NEW INSIGHTS AND PERSPECTIVES ON

INTRACEREBRAL HEMORRHAGE:

A COMPREHENSIVE UPDATE

ISC BFM

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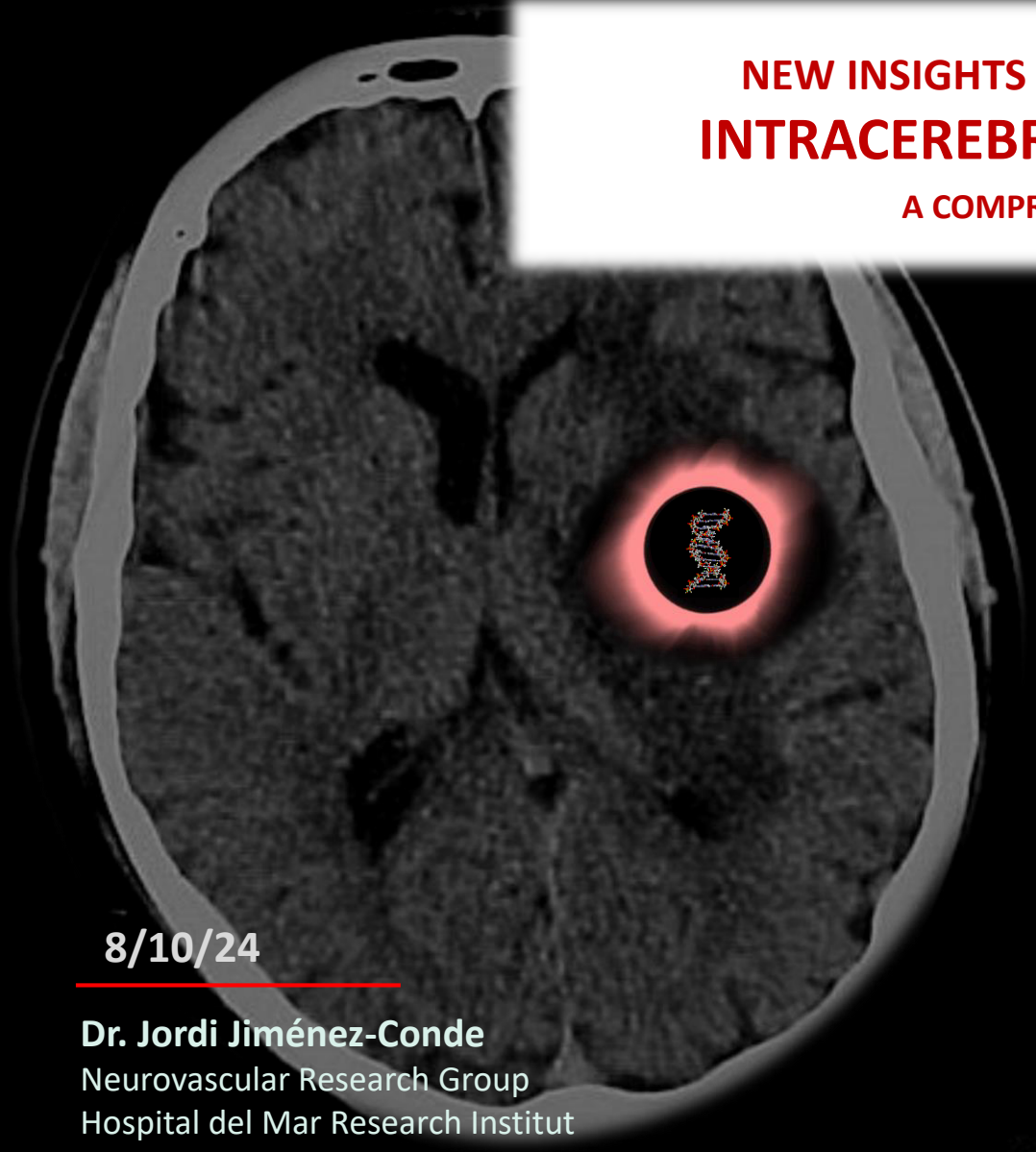
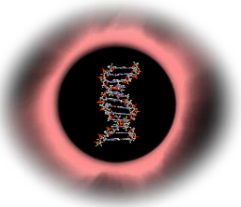
RICORS-ICTUS | Instituto de Salud Carlos III



What is written in the Genes

8/10/24

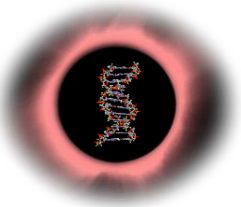
Dr. Jordi Jiménez-Conde
Neurovascular Research Group
Hospital del Mar Research Institut
Barcelona



GENETICS. WHY IT MATTERS

- Heritability of ICH: 30-50%

*Heritability: the proportion of variation in a trait explained by inherited genetic variants. In other words, it's a way to measure how much the differences in people's DNA can explain the differences in their traits.



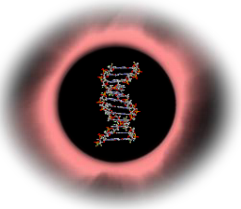
GENETICS. WHY IT MATTERS

- Heritability of ICH: 30-50%
- Understanding disease mechanisms (occurrence, evolution, complications..)
- Identify Individual Risk (Biomarkers)
- Predict outcome. Prognosis. (Biomarkers)
- Discover new pathways and potential therapeutic targets
    - Prevention targets
    - Treatment of the event and complications
- Response and risk of treatments.

GENETICS IN CEREBROVASCULAR DISEASES

BRIEF HISTORICAL JOURNEY

- Monogenic Diseases



Some Mendelian diseases

	Inheritance	Gene	Stroke mechanism	Associated clinical features	Diagnostic test
Ischaemic stroke					
CADASIL	Autosomal dominant	*NOTCH3*	Small-vessel disease	Migraine with aura	Mutational screening, skin biopsy
CARASIL	Autosomal recessive	*HTRA1*	Small-vessel disease	Premature baldness; severe low back pain; spondylosis deformans or disk herniation	Mutational analysis
Fabry's disease	X-linked	*GAL*	Large-artery disease and small-vessel disease	Angiokeratoma; neuropathic pain; acroparaesthesia; hypohydrosis; corneal opacities; cataract; renal and cardiac failure	α galactosidase activity, mutational screening
MELAS	Maternal	mtDNA	Complex (microvascular and neuronal factors)	Developmental delay; sensorineural hearing loss; short stature; seizures; episodic vomiting; diabetes; migraine-like headache; cognitive decline	Muscle biopsy, mutational analysis of mtDNA
Sickle-cell disease	Autosomal recessive	*HBB*	Large-artery disease, small-vessel disease, haemodynamic insufficiency	Pain crises; bacterial infection; vaso-occlusive crises; pulmonary and abdominal crises; anaemia; myelopathy; seizure	Peripheral blood smear, electrophoresis, mutational analysis
Homocystinuria	Autosomal recessive	*CBS* and others	Large-artery disease, cardioembolism, small-vessel disease, arterial dissection	Mental retardation; atraumatic dislocation of lenses; skeletal abnormalities (Marfan-like); premature atherosclerosis; thromboembolic events	Urine analysis, measurement of concentrations of homocysteine and methionine in plasma (mutational screening)
Marfan's syndrome	Autosomal dominant	*FBN1*	Cardioembolism and arterial dissection	Pectus carinatum or excavatum; upper-to-lower-segment ratio <0·86, or arm-span-to-height ratio >1·5; scoliosis >20%; ectopia lentis; dilation or dissection of the ascending aorta; lumbosacral dural ectasia	Clinical diagnosis (mutational screening)
Ehlers-Danlos syndrome type IV	Autosomal dominant	*COL3A1*	Arterial dissection	Easy bruising; thin skin with visible veins; characteristic facial features; rupture of arteries, uterus, or intestines	Biochemical studies, mutational screening
Pseudoxanthoma elasticum	Autosomal recessive	*ABCC6*	Large-artery disease and small-vessel disease	Skin changes (increased elasticity and yellow-orange papular lesions); ocular changes (angioid streaks); hypertension	Skin biopsy, mutational screening
Intracerebral haemorrhage					
Familial cerebral amyloid angiopathy	Autosomal dominant	*APP*	Rupture of cortical cerebral small vessels	Cerebral lobar macrohaemorrhages and microhaemorrhages; white-matter lesions; cognitive impairment	Brain biopsy, mutational screening
COL4A1-related intracerebral haemorrhage	Autosomal dominant	*COL4A1*	Rupture of cortical and subcortical cerebral small vessels	Infantile hemiparesis; congenital porencephaly; white-matter lesions; cerebral macrohaemorrhages and microhaemorrhages (lobar and non-lobar); transient ischaemic attacks	Clinical diagnosis, mutational screening

CADASIL=cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy. CARASIL=cerebral autosomal recessive arteriopathy with subcortical infarcts and leukoencephalopathy. MELAS=mitochondrial myopathy, encephalopathy, lactic acidosis, and stroke. mtDNA=mitochondrial DNA.

GENETICS IN CEREBROVASCULAR DISEASES

BRIEF HISTORICAL JOURNEY

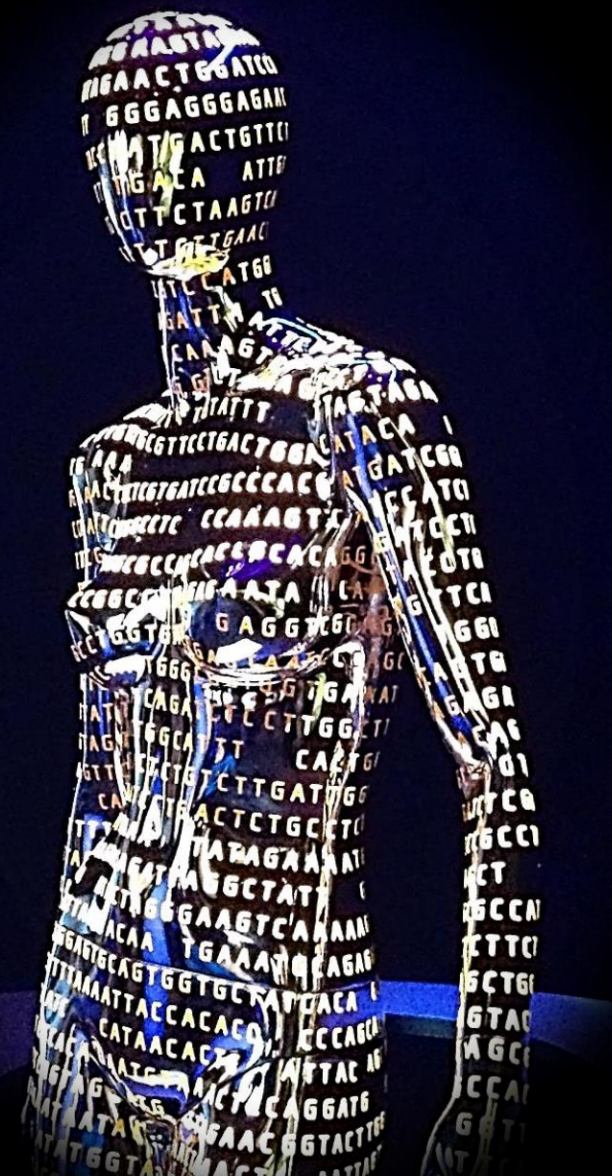
- Monogenic Diseases
- Candidate Genes studies late 90's (many pitfalls).

- 2000. TECHNICAL ADVANCES


TCACCCATGAATGCTCAC
CTGAAACAAGATGCCATTG
CTGCTGCTCTCCGGGGGAACC
CCACTGGAGGGTGGCCCC
GCATATGCAGGAAGCGG
GCCTCCTGACTTTCCTC
TCCCAGGCCAGTGC
AGCTCGGGAGGTGG

THE HUMAN GENOME PROJECT

First complete sequencing
of human genome.
February, **2001**



GENETICS IN CEREBROVASCULAR DISEASES

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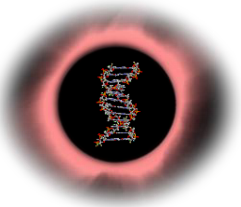


International HapMap Project

Phase I
(2005)
5 populations
1M SNPs

International HapMap Project

Phase II
(2007)
5 populations
3,1M SNPs

International HapMap Project

Phase III
(2009)
11 populations
4,7M SNPs

The data base ***dbSNP*** has **>900 M** human SNPs

Of these, about **10 M** are common (MAF>5%)

1000 Genomes Project
Defining Genetic Variation in People

1st Release
(2012)
1,092 Individuals
12 populations
Whole Genome

1000 Genomes Project
Defining Genetic Variation in People

3rd Release
(2015)
2,504 Individuals
26 populations
Whole Genome

100,000 Genomes Project

(on going)
100,000 Individuals
UK population
Whole Genome

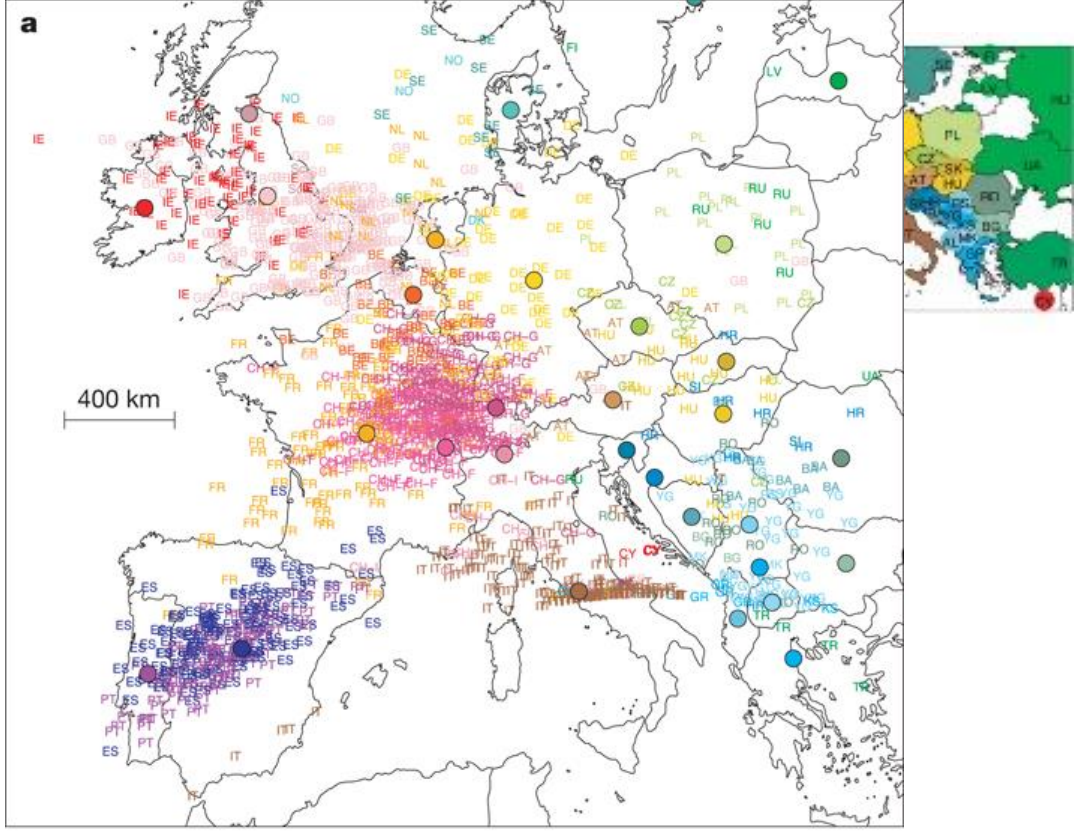
...

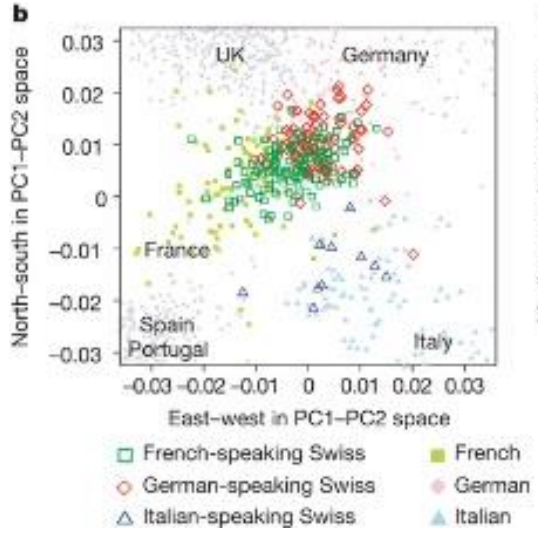
Advances in massive Genotyping

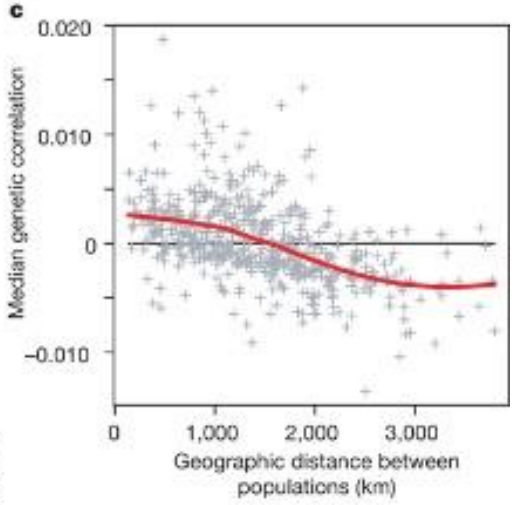
- Chips GWAs (2005): 100.000 SNPs
- Chips GWAs (2015): 5,000.000 SNPs
- Chips GWAS (2017): 5,000,000 SNPs + rare variants + CNVS
- …

