Antiphospholipid Antibody Syndrome

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KEYWORDS

- Antiphospholipid syndrome • Antiphospholipid antibodies
- Recurrent pregnancy loss • Fetal demise • Unfractionated heparin
- Complications of pregnancy

KEY POINTS

- Antiphospholipid antibodies (aPLs) are acquired antibodies directed against negatively charged phospholipids, a group of inner and outer cell membrane antigens found in mammals.
- Obstetric antiphospholipid antibody syndrome (APS) is diagnosed in the presence of certain clinical features in conjunction with positive laboratory findings.
- Although obstetric APS was originally reported in association with slow progressive thrombosis and infarction in the placenta, it is most often associated with a poor obstetric outcome.
- Several pathophysiologic mechanisms of action of aPLs have been described.
- The most common histopathologic finding in early pregnancy loss has been defective endovascular decidual trophoblastic invasion.
- Treatment with heparin and aspirin is emerging as the therapy of choice, with approximately 75% of treated women with RPL and aPL having a successful delivery, compared with less than 30% without treatment.

INTRODUCTION

Antiphospholipid antibodies (aPLs) are acquired antibodies (immunoglobulin [Ig] G, IgM, and/or IgA isotypes) that react against negatively charged phospholipids. They were originally associated with a slow progressive thrombosis and infarction in the placenta[1] and thus have been classified as thrombophilic factors. aPLs should additionally be classified as autoimmune factors when considering implantation and

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pregnancy because of the complex nature of the interaction of aPLs with target tissues. They have been shown to inhibit the release of human chorionic gonadotropin from human placental explants; to block in vitro trophoblast migration, invasion, and multinucleated cell formation; inhibit trophoblast cell adhesion molecules; and to activate complement on the trophoblast surface, inducing an inflammatory response.[2–5]

Antiphospholipid syndrome (APS) is an autoimmune condition characterized by the production of aPL combined with certain clinical features. The presence of aPL (including anticardiolipin [aCL] and lupus anticoagulant [LAC]) during pregnancy is a major risk factor for adverse pregnancy outcome.[6] In large meta-analyses of studies of couples with recurrent pregnancy loss (RPL), the incidence of APS was between 15% and 20% compared with about 5% in nonpregnant women without a history of obstetric complications.[7,8] It is not yet understood how aPLs arise in patients with APS. Genetic factors and infection may play a role. Family studies suggest a genetic disposition to APS, either when it presents as a primary condition or when it is seen in the context of systemic lupus erythematosus (SLE). This genetic disposition is accounted for, at least in part, by the major histocompatibility complex. The antibodies generated in patients with APS seem to recognize epitopes on phospholipid-binding proteins, unlike the antibodies that arise following infections such as syphilis and Lyme disease, which recognize phospholipids directly.[9]

DIAGNOSIS OF APS

APS is a syndrome that is defined based on both clinical and laboratory criteria. The diagnosis can be based on the presence of the clinical manifestation together with the laboratory detection of abnormal antibodies (**Box 1**). APS is designated as primary in patients without clinical or laboratory evidence of an underlying condition or disease, and secondary when it is associated with other diseases or conditions.[10] SLE is the disorder in which APS is most commonly associated but it may be associated with other conditions.[11,12] **Box 2** lists the conditions associated with secondary APS that may be relevant to obstetricians.

Widespread interest in APS among all areas of medicine led to the First International Symposium on Antiphospholipid Antibodies in 1984. The purpose of this group was to bring together the international research and clinical communities with the goal of clinical and scientific sharing and standardization of APS. Meetings have been held periodically with research and clinical data reviewed and discussed at preconference meetings and committees. An article was published with the latest suggested criteria for the diagnosis of APS with the current version referred to as the Sydney criteria.[13] These criteria should be useful for research investigations because the criteria result in specific diagnoses; however, in the clinical setting, these criteria are not very sensitive. These committee recommendations have been widely published, but the committee is not sanctioned or supported by any governing body such as the American College of Obstetricians and Gynecologists. The committee is primarily composed of rheumatologists, hematologists, and internists, with minor representation from obstetricians.

Clinical Criteria

The clinical manifestations of primary and secondary APS are diverse and may involve most organ systems.[10,14] Based on currently available evidence, vaso-occlusive disease is the pathologic basis for many of the complications of primary and secondary APS. Therefore, preconception counseling of the patient with APS regarding the risks

Box 1
Clinical and laboratory criteria established for the research of definite antiphospholipid syndrome: the Sydney criteria

Note: At least 1 clinical and 1 laboratory criterion must be present for definite APS.

Clinical criteria

1. Vascular thrombosis

   One or more clinical episodes of an arterial, venous, or small vessel thrombosis, confirmed by imaging or Doppler studies or histopathology, without significant evidence of inflammation in the vessel wall.

