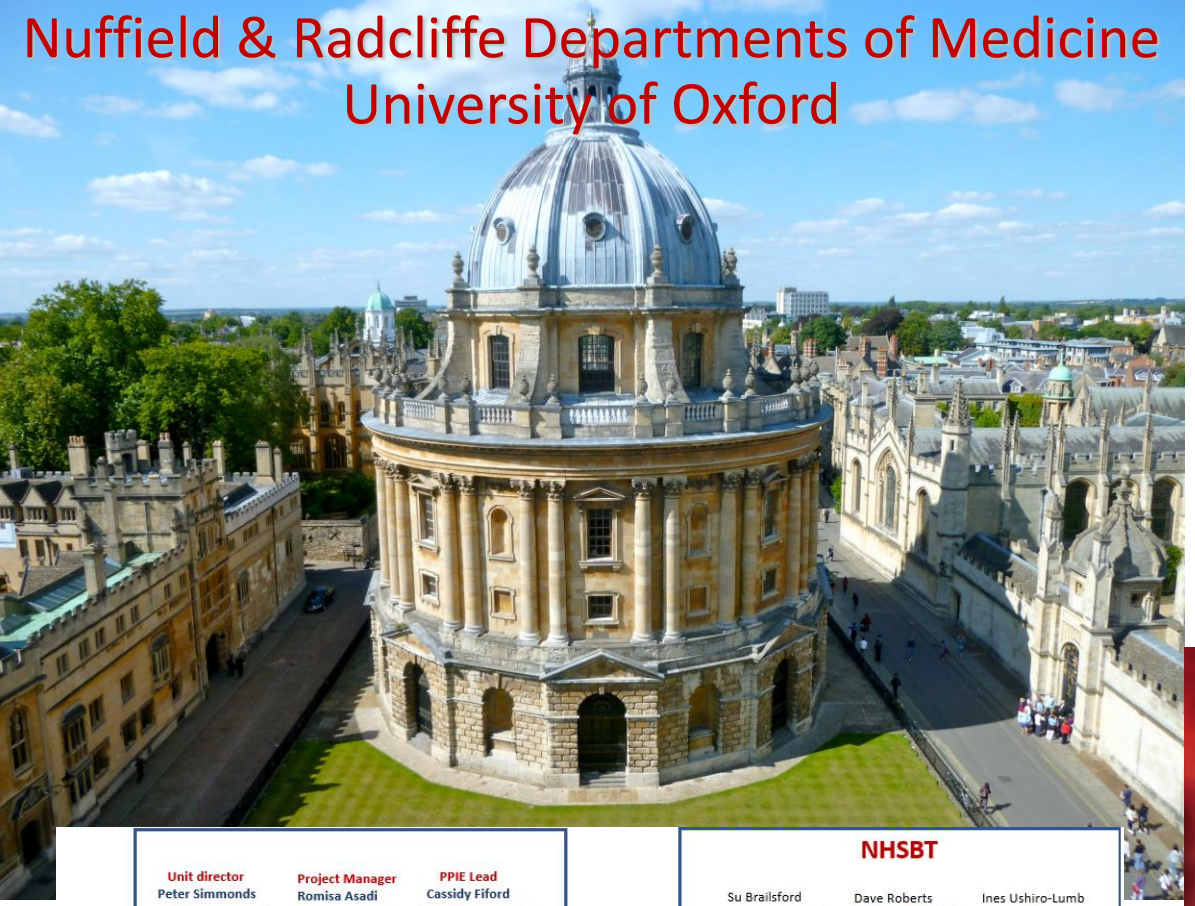
Nuffield & Radcliffe Departments of Medicine
University of Oxford



Blood and transplant microbiological safety the challenges and opportunities ahead

Peter Simmonds

NIHR | Blood and Transplant Research Unit in Genomics to Enhance Microbiology Screening at University of Oxford

Genomics to Enhance Microbiology Screening (GEMS)









UK Health Security Agency

NHS Blood and Transplant

CellPress
OPEN ACCESS

Cell
Leading Edge

Perspective

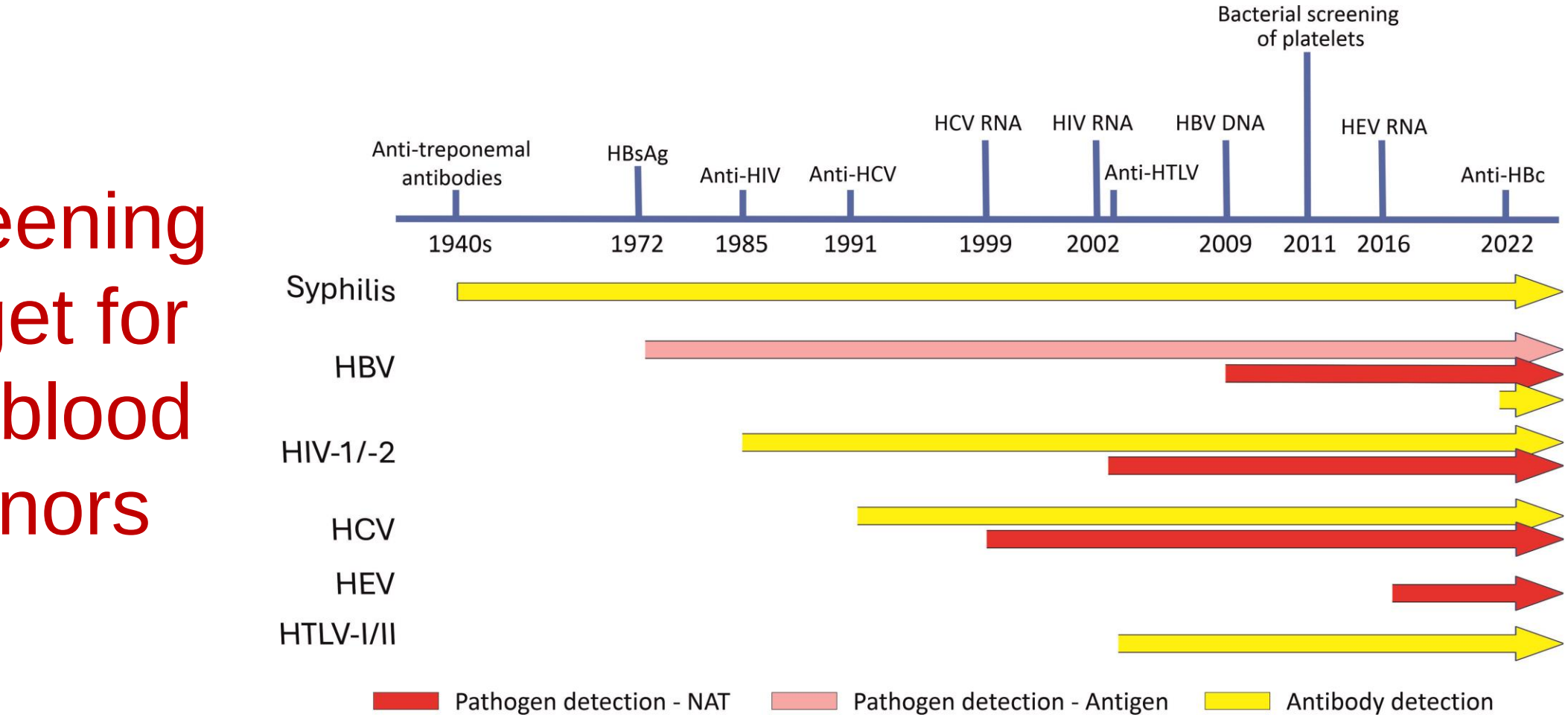
Virology—The next fifty years

Edward C. Holmes,[1,2,*] Florian Krammer,[3,4,5,7,8] and Felicia D. Goodrum[6,8]