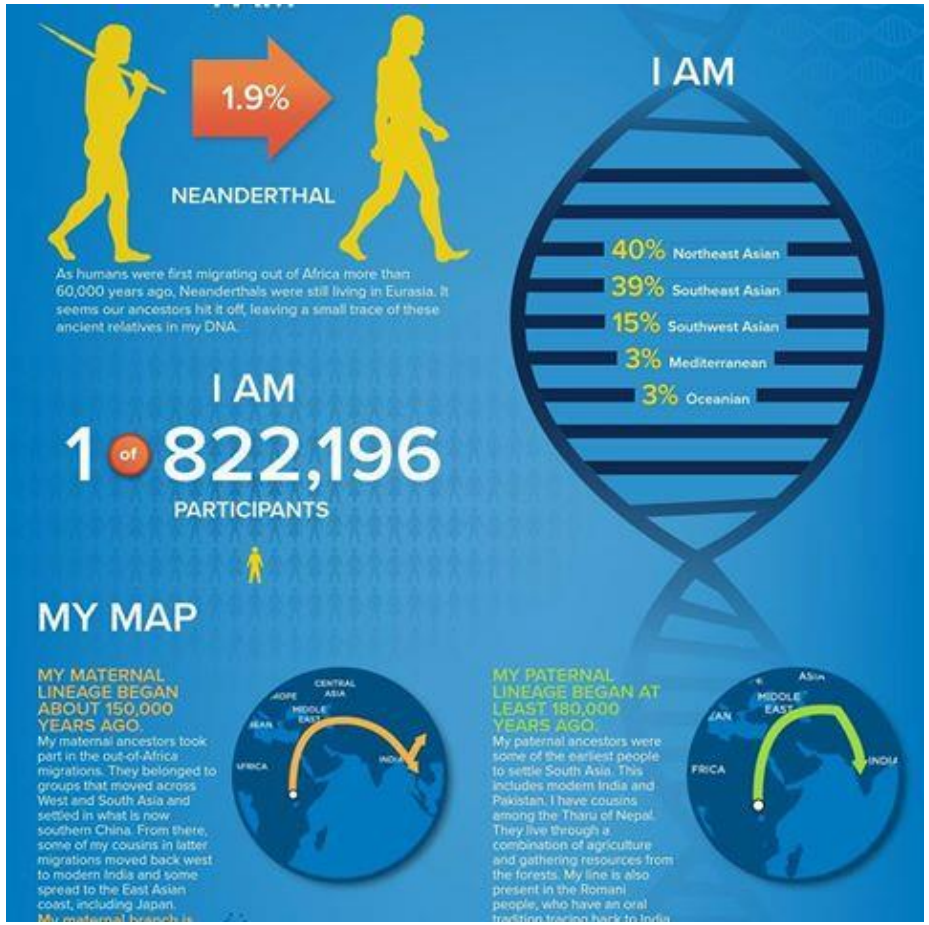
Concepts:

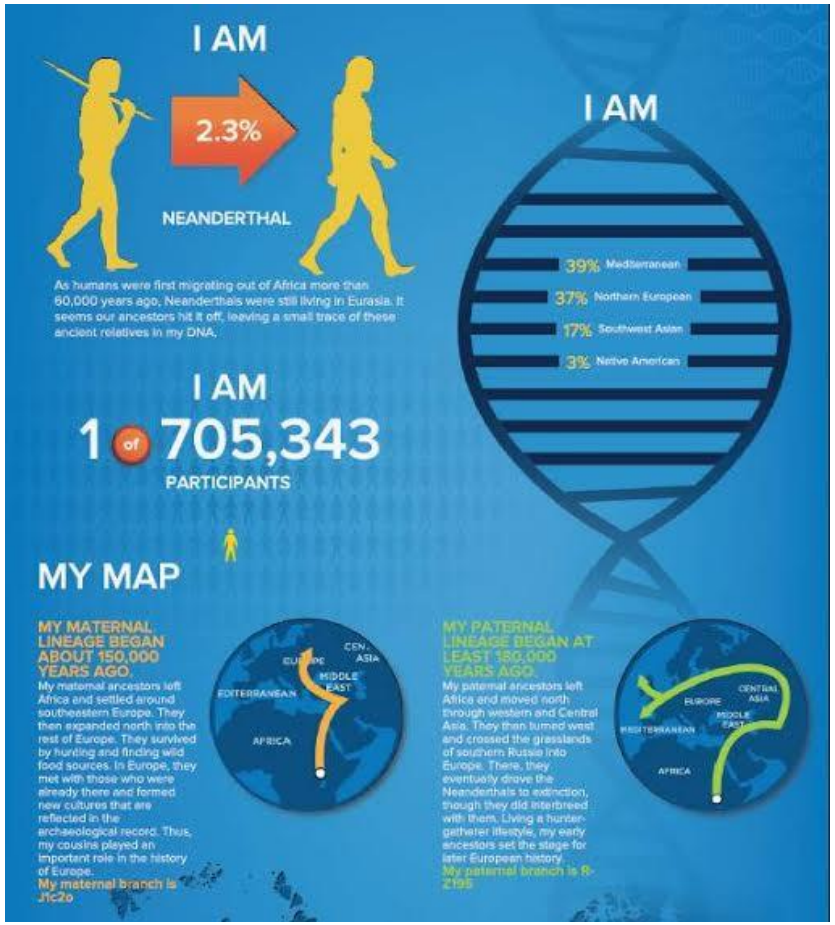
- Identity by State (IBS)
- Identity by Descendent (IBD)
- **Principal Components (PC)**








1.9%
NEANDERTHAL
As humans were first migrating out of Africa more than 60,000 years ago, Neanderthals were still living in Eurasia. It seems our ancestors hit it off, leaving a small trace of these ancient relatives in my DNA.
I AM
1 of 822,196
PARTICIPANTS
I AM
40% Northeast Asian
39% Southeast Asian
15% Southwest Asian
3% Mediterranean
3% Oceanian
MY MAP
MY MATERNAL LINEAGE BEGAN ABOUT 150,000 YEARS AGO.
My maternal ancestors took part in the out-of-Africa migrations. They belonged to groups that moved across West and South Asia and settled in what is now southern China. From there, some of my cousins in latter migrations moved back west to modern India and some spread to the East Asian coast, including Japan.
MY PATERNAL LINEAGE BEGAN AT LEAST 180,000 YEARS AGO.
My paternal ancestors were some of the earliest people to settle South Asia. This includes modern India and Pakistan. I have cousins among the Tharu of Nepal. They live through a combination of agriculture and gathering resources from the forests. My line is also present in the Romani people, who have an oral tradition tracing back to India


I AM
2.3%
NEANDERTHAL
As humans were first migrating out of Africa more than 60,000 years ago, Neanderthals were still living in Eurasia. It seems our ancestors hit it off, leaving a small trace of these ancient relatives in my DNA.
I AM
1 of 705,343
PARTICIPANTS
I AM
39% Mediterranean
37% Northern European
17% Southwest Asian
3% Native American
MY MAP
MY MATERNAL LINEAGE BEGAN ABOUT 150,000 YEARS AGO.
My maternal ancestors left Africa and settled around southeastern Europe. They then expanded north into the rest of Europe. They survived by hunting and finding wild food sources. In Europe, they met with those who were already there and formed new cultures that are reflected in the archaeological record. Thus, my cousins played an important role in the history of Europe.
My maternal branch is J1c2o
MY PATERNAL LINEAGE BEGAN AT LEAST 150,000 YEARS AGO.
My paternal ancestors left Africa and moved north through western and Central Asia. They then turned west and crossed the grasslands of southern Russia into Europe. There, they eventually drove the Neanderthals to extinction, though they did interbreed with them. Living a hunter-gatherer lifestyle, my early ancestors set the stage for later European history.
My paternal branch is R-Z195

GENETICS IN CEREBROVASCULAR DISEASES

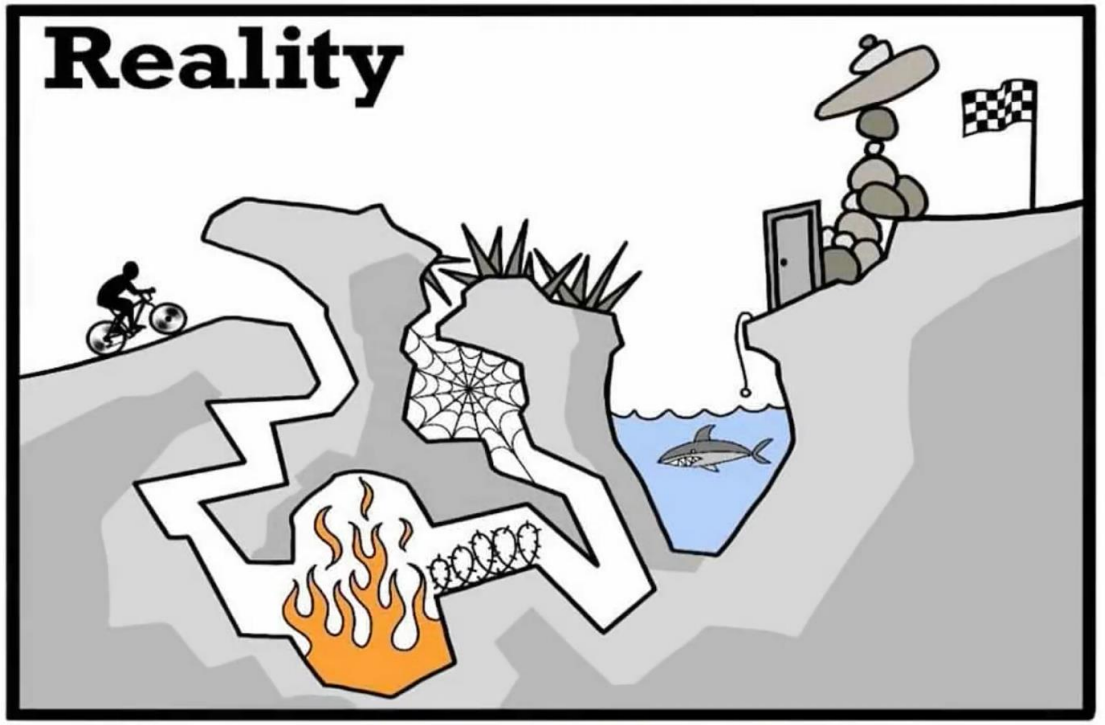
BRIEF HISTORICAL JOURNEY

- Monogenic Diseases

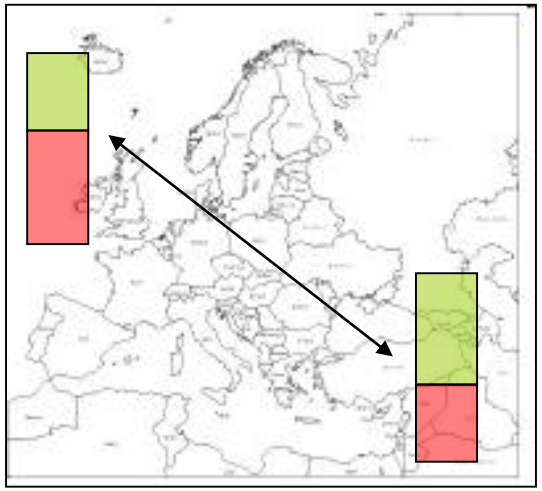
- Candidate Genes studies late 90's (many pitfalls).

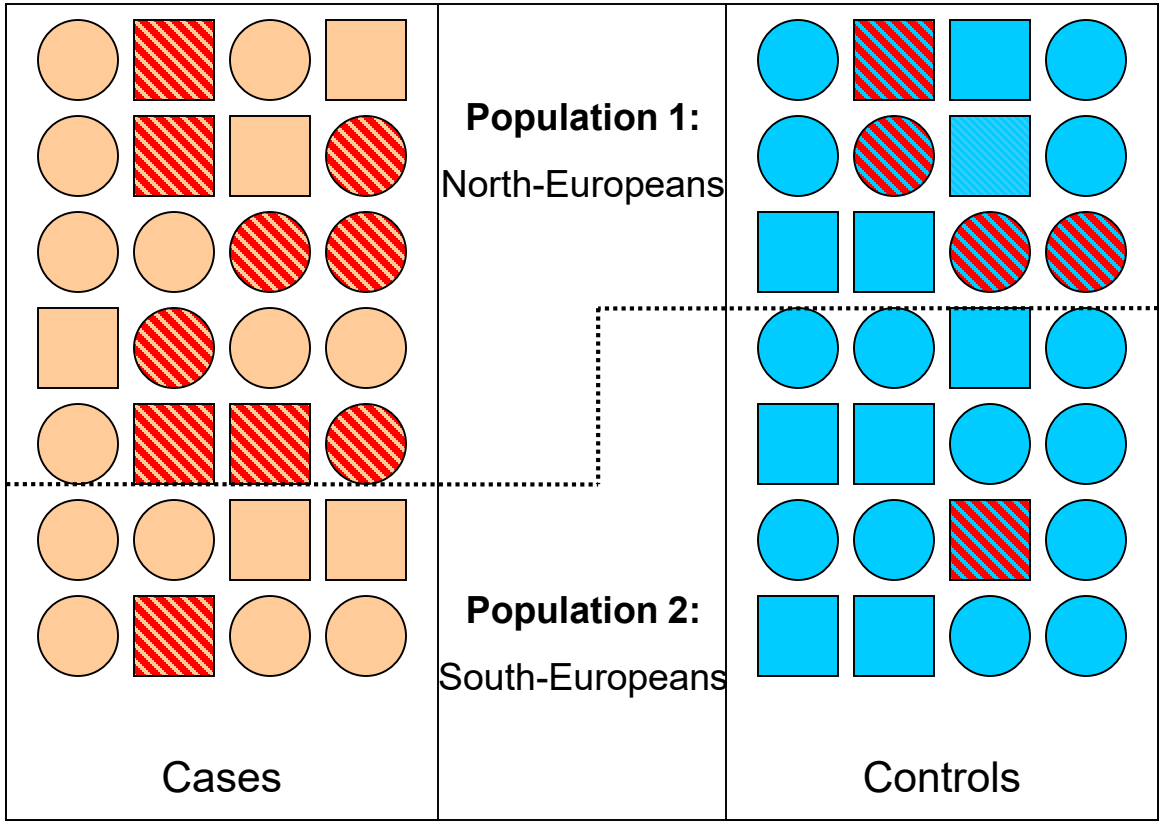
- 2000. TECHNICAL ADVANCES

- PROBLEMS/DIFFICULTIES (Complex designs, sample size, missing Heritability, complex interactions, genomic dynamics….