2. Obstetric morbidity

   a. One or more unexplained demise of a morphologically normal fetus at or beyond 10 weeks of gestation, or

   b. One or more premature births of a morphologically normal neonate at or before 34 weeks of gestation, caused by severe preeclampsia or severe placental insufficiency, or

   c. At least 3 unexplained, consecutive miscarriages of less than 10 weeks of gestation. Known factors associated with recurrent miscarriage, including parental genetic, anatomic, and endocrinologic factors, should be ruled out.

Laboratory criteria

1. aCL IgG and/or IgM in blood, present in medium or high titers (greater than 40 GPL or MPL or greater than the 99th percentile) on 2 or more occasions at least 12 weeks apart, measured by a standardized ELISA.

2. Anti-β2GP1 antibody of IgG and/or IgM isotype in blood (greater than the 99th percentile) on 2 or more occasions at least 12 weeks apart, measured by a standardized ELISA.

3. Lupus anticoagulant present in plasma, on 2 or more occasions at least 12 weeks apart, detected according to the guidelines of the International Society on Thrombosis and Hemostasis, which include the following steps:

   a. Prolonged phospholipid-dependent coagulation using a screening test such as the aPTT, kaolin clotting time, dilute Russell viper venom time, and dilute prothrombin time

   b. Failure to correct the prolonged coagulation time on the screening test by mixing with normal plasma

   c. Shortening or correction of the prolonged coagulation time on the screening test by the addition of excess phospholipid or platelets

   d. Exclusion of other coagulopathies (eg, factor VIII inhibitor) or heparin

Adapted from Werner E, Lockwood C. Thrombophilias and Stillbirth. Clin Obstet Gynecol 2010;53:617–27; with permission.

for serious medical problems is essential. In women referred to reproductive endocrinologists with APS and histories of early loss, the prevalence of prior thromboembolic events is low. However, these patients are still at risk for thromboembolic events. Estrogen-containing oral contraceptive pills should be avoided in women with repeated positive aPL.[15] Furthermore, women should be counseled that a single low-dose aspirin (81 mg) per day may decrease this future risk for thromboembolic event, as suggested by Silver and colleagues.[16] Thus, it is important for clinicians to be cognizant of the red flags, which are indications to perform diagnostic testing for the LAC and aCL antibodies (**Box 3**).

Box 2
Classification of the APS relevant to obstetricians

I. Primary APS
- Obstetric antiphospholipid syndrome

II. Secondary APS

A. Autoimmune disease
- SLE
- Rheumatoid arthritis
- Sjögren syndrome
- Systemic sclerosis
- Diabetes mellitus
- Crohn disease
- Autoimmune thyroid disease

B. Malignancies
- Carcinoma of ovary and cervix
- Lymphoma
- Leukemia

C. Drug-induced conditions
- Oral contraceptives

D. Infectious diseases
- Syphilis
- Human immunodeficiency virus infection

Box 3
Indications to identify LAC and aPL in obstetric patients

- RPL
- Unexplained second-trimester or third-trimester loss
- Fetal demise
- Early-onset, severe preeclampsia
- Pregnancy-related thrombosis (venous or arterial)
- Severe IUGR
- Autoimmune or connective tissue disease
- False-positive serologic test for syphilis (Venereal Disease Research Laboratory or rapid plasma reagin)
- Prolonged coagulation studies
- Positive autoantibody tests

Laboratory Criteria

Lupus anticoagulant is an immunoglobulin (usually IgG, IgM, or both) that interferes with one or more of the phospholipid-dependent tests of in vitro coagulation.[17] The name is a misnomer in 2 ways. Although it is called an anticoagulant, patients with LAC more frequently have a hypercoagulable state. Second, LAC is frequently found in patients without SLE. The most common tests that have been used to identify the LAC include the activated partial thromboplastin time (aPTT), the kaolin clot time (KCT), the dilute Russell viper venom time (dRVVT), and the plasma clot time (PCT).

The advantages, disadvantages, and sensitivity of the screening procedures used for LAC are listed in **Table 1**. aPTT reagents vary widely in their sensitivity to LAC because of variations in the phospholipid present in the reagent. It is not a reliable test to use in pregnancy because coagulation proteins, which increase during pregnancy (fibrinogen, factor VIII, von Willebrand factor, factor VII, and factor X), may mask the LAC. The KCT is reliable and sensitive but cannot be used as a confirming test. The dRVVT is a sensitive test and, because the snake venom activates factor X, this test is less affected by pregnancy.[18]

Regardless of which test is used, there are 3 steps that are necessary to identify an LAC. Once a prolonged in vitro coagulation test has been identified, it is necessary to document the presence of an inhibitor. Most commonly, mixing studies are done with pooled normal platelet-poor plasma and patient plasma in a 1:1 ratio. In general, a marked prolonged aPTT corrects to nearly the value of normal plasma (within 5 seconds) if the abnormal coagulation test was caused by a factor deficiency. To confirm that the persistently prolonged aPTT is caused by the LAC, a confirmatory test, such as the platelet neutralization test, must be performed. Frozen, thawed

