Upalni medijatori i njihova povezanost s dijastoličkom disfunkcijom i remodeliranjem srca u osoba starije životne dobi

Inflammatory Mediators and Their Association with Diastolic Dysfunction and Heart Remodeling in the Elderly

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SAŽETAK: Uvod: Najvažniji indeks za remodeliranje srca u starijoj životnoj dobi, osim hipertrofije lijeve klijetke i dilatacije lijevog atrija, jest progresivna dijastolička disfunkcija lijeve klijetke, što je ujedno i najvažniji znak starenja srca. **Cilj**: Dokazati povezanost i ulogu sustavne upale prisutne u najčešćim pratećim bolestima u starijoj životnoj dobi (arterijska hipertenzija, šećerna bolest, pretilost i kronična bubrežna disfunkcija) uz postojanje dijastoličke disfunkcije pri zatajivanju srca s očuvanom istisnom frakcijom u osoba starije životne dobi. Ustanoviti razinu upalnih posrednika: serumskog IL-6 i hs-CRP-a u testnoj i kontrolnoj skupini te ih povezati s ehokardiografskim parametrima dijastoličke disfunkcije, kao i s razinom karotidne ateromatoze. **Pacijenti i metode:** Istraživanje je provedeno u ukupno 78 pacijenata starijih od 65 godina. U njih 60 kliničkim je i ehokardiografskim testiranjem otkriveno zatajivanje srca s očuvanom istisnom frakcijom, kao i prisutnost jednog ili više istraženih komorbiditeta, dok je 18 pacijenata bilo relativno zdravo i bez komorbiditeta. U svih je ispitanika provedena obrada: klinički pregled, elektrokardiografija, laboratorijske pretrage, ehokardiografija te dopler karotidnih arterija. **Rezultati:** Pacijenti sa zatajivanjem srca uz očuvanu istisnu frakciju imaju abnormalnosti srca i vaskularne strukture u usporedbi s relativno zdravom skupinom starijih pacijenata. Takvi pacijenti imaju značajnije remodeliranje srca (koncentričnu hipertrofiju lijeve klijetke i dilataciju lijevog atrija), kao i dijastoličku disfunkciju (viši E/e', niži e') te abnormalnu vaskularnu disfunkciju (promjene u karotidnim krvnim žilama). Razina seruma IL-6 i hs-CRP-a pokazala je bitnu povezanost s parametrima dijastoličke disfunkcije, kao i s razinom hipertrofije lijeve klijetke, parametrima remodeliranja lijevog atrija, NYHA stupnjem zatajivanja srca te karotidnom ateromatozom. **Zaključak:** Osim najčešćih komorbiditeta u starijih bolesnika, upala i oksidativni stres također imaju ulogu u razvoju dijastoličke disfunkcije, remodeliranja srca i pojave zatajivanja srca s očuvanom istisnom frakcijom.

SUMMARY: Introduction: The most important index for heart remodeling in old age, besides left ventricular hypertrophy and left atrium dilatation, also represents the progressive left ventricular diastolic dysfunction, which seems to be the most important marker for cardiac aging. **Aim:** To prove the association and the role of systemic inflammation present in the most frequent comorbid diseases in old age (arterial hypertension, diabetes mellitus, obesity, and chronic renal dysfunction) with the existence of diastolic dysfunction in heart failure with preserved ejection fraction in the elderly. To establish the level of inflammatory mediators: IL-6 and hs-CRP in the blood of patients in the test and control group and to correlate this with echocardiographic parameters for diastolic dysfunction as well as with the level of carotid atheromathosis. **Patients and Methods:** A total of 78 patients aged >65 years were investigated; cardiac failure with preserved ejection fraction was found in 60 using clinical and echocardiographic tests, as well as the presence of one or several investigated comorbidities, while 18 patients were relatively healthy elderly persons without comorbidities. All patients underwent clinical investigations, electrocardiography, laboratory analyses, echocardiography, and carotid Doppler sonography. **Results:** Patients with heart failure with preserved ejection fraction had abnormalities of the heart and vascular structure, compared with the relatively healthy group of the elderly. These patients had more significant heart remodeling (concentric left-ventricular hypertrophy and left atrial dilatation), as well as diastolic dysfunction (higher E/e', lower e'), and abnormal vascular dysfunction (changes in carotid blood vessels). The serum level of IL-6 and hs-CRP showed significant association with the parameters of diastolic dysfunction, as well as with the level of the left vascular hypertrophy, the parameters of the eft atrial remodeling, the level of the heart failure (NYHA), and carotid atheromatosis. **Conclusion:** Inflammation and oxidative stress, in addition to being the most common comorbidities in elderly, have a role in developing diastolic dysfunction, heart remodeling, and the appearance of heart failure with preserved ejection fraction.

KLJUČNE RIJEČI: upalni medijatori, dijastolička disfunkcija, zatajivanje srca.

KEYWORDS: inflammatory mediators, diastolic dysfunction, heart failure.

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Uvod

Čak i u zdrave populacije srce stari zajedno s ostalim organima. Starenje se može smatrati suvremenom pandemijom povezanom s ozbiljnim društvenim i ekonomskim učincima. Progresivno povećanje očekivane životne dobi povezano je s većom učestalosti kroničnih bolesti starije životne dobi. Stoga je razumijevanje mehanizama toga procesa te promjena koje se fiziološki zbivaju s vremenom ključno za unaprjeđenje kvalitete života starijih te smanjenje tereta koje donose bolesti povezane sa starenjem. Proces starenja nedvojbeno utječe na kardiovaskularni sustav, a učestalost kardiovaskularnih bolesti povećava se s vremenom[1]. Proces starenja uzrokuje strukturne i funkcionalne promjene poput vaskularnog ukrućivanja, hipertrofije miocita i zadebljanja stijenke srca, pojačane fibroze miokarda i remodeliranja izvanstaničnog matriksa, što sve zajedno vodi prema dijastoličkoj disfunkciji obilježenoj smanjenjem aktivnog punjenja lijeve klijetke. Progresivna dijastolička disfunkcija lijeve klijetke važna je, čak i u slučajevima kada se masa lijeve klijetke ne povećava znatno tijekom starenja. No, procjena dijastoličke disfunkcije može biti znak starenja srca[1]. Klinički čimbenici koji mogu ubrzati proces starenja srca uključuju: debljinu intraabdominalnoga masnoga tkiva (pretilost), šećernu bolest, dislipidemiju i arterijsku hipertenziju. Na molekularnoj se razini smatra da su kod srčanih miocita reaktivne kisikove vrste, transformirajući čimbenik rasta beta i mitohondrijska funkcija povezani sa starenjem srca[2]. Također se pokazalo da je dijastolička disfunkcija lijeve klijetke vezana za starenje jedna od glavnih čimbenika rizika za razvoj zatajivanja srca (HF). Najčešće je riječ o zatajivanju srca s očuvanom istisnom frakcijom (HFpEF; prema engl. *heart failure with preserved ejection fraction*). Najvažniji čimbenici rizika za razvoj HF-a uključuju: dob, arterijsku hipertenziju, metabolički sindrom, šećernu bolest, disfunkciju bubrega i nedostatak tjelesne aktivnosti[2,3]. Postoji dosta informacija o ulozi upalnih stanica i putova tijekom akutne ozljede i procesa regeneracije, koji se posljedično aktiviraju. Nažalost, relativno se malo zna o uzrocima koji vode do kronične upale u HF-a kao ishoda početne upalne reakcije, što čini samo jednu putanju za progresiju bolesti[4].

