Original Article

Bioinformatics Approach Reveals Evidence for Impaired Endometrial Maturation Before and During Early Pregnancy in Women Who Developed Preeclampsia

Maria B. Rabaglino,* Emiel D. Post Uiterweer,* Arun Jeyabalan, William A. Hogge, Kirk P. Conrad

Abstract—Impaired uterine invasion by extravillous trophoblast in early gestation is implicated in the genesis of preeclampsia, a potentially lethal malady of human pregnancy. However, reasons for extravillous trophoblast dysfunction remain unclear because of virtual inaccessibility of early placental and uterine tissues from women who develop preeclampsia, and the absence of animal models in which the disease spontaneously occurs. Consequently, the possibility that deficient or defective maturation of the endometrium (decidualization) may compromise extravillous trophoblast invasion in preeclampsia remains unexplored. Using a bioinformatics approach, we tested this hypothesis identifying 396 differentially expressed genes (DEG) in chorionic villous samples from women at ≈11.5 gestational weeks who developed severe preeclampsia symptoms 6 months later compared with chorionic villous samples from normal pregnancies. A large number, 154 or 40%, overlapped with DEG associated with various stages of normal endometrial maturation before and after implantation as identified by other microarray data sets (P=4.7×10^{-14}). One-hundred and sixteen of the 154 DEG or 75% overlapped with DEG associated with normal decidualization in the absence of extravillous trophoblast, ie, late-secretory endometrium (LSE) and endometrium from tubal ectopic pregnancy (EP; P=4.2×10^{-9}). Finally, 112 of these 154 DEG or 73% changed in the opposite direction in microarray data sets related to normal endometrial maturation (P=0.01), including 16 DEG upregulated in decidual (relative to peripheral blood) natural killer cells that were downregulated in chorionic villous samples from women who developed preeclampsia (P<0.0001). Taken together, these results suggest that insufficient or defective maturation of endometrium and decidual natural killer cells during the secretory phase and early pregnancy preceded the development of preeclampsia. **(*Hypertension*. 2015;65:00-00. DOI: 10.1161/HYPERTENSIONAHA.114.04481)**

● **Online Data Supplement**

Key Words: decidualization ■ endometrial cycle ■ natural killer cell ■ pregnancy ■ trophoblast

Preeclampsia, a multiorgan disease affecting 3% to 5% of human pregnancies, is associated with significant maternal, fetal, and neonatal, morbidity and mortality.[1–4] In addition, preeclampsia increases the risk of lifelong cardiovascular and metabolic diseases for both mother and offspring.[5–7] Clear understanding of preeclampsia etiology is lacking, which hampers the identification of early predictive biomarkers and development of specific prophylactic and treatment measures.

Although knowledge of preeclampsia pathogenesis has markedly improved during the past decade, etiology remains less certain and has been only infrequently addressed, largely because of formidable investigative challenges.[8] It is widely thought that insufficient extravillous trophoblast (EVT) invasion of uterine spiral arteries starting in early pregnancy is a causal factor.[9] Consequently, there has been considerable investigation of the cellular and molecular mechanisms of EVT in this biological event. In contrast, little attention has been given to the uterine niche in which EVT invades. Perhaps the "soil," rather than or in addition to the "seed" is aberrant in women destined to develop preeclampsia.[10,11]

A major stumbling block to finding etiological factors in preeclampsia is that the disease is thought to begin in early pregnancy related to inadequate EVT invasion (vide supra). Accordingly, etiology is widely separated in time from the onset of disease symptoms, which does not occur until late-pregnancy.[12] Presently, we are not certain about who will develop preeclampsia because of lack of predictive biomarkers[13] although these are being actively pursued in numerous laboratories using discovery-based approaches. This ignorance precludes identification of women for prospective exploration

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of disease etiology in early pregnancy. Nevertheless, even if we knew who would develop preeclampsia, we cannot readily obtain the relevant tissue in which to investigate potential causes, ie, first trimester placenta and decidua, nor can we study third trimester placentas and basal plate decidua, and necessarily gain insight into disease etiology because one cannot really discern cause from effect at this late stage. Finally, preeclampsia is considered to be a disorder peculiar to human pregnancy, which makes investigation of etiological factors in animal models potentially problematic.[14]

In an attempt to overcome these challenging hurtles, we undertook a unique discovery-based approach to study the etiology of preeclampsia.[11,15] By whole-genome gene expression profiling of a collection of surplus chorionic villous sampling (CVS) tissue obtained for prenatal genetic screening, we unexpectedly noted putative decidualization marker genes, *IGFBP1*, *PAEP,* or glycodelin and *PRL* to be downregulated in women who developed preeclampsia 6 months later. Decidualization is a process of endometrial maturation that begins in the secretory phase of the menstrual cycle (predecidualization) and continues after conception and implantation. An important part of this biological process is the enrichment of decidual natural killer (dNK) cells starting in the secretory endometrium.[16] In essence, predecidualization and decidualization are a biological continuum in the preparation of the soil for the seed (EVT and conceptus, respectively).[17] Dysregulated endometrial maturation is emerging as an important precursor of recurrent pregnancy loss and infertility.[18,19] By analogy, we asked whether insufficient or defective endometrial maturation might also contribute to the etiology of preeclampsia.

The objective of the present work was to use a bioinformatics approach[20] to test the hypothesis rigorously that preeclampsia is antedated by disturbances in endometrial maturation before and after implantation. In turn, accumulating evidence links impaired decidualization and deficient dNK cell number and function to compromised extravillous trophoblast invasion, spiral artery remodeling, and placentation.[10,21–23] We reasoned that, if genes upregulated in the endometrium and dNK cells during the biological processes of (pre)decidualization[24–28] are downregulated in CVS from women destined to develop preeclampsia,[11] then this would provide critical missing, prospective evidence needed to underpin the concept of endometrial antecedents of preeclampsia.

Methods

We reanalyzed publically available microarray data sets to determine differentially expressed genes (DEG), which increase expression in LSE (predecidualization) and during endometrial maturation after implantation (decidualization); the latter in the presence or absence of extravillous trophoblast. In addition, we investigated DEG upregulated in dNK relative to peripheral blood NK cells by reanalyzing other microarray data sets. These upregulated DEG were then compared with DEG downregulated in CVS obtained at ≈11.5 gestational weeks from 4 women who developed severe, late onset preeclampsia 6 months later matched to 8 women with normal pregnancy (Results below and Table S1 in the online-only Data Supplement). This overall approach was chosen because our hypothesis was that genes which increased expression during the process of normal endometrial maturation before and after implantation will be decreased in the endometrium of women destined to develop preeclampsia (detailed Materials and Methods are presented in the online-only Data Supplement).

Results

Differentially Expressed Genes Between Chorionic Villous Samples Obtained From Preeclamptic and Normal Pregnant Women

CVS obtained at ≈11.5 gestational weeks from 4 women who developed preeclampsia 6 months later were matched to CVS from 8 women with normal pregnancy.[11] Each of the 4 CVS specimens from women who developed preeclampsia was matched for parity, gestation age at CVS within 3 days, and race with 2 unaffected control specimens.[11] In addition to fetal chorion, CVS invariably contains maternal tissue that is derived mainly from the adherent decidual basal plate with another potential source being placental septae projecting upward from the basal toward the chorionic plate containing an admixture of decidual and uterine NK cells, and EVT.[29,30] Because surplus CVS was frozen within 10 minutes of extraction in this study, they were not cleaned of maternal decidual tissue.[11] The presence of decidua in the CVS is corroborated by the transcriptomics as revealed in this work being consistent with the molecular signature of decidua (see below). Women with preeclampsia met criteria for severe disease,[4,31] and all delivered >34 weeks. There were no comorbidities except the women with preeclampsia tended to have higher body mass index (Table S1). RNA integrity was evaluated on an Agilent Bioanalyzer (RIN ≥6.0; Agilent, Santa Clara, CA[11]). Casting a wide net, we established DEG by *t* test ($P<0.05$), fold change, and J5 analysis (online-only Data Supplement). There was a total of 396 DEG between CVS obtained from women with preeclampsia compared with CVS from women with normal pregnancy outcome of which 201 were upregulated and 195 downregulated in preeclampsia (Table S2 for gene lists).

Differentially Expressed Genes Downregulated in CVS From Preeclamptic Women Are Upregulated During Normal Endometrial Maturation in the Late-Secretory Phase and Early Pregnancy

We analyzed gene expression in normal endometrium from different phases of the menstrual cycle (GSE4888[28] and GSE6364[24]) to identify the cluster of coexpressed genes strongly increasing expression in the endometrium throughout the menstrual cycle and peaking in the LSE. There was a significant overlap of 38 genes between the LSE cluster of 1315 upregulated DEG and the 195 downregulated DEG in CVS from preeclamptic compared with normal pregnant women (preeclampsia-CVS; $P<0.0001$ by Pearson χ^2 test; Figure 1A; Table S3A).

Gene expression in endometrium from tubal EP showing intermediate-decidualization morphology on H&E stained sections as described by Duncan et al[25] was first compared with gene expression in nondecidualized endometrium obtained from women with EP (E-MTAB-680). The upregulated differentially regulated genes in this decidualized endometrium (873 DEG) were then compared with the downregulated DEG in CVS from preeclamptic compared with unaffected control women. There was significant overlap of 32 genes ($P<0.0001$;

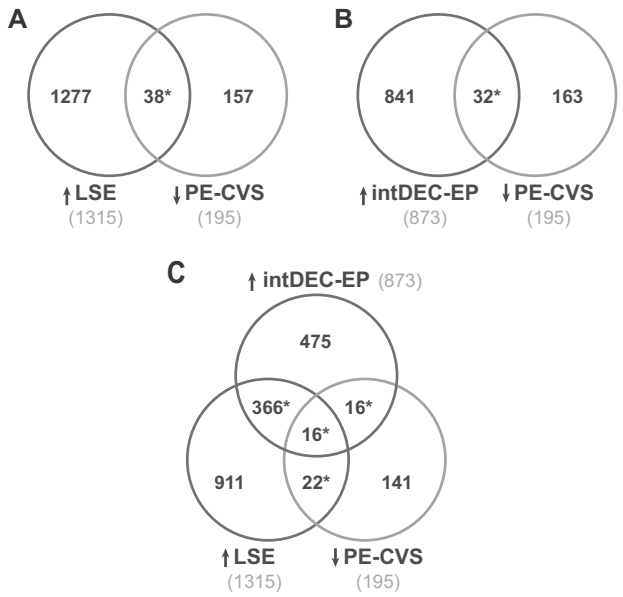

Figure 1. Clusters of genes induced during predecidualization and decidualization in ectopic pregnancy (EP) are downregulated in chorionic villous samples (CVS) from preeclamptic women. The Venn diagrams show significant overlap (*P<0.0001 by Pearson χ^2 test) between differentially expressed genes (DEG) downregulated in CVS from preeclampsia (PE) women (PE-CVS; relative to CVS from women with normal pregnancy) and DEG upregulated in (**A**) late-secretory endometrium (LSE; relative to proliferative endometrium; 38 DEG, Table S3A) and (**B**) EP endometrium with intermediate-decidualization (intDEC) changes (relative to EP endometrium without decidualization changes), which lacks extravillous trophoblast (32 DEG; Table S3B). **C**, There is significant overlap (*P<0.0001) between DEG downregulated in PE-CVS and DEG upregulated in LSE and EP endometrium with intermediate-decidualization changes (16 DEG; Table S3C).

Figure 1B; Table S3B). There was also a large overlap of 382 differentially regulated genes increasing in LSE with DEG upregulated in intermediate-decidualized endometrium (intDEC) from EP (P<0.0001). Of these, 16 significantly overlapped with the DEG downregulated in CVS from preeclamptic women (P<0.0001; Figure 1C; Table S3C). These results suggest an impairment of endometrial maturation in the late-secretory phase and early pregnancy in the women who developed preeclampsia. This impairment is independent of extravillous trophoblast because they are absent in LSE and EP endometrium (the latter verified by histology and cytokeratin immunohistochemistry).[25]

Gene expression in both intDEC and confluent-decidualized endometrium (confDEC) from women with intrauterine pregnancy (IUP) was initially compared with gene expression in non-decidualized endometrium (E-MTAB-680).[25] The upregulated DEG in IUP endometrium with intermediate (1003 DEG) and confluent (1581 DEG) decidualized changes were next compared with the 195 downregulated DEG in CVS from preeclamptic compared with normal pregnant women. 37 and 46 DEG upregulated in intDEC and confDEC from IUP, respectively, overlapped with DEG downregulated in preeclampsia-CVS (for both P<0.0001; Figure 2A and 2B; Tables S4A and S4B, respectively).

Because the decidua from intrauterine but not from EP was populated by extravillous trophoblast, we were able to estimate the potential EVT contribution to the overlap of DEG downregulated in preeclampsia-CVS and upregulated in EP and IUP endometrium matched for the extent of decidualization (intermediate). There was large overlap of 689 DEG upregulated in intDEC from EP and IUP (relative to nondecidualized endometrium; P<0.0001; Figure 2C). As illustrated in Figure 2C, 30 of these 689 DEG overlapped significantly with DEG downregulated in CVS from preeclamptic women relative to normal pregnant women (P<0.0001; Table S4C). The majority of overlapping DEG between those upregulated in intDEC from intrauterine and ectopic pregnancies and downregulated in preeclampsia-CVS were the same genes (30 of 37 for intDEC from IUP and 30 of 32 for intDEC from EP). These results reinforce the notion that the impaired endometrial maturation during early pregnancy in women who developed preeclampsia was a primary event, largely independent of extravillous trophoblast.[25]

Differentially Expressed Genes Downregulated in CVS From Preeclamptic Women Are Not Upregulated in Decidualized Endometrial Stromal Cells by Trophoblast Conditioned Medium

There was no significant overlap of endometrial genes increasing in expression after treatment of decidualized stromal cells in culture with trophoblast conditioned medium and DEG downregulated in CVS from women who developed preeclampsia compared to CVS from women with normal pregnancy outcome, with only 4 in common (Figure S1; P=0.5). These results further support the idea that impaired decidualization in the women who developed preeclampsia may be mostly independent of trophoblast influence (Results in the online-only Data Supplement and Figure S1).

Confluence of Overlapping Genes

We next investigated the confluence of DEG downregulated in CVS from women with preeclampsia relative to normal pregnancy and upregulated in LSE, intDEC from IUP and EP, and confDEC from IUP. As portrayed by the Venn diagram in Figure 3, there were 20 downregulated DEG in preeclampsia-CVS, which were upregulated in LSE but not in intDEC or confDEC; 13 DEG downregulated in preeclampsia-CVS and upregulated in intDEC and confDEC, but not in LSE (P<0.0001), and 16 DEG downregulated in preeclampsia-CVS and upregulated in all data sets related to endometrial maturation (P<0.0001). Individual DEG are presented in Table S5, and mean expression values are illustrated in Figure 4.

Figure 4 depicts $\log_2$ mean expression values for the DEG downregulated in CVS from preeclamptic compared with unaffected control women and upregulated in various states of normal endometrial maturation (Figure 3). Twenty DEG were identified as uniquely upregulated in LSE and downregulated in preeclampsia-CVS; therefore, their average expression did not increase further with decidualization in early pregnancy (Figure 4A). The average gene expression of these 20 DEG was significantly less in preeclampsia-CVS than in midsecretory endometrium (MSE) and LSE (P<0.05), and comparable to proliferative and early secretory endometrium. The 13 DEG downregulated in preeclampsia-CVS and uniquely upregulated in intDEC and confDEC, only increased slightly in LSE (by definition), and mostly rose during decidualization in early

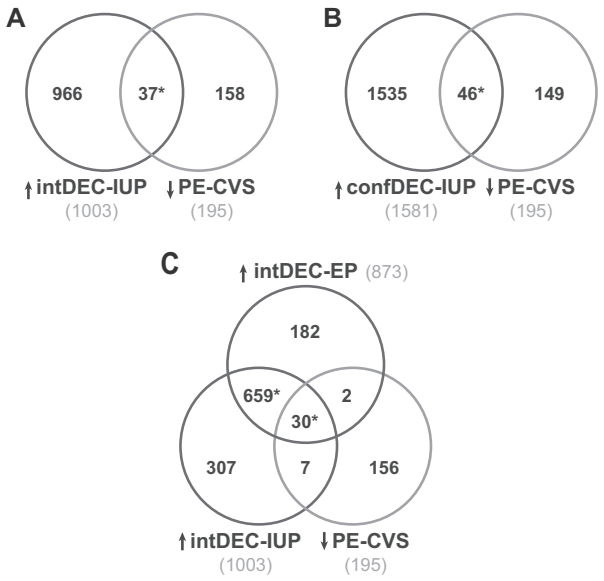

Figure 2. Clusters of genes induced during decidualization in intrauterine pregnancy (IUP) and ectopic pregnancy (EP) are downregulated in chorionic villous samples (CVS) from preeclamptic women. The Venn diagrams show significant overlap (*P<0.0001 by Pearson χ^2 test) between differentially expressed genes (DEG) downregulated in preeclampsia (PE)-CVS relative to CVS from women with normal pregnancy and DEG upregulated in (**A**) intermediate-decidualized endometrium (intDEC; 37 DEG; Table S4A) and (**B**) confluent-decidualized endometrium (confDEC; 46 DEG; Table S4B) both from IUP (relative to EP endometrium without decidualization changes) and containing extravillous trophoblast (EVT). **C**, there are 32 DEG in common between DEG downregulated in PE-CVS and DEG upregulated in EP endometrium with intermediate-decidualization changes and without EVT (Figure 1B) and 37 DEG in common between DEG downregulated in PE-CVS and DEG upregulated in IUP endometrium with intermediate-decidualization changes (EVT present). The majority of these DEG, in turn, are overlapping (30 DEG; Table S4C; *P<0.0001), suggesting minimal EVT contribution to the overlap.

pregnancy (Figure 4B). The average gene expression of these 13 DEG was markedly less in preeclampsia-CVS than in intDEC and confDEC (P<0.05). Finally, the 16 DEG downregulated in preeclampsia-CVS and upregulated in LSE, and intDEC and confDEC increased expression beginning in MSE and rose thereafter progressively (Figure 4C). Again, the average gene expression of these 16 DEG was markedly less in preeclampsia-CVS compared with intDEC and confDEC (P<0.05). The heat map shown in Figure 4D corresponds with the bar graph in Figure 4C. These observations reveal that endometrial maturation was not only impaired in early pregnancy but also during the secretory phase in the women who developed preeclampsia.

