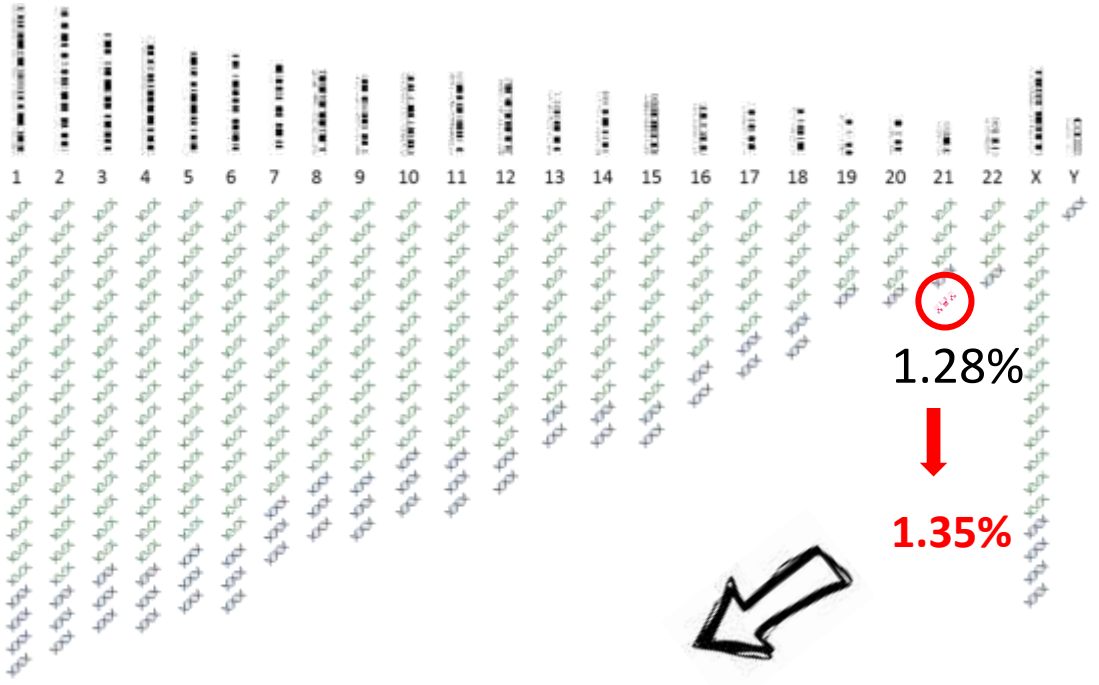
8,31%

Non Invasive Prenatal Testing: counting statistics



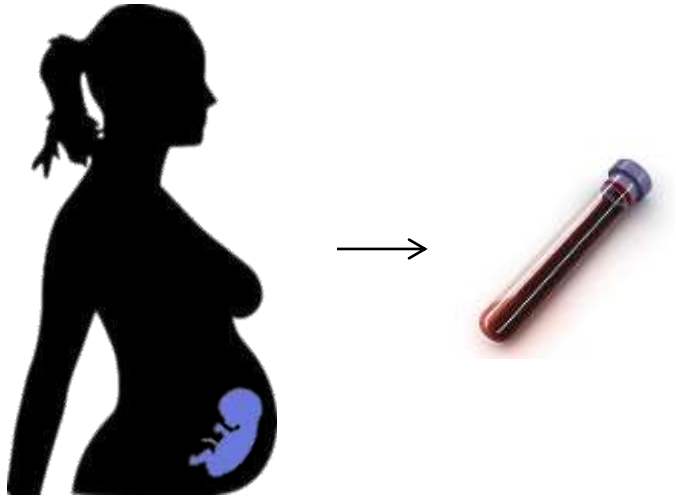
Z-score to define cut-off

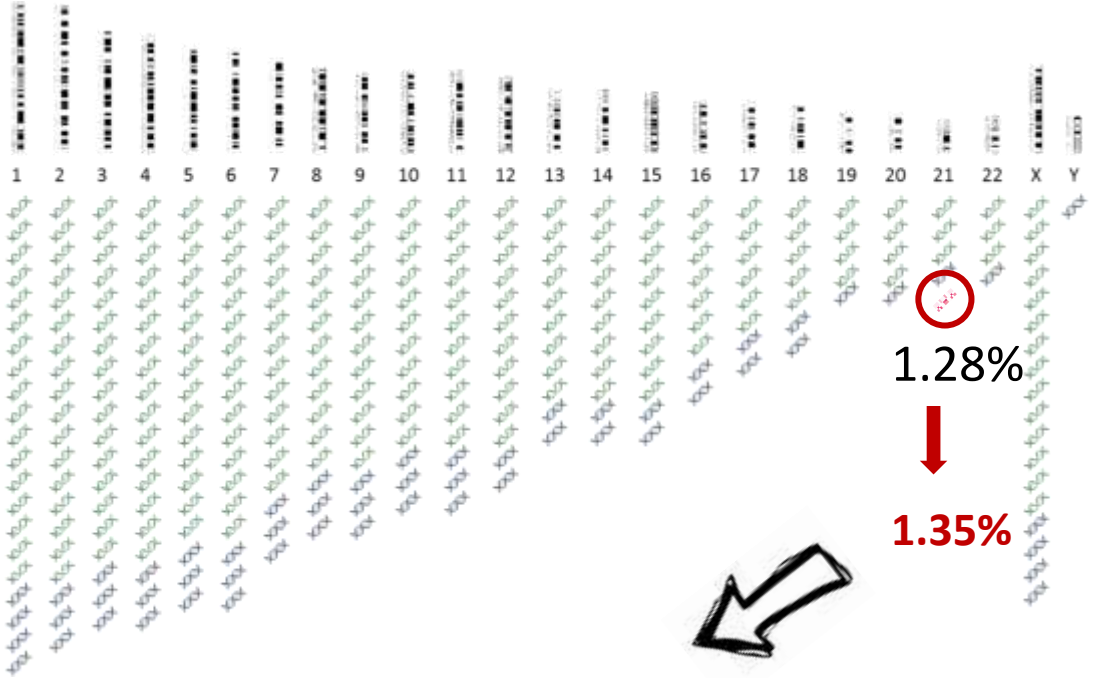




$$\text{Z-score}_{\text{chr21}} = \frac{\text{reads}_{\text{chr 21}} - \overline{\text{reads}_{\text{chr21}}}}{\text{standard deviation}_{\text{ chr 21}}}$$

