

RECOMMENDATIONS AND GUIDELINES

Lupus anticoagulant detection in anticoagulated patients. Guidance from the Scientific and Standardization Committee for lupus anticoagulant/antiphospholipid antibodies of the International Society on Thrombosis and Haemostasis

Armando Tripodi¹ | Hannah Cohen^{2,3} | Katrien M. J. Devreese⁴ 

¹Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Angelo Bianchi Bonomi Hemophilia and Thrombosis Center and Fondazione Luigi Villa, Milano, Italy

²Haemostasis Research Unit, Department of Haematology, University College London, London, UK

³Department of Haematology, University College London Hospitals NHS Foundation Trust, London, UK

⁴Coagulation Laboratory, Department of Diagnostic Sciences, Ghent University Hospital, Ghent, Belgium

Correspondence

Armando Tripodi, Foundation IRCCS Ca' Granda Ospedale Maggiore Policlinico, Angelo Bianchi Bonomi Hemophilia and Thrombosis Center and Fondazione Luigi Villa, Via Pace 9, 20122 Milano, Italy.
Email: armando.tripodi@unimi.it

Abstract

Background: The laboratory detection of lupus anticoagulants (LA) in anticoagulated patients represents a challenge and there is no consensus on the types of assays/procedures to be adopted.

Objectives: This communication of the International Society on Thrombosis and Haemostasis (ISTH), Scientific and Standardization Committee (SSC) aims to give guidance on the procedures to be adopted.

Methods: Members of the ISTH-SSC on Lupus Anticoagulant/Antiphospholipid Antibodies reviewed the literature to search for evidence on the most appropriate assays/procedures to be adopted.

Results: Anticoagulants are able to interfere with the tests used for LA detection, giving rise to occasional false-positive or false-negative LA. Some commercial tests include in their composition heparin-neutralizers able to quench unfractionated or low molecular weight heparin up to 1.0 U/mL. LA tests are less affected by low molecular weight heparin, but caution is needed in the interpretation of results. Vitamin K antagonists (VKAs) may affect LA detection. Dilution of test plasma into pooled normal plasma is not a reliable solution as false-negative or false-positive LA may occur. Direct oral anticoagulants (DOACs) affect LA detection. Hence, it is not recommended to attempt LA detection in those patients. The use of DOAC adsorbents is a promising solution and should be further investigated on LA-positive and LA-negative patient populations. Taipan/Ecarin tests may be a solution for VKAs and anti-FXa DOACs, but independent evidence on their value and standardized kits is needed.

Conclusions: LA detection during anticoagulation remains a challenge, especially for VKAs. DOAC removal by in vitro addition to plasma of appropriate adsorbents is promising.

KEYWORDS

anticoagulation, vitamin K antagonists, direct oral anticoagulants, unfractionated heparin, low molecular weight heparin

1 | BACKGROUND

The laboratory detection of lupus anticoagulants (LA) is one of the criteria to define the antiphospholipid syndrome (APS).¹ In addition, laboratory detection of LA in patients with previous venous and/or arterial thromboembolism or pregnancy morbidity informs clinical decisions on the type of anticoagulation, as well as the optimal duration and intensity to prevent recurrence.² Although detailed recommendations have been issued by the Scientific and Standardization Subcommittee (SSC) on Lupus Anticoagulant/Antiphospholipid Antibodies of the International Society on Thrombosis and Haemostasis (ISTH),³ the British Society for Haematology,⁴ as well as the Clinical and Laboratory Standards Institute,⁵ LA detection is complicated by a number of unresolved issues. Among them, LA detection during anticoagulation is one of the most important and challenging. A recent survey carried out by the ISTH-SSC revealed that there is little agreement on LA detection during anticoagulation. Only 42% of respondents indicated that it would be appropriate to test for LA during anticoagulation and the method(s) of testing suggested was highly variable.⁶ However, although LA testing in anticoagulated samples is generally discouraged because the probability for false-positive/negative results is high,^{7,8} determination of LA is required in certain clinical settings as well as for full characterization of the antiphospholipid status of patients in research studies and registries such as APS-ACTION.^{9,10} To help clear confusion, the SSC of the ISTH sought to prepare guidance based on the current literature, aimed to help laboratory operators, scientists and clinicians to tackle this important issue.