Differentially Expressed Genes Upregulated in Decidual NK Cells are Downregulated in CVS From Preeclamptic Women

16 DEG upregulated in dNK relative to CD56^{dim} and CD56^{bright} peripheral blood natural killer cells were downregulated in CVS from women who developed preeclampsia relative to women who experienced a normal pregnancy (P<0.0001; Figure 5; comprehensive Results in the online-only Data Supplement, Figure S2, and Table S6.)

Systematic Literature Search

Because the biological process of decidualization is not available in public bioinformatic databases for pathway analysis, we conducted a systematic and comprehensive literature search of all 195 DEG downregulated in CVS from preeclampsia relative to normal pregnancy. 31 were previously associated with decidualization/decidua in the literature. Of these 31 DEG, 18 were in common with the 67 DEG identified by the bioinformatics approach (Figure S3, Tables S2B and S5, and comprehensive Results in the online-only Data Supplement.)

Discussion

In the present work, we asked whether the expression of genes downregulated in early placenta (CVS) obtained from women who developed preeclampsia symptoms 6 months later significantly overlaps with expression of genes upregulated during the normal biological processes of predecidualization in the LSE, during decidualization after implantation, and in isolated decidual NK cells (relative to peripheral blood NK cells). If so, then deficient predecidualization, decidualization and dNK cell number and function in women destined to develop preeclampsia may be instrumental in disease etiology. The overall methodology was to capitalize on unique genomics data sets in the public domain, including our own data set from the first trimester placental tissue of women who developed preeclampsia, and to analyze these data sets using a bioinformatics approach to shed light on possible cause(s) of preeclampsia.

With regards to our own microarray data set,[11] we cast a wide net deliberately and identified 396 total upregulated and downregulated DEG in CVS from women who developed preeclampsia compared with those from a normal pregnancy (Table S2). Remarkably, 154 or 40% of these 396 DEG significantly overlapped with DEG associated with various stages of normal endometrial maturation before and after implantation.[24–28] Second, 75% or 116 of these 154 DEG significantly overlapped with DEG associated with normal endometrial maturation in the absence of extravillous trophoblast, ie, LSE and decidualized endometrium from ectopic tubal pregnancy. Finally, 73% or 112 of the 154 DEG either upregulated or downregulated in CVS from preeclamptic women changed in the opposite direction in the microarray data sets related to normal endometrial maturation. These findings implicate impairment of predecidualization and decidualization in the women who developed preeclampsia. Because the overlap of differential expressed genes in CVS from preeclamptic relative to normal pregnant women with DEG linked to normal endometrial maturation was mostly preserved regardless of the presence or absence of extravillous trophoblast, the results further imply that impaired endometrial maturation may be a primary event. This conclusion is underscored by the observation that DEG uniquely upregulated during normal endometrial maturation in the late-secretory phase were significantly downregulated in CVS from women who developed preeclampsia relative to normal pregnancy suggesting that the impairment of decidualization actually began before implantation (Figure 4).

As discussed earlier, we primarily focused on those DEG downregulated in CVS from preeclampsia relative to

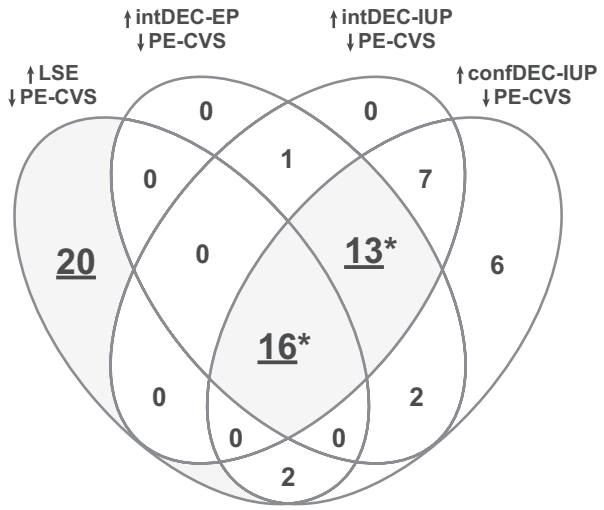

Figure 3. Confluence of gene clusters induced during (pre)decidualization and downregulated in chorionic villous samples (CVS) from preeclamptic women. There are 20 differentially expressed genes (DEG) downregulated in CVS from women that developed preeclampsia-CVS (PE-CVS; relative to CVS from women with normal pregnancy) uniquely upregulated in late-secretory endometrium (LSE, relative to proliferative endometrium). There is significant overlap (*P<0.0001 by Pearson χ^2 test) between DEG downregulated in PE-CVS and DEG upregulated in ectopic pregnancy (EP) and intrauterine pregnancy (IUP) endometrium both with intermediate-decidualization (intDEC) changes, and IUP endometrium with confluent-decidualization (confDEC) changes, but not LSE (13 DEG); and in all 4 of the data sets related to different degrees of endometrial maturation (16 DEG). See Table S5 for individual genes and Figure 4 for average expression levels of these genes.

normal pregnancy, which were upregulated during the biological process of predecidualization in the late-secretory phase (GSE4888[28] and GSE6364)[24] and decidualization after implantation in women with EP (E-MTAB-680)[25] (Figure 1). Notably, 54 of the 195 DEG downregulated in CVS from preeclamptic women were upregulated in LSE or decidualized endometrium from EP (Figure 1C). These results bolster the notion that there is impairment of endometrial maturation in the late-secretory phase and during early pregnancy in the women destined to develop preeclampsia. Included among the genes with diminished expression in decidua of CVS from the women who developed preeclampsia are those classically associated with the biological process of decidualization in the literature, including *IGFBP1*, *PAEP* or glycodelin, and *PRL* (Tables S3A–S3C and S5; Fig. S3). The results also point to a primary defect in predecidualization and decidualization rather than in extravillous trophoblast because EVTs are lacking altogether in the late-secretory phase and EVTs were absent from decidualized endometrium of EP.[25]

Further inspection of the microarray analyses from decidualization in early pregnancy revealed >1000 genes each upregulated in endometrium characterized morphologically as being intDEC or confDEC from IUP compared twith non-decidualized endometrium. 37 and 46 of these upregulated DEG, respectively, overlapped significantly with the 195 DEG downregulated in preeclampsia-CVS (Figure 2A and 2B; Tables S4A and S4B). Of further note, the vast majority of DEG upregulated in intDEC from EP and downregulated in CVS from preeclamptic compared with normal pregnant women (32 DEG), and those upregulated in intDEC from IUP and downregulated in CVS from preeclampsia women (37 DEG) overlapped themselves (30 DEG; Figure 2C; Table S4C). These observations suggest that there may have been little, if any, contribution of extravillous trophoblast to the overlap of DEG downregulated in preeclampsia-CVS and upregulated in intrauterine, and ectopic pregnancy endometrium matched for the degree of decidualization (intermediate) because the vast majority of DEG downregulated in preeclampsia-CVS were upregulated in intDEC regardless of the presence (IUP) or absence (EP) of EVT.

To scrutinize the potential contribution of EVT to impaired decidualization in preeclampsia further, we compared DEG downregulated in CVS from preeclamptic women relative to normal pregnant women with DEG upregulated in endometrial stromal cells decidualized in culture after exposure to trophoblast conditioned medium (TrCM; GSE5809).[32] There was only a nonsignificant overlap of 4 genes (Results in the online-only Data Supplement and Figure S1). This finding is supportive of a minimal EVT contribution to the overlap observed between DEG downregulated in CVS from preeclamptic women and DEG upregulated in either intDEC or confDEC from IUP (Figure 2C and S1, respectively), which is consistent with the concept that there may have been a primary defect of endometrial maturation in the women destined to develop preeclampsia. In fact, there were 47 DEG uniquely downregulated in preeclampsia-CVS, which were upregulated in intDEC or confDEC from IUP; however, the majority, 34 or 72%, were also increased in LSE or decidualized endometrium from EP without extravillous trophoblast (Figure 3).

We examined the confluence of DEG downregulated in CVS from preeclamptic women relative to normal pregnant women with DEG upregulated in LSE, intDEC from intrauterine and ectopic pregnancies, and confDEC from IUP (ie, intersection of all 4 data sets; Figure 3; Table S5). By definition, the mean expression value of 20 DEG downregulated in preeclampsia-CVS and uniquely upregulated in secretory relative to proliferative endometrium was significantly increased in MSE, maintained in LSE, but not increased further during decidualization in early pregnancy (Figure 4A). Of note, the mean expression for these 20 DEG was significantly lower in CVS from preeclamptic women compared with MSE and LSE by ≈5-fold, and comparable with proliferative and early secretory endometrium. Taken together, this analysis reinforces the idea that the impairment of endometrial maturation in the women destined to develop preeclampsia may actually have begun before pregnancy in the secretory phase, a rather startling possibility that we are currently investigating.

The mean expression of the 13 DEG downregulated in CVS from preeclamptic women and upregulated in intDEC from EP and IUP and in confDEC from IUP, but not in LSE, is shown in Figure 4B. Once again, the mean expression level in CVS from preeclamptic women was markedly reduced relative to intDEC and confDEC by ≈15-fold. These results provide further evidence that, in addition to a defect in predecidualization (vide supra), there was also impairment of decidualization during early pregnancy in the women who developed preeclampsia.

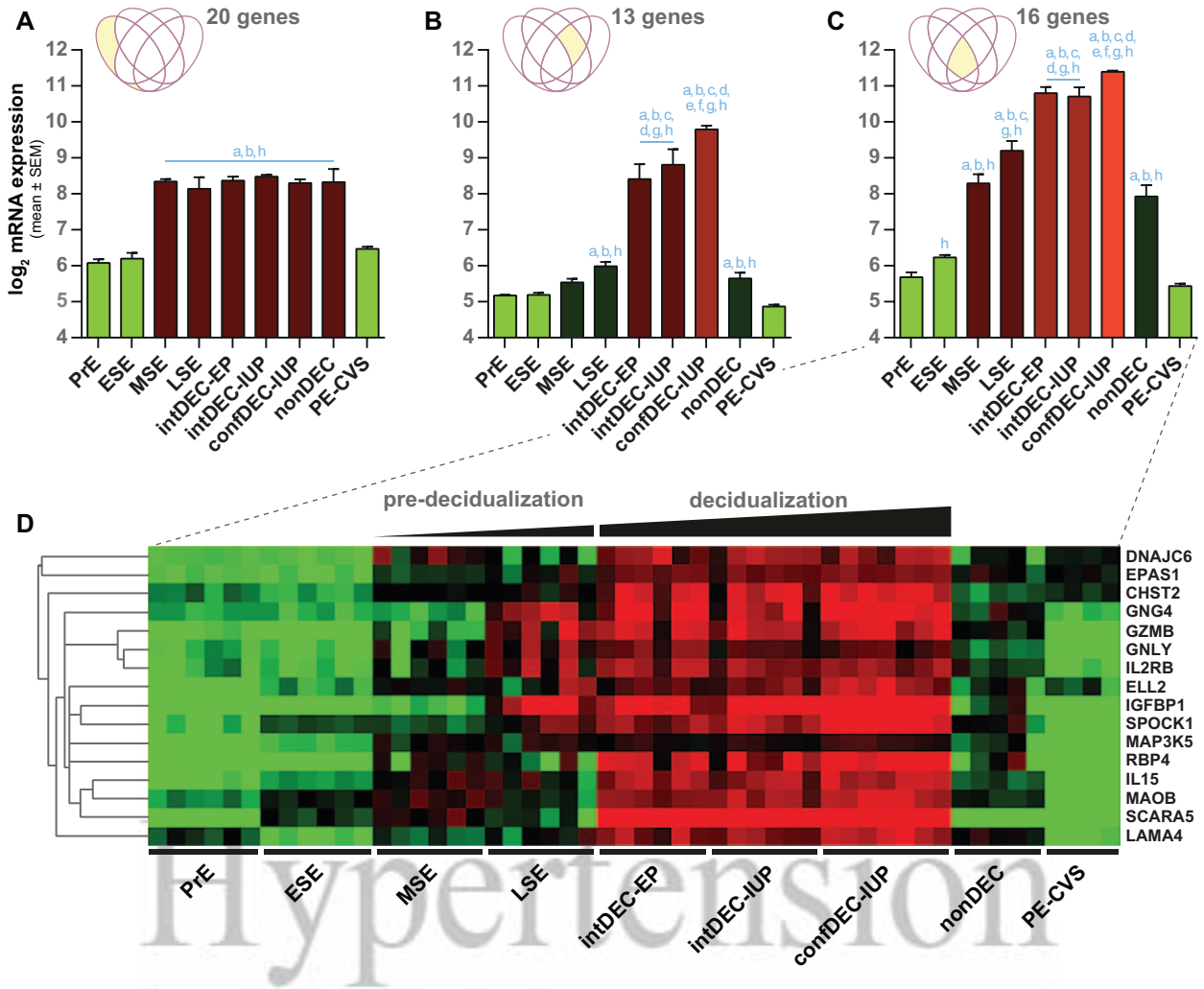

Figure 4. Average gene expression levels (log_2) in endometrium from different stages of endometrial maturation and chorionic villous samples (CVS) from preeclamptic women. **A**, Average expression of 20 differentially expressed genes (DEG) downregulated in CVS obtained from women who developed preeclampsia (PE-CVS; relative to CVS from women with normal pregnancy) and upregulated in mid- and late-secretory endometrium (MSE and LSE, respectively; relative to proliferative endometrium, PrE). **B**, Average expression of 13 DEG downregulated in PE-CVS and upregulated in intrauterine pregnancy (IUP) and ectopic pregnancy (EP) endometrium with intermediate-decidualization (intDEC) changes, and IUP endometrium with confluent-decidualization (confDEC) changes, but not LSE. **C**, Average expression for 16 DEG downregulated in PE-CVS and upregulated in all 4 of the data sets related to different degrees of endometrial maturation. **D**, Heat map corresponding to Figure 4C. The individual DEG in Figure 4A–4C are listed in Table S5. nonDEC indicates nondecidualized endometrium from EP and ESE, early secretory endometrium. Significantly different ($P<0.05$) from: a, PrE; b, ESE; c, MSE; d, LSE; e, intDEC-EP; f, intDEC-IUP; g, nonDEC; h, PE-CVS.

Finally, a core set of 16 overlapping DEG was downregulated in CVS from preeclamptic women compared with normal pregnant women and upregulated in LSE, intDEC from EP and IUP, and confDEC from IUP (Figure 4C and 4D). Relative to proliferative endometrium, the average expression level of the 16 DEG increased progressively beginning with the midsecretory phase peaking in confDEC from IUP. It is noteworthy that the mean gene expression in CVS of preeclamptic women was lower than that of MSE and LSE, and dramatically so compared with intDEC and confDEC, the latter by ≈50-fold. On balance, these data present a composite picture of Figure 4A and B, underscoring the notion that both predecidualization and decidualization were compromised in the women who developed preeclampsia.

Because DEG known to be involved in dNK function emerged from the aforementioned analyses (eg, *IL15* and *IL2RB*), we explored the overlap of DEG upregulated in isolated dNK (relative to $CD56^{dim}$ or $CD56^{bright}$ pbNK) cells and downregulated in CVS from women who developed preeclampsia relative to women with normal pregnancy outcome (Figures 5; Figure S2). Our bioinformatics analysis implicates deficient dNK cell number and function in the women who developed preeclampsia because 16 DEG upregulated in dNK were downregulated in preeclampsia-CVS. This finding is reassuring because dNK cells are an important component of the biological process of (pre)decidualization.[16]

In addition to the evidence provided by bioinformatics approaches linking the DEG downregulated in CVS from women who developed preeclampsia to deficient endometrial maturation, we pursued a different tack to marshal further evidence associating these downregulated DEG to inadequate predecidualization and decidualization. To this end, we found that 31 of the 195 downregulated DEG had been previously

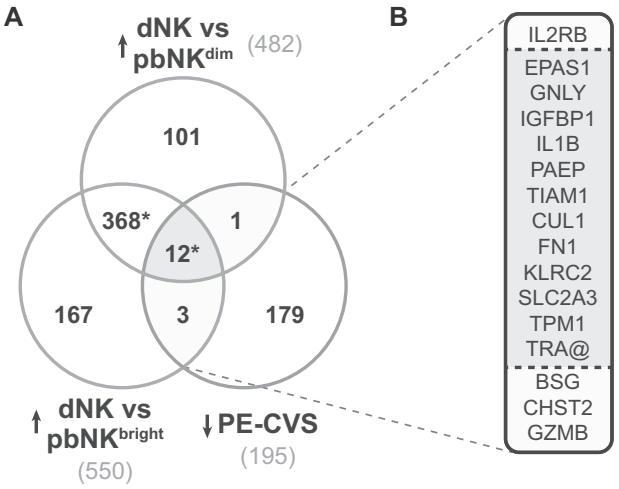

Figure 5. Clusters of genes induced in decidual natural killer cells (dNK) are downregulated in chorionic villous samples (CVS) from preeclamptic women. **A**, The Venn diagram shows significant overlap (*P<0.0001 by Pearson χ^2 test) between differentially expressed genes (DEG) downregulated in preeclampsia (PE)-CVS relative to CVS from women with normal pregnancy and DEG upregulated in decidual natural killer cells (dNK) relative to peripheral blood CD56^{dim} or CD56^{bright} NK cells. The official symbols of the overlapping genes are listed in **B**. pbNK indicates peripheral blood natural killer cells.

linked to decidualization/decidua in the literature (Figure S3; Tables S2B and S5). Thus, the systematic literature search further strengthens the argument that endometrial maturation is inadequate in the women destined to develop preeclampsia.

