Prevalence of cervical insufficiency in polycystic ovarian syndrome

Seth L. Feigenbaum¹, Yvonne Crites², Mohammad K. Hararah³, Miya P. Yamamoto⁴, Jingrong Yang³, and Joan C. Lo³,⁵,*

¹Department of Obstetrics and Gynecology, The Permanente Medical Group, San Francisco, CA 94115, USA ²Department of Obstetrics and Gynecology, The Permanente Medical Group, Santa Clara, CA 95051, USA ³Division of Research, Kaiser Permanente Northern California, Oakland, CA 94612, USA ⁴Department of Obstetrics and Gynecology, Kaiser Permanente Medical Center, Oakland, CA 94611, USA ⁵Department of Medicine, The Permanente Medical Group, Oakland, CA 94612, USA

*Correspondence address. Kaiser Permanente Division of Research, 2000 Broadway, Oakland, CA 94612, USA. Tel: +1-510-891-3682; Fax: +1-510-891-3802; E-mail: joan.c.lo@kp.org

Submitted on April 22, 2011; resubmitted on April 25, 2012; accepted on May 3, 2012

BACKGROUND: Pregnant women with polycystic ovarian syndrome (PCOS) experience a greater rate of adverse obstetrical outcomes compared with non-PCOS women. We examined the prevalence and incidence of cervical insufficiency (CI) in a community cohort of pregnant women with and without PCOS.

METHODS: A retrospective cohort study was conducted within a large integrated health care delivery system among non-diabetic PCOS women with second or third trimester delivery during 2002–2005 (singleton or twin gestation). PCOS was defined by Rotterdam criteria. A non-PCOS comparison group matched for delivery year and hospital facility was used to estimate the background rate of CI. Women were designated as having new CI diagnosed in the index pregnancy (based on cervical dilation and/or cervical shortening) and prior CI based on prior diagnosis of CI with prophylactic cerclage placement in the subsequent pregnancy.

RESULTS: We identified 999 PCOS women, of whom 29 (2.9%) had CI. There were 18 patients with new CI and 11 with prior CI having prophylactic cerclage placement; four CI patients had twin gestation. In contrast, only five (0.5%) non-PCOS women had CI: two with new CI and three with prior CI. The proportion of newly diagnosed incident CI (1.8 versus 0.2%) or prevalent CI (2.9 versus 0.5%) was significantly greater for PCOS compared with non-PCOS pregnant women (both \( P < 0.01 \)). Among PCOS women, CI prevalence was particularly high among South Asians (7.8%) and Blacks (17.5%) compared with Whites (1%) and significantly associated with gonadotropin use (including in vitro fertilization). Overall, the PCOS status was associated with an increased odds of prevalent CI pregnancy (adjusted odds ratio 4.8, 95% confidence interval 1.5–15.4), even after adjusting for maternal age, nulliparity, race/ethnicity, body mass index and fertility treatment.

CONCLUSION: In this large and ethnically diverse PCOS cohort, we found that CI occurred with a higher than expected frequency in PCOS women, particularly among South Asian and Black women. PCOS women with CI were also more likely to have received gonadotropin therapy. Future studies should examine whether natural and hormone-altered PCOS is a risk factor for CI, the role of race/ethnicity, fertility drugs and consideration for heightened mid-trimester surveillance in higher risk subgroups of pregnant women with PCOS.

Key words: polycystic ovarian syndrome / cervical insufficiency / obstetrics / race ethnicity

Introduction

Polycystic ovarian syndrome (PCOS) is a common endocrinopathy of reproductive-aged women and is defined by an international consensus definition that includes at least two of three component criteria: ovarian dysfunction, hyperandrogenism and polycystic ovaries by ultrasound (Rotterdam ESHRE/ASRM-Sponsored PCOS Consensus Workshop Group, 2004). Women with PCOS who conceive and deliver differ from non-affected women in part by the increased use of infertility interventions, and a greater incidence of gestational diabetes (Lo et al., 2006) and hypertensive disorders (Kashyap and Claman, 2000). These features and other associated metabolic perturbations may, in some measure, contribute to the etiopathology of adverse pregnancy outcomes in PCOS women.

