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Original Contribution

Evaluation of Left Ventricular Systolic Function Using Layer-Specific Strain in Rats Performing Endurance Exercise: A Pilot Study

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Objective: The functional characteristics of exercise-induced myocardial hypertrophy were studied in a rat model in conjunction with ultrasound layered strain technique to investigate the hidden changes in the heart brought about by exercise.
Methods: Forty specific pathogen free (SPF) adult Sprague–Dawley rats were selected and randomly divided into two groups of 20 exercise and 20 control rats. The longitudinal and circumferential strain parameters were measured using the ultrasonic stratified strain technique. The differences between the two groups and the predictive effect of stratified strain parameters on left ventricular systolic function were analyzed.
Results: The exercise group had significantly higher global endocardial myocardial longitudinal strain (GLSendo), global mid-myocardial global longitudinal strain (GLSmid) and global endocardial myocardial global longitudinal strain (GCSendo) values than the control group ($p < 0.05$). Even though global mid-myocardial circumferential strain (GCSmid) and global epicardial myocardial circumferential strain (GCSepi) were higher in the exercise group than in the control group, statistical significance was not reached ($p > 0.05$). Conventional echocardiography parameters were well correlated with GLSendo, GLSmid, and GCSendo ($p < 0.05$). GLSendo was the best predictor of left ventricular myocardial contractile performance in athletes determined using the receiver operating characteristic curve, with an area under the curve of 0.97, sensitivity of 95% and specificity of 90%.
Conclusion: Rats performing endurance exercise exhibited subclinical changes in the heart after prolonged high-intensity exercise. A stratified strain parameter, GLSendo, played an important role in the evaluation of LV systolic performance in exercising rats.

Introduction

Endurance sports are defined as aerobic training with the goal of prolonging athletic output over an extended distance or for a long period [1]. The variations in heart size and hemodynamics in individuals engaged in endurance sports have been described since the early 20th century. The type of remodeling observed in an athlete's heart [2,3] can vary by sport. Centripetal hypertrophy occurs mostly in power sports, while centrifugal hypertrophy is characteristic of endurance sports. Cardiac motility remodeling includes both physiological adaptations and pathological changes, which are often hidden and manifest as a decrease in left ventricular (LV) systolic function [4]. The recognition of training-induced physiological hypertrophy in the heart has resulted in a large number of experiments and clinical programs. To investigate the underlying causes of exercise-induced variations in the heart, our research team constructed an experimental model in swimming rats to explore changes in heart structure and function brought about by endurance exercise. The ultrasound layered strain technique, developed on the basis of the 2-D speckle technique, is used to clarify the longitudinal and circumferential strain states of the subendocardial, middle and epicardial myocardium in the left ventricular wall to more accurately evaluate the small changes in each layer of the myocardium in exercising rats

Abbreviations: AUC, Area under the receiver operating characteristic curve; BSA, Body surface area; CI, Confidence interval; EDV, End-diastolic volume; ESV, End-systolic volume; FS, Fractional shortening; GLSendo, Global endocardial myocardial longitudinal strain; GLSmid, Global mid-myocardial longitudinal strain; GLSepi, Global epicardial myocardial longitudinal strain; GCSendo, Global endocardial mycocardial circumferential strain; GCSmid, Global mid-myocardial circumferential strain; GCSepi, Global epicardial myocardial circumferential strain; HR, Heart rate; LVIDd, diastolic left ventricular internal diameter; LVIDdI, diastolic left ventricular internal diameter index; IVSTd, diastolic Interventricular septal thickness; IVSTdI, diastolic Interventricular septal thickness index; LVPWTd, diastolic left ventricular posterior wall thickness; LVPWTdI, diastolic left ventricular posterior wall thickness index; LVM, Left ventricular mass; LVMI, Left ventricular Mass index; EF, Ejection fraction; SV, Stroke volume; ROC, Receiver operating characteristic; RWT, Relative wall thickness

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[5]. The aims of this study were to describe the basic echocardiographic characteristics of the left ventricle in the exercising rat heart and to explore the possibility of stratified strain parameters to assess myocardial function in exercising rats.

Methods

Animals

Forty young adult Sprague–Dawley rats aged 3–4 mo were housed at the animal center for 1 wk on an *ad libitum* diet (feed was the national standard dry feed for rodents) at a room temperature of 20°C–23°C and relative humidity of 40%–60%. After adapting to the environment, the rats were randomly divided into the exercise and control groups, with 20 rats in each group.