Upala je složeni obrambeni mehanizam u kojem leukociti putuju iz vaskularnog sustava u oštećena tkiva kako bi uništili uzročnike koji mogu uzrokovati oštećenje tkiva. Akutna je upala ograničena povoljna reakcija, napose tijekom upalnog podražaja, dok je kronična upala perzistentna pojava koja može uzrokovati tkivno oštećenje. Jedno je obilježje akutne upale činjenica da je infiltrat leukocita prvotno neutrofilan, ali nakon 24 do 48 sati prevladavaju monociti. Za razliku od toga, kronična je upala histološki povezana s pristnošću mononuklearnih stanica, tj. makrofaga i limfocita[4]. Premda postoji nekoliko pokušaja objašnjenja, ipak nedovoljno znamo o mehanizmima koji upravljaju prelaskom neutrofila u monocite tijekom preobrazbe iz akutne u kroničnu upalu. Moguće je da interleukin-6 (IL-6)/topljivi IL-6 receptor α (sIL-6Rα) kompleks ima važnu ulogu u toj preobrazbi. IL-6 ima dvostruki učinak; na nekim razinama djeluje kao obrambeni mehanizam, no u kroničnoj upali ima proupalni učinak.[4,5] Imunosenescencija je sastavni dio ljudskoga starenja, a njezin je ishod smanjenje broja mirujućih limfocita T i B, nakupljanje memorijskih i aktivnih limfocita T i B, proizvodnje nedjelotvornih protutijela, povećanje proizvodnje autoprotutijela te kronična upala

Introduction

Even in the healthy population, the heart ages together with other body organs. Aging may be considered a modern pandemic associated with a serious social and economic impact. The progressive increase in life expectancy is associated with higher prevalence of chronic age-related disease. In this light, understanding the mechanisms underlying this process and the physiological changes that occur with time, is crucial in improving the quality of life of the elderly and reducing the burden of age-related diseases. The aging process undeniably affects the cardiovascular system, and the prevalence of cardiovascular diseases increases over time[1]. The aging process induces structural and functional changes such as vascular stiffening, myocyte hypertrophy and increased wall thickness, increased myocardial fibrosis, and extracellular matrix remodeling, which taken together lead to diastolic dysfunction characterized by reduced active filling of the left ventricle. Progressive left ventricular diastolic dysfunction is important, even in cases when the left ventricular mass is not remarkably increased during aging. However, the estimation of diastolic dysfunction could represent a marker for cardiac aging[1]. Clinical factors which could accelerate the process of cardiac aging include: visceral thickness (obesity), diabetes mellitus, dyslipidemia, and hypertension. At the molecular level, it is believed that in cardiac myocytes – the reactive oxygen species, the transforming growth factor-beta, mitochondrial function – are associated with cardiac aging[2]. Furthermore, the aging-related left ventricular diastolic dysfunction was showed to be one of the main risk factors for developing heart failure, which is most frequently heart failure with preserved ejection fraction (HFpEF). The most important risk factors for its development include: age, hypertension, metabolic syndrome, diabetes mellitus, renal dysfunction, and physical inactivity[2,3]. There is a wealth of information in the literature on the role of inflammatory cells and pathways during the acute injury and the regeneration processes, which are activating subsequently. Unfortunately, relatively little is known about the reasons which lead to chronic inflammation in heart failure as a sum of the initial inflammatory response, which represents only a trajectory for disease progression[4].

Inflammation is a complex defense mechanism in which leucocytes migrate from vasculature into damaged tissues to destroy the agents that can potentially cause tissue injury. Acute inflammation has a limited beneficial response, particularly during infectious challenge, whereas chronic inflammation is a persistent phenomenon that can lead to tissue damage. One hallmark of acute inflammation is that the leucocyte infiltrate is initially mostly neutrophilic, but monocyte cells predominate after 24 to 48 hours. In contrast, chronic inflammation is histologically associated with the presence of mononuclear cells such as macrophages and lymphocytes.[4] Although several explanations have been suggested, the mechanisms that control the transition from neutrophil to monocyte recruitment during the transformation from acute to chronic inflammation are poorly understood. It is possible that the interleukin-6 (IL-6)/soluble IL-6 receptor α (sIL-6Rα) complex plays an important role in this transition. IL-6 has a dual effect; at some levels it acts as a defending mechanism but in chronic inflammation it is rather proinflammatory.[4,5] Immuno-senescence is an integral part of human aging that results in a decrease in the number of naive T and B lymphocytes, the accumulation of memory and

niskoga stupnja.[4,5] Njezina najvažnija obilježja uključuju mala povećanja u koncentraciji proupalnih citokina, kemokina i adipokina poput interleukina-1ß (IL-1ß), IL-6, faktora nekroze tumora-alfa (TNF-alfa) te monocitnoga kemoatraktant protein-1.[4] Proupalni citokini stimuliraju sintezu C-reaktivnog proteina (CRP) u jetri, a pokazalo se da se njegova razina povećava u starijih. Mehanizmi povećanja CRP-a u pacijenata sa zatajivanjem srca nisu potpuno razjašnjeni[6]. Moguće teorije uključuju: kongestiju organa i hipoperfuziju koja uzrokuje lučenje IL-6 iz jetrenih, bubrežnih, endotelnih, mononuklearnih te srčanih miocita. IL-6 uzrokuje proizvodnju CRP-a u jetri. Takva, povećana proizvodnja može biti odraz pojave upalnoga stanja koja uzrokuje razvoj dijastoličke disfunkcije. Premda su klinički znakovi i simptomi upale minimalni ili odsutni, to stanje pridonosi raznim molekularnim patologijama te uzrokuje oštećenje, žila i otpornosti na inzulin te stoga povećava rizik od razvoja dijabetesa tipa 2, kardiovaskularnih bolesti, moždanog udara, raka, sarkopenije, neurodegeneracije te slabosti. Upala niskoga stupnja također je prediktor smrtnosti u starijih pojedinaca koji pate od različitih patologija.[4,6]

Predloženo je nekoliko poveznica između upale i pogoršanja zdravlja u starijoj dobi, a IL-6 i CRP među najčešće su korištenim pokazateljima upale.[5,6] Čak i u odsutnosti kroničnih stanja, cirkulirajući čimbenici upale poput interleukina IL-6, čimbenika nekroze tumora (TNF)-alfa i topljivog TNF receptora-1 (TNFR-1) te C-reaktivnog proteina (CRP), najčešće su dva do četiri puta veći u starijih u usporedbi s mlađim osobama. CRP se sintetizira u jetri, a hs-CRP je osjetljiv biomarker sustavne upale. Razine upalnog biomarkera hs-CRP u plazmi predviđaju vaskularni rizik s procjenom učinka jednako važnim kao kod ukupnog ili LDL kolesterola.[5,7]

Ciljevi su ovoga članka:

- dokazati prisutnost sustavne upale istraživanjem razine upalnih posrednika (IL-6, hs-CRP) u perifernoj krvi u osoba s HFpEF i komorbiditetima
- dokazati povezanost upalnih posrednika s razinom dijastoličke disfunkcije i parametrima remodeliranja srca (hipertrofija lijeve klijetke, indeks volumena lijevog atrija)
- ustanoviti korelaciju između razine upalnih medijatora s razinom HF-a prema NYHA klasifikaciji i razini dijastoličke disfunkcije
- ustanoviti korelaciju između upalnih posrednika s razinom karotidne ateromatoze i endotelne disfunkcije: debljina intima-medije, omjer krajnje sistoličke / krajnje dijastoličke brzine (PSV/EDV) zajedničke karotidne arterije (ACC).

