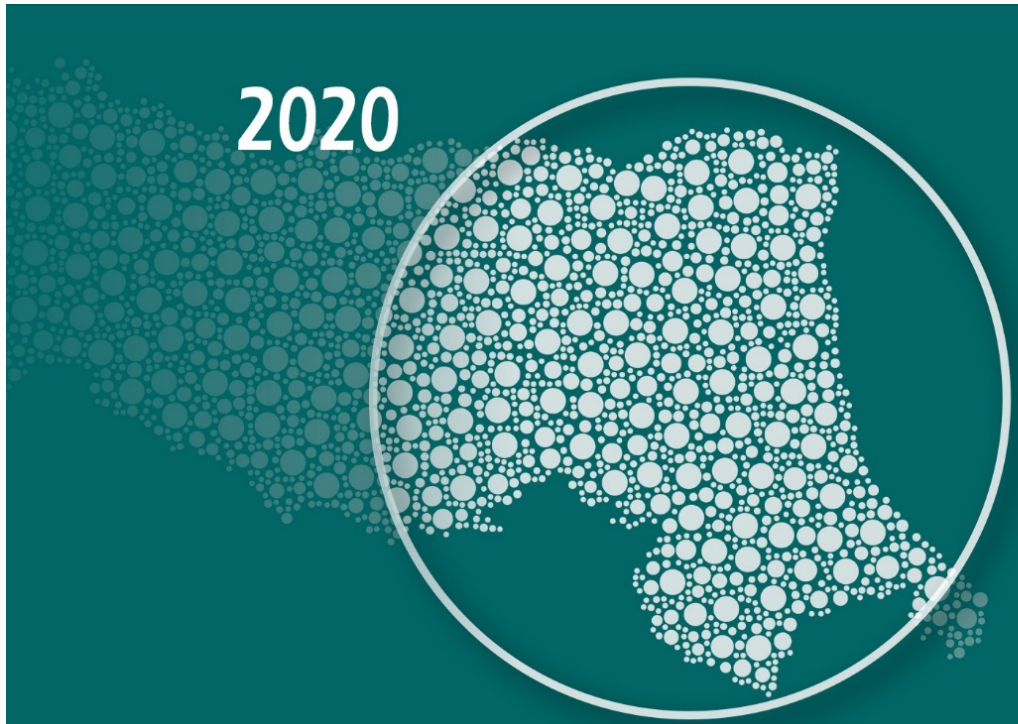


Progetto
Ematologia
Romagna

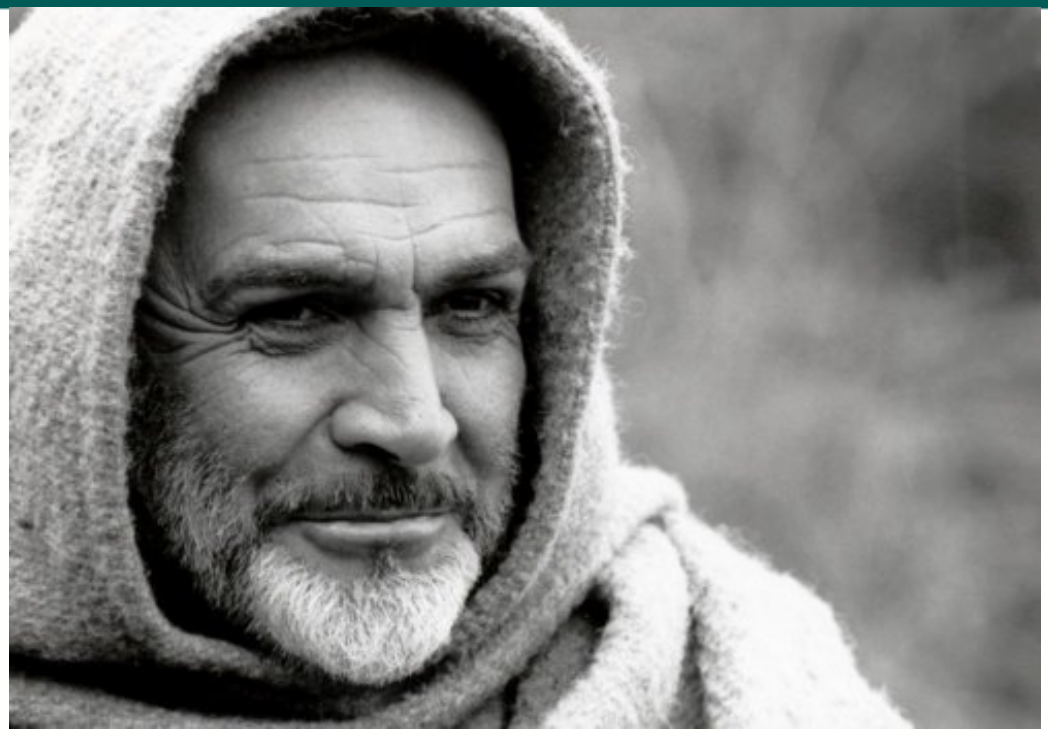
DISORDINI EMOCOAGULATIVI ACQUISITI

Marco Marietta - Modena

Relazioni con soggetti portatori di interessi commerciali in campo sanitario

Ai sensi dell'art. 76 sul Conflitto di Interessi, pag. 34 dell'Accordo Stato-Regione del 2 febbraio 2017, dichiaro che negli ultimi due anni ho avuto i seguenti rapporti anche di finanziamento con soggetti portatori di interessi commerciali in campo sanitario:

- ✓ **Advisory board**: BioVIIIx
- ✓ **Consulenza**: Kedrion, Octapharma Italy
- ✓ **Relazioni a convegni**: BioVIIIx



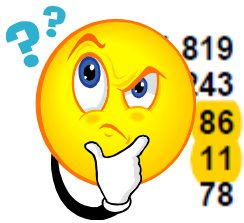
A parità di fattori la spiegazione più semplice è da preferire.