This study was authorized by the ethics committee of Zhengzhou University First Affiliated Hospital (2021-KY-0603-001). The animal experiments were approved by the animal ethics committee of Zhengzhou University Animal Center. The ethical lot number for the present study is 2021-KY-0603-001.

Experimental model

Long-term exercise training rat models of exercise-induced cardiac hypertrophy are usually generated using three types of exercise: running, swimming and wheel running. Swimming training can be performed on several animals at the same time, making it easier to control exercise intensity. The experimental animals in exercise models usually include pigs, rabbits and rodents, such as rats and mice, among which the rat motor training model is the most common. Therefore, the present experiment was intended to replicate the myocardial hypertrophy model in rats using swimming training. The training was carried out by allowing the animal to swim on its own. Specifically, 1% of the tail weight-bearing method was used. The training was performed once a day at 9:00 a.m. for 60 min at a water temperature of 30°C–32°C. The program continued 6 d a week from Monday to Saturday with Sunday off for a total of 8 wk of training. If the rats floated and rested on the water surface during the training time, the pool was agitated or a flow-through pool was used to ensure the rats were actively swimming. The control rats were allowed to float at the same water temperature (30°C–32°C) as the experimental group for the same amount of time (60 min) for a total of 8 wk of training.

Echocardiographic analysis

To obtain images for analysis, transthoracic echocardiography was performed employing a Vivid E95 ultrasound system equipped with a M12S 225 MHz transducer (GE Vingmed Ultrasound, Horten, Norway). All rats underwent echocardiography analysis at the end of the experiment. The rats were anesthetized with 10% chloral hydrate at a dose of 0.3 mL/kg, prepared by removing skin from the thoracic and abdominal walls, placed in the left lateral recumbent position and simultaneously connected to the electrocardiogram. A 12S transducer was used for image acquisition with a depth setting of 2.5 cm and a frame rate of 225 MHz. The long- and short-axis views of the parasternal left ventricle and LV papillary muscle were clearly displayed. Fractional shortening (FS) and LV ejection fraction (LVEF) were determined using the Teicholz formula. All data were measured three times and averaged. LV mass (LVM) was calculated as follows: LVM [6] = 0.8×1.04 (diastolic LV internal diameter [LVIDd] + diastolic left ventricular posterior wall thickness [LVPWTd] + diastolic interventricular septal thickness [IVSTd])^3 − ×[LVIDd]^3) g. Body surface area (BSA) was used for correction to reduce the error caused by individual size differences in rats, such that BSA [7] = 0.09 × (weight [kg])$^{2/3}$, LVIDd index (LVIDdI) = LVIDd/BSA, diastolic LV posterior wall thickness index (LVPWTdI) = LVPWT/BSA, diastolic interventricular septal thickness index (IVSTdI) = IVSTd/BSA, LV mass index (LVMI) = LVM/BSA and relative ventricular wall thickness (RWT) = (IVST + LVPWTd)/LVIDd.

Speckle-tracking echocardiography

The 2-D images were imported into the EchoPAC software system in Q-Analysis mode to perform 2-D strain analysis. Ten consecutive and stable cardiac cycles were selected, the frame was frozen and the image was adjusted until the endocardium was clearly displayed. The image was imported into Echo PAC workstation, the myocardial range of the region of interest (ROI) to be analyzed was selected, the endomyocardial margin curve was outlined and the width of the ROI was adjusted. The system automatically analyzed each segment; "√" represents a successful segment, and "×" represents an unsuccessful segment. The system accepted the "√" segment and automatically displayed the strain map of each segment. ROIs of poor tracking were either manually moved to areas of better speckle quality or deleted. The endocardial surface was then manually outlined in the long-axis view of the left ventricle next to the sternum and in the short-axis view of the papillary muscle of the left ventricle. After approval, the system automatically obtained the strain curves of the corresponding LV section and its layers (Fig. 1). The overall longitudinal strain (endocardial myocardial global longitudinal strain [GLSendo], mid-myocardial global longitudinal strain [GLSmid] and global epicardial myocardial longitudinal strain [GLSepi]) and overall circumferential strain (global endocardial myocardial longitudinal strain [GCSendo], global mid-myocardial circumferential strain [GCSmid] and global epicardial myocardial global circumferential strain [GCSepi]) of each myocardial layer were recorded. All data were measured three times and averaged.

