Retrospective analysis on clinical characteristics and venous thromboembolism outcomes using bridging prophylaxis with low molecular weight heparin for thrombophilic women performing oocyte retrieval for assisted reproductive procedures: 15 years of experience

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ABSTRACT

In vitro fertilization (IVF) procedures have been frequently associated with antithrombotic treatment, particularly aspirin or low molecular weight heparin. Historically, this type of treatment has been intended to increase the success rate of IVF with embryo transfer (IVF-ET) and live births after the procedure, as well as to prevent thrombotic disorders during pharmacological ovarian stimulation. Recurrent IVF failures and venous thromboembolism (VTE) complications during IVF-ET may be related to inherited thrombophilia. However, there aren't many studies in the literature on the frequency of VTE or bleeding in women undergoing thromboprophylaxis for IVF-ET, and reports on the caliber of clinical data vary. Thus, in this report, we describe our clinical experience with early antithrombotic prophylaxis with enoxaparin in women who have had thrombophilic defects and are undergoing IVF-ET over a period of years.

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Introduction

Several medical reasons have been identified as the cause of unsuccessful *in vitro* fertilization failure (IVF) with embryo transfer (ET) or intracytoplasmatic sperm injection (ICSI), and molecular thrombophilia is one of them.[1-4] For this reason anti-thrombotic therapy with aspirin and/or with low-molecular-weight heparins (LMWHs) has been suggested in the last decades as an additional treatment in women with ongoing IVF-ET or ICSI.[2,5]

Yet, anti-thrombotic therapy in this clinical setting should also be considered to prevent thrombotic complications potentially occurring during the pharmacological hormonal ovarian stimulation [*i.e.*, controlled ovarian stimulation (COS)] or the pregnancy following ICSI or IVF-ET.[5-8] Indeed, several studies emphasized that thrombotic complications in women ongoing hormonal therapy for IVF-ET or ICSI may occur either in the case of ovarian hyperstimulation syndrome (OHSS) or in the first trimester of pregnancy.[3,5,9-14] Furthermore, also the presence of a mild thrombophilia may result as an additional risk factor for women developing venous thromboembolism (VTE) during assisted reproductive procedures (ARP).[9,10,15]

Previous reports have already highlighted the role of VTE thromboprophylaxis in women with ongoing IVF-ET or ICSI during COS, due to the presence of many other pro-

thrombotic conditions during the performance of these gynecological procedures.[5,16,17]

Interestingly, the same prothrombotic conditions are also included in the Padua Prediction Score (PPS) for in-patients and should be considered before selecting a proper pharmacological thromboprophylaxis to prevent VTE in these patients.[2,5,7,10,18,19] Even, this clinical approach is considered for in-patients only but not for women ongoing IVF-ET or ICSI, and that is why its routine use is still a matter of discussion in this gynecological setting.[13] Furthermore, randomized clinical trials properly designed to attest the preventive role of LMWHs and their safety in this clinical setting are scarcely represented in the literature.[2,5,20,21]

We aimed to retrospectively analyze our records of thrombophilic patients with ongoing IVF-ET or ICSI receiving a prophylactic dose of LMWH in order to escape the risk of VTE; furthermore, the occurrence of major bleeding or clinically relevant bleeding and death for any causes have also been observed.

Materials and Methods

The study was conducted at the Fertility Center of Federico II University of Naples and data were retrospectively analyzed from 2019 to 2005.

A total of 9500 cycles of ICSI in 6400 female patients were analyzed. Thromboprophylaxis for an increased VTE risk was suggested for 3500 patients because of the presence of thrombophilia and other prothrombotic conditions detected by the application of the PPS, while the remaining 2900 patients did not perform pharmacological thromboprophylaxis because prothrombotic conditions were not identified.

All women underwent controlled ovarian hormonal stimulation with gonadotropins according to standard or personalized pharmacological protocol in order to increase the number of recruitable oocytes for ICSI.

