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Increased Red Cell Aggregation Does Not Reduce Uteroplacental Blood Flow in the Awake, Hemoconcentrated, Late-Pregnant Guinea Pig

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ABSTRACT. The effect of increased red blood cell aggregation on uteroplacental blood flow was studied in 11 awake, late-pregnant guinea pigs. The aggregation of the red cells was increased by administering high molecular weight dextran (HMWD) to the previously hemoconcentrated animal. The purpose of the hemoconcentration before HMWD was *1*) to use a preeclampsia model in which the hemorheology may be impaired because of the combined effect of polycythemia, an increased red cell aggregation, and an increased plasma viscosity and *2*) to potentiate the aggregation-increasing effect of HMWD. Relative to the pre-HMWD condition, arterial blood pressure and systemic vascular resistance increased by 10 and 26%, respectively. The cardiac output fraction shunted across the systemic circulation and the arterial hematocrit decreased by 30 and 4%, respectively. Neither cardiac output nor the weighted organ flows, including those to the placentas, changed in response to the rise in red cell aggregation. We conclude that an imposed increase in red cell aggregation has no appreciable effect on uteroplacental blood flow in the awake and healthy late-pregnant guinea pig. These data do not exclude the possibility that increased red blood cell aggregation potentiates the negative effects on uteroplacental blood flow, *e.g.* in pregnancy-induced hypertension or preeclampsia, where the placenta is not only marginally perfused but also frequently damaged histologically. (*Pediatr Res* 31: 91–93, 1992)

Abbreviations

CO, cardiac output ($mL \cdot min^{-1}$)
Hct, hematocrit (vol%)
HC, hemoconcentration
HMWD, high molecular weight dextran
HR, heart rate (bpm)
MAP, mean arterial pressure (mm Hg)
RBC_{agg}, red blood cell aggregation
SVR, systemic vascular resistance ($mm\ Hg \cdot min \cdot mL^{-1}$)
UBF, uteroplacental blood flow ($mL \cdot min^{-1}$)
SR, shear rate

Under normal macrocirculatory conditions, the local shear forces are always such that whole blood viscosity is of minor importance for blood flow. However, the role of whole blood viscosity upon local blood flow is likely to become more important when the shear forces exerted upon the blood in the microcirculation are not optimized by vasomotor control or when the "Fåhraeus-Lindquist effect" is not operative (1, 2). The Fåhraeus-Lindquist effect is defined as the apparent fall in local viscosity, with a decrease in arterial/arteriolar diameter from 100 to ≈15 μm (3). The small shear forces assumed to prevail in the porous (hemochorial) placental microvasculature, because of the relatively large diameter of the intervillous space, make it unlikely that the perfusion of the intervillous space benefits from the Fåhraeus-Lindquist effect (4). This implies that the negative effects on perfusion of an elevated whole blood viscosity could be larger in the hemochorial placenta than in capillary microcirculations.

In pregnancies complicated by preeclampsia and/or impaired fetal growth, UBF appears to be chronically compromised by the combined effect of inadequate spiral artery growth/dilatation (5–7) and increased whole blood viscosity (8). In these pregnancies, the increased whole blood viscosity is a result of the concerted action of elevated Hct (9–12), decreased red blood cell deformability (10–14), increased RBC_{agg} (10, 15, 16), and, to a lesser degree, increased plasma viscosity (8, 11, 17, 18).

In recent studies, Hct and red cell deformability were found to contribute little to the variation in UBF (19, 20). Inasmuch as Hct and red cell deformability affect primarily the high-shear viscosity, these findings were not surprising because the SR in the intervillous space is likely to be extremely low (4, 21). This fact raises the possibility that particularly a rise in RBC_{agg} could have a more important (negative) effect on UBF than factors that primarily affect high-shear viscosity, such as Hct.

The objective of the present study was to evaluate whether an imposed increase in RBC_{agg} reduces UBF in the awake, healthy, late-pregnant guinea pig. The concomitant response of other maternal hemodynamic variables was used to evaluate the efficacy of the methodology that was used to raise low-shear viscosity. To this end, the CO, HR, MAP, SVR, and regional blood flows were determined in awake, previously hemoconcentrated, late-pregnant guinea pigs before and 30 min after increasing the RBC_{agg} 5-fold by a bolus injection of HMWD. The HC before HMWD was performed *1*) to use a preeclampsia model in which the hemorheology may be impaired because of the combined effect of polycythemia, an increased red cell aggregation, and an increased plasma viscosity; *2*) because the effect of fibrinogen (RBC_{agg}) on low-shear viscosity (placenta) is potentiated by increasing the Hct (22); and *3*) because in guinea pig elevation of the Hct potentiates the increasing effect of HMWD on RBC_{agg}. This was demonstrated *in vitro* before the present study.

