Low molecular weight heparin and first trimester maternal PAPP-A and hCG levels, fetal nuchal translucency in the first trimester of pregnancy

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Summary

Purpose of investigation: We studied the relation of first trimester nuchal translucency and first trimester biochemical markers using low molecular weight heparin (LMWH) at 11-14 weeks of gestation. *Methods:* This retrospective study was conducted at our university between January 2007 and July 2009. Ninety-six patients were treated with low-dose LMWHs (enoxaparine 40 mg) from the beginning of pregnancy to 36 weeks of gestation and low-dose aspirin (100 mg) to 32 weeks (group 1) and 100 control subjects (group 2) were included in the study. Transabdominal ultrasound examination was performed to diagnose any major fetal defects and for measurement of crown rump length (CRL) and fetal nuchal translucency (NT) thickness. Blood samples were drawn from each women PAPP-A and free beta hCG levels. *Results:* There were no significant differences with respect to age, gestational age at the first trimester, and gestational age at birth between the groups. The mean birth weight of babies in the LMWH group was lower than the control ($p = 0.026$). There were also no significant differences with respect to CRL, serum PAPP-A and hCG at 11-14 weeks of gestation. However, NT of group 1 was significantly lower than group 2 ($p = 0.000$). In the Spearman correlation, LMWH was negatively correlated with only NT ($r = –0.298$, $p = 0.000$). NT in the first trimester (95% CI –0.632-0.230, $p = 0.000$) was an independent parameter related to using LMWH. *Conclusion:* Women who used LMWH during pregnancy had decreased NT compared with unaffected women.

Key words: Low molecular weight heparin; First trimester screening; Nuchal translucency.

Introduction

In the first trimester of pregnancy, serum level of pregnancy-associated plasma protein-A (PAPP-A) is used in combination with nuchal translucency (NT), free B-human chorionic gonadotrophin (β-hCG) and maternal age to screen pregnancies for the risk of Down syndrome and other chromosome abnormalities. Experience with this screening program has achieved Down's syndrome detection rates of up to 90% for a false-positive rate of 5% [1].

Independent of the presence of aneuploidy, women undergoing biochemical screening and found to have markedly reduced PAPP-A levels in the first trimester are increasingly being recognized as also at increased risk of other pregnancy complications. They include miscarriage, growth restriction, hypertensive disorders, premature delivery and stillbirth [2-5].

In recent years low molecular weight heparin (LMWH) has been used for women with acquired or inherited thrombophilia. Our aim in this study was to investigate the relation of first trimester fetal nuchal translucency (NT) and first trimester maternal biochemical markers using LMWH at 11-14 weeks of gestation.

Material and Method

This retrospective study was conducted at our university between January 2007 and July 2009. Approval for the study was granted by the local ethical committee. Patients with multiple gestation, preterm delivery, maternal hypertension or proteinuria, pregestational diabetes and known genetic or congenital malformations were excluded from the study.

Ninety-six patients were treated with low-dose LMWH from the beginning of pregnancy to 36 weeks of gestation and low-dose aspirin (100 mg) to 32 weeks (group 1). Patients with a minimum of three consecutive pregnancy losses or one pregnancy loss after ten weeks (with positive lupus anticoagulant and/or APA on at least two occasions, protein C, S or antithrombin III deficiencies, mutations of the factor V Leiden and prothrombin II genes) and an otherwise negative recurrent pregnancy loss workup were included in this study [6-8]. Enoxaparin 40 mg subcutaneous injection was usually preferred for the thromboprophylaxis. It was listed by the FDA in pregnancy category B [9, 10].

A hundred control subjects were generated from patients that were appropriate for gestational age (AGA) term neonates (birth weight > 2500 g and < 4000 g) served as a control (Group 2). This group was also defined as those pregnancies in which a single live fetus was delivered with no anomalies.

Transabdominal ultrasound examination was performed to diagnose any major fetal defects and for measurement of crown rump length (CRL) and fetal NT thickness [11]. Measurement of PAPP-A and maternal serum free β-hCG were carried out by an automated machine Kryptor system, Brams AG, Berlin, Germany). Gestational age was based on the CRL at the time of screening.

Data are expressed as mean ± SD. Differences between the

Revised manuscript accepted for publication June 8, 2010

two groups were analyzed using the independent Student's t-test and the Mann-Whitney U-test. Homogeneity of variance was calculated by Levene's test and Liliefors significance correction test. Correlations between groups were assessed by Pearson's correlation coefficient and multiple regression analyses. All statistical calculations were performed using the program SPSS for Windows (vers. 9.05; SPSS, Inc., Chicago, IL, USA). Differences were considered statistically significant at levels of probability < 0.05.

Results

There were no significant differences with respect to age, gestational age at the first trimester, and gestational age at birth between the groups. The mean birthweight of babies in the LMWH group was lower than the control (p = 0.026) (Table 1).

