EXPERT REVIEWS

The emerging role of multiple antiphospholipid antibodies positivity in patients with antiphospholipid syndrome

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Antiphospholipid syndrome (APS) is an autoimmune disorder characterized by clinical symptoms of vascular thrombosis and/or pregnancy morbidity in the presence of autoimmune antiphospholipid antibodies (aPL). Current laboratory APS criteria include the presence of at least one of the three relevant aPL: lupus anticoagulant, anticardiolipin antibodies and anti-β_2 glycoprotein I antibodies. Therefore, patients could have a single aPL pattern or combinations of aPL. Evidence arising from clinical experience indicates that patients having the highest aPL titer and simultaneous aPL detected by different tests have a worse prognosis and a higher probability of recurrence of the APS clinical features. In recent years, an emerging role of multiple aPL positivity in the identification of high-risk patients with aPL/APS is evident. This paper will review the current knowledge on the clinical relevance of having single or multiple aPL positivity.

Keywords: antiphospholipid antibodies • antiphospholipid syndrome • arterial thrombosis • pregnancy morbidity • venous thrombosis

Current laboratory & clinical criteria for the antiphospholipid syndrome

Antiphospholipid antibodies (aPL) are a heterogeneous family of antibodies detected by diverse coagulation tests and immunologic assays. The first aPL was called lupus anticoagulant (LA) and its diagnosis relies on a set of successive phospholipid-dependent clotting tests. Anticardiolipin (aCL) and anti-β_2 glycoprotein I antibodies (aβ_2GPI) are the other key members and they are measured by different immunologic assays. Most LAs are directed against either β_2GPI or prothrombin, whereas antibodies detected in the aCL assay recognize β_2GPI complexed with cardiolipin or cardiolipin alone [1,2].

Within the field of aPL there are two different groups of patients. One includes subjects having an acute or chronic infection and other disorders such as neoplasms, drug-induced aPL; and the other one include subjects with autoimmune conditions [3,4]. aPL may bind phospholipids directly but most pathogenic aPL require β_2GPI and prothrombin for antibody binding to phospholipids. Positivity on the aCL assay not directed against β_2GPI is frequently found in the setting of several infectious disorders [4]. There are a number of evidences that the most important epitope of aβ_2GPI is situated on the Gly40-Arg43 region in the first domain of β_2GPI [5].

Since the 1980s, the term antiphospholipid syndrome (APS) is used to describe patients characterized clinically by the occurrence of venous or arterial thrombosis in different vascular beds, and/or pregnancy morbidity in association with raised levels of aPL [6,7]. The revised classification clinical criteria for the APS indicates that vascular thrombosis includes one or more clinical episodes of arterial, venous or small vessel thrombosis, in any organ or tissue [8]. On the other hand, pregnancy morbidity includes: one or more

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Table 1. Family of antiphospholipid antibodies and its estimated prevalence in antiphospholipid syndrome.

Antibody	Prevalence (%)	Ref.
Included in laboratory APS criteria		
Lupus anticoagulant	~55	[6]
Anticardiolipin IgG/IgM	~80	[6]
Anti-β_2 glycoprotein I IgG/IgM	~40	[18,21,66]
Not included		
Anticardiolipin IgA	10–40	[26,27]
Anti-β_2 glycoprotein I IgA	10–40	[26,35]
Anti-prothrombin	~30	[48]
Anti-phosphatidylserine/prothrombin	~50	[69]
Anti-phosphatidylethanolamine	~50	[37,41]
Anti-phosphatidylserine	~60	[37]
Anti-phosphatidic acid	~70	[37]
Anti-phosphatidylinositol	~70	[37]
Anti-domain I β_2 glycoprotein I	~40	[51]

APS: Antiphospholipid syndrome.

unexplained deaths of a morphologically normal fetus at or beyond the 10th week of gestation, with normal fetal morphology or one or more premature births of a morphologically normal neonate before the 34th week of gestation because of eclampsia or severe pre-eclampsia, or recognized features of placental insufficiency or three or more unexplained consecutive spontaneous abortions before the 10th week of gestation [8].

aPL can occur in isolation or in association with other autoimmune conditions, particularly systemic lupus erythematosus (SLE). APS requires the combination of at least one clinical and one laboratory criterion. The recent consensus statement suggests avoiding classification of APS in less than 12 weeks or more than 5 years separate the positive aPL tests and the clinical manifestation. In addition, the evaluation of coexisting inherited and acquired thrombosis risk factors in APS patients is highly recommended. Consequently, patients fulfilling APS criteria must be stratified according to the presence or absence of other contributing causes of thrombosis [8].

