Organizado por:

Clínica Universidad de Navarra

PUESTA AL DÍA

HEMATOLOGÍA EN 48H [LO QUE DEBES CONOCER PARA TU PRÁCTICA CLÍNICA]

X EDICIÓN

ACTUALÍZATE 48 HORAS

Procedimiento diagnóstico en las Microangiopatías Trombóticas

Dra. Cristina Pascual Izquierdo, MD, PhD
Jefe Sección Servicio de Hematología y Hemoterapia.
Profesor Asociado UCM
Hospital General Universitario Gregorio Marañón
Instituto de Investigación Gregorio Marañón

SaludMadrid Comunidad de Madrid

Hospital General Universitario Gregorio Marañón

IGM

Universitas Complutensis Matritensis

Disclousures

- **Grant/Research support:** Sanofi, Sobi,
- **Advisory board:** Amgen, Bayer, Boehringer Ingelheim, Novartis, Sanofi, Sobi, Takeda.
- **Speaker bureau:** Amgen, Boehringer Ingelheim, Grifols, Novartis, Sanofi, Takeda, Werfen

Caso clínico

- Mujer de 50 años, sana y sin FRCV
- Enviada al Servicio de Urgencias por aparición de hematomas, y astenia

Caso clínico

- Mujer de 50 años, sana y sin FRCV
- Enviada al Servicio de Urgencias por aparición de hematomas, y astenia

-Hemograma: **Hb 8.8 gr/dl,** Hct 25.5% VCM 96 fl, HCM33, plaquetas **$5x10^9$/L,** leucocitos 3200 (Neu 2.0, Lin 1.0). Retis 7.45%, absolutos 198000/mcL
-Hemostasia: INR 1, Fibrinógeno 392mg/dl, APTT 29.7"/31".
Coomb directo: NEGATIVO
-Bioquímica: **BT 1.4mg/dl, LDH 664U/L,** Cr 2.20 mg/dl, Na 139mmol/L, K 3.7mmol/L, **Haptoglobina <6.** Troponina 15.2ng/L

Caso clínico

- Mujer de 50 años, sana y sin FRCV
- Enviada al Servicio de Urgencias por aparición de hematomas, y astenia

-**Hemograma:** **Hb 8.8 gr/dl,** Hct 25.5% VCM 96 fl, HCM33, plaquetas **5x10^9/L,** leucocitos 3200 (Neu 2.0, Lin 1.0). Retis 7.45%, absolutos 198000/mcL
-**Hemostasia:** INR 1, Fibrinógeno 392mg/dl, APTT 29.7"/31".
Coomb directo: NEGATIVO
-**Bioquímica:** **BT 1.4mg/dl, LDH 664U/L**, Cr 2.20 mg/dl, Na 139mmol/L, K 3.7mmol/L, **Haptoglobina <6.** Troponina 15.2ng/L

Frotis de sangre periférica:

- Anisopoiquilocitosis con presencia de **esquistocitos 8%.**
- Plaquetas de talla intermedia, bien granuladas.
- Fórmula Ne 68%, Lin 27%, Mon 4%, Eos:1%, Bas 1%.

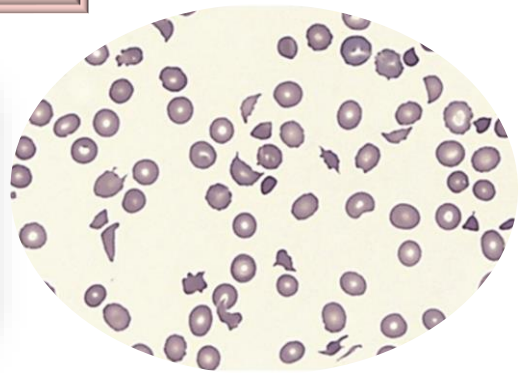
Caso clínico

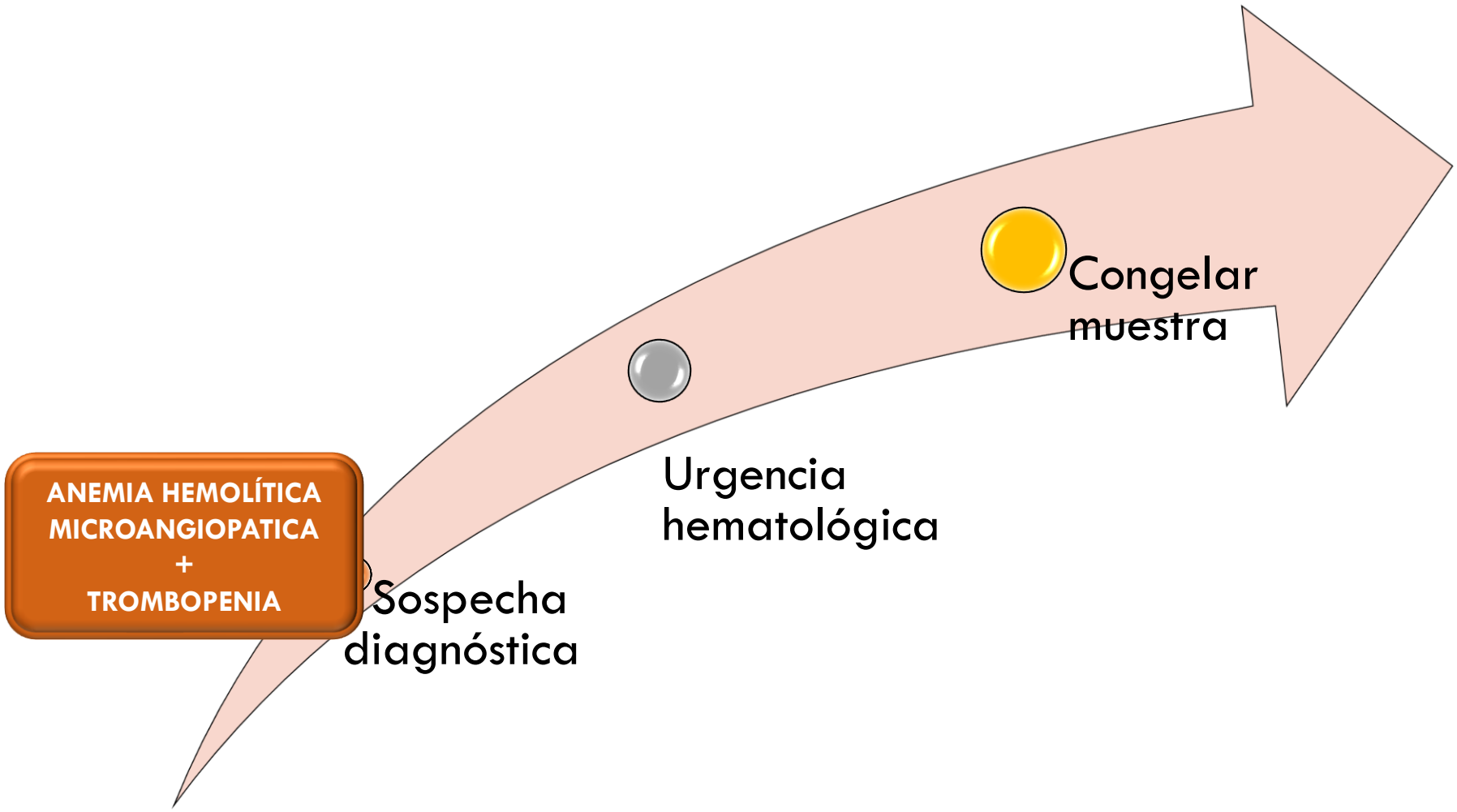
- Mujer de 50 años, sana y sin FRCV
- Enviada al Servicio de Urgencias por aparición de hematomas, y astenia

-**Hemograma:** **Hb 8.8 gr/dl,** Hct 25.5% VCM 96 fl, HCM33, plaquetas **5x10^9/L,** leucocitos 3200 (Neu 2.0, Lin 1.0). Retis 7.45%, absolutos 198000/mcL
-**Hemostasia:** INR 1, Fibrinógeno 392mg/dl, APTT 29.7"/31".
Coomb directo: NEGATIVO
-**Bioquímica:** **BT 1.4mg/dl, LDH 664U/L**, Cr 2.20 mg/dl, Na 139mmol/L, K 3.7mmol/L, **Haptoglobina <6.** Troponina 15.2ng/L

Frotis de sangre periférica:

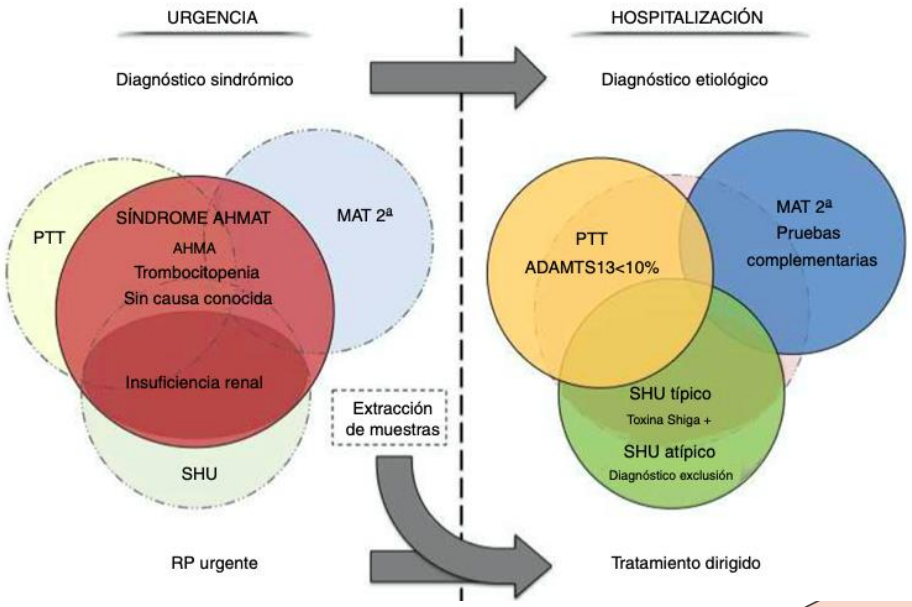
- Anisopoiquilocitosis con presencia de **esquistocitos 8%.**
- Plaquetas de talla intermedia, bien granuladas.
- Fórmula Ne 68%, Lin 27%, Mon 4%, Eos:1%, Bas 1%.





Zheng et al, JTH 2020

S. Romero et al. / Med Clin (Barc). 2017;xxx(xx):xxx–xxx



MAT, PTTa??

ANEMIA HEMOLÍTICA MICROANGIOPATICA + TROMBOPENIA

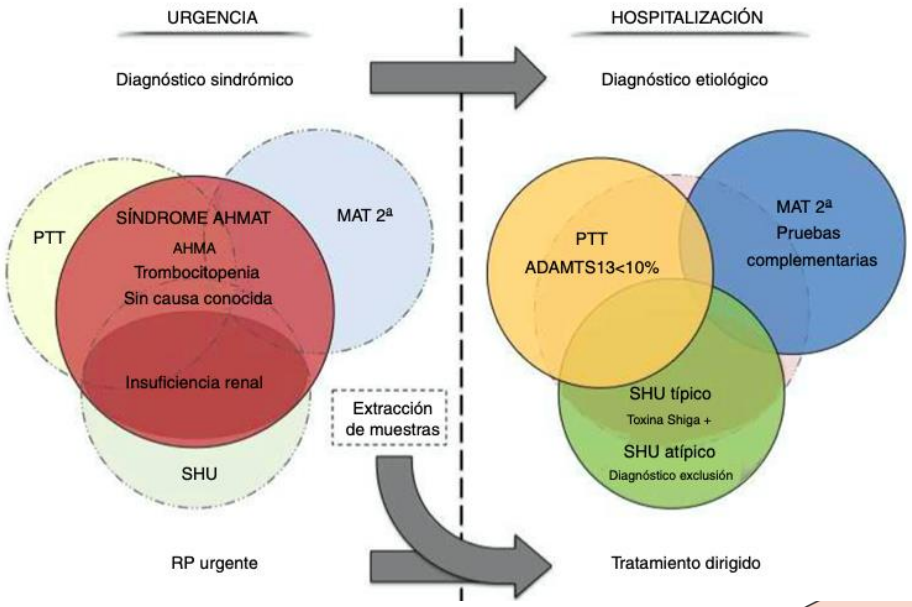
Congelar muestra

Urgencia hematológica

Sospecha diagnóstica

Zheng et al, JTH 2020

S. Romero et al. / Med Clin (Barc). 2017;xxx(xx):xxx–xxx



MAT, PTTa??

ANEMIA HEMOLÍTICA MICROANGIOPATICA + TROMBOPENIA

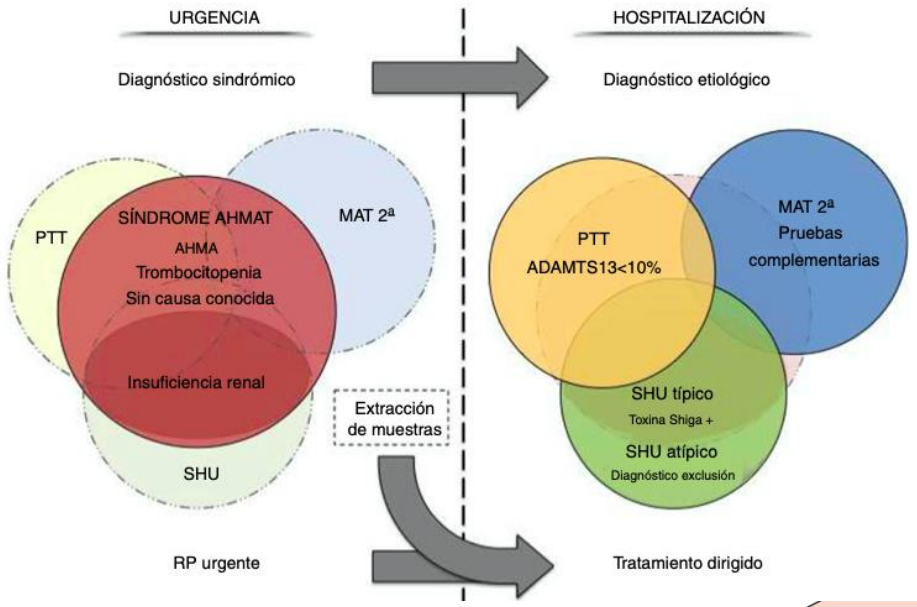
Congelar muestra

Urgencia hematológica

Sospecha diagnóstica

Zheng et al, JTH 2020

S. Romero et al. / Med Clin (Barc). 2017;xxx(xx):xxx–xxx



Programar determinación de actividad ADAMTS13 y otras determinaciones

Tratamiento Urgente

Congelar muestra

MAT, PTTa??

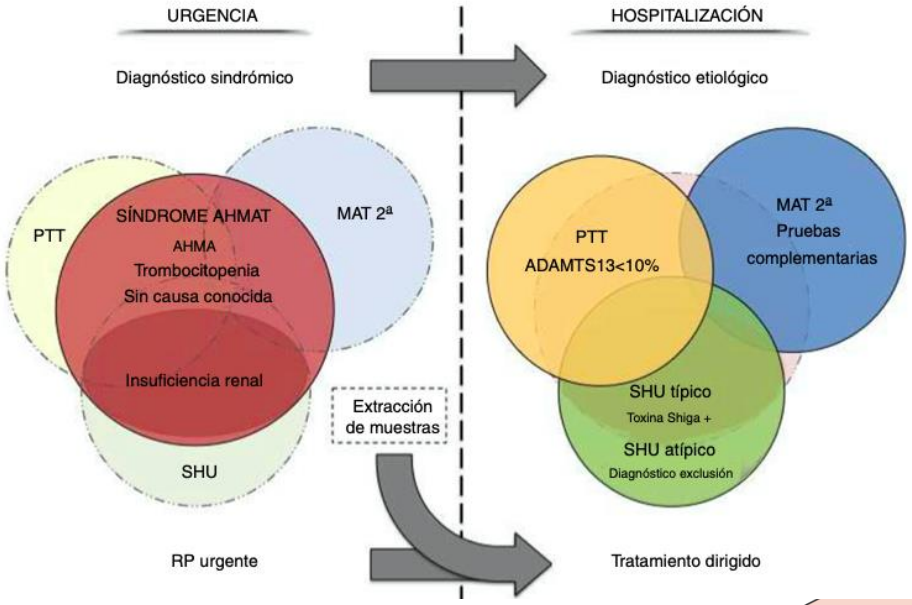
Urgencia hematológica

ANEMIA HEMOLÍTICA MICROANGIOPATICA + TROMBOPENIA

Sospecha diagnóstica

Zheng et al, JTH 2020

S. Romero et al. / Med Clin (Barc). 2017;xxx(xx):xxx–xxx



Programar determinación de actividad ADAMTS13 y otras determinaciones

Tratamiento Urgente

Congelar muestra

MAT, PTTa??

Urgencia hematológica

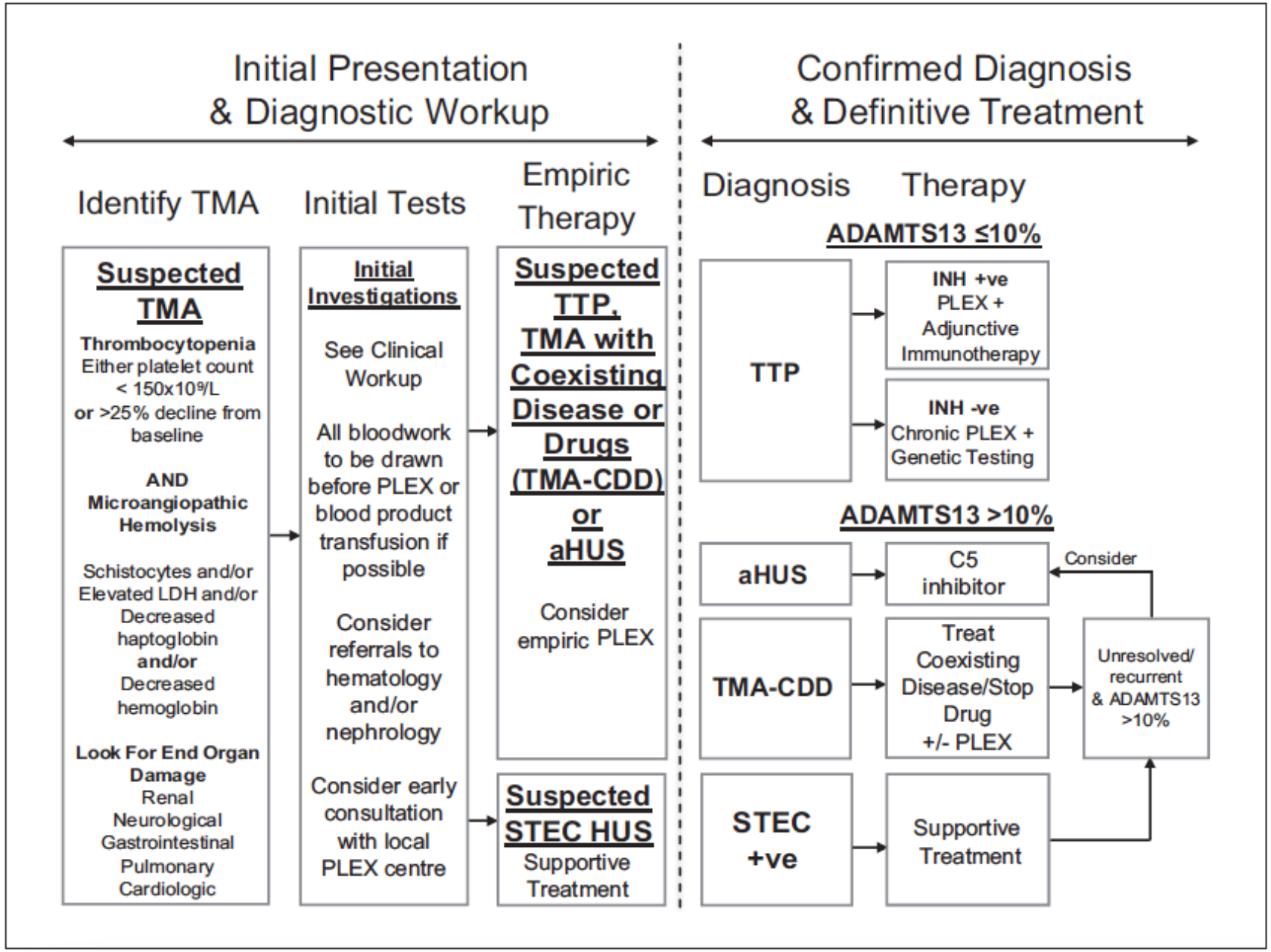
ANEMIA HEMOLÍTICA MICROANGIOPATICA + TROMBOPENIA

Sospecha diagnóstica

Vamos a tener que tratar antes de tener el diagnóstico de confirmación SIEMPRE……

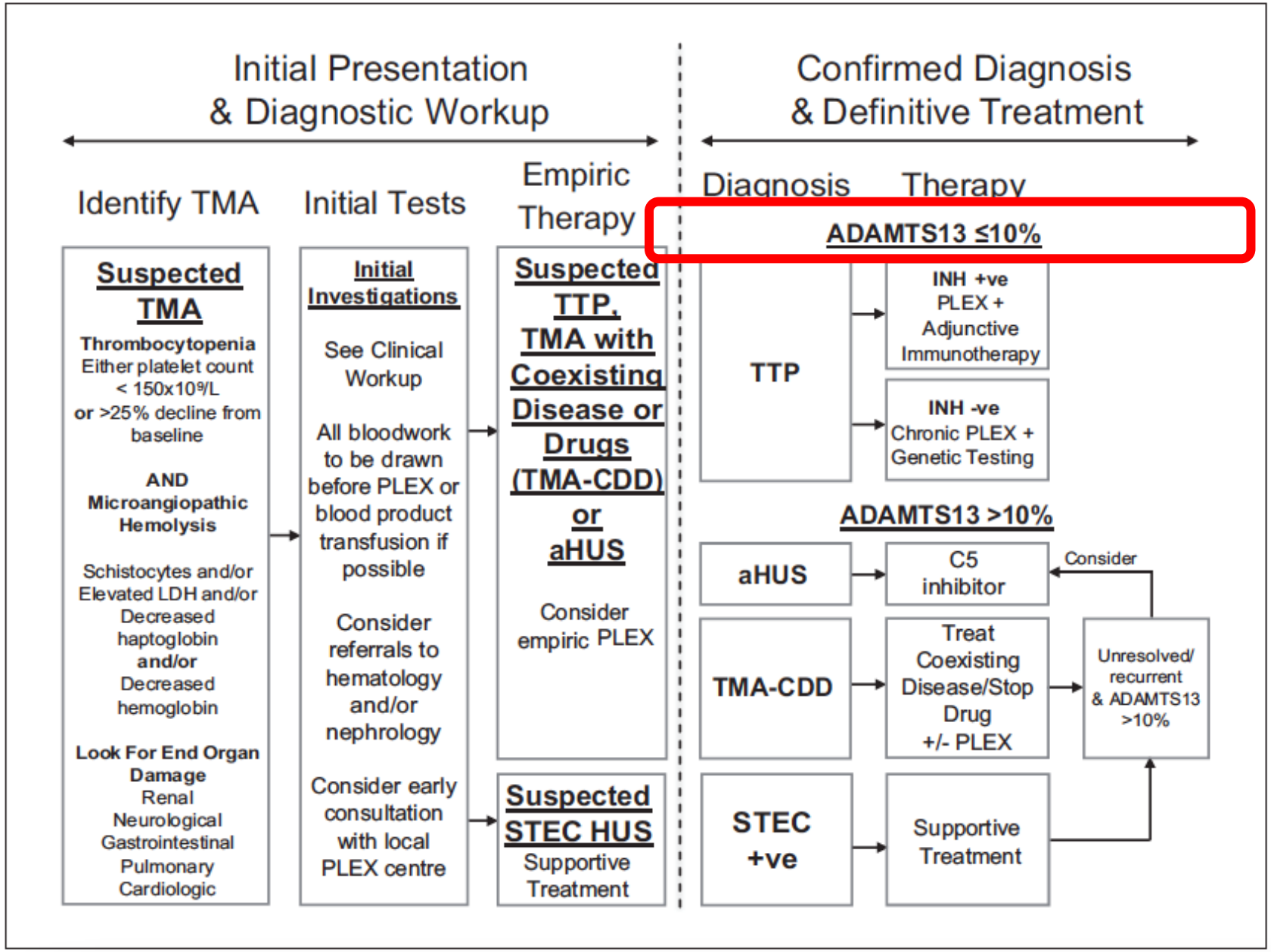
Zheng et al, JTH 2020

Diagnóstico diferencial



Mcfarlan et al, Canadian Journal of Kidney Health and Disease 2021, Volume 8: 1–12

Diagnóstico diferencial



Mcfarlan et al, Canadian Journal of Kidney Health and Disease 2021, Volume 8: 1–12

REVIEW

ISLH International Journal of Laboratory Hematology WILEY

International Council for Standardization in Haematology (ICSH) recommendations for laboratory measurement of ADAMTS13

Ian Mackie[1] | Ilaria Mancini[2] | Joshua Muia[3] | Johanna Kremer Hovinga[4] | Sukesh Nair[5] | Sam Machin[1] | Ross Baker[6]

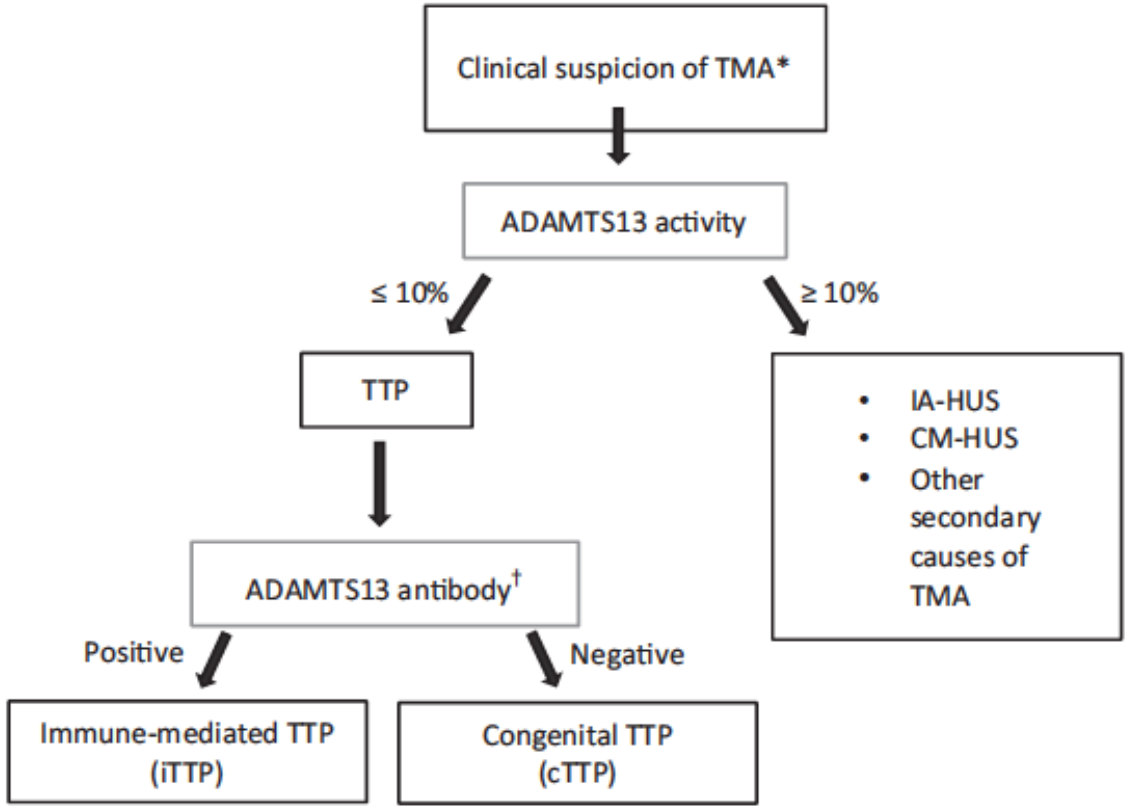


FIGURE 1 ADAMTS13 testing for the diagnosis of TTP and other TMA. *Based on clinical assessment including Clinical Gestalt, PLASMIC score, laboratory investigation (haemoglobin, platelet count, fragments, and creatinine level). †High titre ADAMTS13 antibody (ELISA) or functional inhibitor of ADAMTS13 activity measured by Bethesda assay. CM-HUS, complement mediated haemolytic uremic syndrome; IA-HUS, infection associated haemolytic uremic syndrome; TMA, thrombotic microangiopathy; TTP, thrombotic thrombocytopenic purpura

Mackie I, Mancini I, Muia J, Kremer Hovinga J, Nair S, Machin S, Baker R. International Council for Standardization in Haematology (ICSH) recommendations for laboratory measurement of ADAMTS13. Int J Lab Hematol. 2020 Dec;42(6):685-696.

