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STUDY DESIGN

Effect of melatonin on heart failure: design for a double-blinded randomized clinical trial

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Abstract

Aims Current studies indicate that melatonin can counteract renin–angiotensin–aldosterone system and sympathetic over activity in heart failure (HF) and might have a protective and repairing effect on cardiovascular injuries, skeletal muscle weakness, and metabolic abnormalities, which are common pathological processes in patients with HF. The MeHR trial (Melatonin for Heart Failure with Reduced Ejection Fraction) aims to evaluate the effect of oral melatonin on myocardial, skeletal muscle, and metabolic dysfunctions in HF, which leads to lower quality of life and increased morbidity and mortality in these patients.
Methods and results This is a double-blind randomized clinical trial with two parallel arms of 1:1 allocation, which recruits 90 outpatients with HF with reduced ejection fraction. Participants receive 10 mg tablets of melatonin or placebo for 24 weeks. The primary outcomes are changes in echocardiographic indexes of HF and serum levels of N terminal pro brain natriuretic peptide. Secondary outcome is a composite clinical endpoint score including all-cause mortality, hospitalization for HF, and change in the quality of life during the study. Other outcomes are the evaluation of melatonin attributable adverse effects, flow-mediated vasodilation, skeletal muscle mass, exercise capacity, and serum markers of inflammation, oxidative stress, and metabolism. Statistical analysis will include simple unadjusted analyses for the detection of differences between groups and changes in outcomes and also a generalized linear mixed model to explore potential associations between outcomes and participant characteristics.
Conclusions The results of this comprehensive study might elucidate the safety of oral melatonin in patients with HF and provide some evidence on its effectiveness as an adjunctive therapy to enhance the well-being of these patients.

Keywords Melatonin; Heart failure with reduced ejection fraction; Muscle wasting

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Introduction

Heart failure (HF) is a complex clinical syndrome characterized by the impairment of heart to circulate blood and is the ending continuum of diverse heart diseases such as ischemic heart disease, myocardial abnormalities, and metabolic disorders.[1] HF is classified to reduced (<40%), borderline (40–50%), and preserved (>50%) ejection fraction (EF). HF with reduced EF (HFrEF) accounts for about 50% of all HF patients in USA. These patients usually have frequent hospitalizations and excessive health resource utilization, which is a burden on patient's family and community.[2] Chronic comorbidities of HF like muscle wasting or sarcopenia, which usually precedes cardiac cachexia, diabetes, and metabolic syndrome, contribute to a great degree to the poor outcome of patients. Recent studies try to target these conditions as well as myocardial dysfunction.[3,4]

Melatonin is an endogenous hormone secreted from pineal gland mainly at night and has a variety of physiological functions besides its well-known role in organizing circadian rhythms.[5] The effect of melatonin on cardiovascular health is the topic of ongoing research. It has direct and indirect

beneficial effects on cardiovascular system via the inhibition of oxidative stress, reperfusion injury, apoptosis, fibrosis, pathological remodelling of the myocardium, and immune and metabolic regulation.[6]

In HFrEF, regardless of the primary insult, abnormalities in myocardial structure and energy metabolism lead to myocardial failure, which initiates downstream deterioration of neurohormonal regulation to compensate for pump failure.[7] Up-regulation of renin–angiotensin–aldosterone system (RAAS) has a pivotal role in fluid retention and vasoconstriction, which in a long term leads to cardiac overload, myocardial fibrosis and apoptosis, and left ventricular remodelling.[8] Activation of RAAS inhibits protein synthesis via the inhibition of growth hormone (GH)/insulin-like growth factor 1 (IGF-1) and adenosine monophosphate-activated protein kinase (AMPK) signalling pathways. RAAS activation also causes protein degradation by ubiquitin–proteasome system up-regulation, induces inflammatory state, and increases reactive oxygen species (ROS) formation, which all lead to myocyte degradation in both heart muscles and skeletal muscles and worsen cardiac cachexia.[9]

