Volume 4, Number 9, March 2016
ISSN: 2309-0901
http://cardioprogress.ru

International Heart and Vascular Disease Journal

Journal of the Cardioprogress Foundation

European clinical guidelines for cardiology 2015

The relationship between total epicardial fat volume and atrial fibrillation

Impaired regulation of genome stability may be the key mechanism of left ventricular hypertrophy development in arterial hypertension

The Ministry of Health of the Russian Federation
Russian Academy of Sciences
Russian Society of Cardiology
Foundation for the Advancement of Cardiology «Cardioprogress»

V INTERNATIONAL FORUM OF CARDIOLOGY AND INTERNAL MEDICINE

29–31 March, 2016
Moscow

- Global participation, with 3,000 delegates from Russia, Europe, Asia, Africa and the Americas
- Scientific programme includes plenary sessions, lectures, symposia, round tables discussions, interactive case studies and workshops
- Presentations from leading experts in Russia, Europe and the U.S.
- Collaboration with the European Society of Cardiology and the World Heart Federation
- Exhibition stands and participation from more than 60 pharmaceutical companies and manufacturers of medical equipment
- Exciting cultural programme to include the iconic sights, theatres and museums of Moscow
- Free Registration!

To register your place and for more information, please visit:
www.cardioprogress.ru

International Heart and Vascular Disease Journal
Journal of the Cardioprogress Foundation

The *International Heart and Vascular Disease Journal* is a peer-reviewed open access publication printed quarterly. The journal features original research articles, case reports, clinical reviews, editorials, and letters to the Editor. All published articles are freely accessible from the journal's website.

The publication of articles within the journal is free of charge for authors. Guidelines for authors on submitting manuscripts are available at: www.cardioprogress.ru

Contact details:
Cardioprogress Foundation and Editorial Office:
Room 213, Building 2, Prospect Gostinichny 6, Moscow 127106, Russia
Editorial Office tel.: (+7) 965 236 1600
Official website: www.cardioprogress.ru

Editorial correspondence should be sent to: Mehman Mamedov, Deputy Editor, editor.ihvdj@gmail.com
Articles for publication should be sent to: Anna Arteyeva, Associate Editor, submissions.ihvdj@gmail.com

Printed in Russia

International Heart and Vascular Disease Journal

Journal of the Cardioprogress Foundation

Volume 4, Number 9, March 2016

Contents

Editor's Welcome

Dear colleagues

In the 9th issue of the International Heart and Vascular Disease Journal, there are leading article, review, original articles and clinical case.

The leading article of the issue is dedicated to the review of five clinical guidelines published by the European Society of Cardiology, which include sudden death, management of patients with ACS and ventricular arrhythmias. This review analyses updates and changes of new issues of these guidelines, and it is has high practical interest and importance.

Review article of this issue presents the work of coauthors A.V. Starodubova and A.O. Kislyak. It is dedicated to particular features of coronary heart disease course in women. The review discusses gender differences in importance of risk factors, clinical course and prognosis. Authors highlight the importance of development of Russian criteria of high risk group formation, further investigation of coronary heart disease course features, and search for effective treatment.

In "Original articles" of the 9th issue section we published three papers. The first article discusses the results of a study dedicated to the search for correlation between epicardial fat pad volume and the risk of atrial fibrillation development. Using correlation analysis of body mass index values, epicardial fat pad volume assessed by MRI, and transthoracic echocardiography, Egyptian researchers for one more time proved the hypothesis about local pathogenic influence of epicardial fat pad on arrythmogenic mechanisms that lead to atrial fibrillation. Another article demonstrates the results of comparative study that investigated different treatment regimens efficacy on vascular rigidity characteristics in male patients with arterial hypertension. The third article of Russian scientist is dedicated to investigation of PPAR, PARP, PARG and NOS3 genetic polymorphisms association with left ventricular hypertrophy in patients with arterial hypertension. The group of authors, that included genetics, demonstrated that one of mechanism responsible for left ventricular hypertrophy in patients with arterial hypertension can be impaired balance of processes that lead to genome destabilization/stabilization.

The "Clinical case" section describes unique clinical case of Gitelman's syndrome with severe hypokalemia and pseudoischemic ECG changes. This publication presents a short review about this tubulopathy and highlights clinical significance of possible difficulties of these patients management for cardiologist.

I invite everybody to collaborate with the journal. We are waiting for your original papers, review articles, discussions, opinions about problems, treatment and prophylaxis recommendations.

Rafael G. Oganov

Editor-in-Chief

President of the "Cardioprogress" Foundation

International Heart and Vascular Disease Journal
Volume 4, Number 9, March 2016

Journal of the Cardioprogress Foundation

LEADING ARTICLE

European clinical guidelines for cardiology 2015

Kanorsky S.G.*

Kuban State Medical University, Krasnodar, Russia

Autor:

Sergey G. Kanorsky, MD, Professor, Head of the Department of Therapy № 2 of Faculty of Advanced Training and Professional Retraining of Specialists of Kuban State Medical University, Krasnodar, Russia

Summary

The review presents the main provisions of 5 clinical practice guidelines of the European Society of Cardiology, published in 2015: guidelines for the management of acute coronary syndromes in patients presenting without persistent ST-segment elevation, guidelines for the management of patients with ventricular arrhythmias and the prevention of sudden cardiac death, guidelines for the diagnosis and management of pericardial diseases, guidelines for the diagnosis and treatment of pulmonary hypertension, guidelines for the management of infective endocarditis. This review focuses on changes introduced in the new version of the guidelines.

Keywords

Clinical practice guidelines, acute coronary syndrome, sudden cardiac death, pericarditis, pulmonary hypertension, infectious endocarditis.

In 2015 the European Society of Cardiology published 5 new clinical guidelines developed by working groups of experts and reviewers. These guidelines cover the following topics: acute coronary syndrome without persistent ST segment elevation (ACS| ST), ventricular arrhythmias (VA) and sudden cardiac death (SCD), pericardial diseases, pulmonary hypertension (PH), infective endocarditis (IE). These guidelines summarize all modern scientific data related to the topics of interest, thus being a valuable educational source for clinical practitioners.

Guidelines for ACS/ST treatment

Guildelines for ACS/ST treatment have been prepared by the group of experts of the European Society of Cardiology lead by Roffi M., et al [1]. Previous guidelines were published in 2011. Main changes of the new version are related to ACS/ST diagnostics, cardiac rhythm monitoring, risk stratification and treatment strategy.

In patients with suspected ACS/ST it is necessary to check the levels of cardiac troponin using sensitive or high-sensitivity test and obtaining the results

*Corresponding author: Tel +79172458474. E-mail: kanorskysg@mail.ru

during 60 minutes after it. It is recommended to use high-sensitivity troponin test during 0h/1h timing if it is available in addition to the 0h/3h fast diagnostic algorithm that had been proposed in the previous edition of guidelines, thus deciding if a patient should be admitted to hospital. Additional testing in 3-6 hours is indicated if the results of first two troponin levels tests are not definitive and if clinical manifestations still allow to suspect ACS.

Continuous heart rhythm monitoring is recommended until the ACS/ST diagnosis would be established or excluded. Patients with ACS/ST should be admitted to the intensive care units. Heart rhythm monitoring for 24h or before percutaneous coronary intervention (PCI) should be considered for ACS|ST and low risk of dangerous arrhythmias. Heart rhythm monitoring for ⩾ 24h is indicated for patients with ACS|ST and moderate or high risk of arrhythmias. If manifestations of continuous ischemia are not present heart rhythm monitoring can be necessary only in some patients with unstable angina, so in case of negative high-sensitivity troponin test results.

Guidelines include new criteria of risk stratification in patients with ACS/ST that allow to choose treatment strategy and timing of invasive intervention. Presence of very high risk criteria: hemodynamic instability/cardiogenic shock, ongoing chest pain refractory to medical treatment, life-threatening arrhythmias/cardiac arrest, mechanic complications of myocardial infarction, acute heart failure, dynamic changes of ST-T wave in electrocardiogram (ECG), consider performing of coronary angiography and myocardial revascularization during 2 hours after admission. High-risk criteria (rise in cardiac troponin levels, dynamic ST- or T-wave changes, GRACE (Global Registry of Acute Coronary Events) score >140, require invasive approach during up to 24 hours. Intermediate risk criteria: diabetes mellitus, glomerular filtration rate <60 ml/min/1.73 m^2, early post-infarction angina, LVEF <40% or congestive heart failure, prior PCI or coronary artery bypass grafting, GRACE risk score >109 and <140), consider maximum 72 h window from admission to coronary angiography. Conservative treatment is recommended in case of absence of all above-mentioned risk criteria

This document proposes transition from femoral access during coronary angiography to radial access in the hospitals where the treatment of patients with ACS is performed. It is recommended to use new-generation drug-eluting stents for PCI. Drug-eluting stents may be preferred comparing with bare-metal stents even in patients at high bleeding risk for whom short duration of dual antiplatelet therapy (30 days) is considered.

Guidelines for treatment of patients with VA and SCD prevention

Guidelines for VA and sudden cardiac death (SCD) were prepared by expert group of European Society of Cardiology and were endorsed by Association for European Paediatric and Congenital Cardiology (AEPC) [2]. Comparing with previous guidelines issued in 2006 new document includes updated information aimed to improve efficacy of SCD prevention in patients with VA. Cardiac diseases that lead to SCD the most frequently include channelopathies and cardiomyopathies, myocarditis and substance abuse in young patients and chronic degenerative diseases in older populations. For the first time DNA extraction and analysis has been recommended as a part of standard autopsy in order to determine the presence of channelopathies in sudden death cases.

Authors consider the leading role of detection of life-threatening VA in SCD prevention and propose several approaches for patients' examination. Standard 12-lead ECG registration is recommended in all patients with suspected VA (Class of recommendation I, level of evidence A). Ambolatory ECG screening is reasonable for arrhythmias diagnostics and detection, QT interval length estimation, ST segment deviation (I, A). Cardiac event recorders can be used in patients with sporadic symptoms that allow to suspect VA (I, B). It is reasonable to use implantable loop recorders in case of sporadic symptoms that are suspected to be related to arrhythmia and also if the connection of arrhythmia and symptoms cannot be established by conventional diagnostic techniques (I, B). Signal-averaged ECG can help to diagnose arrhythmogenic right ventricular cardiomyopathy (I B).

Exercise stress testing is indicated for adult patients with VA and intermediate/high probability of having coronary artery disease (CAD) (I, B), for patients with known or suspected exercise-induced VA (I, B) and can be considered in estimating response to medical or ablation therapy in patients with exercise-induced VA (IIa C).

Echocardiography allows estimation of left ventricle function and detection of structural heart disease for patients with suspected/known VA or for patients with the risk of developing serious VA/SCD (I, B). Exercise testing plus imaging is recommended

to detect silent ischaemia in patients with VA and intermediate probability of having CAD at whom ECG testing is less reliable (I B). Pharmacological stress test is reasonable to perform in patients with VA and intermediate probability of having CAD who are unable to perform physical exercise test (I B). Computer tomography or magnetic resonance imaging may be considered for patients with VA when echocardiography doesn't provide precise estimation of left and right ventricle function or cardiac structural changes (IIa, B).

Coronary angiography should be considered to prove or exclude significant obstructive CAD in patients with life-threatening VA or SCD survivors with intermediate or high probability of having CAD (IIa, C). Electrophysiological study is advised to patients who had myocardial infarction with symptoms reminding ventricular tachyarrhythmia (I, B), to patients with syncopes and suspected brady- or tachyarrhythmias (I, C). It can be recommended for the differential diagnosis of arrhythmogenic right ventricular cardiomyopathy and comparably benignant conditions like right ventricular outflow tract tachycardia or sarcoidosis (IIb, B).

Recommended device therapy for patients with VA includes implantable cardioverter defibrillators (ICD), subcutaneous implantable cardioverter defibrillators (SICD) and wearable cardioverter defibrillators. ICD are advised for secondary prevention of SCD and ventricular tachyarrhythmia treatment, primary SCD prevention in patients with severe left venricular dysfunction. New version of guidelines allows to consider SICD as alternative treatment of VA in young patients, people with difficult transvenous access or with infections. But this device is not suitable for patients who need bradycardia pacing or cardiac resynchronization therapy and also for patients with tachyarrhytmias that can be easily terminated by antitachychardia pacing. Wearable cardioverter defibrillators now can be used in patients with short-term risk of SCD for whom ICD are not suitable.

Catheter ablation should be considered in patients with continuous ventricular tachycardia or "electrical storms" because of myocardial scarring in case of CAD and repeated appropriate ICD shocks due to recurrent sustained ventricular tachycardia. Statement about making ablation after the first episode of sustained ventricular tachycardia in patients with CAD and ICD has been added to the new guideline.

Resynchronization therapy nowadays is recommended for primary SCD prevention in selected patients with sinus rhythm and NYHA functional class II/III and ambulatory class IV chronic heart failure.

This guideline contains separate table dedicated to treatment of patients with cardiomyopathy for SCD prevention.

Diagnostic criteria and guidelines for treatment of inherited primary arrhythmia syndromes were updated. It is recommended to use ICD implantation in patients with long QT syndrome who survived cardiac arrest, in the group of high risk prophylactic implantation of ICD can be considered. ICD implantation for secondary prevention is recommended in patients with short QT syndrome. ICD should be used in patients with Brugada syndrome or catecholaminergic polymorphic ventricular tachycardia who survived cardiac arrest. Differentiated pharmacological therapy (beta-blockers and I class antiarrhythmic drugs) also can be recommended for patients with these syndromes.

Guidelines for the diagnosis and management of pericardial diseases

Guidelines for the diagnosis and management of pericardial diseases have been prepared by the expert group of the European Society of Cardiology and have been endorsed by the European Association for Cardio-Thoracic Surgery [3]. Previous guidelines for this problem were published in 2004. These guidelines are particularly concentrated on diagnostics and treatment strategies in pericardial diseases.

Simple aetiological classification of pericardial diseases splitting them into infectious and non-infectious has been proposed. In developed countries, viruses and bacteria (mostly mycobacterium tuberculosis) are the most frequent causes of pericarditis, tumoral pericarditis and pericarditis related to systemic (usually autoimmune) disease occur more rarely. Classic pericardial symptoms include pericarditis, pericardial effusion, cardiac tamponade and constrictive pericarditis. Cardiac tamponade and pericardial effusion may occur in absence of pericarditis.

The diagnosis of acute pericarditis can be made with at least two following criteria: chest pain typical for pericarditis, pericardial friction rub, ECG changes – new expanded ST elevation or PR depression, pericardial effusion. Incessant pericarditis is defined as pericarditis lasting for more than 4 (up to 6) weeks but less than 3 months. Recurrent pericarditis characterized by the recurrence of pericarditis after a documented first episode of acute pericarditis and a symptom-free interval of 4–6 week. Pericarditis

without remission.and lasting more than 3 months is defined as chronic one.

Authors of recommendations listed predictors of poor prognosis of pericarditis. Major risk factors inclue fever >38 ºC, sub-acute onset, large pericardial effusion, cardiac tamponade, lack of reaction to aspirin or non steroid anti-inflammatory drugs (NSAID) after 1 week of administrations Minor risk factors include myopericarditis, immunodepression, trauma, oral anticoagulant therapy. When pericarditis is suspected, first stage of diagnostics requires assessment of inflammation markers – leucocytosis, C-reactive protein and others and markers of myocardial injury – cardiac troponins, creatine kinase, estimation of kidney, liver and thyroid function, chest X-ray, ECG registration, echocardiography. Second level of diagnostic is required in case of insufficient information value of the first stage and it can include computer tomography or MRI (magnetic resonance imaging) of the heart, pericardial fluid analysis in order to detect bacteria and tumor cells in case of large effusion not responsive to standard anti-inflammatory therapy. Additional diagnostic procedures aiming to define pericarditis etiology should be performed being based on clinical symptoms and presence of the high risk of poor outcome predictors.

Pericardial effusion is classified according with the mechanisms of its onset – acute, subacute or chronic, its size – mild (<10mm), moderate (10–20 mm) or large (>20mm), its distribution – circumferential or loculated, and composition – transudate or exudates. Etiologically it is classified to idiopathic, cancer, infectious, iatrogenic, and related to connective tissue diseases. Complex evaluation of possible pericardial effusion should include chest x-ray, inflammation markers assessment, transthoracic echocardiography, computer tomography or heart MRI in patients with loculated effusion, pericardial thickening and masses, as well as associated chest abnormalities.

The most frequent causes of cardiac tamponade are pericarditis, tuberculosis, iatrogenic causes, traumas and tumors. Echocardiography is the first choice visualization technique for evaluation of size, localization and grade of hemodynamic changes of pericardial effusion. If it was found, cardiac tamponade requires immediate pericardiocentesis or surgical drainage.

Constrictive pericarditis can occur after almost every pericardial disease, but it rarely follows recurrent pericarditis. Idiopathic constrictive pericarditis is the most common one, other frequent causes are viral infection, cardiac surgery, radiotherapy, connective tissue diseases, post-infectious causes not related to viral infections. Transthoracic echocardiography and chest X-ray are recommended for all the patients with suspected constrictive pericarditis. Computer tomography and heart MRI are indicated as second stage visualization techniques for evaluation of pericardial calcification, thickness and degree of extension. Heart catheterization is reasonable when non-invasive diagnostic techniques don't provide a definitive diagnosis of constriction.

Hospital admission is recommended for treatment of acute and recurrent pericarditis in patients with high risk (I B). Colchicine use (0.5 mg twice or once daily for patients < 70 kg or intolerant to higher doses) is recommended as first-line therapy for acute pericarditis as an addition to aspirin/NSAID therapy (3 months) and is also recommended for recurrent pericarditis (6 months therapy) (I A). Corticosteroids are not recommended as first-line therapy of acute and recurrent pericarditis (III C). Serum C-reactive protein levels can be used to determine treatment duration and to estimate response to therapy (IIa C). Aspirin, NSAID or colchicines are recommended for treatment of exudative pericarditis if pericardial effusion is associated with systemic inflammation (I C). Pericardiocentesis or surgical drainage are reasonable to use in case of cardiac tamponade, or symptomatic moderate/large cardiac effusion, non responsive to pharmacological therapy, or suspected unknown bacterial or tumoral etiology (I C). If etiology of pericardial effusion is defined, it is recommended to target the therapy (I C). Pericardiectomy is the main treatment of chronic constrictive pericarditis (I C). Pharmacological treatment of defined causes of pericarditis is recommended to prevent the progression of constriction (I C). Empiric anti-inflammatory therapy can be considered in case of transient or new diagnosis constriction when there are the evidences of concomitant pericardial inflammation (IIb C).

Guidelines for the diagnosis and treatment of pulmonary hypertension

Guildelines for the diagnosis and treatment of pulmonary hypertension (PH) have been prepared by the European Society of Cardiology and the European Respiratory Society [4]. Previous guidelines were published in 2009.

It is mentioned that PH can include multiple clinical conditions and be a complication of several cardiovascular and respiratory diseases. PH is defined as an increase in mean pulmonary arterial pressure

(PAPm) ⩾ 25 mmHg at rest as assessed by right heart catheterization. Definition of PH and pre-capillary PH didn't change, but the definition of post-capillary PH has been modified.

Proposed PH clinical classification includes new conditions, recently identified gene mutations and some other changes. New main positions of this classification:

- pulmonary arterial hypertension (PAH);
- PH due to left heart disease;
- PH due to lung diseases or hypoxia;
- Chronic thromboembolic PH and other pulmonary artery obstructions;
- Pulmonary hypertension with unclear and/or multifactorial mechanisms.

PH diagnosis is based on evaluation of symptoms, physical examination, analysis of examinations defining hemodynamic criteria, etiology and severity of functional and hemodynamic condition. The main cause of PH should be identified according with the clinical classification.

Right heart catheterization is recommended to confirm the diagnosis of PAH and to explain the decision of treatment choice. It is also recommended for patients with PH due to left heart disease, lung disease or thromboembolic PH. Vasoreactivity testing during right heart catheterization is recommended for patients with idiopathic, hereditary, drug or toxin induced PAH in order to choose the patients who can be treated with high dose slow calcium channels blockers. PAH severity should be estimated using clinical data, physical exercise test results, biochemical markers, echocardiography and hemodynamic assays with subsequent dynamic control in stable patients each 3–6 months. Patients with PAH should avoid pregnancy.

In the beginning of PAH treatment monotherapy or drug combination are recommended for patients who didn't receive therapy before and for patients with low or intermediate risk .Initial combined therapy including intravenous administration of prostacyclin analogue is recommended for patients with high risk. Established approaches of PAH treatment are not recommended for patients with PH due to left heart or pulmonary diseases. Surgical pulmonary endarterectomy in condition of deep hypoxia and circulation arrest is recommended for patients with chronic thromboembolic PH.

Guidelines for the management of infective endocarditis

IE recommendations have been prepared by the group of experts of the European Society of Cardiology and have been endorsed by European Association for Cardio-Thoracic Surgery, the European Association of Nuclear Medicine [5]. Previous guidelines dedicated to this problem were published in 2009. Authors of this new edition concentrated on the increase of a role of prevention, principles of teamwork of multidisciplinary "endocarditis team", multimodal visualization techniques, new diagnostic criteria and IE surgical treatment.

New guidelines highlight the key role of general IE prophylaxis and not only of antimicrobial prophylaxis. Prophylaxis is still recommended in patients with predisposing cardiological conditions, and also in patients who are undergoing procedures with high risk of developing IE.

High risk of IE group includes patients with valve replacement, with previous episode of IE, and with congenital heart disease. Antimicrobial prophylaxis should be considered only in dental procedures requiring manipulation of the gingival or periapical region of the teeth or perforation of the oral mucosa. Good oral hygiene and regular dentist visits are more important for reducing IE risk. Vulnerable patients undergoing high-risk dental procedures should receive amoxicillin, ampicillin or clindamycin in case of allergy to penicillin. Antimicrobal prophylaxis is not recommended for procedures involving airways, gastrointestinal tract, urogenital system, skin and soft tissues.

Work of multidisciplinary surgical team using standard protocol of IE treatment provides significant reduction of mortality risk. It is recommended to examine patients with complicated IE during early stage of disease in hospital with possibility of surgical intervention and presence of qualified team that includes infectious disease specialist, microbiologist, cardiologist, imaging specialist, cardiac surgeon, and, if necessary, specialist in CAD. Transthoracic echocardiography is recommended as the first-line imaging procedure for diagnostics of suspected IE. In addition, transesophageal echocardiography can be used. The last one should be initial imaging approach in patients with valve replacement or implanted intracardiac material. Diagnostic algorithm and modified diagnostic criteria of IE are present in the text of the guidelines.