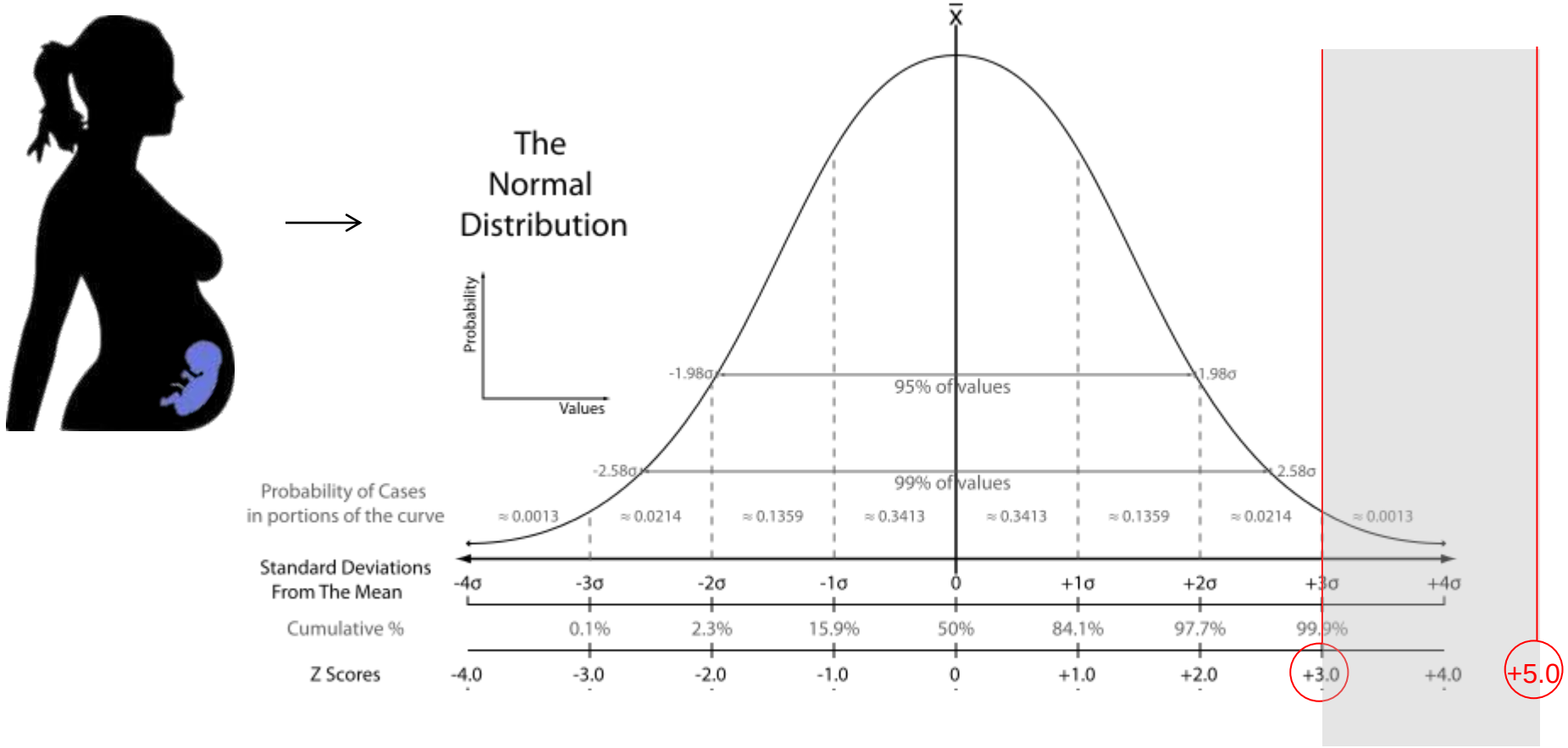
Z-score to define cut-off



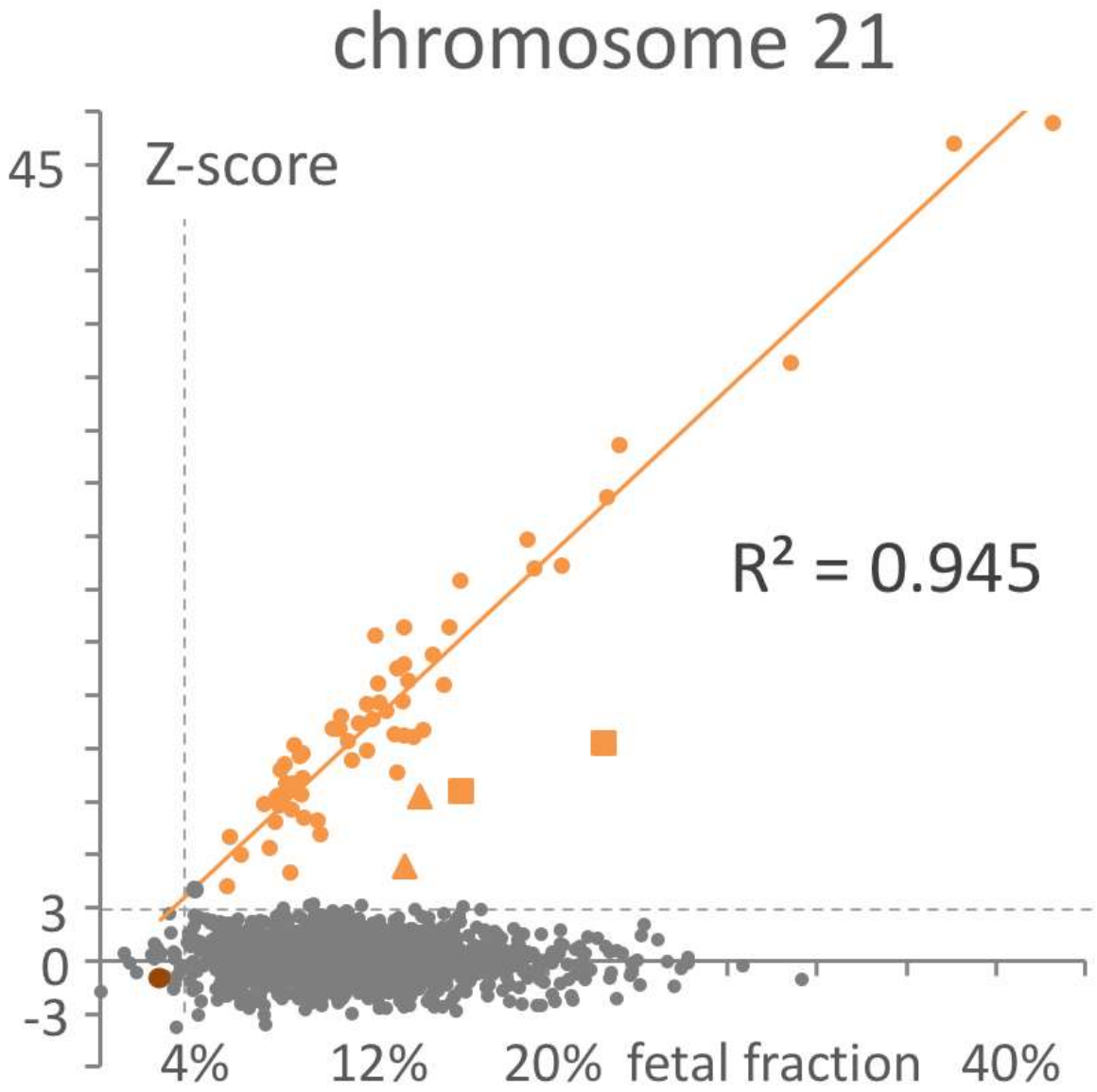


$$\text{Z-score}_{\text{chr21}} = \frac{1.35\% - 1.28\%}{0.005\%} = 14$$

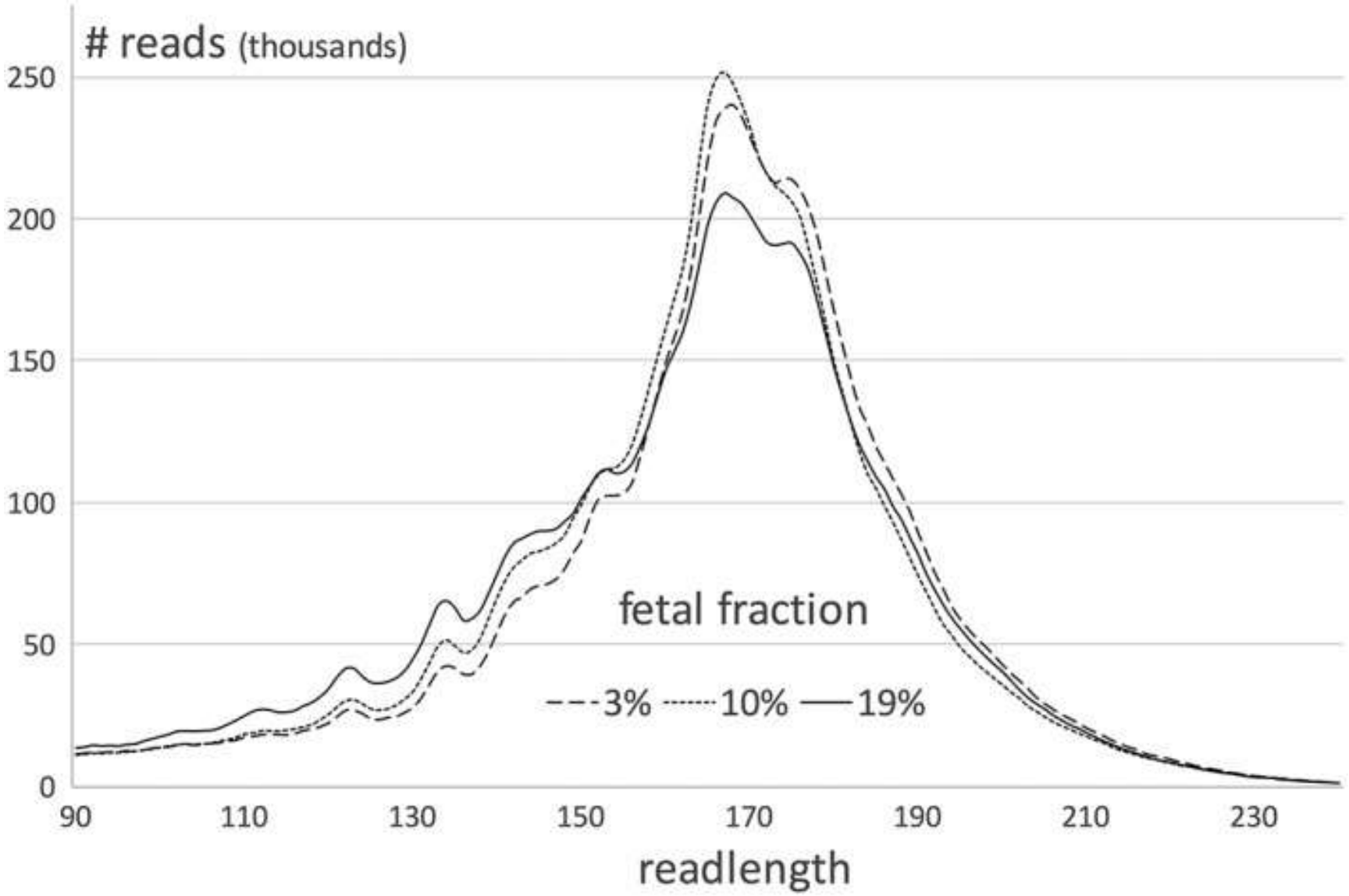
Z-score 3 <> 5 = grey-zone
> 5 = high risk for trisomy



high correlation between Z-score and fetal fraction

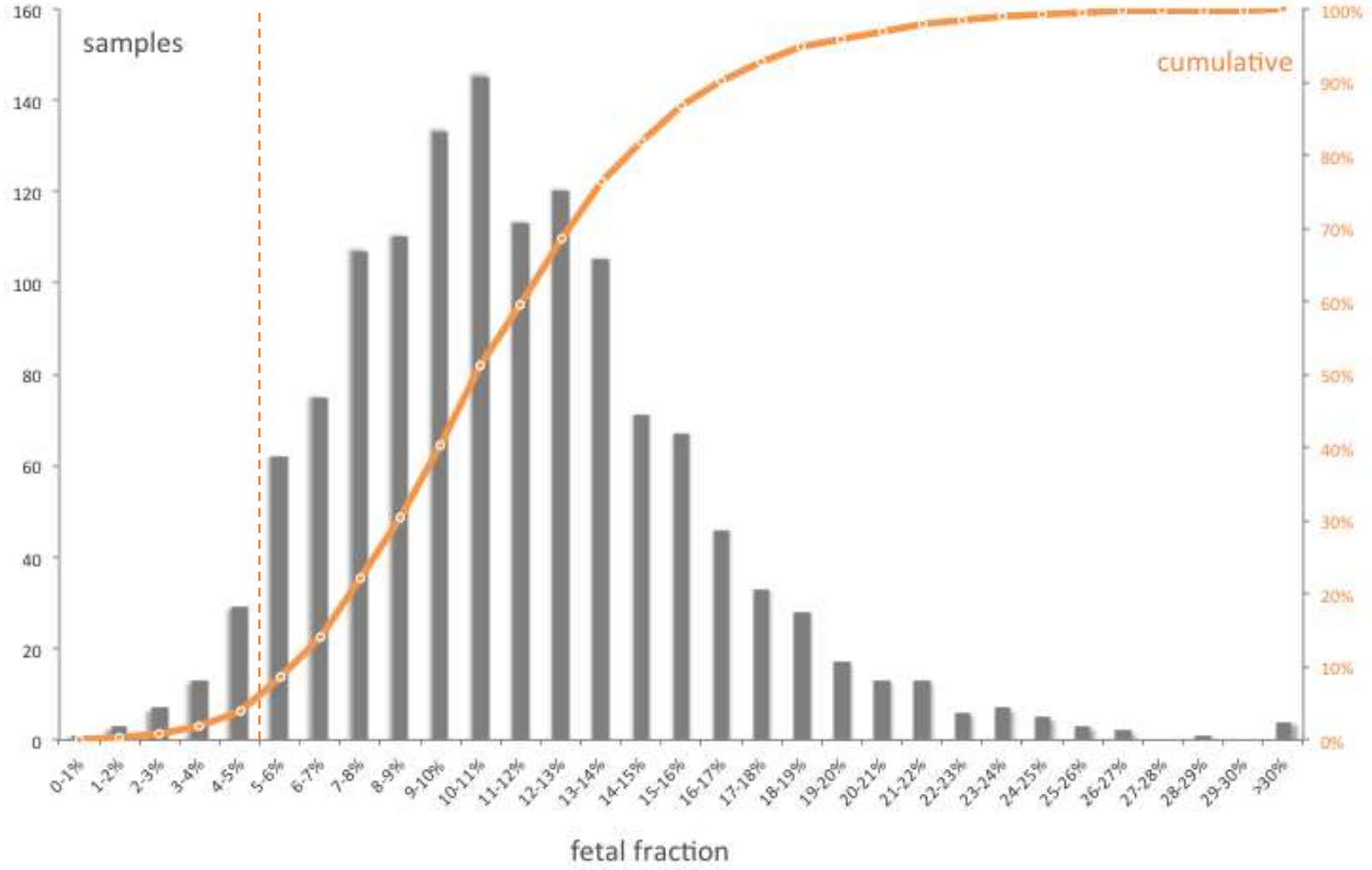


Fetal fraction calculation



Dheedene et al. 2016

Fetal fraction > 4% in most pregnancies



NIPT semi-automated workflow

3 a 4 runs/week

wisecondorX / PREFACE + vivar

6 – 7 days

- ‘fetal’ cell free DNA originates from syncythiotrophoblasts (placenta)
- strongly fragmented DNA: ~169 bp in size
- **1/5 of the genome is sequenced**

WETENSCHAP

Downtest vanaf juli (bijna) gratis voor iedereen

29-05-17, 04.30u - IB

Een jongetje met het syndroom van Down. © AP 1

Vanaf 1 juli zal de NIP-test in België volledig worden terugbetaald voor zwangere vrouwen met een voorkeursregeling. Vrouwen zonder voorkeursregeling zullen nog maximaal 8,68 euro betalen. Dat schrijven de kranten van Mediahuis vandaag. België is het eerste land in Europa dat de test vrijwel gratis maakt.