Se senti rumore di zoccoli, pensa a dei cavalli, non a delle zebre.

...a caccia di zebre...

Tabella A6. Totale dei pazienti inseriti nel Registro, distinti per patologia e sesso (2014)

Patologia	Maschi	Femmine	Totale
Emofilia A grave	1.798	2	**1.800**
Emofilia A moderata	520	2	**522**
Emofilia A lieve	1.559	25	**1.584**
Emofilia B grave	292	1	**293**
Emofilia B moderata	176	2	**178**
Emofilia B lieve	341	9	**350**
Malattia di von Willebrand tipo 1	897	1.277	**2.174**
Malattia di von Willebrand tipo 2	246	296	**542**
Malattia di von Willebrand tipo 3	49	64	**113**
Difetti di altri fattori della coagulazione	902	917	**819**
Piastrinopatie	101	142	**243**
Emofilia A acquisita	37	49	**86**
Malattia di von Willebrand acquisita	5	6	**11**
Altro	37	41	**78**
Carrier Emofilia A	-	513	**513**
Carrier Emofilia B	-	128	**128**
Totale	**6.960**	**3.474**	**10.434**



Zebra #1

- ✓ *Donna, 74 anni, con storia di sclerosi multipla parzialmente invalidante da circa 15 anni; concomita ipotiroidismo in trattamento sostitutivo, ipercolesterolemia famigliare ed ipertensione arteriosa essenziale.*
- ✓ *Da alcuni mesi comparsa di ematomi agli arti sia spontanei che in relazione a traumi e/o tensione muscolare prolungata (braccio sinistro utilizzato per sollevare il corpo da letto) associati ad alterazione dell' aPTT (modesto allungamento) e positività per Lupus Anticoagulant (6/8/2020)*
- ✓ *11.9: accesso in PS Ospedale per accertamenti (caduta ed ematomi), esegue emogas radiale a sinistra.*

Zebra #1

13.9.2020

✓*Nuovo accesso in PS per ecchimosi arto superiore dx diffuse e notevole tumefazione ecchimotica dell'arto superiore sn, la pz si scompensa e viene centralizzato su Ospedale 2° Livello, dove giunge normotesa , cosciente e collaborante con sacca di O negativo in corso.*

✓*ore 14.23: TC arto superiore sinistro: discreta tumefazione dei tessuti molli sottofasciali del versante volare di tutto l'avambraccio sinistro, a densità disomogenea, come da ematoma, mal delimitabile rispetto al tessuto muscolare in relazione ai limiti di risoluzione di contrasto della metodica, senza rilevabili contestuali spandimenti di mdc nelle acquisizioni post-contrastografiche ottenute.*

Zebra #1

13.9.2020

- ✓ *ore 14.41: Cons. Chirurgia Vascolare: ematoma sottofasciale avambraccio sinistro in paziente con sospetta diatesi emorragica.*
- ✓ *ore 17: fasciotomia*
- ✓ *ore 22.17: Hb 7.9 g/dl, NO APTT*