Updated recommendations approve early surgical intervention for IE treatment. Heart failure is the most frequent IE complication and common indication for cardiac valvular operations. Second and third indications for the operation are uncontrollable infection and necessity of emboly , respectively.

Restricted volume of this article doesn't allow to expound all statements issued in new clinical guidelines of the European Society of Cardiology in 2015. Full texts are available on the cite: http://www.escardio.org/Guidelines-&-Education/Clinical-Practice-Guidelines/ESC-Clinical-Practice-Guidelines-list/listings.

Conflict of interest: None declared

References

1. Roffi M, Patrono C, Collet JP, et al. 2015 ESC Guidelines for the management of acute coronary syndromes in patients presenting without persistent ST-segment elevation: Task Force for the Management of Acute Coronary Syndromes in Patients Presenting without Persistent ST-Segment Elevation of the European Society of Cardiology (ESC). Eur Heart J. 2015 Aug 29. [Epub ahead of print].
2. Priori SG, Blomström-Lundqvist C, Mazzanti A, et al. 2015 ESC Guidelines for the management of patients with ventricular arrhythmias and the prevention of sudden cardiac death: The Task Force for the Management of Patients with Ventricular Arrhythmias and the Prevention of Sudden Cardiac Death of the European Society of Cardiology (ESC) Endorsed by: Association for European Paediatric and Congenital Cardiology (AEPC). Eur Heart J. 2015 Aug 29. [Epub ahead of print].
3. Adler Y, Charron P, Imazio M, et al. 2015 ESC Guidelines for the diagnosis and management of pericardial diseases: The Task Force for the Diagnosis and Management of Pericardial Diseases of the European Society of Cardiology (ESC) Endorsed by: The European Association for Cardio-Thoracic Surgery (EACTS). Eur Heart J. 2015; 36(42):2921-64.
4. Galiè N, Humbert M, Vachiery JL, et al. 2015 ESC/ERS Guidelines for the diagnosis and treatment of pulmonary hypertension: The Joint Task Force for the Diagnosis and Treatment of Pulmonary Hypertension of the European Society of Cardiology (ESC) and the European Respiratory Society (ERS) Endorsed by: Association for European Paediatric and Congenital Cardiology (AEPC), International Society for Heart and Lung Transplantation (ISHLT). Eur Heart J. 2015 Aug 29. [Epub ahead of print].
5. Habib G, Lancellotti P, Antunes MJ, et al. 2015 ESC Guidelines for the management of infective endocarditis: The Task Force for the Management of Infective Endocarditis of the European Society of Cardiology (ESC) Endorsed by: European Association for Cardio-Thoracic Surgery (EACTS), the European Association of Nuclear Medicine (EANM). Eur Heart J. 2015 Aug 29. [Epub ahead of print].

International Heart and Vascular Disease Journal
Volume 4, Number 9, March 2016

Journal of the Cardioprogress Foundation

Coronary heart disease in women

Starodubova A.V.[1, 2]*, Kislyak O.A.[1], Chervyakova Y.B.[1]

[1] Russian National Research Medical University named after N.I. Pirogov, Moscow, Russia
[2] Federal State Scientific Institution "Research Institute of Nutrition", Moscow, Russia

Authors:

Antonina V. Starodubova, MD, Head of Department of therapeutic and preventive nutrition, Federal State Scientific Institution "Research Institute of Nutrition". Associate Professor of Faculty Therapy, Russian National Research Medical University named after N.I. Pirogov, Moscow, Russia;
Oksana A. Kislyak, MD, Professor, Head of the Department of Faculty Therapy, Russian National Research Medical University named after N.I. Pirogov, Moscow, Russia;
Julia B. Chervyakova, MD, PhD, Assistant of the Department of Faculty Therapy, Russian National Research Medical University named after N.I. Pirogov, Moscow, Russia.

Summary

Cardiovascular disease (CVD) represents the leading cause of death among women as well as men. The number of deaths due to CVD in women are greater than in men. There are significant gender-related differences concerning CVD. It is less known about CHD in women than in men. There is a need to develop a risk score scale for women in Russia, and for further investigations in the field of treatment and prevention of CVD in women.

Keywords

Coronary heart disease, cardiovascular disease, gender differences, women

Introduction

Coronary heart disease (CHD) is the leading cause of death as in men as in women, and absolute numbers of cardiovascular disease (CVD) mortality are greater in women than in men [1, 2]. During lifetime the risk of developing CVD in men is higher than in women [3]. During last years in developed countries the risk of CVD in men is reducing, together with the increase of CVD in women [4].

Common risk factors are the same both for men and women, but some of them like smoking, diabetes mellitus type 2 and arterial hypertension (AH) have bigger importance in women [5]. If young women don't have 5 risk factors: smoking, AH, diabetes mellitus, hypercholesterolemia, body overweight, they rarely develop CHD and CVD. Only 20% of women <40 years fit these low risk criteria, and at the same time 48% have ⩾ 3 metabolic risk factors of CHD [6]. In Russia the occurrence of risk factors, including metabolic ones, in women is a bit higher than in men: high blood pressure (BP) – 48,4% and 46,6%, body overweight – 48,4% and 46,6%, obesity – 32,9% and 18,6%, total cholesterol levels >5mmol/L – 56,4% and 47,8% respectively [7].

* Corresponding author. Tel. +79262068621. E-mail: lechebnoedelo@yandex.ru

It is well known that increased number and combination of several risk factors has cumulative effect on the risk of CVD development both in men and women [8]. The study, that lasted more than 30 years and involved women of age 18-39 years who didn't have CVD initially, revealed that women without CVD risk had the lowest occurrence of CHD. At the same time women who had one risk factor had 2,4-fold level of morbidity, and women with ≥ 2 risk factors had CHD 5,4 times more often [6]. The SCORE (Systematic Coronary Risk Evaluation) scale is more common in Europe and in Russia, and Framingham Risk Score is more used in the USA. It was demonstrated that according with the Framingham Risk Score, that takes age, AH, smoking, diabetes mellitus and hyperlipidemia into account, the majority of middle age patients are classified as patients of low or moderate risk and > 3/4 of women below 80 years have 10-years Framingham Risk < 10%, and it is not a precise reflection of a real situation [9]. Therefore experts say more and more often that it is necessary to include more women in cardiological studies and it is necessary to take into account specific for female risk factors for prediction of CVD prognosis. In the USA the Reynolds Risk Score was developed especially for CVD risk estimation in women. The most important difference between this score and Framingham Risk score is considering the information about family history of CVD, the levels of high-sensitive C-reactive protein and the levels of glycated hemoglobin in female patients with diabetes mellitus. The Women's Health Study used Reynolds Risk Score and it reclassified 15% of women with the moderate risk into the high risk patients [10].

Gender features of CHD

There are gender differences in complaints and symptoms of unstable CHD, quite often female patients, especially the ones below 55 years, are presented with "atypical" complaints, but due to low awareness of CVD these complaints can be interpreted in a wrong way and acute coronary syndrome (ACS) diagnosis can be not established or established too late [11]. In all age groups women with ACS less frequently have typical chest pain and more often – vasomotor and vegetative symptoms comparing with men [12-14].

It was found that the prognosis for women with recurrent pain and nonocclusive coronary disease is less benignant that it was considered before, and it strongly depends on the number of existing cardiovascular risk factors. 5-years risk of cardiovascular events in women presented with complaints and nonocclusive coronary disease is ~ 50% higher than in women presented with complaints and normal coronary arteries [15].

Women of all age groups have obstructive lesions of coronary arteries more rarely than men [16]. It was described that morphology of atherosclerotic plaques (AP) of male and female is different [17]. AP composition changes during menopause. Women have more inflammatory lesions in coronary arteries than men. Nevertheless, it is supposed that atherosclerosis in middle age women develops slower than in men, atherosclerosis has more diffuse character, and superficial remodeling is common [18]. AP erosion occurs more often in female patients of younger age with ACS, and for male patients and elderly women AP rupture with future thrombus formation [19]. AP erosions can lead to distal embolization with microemboles and dysfunction of microvascular coronary system. Females have ACS without coronary arteries' occlusion more often. Probably, microvascular dysfunction and subendocardial ischemia in case of non-occluded coronary arteries have more importance in women than in men. Women have AP in carotid arteries more rarely and these plaques are more stable than the male ones [20]. At the same time a small prospective study WISE (Women's Ischaemia Syndrome Evaluation) demonstrated that impaired endothelial function is a negative prognostic factor [21]. There is an opinion, that microvascular lesion is the consequence of impaired vasomotor and metabolic regulation of small coronary arterioles and it is one of important CHD risk factors in women and it determines the presence of angina if there is no significant coronary arteries' occlusion [23-25].

CVD progression depends on relation between damage and reparation processes. Endogenous mobilization of endothelial cell precursors playing an important role in reparation processes is associated with improved restoration of endothelium, improved endothelial function and reduced atherosclerotic lesion of vessels. In healthy women of reproductive age stable number of these cells (CD3+KDR+) was bigger than in males, and it didn't differ that much between women in post menopause and men of the same age. These differences reflect gender characteristics of cardiovascular profile, vascular function (endothelial dysfunction) and thickness of intima-media complex of common carotid artery. Endothelial cell progenitors in females are activated according with menstrual cycle and is synchronized with the levels of circulating 17-betaestradiol and it is possible that

they participate actively in protective processes in females before menopause. Experimental works in animal models prove an important role of estrogens in stimulation of vascular inflammation [26].

Vegetative nervous system has an important role in the regulation of cardiovascular system. It is supposed, that activity of sympathetic nervous system is higher in males, and parasympathetic nervous system activity prevails in females. These differences can be explained with the type of fat tissue distribution, hormonal differences, age, presence of obesity, inflammation and psychosocial features. Abnormal vegetative nervous system activity measured by variability of cardiac rhythm is associated with prothrombogenic changes in women with CHD [27].

Coronary angiography is the golden standard for diagnostics of coronary arteries' diseases, but it is not completely appropriate for diagnostic use in women of middle age, because the same symptoms in this category of patients can appear due to abnormal reaction of vessels and vascular reactivity and not because of stenosis. Some studies demonstrated that additional measurement of coronary flow reserve can reveal abnormal vascular reactivity in female patients with angina complaints and nonocclusive coronary artery disease. Intravascular echography allowed to reveal increased thrombotic activity in women with stable and unstable CHD. Therefore to improve CHD diagnostics in female patients it is necessary to use not only coronary angiography but also estimation of coronary flow reserve and intravascular echography, but it is not always possible. Non-invasive techniques like perfusion magnetic resonance imaging, radioscintigraphy, computer tomography-angiography are considered as diagnostic tools for CHD detection in women [28, 29].

Some gender differences in ACS treatment and outcomes are described. In case of myocardial infarction with ST segment elevation percutaneous coronary interventions have equal advantages in men and women. Treatment strategies differ in patients with low risk and myocardial infarction without ST segment elevation. In the FRISCII (The Framingham and Fast Revascularization During Instability in Coronary Artery Disease) and RITA 3 (The Third Randomized Intervention Treatment of Angina trials) studies early invasive intervention in patients with unstable angina and negative biomarkers or in patients with low risk and myocardial infarction without ST elevation led to decrease of mortality in men and not in women [30,31]. In the WISE study increased levels of inflammation markers was associated with unfavorable outcome of CHD in women and they didn't depend on traditional cardiovascular risk factors. Women with ACS usually are older and they have more risk factors. More than that, women have less developed coronary collateral network, less coronary flow reserve, they have more prominent microvascular dysfunction that negatively influences the prognosis. In case of nonocclusive coronary artery disease mortality is higher in women [32, 33]. Hospital mortality of women with ACS is higher than of the same age men [34]. Women develop hemorrhagic complications after coronary interventions especially in case of therapy with glycoprotein IIb/IIIa more often than men [37, 38].

Conclusion

The problem of cardiovascular and metabolic risk in woman of the Russian Federation is very important. Undoubtedly, there are several gender differences in the features of CVD development and clinical course. It is worth to mention, that CHD development and clinical course in women is less studied than in men. It is necessary to develop Russian criteria for the formation of increased risk of CVD group in women and perform further studies aiming to find effective approach of CVD prevention and treatment in women.

Conflict of interest: None declared.

References

1. Women and health: today's evidence tomorrow's agenda. Geneva: World Health Organization 2009; 91p.
2. Go AS, Mozaffarian D, Roger VL, et al. Heart disease and stroke statistics—2014 update: a report from the American Heart Association. Circulation. 2014;129: e28-292.
3. Mosca L, Benjamin EJ, Berra K, et al. Effectiveness-based guidelines for the prevention of cardiovascular disease in women – 2011 update: a guideline from the American Heart Association. Circulation. 2011; 123:1243-62.
4. Towfighi A, Zheng L, Ovbiagele B. Sex-specific trends in midlife coronary heart disease risk and prevalence. Arch Intern Med. 2009; 169: 1762-6.
5. Maas A, Van der Schouw Y, Regitz-Zagrosek V, et al. Red alert for women's heart: the urgent need for more research and knowledge on cardiovascular disease in women. Eur Heart J. 2011; 32:1362-8.
6. Daviglus ML, Stamler J, Pirzada A, et al. Favorable cardiovascular risk profile in young women and long-term risk of cardiovascular and all-cause mortality. JAMA. 2004; 292:1588-92.
7. Nichols M, Townsend N, Luengo-Fernandez R, et al. European cardiovascular disease statistics 2012: European Society of Cardiology. Brussels: European Heart Network, Sophia Antipolis; 2012. 122 p.

8. Goff D, Lloyd-Jones D, Bennett G, et al. ACC/AHA guideline on the assessment of cardiovascular risk 2013: a report of the American College of Cardiology. American Heart Association Task Force on Practice Guidelines. Circulation. 2014; 129:S49-73.
9. Yusuf S, Rangarajan S, Teo K, et al. Cardiovascular risk and events in 17 low-, middle-, and high-income countries. N Engl J Med. 2014; 371(9): 818-27.
10. Johnson BD, Shaw LJ, Buchthal SD, et al. Prognosis in women with myocardial ischemia in the absence of obstructive coronary disease. Circulation. 2004; 109: 2993-9.
11. Shaw LJ, Bugiardini R, BaireyMerz CN. Women and ischemic heart disease. Evolving knowledge. J Am Coll Cardiol. 2009; 54:1561-75.
12. Dey S, Flather MD, Devlin G, et al. Sex-related differences in the presentation, treatment and outcomes among patients with acute coronary syndromes: the Global Registry of Acute Coronary Events. Heart. 2009; 95:20-6.
13. Kislyak OA, Starodubova AV, Hautieva FM, Kopelev AA. Myocardial infarction in overweight women and women with obesity. Consilium Medicum. 2010; 10:26-31. Russian.
14. Kotova DP, Starodubova AV. Age related changes of arteries in obese females. Lechebnoe delo. 2010; 4: 82-7. Russian.
15. Berger JS, Elliott L, Gallup D, et al. Sex differences in mortality following acute coronary syndromes. JAMA. 2009; 302:874-82.
16. Nicholls SJ, Wolski K, Sipahi I, et al. Rate of progression of coronary atherosclerotic plaque in women. J Am Coll Cardiol. 2007; 49:1546-51.
17. Heer T, Schiele R, Schneider S, et al. Gender differences in acute myocardial infarction in the era of reperfusion (the MITRA registry). Am J Cardiol. 2002; 89: 511-7.
18. Frink RJ. Gender gap, inflammation and acute coronary disease: are women resistant to atheroma growth? Observations at autopsy. J Invasive Cardiol. 2009; 21: 270-7.
19. Shaw LJ, Bugiardini R, BaireyMerz CN. Women and ischemic heart disease. Evolving knowledge. J Am Coll Cardiol. 2009; 54:1561-75.
20. Hellings WE, Peeters W, Moll FL, et al. Composition of carotid atherosclerotic plaque is associated with cardiovascular outcome. A prognostic study. Circulation. 2010; 121:1941-50.
21. Von Mering GO, Arant CB, Wessel TR, et al. Abnormal coronary vasomotion as a prognostic indicator of cardiovascular events in women. Circulation. 2004;109: 722-5.
22. Lanza GA, Crea F. Primary coronary microvascular dysfunction: clinical presentation, pathophysiology, and management. Circulation. 2010; 121: 2317-25.
23. Nugent L, Mehta PK, BaireyMerz CN. Gender and microvascular angina. J Thromb Thrombolysis. 2011; 31:37-46.
24. Wang J, Bingaman S, Huxley VH. Intrinsic sex-specific differences in microvascular endothelial cell phosphodiesterases. Am J Physiol Heart Circ Physiol. 2010; 298:H1146-54.
25. Podzolkov V, Vasilyeva L, Matveev V. Gender-specific microcirculatory features in healthy individuals. Vrach (The Doctor). 2013; 3: 55-7. Russian.
26. Lemieux C, Cloutier I, Tanguay JF. Menstrual cycle influences endothelial progenitor cell regulation: a link to gender differences in vascular protection? Int J Cardiol. 2009; 136:200-10.
27. Von Känel R, Orth-Gomér K. Autonomic function and prothrombotic activity in women after an acute coronary event. J Womens Health. 2008; 17:1331-7.
28. Doyle M, Weinberg N, Pohost GM, et al. Prognostic value of global MR myocardial perfusion imaging in women with suspected myocardial ischemia and no obstructive coronary disease. J Am Coll Cardiol. 2010; 3:1030-6.
29. Shaw LJ, Min JK, Narula J, et al. Sex differences in mortality associated with computed tomographic angiographic measurements of obstructive and nonobstructive coronary artery disease. Circ Cardiovasc Imaging. 2010; 3:473-81.
30. Clayton TC, Pocock SJ, Henderson RA, et al. Do men benefit more than women from an interventional strategy in patients with unstable angina or non-ST-elevation myocardial infarction? The impact of gender in the RITA3 trial. Eur Heart J. 2004; 25:1641-50.
31. O'Donoghue M, Boden WE, Braunwald E, et al. Early invasive versus conservative treatment strategies in women and men with unstable angina and non-ST-elevation myocardial infarction: a meta-analysis. JAMA. 2008; 300: 71-80.
32. Arant CB, Wessel TR, Ridker PM, et al. Multimarker approach predicts adverse cardiovascular events in women evaluated for suspected ischemia. Clin Cardiol. 2009; 32: 244-50.
33. Gulati M, Cooper-DeHoff RM, McClure C, et al. Adverse cardiovascular outcomes in women with nonobstructive coronary artery disease. Arch Intern Med. 2009; 169: 843 – 50.
34. Hochman JS, Tamis JE, Thompson TD, et al.Sex, clinical presentation, and outcome in patients with acute coronary syndromes. N Engl J Med. 1999; 341:226-32.
35. Alexander KP, Chen AY, Newby LK, et al. Sex differences in major bleeding with glycoprotein IIb/IIIa inhibitors: results from the CRUSADE initiative. Circulation. 2006; 114:1380-7.
36. Kruk M, Pregowski J, Mintz GS, et al. Intravascular ultrasonic study of gender differences in ruptured coronary plaque morphology and its associated clinical presentation. Am J Cardiol. 2007; 100: 185-9.
37. Poirier P, Giles TD, Bray GA, et al. Obesity and cardiovascular disease: pathophysiology, evaluation, and effect of weight loss: an update. Circulation. 2006; 113:898-918.
38. Shalnova SA, Evstifeeva SE, Deev AD, et al. Impact of the inflammatory and ischemic heart disease markers into the overall cardiovascular mortality in senile citizens of a large city (the data from SAHR trial). Russ J Cardiol 2015; 6 (122): 7-13. Russian.

International Heart and Vascular Disease Journal
Volume 4, Number 9, March 2016

Journal of the Cardioprogress Foundation

The relationship between total epicardial fat volume and atrial fibrillation

Mohammed Abdelshafy MD[1*], Ahmad Torky MD[2] , Ahmad Farid MD[2]

[1] Cardiology department , [2] Radiology department , Faculty of medicine, Benha University, Benha, Egypt.

Authors:

Mohamed Abdelshafy Tabl, MD, Cardiology department, Faculty of medicine, Benha University, Benha, Egypt;

Ahmad Torky, MD, Radiology department, Faculty of medicine, Benha University, Benha, Egypt;

Ahmad Farid, MD, Professor of radiology, Radiology department, Faculty of medicine, Benha University, Benha, Egypt.

Abstract

Background

Obesity is an important risk factor for atrial fibrillation (AF). Local epicardial fat enclosed by the visceral pericardial sac has been hypothesized to exert local pathogenic effects on cardiac structures. We aimed to characterize the relationship between total epicardial fat volume assessed by non contrast cardiac CT and AF.

Methods

This case control study conducted from May 2013 to December 2014 in cardiology and radiology departments of Benha University Hospitals. Fifty patients with a history of AF were taken up plus control group of 50 reference patients without history of AF. All patients underwent cardiac CT imaging to measure total epicardial fat volume (EFV), together with systemic obesity indices as body mass index (BMI), waist circumference and body weight plus echocardiographic parameters as left atrium (LA) volume index, left ventricular ejection fraction. All these were examined in relation to the presence and chronicity of AF.

Results

EFV was significantly associated with the presence of AF (p values<0.05).Significant positive correlation between EFV and AF chronicity was denoted. Patients with persistent AF had significantly larger EFV versus patients with paroxysmal AF (p value = 0.002). EFV was positively correlated with LA volume index (r = +0.45, p<0.001) Multivariate logistic regression model for AF risk factors revealed that EFV was the strongest independent risk factor for AF with highest odds ratio (2.13,95%CI: 1.01 to 3.06) followed by odds ratio (1.81,1.55 and 0.8) for LA volume index ,waist circumference and BMI respectively.

* Corresponding author. Tel. +2001223723050. E-mail: mshafytabl@yahoo.com

Conclusion

Epicardial fat is associated with the presence of AF and predicts chronicity. These associations are independent to systemic measures of adiposity and sensitive echocardigraphic parameters as LA volume index. These findings are consistent with the hypothesis of a local pathogenic effect of epicardial fat on the arrhythmogenic substrate supporting AF

Key words

Atrial fibrillation, cardiac CT imaging, obesity, epicardial fat.