Numerous *in vivo* and *in vitro* studies propose that melatonin antagonizes angiotensin II effects on cardiovascular system by inducing vasodilation via its central and peripheral receptors and by reversing the catabolic state developed by angiotensin II.[9] Melatonin activates GH/IGF-1 signalling by the induction of PI3k/AKT/mTOR pathway, activates AMPK, and regulates mitochondrial biogenesis to allow protein synthesis and apoptosis reduction.[10]

Furthermore, melatonin suppresses inflammation by modulating key transcription factors such as nuclear factor kappa B and signal transducer and activator of transcriptions, and by the suppression of pro-inflammatory cytokines, isoforms of inducible nitric oxide synthase and cyclooxygenase.[11] It is shown that melatonin exerts anti-oxidant effects by directly scavenging ROS as well as up-regulating antioxidant enzymes.[12] Therefore, melatonin can enhance effects of RAAS inhibitors in HF and cardiac cachexia.

Other up-regulated neurohormones in HF are catecholamines and glucocorticoids, which are stress hormones, enhancing catabolic state and deteriorating myocardial failure. Melatonin is supposed to reduce catecholamine and cortisol levels in experimental studies and might reverse their effects through its antioxidant properties and by activating anabolic signalling pathways.[9]

Overall, melatonin seems to be beneficial not only in preventing HF deterioration and improvement of myocardial function but also by potential reversal of its complications such as muscle wasting and cardiac cachexia.[13] Predisposing factors such as high blood pressure and abnormal blood lipid profile are other supposed targets of melatonin in HF.[14,15] The neurohormonal regulating role of melatonin besides its safety profile and possible beneficial effects on comorbidities of HF makes it a reasonable candidate in this disease; however, no clinical data are available in this regard.

Our main objective in Melatonin for Heart Failure with Reduced EF (MeHR) trial is to evaluate the effect of melatonin supplementation on echocardiographic indexes and serum levels of N terminal pro brain natriuretic peptide (NT-proBNP) in patients with HFrEF (with any underlying aetiology) after 24 weeks of intervention. As mentioned earlier, melatonin might reverse effects of RAAS over-activation on cardiovascular system via different pathways; thus, its effect in reducing myocardial stress should be ideally reflected in echocardiography and NT-proBNP levels.

Concurrently, we will evaluate the effect of melatonin on a composite clinical outcome, which includes mortality, morbidity, and patients' health-related quality of life, melatonin-related adverse effects, and whether melatonin supplementation will improve endothelial dysfunction, muscle mass and function, and psychological status of the patients. Also, changes in mediators of muscle and heart deterioration, including inflammatory, metabolic, and oxidative biomarkers of the serum will be assessed.

Study design

MeHR trial is a double-blind prospective phase II randomized placebo controlled clinical trial with two parallel arms of 1:1 allocation conducted on patients with HFrEF.

Ethics

This investigation conforms to the principles outlined in the *Declaration of Helsinki*, which have been approved by Ethical Committee of Isfahan University of Medical Sciences (Ethics code: IR.MUI.MED.REC.1397.067). The project and its aims are clearly explained to the patients, and the confidentiality of the data is ascertained; thereafter, all participants sign an informed consent. Patients receive appropriate medical management during the study, and they are informed that discontinuing participation in the project will not interfere with their medical management. The trial protocol has been registered at ClinicalTrials.gov [NCT03894683].

Study setting and participants

MeHR trial enrols patients with HFrEF from outpatient clinics of the Chamran cardiology hospital, which is the primary referral hospital of Isfahan province, Iran. The trial takes place in Isfahan Cardiovascular Research Institute, which is located in the vicinity of the hospital.

