MEDLAB BASICS · STUDY SHEET

Hemostasis and Coagulation Testing

Platelets and the cascade, factors and inhibitors, PT/INR, aPTT, TT, fibrinogen, D-dimer, mixing studies, platelet function, anti-Xa, specimen rules and the bleeding and clotting disorders

40 sources + more detail
medlabbasics.com/sheets/lab-coagulation.html
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1 Hemostasis in one table

STEP	WHAT HAPPENS	KEY PLAYERS
Adhesion	VWF anchors to exposed collagen and binds platelets	VWF, GP Ib/IX (absent in Bernard-Soulier)
Activation	Platelets release ADP; COX makes thromboxane A2	ADP (P2Y12 receptor), TXA2 (blocked by aspirin)
Aggregation	Fibrinogen bridges GP IIb/IIIa on neighboring platelets	GP IIb/IIIa (absent in Glanzmann), fibrinogen
Coagulation	Tissue factor/VIIa starts it; tenase and prothrombinase make a thrombin burst; thrombin turns fibrinogen into fibrin	TF, VIIa, IXa/VIIIa, Xa/Va, thrombin
Stabilization	XIIIa cross-links fibrin	Factor XIII
Regulation	Natural anticoagulants switch the process off	Antithrombin, protein C/S, TFPI
Fibrinolysis	Plasmin digests fibrin into degradation products, including D-dimer	Plasminogen, tPA, PAI-1, alpha2-antiplasmin

2 Screening tests and what they cover

TEST	MEASURES	PROLONGED BY
PT / INR	VII, X, V, II, fibrinogen (extrinsic + common)	Warfarin, vitamin K deficiency, liver disease, VII deficiency, DIC
aPTT	XII, XI, IX, VIII, X, V, II, fibrinogen, prekallikrein, HMWK (intrinsic + common); not VII or XIII	Heparin, hemophilia A/B, XI or XII deficiency, factor VIII inhibitor, lupus anticoagulant
Thrombin time	Thrombin converting fibrinogen to fibrin	Heparin, dabigatran, low or abnormal fibrinogen
Reptilase time	Same step, snake venom instead of thrombin	Low or abnormal fibrinogen; NOT heparin
Fibrinogen (Clauss)	Clotting time of diluted plasma with excess thrombin, read from a calibration curve	Low in severe liver disease and DIC; high as an acute-phase reactant
D-dimer	Cross-linked fibrin broken down by plasmin	High in DIC and venous thrombosis

INR = (patient PT ÷ MNPT)^ISI. MNPT is the geometric mean PT of at least 20 healthy people on the same reagent; ISI is the reagent's sensitivity compared with the WHO reference thromboplastin. Example: PT 24 s, MNPT 12 s, ISI 1.2 gives 2^1.2 = 2.3. INR is built for warfarin and is not used for DOACs.

Typical intervals (each laboratory sets its own; the ASCP composite ranges list none): PT 10–13 s, aPTT 28–34 s, fibrinogen 180–400 mg/dL (1.8–4.0 g/L). The PT and aPTT do not prolong until a factor falls to about 30% of normal.

PT	APTT	PLATELETS	THINK OF
Normal	Long	Normal	Hemophilia A or B, XI or XII deficiency, factor VIII inhibitor, lupus anticoagulant, heparin
Long	Normal	Normal	Factor VII deficiency, early warfarin, early vitamin K deficiency, early liver disease
Long	Long	Low	DIC, cirrhosis
Long	Long	Normal	Liver disease, vitamin K deficiency, warfarin, heparin, DOACs; X, V, II or fibrinogen deficiency
Normal	Normal	Normal	VWD, platelet function disorder, factor XIII deficiency

3 Mixing studies

- Patient plasma is mixed 1:1 with pooled normal plasma, which guarantees at least 50% of every factor: enough to normalize the PT or aPTT if a factor is simply missing.
- Test the mix at once and again after incubation at 37 °C (Merck: 1 hour).
- Corrects immediately and after incubation: factor deficiency.
- Corrects or nearly corrects at once, then prolongs after incubation: time- and temperature-dependent inhibitor, classically an anti-factor VIII antibody (acquired hemophilia or an alloantibody in hemophilia A).
- Does not correct, immediately or after incubation: lupus anticoagulant (confirm with a phospholipid-dependent test) or heparin (check TT, reptilase time or anti-Xa).

EXAM TIP Exams pair the patterns: bleeding plus a non-correcting aPTT that worsens with incubation is a factor VIII inhibitor; clotting or pregnancy loss plus a non-correcting aPTT is lupus anticoagulant. LA prolongs the test tube but causes thrombosis, not bleeding.

4 Specimen rules

- 3.2% buffered sodium citrate (105–109 mmol/L), 9 parts blood to 1 part citrate. 3.8% citrate lengthens PT and aPTT against 3.2% reference ranges.
- Fill to the mark (at least 90%). Under about 89% fill the aPTT is biased; underfilled tubes are rejected. Clotted samples are always rejected.
- Order of draw: blood cultures, then citrate, then serum and additive tubes. A discard tube is needed only with a winged (butterfly) set, to fill the tubing's dead space.