Bolesnici i metode

Način istraživanja. Istraživanje je provedeno u osoba starijih od 65 godina, od kojih je njih 60 imalo znakove HFpEF-a i komorbiditete, dok se zdrava, kontrolna skupina bez komorbiditeta sastojala od njih 18. Najčešći su komorbiditeti bili: arterijska hipertenzija, šećerna bolest, pretilost i kronična bubrežna disfunkcija. Iz istraživanja su isključeni bolesnici s dijagnosticiranom kroničnom koronarnom bolesti srca, kroničnom opstruktivnom plućnom bolesti, progresivnom anemijom, akutnim i kroničnim upalnim, zloćudnim, progresivnim neurološkim bolestima te demencijom, osobe na dugotrajnoj protuupalnoj terapiji i oni u kojih su vrijednosti hs-CRP-a bile >10 mg/L.

effect of T and B cells, the production of defective antibodies, an increase in the production of autoantibodies, and in chronic low-grade inflammation.[4,5] Among its most important features are slight elevations of the concentrations of pro-inflammatory cytokines, chemokines, and adipokines, such as interleukin-1ß (IL-1ß), IL-6, tumor necrosis factor-α (TNF-α), and monocyte chemoattractant protein-1.[4] Pro-inflammatory cytokines stimulate the synthesis of C-reactive protein (CRP) in the liver, the level of which has been shown to increase in elderly individuals. The mechanisms for CRP elevation in patients with heart failure have not been completely defined[6]. Possible theories include organ-congestion and hypoperfusion which causes secretion of IL-6 from hepatic, renal, endothelial, mononuclear, and, of course, cardiac myocytes. IL-6 causes CRP production in liver. This increased production could reflect a phenomenon of inflammatory state which causes development of diastolic dysfunction. While clinical signs and symptoms of inflammation are minimal or absent, this condition contributes to various molecular pathologies, leading to vascular damage and insulin resistance and, therefore, increases the risk of developing type 2 diabetes, cardiovascular disease, stroke, cancer, sarcopenia, neurodegeneration, and frailty. Furthermore, low-grade inflammation predicts mortality in elderly individuals who are affected by various pathologies.[4,6]

A number of links have been proposed between inflammation and deterioration of health at older ages. IL-6 and CRP are among the most commonly used indicators of inflammation.[5,6] Even in absence of chronic conditions, circulating inflammatory factors, such as interleukin IL-6, tumor necrosis factor (TNF)-α and soluble TNF receptor-1 (TNFR-1), and C-reactive protein (CRP) are usually two to four times higher in the elderly compared to young subjects. CRP is synthesized in the liver, and high-sensitivity CRP (hs-CRP) is a sensitive biomarker of systemic inflammation. Plasma levels of the inflammatory biomarker hs-CRP predict vascular risk with an effect estimate as large as that of total or high-density lipoprotein cholesterol.[5,7]

The aims of this article were:

- to prove the presence of systemic inflammation by investigation of the level of inflammatory mediators (IL-6, hs-CRP) in peripheral blood in patients with development of HFpEF and comorbid diseases,
- to demonstrate the association of the inflammatory mediators with the level of diastolic dysfunction and the parameters of heart remodeling (left ventricular hypertrophy, left atrial volume index),
- to establish the correlation between the level of the inflammatory mediators and the level of heart failure according to the New York Association (NYHA stadium) and the level of diastolic dysfunction,
- to establish the correlation between the inflammatory mediators and the level of carotid atheromathosis and endothelial dysfunction: intima-media thickness, the end-systolic/end-diastolic velocity (PSV/EDV) ratio of the common carotid artery (ACC).

Patients and Methods

Study design. This study examined elderly people aged >65 years, 60 of whom had signs of HFpEF and comorbid diseases, while 18 comprised the healthy control group with no comorbidities. Of the most common comorbid diseases, the follow-

Osnovna ispitivanja. Svi su ispitanici odgovorili na prethodno pripremljene upitnike o anamnezi, prisutnosti komorbiditeta, pušenju, primjeni lijekova te na *Minnesota Living With Heart Failure Questionnaire*, koji je dijelom prilagođen za dobnu skupinu stariju od 65 godina. U Institutu za gerontologiju u Skopju provedena su mjerenja tjelesne težine i visine, indeksa tjelesne mase (BMI), arterijskoga tlaka i frekvencije srca, 12-kanalni elektrokardiogram te vađenje krvi za laboratorijske pretrage.

Upalni medijatori. Vrijednost hs-CRP analizirana je s pomoću turbidimetrije (mg/L; Integra 400 Roche) na uzorcima periferne venske krvi. Vrijednost IL-6 analizirana je s pomoću metode ECLIA (pg/mL) na uzorcima periferne venske krvi. Analiza medijatora provedena je u privatnome biokemijskom laboratoriju „Adrialab" (Sinlab).

Ehokardiografija i obojeni dopler karotidnih krvnih žila. Svim je pacijentima učinjen 2-D ehokardiografski pregled i dopler karotida s pomoću ultrazvuka (*SonoSite MicroMaxx*, SAD) s 2,5-MHz-nim sondama za sve pretrage. Sva su ehokardiografska mjerenja provedena prema Preporukama za ehokardiografsku kvantifikaciju srčanih šupljina u odraslih iz 2015.[8] i Preporukama za ehokardiografsko mjerenje dijastoličke funkcije lijeve klijetke iz 2016[9]. Stupnjevanje je provedeno na temelju nekoliko prethodno ustanovljenih kriterija: prvi stupanj – kasna relaksacija, drugi stupanj – pseudonormalizacija, te treći stupanj – restriktivni tip[9].

Razina zatajivanja srca procijenjena je prema klasifikaciji *New York Classification for Heart Failure* (NYHA) sukladno podatcima iz upitnika. Pacijenti su u skupinama analizirani prema klasifikaciji NYHA (II. –IV.).

Razina promjena na karotidama. Stupanj 0.: nema plaka ni zadebljanja intime-medije (IMT) <1 mm; stupanj 1.: zadebljana ((≥1mm) IMT; stupanj 2: nestenotski plak (sa zadebljanjen IMT-a ili bez njega); stupanj 3.: stenotski (≥50 %) plak. Određene su maksimalne sistoličke brzine i krajnje dijastoličke brzine (PSV/EDV) u zajedničkoj karotidnoj arteriji.

Rezultati

U ispitanoj skupini ispitanika starijih od 65 godina s kliničkom i ehokardiografskom dijagnozom HFpEF-a otkriveno je da manji dio oboljelih, njih šest, ima samo jedan komorbiditet (12,2 %), njih 18 (36,7 %) imalo je dva komorbiditeta, jednak broj ispitanika imao je tri komorbiditeta, dok su u njih sedam (14,3 %) pronađena četiri komorbiditeta (**slika 1**). Broj komorbiditeta nije znatno ovisio o bolesnikovu spolu (dvostrani Fisherov egzaktni test, p = 0,53). Što se tiče tipova komorbiditeta, svi su pacijenti iz istražene skupine imali arterijsku hipertenziju, i to 36 (75,0 %) žena i 13 (26,5 %) muškaraca.

