Faster thrombin generation in women with polycystic ovary syndrome compared with healthy controls matched for age and body mass index

Maria Raquel Marques Furtado de Mendonça-Louzeiro, M.D.,[a]
Joyce Maria Annichino-Bizzacchi, M.D., Ph.D.,[b,c] Luís Alberto Magna, M.D., Ph.D.,[d]
Susan Kelly Picoli Quaino, M.D.,[c] and Cristina Laguna Benetti-Pinto, M.D., Ph.D.[a]

[a] Department of Gynecology and Obstetrics, School of Medical Sciences, [b] Department of Clinical Medicine, [d] Department of Medical Genetics, School of Medical Sciences; and [c] Hemostasis Laboratory, Campinas Blood Center, University of Campinas, São Paulo, Brazil

Objective: To evaluate hemostatic markers in women with polycystic ovary syndrome (PCOS) compared with healthy controls matched for age and body mass index.
Design: Cross-sectional study.
Setting: Tertiary teaching hospital.
Patient(s): Forty-five women with PCOS and 45 controls paired for age (±2 years) and body mass index (±2 kg/m^2).
Intervention(s): Clinical evaluation and venipuncture.
Main Outcome Measure(s): Thrombin activatable fibrinolysis inhibitor, D-dimer, plasminogen activator inhibitor-1, and the thrombin generation test.
Result(s): Thrombin generation lag-time was significantly shorter in women with PCOS compared with controls. The other hemostatic parameters were similar in both groups.
Conclusion(s): Thrombin generation is faster in young and overweight women with PCOS, suggesting a greater risk of hypercoagulability. (Fertil Steril® 2013;99:1786–90. ©2013 by American Society for Reproductive Medicine.)
Key Words: Polycystic ovary syndrome, hemostatic markers, coagulation, fibrinolysis, venous thrombosis

Discuss: You can discuss this article with its authors and with other ASRM members at http://fertstertforum.com/mrmendoncalouzeiro-venous-thrombosis-polycystic-ovary-syndrome/

Use your smartphone to scan this QR code and connect to the discussion forum for this article now.*

* Download a free QR code scanner by searching for "QR scanner" in your smartphone's app store or app marketplace.

Polycystic ovary syndrome (PCOS) is a highly prevalent condition, affecting 5%–10% of women of reproductive age (1–3). Women with PCOS often have metabolic abnormalities, including insulin resistance, dyslipidemia, glucose intolerance, type 2 diabetes, and metabolic syndrome (4–9).

These typical features of PCOS are well-known risk factors for cardiovascular disease. Other factors, such as suppression of fibrinolysis, might also contribute to increased risk of myocardial infarction (10). However, in PCOS, although it is assumed that the disorders of the hemostatic system and coagulation may contribute toward increasing the risk of cardiovascular events, evidence in the literature is conflicting and based on only a few studies (10–14), with some questions remaining to be answered, particularly regarding pathogenesis and risk factors (15). Studies also suggest a proinflammatory and prothrombotic state, particularly in the presence of the metabolic syndrome (16).

Hemostatic system disorders may be the consequence of a large number of diseases, but the evaluation of these disorders is not easy (17). The literature

Received September 18, 2012; revised January 6, 2013; accepted January 14, 2013; published online February 15, 2013.
M.R.M.F.d.M.-L. has nothing to disclose. J.M.A.-B. has nothing to disclose. L.A.M. has nothing to disclose. S.K.P.Q. has nothing to disclose. C.L.B.-P. has nothing to disclose.
Financial support provided by the São Paulo State Foundation for the Support of Research (project no. 2010/11707-0).
Reprint requests: Cristina Laguna Benetti-Pinto, M.D., Ph.D., Departamento de Ginecologia e Obstetrícia, Universidade Estadual de Campinas (UNICAMP), Faculdade de Ciências Médicas, Av. Alexander Fleming 101, Cidade Universitária, 13083-881 Campinas, São Paulo, Brazil (E-mail: laguna.unicamp@gmail.com).

Fertility and Sterility® Vol. 99, No. 6, May 2013 0015-0282/$36.00

http://dx.doi.org/10.1016/j.fertnstert.2013.01.105

shows that various laboratory tests have been developed to identify alterations in the functioning of the coagulation system; however, a single laboratory parameter that would be increased in all forms of hypercoagulability and, likewise, would be lower in all forms of hypocoagulability, is yet to be determined. Such a test would be extremely useful for detecting a tendency toward thromboembolism and for discovering drug-induced hypocoagulability (17). In this respect, endogenous thrombin potential (ETP), measured by the thrombin generation test (TGT), has been suggested as a good indicator of coagulability (17–19).