- Current screening practice for blood-borne pathogens
- Safety threats from emerging and pandemic viruses
- Evaluation of risk, haemovigilance
- The human virome
- Proteinopathies
- Pathogen reduction
- The health economic background for screening
- Screening for “everything”, impact of new technologies

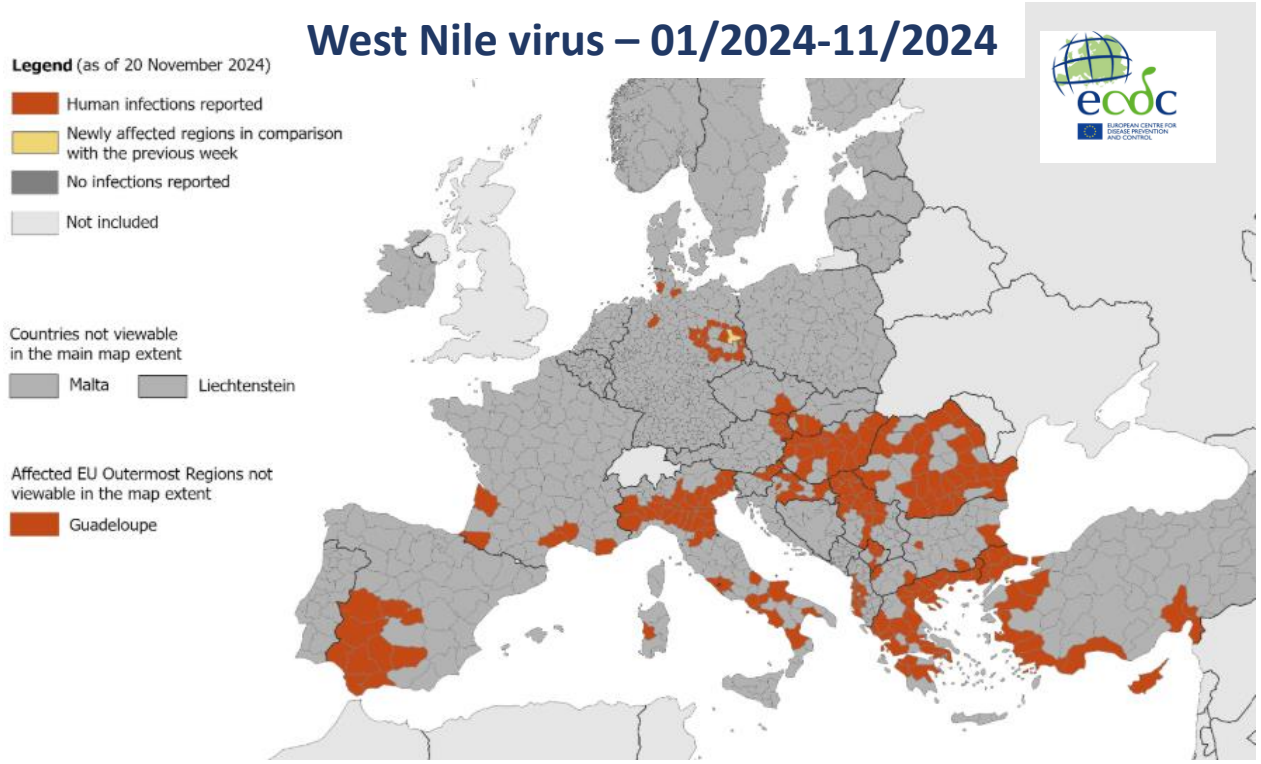
→ An uncertain outlook, even for 10 years

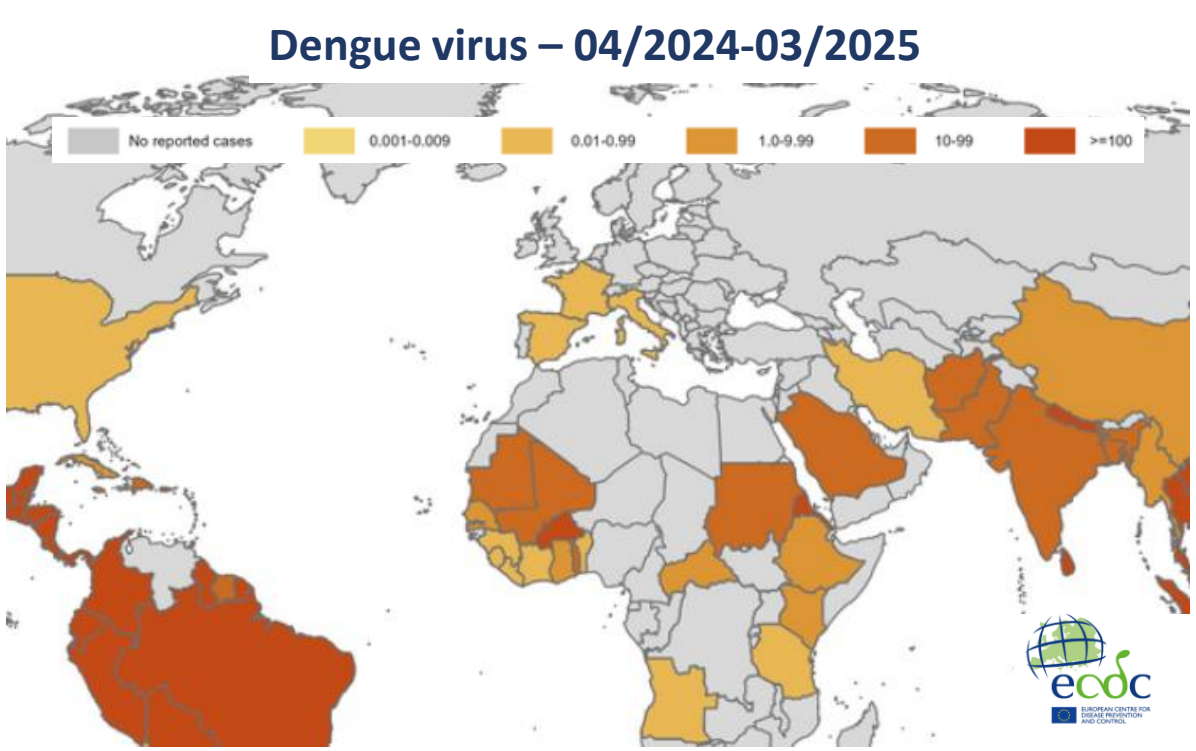
Screening target for UK blood donors



- Selected based on their established transmission risk and health impact for the recipient
- Discretionary: WNV, malaria
- Transplant: As above + HCMV and HHV-8