Aberrant endometrial maturation has been previously linked to infertility[19] and recurrent pregnancy loss.[18] Women with polycystic ovary syndrome (PCOS) have infertility and increased miscarriage rates. One potential explanation is that PCOS is associated with defective endometrial function possibly mediated through insulin resistance, hyperinsulinemia and androgen excess.[33] Dehydroepisandrosterone, which is elevated in PCOS, has been shown to inhibit the pentose phosphate pathway, thereby impairing decidualization.[34] By analogy, one might predict that defective (pre)decidualization could also predispose to other adverse pregnancy outcomes, including placental syndromes, such as preeclampsia. In fact, women with PCOS and insulin resistance are at increased risk of developing preeclampsia.[35,36] However, direct evidence to support this linkage is more difficult to obtain in the case of preeclampsia because we do not know in early pregnancy who will develop disease, first trimester chorionic and decidual tissue is not easily accessible for investigation, and molecular interrogation of delivered placenta, basal plate decidua and cells derived thereof, cannot discern between causes or consequences of the disease. The latter is highlighted by further bioinformatics analysis (data not shown), which revealed only 5 DEG in common between the 396 DEG in CVS from preeclamptic women relative to normal pregnant women and 457 DEG in decidua basalis of third trimester placentas obtained from preeclampsia compared with normal pregnant women (E-TABM-682).[37]

Nevertheless, recent evidence is consistent with the concept of endometrial antecedents of preeclampsia. First, women with high uterine vascular resistance as determined by ultrasound before elective termination in early pregnancy demonstrated impairment of dNK cell function, which is considered to be critical for spiral artery remodeling. However, pregnancy termination obviously precluded knowledge of pregnancy outcome in this study. Second, trophoblast isolated from placentas of severe preeclamptic women demonstrated increased expression of *SEMA3B*, a cytokine that impairs trophoblast invasion in vitro. After 48 hours in culture, *SEMA3B* expression spontaneously returned to normal levels suggesting that the in vivo milieu was responsible for the elevation.[38] Although consistent with the idea that defective decidualization may perturb EVT *SEMA3B* expression during early pregnancy in women destined to develop preeclampsia, once again, this interpretation is problematic because trophoblasts were isolated from delivered placentas. As such, it is not possible to discern whether the reported findings caused or resulted from the disease. Finally, women with endometriosis may have decreased risk of preeclampsia,[39] although not all agree.[40,41]

Potential Study Limitations

The validity of our conclusions mainly rests on the reliability of the original investigations, which generated the DNA microarray data sets analyzed herein. Whenever possible, we built-in redundancy or overlap by incorporating more than one data set, eg, GSE4888[28] and GSE6364[24] for endometrial gene expression in the menstrual cycle. Similarly, we tested the potential contribution of extravillous trophoblast using 3 approaches: (1) analysis of LSE obviously devoid of EVT (GSE4888[28] and GSE6364),[24] (2) comparison of endometrium from IUP and EP matched for the degree of decidualization with and without EVT influence, respectively (E-MTAB-680),[25] and (3) incubation of endometrial stromal cells decidualized in culture with trophoblast conditioned medium [GSE5809 (GEO database) or E-GEOD-5809 (EMBL-EBI database)].[32]

To our knowledge, our genome-wide gene expression study on chorionic villous sampling of women who developed preeclampsia is the only one available study in the public domain (GSE12767).[11] Surplus villi were snap frozen within 10 minutes of extraction from women undergoing CVS for prenatal genetic screening. The tremendous labor and time involved in CVS collection and the rigorous inclusion criteria affected our sample size; however, this potential limitation may be at least partially offset by the validation methods of class predictions through cross-validation (leave-one-out cross-validation) and by the rigorous statistical methods specifically designed to mitigate the potential limitation of small sample size frequently encountered in microarray studies by scarcity of tissue used[42] (Materials and Methods in the online-only Data Supplement).

Other potential limitations to the study of CVS are detailed in the original publication.[11] However, to date, CVS is the only approach to obtain first trimester chorionic tissue and decidua in women with known pregnancy outcome. Therefore, the specimens for this study provided a rare glimpse into the transcriptomics of early placenta in women who developed severe preeclampsia. Moreover, CVS is decreasing dramatically in the United States because of the emerging practice of noninvasive prenatal screening.

We cannot exclude the possibility of less decidua in CVS from the preeclamptic women compared with normal pregnant

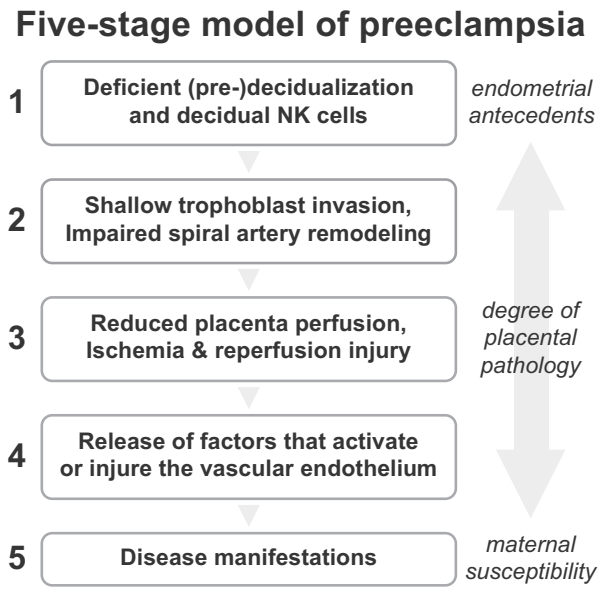

Figure 6. Five-stage model of placental preeclampsia. Based on a bioinformatics approach, the findings of this study raise the possibility that impaired endometrial maturation and deficient decidual natural killer (NK) cell number and function in the secretory phase (predecidualization) and during early pregnancy (decidualization) precede the development of preeclampsia. As (pre)decidualization and associated decidual NK cell function are emerging as important players in successful placentation, perturbation of these biological processes may contribute to the etiology of preeclampsia at least in a subset of women who develop the disease. Preeclampsia may arise in some women with little or no endometrial or placental pathology.

women as an etiological factor in the disease, ie, a quantitative difference rather than a qualitative difference or both; conceivably, this deficiency could also compromise placentation. An assumption built into this work is that extravillous trophoblast invasion and spiral artery remodeling depend on normal decidualization and dNK function (vide supra). Although, this linkage is not proven, there is growing evidence supporting the concept.[10,21–23] Moreover, because histiotrophic nutrition of the placenta and fetus in early pregnancy depends on healthy, optimally decidualized endometrial gland epithelium,[43] it seems possible that this physiological process could also have been compromised in the women who developed preeclampsia. As a final cautionary note, preeclampsia is likely to be a disease of heterogenous etiology; consequently, the evidence revealed herein of inadequate or defective (pre)decidualization may only pertain to a subset of women who develop the disease.

Perspectives

A bioinformatics approach implicates deficient or defective (pre) decidualization and decidual NK cells in the secretory phase and early pregnancy in women who developed severe preeclampsia (Figure 6). Maternal constitutional factors, such as PCOS, obesity, diabetes mellitus, and poor cardiovascular health etcetera, may compromise endometrial maturation before and during early pregnancy, thereby predisposing to preeclampsia. Conceivably, aberrant endometrial gene expression could inform targeted investigation and discovery of protein biomarkers in blood, urine or uterine fluid for women at increased risk of preeclampsia even before conception. Ultimately, designing interventions that improve endometrial maturation to facilitate normal placentation and reduce preeclampsia risk might be a logical therapeutic course of action. At the least, the present study should motivate further inquiry into the concept that deficiency or defects in (pre)decidualization and uterine NK cells antedate preeclampsia.

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Disclosures

None.

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Novelty and Significance

What Is New?

- Using a bioinformatics approach, comparison of whole-genome expression profiles of early placentas from women who developed severe preeclampsia in the context of other microarrays in the public domain related to normal endometrial maturation or decidualization revealed insufficient or defective decidualization during the secretory phase and early pregnancy in the women who developed preeclampsia.
- Further evidence that genes related to uterine natural killer cells were also adversely affected corroborates the concept of impaired endometrial maturation as a cause of preeclampsia.

What Is Relevant?

- By implicating impaired decidualization as an etiological factor in women who developed preeclampsia, this work opens up new avenues for investigation of the specific endometrial molecular defects and underlying causes, targeted biomarkers, and prophylactic or therapeutic measures to improve or repair endometrial maturation, thereby reducing preeclampsia risk.

Summary

Capitalizing on rare, early placental tissues, a bioinformatics analysis identified the impairment of endometrial maturation and decidual natural killer cells in the secretory phase and during early pregnancy in women who developed preeclampsia.

ONLINE DATA SUPPLEMENT

A bioinformatics approach reveals evidence for impaired endometrial maturation before and during early pregnancy in women who developed preeclampsia

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Materials and Methods

Microarray Datasets

Microarray dataset searches was performed in two public functional genomics data repositories: Gene Expression Omnibus (GEO) from the National Center for Biotechnology Information (NCBI) (http://www.ncbi.nlm.nih.gov/geo/) and the European Bioinformatics Institute from the European Molecular Biology Laboratory (EMBL-EBI) (http://www.ebi.ac.uk/arrayexpress/). Both data repositories support MIAME-compliant data submissions.

An important pre-condition for microarray dataset searches was the selection of microarray data in which RNA was hybridized to the Affymetrix Human Genome U133 Plus 2.0 Array (GPL570 for GEO; A-AFFY-44 for EMBL-EBI), the same platform used for the interrogation of CVS from PE and NP women (GSE12767, [1]). This pre-condition is necessary because it enables the direct comparison of the microarray data in the present work.

To search for pre-decidualization data, the keywords entered were “endometrium” AND “menstrual cycle” AND GPL570 (or A-AFFY-44). Two datasets were selected from the GEO database: GSE4888 [2] and GSE6364 [3]. The dataset GSE4888 consisted of 27 samples obtained from women with normal ovulatory cycles. Twenty-one had histologic phenotypes of proliferative (PrE; n=4), early-secretory (ESE; n=3), mid-secretory (MSE; n=8) or late-secretory (LSE; n=6) endometrium, while 6 had ambiguous histological reading. The dataset GSE6364 consisted of 37 endometrial biopsies obtained from women without pathology (n=16) or diagnosed with some degree of endometriosis (n=21). Biopsy samples of the former were from PrE (n=5), ESE (n=3), and MSE (n=8). The 21 and 16 normal endometrial samples from GSE4888 and GSE6364, respectively, were pooled. The LSE phase was only represented by the 6 samples from GSE4888. To maintain equal number of replicates per stage of the endometrial cycle, 3 samples were randomly selected from each dataset for PrE, ESE and MSE. Thus, each of the 4 menstrual cycle phases was comprised of 6 endometrial samples (n=24 total).

To search for decidualization data, the keywords employed were “endometrium” AND “decidualization” AND GPL570 (or A-AFFY-44). One dataset was selected from EMBL-EBI database: E-MTAB-680 [4]. This dataset consisted of 24 endometrial samples collected at approximately 59 days of gestation. Of these, 13 were obtained from intrauterine pregnancies (IUP) and 11 from ectopic tubal pregnancies (EP). As reported by the authors, these samples presented different degrees of endometrial decidualization as assessed by morphology in H&E stained sections. The IUP samples were classified as confluent-decidualization (confDEC-IUP, n=7) or intermediate-decidualization (intDEC-IUP, n=6), while the EP endometrial samples were intermediate-decidualization (intDEC-EP, n=6) or without decidualization changes (nonDEC, n=5). The presence or absence of trophoblast was determined by cytokeratin staining.

The keyword employed to evaluate the trophoblast influence on the decidualization process were “trophoblast” AND “decidualization” AND “endometrium” AND GPL570 (or A-AFFY-44). One dataset met the search criteria GSE5809 (GEO database) or E-GEOD-5809 (EMBL-EBI database) [5]. Human endometrial stromal cells were

decidualized in culture or left untreated serving as a control. The decidualized and non-decidualized cells were then incubated with conditioned media from human trophoblast (TrCM) for 0 (n=3), 3 (n=6) and 12 (n=5) hours. Cytotrophoblasts were isolated from placentae obtained after elective pregnancy termination (6-22 gestational weeks), and they were cultured on Matrigel-coated matrix for 48 hours before harvesting of the conditioned media.
To approximate DEG up-regulated in NK cells during the pre-decidualization or decidualization process, we wanted to compare gene expression between decidual (d)NK or endometrial (e)NK cells and peripheral blood (pb)NK cells. To this end, Koopman et al. generously provided the microarray datasets performed on dNK cells (n=9) and pbNK cells (n=10) cells [6]. In this study, decidual samples were collected from pregnant woman between 6 to 12 weeks of gestation after elective termination, and dNK cells were isolated by fluorescence-activated cell sorting. The same technique was employed to isolate pbNK cells from peripheral blood mononuclear cells of healthy donors (n=5 $CD56^{bright}$ pbNK and n=5 $CD56^{dim}$ pbNK). The isolated RNA was amplified, labeled and hybridized to the Affymetrix Human Genome U95 Version 2 Array (GPL8300). This platform is not the same as the others employed for the datasets described above. However, results from the analysis of this dataset are comparable, insofar as data imputation, normalization and transformation are the same.

Data Analysis

Data input

The Bioconductor software (http://www.bioconductor.org) for the R software environment (http://www.r-project.org) was employed for all the analyses. The gcRMA package [7] was employed to import the raw data into R, perform background correction, as well as normalize and summarize the data. Then, rows of each data set were collapsed, in order to retain the microarray probe with the highest mean value (Max mean) from the group of the genes with the same official symbol. The function applied was the “collapseRows” from the WGCNA package [8]. The purpose of row collapsing is to obtain unique identifiers for each gene in the working data set. Thus, from the original platform GPL570, containing 54675 probes, 21049 probes belonging to unique genes were retained for further analysis. For the platform GPL8300 employed in the NK cell dataset, 9127 probes related to unique genes were retained from 12625 probes.

Statistical analysis

Statistical analysis of microarray data is challenging, because often there are very few replicates per gene and thousands of genes in an experiment, which can result in high false positive (Type I error) and false negative (Type II error) rates [9] with multiple comparisons [10]. Thus, several authors adapted or developed statistical methods to overcome this potential deficiency in microarray data analysis. In this work, we applied well-known and proven statistical methods to identify DEG in the datasets. For two group comparisons, we employed moderated t-statistics, a variation of the classical t-test, developed by Smyth [11]. This algorithm has robust behavior even for small numbers of arrays. Also, compared to classical tests, the method results in more stable inference when the number of arrays is limited. For the time course experiments, we applied the

bayesian estimation of temporal regulation method, developed by Aryee et al. [12]. This algorithm uses the time dependent structure of the data and employs an empirical Bayes procedure to stabilize estimates derived from the small sample sizes. This method is also suitable for cross sectional time course experiments with one or more conditions, comparable to the datasets analyzed in this report. Thus, by choosing the right statistical methods for the analysis of microarray data, the potential limitation of small sample size frequently encountered in microarray studies by scarcity of tissue, is mitigated.

LSE (or pre-decidualization): Data from biopsy samples in GSE4888 and GSE6364 (n=24) were analyzed using time as an ordinal variable. The Bayesian Estimation of Temporal Regulation (BETR) [12] algorithm was used to identify the DEG at a False Discovery Rate (FDR) of <0.05. The first phase of the endometrial cycle (PrE) was considered as the baseline measurement and was compared to subsequent stages of the endometrial cycle, in order to correlate the differential expression among the various stages. This method, which is applied with the BETR package, provides the probabilities of differential expression for each gene in the data set. Genes with a probability higher than 99.99% were considered as differentially expressed genes (DEG).

Next, DEG selected by the BETR analysis were subjected to a supervised weighted gene co-expression network analysis employing the WGCNA package. The automatic method was employed for block-wise network construction and module detection. The co-expression similarity was raised to a soft thresholding power (β) of 12 to calculate adjacency. The adjacency for the signed network is defined as $a_{ij}=|(1+cor(xi,xj))/2|^{\beta}$ [13]. The resulting modules for each network were related to the phase of the endometrial cycle in order to identify modules or clusters of co-expressed genes showing increasing expression pattern with progression through the endometrial cycle and peaking in the late secretory phase. Gene significance (GS) was defined as the correlation of i-th gene with a temporal pattern. Module membership (MM) was defined as the correlation of the i-th gene with respect to its corresponding module (the higher the MM the more connected is the i-th gene with the other genes of the corresponding modules). The correlation coefficient of MM and GS was measured for each module, plotting MM versus GS. Higher correlation between MM and GS indicates that genes that are highly associated in a temporal pattern are also the central elements of a given module [13]. The module with the highest positive correlation between MM and GS was selected for further comparison with DEG down-regulated in PE- vs NP-CVS.

Decidualization: Data from the intDEC-EP (n=6), intDEC-IUP (n=6) and confDEC-IUP (n=7) endometrium were compared to nonDEC endometrium (n=5) from EP, all from the E-MTAB-680 database, to determine DEG up-regulated during the biological process of decidualization. The limma package was used for the statistical analysis, applying the empirical Bayes method proposed by Smyth [11]. This method calculates a moderated t-statistic for differential expression of each gene by performing a linear model fit of the data. Then, an empirical Bayes step is applied to moderate the standard errors of the estimated log-fold changes in order to produce more stable estimates, especially when the number of replicates is small. A gene was considered to be significantly differentially expressed, if both of the following conditions were met: 1) the ratio of the normalized intensity of the intermediate- or confluent-decidualization to normalized intensity of the

non-decidualized endometrial samples was higher than a 2-fold change; and 2) differences were considered statistically significant at $P \leq 0.05$.

