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Blood Viscosity and its Relationship to Iron Deficiency, Symptoms, and Exercise Capacity in Adults With Cyanotic Congenital Heart Disease

Craig S. Broberg, MD,* Bridget E. Bax, PHD,‖ Darlington O. Okonko, BSC, MRCP,† Michael W. Rampling, MD,¶ Stephanie Bayne, BS,‡ Carl Harries, BS,* Simon J. Davidson, BS,§ Anselm Uebing, MD,* Arif Anis Khan, MD,* Swee Thein, MD,# J. Simon R. Gibbs, MD,** John Burman, MD,‡ Michael A. Gatzoulis, MD, PHD*

London, England

OBJECTIVES This study sought to determine the relationship between blood viscosity and iron deficiency and their impact on symptoms and exercise function in adults with cyanotic congenital heart disease.

BACKGROUND Iron deficiency is believed to raise whole blood viscosity in cyanotic congenital heart disease, although available data are inconsistent.

METHODS Thirty-nine cyanotic adults were prospectively assessed for iron deficiency (transferrin saturation ≤5%), hyperviscosity symptoms, and exercise capacity. Same-day measurement of whole blood viscosity and hematocrit (Hct) adjusted viscosity (cells resuspended in autologous plasma to Hct of 45%) was performed at shear rates ranging from 0.277 s^{-1} to 128.5 s^{-1}.

RESULTS Viscosity did not differ between patients with iron deficiency (n = 14) and those without (n = 25). Whole blood viscosity correlated with Hct ($r = 0.63$, $p < 0.001$ at low shear and $r = 0.84$, $p < 0.001$ at high shear) but not with red blood cell size or iron indices. Hyperviscosity symptoms were independent of iron indices but directly correlated with increased Hct-adjusted viscosity ($r = 0.41$, $p = 0.01$). Exercise capacity did not differ in iron-deficient patients. However, peak oxygen consumption was higher in those with Hct ≥ 65% (12.6 ± 3.4 ml/kg/m^2 vs. 9.8 ± 2.6 ml/kg/m^2, mean ± SD, $p = 0.036$) despite higher whole blood viscosity in these same individuals ($p < 0.01$ for all shear rates).

CONCLUSIONS Iron deficiency is common in cyanotic adults but does not alter viscosity. Hyperviscosity symptoms are associated with a higher Hct-adjusted viscosity independent of cell size or iron stores. Higher Hct is associated with better exercise capacity. Further work to understand the origin of hyperviscosity symptoms is warranted. (J Am Coll Cardiol 2006;48:356–65)

Increased whole blood viscosity in adults with cyanotic congenital heart disease is an unavoidable result of secondary erythropoiesis. Little is known about the role of nonerythrocytotic factors in the genesis of hyperviscosity. Improved understanding of the determinants of blood viscosity in cyanotic congenital heart disease is of substantial clinical importance.

Data showing that iron deficiency and microcytosis increase whole blood viscosity are inconsistent. This relationship, based on red blood cell (RBC) studies from the 1970s and 1980s (1,2), has been shown in animals (3), polycythemic adults (4), iron-deficient children (5), and cyanotic children (6), although never in cyanotic adults. In contrast, more recent research showed viscosity to be independent of mean corpuscular volume (MCV) in mammals (7), polycythemic adults (8–10), and iron-deficient adults, in whom RBC filtration resistance was in fact lower (11). Such conflicting results may in part be explained by the heterogeneity of subjects and methodology. Viscosity of whole blood varies according to shear rate (Fig. 1). Moreover, iron deficiency per se can also cause hyperviscosity-like symptoms such as headache, paresthesias, irritability, and exercise intolerance (12), adding complexity to the clinical care of cyanotic patients.

Iron deficiency is a spectrum that in its extreme will sufficiently limit production of hemoglobin (Hb). Lower Hb and hematocrit (Hct) result in lower viscosity and systemic oxygen transport (13,14). Because Hct is the strongest determinant of whole blood viscosity (5,15), it is logical to hypothesize that lower viscosity rather than higher viscosity would result from iron deficiency in cyanotic congenital heart disease. To test this hypothesis, we quantified whole blood viscosity, iron indices, hyperviscosity symptoms, and exercise capacity specifically in adults with cyanotic congenital heart disease.

From the *Adult Congenital Heart Centre, †Department of Cardiac Medicine, ‡Department of Exercise Physiology, and the §Department of Haematology, Royal Brompton Hospital and National Heart and Lung Institute, Imperial College School of Medicine; ‖Child Health, Department of Clinical Developmental Sciences, St. George's Hospital, University of London; ¶Division of Biomedical Sciences, Department of Physiology and Biophysics, Imperial College School of Medicine; #Department of Haematological Medicine, King's College London Medical School, King's College Hospital; and **Hammersmith Hospital, London, England. This study was funded by a grant from the Royal Brompton Hospital Clinical Research Committee. Salary support for Dr. Broberg was provided by the Waring Trust, as well as an unrestricted grant from Actelion, United Kingdom. Dr. Gatzoulis and the Royal Brompton Adult Congenital Heart Centre and Centre for Pulmonary Hypertension have received support from the British Heart Foundation. Portions of this data were presented orally at the American College of Cardiology Scientific Sessions, March 2005, in Orlando, Florida.

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Abbreviations and Acronyms

Hb = hemoglobin
Hct = hematocrit
MCH = mean corpuscular hemoglobin
MCHC = mean corpuscular hemoglobin concentration
MCV = mean corpuscular volume
RBC = red blood cell
Tsat = transferrin saturation
V_{O_2} = oxygen consumption
VSD = ventricular septal defect

METHODS

Patients. Consecutive patients (age >16 years) with cyanotic congenital heart disease (transcutaneous oxygen saturation ≤92% at rest or ≤87% with exertion) were prospectively enrolled from a single tertiary care facility. Patients with decompensated congestive heart failure, acute hemoptysis, endocarditis, or recent surgery were excluded. The study was approved by the institutional ethics review committee, and all patients gave written consent before participation. For patients with Down syndrome or other forms of developmental delay, parental consent was obtained. All tests were performed within a 24-h period.