The TGT is a recently developed test that measures the interaction capacity between all the plasmatic components that result in the formation of the fibrin clot. For this reason it is considered a good indicator of coagulation. Therefore, the pioneering evaluation of TGT, and hence ETP, in women with PCOS is extremely interesting, because it permits the behavior of the hemostatic system to be evaluated as a whole in this population group (17–19).

Other markers also used to evaluate alterations in the hemostatic system include thrombin activatable fibrinolysis inhibitor (TAFI), D-dimer, and plasminogen activator inhibitor-1 (PAI-1). With respect to TAFI, its role as a possible risk factor for thrombotic disease has yet to be completely clarified (20–23). Although some studies report that high levels of TAFI represent a risk factor for deep vein thrombosis and ischemic stroke, others have failed to confirm these findings (24, 25). D-dimer is a product of fibrin degradation and acts as a marker of fibrinolysis and hypercoagulability. Elevated D-dimer levels are indicative of fibrinogen and fibrin turnover. An increase in the levels of this marker is suggestive of an increase in thrombogenesis (26). Last, an increase in PAI-1 activity is associated with hypofibrinolysis and may contribute to the development of deep vein thrombosis (27, 28).

Hence, the objective of the present study was to compare hemostatic markers, including a global coagulation test (TGT), between women with and without PCOS.

MATERIALS AND METHODS

Subject Selection

A cross-sectional study was conducted with 45 women with PCOS (Rotterdam Consensus, 2003) (29) and 45 women with normal ovarian function (control group). Women with PCOS were paired with controls for age (±2 years) and body mass index (BMI) (±2 kg/m^2). The study participants were 18–35 years of age, had not been using any hormonal methods of contraception for 3 months or more before enrollment, and were receiving care at the gynecological endocrinology (cases) and family planning (controls) outpatient clinics at the Department of Gynecology and Obstetrics, School of Medical Sciences, University of Campinas, Brazil.

The women in the control group were selected at the family planning clinic from a group of women scheduled to have an intrauterine device inserted. All had regular menstrual cycles (24–35 days) and had not been in use of any hormonal contraceptive method or any other type of medication that could alter the menstrual cycle for at least 3 months. None of these women had hirsutism, and at clinical examination all were evaluated according to the Ferriman-Gallwey index (30). They were also submitted to ultrasonography to evaluate the intrauterine device, and polycystic ovaries were not found in any cases. Therefore, we consider that a diagnosis of PCOS was excluded in the women in the control group.

The following clinical variables were evaluated in the two groups: age, BMI, waist and hip measurements, waist/hip ratio, and the Ferriman-Gallwey index. Laboratory variables included fasting glucose, fasting insulin, insulin resistance according to the homeostasis model of assessment–insulin resistance (HOMA-IR) (31), total T, and free T, as well as the hemostatic markers TAFI, D-dimer, PAI-1, and TGT.

Exclusion criteria consisted of pregnancy, chronic diseases such as hypothyroidism, kidney failure, and liver failure, BMI $\geq$40 kg/m^2 (morbid obesity), a history of cancer or thromboembolic disease, and the use of any medication that could interfere with coagulation or fibrinolysis, such as aspirin, heparin, anticoagulants, antiplatelet drugs, or hormones.

The study was approved by the institution's internal review board under reference number 800/2010, and all the participants signed an informed consent form.

Laboratory Tests

Blood sampling. All samples were collected between 7:30 and 10:00 AM (to avoid the effect of the daily variation in the hemostatic system), after at least 12 hours of fasting, from the antecubital vein in the left arm, with minimal or no venous occlusion (protracted venous occlusion may stimulate PAI-1 production by the endothelial cells and alter the results of other markers of hemostasis). Blood sampling was performed between the 3rd and 9th days of the menstrual cycle, or more than 60 days after the last menstrual period for the women with PCOS.

To measure the markers of hemostasis (TAFI, D-dimer, PAI-1, and TGT), 14 mL of peripheral blood were collected in 3.8% sodium citrate in a proportion of 9:1 and immediately centrifuged at 3,500 rpm for 15 minutes, after which the plasma was divided into aliquots of 400 μL and stored in a freezer at −80°C until analysis. For the biochemical and hormonal measurements (glucose, insulin, total T, and free T), a 14-mL sample of peripheral blood was collected.

Analyses. Fasting glucose was evaluated using a colorimetric enzymatic method (Roche/Hitachi 904/911 Modular ACN 249). To evaluate insulin levels, a solid-phase two-site sequential chemiluminescent immunometric assay (Immulite/Immulite 1000; Siemens) was used. Total T was measured using an electrochemiluminescence assay (Cobas E-411; Roche), whereas free T was measured using solid-phase radioimmunoassay (Beckman Coulter DSL 4900).

Markers of Hemostasis

Thrombin activatable fibrinolysis inhibitor was measured in an STA compact coagulometer (Diagnostica STAGO) using a chromogenic assay with a Stachrom TAFI kit (Diagnostica