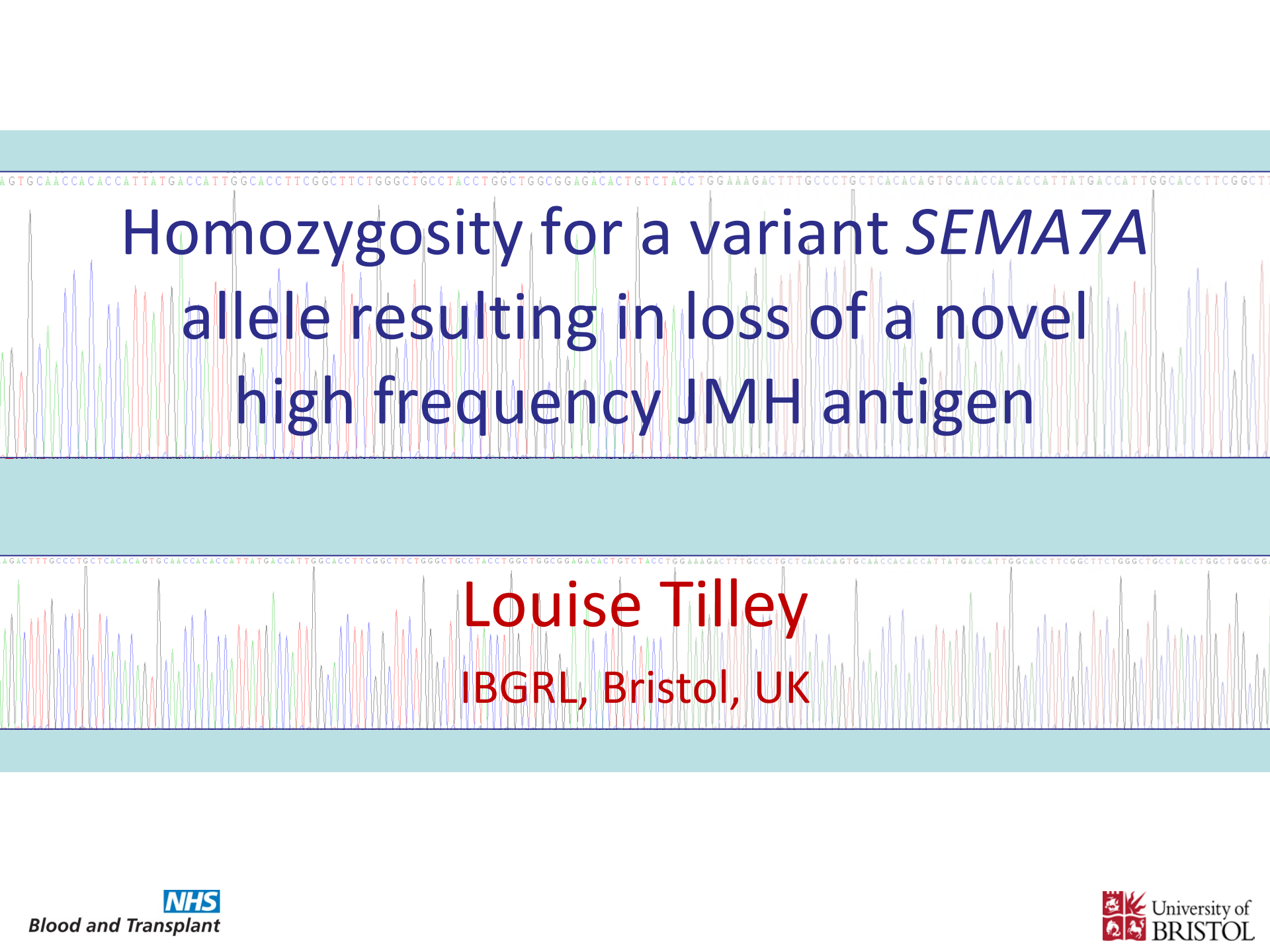
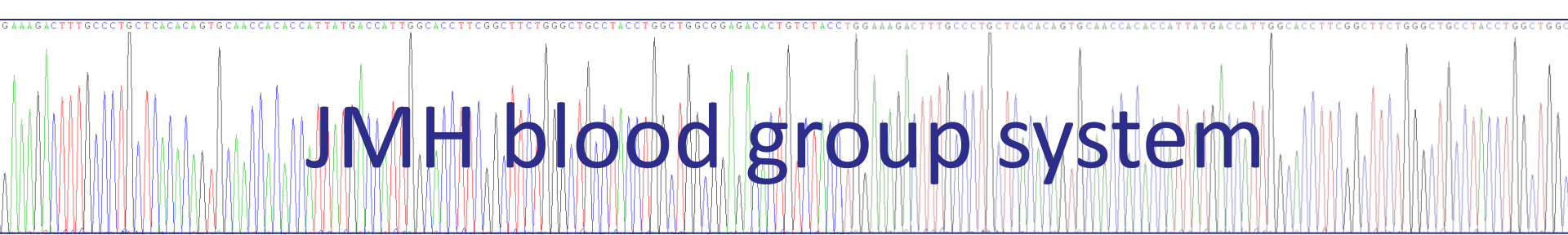
The background of the slide features a DNA sequence at the top and a corresponding chromatogram below it. The sequence is: A G T G C A A C C A C A C C A T T A T G A C C A T T G G C A C C T T C G G C T T C T G G G C T G C C T A C C T G G C T G G C G G A G A C A C T G T C T A C C T G G A A A G A C T T T G C C C T G C T C A C A G T G C A A C C A C A C C A T T A T G A C C A T T G G C A C C T T C G G C T. The chromatogram shows four colored traces (blue, green, red, black) representing the DNA sequence data.