DeMorgen. Cult. Muziek, film, tv, expo Zine. Interview, foto, lifestyle

WETENSCHAP

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vrt NWS Hoofdpunten Regio Kijk Luister Net binnen

Derde minder kinderen met downsyndroom geboren sinds terugbetaling NIP-test voor zwangere vrouwen

Ferre Windey
vr 19 feb 14:53

Sinds de volledige terugbetaling van de NIP-test voor zwangere vrouwen zijn een derde minder kinderen met het syndroom van Down geboren. De test werkt erg nauwkeurig waardoor ook veel minder vrouwen onderworpen worden aan risicovolle testen zoals een vruchtwaterpunctie. Dat blijkt uit een onderzoek van alle genetische centra in België. Professor Humane Genetica Joris Vermeesch: "België was het eerste land, en is nog altijd een van de weinige, waar de test volledig wordt terugbetaald."

2 years of NIPT reimbursement

8 Centers for Human Genetics: 153 575 NIPT samples

- Trisomy 21: 1/300 (<> 1/800 live births without testing)
- Trisomy 13: 1/1650
- Trisomy 18: 1/1450

Low Fetal Fraction (FF < 4%): ~2%

Van Den Bogaert et al. (2021)

2 years of NIPT reimbursement

8 Centers for Human Genetics: 153 575 NIPT samples

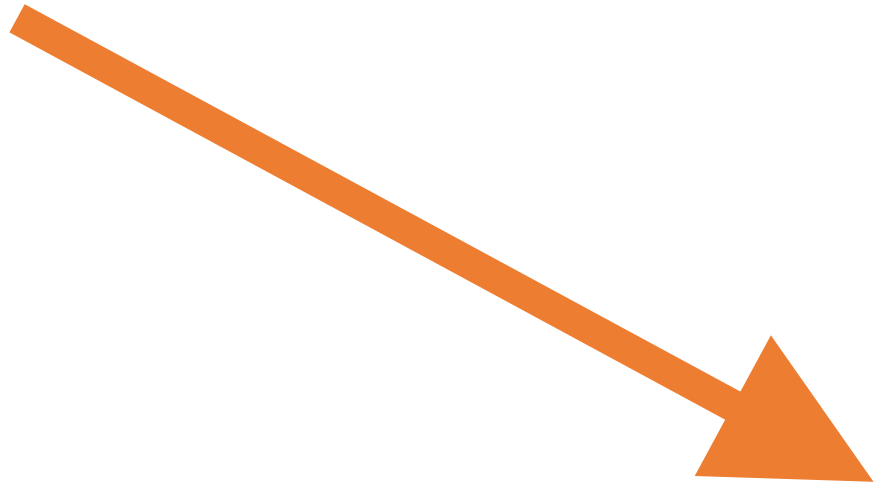
- Trisomy 21: 1/300 92,39%
- Trisomy 13: 1/1650 **43,90%** **Confirmed on amniocentesis**
  → Confined Placental Mosaicism?!
- Trisomy 18: 1/1450 84,62%

Van Den Bogaert et al. (2021)

Invasive Testing - numbers

~7600

2018



2011

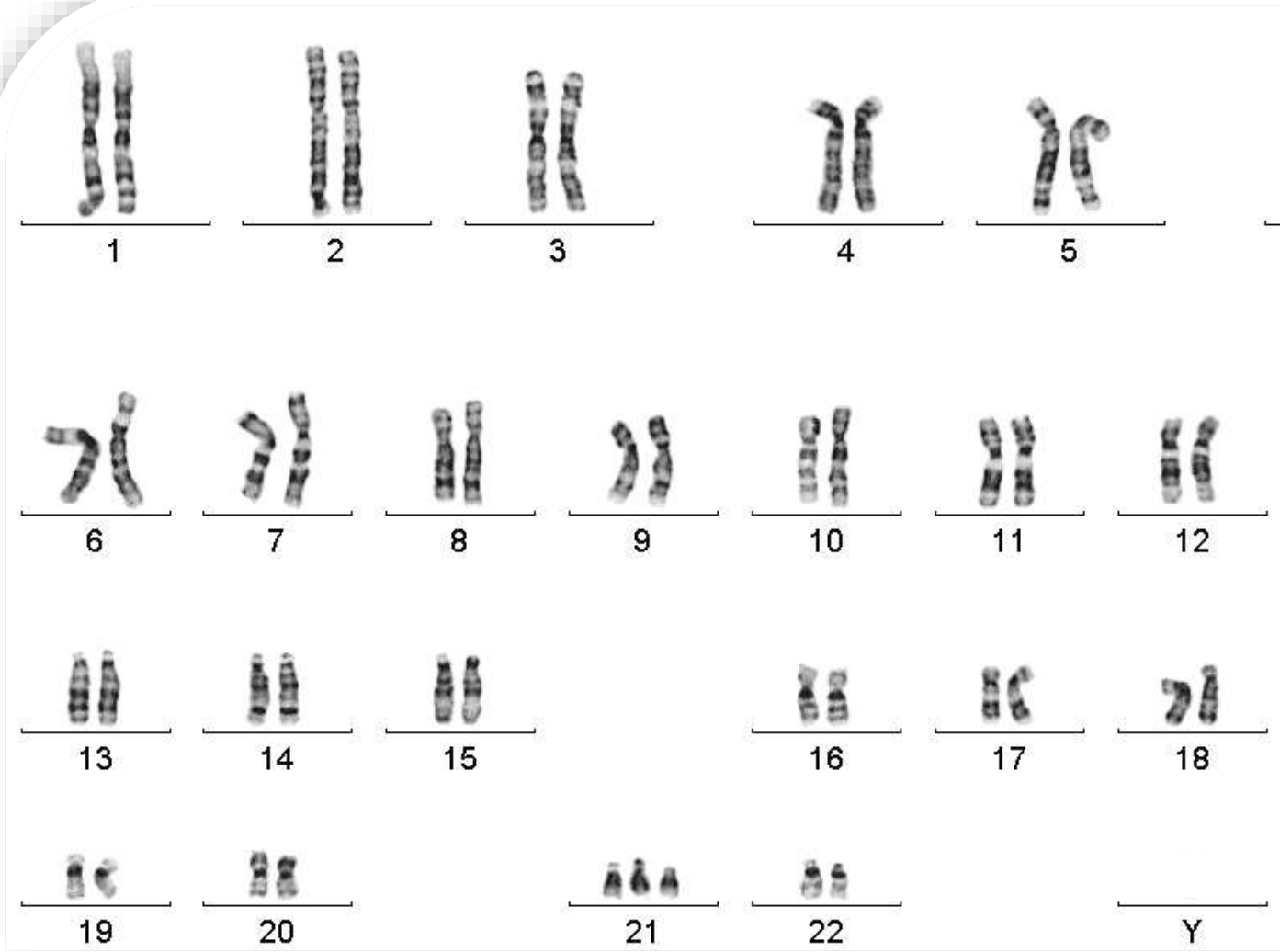
~3000

Van Den Bogaert et al. (2021)

NIPT: moeilijkheden in de dagelijkse praktijk

Dr. Ellen Roets

What holds the future?



RARE DISEASE DAY®

#notaunicorn

I'M NOT A UNICORN

Lou is 4 en lijdt aan ataxia telangiectasia (AT), ook wel de ziekte van Louis-Bar genoemd, een ongeneeslijke neurodegeneratieveziekte. Dit vrolijke meisje heeft een levensverwachting van zo'n 20 jaar. Op termijn zal zij niet meer kunnen stappen, misschien niet meer kunnen praten, niet meer zelfstandig kunnen eten of drinken, ... Er staat kinderen met A-T een intense medische opvolging te wachten, want de ziekte tast de immuniteit aan en veroorzaakt bijgevolg veel infecties. AT gaat bovendien gepaard met een sterk verhoogd risico op kanker. De opvolging van de ziekte vereist talloze bezoeken aan therapeuten en specialisten met een grote impact op het dagelijkse leven.

500.000 BELGEN MET EEN ZELDZAME ZIEKTE BESTAAN ECHT.

LOU (4)
AT, NEURODEGENERATIEVE AANDOENING

RaDiOrg
RARE DISEASES BELGIUM

RADIORG.BE/NOTAUNICORN

Frequencies of Genetic Disorders in 1,169,873 Births, 1952-83

>= 53 / 1000 individuals (younger than 25) carry a genetic disorder

1.8 / 1000 chromosomal disorder (1.2/1000 = downsyndrome)

3.6 / 1000 single gene disorder

1.7 autosomal recessive
0.5 X-linked condition
1.4 autosomal dominant

46.4 / 1000 multifactorial

Baird et al. (1988)

Belgian G**e**netics **E**xpanded C**a**rrier S**c**reening

- 1400 mild to severe recessive disorders
- Everybody carries 3 -5 recessive (lethal) alleles
- 1-2% of all couples at risk

federale overheidsdienst
VOLKSGEZONDHEID, VEILIGHEID VAN DE VOEDSELKETEN EN LEEFMILIEU

https://www.health.belgium.be/nl/node/31149

Advisory Report 9240 - Carrier screening

10/04/2017

Beschrijving Document downloaden

Expanded carrier screening in a reproductive context. Towards a responsible implementation in the healthcare system (SHC 9240) (March 2017)

In this advisory report, the Superior Health Council of Belgium provides recommendations on the criteria that need to be applied in preconceptual genetic testing for severe autosomal and X-linked recessive diseases for couples planning a pregnancy. This report aims at providing healthcare authorities and healthcare professionals with specific recommendations on the scientific and ethical issues that need to be considered in view of a responsible implementation of preconceptual genetic testing in a reproductive context. The report specifically discusses the framework underpinning the appropriate introduction of such testing and suggests inclusion criteria for diseases that could be targeted by the screening process: (i) severity, (ii) age of onset, (iii) prevalence, (iv) selection of mutations based on clinical significance and (v) treatability.