2 | EVIDENCE THAT LA DETECTION IS AFFECTED BY ANTICOAGULATION

Anticoagulation with any drug, including unfractionated heparin (UFH), low molecular heparin (LMWH), vitamin K antagonists (VKAs), direct oral anticoagulants (DOACs) and others, may potentially complicate LA detection, simply because anticoagulants generally prolong test clotting times (ie, the activated partial thromboplastin time [APTT] and dilute Russell's viper venom time [dRVVT]), currently recommended for LA detection.³ Consequently, the results of screen, mixing, and confirm procedures that are the mainstay of LA detection, may be difficult to interpret. Although clotting time prolongation is generally observed with all anticoagulants, there may be exceptions. Some tests used for LA detection, such as the APTT-derived silica clotting time or the dRVVT, include in their composition heparin neutralizers (ie, heparinase or polybrene), which are able to quench UFH and LMWH up to 1.0 U/mL. Some brands of LMWH, depending on their anti-factor (F) Xa/FIIa ratio, may result in sizeable prolongation of the APTT and congener tests and may therefore affect LA detection.¹¹ UFH clearly affects LA assays, especially APTT-based tests, with false-positive screening and mixing results.¹² However, at low anti-FXa UFH activity levels, application of the three-step procedure does not produce false-positive LA.¹¹

Essentials

- Lupus anticoagulant detection for anticoagulated patients is not well established.
- We reviewed literature and expert practice to guide lupus anticoagulant detection in this setting.
- Although there are no easy solutions, some options can be implemented.

LMWH (enoxaparin) causes false-positive APTT-based LA results at supra-therapeutic anti-FXa activity levels.^{11,13} The dRVVT is also influenced, albeit at higher anti-FXa activity levels compared with the APTT and may lead to false-positive results.^{11,13} It is therefore essential that the laboratory is informed on the composition of the reagents used for testing, the anti-FXa level up to where UFH and LMWH are neutralized, and possibly on the effect brought about on the local APTT/dRVVT by the brand(s) of LMWH used to treat patients.

Deficiency of vitamin K-dependent coagulation factors may influence the results of the APTT and dRVVT, depending on the reagent used, with variable impact on the results of the mixing test.¹⁴ Interference by DOACs in coagulation assays resulting in false-positive results for LA has been extensively studied and increased rates of false-negative or false-positive results have been reported.¹⁵ Finally, test results (especially with the APTT) may affect LA detection during acute thrombosis, during which patients are also generally treated with LMWH or DOACs. This effect is also dependent on the composition of reagents used for testing and on the presence of acute phase reactants.^{16,17} Overall, LA testing is not recommended during the acute phase unless strictly required by the clinical situation (eg, in patients with suspected catastrophic APS).

3 | THE NEED TO DETECT LA DURING ANTICOAGULATION

There are a number of reasons indicating that LA detection is useful during anticoagulation. First, current guidelines advise consideration of indefinite anticoagulation for patients with a first unprovoked venous thromboembolism (VTE),¹⁸ for which anticoagulation with DOACs has become the standard of care. Second, the European Medicines Agency has recommended avoidance of the use of DOACs in APS, especially in triple-positive patients (those with LA, anti-cardiolipin and anti- β 2glycoprotein I antibodies).¹⁹ This recommendation followed a risk assessment triggered by a recent clinical trial of APS patients triple positive for antiphospholipid antibodies, who were randomized to receive rivaroxaban vs standard-intensity warfarin and that was prematurely stopped because of excess thrombotic events in the rivaroxaban arm of the study.²⁰ The European Medicines Agency recommendation and the results stemming from another randomized clinical trial²¹ strengthen the need

for LA testing during anticoagulation. Third, some authors suggest that thrombophilia testing, including LA, may be useful in selected patients (ie, extensive VTE; recurrent arterial thrombosis and/or deep vein thrombosis or pulmonary embolism; thrombosis in the cerebral or splanchnic veins).²² Fourth, among patients with unprovoked VTE, LA positivity is associated with an increased risk of recurrence.²³ Finally, triple positivity identifies patients at the highest risk of thrombosis.²⁴

The detection of solid-phase antibodies anti-cardiolipin and anti- β 2glycoprotein I antibodies, the other two components of the laboratory APS triad, are unaffected by anticoagulation, but their detection will not enable identification of triple-positive patients. Confirmation of triple-positive patients might be anticipated without relying on the confirmation of diagnoses after twelve weeks, as required by guidelines,^{1,3} because the majority of these patients persist in positivity,^{25,26} but this approach necessitates testing during anticoagulant therapy.