Introduction

Atrial fibrillation (AF) is the most common arrhythmia found in clinical practice (1). It also accounts for 1/3 of hospital admissions for cardiac rhythm disturbances (2). Systemic obesity is a common modifiable risk factor for different cardiovascular disorders including AF. Above and beyond hazardous effect of obesity, Epicardial fat defined as the local visceral fat depot enclosed by the visceral pericardial sac shares the same blood supply as adjacent myocardium and also show paracrine functions. This is the risky fat that is metabolically active and has been hypothesized to exert a local pathogenic inflammatory effect on nearby cardiac structures (3). In recent years, several studies have shown that an increased epicardial fat volume noninvasively measured by CT or MRI images was strongly associated with the presence of coronary artery disease and atrial fibrillation, and adverse cardiovascular events (4). Because multiple factors are related to epicardial fat, we hypothesized whether epicardial fat could be independently associated with AF after adjusting multiple factors potentially related to epicardial fat. Therefore, this study was conducted to assess the relationship between epicardil fat volume and the presence and progression of AF when considering co variables related to epicaerdial fat.

Materials and Methods

Study population

This study was a case control study conducted from May 2013 to December 2014 in cardiology and radiology departments in Benha University Hospitals and local private centers. One hundred patients selected from cardiology department in Benha University Hospital. Their age ranged from 45 to 65 years and their body mass index (BMI) ranged from 25 to 32. They were divided into two groups as follows: AF group included 50 patients with a documented history of AF. Control group included 50 patients with intermediate risk and had no history of AF. They had age and sex matching to AF group.

All patients in both groups were referred for non-contrast CT for the evaluation of the volume of the total epicardial fat (EFV). This study was approved by the ethical committee in the faculty of medicine, Benha University.

Methods

All patients were subjected to the following:

History taking: Age, Sex, Smoking, hypertension, diabetes mellitus, thyrotoxicosis and documented history of AF.

Anthropometric measurements: Weight in kilograms, height in meters, waist circumference and body mass index (BMI) ranged from 25 to 32.

Clinical examination: Full general and local cardiac examination.

Echocardiography: Transthoracic echocardiography was performed with a commercially available system (Vivid Seven, General Electric, Milwaukee, WI). Left atrial volume was calculated using the modified biplane Simpson's method from the apical 2-chamber and 4-chamber views. (Figure 1) LA volume index was calculated (LA volume (ml)/BSA (m^2). Left atrial enlargement was defined as LA volume index > 22±6 (ml/m^2) for both men and women. Left ventricular ejection fraction (LVEF) was measured using the Simpson method. An LVEF <50% was considered abnormal. Structural heart disease was defined as moderate or greater amount of valvular regurgitation or left ventricular hypertrophy (5).

Non contrast CT. Imaging Technique: All CT scans were performed on a different local radiation centers including CT unit in Benha University Hospital with The Aquilion ONE ViSION CT scanner , Toshiba America Medical Systems , USA. All images were interpreted by a single radiologist who had more than 15 years of experience in the interpretation of CT scanning field and he was blinded to the history of AF. Tomogram was taken from tracheal bifurcation to the diaphragm in a single breath-hold in the craniocaudal direction. The superior heart limit slice is typically chosen at the split of the pulmonary artery. The

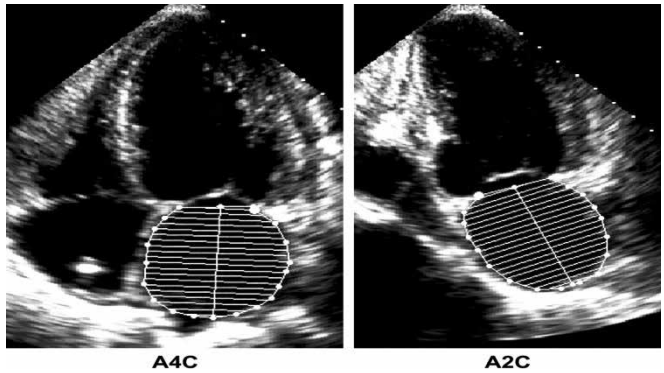

Figure 1. Left atrial volume was calculated using the modified biplane Simpson's method.

anatomic landmark for the inferior limit of the heart is typically the most inferior slice of the myocardium or the most inferior slice with the posterior descending artery (6). Image reconstruction was performed using retrospective ECG-gated acquisition spiral mode. Since epicardial fat is a compressible structure, End systolic frames are used to avoid suspected attenuation of fat during diastole by myocardial mass. A 3-D workstation was used to reconstruct axial images retrospectively at an optimal window. The image data sets were analyzed by means of Multiplanar reformatted images (vertical, long-axis, and short-axis views), curved Multiplanar reformatted images, thin-slab maximum-intensity projection images, and volume-rendered images (6).

Measurement of epicardial fat volume (EFV): Using the 5.0-mm-thick axial slices the parietal pericardium was manually traced in every fourth slice starting from the bifurcation of pulmonary artery to the diaphragm. The computer software (Toshiba - Aquilion ONE 640) then automatically interpolated and traced the parietal pericardium in all slices interposed between the manually traced slices to measure the EFV in cm^3. The total number of slices was 30 to 40 per heart. All automatically traced slices were examined and verified for accuracy. To ensure adequate gating and minimal motion artifact, patients in AF received beta-blockers and have CT scanning only if the ventricular response was <80 beats/min. The typical processing time for this method is 7–10 minutes. Standard fat attenuation values are used to define fat attenuation by CT; for non-contrast CT typically an attenuation range of (–30, –190) Hounsfield Units is used. Fat voxels within this attenuation range within the visceral pericardium are classified as epicardial fat, and within the inner thoracic cavity classified as thoracic fat (7) (Figure 2).

Statistical analysis of the collected data

Results were collected, tabulated, statistically analyzed using statistical Package of Social Science (SPSS) version 11 by Department of University Academic Computing Technologies (UACT) (American University in Cairo).

Two main statistical methods were used to present data:

Descriptive statistics: in which quantitative data were presented in the form of mean (x), standard de-

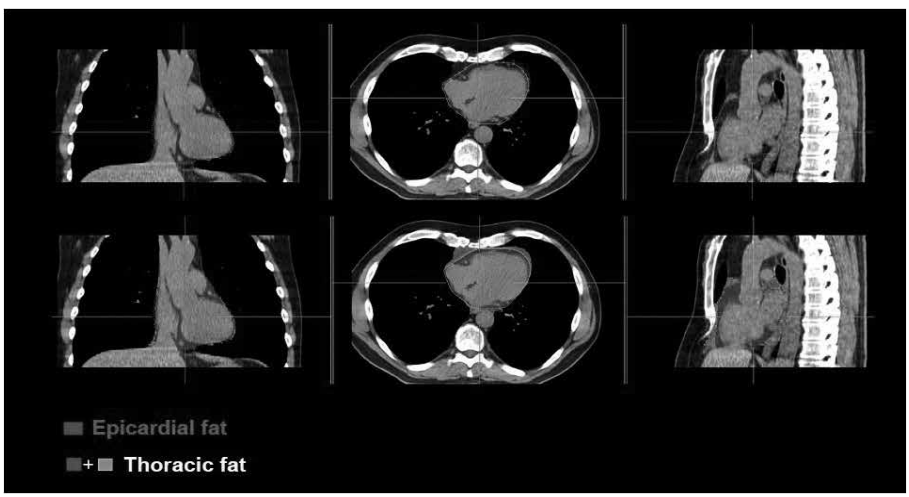

Figure 2. Measurement of EFV and thoracic fat From Noncontract CT

viation and range, and qualitative data were presented in the form of number and percentage (%).

Analytical (inferential) statistics: used to find out the possible association between many factors and the targeted disease. The following statistical tests were applied.

Chi-Squared test (χ^2), Fischer exact test, t-test, Mann-Whitney test.

Correlation: Correlation is a statistical technique that can show whether and how strongly pairs of variables are related.

Binary logistic regression: A statistical measure that attempts to determine the strength of the relationship between one dependent variable (usually denoted by Y) and a series of other changing variables (known as independent variables).

P-value of <0.05 was considered statistically significant.

Results

Patient characteristics

There was age and sex matching between AF and control groups (P value = 0.15). Mean age equal 58.56 ±8.79 and 55.80±7.13 for AF and control group respectively.

As regards clinical history, rate of DM and thyrotoxicosis was significantly higher (P value = 0.03 & 0.01) among the AF group. Hypertension and smoking showed no significant differences in both groups (p value > 0.05). As regard systemic obesity indices, AF group were of significantly larger weight, BMI and waist circumference (P value = 0.038, 0.01 and 0.001) respectively.

As regards different echocardiographic parameters, AF group showed significantly larger LA volume indices & LA diameters (P value <0.001). The results showed insignificant difference in both studied groups as regards LVEF% and Lt. ventricular thickness (P value = 0.2 & 0.59).

Relation between EFV and different parameters

The AF group had a significantly larger EFV than the control group (170.34±44.58 cm^3 versus 107.28±40.08 cm^3, p < 0.001) (Table 1). EFV was positively correlated with body weight (r = 0.25, p = 0.08), BMI (r = 0.38, p = 0.007), and LA volume index (r = 0.39, P = 0.005). There were no correlations between EFV and weight (r = 0.25, P = 0.08), waist (r = 0.01, P = 0.95), age, left ventricular ejection fraction, and left ventricular wall thickness (P > 0.05 for all) (Table2).

Table 1. **Comparison of demographic, echocardiographic and CT data in the two groups**

Variable	AF group	Control group	P value
Age	58.56 ±8.79	55.80±7.13	0.15
Men	35(70%)	33(68%)	0.15
Weight (kg)	89.48±10.94	89.48±10.94	0.038
BMI	29.21±4.76	27.02±3.60	0.01
Waist circumference (cm)	103.20±7.94	98.10± 6.94	0.001
HTN	25%	20%	>0.05
DM	20%	6%	0.03
Thyrotoxicosis	12%	0%	0.01
smoking	30%	25%	>0.05
Lt atrial diameter (mm) X ± SD Range	43.36±7.11 31 – 59	31.26±6.66 20 – 42	<0.001
LA volume index (ml/m^2) X ± SD Range	33.5±6.11 29-36	22.56±6.8 20-24	<0.001
LV EF % X ± SD Range	50.36±8.19 32 – 62	63.32±6.33 54 – 75	0.20
Lt. ventricular thickness /cm X ± SD Range	1.08±0.31 0.60 – 1.90	1.03 ± 0.37 0.60 – 1.6	0.59
Epicardial fat volume (EFV) X ± SD Range	170.34±44.58 cm^3 90 – 259	107.28±40.08 cm^3 45 – 211	<0.001

BMI=body mass index, HTN=hypertension,DM=diabetes mellitus, LA=left atrium , LV=left ventricle , EF = ejection fraction.
SD = slandered deviation.

Table 2. **Correlation between pericardial fat volume and other parameters among AF group**

	pericardial fat volume (AF group)	
	Correlation coefficient - r-	P value
Age	- 0.32	0.02
Weight /year	+ 0.25	0.08
BMI	+ 0.38	0.007
Waist circumference	+ 0.01	0.95
Left atrial volume index	+ 0.39	0.005
LVEF %	+ 0.16	0.27
Left ventricular wall thickness	+ 0.21	0.14

BMI=body mass index, LVEF=left ventricle ejection fraction.

Multivariate regression model for risk factors of AF

Using Wald test for multiple parameters , this model revealed that EFV and LA volume index were independent risk factors for occurrence of AF with highest odds ratio (2.13 & 1.81) & highest Wald x2 values (10.34 & 11.94) respectively. While obesity indices as

Table 3. **Multivariate logistic regression model for risk factors of AF**

Risk factors	Wald x^2	P value	Odds Ratio (Exp. Beta)	95% CI	
				Lower	Upper
DM	0.03	0.85	0.88	0.07	9.13
Thyrotoxicosis	0.06	0.81	0.91	0.13	8.70
Weight	0.18	0.67	1.02	0.93	1.12
BMI	1.11	0.29	0.87	0.66	1.13
Waist circumference	5.65	0.02	1.55	1.02	1.99
Lt. atrial volume index	11.94	0.001	1.81	1.11	2.45
Epicardial fat volume	10.34	0.001	2.13	1.01	3.06

Wald x^2 = Wald Test on multiple parameters, CI = Confidence interval, DM=diabetes mellitus, BMI=body mass index.

BMI, waist circumference and weight showed lower odds ratio (0.87, 1.55 & 1.02) and lower Wald x^2 values (1.11, 5.6, 0.18 respectively) (Table 3). This data concluded that EFV by CT and LA volume index by echocardiography were the strongest independent risk factors for AF occurrence.

Impact of EFV on the progression of AF

66% of the AF group had paroxysmal AF and 34% had persistent AF. There was no significant difference between persistent and paroxysmal AF as regards age (P value = 0.34). There was significant association between persistent AF and male sex as 88.2% of persistent AF subjects were males versus only 60% among paroxysmal AF cases. Hypertension, DM, smoking and thyrotoxicosis showed no significant differences among paroxysmal AF and persistent AF cases (P value > 0.05). Persistent AF group were of significantly larger weight and BMI than paroxysmal AF group (P value <0.05).

Persistent AF group showed significantly larger LA volume indices than paroxysmal AF group (P value = 0.005). No significant difference between two groups as regards LVEF (%) and Left ventricular thickness (P value >0.05). The group with persistent AF had significantly larger EFV than those with paroxysmal AF group (196.29±49.48 cm^3 versus 156.97±35.88 cm^3 and P value = 0.002) (Table 4).

Table 4. **Comparison between paroxysmal AF subgroup and persistant AF subgroup as regard different risk factors.**

variable	Paroxysmal AF N=33	Persistant AF N=17	T test	P value
Age /year X ± SD Range	59.42 ±8.22 38 – 78	56.88±9.85 40 – 74	0.97	0.34
Male	20(60%)	15(88%)	4.02	0.04
HTN	17(51.5%)	8(47.1%)	0.09	0.76
DM	4(12.1%)	6(35.5%)	3.77	0.07
Thyrotoxicosis	5(15.2%)	1(5.9%)	0.91	0.65
smoking	14(32.4%)	10(58.9%)	1.46	0.48
Weight /kg X ± SD	85.51±10.40	97.18 ±7.38	4.58	<0.001
BMI X ± SD	26.68 ±2.99	34.13±3.55	7.82	<0.001
Waist circumference X ± SD	101.97±6.94	105.59±9.37	1.41	0.17
Lt atrial volume index (ml/m^2) X ± SD	31.39±6.25	37.18±7.28	2.93	0.005
LVEF (%) X ± SD	50.51±8.72	50.06±7.29	0.18	0.85
Lt. ventricular thickness /cm X ± SD	1.10±0.32	1.04±0.29	0.70	0.49
Epicardial fat volume X ± SD	156.97±35.88 cm^3	196.29±49.48 cm^3	Mann Whitney U 3.23	0.002

SD=standerd deviation , DM=diabetes mellitus, BMI=body mass index.

Discussion

Major findings

In the current study, detailed echocardiography and non contrast CT examination were used to present new information regarding the interrelationships between localized epicardial fat depots and AF. The results revealed that, patients with AF had significantly larger EFV compared with control group (P <0.001). (Table 1) There was a strong association between EFV and AF chronicity. Persistent AF patients had a significantly larger EFV compared with paroxysmal AF patients (P = 0.002). (Table 4) These results were in aggrement with Chekakie et al. 2010 who examined the association between epicardial fat and AF chronicity using non contrast CT and demonstrated a significant association of EFV with both paroxysmal and persistent AF. EFV was associated with both paroxysmal AF (odds ratio 1.11, 95% CI: 1.01 to 1.23; p=0.04) and persistent AF (odds ratio 1.18, 95% CI: 1.05 to 1.33; P=0.004) (8). In current study, epicardial fat was strongely associated with the presence of AF (odds ratio: 2.13; 95% confidence interval: 1.01 to 3.06, P = 0.04) and this association was completely independent to DM, thyrotoxicosis, weight, waist circumference, BMI.(Table 2) Finally, strong positive correlations between EFV and LA volume indices were documented (r = +0.39, p value = 0.005). While more systemic measures of adiposity as waist, weight had a lack of same close positive correlations to EFV (r = +0.01 and +0.25, P value = 0.95 and 0.08).(Table 2) Our data suggest that epicardial fat may have a pathogenic effect on the anatomically contiguous atria, above and beyond systemic effects of generalized adiposity. Such effect could promote an arrhythmogenic substrate initiating AF.

The difference and novelty of the study

In spite our results were in agreement with many previous studies, to our knowledge the present study is unique in using both CT to assess epicardial fat volume plus echocardiographic measures of LA volume in AF patients. This study provides the first report of a clear association between epicardial fat, LA volume index and AF occurrence. Previous studies as Thanassoulis et al. 2010 who reviewed the Framingham Heart Study and revealed association between epicardial fat and AF occurrence (P=0.02) not used the atrial dimension as a covariate in the multivariable model, while in the present study we aimed to use LA volume parameters side by side to different obesity indices in the multivariable model and showed that EFV and LA volume index were independent risk factors for occurrence of AF (9). Iacobellis et al. 2007, who tested the relation between epicardial adipose tissue and atrial dimensions by echocardiography in morbidly obese subjects, found that epicardial fat has been strongly associated with LA dimensions in AF patients. They had two limitations; first they analyzed epicardial fat thickness using echocardiography instead of epicardial fat volume assessed by cardiac CT which is known to be more accurate measure. Second they used LA volume instead of LA volume index which could limit the strength of the results and the conclusion obtained from their study (10).

Hypothesis of a local pathogenic effect of epicardial fat

At a local level, pericardial fat has been associated with increased expression of numerous inflammatory markers (11-12). Intracardiac inflammatory markers have also been observed to be greater than peripheral inflammatory markers, and greatest in the left atrium, which plays a critical role in AF genesis (13). Cytokines have also been shown to activate fibroblasts, with the extracellular matrix deposition and fibrosis causing electro anatomical remodeling (14). Therefore, the present finding supports the notion that epicardial fat may exert deleterious effects on the anatomically contiguous atria and promote arrhythmogenesis.

Clinical Implications

With the increasing use and availability of CT scan, EFV assessed by CT scan may be used to identify the patients with undetected AF or asymptomatic AF.

Study limitations

The relatively limited number of the patients could limit the strength of results and conclusion obtained from this study.

Conclusion

Epicardial fat is associated with the presence of AF and predicts chronicity of AF. These associations are both independent of systemic measures of adiposity. EFV and LA volume index were independent risk factors for occurrence of AF**.** Associations between EFV and LA volume changes could explain the mechanism of AF initiation. These findings are consistent with the hypothesis of a local pathogenic effect of epicardial fat on the arrhythmogenic substrate supporting AF.

Conflict of interest:

The authors declare no conflict of interests.

Acknowledgment

The authors would like to thank all the staff members of cardiology and radiology departments in Benha University Hospitals.

References

1. Hylek EM, Phillips KA, Chang Y, Henault LE, Selby JV ,Singer DE , et al: Prevalence of diagnosed atrial fibrillation in adults: national implications for rhythm management and stroke prevention: the AnTicoagulation and Risk Factors in Atrial Fibrillation (ATRIA) Study. JAMA 2001 285 (18): 2370–5.
2. Friberg L., Hammar N, Pettersson H. and Rosenqvist M.: Increased mortality in paroxysmal atrial fibrillation: report from the Stockholm Cohort-Study of Atrial Fibrillation (SCAF). Oxford Journals- Medicine - European Heart Journal. 2007 Vol. 28 (19); Pp. 2346-53.
3. Iacobellis G, Corradi D & Sharma AM: Epicardial adipose tissue: anatomic, biomolecular and clinical relationships with the heart. Nat Clin Pract Cardiovasc Med. 2005 Oct; 2(10):536-43.
4. Damini Dey,Ryo Nakazato,Debiao Liand & Daniel S Berman : Epicardial and thoracic fat - Noninvasive measurement and clinical implications. Cardiovasc Diagn Ther. 2012 Jun; 2(2): 85–93.
5. Lang RM, Bierig M, Devereux RB, Flachskampf FA, Foster E, Pellikka PA, et al : American Society of Echocardiography's Nomenclature and Standards Committee, Task Force on Chamber Quantification, American College of Cardiology Echocardiography Committee, American Heart Association, European Association of Echocardiography, European Society of Cardiology . Eur J Echocardiogr. 2006 Mar; 7(2):79-108.
6. Shmilovich H, Damini D, Victor Y , Ronak R, Nakazato R, Otaki Y, et al: Threshold for the Upper Normal Limit of Indexed Epicardial Fat Volume Derivation in a Healthy Population and Validation in an Outcome-Based Study Am J Cardiol. 2011 Dec 1; 108(11): 1680–1685.
7. Dey D., Suzuki Y., Suzuki S. Ohba M, Slomka PJ, Polk D, et al: Automated quantitation of pericardiac fat from noncontrast CT. Invest Radiol. 43 2008:145-153
8. Al Chekakie M, WellesChristine C, Raymond M, ShapiraAdam R, Joseph C, WilberDavid J, et al: Pericardial fat is independently associated with human atrial fibrillation.J. Am. Coll. Cardiol.2010; 56 (10):784–8.
9. Thanassoulis G, Massaro JM, O'Donnell CJ, Hoffmann U, Levy D, Ellinor PT, et al: Pericardial fat is associated with prevalent atrial fibrillation: the Framingham Heart Study. Circ Arrhythm Electrophysiol.2010;3(4):345–50.
10. Iacobellis G, Leonetti F, Singh N & Sharma AM.: Relationship of epicardial adipose tissue with atrial dimensions and diastolic function in morbidly obese subjects. Int J Cardiol 2007; 115:272–3.
11. Issac TT, Dokainish H & Lakkis NM.: Role of inflammation in initiation and perpetuation of atrial fibrillation: a systematic review of the published data. J Am Coll Cardiol 2007;50:2021– 8.
12. Mazurek T, Zhang L, Zalewski A, Mannion JD, Diehl JT, Arafat H, Sarov-Blat L,et al.: Human epicardial adipose tissue is a source of inflammatory mediators. Circulation 2003; 108: 2460–6.
13. Marcus GM, Smith LM, Ordovas K, R J Lee , N Badhwar , A M Kim et al. Intracardiac and extracardiac markers of inflammation during atrial fibrillation. Heart Rhythm 2010; 7:149 –54.
14. Nguyen BL, Fishbein MC, Chen LS, Chen PS & Masroor S.: Histopathological substrate for chronic atrial fibrillation in humans. Heart Rhythm 2009; 6:454–60.