The new criteria are very useful in research and besides in real practice because they limit the inclusion of a diverse group of patients. In addition, the assessment of concurrent prothrombotic factors provides a tool for risk stratification that is in fact very important because some of them may be treatable.

The detection of persistently elevated levels of aPL is a requisite laboratory feature for the diagnosis of APS. The positivity for at least one aPL test, LA and/or IgG/IgM aCL and/or aβ_2GPI, must be detected. They have to be found on two or more occasions at least 12 weeks apart. In the 2006 updated APS criteria [8], it is advised to classify APS patients for clinical studies into one of the following categories: I, more than one laboratory criteria present (any combination); IIa, LA present alone; IIb, aCL present alone; IIc, aβ_2GPI present alone [9,10].

The catastrophic APS is a rare (<1%) variant of APS characterized by microthrombi in multiple organs and associated systemic inflammatory response syndrome and a life-threatening clinical course [11,12]. Sometimes aPL may clinically start with a hemorrhagic syndrome when a severe thrombocytopenia, or an acquired thrombocytopathy, factor VIII inhibitor, or prothrombin deficiency is present [13].

Clinical evidence on the thrombotic risk of the different aPL

LA, aCL & aβ_2GPI

The list of the different aPL evaluated in several studies in the APS field and an estimate of its prevalence are shown in Table 1. However, up to now only the three first antibodies are included in the laboratory classification criteria for APS. They have been the most studied antibodies through the years. In a large cohort of 1000 patients with APS, the prevalence of LA and aCL were 88 and 54%, respectively [6]. In addition, an increasing amount of clinical evidence coming from several retrospective but also prospective studies indicates that they correlated with the main clinical features of the APS [14–19].

Among them, LA better correlates with thrombosis and pregnancy morbidity than aCL and aβ_2GPI do [14]. The last systematic review published in 2003 [15] included data from 25 evaluable studies recruiting 4184 patients and 3151 controls. Overall, the review established LA is a strong risk factor for thrombosis, irrespective of the site and type of thrombosis, and for the presence of SLE. aCL were not such strong risk factors for thrombosis as LA, and only 50% of their associations with thrombosis reached statistical significance. Though, when IgG aCL titers are above 40 units, significant associations were found. Data from a prospective study showed that subsequent thrombo-occlusive events and death after focal cerebral ischemia associated with IgG aCL may occur sooner and more frequently with GPL >40 [20]. In addition, in a 4-year prospective study from the Italian Registry [16] an IgG aCL titer >40 units was an independent predictor of thrombosis in 360 patients with LA and/or aCL. Most of the authors agree that aβ_2GPI are closely associated with thrombosis and other clinical features of the APS [14,18,21,22]. In other systematic review performed by the same authors, the clinical role of aβ_2GPI and anti-prothrombin antibodies in 5102 patients and 1973 controls from 32 evaluable studies were included [17]. The aβ_2GPI assay shows higher specificity than aCL in the recognition of patients with APS. Since the first studies [18,21], IgG aβ_2GPI is the only isotype significantly associated with a history of venous thromboembolism.

The titer and also the isotype of aPL is very important. Only medium and high but not low aPL titer are related to the thrombotic risk. It is important to remember that low titer aPL can be transient and in numerous occasions linked to infectious disorders [23]. Some authors have claimed that IgM

aCL tends to be less important in the APS and that the IgM isotype must be excluded from the laboratory APS criteria [24]. Nevertheless, its measurement is still within the laboratory criteria. Probably, the lesser importance of IgM is in the lower range of positivity. When titer is high and persistent likely the diagnostic power is greatly increased.

IgA isotype

Regarding the IgA isotype, most studies indicate that they are usually detected together with either IgG and/or IgM isotypes [25]. This is true for the aCL as well as for the aβ_2GPI. In addition, titers of IgA isotype seem to be lower than those for the other isotypes. Some studies [26,27] found association of IgA aCL with thrombosis but others did not [28,29]. Afro-American, Afro-Caribbean and Japanese patients have been reported showing the highest prevalence of IgA aCL [30,31]. IgA aβ_2GPI have been reported with high prevalence in SLE patients, and showed a high association with thrombosis in this group of patients [26,32,33]. However, a lack of consensus is also demonstrated because other reports failed in demonstrating such an association [34,35]. Isolated positivity of IgA aCL and aβ_2GPI is rare. Consequently, its utility is largely restricted to those patients with a strong suspicion of APS but negative conventional aPL tests [25] and also in those groups of patients having IgA as the dominant isotype.