In contrast, cervical insufficiency (CI) is a relatively uncommon complication of pregnancy. In a 10-year registry study of all hospitalized patients in Denmark, Lidgård reported a CI incidence rate of 4.6 per 1000 births, (Lidgård, 1994) similar to more recent estimates from Massachusetts, USA, where the prevalence of CI was estimated at 0.41% of 157 066 births not conceived with infertility medications.
or assisted reproductive technology (Schieve et al., 2007). Since the initial characterization of CI in 1959 (Easterday and Reid, 1959), recognition of specific risk factors including cervical conization, higher order multiple gestation, and exclusion of patients with preceding pre-term labor and infections has narrowed the spectrum for a more specific diagnosis, although a clear definition remains lacking (American College of Obstetrics and Gynecology, 2003). The infrequency of CI in conjunction with a lack of consensus criteria and advances in molecular and imaging technology has contributed to the challenges in examining the epidemiology of CI (McElrath, 2010). Furthermore, very few studies have investigated the risk of CI among selected clinical subsets.

As part of larger study examining the epidemiology of pregnancy in PCOS in Northern California, we utilized data from an integrated healthcare delivery system to examine the prevalence and incidence rate of CI, within a large community population cohort of PCOS women with delivered pregnancies and compared these rates with a reference population of non-PCOS women who delivered in the same period.

Methods

Kaiser Permanente Northern California (KPNC) is a large, integrated healthcare delivery system that provides comprehensive medical care for at least 30% of insured lives in the San Francisco and Greater Bay Area. Membership consists of ~3.2 million insured lives, with coverage spanning more than 14 counties in Northern California. There is substantial racial and ethnic diversity, particularly among the women with pregnancy and delivery (Lo et al., 2006).

Electronic databases were used to identify a large sample of non-diabetic pregnant women with and without diagnosed PCOS who received prenatal care and had a second or third trimester delivery within the KPNC geographic region during 2002–2005. The first delivered pregnancy during this interval was ascertained and designated the index pregnancy. The PCOS cohort was further qualified by retrospective verification of PCOS status by the ASRM-ESHRE (Rotterdam) criteria using ambulatory, laboratory, and radiologic records. Women with PCOS were required to have at least two of the following three conditions: menstrual dysfunction, androgen excess and polycystic-appearing ovaries, in the absence of other etiologies. Determination of clinical androgen excess was based on a clinical diagnosis of hirsutism, hyperandrogenism, adult acne, androgenic alopecia and/or any elevated free or total testosterone or free androgen index as determined in the Kaiser Permanente Regional Laboratory or commercial reference laboratory. Radiographic images were not available for 5% of the cohort in whom polycystic-appearing ovaries were repeatedly noted during reproductive endocrinology clinical evaluation. For these analyses, PCOS and non-PCOS women with triplet or higher gestation and prior cold-knife cervical conization were excluded.

A non-PCOS cohort of pregnant women with singleton or twin pregnancy matched by hospital facility and delivery year with the PCOS women was used for comparison and to estimate the background prevalence and incidence of CI among non-PCOS pregnant women.

Information on age, race/ethnicity and comorbidities were obtained from clinical records and electronic databases. Gestational diabetes mellitus was determined by standard glucose tolerance test criteria. Prior reproductive history, CI diagnosis and cerclage placement were ascertained by detailed chart review, including visit notes, ultrasound and operative reports, discharge summaries, pathology and microbiology reports to exclude patients with cervical changes attributable to infection or preterm labor. The CI status was investigated for both the index pregnancy (first pregnancy during 2002–2005) and, for ‘prior CI’ patients, the first pregnancy in which CI was originally diagnosed. The term ‘new CI’ was used if the first CI diagnosis occurred during the index pregnancy. The term ‘prior CI’ was used for patients with a prior confirmed CI pregnancy in whom a prophylactic cerclage was placed in the subsequent pregnancy. The CI diagnosis was defined by the presence of painless cervical dilation (according to traditional ACOG criteria) or progressive cervical shortening with funneling to a residual cervix of ≤1.5 cm. All identified CI cases were confirmed by a perinatologist (Y.C.). Cases with insufficient data, selective fetal reduction during the CI pregnancy or those with evidence of proximate infection, preterm labor, preterm premature rupture of membranes or other such causes, were not classified as CI in this study.