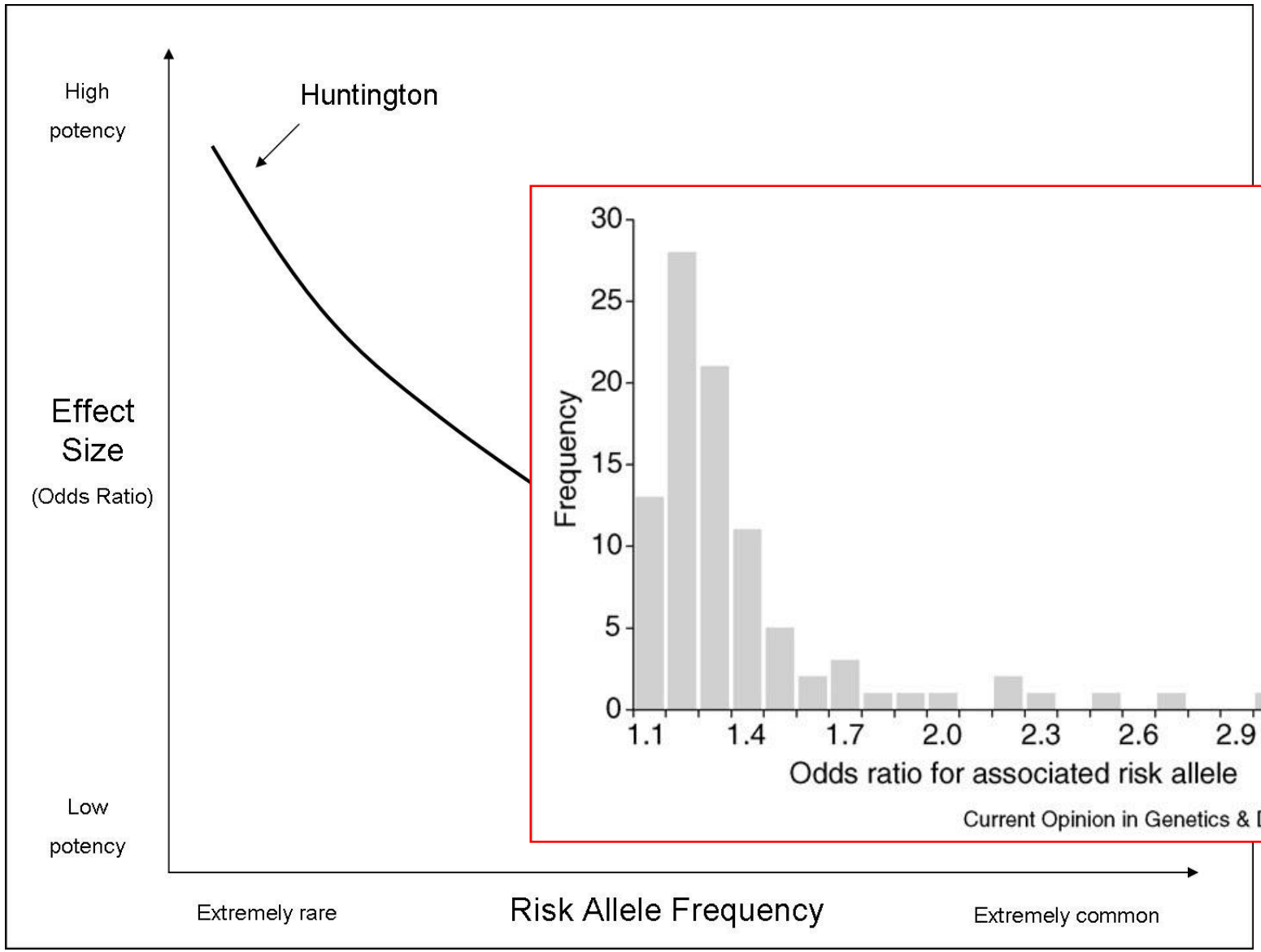
Population Stratification





Genetics in Complex Diseases

Common Variants. Low effects



CONSORTIA

GIANT CONSORTIUM

Genetic Investigation of ANthropometric Traits

ICBP

International Consortium for Blood Pressure

GeneStroke

CONSORCIO ESPAÑOL DE GENÉTICA DEL ICTUS

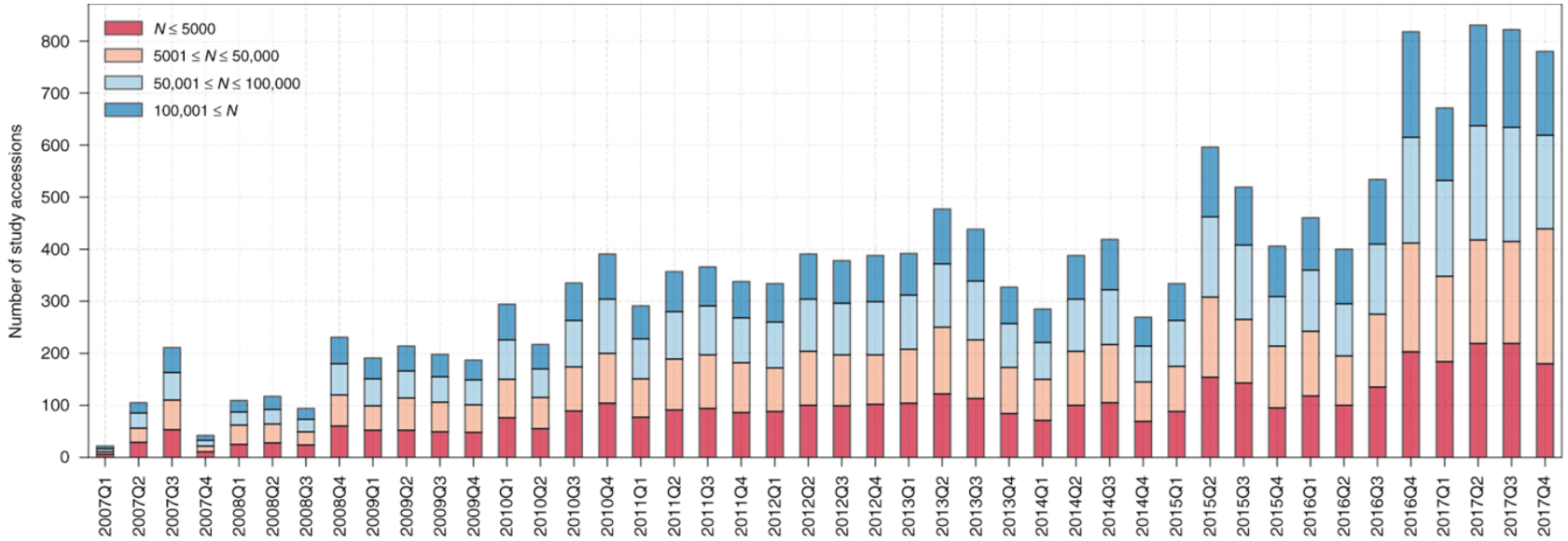
ISGC

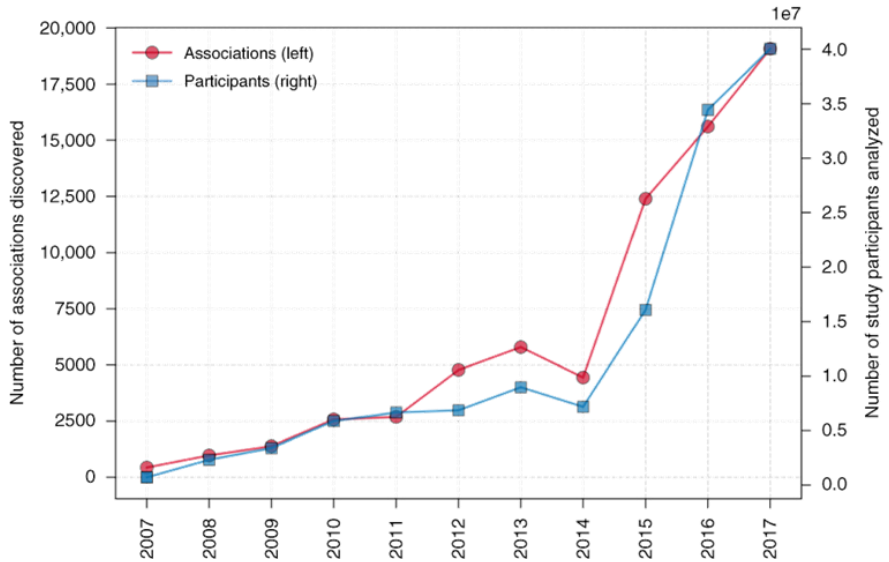
International Stroke Genetics Consortium

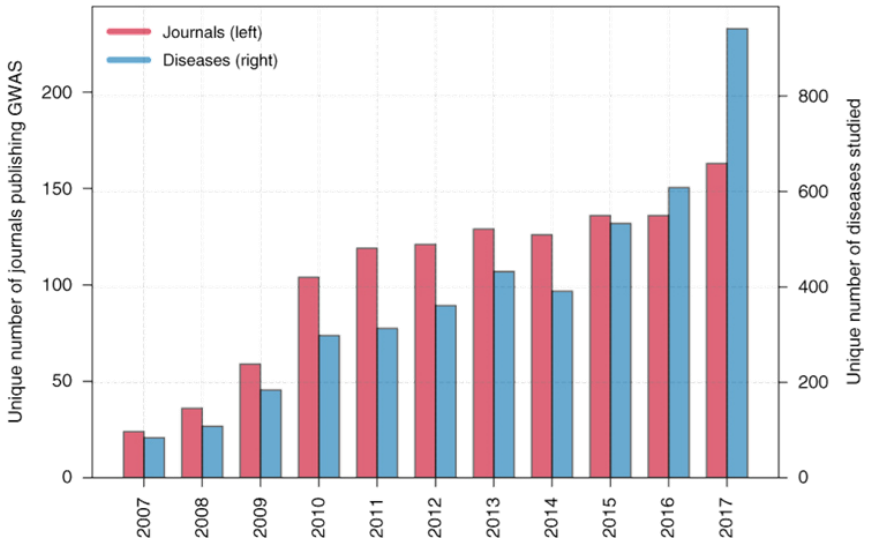
CARDIoGRAMplusC4D

MIGen

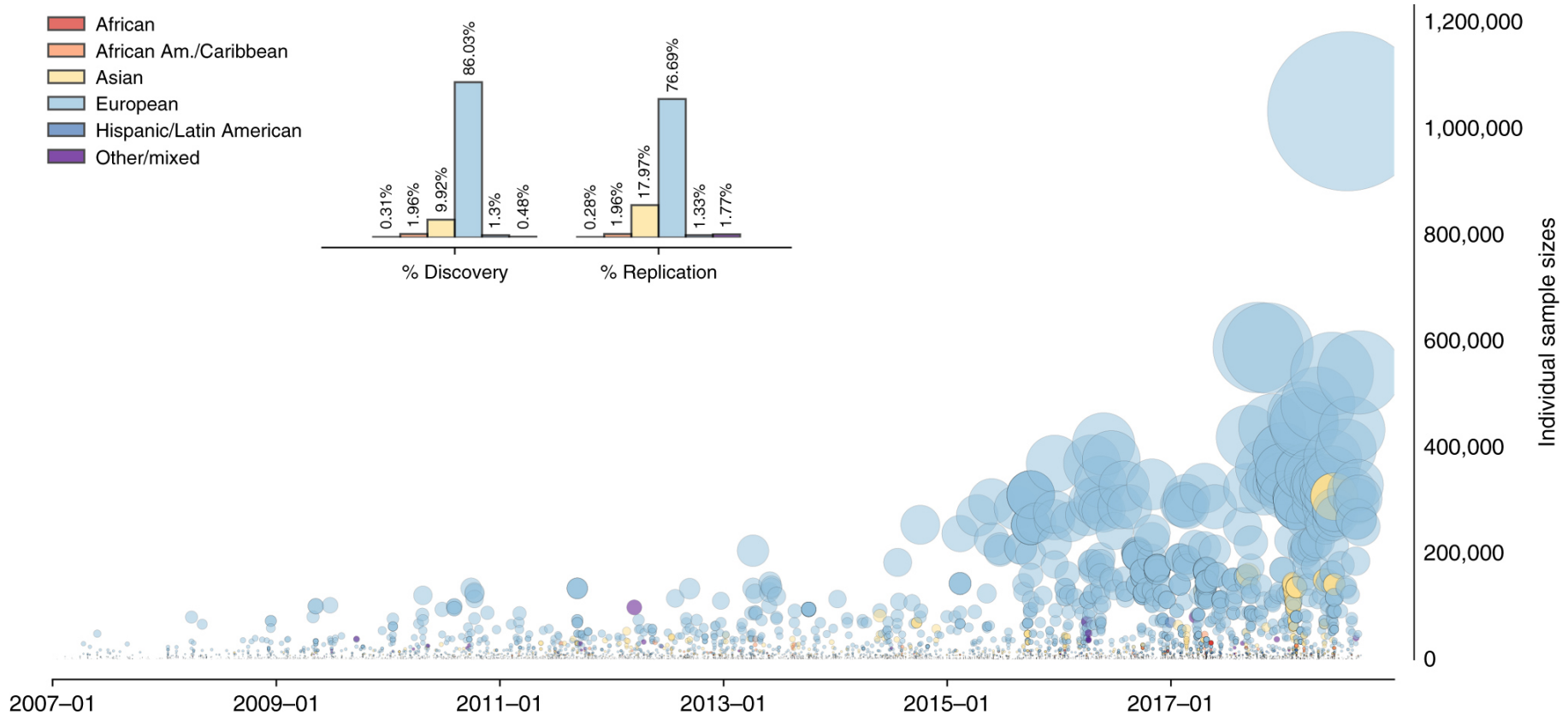
Myocardial Infarction Genetics Consortium







Melinda C Mills, et al. Commun. Biol. 2019 Jan 7;2:9. doi: 10.1038/s42003-018-0261-x. eCollection 2019.



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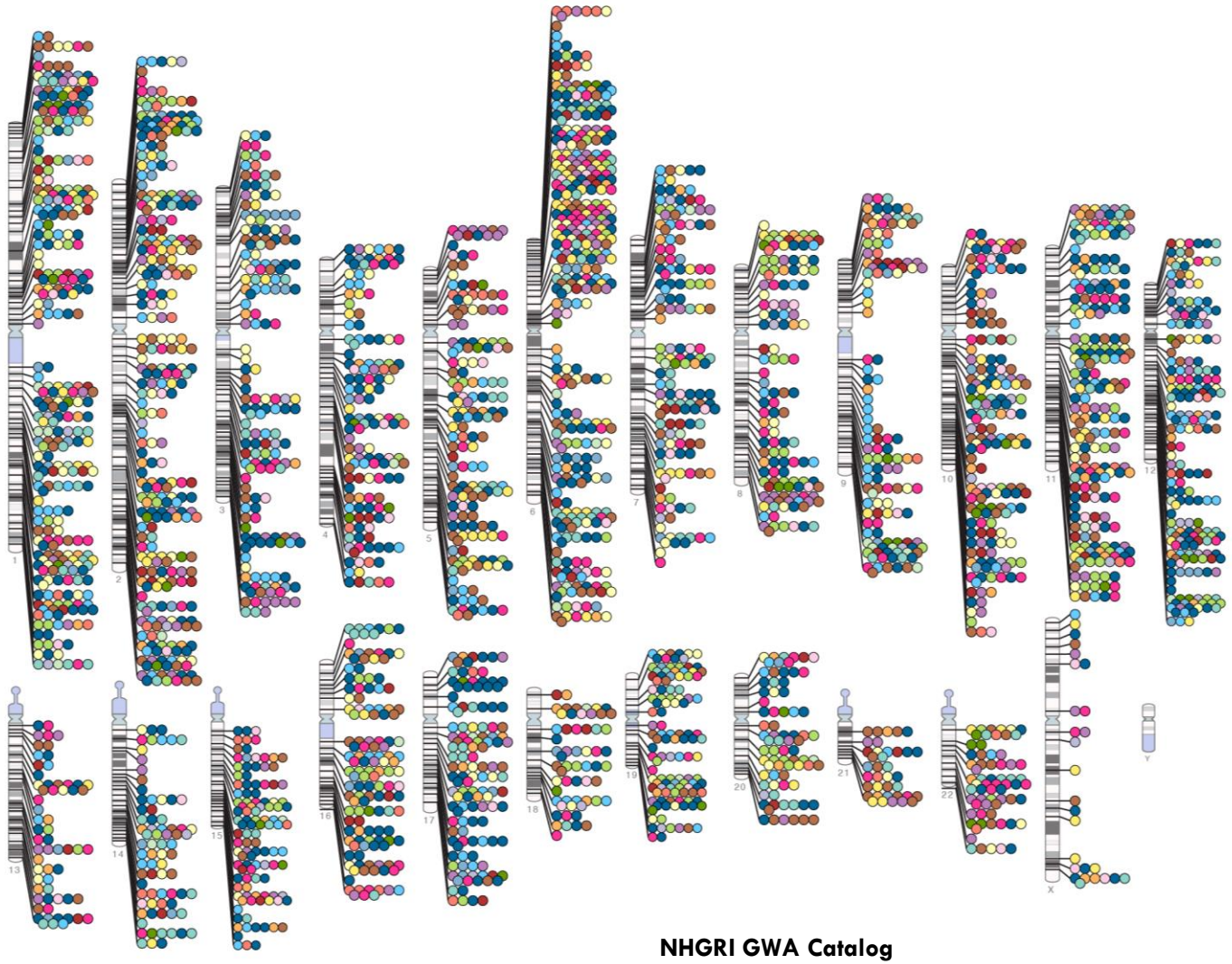


NHGRI GWA Catalog
www.genome.gov/GWAStudies
www.ebi.ac.uk/fgpt/gwas/

National Human Genome Research Institute

EMBL-EBI

http://www.ebi.ac.uk/gwas/

http://www.ebi.ac.uk/gwas/diagram

What we know until now…

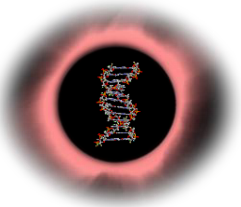
GENETICS ON RISK OF ICH

MONOGENIC DISEASES

TABLE 5 | Genetics variants/genes related to perinatal and pediatric ICH and ICH of Mendelian forms.

Ref.	Gene name/Loci and Abb.	Variants	OR (95% CI)	Study population	Cases/Control	Study type	Notes
He et al. (2019)	Factor VII (*F7*)	IVS7 + 1G	–	Asian	2 cases	Case report	P-ICH
Cheng et al. (2019)	μ-opioid receptor (*OPRM1*)	A118G	1.55 (1.00–2.39)	Asian	167/163	Candidate gene: CCS	P-ICH
Demmert et al. (2015)	Fucosyltransferase 2 (*FUT2*)	G428A	1.20 (0.99–1.40)	European	2404	Candidate gene: PCS	P-ICH
Unal et al. (2014)	Protein C (*PROC*)	T903C	–	European	1case	Case report	P-ICH
Berber et al. (2018)	Vitamin K epoxide reductase complex subunit 1 (*VKORC1*)	G1639A	3.63 (1.32–9.94)	European	51/51	Candidate gene: CCS	P-ICH
Herrmann et al. (2005)	Factor X (*F10*)	Gly380Arg	–	European	6 cases	Case report	P-ICH
Göpel et al. (2002)	Factor XIII (*F13*)	Val34Leu	–	European	832	Candidate gene: PCS	P-ICH
Van Broeckhoven et al. (1990)	Amyloid precursor protein (*APP*)	–	–	European	2 families (20 cases)	Case series	HCHWA-D
Palsdottir et al. (1988) and Jensson et al. (1989)	Cystatin C (*CST3*)	–	–	European	8 families (22 cases)	Case series	HCCAA and ICH
Denier et al. (2004)	Krev interaction trapped protein 1 (*KRIT1*)	–	–	European	64 families (202 cases)	Case series	CCM and ICH
Cottin et al. (2007)	Activin receptor-like kinase 1 (*ACVRL1*) and Endoglin (*ENG*)	–	–	European	126 cases	Case series	HHT and ICH
Gould et al. (2006)	Collagen type IV alpha 1 chain (*COL4A1*)	–	–	European	1 family (11 cases)	Case series	BSVD1 and ICH

CCS, case control study; PCS, prospective cohort study; ICH, intracerebral hemorrhage; P-ICH, perinatal and pediatric ICH; HCHWA-D, human hereditary cerebral hemorrhage with amyloidosis of the Dutch type; HCCAA, hereditary cystatin C amyloid angiopathy; CCM, cerebral cavernous malformations; HHT, hereditary hemorrhagic telangiectasia; BSVD1, brain small vessel disease 1; Ref., reference; Abb., Abbreviation; OR, odds ratio.



GENETICS ON RISK OF ICH

SPORADIC COMPLEX DISEASE

TABLE 1 | Genetic variants related to ICH risk in different location (Candidate gene approach).

Ref.	Gene name/Loci and Abb.	Variants	OR (95% CI)	Study population	Cases/Control	Study type	Notes
Pera et al. (2008)	Glutathione peroxidase 1 (*GPX1*)	C593T	2.36 (1.31–4.26)	European	192/192	Candidate gene: CCS	LICH
Biffi et al. (2012)	Complement C3b/C4b receptor 1 (*CR1*)	rs6656401	1.61 (1.19–2.17)	American	369 (89 CAA-ICH)/324	Candidate gene: CCS	CAA-ICH
Dardiotis et al. (2017)	Integrin subunit alpha V (*ITGAV*)	rs7565633	0.56 (0.37–0.86) (Dom)	European	443/572	Candidate gene: CCS	LICH
Chen et al. (2012)	Protein kinase C eta (*PRKCH*)	1425 G/A	1.73 (1.01– 2.9)	Asian	303 (266 DICH/37 LICH)/381	Candidate gene: CCS	LICH
Chen et al. (2008)	Angiotensin I converting enzyme (*ACE*)	*ACE* I/D and A240T (T-D)	2.7 (1.1–6.5)	Asian	217 DICH/283	Candidate gene: CCS	DICH in females
Chen et al. (2015)	Tissue metalloproteinase inhibitor 2 (*TIMP2*)	rs7503607	2.45 (1.37–4.38) (Add)	Asian	396 DICH/376	Candidate gene: CCS	DICH in subjects ≥ 65 years, especially in males
		rs7503726	0.29 (0.10–0.84) (Rec)				DICH in females ≥ 65 years
Chen et al. (2015)	Matrix Metalloproteinase 2 (*MMP2*)	rs2285053	2.91 (1.02–8.31) (Rec)	Asian	396 DICH/376	Candidate gene: CCS	DICH in subjects ≥ 65 years
Ho et al. (2015)	*MMP9*	rs3787268	0.48 (0.27–0.86)	Asian	326 DICH/439	Candidate gene: CCS	DICH in subjects ≥ 65 years
		rs2250889	0.48 (0.27–0.84)				DICH in males < 65 years
Ho et al. (2015)	*TIMP1*	rs4898	0.35 (0.15–0.81)	Asian	326 DICH/439	Candidate gene: CCS	DICH in males ≥ 65 years
Chen et al. (2010)	Tumor necrosis factor (*TNF*)	G-308A	2.6 (1.3– 5.3)	Asian	260 DICH/368	Candidate gene: CCS	DICH in males
		C-863A	0.5 (0.2–0.9)				DICH in females

CCS, case control study; Dom, dominant; Rec, recessive; Add, additive; LICH, lobar spontaneous intracerebral hemorrhage; DICH, deep spontaneous intracerebral hemorrhage; CAA-ICH, cerebral amyloid angiopathy-related intracerebral hemorrhage; Ref., reference; Abb., Abbreviation; OR, odds ratio.