aPL to non-cardiolipin antigens

The clinical relevance of antibodies directed against negatively charged phospholipids such as antibodies against phosphatidylserine, phosphatidic acid and phosphatidylinositol is uncertain. It is important to keep in mind that aCL greatly cross-react to antibodies against phosphatidylserine and phosphatidylinositol. Some authors have reported that testing for these antibodies may help identify patients with APS who had repeatedly negative results for the aPL included in the laboratory APS criteria [36]. In a more recent study assessing the performance of commercially available kits for these antibodies, the authors conclude that testing for multiple phospholipid specificities does not improve diagnostic yield for APS [37]. The prevalence rates of these antibodies are shown in Table 1. With respect to antibodies to phosphatidylethanolamine, they can be found as the only positive aPL in patients with a high suspicion of APS. Its clinical importance in pregnancy morbidity is not clear because results from different studies are inconclusive or even contradictory [25,38,39]. A large study underlines the strength of the association between the presence of antibodies to phosphatidylethanolamine and thrombosis and suggests their measurement in thrombotic patients, particularly when conventional aPL are not detected [40]. However, other study failed to find such an association [41].

Antibodies to prothrombin

There are two types of antibodies to prothrombin, one reacting with prothrombin alone (aPT) and the other reacting with the complex between phosphatidylserine and prothrombin (aPS/PT). Recently, it was demonstrated that antibodies detected by the aPT and aPS/PT assays are in fact two distinct populations with partial overlapping activity [42]. The results of a 15-year longitudinal study showed the IgG aPT to be the most useful predictor of thrombosis in patients with SLE [43]. A systematic review of the literature in the last years was recently published [44]. A retrospective design was used in the majority of the studies with very few case–control or prospective clinical studies. Overall, IgG aPT seemed more constantly associated with thrombosis than IgM antibodies. The evaluation of aPS/PT reached significance with thrombosis as a whole and mainly with venous thrombosis.

Elevated levels of aPT were found in 34% of patients from a series of 139 SLE patients and were significantly associated with deep venous thrombosis [45]. A retrospective study recruiting 233 patients with LA and/or aCL demonstrated that aPT were related to venous thrombosis in the univariate but not in the multivariate analysis [46]. In the setting of SLE, patients with aPT had a history of thrombosis more frequently than those without aPT [47].

One study showed that LA and of IgG and/or IgM aPS/PT were independent risk factors for thrombosis and pregnancy loss. aPS/PT, but not aPT, were more frequently found in patients with LA and their association with thrombosis seems to be independent of LA [48]. In other cohort of 295 patients with APS or APS-related diseases, it was found aPS/PT to be highly associated with venous thrombosis (odds ratio [OR]: 7.4 for IgG and OR 2.5 for IgM) and obstetric complications (OR 2.4 for IgG), but not with arterial thrombosis [49].

At the present time, the measurement of aPS/PT and less the detection of aPT might be useful in assessing the thrombotic risk in APS patients. However, more studies (prospective design) are needed before consider its inclusion as laboratory criteria for the APS [25].

Antibodies to domain I of β_2GPI

As the main epitope of pathogenic aβ_2GPI seems to be located in the domain I of β_2GPI, a number of groups have recently focused in evaluating the clinical relevance of specific anti-domain I β_2GPI. The larger study (multicenter) published until now shows in 442 patients all positive for aβ_2GPI that those patients having IgG anti-domain I β_2GPI-positive results were more likely to develop vascular thrombosis or pregnancy morbidity than those having negative results [50]. The prevalence reported was of 55% in APS patients having aβ_2GPI. In that report, IgM was not associated with any of the clinical features of the APS.

In a very recent report, the IgG anti-domain I β_2GPI assay was positive in a significant proportion of seropositive patients with APS, but also in a small proportion (3/40) of seronegative APS patients (clinical features of APS but repeatedly negative in conventional aPL assays) [51]. The evaluation of anti-domain I β_2GPI is very promising, but its clinical relevance must be confirmed in longitudinal, prospective studies to clarify its role [52].