International Heart and Vascular Disease Journal
Volume 4, Number 9, March 2016

Journal of the Cardioprogress Foundation

Possibilities of using two treatment regimen for vascular stiffness correction

Drozdetsky S.I., Kuchin K.V.*

Nizhny Novgorod State Medical Academy, Nizhny Novgorod, Russia

Authors:

Kirill V. Kuchin, M.D. Ph.D., assistant of the department of hospital and outpatient therapy, Nizhny Novgorod State Medical Academy, Nizhny Novgorod, Russia;

Sergei I. Drozdetsky, M.D., Ph.D, professor of the department of hospital and outpatient therapy, Nizhny Novgorod State Medical Academy, Nizhny Novgorod, Russia.

Summary

Objective

To find the optimal regimen of antihypertensive therapy with the most evident effect on elastic properties of blood vessels.

Materials and methods

This study involved male patients 20–70 years old with arterial hypertension without severe somatic diseases and compared vasculoprotective activity of two therapeutic regimens based either on long acting metaprolol tartrate or on fixed combination of amlodipine and lisinopril.

Results

Although both treatment regimen had comparable antihypertensive effect, using fixed combination of amlodipine and lisinopril as a basis therapy demonstrated better vasculoprotective activity.

Conclusion

The results of this study allow to recommend fixed drug combination of amlodipine and lisinopril as the preferable one for the treatment of male patients with arterial hypertension and abnormalities of vessel wall elasticity.

Keywords

Arterial hypertension, augmentation index, aortic pulse wave velocity, vessel wall

* Corresponding author. Tel. +79307052373. E-mail: kuchinkv@yandex.ru.

Introduction

Slowing down the increase of vascular rigidity and its involution in arterial hypertension (AH) has significant interest in clinical practice. Results of various studies demonstrated positive effect of many non-pharmacological approaches like physical exercises, reducing body weight, low-salt diet, reduced alcohol consumption, addition of garlic, fish oil, α-linoleic acid [1].

Between pharmacological agents angiotensin-converting enzyme inhibitors (ACE inhibitors), angiotensin receptor type II inhibitors (AR inhibitors), calcium channel blockers (CB), some beta-blockers (β-B), indapamide, nitrates and statins have proved effect on vascular remodeling [2–4]. Reaching the target blood pressure (BP) levels for these drugs is the necessary condition of their effect on elastic properties of blood vessels [3]. The COMPLIOR study for the first time raised the question about additional (pleiotropic) effects of different antihypertensive drug classes on vascular rigidity that are not related to BP reduction [5]. Combined therapy of ACE inhibitors and indapamide reduced aortic pulse wave velocity (PWVVao) by 9 % and there was no correlation between the grade of BP reduction and PWVVao. These data allow to consider some additional factors that influence vascular elasticity besides the grade of BP reduction in patients with AH who receive ACE inhibitors therapy. Another study demonstrated that although the grade of BP reduction was comparable in case of therapy with ACE inhibitors, CB and AR inhibitors only ACE inhibitors therapy allowed to achieve significant reduction of PWVVao [6]. Another study that aimed to prove high vasculoprotective activity of ACE inhibitors compared four classes of antihypertensive drugs (ACE inhibitors, AR inhibitors, β-blockers, CB [7]. Patients who received ACE and AR inhibitors had better characteristics of vascular elasticity after 4 months of antihypertensive therapy comparing with patients who received ß-blockers. Patients with AH who received CB demonstrated intermediate characteristics of vascular elasticity.

Lisinopril is one of the best studied drugs of ACE inhibitors class [8]. Its efficacy is investigated in >50 studies with more than 30000 patients involved. It has been demonstrated that combined therapy with lisinopril and simvastatin has significant positive effect on PWVao and augmentation index (AI) [9]. It necessary to notice that although target levels of BP and lipid characteristics in this patients have been achieved during first 2 month of treatment, PWVao and AI reached normal levels only after 6 and 12 months of treatment. These data prove the necessity of prolonged therapy for slowing down or involution of vascular wall remodeling. Lisinopril administration instead of any other ACE inhibitor in treatment of patients with congestive heart failure (CHF) during 6 months led to significant increase endothelium-dependent vasodilatation and demonstrated positive influence on PWVao and AI [10].

CB have proved its efficacy in reaching target levels of BP and organ protection during AH treatment [11]. Vasculoprotective effect of these drugs is caused by their direct relaxing action on vessels and their ability to regulate collagen metabolism in smooth muscle cells [12]. Amlodipine is the most frequently used dihydropyridine CB. Monotherapy with amlodipine allows to reach target levels of BP in 75–87 % of patients [13]. Major trials demonstrated that amlodipine and ACE inhibitors have the most prominent ability to cause right ventricular hypertrophy (RVH) regression [14]. The PREVENT (Prevention of Recurrent Venous Thromboembolism) study showed the amlodipine ability to reduce the thickness of intima-media complex layer of carotid arteries [15]. Smaller study demonstrated the ability of amlodipine to significantly reduce PWVao during 6-months treatment [16].

Amlodipine and lisinopril combination for treatment of patients with AH allows to increase antihypertensive and pleotropic effects and also to reduce the risk to develop unfavorable reactions. The ASCOT (Anglo-Scandinavian Cardiac Outcomes Trial) demonstrated significant reduction of total mortality by 11 %, cardiovascular mortality — by 24 %, the risk of stroke development — by 23 % in patients who received ACE inhibitor/CB comparing with patients who received β-B/thiazide diuretic combination [17]. The CAFÉ (The Conduit Artery Functional Endpoint Study) found out that the reason of this difference is less prominent reduction of central blood pressure in patients who received β-B/thiazide diuretic combination and absence of its influence on elastic properties of vessels [18]. The ACCOMPLISH (Avoiding Cardiovascular events through Combination therapy in Patients Living with Systolic Hypertension) study showed that ACE inhibitor/CB combined therapy has an advantage over ACE inhibitor/thiazide diuretic combination [19]. It was possible to reduce the risk of developing mixed primary endpoint (cardiovascular mortality, acute myocardial infarction, hospital admission with unstable angina, recanalization of coronary arteries procedures) by 19 % in the group of

patients who took ACE inhibitor/CB comparing with ACE inhibitor/thiazide diuretic combination.

Fixed combination of amlodipine and lisinopril allows to reach BP target levels in 77–99 % of patient with AH stage 1–3 according with results of different studies [20–22]. This drug combination also demonstrated good vasculoprotective activity. It was more effective in PWVao reduction after 6-month therapy comparing with enalapril/hydrochlorotiazide combination [23]. Although β-B are the first line therapy of AH, nowadays there is no unambiguous opinion about β-B effect on vascular wall rigidity. Some studies demonstrated lack of this class's influence on parameters of vascular elasticity and central BP and showed even their negative effect — AI increase [24]. Some authors associated with this the certain worsening of prognosis of patients with AH and without comorbid coronary artery disease (CAD) during β-B therapy comparing with other classes of antihypertensive drugs [18, 25]. Nevertheless, therapy with extended release metoprolol demonstrated significant decrease of central BP and PWVao [26].

According with this information, the ACE inhibitor/CB therapeutic regimen is considered to be optimal for vascular "organ protection" but it has to gain further confirmation, particularly to be compared with long acting metaprolol tartrate.

The aim of this study is to estimate vasculoprotective activity of two therapeutic regimen: lisinopril/amlodipine fixed drug combination and long acting metoprolol tartrate as basis therapy.

Materials and methods

This study included male patients with AH who gave their voluntary informed consent to partecipation in this study. Patients with acute diseases or acute conditions of chronic diseases that occurred < 3 months before, acute coronary syndrome, decompensated CHF and acute cardiovascular collapse, acute cerebral circulation disorders (ACCD) that happened < 1 year ago, diabetes mellitus, atrial fibrillation, stable angina of III/IV functional classes (NYHA). Stratified block randomization allowed to distribute the patients who were involved in the study between two groups (n=30 in each one). The first group ("Group 1") received fixed combination of amlodipine and lisinopril as the basis antihypertensive therapy with the starting dose of 5–10 μg Therapeutic regimen in the second group of patients ("Group 2") was based on the long-acting form of metoprolol tartrate with the starting dose of 50μg twice per day.

Examination and treatment of patients has been done according with the guidelines of The Russian Medical Society on Arterial Hypertension [27]. All patients underwent 24-hours BP monitoring (24h-BPM) and parameters of vascular rigidity were assessed by oscillometric technique with 24-h BP monitor "MiSDP-2" and Vasotens software (BPLab company,

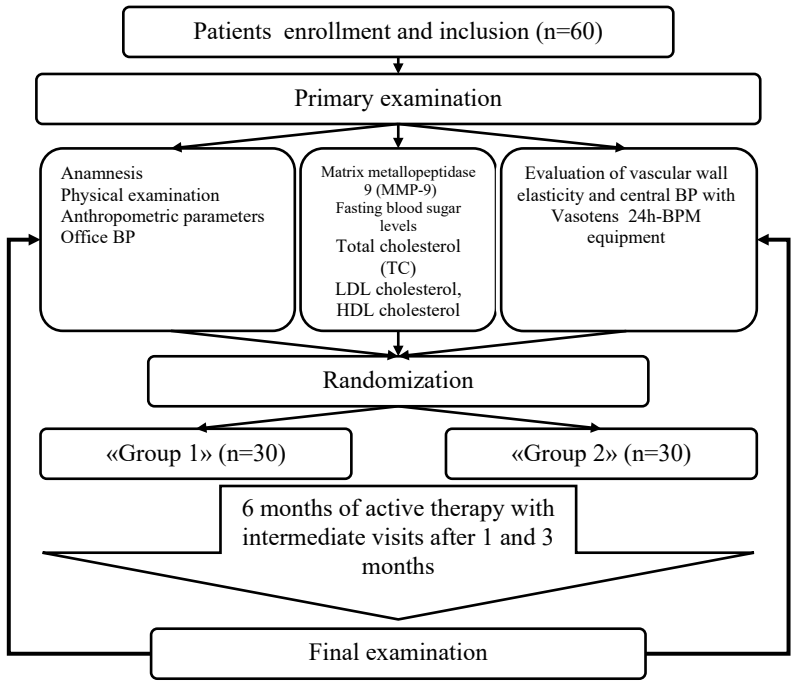

Figure 1. Study design

Nizhny Novgorod) [28]. Study program included investigation of matrix metalloproteinase-9 (MMP-9), the enzyme that participates in collagen and elastic fibers [29].

We used nonparametric methods of statistical analysis. Quantitative data are represented as median with demonstration of 25th and 75th quantiles (M [25, 75]). Qualitative and ordinal data are present as percentage. Mann-Whitney test was used to evaluate significance of differences between two independent groups of quantitative characters. To estimate the differences between three and more groups Kruskal-Wallis was used. χ^2 test was used for evaluation of differences between the groups of qualitative and ordinal characters. Significance of differences in the same group of patients before and after treatment was estimated with McNemar test in case of quantitative characters, and in case of qualitative characters. χ^2 test was used. Multiple comparisons adjustments were performed if it was necessary. Correlation analysis was done with Spearman's rank correlation test.H0 was rejected if p-value was less than 0.05. All statistical analysis was done using IBM SPSS Statistics 22 software.

Results and discussion

A comparative characteristic of two groups of patients before the start of active therapy is demonstrated in Table 1. Studied groups were comparable in age of patients, AH duration, waist circumference. body mass index, systolic blood pressure (SBP) and pulse rate, number of smokers and the number of pack years, occurrence of alcohol consumption and sedentary life, and also the frequency LVH, CAD and ACCD in anamnesis. Diastolic blood pressure (DBP) "Group 2" was 98 [90; 102] mm Hg (here and further data are presented as median, 25th and 75th quantiles), and in "Group 1" it was 100 [93; 104] mm Hg (p=0.037). Groups of patients were comparable in all blood test characteristics: total cholesterol levels (TC), low density lipids cholesterol (LDL cholesterol), high density lipids cholesterol (HDL cholesterol), triglycerides (TG), creatinine, glomerular filtration rate (GFR), MMP-9 concentration. Initial 24h monitoring of peripheral and central BP and of parameters of vascular rigidity didn't show the differences between the groups of patients.

It is worth to mention that in past all patients didn't receive CB regularly, although high antihypertensive activity and a lot of evidences of this class of drugs is well known. In general, previous antihypertensive therapy didn't differ in the groups of patients.

Table 1. **Comparison of main parameters before the beginning of therapy in the groups of patients (men with AH)**

Parameter	«Group 1» (n=30)	«Group 2» (n=30)	p
Age (years)	50 [42; 59]	50 [39; 58]	0,359
AH duration (years)	8 [5; 10]	5 [3; 15]	0,386
Waist circumference (cm)	105 [100;118]	106 [96; 119]	0,882
Body mass index	29,4 [28,4; 34,7]	29,7 [26,3; 34,5]	0,515
SBP (mm Hg.)	154 [148; 170]	152 [146; 158]	0,358
DBP (mm Hg.)	100 [93; 104]	98 [90; 102]	0,037
Pulse (beats per minute)	72 [62; 75]	70 [65; 78]	1,000
Frequency of smoking	6 (20%)	10 (33%)*	0,191
Number of pack-years	22 [5; 38,3]	30 [18,5; 38,8]	0,408
Frequency of alcohol consumption	24 (80%)	20 (67%)	0,191
Frequency of regular physical activity	0 (0%)	2 (7%)	0,246
Frequency of LVH registration	22 (73%)	16 (53%)	0,090
Frequency of CAD registration	12 (40%)	14 (47%)	0,397
Frequency of регистрации of ACCD	0 (0%)	0 (0%)	1,000
Glucose (mmol/L)	5,1 [3,8; 5,5]	4,9 [4,4; 5,4]	0,614
TC (mmol/L)	5,8 [4,9; 6,7]	5,1 [3,9; 6,3]	0,123
HDL (mmol/L)	1 [0,9; 1,3]	1,04 [0,9; 1,4]	0,572
LDL (mmol/L)	3,2 [2,2; 3,6]	2,8 [2,2; 4,2]	0,836
TG (mmol/L)	2,2 [1,6; 3,2]	1,8 [1,47; 2,6]	0,328
Creatinine (umol/L)	92 [76; 105]	85 [68; 100]	0,076
GFR (mmol/L)	77,5 [64,4; 101,5]	89,7 [65; 112,9]	0,391
MMP-9 (ng/mL)	93,2 [65,1; 125,4]	64 [42,4; 100]	0,051
SBP_{24} (mm Hg.)	139 [132; 156]	138 [128; 145]	0,700
DBP_{24} (mm Hg.)	92 [80; 104]	87 [83; 100]	0,745
PP_{24} (mm Hg)	52 [46; 63]	51 [44; 57]	0,574
Pulse $rate_{24}$ (beats per.)	67 [64; 73]	72 [66; 73]	0,389
AI_{24} (%)	–13 [–20; 7]	–16 [–37; –4]	0,110
$SBPao_{24}$ (mm Hg.)	131 [124; 146]	128 [119; 134]	0,359
$DBPao_{24}$ (mm Hg	94 [80; 105]	88 [85; 102]	0,813
$PPao_{24}$ (mm Hg.)	41 [35; 47]	39 [32; 45]	0,359
$Alao_{24}$ (%)	22 [14; 35]	25 [10; 33]	0,407
$PWVao_{24}$ (m/sec)	9,44 [9,1; 9,9]	9,34 [8,7; 10,7]	0,953

* Here there is relative number of patients in %, in relation to the total number of patients together with the absolute number.

Patients from the "Group 1" received 5+10 mg starting dose of amlodipine and lisinopril with possible increase of dosage up to 10+20 mg respectively. Therapeutic regimen of the "Group 2" considered administration of 50 mg starting dose of extended-release metaprolol tartrate twice per day with further increase of dosage up to 200 mg and higher. During the first week of therapy (during hospital treatment) correction of dosage and, if necessary, addition of other antihypertensive drugs was performed. Characteristic of prescribed therapy is present in

Table 2. **Therapy of patients with AH involved in this study**

Parameter	«Group 1» (n=30), abs.	«Group 2» (n=30), abs.	p
CB	30	0	0,000
β-B	0	30	0,000
Diuretics	6	8	0,753
ACE inhibitors	30	0	0,016
Statins	30	26	0,112
Central-acting agents	1	2	0,862

Table 2. It demonstrates that significant differences between groups exist only in basis therapy provided with the study protocol.

After 6 months of treatment the level of office SBP in the "Group 2" reduced significantly from 152 [145; 158] mm Hg. to138 [128; 144] mm Hg. (p=0.08). DBP levels reduced from 98 [90; 102] mm Hg. to 82 [80; 90] mm Hg. At the same time significant pulse rate reduction from 70 [65; 78] beats per minute to 64 [63; 74] beats per minute existed only during the first months of therapy. The most prominent antihypertensive effect was present after 1 month after the start of active therapy. And some reduction of antihypertensive effect was reported between the third and fourth visits: SBP increased from 128 [120; 138] mm Hg to 138 [128; 144] mm Hg, but nobody had SBP higher than 140 mm Hg.

In the "Group 1" during 6 months of therapy SBP levels reduced from 154 [148; 170] mm Hg to 138 [126; 154] mm Hg, and DBP levels reduced from 100 [93; 10] mm Hg to 82 [80; 96] mm Hg. There was no significant difference in pulse rate. As in the "Group 2", that used extended release metoprolol tartrate, maximal SBP reduction occurred between the first and the second visit. And also there was some "loss" of antihypertensive activity between the third and the fourth visits. During this period of time SBP increased from 120 [118; 136] mm Hg to 38 [126; 154] mm Hg. DBP levels in this group reduced significantly between the first and the second visits and further remained stable in the borders of normal blood pressure.

Partial "loss" of antihypertensive effect on SBP between 3 and 4 visits in both groups can be related to the violation of treatment regimen and not precise following other medical adivices.

It is necessary to mention that there were no significant differences in the grade of BP reduction between visits in both groups (Table 3); it allows to consider that the use of both extended release metoprolol tartrate and amlodipine/lisinopril combination as basic pharmacological agents has comparable efficacy in influencing office BP during 6 months of active therapy. as basic pharmacological agents.

Table 3. **Comparison of antihypertensive effect dynamics in the groups of patients involved in the study**

Parameter	«Group1» (n=30) (mm Hg.)	«Group2» (n=30) (mm Hg.)	p
SBP reduction between 1 and 2 visits	27 [16; 40]	28 [10; 42]	0,554
SBP reduction between 2 and 3 visits	0 [–4; 10]	4 [0; 8]	0,744
SBP reduction between 3 and 4 visits	–12 [–28; –4]	–6 [–16; 0]	0,103
SBP reduction between 1 and 4 visits	16 [–4; 38]	18 [4; 24]	0,882
DBP reduction between 1 and 2 visits	18 [14; 24]	18 [12; 24]	0,406
DBP reduction between 2 and 3 visits	0 [–2; 8]	2 [-8; 10]	0,882
DBP reduction between 3 and 4 visits	–4 [–17; 0]	–4 [–10; 4]	0,744
DBP reduction between 1 and 4 visits	18 [10; 23]	12 [8; 20]	0,172

The results of 6-months treatment with chosen therapeutic regimen influence on 24h-BPM, BP, condition of vascular wall, central BP and some laboratory parameters in the "Group 2" are presented in the Table 4. It was possible to reduce average daily value of SBP and DBP both in brachial artery and aorta. The levels of peripheral average daily BP reduced significantly from 138 [128; 145] / 87 [83; 100] mm Hg. to 129 [125; 136]/ 82 [79; 93] mm Hg. (p<0.05). The levels of average daily central BP reduced sig-

Table 4. **Influence of 6-month therapy in the "Group 1" on 24-hour blood pressure monitoring and some laboratory tests characteristics (n=30)**

Parameter	1st visit	4st visit	p
SBP_{24} (mm Hg)	138 [128; 145]	129 [125; 136]	0,000
DBP_{4} (mm Hg.)	87 [83; 100]	82 [79; 93]	0,003
PP_{24} (mm Hg.)	51 [44; 57]	47 [44; 54]	0,243
$Pulse_{24}$ (beats per minute)	72 [66; 73]	68 [64; 74]	0,041
AI_{24} (%)	–16 [–37; –4]	–21 [–38; –6]	0,194
$SBPao_{24}$ (mm Hg.)	128 [119; 134]	119 [116; 128]	0,000
$DBPao_{24}$ (mm Hg.)	88 [85; 102]	84 [81; 94]	0,005
$PPao_{24}$ (mm Hg.)	39 [32; 45]	36 [33;40]	0,135
$PWVao_{24}$ (m/s)	9,34 [8,7; 10,7]	11,02 [9,7; 11,4]	0,003
$Alao_{24}$ (%)	25 [10; 33]	21 [10; 28]	0,105
Glucose (mmol/L)	4,9 [4,4; 5,4]	4,8 [4,5; 5,4]	0,403
TC (mmol/L)	5,1 [3,9; 6,3]	4,8 [4,3; 5,6]	0,873
TG (mmol/L)	1,8 [1,5; 2,6]	1,4 [1; 2,3]	0,000
HDL (mmol/L)	1,04 [0,9; 1,4]	1,2 [1; 1,9]	0,016
LDL (mmol/L)	2,8 [2,2; 4,2]	2,7 [2; 3]	0,005
Creatinine (μmol/L)	87 [70; 100]	98 [90; 106]	0,096
GFR (ml/min/m^2)	89,7 [65; 112,9]	76,5 [67,5; 88,6]	0,289
MMP-9 (ng/mL)	64 [42,4; 100]	72,76 [42,5; 132,5]	0,232

Note: Negative IA24 value indicates more favorable condition of vessel wall

nificantly from 128 [119; 134]/ 88 [85; 102] mm Hg. to 119 [116; 128]/ 84 [81; 94] mm Hg. ($p<0.05$). Average daily pulse rate also decreased significantly from 72 [66; 73] beats per minute to 68 [64; 74] beats per minute ($p=0.041$). At the same time it was impossible to affect such parameters of vascular rigidity like pulse pressure (PP_{24}), AI24, $AIao_{24}$. More than that, $PWVao_{24}$ significantly increased ($p=0.03$) from 9.34 [8.7; 10.7] m/sec to 11,02 [9,7; 11,4] m/sec (normal value ⩽10 m/sec). Thus, although the antihypertensive effect of extended release metoprolol tatrate 6-months therapy is sufficient, in this study suc h treatment didn't have positive effect on vascular wall properties. The results of other studies prove that metoprolol tartrate is not enough effective for correction of arterial rigidity comparing with other classes of antihypertensive drugs including β-blockers with additional vasodilating properties [24, 25, 30].