GENETICS ON RISK OF ICH

SPORADIC COMPLEX DISEASE

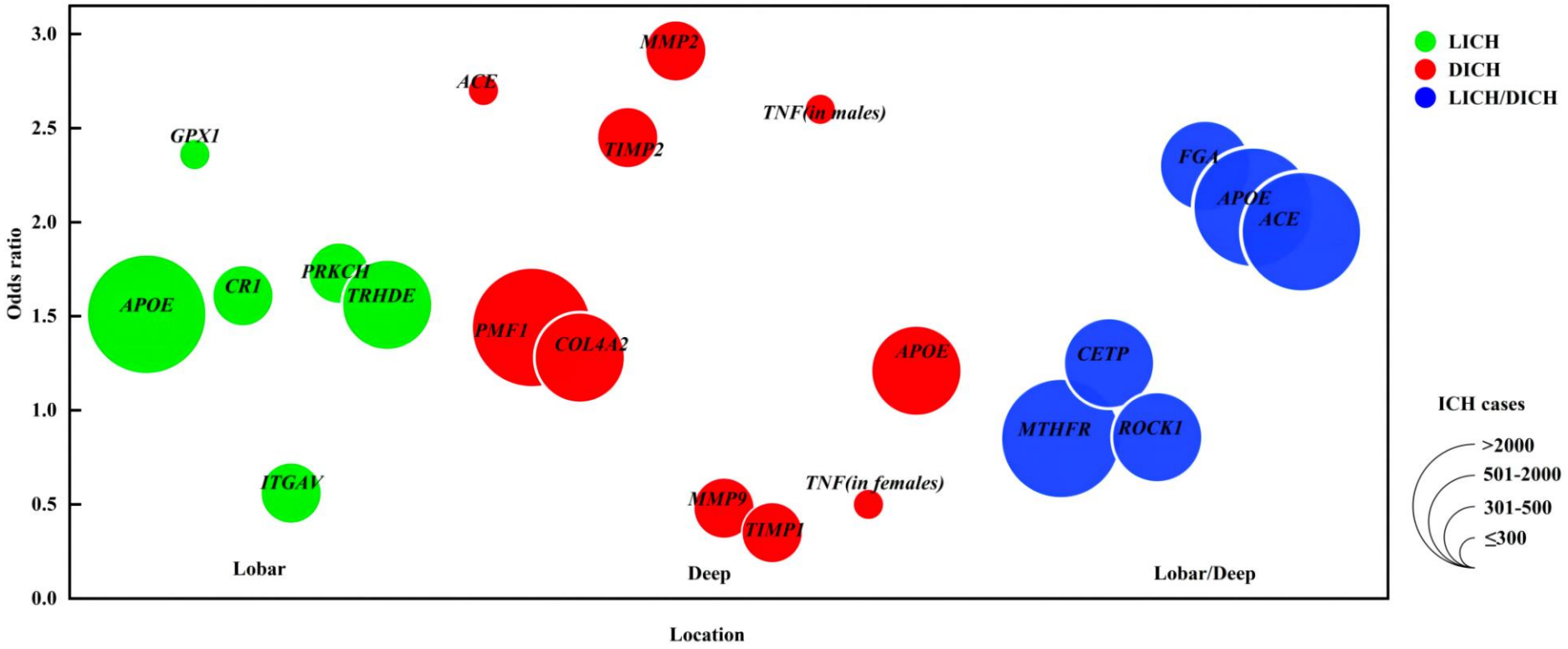
TABLE 2 | Genetic variants related to ICH risk in different location (GWAS).

Ref.	Gene Name/Loci and Abb.	Variants	OR (95% CI)	Study population	Cases/Control	Study type	Notes
Marini et al. (2019)	Apolipoprotein E (*APOE*)	*APOE* ε4 *APOE* ε2	1.51 (1.23–1.85) 1.49 (1.24–1.80)	American, European	6195 (2305 LICH)/6929	GWAS with meta-analysis	LICH
Biffi et al. (2010)	*APOE*	*APOE* ε4	1.21 (1.08–1.36)	European	1081 DICH/3657	GWAS with meta-analysis	DICH
Woo et al. (2014)	*12q21.1*	rs11179580	1.56	European, American	1545 (664 LICH/881 DICH)/1481	GWAS	LICH
Woo et al. (2014)	*1q22*	rs2984613	1.44	European, American	3226/3742	GWAS with meta-analysis	DICH
Rannikmäe et al. (2017)	Collagen type IV alpha 2 chain (*COL4A2*)	rs4771674	1.28 (1.13–1.44)	European	1878/2830	GWAS with meta-analysis	DICH

GWAS, genome-wide association studies; LICH, lobar spontaneous intracerebral hemorrhage; DICH, deep spontaneous intracerebral hemorrhage; Ref., reference; Abb., Abbreviation; OR, odds ratio.

GENETICS ON RISK OF ICH

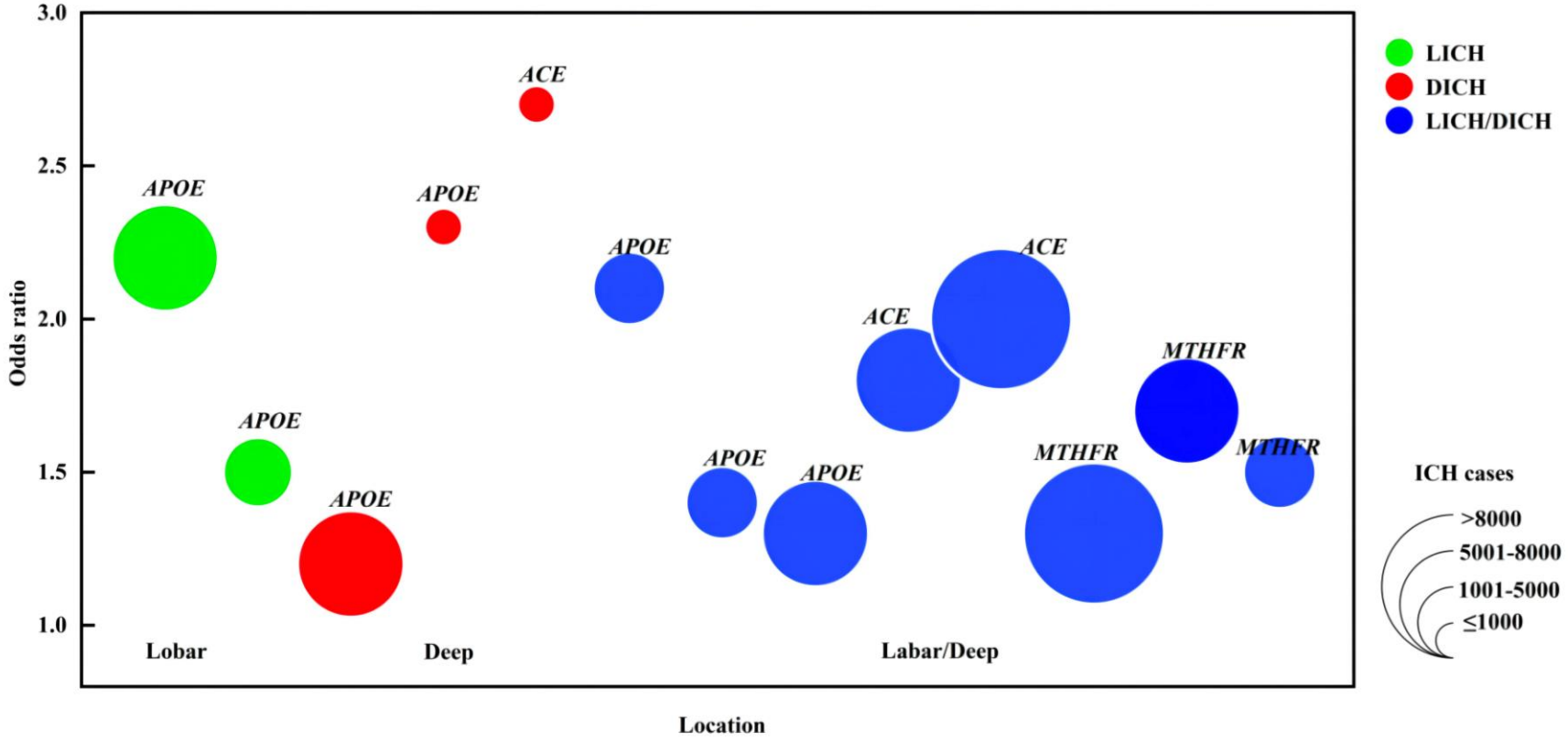
Gene variants associated with ICH risk



Genetic loci for intracerebral hemorrhage. Colors indicate ICH location. The size of bubbles indicates ICH population.

GENETICS ON RISK OF ICH

*Gene variants associated with ICH risk & **replicated** in multiple studies/populations*



Genetic loci for intracerebral hemorrhage. Colors indicate ICH location. The size of bubbles indicates ICH population.

GENETICS ON RISK OF ICH

TABLE 3 | Well-proved genetic variants related to LICH/DICH risk.

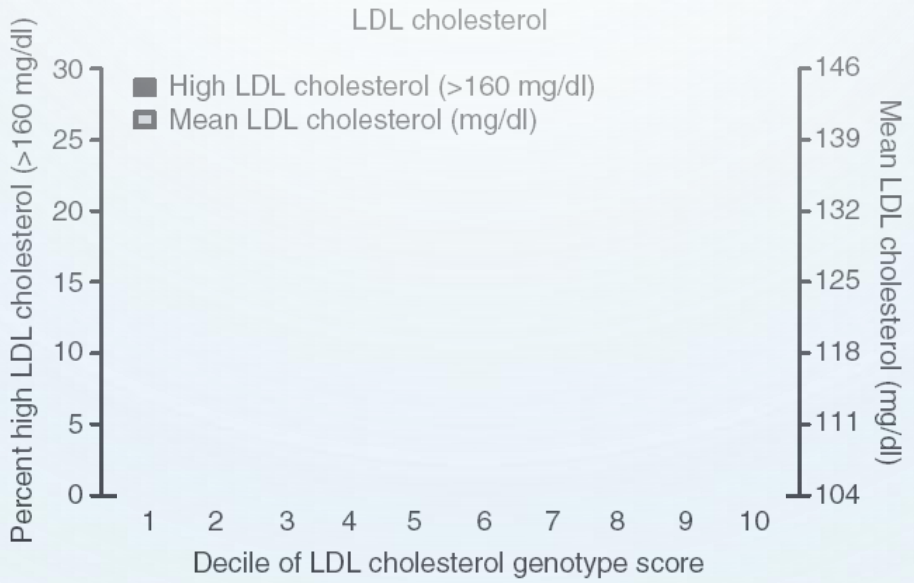
Ref.	Gene name/Locus and Abb.	Variants	OR (95% CI)	Study population	Cases/ Control	Study type (No.)	Notes
Wang et al. (2021)	Methylenetetrahydrofolate reductase (*MTHFR*)	C677T C	0.85	European, American, African, and Asian	3679/9067	Candidate gene: Meta-analysis	ICH
Anderson et al. (2016)	Cholesteryl ester transfer protein (*CETP*)	rs173539	1.25	European, American	1149/1238	GWAS with replication	ICH
Yang et al. (2018)	Rho-associated kinase 1 (*ROCK1*)	rs288980	0.857 (Add)	Asian	607/2443	Candidate gene: CCS	ICH
Jagiełła et al. (2014)	Fibrinogen alpha chain (*FGA*)	Thr312Ala	2.3 (1.1–4.8) (Dom)	European	503/774	Candidate gene: Meta-analysis	ICH
Yamada et al. (2017)	Serine threonine tyrosine kinase 1 (*STYK1*)	rs138533962	111.3 (33.0–694.6)	Asian	673/9158	GWAS	ICH
Chen and Hu (2016)	*APOE*	*APOE* ε4	2.08 (1.57–2.75)	Asian	2018/2143	Candidate gene: Meta-analysis	ICH
Nie et al. (2019)	*APOE*	*APOE* ε2	1.21 (1.07–1.37)	European, American, African, and Asian	1642/5545	Candidate gene: Meta-analysis	ICH in European and American
		APOE ε4	1.32 (1.14–1.52)				
Li et al. (2020)	*ACE*	*ACE* I/D	1.95 (1.57–2.43) (Rec); 0.7 (0.6–0.82) (Dom); 0.68 (0.6–0.7) (All)	European, American, African, and Asian	3839/5353	Candidate gene: Meta-analysis	ICH in Asians

APOE, apolipoprotein E; ACE, angiotensin I converting enzyme; Dom, dominant; Rec, recessive; Add, additive; All, allelic; GWAS, genome-wide association studies; ICH, spontaneous intracerebral hemorrhage; CCS: case control study; Ref., reference; Abb., Abbreviation; OR, odds ratio.

Genetic Scores

Genetic Scores

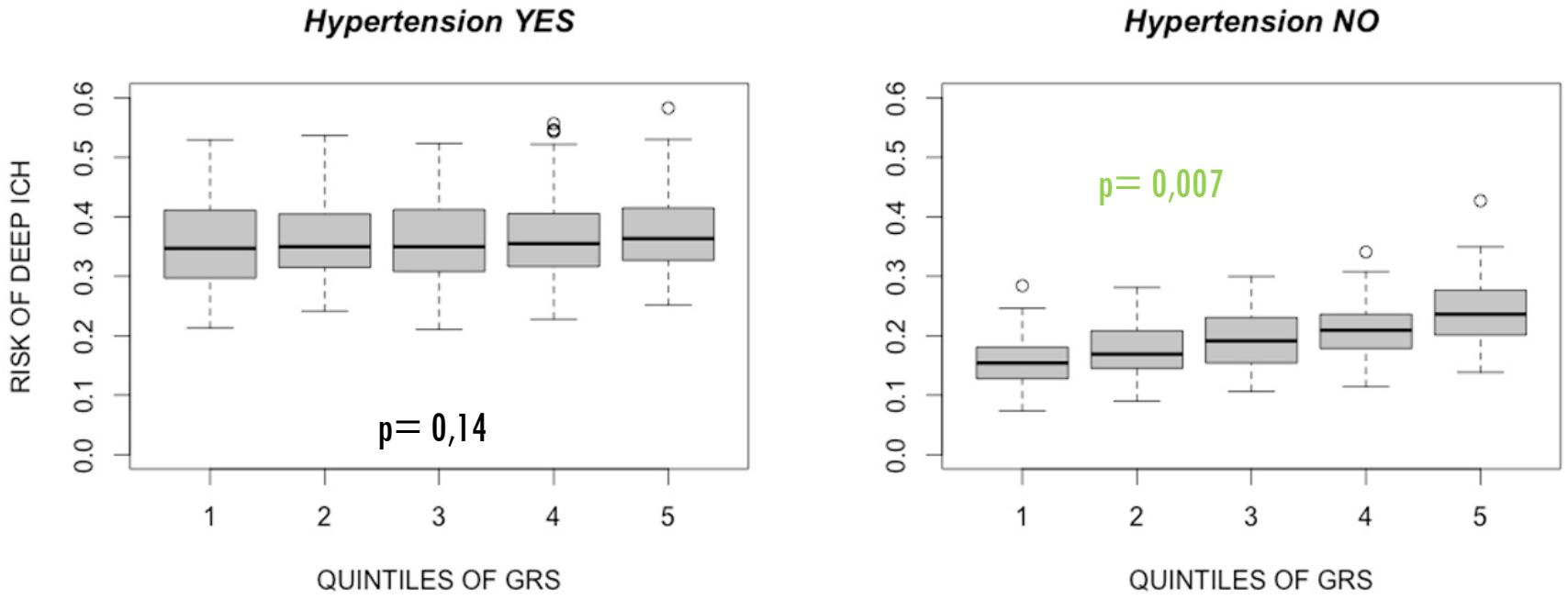
Kathiresan et al.
Nature Genetics
2009



GENETICS ON RISK OF ICH

GRS with Alleles for **Hypertension** Increases Risk **of Deep ICH**.

This effect is predominantly observed in non-HTN subjects.



Falcone et al. 2012

GENETICS ON RISK OF ICH

GRS with Alleles for **Hypertension** Increases Risk **of Deep ICH.**

Georgakis M, Neurology 2020

Figure 2 Mendelian randomization associations between genetically determined blood pressure and risk of stroke and stroke subtypes

Stroke subtype	SBP (10 mmHg increment) OR (95% CI)	*p* value	DBP (5 mmHg increment) OR (95% CI)	*p* value
Any stroke	1.39 (1.33, 1.44)	1.9E–60	1.27 (1.23, 1.32)	1.2E–42
Ischemic stroke	1.41 (1.35, 1.47)	1.3E–53	1.28 (1.24, 1.33)	2.6E–40
Large artery stroke	1.68 (1.54, 1.84)	5.2E–30	1.34 (1.25, 1.44)	1.0E–14
Cardioembolic stroke	1.24 (1.16, 1.34)	9.9E–09	1.17 (1.10, 1.24)	2.7E–06
Small vessel stroke	1.47 (1.36, 1.58)	3.5E–22	1.36 (1.27, 1.45)	7.8E–19
Intracerebral hemorrhage	1.41 (1.11, 1.79)	8.3E–03	1.29 (1.05, 1.57)	0.019
Lobar intracerebral hemorrhage	1.04 (0.77, 1.40)	0.389	0.97 (0.76, 1.25)	0.391
Deep intracerebral hemorrhage	1.73 (1.30, 2.32)	8.3E–04	1.54 (1.21, 1.97)	8.2E–04

Results from the fixed-effects inverse variance weighted analysis. CI = confidence interval; DBP = diastolic blood pressure; OR = odds ratio; SBP = systolic blood pressure.