REVIEW

ISLH International Journal of Laboratory Hematology WILEY

International Council for Standardization in Haematology (ICSH) recommendations for laboratory measurement of ADAMTS13

Ian Mackie[1] | Ilaria Mancini[2] | Joshua Muia[3] | Johanna Kremer Hovinga[4] | Sukesh Nair[5] | Sam Machin[1] | Ross Baker[6]

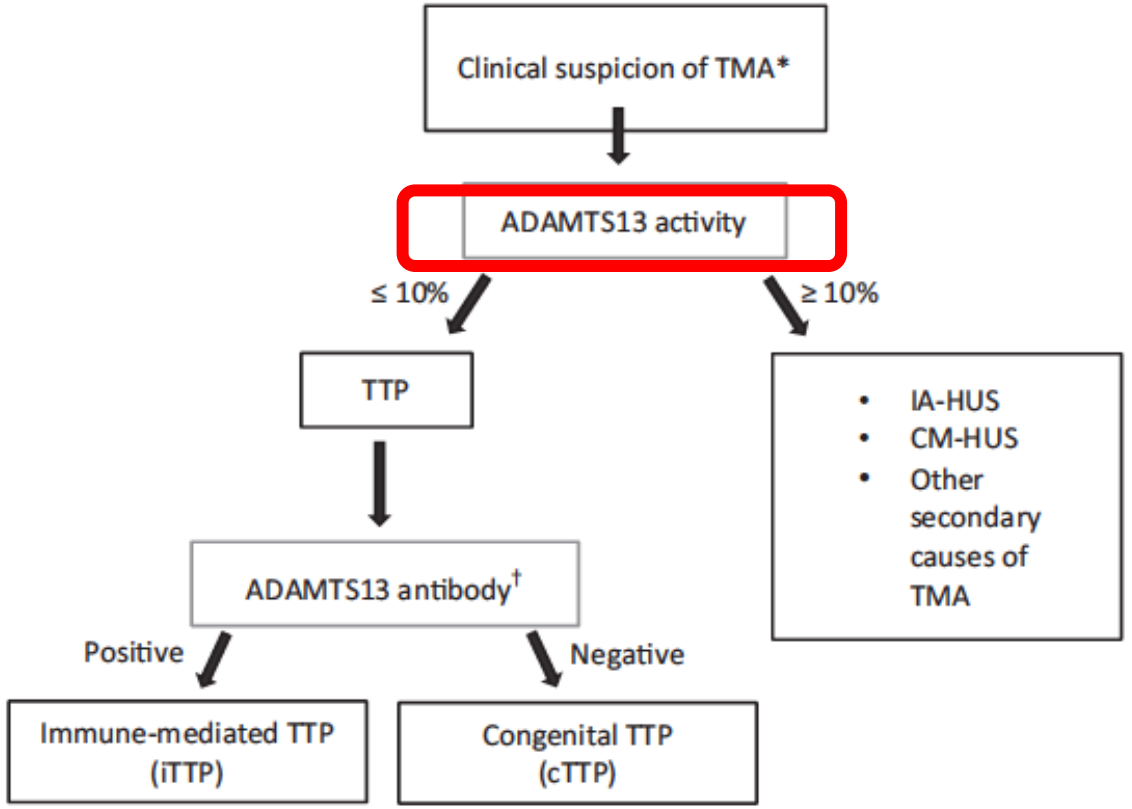


FIGURE 1 ADAMTS13 testing for the diagnosis of TTP and other TMA. *Based on clinical assessment including Clinical Gestalt, PLASMIC score, laboratory investigation (haemoglobin, platelet count, fragments, and creatinine level). †High titre ADAMTS13 antibody (ELISA) or functional inhibitor of ADAMTS13 activity measured by Bethesda assay. CM-HUS, complement mediated haemolytic uremic syndrome; IA-HUS, infection associated haemolytic uremic syndrome; TMA, thrombotic microangiopathy; TTP, thrombotic thrombocytopenic purpura

Mackie I, Mancini I, Muia J, Kremer Hovinga J, Nair S, Machin S, Baker R. International Council for Standardization in Haematology (ICSH) recommendations for laboratory measurement of ADAMTS13. Int J Lab Hematol. 2020 Dec;42(6):685-696.

PTTi- generalidades

- La púrpura trombótica trombocitopénica inmune (PTTi) es una microangiopatía trombótica (MAT) rara y potencialmente mortal caracterizada por una deficiencia severa de la actividad de ADAMTS13 (<10%).
- PTTa es una microangiopatía trombótica con un potencial pronóstico fatal.
- Urgencia hematológica.
- Mortalidad de >90% sin tratamiento.
- 10-20% mortalidad con tratamiento SOC
- <5% desde introducción del caplacizumab
- 5-10% refractariedad, 8-12% exacerbaciones
- Recurrente, 40% de los casos

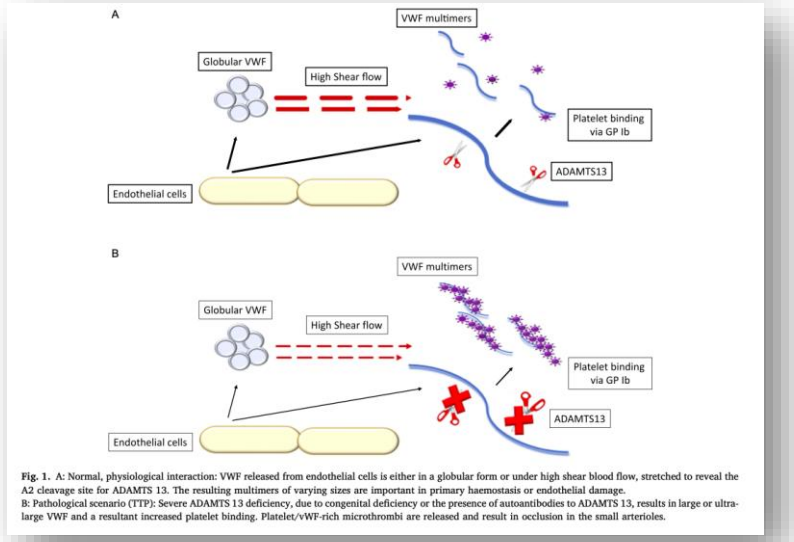


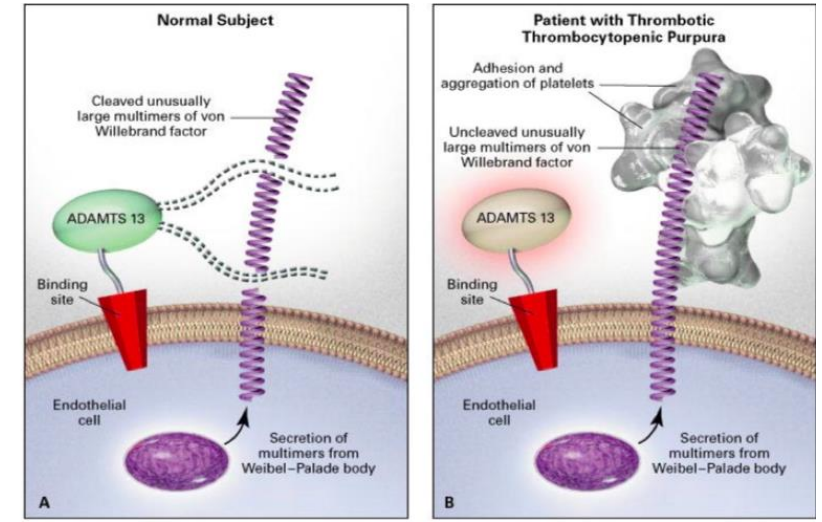
Fig. 1. A: Normal, physiological interaction: VWF released from endothelial cells is either in a globular form or under high shear blood flow, stretched to reveal the A2 cleavage site for ADAMTS 13. The resulting multimers of varying sizes are important in primary haemostasis or endothelial damage. B: Pathological scenario (TTP): Severe ADAMTS 13 deficiency, due to congenital deficiency or the presence of autoantibodies to ADAMTS 13, results in large or ultra-large VWF and a resultant increased platelet binding. Platelet/vWF-rich microthrombi are released and result in occlusion in the small arterioles.

- La rápida determinación de su actividad es un elemento clave para el diagnóstico precoz y manejo óptimo de los pacientes con PTTi y otras MAT.

Subhan M, Scully M. Advances in the management of TTP. Blood Rev. 2022 Feb 17:100945. doi: 10.1016/j.blre.2022.100945. Epub ahead of print. PMID: 35216839. Joly BS, Coppo P, Veyradier A. Thrombotic thrombocytopenic purpura. Blood. 2017;129:2836-2846.

PTTi_ Fisiopatología

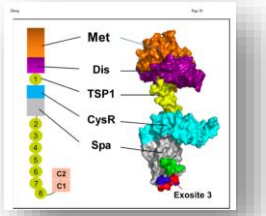
En condiciones normales ADAMTS-13 fragmenta los multímetros de alto peso molecular del FvW en monómeros con menor capacidad para inducir adhesión y agregación plaquetaria.



Moake JL. N Engl J Med 2002;347:589-600.

Anticuerpos anti ADAMTS 13

Actividad ADAMTS13 < 10%



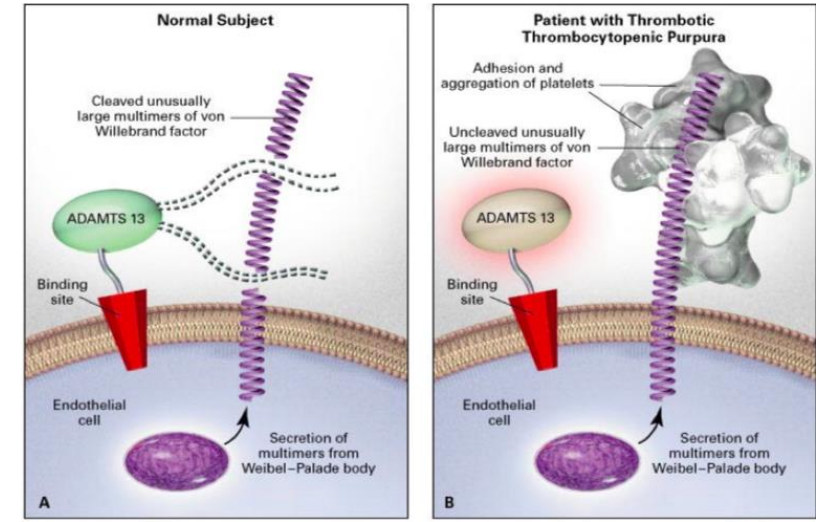
No escisión FvW

Aumento agregación plaquetaria con formación de microtrombos → TROMBOPENIA POR CONSUMO

Fragmentación de los hematíes al pasar por vasos estrechos con trombos → ANEMIA HEMOLÍTICA

PTTi_ Fisiopatología

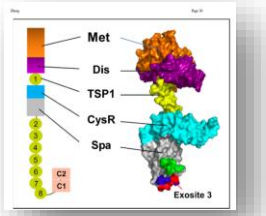
En condiciones normales ADAMTS-13 fragmenta los multímetros de alto peso molecular del FvW en monómeros con menor capacidad para inducir adhesión y agregación plaquetaria.



Moake JL. N Engl J Med 2002;347:589-600.

Anticuerpos anti ADAMTS 13

Actividad ADAMTS13 < 10%



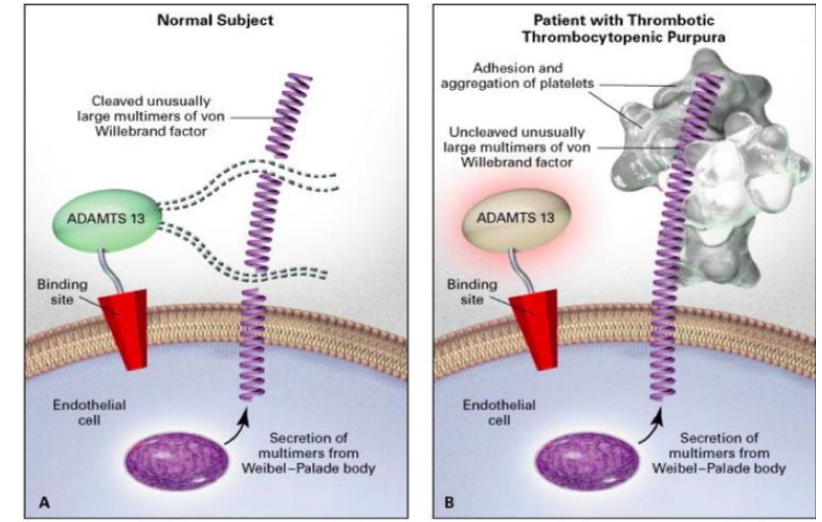
No escisión FvW

Aumento agregación plaquetaria con formación de microtrombos → TROMBOPENIA POR CONSUMO

Fragmentación de los hematíes al pasar por vasos estrechos con trombos → ANEMIA HEMOLÍTICA

PTTi_ Fisiopatología

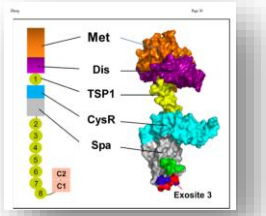
En condiciones normales ADAMTS-13 fragmenta los multímetros de alto peso molecular del FvW en monómeros con menor capacidad para inducir adhesión y agregación plaquetaria.



Moake JL. N Engl J Med 2002;347:589-600.

Anticuerpos anti ADAMTS 13

Actividad ADAMTS13 < 10%



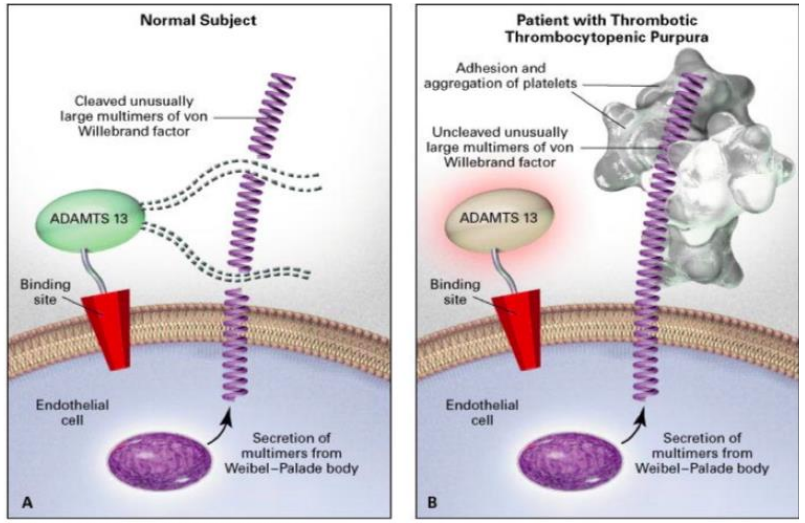
No escisión FvW

Aumento agregación plaquetaria con formación de microtrombos → TROMBOPENIA POR CONSUMO

Fragmentación de los hematíes al pasar por vasos estrechos con trombos → ANEMIA HEMOLÍTICA

PTTi_ Fisiopatología

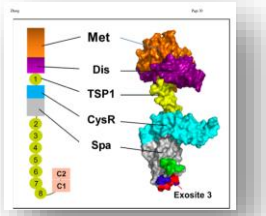
En condiciones normales ADAMTS-13 fragmenta los multímetros de alto peso molecular del FvW en monómeros con menor capacidad para inducir adhesión y agregación plaquetaria.



Moake JL. N Engl J Med 2002;347:589-600.

Anticuerpos anti ADAMTS 13

Actividad ADAMTS13 < 10%



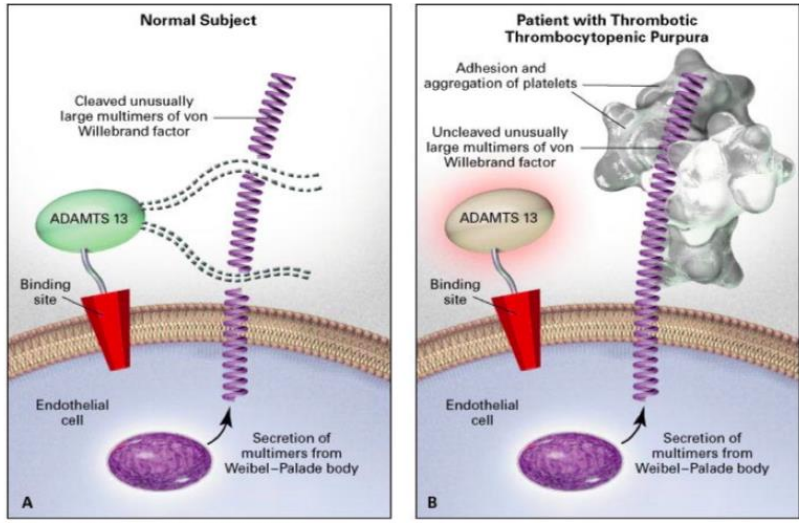
No escisión FvW

Aumento agregación plaquetaria con formación de microtrombos → TROMBOPENIA POR CONSUMO

Fragmentación de los hematíes al pasar por vasos estrechos con trombos → ANEMIA HEMOLÍTICA

PTTi_ Fisiopatología

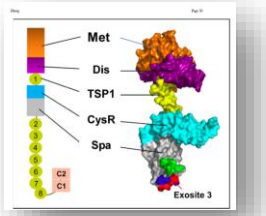
En condiciones normales ADAMTS-13 fragmenta los multímetros de alto peso molecular del FvW en monómeros con menor capacidad para inducir adhesión y agregación plaquetaria.



Moake JL. N Engl J Med 2002;347:589-600.

Anticuerpos anti ADAMTS 13

Actividad ADAMTS13 < 10%



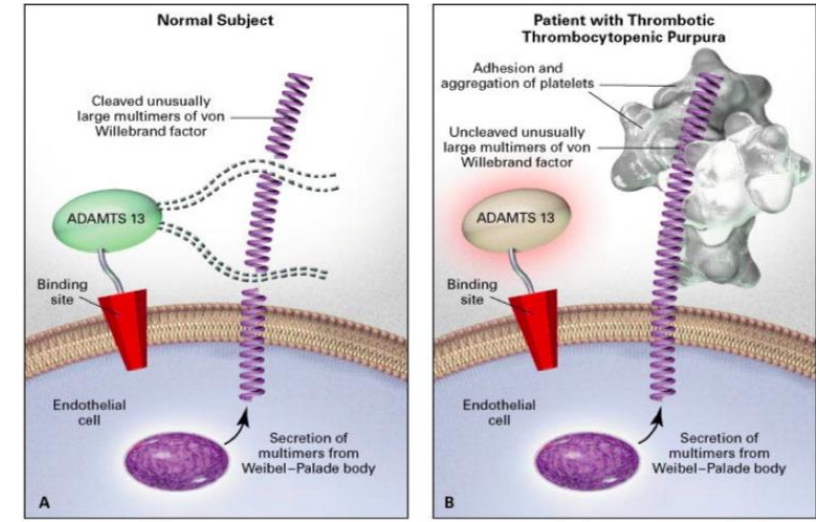
No escisión FvW

Aumento agregación plaquetaria con formación de microtrombos → TROMBOPENIA POR CONSUMO

Fragmentación de los hematíes al pasar por vasos estrechos con trombos → ANEMIA HEMOLÍTICA

PTTi_ Fisiopatología

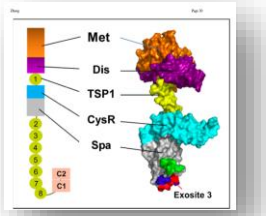
En condiciones normales ADAMTS-13 fragmenta los multímetros de alto peso molecular del FvW en monómeros con menor capacidad para inducir adhesión y agregación plaquetaria.



Moake JL. N Engl J Med 2002;347:589-600.

Anticuerpos anti ADAMTS 13

Actividad ADAMTS13 < 10%



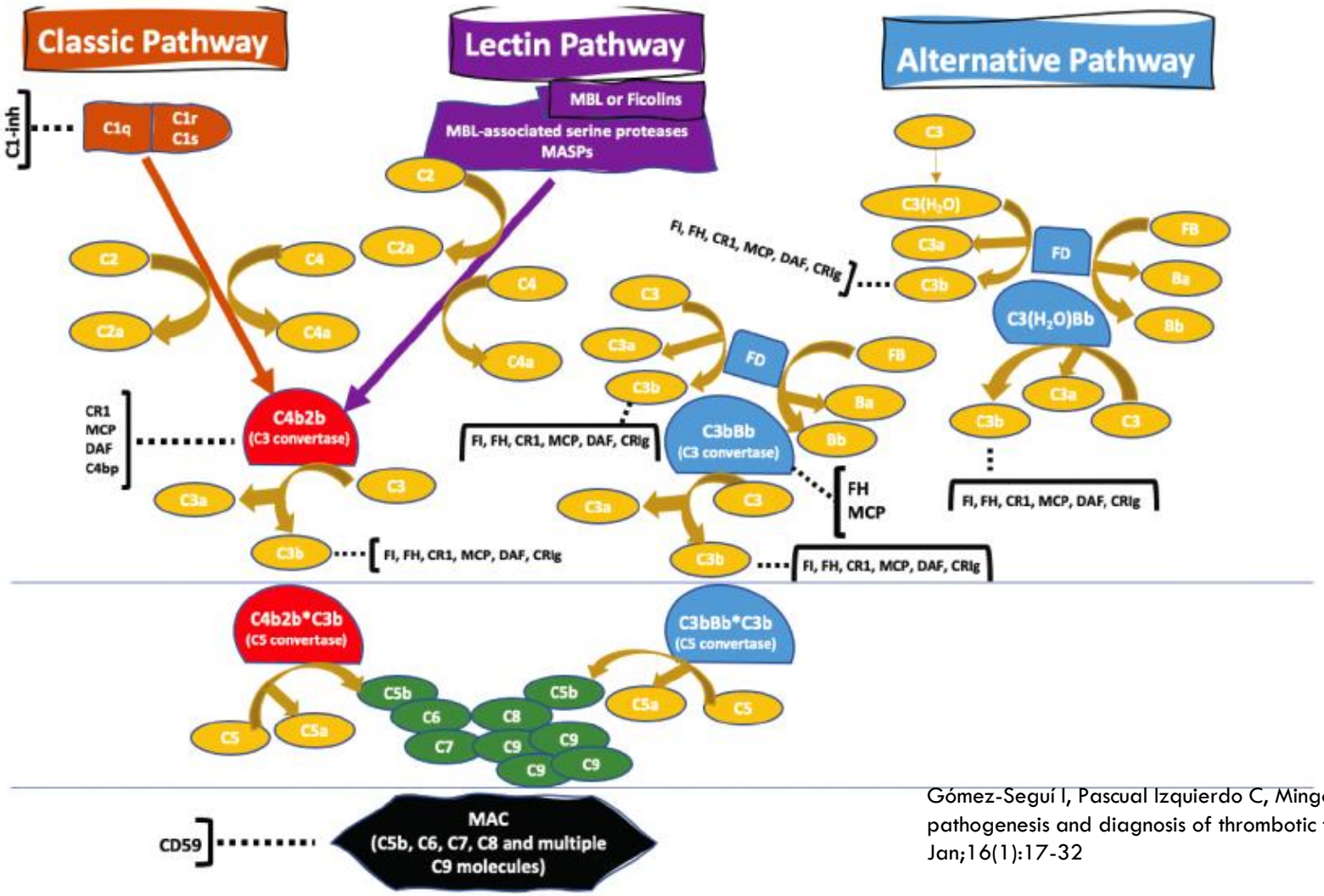
No escisión FvW

Aumento agregación plaquetaria con formación de microtrombos → TROMBOPENIA POR CONSUMO

Fragmentación de los hematíes al pasar por vasos estrechos con trombos → ANEMIA HEMOLÍTICA

Sin tratamiento es mortal

Complement system pathways activation and control steps.

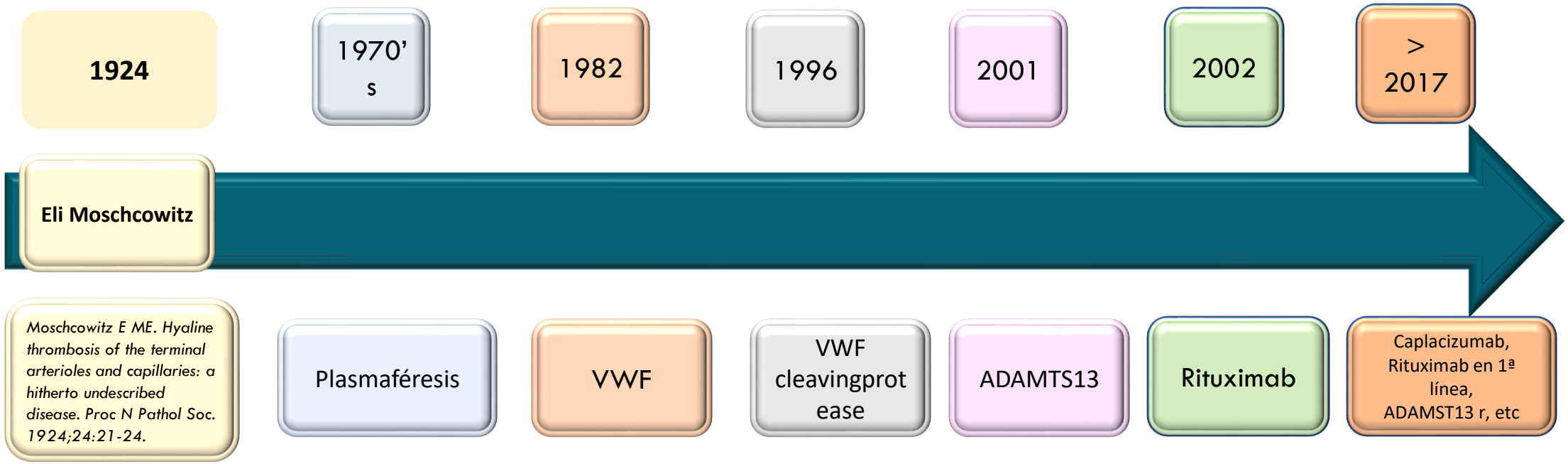


C5 podría jugar un papel primordial en la fisiopatología de la PTT

Gómez-Seguí I, Pascual Izquierdo C, Mingot Castellano ME, de la Rubia Comos J. An update on the pathogenesis and diagnosis of thrombotic thrombocytopenic purpura. Expert Rev Hematol. 2023 Jan;16(1):17-32

Historia de la PTTi

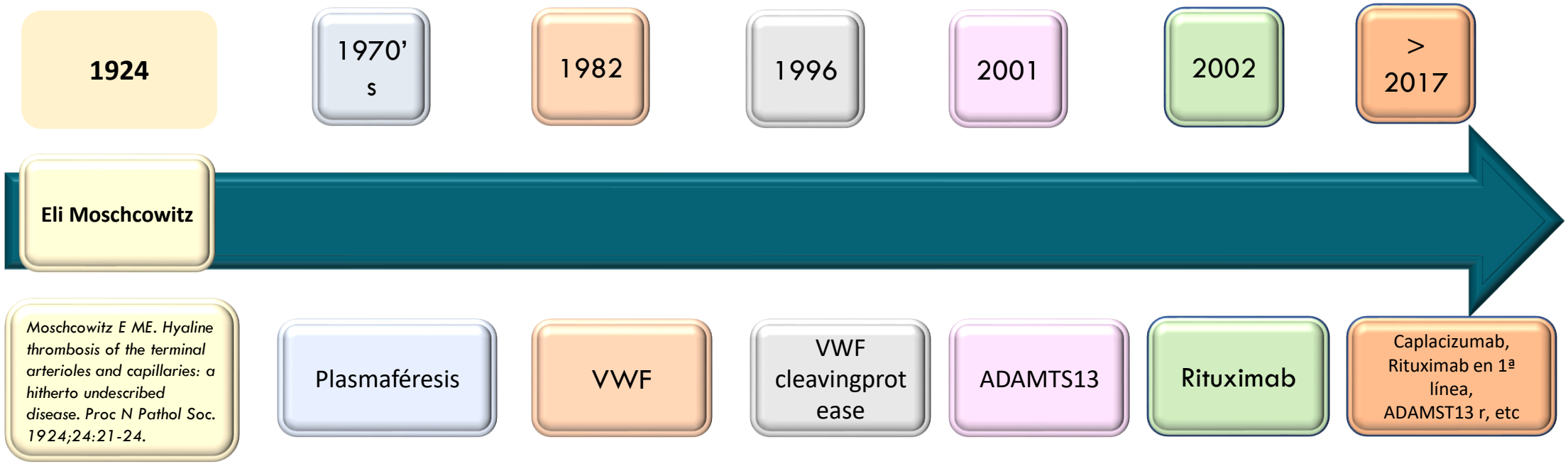
La púrpura trombótica trombocitopénica (PTT) es una microangiopatía trombótica (MAT), definida por trombopenia, anemia hemolítica microangiopática y trombosis en los pequeños vasos, que se produce por una deficiencia de la actividad ADAMTS 13 (metaloproteasa responsable de la ruptura del factor de von Willebrand).



1.Proc N Y Pathol Soc. 1924;24:21. 2. Medicine 1966;45:139. 3. NEJM 1982;307:1432. 4. NEJM 1991;325:393, NEJM 1991;325:393. 5. Blood 1996;87:4223, Blood 1996;87:4235. 6. Blood Cells, Molecules, and Diseases 2002;28(3):385. 7. NEJM 2016;374:511, NEJM 2019;380:335. https://clinicaltrials.gov/ct2/show/NCT03922308?term=rADAMTS13&draw=2&rank=1 último acceso marzo 2022
Rituximab no está aprobado por ningún organismo internacional para el tratamiento de la PTTa
Recombinant ADAMTS13 is an investigational product in phase 2 of clinical development for TTP. ClinicalTrials.gov. Available from: https://clinicaltrials.gov/ct2/show/NCT03922308. Accessed March 2022.

Historia de la PTTi

La púrpura trombótica trombocitopénica (PTT) es una microangiopatía trombótica (MAT), definida por trombopenia, anemia hemolítica microangiopática y trombosis en los pequeños vasos, que se produce por una deficiencia de la actividad ADAMTS 13 (metaloproteasa responsable de la ruptura del factor de von Willebrand).



1.Proc N Y Pathol Soc. 1924;24:21. 2. Medicine 1966;45:139. 3. NEJM 1982;307:1432. 4. NEJM 1991;325:393, NEJM 1991;325:393. 5. Blood 1996;87:4223, Blood 1996;87:4235. 6. Blood Cells, Molecules, and Diseases 2002;28(3):385. 7. NEJM 2016;374:511, NEJM 2019;380:335. https://clinicaltrials.gov/ct2/show/NCT03922308?term=rADAMTS13&draw=2&rank=1 último acceso marzo 2022
Rituximab no está aprobado por ningún organismo internacional para el tratamiento de la PTTa
Recombinant ADAMTS13 is an investigational product in phase 2 of clinical development for TTP. ClinicalTrials.gov. Available from: https://clinicaltrials.gov/ct2/show/NCT03922308. Accessed March 2022.

