Article

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The Action and Mechanism of Trehalose on GATA4 Autophagy Degradation and Ventricular Remodeling

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Objective: To probe the effect of trehalose on myocardial hypertrophy and its specific molecular mechanism.
Methods: C57BL/6J male mice were divided into four subgroups: Sham operation subgroup (Sham), negative sham subgroup (Sham+Trehalose), transverse aortic constriction (TAC), and trehalose treatment subgroup (TAC+Trehalose). Immediately after the TAC operation, trehalose at a dose of 10 mg/kg was given daily via gavage. After four weeks, changes in cardiac function were evaluated using ultrasound to measure EF (ejection fraction), FS (fractional shortening), IVRT (isovolumic relaxation time), MPI (myocardial performance index), Tau (isovolumic relaxation time constant), LVESP (left ventricular end-systolic pressure), and EDPVR (end-diastolic pressure-volume relationship). The profiles of autophagy-associated proteins (p62, LC3II/I, and Beclin-1) and GATA4 protein in mice myocardial tissues were assessed by Western blotting (WB). Myocardial cells were classified from TAC mice into five groups: Control, Trehalose, Phenylephrine (PE), PE+Trehalose, and PE+Trehalose+autophagy inhibitor chloroquine groups. In the PE group, cardiomyocytes were treated with 50 μmol/L PE. Then, the cells were treated with trehalose (100 μmol/L), trehalose (100 μmol/L)+autophagy (20 μmol/L) for 24 hours respectively. The Control group was treated with the same amount of normal saline. Flow cytometry was utilized to detect myocardial cell apoptosis in each subgroup. The alterations in apoptosis and autophagy-correlated proteins (p62, LC3II/I, and Beclin-1) were assessed by WB. Additionally, the level of GATA4 protein upstream of autophagy was estimated. Furthermore, the expression levels of pro-apoptotic proteins Bad, BAX, Cleaved-caspase-3, and anti-apoptotic protein Bcl-2 were examined by WB.
Results: The TAC operation significantly augmented myocardial hypertrophy, heart weight-to-body weight ratio, and myocardial cell apoptosis in mice ($p < 0.05$). Trehalose significantly improved cardiac hypertrophy, cardiomyocyte apoptosis, and cardiac function decline in mice. Additionally, it also significantly enhanced autophagy in mouse cardiac tissues ($p < 0.05$). At the cellular level, trehalose significantly decreased PE-elicited apoptosis and promoted the protein expressions of Beclin-1 and LC3 II/I in cardiomyocytes while significantly dampening the profiles of p62 and GATA4 in cells. The effect of trehalose and chloroquine treatment was significantly greater than that of the trehalose group.
Conclusions: Trehalose significantly abates myocardial hypertrophy and pressure overload-induced cardiomyocyte apoptosis in mice. The cardioprotective effect of trehalose on enhanced autophagy is attributed, at least in part, to the promotion of autophagic degradation of GATA4.

Keywords: trehalose; autophagy; GATA4; pressure overload; ventricular remodeling; coarctation of the aorta

Introduction

Cardiovascular disease is a leading cause of mortality and morbidity among adults worldwide. Hypertension is a prevalent chronic disease that poses a severe threat to human health, particularly in the context of cardiovascular disease [1]. The apoptosis of many myocardial cells triggered by compensatory myocardial hypertrophy during the early stages of hypertension is the principal pathological mechanism underlying the transition from compensatory hypertrophy to decompensation [2,3]. Consequently, early intervention in the ventricular remodeling induced by cardiomyocyte apoptosis is critical to delaying heart failure.

Autophagy is a highly conserved metabolic process commonly found in eukaryotic cells. Under physiological conditions, autophagy is vital for maintaining intracellular homeostasis, by selectively degrading intracellular proteins and damaged organelles. However, excessive autophagy can result in self-digestion and worsen cellular damage [4]. Studies suggest that during cardiac hypertrophy, decreased ATP (Adenosine triphosphate) synthesis can increase autophagic activity, which can improve myocardial hypertrophy, but persistent activation can further damage mitochondria, resulting in abnormal heart structure and function [5]. GATA transcription factors are DNA-binding proteins and transcription factors with a zinc finger structure. It simu-

lates the development of many kinds of tissues by activating or suppressing transcription [6]. The GATA family consists of six members (GATA1–GATA6), with GATA4 being one of the most studied transcription factors, intimately connected to heart development and ventricular remodeling [7]. A study found slow cardiac hypertrophy in *GATA4* transgenic rats, indicating that *GATA4* is directly involved in regulating myocardial hypertrophy [8].

Trehalose is a non-reducing disaccharide, consisting of two glucose molecules, that is naturally synthesized by lower organisms, but not by mammals. It rapidly accumulates in lower organisms, such as yeast and slow-moving animals, allowing them to be under dehydration, heat shock, oxidative stress, and protein aggregation. Numerous studies demonstrated that trehalose can accelerate disease progression by activating autophagy *in vivo*. Trehalose promotes the clearance of β-amyloid and the aggregation of Huntington protein by activating autophagy, which plays a beneficial role in mouse models of neurodegenerative diseases [9,10]. Additionally, trehalose was been shown to reduce liver steatosis by promoting the clearance of intracellular lipid droplets [11]. Moreover, trehalose can activate macrophage autophagy by promoting the nuclear translocation of transcription factor EB (end-binding protein), resulting in a reduction in the burden of atherosclerotic plaque and a protective effect against atherosclerosis [12]. Similar studies found that trehalose can reduce myocardial injury and cardiac function in response to myocardial ischemia-reperfusion [13]. Favorable evidence supports the role of trehalose in improving myocardial remodeling induced by ischemia and hypoxia [14].