Potential influence of extravillous trophoblast: Data from cultured endometrial stromal cells in GSE5809 (n=14) were analyzed over time (0, 3 and 12 hours incubation with TrCM) and by two conditions (decidualized and non-decidualized). The BETR algorithm [12] was used to identify DEG between decidualized endometrial cells treated with TrCM and non-decidualized endometrial stromal cells treated with TrCM at a FDR <0.05 as a function of TrCM incubation time (0, 3 and 12 hours). This method yields the probability of differential expression for each gene in the data set. Genes with a probability of 99.9% were considered as DEG. Co-expressed genes as determined by WGCNA (see above) increasing in expression by 12 hours of incubation with TrCM were selected for further comparison with DEG down-regulated in CVS from PE women relative to CVS from NP women.

Decidual NK cells: Data from dNK (n=9) were compared to $CD56^{dim}$ pbNK (n=5) or $CD56^{bright}$ pbNK (n=5) by the empirical Bayes method [11] as described above. A gene was considered to be significantly differentially expressed if both of the following conditions were met: 1) the ratio of the normalized intensity of the dNK to normalized intensity of the pbNK samples was higher than a 2-fold change; and 2) differences were considered statistically significant at $P \leq 0.05$.

Chorionic villous samples: Microarray data of CVS from PE (n=4) and NP (n=8) women in the dataset GSE12767 were compared by the empirical Bayes method [11]. The DEG were considered if both of the following conditions were met: 1) the ratio of normalized intensity in PE-CVS to normalized intensity in NP-CVS samples exceeded a 1.5-fold change; and 2) differences were considered statistically significant at $P \leq 0.05$. To expand the number of genes, down-regulated DEG determined by J5 and FC analysis were also included from Founds and coworkers (see Table 2 and table S1 in [1]). The results of the J5 analysis were taken from our original publication [1], those from t-test were obtained by re-analyzing the original Affymetrix data GSE12767, and FC data stemmed from both the original (< or > 2.0) and re-analysis (< or > 1.5).

Class prediction

In order to evaluate the performance of the selected DEG in each dataset, we performed class prediction applying the k-Nearest Neighbors (kNN) algorithm for classification and the K-fold cross validation method as classifier. The methodology was performed with the RWeka package for R [14]. Specifically, after gene selection by the corresponding statistical method, we evaluated if each sample for that dataset would be able to predict to which class it belongs according the Euclidean distance to its kNN. For this, the K-folds number was set to the n samples for each dataset, known as leave-one-out cross validation (LOOCV). The k number for KNN was set as n_i-1, for n_i being the number of samples in the class of interest. The corresponding K-fold and k numbers, and the number of correct classifications for each dataset are shown below:

Dataset	LSE	intDEC-EP	intDEC-IUP	confDEC-IUP	PE-CVS	dNK
Classes	PrE, ESE, MSE, LSE (n=24)	intDEC-EP, NonDEC (n=11)	intDEC-IUP, NonDEC (n=11)	confDEC-IUP, NonDEC (n=13)	PE-CVS, NP-CVS (n=8)	dNK, pbNK (n=19)
Class of interest	LSE (n_i = 6)	intDEC-EP (n_i = 6)	intDEC-IUP (n_i = 6)	intDEC-IUP (n_i = 7)	PE-CSV (n_i = 4)	dNK (n_i = 9)
K-fold	24	11	11	13	12	17
kNN	5	5	5	6	3	9
Number of correct classifications	24 (100%)	11 (100%)	11 (100%)	13 (100%)	12 (100%)	13 (100%)

Data comparison

The DEG down-regulated in CVS from PE relative to NP women were compared to: (i) the cluster of co-expressed endometrial genes increasing expression by the late-secretory phase of the menstrual cycle (pre-decidualization); (ii) DEG up-regulated in intermediate decidualization endometrium from IUP or EP with and without extravillous trophoblast, respectively, and confluent decidualization from IUP; (iii) the cluster of co-expressed genes increasing expression in decidualized endometrial stromal cells in culture after 12 hours of incubation with TrCM; and (iv) DEG up-regulated in decidual relative to peripheral blood NK cells. Statistical comparisons were made by the test of independence (Pearson's chi-square test) to determine the relatedness between down-regulated DEG in CVS from PE relative to NP women, and up-regulated DEG in LSE, intermediate decidualized (IUP or EP) and confluent decidualized endometrium (IUP), decidualized stromal cells in culture treated with TrCM, and decidual NK cells.

Systematic Literature Search

Systematic review of the literature was undertaken on June 1, 2014 by electronic searches in Medline through PubMed without language or publication date restrictions. The goal was to identify all publications related to decidualization that also reported one or more of the DEG *down-regulated* in CVS from PE compared to NP women (195 down-regulated DEG). To enable identification of all relevant publications, Human Genome Organization (HUGO) approved gene symbols were searched, as well as previous symbols and synonyms as listed by HUGO (http://www.genenames.org/cgi-bin/hgnc_downloads). The electronic search strategy was based on the Medical Subject Heading (MeSH) for each gene name, and when applicable, combined with title/abstract searches with all gene symbol synonyms. Synonyms that were not specific for a gene and generated too many irrelevant abstracts were omitted from the search string. By the use of Boolean operators individual gene searches (n=195) were combined with a search strategy identifying titles/abstracts related to "decidua/decidualization" based on the MeSH "decidualization" or a title/abstract search for "decidualization*". Retrieved references reporting DEG(s) down-regulated in PE-CVS and "decidua/decidualization" in the title or abstract were selected by two reviewers (EPU and KPC) who independently scrutinized the titles and abstracts. Full-text articles of any ambiguous

references were selected by one reviewer (EPU) and further scrutinized by two reviewers (EPU and KPC) to determine whether there was a clear relationship between the DEG(s) down-regulated in PE-CVS with decidua/decidualization. As a reference we included the PubMed identifier (PMID) of one of the most relevant publications for each gene related to decidualization (Table S2B). For all the DEG down-regulated in CVS from PE relative to NP women (n= 195), we applied the test of independence (Pearson's chi-square test) to determine the relatedness between DEG identified by the system biology approach (n= 67, Figure 3), and genes identified by the literature search in Pubmed (n=31).

Results

Differentially expressed genes down-regulated in CVS from preeclamptic women are not up-regulated in decidualized endometrial stromal cells by trophoblast conditioned medium (TrCM)

DEG down-regulated in PE-CVS were compared to genes induced by TrCM in cultured endometrial stromal cells decidualized *in vitro*. TrCM was obtained from cytotrophoblasts isolated from placentae between 6 and 22 weeks of gestation after elective termination and cultured on Matrigel-coated substrate for 48 hours (GSE5809 [5]). As expected, there was significant overlap between the cluster of 304 DEG increasing expression in decidualized endometrial cells incubated with TrCM and 1581 DEG up-regulated in confluent-decidualized endometrium from IUP containing EVT (69 DEG, $p<0.0001$; Figure S1). There was also significant overlap of DEG down-regulated in CVS from PE compared to NP women with DEG up-regulated in confluent-decidualized IUP endometrium (46 DEG, $p<0.0001$; Figure S1). However, there was no significant intersection of endometrial genes increasing in expression after treatment with TrCM with DEG down-regulated in PE-CVS with only 4 in common (Figure S1, $p=0.5$). These results further support the idea that impaired decidualization in the women who developed PE may be mostly independent of trophoblast influence.

Differentially expressed genes down-regulated in CVS from preeclamptic women are up-regulated in decidual NK cells

In contrast to published gene expression of peripheral blood (pb) and endometrial NK cells derived from different microarray platforms [6,15], we were able to compare gene expression between dNK and $CD56^{dim}$ pbNK or $CD56^{bright}$ pbNK cells [6], because the same microarray platform was employed. As expected, there was a large confluence of 380 DEG up-regulated in dNK relative to $CD56^{dim}$ and $CD56^{bright}$ pbNK cells ($p<0.000001$, Figure S2). There was also high overlap (112 DEG; demarcated by dotted line in Figure S2A) between DEG up-regulated in dNK relative to $CD56^{dim}$ and $CD56^{bright}$ pbNK cells, and DEG up-regulated in LSE (relative to proliferative endometrium) plus intermediate-decidualized endometrium from EP (relative to non-decidualized endometrium) in the *absence* of EVT influence ($p<0.00001$); and a high number of overlapping DEG (93 DEG; demarcated by dotted line in Figure S2B) up-regulated in dNK relative to $CD56^{dim}$ and $CD56^{bright}$ pbNK cells, and intermediate- plus confluent-decidualized endometrium from IUP (relative to non-decidualized endometrium) in the

presence of EVT influence ($p<0.00001$). The majority of these 112 and 93 DEG were the same (74 DEG, $p<0.00001$, Table S6) suggesting little contribution of EVT to the overlap. Of particular note, 16 DEG up-regulated in dNK relative to $CD56^{dim}$ and $CD56^{bright}$ pbNK cells were down-regulated in CVS from PE relative to NP women ($p<0.0001$; Figure S2A-B and Figure 5). Further, there was a significant overlap of 14 differentially expressed genes between DEG up-regulated in pbNK relative to dNK cells and DEG up-regulated in PE-CVS compared to NP-CVS ($p<0.001$; AOAH, ARFGEF2, CCL3, DDX3Y, DGKD, DHX30, GPR183, ISG20, RFTN1, RPGR, RPS4Y1, SCML2, SRF, ZNF101). This finding suggests that, if dNK cells wholly or partly derive from differentiation of pbNK cells, this process may not be optimal in the women who developed preeclampsia, because non-decidualized endometrium lacks the cues necessary for dNK differentiation.

Systematic literature search

Because the biological process of "decidualization" is not available in public bioinformatic databases for pathway analysis, we conducted a systematic and comprehensive literature search of all 195 DEG down-regulated in CVS from PE compared to NP women. Thirty-one were previously associated with decidualization/decidua in the literature (Figure S3, Table S2B and Table S5). We also evaluated the overlap of these 31 DEG identified by literature search with the overlapping DEG determined by systems biology approach, i.e., those down-regulated in PE-CVS and up-regulated in: LSE (38 DEG; Figure 1A in the text), intermediate-decidualized endometrium from EP (32 DEG; Figure 1B) and IUP (37 DEG, Figure 2A) and confluent-decidualized endometrium from IUP (46 DEG; Figure 2B), all together yielding 67 unique genes (refer to Figure 3). We found that 18 of the 31 DEG identified in the literature were in common with 67 DEG identified by systems biology ($p=0.001$). The majority (15 of 18, $p=0.03$) was up-regulated in LSE or intermediate- decidualized endometrium from EP in which EVT influence is absent.

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Table S1. Chorionic villous sampling: Clinical characteristics of subjects.

Maternal characteristics	Control (*N*= 8)	Preeclampsia[*] (*N*= 4)
Maternal age (years)	38.1 ± 3.1[†]	36.5 ± 0.6
Nulliparous	7	4
BMI at CVS	24.5 ± 4.0	29.9 ± 4.2
Gestational weeks at CVS	11.3 ± 0.6	11.4 ± 0.7
Gestational weeks at delivery	39.8 ± 1.2	37.5 ± 2.9
Birth weight (g)	3347 ± 437	2718 ± 643
Infant sex		
Female	4	2
Male	4	2
Smoking	1	0

PE case (Patient ID)	Maternal age at CVS	Gravidity	Parity	Race	BMI at CVS	Gestational age at CVS	Gestational age at delivery	Mode of delivery	Average systolic BP (prior to 20 weeks)	Average diastolic BP (prior to 20 weeks)
1 (19)	37	2	0	W	27.5	11.4	38	C-section	109	69
2 (21)	36	2	0	W	36.0	12.4	35.7	Vaginal	116	69
3 (58)	36	1	0	W	26.8	10.7	41.3	Vaginal	129	77
4 (147)	37	1	0	W	29.1	11.0	34.9	C-section	102	69

PE case (Patient ID)	Average systolic BP (at admission)	Average diastolic BP (at admission)	Proteinuria (grams per 24 h urine)	Uric acid (> 1SD for gestational age [+], mg/dL)	Birth weight (grams)	Birth weight percentile	Other
1 (19)	145	80	1.06	10.2 (+)	3241	>10	Max BP 155/90, low platelets to 87 K, creatinine 1.1 mg/dL
2 (21)	155 (145-164)	90 (80-88)	4.76	8.1 (+)	1965	<3	Max BP 164/88, IUGR
3 (58)	163 (149-177)	80 (69-95)	0.77	4.0 (-)	3265	>10	Max systolic BP 184
4 (147)	151 (134-176)	93 (84-115)	0.74	5.2 (+)	2400	>10	Max BP 176/115

[*] Preeclampsia definition is based on the National High Blood Pressure Education Program Working Group.
[†] Mean ± SD.
Adapted from [1].

Table S2. Differentially expressed genes: **(A)** up-regulated and **(B)** down-regulated in CVS obtained from women who developed late onset, severe preeclampsia compared to CVS from women who experienced normal pregnancy.

A)

Approved symbol	Approved name	HGNC ID	Location
ABL1	c-abl oncogene 1, non-receptor tyrosine kinase	HGNC:76	9q34.1
ABLIM2	actin binding LIM protein family, member 2	HGNC:19195	4p16.1
ACOT8	acyl-CoA thioesterase 8	HGNC:15919	20q13.12
ACP5	acid phosphatase 5, tartrate resistant	HGNC:124	19p13.2
ACSS1	acyl-CoA synthetase short-chain family member 1	HGNC:16091	20p11.23-p11.21
ADCY4	adenylate cyclase 4	HGNC:235	14q11.2
AHSG	alpha-2-HS-glycoprotein	HGNC:349	3q27.3
ANKRD20A1	ankyrin repeat domain 20 family, member A1	HGNC:23665	9p12
ANXA13	annexin A13	HGNC:536	8q24.13
AOAH	acyloxyacyl hydrolase (neutrophil)	HGNC:548	7p14-p12
AP1S2	adaptor-related protein complex 1, sigma 2 subunit	HGNC:560	Xp22
AP3M2	adaptor-related protein complex 3, mu 2 subunit	HGNC:570	8p11.2
ARFGEF2	ADP-ribosylation factor guanine nucleotide-exchange factor 2 (brefeldin A-inhibited)	HGNC:15853	20q13.13
ARL2	ADP-ribosylation factor-like 2	HGNC:693	11q13
ATP7B	ATPase, Cu++ transporting, beta polypeptide	HGNC:870	13q14.3
AUTS2	autism susceptibility candidate 2	HGNC:14262	7q11.22
BCL2A1	BCL2-related protein A1	HGNC:991	15q24.3
BICD1	bicaudal D homolog 1 (Drosophila)	HGNC:1049	12p11.2-p11.1
BOLA2	bolA family member 2	HGNC:29488	16p11.2
C11orf45	chromosome 11 open reading frame 45	HGNC:28584	11q24.3
DHRS4-AS1	DHRS4 antisense RNA 1	HGNC:23175	14q11.2
C2orf44	chromosome 2 open reading frame 44	HGNC:26157	2p23.3
NOP14-AS1	NOP14 antisense RNA 1	HGNC:20205	4p16.3
NDNF	neuron-derived neurotrophic factor	HGNC:26256	4q27

CARD16	caspase recruitment domain family, member 16	HGNC:33701	11q23
CASK	calcium/calmodulin-dependent serine protein kinase (MAGUK family)	HGNC:1497	Xp11.4
CCBL2	cysteine conjugate-beta lyase 2	HGNC:33238	1p22.2
CCDC159	coiled-coil domain containing 159	HGNC:26996	19p13.2
CCK	cholecystokinin	HGNC:1569	3p22.1
CCL3	chemokine (C-C motif) ligand 3	HGNC:10627	17q12
CD52	CD52 molecule	HGNC:1804	1p36
CD58	CD58 molecule	HGNC:1688	1p13
CD83	CD83 molecule	HGNC:1703	6p23
CDH15	cadherin 15, type 1, M-cadherin (myotubule)	HGNC:1754	16q24.3
CDH26	cadherin 26	HGNC:15902	20q13.33
CDH3	cadherin 3, type 1, P-cadherin (placental)	HGNC:1762	16q22.1
CDH6	cadherin 6, type 2, K-cadherin (fetal kidney)	HGNC:1765	5p13.3
CDK16	cyclin-dependent kinase 16	HGNC:8749	Xp11
CENPBD1	CENPB DNA-binding domains containing 1	HGNC:28272	16q24.3
CHST15	carbohydrate (N-acetylgalactosamine 4-sulfate 6-O) sulfotransferase 15	HGNC:18137	10q26
COL5A1	collagen, type V, alpha 1	HGNC:2209	9q34.2-q34.3
COL9A3	collagen, type IX, alpha 3	HGNC:2219	20q13.3
CR1	complement component (3b/4b) receptor 1 (Knops blood group)	HGNC:2334	1q32
CTAG2	cancer/testis antigen 2	HGNC:2492	Xq28
CXCL9	chemokine (C-X-C motif) ligand 9	HGNC:7098	4q21
DDX3Y	DEAD (Asp-Glu-Ala-Asp) box helicase 3, Y-linked	HGNC:2699	Yq11
DGKD	diacylglycerol kinase, delta 130kDa	HGNC:2851	2q37
DHX30	DEAH (Asp-Glu-Ala-His) box helicase 30	HGNC:16716	3p24.3-p22.1
DLC1	deleted in liver cancer 1	HGNC:2897	8p22
DOK4	docking protein 4	HGNC:19868	16q13
DPYSL3	dihydropyrimidinase-like 3	HGNC:3015	5q32
DPYSL4	dihydropyrimidinase-like 4	HGNC:3016	10q25.2-q26
EGR1	early growth response 1	HGNC:3238	5q23-q31
ELOVL4	ELOVL fatty acid elongase 4	HGNC:14415	6q14
EVC2	Ellis van Creveld syndrome 2	HGNC:19747	4p16.2-p16.1

