ADAMTS13 – 10 Years Later


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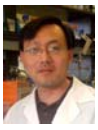
Elaine Majerus


Weiqiang Gao


Hendrik Feys

- Pathophysiology of thrombotic thrombocytopenic purpura (TTP)
- Proteolysis of von Willebrand factor (VWF) by ADAMTS13
- Clinical uses of ADAMTS13 data

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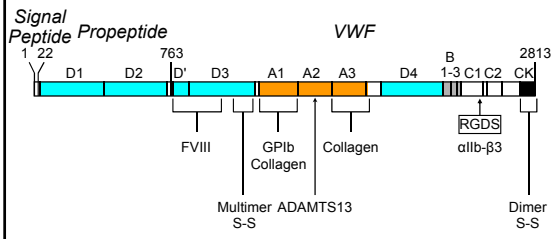
The Von Willebrand Factor Precursor



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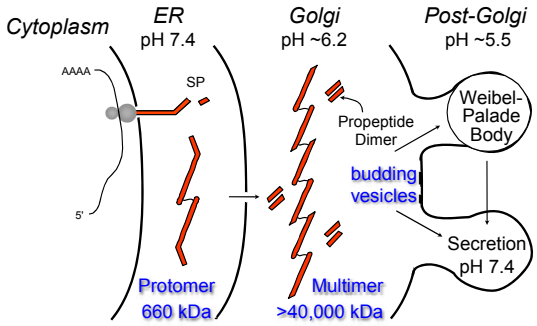
Biosynthesis of VWF



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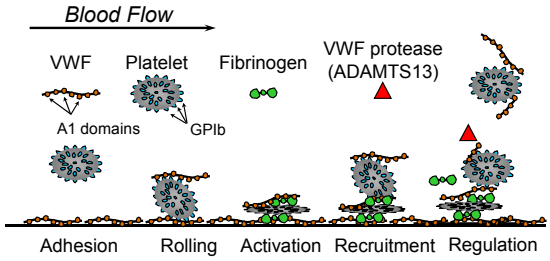
VWF and Platelet Adhesion



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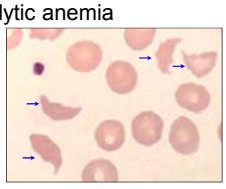
Thrombotic Thrombocytopenic Purpura

A Disorder of VWF Proteolysis

A classic pentad of signs:

- Microangiopathic hemolytic anemia
- Thrombocytopenia
- Neurologic dysfunction
- Renal disease
- Fever



≈ 4 per million incidence

Strikes mainly young adult women

Untreated, mortality >90%

Treated with plasma exchange, mortality <20%

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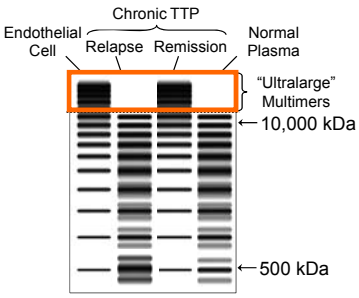
VWF Multimers in TTP



Failure to cleave ultralarge VWF causes TTP?

Moake et al, *N Engl J Med* 1982; **307:** 1432-1435

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VWF Cleaving Protease in Plasma

- 1982: Predicted by Moake
- 1996: Discovered by Tsai and Furlan
- 1997: Absent in congenital TTP
- 1998: Absent in most idiopathic TTP (acquired IgG autoantibody inhibitor)

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VWF Cleaving Protease (ADAMTS13)

A Disintegrin-like And Metalloprotease with Thrombo Spondin-1 repeats

Zheng et al, *J Biol Chem* 2001; **276**: 41059-63
Levy et al, *Nature* 2001; **413**: 488-94
Soejima et al, *J Biochem (Tokyo)* 2001; **130**: 475-80

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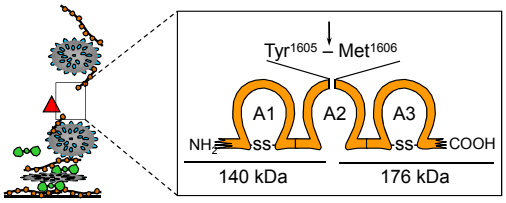
Shear and VWF Proteolysis