Based on previous relevant research and the preliminary results of this study, we propose a hypothesis that trehalose can promote the autophagic degradation of GATA4, by activating autophagy and reducing pressure overload-elicited ventricular remodeling. Furthermore, oral administration of trehalose can reverse ventricular remodeling and cardiac dysfunction in spontaneously hypertensive mice. The aim of this study was to investigate the potential relationship between trehalose inhibiting myocardial hypertrophy and enhancing autophagy in pressure-overloaded mice.

Materials and Methods

Animals

A total of 48 specific-pathogen-free (SPF) male mice, aged 8 weeks and weighing on average 25–30 g, were obtained from the Experimental Animal Center at Hebei Medical University, Shijiazhuang, China. All mice were acclimatized for one week prior to experimentation. The mice were housed in an animal facility with controlled temperature (23–25 °C), humidity (55–65%), and at a 12 h/12 h day/night cycle. The animal house was well-ventilated with no significant noise. The mice were provided standard feed and water ad libitum throughout the study.

Drug Preparation and Treatment Methods

The mice were randomly assigned to four subgroups (Sham, Sham+Trehalose, transverse aortic constriction (TAC), and TAC+Trehalose) after one week of adaptive feeding. Trehalose (Sigma, T9531, St. Louis, MO, USA) was dissolved in 0.9% saline solution to prepare a 2 mg/mL solution, which was the optimal concentration determined from pre-tests.

Establishment of a Mouse Model of Ventricular Remodeling Induced by TAC Operation and Trehalose Intervention

The TAC operation was performed to establish the model of ventricular remodeling in pressure-overload mice. After an intraperitoneal injection of 0.45% pentobarbital sodium (45 mg/kg, H31021724, Shanghai Shangyao Xinya Pharmaceutical Co., Ltd., Shanghai, China) for anesthesia, the thoracic cavity of the mice was opened. The aortic arch of the TAC subgroup was ligated with a 0.5 mm-diameter pad needle, while it of the Sham subgroup was not ligated, just opening the chest for threading. The Trehalose and TAC+Trehalose subgroups received intragastric administration of trehalose (20 $mg \cdot kg^{-1} \cdot d^{-1}$) one week before the operation and 8 weeks after the operation, while mice in the Sham and TAC subgroups were given an equal volume of normal saline. Four mice in the TAC and TAC+Trehalose subgroups died during and after the operation, but no mice in the Sham subgroup and the Sham+Trehalose subgroup died within 8 weeks. The remaining mice were included in follow-up experiments. Echocardiography was performed on the mice in each subgroup at the 8th week after the operation, followed by sample collection and estimation of the heart weight ratio.

Echocardiography

At 8 weeks after TAC, mice in each subgroup were anesthetized with isoflurane (dosage Sham: 0%~5%) inhalation and underwent cardiac ultrasound (MYLAB™ Sigma VET, St. Louis, MO, USA) examination [15]. The cardiac function indexes of the mice that were measured included: Ejection fraction (EF), fractional shortening (FS), isovolumic relaxation time (IVRT), and myocardial performance index (MPI).

Analysis of Left Ventricular Pressure-Volume Loop

The left ventricular pressure-volume loop analysis was performed according to the previously described protocol [16]. Briefly, mice were anesthetized with urethane (1g/kg, i.p.). After a catheter (RenaPulse catheter, 120RPT040, Braintree, MA, USA) was implanted into the internal jugular vein for hypertonic saline injection, a catheter (Millar 1.4F, SPR 839, Millar Instruments, Houston, TX, USA) was inserted into the left ventricle through the cardiac apex. Hypertonic saline (15% NaCl, 5 μL) was injected to achieve parallel conductance.