# Homozygosity for a variant *SEMA7A* allele resulting in loss of a novel high frequency JMH antigen

The background of the slide features a DNA sequence at the top and a corresponding chromatogram below it. The sequence is: A G A C T T T G C C C T G C T C A C A G T G C A A C C A C A C C A T T A T G A C C A T T G G C A C C T T C G G C T T C T G G G C T G C C T A C C T G G C T G G C G G A G A C A C T G T C T A C C T G G A A A G A C T T T G C C C T G C T C A C A G T G C A A C C A C A C C A T T A T G A C C A T T G G C A C C T T C G G C T T C T G G G C T G C C T A C C T G G C T G G C G. The chromatogram shows four colored traces (blue, green, red, black) representing the DNA sequence data.

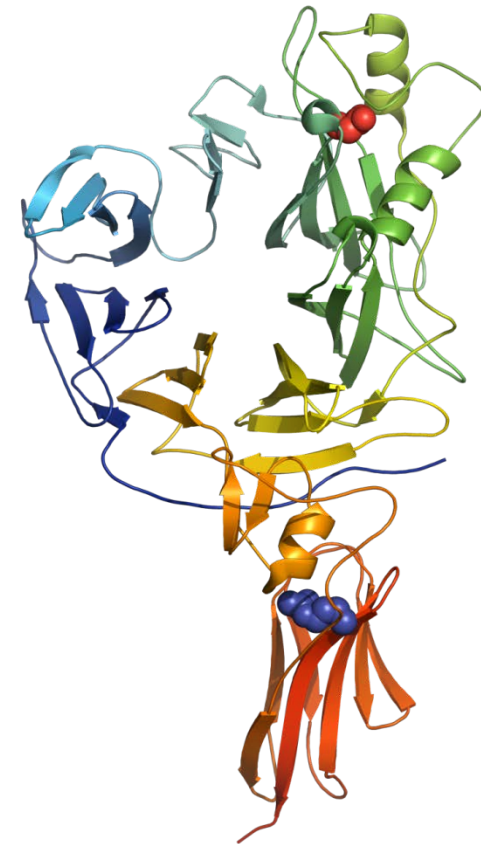
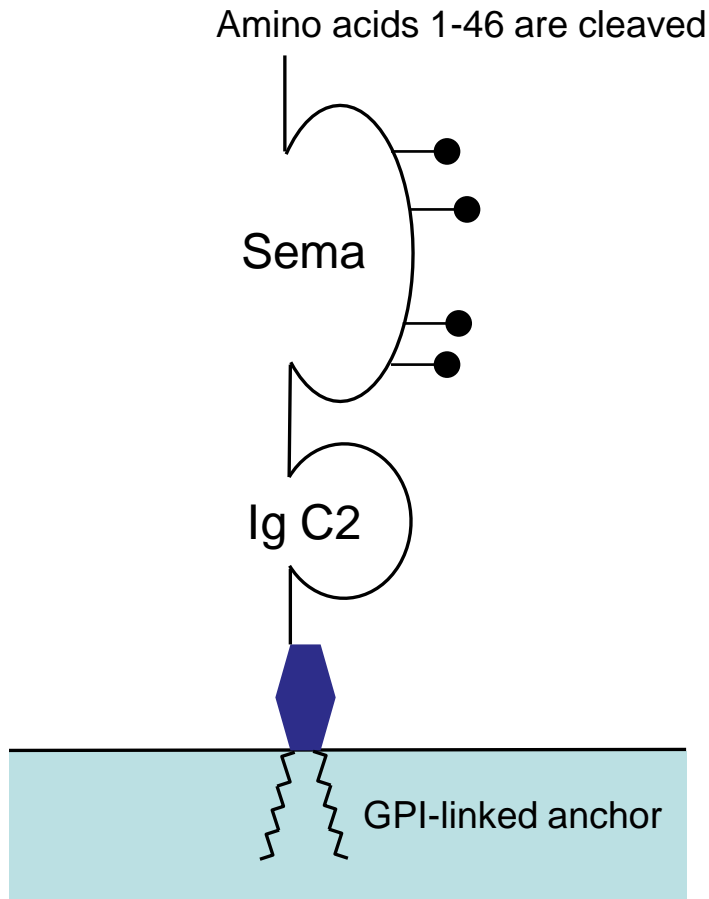
**Louise Tilley**  
IBGRL, Bristol, UK



# JMH blood group system

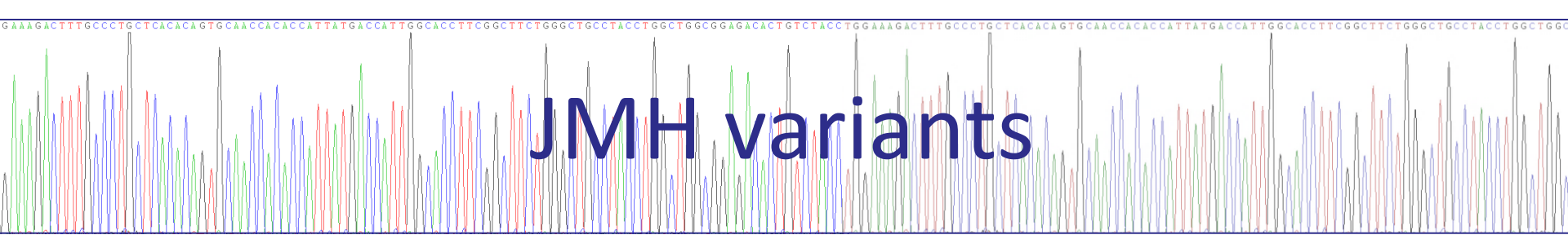
- JMH antigens carried on signalling protein semaphorin 7A (Sema7A, CD108)
- Semaphorins are glycoproteins, may be secreted, membrane-spanning or GPI-linked
- Sema domain, highly conserved seven-blade  $\beta$ -propeller fold, linked through cysteine-rich region to immunoglobulin domain
- Sema7A is widely expressed, with both immune and neurological functions, although function in red cells unknown

# Sema7A protein



Adapted from:  
*The Blood Group Antigen Facts Book 2012*  
M. Reid, C. Lomas-Francis & M. Olsson