Table 1
Screening tests for lupus anticoagulants

Test	Variables Affected by Pregnancy	Advantages	Disadvantages	Sensitivity
aPTT	Increased factor VIII may mask	Readily available, easily automated, can be used in PL confirmatory step	Reagents vary widely in sensitivity to LAC	Least
KCT	Not significantly affected by pregnancy	Very sensitive to LAC if patient is on oral anticoagulants	Very sensitive to residual platelets; not readily automated; cannot use in PL confirmation step	Most
dRVVT	Not significantly affected by pregnancy	Easy to perform; readily available	Manual techniques, affected by heparin or oral contraceptives	Good
PCT	Increased factor VII may blunt LAC effect	Requires no reagents or equipment	Must be performed on freshly drawn blood; manual technique	Good

Adapted from Branch DW, Silver RM, Blackwell JL, et al. Outcome of treated pregnancies in women with antiphospholipid syndrome: an update of the Utah experience. Obstet Gynecol 1992;80:614–20; with permission.

platelets contain and release excess phospholipid, which is added to the patient plasma. The LAC is absorbed to the phospholipid and the in vitro coagulation test returns toward normal.[17]

aPLs are acquired antibodies (IgG, IgM, and/or IgA) that belong to the large family of antibodies that react against negatively charged phospholipids, including cardiolipin (diphosphatidyl glycerol), phosphatidylserine, phosphatidylinositol, and phosphatidylglycerol. In 1983, Harris introduced a solid phase immunoassay for cardiolipin.[19] Two years later, he developed an enzyme-linked immunosorbent assay (ELISA) for cardiolipin, which remains the gold standard. In this immunoassay, cardiolipin, phosphatidylserine, and other phospholipids are used as coating antigens on microtiter plates. There have been 3 international workshops to standardize reporting. Standards for cardiolipin IgG, IgM, and IgA are commercially available through the Antiphospholipid Laboratory, Inc. These standard sera have been assigned numerical values based on their ability to bind 1 μg of cardiolipin and the units are reported as IgG phospholipid units (GPL). Based on the original studies and recommendations of Harris,[7] positive values are considered greater than or equal to 20 GPL, greater than or equal to 20 IgM phospholipid units (MPL), and greater than or equal to 30 IgA phospholipid units (APL), respectively. It has been recommended that values be reported in a semiquantitated fashion such as negative, borderline, positive, and high positive. Positive tests should ideally be confirmed in 6 to 8 weeks. In general, aPLs decline during pregnancy, perhaps because of increase maternal blood volume and dilution. However, aPLs may increase in as many as 20% of women during pregnancy.

A major target molecule for aPL binding seems to be beta-2 glycoprotein-1 (β2GP1) present on the surface of trophoblastic cell membranes.[20,21] β2GP1 is a cationic plasma protein that is bound to phosphatidylserine, which is exposed on the surface of trophoblastic cell membranes undergoing syncytial formation. Although the physiologic role of β2GP1 is unclear, the molecule seems to inhibit thrombosis by reducing the conversion of prothrombin to thrombin on platelets and inhibiting the activation of the intrinsic coagulation cascade.[22] β2GP1 was added to the criteria for the definitive diagnosis of APS in the Sydney criteria[13]; it is thought to be a more specific indicator of APS but several studies indicate that it is less sensitive than aCL or aPL for the diagnosis of APS.

OBSTETRIC COMPLICATIONS OF APS

Determining the diagnosis of APS in an individual patient requires both laboratory and clinical criteria to be met. The most recent compelling data for a link between aPLs and pregnancy complications come from studies in women with recurrent miscarriage,[23,24] although data associated with fetal loss, severe preeclampsia before 34 weeks' gestation, and severe placental insufficiency are also increasing.[25]

Recurrent Pregnancy Loss

RPL has been defined as 2 or 3 or more consecutive losses before the 20th week of gestation with the same partner, and affects up to 2% to 4% of pregnancies.[26,27] However, requiring 3 failed pregnancies before initiating a work-up may offer little additional clinical insight compared with testing after the second loss.[27,28] The risk of miscarriage in subsequent pregnancies is 30% after 2 losses and 45% after 3 losses in patients without a history of live birth, therefore testing after 2 consecutive losses may spare the affected couple the difficulty of dealing with an additional failed pregnancy, and may minimize excessive and unproductive evaluation. The American