EXD2	exonuclease 3'-5' domain containing 2	HGNC:20217	14q24.1
EXOSC6	exosome component 6	HGNC:19055	16q22.1
F8A1	coagulation factor VIII-associated 1	HGNC:3547	Xq28
FAM132B	family with sequence similarity 132, member B	HGNC:26727	2q37.3
FAM189A2	family with sequence similarity 189, member A2	HGNC:24820	9q21.11
FAM57A	family with sequence similarity 57, member A	HGNC:29646	17p13.3
FAT1	FAT atypical cadherin 1	HGNC:3595	4q35.2
FIZ1	FLT3-interacting zinc finger 1	HGNC:25917	19q13.42
FJX1	four jointed box 1 (Drosophila)	HGNC:17166	11p13
FKBP1A	FK506 binding protein 1A, 12kDa	HGNC:3711	20p13
FLRT2	fibronectin leucine rich transmembrane protein 2	HGNC:3761	14q24-q32
FOS	FBJ murine osteosarcoma viral oncogene homolog	HGNC:3796	14q24.3
FOSB	FBJ murine osteosarcoma viral oncogene homolog B	HGNC:3797	19q13.3
FPR3	formyl peptide receptor 3	HGNC:3828	19q13.3-q13.4
FUT6	fucosyltransferase 6 (alpha (1,3) fucosyltransferase)	HGNC:4017	19p13.3
FZD5	frizzled class receptor 5	HGNC:4043	2q33.3
GBP5	guanylate binding protein 5	HGNC:19895	1p22.2
GDNF	glial cell derived neurotrophic factor	HGNC:4232	5p13.1-p12
GPR183	G protein-coupled receptor 183	HGNC:3128	13q32.3
HBEGF	heparin-binding EGF-like growth factor	HGNC:3059	5q23
HCFC1	host cell factor C1 (VP16-accessory protein)	HGNC:4839	Xq28
HEXA	hexosaminidase A (alpha polypeptide)	HGNC:4878	15q24.1
HINFP	histone H4 transcription factor	HGNC:17850	11q23.3
HLA-DQA1	major histocompatibility complex, class II, DQ alpha 1	HGNC:4942	6p21.3
HP	haptoglobin	HGNC:5141	16q22.2
LGALSL	lectin, galactoside-binding-like	HGNC:25012	2p14
IKZF1	IKAROS family zinc finger 1 (Ikaros)	HGNC:13176	7p12.2
IL18BP	interleukin 18 binding protein	HGNC:5987	11q13
IPCEF1	interaction protein for cytohesin exchange factors 1	HGNC:21204	6q25.2
ISG20	interferon stimulated exonuclease gene 20kDa	HGNC:6130	15q26
ISL1	ISL LIM homeobox 1	HGNC:6132	5q11.2

ITFG2	integrin alpha FG-GAP repeat containing 2	HGNC:30879	12p13.33
ITGA9	integrin, alpha 9	HGNC:6145	3p21.3
KIF22	kinesin family member 22	HGNC:6391	16p11.2
KLHL6	kelch-like family member 6	HGNC:18653	3q27.3
ERAP2	endoplasmic reticulum aminopeptidase 2	HGNC:29499	5q15
LRP1	low density lipoprotein receptor-related protein 1	HGNC:6692	12q13.3
PPP1R37	protein phosphatase 1, regulatory subunit 37	HGNC:27607	19q13.32
LRRC8D	leucine rich repeat containing 8 family, member D	HGNC:16992	1p22.2
LY96	lymphocyte antigen 96	HGNC:17156	8q13.3
MAGEL2	MAGE-like 2	HGNC:6814	15q11-q12
MAP1S	microtubule-associated protein 1S	HGNC:15715	19p13.12
MAP2K7	mitogen-activated protein kinase kinase 7	HGNC:6847	19p13.3-p13.2
MED21	mediator complex subunit 21	HGNC:11473	12p12
MED22	mediator complex subunit 22	HGNC:11477	9q34.1
MGA	MGA, MAX dimerization protein	HGNC:14010	15q15
MGMT	O-6-methylguanine-DNA methyltransferase	HGNC:7059	10q26
MIIP	migration and invasion inhibitory protein	HGNC:25715	1p36.22
MPPED2	metallophosphoesterase domain containing 2	HGNC:1180	11p13
MRVI1	murine retrovirus integration site 1 homolog	HGNC:7237	11p15
MSR1	macrophage scavenger receptor 1	HGNC:7376	8p22
MTRR	5-methyltetrahydrofolate-homocysteine methyltransferase reductase	HGNC:7473	5p15.31
MYL9	myosin, light chain 9, regulatory	HGNC:15754	20q11.23
NAAA	N-acylethanolamine acid amidase	HGNC:736	4q21.1
NAP1L3	nucleosome assembly protein 1-like 3	HGNC:7639	Xq21.3-q22
NDN	necdin, melanoma antigen (MAGE) family member	HGNC:7675	15q11-q12
NINJ2	ninjurin 2	HGNC:7825	12p13
NKX2-5	NK2 homeobox 5	HGNC:2488	5q34
NMNAT3	nicotinamide nucleotide adenylyltransferase 3	HGNC:20989	3q23
NNAT	neuronatin	HGNC:7860	20q11.2-q12
NOTCH4	notch 4	HGNC:7884	6p21.3
NUB1	negative regulator of ubiquitin-like proteins 1	HGNC:17623	7q36

OPRL1	opiate receptor-like 1	HGNC:8155	20q13.33
ADCYAP1	adenylate cyclase activating polypeptide 1 (pituitary)	HGNC:241	18p11
PDE4B	phosphodiesterase 4B, cAMP-specific	HGNC:8781	1p31
PDE9A	phosphodiesterase 9A	HGNC:8795	21q22.3
PDXP	pyridoxal (pyridoxine, vitamin B6) phosphatase	HGNC:30259	22q12.3
PGA3	pepsinogen 3, group I (pepsinogen A)	HGNC:8885	11q13
PLA2G4A	phospholipase A2, group IVA (cytosolic, calcium-dependent)	HGNC:9035	1q25
PLEKHG4B	pleckstrin homology domain containing, family G (with RhoGef domain) member 4B	HGNC:29399	5p15.33
POMZP3	POM121 and ZP3 fusion	HGNC:9203	7q11.2
PPM1H	protein phosphatase, Mg2+/Mn2+ dependent, 1H	HGNC:18583	12q14.1
PPM1M	protein phosphatase, Mg2+/Mn2+ dependent, 1M	HGNC:26506	3p21.31
PPP1R13B	protein phosphatase 1, regulatory subunit 13B	HGNC:14950	14q32.33
PPP1R9A	protein phosphatase 1, regulatory subunit 9A	HGNC:14946	7q21.3
PTPRN	protein tyrosine phosphatase, receptor type, N	HGNC:9676	2q35-q36.1
RAD52	RAD52 homolog (S. cerevisiae)	HGNC:9824	12p13-p12.2
REST	RE1-silencing transcription factor	HGNC:9966	4q12
RFTN1	raftlin, lipid raft linker 1	HGNC:30278	3p24.3
RGPD1	RANBP2-like and GRIP domain containing 1	HGNC:32414	2p11.2
RIN1	Ras and Rab interactor 1	HGNC:18749	11q13.2
RPGR	retinitis pigmentosa GTPase regulator	HGNC:10295	Xp11.4
RPL31	ribosomal protein L31	HGNC:10334	2q11.2
RPS4Y1	ribosomal protein S4, Y-linked 1	HGNC:10425	Yp11.3
S100A12	S100 calcium binding protein A12	HGNC:10489	1q21
S100A8	S100 calcium binding protein A8	HGNC:10498	1q12-q22
SACS	sacsin molecular chaperone	HGNC:10519	13q11
SCARB2	scavenger receptor class B, member 2	HGNC:1665	4q21.1
SCML2	sex comb on midleg-like 2 (Drosophila)	HGNC:10581	Xp22
SEZ6L	seizure related 6 homolog (mouse)-like	HGNC:10763	22q12.1
SH3BP1	SH3-domain binding protein 1	HGNC:10824	22q13.1
SH3BP5L	SH3-binding domain protein 5-like	HGNC:29360	1q44

SIRT5	sirtuin 5	HGNC:14933	6p23
SLC13A5	solute carrier family 13 (sodium-dependent citrate transporter), member 5	HGNC:23089	17p13.1
SLC22A7	solute carrier family 22 (organic anion transporter), member 7	HGNC:10971	6p21.1
SLC25A29	solute carrier family 25 (mitochondrial carnitine/acylcarnitine carrier), member 29	HGNC:20116	14q32.2
SNRNP25	small nuclear ribonucleoprotein 25kDa (U11/U12)	HGNC:14161	16p13.3
SNRPN	small nuclear ribonucleoprotein polypeptide N	HGNC:11164	15q11.2
SNX16	sorting nexin 16	HGNC:14980	8q21.13
SORD	sorbitol dehydrogenase	HGNC:11184	15q15-q21.1
SPDEF	SAM pointed domain containing ETS transcription factor	HGNC:17257	6p21.3
SPRR2B	small proline-rich protein 2B	HGNC:11262	1q21-q22
SPRY1	sprouty homolog 1, antagonist of FGF signaling (Drosophila)	HGNC:11269	4q
SRF	serum response factor (c-fos serum response element-binding transcription factor)	HGNC:11291	6p
STAG3	stromal antigen 3	HGNC:11356	7q22
STXBP2	syntaxin binding protein 2	HGNC:11445	19p13.3-p13.2
TBC1D7	TBC1 domain family, member 7	HGNC:21066	6p23
TGIF2	TGFB-induced factor homeobox 2	HGNC:15764	20q11.23
TMEM100	transmembrane protein 100	HGNC:25607	17q23.1
TMEM106C	transmembrane protein 106C	HGNC:28775	12q13.1
TMEM216	transmembrane protein 216	HGNC:25018	11q13.1
TMEM229B	transmembrane protein 229B	HGNC:20130	14q23.3-q24.1
TMSB15A	thymosin beta 15a	HGNC:30744	Xq21.33-q22.3
TNFSF12	tumor necrosis factor (ligand) superfamily, member 12	HGNC:11927	17p13.1
TPTE	transmembrane phosphatase with tensin homology	HGNC:12023	21p11
TRIM3	tripartite motif containing 3	HGNC:10064	11p15.5
TRIM55	tripartite motif containing 55	HGNC:14215	8q13.1
TRMT2B	tRNA methyltransferase 2 homolog B (S. cerevisiae)	HGNC:25748	Xq22.1
TRPV2	transient receptor potential cation channel, subfamily V, member 2	HGNC:18082	17p11.2
TRRAP	transformation/transcription domain-associated protein	HGNC:12347	7q21.2-q22.1
TSSC1	tumor suppressing subtransferable candidate 1	HGNC:12383	2p25.3

TUBB1	tubulin, beta 1 class VI	HGNC:16257	20q13.32
UGT2B7	UDP glucuronosyltransferase 2 family, polypeptide B7	HGNC:12554	4q13
ULK3	unc-51 like kinase 3	HGNC:19703	15q24.1
WNT10B	wingless-type MMTV integration site family, member 10B	HGNC:12775	12q13
ZFP57	ZFP57 zinc finger protein	HGNC:18791	6p22.1
ZKSCAN2	zinc finger with KRAB and SCAN domains 2	HGNC:25677	16p12.1
ZMYM3	zinc finger, MYM-type 3	HGNC:13054	Xq13.1
ZNF101	zinc finger protein 101	HGNC:12881	19p13.11
ZNF383	zinc finger protein 383	HGNC:18609	19q13.13
ZNF385A	zinc finger protein 385A	HGNC:17521	12q13.13
ZNF469	zinc finger protein 469	HGNC:23216	16q24
ZNF542P	zinc finger protein 542, pseudogene	HGNC:25393	19q13.43
ZNF571	zinc finger protein 571	HGNC:25000	19q13.12
ZNF581	zinc finger protein 581	HGNC:25017	19q13.42
ZP3	zona pellucida glycoprotein 3 (sperm receptor)	HGNC:13189	7q11.23
LOC100131366	……..	……..	……..
LOC100132147	……..	……..	……..
LOC100132999	……..	……..	……..
LOC100271836	……..	……..	……..
LOC100505956	……..	……..	……..
LOC151146	……..	……..	……..
LOC643529	……..	……..	……..
LOC728377	……..	……..	……..
MGC34034	……..	……..	……..
PK155	……..	……..	……..

B)

Approved symbol	Approved name	HGNC ID	Location	Literature DEC PMID	Hyperlink
ACACA	acetyl-CoA carboxylase alpha	HGNC:84	17q21		
ACOT1 /// ACOT2	acyl-CoA thioesterase 1 /// 2	HGNC:33128 /// HGNC:18431	14q24.3		
AGAP3	ArfGAP with GTPase domain, ankyrin repeat and PH domain 3	HGNC:16923	7q36.1		
AIF1L	allograft inflammatory factor 1-like	HGNC:28904	9q34.13-q34.3		
AKR7A3	aldo-keto reductase family 7, member A3 (aflatoxin aldehyde reductase)	HGNC:390	1p36.13		
ALDH1A2	aldehyde dehydrogenase 1 family, member A2	HGNC:15472	15q21.2		
ALS2CL	ALS2 C-terminal like	HGNC:20605	3p21.31		
AOC1	amine oxidase, copper containing 1	HGNC:80	7q36.1	20668027	http://www.ncbi.nlm.nih.gov/pubmed/?term=20668027
APC	adenomatous polyposis coli	HGNC:583	5q21-q22		
AQP2	aquaporin 2 (collecting duct)	HGNC:634	12q12-q13		
ART1	ADP-ribosyltransferase 1	HGNC:723	11p15		
ASCL2	achaete-scute family bHLH transcription factor 2	HGNC:739	11p15.5		
AXIN1	axin 1	HGNC:903	16p13.3		
BAIAP2L1	BAI1-associated protein 2-like 1	HGNC:21649	7q22.1		
BDKRB2	bradykinin receptor B2	HGNC:1030	14q32.1-q32.2		
BEAN1	brain expressed, associated with NEDD4, 1	HGNC:24160	16q21		
BEX1	brain expressed, X-linked 1	HGNC:1036	Xq22.1		
BLNK	B-cell linker	HGNC:14211	10q23.2-q23.33		
BSG	basigin (Ok blood group)	HGNC:1116	19p13.3	12141934	http://www.ncbi.nlm.nih.gov/pubmed/?term=12141934
C12orf75	chromosome 12 open reading frame 75	HGNC:35164	12q23.3		
C3	complement component 3	HGNC:1318	19p13.3-p13.2	8311932	http://www.ncbi.nlm.nih.gov/pubmed/?term=8311932

C4BPA	complement component 4 binding protein, alpha	HGNC:1325	1q32		
C7orf71	chromosome 7 open reading frame 71	HGNC:22364	7p15.2		
CA12	carbonic anhydrase XII	HGNC:1371	15q22		
CA2	carbonic anhydrase II	HGNC:1373	8q21.2	9692790	http://www.ncbi.nlm.nih.gov/pubmed/?term=9692790
CC2D2B	coiled-coil and C2 domain containing 2B	HGNC:31666	10q23.33		
CCDC113	coiled-coil domain containing 113	HGNC:25002	16q21		
CCDC125	coiled-coil domain containing 125	HGNC:28924	5q13.2		
CFH /// CFHR1	complement factor H /// complement factor H-related 1	HGNC:4883 /// HGNC:4888	1q32		
CHERP	calcium homeostasis endoplasmic reticulum protein	HGNC:16930	19p13.1		
CHRDL1	chordin-like 1	HGNC:29861	Xq23		
CHST2	carbohydrate (N-acetylglucosamine-6-O) sulfotransferase 2	HGNC:1970	3q24		
CHST6	carbohydrate (N-acetylglucosamine 6-O) sulfotransferase 6	HGNC:6938	16q22		
CLASP2	cytoplasmic linker associated protein 2	HGNC:17078	3p24.3		
CLCN7	chloride channel, voltage-sensitive 7	HGNC:2025	16p13		
CLDN6	claudin 6	HGNC:2048	16p13.3		
CMAHP	cytidine monophospho-N-acetylneuraminic acid hydroxylase, pseudogene	HGNC:2098	6p23-p22		
CMTM4	CKLF-like MARVEL transmembrane domain containing 4	HGNC:19175	16q22.1-q22.3		
COL27A1	collagen, type XXVII, alpha 1	HGNC:22986	9q33.1		
COTL1	coactosin-like F-actin binding protein 1	HGNC:18304	16q24.1		
CPM	carboxypeptidase M	HGNC:2311	12q15		
CPXM2	carboxypeptidase X (M14 family), member 2	HGNC:26977	10q26		
CRH	corticotropin releasing hormone	HGNC:2355	8q13	159239	http://www.ncbi.nlm.nih.gov/pubmed/?term=159239
CRYBB1	crystallin, beta B1	HGNC:2397	22q12.1		
CUL1	cullin 1	HGNC:2551	7q36.1		

CYP4A11	cytochrome P450, family 4, subfamily A, polypeptide 11	HGNC:2642	1p33		
CYTH2	cytohesin 2	HGNC:9502	19q13.32		
DEPDC7	DEP domain containing 7	HGNC:29899	11p13		
DHRS2	dehydrogenase/reductase (SDR family) member 2	HGNC:18349	14q11.2		
DLGAP1	discs, large (Drosophila) homolog-associated protein 1	HGNC:2905	18p11.3		
DNAJC6	DnaJ (Hsp40) homolog, subfamily C, member 6	HGNC:15469	1p31.3		
DSC2	desmocollin 2	HGNC:3036	18q12.1		
DUXAP10	double homeobox A pseudogene 10	HGNC:32189	14q11.2		
EFCAB2	EF-hand calcium binding domain 2	HGNC:28166	1q44		
EGLN3	egl-9 family hypoxia-inducible factor 3	HGNC:14661	14q12		
ELL2	elongation factor, RNA polymerase II, 2	HGNC:17064	5q15		
EPAS1	endothelial PAS domain protein 1	HGNC:3374	2p21-p16		
ERAP2	endoplasmic reticulum aminopeptidase 2	HGNC:29499	5q15	24331737	http://www.ncbi.nlm.nih.gov/pubmed/?term=24331737
ERO1L	ERO1-like (S. cerevisiae)	HGNC:13280	14q22.1		
F11R	F11 receptor	HGNC:14685	1q21.2-q21.3		
F2R	coagulation factor II (thrombin) receptor	HGNC:3537	5q13	12549865	http://www.ncbi.nlm.nih.gov/pubmed/?term=12549865
FABP7	fatty acid binding protein 7, brain	HGNC:3562	6q22-q23		
FAM3B	family with sequence similarity 3, B	HGNC:1253	21q22.3		
FHL2	four and a half LIM domains 2	HGNC:3703	2q12.2		
FKBP11	FK506 binding protein 11, 19 kDa	HGNC:18624	12q13.12		
FLT4	fms-related tyrosine kinase 4	HGNC:3767	5q34-q35	11297624	http://www.ncbi.nlm.nih.gov/pubmed/?term=11297624
FN1	fibronectin 1	HGNC:3778	2q34	14611684	http://www.ncbi.nlm.nih.gov/pubmed/?term=14611684
FSTL3	follistatin-like 3 (secreted glycoprotein)	HGNC:3973	19p13	15130517	http://www.ncbi.nlm.nih.gov/pubmed/?term=15130517
GATA1	GATA binding protein 1 (globin transcription factor 1)	HGNC:4170	Xp11.23		
GDA	guanine deaminase	HGNC:4212	9q21.13		