Assessment of symptoms and cyanosis. A thorough medical history was obtained and included details of hemoptysis or other bleeding, therapeutic phlebotomy, menses, diet, and use of any iron supplementation. Patients were asked in simple language about 12 common symptoms of hyperviscosity adapted from Perloff et al. (16). These included headache, faintness/dizziness, visual disturbances, fatigue, muscle aches, joint aches, paresthesias, easy bruising, epistaxis, gingival bleeding, gout, and poor mental function. Patients were asked to give each symptom a 0 to 3 rank (0 being nonexistent and 3 being severe and debilitating). Because symptoms often vary depending on coexisting illness (such as common colds), medication use (e.g., allopurinol), or weather, patients were specifically asked to score symptoms based on their current clinical state. The sum of the ranks gave a total viscosity symptom score (0 to 36 scale).

Oxygen saturation was measured in a seated position after a minimum of 5 min of rest using a standard transcutaneous pulse oximeter at the finger. Saturation was measured in the toe of any patient suspected of having differential distal cyanosis, and this saturation was used for subsequent analysis.

Laboratory tests. Hematologic measurements included Hb concentration, packed cell volume, MCV, mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC) as well as RBC, leukocyte, platelet, and reticulocyte counts (laser-based counter, Advia 120, Bayer, Tarrytown, New York). Serum tests included serum creatinine, urea, total bilirubin, albumin, thyroid stimulating hormone, RBC folate, and vitamin B_{12} levels. Plasma fibrinogen was also measured. In addition, P_{50} of the oxygen-Hb dissociation curve was measured on the same day (Hem-O-Scan, American Instrument Company, Silver Spring, Maryland). Laboratory values were reviewed by a hematologist blinded to other data. Patients were assigned to one of two groups: iron-deficient or iron-replete. Iron deficiency was defined by a transferrin saturation (Tsat) ≤ 15% (17) and a ferritin level ≤15 μg/l.

Viscosity measurement. Venous blood for viscosity assessment was obtained in the early morning, preprandially, and before any significant exertion or planned intervention. Samples were collected into heparinized tubes (10 ml) and analyzed within 4 h of collection. Viscosity measurements were performed at 37°C using a rotational viscometer fitted with a bob-in-cup system (Contraves Low Shear 30, Contraves AG, Zurich, Switzerland), according to the recommendations of the International Committee for Standardization in Hematology (18). Whole blood viscosity was measured at eight predefined shear rates (0.277, 0.69, 1.74, 4.39, 11.02, 27.7, 69.5, and 128.5 s^{-1}). Thereafter, Hct was adjusted to 45% (8) by the addition or subtraction of autologous plasma (rechecked by centrifugation) and viscosity at all shear rates was remeasured (Hct-adjusted viscos-

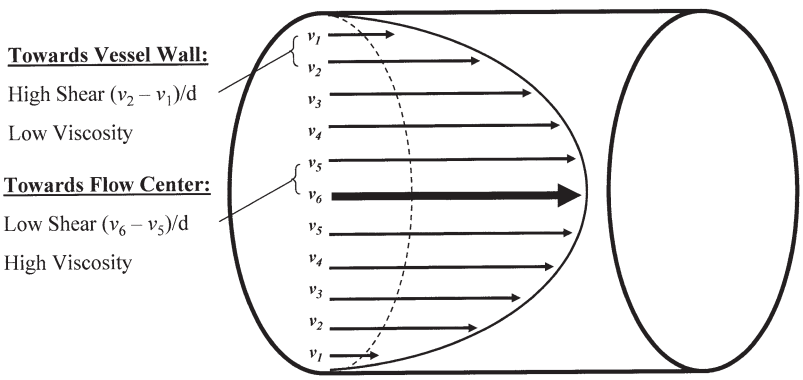

Figure 1. The inverse relationship between viscosity and shear rate is shown for a standard Poiseuille flow curve. Shear is defined as $(v_2 - v_1)/d$, where $(v_2 - v_1)$ is the difference in velocity between one flow layer and its adjacent layer and d is the distance between layers. Viscosity, the resistance to flow, is inversely proportional to shear rate. Thus, toward the wall of the vessel there is a larger difference in velocity (shear rate) but relatively low viscosity (i.e., less resistance). In contrast, toward the center of flow the difference in velocity is much smaller $(v_6 - v_5)/d$, hence a low shear rate, and viscosity is logarithmically higher (also see Fig. 2C).

ity). Plasma viscosity was measured at shear rates of 69.5, 94.5, and 128.5 s^{-1}, and then averaged.

Erythrocyte aggregation. Erythrocyte aggregation indices were also obtained using an erythrocyte aggregometer (Myrenne GmbH, Roetgen, Germany), a technique based on the increased transmission of light through gaps in a suspending medium when erythrocytes aggregate. Twenty-five microliters of Hct-adjusted blood (Hct 45%) were placed between a transparent cone and plate device at 22°C and subjected to a high shear rate to disaggregate the cells. After a short delay, a digital readout was obtained, either in stasis or at a low shear rate (3 s^{-1}), which reflected erythrocyte aggregation both in the absence of shear and in the presence of fluid movement, respectively. The final aggregation index represented the mean of three readings.

Exercise testing. All patients were asked to perform a 6-min walk test using standard guidelines (19). In addition, capable patients performed a maximal treadmill exercise test using a modified Bruce protocol. Oxygen saturation during and after peak exercise was recorded (sites as above), in addition to all standard measurements including oxygen consumption (V_{O_2}) measured spontaneously using a respiratory mass spectrometer (Amis 2000, Innovision, Odense, Denmark).

