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PHYSIOLOGICAL MODIFICATIONS OF BLOOD VISCOSITY AND RED BLOOD CELL AGGREGATION DURING LABOR AND DELIVERY

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ABSTRACT

Since most stresses are known to modify blood rheology, we studied hemorheological parameters during labor and delivery, which are highly stressful physiological events. In 80 pregnant women we measured blood viscosity, plasma viscosity and red blood cell (RBC) aggregation during labor (before and after 4 cm dilatation), during delivery, and during delivery of the placenta. Blood viscosity at both native and corrected hematocrit increases ($p<0.001$) with a peak during delivery followed by a rapid normalization during delivery of the placenta ($p<0.01$). Hematocrit and plasma viscosity do not change during labor and delivery, but decrease during delivery of the placenta. RBC aggregation (physiologically increased during late pregnancy) acutely returns to normal during delivery ($p<0.01$). The transient increase in viscosity during delivery is explained by an increase in RBC rigidity as measured by Dintenfass' 'Tk' ($p<0.01$) and a similar (nonsignificant) tendency to decrease RBC filterability measured by the hemorheometre. Thus, delivery is associated with a transient hyperviscosity syndrome which is mainly due to a decrease in RBC flexibility.

Key words: labor, delivery, pregnancy, rheology, erythrocyte deformability, erythrocyte aggregation.

INTRODUCTION

Physiological modifications of blood rheology during normal pregnancy are well documented (1-6). The fundamental deviation of the maternal blood from the flow behavior found in non-pregnant females was clearly described by R. Fahraeus (7): there is a high tendency to erythrocyte aggregation, despite an overall low blood viscosity and normal plasma viscosity. This hyperaggregation is increased in preeclampsia and may be involved in the vascular risk of this disease. By contrast, little has been reported on the modifications of blood rheology during labor. Labor is a very stressful situation for both mother and fetus, and is accompanied by an increase in many stress hormones like ACTH (8) and beta-endorphin (9). Since several kinds of stresses are known to modify blood rheology, it was interesting to determine whether labor was also associated with such modifications.

METHODS

Patients

First study. We have been studying 80 normal pregnant women. Women were 26.9 $\pm$ 3.2 yr old (extreme values: 18-41 yr) and mensural age at the onset of labor was 39.3 $\pm$ 1.58 wks (extreme values: 35-42.5 wks). All patients underwent 7 blood drawings: 2 before 4 cm dilatation, 2 after 4 cm dilatation, one at the moment the newborn was expelled, 2 during delivery of the placenta. Second study. 19 women (age 20-35 yr; mean age 28 yr) underwent a blood sampling during a single uterine contraction. Uterine contractions being monitored by manometry, a sample was drawn at rest, another when contraction reached its maximum, and the third when uterine muscular activity had stopped. In 4 cases this was done after 4 cm dilatation, in 15 cases this was done before 4 cm dilatation. Total duration of labor ranged between 1 and 10 hr.

Hemorheological measurements.

8 ml of peripheral blood from antecubital veins was collected by vacutainer into potassium EDTA. Measurements were performed within two hours. Hematocrit (packed cell volume) was measured by microcentrifugation. Erythrocyte rigidity was measured by filtration of erythrocytes resuspended at 8% hematocrit in Tris-Albumin buffer, with the Hemorheometre MK-1 (from IMH, 2, allee du Jardin de la Cure, 95470 Saint Witz, France). This apparatus measures the the initial flow rate of a suspension of erythrocytes (10) through 5 μm Nuclepore sieves. Results were expressed as a relative viscosity of filtration (μfr) and corrected by hematocrit:

$\mu fr = (ts/tb)/h$

where ts is the time of passage of the suspension of erythrocytes at 8% hematocrit, tb the time of passage of the buffer alone, and h the packed cell volume (%). Blood viscosity and plasma viscosity were measured at very high shear rate (2000 s^{-1}) with the MT90 falling ball viscometer (Medica-test, 37 rue de l'Ermitage F-86280 Saint Benoit) (11). The coefficient of variation of this method ranges between 0.6 and 0.8 % (10 repetitive measurement of the same sample). The results of viscometric measurements were expressed as apparent viscosity at native hematocrit μb, viscosity for corrected hematocrit 45% μ_{45}, and erythrocyte rigidity index 'Tk'. Correction of blood viscosity for hematocrit was calculated according to Quemada's equation (12):

