

Complications After Coronary Artery Bypass Grafting.

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Abstract

Complications following coronary reperfusion, a process where blood flow is restored to previously ischemic myocardium, are largely attributed to reperfusion injury. The revealed changes in the iron pool parameters in patients with coronary artery bypass grafting indicate the effect of iron on the activity of oxidative processes, predisposing to the development of complications. This is confirmed by the nature of the change in the concentration of nitrites and nitrates, which depended on the severity of intraoperative hemolysis, indicating the effect of hemolysis products, including iron, on the processes of formation and utilization of nitric oxide. According to the degree of intraoperative hemolysis (IOH), determined by the level of free hemoglobin, Hbfree, patients after coronary artery bypass grafting (CABG) were divided into 3 groups: group 1 - without IOH (Hbfree \leq 0.1 g/l), n=43, group 2 - with low IOH (lIOH) with Hbfree >0.1 g/l and <0.5 g/l, n=42, group 3 with high IOH (hIOH) - Hbfree \geq 0.5 g/l, n=38. The study of changes in iron levels was carried out on the basis of determining the content of serum iron (Fesvv.), transferrin (Tr), total iron-binding capacity of serum (TIBC), latent iron-binding capacity of blood, LIBC, ferritin (F) in blood plasma. The NO level was determined by the total concentration of nitrites and nitrates in blood plasma – NOx.

Keywords: coronary bypass surgery; iron; nitric oxide; CABG; IOH, TIBC

Introduction

Relevance

A key pathophysiological mechanism underlying this injury is oxidative stress. This involves an imbalance between the production of reactive oxygen species (ROS) and the body's ability to neutralize them. The restoration of blood flow after a period of ischemia leads to a surge in ROS production, exceeding the antioxidant capacity of the tissues. This oxidative stress damages cellular components, including lipids, proteins, and DNA, contributing to inflammation and cell death. Furthermore, the process also involves reactive nitrogen species (RNS), such as nitric oxide (NO). While NO has important physiological roles, excessive production or altered NO metabolism during reperfusion can contribute to nitrosative stress, exacerbating the damage caused by oxidative stress. The interplay between ROS and RNS in the context of reperfusion injury is complex and remains an area of ongoing research. Nitrogen oxide is one of the key signal molecules in vascular homeostasis, which has in small concentrations due to its basal products, the ability to inhibit platelet aggregation, proliferation of smooth muscle cells, as well as their migration, reduce the expression of cellular adhesion molecules, suppress the migration of the activated Leukocytes in the intimacy of the arteries [13, 16]. The playotrophic nature of the effects of NO proves the importance of its adequate products for the prevention of the development of cardiovascular complications of the

operation. NO deficiency is the cause of endothelial dysfunction, manifested by excessive vasoconstriction, proadgesic and pro-aggregation effects in relation to platelets and leukocytes, contributes to the development of oxidative stress, activation of inflammation and proapoptotic effects. The transformation of positive NO effects into negative is facilitated by a significant increase in the products of active forms of oxygen that convert NO into highly toxic peroxinitrite (ONOO-) [14].

Cardiopulmonary bypass (CPB), while essential for facilitating coronary artery bypass grafting (CABG) surgery on a "bloodless" heart, introduces the risk of several complications, many with a vascular etiology, including myocardial infarction, arrhythmias, heart failure, and acute cerebrovascular events. The pathogenesis of these complications is complex but often involves disruptions in nitric oxide (NO) homeostasis. Changes in NO balance can trigger a cascade of pathophysiological mechanisms, including oxidative stress, inflammation, and thrombosis [3]. While CPB optimizes surgical conditions by maintaining organ perfusion, several intraoperative factors contribute to hemolysis. These include the inherent hemodynamic stresses of the CPB circuit: turbulent blood flow, high shear stress, fluctuating pressures (both positive and negative), and interfacial forces such as surface tension. These mechanical stressors damage red blood cells, leading to hemolysis. Furthermore, patient-specific risk factors, such as

smoking and alcohol consumption, significantly augment the susceptibility to CPB-induced hemolysis. The resulting increase in free hemoglobin and other breakdown products can further exacerbate oxidative stress and contribute to post-operative complications. Therefore, understanding and mitigating the effects of CPB-induced hemolysis and the subsequent impact on NO balance are crucial for minimizing post-CABG morbidity and mortality [17].

The use of the AC apparatus, in the contours of which the conditions for hemolysis are created, indicates a possible change in the iron pool released from the hem of free hemoglobin [4]. As you know, an increase in free hemoglobin, hem and iron in blood serum is a risk factor for cardiovascular diseases [9, 10]. At the same time, the level of free hemoglobin and iron - the hem component can influence the metabolism of nitrogen oxide and its level of metabolites. The inflammatory process initiated by the fragments of the cellular membranes of red blood cells can also contribute to a change in level of NO due to the activation of inducible NO-synthase [14]. It is obvious the need to study violations of the homeostasis of iron and nitrogen oxide, the marker of which is the content of its stable metabolites in the blood of nitrites and nitrates (NOx) in patients who underwent the Operation of KSh in the IR.

