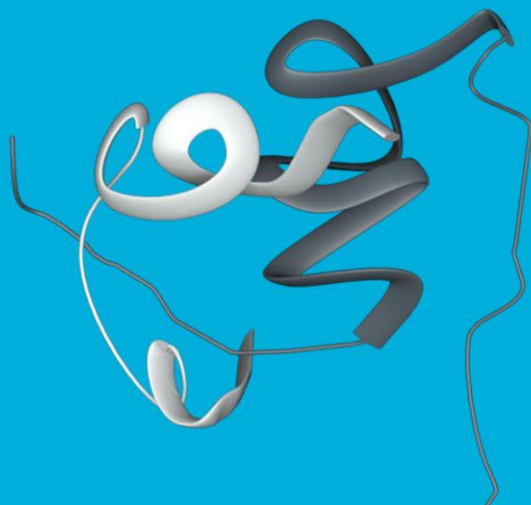


Quality assays = quality diagnostics

Elena B. Volokhina, PhD,
Nijmegen, The Netherlands
31-10-2019

TRANSLATIONAL METABOLIC LABORATORY

"Expert diagnostics of metabolic disorders"



TRANSLATIONAL METABOLIC LABORATORY

Expert diagnostics of metabolic disorders

"Functional testing of unsolved variants"



About us:

The Translational Metabolic Laboratory of the Radboud University Medical Center offers an extensive portfolio of functional tests for diagnosis of metabolic diseases and the characterization of genetic variants of unknown significance. The laboratory is ISO 15189 accredited. Please contact us for further information on how we can help you to put unknown genetic variants into functional context.

Our laboratory offers diagnostics for:



Metabolic diseases



Complement mediated disorders



Mitochondrial disorders



Neurodegenerative disorders



Iron disorders



Congenital disorders of glycosylation

Innovative translational technologies used:

- Functional genomics
- Metabolomics
- Glycoproteomics
- Fluxomics



Ordering and practical questions:

www.metadiagnostics.nl

TML-diagnostics.labgk@radboudumc.nl



The art of quality assays... .

Wat does it mean at a University hospital?

1. Of course all of the usual: sensitivity, specificity, reproducibility, linearity, recovery etc. .

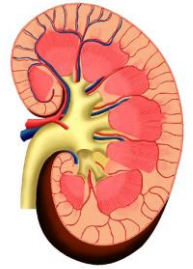
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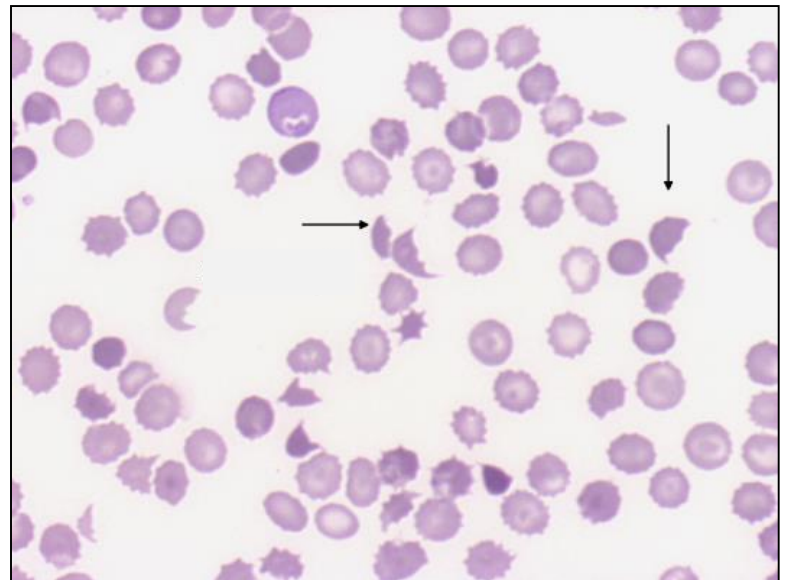
But there is more.

Hemolytic uremic syndrome (HUS)



Thrombotic microangiopathy characterized by:

- Hemolytic anemia
Abnormal breakdown of red blood cells
- Thrombocytopenia
Low platelet count
- Acute renal failure



Hemolytic uremic syndrome (HUS)

Two types of HUS:

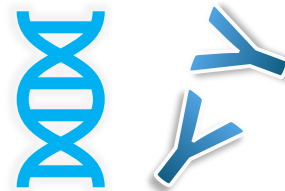
- 90-95%: *STEC*-HUS: infection with Shiga toxin producing *E. coli* (*STEC*)



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- 5-10% atypical HUS (aHUS): genetic defects and autoantibodies in complement (innate immunity). Prevalence 5/1000 000.