Men or women aged >18 years with systolic dysfunction [diagnosed in agreement with the 2016 European Society of

Cardiology (ESC) guidelines[1]] who are willing to participate in the study and provide informed consent are eligible for participation. Patients have to be symptomatic (New York Heart Association class II–III), and their symptoms and medications of HF have been stable for at least 3 months. Participants have to be on a guideline-directed medical therapy in concordance with the 2016 ESC guidelines before and during the study. They have to be on maximum tolerated dose of an angiotensin-converting enzyme inhibitor or an angiotensin-receptor blocker as well as a beta-blocker as tolerated (regarding patient's heart rate and blood pressure) and spironolactone/eplerenone unless contraindicated. Diuretics are given as needed, and their doses can be adjusted during the study, guided by the patient's symptoms. Device-based treatment (implantable cardioverter defibrillator/cardiac resynchronization therapy) has to be implemented at least 3 months prior to the enrolment. Otherwise, the patient has not been a candidate for device implantation based on pre-enrolment evaluations.

Exclusion criteria are chronic comorbidities or conditions suspected to influence the desired outcomes or to put the patients at risk for melatonin adverse effects. We will exclude patients with end stage and overt comorbidities (renal, liver, or pulmonary diseases), as these conditions need specific care and frequently limit medication prescription. Also, patients with untreated or uncontrolled metabolic, endocrine, and autoimmune diseases are not enrolled as the commencement of their treatment during the study will interfere with the trial endpoints. Details of the eligibility criteria are listed in *Table* 1.

Randomization and blinding

The method of randomization is block randomization with a fixed block size of four using the web site Randomization.com (http://www.randomization.com). An investigator who is not involved in any of the steps of patient assessments, patient enrolment, and data analysis codes the drug and the placebo and generates the sequence of codes.

Allocation concealment is performed by the same investigator using opaque, sealed envelopes, which are coded sequentially. After assessing for eligibility criteria and signing informed consent, patients are coded in the order of enrolment and assigned to the treatment according to the content of the sealed envelopes.

The main investigator who enrols the patients, all of the outcome assessors, the statistician, and the participants are kept blinded to the study groups.

Intervention

Patients are randomized to either melatonin group or placebo group. The treatment group receives oral melatonin (10 mg tablets), ingested every night at bedtime for 24 weeks. Melatonin and placebo tablets are manufactured by the Sepidteb Pharmaceutical Company. Placebo tablets are the same as melatonin tablets regarding their shape, smell, and taste and are prescribed the same as the melatonin tablets. Possible adverse effects of the treatment and adherence to the drug is monitored closely by calling patients at 1 week

Table 1 Eligibility criteria for MeHR trial participation

Inclusion criteria
1. Age $>$ 18 years
2. Ejection fraction <40 due to either ischemic cardiomyopathy or dilated cardiomyopathy
3. NYHA class II–III
4. Documented diagnosis of HFrEF and receiving guideline directed treatment for at least 3 months
5. Willing to participate in the study and providing informed consent
Exclusion criteria
1. Renal failure on haemodialysis or anticipated to require dialysis in the next 6 months
2. End-stage liver diseases (infectious, alcoholic, autoimmune, idiopathic)
3. Chronic obstructive pulmonary disease (Class D according to GOLD classification) or uncontrolled asthma
4. Untreated or uncontrolled diabetes mellitus, thyroid disease, RA, SLE, or any chronic disease requiring specific treatment prior to enrolment in the study.
5. Waiting for a procedure (CABG, PCI, valvular repair, implantation of a CRT or ICD, or cardiac transplantation) within the next 6 months
6. Pregnancy or planning to be pregnant in the next 6 months
7. Morbid obesity (BMI $>$ 35)
8. Acute ischemic heart event or revascularization procedure in the last 3 months
9. Cognitive or psychological disorders interfering with medication adherence
10. Regular supervised exercise or ingestion of muscle hypertrophy supplementations in the last 3 months
11. Vegetarian diet or sever restriction of protein in the diet in the last 3 months
12. Inability to attend follow-ups (distance and travelling problems, advanced medical disease such as cancer with a suspected survival of less than 6 months).
13. Increased risk of falling due to musculoskeletal or neurologic disorders
14. Previous known hypersensitivity to melatonin or a history of angioedema
15. Participation in another research