Statistical analysis. Data for continuous variables are expressed as mean ± standard deviation or median/interquartile range if the distribution was non-normal. Binary variables are expressed as frequencies (%). Comparisons between iron-deficient and iron-replete groups were made using the Student *t* test, Mann-Whitney *U* test, Fisher exact test, or chi-square testing as appropriate. Correlations were made using the Pearson or Spearman rho coefficient. Stepwise forward bivariate and multiple regression analyses were subsequently performed using significant univariate correlates ($p < 0.05$) to determine independent predictors of viscosity (separate models for whole blood and Hct-adjusted viscosity at high and low shear were constructed), total viscosity symptom score, treadmill exercise duration, and peak V_{O_2}. Statistical analysis was done using SPSS for Windows version 11.0 (SPSS Inc., Chicago, Illinois). A p value < 0.05 was considered statistically significant. The risk of type I error was 0.05 for each individual comparison.

RESULTS

Thirty-nine patients were studied (28 women, 11 men). Anatomical diagnoses were as follows: 2 patients (5%) had an atrial septal defect, 14 (36%) had a ventricular septal defect (VSD), 4 (10%) had a patent ductus arteriosus, 3 (8%) had an atrioventricular septal defect, 7 (18%) had truncus arteriosus or aortopulmonary window, 3 (8%) had transposition of the great arteries plus VSD, 2 (5%) had pulmonary atresia with VSD, 2 (5%) had double-outlet right ventricle, and 2 (5%) had complex single ventricle anatomy. All but 2 patients had Eisenmenger physiology (increased pulmonary vascular resistance with reversed or bidirectional shunt).

Iron deficiency. Comparative data between iron-deficient and iron-replete groups are shown (Table 1). Fourteen patients (36%) were iron deficient. Potential etiologies were significant hemoptysis (defined as ≥1 episode of at least one-half cup of bright red blood within the last year), therapeutic phlebotomy within the last year, or menorrhagia, but none of these factors were statistically different in frequency from the iron-replete group, except when more than one risk factor was present (Table 1). Two iron-deficient patients were taking warfarin but had no symptoms suggestive of gastrointestinal bleeding. No patients were vegetarian.

Iron-deficient patients had a lower resting oxygen saturation, lower Hb, and higher RBC count, in addition to expected differences in MCV, MCH, and MCHC (Table 1). There was no difference in spun Hct or packed cell volume, leukocyte, platelet, or reticulocyte counts. There were no differences in any blood chemistries, but there was a nonsignificant trend toward lower fibrinogen in those with iron deficiency. There was a significant rightward shift of the oxygen-Hb dissociation curve in the iron-deficient group.

Whole blood viscosity was not different between the iron-replete and iron-deficient groups at any shear rate (Table 2). Viscosity corrected to a Hct of 45% (Hct-adjusted viscosity) was also not different. There was no difference in plasma viscosity. The RBC aggregation index at low shear was significantly lower in those with iron deficiency.

Correlations with viscosity. Correlation coefficients for viscosity by univariate regression are shown. There was a positive correlation between whole blood viscosity and Hct, Hb, packed-cell volume, and RBC count at both low and high shear, and the strength of correlation was similar in those with or without iron deficiency (Fig. 2). In contrast, MCV was unrelated to whole blood viscosity at any shear rate. There was no correlation between viscosity and MCH, MCHC, ferritin, or Tsat at any shear rate (Table 3). On bivariate regression analyses, Hct remained the only predictor of whole blood viscosity at high and low shear in models that incorporated Hct and either Hb, packed cell volume, or RBC counts (all $p < 0.001$). In multiple regression models that incorporated all 4 univariate predictors, Hct remained the only predictor of whole blood viscosity at low shear (partial $r = 0.63$, $p < 0.001$) and high shear (partial $r = 0.84$, $p < 0.001$).

Because Hct is such a powerful determinant of viscosity, it was essential to measure viscosity with Hct corrected to 45%. The Hct-adjusted viscosity did not correlate with MCV (Fig. 2), nor with MCH, MCHC, ferritin, or Tsat. By regression, Hct-adjusted viscosity was related to plasma viscosity at low shear only ($r = 0.38$, $p = 0.02$), whereas there were no significant correlations at high shear (Table 3). Plasma viscosity itself correlated positively with age ($r = 0.47$, $p = 0.002$) and fibrinogen ($r = 0.41$, $p = 0.044$). By bivariate

Table 1. Comparisons Between Iron-Replete and Iron-Deficient Patients

	Iron-Replete	Iron-Deficient	p Value
General	n = 25	n = 14	
Age (yrs)	37.8 ± 13.1	40.0 ± 13.0	0.63
Female gender (n)	19 (76%)	9 (64%)	0.48
Developmental delay (n)	6 (24%)	3 (21%)	1.00
New York Heart Association functional class III to IV (n)	9 (36%)	5 (36%)	1.00
Total viscosity symptom score	10.3 ± 3.7	9.0 ± 5.4	0.42
Heart rate (beats/min)	85 ± 17	85 ± 15	0.95
Resting O_2 saturation (%)	82 ± 7	76 ± 7	0.019
Factors related to iron deficiency			
Current iron supplementation (n)	4 (16%)	3 (21%)	0.69
Hemoptysis within 1 yr (n)	7 (28%)	7 (50%)	0.29
Phlebotomy within 1 yr (n)	2 (8%)	5 (36%)	0.075
Menorrhagia (n)*	4 (21%)	4 (44%)	0.40
Any risk factor†	12 (48%)	11 (79%)	0.063
>1 risk factor†	1 (4%)	4 (29%)	0.047
Hematology			
Hemoglobin (g/dl)	20.6 ± 2.5	18.5 ± 2.8	0.021
Hematocrit (spun, %)	64 ± 9	64 ± 10	0.99
Packed cell volume (%)	62 ± 8	61 ± 9	0.84
RBC count ($\times 10^9$/l)	6.16/1.73	7.40/4.41	0.017
MCV (fl)	95 ± 7	81 ± 9	<0.001
MCH (g)	30.5/3.4	25.7/8.2	<0.001
MCHC (g/dl)	32.3 ± 1.5	30.1 ± 1.3	<0.001
Biochemistry			
Fibrinogen (g/l)	3.8 ± 1.5	2.8 ± 0.6	0.095
Serum iron (μmol/l)	22.5/15.0	7.9/6.2	<0.001
Total iron-binding capacity (μmol/l)	63 ± 15	79 ± 18	0.005
Transferrin (g/l)	2.6 ± 0.5	3.3 ± 0.6	0.001
Transferrin saturation (%)	33/20	9/9	<0.001
Ferritin (μg/l)	39/54	7/10	<0.001
P_{50} of O_2-Hb curve (mm Hg)	28.5 ± 2.1	31.1 ± 2.8	0.021