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MATERIALS AND METHODS

The study was performed in 11 pregnant albino guinea pigs, with known postconceptional pregnancy length, supplied by a commercial breeder (Charles River, Sulzfeld, Germany). Using aseptic techniques (23), polyethylene catheters (outer diameter/inner diameter = 0.96/0.58 mm) were inserted into the left ventricle and abdominal aorta around the 48th d of gestation. After complete recovery (± 6 d) and as soon as steady state was established (stable MAP and HR for at least 20 min), the experiments were started. The first experiment (measurement 1: baseline) involved the measurement of CO and organ flows with 15-μm labeled microspheres (^{95}Nb, ^{103}Ru, ^{113}Sn, or ^{141}Ce) (24), the measurement of arterial Hct (in duplicate, microcapillary method), and the quantitation of the RBC_{agg} (MA1, Myrenne Gmbh, Roetgen, Germany). The aggregometer has two options for measuring the extent of aggregation: at stasis (SR_0) and at a low SR (SR_{10}). For both options the blood is rotated at an SR of 600 s^{-1} for 10 s, whereafter the aggregation is determined during 15 s, either at zero SR (SR_0) or at an SR of 10 s^{-1} (SR_{10}). The quantity of transmitted infrared light, measured with photosensors, is proportional to the degree of aggregation and is digitally displayed in arbitrary units. After reference sampling (sample rate 0.65 mL·min^{-1}, from 10 s before the intraventricular bolus injection of the microspheres until 30 s after its completion), the animals were gradually hemoconcentrated in six steps by consecutive isovolemic exchange transfusions. Each transfusion involved the substitution of 5 mL of blood of the recipient animal by an equal amount of packed red cells from a nonpregnant donor guinea pig. Twenty h after the administration of packed cells, MAP, HR, CO, organ flows, Hct, and RBC_{agg} were remeasured (measurement 2: preaggregation). Subsequently, 400 mg of HMWD (500 000 g·mol^{-1}) dissolved in 2 mL of saline were injected into the left ventricle. It can be estimated (25) that this quantity will result in an HMWD plasma concentration of 1% wt/vol. Thirty min after the bolus injection, all variables were remeasured (measurement 3: postaggregation).

Finally, the animals were killed with an overdose of pentobarbital. All organs were dissected, weighed, and placed in counting vials. The accumulated radioactivity was determined in a sodium crystal scintillation counter (Packard, Delft, The Netherlands). The microsphere data obtained in baseline, preaggregation, and postaggregation were analyzed as described previously (26). Data from different experimental conditions were compared with each other using the Wilcoxon Rank Sign Test. A probability of less than 5% (two-sided) was considered significant. The results are expressed as means ± SD throughout the text.

RESULTS

The average maternal weight at the day of the first experiment was 721 ± 108 g, including 137 ± 64 g fetal mass. The litter size ranged from one to four fetuses. The gestational age ranged from 52 to 63 d.

The hemodynamic changes in response to the HC procedure can be deduced by comparing the results listed in column 2 with those in column 1 of Table 1. CO and thus also organ blood flows could only be determined succesfully in eight out of 11 animals because of failure to obtain a reliable reference sample in three animals. HC had no effect on CO, HR, systemic shunting of the microspheres (*i.e.* lung fraction), or any weight-specific organ flows except for renal flow, which increased consistently. In addition to the obvious rise in Hct, the MAP had also increased markedly. The increase in MAP was, however, not paralleled by an increase in SVR.

The cardiovascular effects associated with the rise in RBC_{agg} emerge when the results listed in column 3 of Table 1 (postaggregation) are compared with those in column 2 (preaggregation). The administration of HMWD raised RBC_{agg} 5-fold in both SR_0 and SR_{10}. An increase in both MAP and SVR was paralleled by a decrease in both arterial Hct and the CO fraction distributed to shunts in the systemic circulation. Neither CO nor any weight-specific organ flow, including UBF, changed in response to the increase in RBC_{agg}.

Table 1. *RBC_{agg} at SR_0 and SR_{10} (arbitrary units), CO (mL·min^{-1}), HR (bpm), MAP (mm Hg), SVR (mm Hg·min·mL^{-1}), Hct (vol%), and weighted organ flows (mL·min^{-1}·100 g^{-1}) (n = 8) before (baseline), 20 h after packed cell transfusion (preaggregation), and 30 min after increasing RBC_{agg} (postaggregation)*