Historia de la PTTi

La p
trom
una
Will

ngiopatía trombótica (MAT), definida por
sis en los pequeños vasos, que se produce por
a responsable de la ruptura del factor de von

19

Eli Mosc

Moschcowitz
thrombosis of
arterioles and
hitherto undes
disease. Proc
1924;24:21-

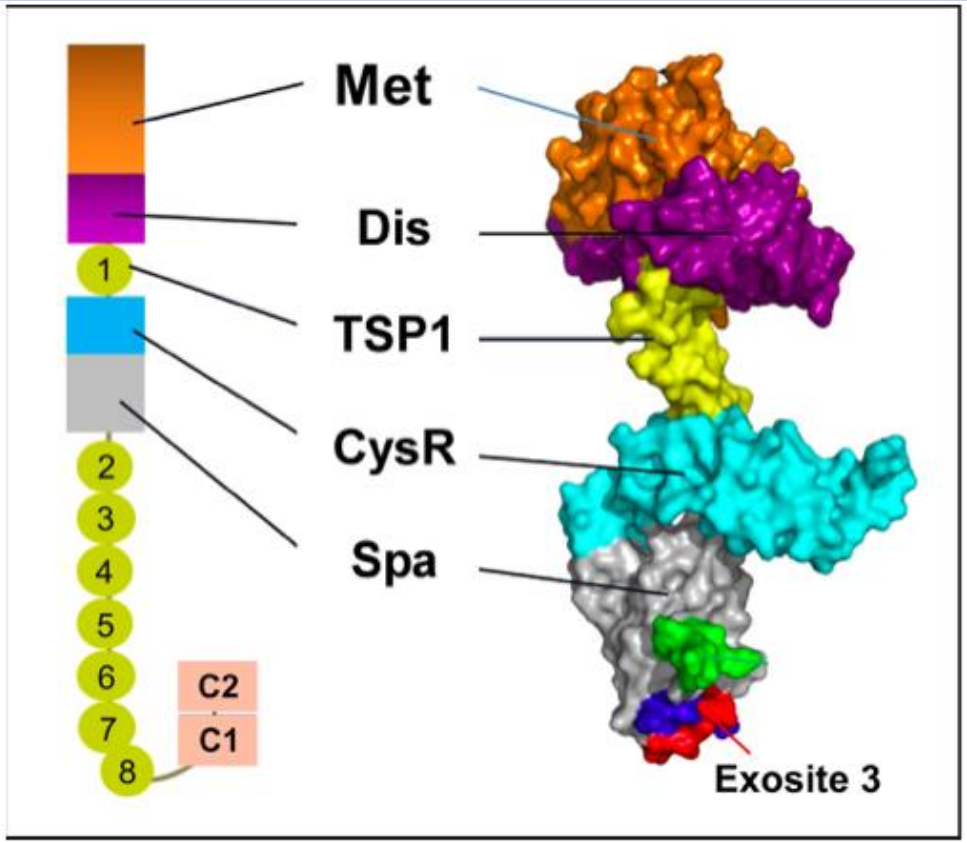


Fig. 2. Domain organisation and partial crystal structure of ADAMTS13
On the left, the domain organization of human mature ADAMTS13 is shown, which consists of a metalloprotease domain (M), a disintegrin-like domain (D), the first TSP1 repeat, a Cys-rich domain (C), and a spacer domain (S). In addition, the C-terminus contains 7 more TSP1 repeats (2–8) and two CUB domains (C1 and C2). On the right, the surface and carton presentation of the crystal structure of ADAMTS13 disintegrin-like domain (Dis), first TSP1 repeat (TSP1), Cys-rich (Cys) and spacer domain (Spa) in addition to a modelled metalloprotease domain (based on the metalloprotease domains of ADAMTS4 and ADAMTS5).

2001

2002

> 2017

VF
gpro
se

ADAMTS13

Rituximab

Caplacizumab, Rituximab en 1ª línea, ADAMST13 r, etc

1991;325:393. 5. Blood 1996;87:4224. Blood 1996;87:4235. 6. Blood Cells, Molecules, and Diseases 2002;28(3):385. 7. NEJM
raw=2&rank=1 último acceso marzo 2022

Available from: https://clinicaltrials.gov/ct2/show/NCT03922308. Accessed March 2022.

ADAMTS13

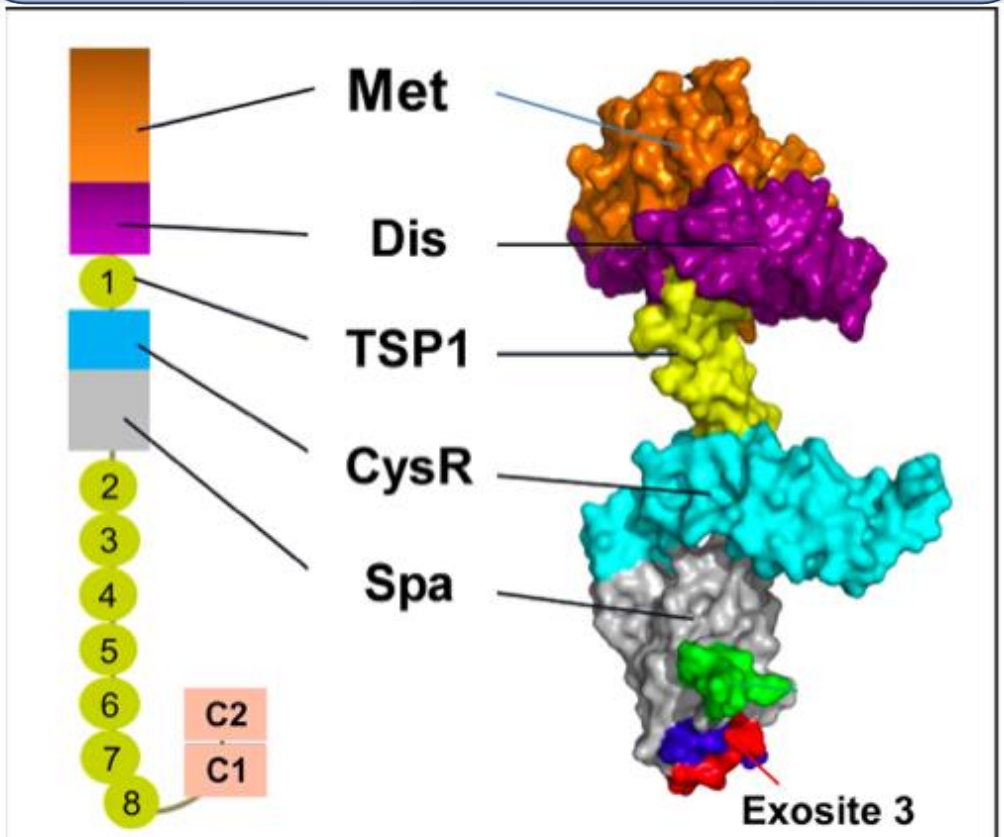
- Durante mucho tiempo se ha pensado que ADAMTS13 es una enzima constitutivamente activa.
- Los últimos descubrimientos muestran que ADAMTS13 necesita una activación alostérica a través de la interacción con su sustrato VWF para volverse activo.

Pasa de configuración cerrada a abierta

Muia J, Zhu J, Gupta G, et al. Allosteric activation of ADAMTS13 by von Willebrand factor. Proc Natl Acad Sci U S A 2014; 111:18584–18589.
South K, Luken BM, Crawley JTB, et al. Conformational activation of ADAMTS13. Proc Natl Acad Sci U S A 2014; 111:18578–18583.
Petri A, Kim HJ, Xu Y, et al. Crystal structure and substrate-induced activation of ADAMTS13. Nat Commun 2019; 10:1–16.

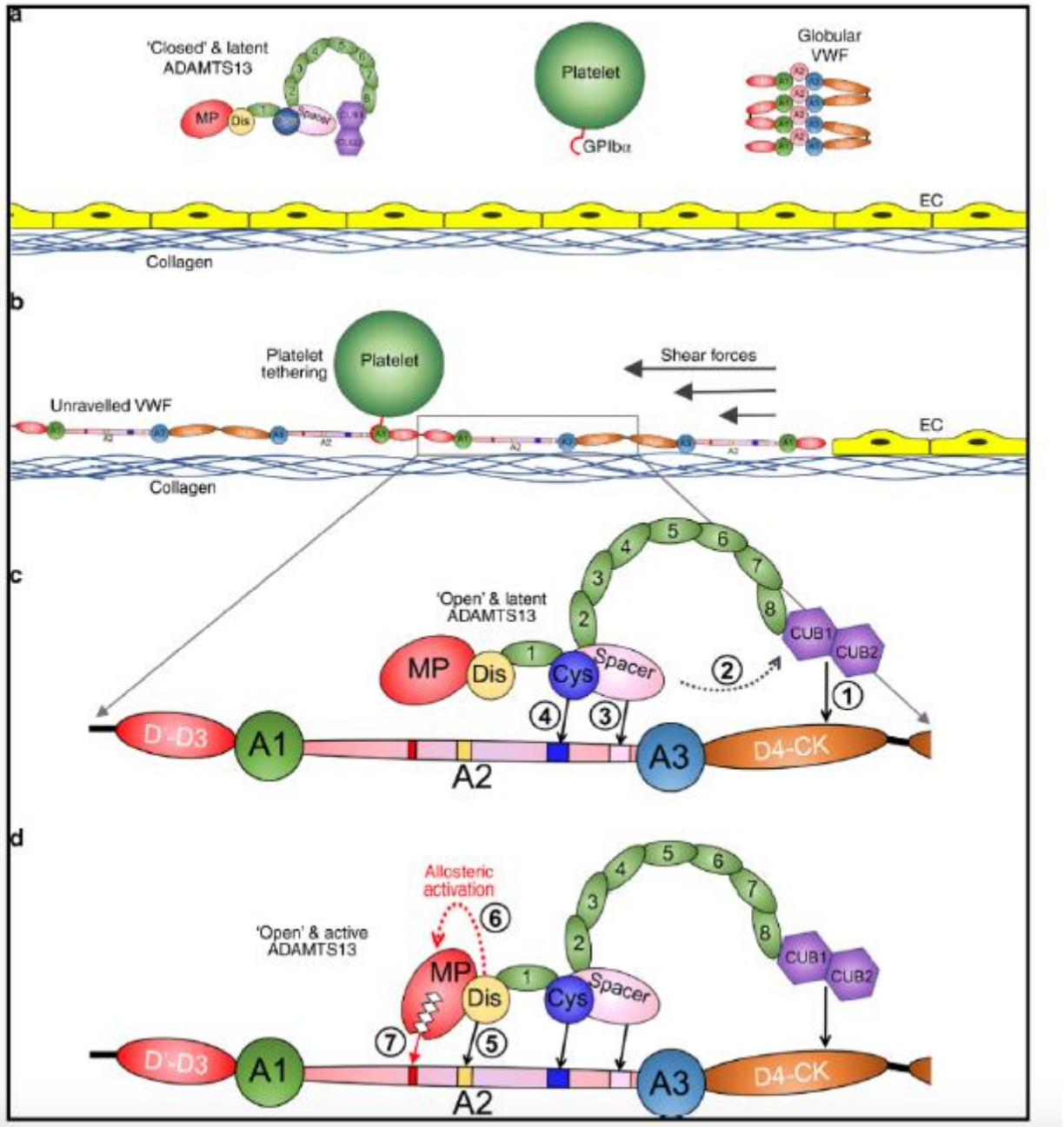
Metaloproteasa responsable de la ruptura del factor de von Willebrand, clonada en 2001.

Act ADAMTS13<10%=PTTa



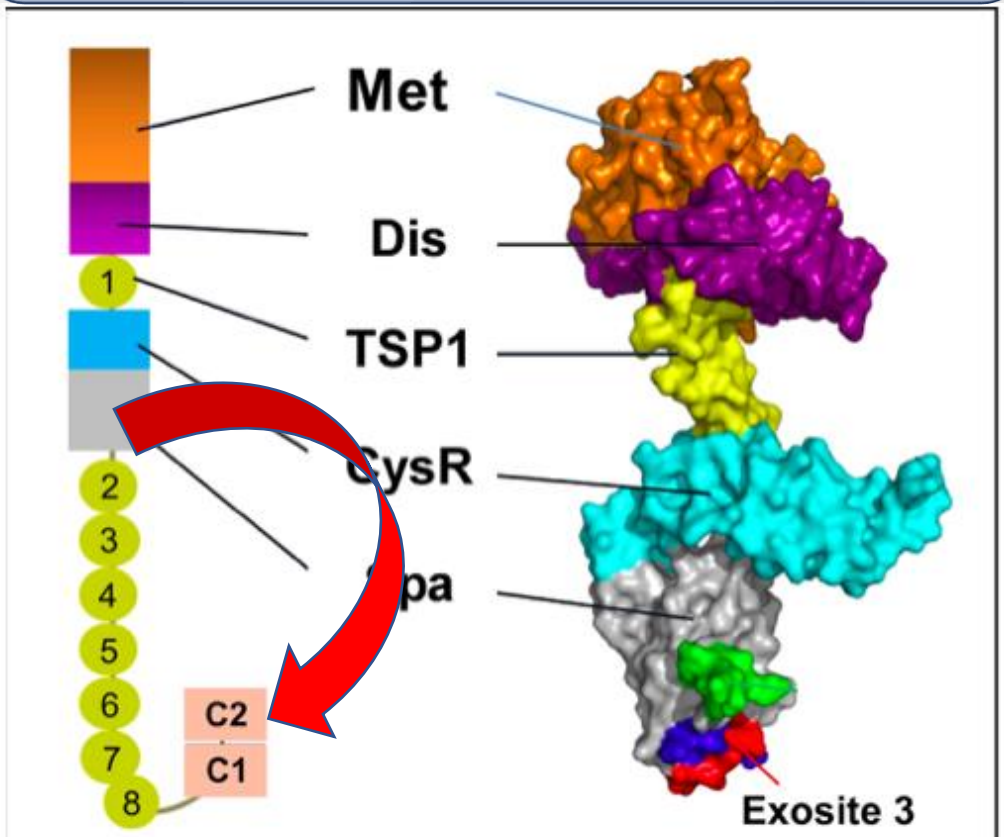
ADAMTS-13 consists of 14 domains:

- ✓ a metalloprotease (M),
- ✓ a disintegrin-like (D),
- ✓ a thrombospondin type-1 repeat (T),
- ✓ a cysteine-rich (C),
- ✓ a spacer (S),
- ✓ seven additional T,
- ✓ and two CUB domains



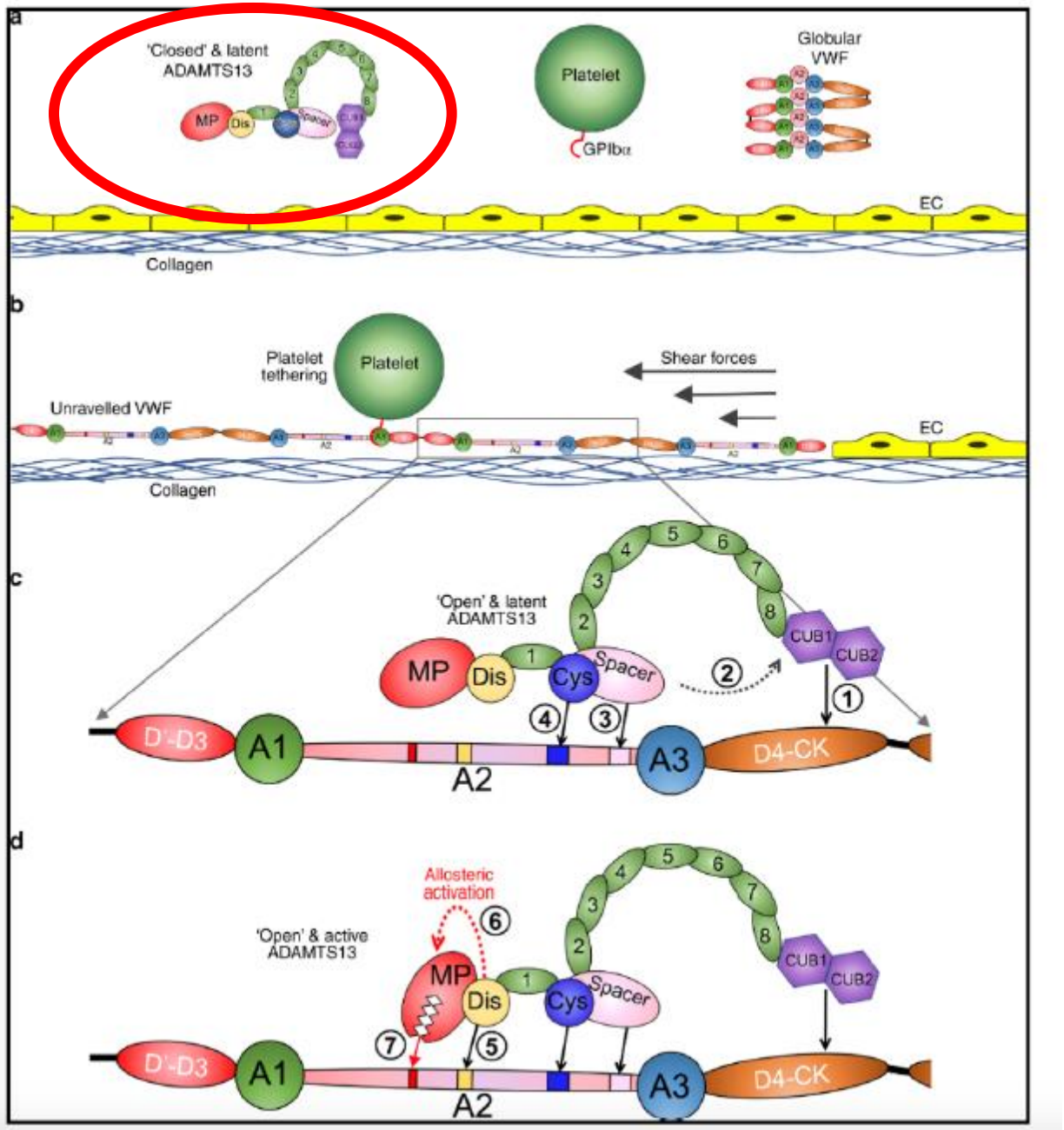
Metaloproteasa responsable de la ruptura del factor de von Willebrand, clonada en 2001.

Act ADAMTS13<10%=PTTa



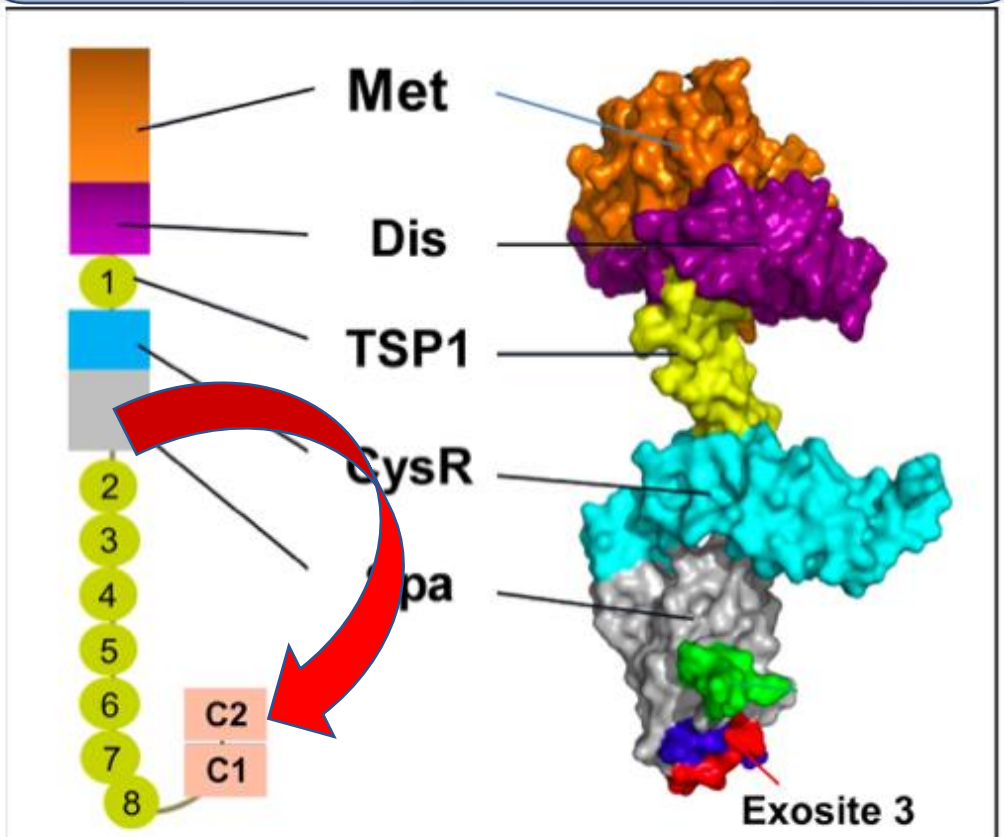
ADAMTS-13 consists of 14 domains:

- ✓ a metalloprotease (M),
- ✓ a disintegrin-like (D),
- ✓ a thrombospondin type-1 repeat (T),
- ✓ a cysteine-rich (C),
- ✓ a spacer (S),
- ✓ seven additional T,
- ✓ and two CUB domains



Metaloproteasa responsable de la ruptura del factor de von Willebrand, clonada en 2001.

Act ADAMTS13<10%=PTTa



ADAMTS-13 consists of 14 domains:

- ✓ a metalloprotease (M),
- ✓ a disintegrin-like (D),
- ✓ a thrombospondin type-1 repeat (T),
- ✓ a cysteine-rich (C),
- ✓ a spacer (S),
- ✓ seven additional T,
- ✓ and two CUB domains

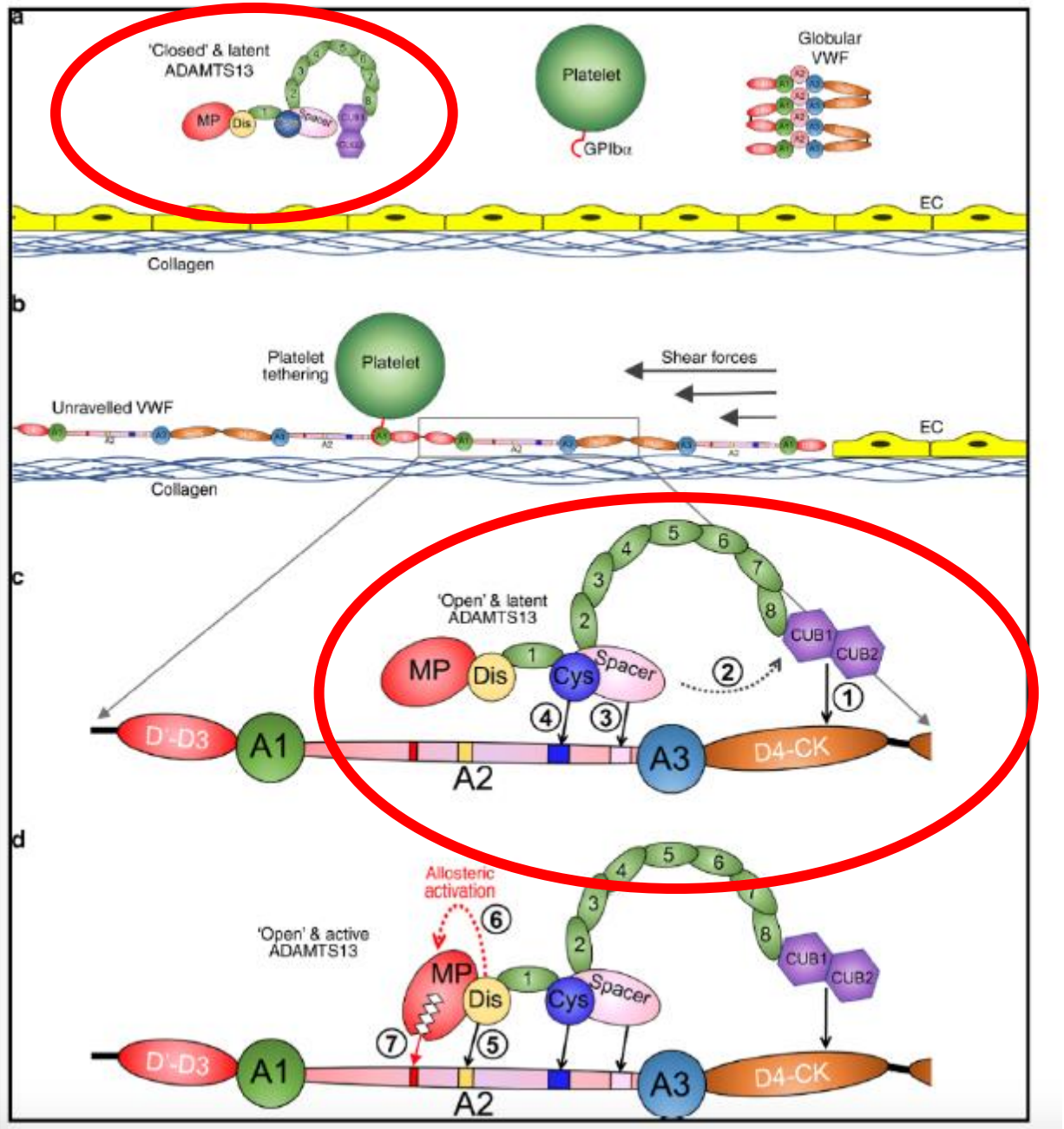


Table 1. Physiological and Pathological conditions associated with ADAMTS13 deficiency

Physiological conditions (Mild to moderate deficiency)	Pathological conditions (Moderate to severe deficiency) *
Pregnancy	Preeclampsia/HELLP syndrome
Newborn	Severe infections (e.g.: bacterial sepsis, SARS-CoV-2 severe infection, cerebral malaria)
Elderly	Disseminated Intravascular Coagulation
Black ethnicity	Liver disease
Non-0 blood	Metastatic cancer
	Hereditary or immune TTP

*Although severe deficiency, defined as <10% of normal activity, is characteristic of TTP, few cases of these pathological conditions might have very low levels of ADAMTS13 activity and, infrequently, even close to or <10%. The clinical picture should help clinicians to correctly diagnose and treat these patients.

Gómez-Seguí I, Pascual Izquierdo C, Mingot Castellano ME, de la Rubia Comos J. An update on the pathogenesis and diagnosis of thrombotic thrombocytopenic purpura. Expert Rev Hematol. 2023 Jan;16(1):17-32

Determinación de la actividad de ADAMTS13

Table 4. Recommendations on pre-analytical phase for ADAMTS13 assays

ISCT consensus recommendations on pre-analytical phase for ADAMTS13 assays
Citrated plasma should normally be used.
Heparin plasma or serum samples may be used depending on the assay type and if it is validated for these samples' types.
Never use EDTA plasma for ADAMTS13 activity determination.
Samples should be centrifuged and plasma separated from cells as rapidly as possible after blood collection to avoid *in vitro* changes.
Plasma samples should be stores and shipped below -40ºC, unless assays are performed immediately.

Gómez-Seguí I, Pascual Izquierdo C, Mingot Castellano ME, de la Rubia Comos J. An update on the pathogenesis and diagnosis of thrombotic thrombocytopenic purpura. Expert Rev Hematol. 2023 Jan;16(1):17-32

Consensus recommendations on assays for ADAMTS13, autoantibodies and inhibitors, Mackie et al

Functional FRET-based assays or chromogenic activity ELISA methods are recommended as front line assays as they are sensitive, show good precision and are simpler to use, being completed in a few hours.
Rapid point of care assays may have utility as screening methods or “out of hours” emergency tests, but there is currently limited performance data.
Every calibrator should be traceable to the International Standard Plasma (12/252) for assaying ADAMTS13 activity in citrated plasma samples.
When reporting results: indicate the type of assay performed and use the correct units (eg IU/dL) for activity and antigen assays. If calibrants traceable to the IS are not available, use percentage of pooled normal plasma. State the reference range for the method.
High and low activity controls should be included in each assay run. Do not use commercial controls in methods other than those intended for their use.
If gross icterus interferes in some FRET assay methods, the problema can sometimes be resolved by assaying at a higher dilution, treatment with bilirubin oxidase, or using a chromogenic activity ELISA. A comment regarding potential assay interference should be added to the laboratory report.
If decreased ADAMTS13 activity (<20 IU/dL) is detected in a new patient, an ADAMTS13 antibody assay or inhibitor test should be performed. If the results do not match the clinical picture, potential EDTA contamination should be considered and where possible, fresh blood samples obtained.
Wherever possible, use the same ADAMTS13 assay when studying a patient longitudinally to manage treatment.

Mackie I, Mancini I, Muia J, Kremer Hovinga J, Nair S, Machin S, Baker R. International Council for Standardization in Haematology (ICSH) recommendations for laboratory measurement of ADAMTS13. Int J Lab Hematol. 2020 Dec;42(6):685-696.