14.9.2020

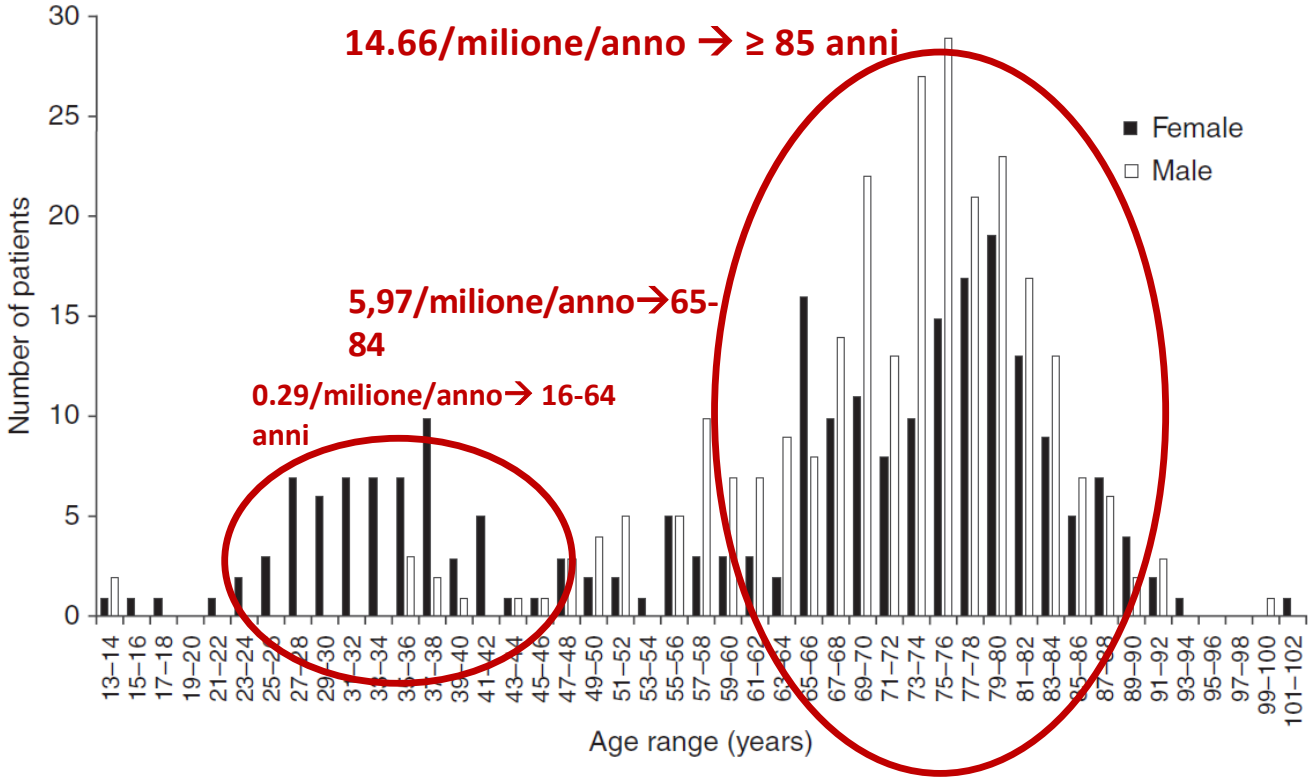
- ✓ *ore 6.25: Hb 7 g/dl, NO APTT*
- ✓ *ore 10.30: consulenza Ematologica*
- ✓ *ore 14.56: fattore VIII 14%*
- ✓ *Ore 17.26: Hb 8.4 g/dl (2 U di EC trasfuse)*
- ✓ *ore 18: inizia rFVIIa 7 mg ogni 3 ore + Urbason 70 mg/die*

Emofilia A acquisita: incidenza di 1.5 casi /milione/ anno

ORIGINAL ARTICLE

J Thromb Haemost 2012; **10**: 622–31.

Demographic and clinical data in acquired hemophilia A: results



ORIGINAL ARTICLE

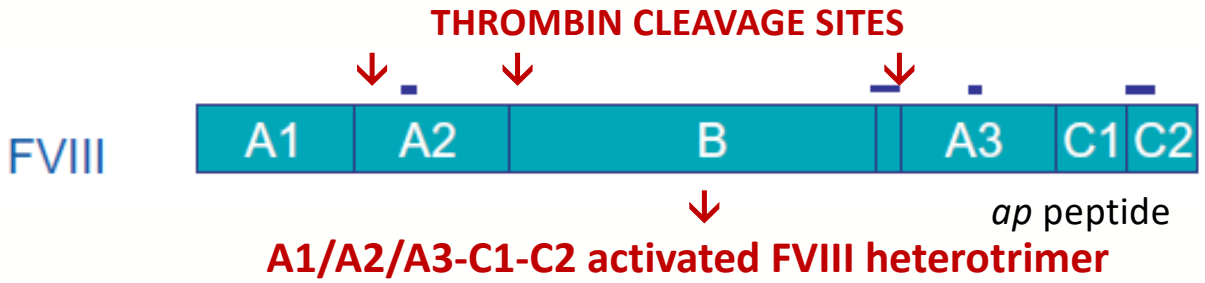
J Thromb Haemost 2012; **10**: 622–31.

Demographic and clinical data in acquired hemophilia A: results from the European Acquired Haemophilia Registry (EACH2)

P. KNOE
F. PELLE
CONTRI

Idiopathic (no underlying disorder reported)	260 (51.9)	
Malignancy	59 (11.8)	
Solid tumors		40 (67.8)
Hematologic neoplasia		19 (32.2)
Pregnancy	42 (8.4)	
Infections	19 (3.8)	
Drug induced	17 (3.4)	
Beta-lactam antibiotics		4 (23.5)
Clopidogrel		3 (17.6)
Non-beta-lactam antibiotics		2 (11.8)
Interferon		2 (11.8)
NSAID		2 (11.8)
MGUS	13 (2.6)	
Polymyalgia rheumatica	11 (2.2)	
Dermatology	7 (1.4)	