GENETISCHE TEST

Vanaf 2019 genetische test voor koppels met kinderwens

20-12-18, 06.00u - Sara Vandekerckhove - Bron: Eigen berichtgeving

© REUTERS 1

Vanaf volgend jaar zullen koppels met een kinderwens een genetische test kunnen laten uitvoeren die speurt naar 400 genetische aandoeningen. BEGECS (Belgian Genetics Expanded Carrier Screening) heet het en het werd ontwikkeld door de universitaire centra in ons land.

MEEST GELEZEN

1 Bye bye confederale droom van N-VA? Atoma-schriftjes gaan versnipperaar in

2 "De arrogantie van de macht, dat is het enige wat Bart De Wever en zijn adviseurs kennen"

3 Sektewaakhond vraagt onderzoek naar kindermisbruik bij Jehova's getuigen

4 Niet eerder in zijn leven was Charles Michel zoveel Belg, zoveel leider, zoveel attractie

5 'Dat ze maar eens goed verkracht moeten worden': vrouwen over de Twitter-bagger die ze dagelijks incasseren

Preimplantation Genetic Testing (PGT)

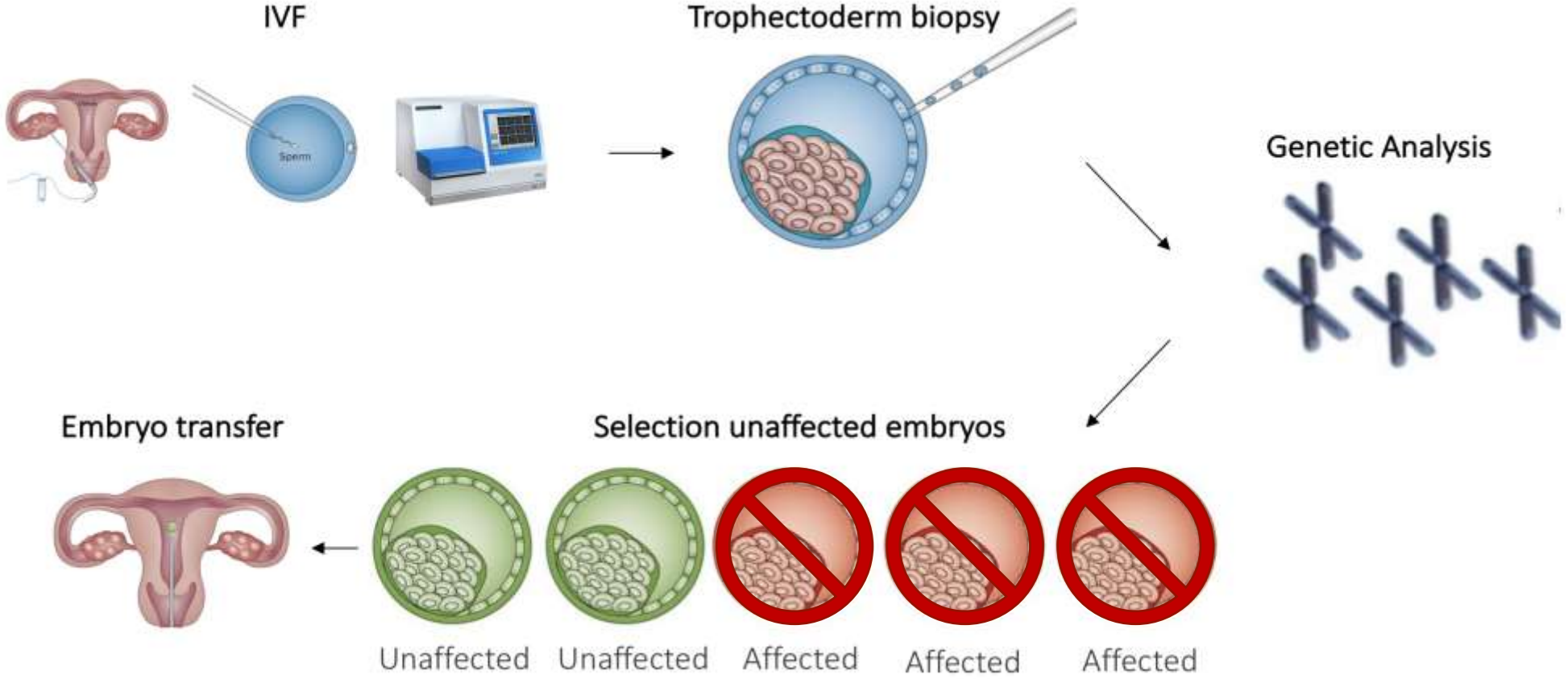


Figure adapted from Kaspa et al. 2014, Nat Rev Neur.

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- 1.7 autosomal recessive
- 0.5 X-linked condition
- 1.4 autosomal dominant (~de novo)

46.4 / 1000 multifactorial

Baird et al. (1988)

MENU

nature medicine

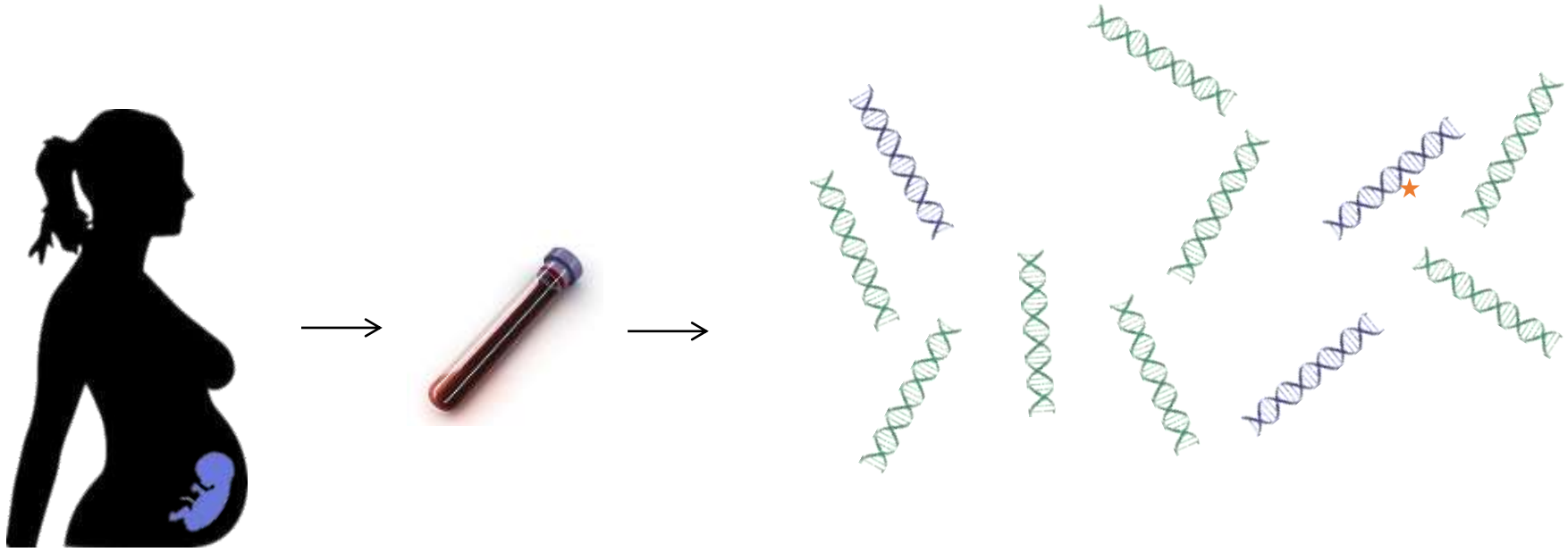
Letter | Published: 28 January 2019

Non-invasive prenatal sequencing for multiple Mendelian monogenic disorders using circulating cell-free fetal DNA

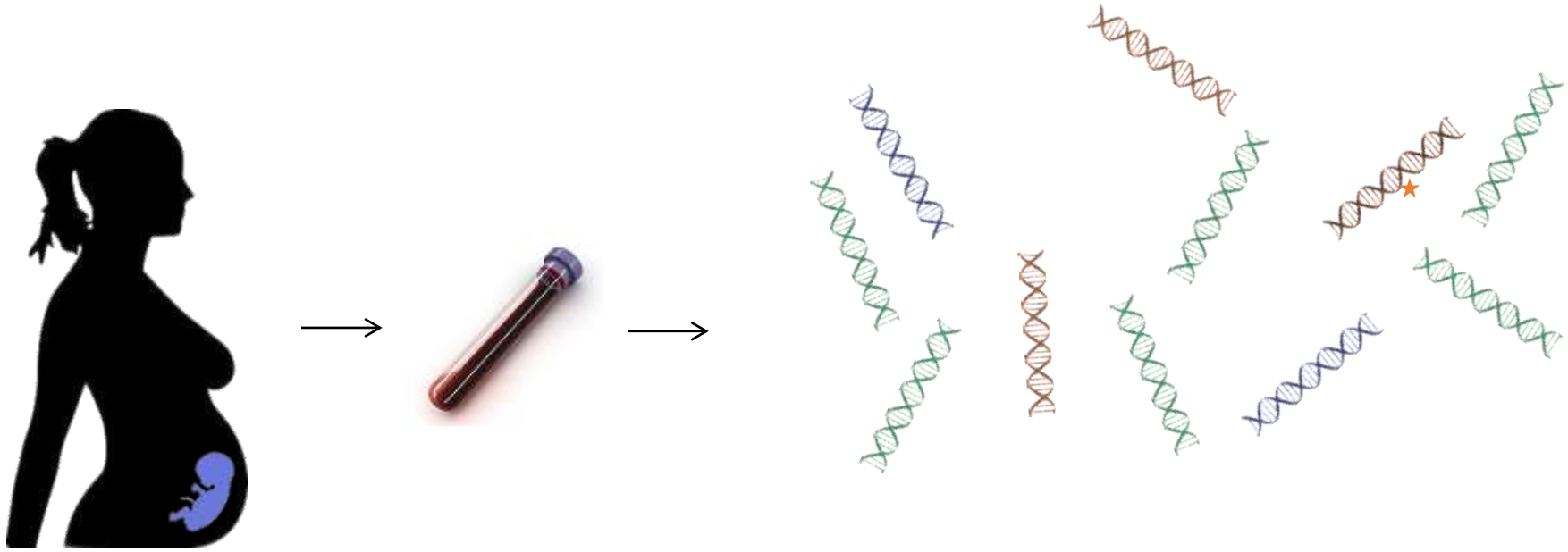
Jinglan Zhang ✉, Jianli Li, Jennifer B. Saucier, Yanming Feng, Yanjun Jiang, Jefferson Sinson, Anne K. McCombs, Eric S. Schmitt, Sandra Peacock, Stella Chen, Hongzheng Dai, Xiaoyan Ge, Guoli Wang, Chad A. Shaw, Hui Mei, Amy Breman, Fan Xia, Yaping Yang, Anne Purgason, Alan Pourpak, Zhao Chen, Xia Wang, Yue Wang, Shashikant Kulkarni, Kwong Wai Choy, Ronald J. Wapner, Ignatia B. Van den Veyver, Arthur Beaudet, Sheetal Parmar, Lee-Jun Wong & Christine M. Eng - Show fewer authors