$$\mu b = \mu pl\ (1\text{-}1/2\ k.h)^{-2}$$

where μpl is plasma viscosity, h hematocrit, and k a structural parameter of blood viscosity which depends at high shear rate on erythrocyte flexibility. A viscometric index 'Tk' of erythrocyte rigidity (as reflected by shear-induced erythrocyte elongation) was calculated from this viscometric measurement according to Dintenfass (13, 14). 'Tk' is given by the following equation:

$$Tk = (\mu r^{0.4}\text{-}1)/(\mu r^{0.4}.\ h)$$

where μr is relative blood viscosity (i.e. $\mu b/\mu pl$).

erythrocyte aggregation was measured using a transparent cone-plate shearing instrument (Model MA-1 Aggregometer, Myrenne GmbH, Roetgen, FRG) which employs the light transmission method of Schmid-Schönbein et al (15). This technique is based upon the increase of light transmission through a erythrocyte suspension which occurs when individual erythrocytes aggregate into rouleaux or rouleaux-rouleaux complexes. Increased light transmission (through plasma gaps between the aggregates) is proportional to erythrocyte aggregation. The sample is placed between a transparent cone and a transparent plate and sheared, at 600 s^{-1}, to disperse all pre-existing cell aggregates. The shearing is then instantly stopped, and the light transmission increases at a rate proportional to the rate of erythrocyte aggregate formation during stasis (M index). Another measurement is also performed after similar shearing at 600 s^{-1} during 10 s: shearing is reduced to 3 s^{-1} and light tansmission (low shear conditions) is measured in this situation which stimulates aggregation (M1 index).

Statistics.

Results are presented as mean $\pm$ the SE of the mean. A value of $p<0.05$ was considered as significant. Correlations were performed using the method of least squares. Comparisons were made with Mann-Whitney and Wilcoxon tests. Significance was defined as $p<0.05$. The choice of nonparametric tests was done in order to adhere the guidelines of J. Stuart (16) and the ICSH expert

pannel for blood rheology (17), since hemorheological parameters usually appear to exhibit a nonnormal distribution.

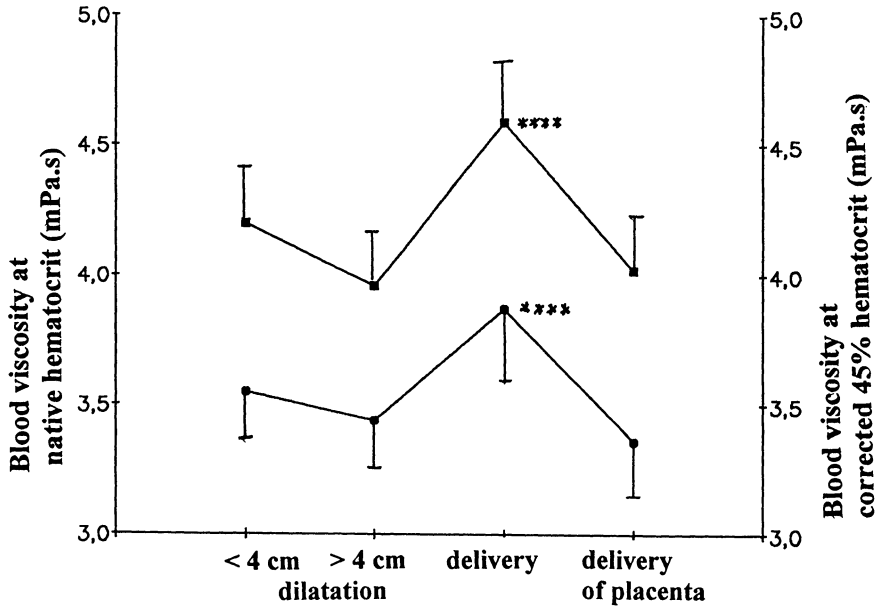

FIG. 1.

*Modifications of blood viscosity (mPa.s) measured at 1000 s-1 . Lower curve: viscosity at native hematocrit. Upper curve: viscosity at corrected hematocrit 45%; Comparison with the preceding value **** p< 0.001.*

RESULTS

During delivery, drammatic transient modifications of hemorheological parameters were observed. Although individual changes seemed to be erratic, statistical analysis detected the following events.