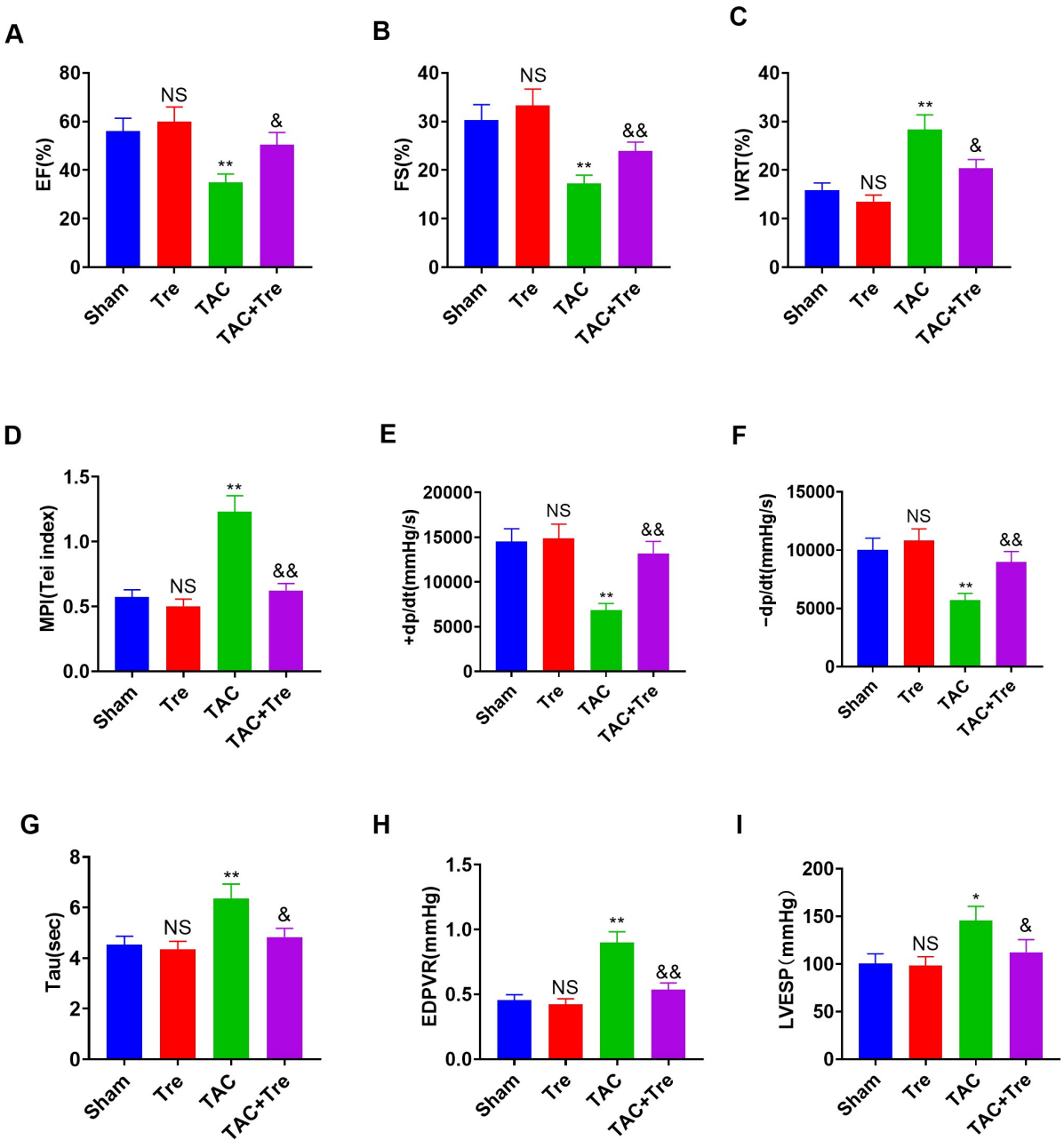

Fig. 1. Trehalose can alleviate myocardial hypertrophy in the TAC mouse model. (A–D) Transthoracic echocardiography showed EF (A), FS (B), IVRT (C), and MPI (D) of different subgroups of hearts. (E–I) Cardiac function, as assessed by direct intraventricular pressure measurement. The results were expressed as mean ± SD ($n = 6$). NS $p > 0.05$, *$p < 0.05$, **$p < 0.01$ (vs. the Sham group). &$p < 0.05$, &&$p < 0.01$ (vs. the TAC group). Tre, Trehalose; TAC, The aortic coarctation.

The parameters measured included heart rate (HR), maximum Left ventricular pressure rise time derivative (dp/dt) (+dp/dtmax), minimum dp/dt (–dp/dtmin), isovolumic relaxation time constant (Tau), end-diastolic pressure-volume relationship (EDPVR), and left ventricular end-diastolic pressure (LVEDP).

Primary Isolation and Culture of Mouse Cardiomyocytes

The hearts of healthy C57BL/6 mice with SPF grade, aged 1–3 days, were aseptically obtained. Ventricular myocytes and cardiac fibroblasts were isolated, using the differential adhesion method, after digestion with 0.125% trypsin. The isolated myocardial cells were cultured in 12-well plates pre-coated with 1% gelatin, while the isolated