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From single to multiple aPL positivity

The first publications from the 50s reported clinical associations between LA and thrombosis and/or pregnancy losses. Soon, after the evaluation of aCL in the 80s, a number of studies indicated the classical APS clinical features have also linked to aCL. Consequently, both aCL and LA were detected simultaneously in a high number of patients. In the 90s, when aβ_2GPI was included in the aPL panel by several researchers, there were clinical studies with patients carrying LA, aCL and/or aβ_2GPI. In the Sapporo criteria but also in the 2006 criteria for the classification of APS, a single positivity of any of the three aPL tests mentioned before fulfill the serologic criteria [8]. However, the current APS criteria also advised to classify APS patients in clinical studies into four categories taking into account the aPL pattern as mentioned before in this review.

Evidence arising from clinical experience from several groups in the world indicates that patients having the highest aPL titer and also simultaneously aPL detected by different tests have a worse prognosis and a higher probability of recurrence of the APS clinical features.

There is growing published evidence that the risk of thrombosis increases with the number of positive tests in APS patients and also in asymptomatic carriers of persistent aPL. The concept of triple positivity conferring a higher risk for thromboembolic events and pregnancy morbidity has been proposed by Pengo *et al.* [53]. In their proposal, the triple positivity comprises LA + aCL + aβ_2GPI, the double positivity involves LA negative with positive aCL and aβ_2GPI of the same isotype, and single positivity when only LA, or aCL, or aβ_2GPI were positive. The proposal was based on several retrospective and prospective studies in patients with or without SLE [54–57].

In 2010, a large and prospective multicenter study was carried out in Italy. One hundred and sixty definite APS patients were enrolled and all of them tested positive for LA/aCL/aβ_2GPI. This cohort of triple aPL-positive APS patients had a cumulative incidence of thrombosis of 12.2, 26.1 and 44.2% after 1, 5 and 10 years of follow-up, respectively [58]. Notably, recurrence remained high despite the use of oral anticoagulant therapy. In 2011, it was reported that the annual incidence of the first thrombotic event was 5.3% per year among 104 triple aPL-positive patients (mean age of 45 years) with no history of thrombosis (aPL carriers) followed-up for a mean of 4.5 years [59]. In comparison, in normal white population at 35–55 years of age, the annual rate was 0.4% [60]. In 125 single aPL-positive carriers with a mean age of 41 years, the annual rate of first cardiovascular events was 1.36% [61].

In our experience as well as in some published studies [54,62], the single positivity of LA (single profile) is not associated with a high risk for thrombosis or other clinical features of APS.

These findings strengthen the concept that the triple aPL-positive population has a more severe course of the disease. It is likely that the presence of triple positivity is due to antibodies directed the first domain of β_2GPI. Some data indicate that antibodies directed against the epitope situated on the Gly40-Arg43 region in the first domain of β_2GPI highly correlate with thrombosis and cause LA activity [5]. β_2GPI undergoes a conformational change after binding to phospholipid surfaces and thus uncover the crucial sequence which is necessary to allow the binding of pathogenic aβ_2GPI [63]. Antibodies to β_2GPI not directed against domain I seem to be non-pathogenic. The detection of anti-domain I β_2GPI antibodies seems to be a promising biomarker in diagnosis/risk assessment of APS [52]. In a recent cross-sectional study, it was demonstrated that anti-domain I β_2GPI antibodies correctly classify patients at risk [64]. These antibodies were significantly more frequent and titers were higher in patients having IgG aβ_2GPI and triple (n = 32) as compared with double (n = 23) or single (n = 10) positivity. In addition, negative and positive values of anti-domain I β_2GPI antibodies were confirmed after 12 weeks. The same group has further evaluated the presence of antibodies to domain 4/5 of β_2GPI in the same group of patients and found that patients in the single positive group have significantly higher values of anti-domain 4/5 β_2GPI antibodies with respect to triple and double positive groups [65]. However, only 5 out of 65 patients had positivity of this type of antibodies.

Testing for other aPL such as aPT and aPS/PT has been proposed to be relevant to APS [25,43]. Studies on the significance of aPT in thrombosis have shown controversial results [17]. We assessed the contribution of aβ_2GPI and aPT to the thrombotic risk in a cohort of 194 consecutive patients with persistent LA and/or aCL [66]. Patients were prospectively followed during a median follow-up of 45 months (range 3–120). A total of 39 patients had one episode of thrombosis during follow-up. In 28 cases, there were recurrences of thrombosis but in 11 individuals the thrombotic event was the first. The overall incidence of venous/arterial thrombosis was 5.6% per patient-year. Our findings showed that the triple positivity for LA, IgG aβ_2GPI, concurrent with the presence of IgG aPT, gave the highest annual rate of thrombosis (8.4%), which was statistically significant in multivariate analysis (OR: 2.6; 95% CI: 1.35–5.01). The presence of IgM aPL was not a predictor of thrombosis. As in a more recent paper [59], the male sex was also independently associated with an increased risk of thrombosis.