4 | OPTIONS AVAILABLE FOR LA DETECTION DURING ANTICOAGULATION

Although the best option would be to collect blood for testing before starting anticoagulation, even when this is possible, most patients would be within the acute phase of thrombosis and therefore interpretation of results could be difficult. Alternative options with their inherent limitations are discussed next.

1. Dilution of the patient plasma into a pooled normal plasma.

The rationale for this procedure rests on the concept that the acquired deficiency of coagulation factors subsequent to anticoagulation will be reasonably corrected by the pooled normal plasma, thus allowing LA detection. This procedure is typically applied to patients on VKAs in whom there is a partial deficiency of vitamin K-dependent coagulation factors. It is of little value with those drugs (eg, DOACs, UFH, LMWH) that directly or indirectly inhibit coagulation factors. The procedure is relatively simple, but has some disadvantages. First, the pooled normal plasma must be suitable for the intended use (ie, platelet-free and activity of each individual coagulation factors close to 100%). In-house pooled normal plasma could be suitable, but relatively few laboratories have facilities to prepare and store this material. Second, the dilution (1:1) of the test plasma into pooled normal plasma will reduce by 50% the LA potency of the patient plasma. It is therefore likely that weak LA would be lost at diagnosis.²⁷ Third, empirical observations and data from the literature¹⁴ show that the degree of correction of the acquired coagulopathy supported by the pooled normal plasma is likely dependent on the composition of the commercial APTT or dRVVT used for testing. Hence, any information derived from local reagents cannot be generalized to others. Fourth, result interpretation after this procedure may still be problematic. Finally, there are no conclusive studies on the value of the

procedure, because there is no gold standard to detect LA and results from the literature are sparse and mainly comprise small series of patients reported as being historically positive for LA before starting anticoagulation. The use of well-characterized LA-positive plasma at graded potency²⁸ might help to understand the value of the procedure. Overall, it should be realized that, although dilution of the test plasma into pooled normal plasma is widely used, it is not robust enough to help making diagnosis of LA and both false-negative or false-positive results may occur.¹⁴

2. **Use of integrated assays for LA detection compared with three-step procedure.** There are commercial platforms for LA detection that include dual tests performed on two aliquots of the same sample at low (screen) and high (confirm) phospholipid concentrations (here called integrated LA tests). They rest on the concept that in the presence of LA, the screen clotting time is prolonged, whilst the confirm is shortened. The ratio of screen/confirm higher than the cut off value (usually 1.2) is indicative of LA. Earlier reports suggested that LA detection based on such integrated tests as silica clotting time or dRVVT is reliable for the majority of patients, even in the presence of UFH or VKAs.²⁹ However, subsequent reports showed that the screen and confirm clotting times in the presence of DOACs are not proportionally prolonged.³⁰ The screen clotting time tends to be more prolonged than the confirm. Consequently, the ratio screen/confirm tends to be higher than expected and may therefore lead to false-positive LA during DOACs.^{13,30} Similar results were observed applying the three-step procedure, also for enoxaparin.¹³
3. **Use of test procedures (reportedly) less affected by anticoagulants.** Protein extracts from snake venoms such as Taipan (*Oxyuranus scutellatus*) or Ecarin (*Echis carinatus*) might be useful for LA detection in anticoagulated patients.³¹ Both venom extracts are able to activate partially carboxylated FII (induced by VKA) as well as native FII. Interestingly, Taipan is a phospholipid and calcium-dependent activator, whereas Ecarin is not. Because of these properties, if used in combination, the two venoms extracts may help to detect LA during anticoagulation. There is information from published literature on the Taipan/Ecarin^{32,33} use, but it is mainly based on small series of patients (reportedly) positive for LA before starting anticoagulation with VKAs, with no conclusive independent evidence reported on their diagnostic efficacy. A collaborative study supported by the ISTH-SSC to assess the value of these snake venoms is ongoing,³⁴ with the results awaited. Scanty information is available on the Taipan/Ecarin diagnostic efficacy in patients on DOACs other than rivaroxaban.^{35,36} Finally, availability of commercial kits is very limited and standardization remains an issue. This is likely to limit widespread use and demonstration of independent evidence on their efficacy.
4. **Use of antidotes or neutralizers to quench in vitro the activity of anticoagulants.** In addition to heparin neutralizers (heparinase or polybrene), which are added to some commercial kits specifically designed for LA detection, there are other options that can theoretically be used to quench in vitro the activity of DOACs