The method of conception was categorized as spontaneous, ovulation induction using oral agents (clomiphene citrate and/or metformin, thiazolidinediones or aromatase inhibitors), ovulation induction by injectable gonadotropins (HMG) with or without adjunctive agents and in vitro fertilization (IVF). In addition, we separately classified the HMG exposure to include women who conceived using injectable ovulation induction and/or IVF. Gestational age was based on the prenatal assignment of the estimated date of confinement, confirmed or adjusted by first and/or second trimester ultrasound. The Kaiser Foundation Research Institute’s Institutional Review Board approved the study.

Statistical approach

Comparison between groups were conducted using χ² or Fisher’s exact tests for categorical variables and Student’s t-test or Wilcoxon rank sum test for continuous variables. The prevalence of CI (current or prior) was reported as a point estimate with 95% confidence intervals (CI) overall and within the PCOS subset, stratified by race/ethnicity and singleton versus twin gestation status. We calculated the incidence rate of CI per 1000 births among the subset of PCOS and non-PCOS women without prior CI. Multivariable logistic regression was used to examine the independent association of PCOS and CI pregnancy. All analyses were conducted using STATA version 10.1 (StataCorp LP, College Station, TX, USA). A two-sided P value of <0.05 was considered statistically significant.

Results

The final analytic cohort of women with PCOS included 901 with singleton and 98 with twin delivered pregnancies after the exclusion of women with a history of cone biopsy and those with higher order multiple gestation in the index pregnancy. Twenty-nine cases of CI were identified among the PCOS women and further subdivided for better characterization as shown in Figure 1. These included 11 patients with prophylactic cerclage placed in the index (current) pregnancy based on confirmed CI diagnosis in a prior pregnancy (all with painless cervical dilation in the prior pregnancy) and 18 patients with newly diagnosed CI, 6 of whom subsequently had a cerclage placed. Among the 18 PCOS women with newly diagnosed CI, 16 presented with painless cervical dilation and only 2 received their diagnosis based on progressive cervical shortening to a cervical length of ≤1.5 cm (without cervical dilation). Based on these numbers, the proportion of newly diagnosed CI among the PCOS pregnancies was 1.8% or 18.2 (95% confidence interval, CI 10.8–28.6) per 1000 births overall. For singleton PCOS pregnancies, the rate of new CI was 16.8 per 1000 births (95% CI 9.5–27.6) and for twin PCOS pregnancies, the rate was 30.9 per 1000 (95% CI 6.4–87.7) births.
Among the reference non-PCOS group of 1020 delivered pregnancies matched for delivery facility and delivery year to the PCOS women, only 5 had CI, of whom 2 had newly diagnosed CI in the index pregnancy and 3 had prior CI (two with prophylactic cerclage placement in the index pregnancy and one declining cerclage placement). All cases involved singleton pregnancies. This rate of newly diagnosed CI in the non-PCOS subgroup was substantially lower at 0.2% or 2.0 (95% CI 0.2–7.0) per 1000 births compared with the PCOS group.

The overall prevalence of current or prior CI among the PCOS cohort was 2.9% (95% CI 2.0–4.1%), significantly higher than 0.5% (95% CI 0.2–1.1%) among the non-PCOS reference group (P < 0.01). Even after adjustment for maternal age, race/ethnicity, nulliparity, body mass index and fertility treatment, the PCOS status was associated with a significantly increased odds of new CI pregnancy (adjusted OR 4.8, 95% CI 1.5–15.4). These results were similar when restricting to the subset of PCOS and non-PCOS women with singleton pregnancy (adjusted OR 4.6, 95% CI 1.4–14.6).

Table I shows the demographic and clinical characteristics of PCOS and non-PCOS women, including the subset of PCOS women with current or prior CI. Women with PCOS had a slightly different race/ethnicity distribution compared with non-PCOS women, particularly a higher proportion of South Asian women. The PCOS women were also more likely to be older, obese, and have fewer prior pregnancies. By definition, most of the PCOS women had evidence of clinical androgen excess.