	Baseline	Preaggregation	Postaggregation
RBC_{agg} at SR_0	0.2 ± 0.6	0.9 ± 1.3	5.1 ± 2.3*
RBC_{agg} at SR_{10}	2.6 ± 0.7	2.2 ± 1.5	12.5 ± 1.6*
CO	188 ± 49	219 ± 79	191 ± 55
HR	256 ± 8	254 ± 25	272 ± 43
MAP	52 ± 9	60 ± 11†	66 ± 12*
SVR	6.1 ± 1.9	6.1 ± 2.5	7.7 ± 2.6*
Hct	37 ± 3	56 ± 4†	54 ± 4*
Systemic shunting	12 ± 3	10 ± 3	7 ± 3*
Weighted organ flows			
Myocardium	324 ± 66	326 ± 96	298 ± 73
Brain	94 ± 28	72 ± 33	72 ± 22
Kidneys	326 ± 111	480 ± 144†	479 ± 64
Gastrointestinal tract	116 ± 44	122 ± 47	104 ± 24
Skin	13 ± 5	13 ± 4	12 ± 4
Carcass	15 ± 6	15 ± 4	15 ± 5
Myometrium	11 ± 6	19 ± 10	18 ± 7
Placentas	124 ± 59	144 ± 82	139 ± 43

* $p < 0.05$ postaggregation different from preaggregation.
† $p < 0.05$ preaggregation different from baseline.

DISCUSSION

To study the effects of RBC_{agg} on the systemic circulation, we believed it was necessary to adopt a procedure that would have a clear impact on red cell aggregation. By combining the aggregation-increasing effects associated with a high Hct with those induced by administering HMWD, we managed to increase the RBC_{agg} 5-fold, a rise expected to be adequate to discern specific hemodynamic effects caused by an elevated RBC_{agg}. Before the RBC_{agg} was raised, the Hct was increased by isovolemic HC to a much higher level than had been reached in a previous study from our laboratory [56 vol% *versus* 38 vol%; (19)]. An elevated Hct may potentiate HMWD-induced RBC_{agg} particularly in the guinea pig, where basal RBC_{agg} (Table 1) is extremely low as compared with man (RBC_{agg} at SR_{10} ± 12, personal observation). The marked rise in MAP that was observed after the more intensive HC in the present study seems to indicate that the effects of increased high-shear viscosity on the circulation become manifest only when the Hct is raised above a certain threshold. Theoretically, it seems unlikely that the induced "hypertension" will interfere with the subsequently induced rise in RBC_{agg} inasmuch as changes in high-shear viscosity have their largest impact on the arterial side of the circulation, where shear rates are highest (small arteries and arterioles), as opposed to changes in RBC_{agg} which influence primarily the viscosity of blood in vessels with a low SR (venules, small veins, intervillous space). The lack of increase in SVR in response to the HC procedure was the result of the increasing trend in CO with the rise in MAP. In contrast, a rise in low-shear viscosity primarily increases venous resistance, imposing a negative effect on venous return (27). The increase in renal flow after the HC was also observed in our previous study (19). The underlying mechanism appears to be maintenance of renal plasma flow (28). Neither the SVR nor the CO, HR, or regional flows changed in response to HC, supporting the hypothesis that the postaggregation condition represents a selective effect of the imposed increase in RBC_{agg}.

The observed changes in response to the rise in RBC_{agg} were an increase in both MAP and SVR by 10 and 26%, respectively,

together with a fall in systemic shunting and arterial Hct by 30 and 4%, respectively. CO and all organ flows did not change consistently. It is not clear whether the increase in SVR after HMWD infusion is related to an increase in plasma viscosity (28) or can be attributed to increased venous viscosity associated with enhanced formation of RBC aggregates. The increasing trend in CO after HC, followed by a decreasing trend after HMWD, suggests that HMWD affects primarily the venous compartment, *i.e.* venous return. Assuming that the rise in RBC_{agg} caused an increase in venous resistance, the modest increase in MAP could be a compensation serving the purpose of maintaining the perfusion pressure across the systemic microcirculation. The marked decline in fractional systemic shunting could represent an additional compensation for the apparent fall in intraarterial volume (30). That is to say, an induced rise in venous resistance may lead to pooling of blood in the venous compartment, thus interference with venous return. The associated negative impact on CO can be expected to activate the baroreceptor and sympathetic nervous system, resulting in a decrease in the CO fraction shunted across the microcirculation. The arterial Hct may be reduced in the postaggregation state partly as a result of reference sampling. However, theoretically, increased formation of aggregates in the venous compartment will increase the venous Hct at the cost of the arterial Hct.

Interestingly, UBF was not compromised by the marked rise in RBC_{agg}. The lack of change in UBF in response to both moderate (19) and severe isovolemic HC (present study) supports the concept that high-shear viscosity has a negligible effect on the perfusion of the intervillous space. Meanwhile, the results of the present study support the concept that a rise in low-shear viscosity also fails to have a measurable effect on the perfusion of the hemochorial placenta in this particular model. One could speculate that such an effect is prevented by the action of some form of vasomotion or autoregulation operative in the supplying arteries.

From these data we conclude that an imposed increase in red cell aggregation has no appreciable effect on UBF in the awake and healthy late-pregnant guinea pig. These data do not exclude the possibility that RBC_{agg} potentiates the negative effects on UBF, *e.g.* in pregnancy-induced hypertension or preeclampsia, where the placenta is not only marginally perfused but also frequently damaged histologically.

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