Society of Reproductive Medicine recently defined RPL as 2 or more failed clinical pregnancies as documented by ultrasonography or histopathologic documentation of products of conception to address this point, and recommended a thorough evaluation of RPL after two or more clinical losses.[27] It has been observed that circulating aPLs is the main risk factor in 7% to 25% of early RPL (loss in the first trimester), whereas prevalence studies show that between 1% and 5% of patients have LAC.[29,30] The failure rate of pregnancies when APS is strictly defined as aPL levels of greater than the 99% of a normal population is estimated to be up to 90% (52% of early miscarriages and 38% of late fetal loss). Patients positive for aPL with a first unexplained pregnancy loss before the 10th week of gestation are likely to experience a high-risk second pregnancy.[31] Although all 3 of the laboratory criteria for diagnosis of APS (the presence of LAC, increased levels of aCL antibodies, and increased levels of β2GP1[13]) have been linked with RPL, the risk varies with the different types of antibodies. For example, the presence of aCL antibodies is associated with an odds ratio (OR) of 22.6 (95% confidence interval, 5.7–8.9) for subsequent pregnancy loss, and the presence of β2GP1 antibodies increases the chance of recurrent miscarriage from 6.8% to 22.2% compared with women with LAC or aCL.[32]

As the literature confirmed the strong association between aPL antibodies and RPL, and as a better understanding of the pathogenic concepts of APS and fetal loss have been reported,[33] the obvious results have been attempts to use this knowledge to develop therapies for this common complication of pregnancy. Although low-dose aspirin plus heparin has been shown to reduce pregnancy loss rates in patients with RPL, the optimal dose and regimen continue to be investigated as recent reports indicate.[34,35] Based on these recent studies, it is becoming clear that, although live birth rates are improving with therapy, older studies suggest that these patients are still at risk for late pregnancy complications including stillbirth, early-onset preeclampsia, and intrauterine growth restriction (IUGR).[36,37]

Fetal loss

Late fetal loss (after 10 weeks of pregnancy) associated with aPL antibodies is one of the diagnostic criteria for APS,[13] with aCL antibodies and β2GP1 antibodies, but not LAC, being strongly associated with intrauterine fetal death.[32,38,39] Late fetal loss is also more common as the number of aPL antibody tests become positive. In a recent report from Italy involving 97 pregnancies in 79 patients with APS and without hereditary thrombophilia, triple positivity conferred a risk of late fetal loss of 52.6% compared with a loss rate of 2.2% when only 2 tests were positive.[40] This observation again highlights the potential importance of other aPLs as a part of the diagnostic evaluation for RPL.

Information regarding the association between APS and stillbirth (defined as an intrauterine fetal death at a minimum gestational age of 20 weeks) is less well defined, and only a few articles have specifically commented on APS and stillbirth. Therapy for patients with a prior stillbirth and APS with low-dose aspirin and heparin is commonly used but not supported by large-scale randomized controlled trials.[41]

Severe preeclampsia before 34 weeks

Another clinical criterion that defines APS is a preterm birth resulting from severe preeclampsia at or before 34 weeks of gestation, and different methodologies have been used to document this association. When using the International Classification of Diseases, Ninth Revision, Clinical Modification (ICD-9-CM) code for the aPL syndrome (795.79) in a hospital discharge dataset to investigate the presence of APS in a large (141,286) group of women who delivered in Florida in 2001, APS had an

increased adjusted OR for preeclampsia and eclampsia at any gestational age of 2.93 (95% confidence interval, 1.51–5.69).[42] A systematic review and meta-analysis of 64 full-text studies that yielded 12 studies for analysis concluded in 2010 that the pooled OR for the association of aCL antibodies alone with preeclampsia was 2.86 (95% confidence interval, 1.37–5.98), and for severe preeclampsia (95% confidence interval, 2.66–46.75) was 11.15.[43]

This study again reported preeclampsia at all gestational ages and made no recommendations regarding screening for aCL antibodies in the presence of preeclampsia. A recent review in 2011 of APS and preeclampsia reported a rate of 20% of patients with severe preeclampsia before 34 weeks having at least 1 aPL test positive compared with 6% in late-onset (after 34 weeks' gestation) preeclampsia.[44] These results compare favorably with controlled cohort studies reviewed in that article, and the investigators conclude that, in the patient with severe preeclampsia at less than 34 weeks' gestation, testing should be done for LAC, aCL antibodies, and β2GP1 antibodies. Reinforcing the concern expressed earlier, Branch and colleagues[45] observed that 50% of women with APS develop preeclampsia despite treatment of early-onset pregnancy complications. In the past, many of these patients were treated with steroids, which have been shown to increase the frequency of gestational hypertension, gestational diabetes, and preterm delivery.[46,47] More recent studies that used heparin for the treatment of women with APS have not reported the same high frequency of late pregnancy complications.[34,35,48,49]

Severe Placental Insufficiency

Severe placental insufficiency, another clinical criterion of the APS, is the least well-defined criterion, and usually includes IUGR and abruptio placenta at or before 34 weeks' gestation. Using the 2001 Florida dataset described earlier, placental insufficiency (as defined by ICD-9-CM codes) presented an adjusted OR of an association with APS of 4.58 (95% confidence interval, 2.00–10.51).[42] Older studies reported IUGR to occur in approximately one-third of patients diagnosed with APS,[45,50] and aCL antibodies are significantly associated with IUGR as well.[38,51] Because IUGR is most commonly seen in the presence of preeclampsia, causality is difficult to establish.