Emerging pathogens threaten blood safety

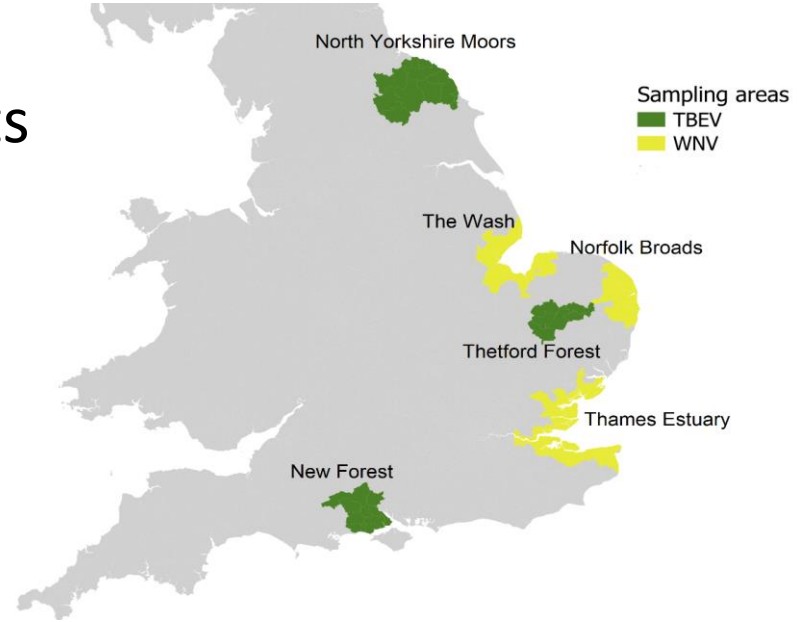




- Vector-borne pathogens – arboviruses, *Borrelia burgdorferi*, malaria
  - Emergence often associated with a change in vector density and distribution
- Pandemic viruses - Influenza A viruses, SARS-CoV-2, HIV-1, smallpox, plague

Proactive response to emerging infections

- CODONET (Harvala, Secret *et al.*)
  - BTRU-GEMS / NHSBT UKHSA collaboration
  - Archiving of consented of donor samples
  - Targeted geographical areas
  - Detailed questionnaire of donor risk
  - Applied to monitoring entry of WNV and Usutu virus
- Implementation
  - 1,400+ donor consents and 1,100 completed surveys
  - Target: 5000
  - PCR and serology applied in 3rd quarter, 2025



Subject	Information parameters
Age	Age (in years)
Biological sex & gender	Male / Female and Gender identify with
Ethnic Group	White
	Mixed or multiple ethnic groups
	Asian or Asian British
	Black, Black British, Caribbean or African
	Other
Residency	Current residential postcode
	Duration of residence
	Country of birth (if not UK)
	Other countries of residence
Travel	Travel outside the UK in the last 5 years: Continents and countries visited, frequency, urban or rural areas
Vaccinations	Vaccinations for unusual or travel-related diseases Vaccine type: Tick-borne encephalitis (TBE), Yellow fever, Japanese encephalitis (JE), Dengue fever, Q fever, Rabies, Hepatitis A, Hepatitis B, Lyme disease (vaccine discontinued), Smallpox, Monkeypox (mpox)
	Timing of most recent vaccination
Medical history	Previous deferral from blood donation
	Previous zoonotic diseases and recency of diagnosis
Exposure to ticks	History of tick bites in last 5 years
	Location(s) of tick bites, UK, abroad
	How frequently bitten
Exposure to mosquitoes	History of mosquito bites in last 5 years
	Location(s) of mosquito bites, UK, abroad
	How frequently bitten
Exposure to animals	History of animal exposure in last 10 years
	Reason and frequency of contact with wild, farm or pet species: Pigs, cattle, sheep or goats, deer, poultry or gamebirds, wild birds, pet birds, camels, llamas or alpacas, bats, dogs, cats, rabbits, guinea pigs, mice, rats, hamsters or gerbils, reptiles, horses, non-human primates, insects, fish
	History of animal bites
Food products	List of dietary exclusions
	Consumption of unpasteurised milk
Outdoor Exposure	Outdoor employment in last 10 years, including: Wildlife ranger, deer management, Forestry worker, Countryside management, Gamekeeper, Farm worker, Small holder, Gardener, Outdoor pursuits instructor, Environmental scientist or ecologist, Veterinarian, Water worker, Fisherman, Other
	Duration of outdoor occupation(s)
	Outdoor hobbies or leisure activities in last 10 years, including: Urban walking or running, Rural walking or running, Urban dog walking, Rural dog walking, Orienteering, Climbing, Cycling, road or off-road. Horse riding, Golf, Field sport, Gardening, Camping, Conservation volunteer, Hunting, Freshwater sports (swimming, sailing, rowing, caving, fishing), Sea sports
	Duration of outdoor activities