and that might be useful to test for LA in anticoagulated patients. It has been reported that the specific antidote for dabigatran (idarucizumab) can be used to neutralize the *in vitro* anticoagulant effect of the drug with minimal effect on clotting times.³⁷ In principle, idarucizumab could be used to test for LA in patients on dabigatran. However, scanty information is currently available³⁸ and the cost incurred for the antidote should be considered. Similarly, andexanet-alfa, recently made available as an antidote for anti-FXa drugs, could in principle be suitable to quench the anticoagulant activity of rivaroxaban, apixaban, or edoxaban *in vitro*.³⁰ More recently, new promising compounds have emerged to quench DOAC anticoagulant activity. They are chemical substances able to bind and inactivate DOACs when added *in vitro* to plasma with minimal effect on clotting times. After centrifugation, the supernatant plasma free from DOACs can be used to measure clotting times. Two substances are commercially available, DOAC-Stop™ (Haematex Research, Sydney, Australia) and DOAC-Remove™ (5-Diagnostics, Quadrage, Switzerland) as mini-tablets to be added to plasma before testing. These charcoal substances are hydrophobic-binding agents, which are apparently able to neutralize any type of DOACs including dabigatran, rivaroxaban, apixaban, or edoxaban.³⁹ DOAC-Stop has been used in a recent international collaborative study by Favaloro et al,³⁰ who spiked plasma samples with rivaroxaban. A number of interesting observations emerged from that study. First, rivaroxaban when added to pooled normal plasma caused clotting time prolongation for most LA tests performed by participants and generated falsely elevated dRVVT screen/confirm ratio results that mimicked the presence of LA. Second, the rivaroxaban plasma samples when treated with DOAC-Stop showed correction of the prolongation of the clotting time and the screen/confirm ratio for most LA tests. Interestingly, all the participants in the study correctly identified the rivaroxaban plasma treated with DOAC-Stop as LA-negative. Third, andexanet-alfa when added to the rivaroxaban plasma was able to correct the prolonged clotting time induced by rivaroxaban. It also corrected the screen/confirm ratio, but to such an extent (overcorrection) that may lead to a potential false-negative LA in those patients with weak positive LA whilst on rivaroxaban. There are other single-center reports on the efficacy of DOAC adsorbents to quench the *in vitro* anticoagulant activity of other DOACs³⁹⁻⁴⁴ and their results are promising. Cox-Morton et al⁴⁵ recently reported results on another charcoal product (DOAC-Remove) used to remove DOACs from plasma samples before LA testing. Overall, DOAC adsorbents seem promising tools to help detect LA in patients anticoagulated with DOACs. Clinical and laboratory experience will guide whether the use of these DOAC adsorbents should eventually become standard practice. So far, no attempt has yet been made to test large numbers of patients, who are positive for LA while on DOACs to see whether DOAC adsorbent yields consistent results. In contrast, the studies carried out so far aimed mainly to show if the adsorbents were able to correct the prolonged clotting time induced by DOACs and some of them show that pretreatment of plasma with DOAC

adsorbents may interfere with clotting times and hence influence the conclusion on LA positivity. Indeed, in a normal (non-anticoagulated) plasma treated with DOAC-Stop, a dose-dependent procoagulant effect was observed,⁴¹ suggesting that that DOAC-Stop removes one or more anticoagulant proteins, especially when used in combination with small plasma volumes; reduction of free tissue factor pathway inhibitor was observed after DOAC-Stop.⁴¹ Also, false-negative LA have been described after treatment with DOAC adsorbents,^{40,44} although in weak LA positive results approaching the cutoff value. Parallel treatment with the DOAC adsorbent of the normal pooled plasma, used for calculating the normalized ratio, may avoid false-negative LA results.⁴⁰ In normal plasma added with dabigatran, treatment with DOAC-Stop had no significant effect on the dRVVT results, but a small prolongation in APTT results was described.³⁸ APTT prolongation was also reported in heparinized samples treated with DOAC-Stop.^{11,42} Moreover, care should be taken in the interpretation of results because complete reversal of the anti-FXa effect does not occur in every sample.⁴⁴ Therefore, based on current knowledge, care should be taken in using DOAC adsorbents according to the manufacturer's recommendation, especially regarding plasma volume.⁴¹ Because DOAC adsorbents may influence clotting times in plasma of non-anticoagulated or heparinized plasma, pretreatment of plasma with the adsorbents is only advised in DOAC-treated patients. Activated carbon substances cannot be used to overcome the effect of heparins on LA testing because they proved unable to neutralize the anti-FXa activity of heparins.^{11,40}