Although there are no prospective trials studying the association between abruptio placenta and APS, Alfirevic and colleagues[39] noted an association between abruptio and aCL IgG antibodies in a systematic review in 2002.

Obstetric complications abound in the presence of APS, but therapeutic recommendations are either incomplete or inadequate. However, results from European registries of infants born to mothers with APS seem to fare well with no cases of neonatal thrombosis[52] and rare long-term complications. It is hoped that advances in understanding of the pathophysiology of APS will lead to more successful management schemes and therapy.

PATHOPHYSIOLOGY OF APS

The diagnosis of APS requires both the presence of aPL and specific clinical manifestations, including RPL, fetal demise, and obstetric complications such as preeclampsia and IUGR. The presence of aPL is the most common autoimmune risk factor for a treatable cause of RPL, fetal demise, and early, severe preeclampsia.[53,54] The presence of aPL is not simply diagnostic in obstetric APS, it is also pathogenic. Animal studies have indicated that the passive transfer of aPL promotes fetal loss and placental thrombosis, and also inhibits trophoblast and decidual cell functions

in vitro.[55–57] Cross-species investigations have shown that the exposure of pregnant rats to a purified IgG fraction from women with APS directly inhibits embryo growth.[58] Histologic studies on the placentas from aPL-treated mice showed thrombotic lesions of decidua basalis maternal blood vessels and necrosis of the placental tissues. aPLs interact with phospholipid-binding plasma proteins to cause arterial and/or venous thrombosis in the absence of other conditions that promote blood coagulation. The two most clinically relevant phospholipid-binding proteins that react with aPL autoantibodies are β2GP1 and prothrombin.[53] aPLs impair fetal growth and development via mechanisms of thrombosis, inhibition of normal placentation, and local inflammatory destruction.

Thrombosis and aPLs

The primary hypothesis for the major pathogenic mechanism for aPL-induced pregnancy loss and fetal growth restriction is placental thrombosis. Thrombosis of vessels causes placental infarction with resultant impairment of maternal-fetal nutrient and oxygen exchange. This hypothesis is supported by the finding of thrombosis and infarction in the placentas of women with APS who had miscarriages in the first and second trimesters.[1,59,60] The scientific plausibility for placental thrombosis as a cause of obstetric complications in APS is supported by the evidence that increasing aCL titers are correlated with an increased risk of peripheral thrombosis.[61,62] The presence of LAC seems to cause a higher risk of thrombosis than aCL.[63] IgG from the sera of LAC-positive patients increased thromboxane synthesis in placental explants compared with controls. This relative increase in thromboxane activity would produce a thrombophilic state.[64] Other studies have indicated that aPL could promote thrombosis by interfering with protein C activation[65] and diminishing antithrombin activity.[66]

A major target molecule for aPL binding seems to be β2GP1, which is present on the surface of trophoblastic cell membranes.[20,21] β2GP1 is a cationic plasma protein that is bound to phosphatidylserine (PS), which is exposed on the surface of trophoblastic cell membranes undergoing syncytial formation. Although the physiologic role of β2GP1 is unclear, the molecule seems to inhibit thrombosis by reducing the conversion of prothrombin to thrombin on platelets and inhibiting the activation of the intrinsic coagulation cascade.[22] Polyclonal IgG from the sera of patients with APS binds to β2GP1 on trophoblastic cells in culture with potential thrombogenic consequences.[3,67] However, observations in murine models and humans are not consistent with the obligate involvement of β2GP1 as a natural inhibitor of placental thrombosis in APS. Mice deficient in β2GP1 have smaller litter sizes and placental insufficiency but otherwise have similar reproductive outcomes compared with control mice.[68] In addition, individuals have been identified who are deficient in β2GP1 but do not show the clinical signs of APS.[69]

Another mechanism for aPL-mediated thrombosis may involve a disruption of the normal function of a plasma protein called annexin V.[70] The binding of cationic annexin V molecules to the anionic PL molecules on the surface of the trophoblastic membrane seems to prevent thrombosis by forming a protective protein coat. This protective coat might prevent the activation of the clotting cascade by blocking the binding of activated factor X and prothrombin. In vitro studies indicate that aPL could displace annexin V from the surface of endothelial and trophoblastic cells in culture, thus activating the clotting cascade. This mechanism is supported by the finding that women with aPLs have diminished annexin V binding to the surface of trophoblastic cells of the intervillous space compared with controls.[71]

Mechanisms besides placental vascular thrombosis are likely to play a role in the RPL and pregnancy complications associated with obstetric APS. Histopathologic studies have not always confirmed thrombosis in the placentas from women with obstetric APS.[7,72,73] Meta-analyses examining the association of aPLs with placental thrombosis have evaluated index women with systemic thrombosis who were subsequently tested for aPLs, potentially introducing selection bias into these investigations. There have been no large prospective studies of unselected patients whose aPL status was determined before objective documentation of thrombotic complications. As a result, the temporal association between aPLs and the subsequent development of placental thrombosis has not been definitively documented.[74]