Nature Medicine (2019) | Download Citation ⬇

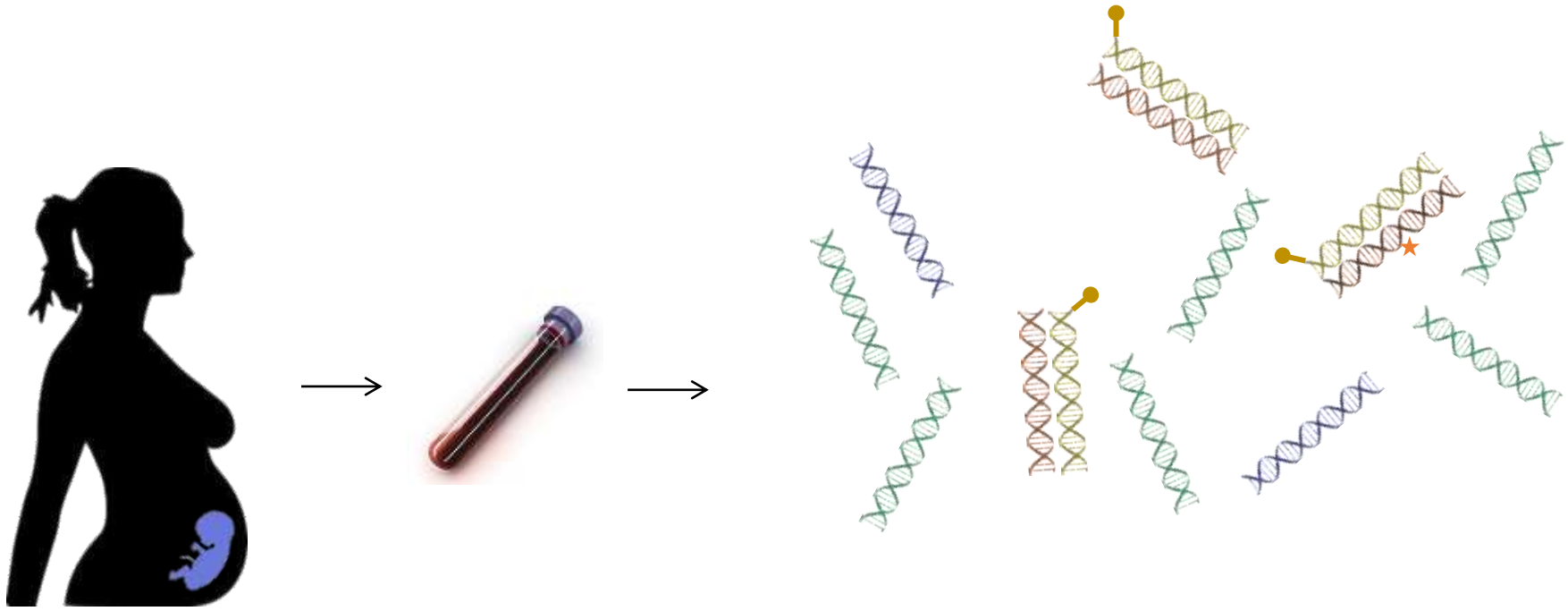
Target enrichment



Target enrichment



Target enrichment



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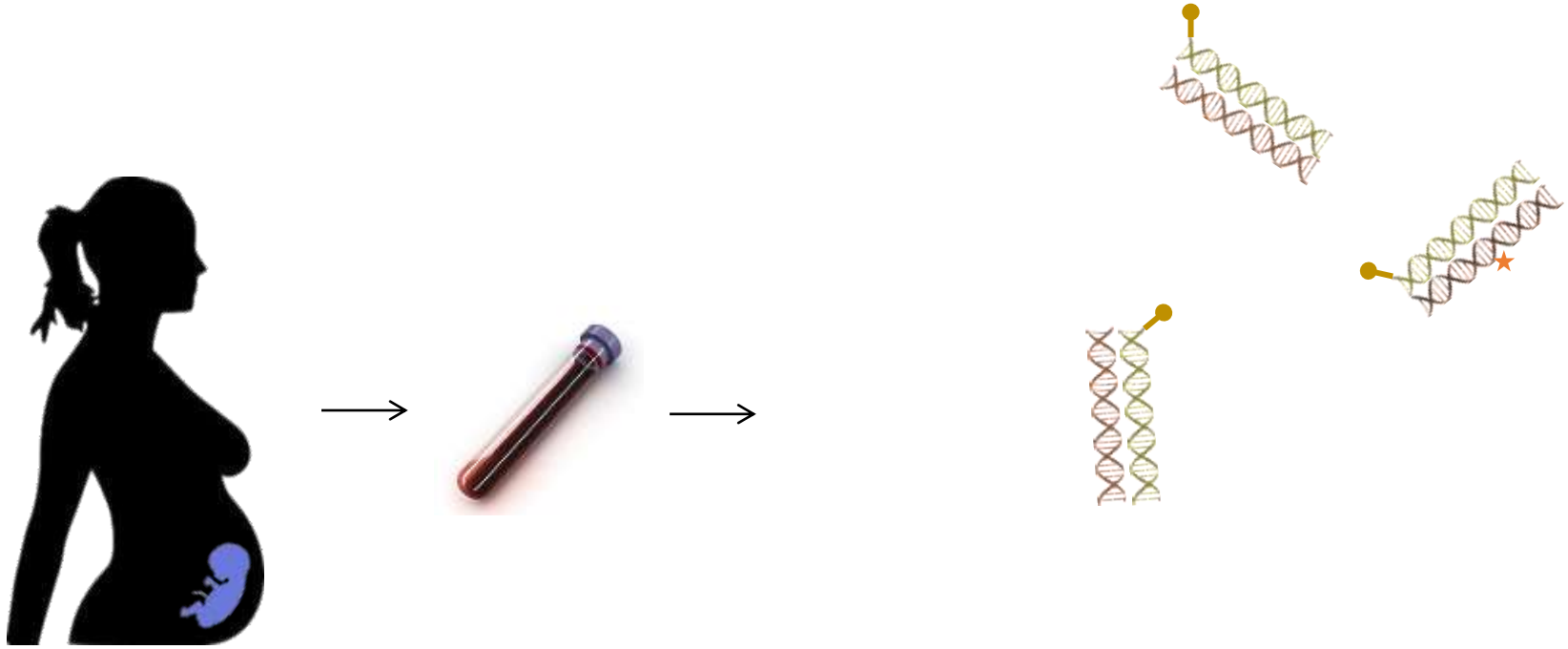

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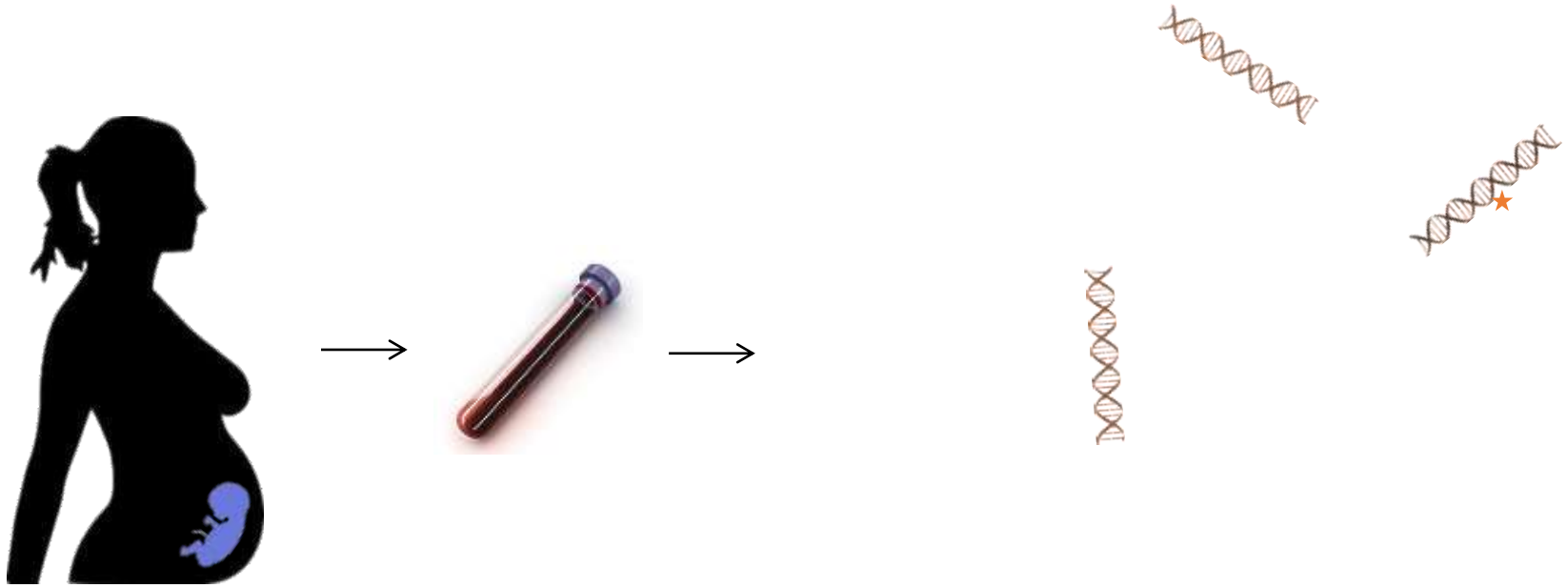