In a recent retrospective study, a large series of SLE patients was analyzed, and assessed the potential clinical usefulness of combining routinely tested aPL with new aPL specificities in an attempt to find a profile that will identify patients at higher risk of APS. The study included 230 patients with SLE and all of them were tested for LA, aCL, aβ_2GPI, aPT, aPS/PT and antibodies to phosphatidylethanolamine. Among the 23 possible combinations of the six aPL tested, LA + aβ_2GPI + aPS/PT had the best diagnostic accuracy for APS as a whole, and for both thrombosis and pregnancy loss. When comparing it with the combination suggested by the current criteria and all the other tested combinations, triple positivity for LA/aβ_2GPI/aPS/PT had the best diagnostic performance in terms of specificity and predictive value in this SLE cohort [67].

In a very recent study, the contribution of IgG and/or IgM aPS/PT in the context of clinical criteria for APS was evaluated [68]. The total cohort included 160 APS patients and 128 patients with clinical criteria for APS but tested negative for the three aPL included in the last APS laboratory consensus criteria. Both IgG and IgM aPS/PT were significantly more frequent in triple than in double and in single positivity. According to multivariate analysis, IgG and/or IgM aPS/PT were independent risk factors for LA. In addition, aPS/PT were found in 9.4% of the patients with clinical features of APS and negative classical aPL results versus 2% of healthy controls. They were also significantly more frequent in the thrombosis with respect to the pregnancy morbidity group. These data attribute a clinical relevance to both IgG and IgM aPS/PT. In particular, the significant prevalence of aPS/PT in conventional aPL-negative patients suggests including them as additional laboratory criterion for APS.

Preliminary data from our laboratory also show the contribution of aPS/PT to the diagnosis of APS (unpublished data). Until now we have recruited 50 APS patients with thrombosis, 35 with positive aPL but none clinical criteria for APS and 50 patients with thrombosis but negative aPL tests. The prevalence was significantly greater in the APS group (45%) than in the second group (15%) and in the group with thrombosis but negative aPL (5%). Preliminary analysis after a median follow-up of 20 months seems to indicate that aPS/PT is frequently found in APS patients with thrombosis but not an independent predictor of thrombosis.

With the objective to ascertain the value of aPS/PT for APS diagnosis, a cross-sectional multicenter study was carried out [69]. The initial study comprised 247 subjects and 214 were included in the confirmation study. aPS/PT were significantly more prevalent in APS patients (51, 47%) than in those without (9, 12%) in the initial and also in the confirmation study, respectively. Sensitivity, specificity, positive and negative likelihood ratio of IgG aPS/PT for APS diagnosis in the initial study was 51, 94, 8.9 and 0.5%, respectively. The same parameters were 47, 93, 7.2% and 0.6% in the confirmation study. The main conclusion is that aPS/PT is a laboratory parameter that may help in the diagnosis of APS.

In the last years, some score systems have been proposed to quantify the risk of clinical events in APS [70–72]. The 'Risk scale' model [70] is based on aPL positivity (LA, aCL, $a\beta_2$GPI), their titers and the methodology used for LA. The second model called the aPL score (aPL-S) was designed testing multiple aPL (LA, aCL, $a\beta_2$GPI, aPS/PT) to evaluate its efficacy for the diagnosis of APS and predictive value for thrombosis [71]. For each assay, a different score weighted on the relative risk of having clinical manifestations of APS was assigned. Two groups of patients were analyzed. In the first group of 233 patients, they evaluated the aPL profiles and in the second group of 411 patients the predictive value of aPL-S for thrombosis was evaluated. The occurrence of APS clinical features increased in agreement with increasing aPL-S. It was concluded that the aPL-S is a useful quantitative index for diagnosing APS and may be a predictive marker for thrombosis in the setting of autoimmune diseases. The usefulness of this score was independently validated in a cohort of 211 SLE patients [73]. The third score is called GAPSS (Global APS Score) and it takes into account the aPL profile (LA, aCL, $a\beta_2$GPI, aPS/PT) an also the conventional cardiovascular risk factors and the autoimmune antibodies profile [72]. A total of 211 SLE patients were used to develop and validate the score. The first group of 106 patients was utilized to develop the GAPSS assigning the risk factors identified by multivariate analysis weighted points. The second set of 105 patients was used to validate the score showing higher values of GAPSS in those patients with a history of thrombosis and/or pregnancy loss compared to those without clinical events. In an independent prospective multicenter study of 137 patients followed-up for a mean duration of 43 months, the GAPSS could predict thrombosis in patients with aPL and associated autoimmune diseases [74]. In the setting of primary APS, the GAPSS values were higher in patients who experienced thrombosis alone when compared with those with pregnancy loss alone. Higher values of GAPSS were also seen in patients who experienced recurrent thrombosis [75]. The GAPSS was also a valid tool for accurate prediction of vascular events in SLE patients with aPL as demonstrated in a prospective study of 51 patients with SLE followed-up for a mean of 33 months [76]. Therefore, the score models seem to be clinically useful but results must be confirmed in larger studies.