CABG, coronary artery bypass grafting; CRT, cardiac resynchronization therapy; GFR, glomerular filtration rate; GOLD, The Global Initiative for Chronic Obstructive Lung Disease; HFrEF: heart failure with reduced ejection fraction; ICD, implantable cardioverter defibrillator; NYHA, New York Heart Association; PCI, percutaneous coronary intervention; RA, rheumatoid arthritis; SLE, systemic lupus erythematosus.

after randomization and then every other week up to 12 weeks and every 4 weeks thereafter, up to 24 weeks. Patients are requested to return unconsumed pills at the scheduled interviews (Weeks 12 and 24), and pill count is carried out to determine adherence.

Outcomes

Participants are evaluated at Weeks 12 and 24 for the following outcomes.

Primary outcome

The main outcomes of the study are (i) changes in echocardiographic parameters including left ventricular EF (LVEF) and left ventricular end diastolic diameter and (ii) changes in serum concentration of NT-proBNP from baseline to Week 24. NT-proBNP levels will be corrected according to the age, sex, body mass index, renal function, and comorbidities of the patients.

Secondary outcomes

We will use a composite clinical endpoint as a secondary outcome. This is described previously and has been used in several clinical trials with promising results.[16] We will use a modification of this outcome according to the A-HeFT study.[17] Components of this scoring system are all-cause mortality, hospitalization for HF during the study, and change in the quality of life by Minnesota Living with Heart Failure Questionnaire (MLHFQ) at Week 24 (*Table* 2).

We will also measure changes from baseline in the mean of lean body mass by bioelectrical impedance analysis (BIA), grip strength, and 6 mi walk distance at Weeks 12 and 24, and lean body mass by dual-energy X-ray absorptiometry (DXA) at baseline and Week 24. Endothelial dysfunction [by changes in flow-mediated dilation (FMD)] will be evaluated from baseline to Week 24.

To evaluate the effect of melatonin on probable intermediate pathways common in the pathophysiology of both HF and sarcopenia, changes in the mean serum concentration of the following markers will be measured from baseline to Week 24: high-sensitivity C-reactive protein (hs-CRP), interleukin-1beta (IL-1β), tumour necrosis factor alpha (TNF-α), myostatin, malondialdehyde, total antioxidant capacity, fasting blood glucose, lipid profile, insulin, and IGF-1. Liver and renal function tests are also monitored at baseline and Week 24.

Changes in the psychological status of the patients including anxiety, depression, and sleep quality will also be studied. The socio-demographic and clinical data, level of physical activity, and nutritional status of the patients are documented as confounding factors, which will be used as adjusting variables in the final analysis. Possible adverse effects of melatonin will be monitored throughout the study. The timetable of the study according to the Standard Protocol Items: Recommendations for Interventional Trials (SPIRIT) 2013[18] is presented in *Table* 3.

Data collection methods

Echocardiography and flow-mediated dilation

All echocardiography and FMD measurements are performed by the same cardiologist who is unaware of the patients grouping. Transthoracic 2D colour echocardiography is performed using a multi-frequency (12 MHz) linear array transducer (GE Vivid 3.0, General Electric Vingmed Ultrasound), and echocardiographic parameters including the diameter of LV in both systole and diastole are measured. The LV systolic function and EF are measured adopting the Simpson method. The reference limits of all echocardiographic parameters are defined according to the guidelines of the American Society of Echocardiography and the European Association of Cardiovascular Imaging.[19] To determine intra-observer variability, echocardiographic parameters of a random sample of 30 experiments will be reanalysed by the same cardiologist at least 1 month later based on the recorded images in the echocardiographic system, and intra-class correlation coefficient will be calculated.