Whole blood viscosity at native hematocrit as well as blood viscosity for hematocrit corrected at 45% (see fig. 1) increased during delivery ($p<0.001$ and $p<0.0001$) and rapidly returned to baseline values during delivery of the placenta ($p<0.0001$ and $p<0.01$). A moderate reduction in plasma viscosity and hematocrit occurs during delivery of the placenta ($p<0.05$) while those parameters remain constant during labor and delivery (fig.2). erythrocyte aggregation remains unchanged during labor and significantly decreases ($p<0.01$) during delivery and delivery of the placenta (fig.3). The most prominent finding was an increase in 'Tk' erythrocyte rigidity index ($p<0.01$) during delivery, which almost reversed to normal afterwards ($p<0.02$), during

delivery of the placenta. A similar nonsignificant tendency was found for erythrocyte filterability measured by the hemorheometre (fig.4).

Linear correlations were found between gestational age at the onset of labor and the following factors: baseline erythrocyte rigidity measured by 'Tk' ($r=0.620$) and filterability ($r=0.348$) and baseline erythrocyte aggregability (M index: $r=0.403$). Fibrinogen was also correlated with baseline 'M' ($r=0.311$) and baseline 'Tk' ($r=0.547$). Baseline 'Tk' and 'M' were correlated ($r=0.263$). At all the times of the study, aggregation indices 'M' and 'M1' were highly correlated (labor $r=0.856$; delivery $r=0.898$; delivery of placenta $r=0.840$). Finally, the variations of 'Tk' and 'M' during delivery were negatively correlated ($r=-0.314$ $p<0.02$).

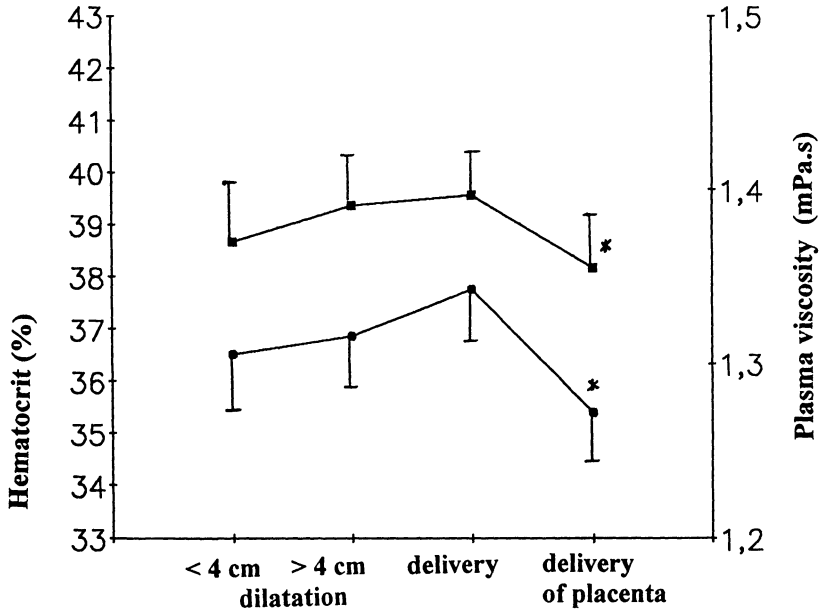

FIG. 2.

*Modifications of plasma viscosity (mPa.s, upper curve) and hematocrit (%, lower curve) during labor. Comparison with the preceding value * $p< 0.05$.*

The study of 19 uterine contractions demonstrated no significant change in either hematocrit or plasma viscosity. As shown on fig. 5, during contraction itself, erythrocyte rigidity indices measured by viscometry were increased ($p<0.05$). Erythrocyte aggregation was unchanged during contraction but decreased after ($p<0.05$). Some correlations were found among parameters studied in this experiment. Changes in aggregation indices exhibited negative correlations with changes in erythrocyte rigidity: variations of 'M' were negatively correlated to those of 'Tk' ($r=-0.581$ $p<0.01$), 'k' ($r=-0.632$

$p<0.01$, see fig. 6), and those of relative viscosity at corrected hematocrit 45% ($r=-0.585$ $p<0.01$). Variations of 'M1' were negatively correlated to those of 'Tk' ($r=-0.528$ $p<0.05$), 'k' ($r=-0.568$ $p<0.01$), and those of relative viscosity at corrected hematocrit 45% ($r=-0.561$ $p<0.01$). Duration and intensity of the contraction were not correlated with any parameter measured in this protocol, but there was a nonsignificant tendency to a correlation with changes in 'M' ($r=0.432$) and changes in 'M1' ($r=0.425$).