Dijabetes je registriran u 34 (69,4 %) bolesnika, nešto izraženije, no neznačajno, u muškaraca – 10 (76,9 %) nasuprot 24 (66,67 %) žene (dvostrani Fisherov egzaktni test, p = 0,7). Dislipidemija nije pronađena u ovoj skupini pacijenata. Kroničnu bubrežnu insuficijenciju imala su 24 (49,0 %) pacijenta, što je u muškaraca nešto učestalije, ali neznačajno – 7 (53,9%) nasuprot 17 (47,2%) (hi-kvadrat test, p = 0,68). Šesnaest je pacijenata (32,7 %) bilo pretilo, što je među ženama nešto učestalije, ali neznačajno – 12 (33,3 %) nasuprot 4 (30,8 %) (dvostrani Fisherov egzaktni test, p = 1,0). Glavna glavna kardiovaskularna obilježja ispitane skupine prikazana su u **tablici 1**.

ing were investigated: arterial hypertension, diabetes mellitus, obesity, and chronic renal dysfunction. Patients with verified chronic coronary arterial diseases, chronic obstructive pulmonary disease, progressive anemia, acute and chronic inflammatory diseases, malignant diseases, and also patients with progressive neurologic diseases and dementia as well as patients being on long-term anti-inflammatory therapy, and patients in whom the values of hs-CRP was >10mg/L were excluded.

Basic investigations. All examinees responded to previously prepared questionnaires addressing their medical history, presence of comorbidities, smoking, medication use, and Minnesota Living With Heart Failure Questionnaire, partly adjusted for the age group above 65 years. Measurements of body weight and height, measurement of the body mass index (BMI), measurement of blood pressure, cardiac frequency, 12-channel ECG, and taking blood for laboratory analyses were performed at the Gerontology Institute "13 November", Skopje.

Inflammatory mediators. hs-CRP was analyzed with the following method: Turbidimetry, Integra 400 Roche) (mg/L) from samples of venous peripheral blood. IL-6–was tested using the ECLIA (pg/mL) method from samples of venous peripheral blood. The investigation of these mediators was performed in the "Adrialab" (Sinlab) private biochemical laboratory.

Echocardiography and color Doppler of carotid blood vessels. All patients underwent two-dimensional color Doppler echocardiography and color Doppler of carotid blood vessels with SonoSite MicroMaxx (USA), with 2.5-MHz probes for all examinations. All echocardiographic measures were performed according to the Recommendations for Cardiac Chamber Quantification by Echocardiography in Adults: An Update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging 2015[8] and Recommendation for the Evaluation of Left Ventricular Diastolic Function by Echocardiography**:** an Update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging 2016[9]. Gradation was performed according to several already established criteria: 1st degree – postponed relaxation, 2nd degree – pseudonormalization, and 3rd degree – restrictive type[9].

The level of the heart failure was estimated using the functional New York Classification for Hearth Failure (NYHA) according to the patients' data given in their questionnaires. The patients in the groups were investigated according to NYHA (II-IV).

Carotid score. score 0: no plaques and thickness of intima-media complex (IMT) < 1 mm; score 1: thickened (≥1 mm) IMT; score 2: non-stenotic plaque (with or without IMT thickening); and score 3: stenotic (≥50% stenosis) plaque. Determination of the pick-systolic velocity and end-diastolic velocity (PSV/EDV) of the associated carotid artery (ACC).

Results

In our investigated group of patients older than 65 years with clinical and echocardiographic diagnosis of HFpEF, it was found that a minority of patients had only one comorbidity 6 (12.2%), 18 (36.7%) had two comorbidities, and the same number of patients had three comorbidities, while 4 comorbid states were registered in 7 (14.3%) patients (**Figure 1**). The number of comorbid conditions did not depend significantly on the patients' sex (Fisher exact, two tailed, p=0.53).

Regarding the type of the comorbidities, all patients from the investigated group had arterial hypertension, 36 (75.0%) female and 13 (26.5%) male patients.

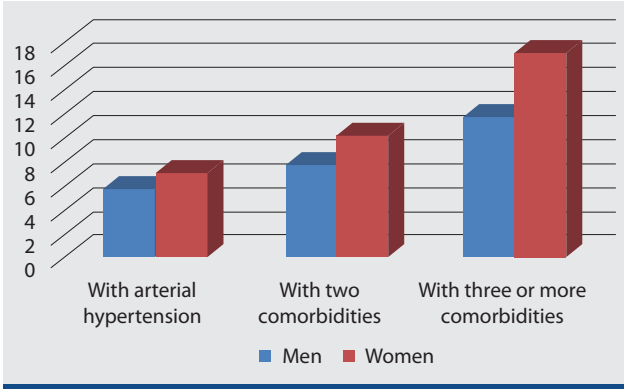

FIGURE 1. Multimorbidity in the investigated group.

Vrijednosti hs-CRP-a znatno su se pozitivno podudarale s brojem komorbiditeta u ispitanoj skupini (Spearmanov test, R = 0,473; p = 0,0006), odnosno više serumske vrijednosti hs-CRP-a bile su povezane s većim brojem komorbiditeta i obrnuto (**slika 2**). U skupini bez komorbiditeta 11 (36,7 %) pacijenata imalo je vrijednosti hs-CRP-a niže od 1 mg/L, a ostalih 19 (63,3 %) imalo je vrijednosti između 1 i 5 mg/L. Nije bilo ispitanika s komorbiditetima i vrijednostima hs-CRP-a nižima od 1 mg/L. Vrijednosti hs-CRP-a više od 5 mg/L registrirane su u 2/6 ispitanika s jednim komorbiditetom, u 10 (55,6 %) s dvama komorbiditetima te u 21 (84,0 %) s trima ili više komorbiditeta. Bolesnici bez komorbiditeta imali su mnogo niže vrijednosti hs-CRP-a u usporedbi s onima s dva, tri i više komorbiditeta (p = 0,00014, odnosno p = 0,00015). Statistički mnogo niže vrijednosti hs-CRP-a zabilježene su i u skupini oboljelih s jednim komorbiditetom u uspordbi s bolesnicima s tri i više komorbiditeta (p = 0,0003).

Vrijednost IL-6 značajno se pozitivno podudarala s brojem komorbiditeta u istraženoj skupini (Spearmanov test, R = 0,445, p = 0,001), tj. više serumske vrijednosti interleukina povezane su s većim brojem komorbiditeta i obrnuto (**slika 3**). U skupini bez komorbiditeta svi su bolesnici imali vrijednosti IL-6 nižu od 5,5 pg/L, dok su vrijednosti IL-6 bile više od 10 pg/L u jednog ispitanika s arterijskom hipertenzijom kao komorbiditetom, u njih 7 (38,89 %) s dvama komorbiditetima te u

TABLE 1. Basic major characteristics in the investigated group.

	Investigated sample
Women (%)	56%
Height (cm)	158 ± 7
Weight (kg)	62 ± 4
BMI (kg/m^2)	24 ± 3
Blood pressure – systole (mmHg)	150 ± 6
Blood pressure – diastole (mmHg)	79 ± 5
Heart rate (beats per minute)	62 ± 8
Serum cholesterol (mmol/L)	5.4 ± 1.0
HDL (mmol/L)	1.5 ± 0.5
LDL (mmol/L)	3.3 ± 0.4
Triglycerides (mmol/L)	1.3 ± 0.60
Fasting glycaemia (mmol/L)	5.3 ± 1.6
LAVI ml/m^2	37 ± 2.1
LVHI g/m^2	115 ±
E/A-ratio	0.95 ± 0.28
e' laterally (cm/s)	7.85 ± 0.5

LAVI (left atrial volume indexed on body mass), LVHI (left ventricular hypertrophy indexed on body mass), E/A (ratio between early and late diastolic filling of the left ventricle), e'laterally (early diastolic velocity of mitral ring laterally), BMI (body mass index).