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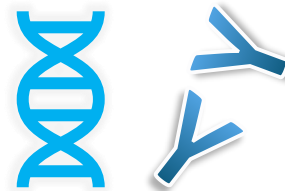
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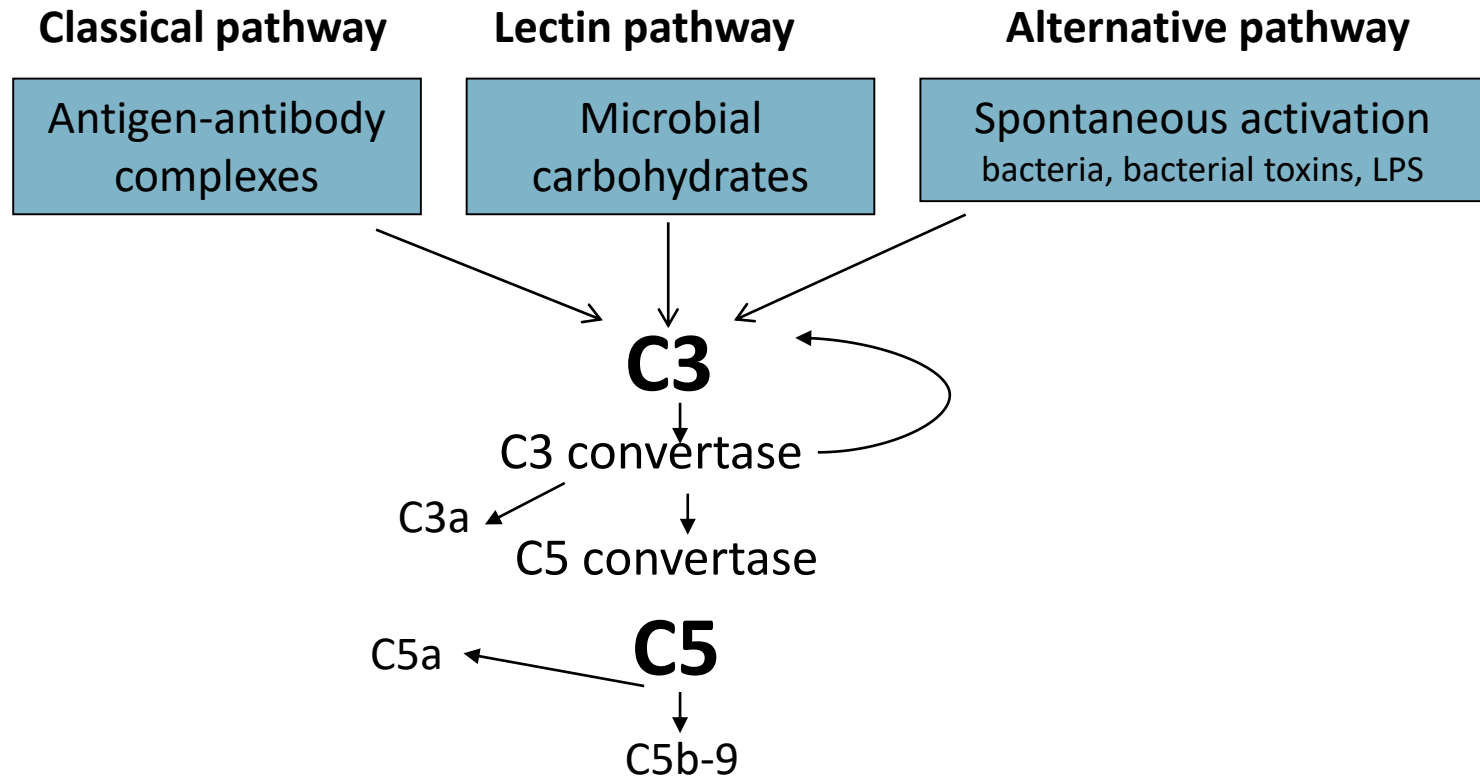