ICH, GENETICS AND LIPIDS PROFILE

- **Genetically Elevated LDL** Associates with **Lower Risk of ICH.**

With a **causal relationship** after Mendelian Randomization (MR) analyses.

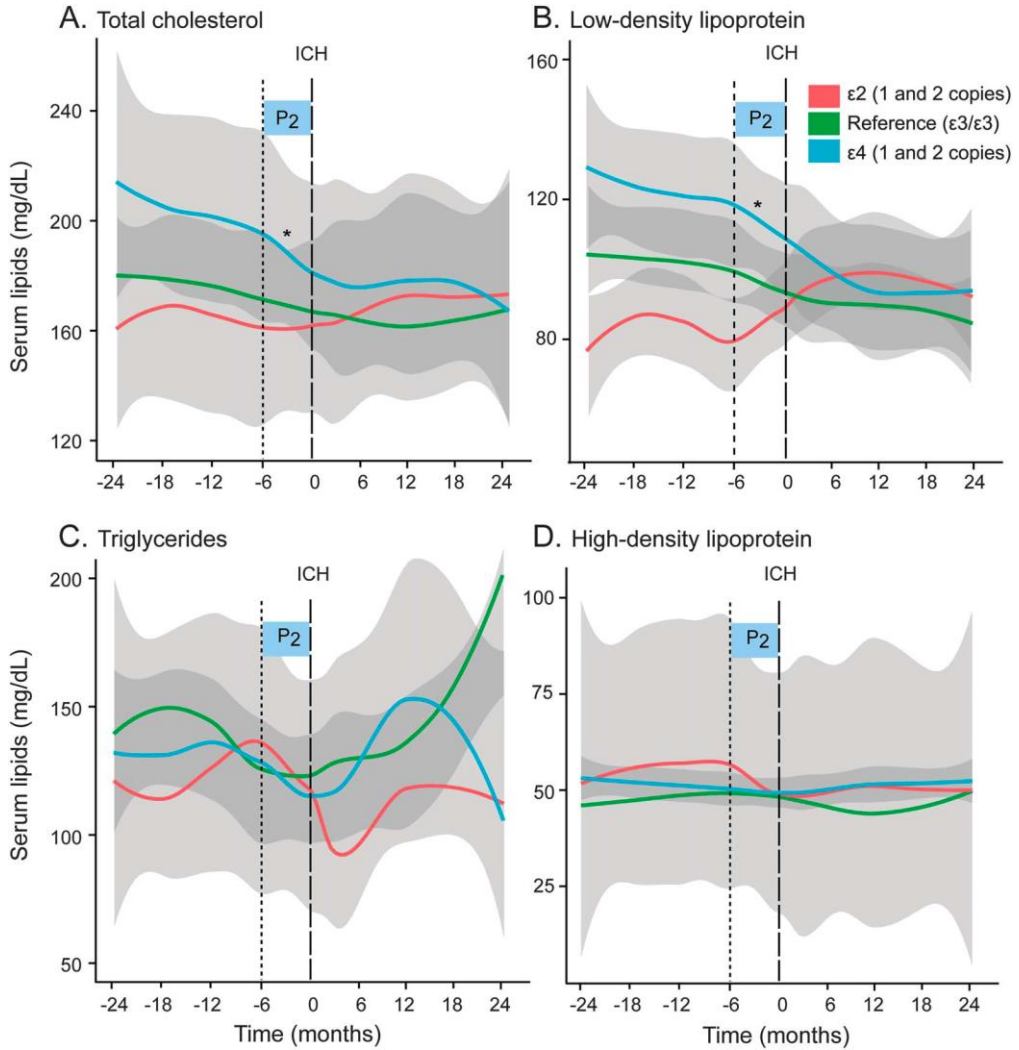
Falcone G, Ann Neurol. 2020
Sun L, Nat. Med 2019

Location-specific results for ICH risk.

Lipid trait	Lobar-ICH = 539 cases			Deep-ICH = 704 cases		
	OR (95%CI)	P	Metanalysis Heterogeneity P	OR (95%CI)	P	Metanalysis Heterogeneity P
Polygenic risk score analysis *						
Total cholesterol	0.89 (0.80 – 0.99)	0.03	0.42	0.94 (0.85 – 1.08)	0.20	0.96
LDL cholesterol	0.81 (0.73 – 0.89)	<0.001	0.96	0.90 (0.82 – 0.99)	0.04	0.99

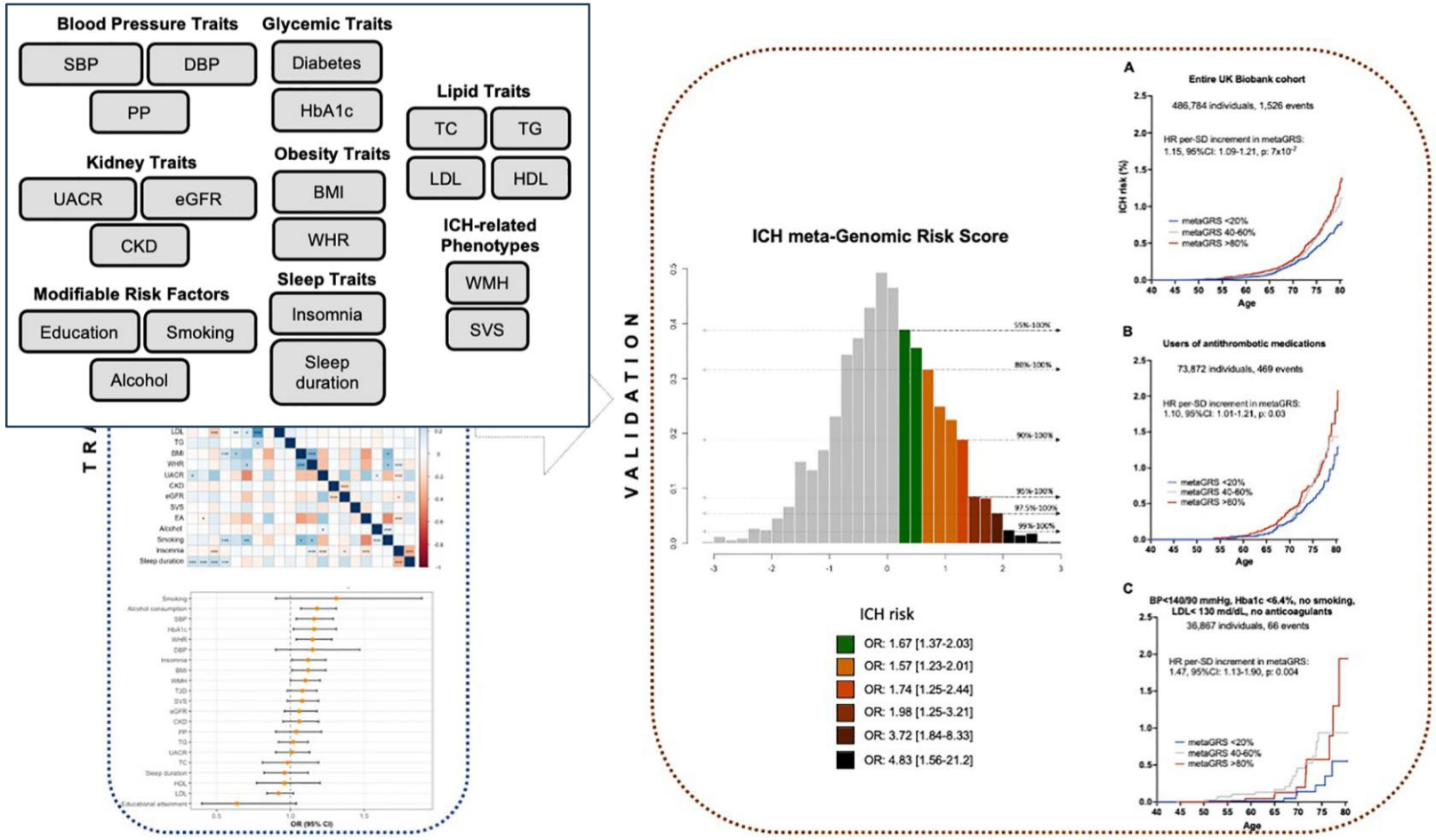
ICH, GENETICS AND LIPIDS PROFILE

- APOE E4 had **decreasing** levels of **TC** and **LDL** 6 months **preceding ICH** (Anderson, 2016)



GENETICS ON RISK OF ICH

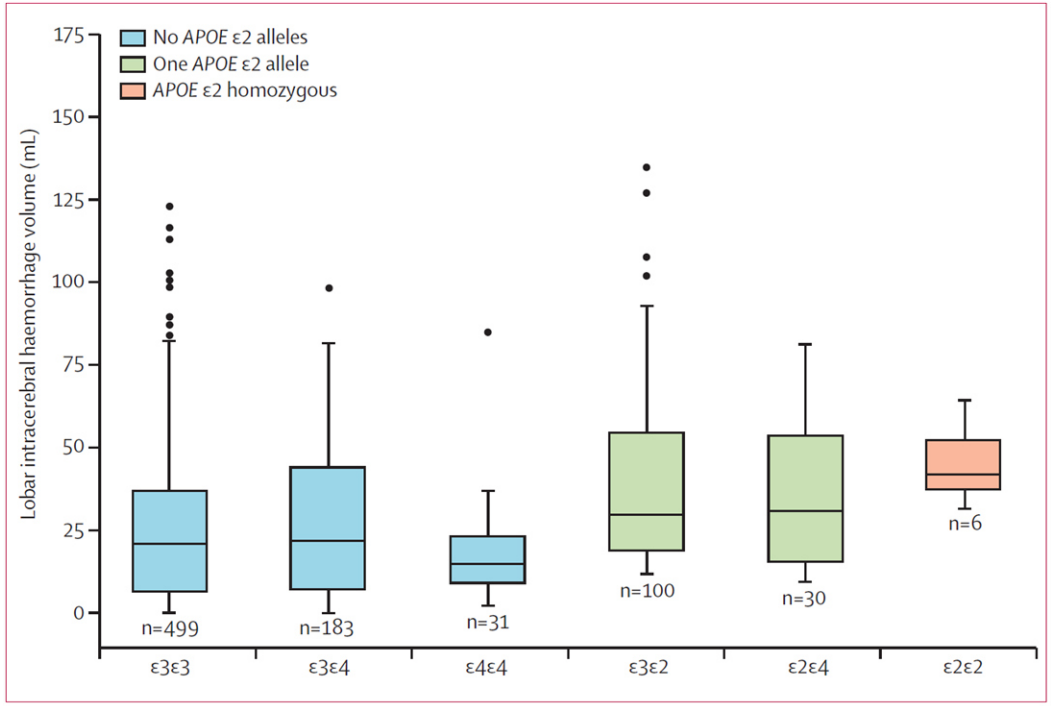
Meta-GRS with Alleles for 21 traits associated with ICH risk better stratify risk of ICH



Myserlis P, Stroke 2023

APOE

HAPLOTYPES:



- E3: Reference
- E2: Risk L-ICH — Larger Volume (mortality, poor outcome) — Protect Alzheimer's D
- E4: Risk L-ICH, Brain MicroBleeds — Decrease LDL pre ICH — Risk Alzheimer's D

Anderson, 2016
Biffi, 2012
Knol, Neurology, 2020

GENETICS ON ICH EVOLUTION AND OUTCOME

TABLE 4 | Genetic variants related to ICH hematoma and outcome.

Ref.	Gene name/Locus and Abb.	Variants	OR (95% CI)	Study population	Sample size	Study type	Notes	
Brouwers et al. (2012a)	*APOE*	APOE ε2	2.72 (1.19–6.23)	European	510 (265 LICH)	Candidate gene: PCS	Hematoma expansion after LICH	L-ICH Expansion
Brouwers et al. (2012b)	*APOE*	APOE ε2	2.09 (1.05–4.19)	European	371 (196 LICH)	Candidate gene: PCS	Spot sign in LICH	
Biffi et al. (2011)	*APOE*	APOE ε2	1.52 (1.25–1.85) for disability; 1.50 (1.23–1.82) for mortality	European, American	2,025 (849 LICH)	GWAS with meta-analysis	Hematoma volume, poor outcome and mortality at 3 m in LICH	L-ICH Volume Bad outcome, Mort
Math et al. (2019)	*APOE*	APOE ε4	2.60 (1.25–5.41)	American, European	192	Candidate gene: meta- analysis	Poor outcome	Bad Outcome
Appelboom et al. (2013)	von Willebrand Factor (*VWF*)	rs216321	–	American	82	Candidate gene: PCS	Relative hematoma growth	ICH Expanison
Marini et al. (2018)	*17p12*	rs11655160	0.17 for aGCS; 1.94 for disability	European	634 (335 DICH)	GWAS with meta-analysis	Hematoma volume, aGCS and poor outcome at 3 m in DICH	D-ICH Volume Bad outcome, Mort
Marini et al. (2018)	*22q13*	rs9614326	–	European	394	GWAS	Hematoma volume	ICH Volume
Murthy et al. (2015)	Haptoglobin (*HP*)	HP2-1/2-2	0.13 (0.03–0.71)	American	94	Candidate gene: OCS	Favorable outcome at 30 days	Good Oucome
El Husseini et al. (2018)	Interleukin 6 cytokine family signal transducer (*IL6ST*)	rs10940495	0.16 (0.03–0.87)	American	54	Candidate gene: OCS	Poor outcome at 6 m	Bad outcome
Appelboom et al. (2011)	Complement factor H (*CFH*)	rs1061170	7.62 (1.40–41.6) for mortality at discharge; 1.822 (1.025–3.239) for mortality at 6 m; 1.822 (1.025–3.239) for survival duration	American, Asian, Hispanic	82	Candidate gene: OCS	Mortality at discharge and 6 m, and survival duration	Bad outcome
Xia et al. (2018)	Collagen type IV alpha 2 chain (*COL4A1*)	rs532625	3.557 for disability at 3 m; 4.264 for disability at 6 m; 3.568 for mortality/disability at 6 m	Asian	181 HICH	Candidate gene: PCS	Disability at 3 and 6 m; poor outcome at 6 m	Bad outcome
Falcone et al. (2013)	Blood pressure-related alleles	42 SNPs	1.71 (1.05–2.80)	European	323 (135 DICH)	GWAS	Hematoma volume, poor outcome at 3 m in DICH	D-ICH Volume Bad outcome, Mort

GENETICS ON ICH EVOLUTION AND OUTCOME

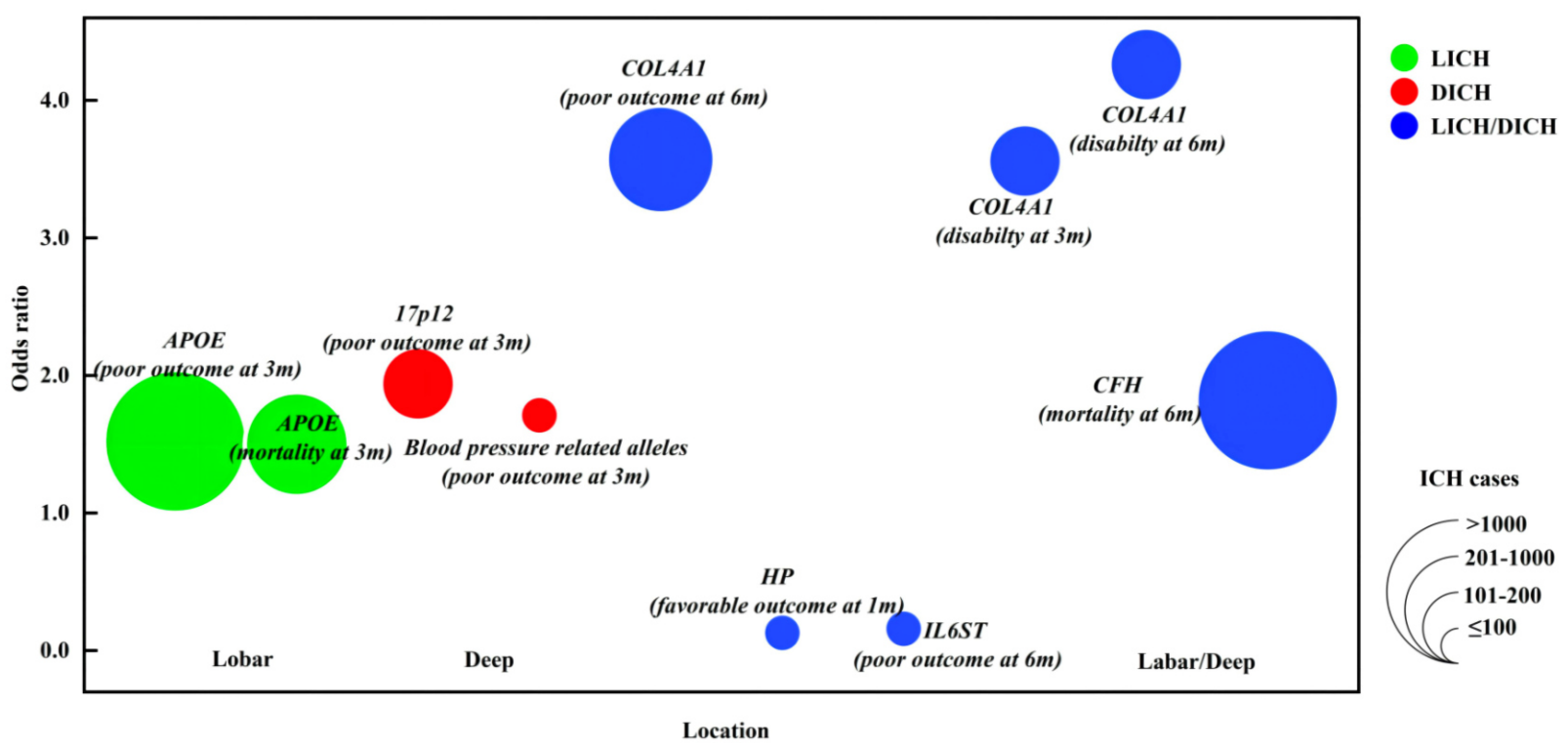
GRS constructed with **Hypertension** related **alleles** was associated with **D-ICH Prognosis**.