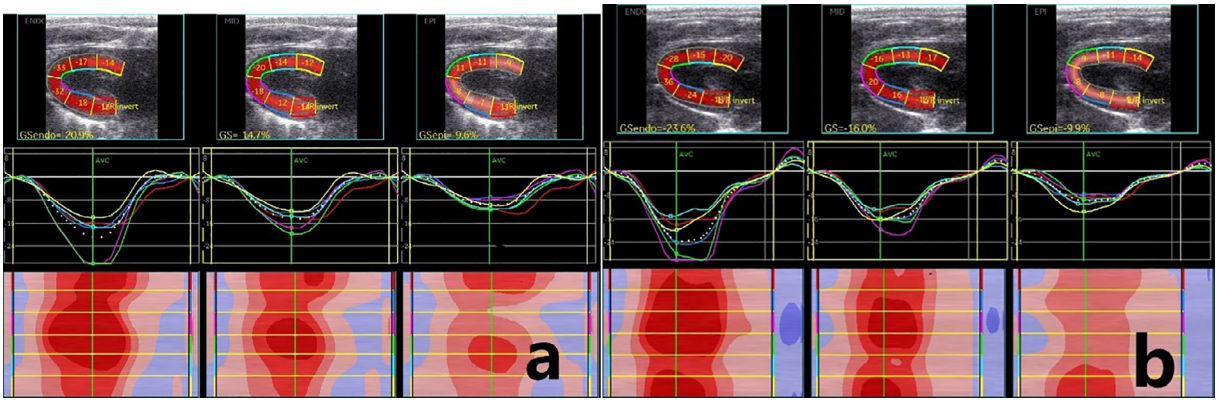

Figure 1. Longitudinal strain of all three myocardial layers. (a) Exercise group rat. (b) Control group rat. GLSendo, global endocardial myocardial longitudinal strain; GLSmid, global mid-myocardial longitudinal strain; GLSepi, global endocardial myocardial longitudinal strain.

Table 1
Basic characteristics and conventional echocardiographic parameters of the study groups

Variable	Exercise rat group (n = 20)	Control group (n = 20)	*p* Value
LVIDd (mm)	9.27 ± 0.74	5.5 ± 0.62	<0.001[a]
LVIDdI (mm/m²)	16.77 ± 1.6	14.22 ± 1.59	<0.001[a]
IVSTd (mm)	4.35 ± 0.75	1.85 ± 0.14	<0.001[a]
IVSTdI (mm)	7.89 ± 1.55	4.79 ± 0.37	<0.001[a]
LVPWTd (mm)	5.37 ± 0.45	2.76 ± 0.4	<0.001[a]
LVPWTdI (mm/m²)	9.73 ± 1.11	7.13 ± 1.08	<0.001[a]
RWT	1.0265 (0.9835–1.1649)	0.842 (0.7493–0.9096)	<0.001[a]
LVM (g)	18.99 ± 1.35	10.11 ± 0.72	<0.001[a]
LVMI (g/ m²)	0.98 ± 0.17	0.75 ± 0.18	<0.001[a]
End-diastolic volume (mL)	0.6 ± 0.13	0.41 ± 0.11	<0.001[a]
End-systolic volume (mL)	0.08 (0.06–0.11)	0.07 (0.06–0.08)	0.201
Stroke volume (mL)	0.51 ± 0.11	0.33 ± 0.1	<0.001[a]
LVEF (%)	84.26 ± 4.34	81.42±5.14	0.066
FS (%)	48.49 ± 4.97	45.15 ± 5.39	<0.001[a]
Weight	0.47 ± 0.04	0.28 ± 0.01	<0.001[a]
Body surface area (m²)	0.55 ± 0.03	0.39 ± 0.01	<0.001[a]
Heart rate (bpm)	398.5 (350.5–427)	443.5 (422–472.75)	<0.001[a]

[a] $p < 0.05$, significantly different from control group.

Statistical analysis

The analyses were performed using the statistical software SPSS 21.0 (IBM, Armonk, NY, USA). Continuous normal variables were determined using the Kolmogorov–Smirnov test, and normally distributed continuous data with equal variance between the two groups were tested using the independent sample *t*-test. Otherwise, the corrected *t'*-test was used. The Mann–Whitney *U*-test was used for comparison of the two groups of non-normally distributed variables. Pearson correlation analysis was performed between conventional echocardiography and layer-specific strain parameters. The subject receiver operating characteristic (ROC) curve was used to determine the best parameters for predicting LV systolic function in athletes and the most sensitive and specific area under the ROC curve (AUC) values.