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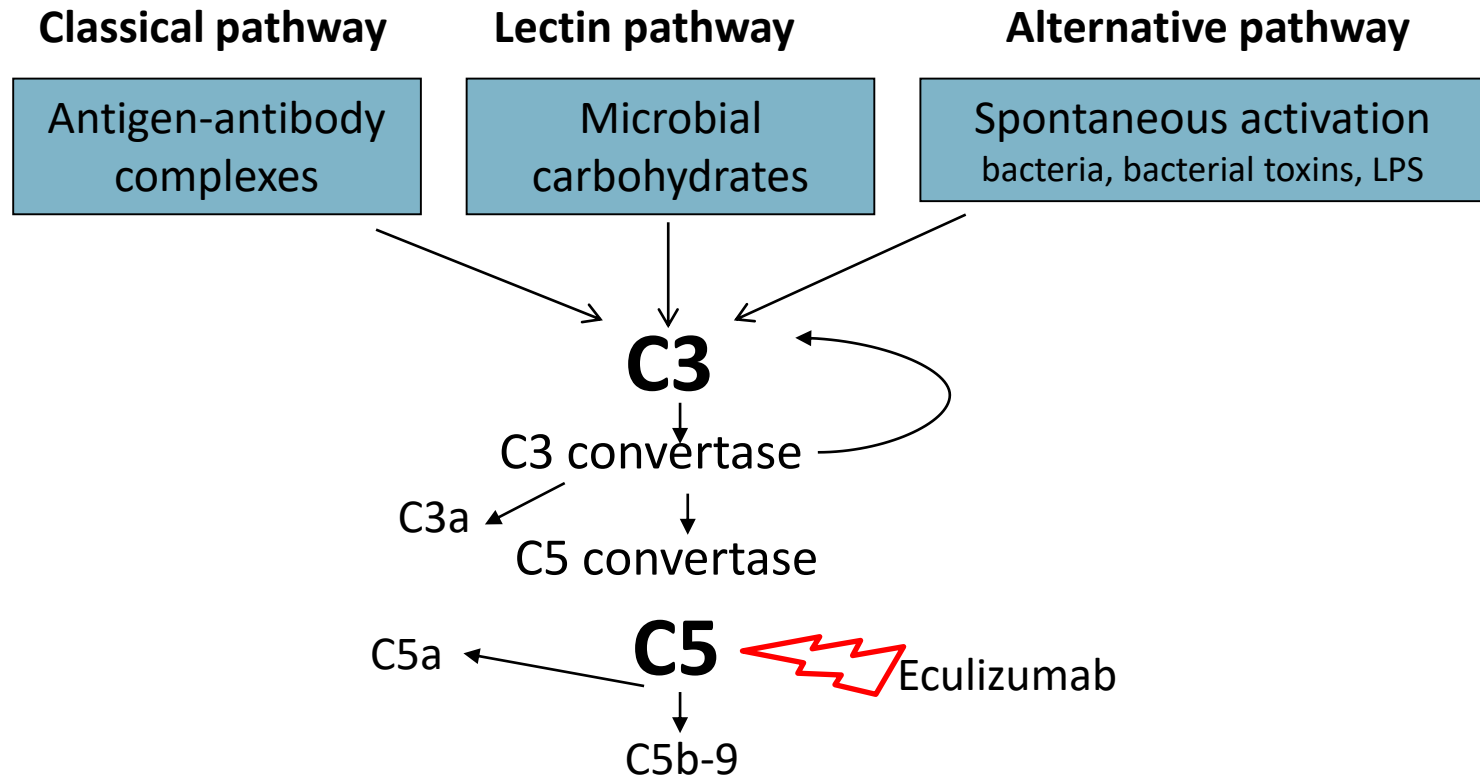
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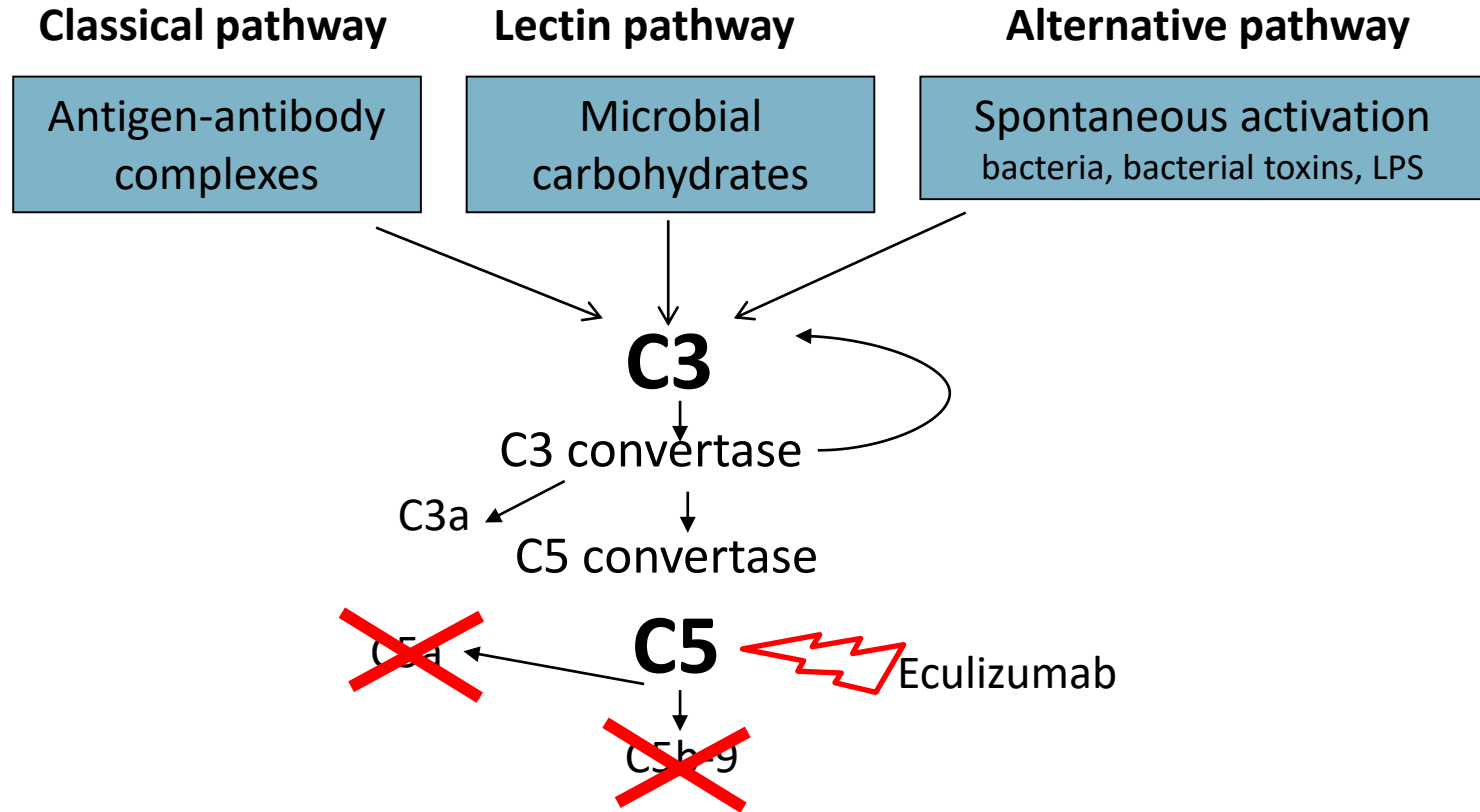
Complement system