Assessment of risk

- Criteria used to evaluate risk

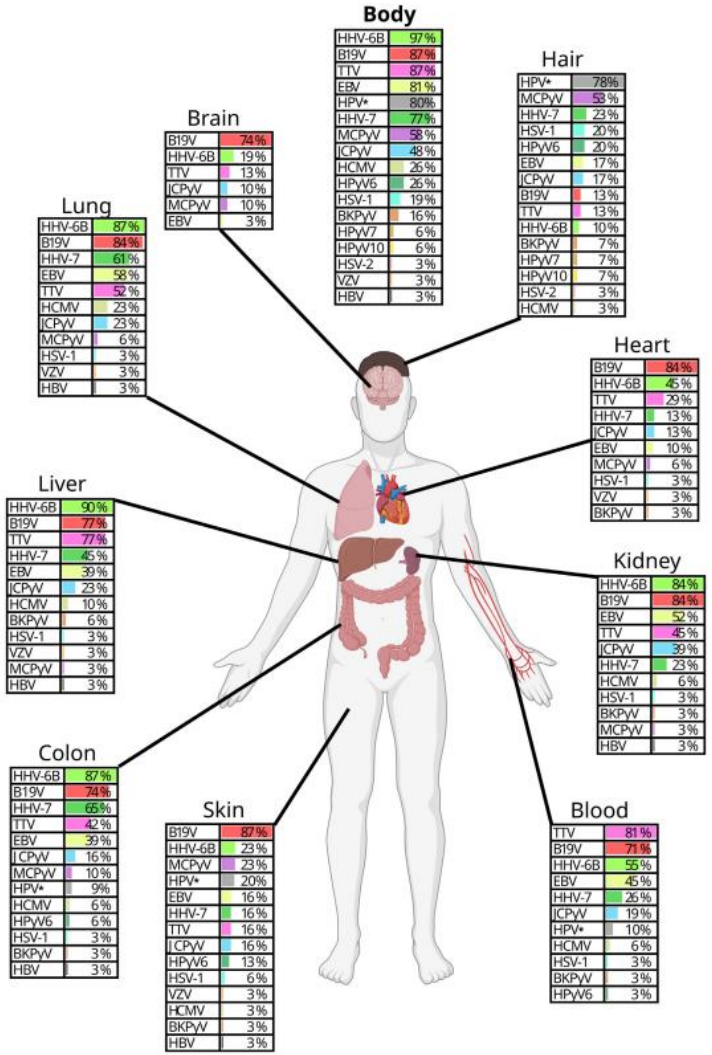
a) Defined agent pathogenicity
b) Demonstrated presence in the donor population
c) Absence of identifiable risk factors that would otherwise exclude donors
d) Demonstrated transmissibility and recipient susceptibility
e) Evidence for post-transfusion disease assessed by haemovigilance
f) Economic assessment costs of screening compared to harm (QALYs)

- Regular horizon scanning
  - UKHSA / SACTTI in the UK
  - European Blood Alliance
  - ECDC SoHO

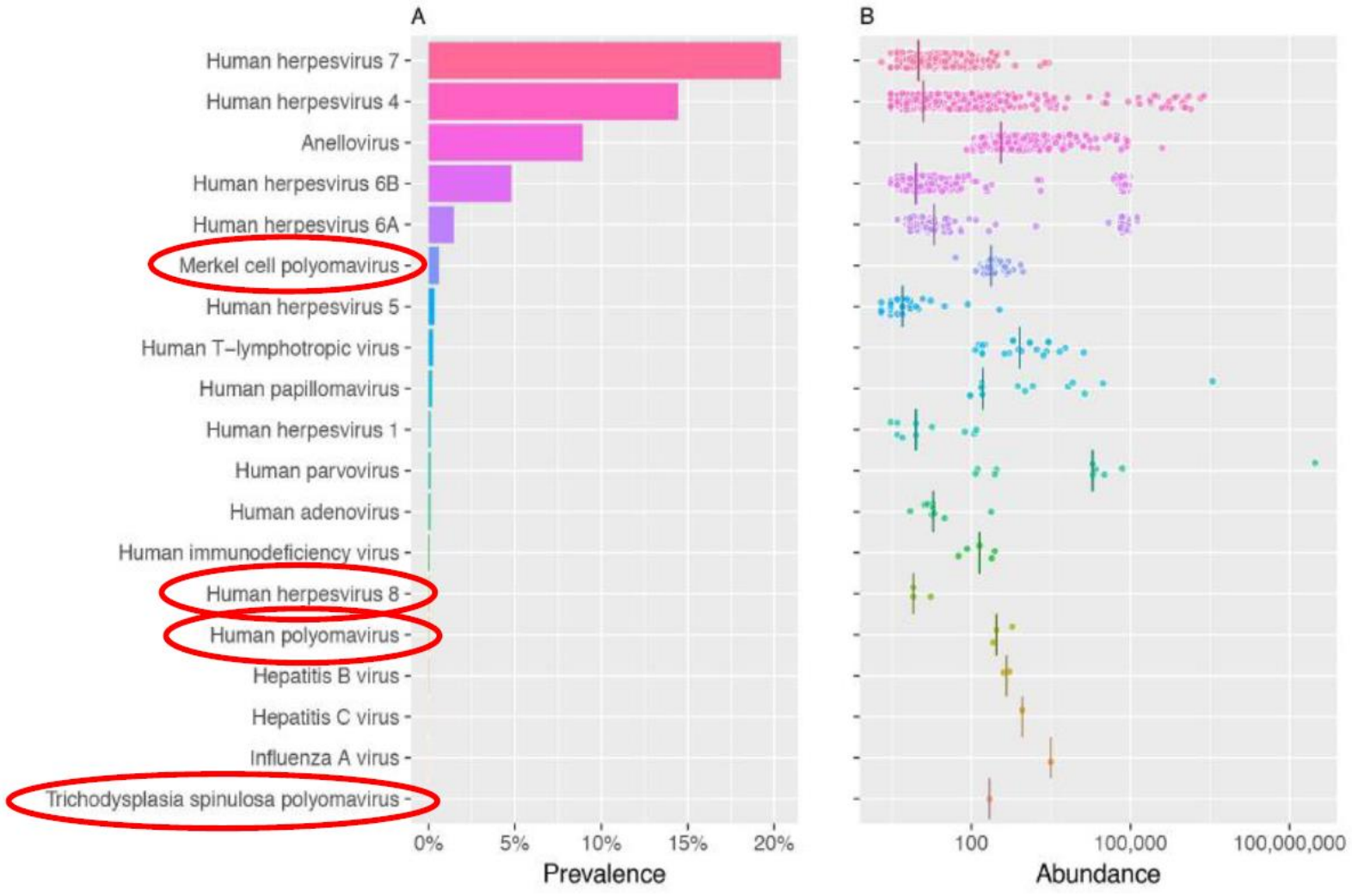
Risk factor	HIV-1	WNV	MPXV	EVs	HPgV-1
Epidemiological factors					
a) Infection incidence	Low	Low	Low	Moderate	Moderate
b) Recipient immunity	None	Low	None	Moderate	Low
c) Risk profile[1]	Partly[3]	None	Shared HIV	None	Low
d) Vector exposure risk		Unreliable			
Pathogen properties					
e) Viraemia duration	Long	Short	Long	Short	Long
f) Blood compartment	All	Plasma	All	Plasma	Plasma
g) Disease severity	Severe	Severe	Moderate	Low	None
h) Asymptomatic viraemia	Yes	Yes	Yes	Yes	Yes
i) Pre-disease viraemia	Yes	Yes	Yes	Yes	Yes
j) Haemovigilance[2]	Yes	Yes	No	No	No

Colour key: Favourable | Some risk | Unfavourable | Non-contributory

The human virome



The tissue-resident DNA virome in humans
Pyöriä et al., Nucleic Acid Research 2023