Within the PCOS subset, the mean age was 31.4 ± 4.4 years; 41.3% were White, 25.8% Asian (14.3% East Asian and 11.5% South Asian), 25.5% Hispanic, 4.0% Black and 3.3% other or mixed race. A total of 577 (57.8%) conceived with the assistance of fertility drugs (clomiphene citrate or gonadotropins), including 7% who underwent IVF. There were no differences in age, mean gravidity or obesity status by CI status, even after adjusting for an Asian-specific high-risk body mass index threshold ≥ 27.5 kg/m² (World Health Organization Expert Consultation, 2004). We also saw no difference in clinical androgen excess status, although androgen levels were not systematically obtained to enable more detailed stratification. Of interest, we found that the CI status varied widely depending on race/ethnicity, with prevalence of CI highest among Black (17.5%) and South Asian (7.8%) women compared with White women (1.0%) with PCOS (P < 0.01, Table II). We also noted that among East and South Asian women with CI, more than two-thirds presented with new CI in the index pregnancy.

Women with PCOS who experienced a CI pregnancy were more likely to have used HMG (either alone or with IVF) for their first CI pregnancy (current or prior) compared with those without CI (55.2

---

**Figure 1** Classification of CI among women with polycystic ovarian syndrome.
CI group compared with no cerclage (30.6 versus 21.5 weeks, associated with significantly lengthier gestation in the newly diagnosed women with newly diagnosed CI. Cerclage placement was also associated with significantly lengthier gestation in the newly diagnosed women (maternal age 34.4 versus 30.7 years, median gravidity 4 versus 1, \( P = 0.01 \) comparing PCOS and non-PCOS women. Among the confirmed non-PCOS women, 88% had acne and none had biochemical hyperandrogenism. Of the confirmed non-PCOS women, 88% had acne and none had biochemical hyperandrogenism. Compared to the PCOS cohort, PCOS women were more likely to receive a prophylactic cerclage (72.4% among CI versus 57.3% non-CI, \( P = 0.05 \) comparing PCOS and non-PCOS women). Among the PCOS women with a prior CI who were treated with any cerclage, 71% had CI diagnosed at the time of cerclage placement (Lidegaard, 1994; Schieve et al., 2007; Anum et al., 2010) and it is well known that PCOS women are prominently represented in large studies of recurrent pregnancy loss among different ethnic groups (Clifford et al., 1994; Diejomaoh et al., 2002). However, the extent to which PCOS patients experience late pregnancy loss attributable to CI has not been carefully investigated. We sought to minimize the effects of known or potential risk factors for CI by limiting analyses to women without prior cervical delivery.

**Discussion**

In this large, diverse community cohort of pregnant PCOS women, we found a surprisingly high frequency of CI, with a prevalence in the range of 3% and incidence rate of 18 per 1000 births, compared to lower reported estimates of CI in the general obstetric population (Lidegaard, 1994; Schieve et al., 2007; Anum et al., 2010) and estimates from a reference sample of non-PCOS pregnant women within our health plan. This study provides one of the first observations of CI among women with PCOS. Thatcher and Jackson (2006) described 2 preterm losses attributed to CI among 171 metformin-treated PCOS patients (1.2%), but broader population data with regard to the prevalence and incidence of this significant obstetrical complication among PCOS women has, to date, remained limited.

Our incidence of CI may underestimate the actual rate of CI, since the cohort was limited to ‘delivered’ pregnancies based on hospital diagnosis, thus excluding earlier CI-related pregnancy loss recorded as spontaneous abortions. Indeed, CI was identified in 8% of women in a large recurrent pregnancy loss study (Drakeley et al., 1998), and it is well known that PCOS women are prominently represented in large studies of recurrent pregnancy loss among different ethnic groups (Clifford et al., 1994; Diejomaoh et al., 2002). However, the extent to which PCOS patients experience late pregnancy loss attributable to CI has not been carefully investigated. We sought to minimize the effects of known or potential risk factors for CI by limiting analyses to women without prior cervical delivery.

\[ \text{P} = 0.02 \] in the PCOS cohort. However, in the latter case where no cerclage was placed, CI was diagnosed at the time of inevitable delivery.

### Table I Demographic and clinical features of women by polycystic ovarian syndrome (PCOS) and current or prior CI (PCOS-CI) status.