Complement system



Complement system



Eculizumab (Soliris) for atypical aHUS



study 2016-2020

Cost-effective treatment for aHUS patients.

<http://cureihus.nl/>



Eculizumab fails to inhibit generation of C5a in vivo.

!!??

Single HELLP syndrome patient

Commercial assay to detect C5a (BD Biosciences)

Question from the bedside

With eculizumab:

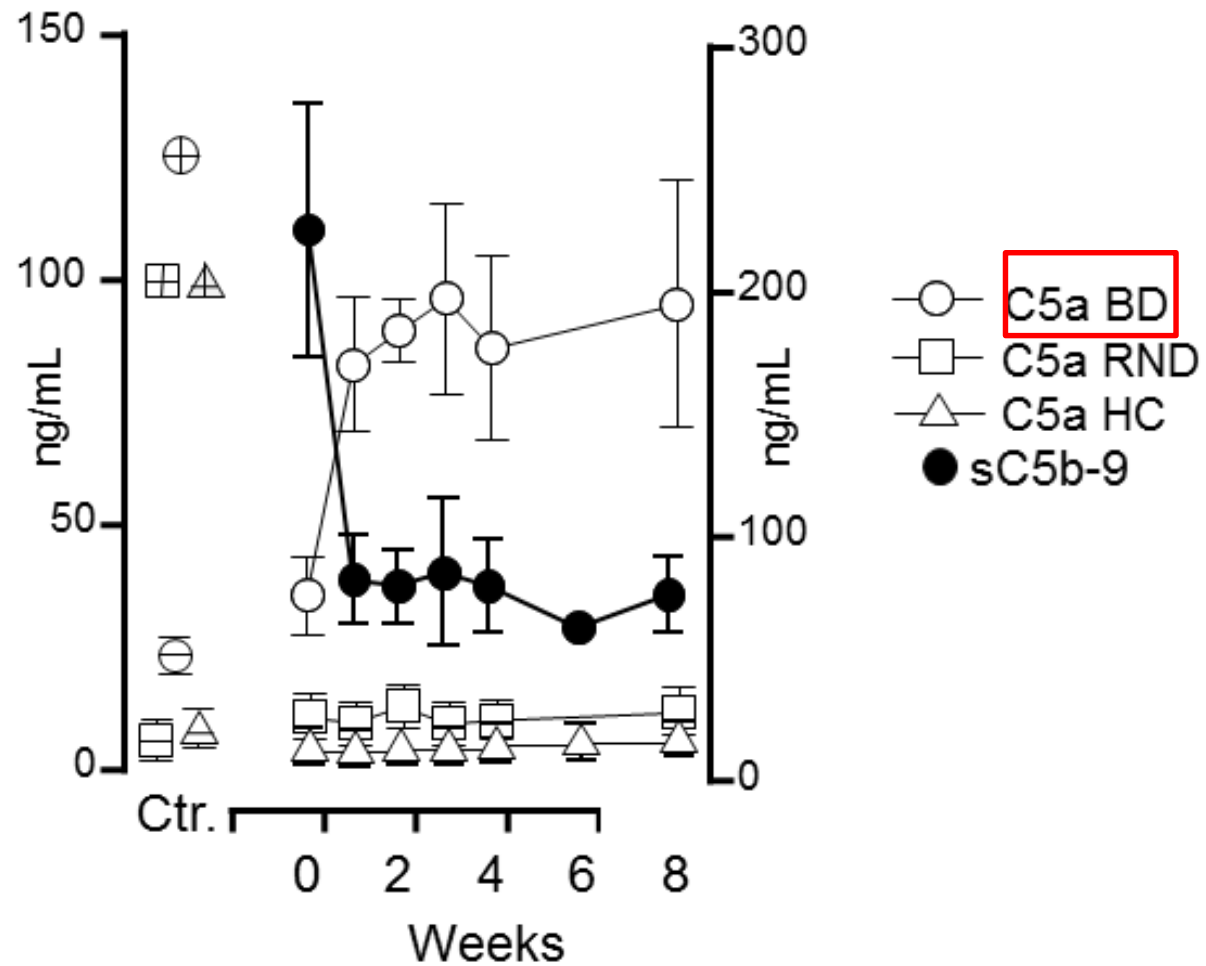
Is my patient still ill due to active C5a??

- 64-73% complete recovery within 26 weeks.