There were also no significant differences with respect to CRL, serum PAPP-A and hCG at 11-14 weeks of gestation. However, NT of group 1 was significantly lower than group 2 (p = 0.000) (Table 2).

In the Spearman correlation, using LMWH was negatively correlated with only NT (r = –0.298, p = 0.000) (Table 3). NT in the first trimester (95% CI -0.632-0.230, p = 0.000) was an independent parameter related to using LMWH.

Table 1. — *Characteristics of the groups*.

	Group 1 (n = 96)	Group 2 (n = 100)	*p*
Age	30.18 ± 5.16	29.46 ± 4.20	0.287
Gestational age (wks) at the first trimester	12.39 ± 0.56	12.47 ± 0.59	0.366
Gestational age (wks) at birth	38.02 ± 1.14	38.27 ± 0.95	0.113
Birth weight (grams)	3199 ± 449	3348 ± 333	0.026

Table 2. — *Comparison of the groups according to the first trimester CRL, NT, and biochemical parameters*.

	Group 1 (n = 96)	Group 2 (n = 100)	*p*
NT (mm)	1.63 ± 0.32	1.84 ± 0.34	0.000
PAPP-A (MoM)	1.00 ± 0.70	1.05 ± 0.59	0.603
Free β-hCG (MoM)	1.36 ± 1.45	1.12 ± 0.65	0.148
CRL (mm)	62.91 ± 7.49	62.57 ± 8.23	0.762

Table 3. — *Spearman correlation analysis of the LMWH group with different parameters in the first trimester.*

	r	*p*
Free β-hCG (MoM)	0.106	0.148
PAPP-A (MoM)	–0.038	0.603
NT (mm)	–0.298	0.000
CRL (mm)	0.022	0.148
Age	0.076	0.287

Discussion

There have been many studies that showed the beneficial effects of LMWH on inherited trombophilia or antiphospolipid syndrome [6-8]. However there were no reports on the relation of first trimester biochemical markers and LMWH during pregnancy in the literature. This study evaluated the relation of using LMWH and first trimester maternal biochemical markers and fetal NT.

LMWH exerts anticoagulant activity by binding antithrombin III, thereby accelerating the inhibition of thrombin (factor IIa) and factor Xa. Because the ability to inactivate thrombin is related to molecular size, LMWH has reduced anti-IIa activity compared to anti-Xa activity and thus a lower risk of bleeding at similar levels of anticoagulation [6].

Therefore, the reliable pharmacokinetics of LMWH and the long half-life, resulting in the need for less frequent injections, made them attractive for practical use during the nine months of pregnancy [9]. Widespread use over the last ten years has shown that LMWH is safer than unfractionated heparin (UFH) in pregnancy.

PAPP-A is a high molecular weight, zinc-binding metalloproteinase. It is thus a potentially proatherosclerotic molecule and has recently been shown to be a specific activator of insulin-like growth factor 1 (IGF-1), a mediator of atherosclerosis [12, 13]. There is evidence suggesting that PAPP-A could play a role in the development of atherosclerotic lesions and may represent a marker of atheromatous plaque instability and extent of cardiovascular disease [12]. Intravenous administration of heparin induces an intense and rapid increase in free PAPP-A in the serum [14]. However, we could not find any significant change with the administration of subcutaneous LMWH. This effect might be seen by a high dose and intravenous route.

NT in the LMWH group was surprisingly lower than the control group. The heterogeneity of conditions associated with increased NT suggests that there may not be a single underlying mechanism for the collection of fluid under the skin of the fetal neck. Possible mechanisms include cardiac dysfunction, venous congestion in the head and neck, altered composition of the extracellular matrix, failure of lymphatic drainage, fetal anemia or hypoproteinemia and congenital infection [11]. The relation of the coagulation system and NT have not been studied before.

Despite the lower mean molecular weight, LMWH does not cross the placenta in any trimester. Women undergoing first or second trimester abortions or normal term delivery did not have detectable anti-Xa levels in the fetal circulation two to seven hours after injection [15-17].

Oberkersch *et al*. showed that LMWH inhibits the classical complement activation pathway in vitro, although it is slightly less effective than UFH. The complement cascade is a powerful effector mechanism of the immune system which, upon activation by the classical, alternative or lectin pathways, generates fragments (C3a, C5a) responsible for the initiation of a local inflammatory response by recruitment of leukocytes to the area of infection or injury [18]. Decreased NT can be related indirectly with the anticoagulant or antiinflammatory effects on maternal circulation.

Our data may provide insights on negative effects of LMWH on NT. The limitation of this study was the lack of a real control group (patients who have acquired or inherited thrombophilia using any medication). However, these limitations can be addressed by further studies that include heparins and pregnancy. Understanding the mechanism(s) by which LMWH decreases NT may lead to the understanding of many different mechanisms for nuchal edema.