In the "Group 1" 6-month therapy reached significant reduction of peripheral and central BP (Table 5). The level of average daily peripheral BP significantly decreased from 139 [132; 156]/ 92 [80; 104] mm Hg to 133 [123; 141]/ 81 [79; 89] mm Hg ($p<0.05$). The level of average daily central BP reduced from 131 [124; 146]/ 94 [80; 105] mm Hg to 126 [115; 127]/83 [80; 90] mm Hg ($p<0.05$). Combined therapy with lisinopril/amlodipine had no significant influence on pulse rate. In this group the parameters of vascular wall properties changed positively: PP24 reduced from 52 [46; 63] mm Hg to 47 [40; 64] mm Hg ($p=0.038$). $PPao_{24}$ reduced from 41 [35; 47] mm Hg to 37 [31; 46] mm Hg ($p=0.042$).

Table 5. **Influence of 6-month therapy in the "Group 1" on 24-hour blood pressure monitoring and some laboratory tests characteristics (n=30)**

Parameter	1st visit	4th visit	p
SBP_{24} (mm Hg)	139 [132; 156]	133 [123; 141]	0,000
DBP_{4} (mm Hg.)	92 [80; 104]	81 [79; 89]	0,001
PP_{24} (mm Hg.)	52 [46; 63]	47 [40; 64]	0,038
$Pulse_{24}$ (beats per minute)	67 [64; 73]	63 [58; 68]	0,212
AI_{24} (%)	–13 [–20; 7]	–16 [–20; –7]	0,006
$SBPao_{24}$ (mm Hg.)	131 [124; 146]	126 [115; 127]	0,001
$DBPao_{24}$ (mm Hg.)	94 [80; 105]	83 [80; 90]	0,001
$PPao_{24}$ (mm Hg.)	41 [35; 47]	37 [31; 46]	0,042
$PWVao_{24}$ (m/s)	9,44 [9,1; 9,9]	9,69 [9,3; 10,6]	0,094
$AIao_{24}$ (%)	22 [14; 35]	20 [7; 32]	0,001
Glucose (mmol/L)	5,1 [3,8; 5,5]	5,4 [4,7; 5,7]	0,101
TC (mmol/L)	5,8 [4,9; 6,7]	5,5 [4,7; 6]	0,314
TG (mmol/L)	2,2 [1,6; 3,2]	2,2 [1,1; 3,4]	0,584
HDL (mmol/L)	1 [0,9; 1,25]	1,6 [1,31; 1,95]	0,000
LDL (mmol/L)	3,2 [2,2; 3,6]	2,9 [2,6; 3,2]	0,112
Creatinine (**μmol/L**)	92 [76; 105]	106 [91; 122]	0,102
GFR (ml/min/m2)	77,5 [64,4; 101,5]	68,5 [58,1; 78,1]	0,110
MMP-9 (ng/mL)	93,2 [65,1; 125,4]	54,79 [43,2; 100,9]	0,015

Note: Negative IA24 value indicates more favorable condition of vessel wall

Average daily AI in aorta also decreased significantly from 22 [14; 35]% to 20 [7; 32]% ($p=0.001$). Average daily AI in brachial artery reduced significantly from $_{(minus)}$ –13 [–20; 7]% to $_{(minus)}$ –16 [–20; –7]% ($p=0.006$). Nevertheless, it was impossible to reach significant PWVao reduction that can be related to short duration of active therapy (6 months). Thereby, fixed combination of amlodipine and lisinopril can lead to significant decrease of average daily peripheral and central BP and melioration of some average daily values of vascular wall properties like PP and AI during 6-month therapy.

Comparison of 6-month treatment with both therapeutic regimens of antihypertensive therapy and its influence on 24-h BPM and characteristics of vascular rigidity is presented at Figure 2.

Characteristics of lipid spectrum improved significantly during treatment (Table 4 and 5). It is worth

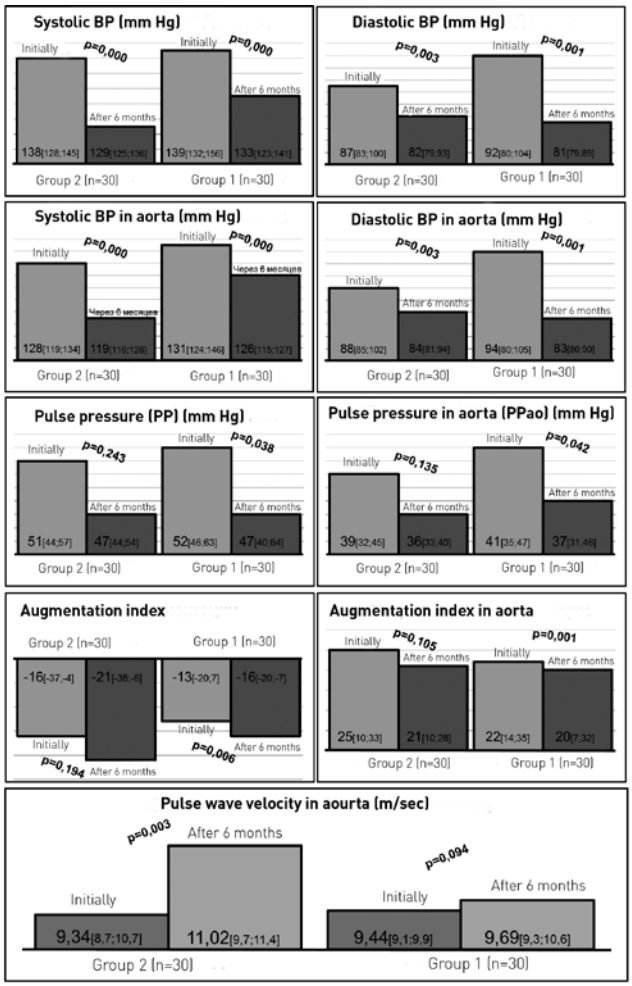

Figure 2. Influence of 6-month therapy on parameters of 24-h monitoring of peripheral and central BP and characteristics of vascular rigidity in both groups of patients with AH

to mention that part of patients received statins if it was indicated. It was not forbidden according with the study protocol and it corresponds clinical guidelines [27].

In addition it is necessary to mention that MMP-9 concentration didn't change significantly during active therapy in the group of patients who received extended release metoprolol tartrate ("Group 2"), whereas the decrease of MMP-9 (from 93.2 [65.1; 125.4] to 54.79 [43.2; 100.9] (p=0.015)) concentration was achieved in the "Group 1" (amlodipine/lisinopril). This correlates with the results of positive influence of fixed drug combination on vascular wall condition (Table 5, Figure 2).

It is important that negative metabolic effects were not registered in both groups during 6-month therapy of AH with chosen drugs. Particularly, starving levels of glucose, creatinine concentration and GFR didn't change negatively (Table 4 and 5).

Quantitative comparison of registered clinically significant adverse reactions that had developed during therapy revealed that they appeared more often in the "Group 1" than in the "Group 2". Particularly, two patients from the "Group 1" developed shin edema during increase of dosage of fixed combination amlodipine/lisinopril (up to 10+20 mg/day) that aimed to reach target BP levels. This adverse reaction required dose reduction up to initial one (5+10 mg/day). One patient of this group had complaints of palpitation that disappeared without assistance after two weeks of therapy without changing the drug dosage. Another patient felt discomfort in epigastrium after drug intake, these adverse reactions didn't require the cancellation of treatment and disappeared without assistance. At the same time no clinically significant adverse reactions were registered in the group of extended release metoprolol tartrate during 6 months of therapy.

There were no fatal outcomes in both groups. At the same time 16 surrogate endpoints were registered. According with the study protocol, these endpoints included: admission to hospital with cardiovascular diseases, death for cardiovascular causes, development of acute coronary syndrome, including AMI, ACCD, atrial fibrillation. 15 cases were related to previously scheduled admission to hospital with cardiovascular diseases and one case was connected with atrial fibrillation paroxysm (In the "Group 2"). In the second group 12 endpoints were registered during six months, whereas in the first one only 4 endpoints were registered (p=0.020).

Conclusion

This study allows to consider more prominent efficacy of hypertensive therapy based on fixed combination of lisinopril and amlodipine comparing with extended-release metoprolol tartrate therapy as a basis antihypertensive medicine. Although both therapeutic regimen were led to reaching similar levels of target BP, the advantage of combined therapy was characterized with positive influence on vascular elasticity and more rare development of "surrogate" endpoints. Particularly, the group that had been treated with fixed combination of amlodipine and lisinopril demonstrated upregulation of MMP-9 linked with AI reduction. The results of our study together with literature analysis allow to recommend fixed combination of lisinopril and amlodipine as the preferable one in treatment of male patients with AH and impaired vascular wall elasticity.

Conflict of interest: None declared.

References

1. Lopatin YM, Ilyukhin OV. Control of vascular stiffness. The clinical significance and methods of correction. Heart. 2007;6 (3): 128–32. Russian
2. Nedogoda SV, Chalabi TA. Vascular stiffness and the propagation velocity of the pulse wave: emerging risk factors for cardiovascular disease and targets for drug therapy. Current issues diseases of the heart and blood vessels. 2006;4:33–49. Russian
3. Ichihara A, Hayashi M, Koyra Y, et al. Long-term effects of intensive blood pressure lowering on arterial wall stiffness in hypertensive patients. Am J Hypertens. 2003;16 (11): 959–65.
4. Laurent S, Cockroft J, Van Bortel L, et al. Expert consensus document on arterial stiffness: methodological issues and clinical application. Eur Heart J. 2006;27 (21): 2588–605.
5. Asmar R, Topouchian J, Pannier B, et al. Pulse wave velocity as endpoint in large-scale intervention trial. The Complior study. Scientific, Quality Control, Coordination and Investigation Committees of the Complior study. J Hypertens. 2001;19 (4): 813–8.
6. Rajzer M, Klocek M, Rawecka-Jaszcz K. Effect of amlodipine, quinapril, and losartan on pulse wave velocity and plasma collagen markers in patients with mild-to-moderate arterial hypertension. Am J Hypertens. 2003; 16 (6): 439–44.
7. Polonia J, Barbosa L, Silva JA, et al. Different influences on central and peripheral pulse pressure, aortic wave reflections and pulse wave velocity of three different types of antihypertensive drugs. Rev Port Cardiol. 2003;22 (12): 1485–92.
8. Kutishenko NP, Martsevich SY. Lisinopril in cardiology practice: evidence-based data. Rational pharmacotherapy in cardiology. 2007;5:79–82. Russian

9. Isakova VN, Garbuzova OG, Klinkova EV, et al. Parameters of arterial stiffness in patients with medium/high risk for cardiovascular disease during therapy with lisinopril and simvastatin. Rational Pharmacotherapy in Cardiology. 2009;3:14–8. Russian
10. Kosheleva NA, Rebrov AP. Peculiarities of the processes of remodeling of heart and vessels in patients with heart failure on a background of 6-month therapy with lisinopril. Rational Pharmacotherapy in Cardiology. 2010;6 (3): 323–8. Russian
11. Shilova EV, Martsevich SY. Dihydropyridine calcium antagonists: their role in modern therapy of cardiovascular diseases. Rational pharmacotherapy in cardiology. 2008;2:53–7. Russian
12. Kharkevich DA. Pharmacology. Tutorial. The tenth edition. Moscow.: GEOTAR-Media; 2010. Russian
13. Runchina NK, Tkacheva ON. Amlodipine: the ability to reduce the risk of complications of hypertension. Systemic hypertension. 2009;4:15–20. Russian
14. ALLHAT Officers and Coordinators for the ALLHAT Collaborative Research Group. The antihypertensive and lipid-lowering treatment to prevent heart attack trial. Major outcomes in high-risk hypertensive patients randomized to angiotensin-converting enzyme inhibitor or calcium channel blocker vs diuretic: the antihypertensive and lipid-lowering treatment to pre-vent heart attack trial (ALLHAT). JAMA. 2002;288:2981–97.
15. Walter MF, Jacob RF, Bjork RE, et al. Circulating lipid hydroperoxides predict cardiovascular events in patients with stable coronary artery disease: the PREVENT study. JACC. 2008;51 (12): 1196–202.
16. Karoli NA, Rebrov AP, Roshchina AA. Efficacy and safety of amlodipine maleate in patients with chronic obstructive pulmonary disease and bronchial asthma with concomitant arterial hypertension. Rational Pharmacotherapy in Cardiology. 2010;6 (2): 173–8. Russian
17. Dahlof B, Sever PS, Poulter NR, et al. Prevention of cardiovascular events with an antihypertensive regimen of amlodipine adding perindopril as required versus atenolol adding bendroflumethiazide as required, in the Anglo–Scandinavian Cardiac Outcomes Trial–Blood Pressure Lowering Arm (ASCOT–BPLA): a multicenter randomized controlled trial. Lancet. 2005;366:895–906.
18. Williams B, Lacy PS, Thom SM, et al. Differential impact of blood pressure-lowering drugs on central aortic pressure and clinical outcomes. Principal results of the Conduit Artery Function Evaluation (CAFÉ) study. Circulation. 2006;113 (9): 1213–25.
19. Jamerson K, Weber MA, Bakris GL, et al. Benazepril plus amlodipine or hydrochlorothiazide for hypertension in high–risk patients. N Engl J Med. 2008;359:2417–28.
20. Boytsov SA, Linchak RM. Combination antihypertensive therapy: ACE inhibitor plus calcium antagonist. New advantages of the known combination. Rational Pharmacotherapy in Cardiology. 2010;6 (1): 89–94. Russian
21. Zadionchenko VS, Shehan GG, Timofeeva NY, et al. Results of clinical studies of the drug Equator in the treatment of hypertension. Russian medical J. 2012;11:554–9. Russian
22. Ostroumova OD, Smolyarchuk EA, Hvorostianaya IV. New approaches to the treatment of arterial hypertension: the choice of the optimal preparation to the selection of optimal combinations. Rational pharmacotherapy in cardiology. 2010;6 (5): 709–16. Russian
23. Kobalava JD, Kotovskaya JV, Troitskaya EA. Combination of the blocker Renin-Angiotensin system and of the dihydropyridine calcium antagonist in the treatment of hypertension. Russian medical J. 2010;18 (3): 123–6. Russian
24. Luca ND, Asmar RG, London JM, et al. Improvement in blood pressure, arterial stiffness and wave reflection with a very-low-dose perindopril/indapamide in hypertensive patients: a comparison with atenolol. Hypertension. 2001;38:922–6.
25. Bangalore S, Sawhney S, Messer-li FH. et al. Relation of beta-blocker-induced heart rate lowering and cardioprotection in hypertension. JACC. 2008;52:1482–9.
26. Oleynikov VE, Matrosova IB, Tomashevskaya YA, et al. Influence of treatment with metoprolol on arterial stiffness. Rational Pharmacotherapy in Cardiology. 2011;7 (6): 685–9. Russian
27. Chazova IE, Oschepkova EV, Zhernakova Yu.V, et al. Diagnosis and treatment of hypertension (clinical practice guidelines). Kardiologicheskij vestnik. 2015;1:3–30. Russian
28. Rogoza AN, Kuznetsov AA. Central aortic blood pressure and augmentation index: comparison between Vasotens and SphygmoCor. Research Reports in Clinical Cardiology. 2012;3:27–33.
29. Rogova LN, Shesternina NV, Zamecnik TV, et al. Matrix metalloproteinases, and their role in physiological and pathological processes (review). Bulletin of new medical technologies. 2011;18 (2): 86–9. Russian
30. Sirenko YN, Recovec OL, Kushnir SN, et al. Comparative efficacy of nebivolol and bisoprolol in terms of influence on Central blood pressure, and elastic-elastic properties of arteries in patients with mild and moderate arterial hypertension. Arterial hypertension. 2013;1 (27): 9–19. Russian

International Heart and Vascular Disease Journal
Volume 4, Number 9, March 2016

Journal of the Cardioprogress Foundation

Impaired regulation of genome stability may be the key mechanism of left ventricular hypertrophy development in arterial hypertension

Minushkina L.O.[1*], Brazhnik V.A.[2], Nikitin A.G.[3], Nosikov V.V.[3], Zateishchikov D.A. [1,2,3]

[1] Central State Medical Academy of the Department for Presidential Affairs of the Russian Federation, Moscow, Russia
[2] City Clinical Hospital №51, Moscow, Russia
[3] Federal Clinical Research Center of Specialized Types of Health Care and Medical Technologies, Federal Biomedical Agency of Russia, Moscow, Russia

Authors:

Larisa O. Minushkina, M.D., doctor of sciences, professor of therapy, cardiology and functional diagnostics department of Central State Medical Academy of the Department for Presidential Affairs of the Russian Federation, Moscow

Victoria A.Brazhnic, M.D., Ph.D., head of City Clinical Hospital №51, assistant professor of therapy, cardiology and functional diagnostics department of Central State Medical Academy of the Department for Presidential Affairs of the Russian Federation, Moscow

Alexei G. Nikitin, Ph.D., head of the laboratory of genetics of Federal Clinical Research Center of Specialized Types of Health Care and Medical Technologies, Federal Biomedical Agency of Russia, Moscow

Valery V. Nosikov, doctor of sciences, professor, head of the laboratory of genetics of Federal Clinical Research Center of Specialized Types of Health Care and Medical Technologies, Federal Biomedical Agency of Russia, Moscow

Dmitry A. Zateishchicov, M.D., doctor of sciences, professor, head of primary vascular department of City Clinical Hospital №51, professor of therapy, cardiology and functional diagnostics department of Central State Medical Academy of the Department for Presidential Affairs of the Russian Federation, leading researcher of the laboratory of genetics of Federal Clinical Research Center of Specialized Types of Health Care and Medical Technologies, Federal Biomedical Agency of Russia, Moscow

* Corresponding author. Tel. 84955300517, 89036738976. E-mail: minushkina@mail.ru

Summary

Objective

To investigate association between PPAR gene family polymorphisms and PARP, PARG and NOS3 genes with left ventricular hypertrophy (LVH) in patients with arterial hypertension (AH).

Materials and methods

This study involved 2012 patients, 127 of them had LVH. We performed transthoracic echocardiography and used determination of alleles and genotypes of polymorphic candidate genes using phenol-chloroform DNA extraction from venous blood of patients. Amplificator "Tercic" ("DNA-technology, Russia) has been used for polymorphic genetic loci amplification. Statistical analysis has been performed with SPSS software.

Results

We demonstrated the association of LVH with 4a allele of NOS3 (OR 1,68, p=0.016) and GC genotype of PARG gene (OR 3.61, p=0.024). Multifactor regression analysis demonstrated independent relationship of left ventricular hypertrophy with 4a NOS3 allele, GG genotype of PARG gene, patient's age and maximal levels of systolic blood pressure.

Conclusion

Impaired balance of processes that lead to genome destabilization/stabilization may be one of the mechanisms responsible for LVH developing in patients with AH.

Key words

PARG, NOS3, arterial hypertension, left ventricular hypertrophy.

Modern guidelines for management of patients with arterial hypertension (AH) mark out target organs lesions like left ventricular hypertrophy (LVH), hypertensive nephropathy as a separate problem and suggest to put much diagnostic efforts into their detection [1]. These lesions are referred to additional risk factors that negatively influence patients' prognosis. Lack of strict correlation between level, severity, duration of AH and the beginning of developing target organ lesions proves that some additional causes influence the formation of these complications. Recently discovered new experimental data demonstrate that DNA (deoxyribonucleic acid) stability regulation can play a key role in this process. It is supposed to think that NO (nitrogen oxide) causes activation of peroxide oxidation that leads to peroxynitrite synthesis. DNA is identified to be one of the targets of peroxynitrite. NO-synthases expression is regulated by PPAR (peroxisome proliferator-activated receptors) family of nuclear receptors. The opposite process of DNA repair starts with involvement of poly ADP(adenosine diphosphate) ribose polymerase I type (PARP I) [2] and poly ADP ribose glycohydrolase (PARG). Changes of genome stability are actively investigated as a possible pathogenetic mechanism of various diseases. There are some evidences proving the role of these mechanisms in development of AH complications [3]. Associative genetic approach allows to test the hypothesis of the role of the protein of interest in pathogenesis of disease by studying patients with different genotypes of this protein that influence differently its activity.

According with this, the current study aimed to investigate possible association of PPAR nuclear receptor family genes polymorphic markers and endothelial NO-synthase with developing LVH in AH.

Characterization of patients and methods

This study has been approved by local ethic commitee. This study involved 212 patients with AH. Exclusion criteria were lack of patient's to participate in study, presence of myocardial scars and evident valvular heart disease.

Clinical characterization of patients 94 male patients (44.3%) and 118 female patients (55.7%). Average age of patients: 60.23 ± 0.74 years, AH duration at the moment of examination:– 14.2±0.79 years. 22 patients(10.4%) at the moment of inclusion into study had AH stage 1, 67 patients (31.6%) had AH stage 2, and 123 patients (58%) had AH stage 3. 115 patients (54.2%) were diagnosed with coronary artery disease (CAD), 35 (16.5%) were diagnosed with diabetes mellitus type 2, 17 (8.1%) survived stroke. Average body mass index (BMI) was 29.2±0.34 kg/m^2, 168 (79.2%) patients had excessive body weight, 37 (17.4%) patients had glomerular filtration rate (GFR) < 60 ml/min.