Flow-mediated dilation measurements are performed after at least 6 h of fasting in the morning in a quiet room with

Table 2 Composite clinical outcome measurement in the MeHR trial[a]

Outcome		Score
Survival during the study	Death	−3
	Survive	0
Hospitalization for heart failure during the study	Any hospitalization	−1
	No hospitalization	0
Change in quality of life measured at the last follow-up[b]	Increase by ≥10 points	+2
	Increase by 5–9 points	+1
	Change by <5 points	0
	Decrease by 5–9 points	−1
	Decrease by ≥10 points	−2
Total score		−6 to +2

[a]Adopted from Taylor AL *et al*.[17]
[b]Measured by Minnesota Living with Heart Failure Questionnaire.

Table 3 Schedule of enrolment, interventions and assessments of the MeHR trial, according to the SPIRIT 2013 statement[18]

TIMEPOINTS	STUDY PERIOD			
	Enrolment Week -1	**Allocation** Week 0	**Follow-up** Week 12	**Close-out** Week 24
ENROLMENT:				
Eligibility screening	X			
Informed consent	X			
Randomization		X		
Allocation		X		
INTERVENTIONS:				
Melatonin (10 mg, oral)		←	—	→
Placebo		←	—	→
ASSESSMENTS:				
Socio-demographic data		X		
Clinical data and physical examination		X	X	X
Echocardiography		X		X
Flow Mediated Dilation		X		X
Health related quality of life		X	X	X
Hospitalization and mortality		X	X	X
Melatonin adverse effects		X	X	X
Bio-impedance		X	X	X
Dual-energy x-ray absorptiometry		X		X
Grip strength		X	X	X
6 Minute walk test		X	X	X
Blood tests		X		X
Depression, anxiety, and sleep quality		X		X
physical activity and nutritional status		X		X

normal temperature (23–25°) following the related guideline.[20] Patients are requested to avoid exercise and consumption of alcohol or caffeine for at least 8 h prior to testing. The diameter of the brachial artery is determined 4–5 cm above the antecubital fossa of the non-dominant hand, with the patient in supine position and arm rested on the table, by a high-frequency vascular probe and the 2D grey scale continuous imaging. Then, ischemia is induced by inflating a blood pressure cuff on the forearm for 5 min, and the diameter of the vessel is measured at the end of the diastolic period within 30 to 90 s after deflation of the cuff. Electrocardiogram monitoring during the echocardiography and FMD studies ensures proper timing of the images regarding cardiac cycle.

Body composition measurements

Lean body mass is evaluated by BIA, as well as DXA method. Those with a cardiac pacemaker or an implantable cardioverter defibrillator are not exposed to BIA. Patients are advised to be fast at least 8 h prior to BIA and not to have excessive physical activity. A single-frequency tetra polar BI analysers (Omron BF511, Omron, Japan) is utilized to determine weight, total body muscle, fat, and visceral fat mass. Height is measured by a stationary stadiometer and used for anthropometric measurements.

Body composition is also measured by DXA (Norland XR-36, Norland Corp., Fort Atkinson, Wisconsin, USA) via whole-body scan with a resolution of 6.5 × 13.0 mm and a scan speed of 260 mm/s in Seyed-al-Shohada Hospital, affiliated to Isfahan University of Medical Sciences. A trained densitometry technologist conducts the tests according to the Norland user's instructions. Patients who have a metallic implant are not subjected to this test.

Muscle function tests

Grip strength is measured using a hydraulic hand dynamometer (Jamar, Jackson, MI). Three consecutive measures of handgrip strength (kg) at both hands are recorded with the patient sitting in an upward position and the shoulder in adduction, elbow flexed to 90°, and forearm and wrist in neutral position.

Individuals participate in 6 min walk test in agreement with the guideline from the American Thoracic Society.[21] The responsible investigator explains the test to the patient, prior to the beginning of the test. Patients are instructed to walk back and forth as far as they can on an indoor track for 6 min, and the investigator observes them carefully during the test and records the tracks. Safety measures for resuscitation are taken, and a physician is present at the site of the test.

