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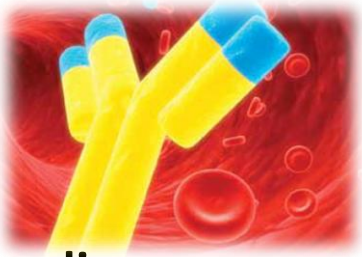
Anti-complement factor H autoantibodies and atypical haemolytic uremic syndrome

Anti-CFH antibodies and aHUS

Haemolytic uremic syndrome (HUS) = microangiopathic haemolytic anaemia and consumptive thrombocytopenia and microvascular glomerular thrombosis, leads to renal failure

Types:

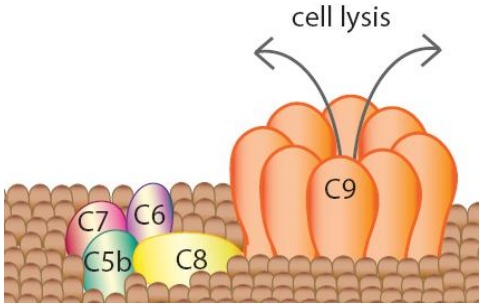
- Typical HUS: **Diarrhea-associated HUS**
  mostly caused by Shiga-like toxin-producing *E.coli* (70-90% of patients)
- Atypical HUS: **non-diarrhea-associated HUS** (**aHUS)**,
  often caused by genetic mutations of complement genes – Factor H, Factor I, Factor B, MCP/CD46,C3
  (10-15% of patients)
- DEAP HUS: **deficiency for CFHR proteins and Factor H autoantibodies**
  (11% of HUS patients)



Anti-CFH antibodies and aHUS

Complement pathway – the defense mechanism of innate immunity against invading microbes and modified self cells

- classical pathway – Ag-Ab complexes (CRP)
- lectin pathway – microbial surfaces (mannose)
- alternative pathway – proactivation condition



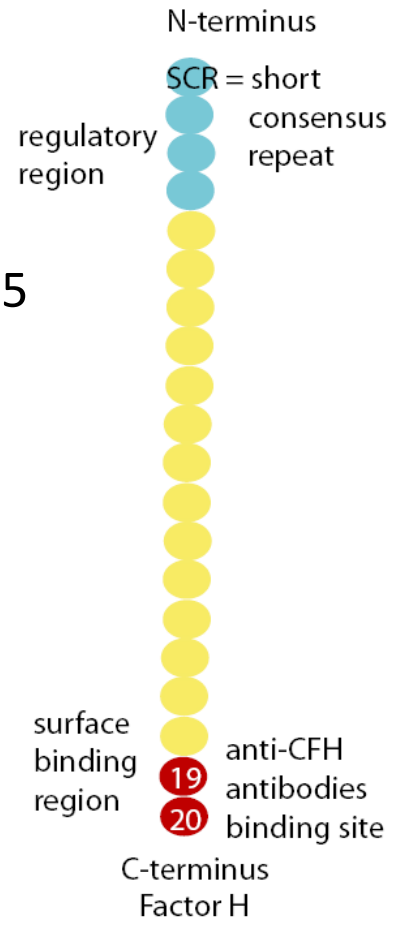
Genetic mutations of complement proteins result in **defective complement regulation**

Type of affected gene is relevant for the disease outcome

Anti-CFH antibodies and aHUS

Factor H & anti-Factor H antibodies

- Factor H is a central inhibitor of alternative complement pathway
- Anti-CFH autoantibodies in patients were first reported in 2005
- binding epitope – C-terminal cell surface attachment region SCRs 19-20
- autoantibodies block cell binding of Factor H which results in enhanced complement activation – enhanced cells damage (e.g. endotelial cells)
- genetic analysis of the patients – CFHR1 and CFHR3 deletion
- DEAP HUS therapy - fresh frozen plasma infusion, plasma exchange, immunosuppresive therapy



Anti-CFH antibodies and aHUS

Anti-CFH antibodies detection

- **ELISA-VIDITEST anti-complement factor H**
- First CE IVD kit available

Kit components:

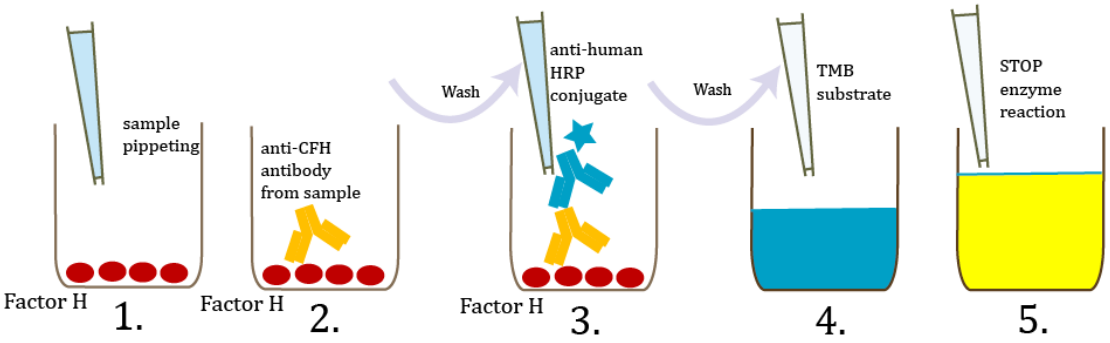
- ELISA Strips coated with Factor H
- Standard 10 000 AU/mL
- Anti-human HRP conjugate
- Dilution buffer
- Wash buffer
- TMB substrate
- STOP solution