Methods. End-diastolic dimensions (EDD), end-systolic dimensions (ESD), interventricular septum thickness (IST), posterior left ventricular wall thickness

Table 1. **Investigated candidate genes**

Candidate gene	Polymorphic marker	Genotype frequency distribution		χ^2, p
		Observed	Expected (according with Hardy Weinberg principle)	
Endothelial NO-synthase gene (NOS3)	4a/4b Glu298Asp	4b4b-68 4a4b-101 4a4a-5	80,7 57,6 17,7	19,65 <0,001
Peroxisome proliferator-activated receptor α gene (PPARA)	C24313G	CC-150 CG-56 GG-6	149,4 57,1 5,5	0,08
Peroxisome proliferator-activated receptor γ2 gene (PPARG2)	Pro12Ala	Pro/Pro-149 Pro/Ala-53 Ala/Ala-8	146,6 57,6 5,67	1,37
Peroxisome proliferator-activated receptor γ3 gene (PPARG3)	C(–681)G	CC -104 CG -48 GG -12	99,9 56,2 7,9	3,49
Peroxisome Proliferator-Activated Receptor Gamma, Coactivator 1 Alpha gene (PPARGC1A)	Gly482Ser	Gly/Gly -71 Gly/Ser- 83 Ser/Ser-10	77,2 70,6 16,2	5,01 <0,05
Peroxisome Proliferator-Activated Receptor Delta gene (PPARD)	T(–87)C	CC -59 CT -26 TT -79	31,6 80,8 51,6	75,4 <0,001
poly(ADP-ribose) polymerase 1gene (ADPRT1)	Leu54Phe	Leu/Leu -44 Leu/Phe -62 Phe/Phe- 58	34,3 81,4 48,3	9,32 <0,005
	Val762Ala	Ala/Ala-127 Ala/Val-28 Val/Val-9	121,2 39,5 3,2	13,98 <0,001
Poly(ADP-ribose) glycohydrolase gene (PARG)	A(–431)G	AA-97 AG-48 GG-19	89,3 68,5 11,2	9,72 <0,005

(PLVWT) were evaluated using transthoracic echocardiography. This measurement was performed in M-mode on the level of mitral valve chords and parasternal long axis view. Ejection fraction (EF) was determined using Simpson's formula in apical 4-chamber position. Left ventricular myocardium mass (LVMM) was measured using Devereus RB formula [5], $LVMM=1,04*[(IST+PLWT+EDD)^3 – EDD^3]-13,6$.

Left ventricular myocardium mass index (LVMMI) was quantified as the LVMM ratio to body surface area. LVMMI >95 g/m^2 was considered as LVH for woman and >110g/m^2 for men respectively.

Phenol-chloroform extraction of genomic DNA from venous blood of patients was used for determination of alleles and genotypes of polymorphic candidate genes Amplificator "Tercic" ("DNA-technology, Russia) was used for polymorphic genetic loci amplification. Agarose gels were stained with ethidium bromide and polyacrilamide gels were stained with silver nitrate. Investigated candidate genes are listed in Table 1.

Statistical analysis Statistical analysis was performed using standard package of SPSS software. For quantitative variables average values and errors of average were quantified. Statistical analysis was done using Mann-Whitney and Kruskal-Wallis tests. Discrete variables were estimated using Pearson's chi-squared test x^2 . When expected number of observations in any square of the contingency table was <5 we used Fisher's exact test and used p-value derived from two-sided test. Independent influence of clinical and genetic factors on LVH degree was estimated with logistic regression. Clinical factors that had significant relation with AH clinical course according with single-factor regression analysis (p<0,05) were included into multifactor regression analysis. Binary logistic regression with Wilks test has been used as multifactor analysis approach. for all tests p-value <0.05 was considered significant. Accordance between observed genotype frequencies and expected ones quantified using Hardy-Weinberg equilibrium was checked with online-calculator. (http://www.oege.org/software/hardy-weinberg.html).

Results

Between observed patients 127 had LVH, 85 patients had no signs of LVH. Patients with LVH were older, there were more female than male between them, these patients had longer AH duration and higher numbers of maximal systolic blood pressure (SBP) (Table 2).

Significant differences in the frequency of alleles and genotypes of polymorphic markers of *PPARG2, PPARG3, PPARA, PPARGC1A, PARP1* genes in the

Table 2. **Clinical characterization of patients**

Parameter	All patients (n=212)	Patients without LVH (n=85)	Patients with LVH (n=127)	p
Gender male/ female	94/118	49/36	45/82	0,001
Age, years	60,2±0,74	54,8±1,04	63,8**±0,**93	0,01
Diabetes mellitus type 2, n(%)	35 (16,5)	9(10,6)	26(20,5)	ns
AH duration, years	14,2±0,79	10,9±0,92	16,7±1,15	0,001
BMI, kg/m	29,2±0,34	28,7±0,44	29,5±0,22	ns
Excessive body weight, n(%)	168 (79,2)	63(74,1)	105(82,7)	ns
SBP max, mm Hg..	198,3±1,53	186,9±3,27	205,9±1,71	0,01
DBP max mm Hg.	110,9±0,79	108,3±1,84	112,8±0,86	ns
GFR, ml\min	81,36±1,43	83,5±2,63	77,2**±1,69**	ns
GFR < 60 ml / min, n(%)	37 (17,4)	12(14,1)	25(19,6)	ns
Stroke, n(%)	17 (8,1)	4(4,7)	13(10,2)	ns
CAD, n(%)	115 (54,2)	40(47,1)	75(59,1)	ns

Comments: DBD – diastolic blood pressure, ns – not significant

groups of patients with and without LVH were not present (Table 1).

Distribution of polymorphic markers of PPARA, PPARG2, PPARG3 genes frequencies corresponded to Hardy-Weinberg equation. Other markers declined from expected distribution (Table 1).

Genotype frequencies of polymorphic markers *PPARG2, PPARG3, PPARA, PPARGC1A, PARP1, ADPRT1* genes had no significant differences between patients with LVH and without LVH (Table 3).

Table 3. **The frequency of polymorphic markers of genes alleles and genotypes polymorphic markers of genes expression products of which participate in metabolic regulation in patients with and without LVH**

	No LVH n= 85	LVH n= 127	p	*OR[95%CI]*
Polymorphic marker *C24313G* of *PPARA* gene				
Genotypes CC CG GG	 61 (71,8%) 23 (27,1%) 1 (1,2%)	 89 (70,1%) 33 (26,0%) 5 (3,9%)	 ns ns ns	 1,01[0,59-2,04] 0,94[0,51-1,76] 3,34[0,39-30,00]
Alleles: *C* *G*	145 (85,3%) 25 (14,3%)	211 (83,1%) 43 (16,9%)	ns ns	0,84[0,49-1,84] 1,18[0,69-2,02]
Polymorphic marker *Pro12Ala* of *PPARG2* gene				
Genotypes Pro/Pro Pro/Ala Ala/Ala	 64 (75,3%) 18 (21,2%) 3 (3,5%)	 85 (67,5%) 36 (28,6%) 5 (4,0%)	 ns ns ns	 0,68[0,36-1,86] 1,48[0,77-2,84] 1,04[0,44-4,48]
Alleles: Pro Ala	146 (85,9%) 24 (14,1%)	206 (81,7%) 46 (18,3%)	ns ns	0,73[0,23-1,45] 1,35[0,79-2,32]
Polymorphic marker *C(–681)G* of *PPARG3* gene				
Genotypes *CC* *CG* *GG*	 44 (64,7%) 19 (27,9%) 5 (7,4%)	 69 (63,3%) 33 (30,3%) 7 (6,4%)	 ns ns ns	 0,94[0,50-1,76] 1,12[0,57-2,18] 0,84[0,26-2,84]
Alleles: *C* *G*	107 (77,5%) 29 (22,5%)	171 (78,4%) 47 (21,6%)	ns ns	0,98[0,58-1,66] 1,01[0,60-1,70]
Polymorphic marker *T(–87)C* of *PPARD* gene				
Genotypes *CC* *CT* *TT*	 23 (33,8%) 18 (26,5%) 27 (39,7%)	 39 (35,8%) 13 (11,9%) 57 (52,3%)	 ns 0,012 ns	 1,09[0,57-0,06] 0,36[0,16-0,81] 1,66[0,90-3,07]
Alleles *C* *T*	64 (47,1%) 72 (52,9%)	91 (41,7%) 127 (58,3%)	ns ns	0,80[0,52-1,24] 1,24[0,80-1,90]
Polymorphic marker *Gly482Ser* of *PPARGC1A* gene				
Genotypes *Gly/Gly* *Gly/Ser* *Ser/Ser*	 29 (42,6%) 36 (52,9%) 3 (4,4%)	 47 (43,1%) 54 (49,5%) 8 (7,3%)	 ns ns ns	 1,01[0,55-1,88] 0,87[0,47-1,60] 1,71[0,44-6,70]
Alleles: *Gly* *Ser*	94 (69,1%) 42 (30,9%)	148 (64,9%) 70 (35,1%)	ns ns	0,94[0,59-1,49] 1,05[0,67-1,66]
Polymorphic marker *Leu64Phe* of *ADPRT1* gene				
Genotypes *Leu/Leu* *Leu/Phe* *Phe/Phe*	 16 (23,5%) 25 (36,8%) 27 (39,7%)	 31 (28,4%) 42 (38,57%) 36 (33,0%)	 ns ns ns	 1,54[0,77-3,06] 1,32[0,72-2,45] 0,74[0,39-1,40]
Alleles *Leu* *Phe*	 57 (41,9%) 79 (58,1%)	 104 (47,7%) 114 (52,3%)	 ns ns	 1,26[0,82-1,94] 0,79[0,51-1,21]
Polymorphic marker *Val762Ala* of *ADPRT1* gene				
Genotypes *Ala/Ala* *Ala/Val* *Val/Val*	 50 (73,5%) 15 (22,1%) 3 (4,4%)	 87 (79,8%) 16 (14,7%) 6 (5,5%)	 ns ns ns	 1,42[0,69-2,90] 0,60[0,27-1,32] 1,26[0,30-5,22]
Alleles *Ala* *Val*	 115 (84,6%) 21 (15,4%)	 180 (86,5%) 28 (13,5%)	 ns ns	 1,17[0,63-2,16] 0,85[0,47-1,57]
Polymorphic marker *A(–431)G* of *PARG* gene				
Genotypes *AA* *AG* *GG*	 44 (64,7%) 21 (30,9%) 3 (4,4%)	 61 (56,0%) 32 (29,4%) 16(14,7%)	 ns ns 0,024	 0,69[0,27-1,29] 0,93[0,48-1,79] 3,61 [1,21-12,91]
Alleles *A* *G*	 109 (80,1%) 27 (19,9%)	 154 (70,6%) 64 (29,4%)	 0,03 0,03	 0,27 [0,07-0,98] 1,64[1,01-2,67]
Polymorphic marker *4a/4b* of *NOS3* gene				
Genotypes *4b/4b* *4b/4a* *4a/4a*	 36 (53,7%) 30 (44,8%) 1 (1,5%)	 38 (33,3%) 72 (63,2%) 4 (3,5%)	 0,005 0,012 ns	 0,43 [0,23-0,79] 2,10 [1,14-3,86] 2,36[0,26-23,53]
Alleles *4b* *4a*	 102 (76,1%) 32 (23,9%)	 148 (64,9%) 80 (35,1%)	 0,016 0,016	0,59 [0,37 – 0,93] 1,68 [1,07-2.62]
Polymorphic marker *Glu298Asp* of *NOS3* gene				
Genotypes *Glu/Glu* *Glu/Asp* *Asp/Asp*	 41 (62,1%) 24 (36,4%) 1 (1,5%)	 62 (52,5%) 52 (44,1%) 4 (3,4%)	 ns ns ns	 0,67[0,36-1,24] 1,37[0,74-2,56] 2,24[0,24-20,84]
Alleles *Glu* *Asp*	 106 (80,3%) 26 (19,7%)	 176 (74,6%) 60 (25,4%)	 ns ns	 0,72[0,42-1,21] 1,39[0,82-2,33]

Patients with LVH had significantly higher frequency of 4a allele of polymorphic marker of NOS3 gene (p=0.016, OR 1.68 [1.07-2.62]). These patients had significantly higher frequency of GG polymorphic marker *A(–431)*G of PARG gene (p=0.024) [OR 3.61 CI 1.21-12.91]. The frequency of A allele was significant-

Table 4. **Echocardiography (EchoCG) results in relation to PARG, PPARA and NOS3 genotypes**

EchoCG parameter	4a|4b of NOS3 gene		C24313G of PPARA gene		A(–431)G of PARG gene	
	Genotype *4b/4b* (n=74)	Genotypes *4a/4a* and *4a/4b* (n=107)	Genotype CC (n=150)	Genotypes CG and GG (n=62)	Genotypes AA and AG (n=158)	Genotype GG (n=19)
PLVWT, cm	1,10±0,050	1,22±0,025	1,19±0,020	1,11±0,024	1,16±0,016	1,23±0,051
p	0,017	0,045	нд			
IST, cm	1,12±0,023	1,21±0,022	1,17±0,017	1,09±0,024	1,14±0,015	1,21±0,048
p	0,004	0,014	ns			
EDD, cm	4,79±0,077	4,85±0,058	4,82±0,047	4,81±0,063	4,82±0,044	5,00±0,154
p	ns	ns	ns			
EF, %	58,5±0,89	56,5±1,04	56,3±0,77	58,5±1,11	55,50±0,72	57,3±02,92
p	ns	ns	ns			
LVMM, g	245,3±9,25	270,6±9,09	262,3±7,54	236,8±9,25	251,9±6,46	298,6±26,50
p	0,053	0,051	0,025			
LVMMI, g/m^2	127,4±4,65	144,6±4,44	138,5±3,70	125,5±4,33	133,8±3,24	157,6±20,02
p	0,032	0,044	0,023			

ly lower [OR 0.27 CI 0.07-0.98], and the frequency of G allele – significantly higher (OR=1.64[1.01-2.67]) comparing with the group of patients without LVH. In the group of patients with LVH the frequency of *T(–87)C* marker heterozygote genotype of PPARD gene was significantly lower.

We also compared main characteristics of left ventricle myocardium in patients with different genotypes of investigated polymorphic markers. Significant differences were obtained just for *NOS3*, *PARG* and *PPARA* genes (Table 4).

It was demonstrated that for polymorphic marker *A(–431)G* of PARG gene patients with rare genotype GG have significantly higher LVMM and LVMMI comparing with the patients with A allele. The association of this marker and systolic and diastolic function parameters was not identified. There were no differences in condition of LV systolic function.

It was shown that for polymorphic marker *C24313G* of *PPARA* gene carriers of CC genotype have significantly more thick walls of LV myocardium, LVMM and LVMMI.

It was demonstrated that in case of polymorphic marker *4a/4b* of NOS3 gene patients who carry 4a allele have significantly more thick walls of LV myocardium and LVMMI.

To evaluate independence of clinical and genetic factors influence on LVH risk we performed regression analysis (Table 5). Single-factor regression analysis demonstrated that male gender, age, SBP levels and NOS3 gene polymorphism were related to LVH development. factors that had significant connection with LVH according with single-factor analysis were included into multifactor analysis.

Multifactor analysis revealed that the presence of 4a allele of 4a/4b polymorphic marker of NOS3 gene, GG genotype of polymorphic marker *A(–431)*G of PARG gene, age of patients and maximal SBP levels in patients with AH are associated independently with LVH

Discussion

According with modern ideas, genome stability is connected with several simultaneous processes. First of them is activity of factors that destabilize DNA, for example peroxynitrite, second – activity of DNA repair, key regulator of which is PARP1 and PARG interaction. Regulation of all these processes is another important factor o genome stability.

Our study demonstrated association of polymorphic markers of NOS3, PPARA, PARG genes with developing LVH in patients with AH. This association proves that LVH development is not only the di-

Table 5. **Clinical and genetic factors that influence independently LVH developing**

Factor	OR (single-factor analysis)	p	OR (multifactor analysis)	p
Male gender	2,59 [1,86-5,72]	0,0001		ns
Age	1,09 [1,02-1,14]	0,0001	1,12 [1,07-1,17]	0,0001
SBP max levels	1,03 [1,01-1,06]	0,001	1,18 [1,02-1,58]	0,023
Allele *4a* of polymorphic marker *4a/4b* of *NOS3* gene	2,32 [1,34-4,11]	0,008	2,58 [1,09-6,09]	0,031
Genotype *GG* of polymorphic marker *A(–431)*G of *PARG* gene	3,72[1,04-13,72]	0,043	8,52 [1,71-42,38]	0,028

rect consequence of increased hemodynamic load on myocardium, but also is the result of impaired balance of factors that maintain genome stability.

NO from one side is considered to be one of the key endothelial factors that regulate vascular tone, from another side it is one of the toxic factors that damage tissues and trigger apoptosis [4]. NO is synthesized from L-arginine by NO-synthase family of enzymes in several tissues. NO-synthase 3 type (NOS3) is responsible for NO production in endothelium where NO activates guanylyl cyclase system and works either as the main vasodilating factor or interacts with peroxide forming peroxynitrite. Peroxynitrite has strong genotoxic effect and it has significant role in poly(ADP-ribose) polymerase expression regulation. Association of polymorphic markers genotypes 4a/4b of NO-synthase gene with LVH development was demonstrated before [7], and in this study it has been proved in a big group of patients. This polymorphism is associated with increased level of basal NO secretion and reduced release of NO as a response to stimuli that activate NOS3, by this creating favorable conditions for peroxynitrite formation [8].

Peroxisome proliferator activating receptors (PPAR) are nuclear receptors that regulate transcription. Apart of it their stimulation can change NO-synthases activity. These receptors are present in 3 isoforms – alpha, gamma and beta/delta. Each of them is coded by its own gene (PPARA, PPARG, PPARD). Each isoform has tissue and substrate specificity. These receptors regulate proliferation, angiogenesis, inflammation, lipid metabolism and lipid peroxidation. PPARA cardioprotective action hasn't been fully understood so far. It has been shown in cell cultures that PPARA reduces cardiomyocyte proliferation in response to endothelin [9]. One of possible mechanisms of this protection, including protection from LVH, can be turning on the mechanism of PPARA-mediated inhibition of apoptosis stimulated with insulin-like growth factor [5, 10]. Another possible way to influence LVH with PPARA activation can be related to sirtuin1(Sirt1), important mediator of energetic metabolism [11]. Sirt1 participates in protein deacetylation and regulates activity of different processes, including NOS3 activity [12]. Important feature of its action is that its substrate NAD+(Nicotinamide adenine dinucleotide) is used also for DNA repair. According with some studies, these processes compete for restricted amount of NAD+. Administration of PPARA blockers SIRT1 effects to LVH development disappear [6]. PPARA activation prevents the development of myocardial fibrosis [13].

One of possible mechanisms of PPARA cardioprotective action in relation to LVH can be its interaction with NO-sythases. PPARA agonist fenofibrate that is used as lipid-lowering agent reduces bronchial response to methacholine, action of which is related with insufficient activity of NO-synthases [14]. Alpha type receptor is expressed mostly in the heart. Gamma type receptors have coactivators, proteins that cause receptor's conformational change and participate in its activation. Alpha1 coactivator is expressed mainly in cardiac tissue and participates in cardiomyocyte energetic metabolism.

PPARA role in LVH is proved with clinical evidences. Previously it has been shown that LVH hypertrophy is associated with CC genotype of C24313G polymorphic marker of PPARA gene [15]. In our study this association has been confirmed for another time in a big group of patients.

Majority of works that investigated LVH development aimed to prove the participation of other nuclear receptors of PPAR family and this relation hasn't been confirmed. Likely it can be explained with low functional significance of selected polymorphisms.

PARP1 is the sensor of DNA damage and starts DNA repair process [16]. PARP1 binds intensively single and double strand DNA breaks that were formed as the result of direct DNA damage or during DNA repair as a result of enzyme action. Further poly(ADP-ribose) synthesis precedes the beginning of damaged DNA repair. At the same time poly(ADP-ribose) promotes apoptosis. Change of poly(ADP-ribose) polymerase activity can lead to hereditary retinal dystrophy. and predisposes to several cancers and autoimmune diseases [17]. PARP family genes activation mediates cell protection from genotoxic, oxidative and other agents. Probably PARP participates in some metabolic processes, particularly in lipid metabolism, Poly(ADP-ribose)polymerases family can be associated with myocardial hypertrophy development [18]. Some myocardial hypertrophy mediators ,like angiotensin II, interleukin-6, are activators of PARP family enzymes, and it is possible that activation of this system mediates LVH development. This fact allowed to consider the association of PARP polymorphism with developing LVH.

PARP1 gene is located in 13q34 chromosome. ADPRT1 gene that codes poly(ADP-ribose)polymerase PARP1 contains two functionally different parts: N-terminal DNA-binding domain and

C-terminal catalytic domain. There is an automodification domain between them. Several polymorphisms are known for this gen, Leu54Phe (located at exon 2) and Val762Ala (located at exon 17 in the beginning of catalytic domain) are the best investigated ones. Val762Ala polymorphic marker is associated with increased risk of several oncologic diseases development [19], Leu54Phe marker is associated with the risk of diabetic nephropathy development [20]. It was shown in experiments that PARP1 can participate in myocardial lesions formation and myocardial hypertrophy [21]. It was demonstrated that PARP1 blockers can prevent LVH development in animal models and in the culture of cardiomyocytes [22, 23]. Clinical data that would be able to prove this hypothesis are still absent. Results related to more studied polymorphic markers didn't demonstrate the association between LVH developing and PARP1 polymorphism.

Poly(ADP-ribose)glycohydrolase is a physiological antagonist of poly(ADP-ribose)polymerase. Poly(ADP-ribose)glycohydrolase is responsible for degradation of poly(ADP-ribose) that is the product of PARP family enzymes. Poly(ADP-ribose) chains that are synthetized in nuclei as a response to mutagenic factors dedrade during 1-2 minutes after termination of their synthesis because of PARG action

This enzyme's function is related to apoptosis system. Poly (ADP-ribose)glycohydrolase slows down apoptosis. The main catalytic center of poly (ADP ribose) glycohydrolase is complementary to ADP-ribose. Poly (ADP ribose) glycohydrolase is located at 10q11.23 chromosome. It is known that PARG activity increases as a response to ischemia. It has been shown that increased expression of this gene in the brain of ischemic mice, and also in abdominal organs if mesenteric artery is ischemic. So far there were no data about PARG gene polymorphic markers association with human disease pathogenesis. This study demonstrated that carrying G allele of polymorphic marker A(-431)G of PARG gene predisposed to developing LVH. Reduced activity of PARG and impaired degradation of ADP-ribose that makes cells more sensitive to growth factor action can be a possible mechanism of this phenomenon.

The limitation of this study was comparably small number of patients. But the results of this study can become a foundation for further studies in this field.

Thus one of the mechanisms responsible for developing LVH in patients with AH can be impaired balance of processes that lead to genome destabilization/stabilization.