Table 2. Personal proposal of classification criteria for the antiphospholipid syndrome according to the antiphospholipid antibodies profile.

Definite APS	Patients with at least triple aPL positivity (high-risk group)
Probable APS	Patients with double aPL positivity (medium-risk group)
Possible- or non-APS	Patients with single aPL positivity (low-risk group)

aPL: Antiphospholipid antibodies; APS: Antiphospholipid syndrome.

Pengo *et al.* [53] proposed to consider only patients with triple positivity (LA + aCL + $a\beta_2$GPI) as definite APS (thrombotic and obstetric). However, taken into account the increasing knowledge and the possibility of different combinations of relevant aPL, it is now suggested that the definition of APS should include a different clinical risk to develop APS-related events. Table 2 shows my personal proposal of classification criteria for the APS according to the aPL profile. Likely this risk stratification would improve the clinical management of patients with aPL. The decision of long-term treatment may be influenced by the type of aPL involved. Some recent evidence indicates that patients with primary or SLE-associated APS and at least triple positivity for aPL are at risk of developing future thromboembolic events.

Expert commentary

It is now well recognized that the thrombosis risk depends on the aPL profile. Patients with multiple aPL positivity have the

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higher risk of developing thromboembolic events. Current diagnosis of APS relies on three aPL laboratory tests that may be positive in a number of combinations. In addition, new tests such as aPS/PT and anti-domain I aβ_2GPI are now increasingly measured in patients with suspicion of APS and thus more aPL combinations are possible. Further, it is important to distinguish which of the possible combinations of aPL tests is best marker in assessing the risk of aPL-related clinical features. Likely the criteria for APS should include clinical risk stratification mainly when taking decisions on the clinical management.

Five-year view

At the present time, there are some groups in the world carrying out prospective studies in order to evaluate the contribution of conventional and new aPL as well as the aPL profiles (particularly multiple profile) to assess the aPL-associated thrombotic risk. An ongoing international project to study the natural course of at least 2000 patients with aPL with or without systemic autoimmune diseases over 10 years has begun in 2012 and key information will be available from this worldwide project (APS ACTION) [77]. It is desirable that some current questions will be answered in the near future.

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Key issues

- The antiphospholipid syndrome (APS) is an autoimmune disease characterized clinically by the occurrence of either venous or arterial thrombosis in different vascular beds, and/or pregnancy morbidity in the presence of antiphospholipid antibodies (aPL).
- The positivity for at least one recommended aPL test: lupus anticoagulant and/or IgG/IgM anticardiolipin and/or anti-β_2 glycoprotein I antibodies are part of the current laboratory APS criteria.
- There is no enough clinical evidence that the measurement of other members of the aPL family could be essential in the APS diagnosis.
- At the present time, the measurement of aPS/PT and anti-domain I anti-β_2 glycoprotein I antibodies might be useful in assessing the thrombotic risk in APS patients.
- Evidence arising from clinical experience indicates that patients having the highest aPL titer and simultaneously aPL detected by different tests have a worse prognosis and a higher probability of recurrence of the APS clinical features.
- The risk of thrombosis increases with the number of positive tests in APS patients and also in asymptomatic carriers of persistent aPL.
- The definition of APS should include a risk stratification to develop APS-related events according to the aPL profile.
- Patients with primary or systemic lupus erythematosus-associated APS and multiple aPL are at risk of developing future thromboembolic events.

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