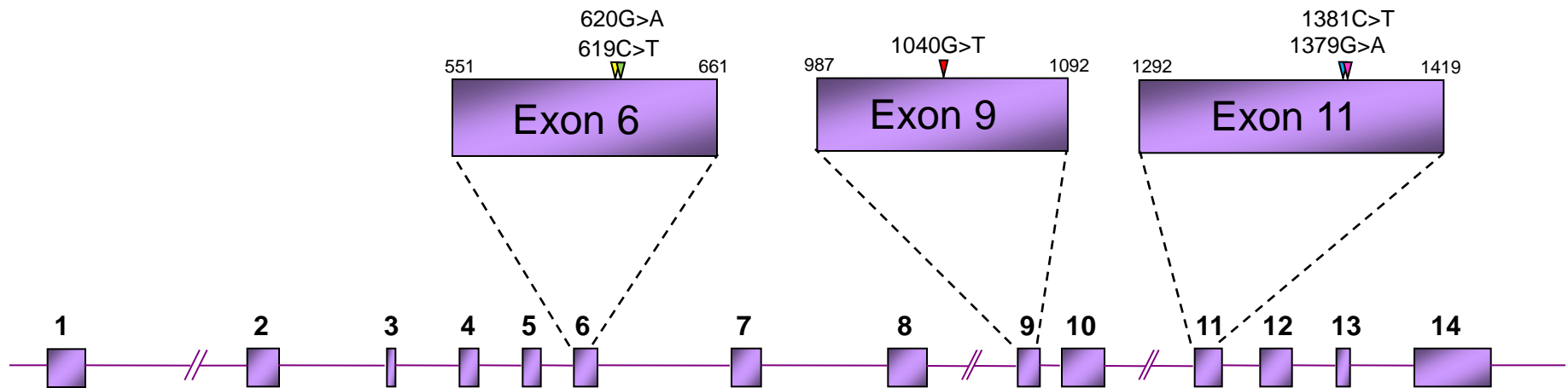
Model courtesy of Nick Burton  
University of Bristol

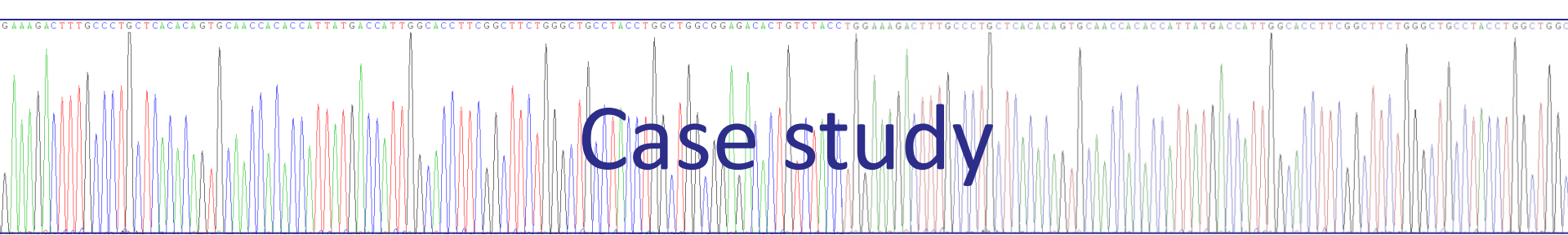


- JMH:-1 phenotype usually acquired & transient
- Five rare inherited variants, lacking JMH2-6
- Identified in JMH+ individuals with alloantibodies to HFA that do not react with JMH- cells
- Variants result from homozygous missense mutations in *SEMA7A*
- *SEMA7A* has 14 exons (all coding) on chr15q22, encoding 666 a.a. protein

# JMH variants

Antigen No.	Name	Frequency	Nucleotide	Exon	Amino acid
JMH1	JMH	High	Not known		
JMH2	JMHK	High	619C>T	6	Arg207Trp
JMH3	JMHL	High	620G>A	6	Arg207Gln
JMH4	JMHG	High	1379G>A	11	Arg460His
JMH5	JMHM	High	1381C>T	11	Arg461Cys
JMH6	JMHQ	High	1040G>T	9	Arg347Leu