Proteolysis increased by:
- Shear stress (aortic stenosis)
- VWD type 2A and 2B mutations

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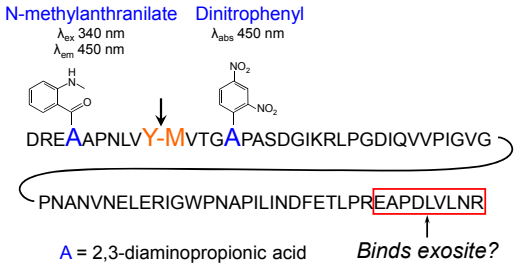
ADAMTS13 Assays: FRETS-VWF73

Asp1596-Arg1668



Kokame et al, *Br J Haematol* 2005; **129**: 93-100

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Why is ADAMTS13 Specific for VWF?

ADAMTS13 is constitutively active

"Stretched" or "sheared" VWF is the only known substrate

No ADAMTS13 inhibitors in vivo

Plasma is a tough environment:
- [ADAMTS13] = 1 µg/ml
- [VWF] = 10 µg/ml
- [Total Protein] = 80 mg/ml (8,000 x higher)

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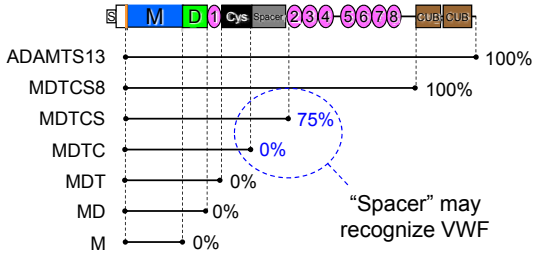
Recombinant ADAMTS13 Activity

Against Plasma VWF



Zheng et al, *J Biol Chem* 2003; **278**: 30136-41

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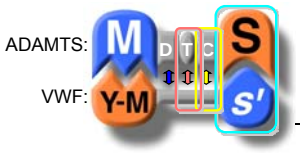
More Substrate-Protease Interactions



	MD	MDT	MDTC	MDTCS
	k_{cat}/K_m (μM^{-1} s^{-1})			
VWF73 Y-M—EAPDLVLQR^{1668}			0.1	2.0
VWF64 Y-M—R^{1659}	0.01	0.01	0.1	0.1
VWF46 Y-M—R^{1641}	0.0003	0.004	0.01	
VWF35 Y-M—V^{1630}	0.0009	0.005	0.01	

Gao et al, *Blood* 2008; **112**: 1713-9

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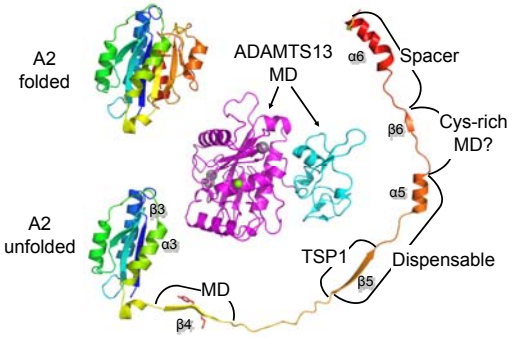
ADAMTS13-A2 Interactions



Gao et al, *Blood* 2008; **112**: 1713-9

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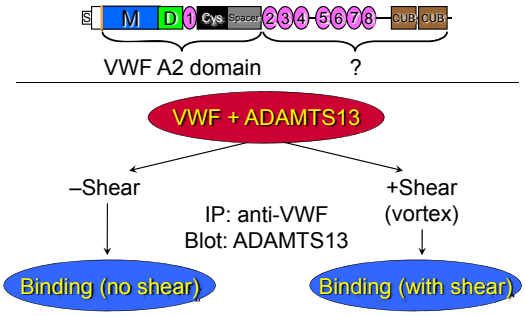
ADAMTS13 Binding to VWF