The blood DNA virome in 8,000 humans
Moustafa et al., PLOS Pathogen 2017

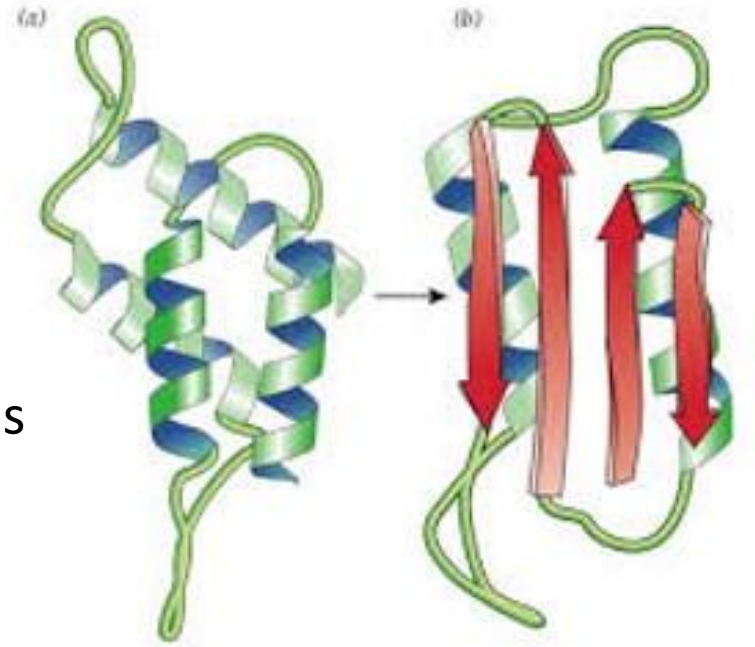
The human virome

- The body is not a sterile site with regards to viruses
- Frequent, long term DNA and RNA virus infections:
  - Human herpesviruses, HCMV, EBV, HHV-6, HHV-7, HSV-1/-2, VZV
  - BKPyV, JCPyV and 12 other recently discovered species of polyomaviruses
  - B19V, PARV4 and 7 other parvoviruses
  - 6 species, and 50+ types of adenoviruses
  - Universal infection with one or more genera of anelloviruses
  - Systemic papillomaviruses
  - Human pegivirus type 1 (3%-5%)
  - Endogenous virus expression

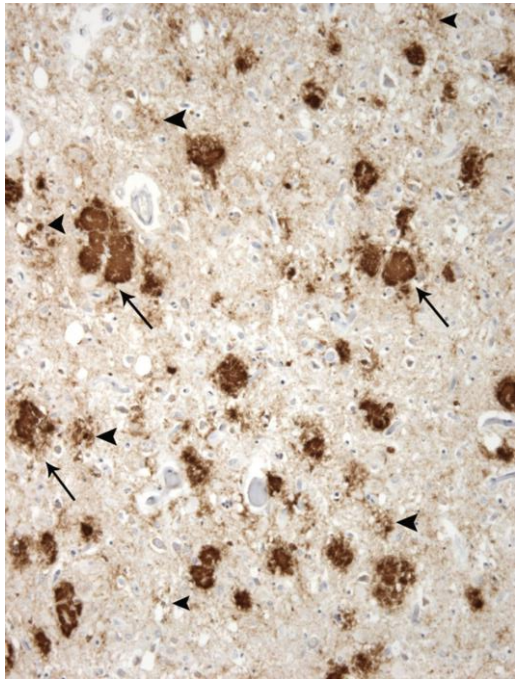
The human virome

- The body is not a sterile site with regards to viruses
- Frequent persistent / latent DNA virus infections
- As with HCMV, they can be pathogenic on transfusion or transplant:
  - Primary infections, severe secondary infections
  - Immunocompromise
- Ubiquity is not the problem
  - Less than universal infection exposes recipients to new potential pathogens
  - Uncertain pathogenic significance of infections with newly discovered viruses
- Haemovigilance could be broadened
  - Use of NGS for universal screening for transmitted / reactivated viruses
  - Association with post-transfusion/ transplant complications and disease

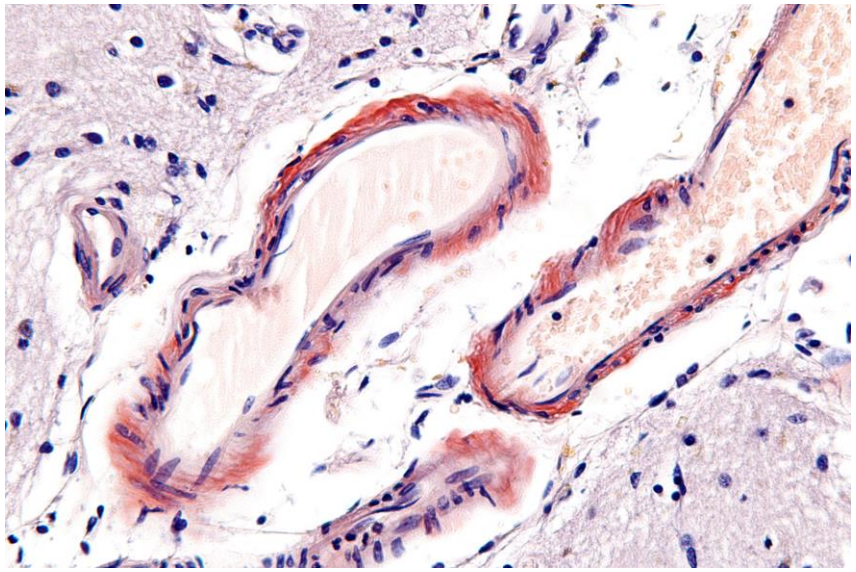
Proteinopathies



- Prion and amyloid deposition diseases in the CNS
- Arise from protein misfolding
  - Genetic predisposition from mutations in PrP or Aβ precursor genes
  - Spontaneous (Alzheimer's disease) or sporadic (Creutzfeldt Jakob disease)
- Transmitted forms of CJD
  - Variant (vCJD) in humans from BSE-contaminated beef in the UK
  - Secondary transmission through blood and plasma donation (5 cases)
  - Kuru from human cannibalism in Papua New Guinea
  - Human pituitary-derived growth hormone, dura mater surgical grafts
- Recently described transmitted form of amyloid disease
  - Chronic amyloid angiopathy (CAA), intracerebral haemorrhage
  - Associated with deposition of misfolded β-amyloid in cerebral arteries
  - Transmitted by growth hormone and dura mater comparable to CJD



Transmission of CAA by transfusion?