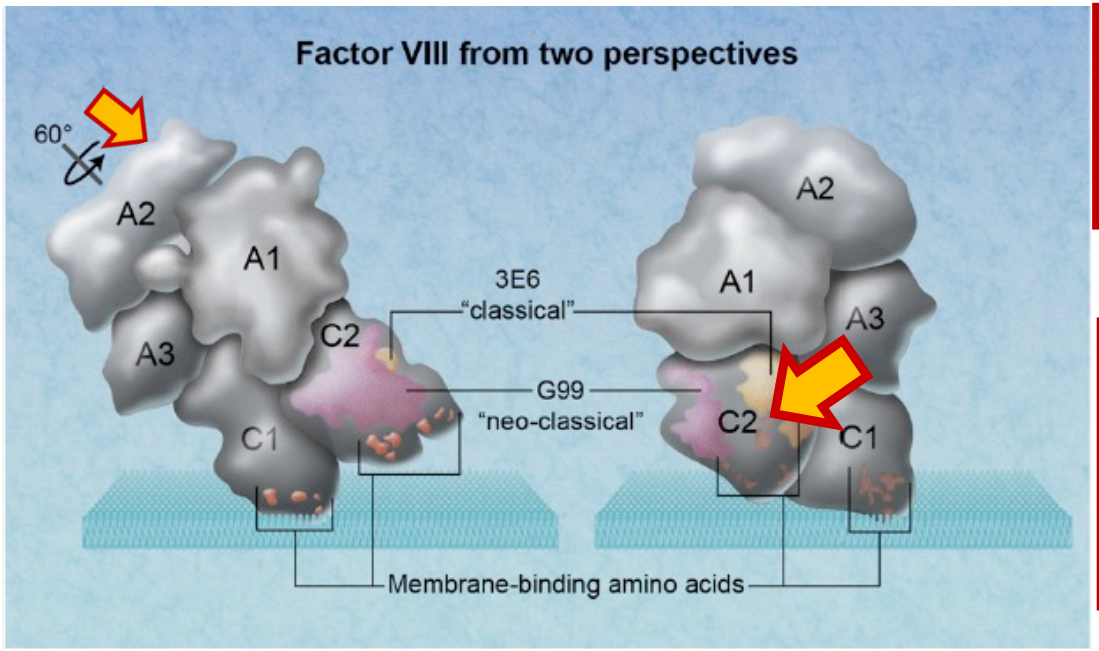
A closer look at the inhibitors in HA and AHA



- ✓ Antibodies in HA and AHA inhibitor plasmas are primarily directed to the **A2 and C2** domains.
- ✓ Intrinsic structural features in the FVIII molecule are an important determinant driving the immune response.
- ✓ Most **AHA plasmas** recognize either the A2 or C2 domain, but not both, with the **C2 domain being more frequently targeted.**

Lollar P. J Thromb Haemost 2004; 2; 1082–95.

Mechanism of action of FVIII inibitors



Anti-C2 type 1 antibodies inhibit FVIIIa binding to PL membranes

Anti-C2 type 2 antibodies bind to VWF and interfere with FVIII activation by thrombin and/or FXa

Anti-A2 inhibitors inhibit intrinsic FXase noncompetitively. Noncompetitive inhibitors do not prevent binding of substrate (FX) to enzyme (FIXa/FVIIIa/phospholipid), but prevent a secondary change when FXa binds the FIXa/FVIIIa complex

How do they work?

Antibodies could inhibit FVIII procoagulant function in several ways:

- ✓ by blocking the binding of FVIIIa to FIXa, FX or phospholipid;
- ✓ by interfering with the proteolytic activation of FVIII;
- ✓ by interfering with binding to VWF, displacing FVIII from VWF in vivo and increasing the clearance of FVIII. These antibodies would not affect the procoagulant activity of FVIII in vitro in the Bethesda assay unless they also interfered with intrinsic pathway FX activation or the activation of FVIII.

Lollar P. J Thromb Haemost 2004; 2; 1082–95.

ORIGINAL ARTICLE

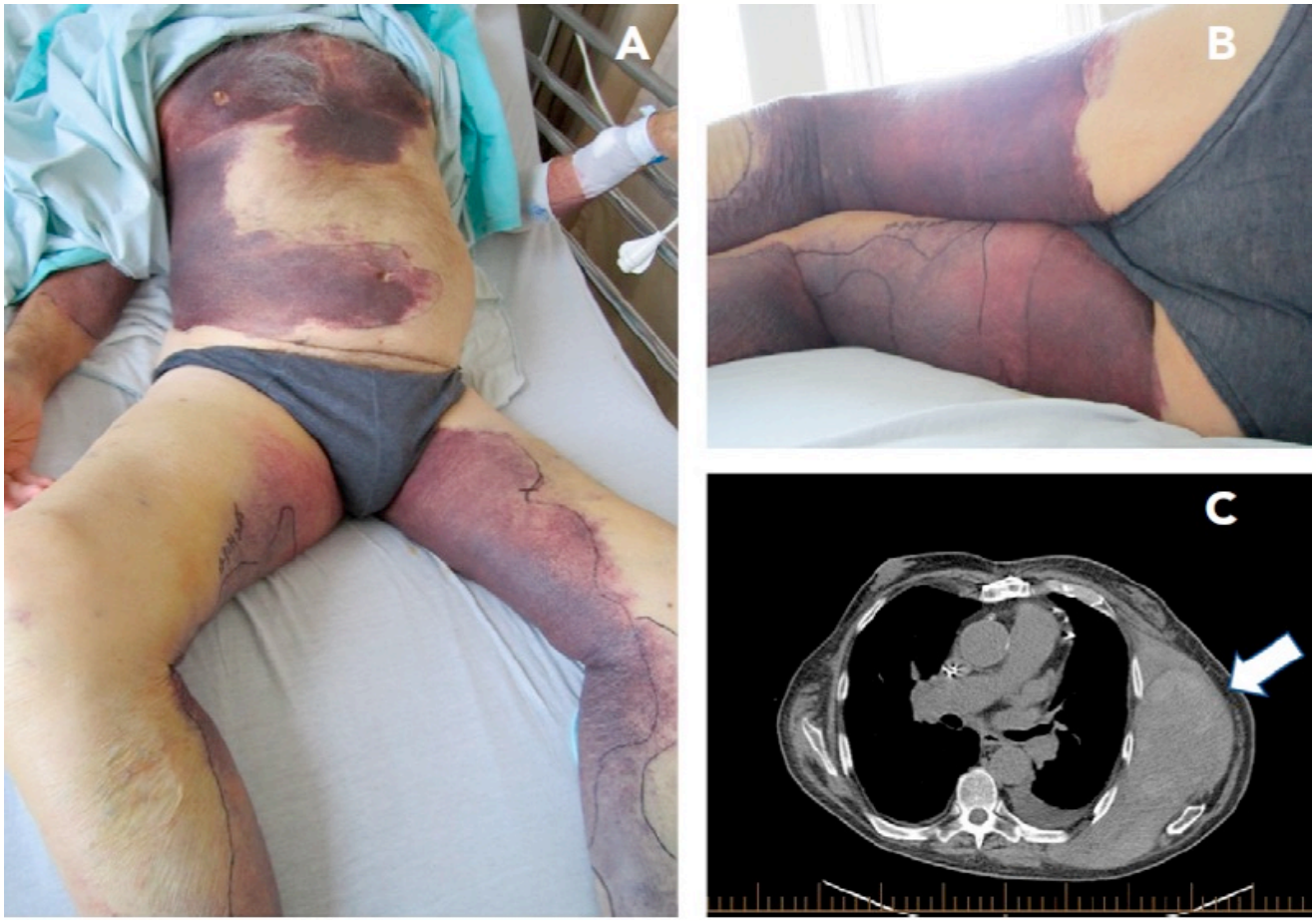
J Thromb Haemost 2012; **10**: 622–31.

