Gender difference in rheologic properties of blood and risk of cardiovascular diseases

M.V. Kameneva *, M.J. Watach and H.S. Borovetz

McGowan Center for Artificial Organ Development, University of Pittsburgh, Pittsburgh, PA, USA

Abstract. According to official statistical data there is a significant difference between pre-menopausal women and age-matched men in morbidity and mortality from cardiac diseases and especially from myocardial infarction. There are several speculations regarding the nature of this phenomenon which have both supporting and refuting evidence.

Our hypothesis was that due to regular physiologic bleeding, rheological properties of blood of pre-menopausal women are superior to those of men, and place such women at a lower risk of cardiovascular diseases than men in any age group. We believe that this difference in hemorheological properties is due to the reduced concentration of red blood cells (RBCs) and due to greater population of younger and less population of older RBCs in female blood. We studied mechanical properties of blood from 47 pre-menopausal women and 50 age-matched men. Compared to female blood, male blood had higher viscosity and RBC aggregation and lower RBC deformability. Oxygen Delivery Index, calculated as a ratio of hematocrit to blood viscosity, was found to be significantly lower in male blood. Decreased oxygen delivery along with increased RBC aggregation and decreased RBC deformability may contribute to the higher risk for the development of cardiovascular diseases. Regular blood donation may reduce hematocrit and blood viscosity, improve rheological properties of blood, and increase oxygen delivery in men.

1. Introduction

Mortality from coronary heart disease (CHD) has declined steadily in the United States for the past three decades. However, CHD is still the leading cause of death in this country. According to official statistical data [1], the mortality of men from CHD and, especially, from myocardial infarction is significantly higher than the mortality of women during reproductive age (Fig. 1).

Acute cardiac events may be caused by an insufficient blood supply due to reduced vessel lumen or altered mechanical properties of blood including increased blood viscosity and RBC aggregation, and decreased RBC deformability. In fact, a number of statistical and experimental studies have demonstrated that increased hematocrit, RBC aggregation and blood viscosity are important risk factors of cardiovascular disorders [2–4]. It is proven statistically [5] that during the reproductive period of female life, there is a significant difference in hematocrit levels in male and female blood (according to [5] hematocrit is equal to 47 ± 5% for adult males and 42 ± 5% for females). This difference may result, in turn, in significantly lower blood viscosity in women during the reproductive period. One hypothesis holds that the difference in hematocrit levels and viscosity of female and male blood is due to women’s menstrual bloodloss; this could explain the reduced risk of cardiovascular diseases in premenopausal women compared to age-matched men [6]. A mathematical model of the hematocrit level development during female and male life was presented in [7]. The model analyzed simple differential equations of red blood cell balance in the body and was based on the assumption that females lose about 50–100 ml of blood every...
menstrual cycle. An additional assumption was that at after age 50, the erythropoesis in both female and males decreases. This mathematical model has proven that the significant difference between hematocrit levels in female and male blood is mostly related to the regular loss of a small amount of blood by females during their reproductive period. The hypothesis that a decreased hematocrit level may reduce the risk of cardiovascular diseases in pre-menopausal women as compared to age-matched men was examined through the study of the relationship between increased hematocrit and hypertension in young men [7,8]. Within the group under investigation, which involved almost 1500 teenagers (17–18 years old) with equal numbers of boys and girls, the number of boys with pressure higher than 140/80 mm Hg was 13 times larger than that of girls. As much as 94% of the boys with high blood pressure possessed an elevated hematocrit level (Ht > 50%), and 95% of boys with high hematocrit possessed an elevated blood pressure. Thus this study demonstrated that there is a strong relationship between high hematocrit (and thus high blood viscosity) and increased blood pressure. So called “juvenile” hypertension is a serious risk factor for the development of atherosclerosis, myocardial infarction and stroke. Overall, a significant positive correlation was found between hematocrit and blood pressure ($r = 0.66$).