Results expressed as mean ± SD for normal continuous variables, mean/interquartile range for non-normal continuous variables, or frequencies (%) for binary variables. *Menorrhagia percentage is of female patients only. †Risk factors include hemoptysis or phlebotomy within the last year, or menorrhagia.

O_2-Hb = oxygen-hemoglobin dissociation; MCH = mean corpuscular hemoglobin; MCHC = mean corpuscular hemoglobin concentration; MCV = mean corpuscular volume; RBC = red blood cell.

regression, the only predictor of plasma viscosity was age (r = 0.54, p = 0.007), and not fibrinogen.

Relationship of symptoms to viscosity and iron deficiency. Total viscosity symptom scores ranged from 3 to 20 and were normally distributed (mean 9.8 ± 4.3, median 9.5). Because questions to patients with developmental delay (n = 9) were usually answered by the parents or caregiver, these patients were initially excluded from this portion of the analysis. Fatigue was ranked highest (mean 1.95 ± 1.0), accounting for 19% of the total points scored. Headache and faintness/dizziness were also common, and each accounted for 12%. Epistaxis, gingival bleeding, and gout were the least common, each contributing only 5% of total points. Viscosity symptom score did not correlate with age, oxygen saturation, Hb, Hct, MCV, MCH, ferritin, Tsat, or whole blood viscosity at any shear rate. We found no association between symptom score and the use of any specific medication, nor with exercise capacity (peak VO_2 or exercise duration). However, there was a positive correlation between total score and Hct-adjusted viscosity at all shear rates between 4.39 s^{-1} and 128.5 s^{-1} (r = 0.37, p = 0.02, and r = 0.41, p = 0.01, respectively).

Using a symptom score cutoff of above or below the median (<10 vs. ≥10), two groups were defined and compared. No difference was found in whole blood viscosity. However, there was a significantly higher Hct-adjusted viscosity in those with higher symptom score (n = 14) for shear rates 1.74 s^{-1} to 128.5 s^{-1} ($p < 0.05$ for each) (Fig. 3).

Exercise capacity. Thirty-five patients completed a 6-min walk test. No significant differences in walk distance were observed between patients with and without iron deficiency, and there was no correlation between any viscosity measurement and distance walked, heart rate attained, or decrease in oxygen saturation from baseline. Twenty-three patients completed a treadmill exercise study with VO_2. Reasons for nonparticipation were orthopedic limitations, difficulty with instructions regarding the mouthpiece, patient unwillingness, or logistical problems with equipment availability on the study day. No differences were found between those who did and did not exercise. The iron-deficient (n = 8) and iron-replete (n = 15) groups did not differ with respect to treadmill exercise duration, decrease in oxygen saturation at peak exercise, peak heart rate, or peak VO_2.

Table 2. Viscosity Data Compared Between Iron-Replete and Iron-Deficient Patients

	Iron-Replete	Iron-Deficient	p Value
Whole blood viscosity (mPa·s)	(n = 25)	(n = 14)	
Shear rate = 0.27 s^{-1}	95/75	134/101	0.59
Shear rate = 0.69 s^{-1}	64/41	80/55	0.61
Shear rate = 1.74 s^{-1}	39/26	46.5/30	0.71
Shear rate = 4.39 s^{-1}	23.1/15.7	27.6/16.3	1.00
Shear rate = 11.02 s^{-1}	15.2/8.9	18.5/10.6	1.00
Shear rate = 27.7 s^{-1}	11.3/6.9	12.5/6.0	0.48
Shear rate = 69.5 s^{-1}	9.1/5.4	9.4/4.4	0.50
Shear rate = 128.5 s^{-1}	8.2/4.8	7.8/3.7	0.44
Hematocrit-adjusted viscosity (mPa·s)			
Shear rate = 0.27 s^{-1}	49.9 ± 9.9	48.6 ± 11.5	0.71
Shear rate = 0.69 s^{-1}	32.2 ± 5.9	30.5 ± 6.6	0.41
Shear rate = 1.74 s^{-1}	20.1 ± 3.6	18.6 ± 4.1	0.26
Shear rate = 4.39 s^{-1}	12.9 ± 2.4	11.6 ± 2.8	0.15
Shear rate = 11.02 s^{-1}	8.5/2.7	8.1/1.6	0.48
Shear rate = 27.7 s^{-1}	6.2/1.4	5.9/1.5	0.15
Shear rate = 69.5 s^{-1}	5.0/1.1	4.7/1.0	0.06
Shear rate = 128.5 s^{-1}	4.3/0.9	4.0/1.0	0.07
Other viscosity parameters			
Plasma viscosity (mPa·s)	1.4 ± 0.2	1.4 ± 0.2	0.68
Red cell aggregation index, low shear	11.9 ± 2.2	9.7 ± 2.8	0.01
Red cell aggregation index, stasis	3.5 ± 0.9	3.1 ± 1.4	0.25

Results expressed as mean ± SD for normal variables or mean/interquartile range for non-normal variables.