Demographic and clinical data in acquired hemophilia A: results from the European Acquired Haemophilia Registry (EACH2)

P. KNOEBL,* P. MARCO,† F. BAUDO,‡ P. COLLINS,§ A. HUTH-KÜHNE,¶ L. NEMES,**
F. PE
CON

Table 3 Bleeding before and after diagnosis

	Entire collective
Bleeding as trigger for diagnosis [*n* (%)]	467 (89.0)*
Time from bleeding event to definite diagnosis	
Median [days (IQR)]	3 (0–12)
More than 6 months [*n* (%)]	6 (1.3)
1–6 months [*n* (%)]	46 (9.8)
1 week–1 month [*n* (%)]	105 (22.4)
1 week [*n* (%)]	122 (26.1)
0 (−1 to 1 day) [*n* (%)]	174 (37.2)
Bleeding after diagnosis [*n* (%)]	
1 week–1 month	6 (1.3)
1 month–1year	4 (0.9)
> 1 year	5 (1.1)
No bleeding [*n* (%)]	33 (6.6)



Jean St-Louis blood® 16 JULY 2020 | VOLUME 136, NUMBER 3

Raccomandazione #1

APTT = Schlutzkrapfen

Semin Thromb Hemost. 2019;45:22-35

How to Optimize Activated Partial Thromboplastin Time (APTT) Testing: Solutions to Establishing and Verifying Normal Reference Intervals and Assessing APTT Reagents for Sensitivity to Heparin, Lupus Anticoagulant, and Clotting Factors

Emmanuel J. Favaloro, PhD, FFSc (RCPA)[1,2] Geoffrey Kershaw, FAIMS[2] Soma Mohammed, BSc[1]

Table 1 The APTT test—multipurpose and sensitive, but nonspecific

Main uses of the APTT	APTT may be prolonged also in the following conditions
• Screen for hemostasis disorders • Screen for hemophilia (A, B, C; FVIII, FIX, and FXI deficiency, respectively) • Unfractionated heparin monitoring • Screen for lupus anticoagulant (LA; or as part of a panel of tests to assess for LA)	• Anticoagulant therapy (VKAs[a] including warfarin, DOACs including dabigatran and rivaroxaban[a]) • Liver disease[a] • Vitamin K deficiency[a] • Disseminated intravascular coagulation[a] • Presence of factor inhibitors • VWD (due to loss of FVIII) • Combined FV/FVIII deficiency[a]

Semin Thromb Hemost 2014;40:803–811.

Laboratory Diagnosis of Acquired Hemophilia A: Limitations, Consequences, and Challenges

Andreas Tiede, MD, PhD[1] Sonja Werwitzke, MD, PhD[1] Rüdiger E. Scharf, MD, PhD[2]

Table 3 Differential laboratory diagnosis of AHA

Differential diagnosis	Interference with AHA diagnosis	Appropriate action
Lupus anticoagulant	• Prolongs aPTT • Acts as (fast reacting) inhibitor in aPTT mixing study • May sometimes inhibit FVIII:C assays • May cause false-positive Bethesda assay	• Negative dRVVT-based assay may exclude LA, but up to 20% of patients with FVIII inhibitors are positive in dRVVT-based mixing and confirmation tests • Use more than one dRVVT assay to test for LA • Clinical consideration