Clinical laboratory assays for ADAMST13 activity determination methods

	FRETS-VWF73	TECHNOZYM® ADAMTS13 Activity ELISA	HemosIL AcuStar ADAMTS13 Activity Assay
Substrate	FRETS-VWF73	GST-VWF73	GST-VWF73-His
Detection method	Fluorescent: direct detection of VWF cleavage products	Chromogenic: immune detection of VWF cleavage products by specific mAb	Chemiluminescent: Immune detection of VWF cleavage products by specific mAb
Detection limit	3.3%	0.002 IU/mL (0.2%)	0.2%
Assay range	3.3–105%	0.003–1.05 IU/mL (0.3–105%)	0.2–150%
Intra-assay precision (CV)	6.0%	< 5.4%	< 4.4%
Interassay precision (CV)	9.5%	< 8.0%	< 5.1%
Normal range	45–147%	0.40–1.30 IU/mL (40–130%)	67–129%
Assay Length	1 hour	~ 3.5 hours	33 minutes

Gómez-Seguí I, Pascual Izquierdo C, Mingot Castellano ME, de la Rubia Comos J. An update on the pathogenesis and diagnosis of thrombotic thrombocytopenic purpura. Expert Rev Hematol. 2023 Jan;16(1):17-32

Letter to the Editor

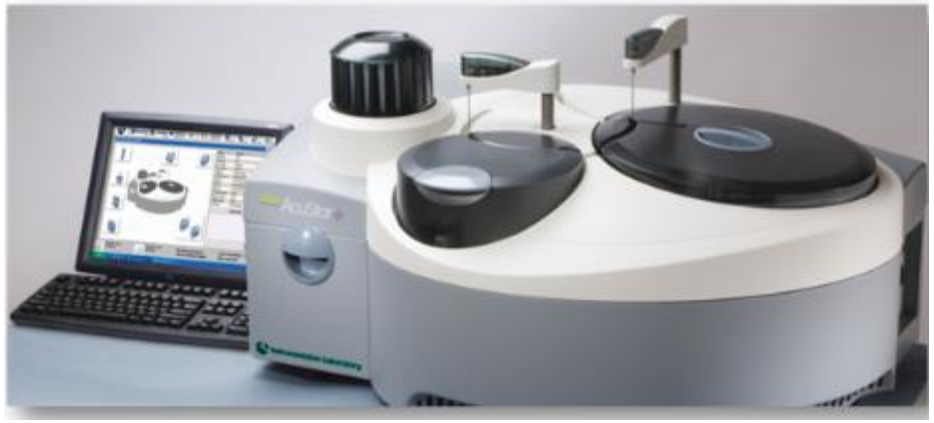
Evaluation of the Fully Automated HemosIL Acustar ADAMTS13 Activity Assay

Julien Favresse[1] Benjamin Lardinois[1] Bernard Chatelain[1] Hugues Jacqmin[1] François Mullier[1]

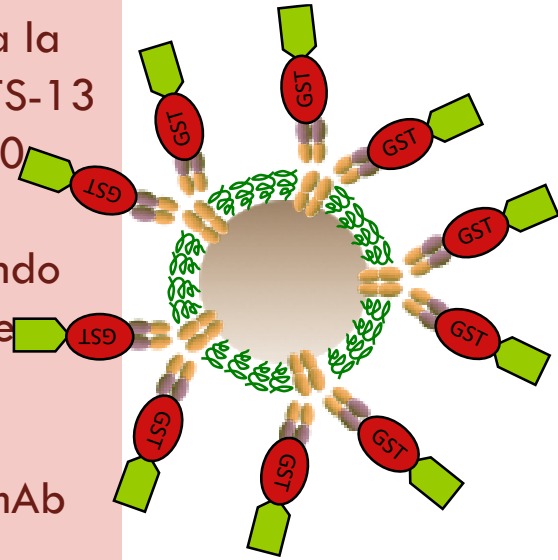
[1]Université catholique de Louvain, CHU UCL Namur, Namur Thrombosis and Hemostasis Center (NTHC), Hematology Laboratory, NARILIS, Yvoir, Belgium

Address for correspondence François Mullier, PharmD, PhD, Université catholique de Louvain, CHU UCL Namur, Namur Thrombosis and Hemostasis Center (NTHC), Hematology Laboratory, NARILIS, Avenue G. Thérasse, 1, Yvoir 5530, Belgium (e-mail: francois.mullier@uclouvain.be).

Thromb Haemost



- Ya comercializada una nueva técnica para la determinación de la actividad del ADAMTS-13 que permitiría obtener los resultados en 30 minutos.
- Se trata de Inmunoensayo en 2 pasos usando como substrato también el GST-VWF73 pe con partículas magnéticas y tecnología de quimioluminiscencia, (HemosIL AcuStar).
- :[4]Trazador: isoluminol-labelled Anti-N10 mAb



Pascual Izquierdo C, Nieto, JM, Fidalgo, T, et al. Multicentric evaluation of the new HemosIL Acustar® chemiluminescence ADAMTS13 activity assay. Int J Lab Hematol. 2021; 43: 485– 493.

Letter to the Editor

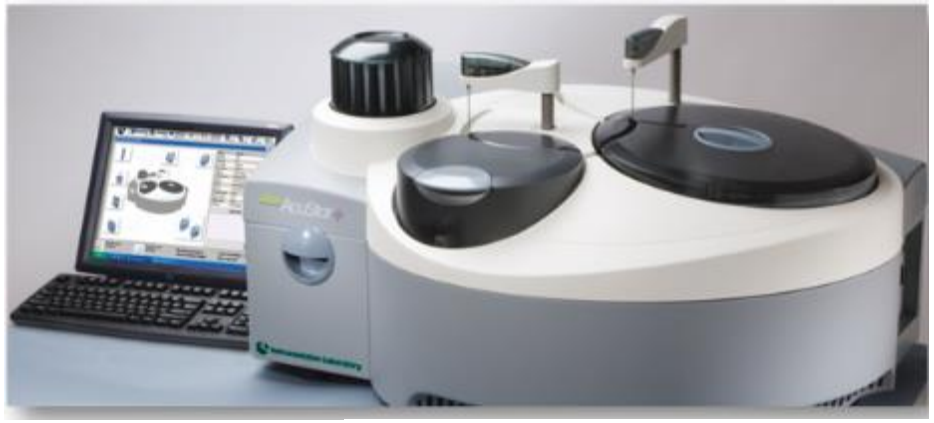
Evaluation of the Fully Automated HemosIL Acustar ADAMTS13 Activity Assay

Julien Favresse[1] Benjamin Lardinois[1] Bernard Chatelain[1] Hugues Jacqmin[1] François Mullier[1]

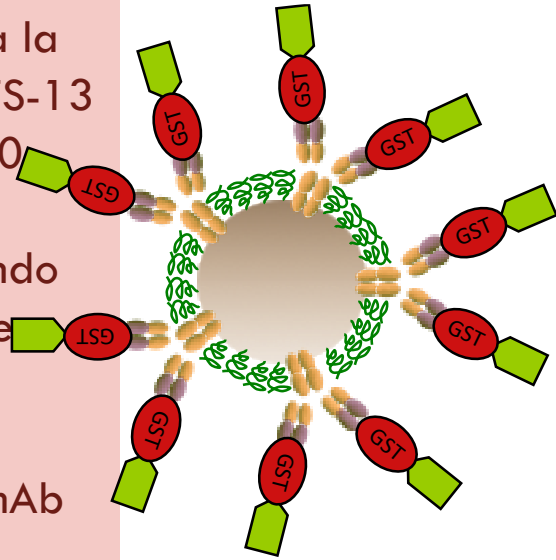
[1]Université catholique de Louvain, CHU UCL Namur, Namur Thrombosis and Hemostasis Center (NTHC), Hematology Laboratory, NARILIS, Yvoir, Belgium

Thromb Haemost

Address for correspondence François Mullier, PharmD, PhD, Université catholique de Louvain, CHU UCL Namur, Namur Thrombosis and Hemostasis Center (NTHC), Hematology Laboratory, NARILIS, Avenue G. Thérasse, 1, Yvoir 5530, Belgium (e-mail: francois.mullier@uclouvain.be).



- Ya comercializada una nueva técnica para la determinación de la actividad del ADAMTS-13 que permitiría obtener los resultados en 30 minutos.
- Se trata de Inmunoensayo en 2 pasos usando como substrato también el GST-VWF73 pe con partículas magnéticas y tecnología de quimioluminiscencia, (HemosIL AcuStar).
- :[4]Trazador: isoluminol-labelled Anti-N10 mAb



Received: 21 September 2020 | Revised: 27 October 2020 | Accepted: 12 November 2020

DOI: 10.1111/ijlh.13414

ORIGINAL ARTICLE

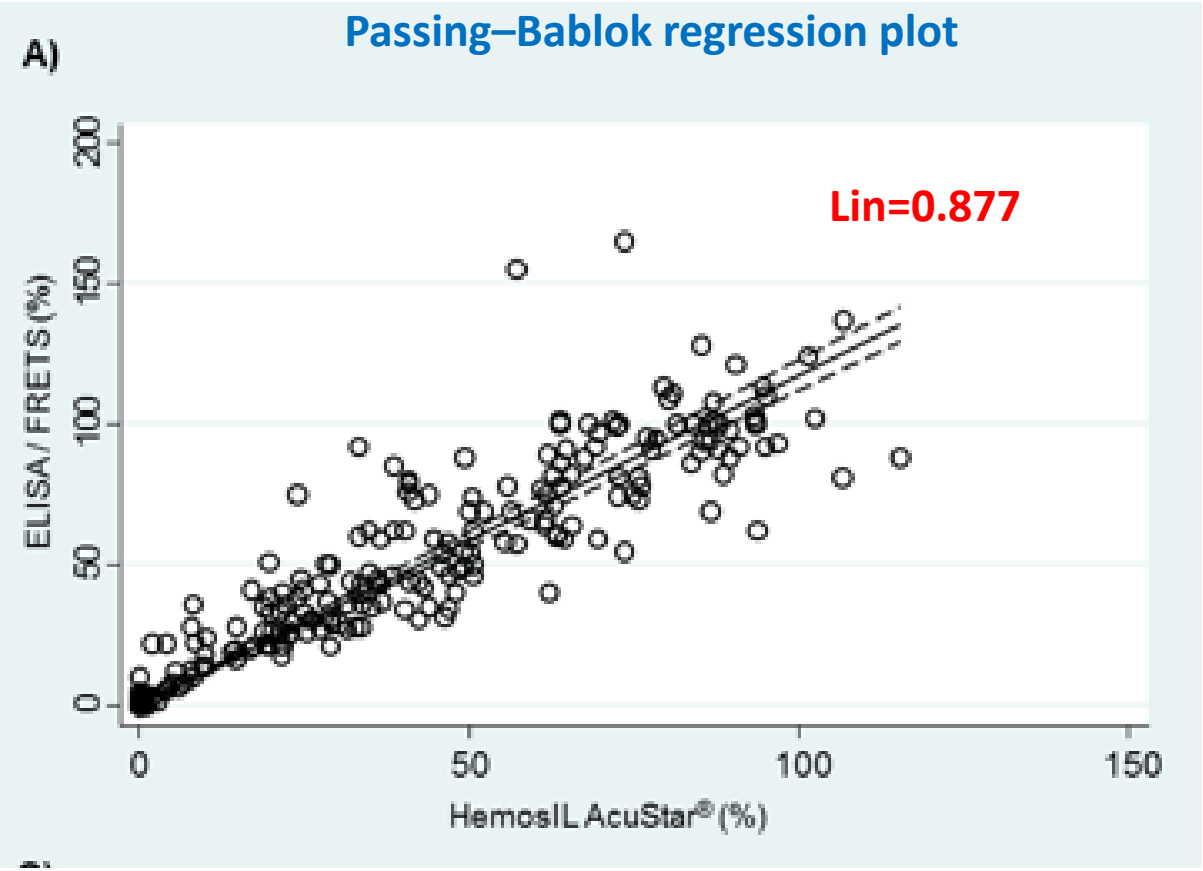
ISLH International Journal of Laboratory Hematology WILEY

Multicentric evaluation of the new HemosIL Acustar® chemiluminescence ADAMTS13 activity assay

Cristina Pascual[1,2] | Jorge M. Nieto[3] | Teresa Fidalgo[4] | Inés Gómez Seguí[5] | Maribel Díaz-Ricart[6] | Marta Fernández Docampo[7] | Julio del Rio[8] | Ramon Salinas[9]

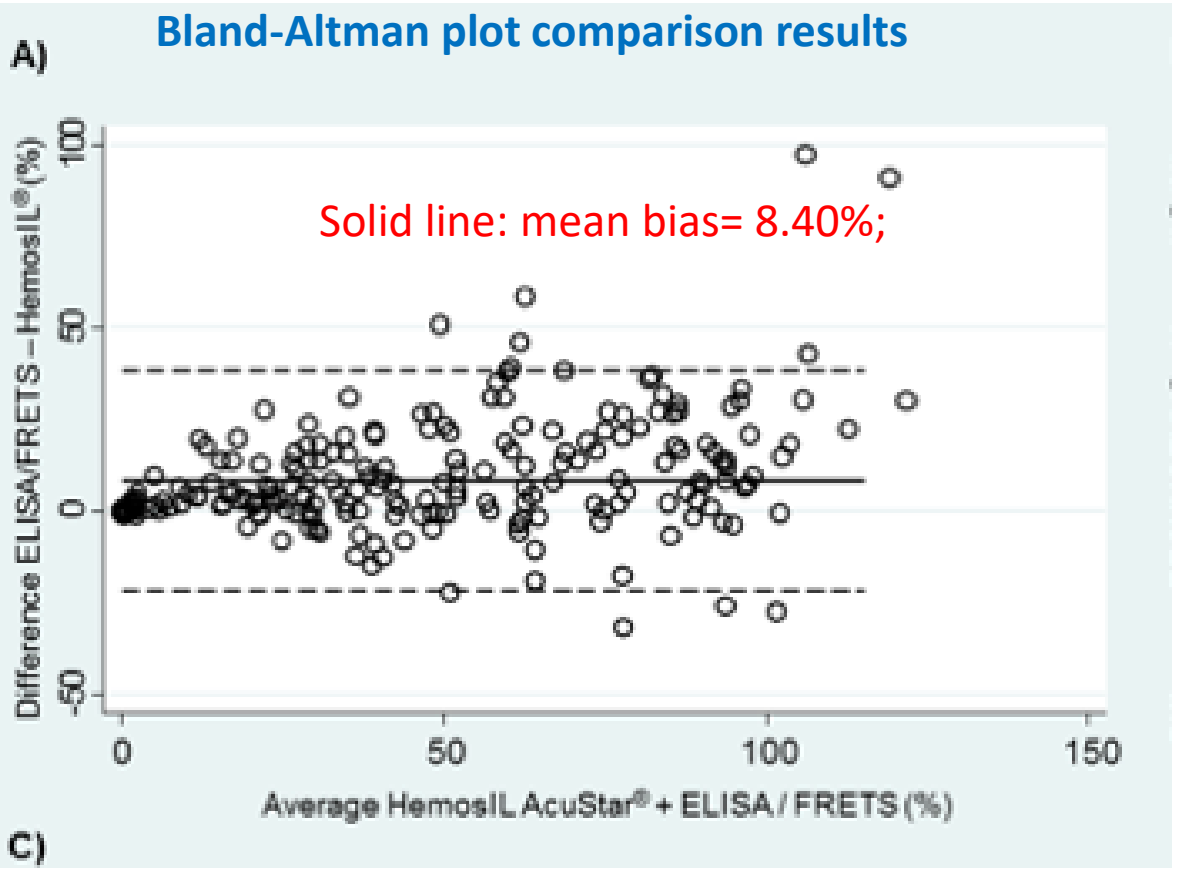
Pascual Izquierdo C, Nieto, JM, Fidalgo, T, et al. Multicentric evaluation of the new HemosIL Acustar® chemiluminescence ADAMTS13 activity assay. Int J Lab Hematol. 2021; 43: 485– 493.

Passing–Bablok regression plot



A) Comparison between HemosIL AcuStar® ADAMTS13 Activity and the other assays. Dashed line: fitted regression line y=0.007+1.175x.

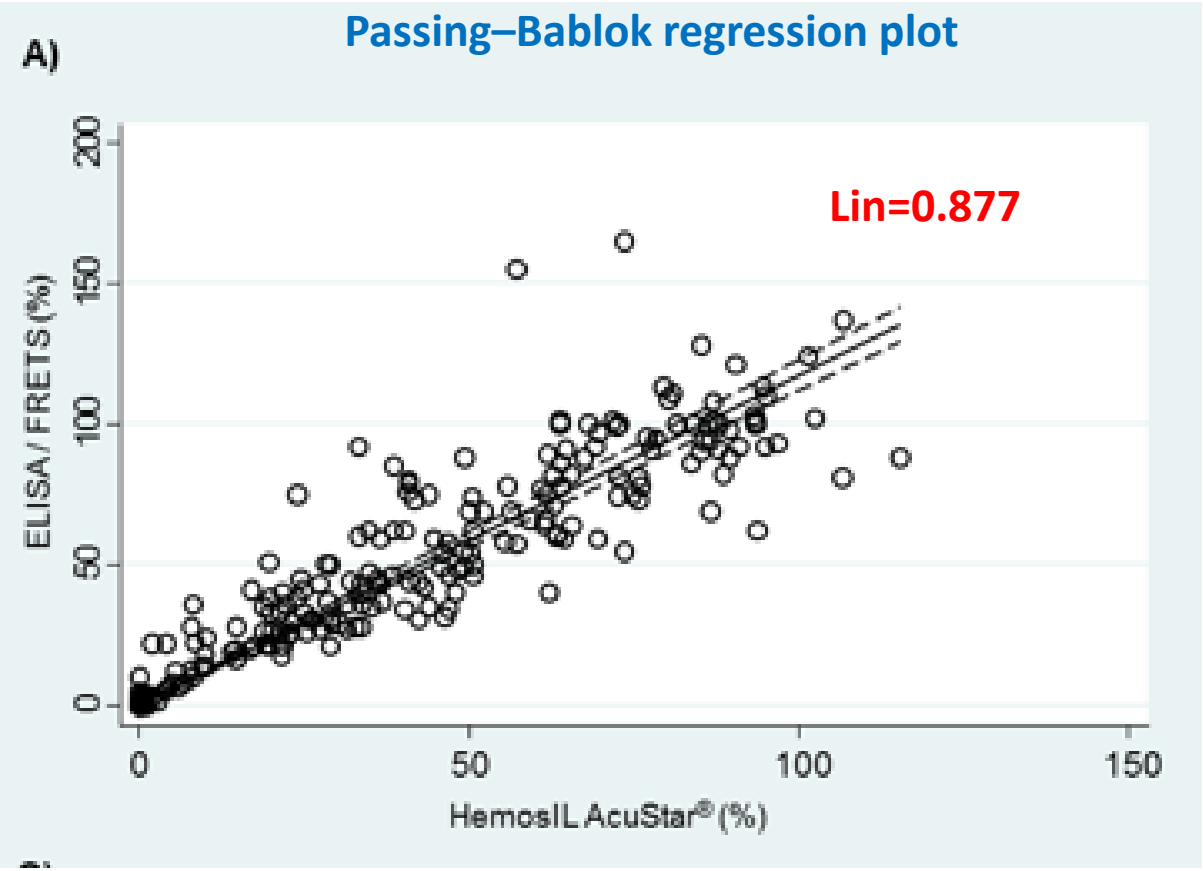
Bland-Altman plot comparison results



B) Comparison between HemosIL AcuStar® ADAMTS13 Activity and the other assays. Solid line: mean bias= 8.40%; dashed lines: 95% limits of agreement –21.60 to 38.50%.

Pascual C, Nieto, JM, Fidalgo, T, et al. Multicentric evaluation of the new HemosIL Acustar® chemiluminescence ADAMTS13 activity assay. Int J Lab Hematol. 2021; 43: 485– 493.

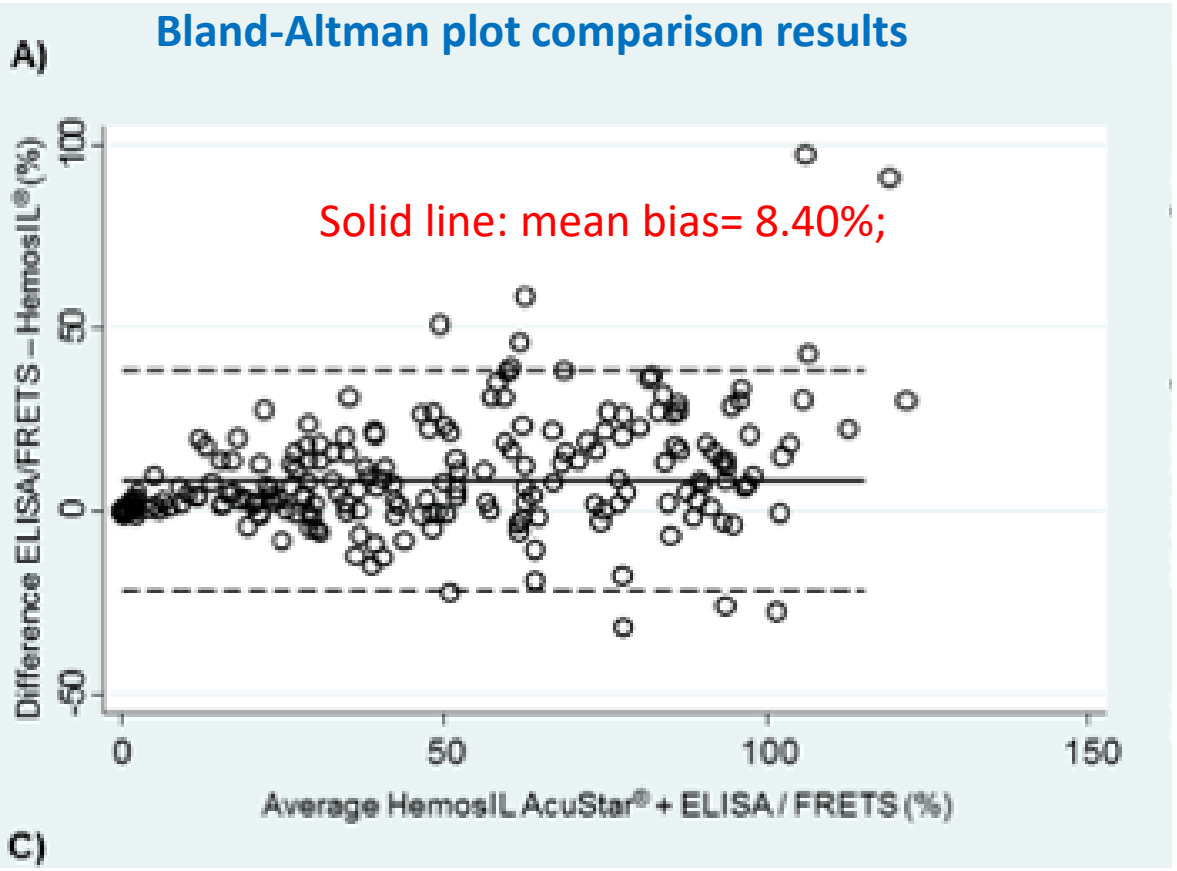
Passing–Bablok regression plot



A) Comparison between HemosIL AcuStar® ADAMTS13 Activity and the other assays. Dashed line: fitted regression line

La técnica CLIA mostró una buena correlación frente al conjunto de técnicas de referencia (Lin=0.877).

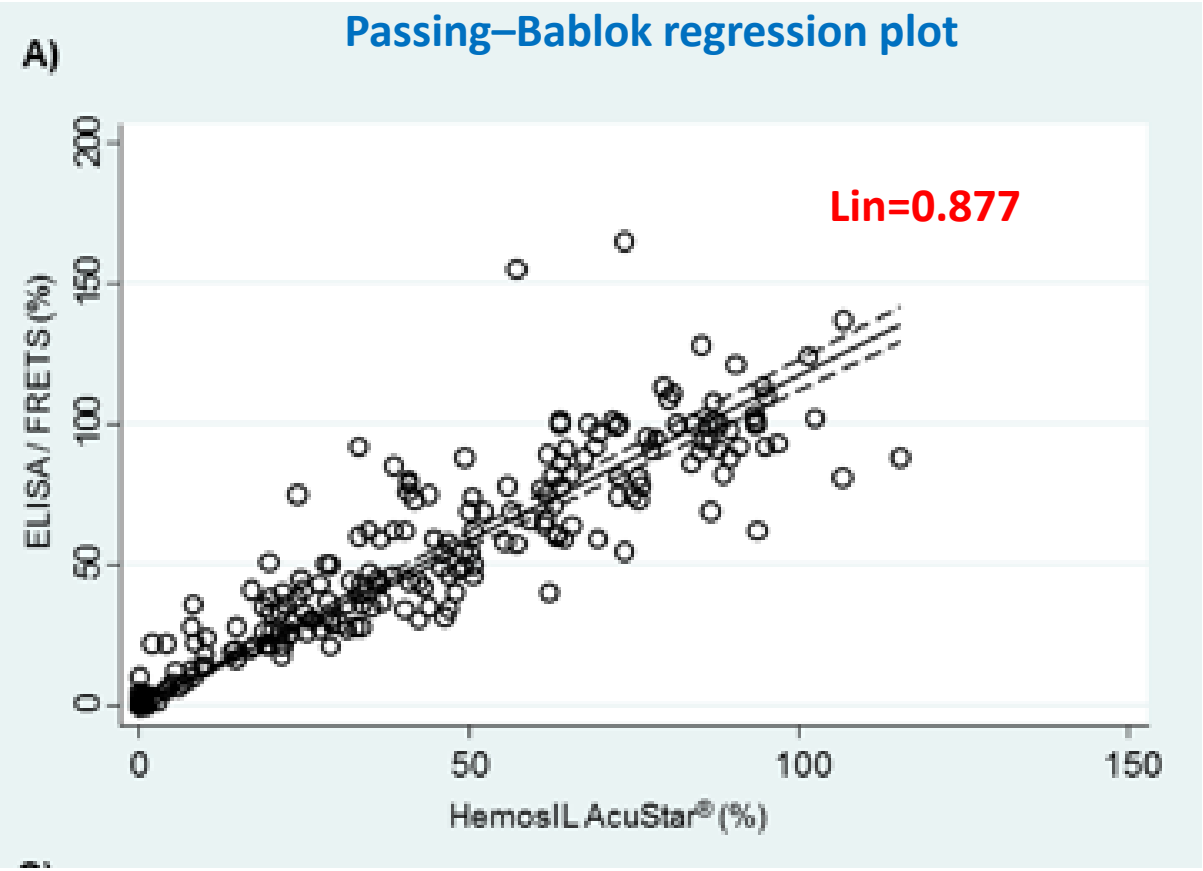
Bland-Altman plot comparison results



B) Comparison between HemosIL AcuStar® ADAMTS13 Activity and the other assays. Solid line: mean bias= 8.40%; dashed lines: 95% limits of agreement –21.60 to 38.50%.

Pascual C, Nieto, JM, Fidalgo, T, et al. Multicentric evaluation of the new HemosIL Acustar® chemiluminescence ADAMTS13 activity assay. Int J Lab Hematol. 2021; 43: 485– 493.

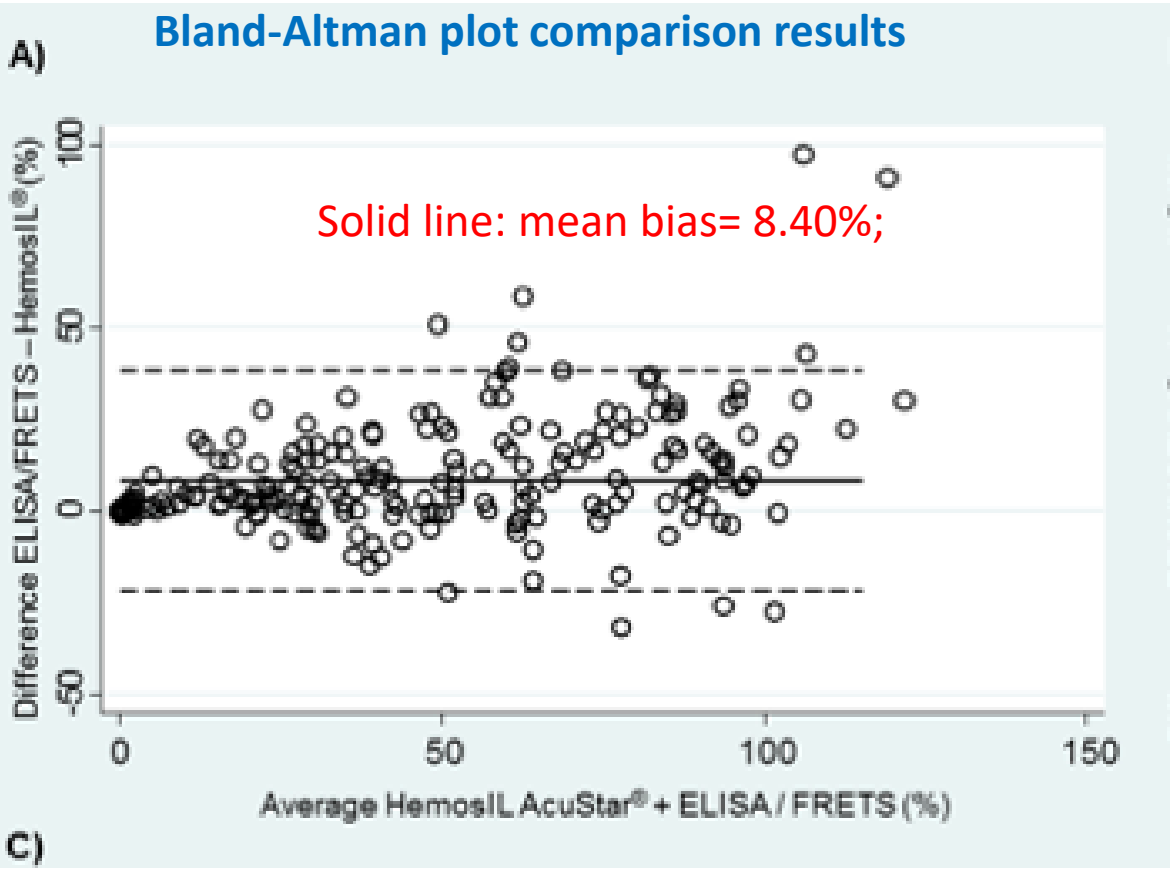
Passing–Bablok regression plot



A) Comparison between HemosIL AcuStar® ADAMTS13 Activity and the other assays. Dashed line: fitted regression line

La técnica CLIA mostró una buena correlación frente al conjunto de técnicas de referencia (Lin=0.877).

Bland-Altman plot comparison results



B) Comparison between HemosIL AcuStar® ADAMTS13 Activity and the other assays.

El análisis de Bland-Altman revela un sesgo medio del 8.4% (95% IC:). Esto indica que CLIA muestra una tendencia a subestimar ligeramente la actividad de ADAMTS13 respecto al resto de técnicas.

