Published in final edited form as:

Clin Hemorheol Microcirc. 2014 ; 56(3): 197–204. doi:10.3233/CH-131685.

The role of hemorheological factors in cardiovascular medicine

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Abstract

Cardiovascular diseases (CVD) are the most frequent cause of death throughout the world. The coronary vessel system is a special part of the circulation since there is a continuous change in blood flow, perfusion pressure and shear rate during each cardiac cycle. It is also the place of the narrowest capillaries in the human body, therefore the role of rheological alterations may be of greater importance than in the other parts of the circulatory system.

During the past decades, our group has investigated hemorheological parameters (HP) in over 1,000 patients diagnosed with various forms of ischemic heart disease (IHD). In one prospective study, we measured the HP of patients with acute coronary syndrome (ACS). On admission, all examined variables were significantly worse than those of control subjects. During the hospital phase, some of the HP showed further deterioration, and HP remained in the pathologic range during the follow-up period. In another study, we showed that HP are in close correlation with the severity of coronary artery disease. In patients treated with percutaneous coronary intervention, changes in HP were very similar to those observed in subjects with ACS. In a recent study, we analyzed HP in patients undergoing CABG surgery. Our data suggest a hemorheological advantage of off-pump surgery. In another study low Hct/WBV ratio can be regarded as a risk factor of cardiac death in IHD. Our data indicate that rheological parameters are significantly altered in patients with IHD: the extent of the alterations is in excellent correlation with the clinical severity of the disease. Our findings prove that HP play a critical role in the pathogenesis of myocardial ischemia.

In recent *in vitro* and *in vivo* studies we have investigated the effects of red wine on hemorheological parameters. Our results show that moderate red wine consumption has beneficial effects on hemorheological parameters which may contribute to the French paradox.

Keywords

ischemic heart disease; acute coronary syndrome; percutaneous coronary intervention; coronary artery bypass grafting; red wine; ethanol; hematocrit per blood viscosity ratio

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1. Introduction

Cardiovascular diseases (CVD) are the leading cause of death and disability among the adult and elderly population in the developed countries as well as in many developing nations. Although mortality rates have decreased in the developed areas, CVD related deaths have risen in the developing countries due to the rapidly increasing level of urbanization and changed lifestyle [26].

Ischemic heart disease (IHD) is the most common cardiovascular related death in the industrialized world. In most cases IHD is caused by coronary artery disease (CAD), originating mainly from atherosclerosis. Both hemodynamic and hemorheologic factors are of crucial importance to consider in the coronary circulation, since there is a continuous change in blood flow, perfusion pressure and shear rate during each cardiac cycle [22]. The myocardium is the highest oxygen consuming organ per one gram of tissue. The oxygen extraction at rest is very high and no further increase is possible, thus only the increased coronary perfusion can maintain the higher oxygen delivery to the myocardium. The coronary system has the narrowest capillaries in the human body with their diameter as small as 3-5 μm , therefore alteration of hemorheological parameters may early lead to impaired microcirculation [22].

The so called “classical” cardiovascular risk factors are well known, but only about half of the cardiovascular events can be explained by the presence of them. Several studies – such as the *Framingham Study*, the *Monica Project* and the *Physicians’ Health Study* – have shown that hemorheological parameters are primary and independent cardiovascular risk factors [3] [5][10][11][12][17][25]. Experimental, clinical and prospective epidemiological studies have provided convincing evidence that atherosclerotic and thrombotic states are associated with alterations in hemorheological and hemostatic profiles [7][9][13][19][20][24]. The fact that rheological alterations appear prior to the development of vascular lesions suggests that they might have a significant role in the pathogenesis of atherosclerosis [22][23].

2. Methods

All of our studies were approved by the Ethics Committees of the study sites (in most cases by the Regional Ethics Committee of the University of Pecs) and written informed consent was signed by all subjects prior to blood donation.

For hemorheological measurements Hevimet 40 capillary viscometer [7], Myrenne MA-1 Aggregometer, LORCA ektacyto- and aggregometer, Carat FT-1 filtrometer [13] and CARAT Tx4 platelet aggregometer [2][4] instruments were used. For detailed descriptions of methods see the cited articles.

3. Results and discussions of the presented studies

3.1. Chronic ischemic heart disease (IHD)

As mentioned in the introduction, hemorheological parameters are independent risk factors in CVD and play an important role in the coronary circulation, thus our working group has thoroughly investigated the relation between hemorheological parameters and IHD.