5. **Discontinuation of anticoagulation.** Whenever LA detection is deemed of special interest for decision-making in individual patients, anticoagulation might be temporarily stopped and patients on VKAs switched to LMWH. This would protect patients from thrombosis, making LA detection possible, provided that the local LMWH brand does not affect LA tests, or when the anti-FXa activity level is low.^{11,13} However, discontinuation of the treatment might be problematic in clinical practice for the following reasons. First, it exposes patients to potential risk of thrombosis, especially during the first few months after a thrombotic event; this risk may be exacerbated in some patients who might be confused about the start and stop timings of LMWH. Second, after LA testing, patients might have to be reestablished on VKAs with bridging LMWH, which is cumbersome because of the need for frequent monitoring during the bridging period and the first 6 or more weeks while the INR stabilizes. Third, this approach may be associated with a potential risk of bleeding. In patients on DOACs, on a pragmatic empirical basis, LA testing may be undertaken at least 48 hours after the last dose, and longer in patients with renal impairment, although DOAC levels should also be checked.¹⁵

5 | CONCLUSIONS

According to the current state of the art, it can be concluded that there is no easy way to reliably detect LA in anticoagulated patients.

TABLE 1 Summary of guidance on lupus anticoagulants (LA) detection during anticoagulation

We recommend that LA testing in patients on anticoagulation should be undertaken with the cognizance that anticoagulants may prolong the clotting time of the tests used for LA detection and that this effect may give rise to false-positive or false-negative LA.

We suggest that whenever possible, blood for LA detection should be collected before initiation of anticoagulation.

Some of the commercial tests specifically designed for LA detection, include in their composition heparin neutralizers that may quench unfractionated heparin (UFH) or low molecular weight heparin (LMWH) up to 1.0 U/mL. We therefore recommend that laboratories be aware of the composition of the reagents used for testing.

LA tests are less affected by LMWH than by UFH. However, we recommend caution in the interpretation of results of LA tests in patients on LMWH, unless the responsiveness to LMWH of the local LA test has been evaluated.

We recommend measurement of anti-FXa activity in patients who are known to be on LMWH or UFH and if the activity is within the therapeutic interval, LA testing can be performed if reagents contain heparin neutralizers.

Dilution of patient plasma into pooled normal plasma is frequently used prior to LA testing in anticoagulated patients. We suggest that this procedure is not a reliable solution in patients on VKAs (false-negative or false-positive LA results may occur) and we recommend against using it in patients on direct oral anticoagulants (DOACs).

Taipan/Ecarin tests that are less affected by VKAs and anti-FXa DOACs may be a solution to LA testing in anticoagulated patients. We propose that their general use is pending upon the provision of independent evidence from collaborative studies with standardized kits.

DOACs invariably affect LA detection. We do not recommend attempting LA detection in patients on DOACs, unless they are removed or neutralized. The use of DOAC adsorbents is a promising solution. We suggest that they should be further investigated in LA-positive and LA-negative patient populations.

Whenever testing for LA in patients, whose pharmacological and/or clinical history is unknown, we recommend routine laboratory testing such as prothrombin time (PT), activated partial thromboplastin time (APTT) and thrombin time (TT). This is to provide background information about unexpected coagulopathies or undocumented anticoagulation. Prolonged TT (suggesting dabigatran or UFH) should be an indication to not perform LA testing, unless the drugs are removed or neutralized.

We recommend incorporating information on the patient's anticoagulation in the request form as it is paramount for interpretation of LA results. Because normal APTT and/or PT do not exclude DOACs or LMWH, we recommend that whenever the laboratory results are suggestive of LA, but there is doubt, results should be reported along with warnings on the possible false positivity because of DOACs or other anticoagulants.

