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European Journal of Obstetrics & Gynecology and Reproductive Biology 127 (2006) 41–49
ejog European Journal of Obstetrics & Gynecology and Reproductive Biology
www.elsevier.com/locate/ejogrb

Phosphodiesterase-5 inhibitors and omental and placental small artery function in normal pregnancy and pre-eclampsia

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Received 13 January 2004; received in revised form 21 April 2004; accepted 13 June 2004

Abstract

Objectives: In pre-eclampsia (PE), endothelium-dependent function of myometrial small arteries is markedly attenuated. The residual PE response is wholly NO mediated. We have previously demonstrated that PDE5 inhibition can improve endothelial function in myometrial small arteries from women with PE. We aimed to assess whether the effect of PDE5 inhibition in PE was myometrial artery specific.
Study design: Small arteries were dissected from omental biopsies obtained at Caesarean section from normal pregnant women (NP, $N = 20$) and women with PE ($N = 11$). Chorionic plate small arteries were dissected from NP ($N = 13$) and PE ($N = 11$) placentae. Vasoconstriction (arginine vasopressin or thromboxane-mimetic U46619) and endothelial-dependent relaxation were assessed by wire and pressure myography. Constriction/relaxation curves were repeated post 1 h incubation with PDE5 inhibitors UK-343664 or sildenafil citrate (0, 10 or 100 nM).
Results: Omental artery constriction was increased in PE. Omental vessel constriction was unaffected by PDE5 inhibition. Sildenafil citrate improved bradykinin-induced but not acetylcholine-induced relaxation of omental small arteries from NP women. PDE5 inhibition did not alter relaxation of omental arteries from women with PE. Placental small arteries were unaffected by PDE5 inhibition.
Conclusion: Use of PDE5 inhibitors does not significantly alter endothelial-dependent relaxation in omental or placental small arteries from PE women.

Keywords: Pre-eclampsia; Phosphodiesterase-5 inhibitors; Human; Omentum; Placenta

1. Introduction

Pre-eclampsia is a complicating factor affecting 2–3% of primigravidae pregnancies in the United Kingdom and is a major cause of perinatal and maternal morbidity [1]. The pathophysiology of pre-eclampsia is thought to involve two interlinked processes: a failure of spiral artery transformation in early pregnancy which leads to a hypo-perfused uteroplacental unit [2,3] and the subsequent release of a factor(s) from the placenta that promotes activation of the maternal vascular endothelium [4,5]. Endothelial involvement/activation in pre-eclampsia is well documented with a number of markers of endothelial cell activation being increased in women with pre-eclampsia [2,3,6–11].

Aberrant endothelium-dependent responses have been demonstrated in a variety of blood vessels from women with pre-eclampsia, compared to vessels isolated from normal pregnant women [12–16]. McCarthy et al. [12] found attenuated endothelial-dependent vasodilatation in subcutaneous vessels from women with pre-eclampsia, a finding supported by Knock and Poston [15] and by Cockell and Poston [14] who demonstrated impaired flow-induced release of nitric oxide in human subcutaneous small arteries from women with pre-eclampsia using the technique of pressure myography. Similarly attenuated endothelium-dependendent vasodilatation has been demonstrated in myometrial [13] and omental [16] small arteries from

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0301-2115/$ – see front matter
doi:10.1016/j.ejogrb.2004.06.014

women with pre-eclampsia. Using pressure myography, we found that the residual relaxation of myometrial resistance arteries isolated from women with pre-eclampsia was almost entirely mediated via nitric oxide [17].

The role of nitric oxide in pre-eclampsia is controversial. Several clinical studies of nitric oxide in pre-eclampsia [18–21] are ambiguous, perhaps reflecting the difficulty in drawing conclusions of nitric oxide production in disparate vascular beds using assessments of overall nitric oxide metabolism. Both nitric oxide production and expression of nitric oxide synthase are increased in cultured endothelial cells after exposure to plasma from women with pre-eclampsia [22,23] whilst an increase in nitric oxide production might be expected to increase vasorelaxation, increased ''total'' nitric oxide production in pre-eclampsia might have deleterious effects; in the presence of oxidative stress, this could promote peroxynitrite formation and possible endothelial cell dysfunction [24,26]. Under such circumstances, pharmacological intervention to increase nitric oxide biosynthesis could be potentially harmful, as suggested by clinical trials investigating the use of nitric oxide donors in pre-eclampsia [27].