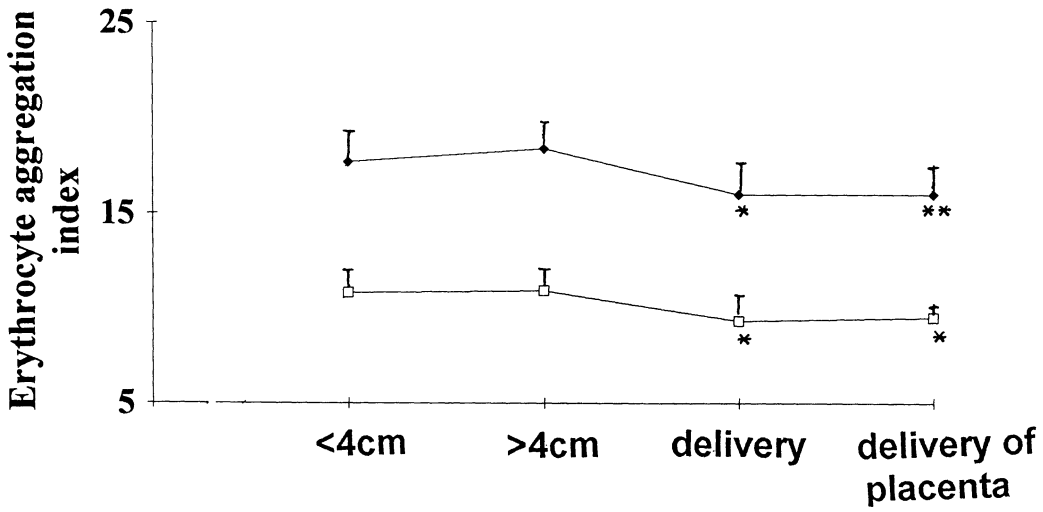

FIG.3.

*Modifications of erythrocyte aggregation during labor. Lower line: 'M' index; upper line: 'M1' index. Comparison with values during dilatation *p< 0.01; ** p< 0.001*

DISCUSSION

Blood rheology, as a factor regulating blood flow, is believed to be an important determinant of the amount of oxygen and nutrients transferred across the placenta. The rheology of pregnancy has been thoroughly studied by several investigators, as well as the peculiar hemorheologic pattern found in newborns. By contrast, we were not aware of sequential studies of the modifications of blood rheology during labor, although this period is known as a 'critical' one.

To our knowledge, the physiological changes in blood rheology during labor and delivery have not been extensively studied. In the paper of Ozanne (4), the last sampling at the end of a careful longitudinal study of pregnancy suggests such data, but the different periods of labor and delivery are not studied.

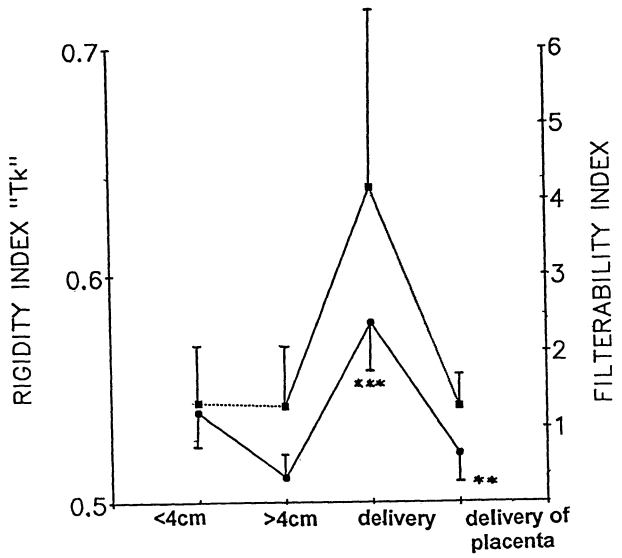

FIG.4.

Changes in erythrocyte rigidity indices during labor and delivery. Lower curve: 'Tk'. Upper curve: filterability with the hemorheometre. Comparison with the preceding value: ** p< 0.02; *** p< 0.01.