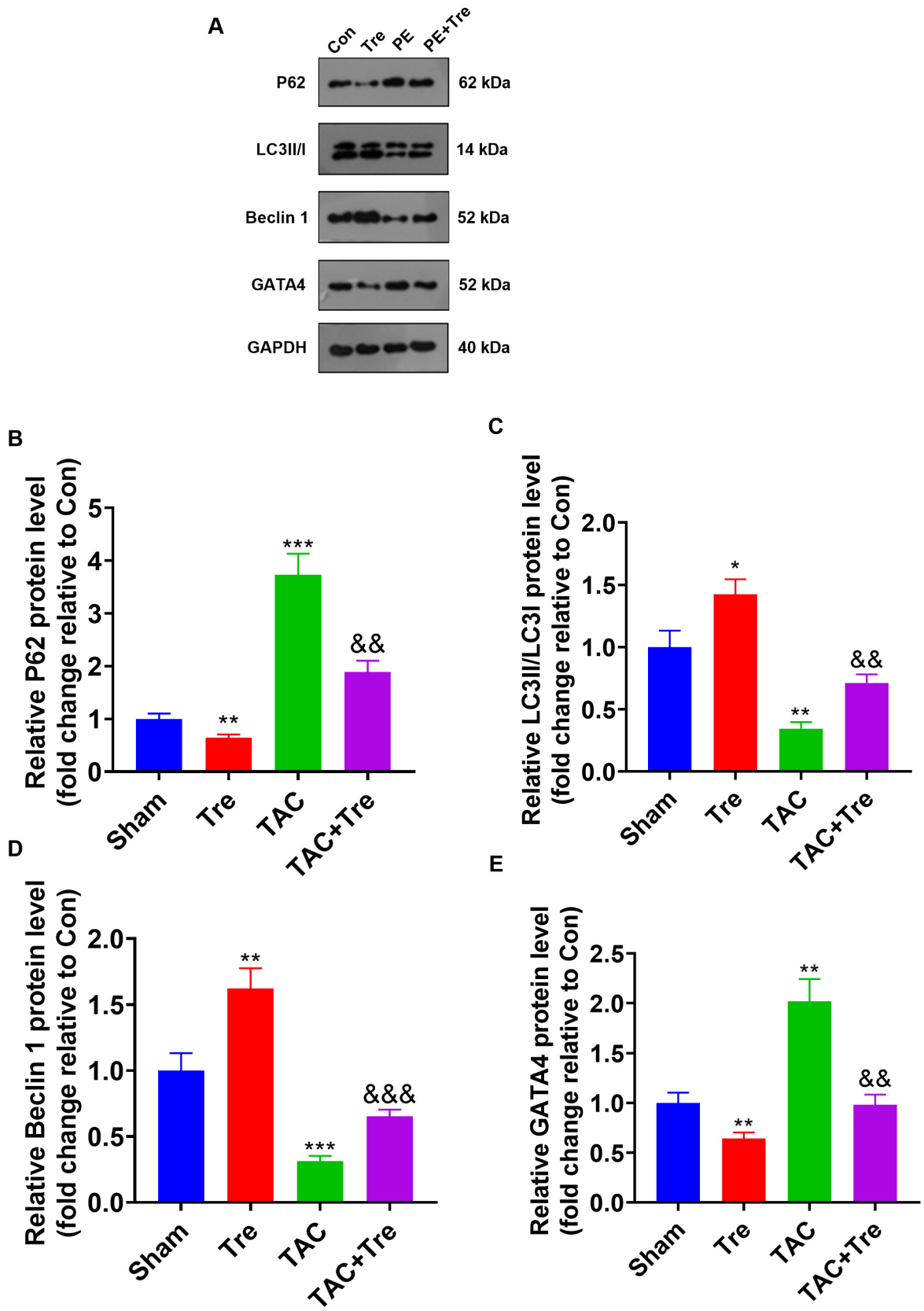

Fig. 2. Trehalose promoted myocardial autophagy and inhibits GATA4 protein expression. (A–E) The protein expression of autophagy-related proteins P62, LC3, Beclin 1, and GATA4 in cardiomyocytes, after PE action, as detected by WB, with GAPDH used as a reference. The results were expressed as mean ± SD ($n = 6$). *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$ (vs. the Sham group). &&$p < 0.01$, &&&$p < 0.001$ (vs. the TAC group). Tre, Trehalose; TAC, The aortic coarctation.

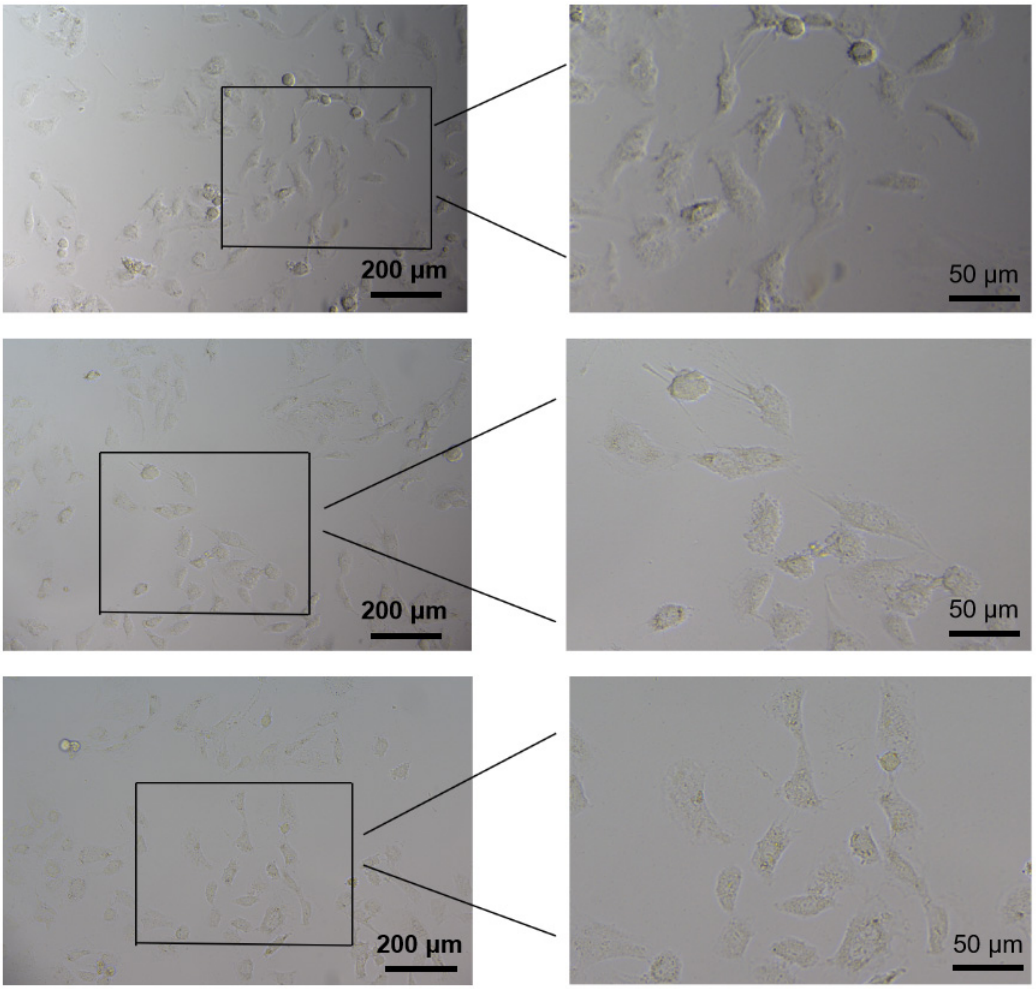

Fig. 3. Identification of primary cardiomyocytes. Observation of the morphology of primary cardiomyocytes under electron microscopy (200×).

cardiac fibroblasts were cultured in DMEM/F12 medium (11320033, Invitrogen, Carlsbad, CA, USA) supplemented with 100 μg/mL penicillin, 100 μg/mL streptomycin, and 10% serum in a 5% CO_2 incubator. After 24 hours, the culture medium was replaced with a complete medium and cultured continuously for 72 hours.