Another reason for the difference in morbidity and mortality from cardiac diseases between pre-menopausal women and men may be associated with the disparity in the mechanical properties of their RBCs related to cell physiological aging process. RBCs have a limited life span due to aging (100–120 days in human blood). New (“young”) RBCs continuously enter the circulation system and senescent (“old”) cells are eliminated by the reticulo-endothelial system. Approximately 0.8% of the total number of RBCs are renewed every 24 hours [9]. Due to monthly blood loss female blood is expected to contain more young and fewer old RBCs. In fact, it has been shown that the age distribution of RBCs is significantly different in males and females [10]. The authors demonstrated that female blood has about 80% more young RBCs and 85% fewer old RBCs than male blood. Our previous study has revealed a significant difference in the mechanical properties of young versus senescent RBCs [11]. A statistically significant increase in RBC mechanical fragility, low shear blood viscosity, erythrocyte sedimentation rate and RBC rigidity for the population of “old” RBCs compared to “young” RBCs were demonstrated. Thus, our previous results showed that a significant elevation of RBC aggregability and a drop of RBC deformability accompany RBC aging. These results were in good agreement with other studies, which have also shown a difference in the mechanical properties of young versus old RBCs [12–17].

In this paper we present the results of the continuation of our studies of the rheological properties of male and pre-menopausal female blood. Oxygen Delivery Index as a function of hematocrit and symp-
Asymptotic blood viscosity is discussed as an additional risk factor for the development of cardiovascular diseases. We also present a hypothesis that relates an increase in erythrocyte mechanical fragility to the increased risk of the development of cardiovascular diseases.

2. Materials and methods

Hematocrit level, blood and plasma viscosity, RBC aggregability, deformability and mechanical fragility were determined in blood samples obtained from 97 healthy volunteers, students and lab personnel, including 47 pre-menopausal women (age 26.1 ± 5.0) and 50 age-matched men (age 26.8 ± 5.2). Smokers and pregnant women were excluded from the study. The applied methods are described elsewhere [18]. In short, viscosity of blood for original and standardized hematocrit (Ht = 40%) was measured by Couette rheometer (Contraves Low Shear 30) over shear rates from 0.277 to 128.5 s\(^{-1}\). Asymptotic viscosity of blood was also measured for original and standardized hematocrit by capillary viscometer (Cannon Instrument Co.) at a shear rate of approximately 500 s\(^{-1}\). Plasma viscosity was measured by Couette rheometer. The ability of RBCs to deform was represented as a RBC Rigidity Index, which is the inverse value of the RBC deformability. The RBC Rigidity Index is calculated according to the ratio:

\[
\text{RBC rigidity index} = \frac{\text{asymptotic blood viscosity}}{\text{plasma viscosity}},
\]

where asymptotic blood viscosity is measured for standard hematocrit of Ht = 40% [17]. An increase in the Rigidity Index signifies a decrease in the RBC deformability. It was shown previously [19] that even a modest increase in the RBC rigidity parameter measured by the same method was accompanied by a very significant elevation in the pressure required for RBC entry into small capillaries. RBC aggregation was estimated by measurement of erythrocyte sedimentation rate (ESR) at standardized hematocrit and low shear blood viscosity. An increase in both low shear viscosity and ESR indicates the increase in aggregation of RBCs [4].

RBC mechanical fragility was determined in 27 male and 20 female blood samples by the previously described method [18].

Amount of oxygen delivered to tissue (bulk O\(_2\) transport) is directly proportional to hematocrit and inversely proportional to blood viscosity; thus, the ratio of hematocrit to blood viscosity can be considered to characterize oxygen delivery. Since the characteristic shear rate range in the arterial region of the vascular system is from 150 to 1500 s\(^{-1}\), the asymptotic blood viscosity is used for calculation of the Oxygen Delivery Index (ODI):

\[
\text{ODI} = \frac{\text{hematocrit}}{\text{asymptotic blood viscosity}}.
\]

Oxygen Delivery Index was calculated for 47 pre-menopausal women and 50 age-matched men by using hematocrit level obtained from blood samples anticoagulated with ethylenediamine-tetraacetic acid (EDTA). Our preliminary tests have shown that this level is normally about 0.5% lower than hematocrit measured in blood anticoagulated with dry heparin. Asymptotic blood viscosity was measured in the same EDTA blood samples by a capillary viscometer at room temperature.
Fig. 2. Asymptotic blood viscosity and Oxygen Delivery Index versus hematocrit for 47 pre-menopausal women and 50 age-matched men.

