Supplemental Material

Table S1. Participant characteristics and pregnancy outcomes for reliability study.

Variable	Repeatability study participants (n=15)
Age (years)	31.9 ± 3.5
Race(% white)	15 (100.0%)
Gravida	2 (1-3)
Parity	
0	7 (46.7%)
≥1	8 (53.3%)
Glycocheck test	
Gestational age (weeks)	33 (29-34)
Delivery	
Gestational age (weeks)	39 (35-40)
Pregnancy outcome	
Normotensive*	10 (66.7%)
Subsequent preeclampsia†	1 (6.7%)
Preeclampsia	1 (6.7%)
Superimposed preeclampsia	1 (6.7%)
Subsequent GH†	2 (13.3%)
Delivery type	
Vaginal	11 (73.3%)
C-section	4 (26.7%)
Systolic blood pressure (mmHg)	129±13
Diastolic blood pressure (mmHg)	81±10
Fetal sex (% male)	10 (66.7%)
Birthweight (g)	3354 ± 1001
Apgar (1 minute)	8 (8-9)
Apgar (5 minutes)	9 (9-9)

Values are presented as mean ± SD, median (interquartile range) or n (%). 10 women had normotensive pregnancies, one

* Two women had a history of preeclampsia, but were normotensive during the pregnancy examined.

† Patient developed the outcome after completing the reliability study

g, grams; GH, gestational hypertension.

Table S2. Trial-to-trial and day-to-day reliability for PBR among vessels in each size category.

Vessel size (μm)*	ICC for PBR		
	Day 1 (Trials 1 to 3)	Day 2 (Trials 1 to 3)	Day1 Average - Day2 Average
3	poor	poor	poor
4	poor	poor	poor
5	poor	poor	poor
6	poor	poor	poor
7	poor	poor	poor
8	poor	poor	poor
9	poor	poor	moderate
10	poor	poor	moderate
11	poor	poor	moderate
12	poor	poor	excellent
13	poor	poor	good
14	poor	poor	moderate
15	poor	poor	moderate
16	poor	poor	moderate
17	poor	poor	moderate
18	poor	poor	moderate
19	poor	poor	poor
20	poor	poor	poor
21	poor	poor	poor
22	poor	poor	poor
23	poor	poor	poor
24	poor	poor	poor
25	poor	poor	poor

ICC, intraclass correlation coefficient; PBR, perfused boundary region. *Vessel size categories, in μm, are defined by the median width of the red blood cell column. Categories for ICCs: < 0.5 poor; 0.5 to 0.75 moderate, 0.75 to 0.9 good; >0.9 excellent. Day 1 and day 2 measurements were performed approximately 24 hours apart in 15 women in the 3rdtrimester of pregnancy.

Table S3. Day-to-day reliability for PBR composite measures.

Vessel size categories (μm)*	ICC (Day 1 Average – Day 2 Average)	
	ICC	Category
5-25	0.788	Good
5-9	0.527	Moderate
10-19	0.868	Good
20-25	0.622	Moderate
10-16	0.836	Good

ICC, intraclass correlation coefficient; PBR, perfused boundary region. *Vessel size categories are defined by the median width of the RBC column (μm). Within each category, values for each day were computed as the average of three trials. Each composite measure was then calculated as the average of the PBR values for all vessels in the included size categories. n=15 women in the 3rd trimester.

Table S4. Test Conditions.

Variable	Normotensive (n = 73)	GDM (n = 21)	Late Onset PE (n = 29)	Early Onset PE (n = 14)
Caffeine intake	23 (31.5%)	4 (19.0%)	6 (20.7%)	3 (21.4%)
Fasted for at least 4 hours	18 (24.7%)	9 (42.9%)	8 (27.6%)	1 (7.1%)
Misoprostol	13 (17.8%)	5 (23.8%)	16 (55.2%)	0 (0%)
Oxytocin	10 (13.7%)	4 (19.0%)	5 (17.2%)	0 (0%)
Active labor	4 (5.6%)	1 (5.0%)	4 (13.8%)	0 (0%)
Epidural	3 (4.1%)	1 (4.8%)	4 (13.8%)	0 (0%)
Anti-hypertensive medications	0 (0%)	1 (4.8%)	7 (24.1%)	6 (42.9%)
Magnesium sulfate	0 (0%)	0 (0%)	9 (31%)	5 (35.7%)
Betamethasone	0 (0%)	0 (0%)	4 (13.8%)	4 (28.6%)
Acetaminophen	2 (2.7%)	2 (9.5%)	11 (37.9%)	8 (57.1%)
Thyroid medications	4 (5.5%)	1 (4.8%)	1 (3.4%)	1 (7.1%)
Glyburide	0 (0%)	8 (38.1%)	1 (3.4%)	1 (7.1%)