Each additional SD of the score was associated with:

- 28% increase in D-ICH **volume**
- 71% increase of **poor clinical outcome** (mRS>2 at 90 days)

Falcone et al. (2013)

GENETICS ON ICH EVOLUTION AND OUTCOME

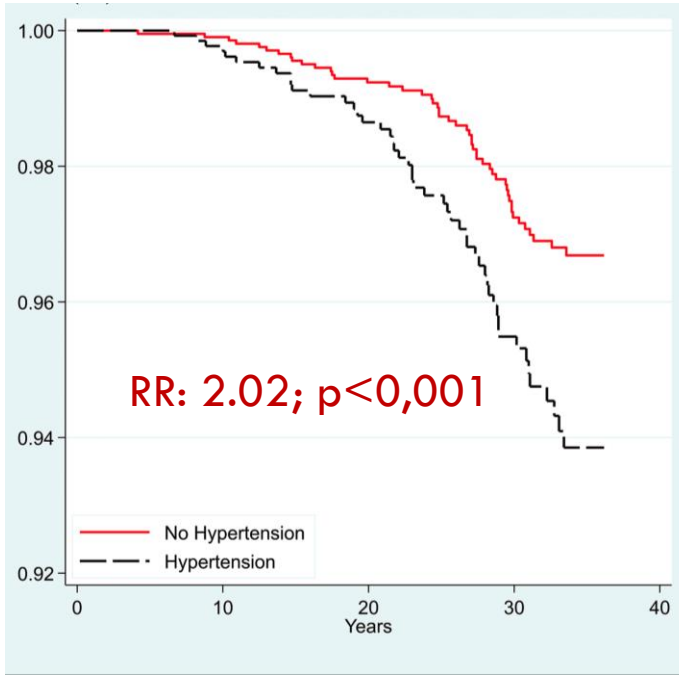


Genetic loci for intracerebral hemorrhage. Colors indicate ICH location. The size of bubbles indicates ICH population.

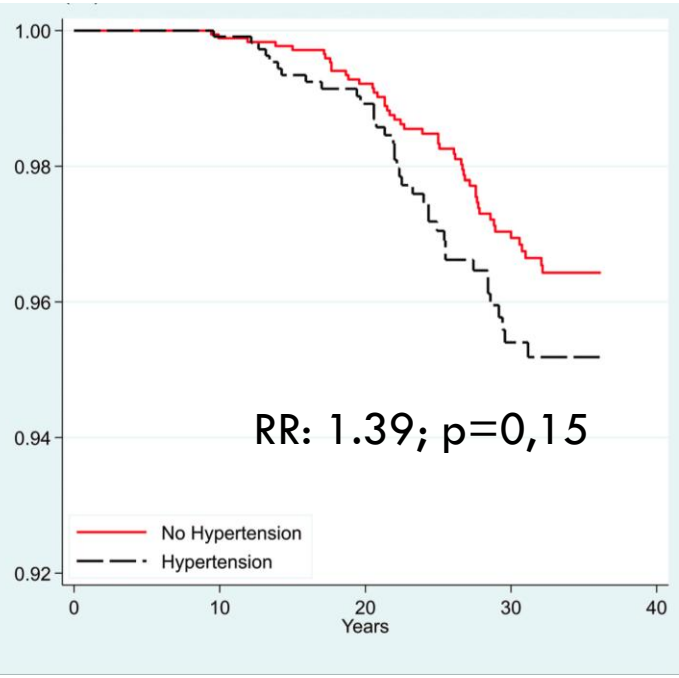
GENETICS MEDIATING ON RISK FACTORS FOR ICH

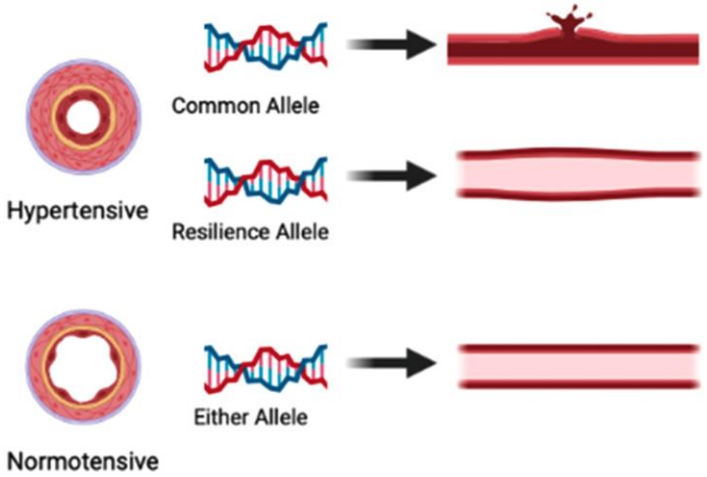
FOXO3 longevity allel (rs2802292) attenuates HT effect on risk of ICH.

FOXO3 (TT) (n=3460)



FOXO3 (TG/GG) (n=3009)





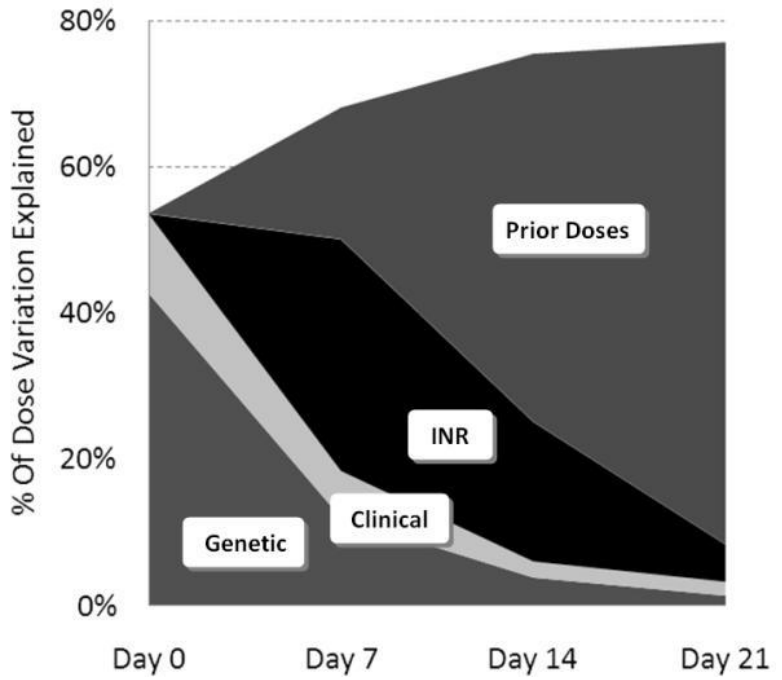
Nakagawa et al. (2022)
(n=6469)

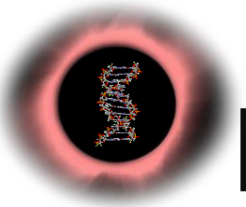
FARMACOGENETICS

ANTICOAGULANTS

- Polymorphisms in **CYP2C9** (encoding cytochrome P450 2C9) and **VKORC1** (encoding vitamin K epoxide reductase complex subunit 1) explain about **30-40% of total variation** in final **dose** of **warfarin.**
- In February 2010, FDA provided **genotype-specific dose ranges,** and suggested that **genotypes be taken into consideration** when prescribing the drug.

Percentage of dose variation explained at weekly time points.





FARMACOGENETICS

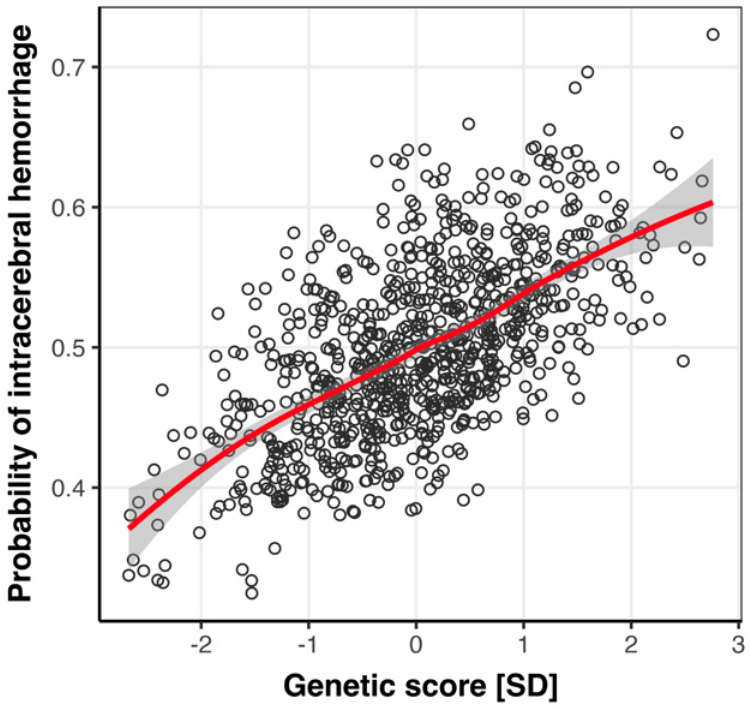
- CYP2C9 (encoding cytochrome P450 2C9) and VKORC1 (encoding vitamin K epoxide reductase complex subunit 1) associated with acenocoumarol (oral anticoagulant) maintenance dose were also associated with ICH risk. (Cullell, 2020)

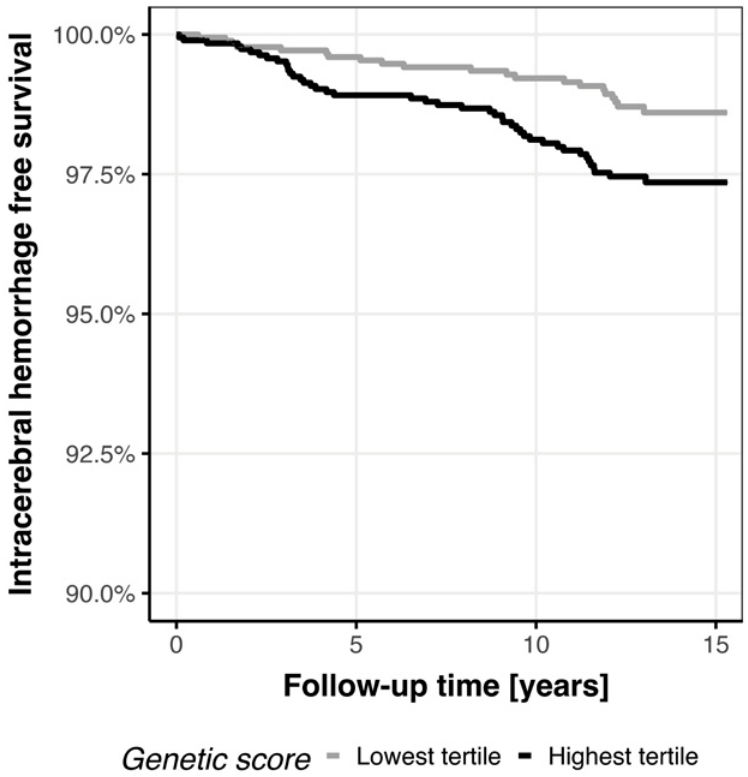
- APOE +2 and APOE +4 were associated with warfarin related L-ICH (Falcone et al., 2014).

FARMACOGENETICS

- GRS from ICH GWAS improves stratification of Anticoagulant risk for ICH. (Mayerhofer et al, 2023)

  - N=5530 Anticoagulant users from UK Biobank





CRS (HAS-BLED) predicted ICH with HR= 1.24 per point

CRS+GRS predicted ICH with HR= 1.33 per point

CRS+GRS had 19% improvement in high-risk classification and a net reclassification improvement of 0.10.

FARMACOGENETICS

DIRECT-ACTING ORAL ANTICOAGULANTS (DOACS)

Variants en ABCB1 and CES1 genes alter concentration of **Dabigatran.**

- CES1
  - **rs2244613:**
    - ↓**active metabolite**
    - ↓ **Bleedings.** No increasing ischemic events
  - **rs8192935 :**
    - ↑**active metabolite**

- ABCB1:
  - **rs4148738:**
    - ↑**active metabolite**

Johanna Raymond , ET AL. J. Pers. Med. 2021, 11, 37. https://doi.org/10.3390/jpm11010037

FARMACOGENETICS

DIRECT-ACTING ORAL ANTICOAGULANTS (DOACS)

Table 2. Pharmacokinetic variations in DOACs based on genetic polymorphisms of *CES1*, *ABCB1*, *CYP3A4*, *CYP3A5*, *ABCG2*, and *SLCO1B1*.

Gene SNP Allelic Change Amino Acid Change Frequency	*DABIGATRAN*	*RIVAROXABAN*	*APIXABAN*	*EDOXABAN*	*BETRIXABAN*
CES1 **rs2244613** intron: C > A - C = 0.266 [13]	↓ [trough] by 15% per mutated allele ($p = 1.2 \times 10^{-8}$) [14] ↓ risk of bleeding ($p = 7 \times 10^{-5}$) [14] ↓ bleeding compared to warfarin for mutated alleles ($p = 0.002$) [14] Not associated with ischemic events [14] ↓ [trough] of dabigatran ($p = 0.04$) HTZ = 2% and MT = 3% [15] No effect on AUC (NS) or [peak] (NS) [16] ↓ [trough] for mutated alleles carriers (NS) [17]	*NI*	*NI*	*NI*	*NI*
CES1 **rs8192935** intron: T > C - T = 0.420 [13]	↓ [peak] by 12% ($p = 3.2 \times 10^{-8}$) [14] Not associated with ischemic or bleeding events [14] ↓ [trough] ($p = 0.033$) HTZ = 3% and MT «TT» = 11% [15]	*NI*	*NI*	*NI*	*NI*
CES1 **rs71647871** 536 G > A 143 Gly > Glu A = 0.014 [13]	Loss of CES1 function: ↓ by 41% of the transformation of the prodrug and metabolites in dabigatran ($p = 0.026$ for BIBR 951) [12]	*NI*	*NI*	*NI*	*NI*
ABCB1 **rs1128503** 1236 C > T 412 Gly > Gly T = 0.46 [13]	Results not significant for AUC and [peak] of dabigatran Haplotype HTZ: $p = 0.61$ Haplotype MT: $p = 0.58$ [16]	Major bleeding under rivaroxaban for three MT patients [18]	No impact on [trough]/dose ratio for apixaban [19]	*NI*	*NI*
ABCB1 **rs2032582** 2677 G > T/A 893 Ala > Ser/Thr T = 0.42 A = 0.08 [13]	Results not significant for AUC and [peak] of dabigatran Haplotype HTZ: $p = 0.61$ Haplotype MT: $p = 0.58$ [16]	One case of rivaroxaban-induced hemorrhage with homozygous mutated genotypes 'TT' [20] No significant increase of rivaroxaban [peak] [16] Major bleeding under rivaroxaban for three MT patients [18]	No impact on [trough]/dose ratio for apixaban [19] One case of highly increased [peak] and concentration 12 h post dose in a homozygous patient (TT), along with other mutations on *ABCB1* (rs1045642, MT), *ABCG2* (rs2231142, HTZ), and *CYP3A5* (rs776746, MT) [21]	*NI*	*NI*

Johanna Raymond , ET AL. J. Pers. Med. 2021, 11, 37. https://doi.org/10.3390/jpm11010037

FARMACOGENETICS

DIRECT-ACTING ORAL ANTICOAGULANTS (DOACS)

Table 2. *Cont.*

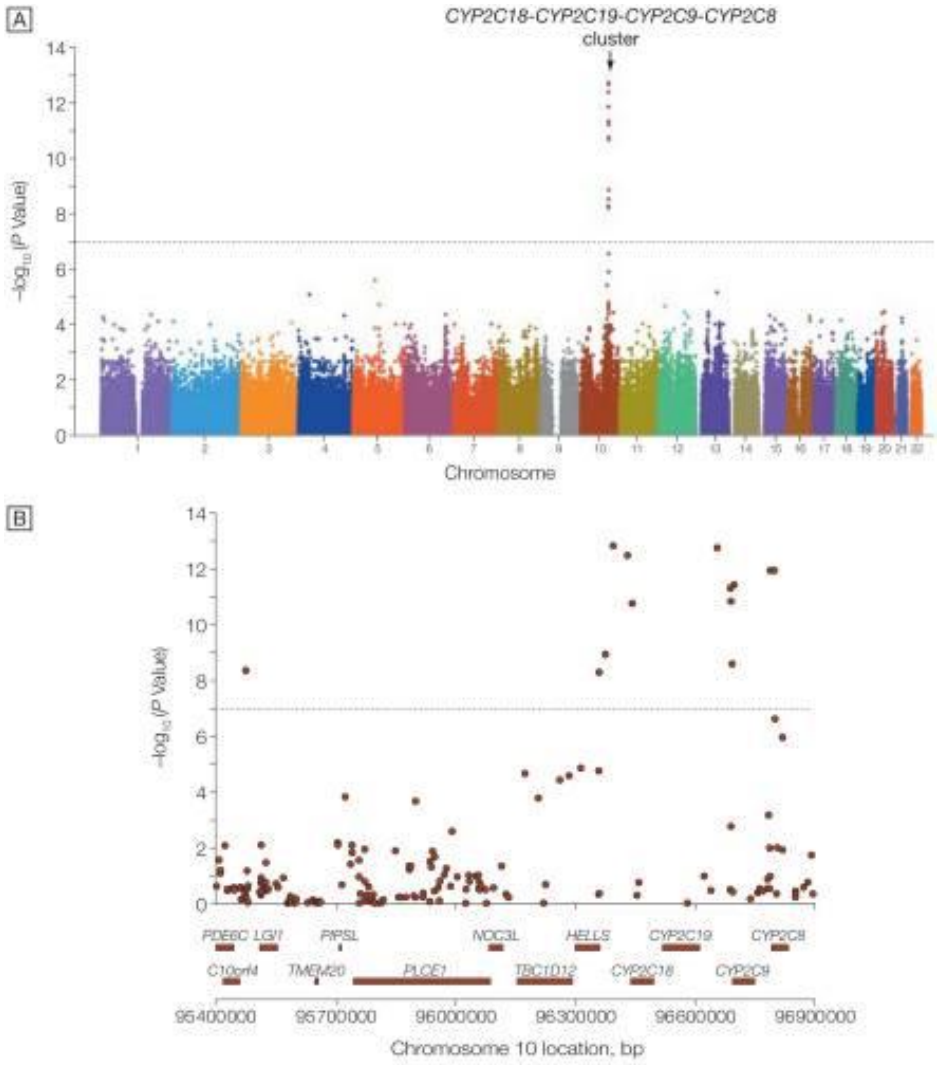
Gene SNP Allelic Change Amino Acid Change Frequency	*DABIGATRAN*	*RIVAROXABAN*	*APIXABAN*	*EDOXABAN*	*BETRIXABAN*
ABCB1 **rs4148738** intron: A > G - G = 0.38 [13]	Associated with ↑ [peak] by 12% ($p = 8.2 \times 10^{-8}$), but not associated with ischemic or bleeding events [14] No effect on [trough] and [peak] of dabigatran [15] Associated with ↑ [peak] of dabigatran [17] No impact on dabigatran pharmacokinetics [25]	Major bleeding under rivaroxaban for three MT patients [18]	Associated with ↑ [peak] of apixaban ($p = 0.048$) [26] No impact on apixaban pharmacokinetics [23]	*NI*	*NI*
CYP3A4 **rs35599367** intron: C > T - T = 0.03 [13]	*NI*	No significant increase of rivaroxaban [peak] in mutated patients compared to wild type (haplotype of ABCB1 rs1045642 and CYP3A4 rs35599367) [22]	*NI*	*NI*	*NI*
CYP3A5 **rs776746** intron: T > C - T = 0.29 [13]	*NI*	*NI*	Significant ↑ of ratio [trough]/dose of apixaban in HTZ or MT patients [19] One case of highly increased [peak] and concentration 12 h post dose in a MT patient, along with other mutations on *ABCB1* (rs2032582 and rs1045642, MT), and *ABCG2* (rs2231142, HTZ) [21] No impact on apixaban pharmacokinetics [23]	*NI*	*NI*
ABCG2 **rs2231142** 421 C > A 141 Gln > Lys A = 0.12 [13]	*NI*	*NI*	Significant ↑ of [trough]/dose ratio of apixaban in MT patients [19] One case of highly increased [peak] and concentration 12 h post dose in an HTZ patient, along with other mutations on *ABCB1* (rs2032582 and rs1045642, MT), and *CYP3A5* (rs776746, MT) [21] ↑ [peak] et [trough] of apixaban [27]	*NI*	*NI*
SLCO1B1 **rs4149056** 521 T > C 174 Val > Ala C = 0.13 [13]	*NI*	*NI*	*NI*	It seems to have no impact on the pharmacokinetics of edoxaban [24]	*NI*

AUC: area under curve; MT: mutated homozygous; HTZ: heterozygous; ↓: decrease; ↑: increase; [peak]: peak concentration; [trough]: trough concentration; NI: no information; NS: non significant.