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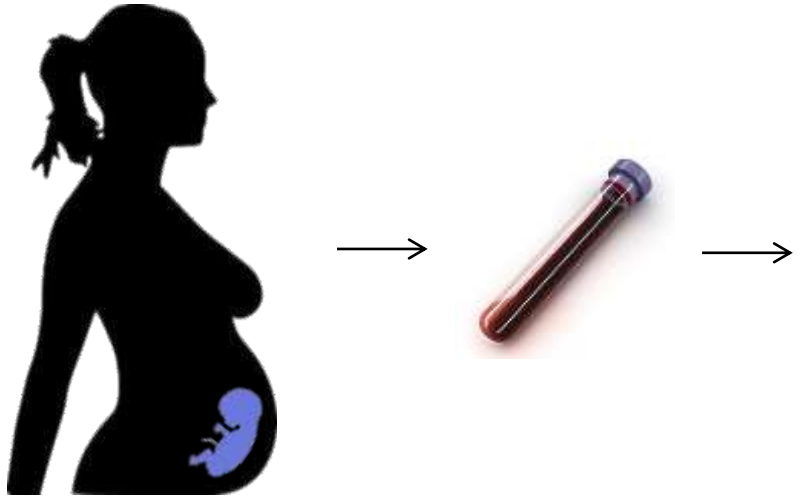

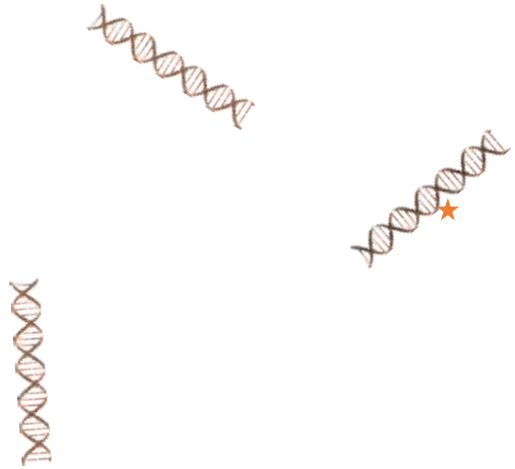

- sequence >1000x

deep, targeted sequencing

Screening panel for 30 genes associated with dominant, monogenic diseases

Gene	OMIM ID	Diseases	Sequencing detection rate[1]	Disease Prevalence[2]
BRAF	164757	cardiofaciocutaneous syndrome 1, LEOPARD syndrome 3, Noonan spectrum disorder	99%	
CBL	165360	Noonan syndrome-like disorder with or without juvenile myelomonocytic leukemia	97%	
HRAS	190020	Costello syndrome, Noonan spectrum disorder	95%	
KRAS	190070	cardiofaciocutaneous syndrome 2, Noonan spectrum disorder	99%	
MAP2K1	176872	cardiofaciocutaneous syndrome 3, Noonan spectrum disorder	99%	
MAP2K2	601263	cardiofaciocutaneous syndrome 4	99%	4-10:10,000
NRAS	164790	Noonan spectrum disorder	99%	
PTPN11	176876	LEOPARD syndrome 1, Noonan spectrum disorder	99%	
RAF1	164760	LEOPARD syndrome 2, Noonan spectrum disorder	99%	
RIT1	609591	Noonan spectrum disorder	99%	
SHOC2	602775	Noonan-like syndrome with loose anagen hair, Noonan spectrum disorder	99%	
SOS1	182530	Noonan spectrum disorder	99%	
SOS2	601247	Noonan spectrum disorder	99%	
COL1A1	120150	Caffey disease, classic Ehlers-Danlos syndrome, Ehlers-Danlos syndrome type VIIA, osteogenesis imperfecta type I, osteogenesis imperfecta type II, osteogenesis imperfecta type III, osteogenesis imperfecta type IV	95%	3-4:100,000
COL1A2	120160	cardiac valvular form of Ehlers-Danlos syndrome, Ehlers-Danlos syndrome type VIIB, osteogenesis imperfecta type II, osteogenesis imperfecta type III, osteogenesis imperfecta type IV	95%	
FGFR2	176943	Antley-Bixler syndrome without genital anomalies or disordered steroidogenesis, Apert syndrome, Beare-Stevenson cutis gyrata syndrome, bent bone dysplasia syndrome, craniofacial-skeletal-dermatologic dysplasia, Crouzon syndrome, Jackson-Weiss syndrome, LADD syndrome, Pfeiffer syndrome, Saethre-Chotzen syndrome	99%	4:100,000
FGFR3	134934	achondroplasia, CATSHL syndrome, Crouzon syndrome with acanthosis nigricans, hypochondroplasia, LADD syndrome, Muenke syndrome, SADDAN, thanatophoric dysplasia type I, thanatophoric dysplasia type II	99%	1-2:10,000
NIPBL	608667	Cornelia de Lange syndrome 1	97%	
SMC1A	300040	Cornelia de Lange syndrome 2	99%	
SMC3	606062	Cornelia de Lange syndrome 3	99%	1-10:100, 000
RAD21	606462	Cornelia de Lange syndrome 4	44%	
HDAC8	300269	Cornelia de Lange syndrome 5	77%	
TSC1	605284	tuberous sclerosis 1	95%	1-2:10,000
TSC2	191092	tuberous sclerosis 2	94%	
CDKL5	300203	early infantile epileptic encephalopathy 2	87%	unknown[3]
CHD7	608892	CHARGE syndrome	94%	10-12:100, 000
MECP2	300005	neonatal severe encephalopathy, Rett syndrome	80%	12:100,000
NSD1	606681	Sotos syndrome 1	48%	7-20: 100,000
JAG1	601920	Alagille syndrome 1, tetralogy of Fallot	89%	1:70,000
SYNGAP1	603384	autosomal dominant mental retardation 5	89%	unknown[3]

[1]Sequencing detection rate = % of mutations caused by small sequence changes [2]Prevalence data was cited from *GeneReviews* (https://www.ncbi.nlm.nih.gov/books/NBK1116/)
[3]No known population prevalence data but multiple individuals were observed in Baylor clinical whole-exome cases with *de novo* mutations in the *CDKL5* and *SYNGAP1* genes.

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1.4 autosomal dominant

46.4 / 1000 multifactorial

Neuropsychiatric disorders (autism, schizophrenia,…)
Coronary artery disease
Type 2 diabetes
Cancer
…

Baird et al. (1988)

RESEARCH ARTICLE SUMMARY

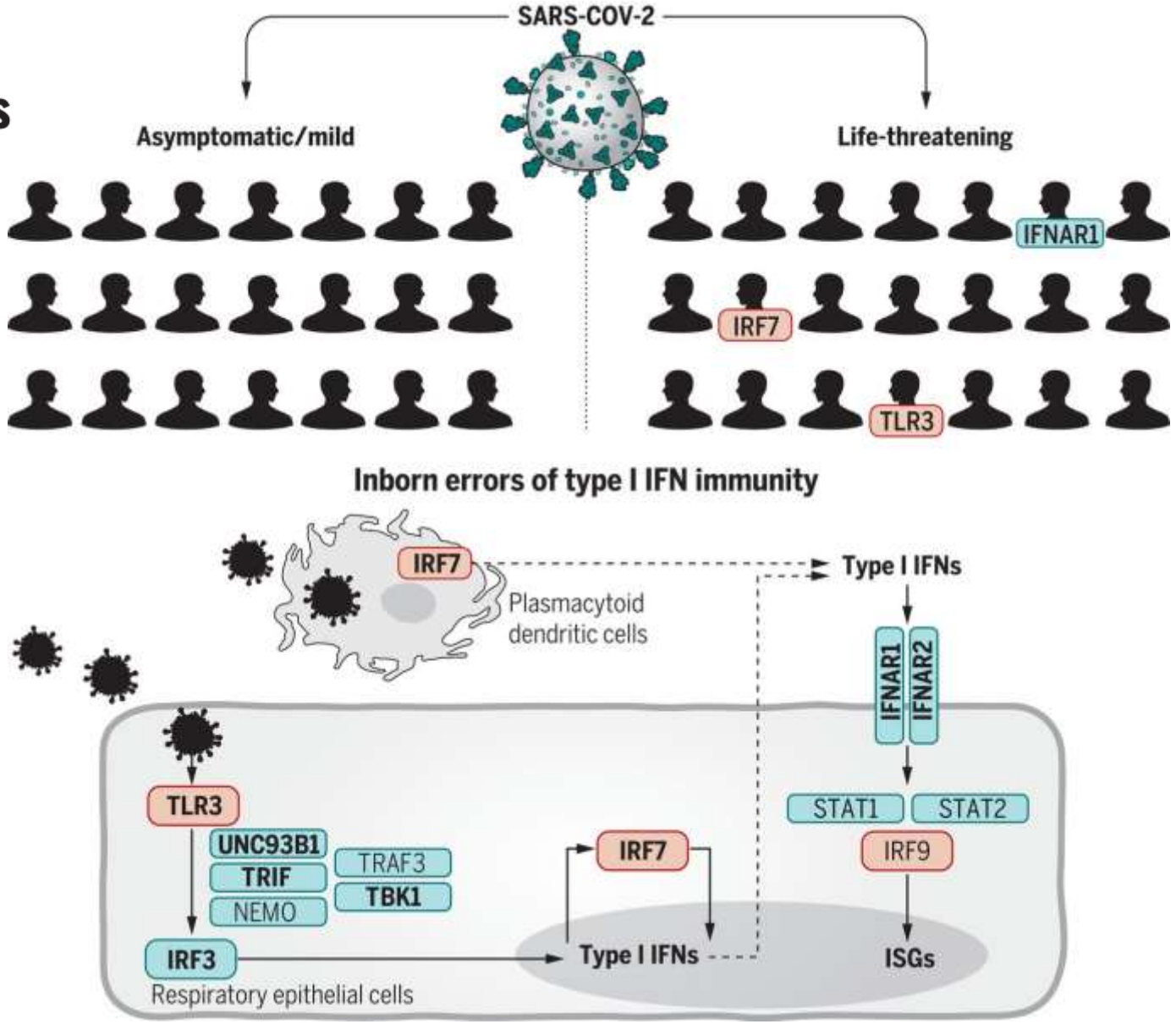
CORONAVIRUS

Inborn errors of type I IFN immunity in patients with life-threatening COVID-19

Zhang *et al.*, *Science* **370**, eabd4570 (2020) 23 October 2020

Prof. Jean-Laurent Casanova (Rockefeller USA)

Filomeen Haerynck
© Thomas Verfaillie



NIPT for pregnancy complications?