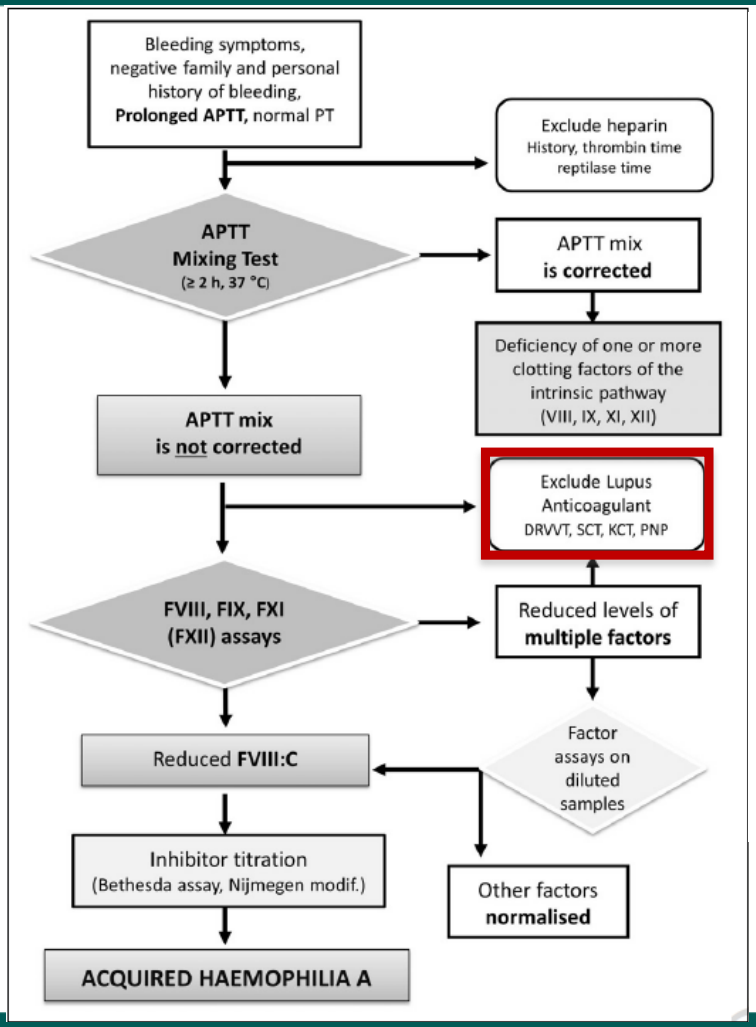
International Journal of Laboratory Hematology
Int. Jnl. Lab. Hem. 2014, 36, 352–363

Pitfalls in LAC testing

To avoid false-positive results	To avoid false-negative results
Apply three step procedure: screen, mix, and confirm	Proper plasma preparation
Calculate cutoff value by 99th percentile	Diluting effect of mixing studies?
Use only two PL-dependent assays: dRVVT and aPTT	
Repeat testing after 12 weeks	
Look for specific coagulation factor inhibitors if suspect bleeding disorder	
Check INR and avoid testing under AVK and new oral anticoagulants	
Do not test in presence of heparin or DOACs	
Do not test in acute phase (check CRP)	Do not test in acute phase (elevated FVIII)

Be compliant to the guidelines
Perform IQC (positive and negative)
Participate in EQC program
Be aware of the performance characteristics, sensitivity, and specificity of your assays
Perform all three aPL tests (LAC, aCL and ab2GPI) and make antibody profiles

Blood Transfus 2015; 13; 498-513



Sometimes a high-titre anti-FVIII inhibitor can interfere non-specifically with assays of other factors of the intrinsic pathway, leading to the finding of falsely reduced activity of FIX, FXI and FXII.

In these cases, diluting the sample being tested with buffer solution reduces this interference, leading to normalization of the activity of the other factors except FVIII, which continues to be inhibited even at lower concentration by the specific autoantibody

Zebra #2

- ✓ *Uomo, 72 anni, noto dal 2005 per doppia componente monoclonale IgG-K e IgM-lambda.*
- ✓ *A settembre 2011 BOM con infiltrato plasmacellulare <10%.*
- ✓ *A marzo 2018 ricovero per angina da sforzo, posizionato stent medicato all'ostio del tronco comune, sulla coronaria di dx distale.*
- ✓ *Ricovero a giugno 2018 per ulteriori multipli posizionamenti di stent medicati (in totale 10). Alla dimissione 7/6/2018: Hb 10.2 g/dl.*
- ✓ *Ottobre 2018: Hb 9.6 g/dl, GB 8400/mmc, Plt 506000/mmc, monociti 1490/mmc, buono l'assetto marziale e b12 e folati, proteinuria 24 ore 243 mg/24 ore, CM IG kappa <0.50 g/dl, IgM lambda 0.85 g/dl, catene k libere 10 mg/L, L 90 mg/L, K/L 0.12.*

Zebra #2

- ✓ *Da quando assume doppia terapia antiaggregante, riferiti multipli episodi di epistassi che hanno necessitato di interventi ORL.*
- ✓ *Agli esami ematici del 24/10 si segnala: Hb 9.4 g/dl,, Plt 473000/mmc, aPTT 1,56, proteinuria 24 ore 108 mg/24 ore, CM IgG kappa < 0.5 g/dl, IgM lambda 0.92 g/dl (stabile), IF urinaria ed elettroforesi urinarie presenza di immunoglobuline monoclonali. Catene k libere 12 mg/L, λ 100 mg/L, K/L 0.12 (stabile).BNP 67 pg/ml.*
- ✓ *Ad approfondimento dell'APTT allungato: fattore VIII 35%, FVW ricof 18%.*
- ✓ *Test alla DDAVP: FVIII 63%, VWF Ag 49%, VWF RiCof 68% (a 4 ore)*
- ✓ *Eseguita BOM (previa somministrazione di DDAVP): marcata infiltrazione linfocitaria 55-60%.*