The absolute values for layer-specific strain parameters were used for Pearson correlation and ROC analyses. Echocardiographic images of 10 rats were randomly selected, and two skilled examiners analyzed each image separately for inter-observer variability. One week later, one of the examiners analyzed each image again for intra-observer variability.

Results

General data and conventional echocardiographic measurements

Statistical data and conventional echocardiographic parameters for the exercise and control groups are outlined in Table 1. Weight and BSA were higher in the exercise group than in the control group, while heart rate (HR) was lower in the control group. The differences in the aforementioned data were statistically significant ($p < 0.05$). LVDd, LVDdI, IVSTd, IVSTdI, LVPWTd, RWT, LVM, LVM index (LVMI), end-diastolic volume (EDV), end-systolic volume (ESV), stroke volume (SV) and fractional shortening (FS) were significantly higher in the exercise group than in the control group ($p < 0.05$).

Speckle-tracking echocardiography

The comparisons of LV stratified strain parameters between the two groups are summarized in Table 2. Compared with the control group, the GLSendo, GLSmid, GLSepi and GCSendo values were decreased in the athlete group ($p < 0.05$) (Fig. 1). Although the levels of GCSmid and GCSepi were higher in the athlete group than in the control group, the differences were not statistically significant ($p > 0.05$). The ROC curve was used to determine whether the stratified strain parameters in the exercising rats could predict the changes in LV systolic function. The AUCs for predicting GLSendo, GLSmid, GLSepi and GCSendo were 0.97, 0.834, 0.75 and 0.819, respectively (Fig. 2). According to the ROC analysis results (Table 3), the optimal critical value for GLSendo was −22.84%, with 95% sensitivity and 90% specificity. The optimal critical value for GLSmid was −17.19%, with 0.95 sensitivity and 0.7 specificity. The optimal critical value for GLSepi was −11.54%, with 0.75 sensitivity and 0.7 specificity. The

Table 2
Layer-specific strain parameters

Variable	Athlete group (n = 20)	Control group (n = 20)	*p* Value
GLSendo	−20.56 ± 2.01	−24.68 ± 1.53	0.000
GLSmid	−15.22 ± 1.67	−17.61 ± 2.06	0.000
GLSepi	−10.85 ± 1.45	−12.44 ± 1.76	0.003
GCSendo	−33.77 ± 5.29	−39.85 ± 4.48	0.000
GCSmid	−18.06 ± 2.61	−16.38 ± 3.72	0.106
GCSepi	−8.76 ± 2.14	−7.88 ± 2.52	0.241

GLSendo, global endocardial myocardial longitudinal strain; GLSmid, global mid-myocardial longitudinal strain; GLSepi, global epicardial myocardial longitudinal strain; GCSendo, global endocardial mycocardial circumferential strain; GCSmid, global mid-myocardial circumferential strain; GCSepi, global epicardial myocardial circumferential strain.

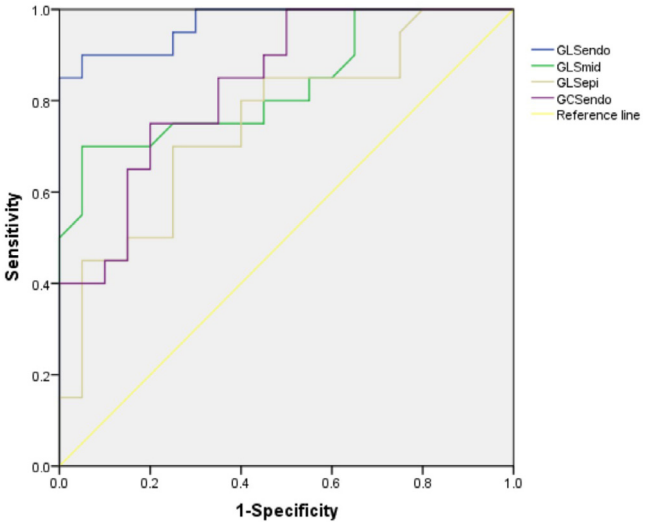

Figure 2. Receiver operating characteristic (ROC) curves were used to assess the predictive capability of left ventricular (LV) layer-specific strain parameters. This ROC was for prediction of LV dysfunction in athlete rats. GLSendo was superior to the other parameters in predicting LV performance (area under the ROC curve = 0.97; 95% CI: 0.926, $p < 0.05$), with a cutoff value of −22.84%, sensitivity of 95% and specificity of 90%.