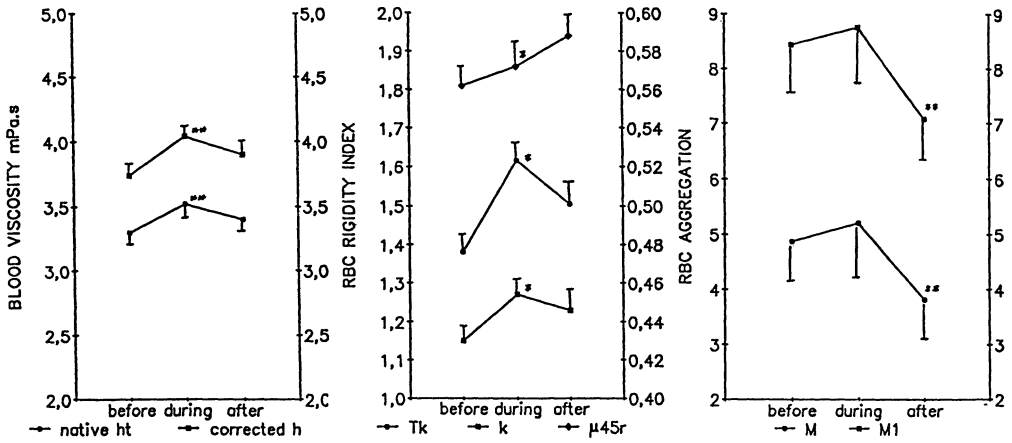

FIG.5.

Study of a single uterine contraction in 19 women. Graph on the left: blood viscosity (upper curve: corrected at Ht 45%; lower curve: at native hematocrit). Graph in the middle: erythrocyte rigidity measured by relative viscosity at 45% hematocrit (upper), "Tk' (intermediate curve, scale on the left), and 'k' (lower curve). Graph on the right: changes in erythrocyte aggregation (upper line: 'M1'; lower line 'M'). * p< 0.05; ** p< 0.02

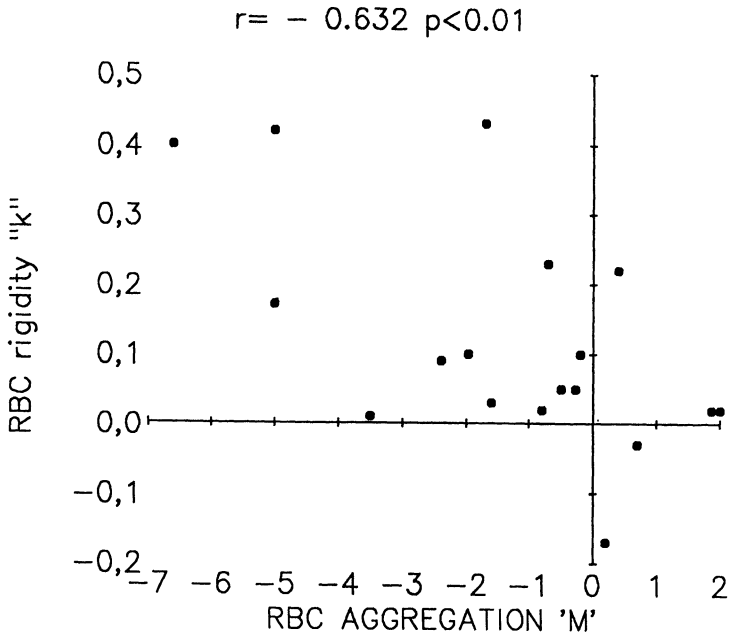

FIG.6.

Correlation between the increase in erythrocyte rigidity index 'k' during uterine contraction and the decrease in erythrocyte aggregability 'M' after this contraction.