In an early study, our group evaluated the hemodynamic and hemorheologic parameters of patients with IHD and healthy controls at rest and peak exercise. The data of 377 patients with IHD and 59 healthy controls at rest; and 32 patients with IHD and 20 healthy volunteers during exercise stress test were analyzed. Cardiac output, cardiac index (CI), whole blood (WBV) and plasma viscosity (PV), fibrinogen level were determined, and – a new index - the circulatory index ($CRI = CI/WBV$) was introduced in order to characterize the hemodynamic and hemorheologic state of the circulation with one formula. At rest all the measured hemorheological parameters were significantly higher in patients with IHD than in healthy controls, while central hemodynamic parameters did not differ significantly from the control group. During exercise, all the measured hemorheological parameters significantly increased in patients with IHD, while this phenomenon was much less expressed in healthy controls. CRI was significantly lower in IHD patients compared to healthy controls both at rest and peak exercise, and was more sensitive than hemorheologic or hemodynamic parameters alone.

These findings underline the pathophysiological role of these parameters as important risk factors in cardiovascular diseases [20].

In another study the correlation of hemorheological parameters and the severity of CAD was examined. The data of 109 patients undergoing coronary angiography and 59 healthy controls were analyzed. Patients were classified into three groups according to their coronary vessel state based on the coronary angiogram: *Group 1*: positive non-invasive tests without significant CAD, *Group 2*: single vessel disease, *Group 3*: multi-vessel disease. Hematocrit (Hct), fibrinogen level, PV, WBV, cardiac output, CI were measured and CRI was calculated. All the hemorheological parameters were higher and CRI was lower in CAD-patients than in healthy controls. Fibrinogen and PV were significantly elevated in *Group 2 and 3* compared to *Group 1*. Hct and WBV were significantly increased in *Group 3* compared to *Group 1 and 2*. CRI was significantly decreased in IHD patients, and it was also lower in *Group 3* than in *Group 2*.

These findings indicate that hemorheological parameters are important risk factors of IHD, and also that they can change along with the severity of the coronary artery disease [7].

3.2. Acute coronary syndrome (ACS)

ACS is the acute imbalance of oxygen supply and demand of the myocardium, caused by the sudden decline of coronary blood flow. Mortality and morbidity of ACS are substantial, therefore ACS has considerable medical, social and economic importance all over the world [13].

Our group studied the changes of hemorheological parameters of 22 patients during the hospital phase of acute myocardial infarction (AMI). Hct, WBV, PV, fibrinogen level and RBC count were measured after the admission to hospital, after the beginning of the mobilization and before the discharge from the hospital. In men significantly poorer starting values were measured than in women and the deterioration of these parameters in this group was also more pronounced [19].

In another study we evaluated the hemorheological effects of a nonionic block copolymer surfactant, *RheothRx* Injection, on the hemorheological parameters in patients with AMI both *in vitro* and *in vivo*. Previous studies had reported that hemorheological parameters, such as RBC aggregation in AMI are significantly worse than those of healthy persons, moreover these parameters showed even a further deterioration during the hospital phase of AMI.

In the *in vitro* study blood samples from 24 patients with chest pain were obtained within 12 hours from hospital admission and if AMI was verified, a second sample was collected 48 hours later. Different concentrations of *RheothRx* were added to blood and RBC aggregation was determined. The results indicated a significant concentration-related decrease of RBC aggregation in both groups in the admission, and in the 48 hour AMI samples.

In a substudy for CORE Trial, the hemorheological effects of *RheothRx* infusion were investigated. Seven patients admitted with AMI and randomized for CORE Trial were involved. Besides standard infarction therapy they received various doses and duration of *RheothRx* infusion. The blood samples were collected after admission, at 12, 24, 48 hours, at day 8 and 35. PV, WBV, RBC aggregation and fibrinogen level decreased during and after the administration of *RheothRx*, but after 2–8 days their values returned to the baseline level. These findings indicate that *RheothRx* can significantly reduce RBC aggregation and other hemorheological parameters, and thus suggest its potential usefulness in clinical states associated with increased RBC aggregation [21].

Later in another prospective study our aim was to investigate almost all hemorheological parameters and the efficacy of the platelet aggregation inhibitor therapy in ACS. 157 patients with ACS and 68 healthy subjects were involved. Fibrinogen level, PV, WBV, RBC aggregation, RBC deformability by filtration method and platelet aggregation were measured after admission, on the 2nd and 6th days, at 1st, 6th and 12th months.