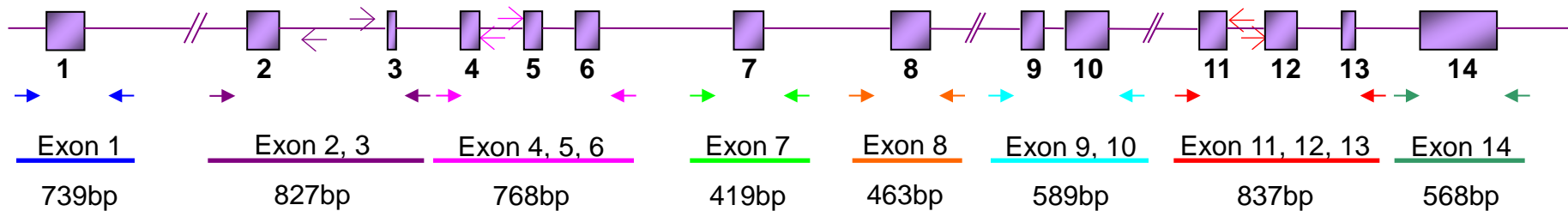


# Case study

- Blood samples from male patient of North African descent with unidentified antibody
- Antibody reacted by IAT with untreated cells, not papain treated cells, identified as anti-JMH
- Plasma compatible with JM<sup>H</sup>- cells, all other cells incompatible
- Patient's own cells clearly negative with antibody
- Patient cells positive with anti-JMH panel (very weak to normal strength)

# SEMA7A sequencing

- All 14 exons amplified using 8 individual PCR reactions
- Sequenced using F&R PCR primers and nested primers as required



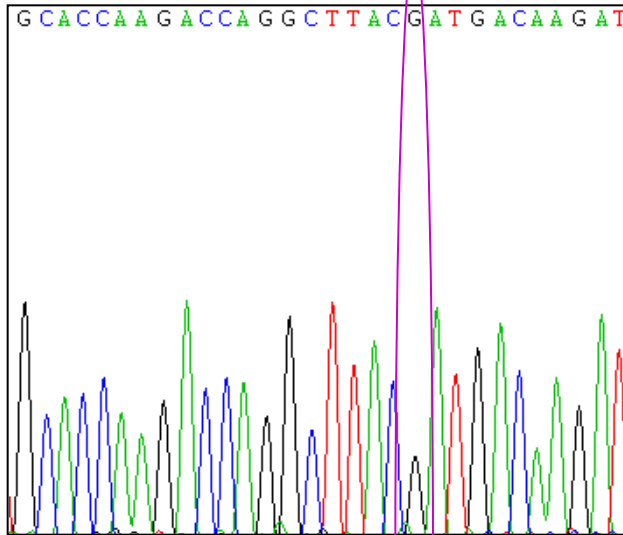


# Results

- Four archived JMH variant samples sequenced, all found to be known variants
- Two homozygous 620G>A (Arg207Gln)  
*JMH\*01.-03* (JMHL- or JMH:-3)
- Two homozygous 1381C>T (Arg461Cys)  
*JMH\*01.-05* (JMHM- or JMH:-5)
- Patient sample homozygous for novel allele;  
709G>A (Asp237Asn), 1545A>G (silent)

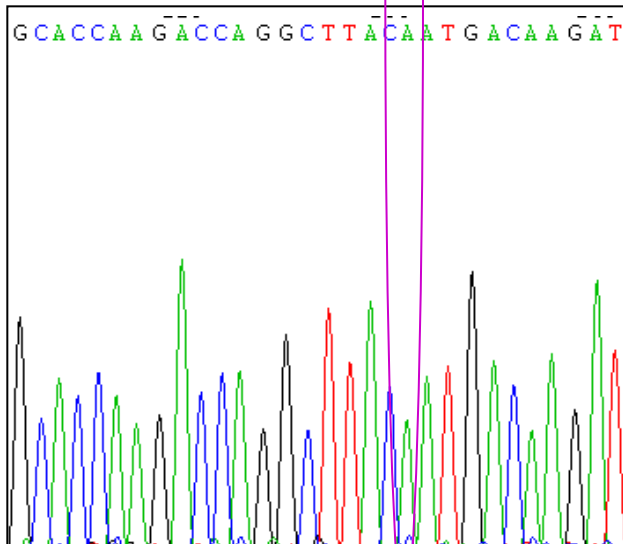


# Sequencing results



CONTROL

Fragment of sequencing  
from *SEMA7A* exon 7 from  
patient and control, showing  
709G>A mutation