Defective Placentation

Abnormalities of early trophoblast invasion caused by aPLs are a likely pathogenic mechanism in obstetric APS.[75] Experimental models indicate that aPLs can directly retard trophoblast invasiveness, impair trophoblastic cellular differentiation and maturation, and diminish human chorionic gonadotropin secretion.[3,76] The histologic abnormality most frequently observed in APS-associated early pregnancy loss was defective decidual endovascular trophoblast invasion, rather than intervillous thrombosis. Exposure of trophoblastic cell monolayers to aPLs resulted in an increased rate of programmed cell death (apoptosis) and inhibition of syncytial formation.[58,77] The sequential expression of cell adhesion molecules between trophoblastic and decidual cells is involved in trophoblastic invasion during normal placentation. Using an in vitro model of trophoblast invasion, it was recently reported that aPLs might affect placental invasion through an abnormal trophoblastic expression of integrins and cadherins.[78]

These findings provide supportive evidence that aPLs may be pathogenic in obstetric APS by directly impairing normal placentation via a mechanism independent of placental or decidual thrombosis.

Local Inflammatory Events

The maternal immune system is transformed during normal pregnancy to prevent immune rejection of the fetoplacental unit. Acute inflammatory responses generally promote adverse pregnancy outcome and chemokine mediators favor a profile of T helper cells type 2 (Th2) responses in early pregnancy which block acute immune rejection.[45] Complement-mediated immune attack is suppressed in normal pregnancies, resulting in viable delivery by complement inhibitory proteins expressed on trophoblast cells.[79] Studies in mice indicate that inhibition of complement C3 activation and deficiencies of C5 and C5a seem to protect against pregnancy loss and fetal growth restriction.[80,81]

The binding of aPL to trophoblast cell membranes promotes complement activation, specifically C3a and C5a, resulting in thrombosis and pregnancy loss.[82] Instigators of inflammatory tissue injury caused by complement activation include the chemokine tumor necrosis factor (TNF) alpha and the cytokine receptor, tissue factor (TF). Exposure of pregnant mice to aPL causes an increase in decidual TNF-α levels, putatively secondary to C5 activation.[83] In an APS mouse model, antibody-mediated reduction of TNF-α helped prevent fetal resorption.[84] Therefore, TNF-α is a likely mediator that links complement activation and aPL to placental inflammatory injury. Complement activation (C5a–C5aR) increases TF expression in neutrophils, which increase generation of reactive oxygen species thus providing a novel mechanism to explain fetal loss.[85] An additional mechanism whereby TF could promote placental injury in APS is its capacity to activate the coagulation pathway, leading to thrombosis.[86]

More evidence is required to confirm the role of complement-mediated pregnancy loss in women with APS. Histopathologic studies reveal that placental tissue from APS gestations show a clustering of inflammatory cells and macrophages around blood vessels compared with controls.[87] Other investigations have not consistently shown the presence of complement complex deposition in placental tissue from women with APS.[88] Important future work will help determine whether therapy that targets interruption of complement-mediated inflammatory injury will be effective in improving pregnancy outcome in women with APS.

Mechanism of Therapeutic Benefit of Heparin

The therapeutic benefit of heparin and aspirin in improving outcomes in women with obstetric APS has been presumed to relate to their anticoagulant properties. However, the efficacy of heparin in the management of obstetric APS is achieved at a dosage lower than is required to achieve clinical anticoagulation. Studies in the murine model indicate that anticoagulants such as hirudin and fondaparinux are ineffective in the treatment of aPL-induced pregnancy loss, despite having anticoagulant effects similar to heparin.[89] Several mechanisms have been identified that may explain the beneficial effects of heparin independently of its anticoagulant action.

Low-molecular-weight heparin (LMWH) directly impedes aPL binding to trophoblast cells and reinstates normal trophoblast invasiveness and differentiation hindered by aPL. Heparin also can potentially regulate apoptosis in placental explants by increasing levels of Bcl-2, an antiapoptotic protein.[90] Furthermore, heparins have been shown to prevent complement activation in vitro, an action that would protect normal placentation from inflammatory injury. Exposure of trophoblastic cells to LMWH results in an increase in matrix metalloproteinases in trophoblastic cells, an action that would promote trophoblastic invasiveness.[91] Heparin sulfate directly binds aPL, providing a novel and alternative mechanism for its therapeutic benefit in treating obstetric APS.[92,93]

Thus, heparin may prevent pregnancy complications in obstetric APS by inhibiting the binding of aPL to trophoblastic cell membranes, modulating trophoblast apoptosis, promoting trophoblast cell invasiveness, and reducing complement activation with the ensuing inflammatory response at the decidual-placental interface. All of these mechanisms are independent of heparin's known anticoagulant property in inhibiting thrombosis.