<table>
<thead>
<tr>
<th>Patient characteristics</th>
<th>Non-PCOS, ( N = 1020 )</th>
<th>PCOS, ( N = 999 )</th>
<th>PCOS-CI, ( N = 29 )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maternal age (years, mean ± SD)</td>
<td>30.9 ± 6.0</td>
<td>31.4 ± 4.4**</td>
<td>32.1 ± 4.8</td>
</tr>
<tr>
<td>Race/ethnicity%</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>41.1%</td>
<td>41.3%</td>
<td>13.8%</td>
</tr>
<tr>
<td>Black</td>
<td>7.1%</td>
<td>4.0%</td>
<td>24.1%</td>
</tr>
<tr>
<td>Hispanic</td>
<td>24.0%</td>
<td>25.5%</td>
<td>13.8%</td>
</tr>
<tr>
<td>East Asian</td>
<td>17.8%</td>
<td>14.3%</td>
<td>17.2%</td>
</tr>
<tr>
<td>South Asian</td>
<td>6.2%</td>
<td>11.5%</td>
<td>31.0%</td>
</tr>
<tr>
<td>Other</td>
<td>3.8%</td>
<td>3.3%</td>
<td>0.0%</td>
</tr>
<tr>
<td>Gravidity (mean ± SD)</td>
<td>2.6 ± 1.6</td>
<td>2.1 ± 1.3*</td>
<td>2.4 ± 1.4</td>
</tr>
<tr>
<td>Obesity (body mass index ≥ 30 kg/m²)</td>
<td>16.5%</td>
<td>42.9%*</td>
<td>44.8%</td>
</tr>
<tr>
<td>Obesity (Asian-specific threshold)*</td>
<td>18.0%</td>
<td>46.7%*</td>
<td>55.2%</td>
</tr>
<tr>
<td>Androgen excess (%)b</td>
<td>5.0%</td>
<td>78.2%*</td>
<td>75.9%</td>
</tr>
</tbody>
</table>

Numbers represent column percent where applicable.
*BMIs ≥ 27.5 kg/m² used for East Asian and South Asian women.
\( b \) Among the confirmed non-PCOS women, 88% had acne and none had biochemical hyperandrogenism.
\( \dagger \) \( P < 0.01 \) comparing PCOS and non-PCOS women.
\( * \) \( P < 0.05 \) comparing PCOS and non-PCOS women.
\( \dagger \) \( P < 0.01 \) comparing PCOS women with new or prior CI to PCOS women without CI.

### Table II Prevalence of CI among PCOS women by race/ethnicity.

<table>
<thead>
<tr>
<th>Race/ethnicity</th>
<th>Prevalent CI (current or prior CI)*</th>
<th>CI case description</th>
</tr>
</thead>
<tbody>
<tr>
<td>White (( N = 413 ))</td>
<td>1.0% (4/413)</td>
<td>4 CI cases: 2 prior CI, prophylactic cerclage 2 new CI in index pregnancy</td>
</tr>
<tr>
<td>Black (( N = 40 ))</td>
<td>17.5% (7/40)</td>
<td>7 CI cases: 3 prior CI, prophylactic cerclage 4 new CI in index pregnancy</td>
</tr>
<tr>
<td>Hispanic (( N = 255 ))</td>
<td>1.6% (4/255)</td>
<td>4 CI cases: 3 prior CI, prophylactic cerclage 1 new CI in index pregnancy</td>
</tr>
<tr>
<td>East Asian/Pacific Islander (( N = 143 ))</td>
<td>3.5% (5/143)</td>
<td>5 CI cases: 1 prior CI, prophylactic cerclage 4 new CI in index pregnancy</td>
</tr>
<tr>
<td>South Asian (( N = 115 ))</td>
<td>7.8% (9/115)</td>
<td>9 CI cases: 2 prior CI, prophylactic cerclage 7 new CI in index pregnancy</td>
</tr>
<tr>
<td>Other race (( N = 33 ))</td>
<td>0.0% (0/33)</td>
<td>No cases</td>
</tr>
</tbody>
</table>

\( * \) Prior PCOS-CI cases had a prophylactic cerclage placed during the current (index) pregnancy.
Cervical insufficiency in polycystic ovarian syndrome

Konization and those with singleton and twin gestations. Nonetheless, our estimate of 30.6 per 1000 births among twin pregnancies contrasts with the CI estimate of 1.1% from a large population of 1451 IVF-derived twin pregnancies (Ombelet et al., 2005). None of our reported CI cases occurred in the context of selective fetal reduction (Silver et al., 1997), known DES exposure, pre-term labor or pre-term premature rupture of membranes (Johanson et al., 2008) or proximate infection that could have explained early cervical change. We also examined the role of maternal age and weight; characteristics associated with very early preterm births (da Silva et al., 2003, Simhan and Bodnar, 2006) and found no association with CI in our PCOS cohort.