Identification of Primary Cardiomyocytes

Cardiomyocytes were successfully isolated, and after 72 hours of culture, the cell density increased to 75%. The morphology of the cardiomyocytes was observed under an inverted phase-contrast microscope and video microscope (Olympus CKX53; Olympus Sales Service Co., Ltd., Beijing, China).

Treatment of Primary Myocardial Cells

After cardiomyocytes were cultured for 48 hours, the cells were randomly divided into five subgroups, according to the random number table. The Control subgroup (Control) received the same amount of normal saline as a treatment. In the Trehalose subgroup, cardiomyocytes were treated with 100 mmol/L trehalose plus an equal volume of saline solution. In the Phenylephrine (PE) subgroup, cardiomyocytes were treated with 50 μmol/L PE (P1240000, Sigma-Aldrich, St. Louis, MO, USA). In the PE+Trehalose subgroup, cardiomyocytes were incubated with 100 μmol/L trehalose for 30 minutes, and then 50 μmol/L PE was added. In the PE+Trehalose+autophagy inhibitor chloroquine (Ch) subgroup, cardiomyocytes were incubated with 100 μmol/L trehalose and 20 μmol/L chloroquine for 30 minutes, followed by adding 50 μmol/L PE.

Flow Cytometry

The cells in each subgroup were collected via centrifugation and washed twice with pre-cooled phosphate buffered saline (PBS) at 4 °C. The cells were then resuspended in 500 μL binding buffer, and the concentration was adjusted to 10^6/mL. A 100 μL cell suspension was added to a 5 mL flow tube and stained with Annexin V-FITC (550911, BD Biosciences, San Jose, CA, USA) and propidium iodide (PI) (042k3655, Sigma, St. Louis, MO, USA). The samples were incubated at room temperature,

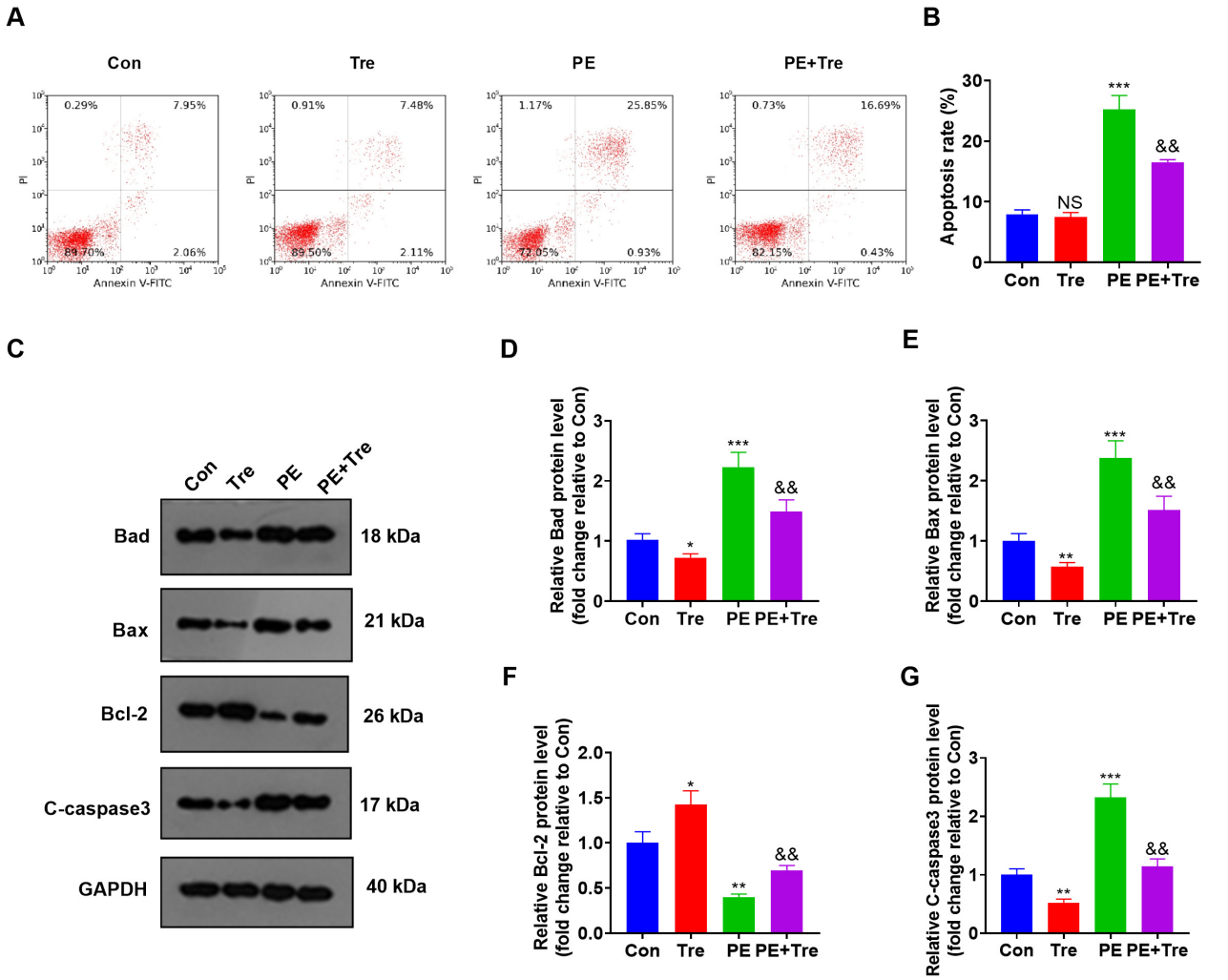

Fig. 4. Trehalose protected cardiomyocytes from PE-induced apoptosis. (A,B) The apoptosis, as estimated by flow cytometry, and statistics were made, after the myocardial cells were treated with trehalose. (C–G) The protein expression levels of apoptosis-related proteins Bad, Bax, Bcl-2, and Cleared Caspase-3, as determined by western blot, with GAPDH utilized as a reference. The results were expressed as mean ± SD ($n = 6$). NS $p > 0.05$, *$p < 0.015$, **$p < 0.01$, ***$p < 0.001$ (vs. the Con group). &&$p < 0.01$ (vs. the PE group). Con, Control; Tre, Trehalose; PE, Phenylephrine.