Note: The method endorsed by the ISTH Guidance and Guidelines Committee Panel on writing guidance has been adopted. Accordingly, the wording "we recommend" indicates a strong consensus among the coauthors; "we suggest," a moderate consensus; and "we propose," the areas where there is limited knowledge.

The members of the ISTH-SSC agreed on the general guidance summarized in Table 1. In addition, the members agreed that before performing LA testing for patients, whose pharmacological and/or

clinical history is unknown, routine laboratory testing such as PT, APTT, and thrombin time should be performed. Anti-FXa activity should be measured in patients who are known to be on LMWH and if the activity is within the therapeutic interval, LA testing can be performed if reagents contain heparin neutralizers. Prolonged thrombin time (suggesting presence of dabigatran or UFH) should be an indication to not perform LA testing. Because normal APTT and/or PT do not exclude DOACs, information on the patient's medication is mandatory. Whenever the laboratory results are suggestive of LA, but there is doubt about the diagnosis, results should be reported along with warnings on the possible false positivity because of DOACs. This recommendation applies also to other anticoagulants.

CONFLICT OF INTEREST

Dr. Tripodi reports honoraria for lectures at educational meetings from Werfen, Stago, and Sobi. Dr. Cohen reports, outside the submitted work, institutional research support and support to attend scientific meetings from Bayer Healthcare, with honoraria for lectures from Bayer Healthcare and consultancy fees from UCB paid to UCLH Charity. Dr. Devreese has nothing to disclose.

AUTHORS' CONTRIBUTIONS

A. Tripodi, H. Cohen, and K.M.J. Devreese conceived the guidance and reviewed the literature. A. Tripodi wrote the first draft of the manuscript. All the authors revised and accepted the manuscript, which was then accepted by the Scientific and Standardization Committee for lupus anticoagulant/antiphospholipid antibodies of the International Society on Thrombosis and Haemostasis (ISTH). The manuscript was reviewed and approved by the Guidelines and Guidance Committee of ISTH.

ORCID

Katrien M. J. Devreese  <https://orcid.org/0000-0002-7559-2579>

REFERENCES

- Miyakis S, Lockshin MD, Atsumi T, *et al.* International consensus statement on an update of the classification criteria for definite antiphospholipid syndrome (APS). *J Thromb Haemost.* 2006;4:295-306.
- Tripodi A, de Groot PG, Pengo V. Antiphospholipid syndrome: laboratory detection, mechanisms of action and treatment. *J Intern Med.* 2011;270:110-122.
- Pengo V, Tripodi A, Reber G, *et al.* Update of the guidelines for lupus anticoagulant detection. *J Thromb Haemost.* 2009;7:1737-1740.
- Keeling D, Mackie I, Moore GW, Greer IA, Greaves M; British Committee for Standards in Haematology. Guidelines on the investigation and management of antiphospholipid syndrome. *Br J Haematol.* 2012;157:47-58.
- CLSI Guideline. *Laboratory Testing for the Lupus Anticoagulant; Approved Guideline. CLSI document H60-A, 2014.* 2014. Clinical and Laboratory Standards Institute, Wayne, PA, USA.
- Cohen H, Mackie IJ, Devreese KMJ; International Society for Thrombosis and Haemostasis Scientific Standardization Committee for Lupus Anticoagulant/Antiphospholipid Antibodies. Clinical and laboratory practice for lupus anticoagulant testing: an International Society of Thrombosis and Haemostasis Scientific Standardization Committee survey. *J Thromb Haemost.* 2019;17:1715-1732.