Diabetes was found in 34 (69.4%) patients, non-significantly more frequently in the male patients – 10 (76.9%) vs 24 (66.67%) (Fisher exact, two tailed p=0.7). Dyslipidemia was not registered in this group of patients. Chronic renal insufficiency was found in 24 (49.0%) patients, non-significantly more frequently in the male patients – 7 (53.9%) vs 17 (47.2%) (Chi-square p=0.68). Sixteen patients (32.7%) were obese, non-significantly more frequently the women – 12 (33.3%) vs 4 (30.8%) (Fisher exact, two tailed p=1.0). Basic major cardiovascular characteristics of the investigated group are shown in **Table 1**.

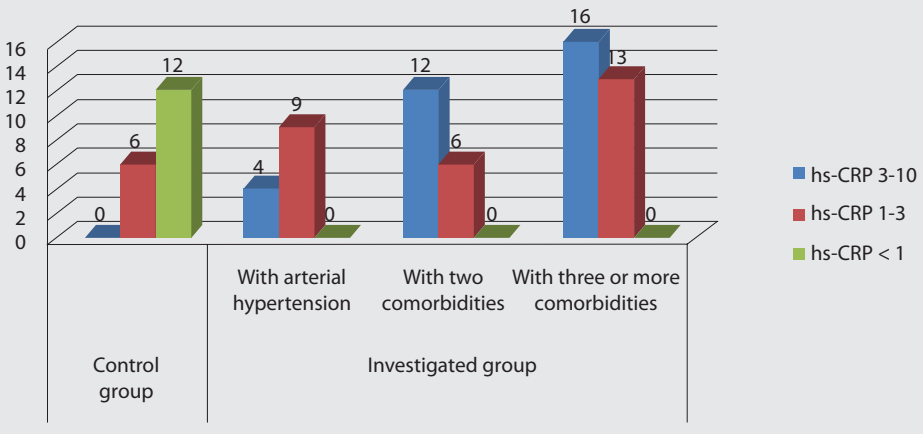

FIGURE 2. Values of hs-CRP in the investigated and in the control group.

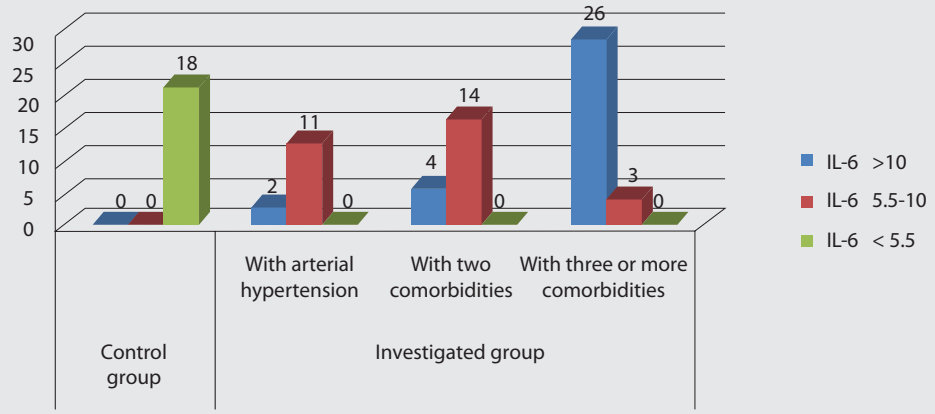

FIGURE 3. Values of IL-6 in the investigated and the control group.

18 (72 %) s tri i više komorbiditeta. Bolesnici bez komorbiditeta imali su mnogo niže vrijednosti interleukina u usporedbi s onima s trima ili više komorbiditeta (p = 0,0001, odnosno p=0,0001). Značajno niže vrijednosti interleukina zabilježene su i u onih s jednim i dva komorbiditeta (p = 0,002 odnosno p = 0,03).

Od ukupnoga broja bolesnika u ispitanoj skupini s HFpEF, povećani indeks volumena lijevog atrija (>34 mL^2) s pomoću oslikavanja ehokardiografijom, registriran je u njih 52 (86,6 %), što je odgovaralo ozbiljnosti dijastoličke disfunkcije, kao i broju i vremenskom trajanju komorbiditeta, dok je u kontrolnoj skupini povećani indeks volumena lijevog atrija registriran u šest bolesnika (10 %). Kod 55 (91,6 %) bolesnika registrirana je hipertrofija lijeve klijetke, dok je u kontrolnoj skupini ona utvrđena u njih 2 (11,1 %), i to u blažem obliku. Bolesnici s drugim stupnjem dijastoličke disfunkcije bili su najbrojniji; njih 46 (76 %) imalo je promjene na karotidi 1. stupnja, dvanaest (20 %) pacijenata imalo je 2. stupanj promjena, dok su dva pacijenta (3,3 %) imala 3. stupanj.

Vrijednosti hs-CRP-a i interleukina značajno su se pozitivno podudarale sa stupnjem HF-a prema NYHA (**tablica 2**). Povišene vrijednosti tih parametara pronađene su u pacijenata s višim stupnjem HF-a i obrnuto.

U ispitanoj je skupini vrijednost hs-CRP-a neznačajno korelirala s masom lijeve klijetke, maksimalnim vrijednostima volumnog indeksa lijevog atrija (LAVI maks.) i indeksom tjelesne mase te je značajno negativno korelirao s vrijednosti e' (R = -0,370; p = 0,009). Serumske vrijednosti hs-CRP-a smanjivale su se povišenjem vrijednosti parametra e' i obratno. U ispitanoj je skupini vrijednost IL-6 neznačajno korelirala s

TABLE 2. Correlation between hs-CRP and IL-6 and the degree of the heart failure (NYHA).

	Spearman R	p-level
hs-CRP / NYHA	0.522	0.000026
IL- 6 / NYHA	0.603	0.000001

The hs-CRP values significantly positively correlated with the number of the comorbidities of the investigated group (Spearman R=0.473; p=0.0006), i.e. the higher serum hs-CRP values were related to a greater (numerous) number of comorbidities, and vice-versa (**Figure 2**). In the group without comorbidities, 11 (36.7%) patients had hs-CRP values lower than 1 mg/L, while the other 19 (63.3%) had values ranging from 1 to 5 mg/L. There were no patients with comorbidities and hs-CRP values lower than 1 mg/L. The serum hs-CRP presented values higher than 5 mg/L in 2/6 patients with one comorbidity, 10 (55.6%) with two comorbidity, and in 21 (84.0%) with three and more comorbidities. The patients without comorbidities had significantly lower hs-CRP values in relation to the patients with two, three, and more comorbidities (p=0.00014, p=0.00015 respectively). Statistically significantly lower hs-CRP values were also registered in the group of patients with one comorbidity in relation to those with three and more comorbidities (p=0.0003).

The IL-6 value significantly positively correlated with the number of comorbidities in the investigated group (Spearman R=0.445; p=0.001), i.e. the higher serum interleukin values were associated with a greater number of comorbidities and vice-versa (**Figure 3**). In the group without comorbidities, all patients had IL-6 values lower than 5.5 pg/L, while the IL-6 values were higher than 10 pg/L in one patient with arterial hypertension as a comorbidity; in 7 (38.89%) patients with two comorbidities, and in 18 (72%) patients with three and more comorbidities. The patients without comorbidities had significantly lower interleukin values compared with the patients with three or more comorbidities (p=0.0001, p=0.0001 respectively). Significantly lower interleukin values were also registered in patients with one and two comorbidities (p=0.002; p=0.03 respectively).