GNG4	guanine nucleotide binding protein (G protein), gamma 4	HGNC:4407	1q42.3		
GNG7	guanine nucleotide binding protein (G protein), gamma 7	HGNC:4410	19p13.3		
GNLY	granulysin	HGNC:4414	2p12-q11	21623991	http://www.ncbi.nlm.nih.gov/pubmed/?term=21623991
GOLGA8B	golgin A8 family, member B	HGNC:31973	15q14		
GPR158	G protein-coupled receptor 158	HGNC:23689	10p12.31		
GTPBP2	GTP binding protein 2	HGNC:4670	6p21		
GZMB	granzyme B (granzyme 2, cytotoxic T-lymphocyte-associated serine esterase 1)	HGNC:4709	14q11.2	16451356	http://www.ncbi.nlm.nih.gov/pubmed/?term=16451356
HBE1	hemoglobin, epsilon 1	HGNC:4830	11p15.5		
HBZ	hemoglobin, zeta	HGNC:4835	16p13.3		
HCAR3	hydroxycarboxylic acid receptor 3	HGNC:16824	12q24.31		
HIVEP2	human immunodeficiency virus type I enhancer binding protein 2	HGNC:4921	6q23-q24		
HOXB7	homeobox B7	HGNC:5118	17q21.32		
HPS3	Hermansky-Pudlak syndrome 3	HGNC:15597	3q24		
HTR2B	5-hydroxytryptamine (serotonin) receptor 2B, G protein-coupled	HGNC:5294	2q36.3-q37.1		
HYDIN	HYDIN, axonemal central pair apparatus protein	HGNC:19368	16q22.2		
IGFBP1	insulin-like growth factor binding protein 1	HGNC:5469	7p13-p12	1385468	http://www.ncbi.nlm.nih.gov/pubmed/?term=1385468
IGKC	immunoglobulin kappa constant	HGNC:5716	2p11.2		
IL15	interleukin 15	HGNC:5977	4q31	10952908	http://www.ncbi.nlm.nih.gov/pubmed/10952908
IL1B	interleukin 1, beta	HGNC:5992	2q14	16860880	http://www.ncbi.nlm.nih.gov/pubmed/?term=16860880
IL1RL1	interleukin 1 receptor-like 1	HGNC:5998	2q12	23300625	http://www.ncbi.nlm.nih.gov/pubmed/?term=23300625
IL2RB	interleukin 2 receptor, beta	HGNC:6009	22q13	21248224	http://www.ncbi.nlm.nih.gov/pubmed/?term=21248224
INPP4B	inositol polyphosphate-4-phosphatase, type II, 105kDa	HGNC:6075	4q31.1		
ITCH	itchy E3 ubiquitin protein ligase	HGNC:13890	20q11.22		

ITGB6	integrin, beta 6	HGNC:6161	2q24.2		
KCNH2	potassium voltage-gated channel, subfamily H (eag-related), member 2	HGNC:6251	7q36.1		
KCNIP3	Kv channel interacting protein 3, calsenilin	HGNC:15523	2q21.1		
KCNQ1	potassium voltage-gated channel, KQT-like subfamily, member 1	HGNC:6294	11p15.5		
KISS1R	KISS1 receptor	HGNC:4510	19p13.3	24225150	http://www.ncbi.nlm.nih.gov/pubmed/?term=24225150
KLRC2	killer cell lectin-like receptor subfamily C, member 2	HGNC:6375	12p13	16488482	http://www.ncbi.nlm.nih.gov/pubmed/?term=16488482
KRT14	keratin 14	HGNC:6416	17q21.2		
LAIR2	leukocyte-associated immunoglobulin-like receptor 2	HGNC:6478	19q13.4		
LAMA4	laminin, alpha 4	HGNC:6484	6q21		
LIAS	lipoic acid synthetase	HGNC:16429	4p14		
LIPH	lipase, member H	HGNC:18483	3q27		
LIPT1	lipoyltransferase 1	HGNC:29569	2q11.2		
LSS	lanosterol synthase (2,3-oxidosqualene-lanosterol cyclase)	HGNC:6708	21q22.3		
LTBR	lymphotoxin beta receptor (TNFR superfamily, member 3)	HGNC:6718	12p13		
MAGEB6	melanoma antigen family B, 6	HGNC:23796	Xp22.12		
MAOB	monoamine oxidase B	HGNC:6834	Xp11.4-p11.3		
MAP3K5	mitogen-activated protein (3)kinase 5	HGNC:6857	6q22.33		
MLIP	muscular LMNA-interacting protein	HGNC:21355	6p12.2-p12.1		
MMD	monocyte to macrophage differentiation-associated	HGNC:7153	17q		
MMP12	matrix metallopeptidase 12 (macrophage elastase)	HGNC:7158	11q22.3	20802175	http://www.ncbi.nlm.nih.gov/pubmed/?term=20802175
MUC15	mucin 15, cell surface associated	HGNC:14956	11p14.3	17720698	http://www.ncbi.nlm.nih.gov/pubmed/?term=17720698
MUC4	mucin 4, cell surface associated	HGNC:7514	3q29		
MVK	mevalonate kinase	HGNC:7530	12q24		
NDP	Norrie disease (pseudoglioma)	HGNC:7678	Xp11.4-p11.3	16035034	http://www.ncbi.nlm.nih.gov/pubmed/?term=16035034

NDUFV2	NADH dehydrogenase (ubiquinone) flavoprotein 2, 24kDa	HGNC:7717	18p11.22		
NOG	noggin	HGNC:7866	17q22	11158592	http://www.ncbi.nlm.nih.gov/pubmed/?term=11158592
NOTUM	notum pectinacetylesterase homolog (Drosophila)	HGNC:27106	17q25.3		
NTN1	netrin 1	HGNC:8029	17p13-p12		
NTN4	netrin 4	HGNC:13658	12q22		
NUDT13	nudix (nucleoside diphosphate linked moiety X)-type motif 13	HGNC:18827	10q22.3		
OXGR1	oxoglutarate (alpha-ketoglutarate) receptor1	HGNC:4531	13q32.2		
P4HA3	prolyl 4-hydroxylase, alpha polypeptide III	HGNC:30135	11q13		
PAEP	progestagen-associated endometrial protein	HGNC:8573	9q34	3194393	http://www.ncbi.nlm.nih.gov/pubmed/?term=3194393
PARP16	poly (ADP-ribose) polymerase family, member 16	HGNC:26040	15q22.2		
PAWR	PRKC, apoptosis, WT1, regulator	HGNC:8614	12q21.2		
PDE4C	phosphodiesterase 4C, cAMP-specific	HGNC:8782	19p13.11	14715868	http://www.ncbi.nlm.nih.gov/pubmed/?term=14715868
PITPNC1	phosphatidylinositol transfer protein, cytoplasmic 1	HGNC:21045	17q24.3		
PLAC8	placenta-specific 8	HGNC:19254	4q21.22		
PLCXD2	phosphatidylinositol-specific phospholipase C, X domain containing 2	HGNC:26462	3q13.2		
PPDPF	pancreatic progenitor cell differentiation and proliferation factor	HGNC:16142	20q13.33		
PPP1R3C	protein phosphatase 1, regulatory subunit 3C	HGNC:9293	10q23-q24		
PRDM1	PR domain containing 1, with ZNF domain	HGNC:9346	6q21		
PRG2	proteoglycan 2, bone marrow (natural killer cell activator, eosinophil granule major basic protein)	HGNC:9362	11q12		
PRKAB2	protein kinase, AMP-activated, beta 2 non-catalytic subunit	HGNC:9379	1q21.2		
PRL	prolactin	HGNC:9445	6p22.3	10611264	http://www.ncbi.nlm.nih.gov/pubmed/?term=10611264

PSG11	pregnancy specific beta-1-glycoprotein 11	HGNC:9516	19q13.2		
PTPRS	protein tyrosine phosphatase, receptor type, S	HGNC:9681	19p13.3		
PVR	poliovirus receptor	HGNC:9705	19q13.2		
RAB12	RAB12, member RAS oncogene family	HGNC:31332	18p11.22		
RBP4	retinol binding protein 4, plasma	HGNC:9922	10q23.33		
RHD	Rh blood group, D antigen	HGNC:10009	1p36.11		
RNF14	ring finger protein 14	HGNC:10058	5q23.3-q31.1		
RORB	RAR-related orphan receptor B	HGNC:10259	9q22		
RSRC1	arginine/serine-rich coiled-coil 1	HGNC:24152	3q25.32		
RUFY3	RUN and FYVE domain containing 3	HGNC:30285	4q13.3		
SART3	squamous cell carcinoma antigen recognized by T cells 3	HGNC:16860	12q24.11		
SCARA5	scavenger receptor class A, member 5 (putative)	HGNC:28701	8p21.1	21858178	http://www.ncbi.nlm.nih.gov/pubmed/?term=21858178
SDHAP1	succinate dehydrogenase complex, subunit A, flavoprotein pseudogene 1	HGNC:32455	3q29		
SEC24D	SEC24 family member D	HGNC:10706	4q26		
SEMA3C	sema domain, immunoglobulin domain (Ig), short basic domain, secreted, (semaphorin) 3C	HGNC:10725	7q21-q31		
SERPINA3	serpin peptidase inhibitor, clade A (alpha-1 antiproteinase, antitrypsin), member 3	HGNC:16	14q32.1	8951488	http://www.ncbi.nlm.nih.gov/pubmed/?term=8951488
SFI1	Sfi1 homolog, spindle assembly associated (yeast)	HGNC:29064	22q12.2		
SGSM1	small G protein signaling modulator 1	HGNC:29410	22q11.23		
SLC13A4	solute carrier family 13 (sodium/sulfate symporter), member 4	HGNC:15827	7q33		
SLC16A6	solute carrier family 16, member 6	HGNC:10927	17q24.2		
SLC25A15	solute carrier family 25 (mitochondrial carrier; ornithine transporter) member 15	HGNC:10985	13q14		
SLC26A7	solute carrier family 26 (anion exchanger), member 7	HGNC:14467	8q23		
SLC2A3	solute carrier family 2 (facilitated glucose	HGNC:11007	12p13.3	12915684	http://www.ncbi.nlm.nih.gov

	transporter), member 3				/pubmed/?term=12915684
SLC36A1	solute carrier family 36 (proton/amino acid symporter), member 1	HGNC:18761	5q33.1		
SLC44A3	solute carrier family 44, member 3	HGNC:28689	1p22.1		
SLCO4A1	solute carrier organic anion transporter family, member 4A1	HGNC:10953	20q13.1		
SNX25	sorting nexin 25	HGNC:21883	4q35.1		
SOWAHC	sosondowah ankyrin repeat domain family member C	HGNC:26149	2q13		
SP140L	SP140 nuclear body protein-like	HGNC:25105	2q37.1		
SPG20	spastic paraplegia 20 (Troyer syndrome)	HGNC:18514	13q13.1		
SPOCK1	sparc/osteonectin, cwcv and kazal-like domains proteoglycan (testican) 1	HGNC:11251	5q31.2		
SSTR1	somatostatin receptor 1	HGNC:11330	14q13		
ST3GAL6	ST3 beta-galactoside alpha-2,3-sialyltransferase 6	HGNC:18080	3q12.2		
ST6GALNAC4	ST6 (alpha-N-acetyl-neuraminyl-2,3-beta-galactosyl-1,3)-N-acetylgalactosaminide alpha-2,6-sialyltransferase 4	HGNC:17846	9q34		
SYCP2L	synaptonemal complex protein 2-like	HGNC:21537	6p24.2		
SYT1	synaptotagmin I	HGNC:11509	12q21.2		
TES	testis derived transcript (3 LIM domains)	HGNC:14620	7q31.2		
THBS4	thrombospondin 4	HGNC:11788	5q13		
THUMPD2	THUMP domain containing 2	HGNC:14890	2p22.2		
TIAM1	T-cell lymphoma invasion and metastasis 1	HGNC:11805	21q22.1		
TLN2	talin 2	HGNC:15447	15q15-q21		
TMC4	transmembrane channel-like 4	HGNC:22998	19q13.42		
TMEM62	transmembrane protein 62	HGNC:26269	15q15.2	24767823	http://www.ncbi.nlm.nih.gov/pubmed/?term=24767823
TOX3	TOX high mobility group box family member 3	HGNC:11972	16q12.1		
TPM1	tropomyosin 1 (alpha)	HGNC:12010	15q22.1		
TRA@	T cell receptor alpha locus	HGNC:12027	14q11.2		

TREML2	triggering receptor expressed on myeloid cells-like 2	HGNC:21092	6p21.1		
TSTD1	thiosulfate sulfurtransferase (rhodanese)-like domain containing 1	HGNC:35410	1q23.3		
TTC18	tetratricopeptide repeat domain 18	HGNC:30726	10q22.3		
USP5	ubiquitin specific peptidase 5 (isopeptidase T)	HGNC:12628	12p13		
WT1	Wilms tumor 1	HGNC:12796	11p13	11739471	http://www.ncbi.nlm.nih.gov/pubmed/?term=11739471
XRCC4	X-ray repair complementing defective repair in Chinese hamster cells 4	HGNC:12831	5q14.2		
ZFP62	ZFP62 zinc finger protein	HGNC:23241	5q35.3		
ZNF165	zinc finger protein 165	HGNC:12953	6p21		
LOC153546	……..	……..	……..		
LOC440157	……..	……..	……..		
FLJ13744	……..	……..	……..		

Table S3. Overlap of DEG down-regulated in CVS from PE relative to NP women, and up-regulated in: **(A)** late secretory endometrium (Figure 1A); **(B)** intermediate-decidualized endometrium from ectopic pregnancy (Figure 1B); **(C)** intermediate-decidualized endometrium from EP and late secretory endometrium (Figure 1C)

A)

Approved symbol	Approved name	HGNC ID	Location
BAIAP2L1	BAI1-associated protein 2-like 1	HGNC:21649	7q22.1
BDKRB2	bradykinin receptor B2	HGNC:1030	14q32.1-q32.2
BLNK	B-cell linker	HGNC:14211	10q23.2-q23.33
C12orf75	chromosome 12 open reading frame 75	HGNC:35164	12q23.3
C3	complement component 3	HGNC:1318	19p13.3-p13.2
C4BPA	complement component 4 binding protein, alpha	HGNC:1325	1q32
CA12	carbonic anhydrase XII	HGNC:1371	15q22
CHST2	carbohydrate (N-acetylglucosamine-6-O) sulfotransferase 2	HGNC:1970	3q24

CMAHP	cytidine monophospho-N-acetylneuraminic acid hydroxylase, pseudogene	HGNC:2098	6p23-p22
DNAJC6	DnaJ (Hsp40) homolog, subfamily C, member 6	HGNC:15469	1p31.3
DSC2	desmocollin 2	HGNC:3036	18q12.1
ELL2	elongation factor, RNA polymerase II, 2	HGNC:17064	5q15
EPAS1	endothelial PAS domain protein 1	HGNC:3374	2p21-p16
ERO1L	ERO1-like (S. cerevisiae)	HGNC:13280	14q22.1
GNG4	guanine nucleotide binding protein (G protein), gamma 4	HGNC:4407	1q42.3
GNLY	granulysin	HGNC:4414	2p12-q11
GTPBP2	GTP binding protein 2	HGNC:4670	6p21
GZMB	granzyme B (granzyme 2, cytotoxic T-lymphocyte-associated serine esterase 1)	HGNC:4709	14q11.2
HPS3	Hermansky-Pudlak syndrome 3	HGNC:15597	3q24
IGFBP1	insulin-like growth factor binding protein 1	HGNC:5469	7p13-p12
IL15	interleukin 15	HGNC:5977	4q31
IL1B	interleukin 1, beta	HGNC:5992	2q14
IL2RB	interleukin 2 receptor, beta	HGNC:6009	22q13
INPP4B	inositol polyphosphate-4-phosphatase, type II, 105kDa	HGNC:6075	4q31.1
ITGB6	integrin, beta 6	HGNC:6161	2q24.2
LAMA4	laminin, alpha 4	HGNC:6484	6q21
MAOB	monoamine oxidase B	HGNC:6834	Xp11.4-p11.3
MAP3K5	mitogen-activated protein kinase kinase kinase 5	HGNC:6857	6q22.33
PAEP	progestagen-associated endometrial protein	HGNC:8573	9q34
PVR	poliovirus receptor	HGNC:9705	19q13.2
RBP4	retinol binding protein 4, plasma	HGNC:9922	10q23-q24
RUFY3	RUN and FYVE domain containing 3	HGNC:30285	4q13.3
SCARA5	scavenger receptor class A, member 5 (putative)	HGNC:28701	8p21.1
SLCO4A1	solute carrier organic anion transporter family, member 4A1	HGNC:10953	20q13.1
SPOCK1	sparc/osteonectin, cwcv and kazal-like domains proteoglycan (testican) 1	HGNC:11251	5q31.2
TES	testis derived transcript (3 LIM domains)	HGNC:14620	7q31.2
TIAM1	T-cell lymphoma invasion and metastasis 1	HGNC:11805	21q22.1
ZNF165	zinc finger protein 165	HGNC:12953	6p21