Aim

The purpose of the study was to identify changes in the level of iron and nitrogen oxide in patients after coronary artery bypass grafting, depending on the degree of intraoperative hemolysis.

Materials and Research Methods

Research was carried out in patients after coronary artery bypass grafting operation: 123 people with varying degrees of IOH (group 1-3). The study corresponded to the principles of the Helsinki declaration of the World Medical Association "Ethical principles of scientific medical research with human participation" and approved by ethical committees of UO "GrSMU" and healthcare institutions "Grodno Regional Clinical Cardiological Center" [<http://www.med-pravo.ru/archives/helsinki.txt>], informed consent received from all patients. Patients of all groups were comparable by age (63.0 (58; 67) years, $p > 0.05$) with the possession of men's sex (81.3 %, $p > 0.05$). According to the degree of intraoperative hemolysis (IOH), determined by the level of free hemoglobin, Hbfree, patients after coronary artery bypass grafting (CABG) were divided into 3 groups: group 1 - without IOH (Hbfree ≤ 0.1 g/l), n=43, group 2 - with low IOH (lIOH) with Hbfree > 0.1 g/l and < 0.5 g/l, n=42, group 3 with high IOH (hIOH) - Hbfree ≥ 0.5 g/l, n=38. [11, 12]. Assessment of the degree of IOH was carried out according to the level of free hemoglobin using the analyzer Nemocue Plasma/Low HB, Sweden [15]. The groups did not differ in age, gender, the number of myocardial infarctions in history ($P > 0.05$), the frequency of ischemic cardiomyopathy ($P > 0.05$) and the frequency of heart rhythm disturbances ($P > 0.05$). Blood took from the AC apparatus at the beginning, before the coronary shunt and at the end of the AC after its imposition, the determination of iron indicators (serum iron, the total iron-binding ability of blood serum, transferrin, ferritin) was carried out on the automatic biochemical analyzer of the Mindray BS-200 with the use of reagents (Diassist RB). The concentration of iron in the blood serum was determined by spectrophotometrically with a wavelength of 560 nm using ferrosine, which forms a painted complex with Fe^{2+} , which is determined after its restoration [18]. The total iron -binding ability of blood serum represents the largest amount of iron, which can attach transferrin to complete saturation [18]. The determination of changes in the content of transferrin in the blood serum is based on measuring changes in the optical density of the test sample

due to agglutination of latex particles of the reagent covered with antibodies, to transferrin, with a wavelength of 340 nm. The determination of the content of ferritin in the blood serum is based on a photometric definition of a change in the optical density of the examined sample due to agglutination of latex particles covered with antibodies to ferritin [5]. A study of the concentration of stable metabolites of nitrogen oxide - nitrites and nitrates [NOx] according to the generally accepted method using the cadmium and the Griss reagent by spectrophotometry [6]. Statistical processing of the received data was carried out using the Statistica 10.0 program. With the use of descriptive statistics for quantitative (method of Kruskalla-Uolis), the method of comparing dependent variables with the use of the Wilcoxon criterion. Data of descriptive statistics are presented in the form of ME (Q25; Q75), where the Median is a variable, Q25 is the value of the lower tenant; Q75 - the meaning of the upper apartment. Differences were considered reliable at $p < 0.05$.

Results and their Discussions

At the beginning of coronary shunting, the differences between the value of free hemoglobin in the studied groups were not noted, $p > 0.05$. After coronary shunting, there was an increase in free hemoglobin in the 2nd (with low IOH) and 3rd (high IOH) groups. At the same time, in a group with low IOH, the concentration of free hemoglobin increased by 3.3 (2.5; 5) times ($p < 0.001$), in a group with a high IOH 12 (7; 15) times ($p < 0.001$). In the 1st group, changes in the level of free hemoglobin in blood plasma were not observed, indicating the absence of IOH ($P > 0.05$). The study of iron indicators in patients with the operation revealed the presence of changes. In a group of patients without IOH, compared with the initial level, there are no excretions of iron indicators at the end of the coronary artery bypass grafting ($P > 0.05$), but there was a change in indications in patients with IOH. Changes in indicators, in comparison with their initial values, were more significant in patients with high IOH - an increase in $[Fe^{2+}]$ in blood plasma by 91.9 (4.8; 117.5) %, $p < 0.001$, ferritin- by 165.9 (135.1; 212.6) %, $p < 0.001$ and reducing the level. Transferrin - at 22.0 (19.7; 30.2) %, $p < 0.001$, and Latent iron-binding ability - at 45.0 (31.3; 68.7) % $P < 0.001$. In blood plasma by 22.9 (14.58; 35.48) % ($p < 0.001$), ferritin - by 37.5 (25.06; 45.9) % ($p < 0.001$), as well as a decrease in the level of transfer - by 8.1 (3.8; 13.8) %, $p < 0.05$ and falsess - by falsess on 9.7 (4.4; 24.9) %, $p < 0.001$. A decrease in the content of the transport protein of transferrin in patients with high IOH may be due to its binding to free iron. Excessive saturation of transferrin with iron leads to its rapid elimination from vascular channel. In turn, an increased saturation of transfer, as a result of an increase in free iron in blood plasma in patients with high IOH, indicate a decrease in Latent iron-binding ability. An increase in ferritin is consistent with the literature on the nature of the change in the depositing iron of protein in patients with sickle cell anemia [17 Hebbel R.P., 2004]. At the end of the coronary shunting, a decrease in $[NOx]$ in the blood plasma of patients of the 1st, 2nd and 3rd groups was noted. Compared to the initial level, the most significant decrease in $[NOx]$ was observed in patients with a high IOH group - by 43.0 (19.4; 58.9) %, $p < 0.001$, while in groups without IOH and with low IOH the decrease was less pronounced (by 12.2 (7.63; 17.001 and 13.8 (9.04; 19.1) %, $p < 0.001$, respectively, a week after the coronary shunting, compared with $[NOx]$ at the end of the operation, an increase in $[NOx]$ was noted in patients of the first group of 1.39 (1.18; 1.66), $p < 0.001$ times, groups with low IOH - 1.69 (1.36; 2.23) times, $p < 0.001$.