On univariate regression analysis, no significant correlations were found with exercise parameters and age, oxygen saturation, MCV, MCH, ferritin, or Hct-adjusted viscosity. Exercise duration correlated with Hct (r = 0.42, p = 0.047), but not significantly with Hb (r = 0.39, p = 0.097). Peak V_{O_2} correlated with Hct (r = 0.54, p = 0.008), Hb (r = 0.46, p = 0.026), whole blood viscosity at low shear (r = 0.42, p = 0.044), and plasma viscosity (r = 0.51, p = 0.014). In bivariate regression analyses, Hct remained the only predictor of peak V_{O_2} in models that incorporated Hct and either Hb or whole blood viscosity at low shear (partial r = 0.54, p = 0.0083 for both). In a model including Hct and plasma viscosity, both parameters remained independently predictive of peak V_{O_2}.

Two groups were subsequently defined by a Hct above or below 65%, a common cut point for symptom generation (20,21) (mean for the study population was 64.3%). Those with Hct ≥65% (n = 10) had a significantly longer exercise duration (6.8 ± 1.6 min vs. 4.8 ± 1.7 min, p = 0.01) and peak V_{O_2} (12.6 ± 3.4 ml/kg/m^2 vs. 9.8 ± 2.6 ml/kg/m^2, p = 0.036) than those with Hct <65% (n = 13, Fig. 4). There was a similar trend for 6-min walk test distance, but this did not reach statistical significance (401 ± 97 m vs. 332 ± 129 m, p = 0.09). In these same individuals, whole blood viscosity was significantly higher (p ≤ 0.003 at all shear rates) (Fig. 4).

DISCUSSION

Principal findings. We show that iron deficiency and microcytosis do not increase whole blood viscosity in adults with cyanotic congenital heart disease. Hypervis-

Table 3. Univariate Correlations for Determinants of Whole Blood Viscosity and Viscosity at Hematocrit of 45% (Hct-Adjusted Viscosity)

	Whole Blood Viscosity				Hct-Adjusted Viscosity			
	Low Shear		High Shear		Low Shear		High Shear	
Variable	r Value	p Value	r Value	p Value	r Value	p Value	r Value	p value
Age (yrs)	−0.03	ns	−0.07	ns	0.28	0.09	0.05	NS
Resting heart rate (beats/min)	−0.19	ns	−0.11	ns	−0.28	0.10	0.05	NS
Oxygen saturation (%)	−0.20	ns	−0.02	ns	0.20	ns	0.10	NS
Hematocrit (%)	**0.63**	**<0.001**	**0.84**	**<0.001**	0.09	ns	−0.17	NS
Packed cell volume (%)	0.52	0.001	0.77	<0.001	0.07	ns	−0.14	NS
Hemoglobin (g/dl)	0.47	<0.002	0.72	<0.001	0.12	ns	−0.13	NS
Red blood cell count (×10^9/l)	0.48	<0.002	0.58	<0.001	0.12	ns	−0.16	NS
Ferritin (μg/l)	−0.20	ns	−0.05	ns	0.02	ns	0.15	NS
Transferrin saturation (%)	−0.54	ns	0.16	ns	0.04	ns	0.26	NS
Fibrinogen (g/l)	0.21	ns	0.28	ns	0.27	ns	−0.28	NS
Plasma viscosity	0.17	ns	0.15	ns	0.38	0.02	0.16	NS

Bold indicates that the variable was a significant predictor by bivariate or multiple regression (see text for r and p values). Low shear = 0.27 s^{-1}, high shear = 128.5 s^{-1}.