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4 Specimen rules CONTINUED

- Hematocrit above 55%: too little plasma for the citrate. Citrate to leave in the tube C (mL) = 0.00185 × (100 − Hct%) × blood volume (mL). Example: Hct 65%, 2.7 mL draw: C = 0.00185 × 35 × 2.7 = 0.17 mL, so remove about 0.13 mL of a 0.3 mL fill.
- Keep capped at room temperature (15–25 °C); do not refrigerate. Centrifuge 1500 g for 15 min to platelet-poor plasma (below 10 × 10⁹/L platelets); double-centrifuge for lupus anticoagulant and for frozen plasma.
- Time limits: PT 24 h; aPTT 4 h; aPTT for heparin monitoring centrifuged within 1 h; factors V and VIII within 3 h; platelet function within 3–4 h.
- Freeze at −70 °C or colder if delayed (−20 °C: about 2 weeks); thaw at 37 °C and do not refreeze.

⚠ **SAFETY** Call critical results promptly to the care team (for example an INR above 5 or fibrinogen below 80 mg/dL [0.8 g/L]; each laboratory sets its own list). An underfilled or clotted tube is rejected and recollected; a line draw is noted on the report, and a long TT with a normal reptilase time points to heparin in the sample.

5 Platelet function and aggregation patterns

DISORDER	ADP	COLLAGEN	EPINEPHRINE	ARACHIDONIC ACID	RISTOCETIN
VWD (types 1, 2A, 2M, 3)	Normal	Normal	Normal	Normal	Reduced (type 1 can be normal); absent in type 3
VWD type 2B (and platelet-type VWD)	Normal	Normal	Normal	Normal	Enhanced at low dose
Bernard-Soulier (GP Ib/IX)	Normal	Normal	Normal	Normal	Reduced or absent; large platelets, low count
Glanzmann (GP IIb/IIIa)	Absent	Absent	Absent	Absent	Preserved
Storage pool (dense granule)	No second wave	Reduced	Reduced	Variable	Normal
Aspirin or aspirin-like (COX) defect	No second wave	Reduced	No second wave	Absent	Normal
P2Y12 defect or inhibitor	Reduced even at high dose	Normal or reduced	Normal or reduced	Normal or reduced	Normal

- Light transmission aggregometry: platelet-rich plasma (200–250 g for 10 min, no brake) clears as platelets clump; finish within 3–4 h; unreliable with platelet counts below 100 × 10³/µL (100 × 10⁹/L); stop aspirin and thienopyridines 10 days and reversible NSAIDs 3 days before, when possible.
- PFA-100/200: whole blood under high shear through a collagen membrane with epinephrine or ADP; the result is the closure time. Long with VWD (types 2 and 3 often no closure) and severe platelet defects; the epinephrine and ADP pair helps separate these from aspirin effect and mild defects. Platelet count and hematocrit also change it, and it is insensitive to mild defects. Reject Hct below 30% or above 50%, or platelets below 100 × 10³/µL.

6 The disorders at a glance

DISORDER	KEY RESULTS
Hemophilia A (VIII) / B (IX)	X-linked; long aPTT, normal PT and platelets; factor assay separates them; severe below 1%
VWD	Low VWF antigen and activity; type 2: activity/antigen ratio below 0.7 (ASH/ISTH 2021); aPTT normal or slightly long
DIC	Low platelets, long PT and aPTT, falling fibrinogen, high D-dimer, schistocytes; factor VIII low
Liver disease	Long PT (then aPTT), low fibrinogen when severe; factor VIII normal or high
Vitamin K deficiency / warfarin	Long PT first (VII has the shortest half-life); corrects with vitamin K
HIT	Platelets fall over 50%, 5–14 days into heparin; thrombosis, not bleeding; anti-PF4/heparin immunoassay, then SRA
TTP	Schistocytes, low platelets, high LD, low haptoglobin, negative DAT, normal PT/aPTT; ADAMTS13 below 10%
ITP	Isolated low platelets, large platelets, normal coagulation; a diagnosis of exclusion
Factor V Leiden	APC resistance assay; confirm by DNA testing
Prothrombin 20210	DNA test only; prothrombin level is high
AT, protein C, protein S deficiency	Activity assays (protein S: free antigen); test off heparin and warfarin and not during acute clot
Antiphospholipid syndrome	LA, anticardiolipin or anti-beta2-GPI positive on two occasions at least 12 weeks apart

💡 **EXAM TIP** MLT candidates: the MLT outline keeps PT/INR, aPTT, fibrinogen, D-dimer, TT, mixing studies, PFA, anti-Xa, protein C/S and FVL/prothrombin 20210. Inhibitor, factor and VWF assays, aggregation, TEG, direct thrombin inhibitors, heparin neutralization, LA and HIT studies are MLS only.

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