In this study, we measured blood viscosity at very high shear rate, a condition which allows to study the behavior of completely disaggregated erythrocytes (13). Values ranging between 1000 and 2000 s^{-1} mimic the high shear rates which are observed at the distal arteriolar level (18) and allow to characterize modifications of erythrocyte rigidity induced by various procedures (19). This study demonstrates that blood viscosity exhibits during delivery an acute, transient increase which cannot be explained by changes in either hematocrit or plasma viscosity. Clearly, it seems to result from a rise in erythrocyte rigidity, which is highly significant when measured by 'Tk' (yet nonsignificant when measured by filtration). The mechanism for such a short term 'rigidification' of erythrocytes is unclear. We hypothesize that hypoxia and lactic acidosis may be involved, but other metabolic and hormonal processes during labor can play also a role. For P. Buchan (personal communication), oxytocin, which *in vitro* decreases erythrocyte deformability, can explain to some extent this

phenomenon. Another hypothesis can be suggested when comparing our findings to the results of Kaibara and coworkers (20). These authors observe that erythrocyte filterability during pregnancy and puerperium is closely related to intracellular ATP levels. Although these authors do not thoroughly describe the period of delivery, there seems to be a decrease in intracellular level of ATP as well as a parallel reduction in erythrocyte filterability between the last point measured during pregnancy and the first after delivery. If such a modification occurs rapidly during delivery, it may be involved in the rigidification of erythrocytes which is evidenciated in our study. However, the precise mechanism of erythrocyte rigidification during delivery remains unclear. It is interesting to notice that indices of erythrocyte rigidity provided by viscometric measurements and by filterability do not give exactly the same results, although they exhibit a similar trend. Discrepancy between these two kinds of measurements is not uncommon (19) because they do not exactly evaluate the same "deformation" of erythrocytes. Viscometry measures shear-induced elongation of the cells as it can occur in large vessels, while filtrometry measures the folding of erythrocytes which pass through narrow channels, a situation which mimics to some extent what happens in capillaries (14). These two deformation processes are not always similarly modified: for instance, it is well known that filterability measurements are very sensitive to changes in erythrocyte surface/volume ratio (16-17). Actually, we observe during delivery some drammatic changes in filterability indices, but with a very large scattering which makes the results statistically nonsignificant. By contrast, modifications of "Tk" are more moderate, but also more reproducible and are statistically very significant.

A second fact which is observed in this study is the reduction in hematocrit and plasma viscosity during delivery of the placenta. The former event is probably explained by hemorrage. The latter may result at least in part from sodium chloride perfusions which have been performed in most women and can not be standardized in this study for obvious ethical reasons. Since hematocrit and plasma viscosity exhibit a parallel behavior, we hypothesize that their modifications result from a combination of hemorrage and dilution by the perfusions.

Erythrocyte aggregation exhibits also significant changes which are different from those of whole blood viscosity, erythrocyte rigidity, plasma viscosity and hematocrit. It remains unchanged during labor and suddenly decreases during delivery (before any changes in hematocrit or plasma viscosity). Therefore, the factors assumed above (hemorrage and perfusions) are not likely to explain this modification, which can be described as a return to nonpregnant conditions, after the well known hyperaggregation of late pregnancy. The mechanism of this latter hemorheological modification is probably related to the well known defibrination which occurs during parturition (21). Since plasma fibrinogen is a major determinant of erythrocyte aggregation, a reduction in fibrinogen may explain this reduction in aggregation.

When looking at a single uterine contraction, with the same hemorheological techniques, we observe an increase in erythrocyte rigidity during the contraction, and a decrease in erythrocyte aggregation after the contraction. It could be suggested that these events result from the stressful effects of the contraction and from fibrinogen consumption before it, as indicated above.

However, in a more recent study using another technique for measuring erythrocyte aggregation, namely laser backscattering, we observed a more complex picture (22). There was an improvement in erythrocyte disaggregability, but also an increase in erythrocyte aggregation index and a reduction in erythrocyte aggregation time. Therefore, the influence of labor on erythrocyte aggregation appears to be a rather complex process, which requires to be further studied. Beside the return to nonpregnant hemorheological conditions, there are some changes which probably reflect the numerous modifications induced by this strongly stressful physiological event.

In conclusion, a short term rise in blood viscosity, resulting from increased erythrocyte rigidity is clearly found during delivery. It probably results from hormonal and metabolic modifications induced by this stressful event. Its mechanism remains to be clarified. Since during this phase, fetal cardiac rhythm is frequently modified, a possible pathophysiological relevance of hemorheologic modifications during labor remains to be investigated. Besides, other hemorheologic modifications (namely a decrease in both erythrocyte aggregation index as assessed by light transmission, and plasma viscosity) are also noticed and may result from the reduction in plasma fibrinogen which has been reported to occur during delivery (21).