Johanna Raymond , ET AL. J. Pers. Med. 2021, 11, 37. https://doi.org/10.3390/jpm11010037

FARMACOGENETICS

- **ANTIPLATELETS (AAS)**
  - **PIA1/A2 (GPIIIa)** PIA2 allele carrier was associated with a higher incidence of stroke
  - **COX1:**
  - **COX2 (rs20417):** Carriers showed decreased CVD



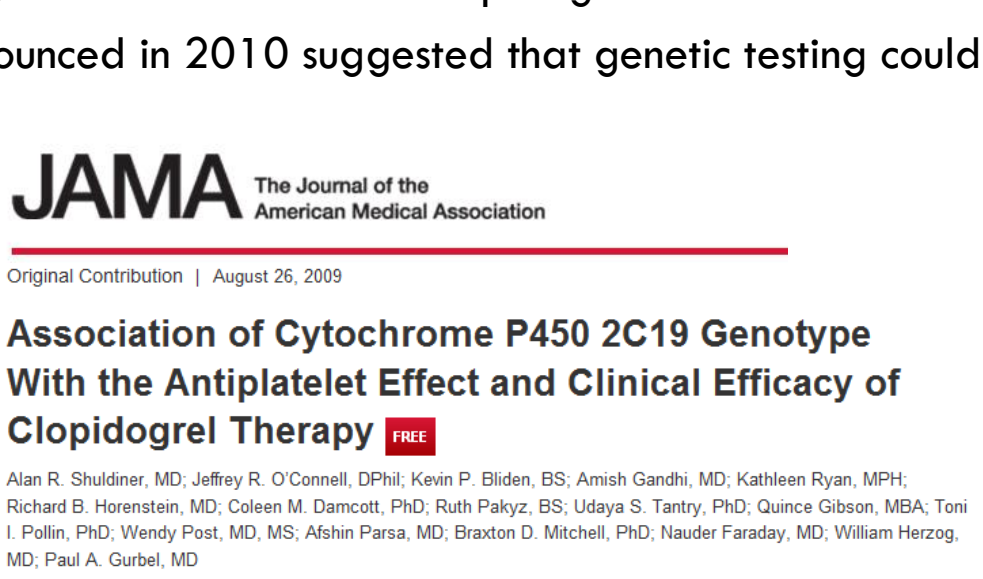
Floyd et al 2014

Ross et al 2014

FARMACOGENETICS

ANTIPLATELETS (Clopidogrel)

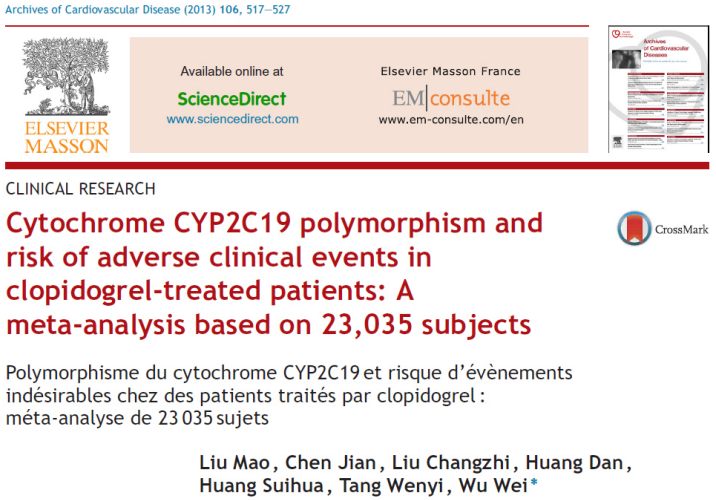
- Carriers of at least one CYP2C19*2 reduced-function allele (about 25 to 30% of the population) have a ↓ **active metabolite** of clopidogrel.
- FDA announced in 2010 suggested that genetic testing could identify individuals slow metabolizers.


JAMA The Journal of the American Medical Association

Original Contribution | August 26, 2009

Association of Cytochrome P450 2C19 Genotype With the Antiplatelet Effect and Clinical Efficacy of Clopidogrel Therapy FREE

Alan R. Shuldiner, MD; Jeffrey R. O'Connell, DPhil; Kevin P. Bliden, BS; Amish Gandhi, MD; Kathleen Ryan, MPH; Richard B. Horenstein, MD; Coleen M. Damcott, PhD; Ruth Pakyz, BS; Udaya S. Tantry, PhD; Quince Gibson, MBA; Toni I. Pollin, PhD; Wendy Post, MD, MS; Afshin Parsa, MD; Braxton D. Mitchell, PhD; Nauder Faraday, MD; William Herzog, MD; Paul A. Gurbel, MD


Archives of Cardiovascular Disease (2013) 106, 517–527

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CLINICAL RESEARCH

Cytochrome CYP2C19 polymorphism and risk of adverse clinical events in clopidogrel-treated patients: A meta-analysis based on 23,035 subjects

Polymorphisme du cytochrome CYP2C19 et risque d'évènements indésirables chez des patients traités par clopidogrel : méta-analyse de 23 035 sujets

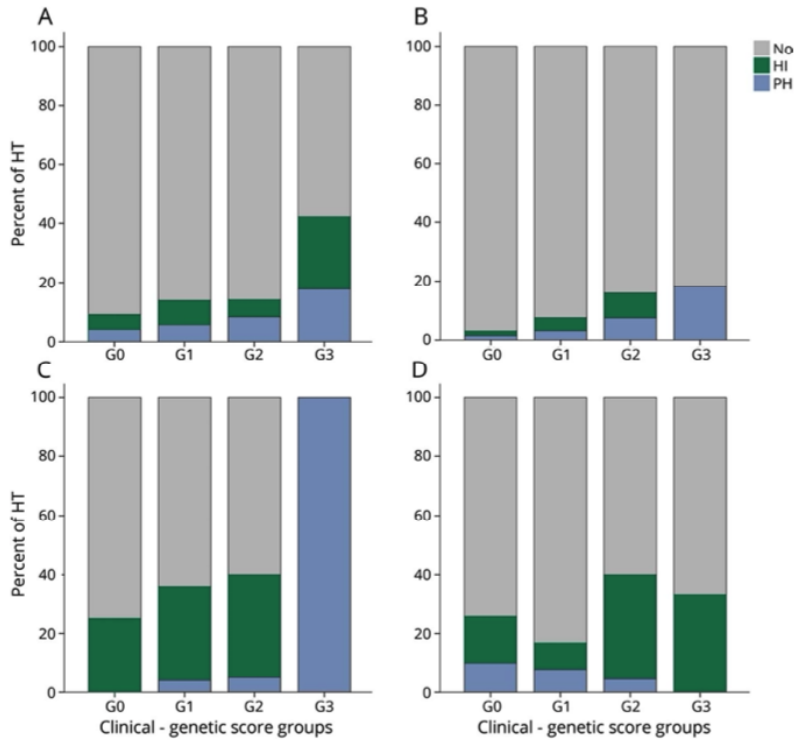
Liu Mao, Chen Jian, Liu Changzhi, Huang Dan, Huang Suihua, Tang Wenyi, Wu Wei*

FARMACOGENETICS

RESPONSE to rTPA

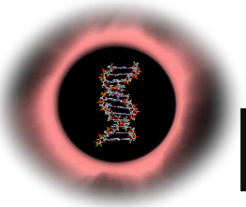
- rs669 in ***A2M gene,*** rs1801020 in ***F12 gene*** and rs1695 in ***GSTP1,*** associated with **hemorrhagic transformation**
- **GenoTPA score** predicts risk if Hemorrhagic Transformation after rTPA.

Figure Occurrence of HT after recombinant tissue plasminogen activator treatment per increasing group of Genot-PA score



(A) Cohort A, (B) cohort B, (C) cohort C, and (D) cohort D. Score groups: G0 ≤3.95 points; G1, 3.95 to 5.10 points; G2, 5.10 to 6.10 points; and G3 ≥6.10 points. HI = hemorrhagic infarct; HT = hemorrhagic transformation; PH = parenchymal hematoma.

Carrera C,et al. Spanish Stroke Genetic Consortium. Neurology. 2019 Aug 27;93(9):e851-e863.

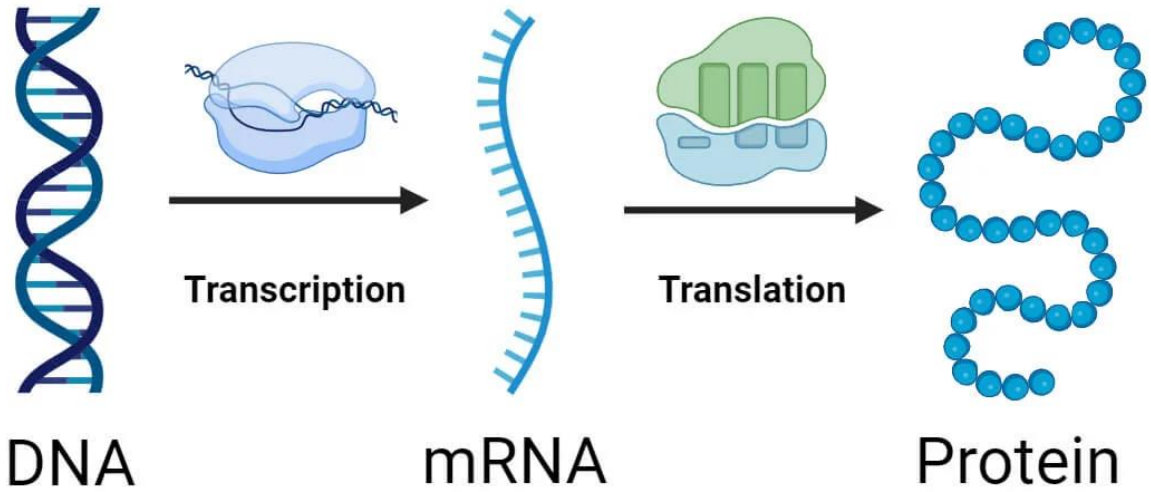


FARMACOGENETICS

RESPONSE to treatment with oral antidiabetic drugs OADs:

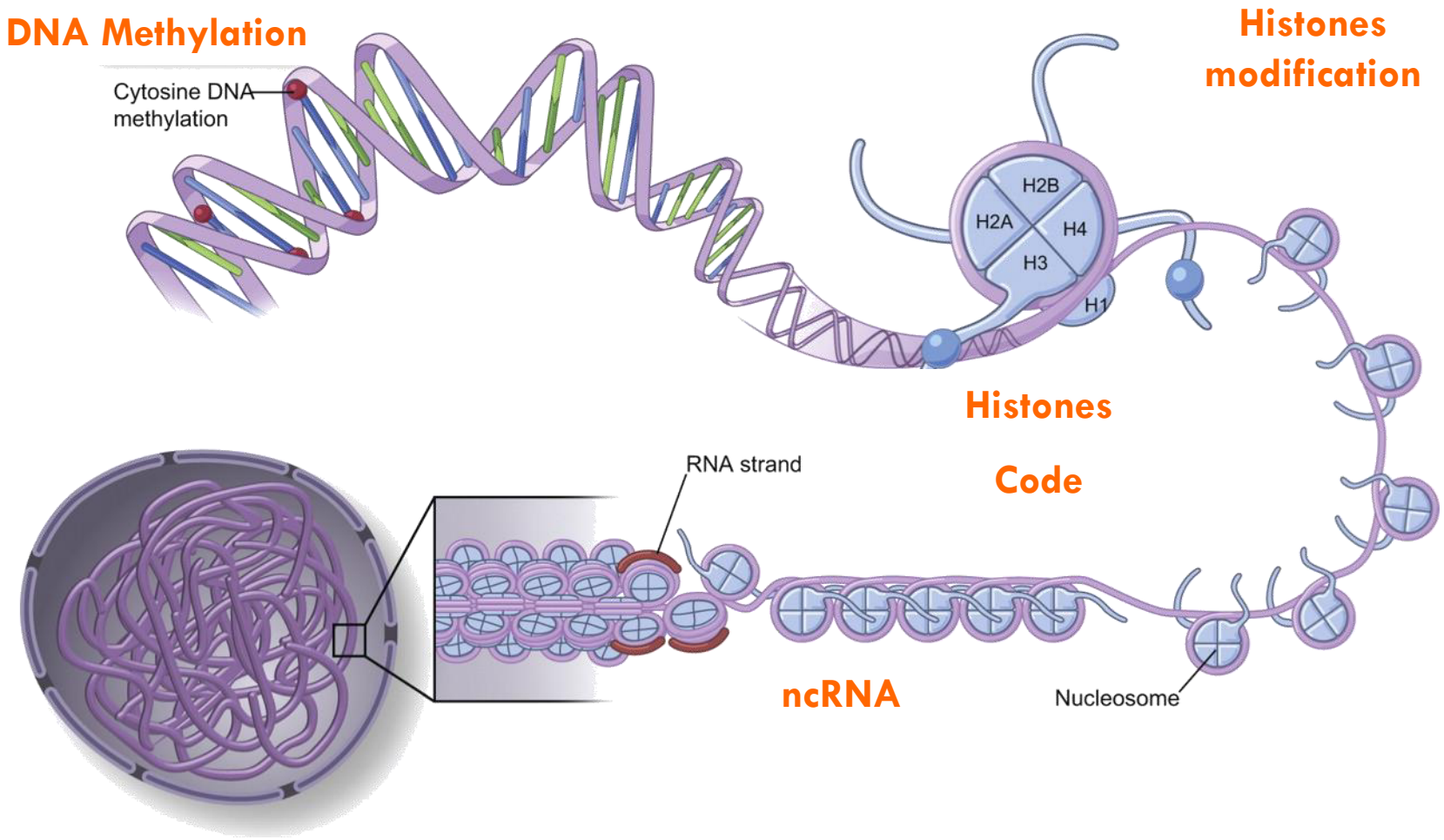
- Metformin: SLC22A1, SLC22A2, SLC47A1, PRKAB2, PRKAA2, PRKAA1, and STK11
- Sulfonylureas: CYP2C9 and TCF7L2
- Repaglinide: KCNJ11, SLC30A8, NEUROD1/BETA2, UCP2, and PAX4 loci
- Pioglitazone: PPARG2 and PTPRD loci
- Rosiglitazone: KCNQ1 and RBP4 loci
- Acarbose: PPARA, HNF4A, LIPC, and PPARGC1A loci.
- …

Gene Expression



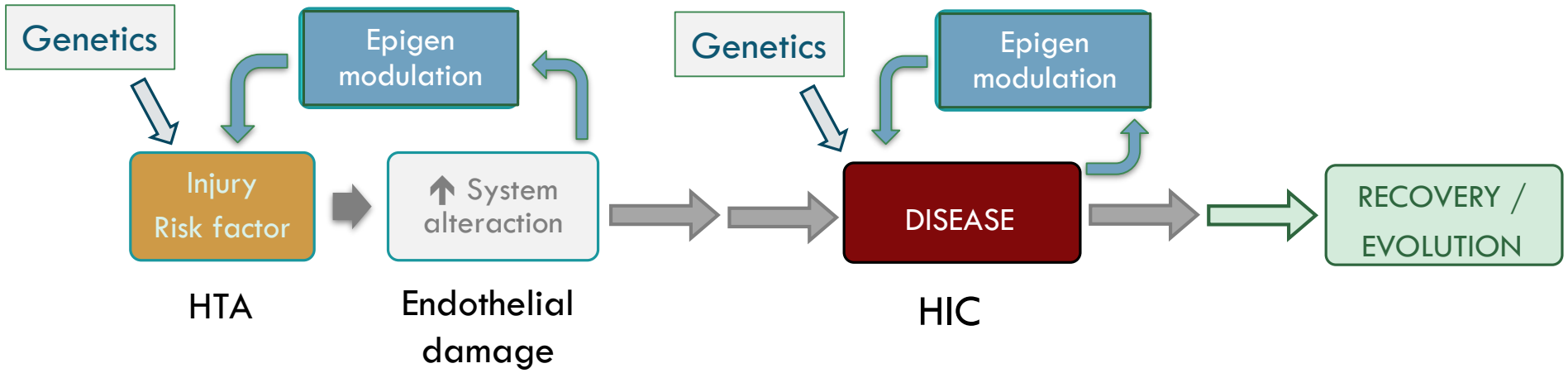
EPIGENETICS

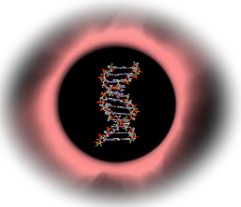
Epigenetic mechanisms in gene regulation



EPIGENETICS

Potential as a dynamic **biomarker**





EPIGENETICS. DNA-Methylation

Some in-vitro and tissue studies suggest candidate genes (m6A hub gene) related with inflammation and ICH (Zhou, Neuroimmun 2023)

No EWAS studies properly powered so far. Some of them retracted.