References

[1] Jaques A.M., Halliday J.L., Francis I., Bonacquisto L., Forbes R., Cronin A., Sheffield L.J.: "Follow-up and evaluation of the victorian first trimester combined screening programme for Down syndrome and trisomy 18". *BJOG*, 2007, *114*, 812.

[2] Spencer K., Cowans N.J., Avgidou K., Molina F., Nicolaides K.H.: "First-trimester biochemical markers of aneuploidy and the prediction of small-for-gestational age fetuses". *Ultrasound Obstet. Gynecol*., 2008, *31*, 15.

[3] Barrett S.L., Bower C., Hadlow N.: "Use of the combined first-trimester screen result and low PAPP-A to predict risk of adverse fetal outcomes". *Prenat. Diagn*., 2008, *28*, 28.

[4] Yaron Y., Heifetz S., Ochshorn Y., Lehavi O., Orr-Urtreger A.: "Decreased first trimester PAPP-A is a predictor of adverse pregnancy outcome". *Prenat. Diagn*., 2002, *22*, 778.

[5] Smith G.C., Stenhouse E.J., Crossley J., Aitken D.A., Cameron A.D., Connor J.M.: "Early pregnancy levels of pregnancy-associated plasma protein A and the risk of intrauterine growth restriction, premature birth, preeclampsia and stillbirth". *J. Clin. Endocrin. Metab*., 2002, *87*, 1762.

[6] Gris J.C., Balducchi J.P., Quere I., Hoffet M., Mares P.: "Enoxaparin sodium improves pregnancy outcome in aspirin-resistant antiphospholipid/antiprotein antibody syndromes". *Thromb. Haemost*., 2002, *87*, 536.

[7] Coppens M., Folkeringa N., Teune M.J., Hamulyak K., van der Meer J., Prins M.H., Buller H.R., Middeldorp S.: "Outcome of the subsequent pregnancy after a first loss in women with the factor V Leiden or prothrombin 20210A mutations". *J. Thromb. Haemost*., 2007, *5*, 1444.

[8] Grandone E., Brancaccio V., Colaizzo D., Scianname N., Pavone G., Di Minno G., Margaglione M.: "Preventing adverse obstetric outcomes in women with genetic thrombophilia". *Fertil. Steril*., 2002, *78*, 371.

[9] Weitz J.I.: "Low-molecular-weight heparins". *N. Engl. J. Med*., 1997, *337*, 688.

[10] Sanson B.J., Lensing A.W., Prins M.H. *et al*.: "Safety of low-molecular-weight heparin in pregnancy: A systematic review". *Thromb. Haemost*., 1999, *81*, 668.

[11] Snijders R.J.M., Noble P., Sebire N., Souka A., Nicolaides K.H.: "UK multicentre project on assessment of risk of trisomy 21 by maternal age and fetal nuchal translucency thickness at 11-14 weeks of gestation. Fetal Medicine Foundation First Trimester Screening Group". *Lancet*, 1998, *352*, 343.

[12] Consuegra-Sanchez L., Fredericks S., Kaski J.C.: "Pregnancy-associated plasma protein-A (PAPP-A) and cardiovascular risk". *Atherosclerosis*, 2009, *203*, 353.

[13] Liu Z.Y., Zhang J.Y., Sun T.W., Zhang Y.J., Zhang L., Wang L.X.: "Levels of pregnancy-associated plasma protein A in patients with coronary artery disease". *Clin. Invest. Med*., 2008, *31*, E85.

[14] Tertti R., Wittfooth S., Porela P., Airaksinen K.E., Metsärinne K., Pettersson K.: "Intravenous administration of low molecular weight and unfractionated heparin elicits a rapid increase in serum pregnancy-associated plasma protein A". *Clin. Chem*., 2009, *55*, 1214.

[15] Andrew M., Ofosu F., Fernadez F., Jefferies A., Hirsh J., Mitchell L., Buchanan M.R.: "A low molecular weight heparin alters the fetal coagulation system in the pregnant sheep". *Thromb. Haemostasis*, 1986, *55*, 342.

[16] Forestier F., Daffos F., Capell-Pavlosky M.: "Low molecular weight heparin does not cross the placenta during the second trimester of pregnancy. Study by direct fetal blood sampling on ultrasound". *Thromb. Res*., 1984, *34*, 557.

[17] Bazzan M., Donvito V.: "Low-molecular-weight heparin during pregnancy". *Thromb. Res*., 2001, *101*, 5175.

[18] Oberkersch R., Attorresi A., Calabrese G.: "Low-molecular-weight heparin inhibition in classical complement activation pathway during pregnancy". *Thromb. Res*., 2010, *125*, e240.

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