Table 3
Receiver operating characteristic curve analysis

Variable	AUC (SE)	AUC (95% CI)	Cutoff value	Sensitivity	Specificity
GLSendo (%)	0.97 (0.85)	0.926–1	−22.845%	95%	90%
GLSmid (%)	0.834 (0.65)	0.706–0.961	−17.195%	95%	70%
GLSepi (%)	0.75 (0.45)	0.599–0.903	−11.545%	75%	70%
GCSepi (%)	0.83 (0.55)	0.717–0.958	−37.19%	80%	75%

AUC, area under the receiver operating characteristic curve; CI, confidence interval; GLSendo, global endocardial myocardial longitudinal strain; GLSmid, global mid-myocardial longitudinal strain; GLSepi, global epicardial myocardial longitudinal strain; GCSepi, global epicardial myocardial circumferential strain; SE, standard error.
[a] $p < 0.05$, significantly different from control group.

Table 4
Correlation analysis of conventional echocardiography and layer-specific strain parameters

Variable	GLSendo	GLSmid	GLSepi	GCSendo	GCSmid	GCSepi
LVIDd	−0.727[a]	−0.536[a]	−0.417[a]	−0.547[a]	0.241	0.153
IVSTd	−0.678[a]	−0.520[a]	−0.404[a]	−0.405[a]	0.424	0.184
LVPWTd	−0.699[a]	−0.513[a]	−0.405[a]	−0.579[a]	0.243	0.177
Ejection fraction	−0.11[a]	0.154	0.184	−0.38[a]	−0.184	−0.156

Values are correlation coefficients (r).
GLSendo, global endocardial longitudinal strain; GLSmid, global mid-myocardial longitudinal strain; GLSepi, global epicardial myocardial longitudinal strain; GCSendo, global endocardial myocardial circumferential strain; GCSmid, global mid-myocardial circumferential strain; GCSepi, global epicardial myocardial circumferential strain; LVIDd, diastolic left ventricular internal diameter; IVSTd, diastolic interventricular septal thickness; LVPWTd, diastolic left ventricular posterior wall thickness.
[a] $p < 0.05$, statistically significant correlation.

best critical value for GCSendo was −22.32%, with a sensitivity of 0.65 and specificity of 0.9. Therefore, GLSendo was superior to parameters such as GLSmid in detecting LV systolic function in endurance exercise rats. In correlation analysis (Table 4), LVIDd, IVSTd and LVPWTd had good correlations with GLSendo, GLSmid, GLSepi and GCSendo, respectively, all with statistical significance ($p < 0.05$).

Repeatability and reproducibility

The intra-observer intra-class correlation coefficients (ICCs) were 0.85, 0.84, 0.85, 0.87, 0.87 and 0.83, and the inter-observer ICCs were 0.84, 0.86, 0.81, 0.93, 0.84 and 0.88 for GLSendo, GLSmid, GLSepi, GCSendo, GCSmid and GCSepi, respectively, indicating good intra- and inter-observer agreement in LV layer-specific strain parameters.