So, an increase in the level of serum iron, a decrease in transferrin and Latent iron-binding ability and an increase in the level of ferritin in patients with a high degree of IOH indicate an increase in plasma of blood levels of free iron, contributing to the development of complications after the coronary

shunting due to activation of oxidative and inflammatory reactions [4, 9]. The studies have shown a decrease in [NOx] in blood plasma at the end of the coronary shunting and its increase within 5-7 days after surgery. The severity of the noted changes depended on the degree of IOH. The decrease [NOx], which is mostly noted in the group with high IOH, may be the result of excessive consumption of NO in the reaction of mutual forms with active forms of oxygen, as the initiators of oxidative stress, in particular, in the reaction of interaction with superoxidanine and its transformation into peroxinitritis, as well as the interaction of ne with oxy and oxy. The hemoglobin deox forms with the formation of metghemoglobin and nitrosyl hemoglobin, respectively [13, 14]. Excessive NO consumption in patients with high IOH can contribute to a decrease in antiadhesive, antiaggregation and antiinflammatory properties of the vascular wall, which predisposes to the development of complications caused by thrombosis. It is important to note that free hemoglobin circulating in the blood, effectively consuming no, reduces its level in blood plasma, contributing to the processes of NO-dependent vasoconstriction and to the develop.

Conclusions

1. Elevated Plasma Nitrites/Nitrates and iNOS Activity: The observed increase in plasma nitrite and nitrate concentrations 5-7 days post-CABG in patients with intraoperative hemolysis (IOH) suggests heightened inducible nitric oxide synthase (iNOS) activity. This enzyme, induced under inflammatory conditions, generates large amounts of nitric oxide (NO). The delayed increase implies a sustained inflammatory response following the surgical procedure.
2. Peroxynitrite Formation and Cellular Damage: Excessive NO production via iNOS contributes to the formation of peroxynitrite (ONOO⁻), a potent oxidant and nitrating agent. Peroxynitrite's nitrosylating effects on proteins cause damage to cell membranes and induce cytotoxic effects within vascular and myocardial cells. This cellular damage triggers a systemic inflammatory response, increasing the risk of thrombosis and ultimately contributing to post-operative complications and adverse outcomes.
3. Paradoxical NOx Changes during CPB: CABG surgery performed with cardiopulmonary bypass (CPB) results in a complex and paradoxical pattern of nitrite/nitrate (NOx) levels. An initial decrease in plasma NOx is observed immediately before the conclusion of surgery, potentially reflecting NO consumption in the context of the surgical stress response and/or altered NO metabolism during CPB. This is followed by a delayed increase 5-7 days post-operatively, consistent with the sustained inflammatory response and iNOS activation described above. The initial decrease and subsequent increase highlight the dynamic nature of NO metabolism during and after CPB.
4. Hemolysis and Altered Iron Metabolism: Intraoperative hemolysis, the destruction of red blood cells within the CPB circuit, leads to alterations in plasma iron parameters, characterized by an increase in free, non-transferrin bound iron. This increase in free iron represents a significant factor as it can contribute to oxidative stress and cellular damage.
5. Correlation between Hemolysis, Free Iron, and NOx: A direct correlation exists between the degree of intraoperative hemolysis, the level of free plasma iron, and the concentration of nitrites and nitrates (NOx). This relationship suggests that hemolysis-induced increases in free iron contribute to the production of reactive nitrogen species, further potentiating oxidative stress and potentially exacerbating the inflammatory response. Consequently, these combined effects of iron dysregulation and altered NO metabolism resulting from hemolysis significantly contribute to the development of post-CABG complications.

These revised statements provide a more nuanced and scientifically accurate description of the findings, emphasizing the complex interplay between oxidative stress, nitrosative stress, hemolysis, and the development of post-operative complications after CABG.

Abbreviations:

CABG: coronary artery bypass grafting

IOH: intraoperative hemolysis

CAD: coronary artery disease

Conflict of Interest:

The authors declare that there are no conflicts of interest.

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