MANAGEMENT OF APS IN PREGNANCY

Overview of Proposed Treatments

Only 20% to 30% of patients with LAC and/or positive levels of aPL who have had unsuccessful previous pregnancies have a successful delivery without treatment.[16,47] Comparison of studies in the literature is unsatisfactory because of differences in study design, patient selection, and treatment regimens. There are few randomized clinical trials, and variations in study definitions affect tabulated outcomes. Pregnancy outcomes are not always defined. Women with low levels of aPL are included in some studies. Moreover, aPL assays performed in laboratories with inappropriate standardization or controls may further complicate interpretation of results.

Several treatments have been proposed, including a single low-dose aspirin per day (81 mg), aspirin and low-dose or high-dose prednisone, aspirin and unfractionated heparin, aspirin and LMWH, and intravenous immunoglobulin.[94] All treatments seem to improve the live birth rate; however, the combination of unfractionated heparin with low-dose aspirin seems to provide the highest success rates.[34,35,48,49] Aspirin

is thought to improve outcome by selective inhibition of thromboxane production thus restoring the balance with prostaglandin. Concomitant use of prednisone and heparin is generally not recommended because this combination has not been shown to be better than either alone and may increase the risk of fractures. In a randomized trial comparing heparin and aspirin versus prednisone and aspirin, Cowchock and colleagues included a small group of 20 women with at least 2 early losses.[46] However, patients were not assigned to treatment until after the documentation of a fetal heartbeat, thus the outcome that showed that the treatments were equal in achieving a successful outcome has been questioned. However, treatment with corticosteroids was associated with increased neonatal morbidity (preterm delivery and low birth weight) and increased maternal morbidity (pregnancy-induced hypertension and gestational diabetes). A more recent prospective randomized clinical trial comparing prednisone and aspirin (100 mg/d) for women with autoantibodies and unexplained recurrent fetal loss showed no benefit in promoting live birth with prednisone and aspirin but an increase in prematurity.[47]

Unfractionated Heparin Compared with LMWH

In our prospective trial of aspirin alone versus aspirin and heparin, a group of well-characterized women with APS ($\geq$3 early RPLs), an otherwise negative evaluation, and positive aPL on 2 occasions were given different treatments.[95] The group treated with aspirin alone had a 44% live birth rate, whereas the group treated with subcutaneous heparin and aspirin had a 78% live birth rate ($P<.05$). The incidence of maternal complications was low and there were no significant differences in the birth weight, incidence of pregnancy- induced hypertension (PIH), gestational diabetes, or cesarean section.

Low-dose heparin is emerging as the treatment of choice for RPL associated with antiphospholipid syndrome.[15,27,34,39,49,92,95] Heparin treatment is generally initiated at 5000 to 7500 U twice a day when the pregnancy test is positive. It is important to obtain a baseline platelet count and partial thromboplastin time. Heparin treatment is associated with heparin-induced osteopenia at total daily doses of more than 15,000 international units (IU) per day which, combined with the normal osteoporosis associated during pregnancy, can be accompanied by increased bone loss. The heparinized pregnant patient should increase her intake of calcium to 600 mg orally twice a day along with vitamin D 400 IU twice daily to optimize absorption of calcium and to reduce the risk of osteopenia (**Box 4**). Heparin is also reported to be associated with thrombocytopenia. However, the incidence in populations of patients treated at our center at daily doses of less than 15,000 IU per day has been less than 1%. During pregnancy, the normal platelet count is more than 100,000/mL compared with the nonpregnant state, in which platelet counts are normally more than 150,000/mL. If counts were normal before conception and decreased dramatically during pregnancy, the heparin dosage should be reduced. It is important to instruct patients carefully on the proper administration of subcutaneous heparin to minimize complications. We use an intensive one-on-one teaching session with a nurse specialist before conception and have found that using this type of therapy reduces the stress level of patients.

The first prospective, randomized trial suggested that low-molecular-weight heparin may be an effective alternative to unfractionated heparin in the treatment of APS.[96] In a 2-centered, nonrandomized study, the use of aspirin 81 mg daily in combination with LMWH during pregnancy for the prevention of RPL in women with APS seemed to be as safe as unfractionated heparin plus aspirin.[97] Large, randomized trials are required to determine differences in outcome with LMWH and low-dose aspirin compared with

Box 4
Guidelines for prophylactic heparin plus aspirin treatment of patients with RPL without a history of thromboembolism but with APS