away from light for 15 min, and apoptosis was detected using flow cytometry (653158, Becton Dickinson, Franklin Lakes, NJ, USA). The data were analyzed using FlowJo v10 software (Tree Star, Inc., Ashland, OR, USA).

Western Blotting

Myocardial tissues were collected on ice and lysed with 500 μL radioimmunoprecipitation assay buffer (RIPA) lysate (P0013B, Beyotime, Shanghai, China) per 100 mg of tissues to extract total protein. The bicinchoninic Acid assay kit (BCA) assay kit (082820201207, Beyotime, Shanghai, China) was utilized to measure the total protein concentration. Protein was separated on a 10% sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS/PAGE) gel and transferred onto polyvinylidene fluoride (PVDF) membranes (IPVH00010, Keygen, Nanjing, China), which were blocked with 5% non-fat milk at 37 °C for 1 h. After the membranes were flushed, the following primary antibodies were added and incubated overnight at 4 °C: Anti-GATA4 (Catalog#ab84593, Abcam, Cambridge, MA, USA, 1:1000), Anti-p62 (Catalog#ab54616, Abcam, Cambridge, MA, USA, 1:1000), Anti-glyceraldehyde-3-phosphate dehydrogenase (GAPDH) (Catalog#ab181602, Abcam, Cambridge, MA, USA, 1:5000), Anti-Beclin1 (Catalog#ab207612, Abcam, Cambridge, MA, USA, 1:2000), all purchased from Abcam (Cambridge, MA, USA). The horseradish peroxidase (HRP)-labeled secondary antibody (1:2000) was then added and incubated at room temperature for 1 h. electrochemiluminescence (ECL) and X-ray film exposure were used for detection. The protein gray level was analyzed, using intraperitoneal pressure (IPP) 6.0 image analysis software (Media Cybernetics Corporation, Rockville, MD, USA), with GAPDH as the internal reference.

Statistical Method

Continuous data were presented as mean ± standard deviation (SD). Statistical analysis was carried out using Statistical package for Social Sciences (SPSS) version 17.0 (SPSS, Chicago, IL, USA). A single-factor analysis of variance (ANOVA) was utilized to compare multiple subgroups. T-Lymphocyte-Specific Defect test was utilized for pairwise comparisons. The difference was statistically significant ($p < 0.05$).

Results

Trehalose Alleviated Myocardial Hypertrophy in TAC Mice

In order to evaluate the cardioprotective effect of trehalose, trehalose was administered to TAC mice. The transthoracic echocardiography showed a decrease of 55.3% and 60.1% in EF and FS, respectively, showing notable alleviation in EF and FS after trehalose treatment. These changes indicated an improvement in cardiac systolic function. Versus the Sham subgroup, TAC significantly increased IVRT and MPI by 85.9% and 100.6%, respectively. These changes were also significantly reversed in trehalose-treated mice ($p < 0.05$, Fig. 1A–D). Although trehalose did not significantly influence left ventricular end-systolic pressure (LVESP) after TAC, it significantly reduced Tau and EDPVR, indicating an improvement in diastolic function.

Trehalose Promoted Myocardial Autophagy and Inhibited the Expression of GATA4 Protein

Compared to the Sham subgroup, the TAC group exhibited significantly increased p62 and GATA4 levels and significantly decreased Beclin 1 and LC3 II/I levels. Compared with the TAC subgroup, Trehalose treatment significantly reversed the expressions of p62 and LC3 II/I proteins ($p < 0.05$, Fig. 2).

Identification of Primary Cardiomyocytes

Under an inverted microscope, the isolated primary cardiomyocytes were found to be spindle-shaped or had many projections, and they were in a regular pulsating state (Fig. 3).

Trehalose Protected Myocardial Cells from PE-Induced Apoptosis

Compared to the control subgroup, the model subgroup showed a significant increase in the apoptosis rate of myocardial cells. Nevertheless, compared to the model subgroup, the trehalose subgroup displayed a significant decrease in the apoptosis rate of myocardial cells ($p < 0.05$, Fig. 4A,B). In addition, compared to the control subgroup, the PE subgroup showed a significant increase in both the apoptosis rate and the expressions of Bad, Bax, and Cleared Caspase-3, whereas the expression levels of Bcl-2 and p-Bad decreased when compared to the control subgroup. In contrast, the trehalose subgroup displayed a significant decrease in the apoptosis rate and the expressions of Bad, Bax, and Cleared Caspase-3, while the expressions of Bcl-2 and p-Bad significantly increased compared to the PE subgroup ($p < 0.05$, Fig. 4C–G).

Trehalose Promoted Autophagic Activity and Inhibited the Level of GATA4 Protein in Cardiac Cells Stimulated by PE

Compared with the Con subgroup, PE treatment significantly increased the expression of P62 and GATA4 in primary cardiomyocytes, and significantly inhibited the expression of LC3II/I and Beclin1. Treatment with Tre significantly reversed the expression of P62, LC3II/I, Beclin1, and GATA4. Compared with PE+Tre, the autophagy inhibitor chloroquine directly and significantly increased the protein levels of P62 and GATA4, and significantly inhibited the expression of LC3II/I and Beclin 1 ($p < 0.05$, Fig. 5A–D). Beclin-1 protein is an indicator of autophagic changes in cells, maintaining the normal structure and function of the heart and protecting myocardial cells from pressure [17]. Therefore, these results denoted that trehalose activated autophagy in mouse cardiomyocytes induced by PE.