Feys et al, *J Thromb Haemost* 2009; in press

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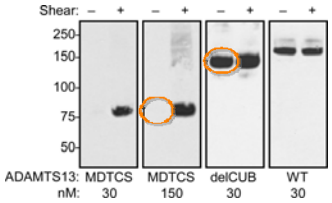
ADAMTS13 Binding to VWF

IP: anti-VWF
Blot: anti- ADAMTS13



- Distal TSP domains bind unsheared VWF
- MDTCS is sufficient to bind sheared VWF

Feys et al, *J Thromb Haemost* 2009; in press

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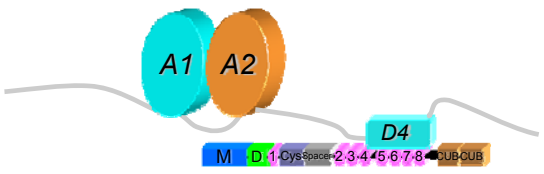
VWF Cleaving Complex



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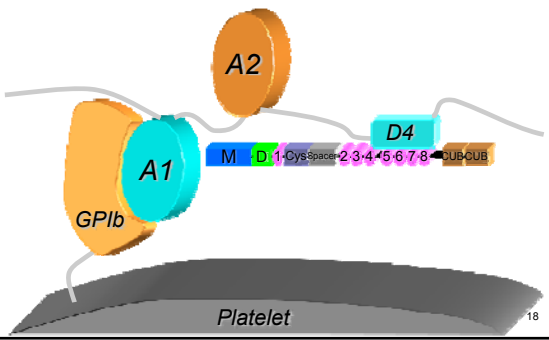
VWF Cleaving Complex



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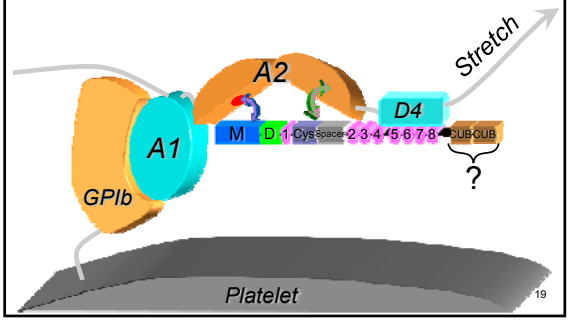
VWF Cleaving Complex



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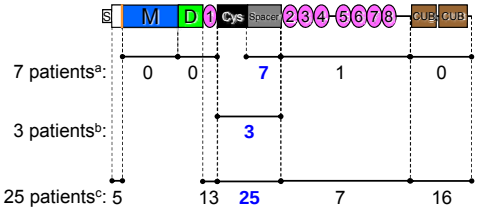
ADAMTS13 Inhibitors in TTP

Epitopes Map to Cys-Rich + Spacer



[a] Luken et al, *Thromb Haemost* 2005; **93:** 267-274
[b] Soejima et al, *Blood* 2003; **102:** 3232-3237
[c] Klaus et al, *Blood* 2004; **103:** 4514-4519

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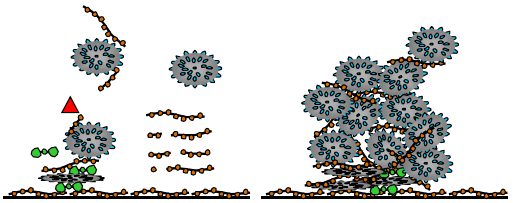
VWF, ADAMTS13 and TTP

With ADAMTS13 *Without ADAMTS13 (<5%)*



Normal VWF Multimers
Normal Hemostasis

Ultralarge VWF Multimers
Microvascular Thrombosis
(TTP)

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Case 1

HPI: 47 year old AA female with 2 days of mild gingival bleeding, severe headache, intermittent visual blurring