7. Aboud M, Morel-Kopp MC, Ward C, Coyle L. False-negative or false-positive: laboratory diagnosis of lupus anticoagulant at the time of commencement of anticoagulant. *J Thromb Haemost.* 2010;8:2070-2073.
8. Tripodi A, Moia M, Pengo V. False-negative or false-positive: laboratory diagnosis of lupus anticoagulant at the time of commencement of anticoagulant: a rebuttal. *J Thromb Haemost.* 2011;9:1435-1436.
9. Erkan D, Lockshin MD; APS ACTION members. APS ACTION—AntiPhospholipid Syndrome Alliance For Clinical Trials and InternatiOnal Networking. *Lupus.* 2012;21:695-698.
10. Efthymiou M, Mackie IJ, Lane PJ, et al. Comparison of real world and core laboratory lupus anticoagulant results from the Antiphospholipid Syndrome Alliance for Clinical Trials and International Networking (APS ACTION) clinical database and repository. *J Thromb Haemost.* 2019;17(12):2069-2080.
11. De Kesel PMM, Devreese KMJ. The effect of unfractionated heparin, enoxaparin, and danaparoid on lupus anticoagulant testing: can activated carbon eliminate false-positive results? *Res Pract Thromb Haemost.* 2019;4(1):161-168.
12. Tripodi A, Biasiolo A, Chantarangkul V, Pengo V. Lupus anticoagulant (LA) testing: performance of clinical laboratories assessed by a national survey using lyophilized affinity-purified immunoglobulin with LA activity. *Clin Chem.* 2003;49:1608-1614.
13. Martinuzzo ME, Barrera LH, D'Adamo MA, Otaso JC, Gimenez MI, Oyhamburu J. Frequent false-positive results of lupus anticoagulant tests in plasmas of patients receiving the new oral anticoagulants and enoxaparin. *Int J Lab Hematol.* 2014;36:144-150.
14. Depreter B, Devreese KM. Dilute Russell's viper venom time reagents in lupus anticoagulant testing: a well-considered choice. *Clin Chem Lab Med.* 2017;55:91-101.
15. Hoxha A, Banzato A, Ruffatti A, Pengo V. Detection of lupus anticoagulant in the era of direct oral anticoagulants. *Autoimmun Rev.* 2017;16:173-178.
16. Ten Boekel E, Böck M, Vrieling GJ, Liem R, Hendriks H, de Kieviet W. Detection of shortened activated partial thromboplastin times: an evaluation of different commercial reagents. *Thromb Res.* 2007;121:361-367.
17. Schouwers SM, Delanghe JR, Devreese KM. Lupus anticoagulant (LAC) testing in patients with inflammatory status: does C-reactive protein interfere with LAC test results? *Thromb Res.* 2010;125:102-104.
18. Kearon C, Akl EA, Ornelas J, et al. Antithrombotic therapy for VTE disease: CHEST guideline and expert panel report. *Chest.* 2016;149:315-352.
19. PRAC recommendations on signals. Adopted at the 8-11 April 2019 PRAC meeting. EMA Pharmacovigilance Risk Assessment Committee (PRAC) https://www.ema.europa.eu/en/documents/prac-recommendation/prac-recommendations-signals-adopted-8-11-april-2019-prac-meeting_en.pdf. Accessed September 2019.
20. Pengo V, Denas G, Zoppellaro G, et al. Rivaroxaban vs warfarin in high-risk patients with antiphospholipid syndrome. *Blood.* 2018;132:1365-1371.
21. Ordi-Ros J, Sáez-Comet L, Pérez-Conesa M, et al. Rivaroxaban versus vitamin K antagonist in antiphospholipid syndrome: a randomized noninferiority trial. *Ann Intern Med.* 2019;171(10):685.
22. Connors JM. Thrombophilia testing and venous thrombosis. *N Engl J Med.* 2017;377:1177-1187.
23. Kearon C, Parpia S, Spencer FA, et al. Antiphospholipid antibodies and recurrent thrombosis after a first unprovoked venous thromboembolism. *Blood.* 2018;131(19):2151-2160.
24. Pengo V, Ruffatti A, Legnani C, et al. Incidence of a first thromboembolic event in asymptomatic carriers of high-risk antiphospholipid antibody profile: a multicenter prospective study. *Blood.* 2011;118:4714-4718.
25. Pengo V, Ruffatti A, Del Ross T, et al. Confirmation of initial antiphospholipid antibody positivity depends on the antiphospholipid antibody profile. *J Thromb Haemost.* 2013;11:1527-1531.
26. Devignes J, Smail-Tabbone M, Herve A, et al. Extended persistence of antiphospholipid antibodies beyond the 12-week time interval: association with baseline antiphospholipid antibodies titres. *Int J Lab Hematol.* 2019;41:726-730.