Background
References Gold estándar vs CLIA

Researcher Group	Sample n	Year	Comparator method	Discrepant AcuStar results <10%	Other data
Prof. Peyvandi	176	Retrospective: 2006-2018	ELISA Technoclone In-house FRET	None	No differences in aHUS and other TMA patients
Prof. Gresele	125	Retrospective	ELISA Technoclone	None	No inteference with APL, possible interference with anti-FVIII inhibitors
Prof. Veyradier	539	Retrospective (316): Jan 2015-Dec 2019 Prospective (223 consec): 1 Jan-30 Apr 2020	In-house FRET	1 SLE, pregnant patient	Sensitivity 90.1%; Specificity 99.7% 14 samples overestimated by the HemosIL AcuStar assay
Prof. Favaloro	733	Retrospective (385): not available Prospective (348): *(Q2-Q4 2019)*	ELISA Technoclone	12 All with clinical diagnosis of TTP	Multicenter. 2 TTP patients would have been missed by ELISA
Dr. Pascual	241	Retrospective: Jul - Oct 2019	IN-house FRET ELISA Technoclone ELISA Grifols	Number of false positives was low for all comparisons. Global comparison with cut-off <10%: 10 false positives; 7 follow-up samples from TTP patients; 1 not TTP; 2 unknown diagnosis Global comparison with cut-off <5%: 5 false positives Highest agreement with TECHNOZYM ELISA, with 3 false positives for cut-off <10% and	Multicenter. Sensitivity 100% Cut-off value <10%: Specificity 95% Cut-off value <5%: Specificity 97% (5 false pos.) 2-fold mean negative bias for sepsis samples. Sepsis could affect the capacity to assess ADAMTS13 activity. At least 1 of the 5 false positives was a case of sepsis. This type of samples may require double-checking with another method for ADAMTS13
Dr. Goetze	706	Retrospective: 706 consecutive over 14 years	In-house FRET	1 DIC sample <10% with both assays Biggest discrepancy for clinical TTP remission samples: 13 <10 % AS, >10% FRET 7 <10% FRET , >10% AS	FRET: Sensitivity 96.4%; Specificity 98.9% AcuStar: Sensitivity 100%; Specificity 98.9% Higher accuracy with cutoff 5%

Improvement of immunologic and molecular techniques for the diagnosis and follow-up of patients with thrombotic thrombocytopenic purpura: a collaborative study proposal of the spanish apheresis group (GEA) in collaboration with the Spanish Society of Hematology and Hemotherapy (SEHH), NCT05046717.

Objetivos

- Evaluar el rendimiento de la técnica de quimioluminiscencia para la determinación de la actividad de ADAMTS13 en pacientes con sospecha de MAT, de manera comparativa con las técnicas gold estándar (ELISA/FRETS) de forma ***prospectiva.***

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

Material y métodos

- Estudio prospectivo, multicéntrico e internacional entre Octubre 2022 y Enero 2024, para comparar la técnica basada en quimioluminiscencia (CLIA, Werfen, Barcelona, España) frente a FRETS-VWF73 y ELISA comercial (TECHNOZYM ADAMTS13 Activity ELISA Kit, Technoclone, Austria Technoclone) en 6 centros de España y Portugal dentro del proyecto "**Improvement of immunologic and molecular techniques for the diagnosis and follow-up of patients with thrombotic thrombocytopenic purpura: a collaborative study proposal of the spanish apheresis group (GEA) in collaboration with the Spanish Society of Hematology and Hemotherapy (SEHH), NCT05046717.**

ESTUDIO INSPIRER

- Se evauó la correlación (Coeficiente de Lin/ Passing Bablock) y el sesgo (Bland- Altman) entre CLIA y las distintas técnicas de referencia.
- Se utilizó el punto de corte de actividad <10% para evaluar la concordancia (Kappa), sensibilidad y especificidad.

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

MATERIAL Y MÉTODOS

Se analizaron un total de 512 muestras de pacientes con sospecha de MAT por las 2 técnicas (ELISA/FRETS) vs CLIA.

- Cristina Pascual-Izquierdo, Jorge Martínez Nieto, Maribel Diaz-Ricart, Inés Gómez- Seguí, Marta Docampo, Manuel Fernández Villalobos, Rafael López Del Amo, Júlia Martinez-Sanchez, Silvia Escribano-Serrat, Ana Belén Moreno-Castaño, Toni Moscardó, Patricia Martinho,
- University Hospital Gregorio Marañón (Madrid, Spain)
- Instituto de Investigación Sanitaria Gregorio Marañon, Madrid (Spain)
- University Hospital Clínico San Carlos (Madrid, Spain)
- Coimbra Hospital and Universitary Center (Coimbra, Portugal)
- University Hospital La Fe (Valencia, Spain)
- Hematopathology, CDB, Hospital Clínic, IDIBAPS, Universty of Barcelona (Barcelona, Spain)
- University Hospital Xestion Xanitaria A Coruña, (A Coruña, Spain)

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

MATERIAL Y MÉTODOS

Se analizaron un total de 512 muestras de pacientes con sospecha de MAT por las 2 técnicas (ELISA/FRETS) vs CLIA.

- Cristina Pascual-Izquierdo, Jorge Martínez Nieto, Maribel Diaz-Ricart, Inés Gómez- Seguí, Marta Docampo, Manuel Fernández Villalobos, Rafael López Del Amo, Júlia Martinez-Sanchez, Silvia Escribano-Serrat, Ana Belén Moreno-Castaño, Toni Moscardó, Patricia Martinho,
- University Hospital Gregorio Marañón (Madrid, Spain)
- Instituto de Investigación Sanitaria Gregorio Marañon, Madrid (Spain)
- University Hospital Clínico San Carlos (Madrid, Spain)
- Coimbra Hospital and Universitary Center (Coimbra, Portugal)
- University Hospital La Fe (Valencia, Spain)
- Hematopathology, CDB, Hospital Clínic, IDIBAPS, Universty of Barcelona (Barcelona, Spain)
- University Hospital Xestion Xanitaria A Coruña, (A Coruña, Spain)

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

MATERIAL Y MÉTODOS

Se analizaron un total de 512 muestras de pacientes con sospecha de MAT por las 2 técnicas (ELISA/FRETS) vs CLIA.

- Cristina Pascual-Izquierdo, Jorge Martínez Nieto, Maribel Diaz-Ricart, Inés Gómez- Seguí, Marta Docampo, Manuel Fernández Villalobos, Rafael López Del Amo, Júlia Martinez-Sanchez, Silvia Escribano-Serrat, Ana Belén Moreno-Castaño, Toni Moscardó, Patricia Martinho,
- University Hospital Gregorio Marañón (Madrid, Spain)
- Instituto de Investigación Sanitaria Gregorio Marañon, Madrid (Spain)
- University Hospital Clínico San Carlos (Madrid, Spain)
- Coimbra Hospital and Universitary Center (Coimbra, Portugal)
- University Hospital La Fe (Valencia, Spain)
- Hematopathology, CDB, Hospital Clínic, IDIBAPS, Universty of Barcelona (Barcelona, Spain)
- University Hospital Xestion Xanitaria A Coruña, (A Coruña, Spain)

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

MATERIAL Y MÉTODOS

Se analizaron un total de 512 muestras de pacientes con sospecha de MAT por las 2 técnicas (ELISA/FRETS) vs CLIA.

- Cristina Pascual-Izquierdo, Jorge Martínez Nieto, Maribel Diaz-Ricart, Inés Gómez- Seguí, Marta Docampo, Manuel Fernández Villalobos, Rafael López Del Amo, Júlia Martinez-Sanchez, Silvia Escribano-Serrat, Ana Belén Moreno-Castaño, Toni Moscardó, Patricia Martinho,
- University Hospital Gregorio Marañón (Madrid, Spain)
- Instituto de Investigación Sanitaria Gregorio Marañon, Madrid (Spain)
- University Hospital Clínico San Carlos (Madrid, Spain)
- Coimbra Hospital and Universitary Center (Coimbra, Portugal)
- University Hospital La Fe (Valencia, Spain)
- Hematopathology, CDB, Hospital Clínic, IDIBAPS, Universty of Barcelona (Barcelona, Spain)
- University Hospital Xestion Xanitaria A Coruña, (A Coruña, Spain)

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

MATERIAL Y MÉTODOS

Se analizaron un total de 512 muestras de pacientes con sospecha de MAT por las 2 técnicas (ELISA/FRETS) vs CLIA.

- Cristina Pascual-Izquierdo, Jorge Martínez Nieto, Maribel Diaz-Ricart, Inés Gómez- Seguí, Marta Docampo, Manuel Fernández Villalobos, Rafael López Del Amo, Júlia Martinez-Sanchez, Silvia Escribano-Serrat, Ana Belén Moreno-Castaño, Toni Moscardó, Patricia Martinho,
- University Hospital Gregorio Marañón (Madrid, Spain)
- Instituto de Investigación Sanitaria Gregorio Marañon, Madrid (Spain)
- University Hospital Clínico San Carlos (Madrid, Spain)
- Coimbra Hospital and Universitary Center (Coimbra, Portugal)
- University Hospital La Fe (Valencia, Spain)
- Hematopathology, CDB, Hospital Clínic, IDIBAPS, Universty of Barcelona (Barcelona, Spain)
- University Hospital Xestion Xanitaria A Coruña, (A Coruña, Spain)

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

MATERIAL Y MÉTODOS

Se analizaron un total de 512 muestras de pacientes con sospecha de MAT por las 2 técnicas (ELISA/FRETS) vs CLIA.

- Cristina Pascual-Izquierdo, Jorge Martínez Nieto, Maribel Diaz-Ricart, Inés Gómez- Seguí, Marta Docampo, Manuel Fernández Villalobos, Rafael López Del Amo, Júlia Martinez-Sanchez, Silvia Escribano-Serrat, Ana Belén Moreno-Castaño, Toni Moscardó, Patricia Martinho,
- University Hospital Gregorio Marañón (Madrid, Spain)
- Instituto de Investigación Sanitaria Gregorio Marañon, Madrid (Spain)
- University Hospital Clínico San Carlos (Madrid, Spain)
- Coimbra Hospital and Universitary Center (Coimbra, Portugal)
- University Hospital La Fe (Valencia, Spain)
- Hematopathology, CDB, Hospital Clínic, IDIBAPS, Universty of Barcelona (Barcelona, Spain)
- University Hospital Xestion Xanitaria A Coruña, (A Coruña, Spain)

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

MATERIAL Y MÉTODOS

Se analizaron un total de 512 muestras de pacientes con sospecha de MAT por las 2 técnicas (ELISA/FRETS) vs CLIA.

- Cristina Pascual-Izquierdo, Jorge Martínez Nieto, Maribel Diaz-Ricart, Inés Gómez- Seguí, Marta Docampo, Manuel Fernández Villalobos, Rafael López Del Amo, Júlia Martinez-Sanchez, Silvia Escribano-Serrat, Ana Belén Moreno-Castaño, Toni Moscardó, Patricia Martinho,
- University Hospital Gregorio Marañón (Madrid, Spain)
- Instituto de Investigación Sanitaria Gregorio Marañon, Madrid (Spain)
- University Hospital Clínico San Carlos (Madrid, Spain)
- Coimbra Hospital and Universitary Center (Coimbra, Portugal)
- University Hospital La Fe (Valencia, Spain)
- Hematopathology, CDB, Hospital Clínic, IDIBAPS, Universty of Barcelona (Barcelona, Spain)
- University Hospital Xestion Xanitaria A Coruña, (A Coruña, Spain)

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

MATERIAL Y MÉTODOS

Se analizaron un total de 512 muestras de pacientes con sospecha de MAT por las 2 técnicas (ELISA/FRETS) vs CLIA.

- Cristina Pascual-Izquierdo, Jorge Martínez Nieto, Maribel Diaz-Ricart, Inés Gómez- Seguí, Marta Docampo, Manuel Fernández Villalobos, Rafael López Del Amo, Júlia Martinez-Sanchez, Silvia Escribano-Serrat, Ana Belén Moreno-Castaño, Toni Moscardó, Patricia Martinho,
- University Hospital Gregorio Marañón (Madrid, Spain)
- Instituto de Investigación Sanitaria Gregorio Marañon, Madrid (Spain)
- University Hospital Clínico San Carlos (Madrid, Spain)
- Coimbra Hospital and Universitary Center (Coimbra, Portugal)
- University Hospital La Fe (Valencia, Spain)
- Hematopathology, CDB, Hospital Clínic, IDIBAPS, Universty of Barcelona (Barcelona, Spain)
- University Hospital Xestion Xanitaria A Coruña, (A Coruña, Spain)

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

MATERIAL Y MÉTODOS

Se analizaron un total de 512 muestras de pacientes con sospecha de MAT por las 2 técnicas (ELISA/FRETS) vs CLIA.

- Cristina Pascual-Izquierdo, Jorge Martínez Nieto, Maribel Diaz-Ricart, Inés Gómez- Seguí, Marta Docampo, Manuel Fernández Villalobos, Rafael López Del Amo, Júlia Martinez-Sanchez, Silvia Escribano-Serrat, Ana Belén Moreno-Castaño, Toni Moscardó, Patricia Martinho,
- University Hospital Gregorio Marañón (Madrid, Spain)
- Instituto de Investigación Sanitaria Gregorio Marañon, Madrid (Spain)
- University Hospital Clínico San Carlos (Madrid, Spain)
- Coimbra Hospital and Universitary Center (Coimbra, Portugal)
- University Hospital La Fe (Valencia, Spain)
- Hematopathology, CDB, Hospital Clínic, IDIBAPS, Universty of Barcelona (Barcelona, Spain)
- University Hospital Xestion Xanitaria A Coruña, (A Coruña, Spain)

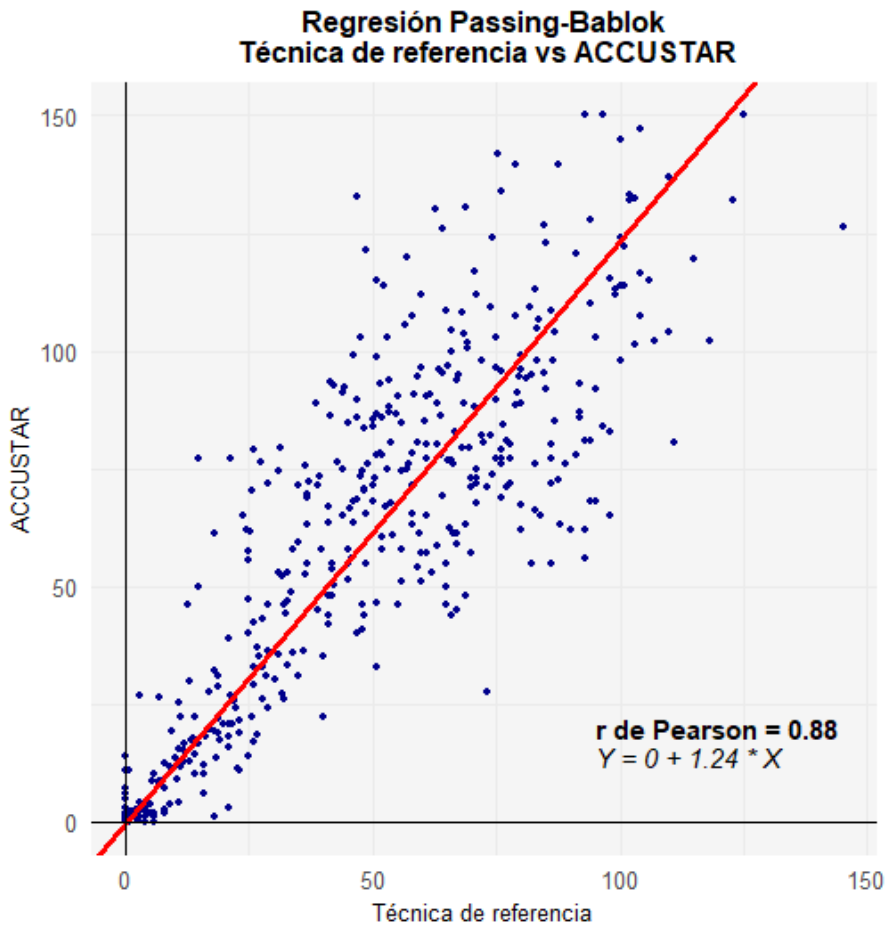
C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

Tabla 1.
Caracterización de las muestras del estudio.

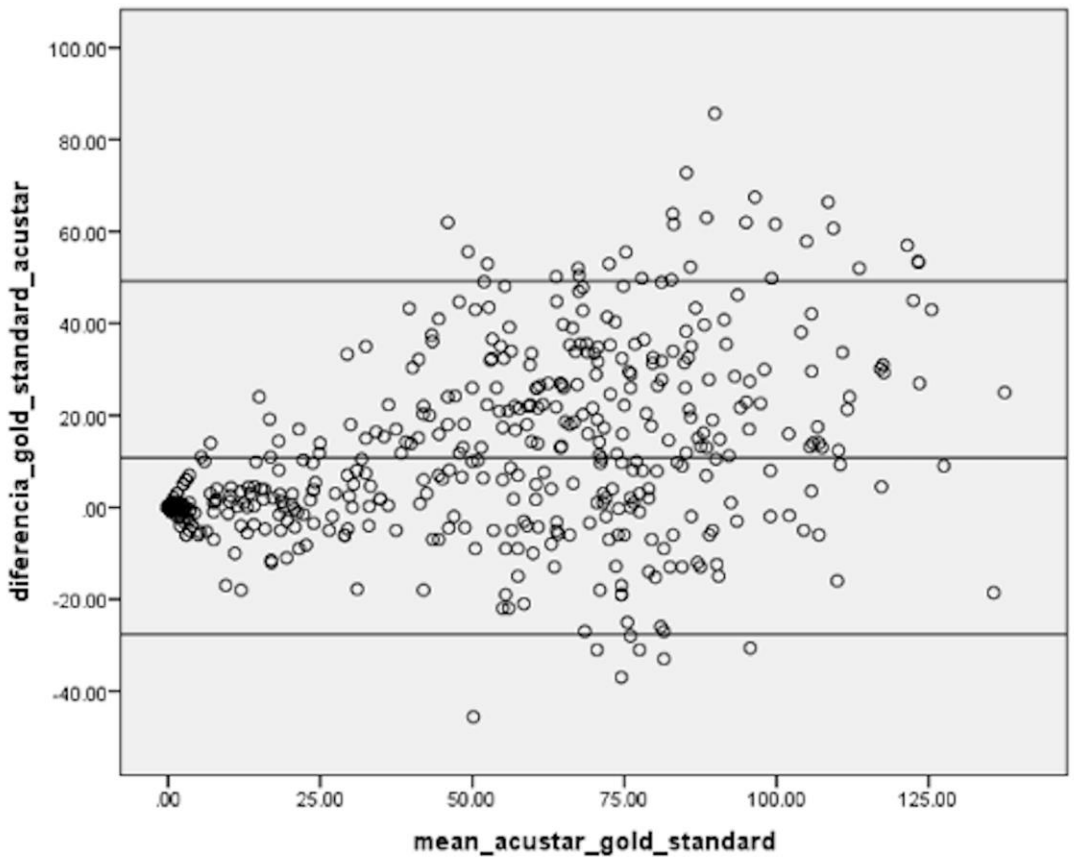
		Número de muestras (%)
Tipo de muestra	Diagnóstico PTTi	74 (14.5%)
	Seguimiento PTTi	365 (71.2%)
	PTTc	38 (7.4%)
	SHU	5 (1.0%)
	Otras MAT	30 (5.9%)
	Total	512 (100%)
Técnica gold standar	TECHNOZYM® ADAMTS13 Activity ELISA	432 (84.4%)
	FRETS-VWF73	80 (15.6%)
	Total	512 (100%)
Método de comparación	ACUSTAR	512 (100%)
	Total	512 (100%)
Rangos de actividad ELISA	0 – 10	86 (20.0%)
	11 – 40	90 (20.9%)
	>40	255 (59.1%)
	Total	432 (100%)
Rangos de actividad FRET	0 – 10	44 (55.0%)
	11 – 40	24 (30.0%)
	>40	12 (15.0%)
	Total	80 (100.0%)
Rangos de actividad ACUSTAR (CLIA)	0 – 10	128 (25.0%)
	11 – 40	78 (15.2%)
	>40	306 (59.8)
	Total	512 (100%)

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA), Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618. DOI:10.1182/blood-2024-209841

Passing–Bablok regression plot



Bland-Altman plot comparison results

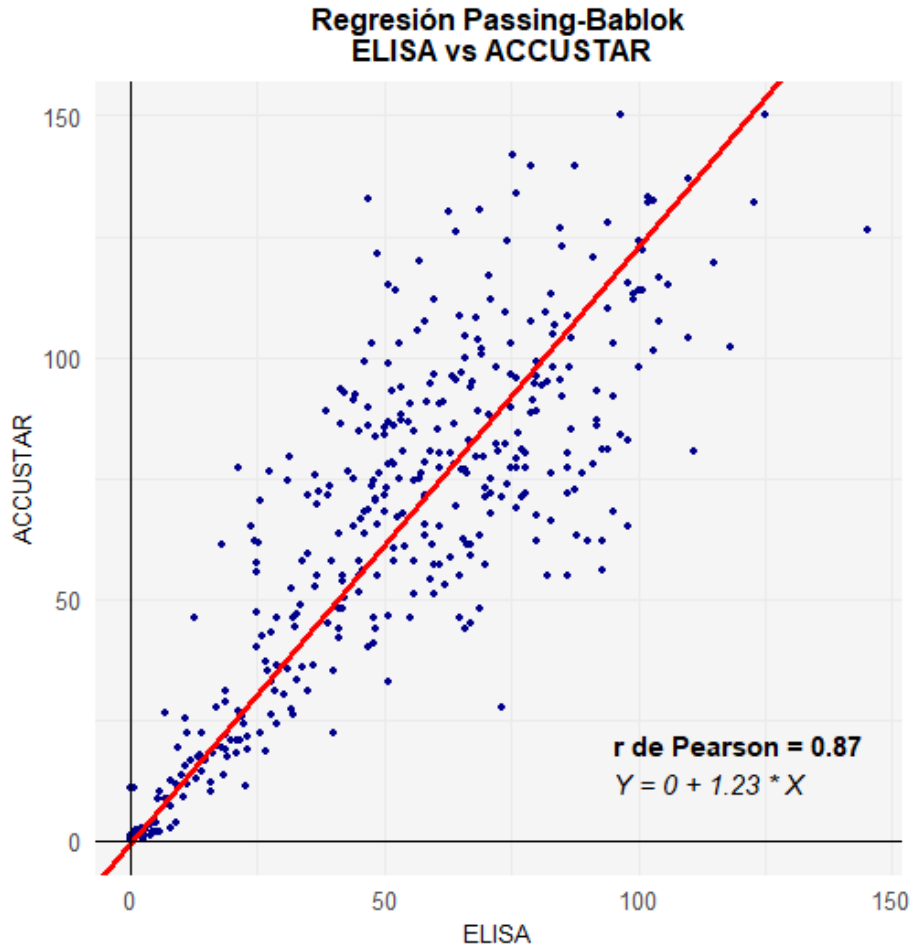


La media de la diferencia entre el gold standard (ELISA o FRETS) y ACUSTAR fue de 10,8 con un límite de acuerdo entre -27,6 y 49,2.

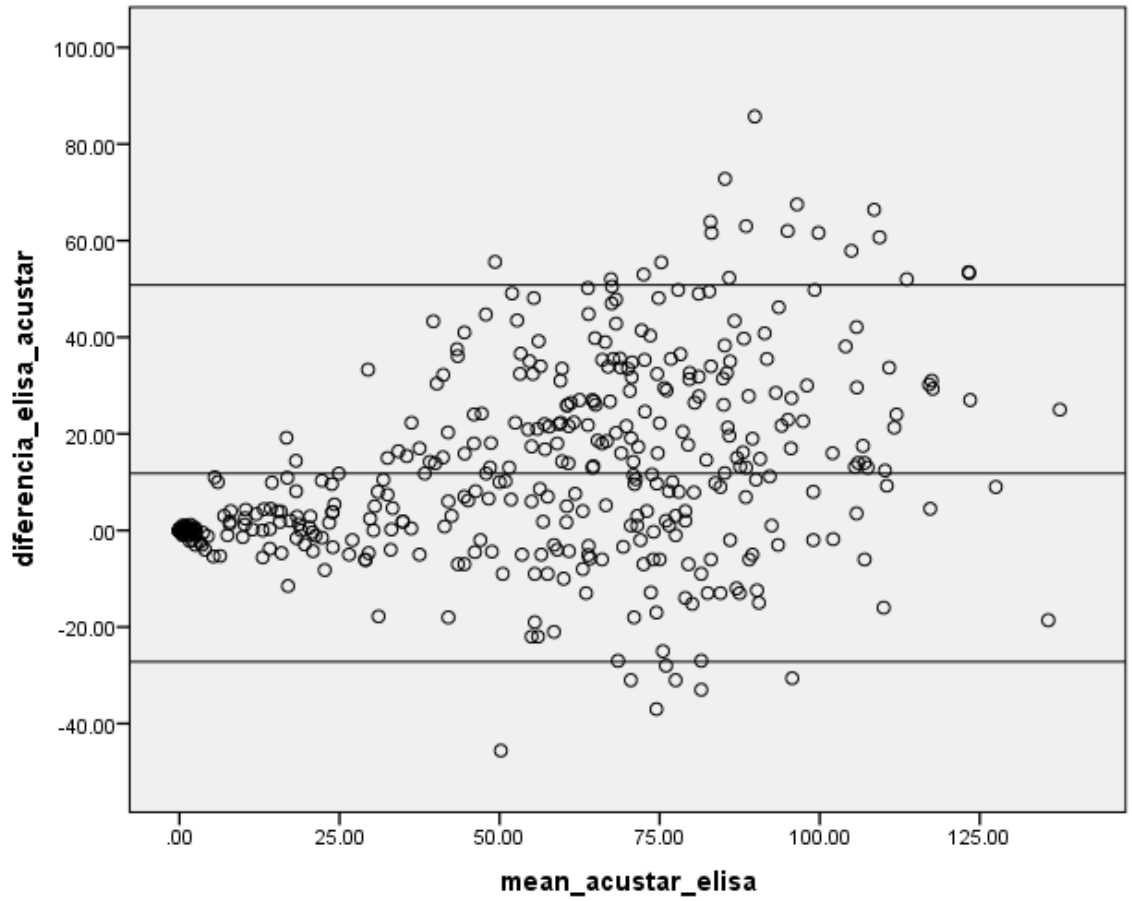
C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

ELISA vs ACUSTAR

Passing–Bablok regression plot



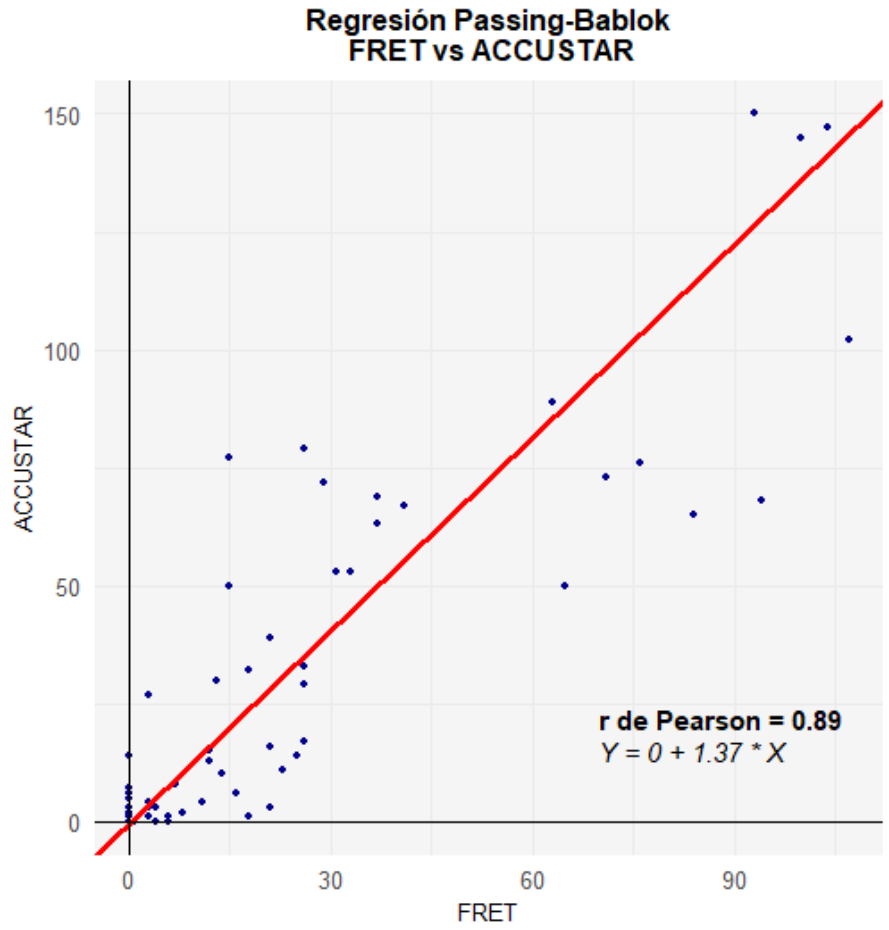
Bland-Altman plot comparison results



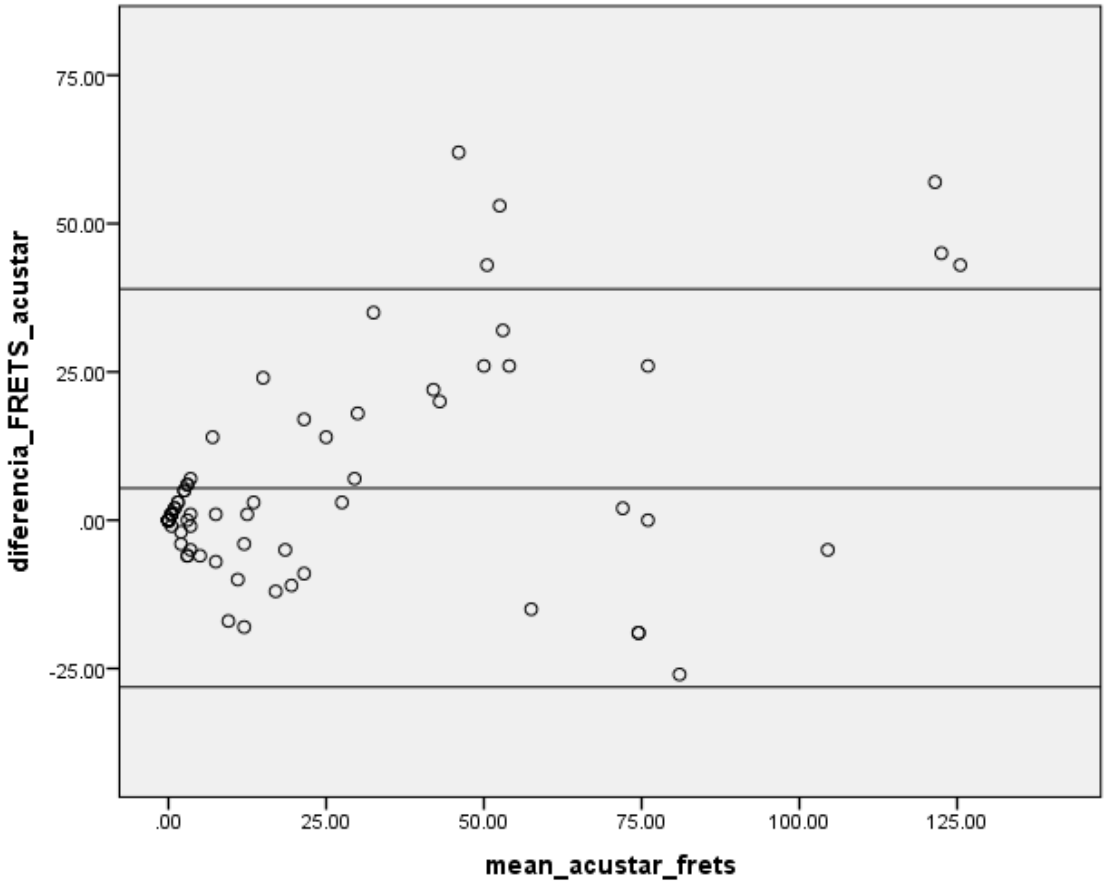
La media de la diferencia entre ELISA y ACUSTAR fue de 11,8 con un límite de acuerdo entre -27,2 y 50,8.