B)

Approved symbol	Approved name	HGNC ID	Location
AIF1L	allograft inflammatory factor 1-like	HGNC:28904	9q34.13-q34.3
CHRDL1	chordin-like 1	HGNC:29861	Xq23
CHST2	carbohydrate (N-acetylglucosamine-6-O) sulfotransferase 2	HGNC:1970	3q24
COTL1	coactosin-like 1 (Dictyostelium)	HGNC:18304	16q24.1
DNAJC6	DnaJ (Hsp40) homolog, subfamily C, member 6	HGNC:15469	1p31.3
ELL2	elongation factor, RNA polymerase II, 2	HGNC:17064	5q15
EPAS1	endothelial PAS domain protein 1	HGNC:3374	2p21-p16
F2R	coagulation factor II (thrombin) receptor	HGNC:3537	5q13
GNG4	guanine nucleotide binding protein (G protein), gamma 4	HGNC:4407	1q42.3
GNLY	granulysin	HGNC:4414	2p12-q11
GZMB	granzyme B (granzyme 2, cytotoxic T-lymphocyte-associated serine esterase 1)	HGNC:4709	14q11.2
HTR2B	5-hydroxytryptamine (serotonin) receptor 2B, G protein-coupled	HGNC:5294	2q36.3-q37.1
IGFBP1	insulin-like growth factor binding protein 1	HGNC:5469	7p13-p12
IL15	interleukin 15	HGNC:5977	4q31
IL1RL1	interleukin 1 receptor-like 1	HGNC:5998	2q12
IL2RB	interleukin 2 receptor, beta	HGNC:6009	22q13
LAMA4	laminin, alpha 4	HGNC:6484	6q21
LIPH	lipase, member H	HGNC:18483	3q27
MAOB	monoamine oxidase B	HGNC:6834	Xp11.4-p11.3
MAP3K5	mitogen-activated protein kinase kinase kinase 5	HGNC:6857	6q22.33
MUC15	mucin 15, cell surface associated	HGNC:14956	11p14.3
NDP	Norrie disease (pseudoglioma)	HGNC:7678	Xp11.4-p11.3
NOG	noggin	HGNC:7866	17q22
P4HA3	prolyl 4-hydroxylase, alpha polypeptide III	HGNC:30135	11q13
PPP1R3C	protein phosphatase 1, regulatory subunit 3C	HGNC:9293	10q23-q24
PRL	prolactin	HGNC:9445	6p22.3
RBP4	retinol binding protein 4, plasma	HGNC:9922	10q23-q24

SCARA5	scavenger receptor class A, member 5 (putative)	HGNC:28701	8p21.1
SLC25A15	solute carrier family 25 (mitochondrial carrier; ornithine transporter) member 15	HGNC:10985	13q14
SNX25	sorting nexin 25	HGNC:21883	4q35.1
SPOCK1	sparc/osteonectin, cwcv and kazal-like domains proteoglycan (testican) 1	HGNC:11251	5q31.2
TLN2	talin 2	HGNC:15447	15q15-q21

C)

Approved symbol	Approved name	HGNC ID	Location
CHST2	carbohydrate (N-acetylglucosamine-6-O) sulfotransferase 2	HGNC:1970	3q24
DNAJC6	DnaJ (Hsp40) homolog, subfamily C, member 6	HGNC:15469	1p31.3
ELL2	elongation factor, RNA polymerase II, 2	HGNC:17064	5q15
EPAS1	endothelial PAS domain protein 1	HGNC:3374	2p21-p16
GNG4	guanine nucleotide binding protein (G protein), gamma 4	HGNC:4407	1q42.3
GNLY	granulysin	HGNC:4414	2p12-q11
GZMB	granzyme B (granzyme 2, cytotoxic T-lymphocyte-associated serine esterase 1)	HGNC:4709	14q11.2
IGFBP1	insulin-like growth factor binding protein 1	HGNC:5469	7p13-p12
IL15	interleukin 15	HGNC:5977	4q31
IL2RB	interleukin 2 receptor, beta	HGNC:6009	22q13
LAMA4	laminin, alpha 4	HGNC:6484	6q21
MAOB	monoamine oxidase B	HGNC:6834	Xp11.4-p11.3
MAP3K5	mitogen-activated protein kinase kinase kinase 5	HGNC:6857	6q22.33
RBP4	retinol binding protein 4, plasma	HGNC:9922	10q23-q24
SCARA5	scavenger receptor class A, member 5 (putative)	HGNC:28701	8p21.1
SPOCK1	sparc/osteonectin, cwcv and kazal-like domains proteoglycan (testican) 1	HGNC:11251	5q31.2

Table S4. Overlap of DEG down-regulated in CVS from PE women compared to NP, and up-regulated in: **(A)** intermediate-decidualized endometrium from IUP (Figure 2A); **(B)** confluent-decidualized endometrium from IUP (Figure 2B); **(C)** intermediate-decidualized endometrium from IUP and EP (Figure 2C).

A)

Approved symbol	Approved name	HGNC ID	Location
ACOT1 /// ACOT2	acyl-CoA thioesterase 1 /// acyl-CoA thioesterase 2	HGNC:33128 /// HGNC:18431	14q24.3
AIF1L	allograft inflammatory factor 1-like	HGNC:28904	9q34.13
CFH /// CFHR1	complement factor H /// complement factor H-related 1	HGNC:4883 /// HGNC:4888	1q32
CHRDL1	chordin-like 1	HGNC:29861	Xq23
CHST2	carbohydrate (N-acetylglucosamine-6-O) sulfotransferase 2	HGNC:1970	3q24
COTL1	coactosin-like 1 (Dictyostelium)	HGNC:18304	16q24.1
CPXM2	carboxypeptidase X (M14 family), member 2	HGNC:26977	10q26
DNAJC6	DnaJ (Hsp40) homolog, subfamily C, member 6	HGNC:15469	1p31.3
ELL2	elongation factor, RNA polymerase II, 2	HGNC:17064	5q15
EPAS1	endothelial PAS domain protein 1	HGNC:3374	2p21-p16
F2R	coagulation factor II (thrombin) receptor	HGNC:3537	5q13
GNG4	guanine nucleotide binding protein (G protein), gamma 4	HGNC:4407	1q42.3
GNLY	granulysin	HGNC:4414	2p12-q11
GZMB	granzyme B (granzyme 2, cytotoxic T-lymphocyte-associated serine esterase 1)	HGNC:4709	14q11.2
HTR2B	5-hydroxytryptamine (serotonin) receptor 2B, G protein-coupled	HGNC:5294	2q36.3-q37.1
IGFBP1	insulin-like growth factor binding protein 1	HGNC:5469	7p13-p12
IL15	interleukin 15	HGNC:5977	4q31
IL1RL1	interleukin 1 receptor-like 1	HGNC:5998	2q12
IL2RB	interleukin 2 receptor, beta	HGNC:6009	22q13
LAMA4	laminin, alpha 4	HGNC:6484	6q21
LIPH	lipase, member H	HGNC:18483	3q27
LSS	lanosterol synthase (2,3-oxidosqualene-lanosterol cyclase)	HGNC:6708	21q22.3
MAOB	monoamine oxidase B	HGNC:6834	Xp11.4-p11.3

MAP3K5	mitogen-activated protein kinase kinase kinase 5	HGNC:6857	6q22.33
MUC15	mucin 15, cell surface associated	HGNC:14956	11p14.3
NDP	Norrie disease (pseudoglioma)	HGNC:7678	Xp11.4-p11.3
P4HA3	prolyl 4-hydroxylase, alpha polypeptide III	HGNC:30135	11q13
PPP1R3C	protein phosphatase 1, regulatory subunit 3C	HGNC:9293	10q23-q24
PRG2	proteoglycan 2, bone marrow (natural killer cell activator, eosinophil granule major basic protein)	HGNC:9362	11q12
PRKAB2	protein kinase, AMP-activated, beta 2 non-catalytic subunit	HGNC:9379	1q21.2
PRL	prolactin	HGNC:9445	6p22.3
RBP4	retinol binding protein 4, plasma	HGNC:9922	10q23-q24
SCARA5	scavenger receptor class A, member 5 (putative)	HGNC:28701	8p21.1
SLC16A6	solute carrier family 16, member 6	HGNC:10927	17q24.2
SLC25A15	solute carrier family 25 (mitochondrial carrier; ornithine transporter) member 15	HGNC:10985	13q14
SNX25	sorting nexin 25	HGNC:21883	4q35.1
SPOCK1	sparc/osteonectin, cwcv and kazal-like domains proteoglycan (testican) 1	HGNC:11251	5q31.2

B)

Approved symbol	Approved name	HGNC ID	Location
ACOT1 /// ACOT2	acyl-CoA thioesterase 1 /// acyl-CoA thioesterase 2	HGNC:33128 /// HGNC:18431	14q24.3
AIF1L	allograft inflammatory factor 1-like	HGNC:28904	9q34.13-q34.3
CFH /// CFHR1	complement factor H /// complement factor H-related 1	HGNC:4883 /// HGNC:4888	1q32
CHRDL1	chordin-like 1	HGNC:29861	Xq23
CHST2	carbohydrate (N-acetylglucosamine-6-O) sulfotransferase 2	HGNC:1970	3q24
COTL1	coactosin-like 1 (Dictyostelium)	HGNC:18304	16q24.1
CPXM2	carboxypeptidase X (M14 family), member 2	HGNC:26977	10q26
DNAJC6	DnaJ (Hsp40) homolog, subfamily C, member 6	HGNC:15469	1p31.3
EFCAB2	EF-hand calcium binding domain 2	HGNC:28166	1q44
ELL2	elongation factor, RNA polymerase II, 2	HGNC:17064	5q15

EPAS1	endothelial PAS domain protein 1	HGNC:3374	2p21-p16
F2R	coagulation factor II (thrombin) receptor	HGNC:3537	5q13
FSTL3	follistatin-like 3 (secreted glycoprotein)	HGNC:3973	19p13
GNG4	guanine nucleotide binding protein (G protein), gamma 4	HGNC:4407	1q42.3
GNLY	granulysin	HGNC:4414	2p12-q11
GZMB	granzyme B (granzyme 2, cytotoxic T-lymphocyte-associated serine esterase 1)	HGNC:4709	14q11.2
HTR2B	5-hydroxytryptamine (serotonin) receptor 2B, G protein-coupled	HGNC:5294	2q36.3-q37.1
IGFBP1	insulin-like growth factor binding protein 1	HGNC:5469	7p13-p12
IL15	interleukin 15	HGNC:5977	4q31
IL1B	interleukin 1, beta	HGNC:5992	2q14
IL1RL1	interleukin 1 receptor-like 1	HGNC:5998	2q12
IL2RB	interleukin 2 receptor, beta	HGNC:6009	22q13
LAMA4	laminin, alpha 4	HGNC:6484	6q21
LIPH	lipase, member H	HGNC:18483	3q27
LSS	lanosterol synthase (2,3-oxidosqualene-lanosterol cyclase)	HGNC:6708	21q22.3
MAOB	monoamine oxidase B	HGNC:6834	Xp11.4-p11.3
MAP3K5	mitogen-activated protein kinase kinase kinase 5	HGNC:6857	6q22.33
MUC15	mucin 15, cell surface associated	HGNC:14956	11p14.3
NDP	Norrie disease (pseudoglioma)	HGNC:7678	Xp11.4-p11.3
NOG	noggin	HGNC:7866	17q22
P4HA3	prolyl 4-hydroxylase, alpha polypeptide III	HGNC:30135	11q13
PPP1R3C	protein phosphatase 1, regulatory subunit 3C	HGNC:9293	10q23-q24
PRG2	proteoglycan 2, bone marrow (natural killer cell activator, eosinophil granule major basic protein)	HGNC:9362	11q12
PRKAB2	protein kinase, AMP-activated, beta 2 non-catalytic subunit	HGNC:9379	1q21.2
PRL	prolactin	HGNC:9445	6p22.3
PVR	poliovirus receptor	HGNC:9705	19q13.2
RBP4	retinol binding protein 4, plasma	HGNC:9922	10q23-q24
RNF14	ring finger protein 14	HGNC:10058	5q23.3-q31.1
RORB	RAR-related orphan receptor B	HGNC:10259	9q22
SCARA5	scavenger receptor class A, member 5 (putative)	HGNC:28701	8p21.1

SERPINA3	serpin peptidase inhibitor, clade A (alpha-1 antiproteinase, antitrypsin), member 3	HGNC:16	14q32.1
SLC16A6	solute carrier family 16, member 6	HGNC:10927	17q24.2
SNX25	sorting nexin 25	HGNC:21883	4q35.1
SPOCK1	sparc/osteonectin, cwcv and kazal-like domains proteoglycan (testican) 1	HGNC:11251	5q31.2
TLN2	talin 2	HGNC:15447	15q15-q21
WT1	Wilms tumor 1	HGNC:12796	11p13

C)

Approved symbol	Approved name	HGNC ID	Location
AIF1L	allograft inflammatory factor 1-like	HGNC:28904	9q34.13-q34.3
CHRDL1	chordin-like 1	HGNC:29861	Xq23
CHST2	carbohydrate (N-acetylglucosamine-6-O) sulfotransferase 2	HGNC:1970	3q24
COTL1	coactosin-like 1 (Dictyostelium)	HGNC:18304	16q24.1
DNAJC6	DnaJ (Hsp40) homolog, subfamily C, member 6	HGNC:15469	1p31.3
ELL2	elongation factor, RNA polymerase II, 2	HGNC:17064	5q15
EPAS1	endothelial PAS domain protein 1	HGNC:3374	2p21-p16
F2R	coagulation factor II (thrombin) receptor	HGNC:3537	5q13
GNG4	guanine nucleotide binding protein (G protein), gamma 4	HGNC:4407	1q42.3
GNLY	granulysin	HGNC:4414	2p12-q11
GZMB	granzyme B (granzyme 2, cytotoxic T-lymphocyte-associated serine esterase 1)	HGNC:4709	14q11.2
HTR2B	5-hydroxytryptamine (serotonin) receptor 2B, G protein-coupled	HGNC:5294	2q36.3-q37.1
IGFBP1	insulin-like growth factor binding protein 1	HGNC:5469	7p13-p12
IL15	interleukin 15	HGNC:5977	4q31
IL1RL1	interleukin 1 receptor-like 1	HGNC:5998	2q12
IL2RB	interleukin 2 receptor, beta	HGNC:6009	22q13
LAMA4	laminin, alpha 4	HGNC:6484	6q21
LIPH	lipase, member H	HGNC:18483	3q27

MAOB	monoamine oxidase B	HGNC:6834	Xp11.4-p11.3
MAP3K5	mitogen-activated protein kinase kinase kinase 5	HGNC:6857	6q22.33
MUC15	mucin 15, cell surface associated	HGNC:14956	11p14.3
NDP	Norrie disease (pseudoglioma)	HGNC:7678	Xp11.4-p11.3
P4HA3	prolyl 4-hydroxylase, alpha polypeptide III	HGNC:30135	11q13
PPP1R3C	protein phosphatase 1, regulatory subunit 3C	HGNC:9293	10q23-q24
PRL	prolactin	HGNC:9445	6p22.3
RBP4	retinol binding protein 4, plasma	HGNC:9922	10q23-q24
SCARA5	scavenger receptor class A, member 5 (putative)	HGNC:28701	8p21.1
SLC25A15	solute carrier family 25 (mitochondrial carrier; ornithine transporter) member 15	HGNC:10985	13q14
SNX25	sorting nexin 25	HGNC:21883	4q35.1
SPOCK1	sparc/osteonectin, cwcv and kazal-like domains proteoglycan (testican) 1	HGNC:11251	5q31.2

Table S5. Down-regulated genes in CVS from PE relative to NP women linked to decidualization by the bioinformatics approach employed in this work and in the literature.