Among patients of the investigated group with HFpEF, elevated LA volume index (>34 mL^2) was found in 52 (86.6%) by echocardiography, which corresponded to the seriousness of diastolic dysfunction, as well as to the number and time duration of the comorbid diseases, while elevated LA volume index was found in 6 (10%) patients in the control group. Left ventricular hypertrophy was found in 55 (91.6%) of the patients, compared with the control group where a low level was found in only 2 (11.1%) patients. Among patients with the second level of diastolic dysfunction, being the most numerous, 46 (76%) had carotid score 1, 12 (20%) patients had carotid score 2, and 2 (3.3%) patients had carotid score 3.

TABLE 3. Correlation between hs-CRP and IL-6 and the parameters of diastolic dysfunction.

	Investigated group		Control group	
	Spearman R	p-level	Spearman R	p-level
hs-CRP / e'	-0.370	**0.009**	-0.124	0.51
IL-6 / e'	-0.233	0.11	-0.054	0.77
hs-CRP / LVHI	0.163	0.28	-0.086	0.66
IL-6 / LVHI	0.108	0.48	-0.203	0.29
hs-CRP / LAVI max	0.021	0.89	-0.006	0.97
IL-6 / LAVI max	0.028	0.85	-0.376	**0.04**
hs-CRP / BMI	0.240	0.096	0.189	0.32
IL-6 / BMI	0.238	0.099	0.011	0.95

LAVI max (left atrial max volume indexed on body mass), LVHI (left ventricular hypertrophy indexed on body mass), e' laterally (early diastolic velocity of mitral ring laterally), BMI (body mass index).

vrijednosti e', masom lijeve klijetke, indeksom LAVI maks. te indeksom tjelesne mase (**tablica 3**).

U kontrolnoj je skupini vrijednost hs-CRP-a neznačajno korelirala s parametrom e', masom lijeve klijetke i indeksom tjelesne mase, dok je vrijednost interleukina neznačajno korelirala s parametrom e', veličinom lijeve klijetke i indeksom tjelesne mase, a negativno korelirala s indeksom LAVI maks. (R = -0,376; p = 0,04). Serumske vrijednosti interleukina u kontrolnoj su se skupini smanjivale povišenjem indeksa LAVI maks. i obrnuto.

Rasprava

Poznato je da je unutar posljednjih deset godina upala postala jedno od središnjih polja u istraživanju sistoličkog HF-a. Od bitne su važnosti i podatci koji upućuju na ulogu upalnih čimbenika u razvoju dijastoličke disfunkcije[10]. Danas je poznato da gotovo svaka stanična vrsta s jezgrom u miokardu, uključujući i srčane miocite, ima sposobnost otpuštanja proupalnih citokina kao odgovor na stres ili razne vrste oštećenja miokarda, čak i u odsutnosti sustavne imunosne aktivacije[11,12]. Razlika u patofiziologiji između zatajivanja srca sa smanjenom istisnom frakcijom (HFrEF) i HFpEF-a i dalje je nerazjašnjena, a učinkovite mogućnosti liječenja trenutačno nisu dostupne za HFpEF[13,14]. Stoga je potrebno pobliže razjasniti patofiziologiju HFpEF-a, što bi s moglo poboljšati ishod liječenja[15,16]. U brojnim je istraživanjima uočeno da su upalni markeri povišeniji u HFpEF-u, dok su u HFrEF-u povišeniji markeri istezanja srca[17]. Postoje brojni dokazi da je kronična upala u vezi s morbiditetom povezanim sa starenjem u oboljelih pojedinaca. Stoga je ovo istraživanje analiziralo skupinu osoba starije životne dobi s nakanom da ustanovi jesu li rezultati jednoga mjerenja razine dvaju proupalnih čimbenika, IL-6 i hs-CRP-a, povezana s kliničkim i ehokardiografskim parametrima te jesu li oni prediktori HFpEF-a.

Bolesnici s HFpEF-om često su stariji, većina njih ima anamnestičke podatke o arterijskoj hipertenziji te najčešće nekoliko komorbiditeta, uključujući dijabetes, pretilost, anemiju, bubrežnu disfunkciju, kroničnu opstruktivnu plućnu bolest i dr.[2,3]. Svaki od tih komorbiditeta utječe na strukturu i funkciju

The hs-CRP values and interleukin significantly positively correlated with the degree of the heart failure, analyzed with NYHA (**Table 2**). Elevated values of these parameters were found in patients with higher degree of heart failure, and vice-versa.

In the investigated group, hs-CRP non-significantly correlated with LV mass, LAVI max, and body mass index (BMI), and significantly negatively correlated with e' parameter (R = -0.370; p=0.009). The hs-CRP serum values decreased with the elevation of the e' parameter values and vice-versa. In the investigation group, IL-6 non-significantly correlated with e', LV mass, LAVI mass, and BMI (**Table 3**).

In the control group, hs-CRP non-significantly correlated with e', LV mass, and BMI, while interleukin non-significantly correlated with e', LV, and BMI, and negatively correlated with LAVI max (R = -0.376; p=0.04). The serum interleukin values in the control group decreased with elevation of the LAVI max parameter, and vice-versa.

Discussion

It is well-known that inflammation became one of the central fields of the investigation of the systolic heart failure over the last decade. There are also significant data which point to the role of inflammatory factors in development of diastolic dysfunction[10]. It is now known that almost every nuclear cell type in the myocardium, including the heart myocytes, is capable of radiating pro-inflammatory cytokines as a response to various damage of the myocardium or stress, even in the absence of systemic immunologic activation[11,12]. The difference in pathophysiology between heart failure with a reduced ejection fraction (HFrEF) and HFpEF remains poorly understood, and effective treatment options are currently not available for HFpEF[13,14]. Therefore, a better understanding of the pathophysiology of HFpEF is required, which may eventually help to improve patient outcomes[15,16]. Numerous studies have concluded that the inflammation markers are more elevated in HFpEF, while the markers of cardiac stretch were more elevated in HFrEF[17]. It is well documented that chronic inflammation is associated with aging-related morbidity in affected individuals. Therefore, the present study evaluated a group

klijetki i žila te je stoga važno ustanoviti je li HFpEF odvojena bolest koja zahtijeva specifičnu terapiju ili, jednostavno, zbroj komorbiditeta koji zahvaćaju populaciju starije životne dobi[3]. U ovom smo istraživanju ustanovili da, u usporedbi s relativno zdravom skupinom osoba starije životne dobi, bolesnici s HFpEF-om imaju abnormalnosti u strukturi srca i krvnih žila. U usporedbi s kontrolnom skupinom, ispitanici s HFpEF-om imaju značajnije remodeliranje srca (koncentričnu hipertrofiju lijeve klijetke i dilataciju lijevog atrija), kao i dijastoličku disfunkciju (povišeni E/e', niži e') te abnormalnu vaskularnu funkciju (zadebljanje IMT-a, nestenotske i stenotske plakove karotidnih krvnih žila). Među njima su komorbiditeti povezani s jedinstvenim kliničkim, strukturno-funkcionalnim te prognostičkim profilom.