Nitric oxide is produced in response to receptor-mediated agonists or via physical forces (shear stress) acting on vascular endothelial cells. In the underlying vascular smooth muscle, this nitric oxide promotes cyclic-GMP (cGMP) production and relaxation of the contractile machinery by a number of mechanisms. The duration of action of cGMP in vascular smooth muscle is controlled by the cytosolic enzyme phosphodiesterase-5 (PDE5; [28–30]). A number of isoform selective phosphodiesterase inhibitors have recently been developed and used in the treatment of erectile dysfunction [30,31].

We have previously demonstrated using isometric wire myography [32] and isobaric pressure myography (Wareing et al., 2004) that the residual nitric oxide-dependent relaxation seen in myometrial small arteries from women with pre-eclampsia [17] can be potentiated by the presence of a PDE5 inhibitor, UK-343664. Pharmacological intervention using PDE5 inhibition will promote nitric oxide-dependent relaxation without stimulating nitric oxide biosynthesis and invoking the possible complications of oxidative stress associated with excess nitric oxide [24–26]. We were interested in using two PDE5 inhibitors since both UK-343664 and sildenafil citrate have similar high specificity for PDE5 over other PDE isoforms in human tissues but UK-343664 has been suggested to be easier to formulate for intravenous usage [33], a factor that may be relevant for use of these compounds therapeutically.

The aim of our current study was to extend our initial findings and to assess, by wire and pressure myography, whether inhibition of PDE5 with UK-343664 or sildenafil citrate could potentiate NO-mediated relaxation and improve endothelial-dependent vasodilatation in omental and placental small arteries from pregnancies complicated by pre-eclampsia.

2. Materials and methods

2.1. General

The ethics committee at St. Mary's Hospital, Manchester, gave approval for this work and written informed consent was obtained for all tissue used in the study. Omental biopsies were obtained at the time of delivery by Caesarean section (LSCS) from normal pregnant women (NP; $N = 35$) with uncomplicated pregnancies. Women with hypertension, diabetes or other significant medical disorders were excluded. Biopsies were also obtained from women with pre-eclampsia (PE as defined by ISSHP guidelines [34]; $N = 27$). All women in this group were delivered by Caesarean section for maternal or fetal reasons. The individualised birth ratio (IBR) was calculated at delivery. IBR relates to a predicted birth-weight centile calculated using independent coefficients for gestation at delivery, fetal sex, parity, ethnic origin, maternal height and booking weight. This method was utilised as it enables a more accurate prediction of pregnancies which end in a poor outcome than birth-weight for gestational age alone [35].

General chemicals were obtained from Sigma–Aldrich (Poole, Dorset, UK) or BDH (Poole, Dorset, UK). Arginine vasopressin (AVP), the thromboxane-mimetic U46619 and bradykinin (BK) were obtained from Sigma–Aldrich (UK). The PDE5 inhibitors UK-343664 and sildenafil citrate were obtained from Pfizer Limited, UK (Sandwich, Kent).

2.2. Samples

Biopsies were taken from normal (NP = 35) and PE ($N =$ 27) patients following delivery by Caesarean section and placed directly into ice-cold physiologic salt solution (PSS) a modified Krebs solution (see below for chemical composition). Omental small arteries were identified under a stereomicroscope and carefully dissected free from the surrounding connective tissue using small dissecting scissors and forceps.

For placentae, biopsies were taken and placed directly into ice-cold PSS. Small arterial branches of the chorionic plate vessels were identified under a stereomicroscope and carefully dissected free from the surrounding connective tissue within 20 min of delivery.

2.3. Wire myography

Short (~2 mm long) sections of omental and placental small arteries were mounted onto a Danish Myotechnology M610 wire myograph as described in detail elsewhere [36]. Initially, the bath contained 7 ml of PSS (in mmol l^{-1}; 127.76 NaCl, 25 $NaHCO_3$, 4.69 KCl, 2.4 $MgSO_4$, 1.6 $CaCl_2$, 1.18 KH_2PO_4, 6.05 glucose, 0.034 EDTA; pH 7.4), warmed to 37 °C and gassed with 95% air/5% CO_2.