FRET vs ACUSTAR

Passing–Bablok regression plot



Bland-Altman plot comparison results



La media de la diferencia entre FRETS y ACUSTAR fue de 5,4 con un límite de acuerdo entre -28,1 y 38,9.

Resultados

5 falsos positivos

10 falsos negativos

	ELISA (N=432)		FRET (N=80)	
	Positive	Negative	Positive	Negative
A. Cut-off value <10% ADAMTS13 activity values				
HemosIL AcuStar® (Positive)	77	1	42	4
HemosIL AcuStar® (Negative)	8	346	2	32
B. Cut-off value <5% ADAMTS13 activity values				
HemosIL AcuStar® (Positive)	67	5	32	7
HemosIL AcuStar® (Negative)	2	357	7	34

Para el *cut-off* de actividad de ADAMTS13 de <10%: 10 falsos negativos con la técnica CLIA, comparando con ELISA/y 2 para FRETS (Todas ellas de muestras de pacientes con PTTi en seguimiento, excepto 1 al diagnóstico.
5 falsos positivos 1 comparando con ELISA y 4 con FRETS, todas PTTi en seguimiento.

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

Resultados

5 falsos positivos

10 falsos negativos

	ELISA (N=432)		FRET (N=80)	
	Positive	Negative	Positive	Negative
A. Cut-off value <10% ADAMTS13 activity values				
HemosIL AcuStar® (Positive)	77	1	42	4
HemosIL AcuStar® (Negative)	8	346	2	32
B. Cut-off value <5% ADAMTS13 activity values				
HemosIL AcuStar® (Positive)	67	5	32	7
HemosIL AcuStar® (Negative)	2	357	7	34

Para el *cut-off* de actividad de ADAMTS13 de <10%: 10 falsos negativos con la técnica CLIA, comparando con ELISA/y 2 para FRETS (Todas ellas de muestras de pacientes con PTTi en seguimiento, excepto 1 al diagnóstico.
5 falsos positivos 1 comparando con ELISA y 4 con FRETS, todas PTTi en seguimiento.

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

Sensibilidad y especificidad

	Gold estándar type 1 (N=512)	
	Positive	**Negative**
Tester (Positive)	119	5
Tester (Negative)	10	378

Correctly classified sample	0.971 (0.956, 0.986)
Misclassified samples	0.029 (0.014, 0.044)
Sensitivity	0.922 (0.876, 0.968)
Specificity	0.987 (0.976, 0.998)
False positive rate	0.013 (0.002, 0.024)
False negative rate	0.078 (0.032, 0.124)
Prevalence	0.252 (0.214, 0.290)
PPV	0.960 (0.926, 0.994)
NPV	0.974 (0.958, 0.990)

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

Sensibilidad y especificidad

	Gold estándar type 1 (N=512)	
	Positive	**Negative**
Tester (Positive)	119	5
Tester (Negative)	10	378

Correctly classified sample	0.971 (0.956, 0.986)
Misclassified samples	0.029 (0.014, 0.044)
Sensitivity	0.922 (0.876, 0.968)
Specificity	0.987 (0.976, 0.998)
False positive rate	0.013 (0.002, 0.024)
False negative rate	0.078 (0.032, 0.124)
Prevalence	0.252 (0.214, 0.290)
PPV	0.960 (0.926, 0.994)
NPV	0.974 (0.958, 0.990)

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

Sensibilidad y especificidad

	Gold estándar type 1 (N=512)	
	Positive	**Negative**
Tester (Positive)	119	5
Tester (Negative)	10	378

Correctly classified sample	0.971 (0.956, 0.986)
Misclassified samples	0.029 (0.014, 0.044)
Sensitivity	0.922 (0.876, 0.968)
Specificity	0.987 (0.976, 0.998)
False positive rate	0.013 (0.002, 0.024)
False negative rate	0.078 (0.032, 0.124)
Prevalence	0.252 (0.214, 0.290)
PPV	0.960 (0.926, 0.994)
NPV	0.974 (0.958, 0.990)

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

Discrepancias ELIS/FRETS vs ACUSTAR

Record ID	Diagnostico	Técnica ADAMTS13 local FRETS	Actividad ADAMTS13 ELISA	ELISA_inferior_10	Actividad ADAMTS13 FRETS	FRETS_inferior_10	ADAMTS13 ACUSTAR	Acustar positvo	
132-92	PTTa seguimiento	ELISA	10.57	Negativo	.	.	9.20	Positivo	FALSO POSTIVO
135-16	PTTa seguimiento	ELISA	7,10	Positivo			26,30	Negativo	FALSO NEGATIVO
135-45	PTTa seguimiento	ELISA	9,50	Positivo			19,40	Negativo	FALSO NEGATIVO
135-53	PTTa seguimiento	ELISA	8,20	Positivo			12,50	Negativo	FALSO NEGATIVO
131-71	PTTa seguimiento	ELISA	9,00	Positivo			11,50	Negativo	FALSO NEGATIVO
136-74	PTTa seguimiento	ELISA	1,00	Positivo			11,00	Negativo	FALSO NEGATIVO
136-62	PTTa dx	ELISA	0,00	Positivo			11,00	Negativo	FALSO NEGATIVO
133-7	PTT congÃ©nita	ELISA	9,60	Positivo			10,80	Negativo	FALSO NEGATIVO
136-64	PTTa seguimiento	ELISA	6,00	Positivo			10,00	Negativo	FALSO NEGATIVO
134-59	PTTa seguimiento	FRET			16,00	Negativo	6,00	Positivo	FALSO POSTIVO
134-60	PTTa seguimiento	FRET			11,00	Negativo	4,00	Positivo	FALSO POSTIVO
134-84	PTTa seguimineto	FRET			21,00	Negativo	3,00	Positivo	FALSO POSTIVO
134-69	PTTa seguimineto	FRET			18,00	Negativo	1,00	Positivo	FALSO POSTIVO
134-32	PTTa seguimineto	FRET			3,00	Positivo	27,00	Negativo	FALSO NEGATIVO
134-66	PTTa seguimineto	FRET			0,00	Positivo	14,00	Negativo	FALSO NEGATIVO

Discrepancias ELIS/FRETS vs ACUSTAR

Record ID	Diagnostico	Técnica ADAMTS13 local FRETS	Actividad ADAMTS13 ELISA	ELISA_inferior_10	Actividad ADAMTS13 FRETS	FRETS_inferior_10	ADAMTS13 ACUSTAR	Acustar positvo	
132-92	PTTa seguimiento	ELISA	10.57	Negativo	.	.	9.20	Positivo	FALSO POSTIVO
135-16	PTTa seguimiento	ELISA	7,10	Positivo			26,30	Negativo	FALSO NEGATIVO
135-45	PTTa seguimiento	ELISA	9,50	Positivo			19,40	Negativo	FALSO NEGATIVO
135-53	PTTa seguimiento	ELISA	8,20	Positivo			12,50	Negativo	FALSO NEGATIVO
131-71	PTTa seguimiento	ELISA	9,00	Positivo			11,50	Negativo	FALSO NEGATIVO
136-74	PTTa seguimiento	ELISA	1,00	Positivo			11,00	Negativo	FALSO NEGATIVO
136-62	PTTa dx	ELISA	0,00	Positivo			11,00	Negativo	FALSO NEGATIVO
133-7	PTT congÃ©nita	ELISA	9,60	Positivo			10,80	Negativo	FALSO NEGATIVO
136-64	PTTa seguimiento	ELISA	6,00	Positivo			10,00	Negativo	FALSO NEGATIVO
134-59	PTTa seguimiento	FRET			16,00	Negativo	6,00	Positivo	FALSO POSTIVO
134-60	PTTa seguimiento	FRET			11,00	Negativo	4,00	Positivo	FALSO POSTIVO
134-84	PTTa seguimineto	FRET			21,00	Negativo	3,00	Positivo	FALSO POSTIVO
134-69	PTTa seguimineto	FRET			18,00	Negativo	1,00	Positivo	FALSO POSTIVO
134-32	PTTa seguimineto	FRET			3,00	Positivo	27,00	Negativo	FALSO NEGATIVO
134-66	PTTa seguimineto	FRET			0,00	Positivo	14,00	Negativo	FALSO NEGATIVO

Conclusiones

- In this prospective study, which includes a high number of samples of suspected TMA, we demonstrate that there is a good agreement in the ADAMTS13 activity results obtained by CLIA with respect to the "gold standard" FRETS/ELISA methods.
- We found discrepancies mostly in some follow-up iTTP samples, which could have therapeutic implications in a small number of patients.
- To date, borderline results with CLIA should be confirm with a validation technique.

C. Pascual-Izquierdo et al. ADAMTS13 Activity Measurement Using a Chemiluminescence-Based Technique. Prospective Multicenter Comparative Study of the Spanish Apheresis Group (GEA)- Spanish Society of Hematology and Hemotherapy (SEHH). Blood 144 (Supplement 1):2618-2618, DOI:10.1182/blood-2024-200941

Otras técnicas en desarrollo

A multi-center evaluation of TECHNOSCREEN® ADAMTS-13 activity assay as a screening tool for detecting deficiency of ADAMTS-13

Gary W Moore [1 2], Daniëlle Meijer [3], Margaret Griffiths [4], Lucy Rushen [1], Alice Brown [1], Ulrich Budde [5], Rita Dittmer [5], Barbara Schocke [5], Anja Leyte [3 6], Sabine Geiter [4], Anneke Moes [3], Jacqueline A Cutler [1], Nikolaus B Binder [4]

TECHNOSCREEN **ADAMTS13**

CEVERON ® S 100

Real-world evaluation of a novel automated method for measuring ADAMTS13 activity using the Ceveron® s100 analyzer.

Bérangère S. Joly, Hélène Deniau, Chloé Doinel, Adeline Brouillard, Virginie Siguret, Paul Coppo, Agnès Veyradier

PII: S1538-7836(25)00137-0

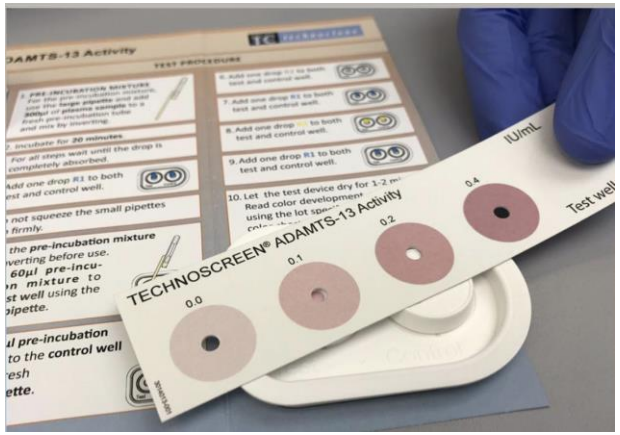
DOI: https://doi.org/10.1016/j.jtha.2025.02.038

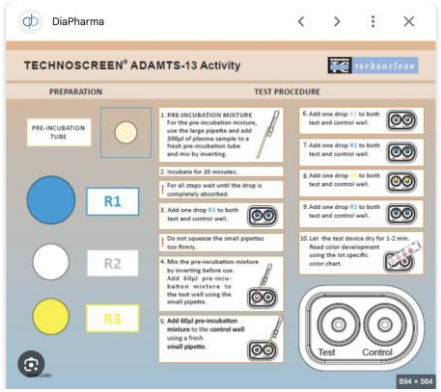
Reference: JTHA 988

Moore GW, Meijer D, Griffiths M, Rushen L, Brown A, Budde U, Dittmer R, Schocke B, Leyte A, Geiter S, Moes A, Cutler JA, Binder NB. A multi-center evaluation of TECHNOSCREEN® ADAMTS-13 activity assay as a screening tool for detecting deficiency of ADAMTS-13. J Thromb Haemost. 2020 Jul;18(7):1686-1694.
Joly BS, Deniau H, Doinel C, Brouillard A, Siguret V, Coppo P, Veyradier A. Real-world evaluation of a novel automated method for measuring ADAMTS13 activity using the Ceveron® s100 analyzer. J Thromb Haemost. 2025 Mar 7:S1538-7836(25)00137-0.

A multi-center evaluation of TECHNOSCREEN® ADAMTS-13 activity assay as a screening tool for detecting deficiency of ADAMTS-13

Gary W Moore [1] [2], Daniëlle Meijer [3], Margaret Griffiths [4], Lucy Rushen [1], Alice Brown, Ulrich Budde [5], Rita Dittmer [5], Barbara Schocke [5], Anja Leyte [3] [6], Sabine Geiter [4], Anneke Moes [3], Jacqueline A Cutler [1], Nikolaus B Binder [4]





Moore GW, Meijer D, Griffiths M, Rushen L, Brown A, Budde U, Dittmer R, Schocke B, Leyte A, Geiter S, Moes A, Cutler JA, Binder NB. A multi-center evaluation of TECHNOSCREEN® ADAMTS-13 activity assay as a screening tool for detecting deficiency of ADAMTS-13. J Thromb Haemost. 2020 Jul;18(7):1686-1694.

TECHNOSCREEN **ADAMTS13**

Abstract

Background: Quantifying A disintegrin-like and metalloprotease with thrombospondin type 1 motif, member 13 (ADAMTS-13) activity enhances thrombotic thrombocytopenic purpura (TTP) diagnosis but most assays are time consuming, technically demanding, and mainly available in reference centers.

Objective: Evaluate a simple, semiquantitative ADAMTS-13 activity screening test for early identification/exclusion of TTP.

Patients/methods: Plasma from 220 patients with suspected thrombotic microangiopathy at three reference centers were tested with TECHNOSCREEN® ADAMTS13 activity screening test in comparison with TECHNOZYM® ADAMTS-13 activity ELISA at two centers, and in-house fluorescence resonance energy transfer assay at the third center. The screening test indicates if ADAMTS-13 activity is at one of four level-indicator points: 0, 0.1, 0.4, or 0.8 IU/mL.

Results: Screen results were interpreted as binary data in that ADAMTS-13 activity was above or below the 0.1 IU/mL TTP clinical threshold. Combining all sites' data, the screen exhibited 88.7% sensitivity, 90.4% specificity, 74.6% positive predictive value, and 96.2% negative predictive value, comparable to published data for quantitative assays. Five samples with quantitative results below the threshold gave screen readings of 0.1 IU/mL and seven marginally above the threshold gave screen readings of zero. All would warrant plasma exchange while the level is quantified. Nine samples with normal/near normal results gave screens of zero and confirmatory quantifications would prompt early treatment withdrawal, as is current practice. One sample generated screen/quantitative results of 0.4/0.00 IU/mL respectively and was the only clear false-negative.

Conclusions: The screening test provides more rapid ADAMTS-13 level evaluation than most currently available assays. Its simple operation renders it suitable for adoption in routine or specialist laboratory environments.

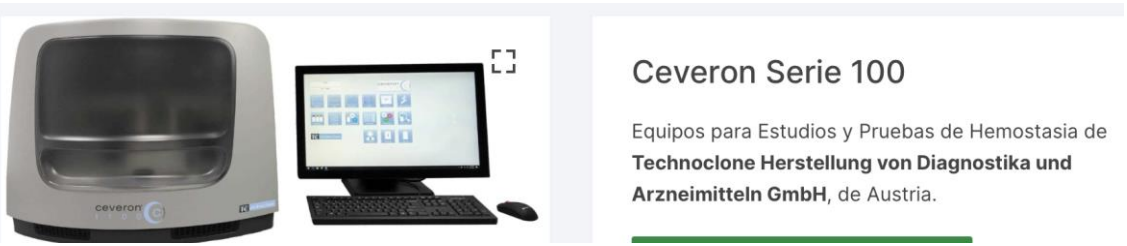
Real-world evaluation of a novel automated method for measuring ADAMTS13 activity using the Ceveron® s100 analyzer.

Bérangère S. Joly, Hélène Deniau, Chloé Doinel, Adeline Brouillard, Virginie Siguret, Paul Coppo, Agnès Veyradier

PII: S1538-7836(25)00137-0

DOI: https://doi.org/10.1016/j.jtha.2025.02.038

Reference: JTHA 988



CEVERON ® S 100

Abstract

Introduction: Thrombotic thrombocytopenic purpura (TTP) is a thrombotic microangiopathy characterized by a severe functional deficiency of ADAMTS13. Measuring ADAMTS13 activity is crucial for diagnosing TTP (<10 IU/dL), monitoring treatments, and detecting relapses (<20 IU/dL). The Technofluor® assay allows a rapid ADAMTS13 activity measurement using the CEVERON® s100 analyzer. This study aims to evaluate the analytical and clinical performance of this new test under real-world conditions.

Materials and methods: ADAMTS13 activity was measured using two fluorometric methods: the Technofluor® ADAMTS13 activity on the CEVERON® s100 (Technoclone, Austria) and our reference FRETS-VWF73 method, in plasma samples collected under real-life conditions (01/12/2024-04/04/2024) and retrospectively selected samples from our biobank. The analytical and clinical performance of the new test was assessed, focusing on the critical low levels (<30 IU/dL).

Results: Four hundred samples were tested under real-world conditions and 100 others tested retrospectively. The Technofluor® assay showed excellent analytical performance, with a detection limit of 0.1 IU/dL, repeatability CV <11%, and reproducibility CV of 6.1% (high level [90 IU/dL]) and 7.5% (low level [40 IU/dL]). Clinical performances were strong at diagnosis (threshold: 10 IU/dL; sensitivity and positive predictive value: 1.0) and during the follow-up (threshold: 20 IU/dL; specificity: 1.0 [95% CI: 0.99;1.00], positive predictive value: 0.96 [0.90;1.01]; negative predictive value: 0.98 [0.97;1.00]). No analytical interference was observed.

Conclusion: The Technofluor® assay on the CEVERON® s100 is a fast, reliable method for measuring ADAMTS13 activity, proving effectiveness for diagnosis and monitoring of TTP patients. However, it remains crucial to interpret ADAMTS13 activity measurement with clinical context to ensure accurate diagnosis and management.

Joly BS, Deniau H, Doinel C, Brouillard A, Siguret V, Coppo P, Veyradier A. Real-world evaluation of a novel automated method for measuring ADAMTS13 activity using the Ceveron® s100 analyzer. J Thromb Haemost. 2025 Mar 7:S1538-7836(25)00137-0.

Valoración del anticuerpo

| TTP 2019: State of the Art |

Clinical and laboratory diagnosis of TTP: an integrated approach

Thita Chiasakul[1] and Adam Cuker[2]

[1]Division of Hematology, Department of Medicine, Faculty of Medicine, Chulalongkorn University and King Chulalongkorn Memorial Hospital, Thai Red Cross Society, Bangkok, Thailand; and [2]Department of Medicine and Department of Pathology and Laboratory Medicine, Perelman School of Medicine, University of Pennsylvania, Philadelphia, PA

Regular Article blood

CLINICAL TRIALS AND OBSERVATIONS

Presenting ADAMTS13 antibody and antigen levels predict prognosis in immune-mediated thrombotic thrombocytopenic purpura

Ferras Alwan,[1] Chiara Vendramin,[2] Karen Vanhoorelbeke,[3] Katy Langley,[2] Vickie McDonald,[4] Steve Austin,[5] Amanda Clark,[6] William Lester,[7] Richard Gooding,[8] Tina Biss,[9] Tina Dutt,[10] Nichola Cooper,[11] Oliver Chapman,[12] Tanya Cranfield,[13] Kenny Douglas,[14] H. G. Watson,[15] J. J. van Veen,[16] Keith Sibson,[17] William Thomas,[18] Lynn Manson,[19] Quentin A. Hill,[20] Sylvia Benjamin,[21] Debra Ellis,[1] John-Paul Westwood,[1] Mari Thomas,[1,22] and Marie Scully[1,22]

- **El diagnóstico de PTTa se realiza mediante la determinación de la actividad ADAMTS13 y presencia de inihibidor.**
- **La presencia/ausencia del inhibidor de ADAMTS13 puede ser un marcador clave en el diagnóstico de la PTTa**

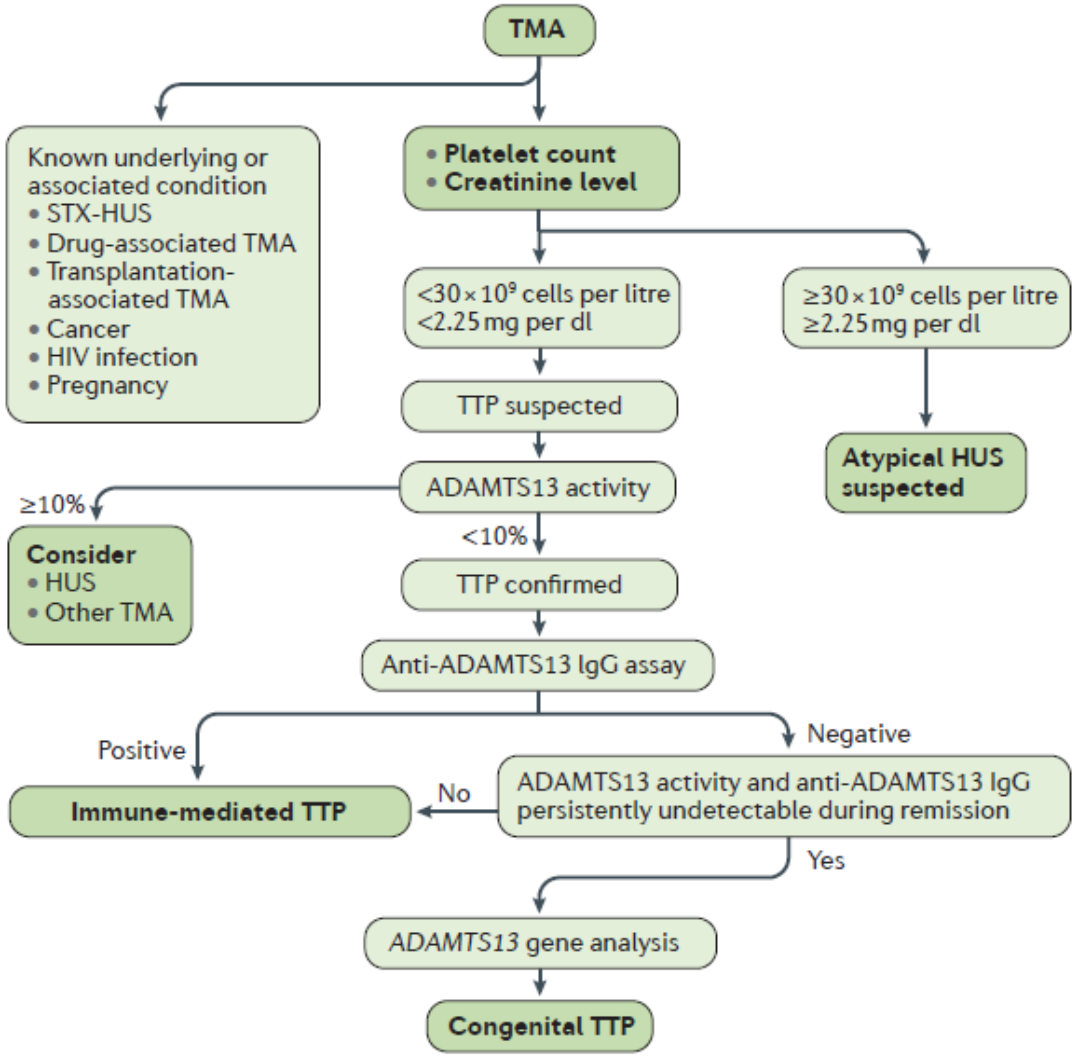
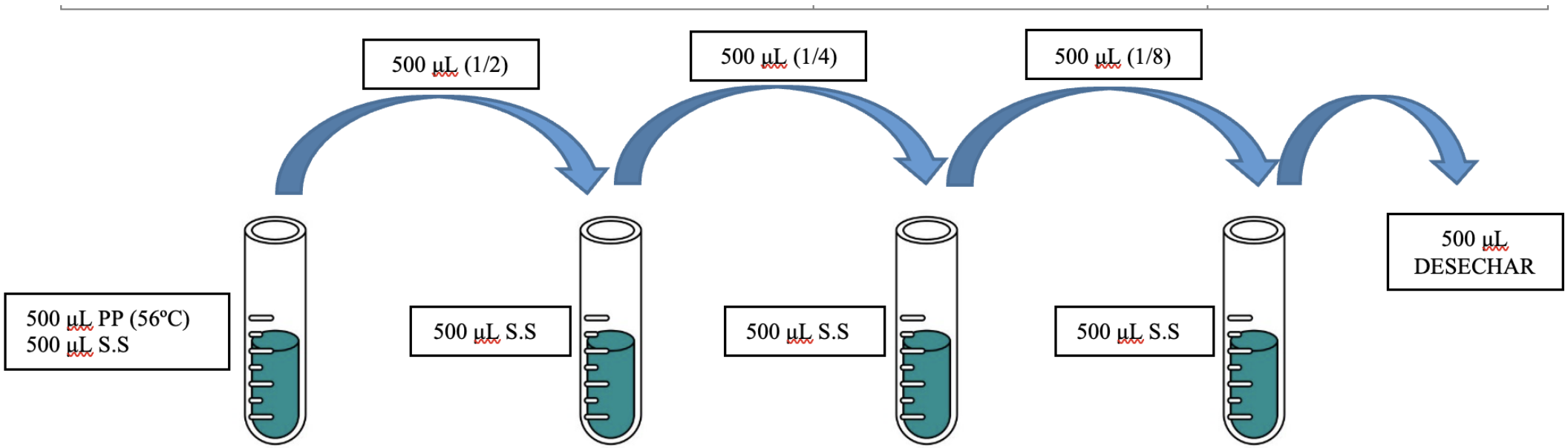


Figure 4 | **Diagnostic algorithm and likelihood of TTP.** Diagnostic flowchart in a

Kremer Hovinga et al. 2017. Nature reviews, Disease primers.

Determinación de Inhibidor



Actividad residual de ADAMTS13= (ADAMTS13 en PP+PC / ADAMTS13 en la muestra control 1/2) x 100%.

ADAMTS13 TUBO 4 ← 100 %
ADAMTS13 TUBO 5-9 → X?

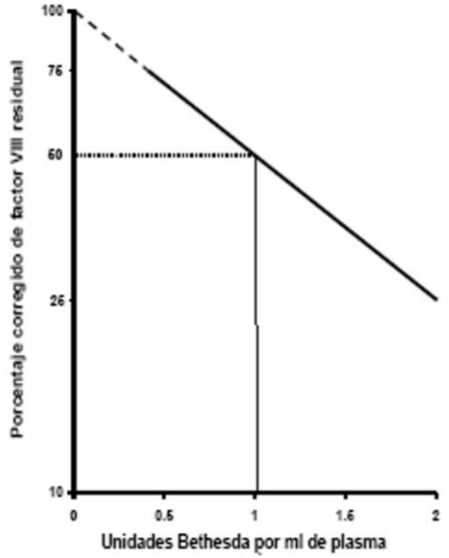
X= Actividad residual de ADAMTS13

Vendramin C, Thomas M, Westwood JP, Scully M. Bethesda Assay for Detecting Inhibitory Anti-ADAMTS13 Antibodies in Immune-Mediated Thrombotic Thrombocytopenic Purpura. *TH Open*. 2018;2(3):e329-e333.

Cálculo de las unidades Bethesda: de acuerdo con la definición, se grafica en escala semilogarítmica, la recta teórica de ADAMTS13 residual (%) vs potencia del inhibidor, considerando los siguientes puntos: el 100% de ADAMTS13 residual corresponde a 0 unidades Bethesda y 50% de ADAMTS13 residual a 1 UB/mL (Fig. 1). Debemos extrapolar el título del inhibidor de la curva teórica y multiplicar por la inversa de la dilución del plasma del paciente, con el cual se logró ese valor de ADAMTS13 residual.