27. Moore GW, Henley A, Greenwood CK, Rangarajan S. Further evidence of false negative screening for lupus anticoagulants. *Thromb Res.* 2008;121:477-484.
28. Gray E. https://www.who.int/biologicals/BS_2244_Lupus_20140702.pdf.
29. Tripodi A, Chantarangkul V, Clerici M, Mannucci PM. Laboratory diagnosis of lupus anticoagulants for patients on oral anticoagulant treatment. Performance of dilute Russell viper venom test and silica clotting time in comparison with Staclot LA. *Thromb Haemost.* 2002;88:583-586.
30. Favalaro EJ, Gilmore G, Arunachalam S, Mohammed S, Baker R. Neutralising rivaroxaban induced interference in laboratory testing for lupus anticoagulant (LA): a comparative study using DOAC Stop and andexanet alfa. *Thromb Res.* 2019;180:10-19.
31. Moore GW. Combining Taipan snake venom time/Ecarin time screening with the mixing studies of conventional assays increases detection rates of lupus anticoagulants in orally anticoagulated patients. *Thromb J.* 2007;5:12.
32. Moore GW, Smith MP, Savidge GF. The Ecarin time is an improved confirmatory test for the Taipan snake venom time in warfarinized patients with lupus anticoagulants. *Blood Coagul Fibrinolysis.* 2003;14:307-312.
33. Parmar K, Lefkou E, Doughty H, Connor P, Hunt BJ. The utility of the Taipan snake venom assay in assessing lupus anticoagulant status in individuals receiving or not receiving an oral vitamin K antagonist. *Blood Coagul Fibrinolysis.* 2009;20:271-275.
34. https://cdn.ymaws.com/www.isth.org/resource/resmgr/subcommittee/tsvt_g_moore.pdf.
35. Arachchilage DR, Mackie IJ, Efthymiou M, Isenberg DA, Machin SJ, Cohen H. Interactions between rivaroxaban and antiphospholipid antibodies in thrombotic antiphospholipid syndrome. *J Thromb Haemost.* 2015;13:1264-1273.
36. van Os GM, de Laat B, Kamphuisen PW, Meijers JC, de Groot PG. Detection of lupus anticoagulant in the presence of rivaroxaban using Taipan snake venom time. *J Thromb Haemost.* 2011;9:1657-1659.
37. Jacquemin M, Toelen J, Schoeters J, et al. The addition of idarucizumab to plasma samples containing dabigatran allows the use of routine coagulation assays for the diagnosis of hemostasis disorders. *J Thromb Haemost.* 2015;13:2087-2092.
38. Jacquemin M, Toelen J, Feyen L, et al. The adsorption of dabigatran is as efficient as addition of idarucizumab to neutralize the drug in routine coagulation assays. *Int J Lab Hematol.* 2018;40:442-447.
39. Exner T, Michalopoulos N, Pearce J, Xavier R, Ahuja M. Simple method for removing DOACs from plasma samples. *Thromb Res.* 2018;163:117-122.
40. Frans G, Meeus P, Bailleul E. Resolving DOAC interference on aPTT, PT, and lupus anticoagulant testing by the use of activated carbon. *J Thromb Haemost.* 2019;17:1354-1362.
41. Kopatz WF, Brinkman HJM, Meijers JCM. Use of DOAC Stop for elimination of anticoagulants in the thrombin generation assay. *Thromb Res.* 2018;170:97-101.
42. Exner T, Ahuja M, Ellwood L. Effect of an activated charcoal product (DOAC Stop™) intended for extracting DOACs on various other APTT-prolonging anticoagulants. *Clin Chem Lab Med.* 2019;57:690-696.
43. Ząbczyk M, Kopytek M, Natorska J, Undas A. The effect of DOAC-Stop on lupus anticoagulant testing in plasma samples of venous

- thromboembolism patients receiving direct oral anticoagulants. *Clin Chem Lab Med.* 2019;57(9):1374-1381.
44. Platton S, Hunt C. Influence of DOAC Stop on coagulation assays in samples from patients on rivaroxaban or apixaban. *Int J Lab Hematol.* 2019;41:227-233.
 45. Cox-Morton S, MacDonald S, Thomas W. A diagnostic solution for haemostasis laboratories for patients taking direct oral anticoagulants using DOAC-Remove. *Br J Haematol.* 2019;187(3): 377-385.

How to cite this article: Tripodi A, Cohen H, Devreese KMJ. Lupus anticoagulant detection in anticoagulated patients. Guidance from the Scientific and Standardization Committee for lupus anticoagulant/antiphospholipid antibodies of the International Society on Thrombosis and Haemostasis. *J Thromb Haemost.* 2020;18:1569-1575. <https://doi.org/10.1111/jth.14846>