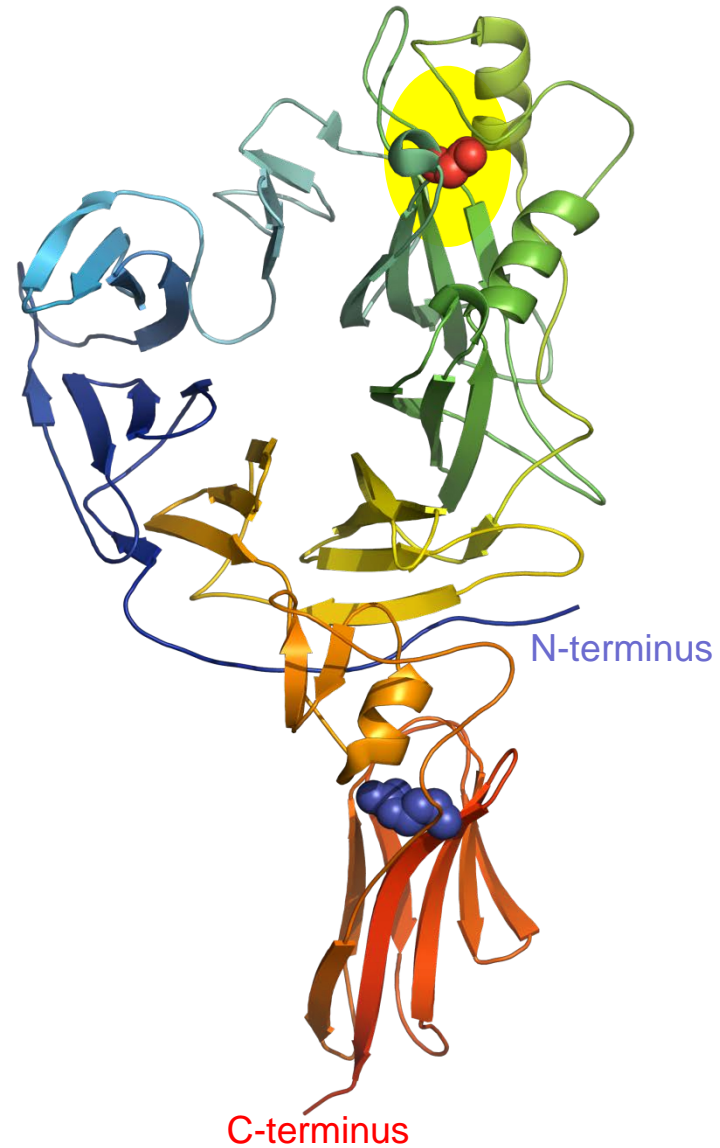


PATIENT

Sequencing results



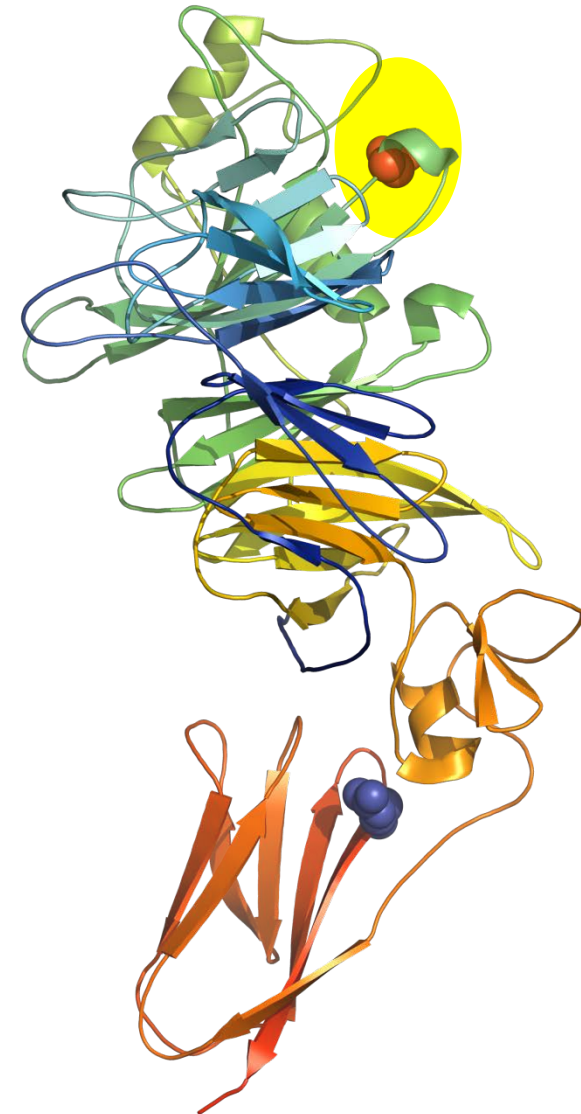
# Sema7A model

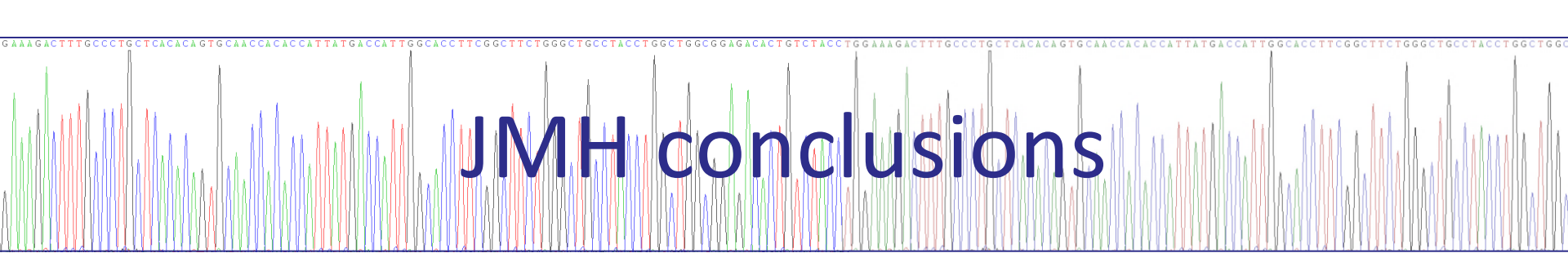


Asp237 sidechain shown  
by red spheres

Solvent-exposed on  
membrane-distal portion  
of protein

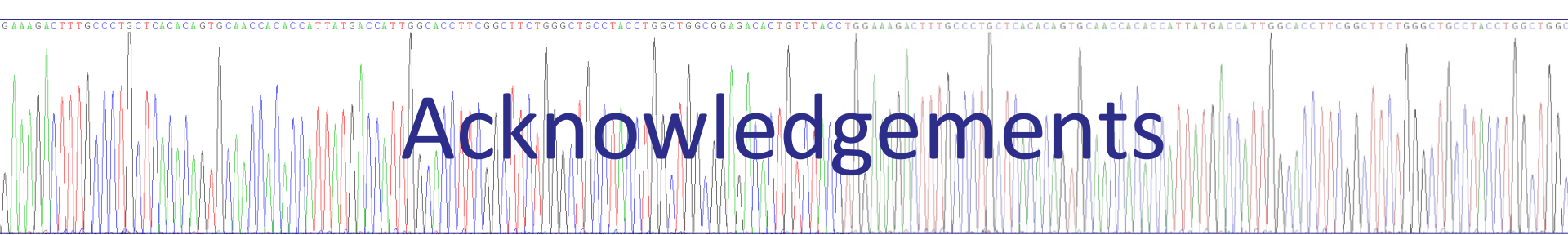
Asp to Asn change leads  
to alteration of charge;  
recognisable by  
antibodies, altering  
antigenicity





# JMH conclusions

- Patient plasma contained anti-JMH, but cells JMH positive (variable reactivity)
- ? Variant JMH antigen
- Sequencing of *SEMA7A* showed homozygosity for 709G>A (Asp237Asn) and 1545A>G (silent)
- Novel allele (*JMH\*01.-07?*), homozygosity results in loss of high frequency JMH antigen (JMH7?) against which patient's antibody is directed



# Acknowledgements

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