By Nephron - Own work, CC BY-SA 3.0, https://commons.wikimedia.org/w/index.php?curid=12142000

JAMA | Preliminary Communication

Intracerebral Hemorrhage Among Blood Donors and Their Transfusion Recipients

Jingcheng Zhao, MD, PhD; Klaus Rostgaard, MSc; Elsa Lauwers, PhD; Torsten Dahlén, MD, PhD; Sisse Rye Ostrowski, MD, PhD, DMSc; Christian Erikstrup, MD, PhD; Ole Birger Pedersen, MD, PhD; Bart de Strooper, MD, PhD; Robin Lemmens, MD, PhD; Henrik Hjalgrim, MD, PhD, DMSc; Gustaf Edgren, MD, PhD

IMPORTANCE Recent reports have suggested that cerebral amyloid angiopathy, a common cause of multiple spontaneous intracerebral hemorrhages (ICHs), may be transmissible through parenteral injection of contaminated cadaveric pituitary hormone in humans.

- Previous observations of vCJD transmission
- Commonality in routes of infectious spread (GH, DM, *etc.*)
- Could CAA also be transmitted through transfusion?
- Incidence of CAA in recipients of blood from donors subsequently diagnosed with CAA in Sweden and Denmark (replication cohort)

Screening for proteinopathies

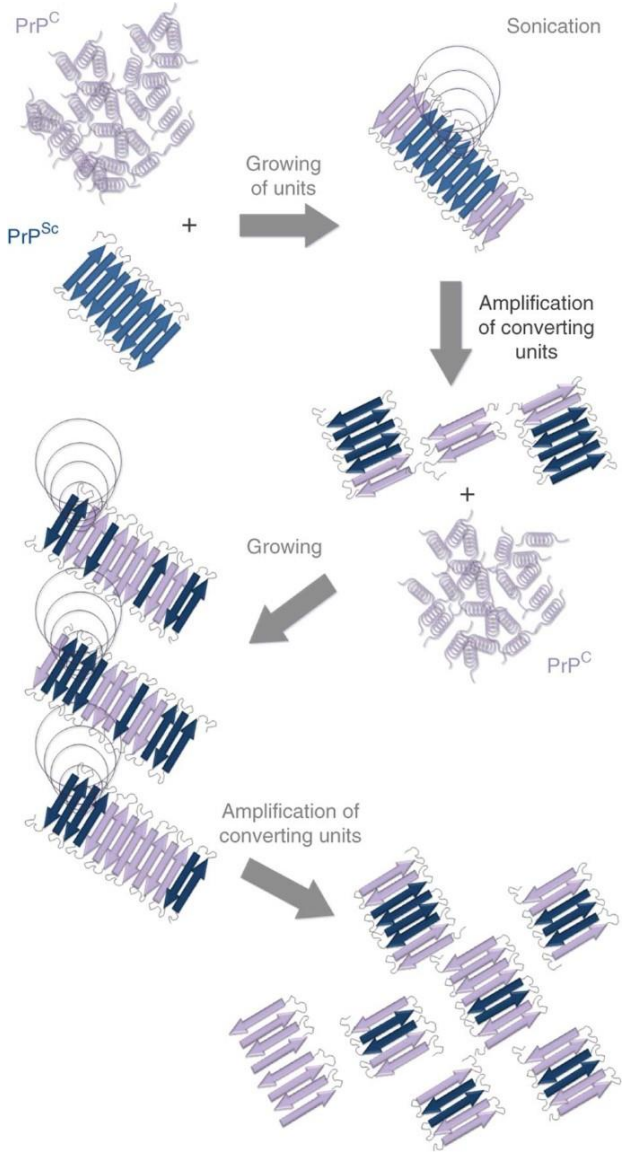
- The agent of both diseases can be detected peripherally pre-neurological onset
  - Potentially present in blood and plasma
  - Supports the use of leucodepletion to mitigate risk
- Screening for vCJD by PrP^{sc} detection
  - Requires extremely high sensitivity and specificity
  - Protein misfolding cyclic amplification (PMCA) - *in vitro* PrP conversion to amplify amounts of misfolded PrP
  - Issues of unknown predictive value of positivity or effectiveness
  - Test practicality for large scale screening, 10 hour cycles
  - No new cases of vCJD after 2017, test never developed for donation screening
- Amyloid screening
- Needs a revolution in understanding the aetiologies of proteinopathies

PROTOCOL

Protein misfolding cyclic amplification of infectious prions

Rodrigo Morales[1], Claudia Duran-Aniotz[1,2], Rodrigo Diaz-Espinoza[1], Manuel V Camacho[1] & Claudio Soto[1]

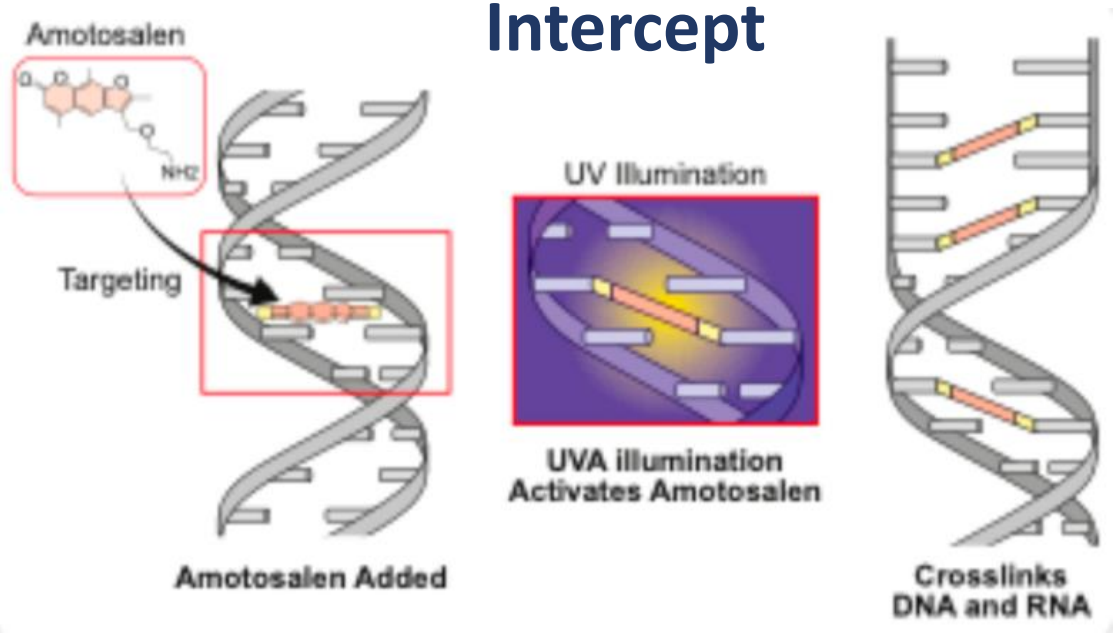
[1]Mitchell Center for Alzheimer's Disease and Related Brain Disorders, Department of Neurology, University of Texas Houston Medical School, Houston, Texas, USA. [2]Facultad de Medicina, Universidad de los Andes, Santiago, Chile. Correspondence should be addressed to C.S. (claudio.soto@uth.tmc.edu).