PE: Temp 36.8 ˚C, Pale

CBC:
- Hgb 7.4
- Platelets 14K
- 20 schistocytes/HPF

Other Labs:
- Cr 0.88 mg/dL
- LDH 1317 IU/L
- Haptoglobin <8 mg/dL

Diagnosis: Thrombotic thrombocytopenic purpura

Treatment: Prednisone, plasma exchange

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TTP – Clinical Course

80% respond to plasma exchange in average of 16 days (range, 3-36)

20% dead within with 5 weeks

40% of responders have exacerbations within one week

30% of responders relapse within 2 years

Rock et al, *New Eng J Med* 1991; **325:** 393-397

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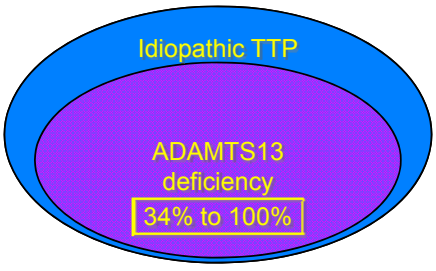
Autoimmune Idiopathic TTP

Acquired ADAMTS13 Deficiency



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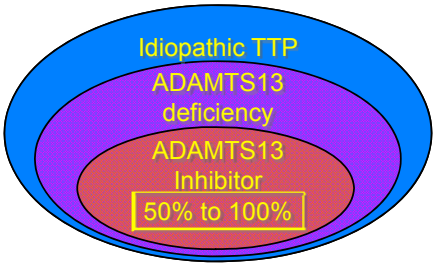
Autoimmune Idiopathic TTP
Acquired ADAMTS13 Deficiency



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ADAMTS13 – Clinical Correlations

ADAMTS13 deficiency (<5-10%) predicts:
- "Idiopathic" (primary) TTP
- Response to plasma exchange
- Survival (long term)
- Relapse

ADAMTS13 inhibitor predicts:
- Prolonged time to complete response
- Death (early)
- Relapse

Reviewed in Coppo et al, *Br J Haematol* 2005; **132:** 66-74
(≈391 total patients)

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ADAMTS13 – Clinical Correlations

Character	Odds Ratio
ADAMTS13 <5%:	
• Survival	≈5
• Primary TTP	≈8
• Female Gender	≈4
• African Ancestry	≈8
• Obesity	High
ADAMTS13 Inhibitor Present:	
• Relapse	≈13

Zheng et al, *Blood* 2004; **103**: 4043-9
Vesely et al, *Blood* 2003; **102**: 60-8
Veyradier et al, *Blood* 2001; **98**: 1765-72

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ADAMTS13 Deficiency and Relapse

At Presentation:
- ADAMTS13 < 5%: 30% relapse in 2 years, and risk continues
- For example, *at WUMS: 11/16 (70%) relapsed, 5/16 (30%) dead, by 6 years of follow up
- ADAMTS13 > 5%: < 9% relapse in 2 years

*(follow up of patients in) Zheng et al, *Blood* 2004; **103**: 4043-9

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ADAMTS13 Deficiency and Relapse

In Remission:
- ADAMTS13 < 10%: 21/35 (60%) relapsed
- ADAMTS13 > 10%: 14/74 (19%) relapsed

At Relapse:
- ADAMTS13 < 5%: all patients

Peyvandi et al. *Haematologica* 2008; **93:** 232-9

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Case 1 (continued)

Clinical Course: Complete response to prednisone 60 mg/d, plasma exchange daily for 12 days. Discharged. Prednisone tapered over 2 weeks.

CBC (at discharge):
- Hgb 10.5
- Platelets 225K
- 2 schistocytes/HPF

Other Labs:
- Cr 0.77 mg/dL
- LDH 189 IU/L
- Haptoglobin 250 mg/dL

ADAMTS13 (at diagnosis):
- Activity <5%
- Inhibitor 2.0 U/ml

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Case 1 (continued)

Follow up: Day 31 after discharge presented with headache, dysarthria, and left arm weakness.