Original Research ajog.org

OBSTETRICS

Preeclampsia: novel insights from global RNA profiling of trophoblast subpopulations

Matthew Gormley, BS; Katherine Ona, BS; Mirhan Kapidzic, MD; Tamara Garrido-Gomez, PhD; Tamara Zdravkovic, PhD; Susan J. Fisher, PhD

Belgian Consortium on Prenatal Genetic Testing

Belgian Consortium on Prenatal Genetic Testing

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François Wilkin

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UZ GENT

Extra slides

Clinical and Societal Impact of National Publicly Funded Firts-Tier Non-Invasive Prenatal Testing

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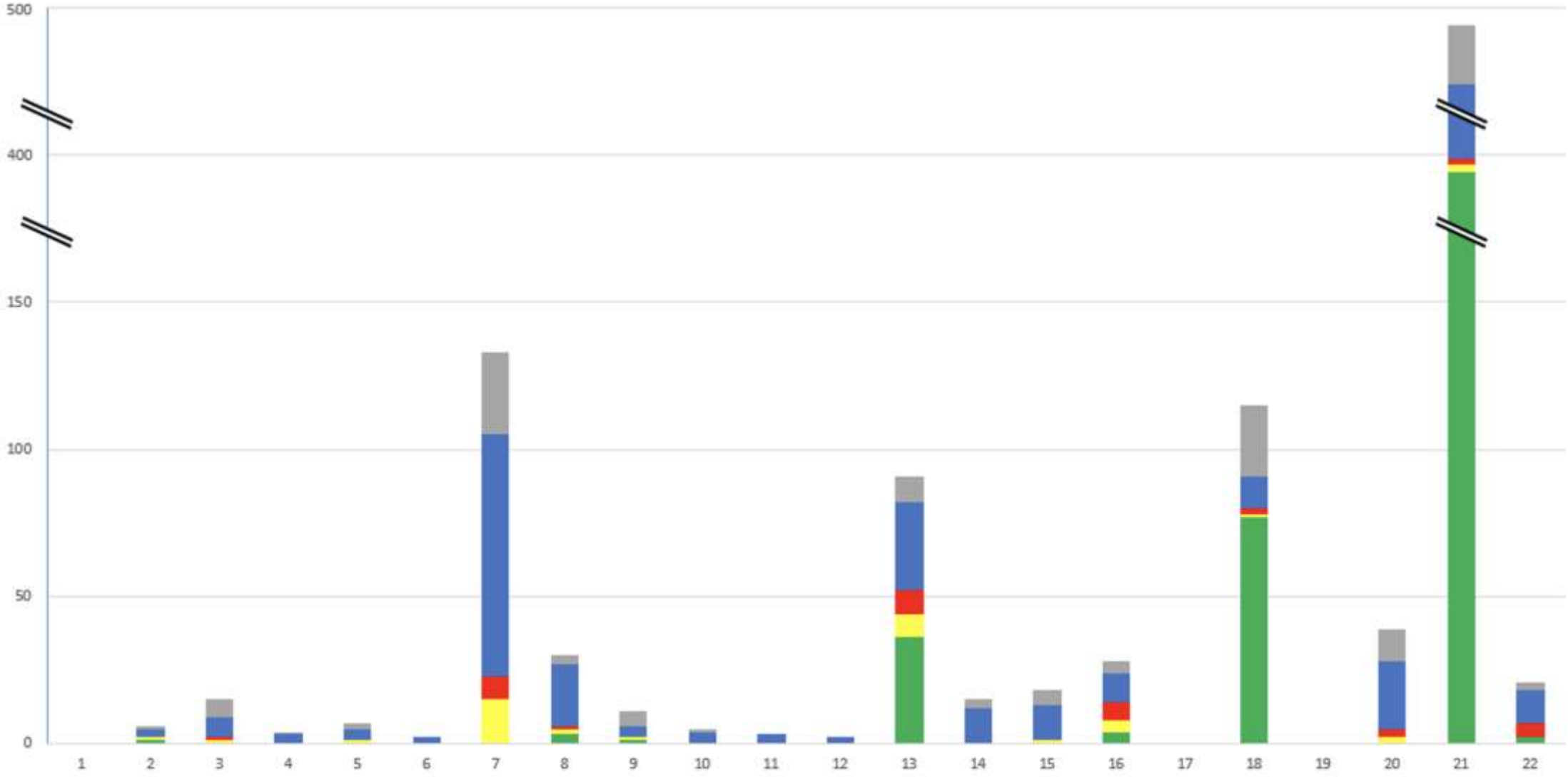
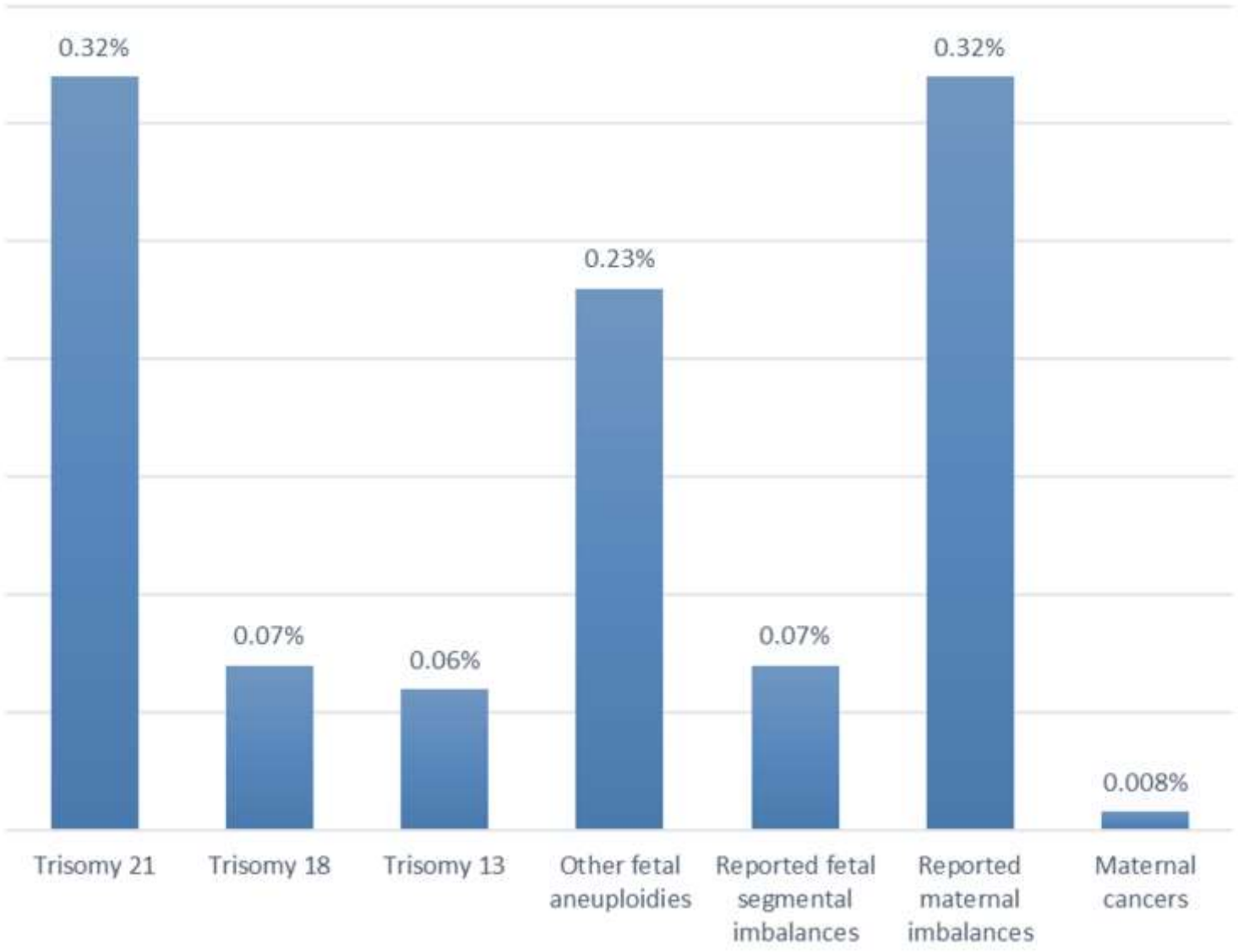
Genetics inMedicine

Reported autosomal trisomies

true positive (green); confined placental mosaicism (yellow); negative on both amniotic fluid and placenta biopsy (red); negative on amniotic fluid but no placenta biopsy available (blue); no invasive follow-up (grey)



incidence

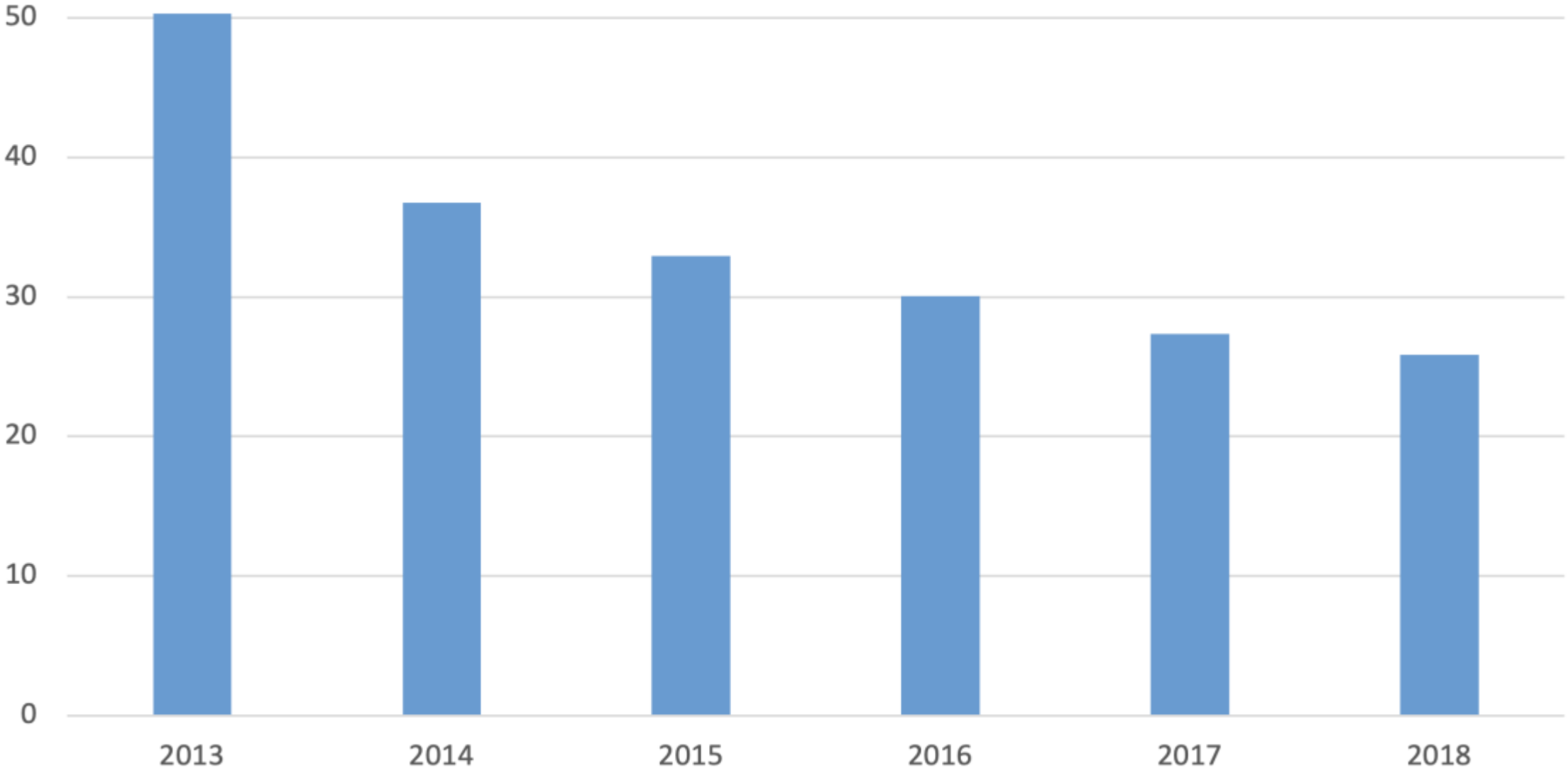

0.32%
0.07%
0.06%
0.23%
0.07%
0.32%
0.008%
Trisomy 21
Trisomy 18
Trisomy 13
Other fetal aneuploidies
Reported fetal segmental imbalances
Reported maternal imbalances
Maternal cancers