PE significantly heightened the profile of GATA4 protein, while trehalose significantly repressed its expression. This result was consistent with *in vivo* findings and suggested that trehalose can suppress GATA4 expression during PE-induced cellular stress. Notably, the autophagy inhibitor chloroquine directly and significantly increased the protein level of GATA4 compared to the PE+Tre subgroup ($p < 0.05$, Fig. 5E).

Discussion

Autophagy is a highly conservative life process, and appropriate autophagy activity is crucial in maintaining the energy metabolism stability of cells [18,19]. However, excessive autophagy can indiscriminately degrade normal mitochondria and mitochondrial-related proteins, worsen mitochondrial damage and energy metabolism disorder, and create a vicious cycle of energy disorder leading to cell death [9]. Autophagy activity in cardiac myocytes is closely associated with cardiac hypertrophy and heart failure development. Proper autophagy can help to inhibit the progress of cardiac hypertrophy, but over-activation of autophagy can ultimately lead to heart failure [20]. Subsequent to TAC in mice, the expression levels of the autophagy-related gene *Beclin-1* and other autophagy markers significantly increased, reflecting the activation of autophagy in TAC-induced myocardial hypertrophy. Restoring the normal expression of autophagy helps inhibit myocardial hypertrophy and improve cardiac function.

Cardiac hypertrophy is a major risk factor for heart disease. Because of the terminal differentiation of cardiac

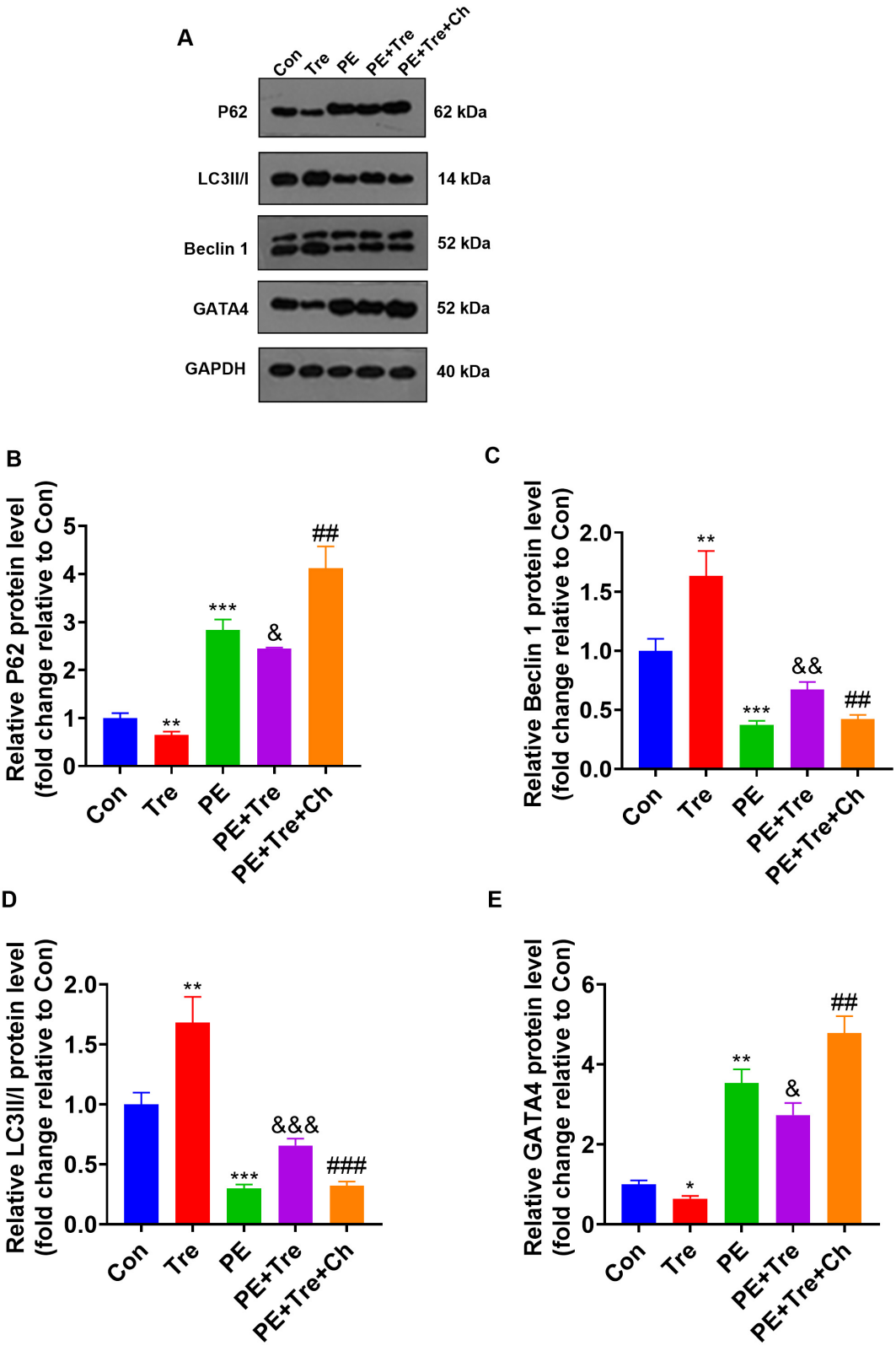

Fig. 5. Trehalose promotes the autophagic activity of cardiac cells induced by PE stimulation and inhibits the level of GATA4 protein. (A–E) Determination of apoptosis-related proteins by western blot. (B) The levels of p62. (C) Beclin 1, (D) LCII/I, and (E) GATA4 data statistics, with GAPDH utilized as a reference. The results were expressed as mean ± SD ($n = 6$). *$p < 0.015$, **$p < 0.01$, ***$p < 0.001$ (vs. the Con group). &$p < 0.05$, &&$p < 0.01$, &&&$p < 0.001$ (vs. the PE group). ##$p < 0.01$, ###$p < 0.001$ (vs. the PE+Tre group). Con, Control; Tre, Trehalose; PE, Phenylephrine; Ch, Chloroquine.