Cálculo para las Unidades Bethesda

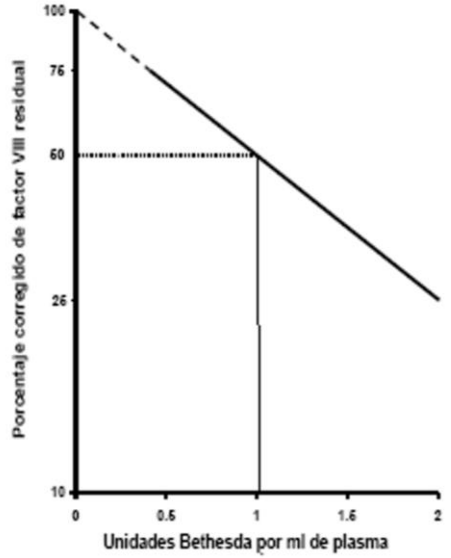
UB = (100 – Actividad residual de ADAMTS13) / 50



gura 1. GRAFICA DE REFERENCIA PARA LA CUANTIFICACIÓN DE INHIBIDORES. METODO BETHESDA

Vendramin C, Thomas M, Westwood JP, Scully M. Bethesda Assay for Detecting Inhibitory Anti-ADAMTS13 Antibodies in Immune-Mediated Thrombotic Thrombocytopenic Purpura. *TH Open*. 2018;2(3):e329-e333.

Cálculo de las unidades Bethesda: de acuerdo con la definición, se grafica en escala semilogarítmica, la recta teórica de ADAMTS13 residual (%) vs potencia del inhibidor, considerando los siguientes puntos: el 100% de ADAMTS13 residual corresponde a 0 unidades Bethesda y 50% de ADAMTS13 residual a 1 UB/mL (Fig. 1). Debemos extrapolar el título del inhibidor de la curva teórica y multiplicar por la inversa de la dilución del plasma del paciente, con el cual se logró ese valor de ADAMTS13 residual.



GRAFICA DE REFERENCIA PARA LA CUANTIFICACIÓN DE INHIBIDORES. METODO BETHESDA

Rango de referencia

La fuerza del efecto inhibitorio corresponde al número de UB; entre mayor sea el número habrá más inhibidores. Como lo recomienda la Sociedad Internacional sobre Trombosis y Hemostasia (ISTH), el valor de corte de lo que constituye la presencia de inhibidores se define como un título ≥ 0.6 UB, usando la modificación Nijmegen de la prueba Bethesda, documentado en 2 ocasiones separadas, generalmente dentro de un periodo de 4 semanas. La prueba Bethesda diferencia inhibidores de título bajo e inhibidores de título alto; los primeros generalmente se definen como inhibidores con un título < 5 BU, mientras que los segundos se definen como inhibidores con un título ≥ 5 BU. Por definición, la prueba Bethesda no detectará anticuerpos no neutralizantes No obstante, los anticuerpos frente a ADAMTS13, tanto neutralizantes como no neutralizantes, pueden detectarse mediante un ensayo por inmunoadsorción ligado a enzimas (ELISA) o un inmunoensayo de fluorescencia.

Vendramin C, Thomas M, Westwood JP, Scully M. Bethesda Assay for Detecting Inhibitory Anti-ADAMTS13 Antibodies in Immune-Mediated Thrombotic Thrombocytopenic Purpura. *TH Open*. 2018;2(3):e329-e333.

Valoración del anticuerpo

331. PATHOPHYSIOLOGY OF THROMBOSIS | NOVEMBER 29, 2018

Adamts-13 Inhibitor Testing Is Often Negative on Initial Testing

Hina Chaudhry, BSc, Michelle Sholzberg, MDM, MSc, FRCPC, Katerina Pavenski, MD

Check for updates

Blood (2018) 132 (Supplement 1): 5051.

https://doi.org/10.1182/blood-2018-99-119399

311. DISORDERS OF PLATELET NUMBER OR FUNCTION: POSTER III | DECEMBER 7, 2017

The Impact of Detectable ADAMTS13 Inhibitor on the Clinical Presentation and Outcome of Patients with Immune Thrombotic Thrombocytopenic Purpura (iTTP): Analysis Using the United States Thrombotic Microangiopathy (USTMA) Clinical Registry

Meera Sridharan, MD , Ana G Antun, MD , Lisa Baumann Kreuziger, Assistant Professor. MD MS , Spero R Cataland, MD , Shruti Chaturvedi, MBBS,MS , Todd Clover, MD , Elizabeth Davis , Andrew Johnson, MD , Keith McCrae, MD , Ming Yeong Lim, MBBChir , J. Evan Sadler, MD PhD , Marshall A. Mazepa, MD , Ronald S Go, MD

Check for updates

- **Existe un porcentaje de pacientes con PTTa y niveles bajos de ADAMTS13 con ausencia de inhibidor (hasta un 20% de pacientes)**

Chaudhry H, et al. 2018. Blood (2018) 132 (Supplement 1): 5051. https://ashpublications.org/blood/article/132/Supplement%201/5051/265664/Adamts-13-Inhibitor-Testing-Is-Often-Negative-on Último acceso octubre 2021
Sridharan M, et al. 2017. Blood. (130) Supplement 1, 3620

Configuración Abierta/Cerrada

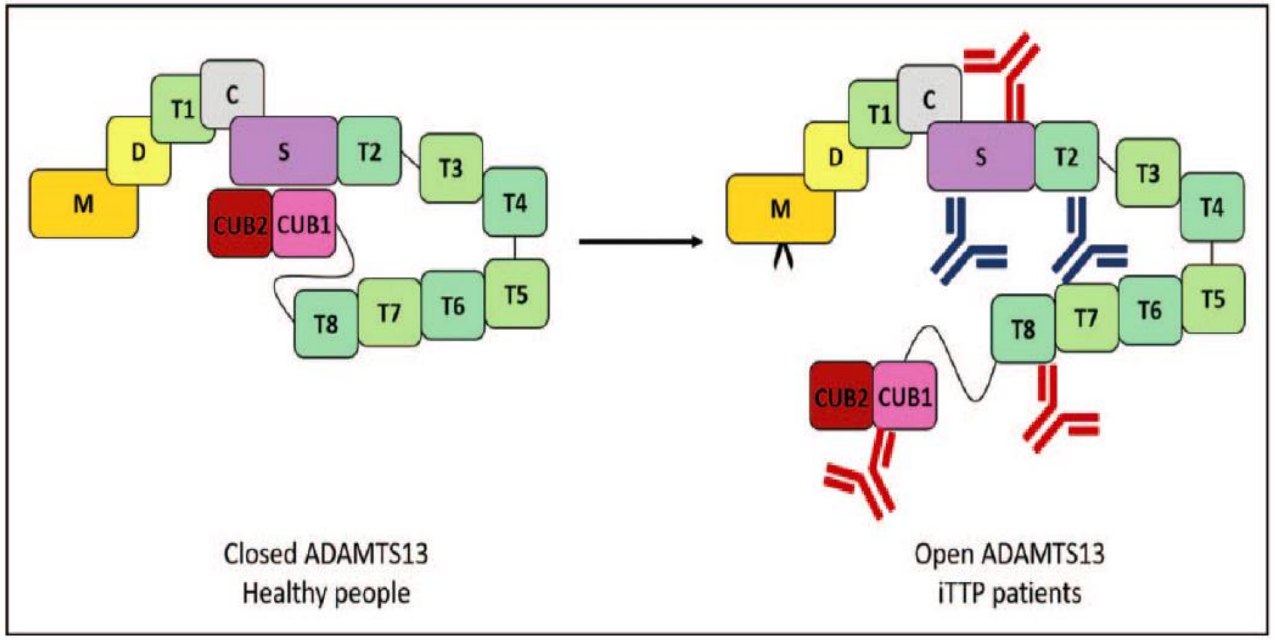


FIGURE 2. ADAMTS13 adopts a closed conformation in healthy people, while iTTP patients have an open ADAMTS13 conformation induced by anti-ADAMTS13 autoantibodies.

Cuando el auto-anticuerpo se liga al ADAMTS13 también adquiere una configuración abierta.

Roose E, et al. Insights into ADAMTS13 structure: impact on thrombotic thrombocytopenic purpura diagnosis and management. Curr Opin Hematol. 2020 Sep;27(5):320-326

Configuración Abierta/Cerrada

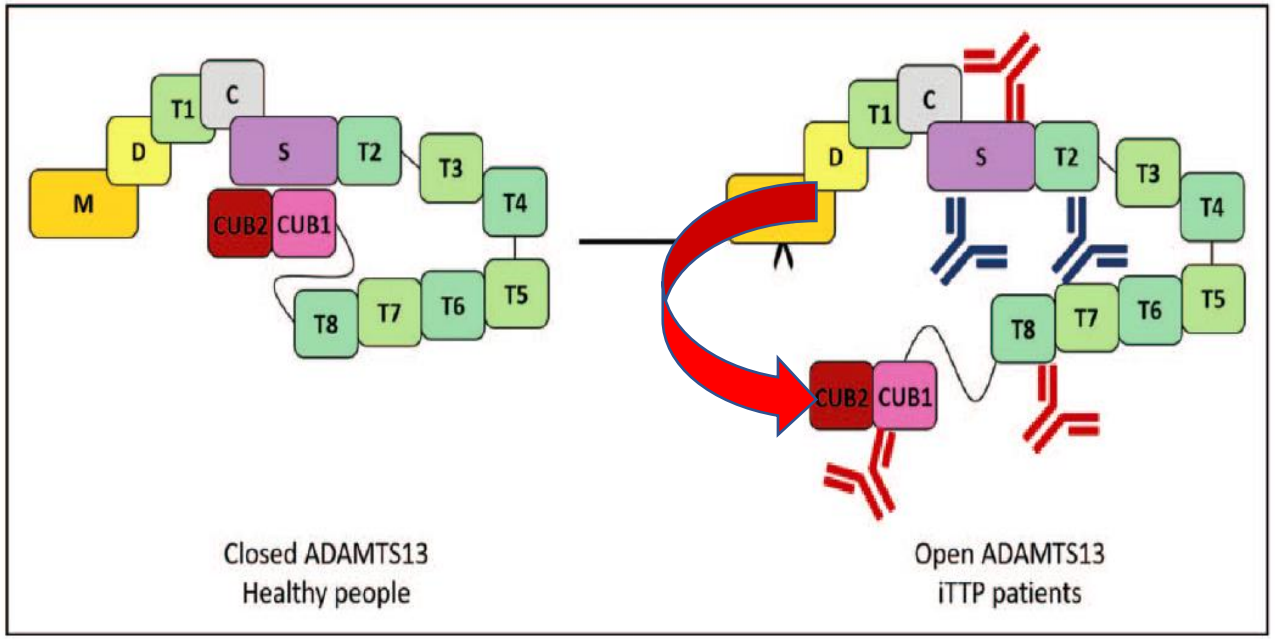


FIGURE 2. ADAMTS13 adopts a closed conformation in healthy people, while iTTP patients have an open ADAMTS13 conformation induced by anti-ADAMTS13 autoantibodies.

Cuando el auto-anticuerpo se liga al ADAMTS13 también adquiere una configuración abierta.

Roose E, et al. Insights into ADAMTS13 structure: impact on thrombotic thrombocytopenic purpura diagnosis and management. Curr Opin Hematol. 2020 Sep;27(5):320-326

blood®
Regular Article

THROMBOSIS AND HEMOSTASIS

Open ADAMTS13, induced by antibodies, is a biomarker for subclinical immune-mediated thrombotic thrombocytopenic purpura

Elien Roose,[1,*] An-Sofie Schelpe,[1,*] Edwige Tellier,[2] György Sinkovits,[3] Bérangère S. Joly,[4] Charlotte Dekimpe,[1] Gilles Kaplanski,[2,5] Maelle Le Besnerais,[6,7] Ilaria Mancini,[8] Tanja Falter,[9,10] Charis Von Auer,[10,11] Hendrik B. Feys,[12,13] Marienn Reti,[14] Heidi Rossmann,[9,10] Aline Vandenbulcke,[1] Inge Pareyn,[1] Jan Voorberg,[15] Andreas Greinacher,[16] Ygal Benhamou,[6,7] Hans Deckmyn,[1] Rob Fijnheer,[17] Zoltan Prohászka,[3] Flora Peyvandi,[8] Bernhard Lämmle,[10,18,19] Paul Coppo,[20] Simon F. De Meyer,[1] Agnès Veyradier,[4] and Karen Vanhoorelbeke[1]

Journal of Thrombosis and Haemostasis, 16: 378–388 DOI: 10.1111/jth.13922

ORIGINAL ARTICLE

An open conformation of ADAMTS-13 is a hallmark of acute acquired thrombotic thrombocytopenic purpura

E. ROOSE,* A. S. SCHELPE,* B. S. JOLY,† M. PEETERMANS,‡ P. VERHAMME,‡ J. VOORBERG,§ A. GREINACHER,¶ H. DECKMYN,* S. F. DE MEYER,* P. COPPO,** A. VEYRADIER† and K. VANHOORELBEKE*

**Laboratory for Thrombosis Research, IRF Life Sciences, KU Leuven Campus Kulak Kortrijk, Kortrijk, Belgium; †Service d'Hématologie biologique, Hôpital Lariboisière, Assistance Publique-Hôpitaux de Paris and EA3518, Institut Universitaire d'Hématologie, Hôpital Saint Louis, Université Paris Diderot, Paris, France; ‡Center for Molecular and Vascular Biology, Department of Cardiovascular Sciences, University of Leuven, Leuven, Belgium; §Department of Plasma Proteins, Sanquin-Academic Medical Center Landsteiner Laboratory, Amsterdam, the Netherlands; ¶Institute for Immunology and Transfusion Medicine, University Medical Center, Greifswald, Germany; and **Département d'hématologie clinique, Hôpital Saint Antoine, AP-HP and Université Pierre et Marie Curie, Paris, France*

Roose E, Schelpe A-S, Joly BS, et al. An open conformation of ADAMTS-13 is a hallmark of acute acquired thrombotic thrombocytopenic purpura. J Thromb Haemost 2018; 16:378–388.
Roose E, Schelpe A-S, Tellier E, et al. Open ADAMTS13, induced by antibodies, is a biomarker for subclinical immune-mediated thrombotic thrombocytopenic purpura. Blood 2020; 2019004221. doi: https://doi.org/10.1182/blood.2019004221

blood®
Regular Article

THROMBOSIS AND HEMOSTASIS

Open ADAMTS13, induced by antibodies, is a biomarker for subclinical immune-mediated thrombotic thrombocytopenic purpura

Elien Roose,[1,*] An-Sofie Schelpe,[1,*] Edwige Tellier,[2] György Sinkovits,[3] Bérangère S. Joly,[4] Charlotte Dekimpe,[1] Gilles Kaplanski,[2,5] Maelle Le Besnerais,[6,7] Ilaria Mancini,[8] Tanja Falter,[9,10] Charis Von Auer,[10,11] Hendrik B. Feys,[12,13] Marienn Reti,[14] Heidi Rossmann,[9,10] Aline Vandenbulcke,[1] Inge Pareyn,[1] Jan Voorberg,[15] Andreas Greinacher,[16] Ygal Benhamou,[6,7] Hans Deckmyn,[1] Rob Fijnheer,[17] Zoltan Prohászka,[3] Flora Peyvandi,[8] Bernhard Lämmle,[10,18,19] Paul Coppo,[20] Simon F. De Meyer,[1] Agnès Veyradier,[4] and Karen Vanhoorelbeke[1]

Journal of Thrombosis and Haemostasis, 16: 378–388 DOI: 10.1111/jth.13922

ORIGINAL ARTICLE

An open conformation of ADAMTS-13 is a hallmark of acute acquired thrombotic thrombocytopenic purpura

E. ROOSE,* A. S. SCHELPE,* B. S. JOLY,† M. PEETERMANS,‡ P. VERHAMME,‡ J. VOORBERG,§ A. GREINACHER,¶ H. DECKMYN,* S. F. DE MEYER,* P. COPPO,** A. VEYRADIER† and K. VANHOORELBEKE*

**Laboratory for Thrombosis Research, IRF Life Sciences, KU Leuven Campus Kulak Kortrijk, Kortrijk, Belgium; †Service d'Hématologie biologique, Hôpital Lariboisière, Assistance Publique-Hôpitaux de Paris and EA3518, Institut Universitaire d'Hématologie, Hôpital Saint Louis, Université Paris Diderot, Paris, France; ‡Center for Molecular and Vascular Biology, Department of Cardiovascular Sciences, University of Leuven, Leuven, Belgium; §Department of Plasma Proteins, Sanquin-Academic Medical Center Landsteiner Laboratory, Amsterdam, the Netherlands; ¶Institute for Immunology and Transfusion Medicine, University Medical Center, Greifswald, Germany; and **Département d'hématologie clinique, Hôpital Saint Antoine, AP-HP and Université Pierre et Marie Curie, Paris, France*

Roose E, Schelpe A-S, Joly BS, et al. An open conformation of ADAMTS-13 is a hallmark of acute acquired thrombotic thrombocytopenic purpura. J Thromb Haemost 2018; 16:378–388.
Roose E, Schelpe A-S, Tellier E, et al. Open ADAMTS13, induced by antibodies, is a biomarker for subclinical immune-mediated thrombotic thrombocytopenic purpura. Blood 2020; 2019004221. doi: https://doi.org/10.1182/blood.2019004221

blood®
Regular Article

THROMBOSIS AND HEMOSTASIS

Open ADAMTS13, induced by antibodies, is a biomarker for subclinical immune-mediated thrombotic thrombocytopenic purpura

Elien Roose,[1,*] An-Sofie Schelpe,[1,*] Edwige Tellier,[2] György Sinkovits,[3] Bérangère S. Joly,[4] Charlotte Dekimpe,[1] Gilles Kaplanski,[2,5] Maelle Le Besnerais,[6,7] Ilaria Mancini,[8] Tanja Falter,[9,10] Charis Von Auer,[10,11] Hendrik B. Feys,[12,13] Marienn Reti,[14] Heidi Rossmann,[9,10] Aline Vandenbulcke,[1] Inge Pareyn,[1] Jan Voorberg,[15] Andreas Greinacher,[16] Ygal Benhamou,[6,7] Hans Deckmyn,[1] Rob Fijnheer,[17] Zoltan Prohászka,[3] Flora Peyvandi,[8] Bernhard Lämmle,[10,18,19] Paul Coppo,[20] Simon F. De Meyer,[1] Agnès Veyradier,[4] and Karen Vanhoorelbeke[1]

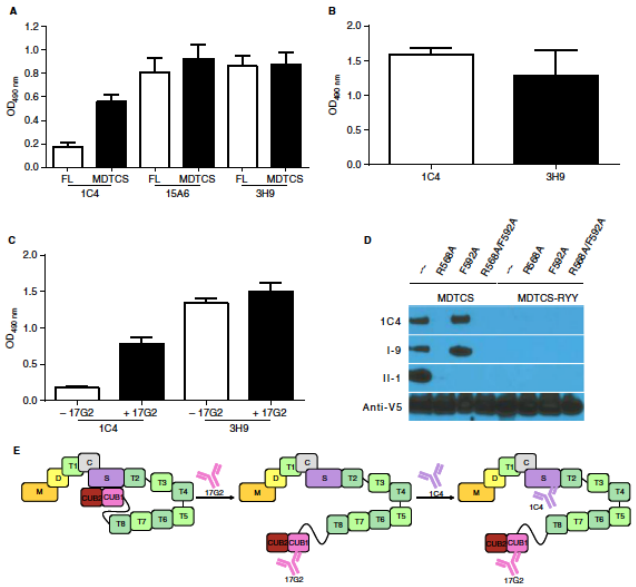


Fig. 1. The epitope of the anti-ADAMTS-13 antibody 1C4 is cryptic. (A) The anti-MDTCS antibodies (1C4, 15A6, and 3H9) were coated (5 μg mL^{-1}), and 5.7 nM full-length (FL) ADAMTS-13 or MDTCS was added. Captured ADAMTS-13 or MDTCS was detected with horseradish peroxidase (HRP)-labeled anti-V5 antibody ($n = 3$). Antibody 1C4 recognizes a cryptic epitope in FL ADAMTS-13, as 1C4 can only capture MDTCS. (B) ADAMTS-13 (15 nM) was coated, and 1C4 and 3H9 (5 μg mL^{-1}) were added. Bound antibody was detected by HRP-labeled goat anti-mouse antibody. Coating of ADAMTS-13 induces a conformational change and exposes the cryptic epitope of 1C4. (C) Antibodies 1C4 and 3H9 (5 μg mL^{-1}) were coated, and ADAMTS-13 (0.5 μg mL^{-1}), preincubated without (–) or with (+) the anti-CUB1 antibody 17G2 (2.5 μg mL^{-1}), was added. Captured ADAMTS-13 was detected with HRP-labeled anti-V5 antibody. The anti-CUB1 antibody 17G2 induces a conformational change in ADAMTS-13, and exposes the cryptic epitope of 1C4. (D) Immunoprecipitation of MDTCS alanine mutants (MDTCS, MDTCS-R568A, MDTCS-F592A, MDTCS-R568/F592A, MDTCS-R660A/Y661A/Y665A [MDTCS-RYY], MDTCS-RYY-R568A, MDTCS-RYY-F592A, and MDTCS-RYY-R568A/F592A) with antibody 1C4 and patient-derived antibodies I-9 and II-1. Anti-V5 antibody was used as positive control. Samples were analyzed with SDS-PAGE and western blotting. HRP-labeled anti-V5 antibody was used for detection. (E) Representation of folded ADAMTS-13 (left), in which the spacer and CUB domains interact. Addition of the anti-CUB1 antibody 17G2 changes ADAMTS-13 towards a more open conformation (center), which exposes a cryptic epitope in the spacer domain to which 1C4 can bind (right). [Color figure can be viewed at wileyonlinelibrary.com]

Journal of Thrombosis and Haemostasis, 16: 378–388 DOI: 10.1111/jth.13922

ORIGINAL ARTICLE

An open conformation of ADAMTS-13 is a hallmark of acute acquired thrombotic thrombocytopenic purpura

E. ROOSE,* A. S. SCHELPE,* B. S. JOLY,† M. PEETERMANS,‡ P. VERHAMME,‡ J. VOORBERG,§ A. GREINACHER,¶ H. DECKMYN,* S. F. DE MEYER,* P. COPPO,** A. VEYRADIER† and K. VANHOORELBEKE*

*Laboratory for Thrombosis Research, IRF Life Sciences, KU Leuven Campus Kulak Kortrijk, Kortrijk, Belgium; †Service d'Hématologie biologique, Hôpital Lariboisière, Assistance Publique-Hôpitaux de Paris and EA3518, Institut Universitaire d'Hématologie, Hôpital Saint Louis, Université Paris Diderot, Paris, France; ‡Center for Molecular and Vascular Biology, Department of Cardiovascular Sciences, University of Leuven, Leuven, Belgium; §Department of Plasma Proteins, Sanquin-Academic Medical Center Landsteiner Laboratory, Amsterdam, the Netherlands; ¶Institute for Immunology and Transfusion Medicine, University Medical Center, Greifswald, Germany; and **Département d'hématologie clinique, Hôpital Saint Antoine, AP-HP and Université Pierre et Marie Curie, Paris, France

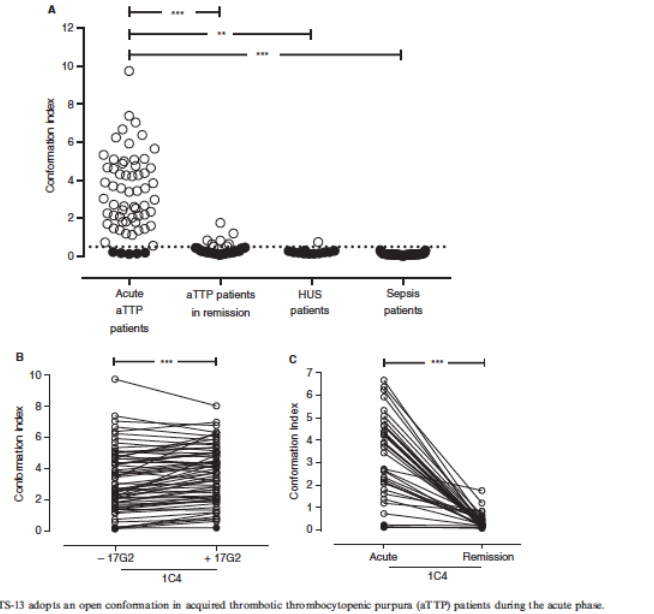


Fig. 4. ADAMTS-13 adopts an open conformation in acquired thrombotic thrombocytopenic purpura (aTTP) patients during the acute phase. (A) Plasma samples of acute aTTP ($n = 63$) patients, aTTP patients in remission ($n = 36$), sepsis ($n = 63$) patients and hemolytic-uremic syndrome (HUS) ($n = 12$) patients were added to wells coated with the anti-ADAMTS-13 antibody 1C4 (recognizing a cryptic epitope in ADAMTS-13), and captured ADAMTS-13 was detected with biotinylated 3H9 and horseradish peroxidase-labeled streptavidin. If ADAMTS-13 was not captured by 1C4, the conformation of ADAMTS-13 was folded (●, conformation index of < 0.5). If ADAMTS-13 was captured by 1C4, the conformation of ADAMTS-13 was open (○, conformation index of > 0.5). ***$P < 0.0001$, **$P < 0.01$, one-way ANOVA. (B) The same plasma samples as in (A) of acute aTTP patients were now preincubated with the anti-CUB1 antibody 17G2 (+ 17G2) (inducing an [additional] open conformation in ADAMTS-13) before addition to the wells coated with 1C4. ***$P < 0.0001$, paired t-test. (C) The link between the conformation of ADAMTS-13 in matched plasma samples of acute aTTP patients and aTTP patients in remission is represented. ***$P < 0.0001$, paired t-test.

Roose E, Schelpe A-S, Joly BS, et al. An open conformation of ADAMTS-13 is a hallmark of acute acquired thrombotic thrombocytopenic purpura. J Thromb Haemost 2018; 16:378–388.
Roose E, Schelpe A-S, Tellier E, et al. Open ADAMTS13, induced by antibodies, is a biomarker for subclinical immune-mediated thrombotic thrombocytopenic purpura. Blood 2020; 2019004221. doi: https://doi.org/10.1182/blood.2019004221

Configuración del ADAMTS13 en la PTTi

- Vanhoorelbeke et al ha desarrollado un TEST-ELISA que puede discriminar entre la configuración cerrada y abierta de ADAMTS13, **utilizando un anticuerpo (1C4)**, que reconoce un epítopo críptico en el dominio dominio S de ADAMTS13, por lo que sólo la conformación abierta de ADAMTS13 abierta, pero no la cerrada, puede ser captada.
- Demuestran que ADAMTS13 circula normalmente en una conformación cerrada, mientras que en los pacientes adultos con iTTP adultos, ADAMTS13 circula en una configuración abierta (92%).

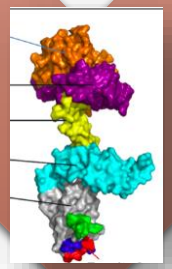
Concluyen:

1. El ADAMTS13 abierto es un marcador específico de iTTP, ya que en otras microangiopatías trombóticas u otras enfermedades en las que la actividad de ADAMTS13 está se reduce (sepsis), ADAMTS13 se encuentra en una conformación cerrada.
2. La determinación de la conformación de ADAMTS13 podría ayudar en el diagnóstico de iTTP .

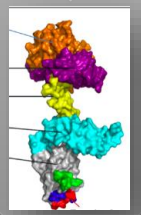
Roose E, Schelpe A-S, Joly BS, et al. An open conformation of ADAMTS-13 is a hallmark of acute acquired thrombotic thrombocytopenic purpura. J Thromb Haemost 2018; 16:378–388.

Impacto en el seguimiento de la determinación configuración ADAMTS13

- En la remisión completa (>50 UI/dl de actividad ADAMTS13) la conformación de ADAMTS13 se volvió a cerrar con frecuencia (62-78%), lo que indica que el factor que abre ADAMTS13 desapareció .
- Los pacientes con iTTP en remisión con una actividad de ADAMTS13 disminuida (<50%) casi todos tenían una conformación abierta de ADAMTS13 abierta, lo que demuestra que la ADAMTS13 abierta es un biomarcador de enfermedad subclínica.



- Si esta la configuración abierta del ADAMTS13 precede a una bajada de actividad ADAMTS13 se sugirió en un estudio de seguimiento a largo plazo, pero se necesitan más estudios para confirmar estos resultados.



- La configuración abierta del ADAMTS13 se debe a la presencia de anticuerpos murinos anti-ADAMTS13 que pueden inducir cambios conformacionales en ADAMTS13.

Roose E, Schelpe A-S, Joly BS, et al. An open conformation of ADAMTS-13 is a hallmark of acute acquired thrombotic thrombocytopenic purpura. J Thromb Haemost 2018; 16:378–388. Roose E, Schelpe A-S, Tellier E, et al. Open ADAMTS13, induced by antibodies, is a biomarker for subclinical immune-mediated thrombotic thrombocytopenic purpura. Blood 2020; 2019004221 doi: https://doi.org/10.1182/blood.2019004221.

Estrategia de diagnóstico, tratamiento y seguimiento actual y futuro

Table 1. Overview of the current strategy to diagnose, treat and follow TTP patients and how the novel biomarker open ADAMTS13 might become useful in those steps in the future

	Diagnosis	Treatment	Follow-up
Current	ADAMTS13 activity Anti-ADAMTS13 autoantibodies *ADAMTS13* sequencing	PEX or recombinant ADAMTS13 Rituximab Caplacizumab → Monitoring through ADAMTS13 activity	ADAMTS13 activity Preemptive rituximab
Future?	Open ADAMTS13 to diagnose iTTP in cases where anti-ADAMTS13 autoantibodies are undetectable?	Open ADAMTS13 to follow treatment response in iTTP patients?	Open ADAMTS13 as a predictor for relapse?