Patients at increased risk of developing VTE used pharmacological thromboprophylaxis (PPS >3-4) using LMWH starting with treatment to obtain controlled ovarian hyperstimulation. Bridging prophylaxis was performed with a transitory withdrawal of LMWH before oocyte retrieval because it is a surgical procedure according to standard protocol (*i.e.*, 12 hours before surgery) with the resumption of treatment 12 hours after the end of surgical procedures) and then continued till the pregnancy test. In particular, administration of LMWH at prophylactic doses (mainly enoxaparin 40 mg daily) began together with pharmacological ovarian stimulation and it was stopped 12-15 hours before the surgical approach for trans vaginal oocytes retrieval (so, managing surgical risk of bleeding) and started again 12-15 hours after trans vaginal oocytes retrieval with same prophylactic doses until pregnancy test.

The investigated thrombophilic defects included: factor V Leiden, prothrombin G20210A mutation, protein S and protein C deficiency, antithrombin deficiency, hyperhomocysteinemia [with or without methylenetetrahydrofolate reductase (MTHFR) c677t gene polymorphism], antiphospholipid syndrome [according to the International Society on Thrombosis and Haemostasis (ISTH) criteria] or combined multiple defects.[22]

Patients with previous VTE as well as patients that developed OHSS were excluded because of their significantly increased risk of VTE complications *per se*.

The occurrence of objectively documented superficial and/or deep vein thrombosis, and pulmonary embolism were investigated; safety of the use of LMWH was evaluated by the incidence of major and clinically relevant non-major bleeding, as assessed by the ISTH criteria.[23]

Deep and superficial vein thrombosis were objectively investigated in all patients undergoing an ET procedure by ultrasonographic assessment. Symptomatic pulmonary embolism, with or without concomitant superficial and/or deep vein thrombosis, was assessed by a computed tomography pulmonary angiography. The rate of thrombosis and bleeding was compared to the rate of patients' ongoing IVF-ET with data available in Literature.[24-26]

Results

The clinical characteristics of study patients are depicted and summarized in Table 1. The mean age was 35±4 years and body mass index >30 was present in 40 patients. 750 patients were smokers. The presence of factor V Leiden was detected in 238 patients (19%; 237 heterozygous and 1 homozygous). Prothrombin G20210A mutation was present in 238 patients (19%; 236 heterozygous and 2 homozygous). Protein S deficiency was found in 28 patients, while protein C deficiency was found in 19 patients. Antithrombin deficiency was found in only one patient. Antiphospholipid syndrome was found in 39 patients, while positivity to lupus anticoagulant or to antiphospholipid antibodies alone was found in 56 and 42 patients, respectively. Combined thrombophilic defects were found in 97 patients.

As an additional finding, homozygous for MTHFR was detected in 550 patients, in 298 of whom it was associated with hyperhomocysteinemia. Pharmacological thromboprophylaxis was planned for all patients with standard doses of enoxaparin (*i.e.*, 4000 international units once a day) in all patients.

Eight deep vein thrombosis (DVT) of lower limbs and one DVT of upper limbs were detected with ultrasound scan while four superficial vein thrombosis of lower limbs were detected in our cohort; two pulmonary embolisms were identified with objective methods and were associated with DVT of lower limbs. VTE related events are reported in Table 2. Thrombotic localizations are summarized in Table 3.

One major bleeding and three clinical relevant bleedings were found in our cohort. Bleeding events are summarized in Table 4.

Discussion

In the last years, the use of antithrombotic drugs in women ongoing IVF-ET has been progressively increased because of the frequent association between thrombophilia and unexplained female infertility,[4,5,26-33] or miscarriage and because of the increased rate of successful pregnancy in women treated with antithrombotic drugs.[2,5,7,8,32-36]

Yet, the improved clinical surveillance of patients ongoing IVF-ET allowed also a better knowledge of the association between assisted reproductive techniques and VTE.[5,37-40] In particular, VTE occurs more frequently in patients in the first trimester of a successful pregnancy obtained with IVF-ET and the risk is estimated doubled compared to regular pregnancy.[5,41]

Table 1. Clinical characteristics and thrombophilic defects in patients that performed thromboprophylaxis during *in vitro* fertilization with embryo transfer or intracytoplasmatic sperm injection.