Zebra #2

- ✓ *Nuovo ricovero 1/09/2019 per RESTENOSI SUBOCCLUSIVA NEL TRATTO MEDIO DI GROSSO RAMO OM TRATTATO CON PTCA e RESTENOSI INTRASTENT NELL'OSTIO DEL VASO CX TRATTATO CON SOLA PTCA.*
- ✓ *Agosto 2020: vWF RiCof 24%, Ag 36%. Fattore VIII 50%. Ripetuto sangue occulto: positivo 1 su 3 campioni.*
- ✓ *Valutazione cardiologica : sospendere Brilique, proseguire con ASA.*
- ✓ *4.9.2020 esegue colonscopia: diverticolosi del colon (DICA2). Non altri reperti. Non effettuate biopsie.*
- ✓ *11.9.2020: GB 4.890/mmc, Hb 11.5 g/dl, VCM 90.8 fl, aPTT Ratio 1.27.*

Ferrata Storti Foundation
Haematologica 2020
Volume 105(8):2032-2037

Acquired von Willebrand syndrome: focused for hematologists

Massimo Franchini[1] and Pier Mannuccio Mannucci[2]

Table 1. Conditions associated with the acquired von Willebrand syndrome.

Conditions	
Cancers	Hematologic: MGUS, multiple myeloma, Waldenstrom macroglobulinemia, chronic lymphocytic leukemia, hairy cell leukemia, lymphomas, essential thrombocythemia, polycythemia vera, chronic myeloid leukemia. Solid: Wilm's tumor, lung and bladder adenocarcinoma.
Autoimmune diseases	Systemic lupus erythematosus, autoimmune thyroid disorders, connective tissue diseases, GvHD.
Drug-induced	Antibiotics (griseofulvin, ciprofloxacin), anticonvulsants (valproic acid), plasma volume expander (HES).
Other	Cardiovascular disorders (aortic stenosis, congenital cardiac defects, mitral valve prolapse, left ventricular assist devices), infections (viral, parasitical), uremia, gastrointestinal angiodysplasia, diabetes.

MGUS: monoclonal gammopathy of unknown significance; GvHD: graft-*versus*-host disease; HES: hydroxyethyl starch.

Haematologica 2020
Volume 105(8):2032-2037

The complex pathophysiology of AvWS involves various and different mechanisms, depending on the underlying disease: :

- ✓ Circulating autoantibodies directed against functional or non-functional vWF domains. Antibody binding to vWF leads to the formation of immune complexes that are cleared from the circulation by the reticulo-endothelial system. (MGUS and multiple myeloma).
- ✓ Inhibitor antibodies that neutralize platelet-related vWF activities (rare)
- ✓ Selective adsorption of HMW multimers on tumor cells leading to their enhanced plasma clearance (lymphoproliferative diseases)
- ✓ Aberrant expression on abnormal plasma cells of the Gp Ib (MGUS)
- ✓ vWF adsorption onto the cell membranes and subsequent plasma clearance (myeloproliferative neoplasms)

...altre zebre...

Advances in managing rare acquired bleeding disorders

- ✓ Autoantibodies against **FV** are the most frequent (~ 150 cases reported) after AE and aVWD
- ✓ The majority of the cases occurred after exposure to bovine thrombin used as topical hemostatic agents in vascular, orthopedic, and neurosurgical procedures
- ✓ The diagnosis of FV inhibitors is established on **prolonged PT and APTT**.
- ✓ Autoantibodies against fibrinogen, prothrombin and factor VII have been reported only sporadically

Franchini M et al. Expert Rev Hematol. 2020 Jun;13(6):599-606.

...altre zebre...

Advances in managing rare acquired bleeding disorders

- ✓ Autoantibodies against **FV** are the most frequent (~ 150 cases reported) after AE and aVWD
- ✓ The majority of the cases occurred after exposure to bovine thrombin used as topical hemostatic agents in vascular, orthopedic, and neurosurgical procedures
- ✓ The diagnosis of FV inhibitors is established on **prolonged PT and APTT**.
- ✓ Autoantibodies against fibrinogen, prothrombin and factor VII have been reported only sporadically

Franchini M et al. Expert Rev Hematol. 2020 Jun;13(6):599-606.