1. Baseline nonpregnant studies of aPLs, complete blood count with platelets, PT, PTT, and lupus anticoagulant should be obtained. aPL assay should be confirmed before pregnancy.
2. Aspirin 81 mg should be initiated before conception and discontinued 4 weeks before the expected delivery date. Aspirin 81 mg should be resumed postpartum and continued for life unless otherwise contraindicated or until better recommendations are available.
3. Subcutaneous heparin (5000 units every 12 hours) should be initiated when pregnancy is confirmed unless instructed otherwise. Patients who weigh more than 80 kg (175 pounds) should use heparin 7500 units every 12 hours. Platelets and PTT tests should be checked every week for 2 weeks initially, 1 week following any adjustment in dose, and each trimester throughout pregnancy to evaluate for heparin-induced thrombocytopenia (patients with prior thromboembolic events should be fully anticoagulated).
4. Calcium carbonate (1200–1500 mg) with vitamin D (800–1000 IU) should be taken daily in divided doses once a patient starts heparin to decrease the bone loss associated with pregnancy and heparin therapy.
5. The pregnancy should be documented by ultrasonography by 7 weeks for the detection of fetal heart motion. Further sonography may be performed at 18 to 20 weeks.
6. Antenatal testing should begin at 28 to 30 weeks, based on the possible increased risk of fetal growth restriction and stillbirth. This testing may include kick counts, nonstress tests, and/or serial biophysical profiles. Serial scans for growth rate may be indicated.
7. Heparin should be continued until the patient initiates spontaneous labor or until the night before any scheduled induction or operative delivery. One heparin dose may be skipped the night before amniocentesis. Heparin should be restarted postpartum at the lowest predelivery dosage and continued for 4 weeks (in those patients with previous thromboembolic events, full anticoagulation should continue for 6 weeks postpartum).
8. For prolonged deliveries and operative deliveries, the use of pneumatic sequential compression devices or hose should be considered until the patient is ambulatory.
9. If the patient is fully anticoagulated and delivery is emergent, 1% protamine sulfate can be administered intravenously over 10 minutes (2.5 mg protamine per 1000 U heparin, maximum 50 mg protamine) if coagulation indicators are increased.
10. Patients should not use estrogen-containing birth control pills for contraception. Aspirin 81 mg daily is advised until further recommendations become available. Patients who smoke should be advised to stop.

Data from Refs.[15,49,95]

treatment with unfractionated heparin combined with low-dose aspirin in this group of patients.

Antepartum Surveillance

Antepartum testing has been recommended, based on the increased risk for poor obstetric outcome. For women who have had only first-trimester losses, it is usual to perform serial ultrasonic assessments weekly until the patients have progressed beyond the point of their prior losses. If the patient had a prior second-trimester or third-trimester fetal loss, serial antepartum testing is recommended.[15] Antepartum assessment using daily fetal kick counts, twice weekly nonstress tests, or weekly biophysical profile has been suggested.

POSTPREGNANCY CONSIDERATIONS

Immediately Postpartum

Women who had a vaginal delivery should ambulate as soon as possible. Women who had a cesarean delivery should continue to use pneumatic compression stockings until they are fully ambulating. Aspirin 81 mg a day can be reinitiated if not contraindicated for other reasons. Authorities recommend that women should receive thromboprophylaxis postpartum for 4 to 6 weeks.[15] In general, this recommendation includes women with and without a prior history of thrombosis. Supplemental calcium should be continued as long as the patient is taking heparin. Breast-feeding is not contraindicated for women taking low-dose aspirin or prophylactic heparin.

Lifelong Consequences of APS

Women with APS have been diagnosed with an acquired autoimmune syndrome similar to SLE. Once the diagnosis is made with appropriate clinical criteria, this should be considered to be a diagnosis that they carry for life. For example, an individual with lupus may have an exacerbation and be severely disabled with multiple abnormal laboratory tests, but, after resolution, her symptoms and laboratory tests may normalize. Many physicians are confused when a patient with APS who previously had positive tests presents years later and the test results have returned to normal; however, in our opinion the patient should still be considered to have the diagnosis of APS. As such, we recommend that, once the diagnosis of APS is made, women should not use estrogen-containing oral contraceptive pills but should use progestin-only pills, barrier methods, or intrauterine devices.[15] They should similarly not use tobacco products and should maintain a normal weight. Any correctable risk factors for future thrombosis, such as increased cholesterol, should also be corrected. We agree with the recommendation that these women should use lifelong aspirin 81 mg per day unless it is contraindicated for other reasons.[98] In general, these individuals should live a healthy lifestyle.

SUMMARY

It is unclear at this time what treatment, if any, is required for women who do not meet all the criteria for diagnosis of APS but are known to have aPL. In some cases, these women were tested because of a prior false-positive test for syphilis with subsequent identification of aPL. Women undergoing in vitro fertilization (IVF) have been tested and found to have an increased incidence of aPL.[99] However, a summary of all published reports on studies of at least 100 patients indicates that positive aPL in patients undergoing IVF does not influence pregnancy rates.[99]

In women who have a diagnosis of APS, meeting both clinical and laboratory criteria, the chance for successful pregnancy is reduced. In these cases, treatment seems to be a clear option, particularly in the case of patients with prior thromboembolic events. Current clinical guidelines support the use of subcutaneous heparin and aspirin. This treatment should begin with a positive pregnancy test, continue throughout pregnancy, and extend postpartum.

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