U ovom je istraživanju serumska razina IL-6 i hs-CRP-a pokazala značajnu povezanost parametara dijastoličke disfunkcije s razinom hipertrofije lijeve klijetke i parametrima remodeliranja lijevog atrija. Ta nam činjenica pruža klinički važne podatke o sustavnim imunosnim abnormalnostima u pacijenata s dijastoličkom disfunkcijom. No, nije bilo značajne razlike u vrijednosti hs-CRP-a < 3 između kontrolne i ispitane skupine sa samo jednim komorbiditetom (arterijska hipertenzija). To je pokazatelj da upala i oksidativni stres mogu biti odgovorni za prelazak pacijenata starijih od 65 godina s dijastoličkom disfunkcijom u simptomatsko HFpEF, posebice s obzirom na to da većina starijih ljudi ima povezane bolesti u kojima je dokazana sustavna upalna priroda. Stupanj promjena na karotidama također je u pozitivnoj korelaciji s razinom dijastoličke disfunkcije koja upućuje na ulogu ateroskleroze velikih krvnih žila u nastalom HFpEF-u. Hipertenzija i dijabetes najveći su mogući prekursori u nastalom HFpEF-u, a prelazak iz asimptomatske u simptomatsku dijastoličku disfunkciju najvećim je dijelom posljedica razvoja dilatacije i disfunkcije lijevog atrija, kao i progresije hipertrofije lijeve klijetke. Istraživanje je pokazalo pozitivnu korelaciju s parametrima remodeliranja lijevog atrija, što podržava pretpostavku da upala nastala remodeliranjem lijevog atrija uzrokuje prelazak u asimptomatsku dilataciju atrija, što je karakteristično za proces starenja u HFpEF-u. Razina dijastoličke disfunkcije značajno je korelirala s razinom karotidne ateromatoze, razinom upalnih posrednika (medijatora) u krvi te razinom HF-a. Vrijednost ranoga mitralnog kretanja u mitralnome prstenu mjerena je na lateralnoj stijenci (e') koja izravno odražava dijastoličku funkciju, a smanjuje se starenjem, što upućuje na smanjenje relaksacije lijeve klijetke. U ovom istraživanju ona je bila u izravnoj korelaciji s brojem i trajanjem komorbiditeta, što upućuje na njihov utjecaj na razvoj dijastoličke funkcije i HFpEF-a. Pacijenti s nižim vrijednostima e' imaju i višu razinu upalnih medijatora u krvi, što upućuje na utjecaj sustavne upale na smanjenje relaksacije lijeve klijetke[17,18].

Nedostatak je našeg istraživanja činjenica da nismo bili u mogućnosti istražiti više upalnih posrednika – TNF alfa, IL-18 itd.

U ovom su istraživanju vrijednosti hs-CRP-a korelirale s varijablama karotidne ateromatoze (zadebljanje intima-medije, prisutnost plaka), ali je u isto vrijeme bila prisutna i korelacija s parametrima dijastoličke disfunkcije u zdravoj, kontrolnoj skupini. To pokazuje da hs-CRP, osim toga što je provjereni marker ukupnoga kardiovaskularnog rizika, također pomaže u stratifikaciji pacijenata s dijastoličkom disfunkcijom koji još nemaju HFpEF, u predviđanju pojave HFpEF-a te pri donošenju terapijske odluke za svakog bolesnika. Na-

of seniors to determine whether results of a single measurement of the levels of two pro-inflammatory factors, IL-6 and hs-CRP, were associated with clinical and echocardiography parameters and whether they were predictors of HFpEF.

Patients with HFpEF are commonly older, and the majority have a history of arterial hypertension and frequently multiple comorbidities including diabetes, obesity, anemia, renal dysfunction, chronic obstructive pulmonary disease, etc.[2,3]. Each of these comorbidities has an impact on ventricular and vascular structure and function, and it is thus important to consider if HFpEF is a separate disease which requires specific therapy or whether it is simply an effect of cumulative comorbidities which follow the older population[3]. Our study demonstrated that, compared with a relatively healthy group of elderly people, HFpEF patients have abnormalities in cardiac and vascular structure. Compared with the heathy control group, these patients had greater cardiac remodeling (concentric left-ventricular hypertrophy and left-atrial dilatation), as well as diastolic dysfunction (elevated E/e', lower e'), and abnormal vascular function (IMT thickness, non-stenotic and stenotic plaques of carotid blood vessels). These comorbidities are associated with a unique clinical, structural, functional, and prognostic profile.

In our study, the serum level of IL-6 and hs-CRP showed significant association with the parameters of diastolic dysfunction, as well as with the level of left-ventricular hypertrophy and the parameters of the left atrial remodeling. This fact provides clinically significant information on the systemic immune abnormalities in patients with diastolic dysfunction. However, for hs-CRP values <3, there was no significant difference between the control and the investigated group, with only one comorbidity (arterial hypertension). This indicates that the progression for the patients >65 years with diastolic dysfunction into symptomatic HFpEF could be caused by inflammation and oxidative stress, especially since the majority of the elderly have associated diseases for which the systemic inflammatory nature has been demonstrated. Carotid score was is also in positive correlation with the level of diastolic dysfunction, which points to the role of atherosclerosis of the major blood vessels in occurring HFpEF. Hypertension and diabetes are the greatest possible precursors for HFpEF, and the progression from asymptomatic into symptomatic diastolic dysfunction is due, to a great extent, to development of left atrial dilatation and dysfunction, as well as to progression of left-ventricular hypertrophy. Our study showed positive correlations with parameters of left atrial remodeling, which corroborates that inflammation, through the left atrial remodeling, leads to progression from asymptomatic atrial dilatation, which is characteristic for the process of aging in HFpEF. The level of diastolic dysfunction significantly correlated with the level of carotid atheromatosis, the level of the inflammatory mediators in blood, and the level of the heart failure. The value of early mitral motion in mitral annulus, measured on the lateral side (e'), and which directly reflects diastolic function, falls with old age, which points to a decrease of the left-ventricular relaxation. In this study, it was in direct correlation with the number and the time-duration of comorbidities, which pointed to their impact on the development of diastolic function and development of HFpEF. Patients with lower e' also had a higher level of inflammatory mediators in the blood, which indicates the impact of systemic inflammation in decreasing the left-ventricular relaxation[17,18].

sumično odabrani rezultati studija vezani za hs-CRP bili su od središnje važnosti za razumijevanje protuupalnih učinaka terapije statinima te su, dosljedno, pokazali da su razine hs-CRP-a tijekom terapije bile jednako bitan prediktor preostaloga kardiovaskularnog rizika kao i vrijednost LDL kolesterola[18]. No, premda je hs-CRP klinički uporabljiv kao biomarker za predviđanje rizika, većina studija o mehanizmu djelovanja pokazuju da sam CRP nije vjerojatan predmet intervencije. Ako je CRP nizvodni biomarker ateromatoze, što usporedivi podatci pokazuju o uzvodnome „sekundarnom glasniku" citokinu IL-6? Kao prvo, kao i hs-CRP, vrijednosti IL-6 izmjerene u navodno zdravoj populaciji također predviđaju budući vaskularni rizik; to je prvotno zamijećeno u muškaraca u istraživanju iz 2000. godine, nakon toga je potvrđeno i u žena te je zatim reproducirano u više od 25 prospektivnih epidemioloških kohortnih studija diljem svijeta[19,20]. IL-6 izravno sudjeluje u preobrazbi srčanih miocita u fibroblaste te su zato promjene u koncentraciji IL-6 izravno povezane s disfunkcijom srca te promjenama kardiovaskularne matrice, a to je i razlog zbog kojeg se IL-6 često spominje kao biomarker remodeliranja koji je neovisan o ostalim upalnim markerima i kao čimbenik rizika za neželjene kardiovaskularne događaje te za razvoj HF-a[20,21]. Uzvodno kretanje u upalnom padu iz CRP-a u interleukin IL-6 te IL-1 pruža nove terapijske prilike za zaštitu od stvaranja plaka usredotočene na središnji signalni sustav IL-6 i u konačnici na sprječavanje proizvodnje IL-1β.