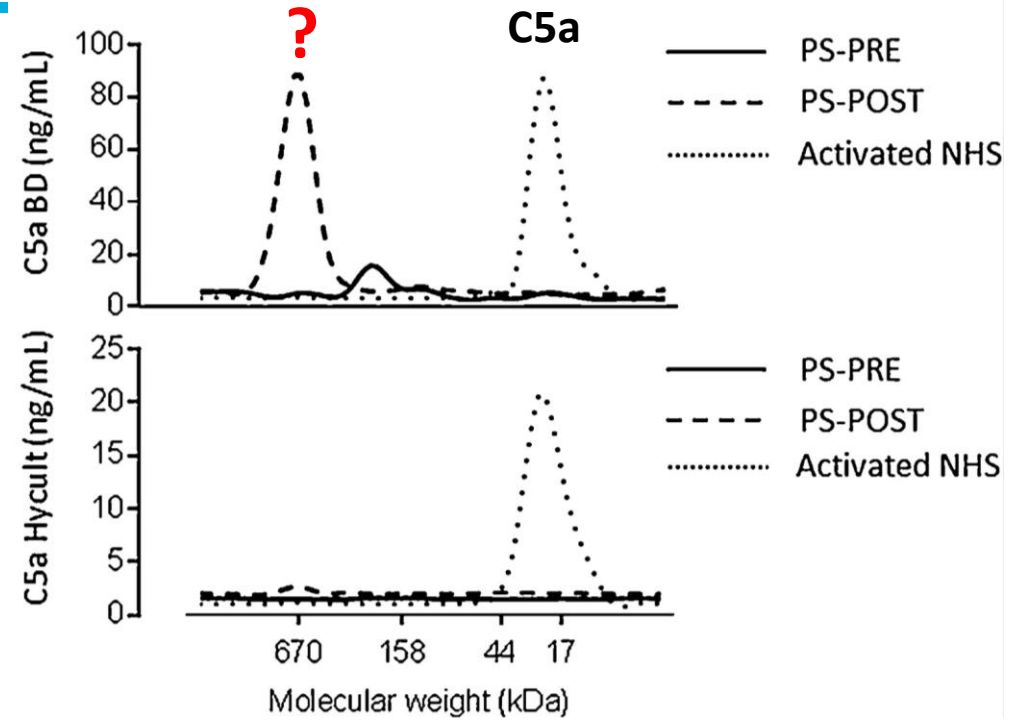
Fakhouri *et al.* Am J Kidney Dis. 2016;68(1):84-93.

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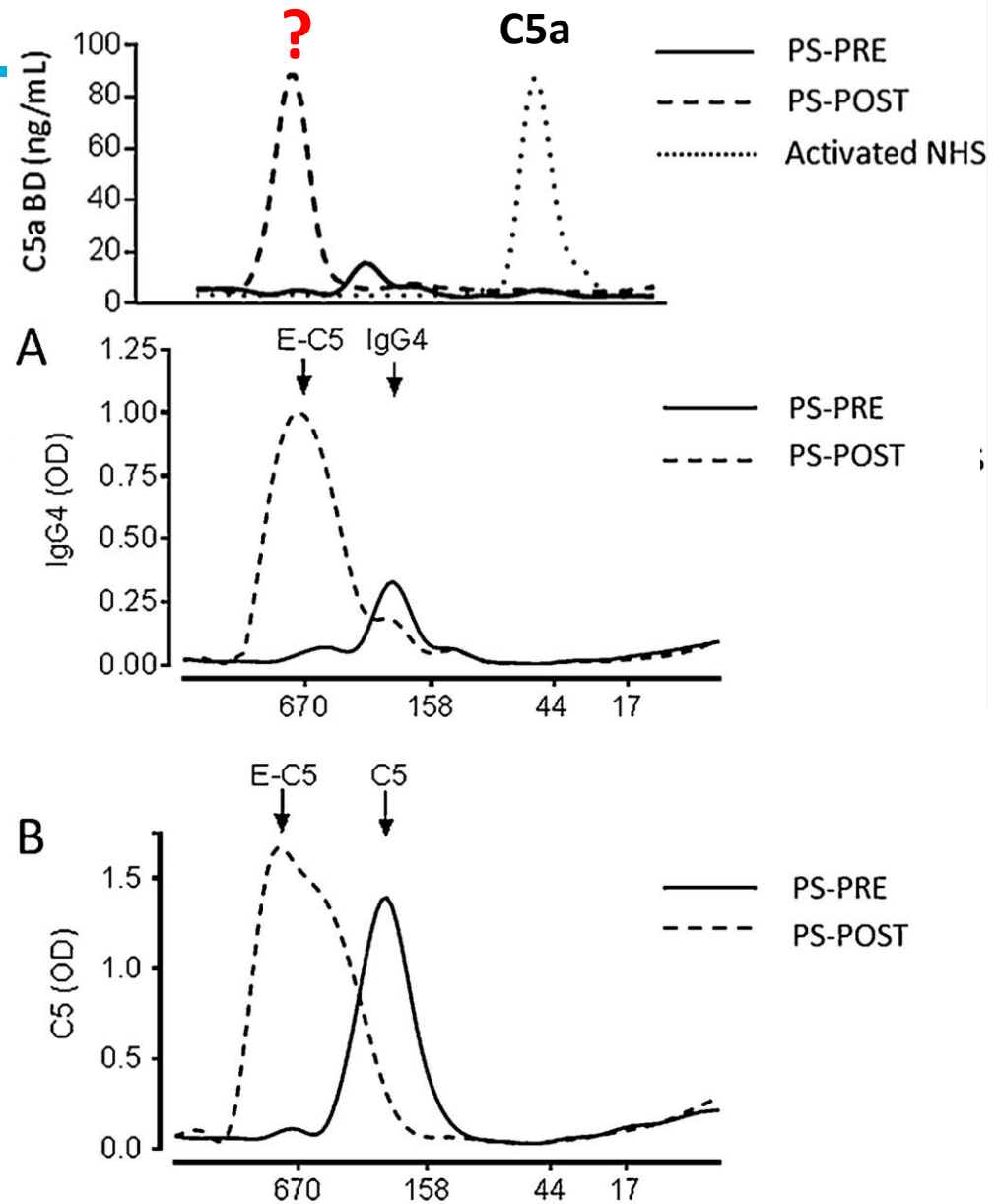
C5a under eculizumab is only detected in BD assay