In this study, we found an association between injectable gonadotropins and subsequent development of CI. An increased relative risk of CI has been associated with IVF singletons (Schieve et al., 2007). However, our study was limited by the relatively small number of patients with IVF-conceived pregnancies, where the proportion with CI was insignificantly higher in the IVF subgroup. A study of 36,062 women with singleton pregnancy initially enrolled between 10 to 13 weeks gestation (Shevell et al., 2005) also showed insignificant differences in fetal loss at <24 weeks gestation between pregnancies spontaneously conceived (0.3%), those conceived using ovulation induction (0.4%) and IVF (0.2%). However, the numbers of patients with PCOS were not reported in that study.

A limitation of this investigation was the uncommon occurrence of CI. While we had the ability to study well-defined PCOS and CI within a large study population, the number of CI cases was too small to effectively examine the selected subgroups in detail. Nonetheless, the larger race/ethnic disparities we observed remain an important area for future investigation. In particular, we found a significant excess of CI among women with South Asian and Black race compared to Whites. Our findings are consistent with the increased risk of CI among Black parturients in the USA, recently described in a study using birth certificate natality data (Anum et al., 2010). Black women are also known to have an increased risk of spontaneous preterm birth, but there is limited data among Asian and South Asian populations, particularly with regard to CI. Perinatal outcomes also differ between race/ethnic groups when studied in immigrant countries or countries of native ethnic origin. Like other immigrant populations, both Chinese and Indian-Pakistani women living in the USA generally do not share the health or pregnancy-related outcomes of those living in their native countries, disparities most often attributable to differences in prenatal care or nutrition (Alexander et al., 2007). In our study, all patients received similar obstetrical care as members of a large prepaid integrated healthcare delivery system. While it is possible that some PCOS pregnancies receive greater surveillance during the early course of pregnancy, our study focused on second and third trimester deliveries where identification of new CI cases based on cervical length criteria alone accounted for only 2 of 18 new PCOS-CI cases who received normal obstetric surveillance. Our somewhat low background incidence of CI is likely based on the fact that cases were not identified through uniform cervical screening practices during the period of our study.

The role of metabolic dysfunction, including hyperandrogenism or glucose intolerance, remains unclear. We saw no such differences between the PCOS-CI and non-CI women in our study with regard to clinical androgen excess or the incidence of gestational diabetes. Mechanical properties of cervical collagen change during pregnancy under the influence of sex steroids (Winkler et al., 1997) and it is tempting to ascribe an association between preterm deliveries in the PCOS patients (as reflected in the higher rate of CI) to contributory effects of the PCOS hormonal milieu. However, no studies have explored these associations in PCOS patients (Petersen and Uldbjerg, 1996, Warren et al., 2007), where more than one mechanism may be operative in a specific patient (Romero et al., 2006).

In conclusion, we found a surprisingly high prevalence of CI among PCOS women, particularly South Asian and Black women. These data contribute to the growing evidence of adverse pregnancy outcomes among women with PCOS (Boomsma et al., 2006) and to existing data pertaining to health disparities and CI risk (Anum et al., 2010). Based on our findings, specifically designed studies should verify whether PCOS is a risk factor for the development of CI and the extent to which selected racial subgroups may be at higher risk warranting heightened mid-trimester obstetrical surveillance. Higher rates of very preterm birth have also been recently reported in PCOS women (Roos et al., 2011), raising the question of whether maternal PCOS status contributes to a higher combined risk of early spontaneous preterm birth.

Acknowledgements
The authors would like to acknowledge Grace Lee MD, Susan Peng MD, Sara Johnson MD and Joel R Gonzalez BS for assistance with data collection.

Authors’ roles
S.F. and J.L. conceived and participated in all aspects of the study and co-wrote the initial draft. Y.C., S.F. and M.Y. validated the pregnancy data and outcomes. J.Y., J.L. and M.H assembled the study cohort and conducted the analyses. All authors contributed substantially to design the study, acquisition and interpretation of data, manuscript review, revision for critical intellectual content and approved the final submission.

Funding
This study was funded by NIH/NICHD Grant R01 HD052966 awarded to Joan C. Lo.

Conflict of interest
None declared.

References