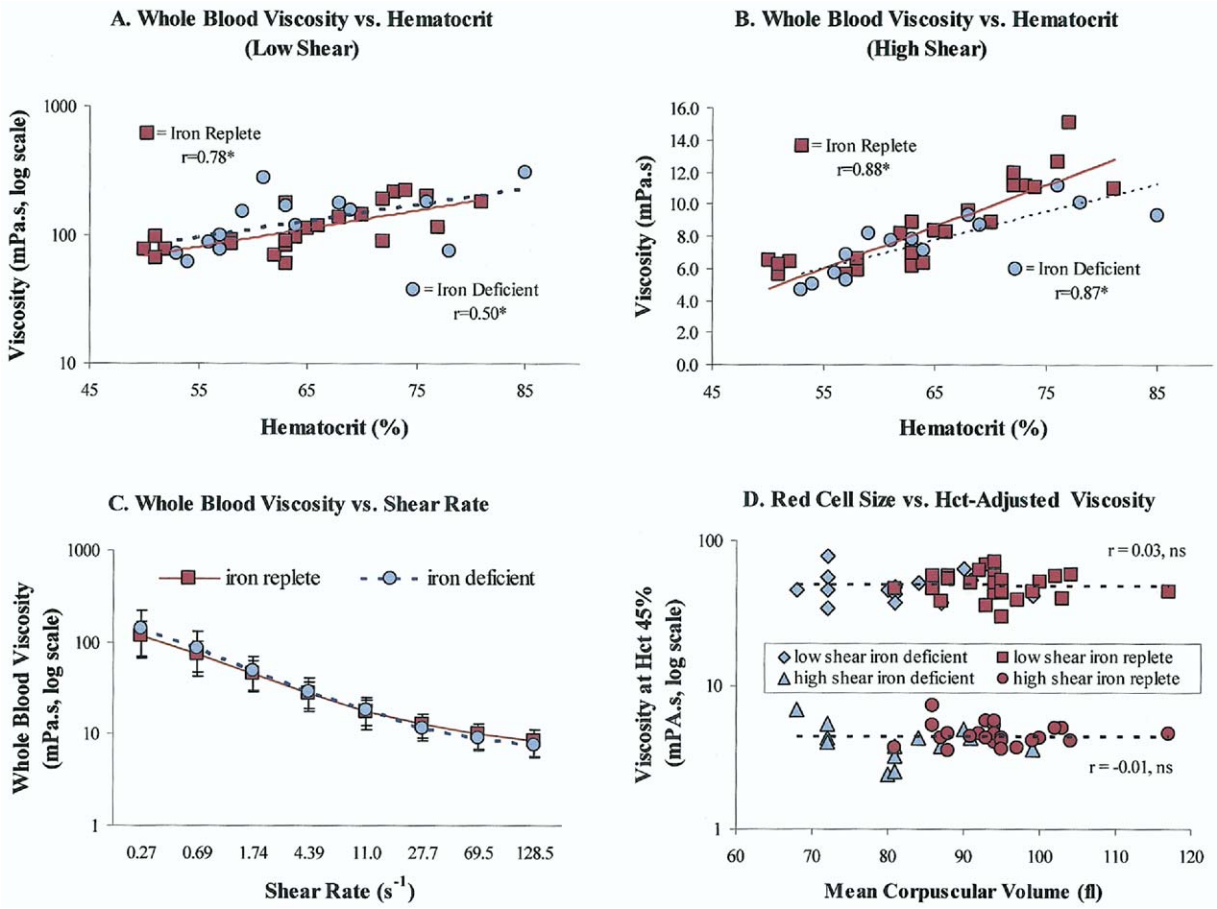

Figure 2. Whole blood viscosity strongly correlates with hematocrit (Hct) at both low shear **(A)** and high shear **(B)**, with a logarithmic increase in viscosity at lower shear rates **(C)**. The same relationship between shear and viscosity (mean ± SD) was seen for both iron-deficient patients **(circles)** and iron-replete patients **(squares)**. In contrast, mean corpuscular volume had no relationship with Hct-adjusted viscosity at any shear rate **(D)**. *$p < 0.001$. fl = femtoliters.

cosity symptoms were common in our cohort and correlated with Hct-adjusted viscosity but not with iron status, Hct, Hb, or MCV. Moreover, higher Hct levels were associated with better exercise capacity despite higher viscosity.

Iron deficiency and viscosity. The proposed link between iron deficiency and hyperviscosity is based on studies with considerable differences in subjects and methods that reach inconsistent conclusions. Comparisons are also difficult because whole blood viscosity cannot be quantified as a single entity. Many studies were conducted in polycythemic subjects (8–10), who, unlike cyanotic patients with secondary erythrocytosis, have elevated leukocyte and platelet counts that contribute significantly to hyperviscosity and symptoms (22,23).

Our finding that iron deficiency does not elevate viscosity is in harmony with the current understanding of erythropoiesis and the general determinants of viscosity. Because of iron's key role in Hb synthesis, reduced availability of iron hinders erythropoiesis. Because Hct is the strongest determinant of viscosity (5,15), it is logical to suspect that lower viscosity, rather than higher, will eventually result from limited iron.

The second most powerful determinant of viscosity is fibrinogen via its role in forming temporary bonds that facilitate RBC aggregation (24). Aggregation is dependent on Rouleaux formation, which is hindered by microcytosis. In iron-deficient patients we found significantly lower RBC aggregation (low shear) and a nonsignificant trend toward lower fibrinogen levels. Phlebotomy-induced iron deficiency has been shown to lower fibrinogen levels in select patients (25), lending further support to our finding that iron deficiency actually decreases RBC aggregation.

We found a significant right-shifted oxygen-Hb dissociation curve in iron-deficient patients, which is consistent with some (26) but not all studies (17). The rightward shift favors tissue oxygen release in response to decreased systemic oxygen transport. It is unclear whether this shift is physiologically advantageous or detrimental (20). Increased oxygen extraction lowers mixed venous oxygen saturation, which when shunted right-to-left will in turn lower arterial saturation, as seen in iron-deficient patients here.

Etiology of iron deficiency. Over one-third of our patients were iron deficient, which is several fold higher than in the general population (12) and consistent with other published data (27). We did not show a significant difference in risk

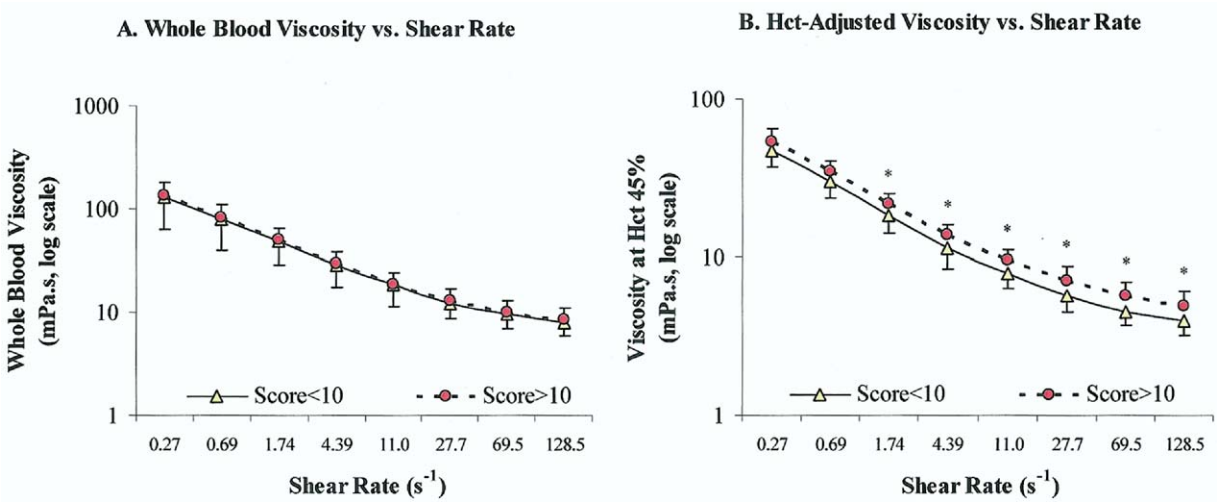

Figure 3. Viscosity (mean ± SD) is plotted against shear rate for patients with a hyperviscosity symptom score <10 compared with those ≥10. There was no difference in whole blood viscosity **(A)**, but a significant difference in viscosity adjusted to a hematocrit (Hct) of 45% at shear rates ≥4.39 s^{-1} **(B)**. *$p < 0.05$.

factors for iron deficiency between iron-deficient and iron-replete patients (although the study was underpowered to show such differences) unless more than one risk factor was present (Table 1). In 3 iron-deficient patients no risk factors were evident. Iron deficiency in this cohort is most likely multifactorial because of: 1) consumption of iron through erythropoiesis; 2) persistent blood loss through normal menses or imperceptible gastrointestinal bleeding; and 3) limited dietary intake and/or absorption. Further work on iron metabolism in cyanotic heart disease may clarify etiology.