Thrombophilic patients with thromboprophylaxis (N. 1211)	Number of patients (%)
Mean age	35
IVF-ET	654 (54)
ICSI	557 (46)
Smoking	750 (61)
BMI >30	40 (3)
MTHFR c677t homozigosity	550 (45)
Factor V Leiden heterozigosity	237 (19)
Factor V Leiden homozigosity	1 (0.0008)
Prothrombin a20210g heterozigosity	236 (19)
Prothrombin A20210G homozigosity	2 (0.0016)
Hyperhomocysteinemia	298 (24)
Protein S deficiency	28 (2)
Protein C deficiency	19 (1)
Antithrombin deficiency	1 (0.0008)
Antiphospholipid syndrome (ISTH criteria)	39 (3)
LAC positivity	56 (4)
Antiphospholipid antibodies positivity	42 (3)
Combined thrombophilic defects	97 (8)

IVF, *in vitro* fertilization; ET, embryo transfer; ICSI, intracytoplasmatic sperm injection; BMI, body mass index; MTHFR, methylenetetrahydrofolate reductase; ISTH, International Society on Thrombosis and Haemostasis; LAC, lupus anticoagulant.

Table 2. Venous thromboembolism-related events in thrombophilic patients that performed thromboprophylaxis for *in vitro* fertilization with embryo transfer or intracytoplasmatic sperm injection.

	Patients with thromboprophylaxis (%) (enoxaparin 4000 IU daily)	Standard rate in patients without thromboprophyalxis (%)
VTE	12	0.08-2.5[24]
Death for VTE	0	-
Major bleedings	1	625
Clinical relevant bleedings	3	0.0826
Death for bleedings	0	-

IU, international units; VTE, venous thromboembolism; -, unknown.

Table 3. Type and localization of venous thromboembolism in patients that performed thromboprophylaxis with enoxaparin 4000 international units daily.

Outcomes	Patients in prophylaxis with enoxaparin 4000 IU daily
Deep vein thrombosis of lower limbs	8
Superficial vein thrombosis of lower limbs	3
Deep vein thrombosis of upper limbs	1
Pulmonary embolism	1

IU, international units.

Table 4. Type and localization of bleedings in patients that performed thromboprophylaxis with enoxaparin 4000 international units daily.

Outcomes	Patients treated with enoxaparin 4000 IU daily
Major bleedings	1 metrorrhagia
Clinical relevant bleedings	3 (2 haemoperitoneum, 1 haematoma of iliopsoas)
Death for bleedings	0

IU, international units.

Therefore although, the clinical association between VTE and IVF-ET is well known, a real frequency of this complication is difficult to be estimated, and it seems to be better quantified only for women that develop VTE associated to OHSS.[42] On the other hand, the risk to develop VTE for thrombophilic women ruled out for IVF-ET or ICSI is known but clearly estimated as far as the usefulness and the safety of LMWH in this clinical setting.

Few studies, in fact, have been focused on VTE prevention or on the efficacy and safety of prophylactic doses of low-molecular weight heparin.[20,43-47]

Our retrospective analysis focused on the primary VTE prevention in thrombophilic women ongoing IVF-ET/ICSI and receiving pharmacological thromboprophylaxis with enoxaparin once-daily. The rate of VTE was low when compared to that reported in the Literature for women not screened for molecular thrombophilia. No life threatening VTE events were recorded in our retrospective analysis. Therefore, the prophylactic use of LMWH in women with increased risk of VTE ruled out for IVF-ET/ICSI is effective also in a cohort of patients with inherited thrombophilia. Furthermore, there was not an increased risk of bleeding in the same cohort and also, in this case, the rate of bleeding complications was lower than that reported in the literature, so testifying that this type of thromboprophylaxis is safe. The bleeding risk in this clinical setting for a primary pharmacological thromboprophylaxis was not negligible because of the surgical approach for oocytes retrieval. In order to consent a safe surgical approach for the oocytes retrieval and for the ET procedures a short withdraw of LMWH administration was planned according to standard protocols for surgical areas.[48] For this reason, since a pharmacological wash out of LMWH was performed, all bleeding events should be considered related to different causes than the use of enoxaparin.

Therefore, for the first time, we reported data about a cohort of thrombophilic patients with thromboprophylaxis with enoxaparin 40 mg daily ongoing ARP with IVF-ET/ICSI starting from gonadotropins administration and with a short bridging time because the surgical approach to perform oocytes retrieval. With this strategy we raised a good efficacy and safety compared to data available in the literature concerning the rate of VTE related complications in this clinical setting.

Of course further data are needed to improve this clinical information because no trials are available in the literature nor are planned in the next future.

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