Blood tests

A 10 mL blood sample is taken from each participant after 12 h of fasting for the routine biochemical tests of fasting blood glucose, triglyceride, LDL and HDL cholesterol, blood urea nitrogen, creatinine (Cr), alanine aminotransferase, and aspartate aminotransferase. Malondialdehyde and total antioxidant capacity are measured by the calorimetric method and testing of NT-proBNP, hs-CRP, IL-1β, TNF-α, myostatin, insulin, and IGF-1 are carried out by the enzyme-linked immunosorbent assay on serum samples stored at −80°.

Questionnaires

Questionnaires used in MeHR trial are MLHFQ for the assessment of health-related quality of life,[22] the state part of Spielberger State–Trait Anxiety Inventory,[23] Beck Depression Inventory II,[24] Pittsburgh Sleep Quality Index to measure the quality and patterns of sleep,[25] International Physical Activity Questionnaire-short form for monitoring physical activity,[26] and 24 h dietary recall questionnaire to record description and quantity of all foods and beverages consumed in the last 24 h.[27] We shall use the Persian validated version of all the mentioned questionnaires and have a trained investigator ask questions and fill the questionnaires.

Clinical data

Clinical data including cardiac comorbidities (past history of myocardial infarction, cardiac surgery, revascularization, valvular diseases, and congenital abnormalities), past history of diseases (history of diabetes mellitus, chronic kidney disease, chronic obstructive pulmonary disease, and current/former smoker), and medications are extracted from medical records of the patients.

Possible adverse effects of melatonin are monitored by calling the patients at regular intervals or visiting them as needed and asking open questions, as well as by a designed questionnaire at the scheduled follow-up visits. Mortality data are gathered by the verbal autopsy, and hospitalization is validated by the hospital documents of the patients, retrieved from the registry systems of the hospitals. An expert committee consisting of three cardiologists, blinded to the study groups, will assess the reasons for hospitalizations and mortalities.

Data management and monitoring

A data management committee including a cardiologist, a physical medicine and rehabilitation specialist, and a psychiatrist monitor mortality, morbidity, and possible adverse effects of the drug on a bimonthly basis. The study will be halted if the mortality and morbidity or the adverse effects attributed to the drug would be significantly higher in one of the study groups.

Sample size

The sample size calculation is based on the expectation to find a clinically relevant difference for LVEF (5%) between the two groups, according to the study of Garakyaraghi *et al.*[28] As this is a pilot study to establish effect sizes, the sample size is planned at the liberal significance level of $\alpha = 0.05$ (two-tailed) and the power of 1-β = 90%. Taking into consideration the two-group design and an expected drop-out rate of about 20%, 90 participants shall be recruited and distributed equally between groups.

Statistical methods

Statistical analysis will be carried out using Stata 14.0 software. Means and frequencies will be used to describe the sample. Simple unadjusted analyses will be performed using two-sample *t*-test for the difference in scores or χ^2 statistics to determine whether outcome and covariates differ between the two groups. A generalized linear mixed model will be adapted to model the dependent variable at any follow-up assessment and explore potential associations between the outcome and demographics and other general characteristics. For the analysis of death from any cause, we will use standard Kaplan–Meier survival methods with the log-rank test. These analyses include all randomized patients. Adverse events will be also compared between groups with the use of χ^2 tests. Sub-group analysis will be done according to the clinical characteristics of the patients and underlying aetiology of HF (ischemic or non-ischemic), if statistically possible. *P* values <0.05 will be defined as statistically significant level.

Discussion

Heart failure with reduced EF is accompanied with high morbidity, mortality, and economic burden. Despite proven treatments with positive effects on survival, many patients still complain of several symptoms of easy fatigability, low functional capacity, and low quality of life while receiving appropriate treatment. MeHR trial aims to test the effect of melatonin on cardiovascular function, skeletal muscle mass and function, and overall quality of life in this group of patients.

Advantageous effects of melatonin on both the heart muscles and the skeletal muscles have been the subject of many experimental studies in the last decade. Despite numerous *in vitro* and *in vivo* experiments, however, clinical studies of melatonin are sparse in both cardiac disease and muscle wasting conditions.