…altre zebre…

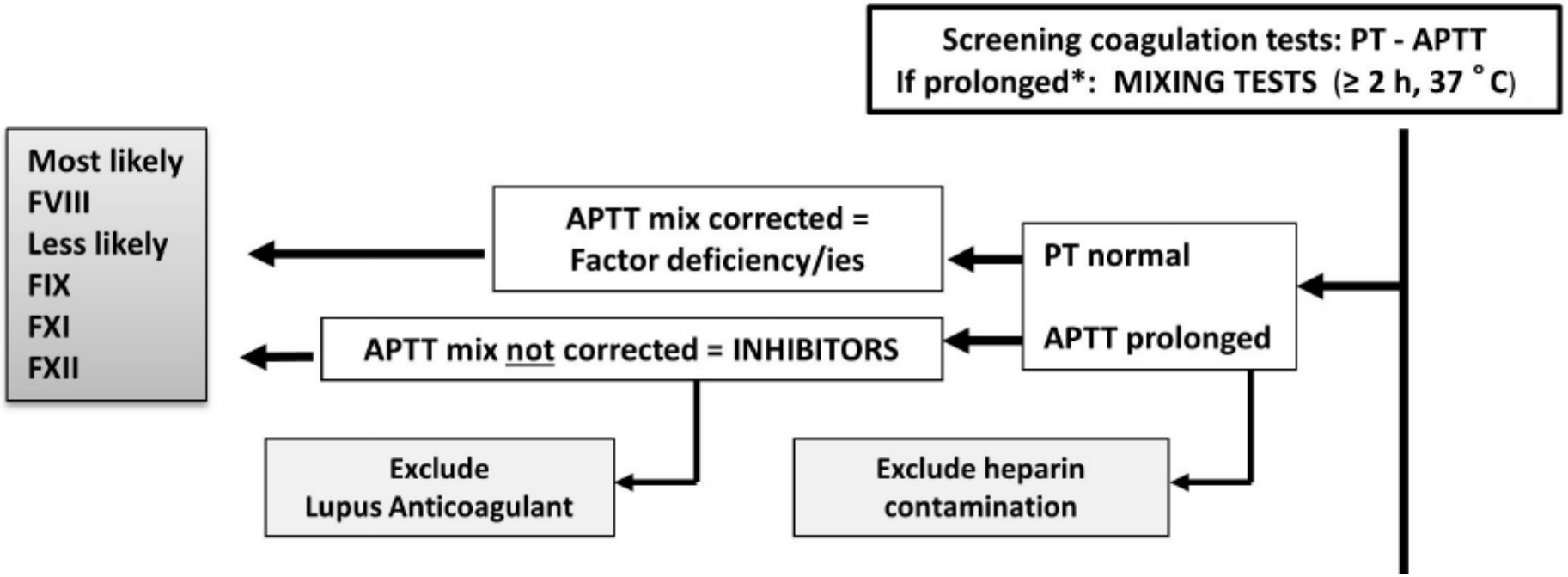
- ✓ **Acquired FX deficiency** due to the presence of **autoantibodies** is rarely reported
- ✓ Most cases of acquired FX deficiency are associated with **amyloidosis**
- ✓ Prolongation of PT and APTT, correctable with the mixing test.
- ✓ Most likely pathogenic mechanism: absorption of FX onto light-chain fibrils, primarily in the liver and spleen, as documented by the recovery of FX activity following splenectomy.

Franchini M et al. Expert Rev Hematol. 2020 Jun;13(6):599-606.

Blood Transfus 2015; 13; 498-513

Acquired inhibitors of clotting factors: AICE recommendations for diagnosis and management

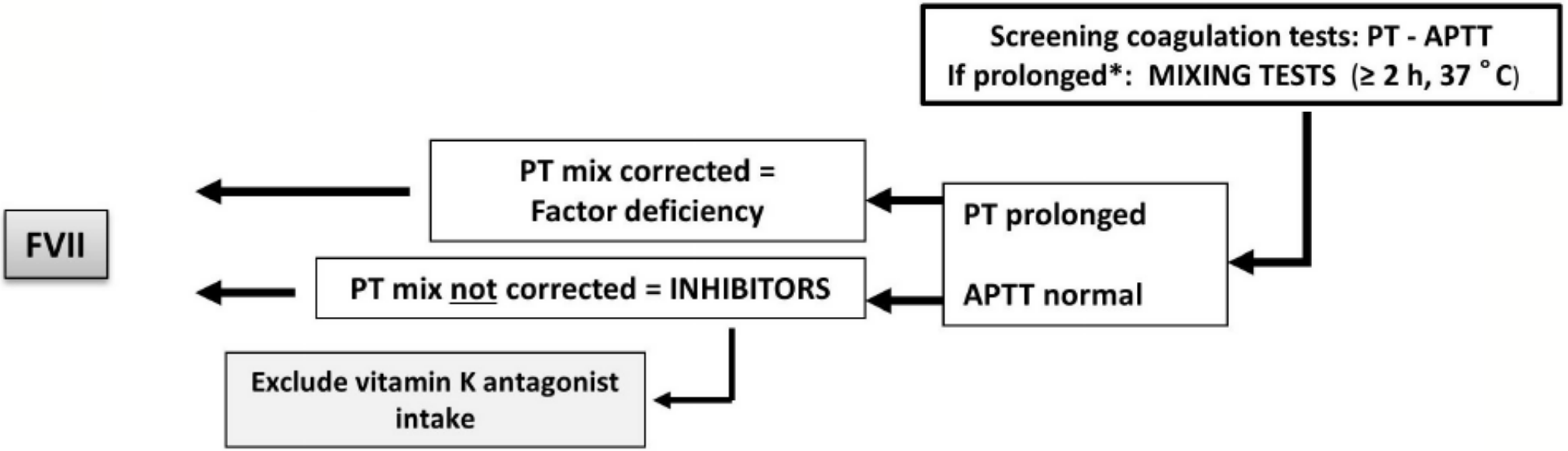
Massimo Franchini[1], Giancarlo Castaman[2], Antonio Coppola[3], Cristina Santoro[4], Ezio Zanon[5],



Blood Transfus 2015; 13; 498-513

Acquired inhibitors of clotting factors: AICE recommendations for diagnosis and management

Massimo Franchini[1], Giancarlo Castaman[2], Antonio Coppola[3], Cristina Santoro[4], Ezio Zanon[5],



Raccomandazione #2

...dipende...

Raccomandazione #3

Pensa(ci) Winnie Pooh, pensa(ci)