Data are presented as mean ± standard deviation. The unpaired Student t-test was performed to estimate the statistical significance of differences between the values of rheological parameters of male and female blood. A value of $p < 0.05$ is assumed to indicate statistical significance.

3. Results

Most of the studied hemorheological parameters were found to be significantly different for male and female blood. Hematocrit level, blood viscosity at both high and low shear rates, and at both original and standardized hematocrit, and ESR at standardized hematocrit were significantly higher in male than in female blood. Viscosity of plasma did not differ significantly (1.74 ± 0.08 cP in male blood vs. 1.73 ± 0.09 cP in female blood, $p > 0.05$). A relatively small (10.7%) but statistically significant difference between male and female RBC Rigidity Index (3.1 ± 0.1 vs. 2.8 ± 0.1, $p < 0.001$) was found. The most pronounced differences between male and female blood were found for hematocrit (45.8 ± 2.7% vs. 40.0 ± 2.4%, $p < 0.001$), asymptotic (6.0 ± 0.8 cP vs. 4.8 ± 0.4 cP, $p < 0.001$) and low shear (58.3 ± 12.1 cP vs. 36.1 ± 4.9, $p < 0.001$) blood viscosity (both measured at the original hematocrit) and erythrocyte sedimentation rate measured at the standardized hematocrit (Ht = 40%) (10.8 ± 4.1 mm/hr vs. 8.4 ± 3.1 mm/hr, $p < 0.005$). RBC mechanical fragility was found to be 28% higher for male blood than for female blood (0.502 ± 0.072 vs. 0.393 ± 0.065, $p < 0.001$).

Oxygen Delivery Index was found to be 8.4 ± 0.5 for female blood and 7.7 ± 0.3 for male blood ($p < 0.001$). Figure 2 presents the whole set of the ODI data for 47 females and 50 males plotted versus hematocrit. In addition, Fig. 2 shows values of the asymptotic blood viscosity used for the calculation of ODI. One can see that the majority of female blood samples possesses much lower viscosity and much higher ODI than do male blood samples.

4. Discussion

There are several speculations regarding the reason for the much lower morbidity and mortality from heart disease in pre-menopausal women than in men and post-menopausal women. The most commonly
accepted hypothesis is that the higher level of some circulating hormones that have a relaxing effect on coronary arteries and aorta [20–24] protects women in reproductive age. However, the increase in risk of heart disease was found to be essentially the same in women with surgical menopause regardless of ovary removal [25]. Another assumption is that the greater incidence of heart disease in men and postmenopausal women is due to higher levels of stored iron in these two groups compared with premenopausal women [25,26]. Recently it was reported that among 2,682 middle-aged men who were followed for about 5 years, the risk of heart attack was 86 percent lower among blood donors [27]. Another epidemiological study, performed on nearly 4,000 people, found that the non-smoking men who had donated blood in the previous three years were at half the risk to have a heart attack or stroke as were those who had never donated blood [28]. The authors hypothesized that the depletion of iron through blood donation may reduce coronary heart disease among donors. At least seven previous epidemiological studies have found a positive association between CHD and various indicators of body iron. On the other hand, 18 epidemiological studies have found a negative or no association [29].

Our hypothesis holds that the reduced morbidity and mortality from cardiovascular diseases in women of reproductive age in comparison to men and postmenopausal women may be associated with the difference in age distribution of red blood cells and the subsequent difference in mechanical properties of blood of menstruating women versus men and postmenopausal women.