Prescription of melatonin before coronary artery bypass grafting increased EF, lowered heart rate, and decreased markers of reperfusion injury in a dose-dependent manner (10 vs. 20 mg, for 5 days before surgery),[29] and in another study, 10 mg melatonin for 1 month before coronary artery bypass grafting intensified antioxidant defence.[30] Furthermore, 12 weeks of 10 mg melatonin in diabetic patients with coronary heart disease had beneficial effects on antioxidant capacity, glycaemic control, blood pressure, and HDL cholesterol.[31]

Considering the effect of melatonin on skeletal muscle, Amstrup *et al*. showed that oral melatonin (1 or 3 mg for 12 months) reduced fat mass and increased lean mass, but had no significant effect on muscle function in postmenopausal women.[32] Another study showed that melatonin supplementation (20 mg for 8 weeks) in HF patients with cachexia has beneficial effects on appetite, fatigue, and the quality of life, and the combination of melatonin with branched chain amino acids enhances these effects.[33] In two observational studies, an inverse association was found between urine melatonin and sarcopenia in post-menopausal women[34] and also between overnight urinary melatonin and muscle strength in elderly people.[35] The effect of melatonin in HF was assessed in one study revealing that melatonin supplementation (3 mg for 2 months) improved LVEF and New York Heart Association functional class in these patients.[28]

Thus, up to our knowledge, the MeHR trial is a unique comprehensive study of both clinical outcomes and biological outcomes of melatonin supplementation in HF. The 6 month duration of the study is also a strength and is critical for long-term evaluation of possible side effects of high-dose melatonin in HF patients.

The justification for use of this dosage of melatonin would be that most of the cardiovascular experiments with melatonin used high doses of this agent, as was noted earlier, and also low levels of melatonin secretion is documented in severe HF patients. Common dose of 3 mg per day of melatonin supplementation for sleep management or circadian rhythm adjustment might have little effect on muscle mass and cardiovascular system.

It is important to consider potential side effects of such a high dose of melatonin. A recently published systematic review on adverse events attributed to oral melatonin administration revealed that the reported adverse events were generally minor and manageable and mostly related to the ingestion of melatonin during daylight hours. A major concern was the interaction of melatonin with antihypertensive drugs in patients with cardiovascular diseases, which was not consistent across different studies and was mainly explained with calcium blocker nifedipine. While calcium blockers, in particular nifedipine, are rarely used in HFrEF,[3] we are extremely cautious about the possible side effects of melatonin in our patients and monitor them closely for any manifestation of melatonin adverse effects.

The main objective of this study is to detect structural changes of myocardium as a result of possible reversing

effects of melatonin on RAAS, inflammatory system, oxidative stress, and sympathetic up-regulation. These effects are expected to be detectable in echocardiography. Moreover, serum levels of NT-proBNP increase in response to myocardial stress. This is because natriuretic peptides are up-regulated to oppose the actions of the RAAS and sympathetic nervous system.[36] Elevated levels of NT-proBNP are also strongly associated with poor prognosis of patients with HFrEF.[37] Thus, NT-proBNP levels seem an appropriate primary endpoint besides echocardiography parameters in this research.

The relatively low sample size is the limitation of this study and may cause non-significant results specifically for clinical outcomes such as mortality and hospitalization, which is tried to be compensated with the application of a composite clinical endpoint score as the secondary outcome of the study. This composite score includes changes in the quality of life as well as mortality and hospitalization throughout the study. Melatonin seems to impact different aspects of HF syndrome, so changes in the quality of life might be a reasonable secondary endpoint. Because this is a Phase II RCT, we decided to lower the sample size in favour of a detailed evaluation of cases, and if there would be a balance between side effect profile and beneficial effects, then a study with larger sample size will be recommended.

In conclusion, in concordance with the large body of experimental data on the benefits of melatonin in HF, results of this unique clinical trial might confirm the role of melatonin as an adjunctive therapy not only for the primary disease but also for the common comorbidities and complications of the HF and overall health status of the patients.

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Conflict of interest

None declared.

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