Performance of NIPT as a first-tier screening test

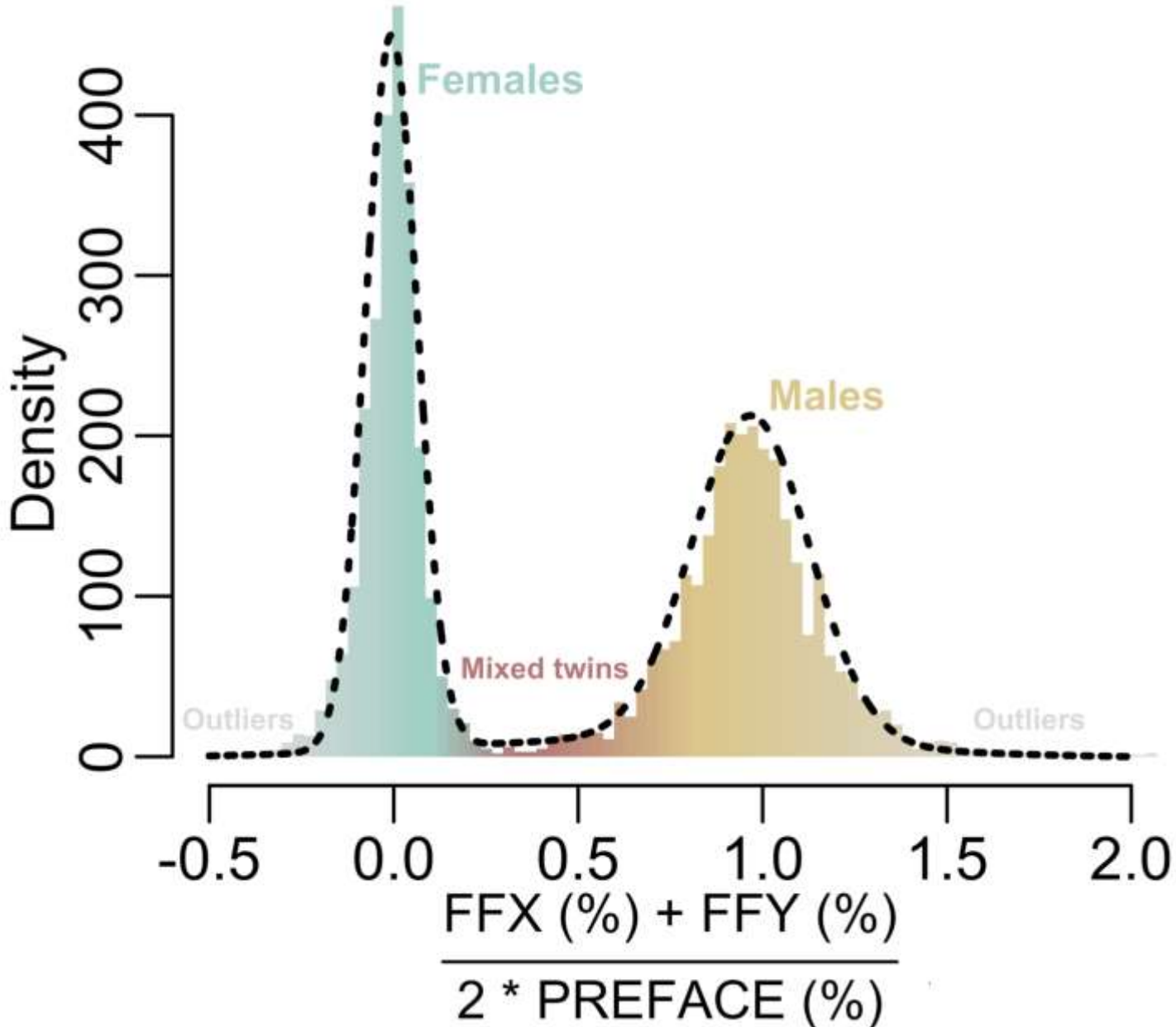
	Incidence	Sensitivity		Specificity		PPV		NPV	
	%	%	CI95%	%	CI95%	%	CI95%	%	CI95%
Trisomy 21	0.32	98.91	97.24 - 99.58	99.98	99.97 - 99.99	92.39	89.34 - 94.61	100.00	99.99 - 100.00
Trisomy 18	0.07	97.47	91.23 - 99.30	99.99	99.98 - 99.99	84.62	75.82 - 90.61	100.00	100.00 - 100.00
Trisomy 13	0.06	100.00	90.36 - 100.00	99.97	99.96 - 99.98	**43.90**	33.67 - 54.68	100.00	100.00 - 100.00

PPV = positive predictive value; NPV = negative predictive value; CI95% = 95% confidence interval

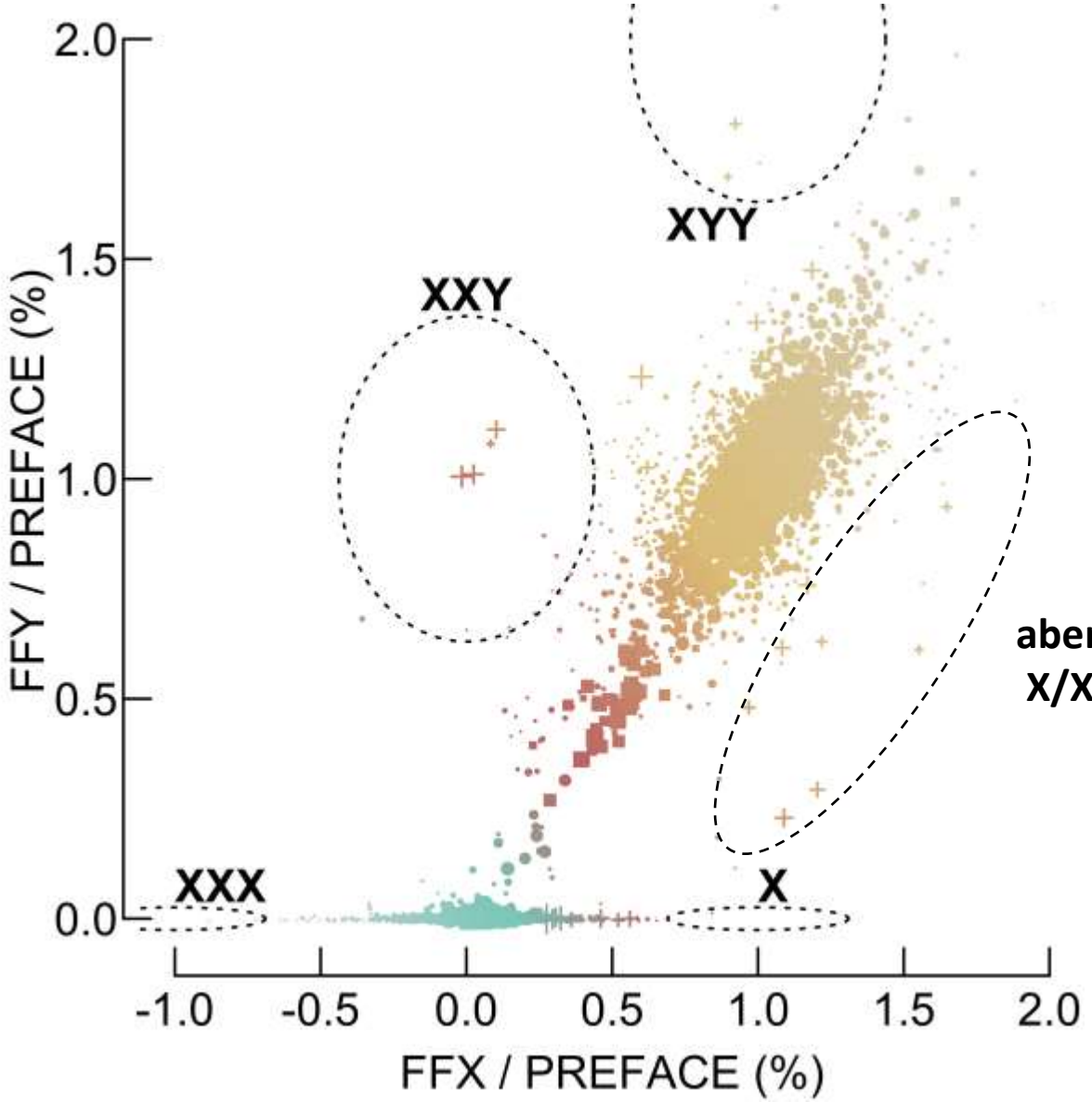
Number of invasive procedures normalized per 1,000 live births



What about Sex Chromosome Abnormalities?



Sex Chromosome Abnormalities: detectable, but with low(er) sensitivity / specificity



aberrant Y?
X/XY mosaicism?

Collaboration between all eight genetic centers (BEGECS)

- consensus panel of clearly pathogenic mutations (class V) (~100 genes/conditions)
- severe, childhood onset disorders
- pre-test counseling by genetic counselor with informed consent
- preferential preconceptional , prenatal possible for high risk groups

federale overheidsdienst
VOLKSGEZONDHEID, VEILIGHEID VAN DE VOEDSELKETEN EN LEEFMILIEU

https://www.health.belgium.be/nl/node/31149