Conflict of interest: None declared

References

1. Mancia G, Fagard R, Narkiewicz K, et al. 2013 ESH/ESC Guidelines for the management of arterial hypertension: The Task Force for the management of arterial hypertension of the European Society of Hypertension (ESH) and of the European Society of Cardiology (ESC). Eur Heart J. 2013; 34(28):2159-219.
2. Ko HL, Ren EC. Functional Aspects of PARP1 in DNA Repair and Transcription. Biomolecules. 2012; 2(4):524-48.
3. Feng X, Koh DW. Roles of poly(ADP-ribose) glycohydrolase in DNA damage and apoptosis. International review of cell and molecular biology 2013; 304:227-81.
4. Nakagawa T, Guarente L: Sirtuins at a glance. J Cell Science. 2011; 124(6):833-8.
5. Devereux RB, Reichek N. Echocardiographic determination of left ventricular mass in man. Anatomic validation of the method. Circulation. 1977; 55(4):613-8.
6. Pacher P, Beckman JS, Liaudet L. Nitric oxide and peroxynitrite in health and disease. Physiol Rev. 2007; 87(1):315-424.
7. Minushkina LO, Zateishchikov DA, Zateishchikova AA, et al. NOS3 gene polymorphism and left ventricular hypertrophy in patients with essential hypertension. Cardiology. 2002; 42(3):30-4. Russian.
8. Wang XL, Mahaney MC, Sim AS, et al. Genetic Contribution of the Endothelial Constitutive Nitric Oxide Synthase Gene to Plasma Nitric Oxide Levels. Arteriosclerosis, Thrombosis, and Vascular Biology. 1997; 17(11):3147-53.
9. Le K, Li R, Xu S, et al. PPARalpha activation inhibits endothelin-1-induced cardiomyocyte hypertrophy by prevention of NFATc4 binding to GATA-4. Archives of biochemistry and biophysics. 2012; 518(1):71-8.
10. El Azzouzi H, Leptidis S, Bourajjaj M, et al. Peroxisome proliferator-activated receptor (PPAR) gene profiling uncovers insulin-like growth factor-1 as a PPARalpha target gene in cardioprotection. The J Biolog Chemistry. 2011; 286(16):14598-607.
11. Planavila A, Iglesias R, Giralt M, Villarroya F. Sirt1 acts in association with PPARalpha to protect the heart from hypertrophy, metabolic dysregulation, and inflammation. Cardiovasc Res. 2011; 90(2):276-84.
12. Canto C, Auwerx J. Targeting Sirtuin 1 to Improve Metabolism: All You Need Is NAD+? Pharmacol Rev. 2012; 64(1):166-87.
13. Ares-Carrasco S, Picatoste B, Camafeita E, et al. Proteome changes in the myocardium of experimental chronic diabetes and hypertension: role of PPARalpha in the associated hypertrophy. J of Proteomics. 2012; 75(6):1816-29.
14. Becker J, Delayre-Orthez C, Frossard N, et al. The peroxisome proliferator-activated receptor alpha agonist fenofibrate decreases airway reactivity to methacholine and increases en-

dothelial nitric oxide synthase phosphorylation in mouse lung. Fundamental & clinical pharmacology. 2012; 26(3):340-6.

15. Minushkina LO, Brazhnik VA, Zateishchikov DA, et al. Genetic predictors of left ventricular hypertrophy: do polymorphisms of peroxisome proliferator activated nuclear receptor genes play any role? Cardiology. 2003; 43(12):71-5. Russian.
16. Luo X, Kraus WL. On PAR with PARP: cellular stress signaling through poly(ADP-ribose) and PARP-1. Genes & development. 2012; 26(5):417-32.
17. Roszak A, Lianeri M, Sowińska A, et al. Involvement of PARP-1 Val762Ala Polymorphism in the Onset of Cervical Cancer in Caucasian Women. Mol Diagn Ther 2013; 17(4):239-45.
18. Pillai JB, Russell HM, Raman J, et al. Increased expression of poly(ADP-ribose) polymerase-1 contributes to caspase-independent myocyte cell death during heart failure. American Journal of Physiology – Heart Circulat Physiol. 2005; 288(2):H486-96.
19. Ye F, Cheng Q, Hu Y, et al. PARP-1 Val762Ala polymorphism is associated with risk of cervical carcinoma. PloS one. 2012; 7(5):e37446.
20. Prasad P, Tiwari AK, Kumar KM, et al. Association analysis of ADPRT1, AKR1B1, RAGE, GFPT2 and PAI-1 gene polymorphisms with chronic renal insufficiency among Asian Indians with type-2 diabetes. BMC medical genetics. 2010; 11:52.
21. Pacher P, Szabo C. Role of poly(ADP-ribose) polymerase 1 (PARP-1) in cardiovascular diseases: the therapeutic potential of PARP inhibitors. Cardiovasc Drug Rev. 2007; 25(3):235-60.
22. Liu M, Li Z, Chen GW, et al. AG-690/11026014, a novel PARP-1 inhibitor, protects cardiomyocytes from AngII-induced hypertrophy. Molec Cell Endocrinol. 2014; 392(1-2):14-22.
23. Deres L, Bartha E, Palfi A, et al. PARP-Inhibitor Treatment Prevents Hypertension Induced Cardiac Remodeling by Favorable Modulation of Heat Shock Proteins, Akt-1/GSK-3beta and Several PKC Isoforms. PloS one. 2014; 9(7):e102148.

International Heart and Vascular Disease Journal
Volume 4, Number 9, March 2016

Journal of the Cardioprogress Foundation

CLINICAL CASE

A case of Gitelman's syndrome with severe hypokalemia and pseudoischemic ECG changes

Grinstein Yu.I.*, Shabalin V.V.

Krasnoyarsk state medical university named after V.F. Voyno-Yasenetsky,
Krasnoyarsk, Russia

Authors:

Yuri I. Grinstein, M.D., professor, head of the therapy department, Institute of Postgraduate Education Therapy (IPT), Krasnoyarsk State Medical University named after V.F. Voyno-Yasenetsky, Krasnoyarsk, Russia;

Vladimir M. Shabalin, M.D., Ph.D, Associate Professor of the therapy department, Institute of Postgraduate Education Therapy (IPT), Krasnoyarsk State Medical University named after V.F. Voyno-Yasenetsky, Krasnoyarsk, Russia.

Summary

A case of Gitelman's syndrome with severe hypokalemia and pseudoischemic ECG changes is presented. A brief review on this kind of primary tubulopathy is also given. Clinical significance of possible difficulties for cardiologist is indicated (pseudoischemic ECG changes, QT-interval prolongation with life-threatening ventricular arrhythmias, risk of myopathy and rabdomyolisis development after statin administration, hypokalemia worsening due to prescribing diuretics).

Key words

Gitelman syndrome, hypokalemia, tubulopathy.

Hypokalemia that appears if serum K+ levels are less than 3/5 mmol/Ll is one of the most frequent electrolyte abnormalities and it occurs in more than 20% of patients admitted to hospital [1]. The most often cause of it is the adverse action of drugs, in particular – diuretics. The role of primary abnormalities of kidney tubular function (tubulopathy) is quite modest, and quite often it affects their opportune diagnostics and treatment.

We describe our observation of one of tubulopathy variants that manifested as severe hypokalemia with pseudoischemic changes on electrocardiogram (ECG).

29 years old female was presented with complaints on severe weakness, weight loss, occasional syncope, dry skin. These symptoms appeared at first 2 years ago and since then developed gradually. During last

* Corresponding autor: Tel. 8-902-990-46-64. E-mail: grinstein.yi@gmail.com

2 months the symptoms have been aggravating. Was examined by physician, neurologist, endocrinologist. Underwent fibrogastroduodenoscopy (FGDS), abdominal ultrasonography (US), head magnetic resonance imaging (MRI). Laboratory tests: cortisol, adrenocorticotropic hormone (ACTG), thyroid-stimulating hormone (TSH), follicle-stimulating hormone (FSH), luteinizing hormone (LH) – normal levels. Diagnosis remained unclear, for further diagnostics was admitted to hospital. No burdened family history. No bad habits. Her occupation is a teacher.

Physical examination revealed evident body mass deficiency: height – 161 cm, weight – 41 kg; signs of connective tissue dysplasia like joint hypermobility; dry skin. Thyroid gland, peripheral lymphatic nodes are not enlarged. Respiratory rate is 18 breaths per minute. Lung auscultation: vesicular breathing, no rales. Heart rate (HR) is 60 beats per minute, blood pressure (BP) is 90/60 mm Hg. Tongue is moist, with white coating. Stomach is soft, no pain during palpation. The lower margin of liver is at the right costal margin, spleen is not palpable. No peripheral edema.

Blood tests: general blood count: normal (hemoglobin – 153 g/L, platelets – 255*10^9/L, leucocytes – 6,1*10^9/L, stab – 4, segmentonuclear – 59, eosinophils – 1, lymphocytes – 33, monocytes – 3, ESR – 10 mm/hour. Biochemical analysis: evident hypokalemia, hypomagnesemia, alkalosis, levels of other markers are normal: glucose – 3 mmol/L, total protein – 16 μmol/L, blood urea – 4.2 mmol/L, creatinine – 73 μmol/L, bilirubin – 16 μmol/L, conjugated bilirubin – 0, alanine-aminotransferase (ALT) – 34 E/L, aspartate-aminotransferase (AST) – 32 E/L, K^+ – 2,0 mmol/L, Mg^{2+} – 0,53 mmol/L (reference levels – 0,66-1,07 mmol/L), Na^+ – 137 mmol/L, Ca^{2+} – 2,5 mmol/L, blood plasma pH – 8,0. Urinalysis: hyposthenuria (1004), protein – negative, leucocytes – 2-3 in visual field. Zimnitsky test: urine's specific gravity: 1006-1008, daily dieresis – 800 ml, nocturnal dieresis – 900 ml. 24-h Ca2+ urine excretion – 0.302 mmol (reference levels – 1.,7 – 3,3, mmol/day).

ECG: evident abnormal repolarization manifested as "pseudoischemic" ST depression in V_4-V_6 leads (Figure 1).

Patient was precisely investigated in order to exclude oncological pathology. Chest X-ray, abdominal and thyroid gland US, abdominal computer tomography (CT). Both US and CT revealed non-homogenous kidney structure and normal kidney size (Figure 2, 3)

The diagnostic version of tubulopathy was raised because of low urine's specific gravity, hypokalemia,

Figure 1. «Pseudoischemic» ECG changes as horizontal ST segment depression in V_4-V_6 lead registered at the time of admission to hospital.

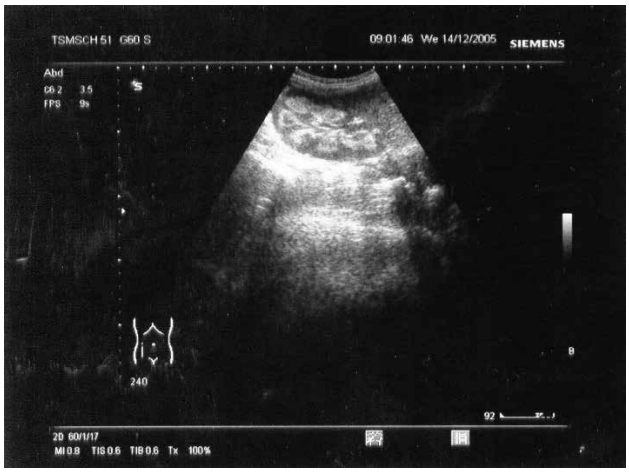

Figure 2. Kidney US (normal dimensions of the kidney, renal pyramids have hyperechoic contour with blurred boundary of different intensity)

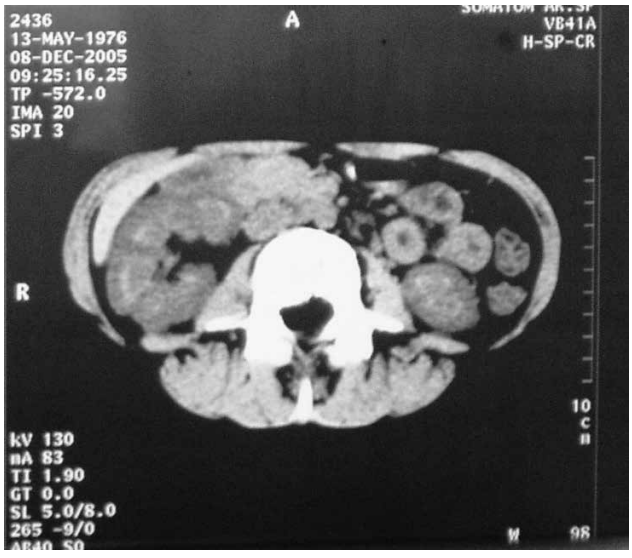

Figure 3. Kidney CT (normal shape, dimensions and position of kidney with distinct boundary. No concrements. No dilation of renal collecting system. Parenchymal structures density is non-homogenous, varies from 35 to 47 Hounsfield units.

hypomagnesemia. Additional tests revealed serum alkalosis, so differential diagnosis of hypokalemic alkalosis was made between Bartter and Gitelman syndromes. But sharply decreased 24-hour Ca2+ urine expression and manifestation of the disease at adult age allowed to establish the diagnosis of Gitelman syndrome, one of distal tubulopathy – distal tubular alkalosis. Not common US and CT signs are considered to be manifestations of nephrocalcinosis, some authors describe calcium deposits in other organs in case of this syndrome. Patient was administered with K+ medications: potassium chloride 3% 50 ml per 10 days intravenously dropwise, after it 3g/day per os, spironolactone 100 mg/day, short course of nimesulide 100 mg/day and magnesium salts with distinct clinical effect: weakness was relieved, work efficiency was restored, patient started to gain weight (after 1 month of therapy she gained 2 kg), no syncope was reported, BP raised up to 110/70 mm Hg, serum pH was normalized. Intravenous potassium chloride administration increased serum K^+ concentration up to intermediate level of 3,0 mmol/L, but it led to adverse reactions: weakness, paresthesia, pain in knee joints. After starting oral administration of potassium containing drugs K+ serum concentration decreased again to 2,3 mmol/L. Because of this spironolactone dose was increased up to 150 mg/day. Patient is under continuous observation.

Discussion

Hypokalemia when levels of serum K+ is <3,5mmol/L is one of the most frequent electrolyte abnormality in clinical practice. Its occurrence in general population is less than 1%, but this pathology can be found in >20% of hospital admitted patients [1].

Since the most frequent cause of hypokalemia is pharmacological therapy, in particular diuretics and laxatives, differential diagnosis should be started from taking precisely patient's history, gaining information about all received drugs, and it is reasonable to subdivide hypokalemia into pharmacological and non-pharmacological one.

Since the patient refused administration of any drugs, it was clear that this hypokalemia was non-pharmacological one. The list of non-pharmacological causes of hypokalemia is big enough [1-3] and includes insufficient intake of electrolyte with food that occurs very rarely because even in case of total starvation organism usually have sufficient compensatory mechanisms that allow to stabilize normal serum K^+ levels; loss of K^+ through gastrointestinal tract (GIT) and kidney, various endocrinological diseases, metabolic alkalosis, hypomagnesemia and some other causes like chronic alcoholism[4] and alcoholic delirium. Patient underwent precise investigation for endocrine pathology in outpatient setting, there were no signs indicating GIT pathology, good social status allowed to exclude dietary and alcoholic causes of potassium deficiency and additional diagnostic techniques that were performed after patient's admission to hospital made it possible to exclude paraneoplastic cause of hypokalemia. Therefore, the diagnostic search was reduced to primary renal causes and in particular to tubulopathies.

Tubulopathies (or tubular dysfunctions) are group of nephropathies that are characterized by partial or generalized loss of renal tubular functions with normal or slightly decreased glomerular filtration. There are primary and secondary tubulopathies, secondary ones appear as a consequence of another systemic disorder like Sjogren's syndrome, Wilson-Konovalov disease, multiple myeloma, paroxysmal nocturnal hemoglobinuria and others.

Primary tubulopathies are classified according with the localization of the lesion (proximal, distal), major clinical syndrome, between them metabolic acidosis or alkalosis have particular importance, main mechanism of transport abnormalities.

Between tubulopathies that are characterized with hypokalemia and metabolic alkalosis the most important ones are Bartter's syndrome and Gitelman's syndrome. Bartter's syndrome is a severe form that manifests very early even during antenatal period and has bad prognosis. Gitelman's syndrome has milder and sometimes asymptomatic course and often it manifests for a first time not only in children and adolescents but also in adults and elderly people [5-7].

There are two hereditary pathological conditions that are related to anomalies of ion transporters in renal tubules (Liddle's syndrome and 11ß-hydroxysteroid dehydrogenase deficiency) that can also cause hypokalemia and metabolic alkalosis, but they are characterized with early development of arterial hypertension and that's why they are excluded in this case.

The first description of Gitelman's syndrome was made in 1966 [8] and because it was considerably different from Bartter's syndrome it got the name of its founder. This inherited tubulopathy with autosome-recessive inheritance mechanism associated with SLC12A3 gene mutation that leads to impaired function of thiazide-sensitive Na^+Cl^- cotransporter in dis-

tal tubules. To date around 100 mutations of SLC12A3 gene have been identified, and occurrence of this pathology can be 1:40000 in Caucasian population [5,6] and even higher occurrence between Japanese [9].

Gitelman's syndrome is characterized with hypokalemia, hypomagnesemia, metabolic alkalosis, hypocalciuria, increased level of renin and aldosterone, weakness, muscle cramps and normal or low BP, with possible polyuria and nocturia. There are evidences of rhabdomyolisis development in case of severe hypokalemia up to formation of acute renal failure [10], cases of statins' intolerance with development of myopathy in patients in whom Gitelman's syndrome wasn't identified on time [11], recurrent syncope [12], possible development of chronic nephropathy with the outcome of chronic renal failure (CRF) [13], choroid and sclerotic calcification [14], paresthesia, depression [15], hypokalemic periodic paralysis [16]. There are many evidences of the combination of Gitelman's syndrome and chondrocalcinosis (pyrophosphate arthropathia) with typical joint syndrome and possible termination of acute arthritis attacks after prescription of magnesium containing drugs [17]. Since both hypokalemia and hypomagnesiema can cause QT interval prolongation, it is expectable that patients with Gitelman's syndrome are prone to more frequent registration of prolonged QT interval [18], development of paroxysmal ventricular arrhythmias [19] and sudden death [20].

Gitelman's syndrome therapy is consisted of several classes of drugs and is enough affordable and easy [5]:

• administration of potassium-containing drugs, preferably potassium chloride per os because this way of administration is more safe, since intravenous administration of potassium chloride is not always well tolerated by patients due to fast increase of potassium serum concentration;

• administration of magnesium-containing drugs (magnesium chloride or magnesium sulphate);

• if the therapeutic effect of mentioned above drugs is not sufficient, potassium-sparing diuretics (spironolactone, triamterene) are prescribed; there are evidences of effective use of eplerenone, antagonist of metalcorticoid receptors [21], and direct rennin inhibitor aliskiren [7].

• non-steroid anti-inflammatory drugs are less effective than in Bartter's syndrome therapy, but there are some evidences of their effective use [5].

Talking about our clinical case, we suppose that although genetic verification of diagnosis has not been performed, relatively late onset of the disease, distinct hypocalciuria together with resistant hypokalemic alkalosis and tendency to hypotension allows to set the diagnosis of Gitelman's syndrome and not a variant of Bartter's syndrome or other primary tubulopathy. Non-homogenous structure of kidney according with US and CT is likely to be the sign of nephrocalcinosis because of deposits of excessively reabsorbed calcium.

As a conclusion it is necessary to notice that, although up to recent times Gitelman's syndrome was considered to be a rare pathology, one of studies made in 1988 demonstrated that occurrence of this syndrome in Sweeden was estimated as 19 per 1 mln of people [22], Japanese researchers [9] investigated the frequency of corresponding genes mutations in 1852 persons on the continent and found out that suppose that occurrence of Gitelman's syndrome should be 10,3 per 10000 people or 1030 per 1 mln. If we assume that real occurrence of this syndrome in Russia is by a factor of ten lower, even in this case there is a big group of people who trying to apply for medical aid with complaints of weakness, fatigability, paresthesia, tendency to hypotension and are discharged with the wrong stereotypic diagnosis of "vegetovascular dystonia", although they could have been diagnosed properly since the diagnostic tactic is not complicated and the treatment is affordable. It is important to notice that, although Gitelman's syndrome is characterized with tendency to hypotension, some patients, especially elderly ones, can be presented with hypertension [23].

The aim of this publication is to attract the attention of doctors to this not very well known and underestimated pathology. Diagnostic algorithm of Gitelman's syndrome requires biochemical proving of resistant hypokalemia, exclusion of such its causes like pharmacological, endocrine, gastrointestinal loss of potassium, evaluation of serum pH (alkalosis is expected), levels of serum magnesium (decrease is expected), calciuria levels (diagnosis is proved with reduced calcium excretion with urine). Diagnosis can be proved with estimation of SLC12A3 gene mutations.

It is necessary to keep in mind such comorbid pathology like Gitelman syndrome. In cardiological practice it is important to remember it during interpretation of pseudoischemic ECG changes, QT interval prolongation, risk of rhabdomyolisis development after statin administration and hypokalemia aggravation after diuretics prescription.

Conflict of interest: None declared.