REFERENCES

1. HUISMAN A, AARNOUDSE JG, KRANS M, HUISJES HJ, FIDLER V, ZIJLSTRA WG. Red cell aggregation during normal pregnancy. Br J Haematol 68, 121-124, 1988

2. OZANNE P, LINDERKAMP O, MILLER FC, MEISELMAN HJ. Erythrocyte aggregation during normal pregnancy. Am J Obstet Gynecol 147, 576-583, 1983

3. GAEHTGENS P, SCHICKENDANTZ S. Rheologic properties of maternal and neonatal blood. Bibl Anat 13, 107-108, 1975

4. FOLEY ME, ISHERWOOD DM, McNICOL GP. Viscosity, haematocrit, fibrinogen and plasma proteins in maternal and cord blood. Br J Obstet Gynaecol 85, 500-504, 1978.

5. FOLEY ME, COLLINS R, McDONALD D. Whole blood viscosity in umbilical cord blood, adult pregnant and non-pregnant blood: the influence of plasma factors. Clin Hemorheol 3, 285, 1983.

6. BUCHAN PC. Evaluation and modification of whole blood filtration in the measurement of erythrocyte deformability in pregnancy and the newborn. J Hematol 45, 97-105, 1980.

7. FAHRAEUS R. Eclampsia - a disease of checked microcirculation. Acta Obstet Gynaec Scand 41, 101, 1962

8. GENNAZZANI AR, FRAIOLI F, HURLIMANN J, FIORETTI P, FELBER JP. Immunoreactive ACTH and cortisol plasma during pregnancy: detection and partial purification of corticotrophin-like placental hormone: human chorionic corticotropin. Clin Endocrinol 4, 1-14, 1974

9. FACCHINETTI F, CENTINI G, PARRINI D, PETRAGLIA F, D'ANTONA N, COSMI EV, GENNAZZANI AR. Opioid plasma levels during labour. Gynecol Obstet Invest 13, 155-163, 1982

10 HANSS M. Erythrocyte filterability measurement by the initial flow rate method. Biorheology 20, 199-211, 1983

11. DOFFIN J, PERRAULT R, GARNAUD G. Blood viscosity measurements in both extensional and shear flow by a falling ball viscometer. Biorheology 21, (suppl.1), 89-93,1984

12. QUEMADA D. Rheology of concentrated disperse systems. II. A model of non newtonian shear viscosity in steady flows. Rheol Acta 17, 632,642, 1978.

13. DINTENFASS L Red cell rigidity, "Tk", and filtration. Clin Hemorheol 5, 241-244,1985

14 DINTENFASS L. Blood viscosity, Hyperviscosity & Hyperviscosaemia. MTP press, 1985.

15 SCHMID-SCHONBEIN H, VOLGER E, KLOSE HJ Microrheology and light transmission of blood III. The velocity of red cell aggregate formation. Pflügers Arch 254, 299-317, 1975

16. STUART J. Design principles for clinical and laboratory studies of erythrocyte deformability. Clin. Hemorheol 5, 159-169, 1985.

17. ICSH expert panel on blood rheology. Guidelines for measurement of blood viscosity and erythrocyte deformability. Clin Hemorheol 6, 439-453, 1986.

18. CHIEN S, SUNG LA. Physicochemical basis and clinical implications of red cell aggregation. Clin Hemorheol 7, 71-91, 1987

19. FONS C, BRUN JF, SUPPARO I, MALLART C, BARDET L, ORSETTI A. Evaluation of blood viscosity at high shear rate with a falling ball viscometer. Clin Hemorheol 13, 651-659,1993

20. KAIBARA M, MARUMOTO Y, TANIGUCHI I , KIGASAWA K, KOBAYASHI T. Erythrocyte filterability and adenosine triphosphate levels in normal pregnancy and puerperium. Int J Microcirc 4, 55-62, 1985

21. KLEINER GJ, MERSKEY C, JOHNSON AJ, MARKUS WB. Defibrination in normal and abnormal parturition. Brit J Haematol 19, 159-165, 1970

22. BRUN JF, BOULOT P, EL BOUHMADI A, ROUSSEAU O, LAFFARGUE F, VIALA JL, ORSETTI A. Modifications of erythrocyte aggregation during labor and delivery. Clin Hemorheol 13, 293,1993