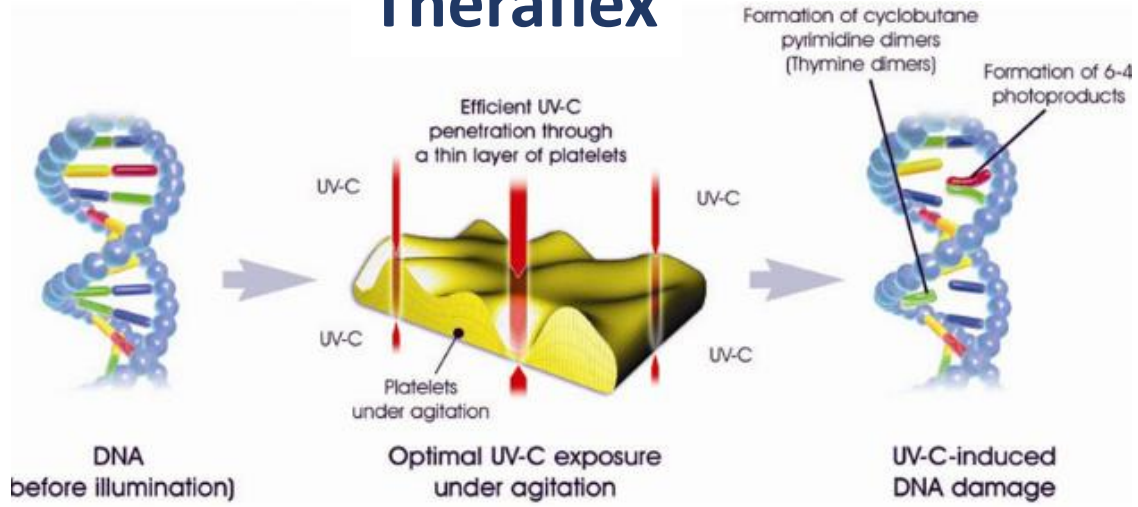
Pathogen inactivation / reduction

- Three psoralen and/or ultraviolet (UV)-based inactivation methods
  - Lead to cross-linking of DNA or RNA sequence preventing replication
  - **Intercept** (Cerus): Amotosalen hydrochloride (AH) & ultraviolet (UV)-A
  - **Mirasol:** Riboflavin + UV-B
  - **Theraflex**: UV-C only
- May achieve blanket reduction or elimination of pathogen infectivity
  - Wide range of RNA and DNA viruses
  - Future potential use for cellular products derived from whole blood donations.
- Backup or replacement of screening?

Intercept



Theraflex



Efficacy of PR

- May achieve blanket reduction or elimination of pathogen infectivity
  - Wide range of RNA and DNA viruses
  - Future potential use for cellular products derived from whole blood donations.
- Backup or replacement of screening?
- Efficacy largely limited to enveloped viruses
  - Reports of HEV and B19V transmission from Intercept treated platelets
- No impact on proteinopathies as no nucleic acid associated with infectivity
- Limited cost-effectiveness (>$1m / QALY)

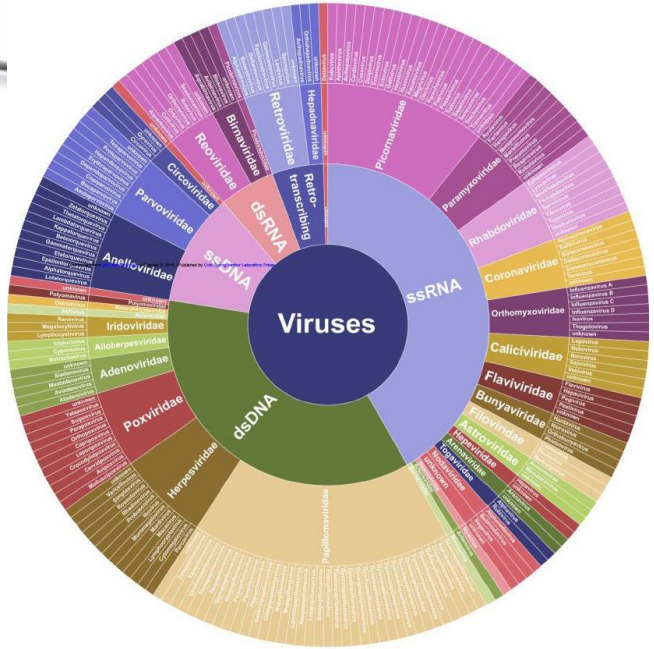
Virus	Species	Genome	LRF
Enveloped			
Retrovirus	HIV-1 cell-associated	ssRNA	>6.64
	HIV-1 cell-free		>6.74
	HIV-1 clinical Z84		>3.94
	HIV-2 clinical CBL20		>5.20
	HTLV-I		4.84
	HTLV-II		5.04
Hepadnavirus	HBV	dsDNA	>5.54
	Duck HBV		>6.74
Flavivirus	HCV	ssRNA	>4.54
	BVDV		>4.71
	West Nile virus		>6.85
	Zika virus		>6.66
	DENV-1		>5.06
	DENV-2		>5.66
	DENV-3		>4.56
	DENV-4		>5.26
	Yellow fever virus		>5.57
Alphavirus	Chikungunya virus	ssRNA	>7.67
	Mayaro virus		>6.97
	Ross River virus		>5.17
Coronavirus	SARS-CoV	ssRNA	>6.27
	MERS-CoV		>4.77
Influenza virus	IAV H5N1	ssRNA	>5.97
Arenavirus	LCMV	ssRNA	>5.67
Herpesvirus	Cytomegalovirus	dsDNA	>6.44
Poxvirus	Vaccinia virus	dsDNA	>5.24
Non-enveloped			
Reovirus	Bluetongue virus 11	dsRNA	5.25
Adenovirus	Human adenovirus 5	dsDNA	5.68
	Simian adenovirus 15		1.52
Calicivirus	Feline calicivirus	ssRNA	1.79
Picornavirus	Hepatitis A virus	ssRNA	0.75
	Hepatitis A virus		0.76
Parvovirus	Human parvovirus B-19	ssDNA	2.14
	Porcine parvovirus		0.38

Screening for everything?