Our study revealed a statistically significant difference in most hemorheological parameters between male and female blood. Male blood had much higher levels of hematocrit, blood viscosity, RBC aggregation and rigidity than did the blood of premenopausal women. Since our previous results demonstrated a significant elevation of RBC aggregability and fragility and a decline in RBC deformability with cell’s aging, the differences in mechanical properties of male RBCs compared to female RBCs may be most likely associated with the difference in age distribution of RBCs in blood of men and pre-menopausal women.

A significant number of experimental and statistical studies show that high levels of hematocrit and blood viscosity, a decrease in the RBC deformability, and increase in the ability of RBCs to aggregate are all important risk factors for cardiovascular disease along with other risk factors such as increased concentration of low density lipoproteins and fibrinogen, obesity, smoking, etc. [4,30]. As mentioned above, men possess a higher hematocrit and greater percentage of old RBCs with suboptimum mechanical properties than do women of reproductive age, who due to menstruation have a lower hematocrit and a higher number of young cells and a lower number of old RBCs than their male counterparts. Thus, our results suggest that an elevated hemorheological risk for males may have a strong association with the age distribution of RBCs in male blood.

It is known that old RBCs are removed from the circulation by the reticulo-endothelial system as they become less deformable with age [31]. We found that the male RBCs have a decreased deformability and increased mechanical fragility compared to that of female cells. Thus, male blood, besides possessing inferior mechanical properties, has a higher number of RBCs which are subjected to phagocytosis, that leads to the release of hemoglobin into plasma. The amount of free hemoglobin released into the plasma from these RBCs after normal metabolic removal is approximately 10% of the daily turnover (6000–8000 RBCs per 1 mm$^3$ of blood) [5]. Plasma free hemoglobin has a strong vasoconstrictive effect due to its ability to bind nitric oxide, an endothelium-derived relaxing factor [32]. Nitric oxide is a known potent vasodilator and an inhibitor of a platelet aggregation [33]. Decrease in nitric oxide concentration is associated with hypertension and atherosclerosis [34]. Thus, we speculate that intravascular nitric oxide concentration may be lowered in male blood by plasma free hemoglobin released from old RBCs. Additionally, since old RBCs have much higher fragility than young ones, they are much more sensitive
to shear stress. As a result, destruction of some old RBCs may occur in any region of the vascular system with high shear stress. The same regions of the vascular system are characterized by high production of NO [33,35,36]. We hypothesize that the blood of pre-menopausal females, upon comparison with male blood, has fewer old RBCs and releases less amount of free hemoglobin from destroyed old RBCs into plasma. Therefore, the average concentration of nitric oxide in the female vascular system is probably higher than that in males. Some proof of this hypothesis can be found in a recently published paper [37]. This may be another reason for the slowed development of atherosclerosis and for the reduced morbidity and mortality from cardiovascular diseases (particularly from myocardial infarction) in women of reproductive age than in men of any age. It seems appropriate to suggest that small, regular bloodletting could significantly improve mechanical properties of male blood and, possibly, increase the intravascular nitric oxide concentration, thus reducing the development of cardiovascular diseases and even sudden death from myocardial infarction.

We hypothesized that remarkable dissimilarity in morbidity and mortality due to cardiovascular diseases in pre-menopausal women versus men and postmenopausal women could be related to a substantial difference in the mechanical properties of their blood. This difference, in turn, relates to the age distribution of RBCs, which is affected by monthly bleeding in pre-menopausal women. Based on these results, one can suggest that the increase in the concentration of young RBCs and decrease in the concentration of old RBCs in the human blood resulting from blood loss is of a great importance for the prevention of cardiovascular diseases. These results implicate the advantage of blood donation, since donation of blood decreases the proportion of old RBCs and increases the proportion of young ones. This, in turn, may significantly improve mechanical properties of the donor’s blood and increase oxygen delivery to donor’s tissues and organs. The recent publications [27,28] mentioned in the discussion above, support our conclusions regarding the benefit of blood donation.

References