Origin of symptoms. Although symptoms such as fatigue, headache, and lightheadedness were common, we did not show a significant correlation between self-reported symptom scores and Hb, Hct, oxygen saturation, whole blood viscosity, or exercise function. Importantly, symptom score as we measured here was unrelated to indices of iron status (i.e., MCV, MCH, Tsat, or ferritin). Similar symptoms occur when systemic oxygen transport is low (14), supporting the overlapping symptomatology between iron deficiency and hyperviscosity.

Although symptoms did not correlate with whole blood viscosity, symptom score was significantly related to Hct-adjusted viscosity (Fig. 3). This is one of the most intriguing findings of our study. None of the hematologic or biochemical variables we measured accounted for this. Several other possibilities are worth exploring. It should be remembered that Poiseuille assumptions about flow at steady state (Fig. 1) rarely apply to blood in the highly complex and pulsatile human circulation. Flow resistance at rest mainly occurs in the arterioles (10 to 20 μm in diameter), where viscosity is more dependent on local RBC interactions such as temporary fibrinogen bonding or Rouleaux formation. In contrast, flow resistance during exercise (hyperemia) occurs at the capillary level in patients with hyperviscosity (28). Because erythrocytes pass single file through the capillary bed, flow may depend more on membrane flexibility. All of these variables are potential targets for further investigation into symptomatology.

Iron deficiency has been associated with an increased risk of stroke in cyanotic children (29–31) and adults (32,33). The theoretical explanation for this is that iron deficiency raises viscosity and impairs cerebral blood flow. Several potential confounders must be considered in light of the data presented here. First, abnormalities of clotting factors known to occur in cyanosis may have coexisted (34,35). Second, platelet abnormalities may have been present given that iron deficiency is associated with both thrombocytopenia (23) and thrombocytosis (36–38). Third, severely cyanotic patients may have been more aggressively treated with phlebotomy. Fourth, poor cerebral oxygenation may be secondary to low Hb levels and reduced systemic oxygen transport. Although cerebrovascular events are more common in cyanotic heart disease, event rates are low (39), making conclusions based on small cohorts difficult. Although phlebotomy improves cerebral blood flow, it does not increase cerebral oxygen delivery (40) and, as several investigators point out (20,21), it should never be practiced as a means of reducing stroke risk in these patients regardless of Hct.

Major determinants of exercise capacity. The predominant factors determining exercise capacity in this cohort were Hct and Hb. Counterintuitively, we found that viscosity was positively related to exercise capacity via its association with higher Hct levels, which is certainly a confounder, as the regression analysis showed. This intriguing finding suggests that erythrocytosis to extreme levels may be worth the cost of hyperviscosity, and that some patients may have found an optimal balance between these two opposing forces. Systemic oxygen transport (cardiac output multiplied by oxygen content) is determined by Hb (14) and not Hct. To optimize oxygen delivery without exacerbating hyperviscosity, one would maximize Hb within

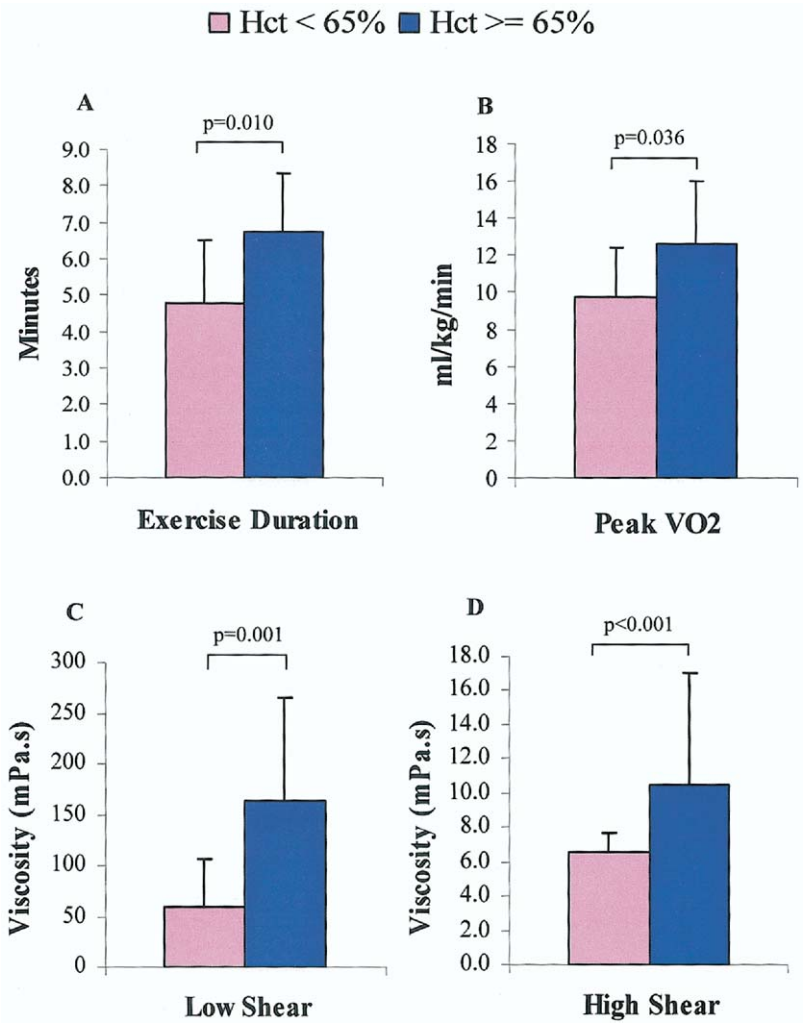

Figure 4. Exercise and viscosity parameters are compared between patients with hematocrit (Hct) ≥65% or <65%. Those with higher Hct had a longer mean treadmill exercise duration **(A)** and higher peak oxygen consumption (V_{O_2}) **(B)** despite higher whole blood viscosity at low shear **(C)** and high shear **(D)** in the same group. V_{O_2} = oxygen consumption; low shear = 0.27 s^{-1}; high shear = 128.5 s^{-1}.