All the measured hemorheological parameters were significantly higher in patients with ACS after admission compared to the control subjects and almost all of them remained in the pathologic range until discharge. Some of the hemorheological parameters showed a slight improvement after 1 month, but Hct and WBV were higher than those after admission and of control subjects. After 6th and 12th months these parameters showed a small, but significant increase.

The results were also evaluated in the respect of the presence of classic cardiovascular risk factors. Pathologically altered hemorheological parameters could be observed in patients with the presence of classic cardiovascular risk factors and after elimination of them hemorheological parameters significantly improved. During the follow-up serum lipid levels and hemorheological parameters were significantly impaired in those subjects with recurring myocardial ischemia and AMI.

On admission antiplatelet therapy was efficient in about 50% of the treated patients, while at discharge 20% improvement of the effectiveness could be seen. Further increase could be detected after one month compared to the admission values.

Our results support the role of abnormal hemorheological parameters in the development of myocardial ischemia and draw attention to the rheological risk of these patients. The results of platelet aggregation measurements show the insufficiency of antiplatelet therapy at some cases and confirm the importance of guided secondary prevention [13].

3.3. Percutaneous coronary intervention (PCI)

PCI is a frequently used method in the treatment of coronary artery disease. The high reocclusion and/or restenosis rate is a main challenge and the mechanism of these processes is still only partially understood. In a study we analyzed the hemorheological and oxygen free radical associated changes during and after PCI.

Nineteen patients undergoing PCI were examined. Blood samples were taken before PCI, 30 minutes and 1, 2, 5 days and 1, 6 months after the procedure. Hct, plasma fibrinogen level, PV, WBV were measured and corrected WBV value was calculated. Oxidative stress and the changes in the antioxidant system were investigated.

Plasma fibrinogen concentration and PV increased significantly during the 1st and 2nd day after PCI. Plasma fibrinogen level returned to the baseline in the 1st month, but in the 6th month it was significantly higher. WBV gradually increased up to the six-month check-up visit. Corrected WBV was significantly elevated on the fifth day and one month later. Superoxide production showed an increasing tendency and was elevated after the 1st day and remained high during the following days. Antioxidant enzymes showed significantly increased activities during the hospital phase and at the end of the first month.

Our findings indicate that PCI may cause significant changes in the hemorheological and free radical associated parameters which can affect the final outcome of this intervention [8].

3.4. CABG surgery

CABG has become a well-established surgical treatment for coronary revascularization. Currently two methods are available: 1) *on-pump* CABG, where the heart is stopped and the circulation is maintained by the heart-lung machine; 2) *off-pump* CABG, in which case the operation is performed on beating heart – being more physiological than using an extracorporeal perfusion.

In our investigation we aimed to compare the two methods in a hemorheological aspect. Blood samples were taken from patients undergoing on-pump or off-pump CABG upon arrival to the operating theatre, after 20 and 40 minutes during the operation, after closing the thorax, on the 1st and 2nd postoperative days, and during the 2nd and 6th month control check-ups. Hct, PV, WBV and RBC aggregation decreased significantly during the early phase of the surgery, started to recover during the postoperative period, and reached the baseline values by the 2nd and 6th month control check-ups. These parameters were significantly lower at 20 and 40 minutes in the *on-pump* group. Scanning electron microscopy showed damaged and malformed RBCs in the samples from the *on-pump* surgeries. RBC deformability measured by ektacytometry showed significant difference neither during the surgeries, nor between the two groups. On the other hand filtrometry showed deformability impairment during *on-pump* surgery. Platelet aggregation decreased

during *on-pump* surgery and by the end of it, it showed a great fall. In the *off-pump* group platelet aggregation did not change during the operation, but decreased significantly by the end of it.

Our results show that changes of hemorheological parameters and RBC damage are less pronounced in case of *off-pump* technique, which seems to be favorable from a hemorheological point of view [14].

3.5. Red wine and hemorheology

Epidemiological evidences indicate that moderate red wine consumption reduces the risk of cardiovascular diseases (French paradox). This favorable effect of red wine can be due to its alcoholic and phenolic components. In *in vitro* and *in vivo* studies we have investigated the effects of red wine and both of its components on the hemorheological parameters.

In the **first *in vitro* experiment** blood was drawn from thirteen healthy volunteers and mixed with red wine to simulate 0.1%, 0.3% and 1% blood alcohol concentrations. Other samples were treated with equivalent amount of AFRW. Erythrocyte aggregation was dose dependently inhibited by both red wine and AFRW. Red wine caused a stronger inhibition than AFRW, which became significant at the highest concentration. Erythrocyte deformability of healthy volunteers did not change significantly for any concentrations of red wine or AFRW [15].