References

1. Cohn JN, Kowey PR, Whelton PK, et al. New guidelines for potassium replacement in clinical practice: a contemporary review by the National Council on Potassium in Clinical Practice. Arch Intern Med. 2000;160(16): 2429-36.
2. Gennari FJ. Hypokalemia. N Engl J Med. 1998; 339: 451-458.
3. Rastergar A, Soleimani M. Hypokalaemia and hyperkalaemia. Postgrad Med J. 2001;77:759-64.
4. Elisaf M, Liberopoulos E, Bairaktari E, et al. Hypokalaemia in alcoholic patients. Drug Alcohol Rev. 2002; 21: 73-6.
5. Knoers NV, Levtchenko EN. Gitelman syndrome. Orphanet J Rare Dis. 2008;3:22.
6. Roser M, Eibl N, Eisenhaber B, et al. Gitelman syndrome. Hypertension. 2009;53(6):893-7.
7. Brambilla G, Perotti M, Perra S, et al. It is never too late for a genetic disease: a case of a 79-year-old man with persistent hypokalemia. J Nephrol. 2013;26(3):594-8.
8. Gitelman HJ, Graham JB, Welt LG. A new familial disorder characterized by hypokalemia and hypomagnesemia. Trans Assoc Am Physicians. 1966;79:221-35.
9. Tago N, Kokubo Y, Inamoto N, et al. A high prevalance of Gitelman's syndrome mutations in Japanese. Hypertens Res. 2004;27(5):327-31.
10. Nishihara G, Higashi H, Matsuo S, et al. Acute renal failure due to hypokalemic rhabdomyolysis in Gitelman's syndrome. Clin Nephrol. 1998;50(5):330-2.
11. Freedman DB, Housley D. Gitelman's syndrome presenting as intolerance to statin therapy. Ann Clin Biochem. 2005; 42 (Pt 3):232-33.
12. Hashida T, Yamada M, Hashimoto K, et al. Loss of consciousness and hypokalemia in an elderly man with a mutation of the thiazide-sensitive Na-Cl cotransporter gene. Endocr J. 2006;53(6):859-63.
13. Bonfante L, Davis PA, Spinello M, et al. Chronic renal failure, end-stage renal disease, and peritoneal dialysis in Gitelman's syndrome. Am J Kidney Dis. 2001; 38(1): 165-8.
14. Vezzoli g, Soldati L, Jansen A, et al. Choroidal calcifications in patients with Gitelman's syndrome. Am J Kidney Dis. 2000;36(4): 855-8.
15. Enya M, Kanoh Y, Mune T, et al. Depressive state and paresthesia dramatically improved by intravenous MgSO4 in Gitelman's syndrome. Intern Med. 2004;43(5):410-4.
16. Saiki S, Yoshioka A, Saiki M, et al. A case of Gitelman's syndrome presenting with the hypokalemic periodic paralysis. Rinsho Shinkeigaku. 2002;42(4):317-9.
17. Ea HK, Blanchard A, Dougados M, et al. Chondrocalcinosis secondary to hypomagnesemia in Gitelman's syndrome. J Rheumatol. 2005; 32(9):1840-2.
18. Foglia PEG, Bettinelli A, Tosetto C, et al. Cardiac work up in primary renal hypokalaemia-hypomagnesaemia (Gitelman syndrome). Nephrol Dial Transplant. 2004;19:1398-402.
19. Nakane E, Kono T, Sasaki Y, et al. Gitelman's syndrome with exercise-induced ventricular tachycardia. Circ J. 2004;68(5): 509-11.
20. Scognamiglio R, Negut C, Calo' LA. Aborted sudden cardiac death in two patients with Bartter's/Gitelman's syndromes. Clin Nephrol. 2007; 67(3):193-7.
21. Blanchard A, Vargas-Poussou R, Vallet M, et al. Indomethacin, amiloride, or eplerenone for treating hypokalemia in Gitelman syndrome. J Am Soc Nephrol. 2015;26(2):468-75.
22. Fava C, Montagnana M, Rosberg L, et al. Subjects heterozygous for genetic loss of function of the thiazide-sensitive cotransporter have reduced blood pressure. Human Molecular Genetics. 2008;17(3):413-8.
23. Balavoine AS, Bataille P, Vanhille P, et al. Phenotype-genotype correlation and follow-up in adult patients with hypokalaemia of renal origin suggesting Gitelman syndrome. Eur J Endocrinol. 2011;165(4):665-73.

International Heart and Vascular Disease Journal
Volume 4, Number 9, March 2016

Journal of the Cardioprogress Foundation

Guidelines for authors

International Heart and Vascular Disease Journal
Requirements for Submission and Publication

The requirements for submission and publication in the **International Heart and Vascular Disease Journal** are based on the 'Uniform Requirements for Manuscripts Submitted to Biomedical Journals', developed by the *International Committee of Medical Journal Editors* (ICMJE), which can be found at www.ICMJE.org

These requirements form the basis for relations between the Editors of the **International Heart and Vascular Disease Journal**, further called "the Editors", and an author who submits a manuscript for publication, further called "the Author".

The **International Heart and Vascular Disease Journal** publishes reviewed articles that cover all aspects of cardiovascular diseases, including original clinical research, experimental research with clinical relevance, reviews on current problems in cardiology, and clinical case studies. Usually 4 issues are published annually (one issue every 3 months).

This is an open access journal, which means that all content is freely available without charge to the user or his/her institution. Users are allowed to read, download, copy, distribute, print, search, or link to the full texts of the articles in this journal without asking prior permission from the publisher or the author. This is in accordance with the *Budapest Open Access Initiative* (BOAI) definition of open access.

1. Submission requirements and publishing policy

1.1. A manuscript should be submitted to the following e-mail address: submissions.ihvdj@gmail.com

Editorial Office tel.: +7(965) 236-16-00

1.2. A manuscript is accepted for further consideration only if the manuscript, or any substantively similar version, has not been submitted to and published in any other journal, or disseminated via any other media, such as the Internet.

1.3. The Author, submitting the manuscript to the Editor, assigns the Editor to publish it. The Editors have the right to incorporate within the manuscript any illustrated or text material, including advertisements. The Editors may allow third parties to put such content into the manuscript.

1.4. Submission of the manuscript to the Editors implies that the Author agrees to transfer the exclusive property rights for the manuscript and other objects of the copyright, like photos, drawings, graphics, tables, etc., to the Editors. The Editors obtain the right to reproduce (partly or fully) all the content submitted, including objects of the copyright, in press and on the Internet; to distribute; to translate the manuscript and other provided content into any language; to export and import copies of the issue where the article of the Author was published; and to revise the manuscript.

1.5. The Author transfers the rights specified in clauses 1.3 and 1.4 to the Editors without any time limitations or territory restrictions, including the territories of the Russian Federation.

1.6. The Editors have the right to transfer the rights received from the author to a third party or to prohibit any use of materials published in the journal by a third party.

1.7. The Author guarantees that he or she holds the copyright to all materials submitted to the **International Heart and Vascular Disease Journal**. In case of violation of this guarantee by the Author and consequent claims to the Editors, the Author is obliged to settle all the claims at his/her own expense. The Editors are not responsible for copyright violation by the Author.

1.8. The Author retains the right to use the published material or its parts for personal use, including scientific and educational purposes. The Author retains the right to publish extracts from the published material or its parts in other journals, on the condition that reference is made to the original publication in the **International Heart and Vascular Disease Journal**.

1.9. The copyright is considered transferred to the Editors once confirmation has been sent to the author confirming the manuscript has been accepted for publication.

1.10. Reprinting of an article published in the **International Heart and Vascular Disease Journal** by third parties is only permitted with written permission from the Editors. If permission is granted, reference to the issue of the **International Heart and Vascular Disease Journal** in which the article was published and to the year of publication is obligatory.

1.11. The Editors are obliged to provide the Author with one copy of the issue in which the article is published. The Author(s) should provide his/her full postal address(es) including post code(s) at the end of the manuscript.

1.12. Manuscripts may be reviewed by independent experts. Manuscripts which are reviewed will be reviewed on a double blind basis: Authors will not know the identity of reviewers and reviewers will not know the identity of Authors. The name of the institution where an Author works or conducts research also remains confidential. The reviewer(s) comments and opinions will be sent to the Author and the Author invited to make any changes and/or corrections. In the case of an Author not returning changes and/or corrections to the Editors by an agreed date, the Editors have the right to make their own changes and/or corrections, or permit changes and/or corrections suggested by the reviewers, or to refuse to publish the manuscript. Editing, shortening and correction of the manuscript, and changes to a graph, picture or table design are made in order they comply the format and standards of the **International Heart and Vascular Disease Journal**.

1.13. The Editors are not responsible for the accuracy of information presented in the manuscripts.

1.14. The Editors recommend that submitted manuscripts conform with the 'Uniform Requirements for Manuscripts Submitted to Biomedical Journals', developed by the *International Committee of Medical Journal Editors* (ICMJE), and available on the **International Heart and Vascular Disease Journal** website www.cardioprogress.ru, in the 'For Authors' section.

1.15. Adhering to the standards outlined in this document will lead to faster reviewing, editing, and publishing of manuscripts accepted for publication. Manuscripts submitted outside the standards on design and formatting for this journal may not be accepted by the Editors.

2. General recommendations for submission of original scientific works

2.1. The Editors recommend that results of randomized controlled trials conform to the 'Consolidated Standards of Reporting Trials' (CONSORT) guidelines. Information on these standards are available on the CONSORT website: www.consort-statement.org

2.2. A manuscript should be typed using the Times New Roman font (12 points, double spacing; with 2 cm at the top, bottom, left and right margins). The length of a manuscript, including references, schedules, drawings and tables, should not exceed 12 standard typewritten pages (1 page is 1800 letters or symbols, including spaces). A case study should not exceed 6 standard pages. Reviews and lectures should not exceed 25 standard pages.

2.3. Manuscripts should be organized as follows: 1) title page; 2) structured summary and keywords; 3) list of abbreviations; 4) text; 5) acknowledgements (if applicable); 6) references; 7) names and legends of pictures, tables, graphics, and photocopies in the order they appear in the manuscript; 8) drawings, tables, graphics, and photocopies should be submitted on separate pages in the order they appear in the manuscript. Numeration of pages should begin from the title page.

2.4. If the manuscript contains pictures, tables, graphics, or photocopies that have been published previously, reference to the author(s) and publication is necessary. It is the Author's responsibility for determining whether permission is required for the duplication of material, and for obtaining relevant permission.

2.5. Manuscripts based on reviews of original research works should contain the following sections: Introduction (reflecting the urgency of a problem and research goals); Material and methods; Results; Discussion of the obtained results and Conclusion. The text should be clear, brief and without repetition.

3. Publication of uncontrolled trials results

3.1. An uncontrolled trial is a research without a control group.

3.2. Manuscripts based on uncontrolled trials results will be accepted for publication in the 'Practical Experience' column only if the uncontrolled design of the study is described in the Material and methods and Discussion sections. It is important not to exaggerate the significance of results in the Conclusion' section.

4. Ethical aspects

4.1. Trials should be conducted in accordance with principles of "good clinical practice". Participants of a trial should be informed about the purpose and main aims of the trial. They must sign to confirm their written informed consent to participate in the trial. The «Material and methods» section must contain details of the process of obtaining participants informed consent, and notifica-

tion that an Ethics Committee has approved conducting and reporting the trial. If a trial includes radiological methods it is desirable to describe these methods and the exposure doses in the «Material and methods» section.

4.2. Patients have the right to privacy and confidentiality of their personal data. Therefore, information containing pictures, names, and initials of patients or numbers of medical documents should not be presented in the materials. If such information is needed for scientific purposes, it is necessary to get written informed consent from the research participant (or their parent, their trustee, or a close relative, as applicable) prior to publication in print or electronically. Copies of written consent may be requested by the Editors.

4.3. Animal trials must conform to the 'International Guiding Principles for Biomedical Research Involving Animals', adopted by the *Council for International Organizations of Medical Sciences* (CIOMS) in 1985.

5. Authorship

5.1. Each author should significantly contribute to the work submitted for publication.

5.2. If more than 4 authors are indicated in the author's list, it is desirable to describe the contribution of each author in a covering letter. If the authorship is attributed to a group of authors, all members of the group must meet all criteria for authorship. For economy of space, members of the group may be listed in a separate column at the end of the manuscript. Authors can participate in the submitted manuscript in the following ways: 1) contributing to the concept and research design or analyzing and interpreting data; 2) substantiating the manuscript or checking the intellectual content; 3) providing final approval for the manuscript. Participation solely in collection of data does not justify authorship (such participation should be noted in the Acknowledgements section). Manuscripts should be submitted with a covering letter containing the following information: 1) the manuscript has not been submitted to any other media; 2) the manuscript has not been published previously; 3) all authors have read and approved the manuscript's content; 4) the manuscript contains full disclosure of any conflict of interests; 5) the author/authors confirm responsibility for the reliability of the materials presented in the manuscript. The author responsible for the correspondence should be specified in the covering letter.

6. Conflict of interests/financing

6.1. It is desirable for authors to disclose (in a covering letter or on the title page) any relationships with industrial and financial organizations, which might be seen as a conflict of interest with regard to the content of the submitted manuscript. It is also desirable to list all sources of financing in a footnote on the title page, as well as workplaces of all authors (including corporate affiliations or employment).

7. Manuscript content

7.1. Title page

7.1.1. It should include the name of the article (in capital letters); initials and last names of the authors; the full name of the institution which supported the manuscript, together with the city and country, and full mailing address with postal code of that institution.

7.1.2. A short title of the article (limited to 45 letters or symbols).

7.1.3. Information about the authors, including full names (last name, first name, patronymic name, if applicable; scientific degrees and titles, positions at main and secondary jobs, including corporate posts).

7.1.4. Full name, full postal address, e-mail address, and telephone number of the "Corresponding author" who will be responsible for any contact with the Editors.

7.1.5. The manuscript (or the covering letter) should be signed by all authors.

7.1.6. It is desirable to provide information about grants, contracts and other forms of financial support, and a statement about any conflict of interests.

7.2. Summary

7.2.1. Summary (limited to 300 words) should be attached to the manuscript. It should include the full title of the article, last names and initials of the authors, the name of the institution that supported the manuscript, and its full postal address. The heading of the summary should contain the international name(s) of any drug(s) mentioned.

7.2.2. Original studies summary should contain the following sections: Aim, Material and methods, Results, and Conclusion. The summary of a review should provide the main themes only. A manuscript must contain all data presented in the summary.

7.2.3. 5-6 keywords of the article should be given at the end of the abstract.

7.3. List of abbreviations and their definitions

7.3.1. To conserve space in the journal, up to 10 abbreviations of general terms (for example, ECG, ICV, ACS) or names (GUSTO, SOLVD, TIMI) can be used in a manuscript. List of abbreviations and their definitions should be provided on a separate page after the structured summary (for example, ACS – aortocoronary shunting). Only words generally accepted in scientific literature should be used.

7.4. Text

7.4.1. Original studies should be structured as follows: Introduction, Material and methods, Results, Discussion and Conclusion.

7.4.2. Case studies, reviews and lectures may be unstructured, but it is desirable to include the following paragraphs: Discussion and Conclusion (Conclusions and Recommendations).

7.4.3. Please, use international names of drugs in the title. Exceptions are possible when use of trade names is well-founded (for example, in studies of bio- or therapeutic equivalence of drugs). It is possible to use a trade name in the text, but not more than once per standard page (1800 symbols including spaces).

7.4.4. You must provide titles and subtitles in the sections: Methods, Results and Discussion. Each reference, image or table should be numbered and specified in order of appearance in the text.

7.4.5. All units of measurement should be provided according to the *International System of Units* (SI) system. No abbreviations, except standard abbreviations of chemical and mathematical terms, are acceptable.

7.4.6. Each image, chart, table, photo, and reference must be indicated in order of appearance in the text.

7.4.7. References in the text must be numbered in Arabic figures, and provided in square brackets.

7.5. Statistics

7.5.1. All submitted materials may be revised to ensure relevance and accuracy of statistical methods and statistical interpretation of results. The Methods section should contain a subsection with detailed description of statistical methods, including those used for generalization of data; and of methods used for testing hypotheses (if those are available). Significance value for testing hypotheses must be provided. Please indicate which statistical software was used to process results and its version if you use more complex statistical methods (besides a t-test, a chi-square, simple linear regression, etc.).

7.6. Acknowledgements

7.6.1. The Acknowledgements section or Appendix should not exceed 100 words.

7.7. References

7.7.1. Please use separate sheets and double spacing for the list of references. Give each source a consecutive number starting on a new line. The list of references should be structured in order of citation. Use *Index Medicus* to search for abbreviations of the names of journals.

7.7.2. All documents referred to in the text, should be included in the list of references.

7.7.3. The list of references should not include any dissertations, theses published more than two years ago, or information that is impossible to check (local conference materials, etc.). If material is taken from a thesis, please, mention that in brackets — (thesis).

7.7.4. It is desirable to refer to periodicals with a high impact factor, if possible.

7.7.5. In order to increase the citing of authors, transliteration of sources in Russian are made in the **International Heart and Vascular Disease Journal** using official coding. Names of authors and journals are transliterated by means of coding, and semantic transliteration (translation) is used for the titles of articles. If a source has an original transliteration, the latter is used. The Editors will be grateful if authors provide the transliterated variant of the list of references. You can use online services: http://translit.ru_for making transliteration.

7.7.6 Authors are responsible for the accuracy of information provided in the list of references.

7.7.7 The list of references should conform to the format recommended by the *American National Information Standards Organization* (NISO), accepted by the *National Library of Medicine* (NLM) for its databases (Library's MEDLINE/Pub Med database) and updated in 2009. Authors should use the official site of the NLM: http://www.nlm.nih.gov/citingmedicine_to find recommended formats for the various types of references. Examples of references provided in accordance with the NLM recommendations are given below:

Periodicals

Go AS, Hylek EM, Phillips KA, et al. Prevalence of diagnosed atrial fibrillation in adults: national implications for rhythm management and stroke prevention: the Anticoagulation and Risk factors in Atrial Fibrillation (ATRIA) Study. JAMA. 2001;285(18):2370-5.

Sources in Russian with transliteration:

Baevskiy RM, Ivanov GG, Chireykin LV, et al. Analiz variabel'nosti serdechnogo ritma pri ispol'zovanii razlichnyh jelektrokardiograficheskih sistem (metodicheskie rekomendacii) [Analysis of heart rate variability using different ECG systems (guidelines)]. Vestnik aritmologii. 2002;24:65-86. Russian.

Please provide initials after the last names of authors. Last names of foreign authors are given in the original transcription. Names of periodicals can be abbreviated. Usually such abbreviations are accepted by the Editors of those periodicals.

These can be found on the Publisher's site or in the list of abbreviations of Index Medicus.

Punctuation in the list of references should be considered. A full stop should be put with a space between the name of the journal and the year of its release. After the year of release a semicolon is put without a space, then a colon follows the volume number, and finally page numbers are given. There are no indications like "volume", "№", "pages". Russian periodicals often have no indication of volume or numbering of pages within a year. In this case the number of an issue should be specified in brackets.

If the total number of authors exceeds four people, please provide the names of the first three authors and put "et al." afterwards. If there are not more than 4 authors, the full list of authors should be provided

Chapters in a book

Swanton RH, Banerjee S. Cardiac Failure. In: Swanton RH, Banerjee S., editors. Swanton's Cardiology: A concise guide to clinical practice. 6th ed. Oxford: Blackwell Publishing; 2008. p. 255-309.

Sources in Russian with transliteration:

Belenkov YuN. Kardiomiopatii [Cardiomyopathies]. In.: Chazov EI, Belenkov YuN., editors. Racional'naja farmakoterapija serdechno-sosudistyh zabolevanij: Rukovodstvo dlja praktikujushhih vrachej [Rationale for drug therapy of cardiovascular diseases: A guide for medical practitioners]. Moscow: Litterra; 2006. p. 431-452. Russian.

Reference to a book chapter should be arranged in the following order: authors of the corresponding chapter; name of the chapter; «In:»; editors (title authors) of the book; name of the book; number of issue, publisher; city of publishing; year of publishing; pages of the corresponding chapter. Punctuation should be considered. There are no quotation marks.

Books

Sources in Russian with transliteration:

Shlyakhto EV, Konradi AO, Tsyrlin VA. Vegetativnaja nervnaja sistema i arterial'naja gipertenzija [The autonomic nervous system and hypertension]. St. Petersburg (Russia): Meditsinskoe izdatel'stvo; 2008. Russian.

Websites

Websites should be provided in the list of references, but not in the text. References to websites should be made only when original text is not available. References should be provided in the following way:

WHO. Severe Acute Respiratory Syndrome (SARS) [Internet]. [place unknown: publisher unknown]; [updated 2010 June 1; cited 2010 June 10]. Available from: http://www.who.int/csr/sars/.

7.8. Diagrams, charts, and figures

7.8.1. Diagrams, charts, and figures should be submitted electronically in the following formats: «MS Excel», «Adobe Illustrator», «Corel Draw» or «MS PowerPoint». Diagrams, charts, and figures must be allocated on separate pages, numbered in order of citation, and have names and notes if necessary. They must not repeat the content of tables. Please indicate the names and units of measurement for graph axes. Provide the legend for each graph (denote lines and filling). If you compare diagrams, provide significance of differences. Do not use 3-D models for histograms. If appropriate, please identify places in the text where you wish graphics, figures and graphs to be inserted.

7.8.2. Photographs must be submitted electronically with a minimum resolution of 300 dots per inch (dpi). Microphotos must be cropped so that only main content is left. Arrows should be used to show main features. All symbols, arrows and legends on gray-scale illustrations should be in contrast with the background.

7.8.3. Size of legends on images and photos should be big enough to be legible after compression for publication. The optimal size is 12 points.

7.8.4. All abbreviations should be defined either after the first citation in a legend, or in alphabetic order at the end of each legend. All symbols (arrows, circles, etc.) must be explained.

7.8.5. If data was published earlier, it is desirable to provide written permission from the publisher for the use of this data.

7.9. Tables

7.9.1. Tables should be typed with double spacing, have numbers in order of citation in the text, and names. Tables should be compact and demonstrative. Names of columns and rows must reflect the content. Data presented in tables should not be repeated in the text or images. Please clearly specify units of measurement of variables and form of data presentation (M±m; M±SD; Me; Mo; percentiles etc.). All figures, sums and percentages must be thoroughly checked and correspond to those in the text. Explanatory footnotes should be provided below the table if necessary.

7.9.2. Abbreviations should be listed in a footnote under the table in alphabetic order. Symbols of footnotes should be given in the following order: *, †, ‡, §, ‖, ¶, #, **, † † etc.

7.9.3. If a table(s) was published earlier, it is desirable to provide written permission from the publisher for use of this table(s).

China Heart Federation

GW-ICC

WHFS

ISCP

GW-ICC 2016
APHC | ICCPR

OCT.13-16
BEIJING, CHINA

The 27th Great Wall International Congress of Cardiology

The World Heart Failure Congress 2016 (WHFC 2016)

The 21st Annual Scientific Meeting of the International Society of Cardiovascular Pharmacotherapy (ISCP)

www.gw-icc.org

FOUNDATION FOR THE ADVANCEMENT OF CARDIOLOGY

"CARDIOPROGRESS"

knowledge, observation, action

Official website: www.cardioprogress.ru

Tel: 007 965 236 1600

Email: inf.cardio@gmail.com

Moscow, Russia