Cat no. VID-ODZ-166

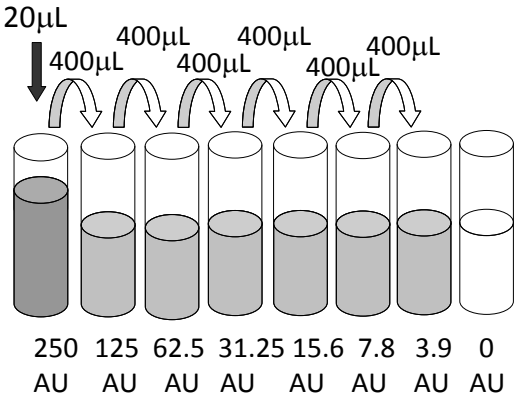
Anti-CFH antibodies and aHUS

Test procedure

- serum samples dilution 1:100
- standard dilution
- ELISA

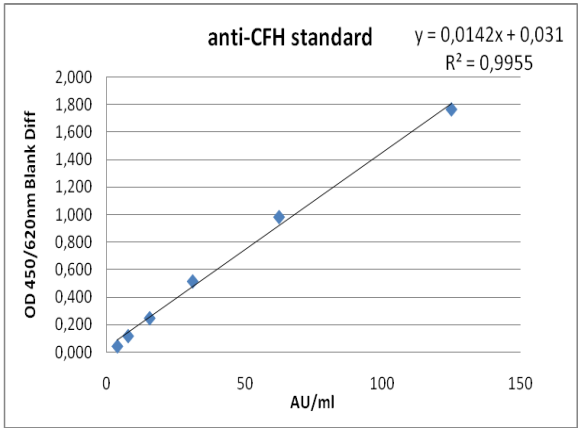


Standard
anti-CFH IgG
10 000 AU/mL



absorbance reading

6.



- data processing

Anti-CFH antibodies and aHUS

ELISA-VIDITEST anti-complement factor H Evaluation study

- Performed in **The Institute of Hematology and Blood Transfusion** (Prague) and **VIDIA spol. s r.o**
- **Samples:**

  Negative samples - serum and plasma samples from blood donors – 130 samples
  Acquired thrombotic thrombocytopenic purpura (TPP) (acute stage) – 20 samples
  Atypical HUS (with CFH or MCP mutation) – 8 samples
  Inherited TPP – 5 samples
  DEAP HUS (deficiency of CFHR plasma proteins and factor H autoantibody positive HUS) – 9 samples

- **Results:**

Cut-off value for serum samples = 27 AU/mL
Cut-off value for plasma samples = 18 AU/mL

Diagnostic sensitivity
DEAP-HUS samples: 9
Positive using anti-Complement factor H test: 9
Diagnostic sensitivity: 9/9 = 100%

Diagnostic specificity
Blood donors samples: 130
Negative using anti-Complement factor H test: 128
Diagnostic specificity: 128/130 = 98.5%

Anti-CFH IgG determination in patients with TPP, aHUS and DEAP HUS

- high anti-CFH IgG concentration in DEAP HUS patients
- low anti-CFH IgG concentration in other diseases clinically close to HUS

- **Final:**

  ELISA-VIDITEST anti-Complement factor H fulfil the requirements for the detection of anti-CFH antibodies and *in vitro* diagnostics of DEAP HUS

Disease	Patient	anti-CFH IgG (AU/mL)	other tests (ELISA, genetic testing)
Acquired TPP	1	10	antibodies to ADAMTS13
	2	10	antibodies to ADAMTS13
	3	10	antibodies to ADAMTS13
	4	19	antibodies to ADAMTS13
	5	17	antibodies to ADAMTS13
	6	10	antibodies to ADAMTS13
	7	9	antibodies to ADAMTS13
	8	9	antibodies to ADAMTS13
	9	16	antibodies to ADAMTS13
	10	13	antibodies to ADAMTS13
	11	8	antibodies to ADAMTS13
	12	11	antibodies to ADAMTS13
	13	<4	antibodies to ADAMTS13
	14	7	antibodies to ADAMTS13
	15	22	antibodies to ADAMTS13
	16	6	antibodies to ADAMTS13
	17	22	antibodies to ADAMTS13
	18	20	antibodies to ADAMTS13
	19	12	antibodies to ADAMTS13
	20	9	antibodies to ADAMTS13
Atypical HUS	1	12	CFH deficiency
	2	11	MCP deficiency
	3	19	CFH deficiency
	4	10	CFH deficiency
	5	18	CFH deficiency
	6	17	CFH deficiency
	7	8	MCP deficiency
	8	8	MCP deficiency
Inherited TPP	1	10	ADAMTS13 deficiency
	2	10	ADAMTS13 deficiency
	3	43	ADAMTS13 deficiency
	4	13	ADAMTS13 deficiency
	5	18	ADAMTS13 deficiency
DEAP-HUS	1	427	del CFHR1 homozyg
	2	251	del CFHR1 homozyg
	3	800	del CFHR1 homozyg
	4	232	del CFHR1 homozyg
	5	316	del CFHR1 homozyg
	6	403	del CFHR1 homozyg
	7	64	del CFHR1 homozyg
	8	53	del CFHR1 heteroyz
	9	79	del CFHR1 homozyg

has to be checked for CFHR 1/3 deletion