In the second phase the protective effect of red wine or AFRW was investigated under PMS induced oxidative stress conditions, which causes RBC deformability impairment. Blood samples from seven healthy subjects were pretreated with red wine or AFRW (0.3%). The deformability impairment caused by PMS was significantly reduced by 0.3% concentration of AFRW, while red wine had no effect [15].

In the **second *in vitro* study** the effect of ethanol on RBC aggregation and deformability was investigated. Blood samples from seven healthy volunteers were mixed with 96% ethanol to gain 0.25%, 0.5%, 1% and 2% alcohol concentrations. RBC aggregation was dose dependently decreased which explains why red wine decreased RBC aggregation more than AFRW in our previous experiment.

The effect of ethanol on RBC deformability was studied in two ways: 1) ethanol was added directly to the whole blood and measured in an ethanol-free LORCA medium; 2) addition of ethanol only to the LORCA medium (ethanol concentration range: 0.25-6%). In the *first* case no changes were found, but in the *second* version RBC deformability dose-dependently increased. This indicates that ethanol improves RBC deformability in a reversible manner [16].

The deformability impairment test from the first study was repeated with ethanol. Ethanol significantly enhanced the negative effect of PMS, which explains why no changes were observed in the first study, when red wine and PMS were added and incubated together. If PMS+blood incubated and measured in an ethanol containing LORCA medium, the PMS-induced deformability impairment was decreased [16].

In a **controlled prospective human study** we aimed to investigate the effects of 3-week red wine consumption on hemorheological parameters in thirty-nine healthy volunteers. They were assigned into two groups: the *control group* drank water, while the *red wine group* consumed 2 dl of red wine each day at dinner for 3 weeks. Hct and PV were not affected. WBV remained unchanged in *controls*, but it considerably decreased in the *red wine group* compared to the 3-week *control group*, while Hct/WBV ratio became significantly higher in the *red wine group* compared to the *control*. RBC aggregation significantly decreased in the *red wine group* and became significantly lower compared to the 3-week *controls*. Red wine significantly increased RBC deformability at high shear stress.

Our results show that moderate red wine consumption has beneficial effects on hemorheological parameters which may contribute to the French-paradox [18].

3.6. Hard clinical end-point studies: Hct/WBV ratio

In healthy subjects Hct/WBV ratio reliably reflects rheological oxygen carrying capacity of blood. Oxygen binding capacity linearly, viscosity exponentially increases with Hct. Increase in Hct results in an inverted U-shape curve of values of Hct/WBV [1].

In a retrospective study our group aimed to investigate the predictive power of Hct/WBV ratio to assess mortality risk of CAD. Seventy-eight patients with CAD were followed for a mean of 8.9 years. Patients who died due to cardiac cause during the follow up were classified as *group C* while others who died due to non cardiac cause or were alive as *group NC*. Hct/WBV ratio was significantly lower in *group C* comparing to *group NC*. Kaplan–Meier survival analysis showed the impact of fibrinogen and Hct/WBV ratio on cardiac mortality. Receiver operating characteristic curves proved that Hct/WBV ratio has the power to predict cardiac death. Hct/WBV ratio showed significant negative correlation with the frequency of hospital admissions. According to this study low Hct/WBV ratio can be regarded as a bad prognostic factor of cardiac death in CAD [6].

4. Conclusion

Our results suggest that rheological parameters play an important role in the coronary circulation and in the development of myocardial ischemia and can have a role also in cardioprotection. Clarifying their exact role in the pathomechanism of ischemic disorders may help in the development of novel, more potent cardiovascular agents.

Acknowledgements

We thank to Jeno Ernst †, Jozsef Tigyi, Miklos Tekeres, Tibor Javor †, Gyula Mozsik, Balazs Sumegi, Kalman Hideg, Erzsebeth Roth, Lajos Bogar, Lajos Kollar, Tamas Habon, Katalin Koltai, Tamas Alexy, Barbara Sandor and all the technicians.

Abbreviations (in the order of appearance)

CVD	cardiovascular disease
IHD	ischemic heart disease

CAD	coronary artery disease
CI	cardiac index
WBV	whole blood viscosity
PV	plasma viscosity
CRI	circulatory index
Hct	hematocrit
AMI	acute myocardial infarction
ACS	acute coronary syndrome
RBC	red blood cell
PCI	percutaneous coronary intervention
CABG	coronary artery bypass grafting
AFRW	alcohol-free red wine extract
PMS	phenazine methosulfate

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