CBC:
- Hgb 11.6
- Platelets 78K
- 2 schistocytes/HPF

ADAMTS13 (at relapse):
- Activity <5%
- Inhibitor 1.9 U/ml

Other Labs:
- Cr 0.78 mg/dL
- LDH 413 IU/L
- Haptoglobin 145 mg/dL
- U/A – no blood
- Troponin 0.94 ng/ml

Diagnosis: Relapsed TTP (CNS, cardiac involvement)

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Case 2

HPI: 25 year old AA male found down with right hemiparesis. Discharged 21 days ago for relapsed TTP (2 yrs), treated with PE, rituximab x 1. Did not keep follow up appointments.

PE: Temp 36.5 ˚C, BP 114/63, Pale, R hemiparesis

CBC:
- Hgb 5.1
- Platelets 346K
- 0-1 schistocytes/HPF

Other Labs:
- Cr 1.31 mg/dL
- LDH 187 IU/L
- Haptoglobin 251 mg/dL

MRI: Acute to subacute infarcts medial L parietal lobe and deep in L cerebral white matter

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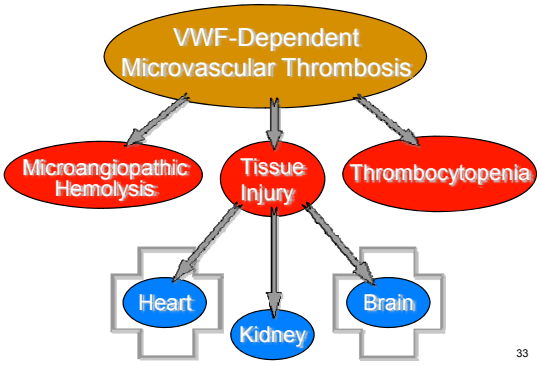
Thrombosis Preceding TTP?



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TTP – What is the Endpoint of Therapy?

Can ADAMT13 assays diagnose atypical presentations of TTP or predict relapses?

Should the goal of treatment be a normal ADAMTS13 level?

Does plasma exchange help patients with normal ADAMTS13?

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TM/H Network STAR Trial
"Study of TTP And Rituximab"

Phase 3, randomized, PE with or without adjuvant rituximab

- >13 institutions, 3 years, 238 patients with "idiopathic TTP"
- Frequency and time to response, relapse
- All cause mortality
- ADAMTS13 data analyzed *post hoc*
- PE failures can receive salvage rituximab

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Case 1 (continued)

Clinical Course: Complete response to prednisone 60 mg/d and plasma exchange after 4 days. Neurological deficits resolved in 3 days.

CBC:
- Hgb 10.8
- Platelets 155K
- 1 schistocytes/HPF

Other Labs:
- Cr 0.92 mg/dL
- LDH 168 IU/L
- Troponin <0.07 ng/ml

Clinical Course (cont): PE every other day. Rituximab 375 mg/m^2 (#1) and discharged 4 days later. Completed rituximab #2,3,4, still in remission, ADAMTS13 pending…

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Case 2 (continued)

Diagnosis: Relapsed TTP (without active MAHA)

Course: PE daily with resolution of UE weakness by day 2, persistent LE weakness requiring a walker. Rituximab #2,3,4 given in hospital...

ADAMTS13:

Date	Activity	Inhibitor	Comment
3/6/07	<5%	<0.4 U/ml	Diagnosis
3/30/07	<5%	3.2 U/ml	Refractory to PE
8/18/09	<5%	3.6 U/ml	Relapse
9/25/09	<5%	<0.4 U/ml	Exacerbation

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ADAMTS13 – 10 Years Later

Look for hemolysis and schistocytes in all patients with anemia and thrombocytopenia

Severe ADAMTS13 deficiency is:

- Exceedingly rare in secondary TTP, etc
- Common (80%) in primary TTP

ADAMTS13 inhibitors correlate with:

- Relapse risk
- Early death

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ADAMTS13 – 10 Years Later

Lethal thrombosis can occur without MAHA

Persistent or recurrent ADAMTS13 deficiency predicts relapse

Treatment to normalize ADAMTS13 may prevent relapses

Should ADAMTS13 testing be routine?

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ADAMTS13 – 10 Years Later

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Elaine Majerus
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