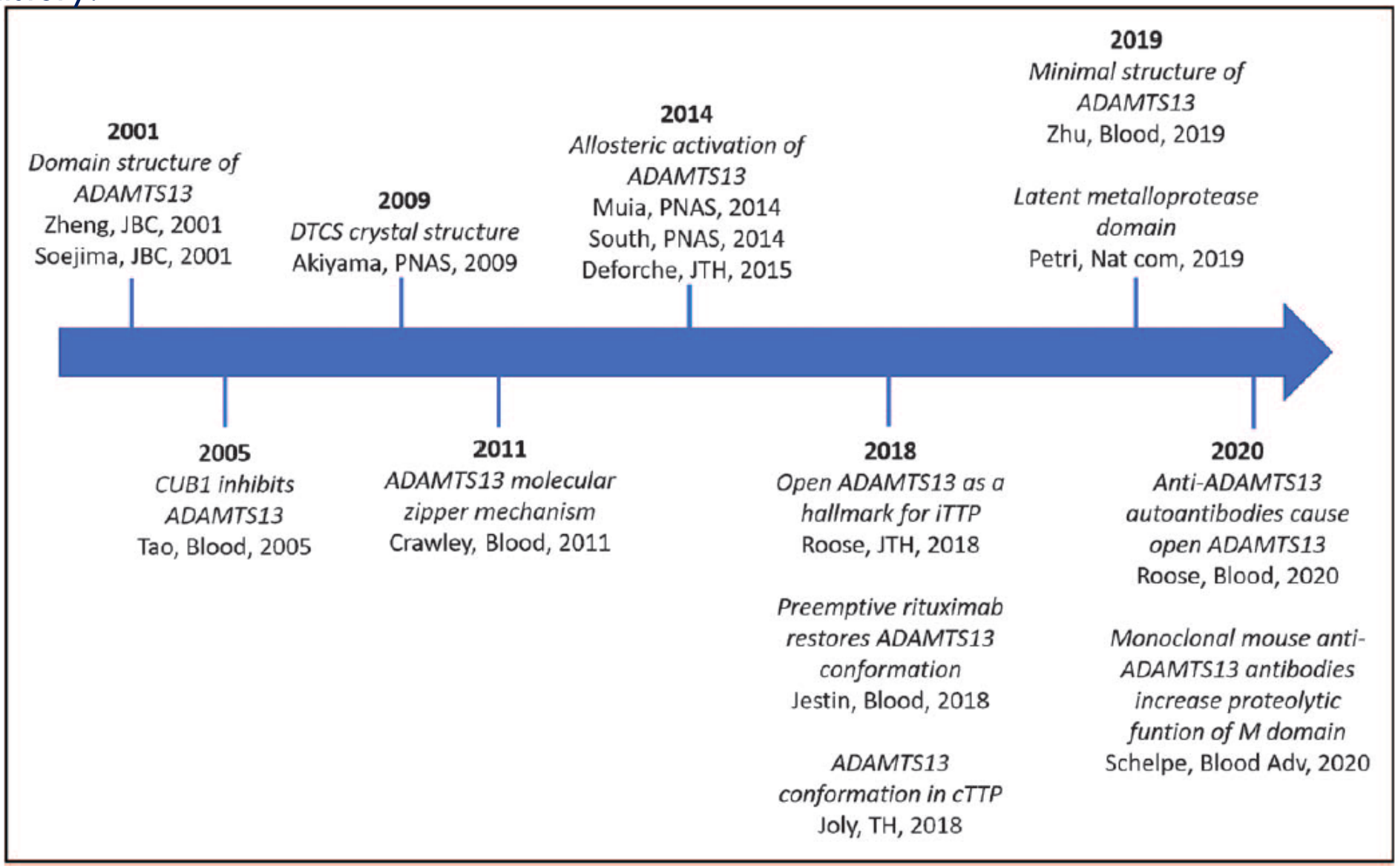
ADAMTS13, a disintegrin and metalloprotease with thrombospondin type one repeats, member 13; iTTP, immune-mediated thrombotic thrombocytopenic purpura; PEX, plasma exchange.

Roose E, Veyradier A, Vanhoorelbeke K. Insights into ADAMTS13 structure: impact on thrombotic thrombocytopenic purpura diagnosis and management. Curr Opin Hematol. 2020 Sep;27(5):320-326

Rituximab no está aprobado por ningún organismo internacional para el tratamiento de la PTTa

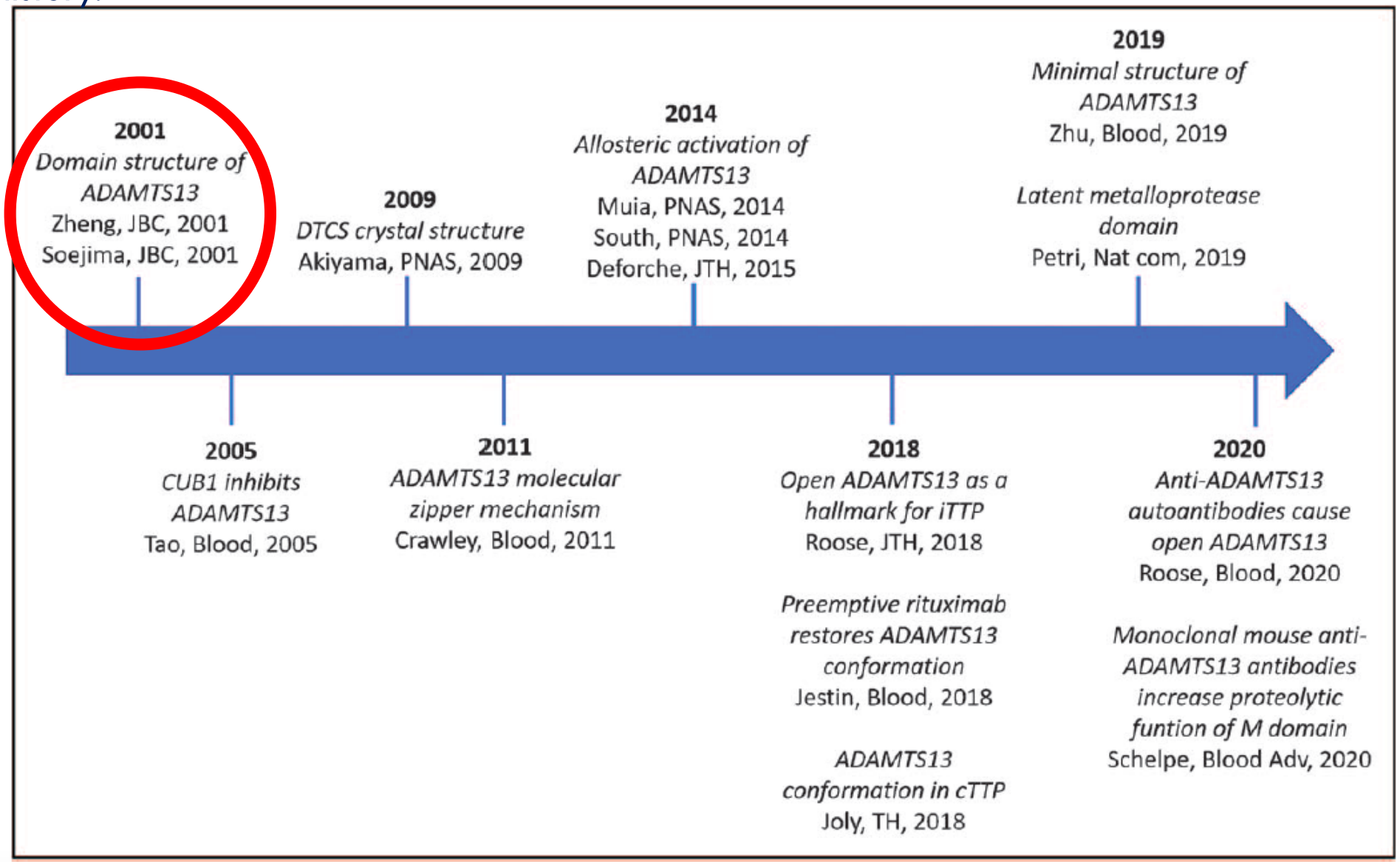
Recombinant ADAMTS13 is an investigational product in phase 2 of clinical development for TTP.[4]ClinicalTrials.gov. Available from: https://clinicaltrials.gov/ct2/show/NCT03922308. Accessed October 2021.

Overview of the most important discoveries on the ADAMTS13 structure and its role in the pathophysiology of TTP during history.



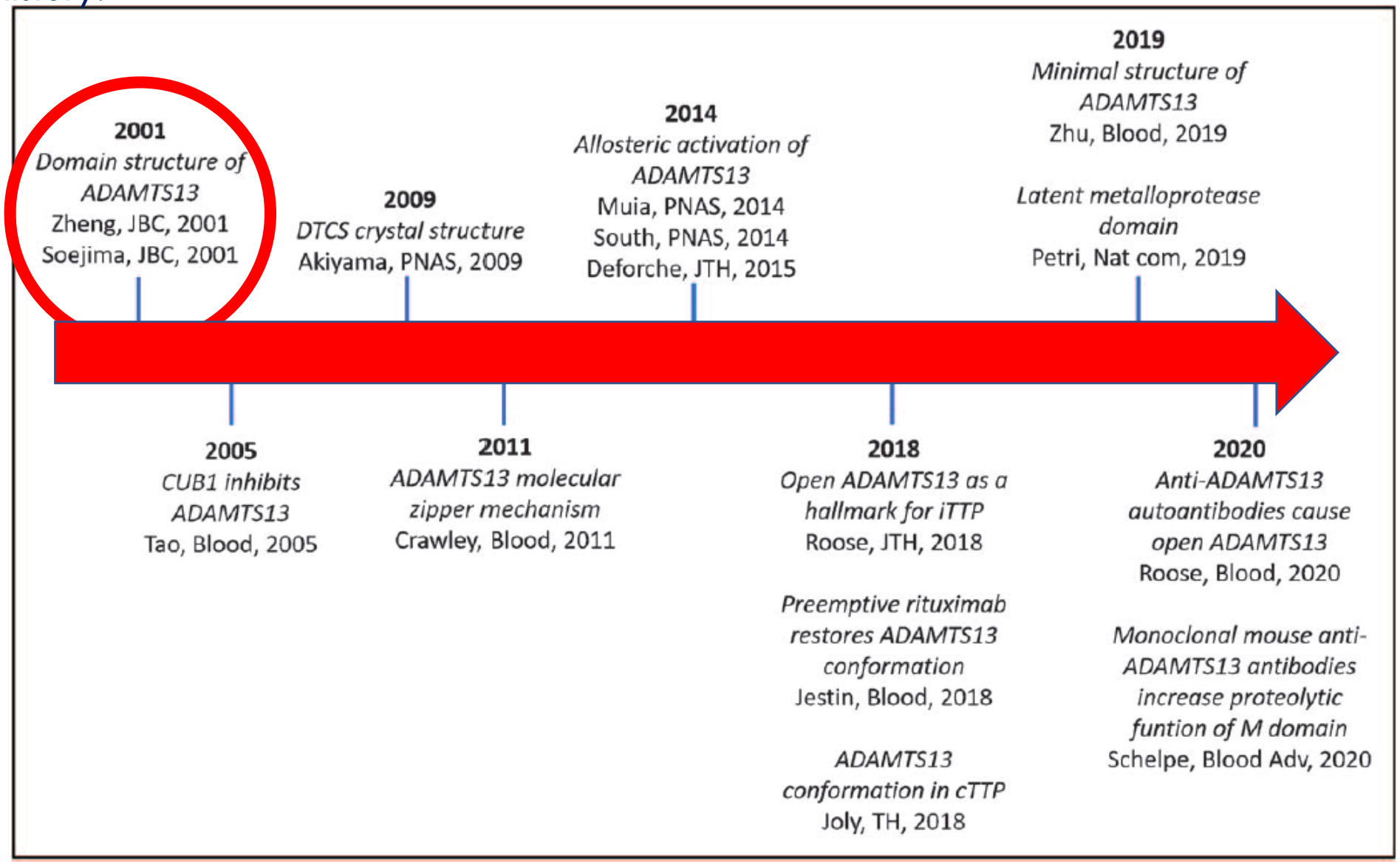
Roose E, Veyradier A, Vanhoorelbeke K. Insights into ADAMTS13 structure: impact on thrombotic thrombocytopenic purpura diagnosis and management. Curr Opin Hematol. 2020 Sep;27(5):320-326. d

Overview of the most important discoveries on the ADAMTS13 structure and its role in the pathophysiology of TTP during history.



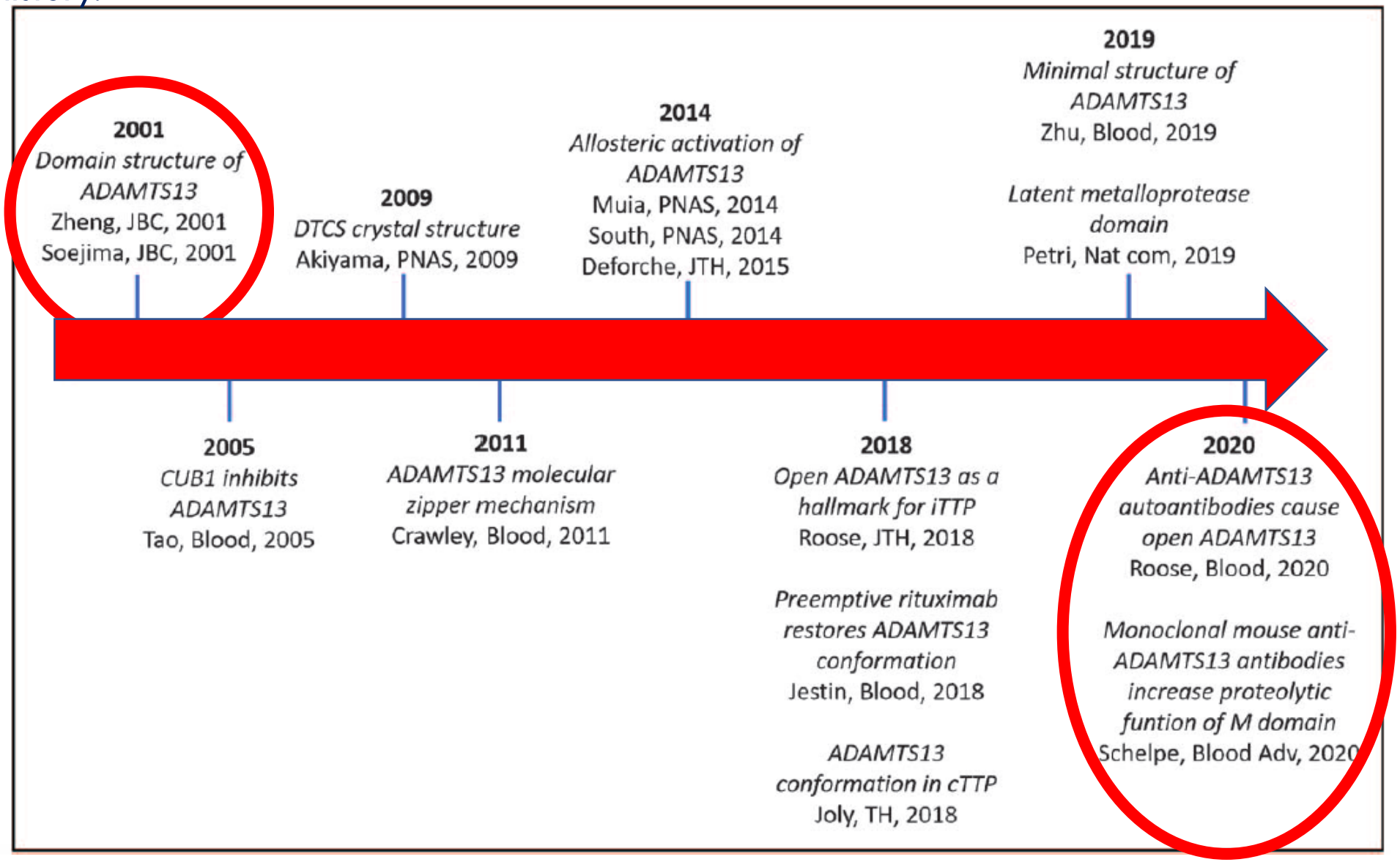
Roose E, Veyradier A, Vanhoorelbeke K. Insights into ADAMTS13 structure: impact on thrombotic thrombocytopenic purpura diagnosis and management. Curr Opin Hematol. 2020 Sep;27(5):320-326. d

Overview of the most important discoveries on the ADAMTS13 structure and its role in the pathophysiology of TTP during history.



Roose E, Veyradier A, Vanhoorelbeke K. Insights into ADAMTS13 structure: impact on thrombotic thrombocytopenic purpura diagnosis and management. Curr Opin Hematol. 2020 Sep;27(5):320-326. d

Overview of the most important discoveries on the ADAMTS13 structure and its role in the pathophysiology of TTP during history.



Roose E, Veyradier A, Vanhoorelbeke K. Insights into ADAMTS13 structure: impact on thrombotic thrombocytopenic purpura diagnosis and management. Curr Opin Hematol. 2020 Sep;27(5):320-326. d

Sid Hemolitico-urémico

Recommendations from COMDORA-SBN for diagnosis and treatment of aHUS

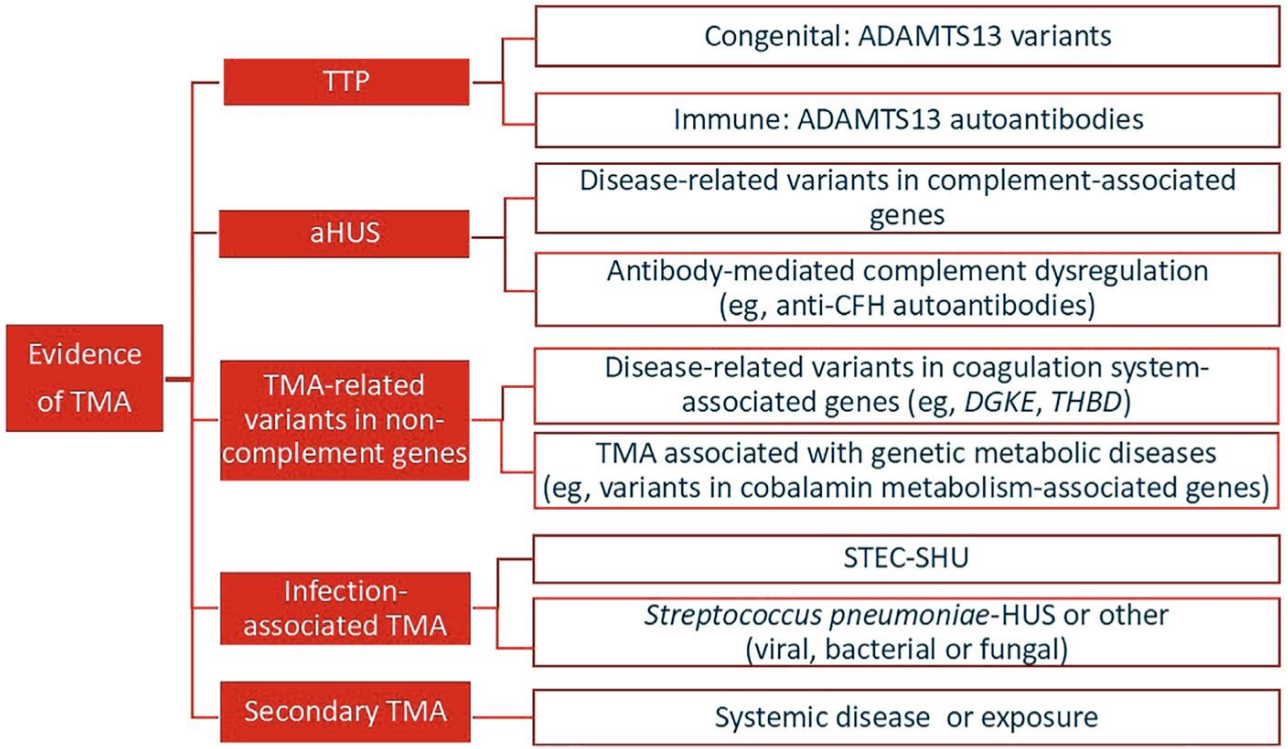


Figure 1. Practical classification of thrombotic microangiopathies. Modified from Genest et al, 2023[17].

Vaisbich MH et al. Recommendations for diagnosis and treatment of Atypical Hemolytic Uremic Syndrome (aHUS): an expert consensus statement from the Rare Diseases Committee of the Brazilian Society of Nephrology (COMDORA-SBN). J Bras Nefrol. 2025 Apr-Jun;47(2):e20240087.

CHART 2 RECOMMENDATIONS FOR DIAGNOSTIC TESTS

	Diagnostic tests
Confirm TMA	Biochemical evaluation
	Hematological exam: hemoglobin, thrombocytes, and reticulocytes
	Serum LDH
	Serum haptoglobin
	Peripheral blood smear (detect the presence of schistocytes)
	Indirect Coombs/direct Coombs
	PT/aPTT/fibrinogen
Complement testing and other tests/Etiology	**Detect STEC**
	Stool or rectal swab culture
	PCR for STEC virulence genes in stool
	Serology: serum (*E. coli*) and Yersinia antibodies (anti-LPS antibodies for prevalent serotypes)
	Detect ADAMTS-13 deficiency
	Von Willebrand protease activity
	Test underlying causes (secondary causes)
	Plasma homocysteine (increased levels are observed in cobalamin disturbances)
	HIV serology, pulmonary cultures, influenza
	ANA/anti dsDNA (Farr)/anti-centromere Ab/antiphospholipid antibodies (anticardiolipin IgG and IgM, anti B/lupus anticoagulant)
	Hemocultures
	Pregnancy testing
	Chest X-ray
	Factor H; factor I antibodies
	Serum CH50
	Serum MAC (C5b-9)
	Serum levels of C3, C4; index C3d/C3
	CD46 expression on leukocytes (poly- or mononuclear leukocytes using a FACS test)
	Blood levels of factor B, factor Bb, C3 convertase, factor H activity, antibodies for factor I, other complement factors and AP50
Genetic testing/ Etiology	Complement factor H (*CFH* gene)
	Complement factor I (*CFI* gene)
	Membrane cofactor protein (*MCP* gene)
	Complement factor B (*CFB* gene)
	Complement C3 (*C3* gene)
	Complement factor H-related proteins (*CFHR* genes)
	CFH-CFHR hybrid gene
	DGKE variants (children under 2 years old, especially if nephrotic syndrome is associated)
	Thrombomodulin (*THBD gene*)
	ADAMTS13 gene: if indicated (PTT)
	MMACHC gene: if indicated to exclude defect in cobalamin deficiency (especially patients under 18 years old)
	Other complement genes, if indicated

Abbreviations – TMA: thrombotic microangiopathy; LDH: lactate dehydrogenase; PT: prothrombin time; aPTT: activated partial thromboplastin time; STEC: Shiga toxin-producing Escherichia coli; PCR: polymerase chain reaction; ANA: antinuclear antibody; CH50: measuring the 50% hemolytic complement; MAC: membrane attack complex; AP50: alternative pathway hemolytic complement.

Sid Hemolitico-urémico

Recommendations from COMDORA-SBN for diagnosis and treatment of aHUS

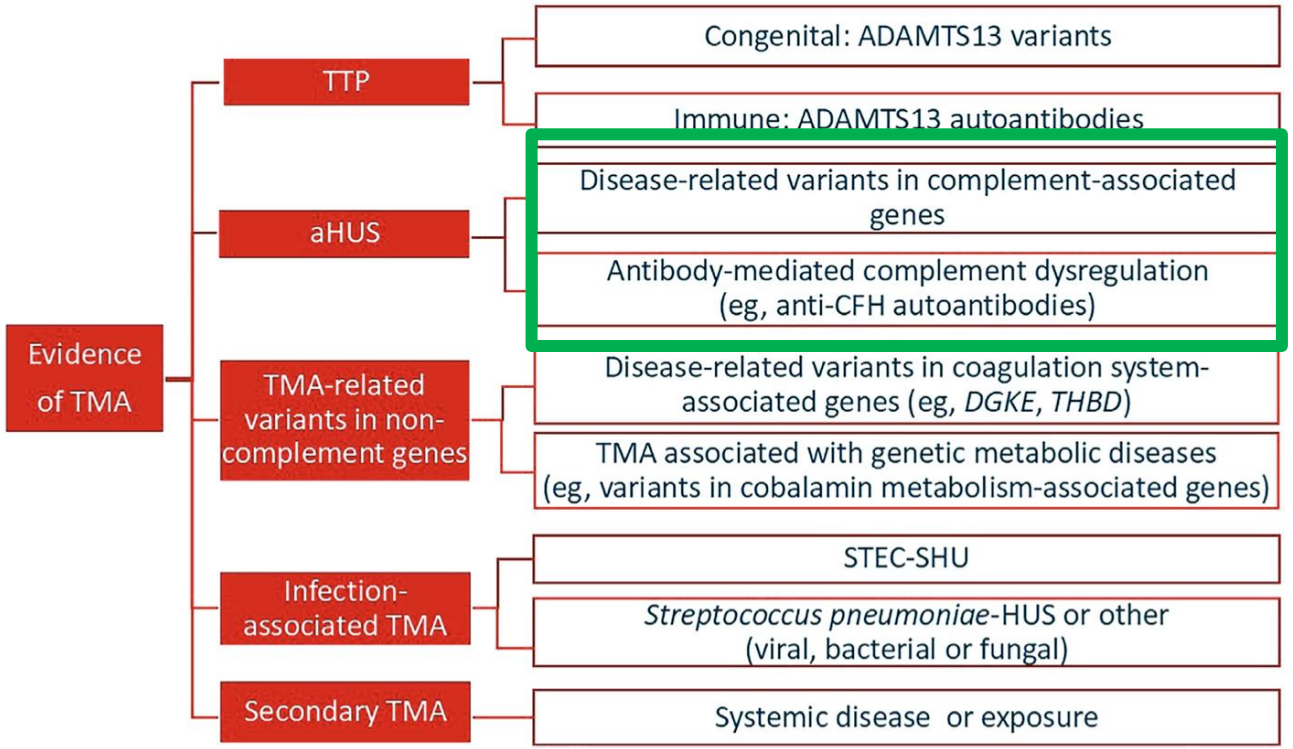


Figure 1. Practical classification of thrombotic microangiopathies. Modified from Genest et al, 2023[17].

Vaisbich MH et al. Recommendations for diagnosis and treatment of Atypical Hemolytic Uremic Syndrome (aHUS): an expert consensus statement from the Rare Diseases Committee of the Brazilian Society of Nephrology (COMDORA-SBN). J Bras Nefrol. 2025 Apr-Jun;47(2):e20240087.

CHART 2 RECOMMENDATIONS FOR DIAGNOSTIC TESTS

	Diagnostic tests
Confirm TMA	Biochemical evaluation
	Hematological exam: hemoglobin, thrombocytes, and reticulocytes
	Serum LDH
	Serum haptoglobin
	Peripheral blood smear (detect the presence of schistocytes)
	Indirect Coombs/direct Coombs
	PT/aPTT/fibrinogen
Complement testing and other tests/Etiology	**Detect STEC**
	Stool or rectal swab culture
	PCR for STEC virulence genes in stool
	Serology: serum (*E. coli*) and Yersinia antibodies (anti-LPS antibodies for prevalent serotypes)
	Detect ADAMTS-13 deficiency
	Von Willebrand protease activity
	Test underlying causes (secondary causes)
	Plasma homocysteine (increased levels are observed in cobalamin disturbances)
	HIV serology, pulmonary cultures, influenza
	ANA/anti dsDNA (Farr)/anti-centromere Ab/antiphospholipid antibodies (anticardiolipin IgG and IgM, anti B/lupus anticoagulant)
	Hemocultures
	Pregnancy testing
	Chest X-ray
	Factor H; factor I antibodies
	Serum CH50
	Serum MAC (C5b-9)
	Serum levels of C3, C4; index C3d/C3
	CD46 expression on leukocytes (poly- or mononuclear leukocytes using a FACS test)
	Blood levels of factor B, factor Bb, C3 convertase, factor H activity, antibodies for factor I, other complement factors and AP50
Genetic testing/ Etiology	Complement factor H (*CFH* gene)
	Complement factor I (*CFI* gene)
	Membrane cofactor protein (*MCP* gene)
	Complement factor B (*CFB* gene)
	Complement C3 (*C3* gene)
	Complement factor H-related proteins (*CFHR* genes)
	CFH-CFHR hybrid gene
	DGKE variants (children under 2 years old, especially if nephrotic syndrome is associated)
	Thrombomodulin (*THBD gene*)
	ADAMTS13 gene: if indicated (PTT)
	MMACHC gene: if indicated to exclude defect in cobalamin deficiency (especially patients under 18 years old)
	Other complement genes, if indicated

Abbreviations – TMA: thrombotic microangiopathy; LDH: lactate dehydrogenase; PT: prothrombin time; aPTT: activated partial thromboplastin time; STEC: Shiga toxin-producing Escherichia coli; PCR: polymerase chain reaction; ANA: antinuclear antibody; CH50: measuring the 50% hemolytic complement; MAC: membrane attack complex; AP50: alternative pathway hemolytic complement.

Conclusiones

- La rápida determinación de la actividad de ADAMTS13 es clave para el diagnóstico de certeza de la PTT y poder descartar otras causas de MAT.
- Se ha producido un avance en el conocimiento de la metaloproteasa ADMTS13.
- La determinación de la actividad de ADMST13 comienza a estar al alcance de de mas centros.
- La investigación dirigida a la determinación del anticuerpo anti-ADAMTS13.
- Mas lenta la determinación del AB (sigue siendo de laboratorios especializados)
- ADAMTS13: configuración ABIERTA vs CERRADA: nuevos biomarcadores para el diagnóstico y seguimiento de PTTi.

Organizado por:

Clínica Universidad de Navarra

PUESTA AL DÍA

HEMATOLOGÍA EN 48H

[LO QUE DEBES CONOCER PARA TU PRÁCTICA CLÍNICA]

X EDICIÓN

ACTUALÍZATE 48 HORAS