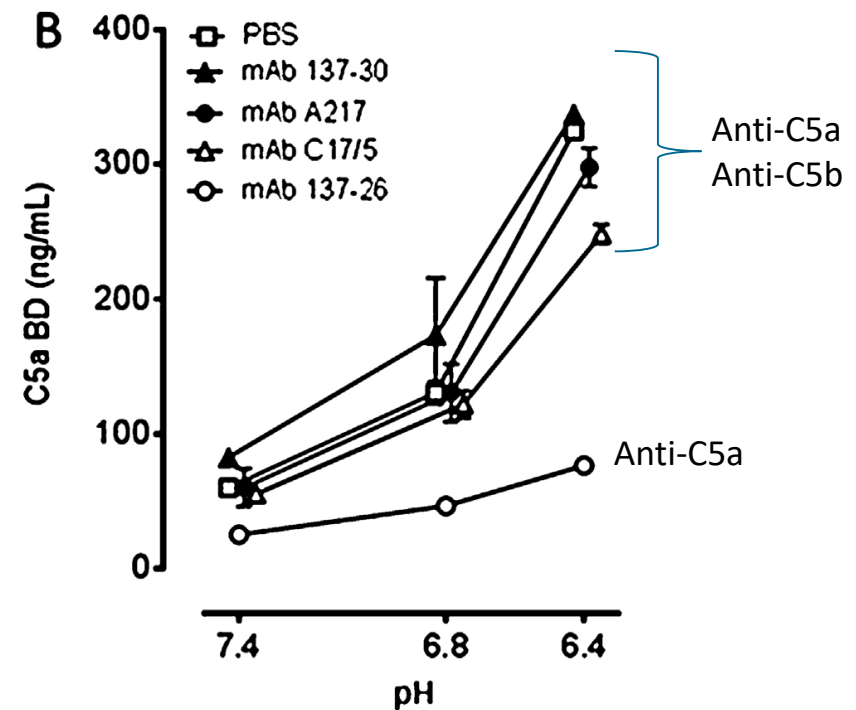
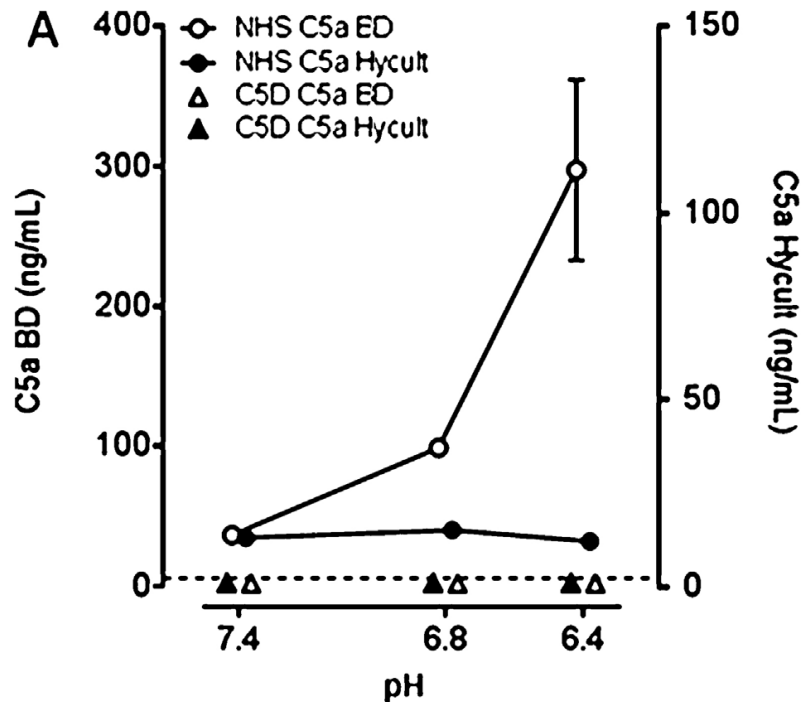
If it is not C5a, what is it???



If it is not C5a, what is it???



Detection of C5a neoepitope in acidified serum

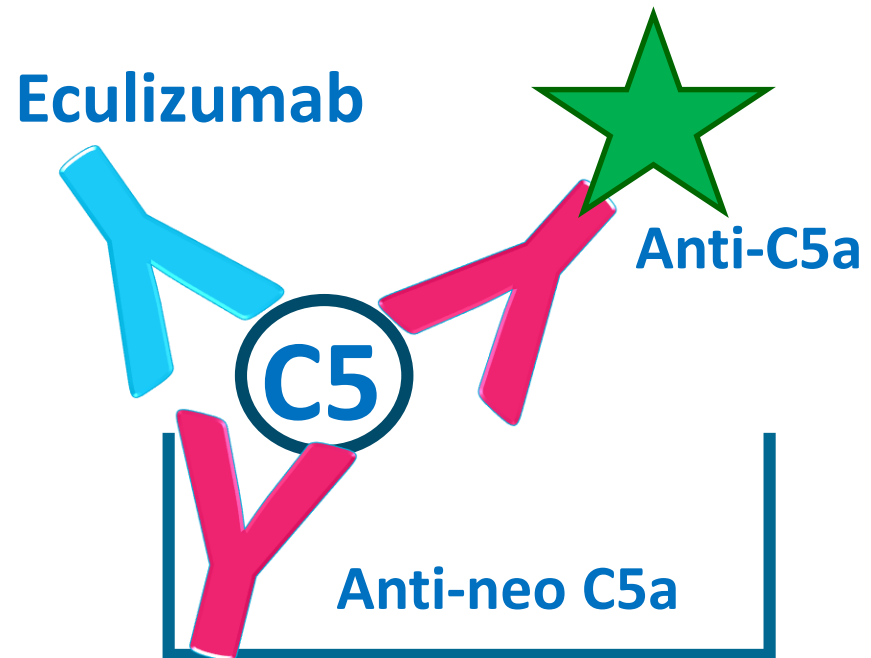


If it is not C5a, what is it???