the smallest volume of Hct. To do so, erythrocytes should be bigger (i.e., higher MCV) with a higher concentration of Hb (i.e., increased MCHC). Macrocytosis in cyanotic heart disease has been described (26,27). Indeed, 3 of our patients were macrocytic (>100 femtoliters) (Fig. 2D) without any evidence of folate or vitamin B_{12} deficiency, liver disease, hypothyroidism, reticulocytosis, or Down syndrome (41). This macrocytic trend may be part of the normal response to cyanosis. Contrarily, iron deficiency lowers Hb, although Hct remains unchanged, as our data show; hence this optimal balance is lost. Increasing Hb with iron supplementation but preserving Hct through phlebotomy has been shown to improve exercise function (42), although we could not show differences in exercise function relative to iron deficiency in this study.

Exercise performance in cyanotic heart disease is complex, and multiple factors likely each play separate roles in limiting exertional capacity (43). Because plasma viscosity was an independent predictor of peak V_{O_2}, this may be another avenue for additional study.

Clinical implications. In the clinical care of patients with cyanotic heart disease, the relationship between iron and hyperviscosity is raised in discussions of the appropriateness of supplemental iron and the indications for therapeutic phlebotomy. Risk of "decompensated" overproduction, defined as a rapid increase in Hct via raised erythropoietin levels as soon as any supplemental iron is given (17), has been used by several investigators as justification to limit oral iron by treating only until an increase in Hct is discernable (20,21). However, those with higher Hct had better exercise duration in our data set. Thus it seems imprudent that any level of Hct should be universally considered too high (20). Iron should be in sufficient supply to allow one to attain steady-state erythropoiesis as determined by the patient's own unique underlying physiology. It has been shown that hyperviscosity can be alleviated with supplemental iron without inducing clinical decompensation (44).

Several findings in our study may be relevant to discussions about the appropriateness of therapeutic phlebotomy. First, we show that iron deficiency lowers Hb but not Hct

because of production of more but smaller RBCs. Thus, one would expect long-term phlebotomy to reduce Hb yet have no lasting effect on Hct and viscosity. Second, patients with lower Hct had a lower exercise capacity, implying that reducing Hct to improve exercise tolerance may be disadvantageous. Third, we found that symptoms attributed to hyperviscosity are just as frequent at lower Hct levels, as have others (44). Again, such symptoms may also be attributable to iron deficiency per se irrespective of Hb level (45), or may relate to poor systemic oxygen delivery. Hence, although this was not a study of therapeutic phlebotomy, our findings suggest the practice may be counterproductive. To our knowledge, no one has yet shown long-term (>2 weeks) (46) symptomatic improvement in cyanotic heart disease from phlebotomy, nor any prognostic advantage.

Study limitations. Any study of viscosity is bound by the limits of the simplified steady-state models used to measure infinitely more complex and dynamic systems in vivo. Our study focused on a representative range of shear. Others have used a similar range (9,47), but this is by no means exhaustive. The clinical relevance of more extreme rates of shear becomes questionable.

Sample size limits the power to detect small differences and is a limitation of this cross-sectional study. Our sample size is adequate to detect a 20% difference in Hct-adjusted viscosity (low or high shear) with >80% power, whereas these values differ by <10% in our data set. We have insufficient power to show that the differences we encountered were different, but we conclude they are not different. In fact, some trends suggest that iron-deficient patients have lower Hct-adjusted viscosity (high shear), which reinforces confidence in the conclusion that they are indeed not higher. Although the study is underpowered to show the significance of smaller differences in viscosity, they arguably would not be clinically relevant. Despite the fact that not all patients participated in treadmill exercise testing, the power in this study to show significantly higher exercise duration in those with Hct ≥65% was 83% (one-tailed). Power to show a higher peak V_{O_2} was only 64%, however. This study reports on a number of statistical analyses. No special technique was used to adjust the p values of individual comparisons. Because the risk of type I error was 0.05 for each, the cumulative risk of there being at least one type I error in the study may be large.

The inherent variability in underlying anatomy and physiology in this patient population further challenges the analysis of data. Self-reported symptom assessment such as we used here is subjective and not formally validated. We used a symptom score designed specifically with hyperviscosity in mind (1), but acknowledge the limitations of making any conclusion based on this unsubstantiated tool. We recognize that exercise capacity is dependent on patient cooperation and motivation. Larger longitudinal studies are likely to shed more light on the complex interactions between viscosity, iron, symptoms, exercise capacity, and treatment effects.

Conclusions. Iron deficiency and microcytosis do not increase blood viscosity in adults with cyanotic congenital heart disease. Symptoms such as fatigue, headache, and dizziness are common in cyanotic patients, but unrelated to Hct, Hb, iron indices, or cell size. However, these symptoms do correlate with higher viscosity when Hct is controlled, although this relationship is unrelated to iron and merits further investigation. Exercise capacity is greater in patients with higher Hct despite their higher blood viscosity. More studies on achieving iron equilibrium and optimal Hb levels will refine the management of these complex patients.

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Reprint requests and correspondence: Dr. Craig S. Broberg, UHN 62, Division of Cardiology, Oregon Health and Science University, 3181 SW Sam Jackson Park Road, Portland, Oregon 97239. E-mail: brobergc@ohsu.edu.

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