Data are presented as n (%). Caffeine intake was recorded as positive if women had consumed food, drinks or medications containing caffeine in the six hours prior to non-invasive glycocalyx measurements. Medications were recorded as positive if women had taken the medication within 24 hours of the non-invasive glycocalyx measurements. No statistical comparisons were performed.

Table S5. Effect of GDM and PE on non-invasive glycocalyx measurements.

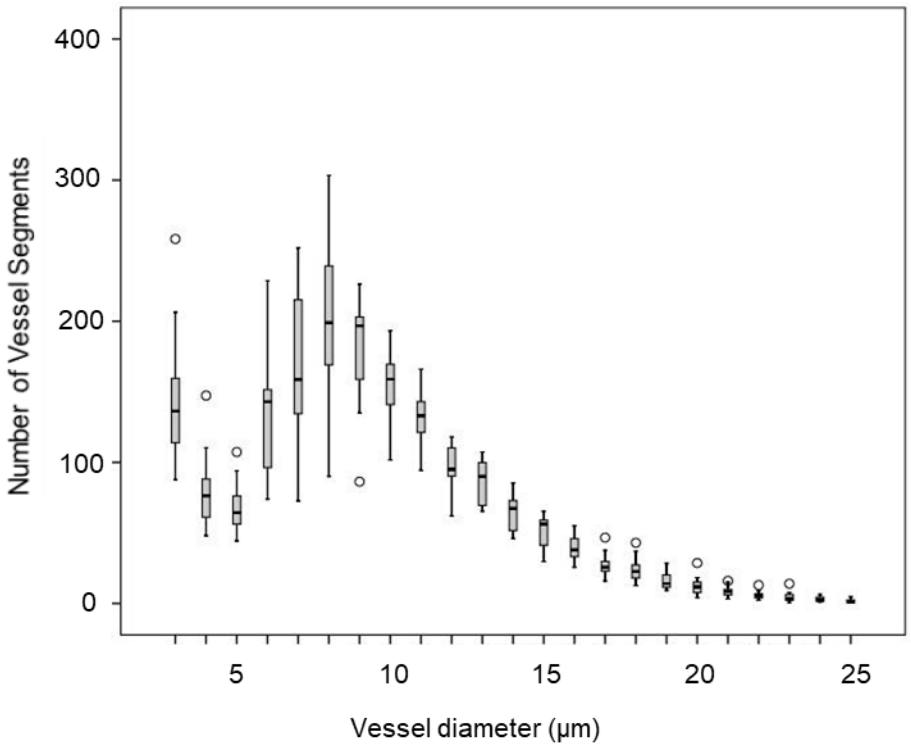
Group	PBR Mean† (95% CI)	Percentage of vessel segments filled with RBCs Mean† (95% CI)
Normotensive (n = 73)	1.989* (1.954-2.024)	0.556* (0.546-0.566)
GDM (n = 21)	1.974* (1.909-2.038)	0.567* (0.547-0.586)
Late onset PE (n = 29)	2.014* (1.959-2.069)	0.563* (0.546-0.580)
Early onset PE (n = 14)	2.124 (2.045-2.203)	0.529 (0.506-0.553)

Data were analyzed by repeated measures ANOVA (Between-subjects factor: group, Within-subjects factor: vessel size, Covariate: caffeine intake, Post-hoc test: least significant differences). Vessel size included 12 categories (5-16 μm), based on the median width of the RBC column. PBR: F = 3.600, df = 3. Percentage of vessel segments filled with RBCs: F = 2.255, df = 3.

CI, confidence interval; df, degrees of freedom; PBR, perfused boundary region; PE, preeclampsia; RBC, red blood cells.

*p<0.05 compared to women with early onset PE; †Adjusted for caffeine intake.

Figure S1. Number of vessel segments for each size category among women in the reliability study.



Average number of vessel segments for six trials (three trials per day), n = 15 women in the third trimester.