C5a neoepitope

in C5-eculizumab complex

Interference of drug
with the assay.



Question from the bedside:

Is my patient still ill due to active C5a??

With eculizumab:

- 64-73% complete recovery within 26 weeks.

Answer from the lab:

No, C5a is blocked, you do not need more C5 inhibition.

Fakhouri *et al.* Am J Kidney Dis. 2016;68(1):84-93.

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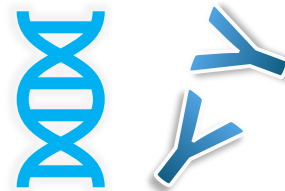
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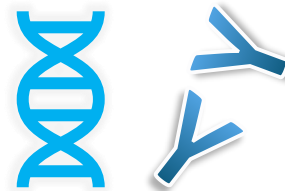
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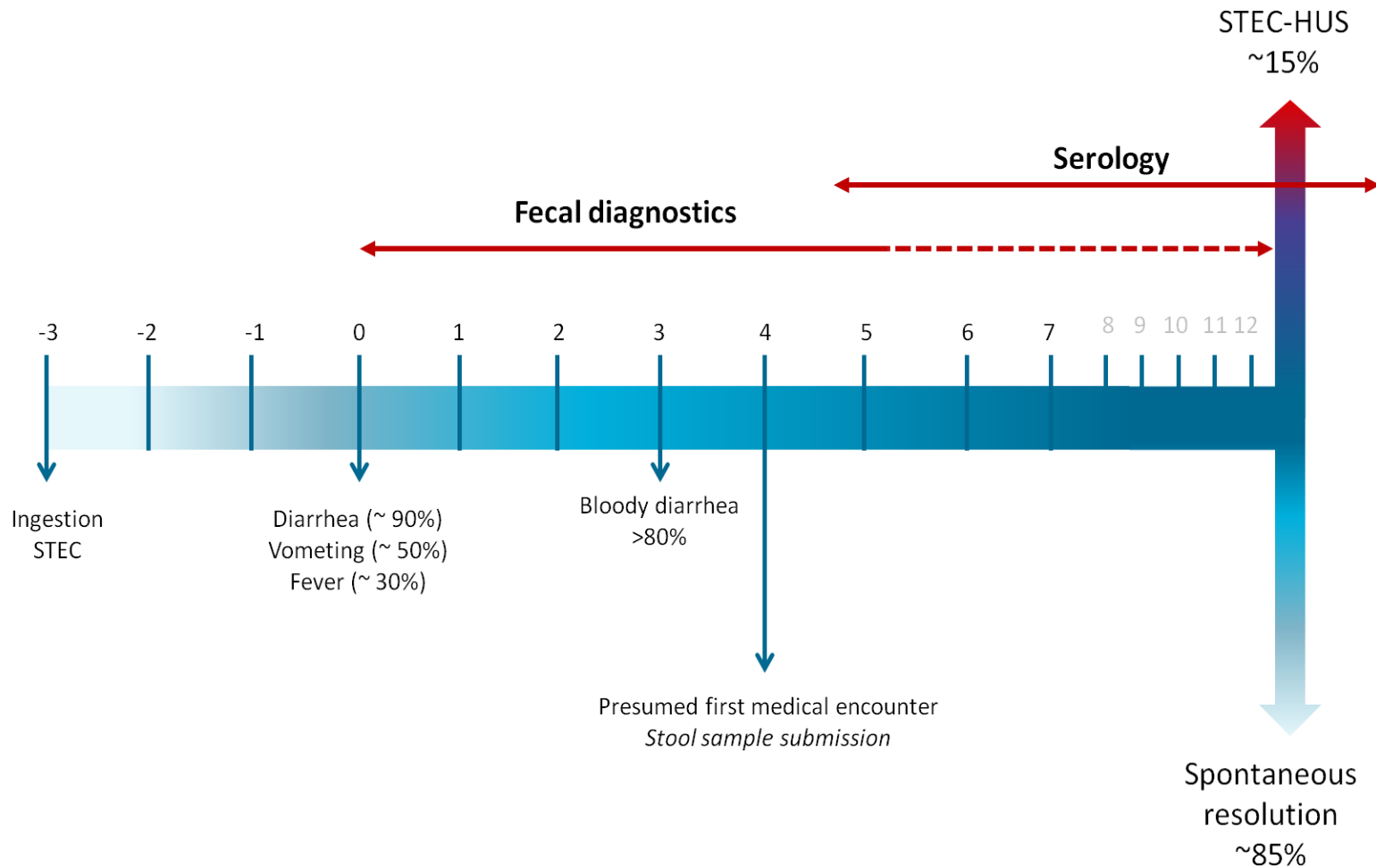


- 5-10% atypical HUS (aHUS): genetic defects and autoantibodies in complement (innate immunity). Prevalence 5/1000 000.