myocytes, cardiac hypertrophy occurs through the increase of the size of cardiac myocytes and the growth of non-cardiac cell components [21]. Moreover, the protein content in hypertrophic heart tissue increases, which is partly caused by the increase in protein synthesis rate. All these structural changes lead to an increase in myocardial volume and mass and impaired heart systolic and diastolic function [22,23]. Cardiac hypertrophy is an adaptive cardiac response to long-term cardiac pressure overload, and hemodynamic overload is the initial stimulus. In its early stage, it is in the "compensation" state that myocardial hypertrophy mainly manifests as the heart maintains or increases its cardiac output. In the later stage, heart failure and its related incidence rate and mortality may follow, which is in the "decompensation" state [24].

Considering the relationship between cardiac function and cardiac hypertrophy, this study unraveled that the degree of cardiac hypertrophy may pertain to cardiac hemodynamic parameters. The LVESP, EDPVR, and Tau of the heart of the model subgroup mice significantly increased, but +dp/dtmax and –dp/dtmax significantly decreased, indicating that with the existence of cardiac hypertrophy, cardiac diastolic function was seriously damaged. As Yun *et al*. [15] found in their study, it was observed that LVESP, EDPVR, and Tau were significantly increased, while +dp/dtmax and –dp/dtmax were significantly decreased in the model subgroup of mice, indicating the presence of myocardial hypertrophy and severe impairment of myocardial diastolic function. These findings are aligned with previous reports [5]. Additionally, the study also found that losartan and ramipril could partially reverse myocardial hypertrophy and improve myocardial diastolic function.

At present, there are no reports on whether trehalose can ameliorate pressure overload-induced myocardial remodeling, such as ventricular remodeling and cardiac dysfunction caused by hypertension. Although human and mouse intestines express trehalase that can reduce trehalose, a small amount of trehalose can still enter the bloodstream and various organs through the intestinal barrier to activate autophagy [8,25–27]. Therefore, oral administration of trehalose can activate autophagy to improve cardiac remodeling and dysfunction.

This study found that trehalose significantly heightened the expression of Beclin-1 protein, denoting its ability to further enhance autophagy function in myocardial hypertrophy. Beclin-1 is a key autophagy protein that participates in the autophagic process of cells, including the engulfment and degradation of harmful or aging proteins, organelles, and cellular waste, to maintain normal cell metabolism. It plays an important role in the process of cellular autophagy [28]. Therefore, research on Beclin-1 has important significance in deepening the understanding of the autophagic process and its role in tumors and other cellular diseases. In addition, Beclin-1 has also become a potential target for developing new treatment strategies. For example, by regulating the expression level or regulatory factors of Beclin-1, it is expected to develop treatments for cardiac remodeling [29].

However, it is unclear how trehalose regulates myocardial autophagy under pressure overload. The function of autophagy is modulated by a variety of intracellular signal pathways [30,31].

Through western blot detection, the study found that the expression of Beclin-1 was significantly lowered, while the p62 and GATA4 protein profiles were significantly increased in the myocardium with pressure overload, which could be significantly reversed by trehalose. These outcomes reflected that trehalose might promote GATA4 degradation and attenuate myocardial hypertrophy by activating autophagy. The current study has some limitations. Based solely on the expression of certain autophagy proteins, it is challenging to discern whether the impact of alglucan on TAC/cardiomyocytes results from a regulatory mechanism of autophagy or merely a correlative relationship. Furthermore, this study did not employ any autophagy-related agonists or inhibitors as interventions. Therefore, future in-depth mechanistic studies will focus on addressing these issues.

Conclusions

This study demonstrated that trehalose can alleviate PE-induced neonatal rat cardiomyocytes (NRCM) hypertrophy and apoptosis at the cellular level. Trehalose can inhibit apoptosis by activating autophagy to promote GATA4 degradation. Nonetheless, further research is required to determine the specific mechanism of action and explore the mechanisms of apoptosis and autophagy, which includes animal experiments and clinical investigations, to provide a theoretical foundation for identifying targets to improve ventricular remodeling and develop more effective treatment strategies.

Availability of Data and Materials

All data generated or analyzed during this study are included in this published article.

Author Contributions

YY, MM, DL, QC, LG, WZ, MZ, LC and SX made substantial contributions to the study design and manuscript writing. QC, LG, WZ, MZ, LC and SX contributed to drafting the manuscript. YY, MM and DL conducted data curation. YY, QC, LG, WZ, MZ, LC and SX performed statistical analyses. All authors have read and approved the final manuscript, and participated sufficiently in the work and agreed to be accountable for all aspects of the work.

Ethics Approval and Consent to Participate

The Animal Ethics Experimentation Committee of the Experimental Animal Center, Hebei Medical University of the Department of Cardiology, approved this study (Ethics number: 2022-l-002).

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Conflict of Interest

The authors declare no conflict of interest.

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