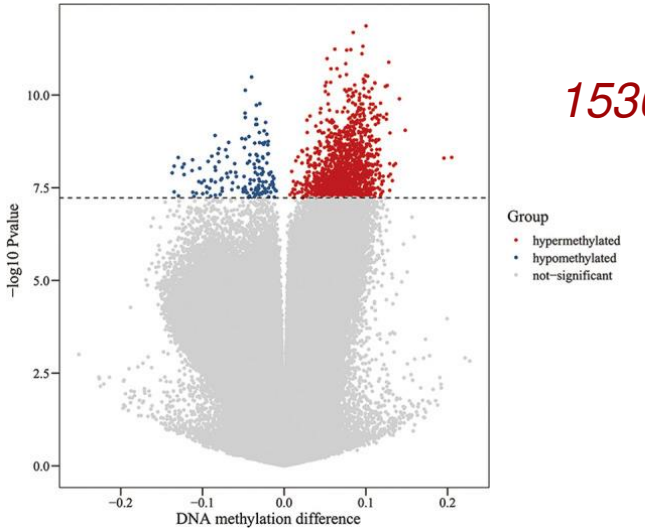
EPIGENETICS. DNA-Methylation

Genome-Wide DNA Methylation Pattern in Whole Blood Associated With Primary Intracerebral Hemorrhage

Yupeng Zhang[1†], Hongyu Long[1†], Sai Wang[1], Wenbiao Xiao[1], Meishan Xiong[1], Jianyi Liu[1], Lei Chen[1], Ruijuan Chen[2], Xueli Wei[2], Yi Shu[3], Yi Zeng[2] and Le Zhang[1]*

[1] Department of Neurology, Xiangya Hospital, Central South University, Changsha, China, [2] Department of Geriatrics, Second Xiangya Hospital, Central South University, Changsha, China, [3] Department of Neurology, Second Xiangya Hospital, Central South University, Changsha, China

Front immunol, 2021



1530 sites (p-value < 5.92E-08)

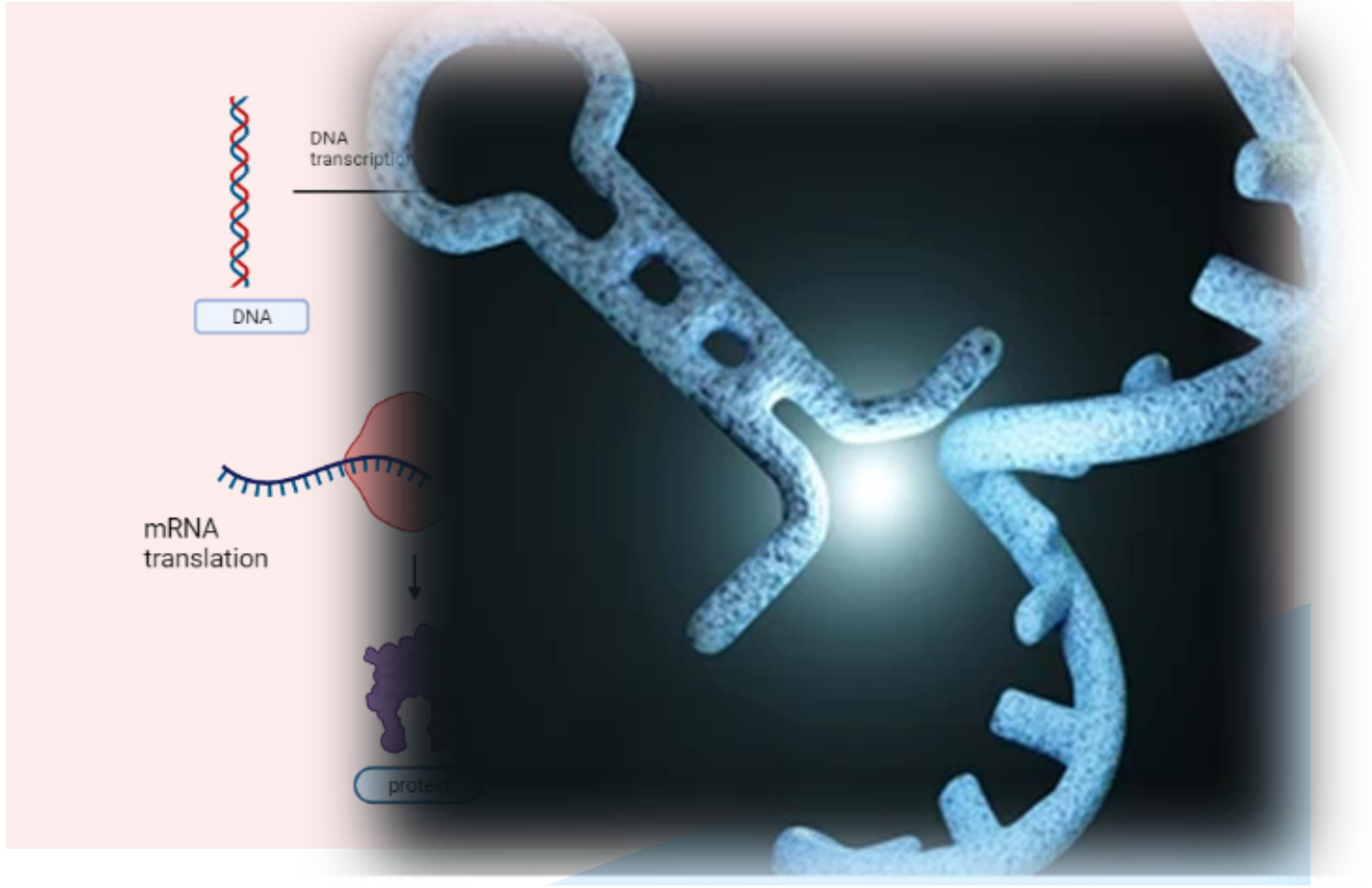
Some design flaws and limitations:

N= 30/34

QC? Popul stratification? Batch Effect?

No adjustments by age or VRF

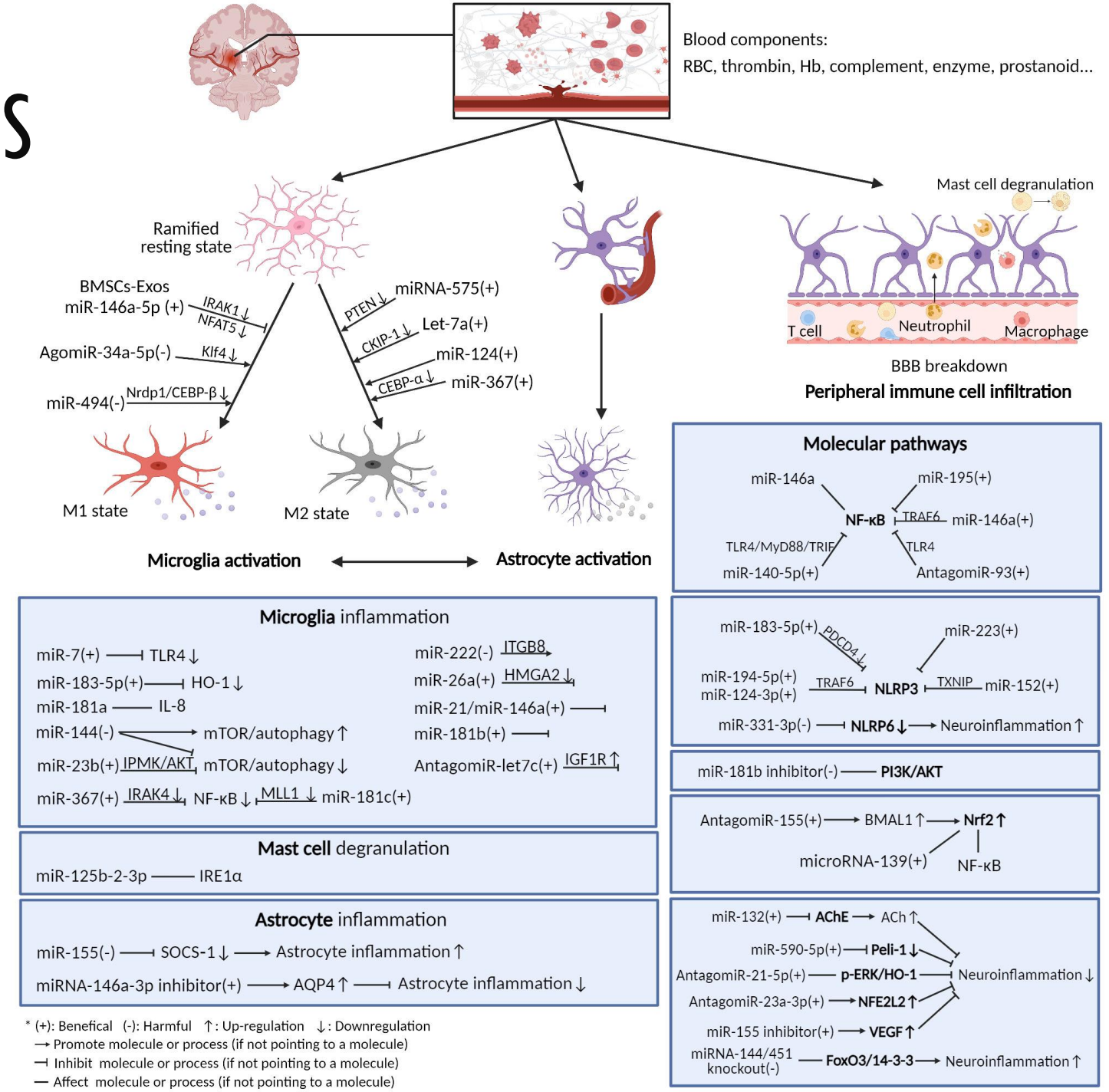
EPIGENETICS. miRNAS



- Risk Factor
- Biomarker
- Therapeutic Target
- Therapeutic agent

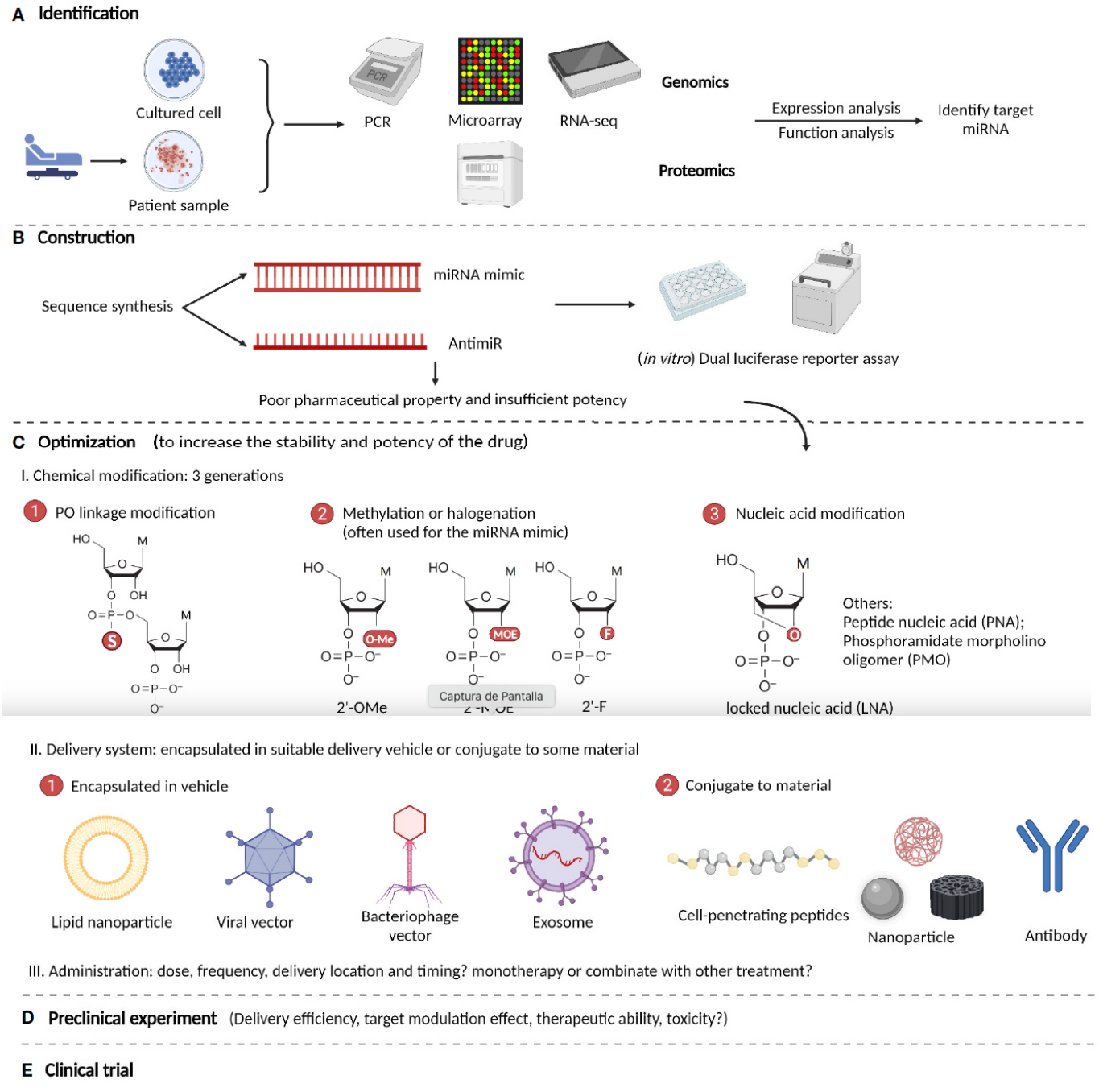
EPIGENETICS. miRNAS

Neuroinflammation after ICH and microRNAs' modulation



EPIGENETICS. miRNAS

Procedures to develop microRNA-based therapeutics for ICH.



Applicability

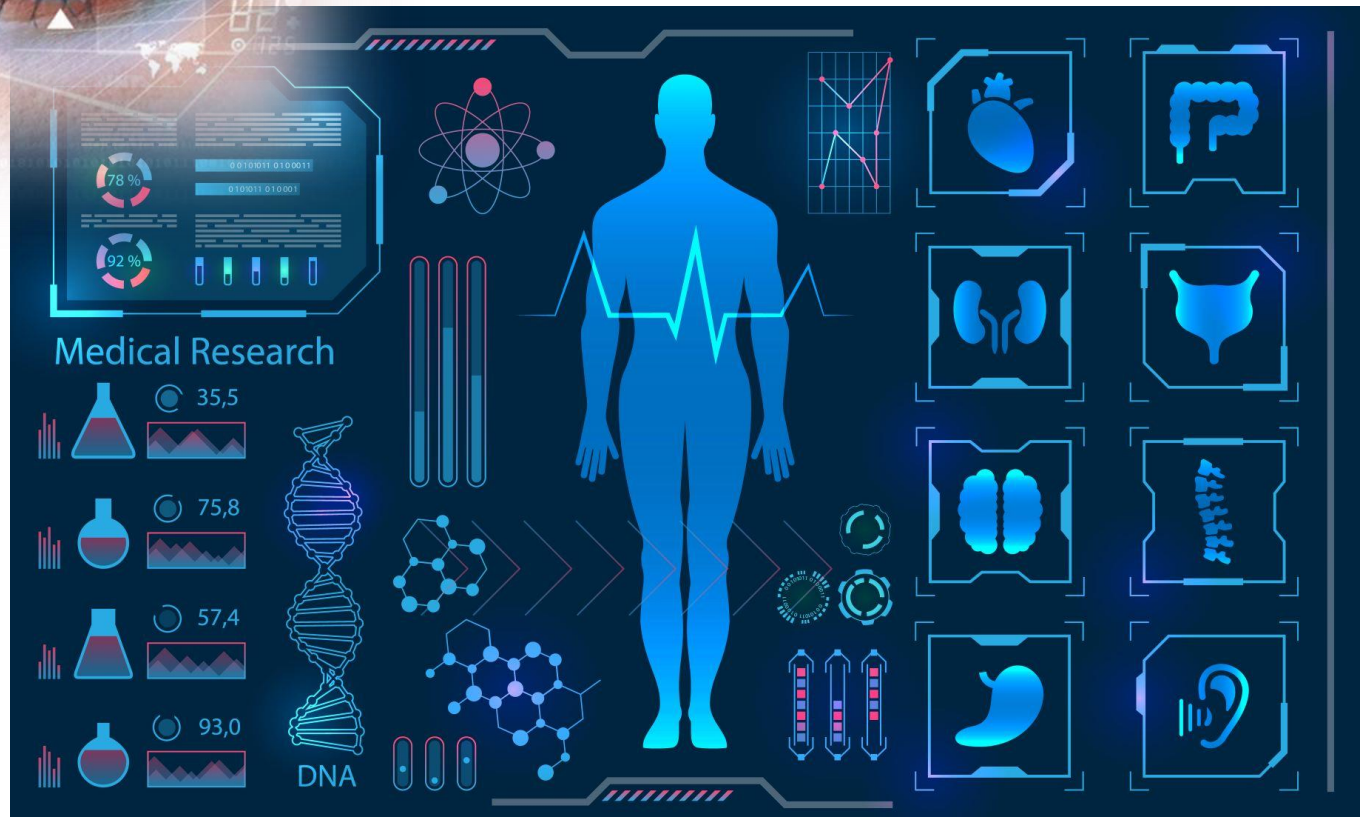
Discover new mechanisms

- Developing new drugs
- New risk factors

Applicability

Personalized Medicine:

- Early diagnosis
- Predicting and preventing risk
- Choosing the best individualized therapy




Thank you