2011: approval of eculizumab for atypical HUS

- Works only for atypical aHUS.
- STEC-HUS must be excluded.



The art of quality assays... .

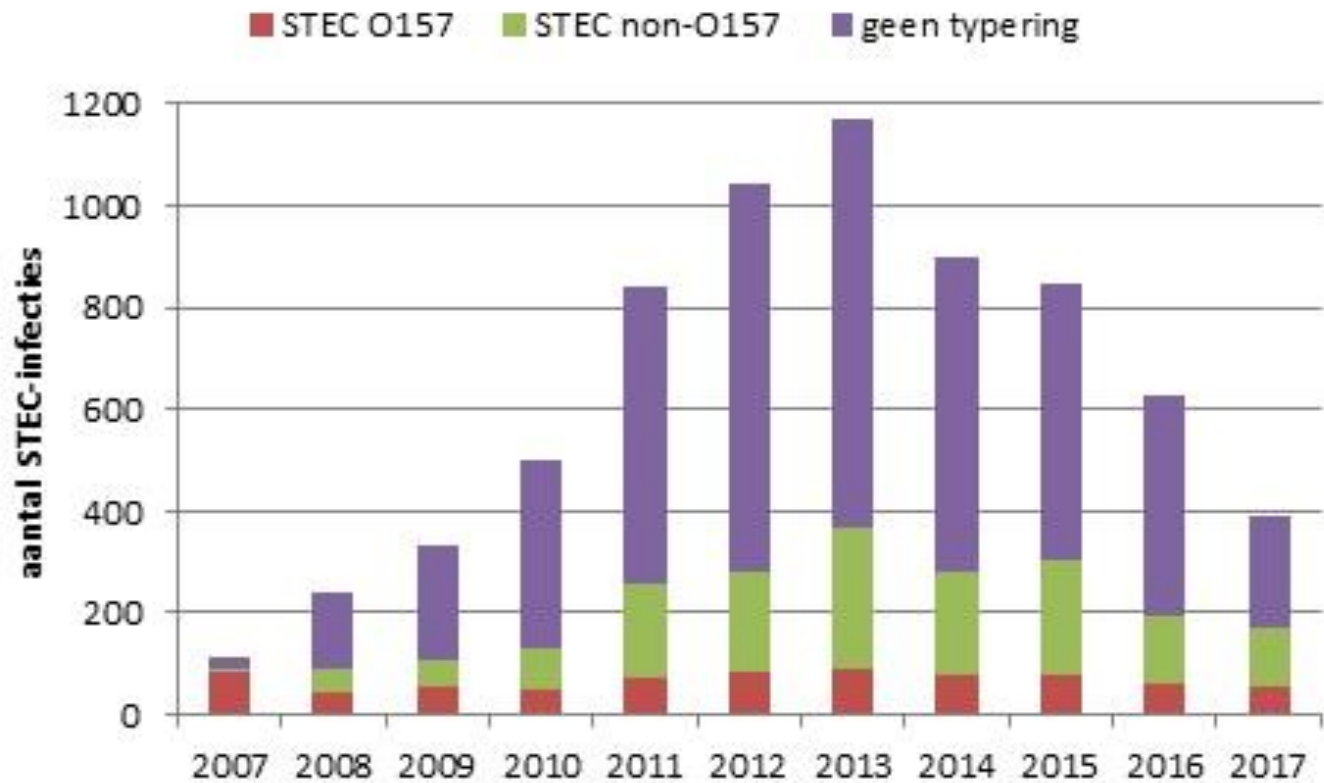
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3. Timing

Fecal diagnostics



RIVM: Surveillance van STEC in Nederland in 2017



Contents lists available at [ScienceDirect](#)

International Journal of Infectious Diseases

journal homepage: www.elsevier.com/locate/ijid



INTERNATIONAL
SOCIETY
FOR INFECTIOUS
DISEASES

Review

Non-O157 Shiga toxin-producing *Escherichia coli*—A poorly appreciated enteric pathogen: Systematic review



SURVEILLANCE AND OUTBREAK REPORT

Outbreak of multiple strains of non-O157 Shiga toxin-producing and enteropathogenic *Escherichia coli* associated with rocket salad, Finland, autumn 2016

INVITED ARTICLE

EMERGING INFECTIONS

James M. Hughes and Mary E. Wilson, Section Editors

The Emerging Clinical Importance of Non-O157 Shiga Toxin-Producing *Escherichia coli*

Development of the STEC serological tests for the market

O157 ready to launch

non-O157 testing feasibility of commercialization



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4. Choice of analytes

The art of quality assays... .

Wat does it mean at a University hospital?

1. Of course all of the usual: sensitivity, specificity, reproducibility, linearity, recovery etc. .

But there is more.

- | | | |
|---------------------------|--|------------------|
| 2. Interfering medication |  | Clinical context |
| 3. Timing | | |
| 4. Choice of analytes | | |

Take home message

- Clinical context: it is all about the patient
- Relevance for particular application
- Be critical to the reagents and data (also your own)
- Work together (clinicians, scientists, industry)



Acknowledgements

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Dr. P. Nilsson

University Medical Center Groningen

Prof. dr. A. Friedrich

Münster University Hospital

Prof. dr. A. Mellmann

Hycult Biotech

Dr. E. Toonen



Dutch national working group on aHUS