Approved symbol	Approved name	HGNC ID	Location	DEG down-regulated in PE-CVS and up-regulated in:				
				LSE	intDEC-EP	intDEC-IUP	confDEC-IUP	Pubmed DEC
ACOT1 /// ACOT2	acyl-CoA thioesterase 1 /// 2	HGNC:33128 /// HGNC:18431	14q24.3			X	X	
AIF1L	allograft inflammatory factor 1-like	HGNC:28904	9q34.13-q34.3		X	X	X	
BAIAP2L1	BAI1-associated protein 2-like 1	HGNC:21649	7q22.1	X				
BDKRB2	bradykinin receptor B2	HGNC:1030	14q32.1-q32.2	X				
BLNK	B-cell linker	HGNC:14211	10q23.2-q23.33	X				
C12orf75	chromosome 12 open reading frame 75	HGNC:35164	12q23.3	X				
C3	complement component 3	HGNC:1318	19p13.3-p13.2	X				X
C4BPA	complement component 4 binding protein, alpha	HGNC:1325	1q32	X				
CA12	carbonic anhydrase XII	HGNC:1371	15q22	X				

CFH /// CFHR1	complement factor H /// complement factor H-related 1	HGNC:4883 /// HGNC:4888	1q32			X	X	
CHRDL1	chordin-like 1	HGNC:29861	Xq23		X	X	X	
CHST2	carbohydrate (N-acetylglucosamine-6-O) sulfotransferase 2	HGNC:1970	3q24	X	X	X	X	
CMAHP	cytidine monophospho-N-acetylneuraminic acid hydroxylase, pseudogene	HGNC:2098	6p23-p22	X				
COTL1	coactosin-like F-actin binding protein 1	HGNC:18304	16q24.1		X	X	X	
CPXM2	carboxypeptidase X (M14 family), member 2	HGNC:26977	10q26			X	X	
DNAJC6	DnaJ (Hsp40) homolog, subfamily C, member 6	HGNC:15469	1p31.3	X	X	X	X	
DSC2	desmocollin 2	HGNC:3036	18q12.1	X				
EFCAB2	EF-hand calcium binding domain 2	HGNC:28166	1q44				X	
ELL2	elongation factor, RNA polymerase II, 2	HGNC:17064	5q15	X	X	X	X	
EPAS1	endothelial PAS domain protein 1	HGNC:3374	2p21-p16	X	X	X	X	
ERO1L	ERO1-like (S. cerevisiae)	HGNC:13280	14q22.1	X				
F2R	coagulation factor II (thrombin) receptor	HGNC:3537	5q13		X	X	X	X
FSTL3	follistatin-like 3 (secreted glycoprotein)	HGNC:3973	19p13				X	X
GNG4	guanine nucleotide binding protein (G protein), gamma 4	HGNC:4407	1q42.3	X	X	X	X	
GNLY	granulysin	HGNC:4414	2p12-q11	X	X	X	X	X
GTPBP2	GTP binding protein 2	HGNC:4670	6p21	X				
GZMB	granzyme B (granzyme 2, cytotoxic T-lymphocyte-associated serine esterase 1)	HGNC:4709	14q11.2	X	X	X	X	X
HPS3	Hermansky-Pudlak syndrome 3	HGNC:15597	3q24	X				
HTR2B	5-hydroxytryptamine (serotonin) receptor 2B, G protein-coupled	HGNC:5294	2q36.3-q37.1		X	X	X	
IGFBP1	insulin-like growth factor binding protein 1	HGNC:5469	7p13-p12	X	X	X	X	X
IL15	interleukin 15	HGNC:5977	4q31	X	X	X	X	X
IL1B	interleukin 1, beta	HGNC:5992	2q14	X			X	X
IL1RL1	interleukin 1 receptor-like 1	HGNC:5998	2q12		X	X	X	X
IL2RB	interleukin 2 receptor, beta	HGNC:6009	22q13	X	X	X	X	X
INPP4B	inositol polyphosphate-4-phosphatase, type II, 105kDa	HGNC:6075	4q31.1	X				

ITGB6	integrin, beta 6	HGNC:6161	2q24.2	X				
LAMA4	laminin, alpha 4	HGNC:6484	6q21	X	X	X	X	
LIPH	lipase, member H	HGNC:18483	3q27		X	X	X	
LSS	lanosterol synthase (2,3-oxidosqualene-lanosterol cyclase)	HGNC:6708	21q22.3			X	X	
MAOB	monoamine oxidase B	HGNC:6834	Xp11.4-p11.3	X	X	X	X	
MAP3K5	mitogen-activated protein kinase kinase kinase 5	HGNC:6857	6q22.33	X	X	X	X	
MUC15	mucin 15, cell surface associated	HGNC:14956	11p14.3		X	X	X	X
NDP	Norrie disease (pseudoglioma)	HGNC:7678	Xp11.4		X	X	X	X
NOG	noggin	HGNC:7866	17q22		X		X	X
P4HA3	prolyl 4-hydroxylase, alpha polypeptide III	HGNC:30135	11q13		X	X	X	
PAEP	progestagen-associated endometrial protein	HGNC:8573	9q34	X				X
PPP1R3C	protein phosphatase 1, regulatory subunit 3C	HGNC:9293	10q23-q24		X	X	X	
PRG2	proteoglycan 2, bone marrow (natural killer cell activator, eosinophil granule major basic protein)	HGNC:9362	11q12			X	X	
PRKAB2	protein kinase, AMP-activated, beta 2 non-catalytic subunit	HGNC:9379	1q21.2			X	X	
PRL	prolactin	HGNC:9445	6p22.3		X	X	X	X
PVR	poliovirus receptor	HGNC:9705	19q13.2	X			X	
RBP4	retinol binding protein 4, plasma	HGNC:9922	10q23.33	X	X	X	X	
RNF14	ring finger protein 14	HGNC:10058	5q23.3-q31.1				X	
RORB	RAR-related orphan receptor B	HGNC:10259	9q22				X	
RUFY3	RUN and FYVE domain containing 3	HGNC:30285	4q13.3	X				
SCARA5	scavenger receptor class A, member 5 (putative)	HGNC:28701	8p21.1	X	X	X	X	X
SERPINA3	serpin peptidase inhibitor, clade A (alpha-1 antiproteinase, antitrypsin), member 3	HGNC:16	14q32.1				X	X
SLC16A6	solute carrier family 16, member 6	HGNC:10927	17q24.2			X	X	
SLC25A15	solute carrier family 25 (mitochondrial carrier; ornithine transporter) member 15	HGNC:10985	13q14		X	X		
SLCO4A1	solute carrier organic anion transporter family, member 4A1	HGNC:10953	20q13.1	X				
SNX25	sorting nexin 25	HGNC:21883	4q35.1		X	X	X	

SPOCK1	sparc/osteonectin, cwcv and kazal-like domains proteoglycan (testican) 1	HGNC:11251	5q31.2	X	X	X	X		
TES	testis derived transcript (3 LIM domains)	HGNC:14620	7q31.2	X					
TIAM1	T-cell lymphoma invasion and metastasis 1	HGNC:11805	21q22.1	X					
TLN2	talin 2	HGNC:15447	15q15-q21		X		X		
WT1	Wilms tumor 1	HGNC:12796	11p13				X	X	
ZNF165	zinc finger protein 165	HGNC:12953	6p21	X					

Table S6. Overlap of DEG up-regulated in decidual NK cells, late secretory endometrium, intermediate-decidualized endometrium from ectopic and intrauterine pregnancy, as well as confluent-decidualized endometrium from IUP.

Approved symbol	Approved name	HGNC ID	Location
ADCY3	adenylate cyclase 3	HGNC:234	2p23.3
ADM	adrenomedullin	HGNC:259	11p15.4
APOBEC3G	apolipoprotein B mRNA editing enzyme, catalytic polypeptide-like 3G	HGNC:17357	22q13.1-q13.2
APOC2	apolipoprotein C-II	HGNC:609	19q13.2
APOD	apolipoprotein D	HGNC:612	3q29
ARHGEF6	Rac/Cdc42 guanine nucleotide exchange factor (GEF) 6	HGNC:685	Xq26
C19orf10	chromosome 19 open reading frame 10	HGNC:16948	19p13.3
C1R	complement component 1, r subcomponent	HGNC:1246	12p13.31
CAMK1	calcium/calmodulin-dependent protein kinase I	HGNC:1459	3p25.3
CAPG	capping protein (actin filament), gelsolin-like	HGNC:1474	2p11.2
CCR1	chemokine (C-C motif) receptor 1	HGNC:1602	3p21
CD38	CD38 molecule	HGNC:1667	4p15.32
CD3E	CD3e molecule, epsilon (CD3-TCR complex)	HGNC:1674	11q23
CD59	CD59 molecule, complement regulatory protein	HGNC:1689	11p13
CD96	CD96 molecule	HGNC:16892	3p13-q13.2
CDHR1	cadherin-related family member 1	HGNC:14550	10q23.1
CLU	clusterin	HGNC:2095	8p21-p12

CORO1A	coronin, actin binding protein, 1A	HGNC:2252	16p11.2
CRYAB	crystallin, alpha B	HGNC:2389	11q22.3-q23.1
CTSA	cathepsin A	HGNC:9251	20q13.12
CTSL	cathepsin L	HGNC:2537	9q21.33
DLEU1	deleted in lymphocytic leukemia 1 (non-protein coding)	HGNC:13747	13q14.3
DOCK10	dedicator of cytokinesis 10	HGNC:23479	2q36.3
DPYSL2	dihydropyrimidinase-like 2	HGNC:3014	8p22-p21
EPAS1	endothelial PAS domain protein 1	HGNC:3374	2p21-p16
FAM49A	family with sequence similarity 49, member A	HGNC:25373	2p24.3
FASLG	Fas ligand (TNF superfamily, member 6)	HGNC:11936	1q23
FCER1G	Fc fragment of IgE, high affinity I, receptor for; gamma polypeptide	HGNC:3611	1q23
FGR	feline Gardner-Rasheed sarcoma viral oncogene homolog	HGNC:3697	1p36.2-p36.1
FKBP1A	FK506 binding protein 1A, 12kDa	HGNC:3711	20p13
GADD45A	growth arrest and DNA-damage-inducible, alpha	HGNC:4095	1p31.2
GAS1	growth arrest-specific 1	HGNC:4165	9q21.3-q22
GLUL	glutamate-ammonia ligase	HGNC:4341	1q31
GNLY	granulysin	HGNC:4414	2p12-q11
GPX3	glutathione peroxidase 3 (plasma)	HGNC:4555	5q23
GZMA	granzyme A (granzyme 1, cytotoxic T-lymphocyte-associated serine esterase 3)	HGNC:4708	5q11-q12
HOPX	HOP homeobox	HGNC:24961	4q12
IGFBP1	insulin-like growth factor binding protein 1	HGNC:5469	7p13-p12
IGFBP2	insulin-like growth factor binding protein 2, 36kDa	HGNC:5471	2q33-q34
IL1B	interleukin 1, beta	HGNC:5992	2q14
ITGA1	integrin, alpha 1	HGNC:6134	5q11.1
ITGAD	integrin, alpha D	HGNC:6146	16p13.1-p11
ITM2A	integral membrane protein 2A	HGNC:6173	Xq13.3
KIR3DL1	killer cell immunoglobulin-like receptor, three domains, long cytoplasmic tail, 1	HGNC:6338	19q13.4
KIR3DL2	killer cell immunoglobulin-like receptor, three domains, long cytoplasmic tail, 2	HGNC:6339	19q13.4
LCP2	lymphocyte cytosolic protein 2 (SH2 domain containing leukocyte protein of 76kDa)	HGNC:6529	5q35.1

LILRP2	leukocyte immunoglobulin-like receptor pseudogene 2	HGNC:15497	19q13.4
MDFIC	MyoD family inhibitor domain containing	HGNC:28870	7q31.1-q31.2
MIR22HG	MIR22 host gene (non-protein coding)	HGNC:28219	17p13.3
MTHFD2	methylenetetrahydrofolate dehydrogenase (NADP+ dependent) 2, methenyltetrahydrofolate cyclohydrolase	HGNC:7434	2p13.1
MYL9	myosin, light chain 9, regulatory	HGNC:15754	20q11.23
NCAM1	neural cell adhesion molecule 1	HGNC:7656	11q23.2
NUCB2	nucleobindin 2	HGNC:8044	11p15.1
NUPR1	nuclear protein, transcriptional regulator, 1	HGNC:29990	16p11.2
OSTF1	osteoclast stimulating factor 1	HGNC:8510	9q13-q21.2
PECAM1	platelet/endothelial cell adhesion molecule 1	HGNC:8823	17q23.3
PLCG2	phospholipase C, gamma 2 (phosphatidylinositol-specific)	HGNC:9066	16q24.1
PNP	purine nucleoside phosphorylase	HGNC:7892	14q11.2
PSTPIP1	proline-serine-threonine phosphatase interacting protein 1	HGNC:9580	15q24-q25.1
PTGIS	prostaglandin I2 (prostacyclin) synthase	HGNC:9603	20q13
PTPN6	protein tyrosine phosphatase, non-receptor type 6	HGNC:9658	12p13.31
RRAS2	related RAS viral (r-ras) oncogene homolog 2	HGNC:17271	11p15.2
SEPT11	septin 11	HGNC:25589	4q21.1
SERPING1	serpin peptidase inhibitor, clade G (C1 inhibitor), member 1	HGNC:1228	11q12.1
SKAP2	src kinase associated phosphoprotein 2	HGNC:15687	7p15.2
SLA	Src-like-adaptor	HGNC:10902	8q24.22
SPINK2	serine peptidase inhibitor, Kazal type 2 (acrosin-trypsin inhibitor)	HGNC:11245	4q12
SPTSSA	serine palmitoyltransferase, small subunit A	HGNC:20361	14q13.1
TGM2	transglutaminase 2	HGNC:11778	20q12
TIMP3	TIMP metallopeptidase inhibitor 3	HGNC:11822	22q12.3
TRAF3IP3	TRAF3 interacting protein 3	HGNC:30766	1q32.3-q41
TRD	T cell receptor delta locus	HGNC:12252	14q11.2
TRGC2	T cell receptor gamma constant 2	HGNC:12276	7p14
TSPAN5	tetraspanin 5	HGNC:17753	4q22.3

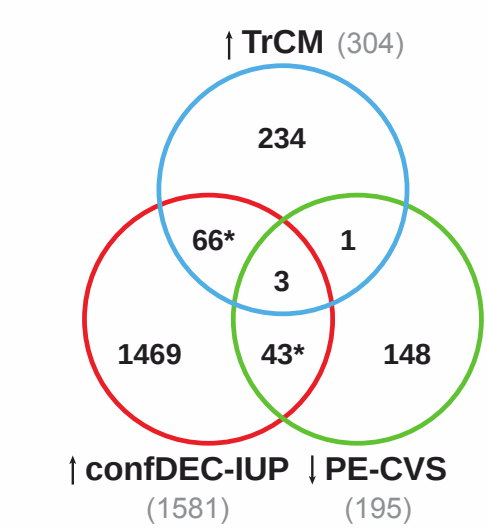

Figure S1. Lack of significant association (n=4 DEG, p=0.5) between DEG down-regulated in PE-CVS and DEG up-regulated by exposure of decidualized stromal cells in culture to trophoblast conditioned medium (TrCM).

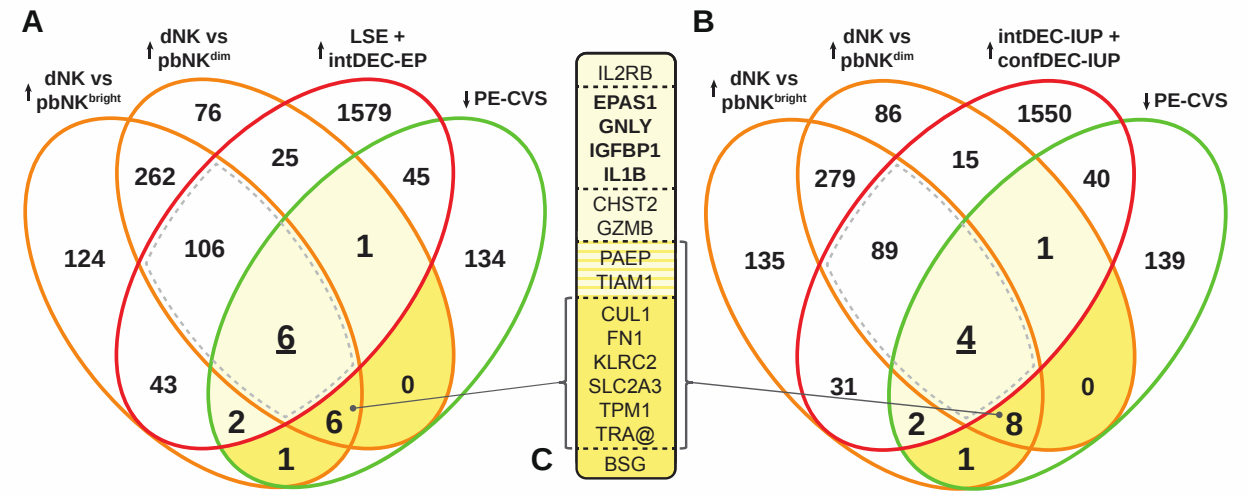

Figure S2. Clusters of genes induced in decidual Natural Killer cells are down-regulated in CVS from preeclamptic women. In **(A)** 112 DEG up-regulated in decidual Natural Killer cells (dNK, relative to peripheral blood CD56^{dim} or CD56^{bright} NK cells), late-secretory endometrium (LSE, relative to proliferative endometrium) and EP endometrium with intermediate-decidualization changes (intDEC; relative to EP endometrium without decidualization changes) are overlapping (highlighted by dotted line). In **(B)** 93 DEG up-regulated in dNK, and IUP endometrium with intermediate- and confluent-decidualization (confDEC) changes are overlapping (highlighted by dotted line). Seventy-four of the 112 and 93 DEG are in common (p<0.0001, **Table S6**). In **(A)** and **(B)** (yellow shading), 16 DEG up-regulated in dNK are down-regulated in PE-CVS (p<0.0001). The gene symbols of the 16 DEG are listed in panel **(C)**.

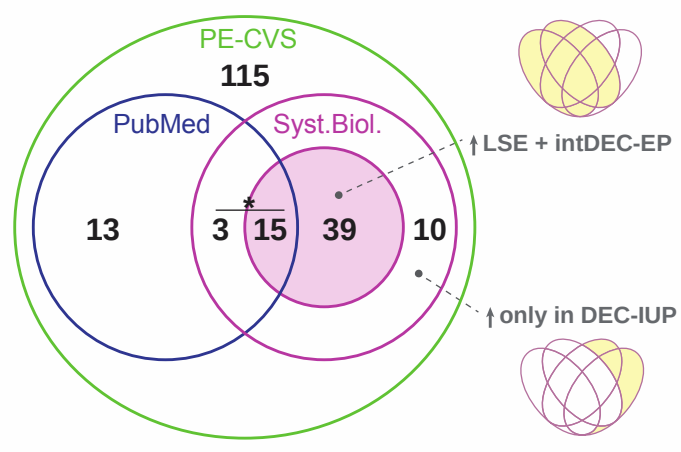

Figure S3. Systematic literature search of decidual genes. Because there is no "biological process of decidualization" available in public bioinformatic databases for pathway analysis, a systematic literature search was conducted to identify genes, which have previously been associated with decidua or the biological process of decidualization. A literature search of all 195 down-regulated DEG in PE-CVS revealed 31 previously identified to be related to decidua or decidualization (Table S2B). Of these 31 genes, 18 (3+15) genes were also identified by the systems biology approach (*p=0.001). The majority (15, p=0.03) is up-regulated in late-secretory endometrium (LSE, relative to proliferative endometrium) or in EP endometrium with intermediate-decidualized changes (intDEC, relative to EP endometrium without decidualization changes), in which extravillous trophoblast influence is absent.

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