Navedeni rezultati idu u korist tvrdnje da je IL-6, citokin koji je ključni medijator u brojnim upalnim, alergijskim i infektivnim bolestima, također važan za stanje kronične upale, a njegova korelacija s razinom dijastoličke disfunkcije i vaskularne ateromatoze može utjecati na prijelaz te kronične upale u fibrozu miokarda koja je dokazana u ispitivanjima na životinjama.

U ovom smo istraživanju pokazali da pacijenti s dijastoličkom disfunkcijom imaju povišene upalne posrednike koji odgovaraju ozbiljnosti bolesti, no pitanje imaju li ti parametri prognostičke implikacije predmet je brojnih drugih istraživanja. Prošli rezultati idu u korist tvrdnji da samo povišene vrijednosti hs-CRP-a mogu biti iskorištene kao čimbenik rizika u starijoj „zdravoj populaciji" za pojavu HF-a i drugih kardiovaskularnih događaja[16]. Dijagnoza HF-a bez simptoma osnova je za unaprjeđenje učinkovitosti terapije. Određenje specifičnih i osjetljivih biomarkera koji odražavaju složenu patofiziologiju HF-a moglo bi se iskoristiti i kao važno kod probira pacijenata zbog HF-a kojima je potrebna dodatna obrada[4,9]. Općenito govoreći, klinička ispitivanja u kojima se nastoji modulirati upalni proces u pacijenata s HF-om najčešće su neuspješna, osim u slučaju nekih manjih podgrupa s opsežnim protuupalnim pristupom. To ne mora značiti kraj razdoblja citokina, ali je pokazatelj izazova toga terapijskog pristupa u moduliranju mreže citokina, što predočuje korisne, ali i štetne učinke na remodeliranje miokarda.

Vrlo je vjerojatno da je HFpEF jedinstvena sinergijska interakcija između učinaka procesa starenja, arterijske hipertenzije i komorbiditeta, a ponajviše kardiovaskularnih, koji uzrokuju srčano i vaskularno remodeliranje i disfunkciju uz izostanak drugih bolesti srca, što zatim vodi do smanjene crpne snage i istisne frakcije[13,14,22]. Dosad je jedini oblik prevencije HFpEF-a bilo liječenje čimbenika rizika. Otkriće novih, višedimenzionalnih strategija koje bi poboljšale endotelnu disfunkciju i remodeliranje srca znači izazov. Nove i nasumično odabrane strategije nužne su za otkrivanje terapije

The limitations of our study are that we were not able to investigate more inflammatory mediators – TNF-alpha, IL-18, etc.

In the present study, hs-CRP was correlated with the parameters of carotid atheromatosis (IMT thickening, plaques presence), but there was also a correlation with the parameter of diastolic dysfunction in the healthy control group. It indicates that hs-CRP, besides being a verified marker for total cardiovascular risk, also helps perform a risk stratification of patients with diastolic dysfunction who still do not have symptoms of HFpEF in order to predict occurrence of HFpEF and to adequately decide an individual therapeutic program for each patient. Furthermore, randomized trial data addressing hs-CRP have been central to understanding the anti-inflammatory effects of statin therapy and have consistently demonstrated on-treatment hs-CRP levels to be as powerful a predictor of residual cardiovascular risk as on-treatment levels of low-density lipoprotein cholesterol[18]. However, although hs-CRP is clinically useful as a biomarker for risk prediction, most mechanistic studies suggest that CRP itself is unlikely to be a target for intervention. If CRP is a downstream biomarker for atheromatosis, what do comparable data for the upstream "secondary messenger" cytokine IL-6 show? First, like hs-CRP, IL-6 levels measured in apparently healthy populations also predict future vascular risk; this observation was initially made in men in 2000, confirmed in women, and subsequently reproduced in more than 25 prospective epidemiologic cohorts worldwide[19,20]. IL-6 directly participates in conversion of cardiac myocytes into fibroblasts, and because of that the changes in IL-6 concentrations are directly related with cardiac dysfunction and changes of the cardiovascular matrix. Therefore, IL-6 is frequently discussed as a "remodeling" biomarker and also as independent from the other inflammatory markers, as a risk-factor for unwanted cardiovascular events and development of heart failure[20,21]. Moving upstream in the inflammatory cascade from CRP to interleukin IL-6 to IL-1 provides novel therapeutic opportunities for atheroprotection that focus on the central IL-6 signaling system and ultimately on inhibition of IL-1β-production.

Our results corroborate that IL-6, a cytokine which is a key mediator in many inflammatory, allergic, and infective diseases, is also significant in the states of chronic inflammation, while its correlation with the level of diastolic dysfunction and vascular atheromatosis could influence progression of this chronic inflammation into myocardial fibrosis that has been demonstrated in animal studies.

In this study, we showed that patients with diastolic dysfunction have elevated inflammatory mediators, which correspond to the seriousness of the disease, but whether these parameters have prognostic implications is the subject of many other studies. Existing results indicate that only the elevated hs-CRP values can be used as a screening risk factor in the senior "healthy population" for heart failure and other cardiovascular events[16]. Diagnosis of heart failure without symptoms is a basic way to improve the therapy efficacy. Determination of specific, sensitive biomarkers, which reflect the complex pathophysiology of heart failure, could also be used as a significant parameter for screening patients with heart failure, who need further extensive diagnostic examinations[4,9]. Generally, clinical studies in which efforts are made to modulate the inflammatory processes in patients with heart failure have to a great extent been disappointing, except for some smaller subgroups with an extensive anti-inflammatory approach. This

HFpEF-a temeljene na dokazima[13]. Povezivanje novih znanja u kardiologiji s aspektom procesa starenja i komorbiditetima u gerijatrijskoj kardiologiji imalo bi veliku važnost u otkrivanju novih oblika liječenja[23,24].

Premda je nekoliko pokušaja liječenja toga stanja primjenom protuupalnih strategija liječenja kako bi se smanjila kardiovaskularna smrtnost doživjelo neuspjeh, usredotočenost na cijelu CRP/IL-6/IL-1 os s monoklonskim protutijelima moglo bi otvoriti nove terapijske putove u kardiovaskularnoj patologiji te ponuditi nove uvide u proces starenja srca[22-24].

does not have to mean the end of the era of cytokine, but only indicates the challenges of this therapeutic approach for modulating the cytokine net, which represent the useful as well as the harmful effects on myocardial remodeling.

It is quite probable than HFpEF represents a unique synergistic interaction between the effect of the aging process itself, hypertension and comorbidities, primarily cardiovascular comorbidities, which cause heart and vascular remodeling and dysfunction in absence of other cardiac diseases, which lead to decreased pump power and ejection fraction[13,14,22]. Up to now, prevention for HFpEF could only be effective by treatment of the risk factors. Discovering novel multidimensional strategies which would improve endothelial dysfunction and heart remodeling poses a challenge. Novel, randomized strategies are needed for discovering evidence-based treatment for HFEF[13]. Connecting discoveries in cardiology with an aspect of the aging process and comorbidities in geriatric cardiology would have larger significance in discovering novel modalities in the treatment[23,24].

While several attempts to target this condition using therapeutic anti-inflammatory regimens to reduce cardiovascular mortality have failed, targeting the whole CRP/IL-6/IL-1 axis with monoclonal antibodies could open new therapeutic lines in cardiovascular pathology and offer novel insights into the aging process of the heart[22-24].

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