- Implementation in practice
  - A fraction of the nucleic acid in sample can be tested (pg's - ng's) by current Illumina or ONT methods
  - Bioinformatic analysis bottlenecks
  - Lack of understanding of the size of the human virome and its significance for health and immunity
  - No established ethical framework for comprehensive NGS in diagnostic testing or population screening
  - Consent and use of human genetic information
- Implementation in theory
  - Is there value in universal virus screening or should we pre-define pathogens for testing?
  - Can donors and recipients be better matched?
  - How can microbiology screening be integrated with genomics programme







Current status of NGS methods (Fu *et al.*)

- Blinded panel sent to expert NGS labs
- Pre-determined viral loads and genotypes
- Methods:
  - PCR + Nanopore or Illumina
  - Target Capture + Illumina
  - Untargeted metagenomics
- Methods assessed for
  - Detection frequency
  - Sequence depth and completeness
  - Cost and assay time

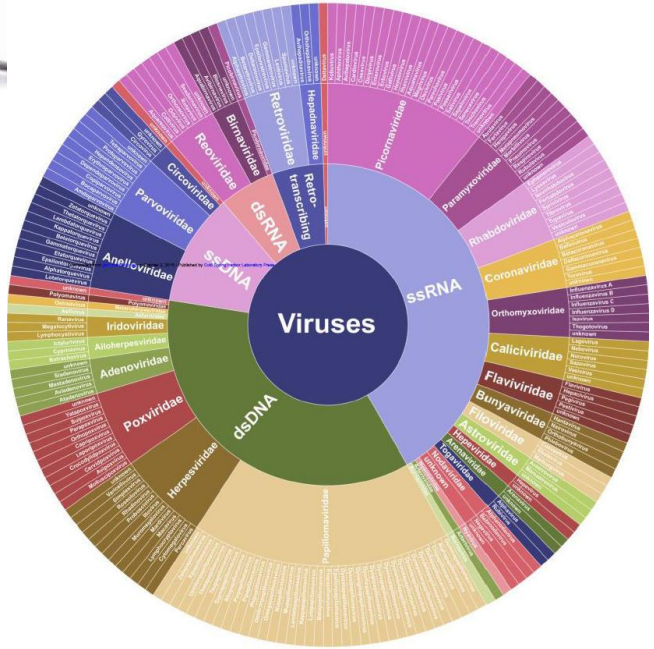
		PCR (Nanopore)		PCR (Illumina)		Target capture			Metagenomics	
Sample	Viral load (IU/mL)	PCR-1	PCR-2	PCR-A	PCR-B	TAC-A	TAC-B	TAC-C	MTG-A	MTG-B
HBV15	6207.0	100%	100%	91%	100%	11%	100%	90%	25%	72%
HBV8	3154.0	100%	100%	98%	100%	91%[c]	97%	99%	50%	14%
HBV2	3108.0	100%	91%		100%	90%[c]	81%	100%		25%
HBV7	1058.0	100%	100%	98%	99%	17%	83%	93%	11%	25%
HBV19	859.8	100%	49%		100%	22%	4%	88%	10%	
HBV16	599.2	100%	85%	59%	100%	9%		75%		
HBV1	551.6	100%	86%	76%	100%	6%	38%	88%	4%	
HBV14	522.3	100%	100%	92%	100%	9%	95%	80%		49%
HBV23	337.1	100%	86%	99%	100%	64%	71%	93%		16%
HBV17	299.9	100%	100%	96%	92%		14%	50%		
HBV9	138.1	100%	100%	33%	99%	9%	8%	35%	2%	
HBV5	128.4	100%	35%		89%			66%		
HBV22	73.6	100%	83%		86%			54%		
HBV4	60.5	100%	85%	59%	100%	35%	7%	100%	12%	
HBV13	60.4	100%	85%		98%			91%	5%	
HBV21	41.0	100%	68%[a]		74%		7%	43%		
HBV6	11.3	100%	33%		90%		3%	28%		3%
HBV18	8.5	60%	67%		88%					
HBV20	3.3	100%	82%[a]		81%			26%	<1%	
HBV12	1.8	15%	67%		97%			25%		
HBV24	1.1	99%	33%[b]		88%					
HBV11	0.3	100%	68%		85%					
HBV3	0.2	71%	68%		91%			5%		
Negative (HBV10)	-	100%[#]	67%[b]	48%	88%					

Screening for everything?

- Implementation in practice
  - A fraction of the nucleic acid in sample can be tested (pg's - ng's) by current Illumina or ONT methods
  - Bioinformatic analysis bottlenecks
  - Lack of understanding of the size of the human virome and its significance for health and immunity
  - No established ethical framework for comprehensive NGS in diagnostic testing or population screening
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  - Is there value in universal virus screening or should we pre-define pathogens for testing?
  - Can donors and recipients be better matched?
  - How can microbiology screening be integrated with genomics programme







The next 50 years?

- Genomic screening
  - Do we go down the route of broader pathogen screening?
  - Universal or an expanded and adaptable range of targets?
- Pathogen reduction
  - Potential contribution towards elimination of non-screened pathogens
- A change in the economic framework?
  - Can costs be better coordinated with other health care interventions?
- Proteinopathies
  - Common, persistent and catastrophically pathogenic
  - Not amenable to genomics or PR approaches
  - A revolution in understanding the nature of the infectious agents is urgently required
- Will we be collecting blood donation? Synthetic blood and platelets

The BTRU-GEMS Team

Unit director
Peter Simmonds



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Cassidy Fiford



NHSBT

Su Brailsford



Dave Roberts



Ines Ushiro-Lumb



Theme 1

Theme 1 Lead
Tanya Golubchik



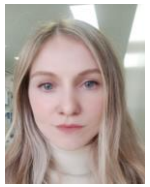
Senior Scientist
Richard Mayne



PhD Students 2023 - 2026
Kaitlin Reid



Kai Kean



Theme 2

Theme 2 Lead
Judy Breuer



Senior Scientists
Oscar Montaguth



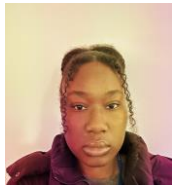
Sofia Morfopoulou



PhD Students 2022 - 2026
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Senior Scientist
Belinda Singleton



Theme 3

Theme 3 Lead
Heli Harvala



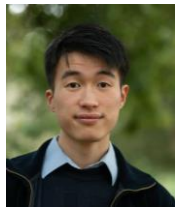
Senior Scientists
Shannah Gates



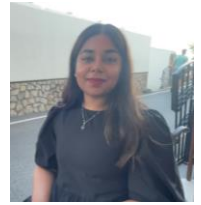
Noreen Mumtaz



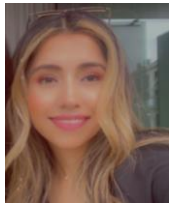
PhD Students 2022 - 2026
Michael Fu



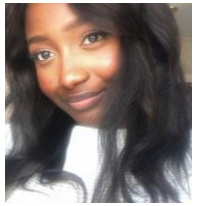
Piya Rajendra



Vidushi Chugh



Jaid Debrah



NIHR | Blood and Transplant Research Unit in Genomics to Enhance Microbiology Screening at University of Oxford

Five MSc students at NHSBT: Courtney Roberts, Jennifer Ingram, Tanya Hughes, Charlotte Washington and Angela Matous (ongoing)
Three professional PhD students at NHSBT: Victoria Maddox (ongoing), Matt Clee (ongoing) and Becky Clare (ongoing)