Discussion

Different types of exercise lead to different changes in the heart, and exercise training can lead to adaptive changes or pathological remodeling of the heart in terms of morphology, structure and function [8,9]. Depending on the endurance (power) and strength (static) components of the exercise process, sports can be divided into different types, such as endurance and strength. During the course of endurance exercise, central output and maximal oxygen consumption increase, while peripheral vascular dilatation and increased volume load manifest as centrifugal hypertrophy. During strength exercise, peripheral vascular resistance increases, and significant increases in cardiac pressure load manifest as centripetal hypertrophy [1,10]. The traditional echocardiographic FS and LVEF, which are used to assess cardiac contractility, are affected by preload and therefore cannot reliably assess the systolic function of models with altered load [11]. Our research team first applied the stratified strain technique to determine the left ventricular characteristics of athymic rats and performed early analysis of longitudinal and circumferential strain in athymic rats, which is the main strength and innovation of this study. The purpose of the ultrasound stratification technique developed using 2-D speckle-tracking echocardiography was to obtain motion parameters in different directions for each myocardial layer by tracking the speckle signal of each layer, allowing simultaneous analysis of the motion of the three myocardial layers in the LV wall and providing new technical support for more detailed assessment of myocardial function in the LV wall [12,13]. This method reflects the overall function of the heart and can be used to analyze partial myocardial function, leading to early detection of abnormal cardiac function. Meta-analysis [14] has revealed that myocardial strain imaging had a better predictive value than LVEF in the prognostic assessment of major adverse events in heart disease. The results of the present study revealed increasing LVDdI, LVPWTdI and LVMI in the exercise group, which is consistent with the results of previous rodent exercise-induced myocardial hypertrophy models [15]. This study found a negative correlation between LVIDd and GLSendo, GLSmid, GLSepi and GCSendo, because the ventricular structure of exercising rats had been changed, and the increase in left ventricular internal diameter led to uneven distribution of blood flow in the subendocardial myocardium and relatively insufficient perfusion in the left ventricle, resulting in a corresponding change in the motion pattern of each layer of the myocardium. This negative correlation is also consistent with Laplace's law [16]. In addition, it was found that IVSTd and LPWTd were negatively correlated with GLSendo, GLSmid, GLSepi and GCSendo, because with the thickening of the left ventricular wall, the myocardium consumes more and more oxygen, which causes the myocardium to be in a state of relative ischemia and the strain of the left ventricular wall is reduced [1]. The primary factor is that aerobic exercise causes contraction of tonic muscles, a decrease in arterial resistance and a rise in venous return resistance, leading to volume overload. In inclusion, left ventricular hypertrophy reflects a mechanism to reduce ventricular wall stress compensation, and these changes may be a cardiovascular adaptation to isotonic exercise. In the trained heart, the determinants of cardiac performance have been studied, and left ventricular systolic function is largely dependent on preload, afterload, left ventricular mass and sinus bradycardia [17–19].Therefore, endurance exercise could cause LV eccentric hypertrophy, likely because aerobic exercise could cause contraction of tonic muscles, a decrease in arterial resistance and a rise in venous return resistance, resulting in LV volume overload. These changes may be a regulation of cardiac response to isotonic motion. The lowered heart rate is also considered to be an independent factor affecting LV systolic function, consistent with the results of the decreased heart rate in the exercise group in the present study ($p < 0.05$), which is an adaptation of the parasympathetic nerves to endurance exercise [20]. Previous studies in animal models have explored the three layers of the LV myocardium as a whole. However, it has been suggested that the inner layer of the LV myocardium is sensitive to volume loading [21] while the middle and outer layers are sensitive to the afterload. It is apparent that each layer of the myocardium responded differently to the same injury. Therefore, the stratification technique could be used to trace the three layers of the LV myocardium layer by layer, which could more accurately evaluate the subtle functional changes in each layer. Thus, the evaluation results have important clinical significance. Prior studies [22] have also found that the rat myocardium is divided into three layers, the same as in humans. These include the inner layer with longitudinal alignment, the middle layer with circular alignment and the outer layer with oblique alignment. GLS and GCS were comprehensively assessed in rats in the present study. The experimental results revealed that the GLS and GCS in both the exercise rats and the control group followed a decreasing trend from the subendocardial layer to the subepicardial layer, which is consistent with a prior study by our team. This may help to explain the early changes in myocardial performance [15]. Li et al. pointed to reduced GLS in athletes as an early indicator of LV systolic dysfunction [8,23]. The reason for the longitudinal and circumferential shortening can be explained. Longitudinal strain reflects downward displacement of the apical fibers to generate the normal ejection fraction of, on average, 60%. In contrast, short-axis shortening arises from predominantly circumferential fiber deformation, but only a 30% ejection fraction occurs. Nevertheless, GLS and GCS differed in rats after exercise, and GLS in the exercise group was lower than that in the control group from the endocardial layer to the epicardial layer. Only t GCS exhibited a decreasing trend in endocardial strain values in the exercise group ($p < 0.05$). GCS values in the mid-myocardial and epicardial layers were higher in the exercise group than in the control group, but the difference was not statistically significant ($p > 0.05$). Russo et al. [24] have suggested that stabilization of GCS function could sustain LVEF in the normal range in the earlier period of impaired longitudinal strain in the GLS. This may be explained by the geometrical differences in the structure of the LV trilaminar myocardium in relation to the physiological functional differences. (i) During cardiac contraction, the epicardium is relatively stationary, while the endocardial layer of the myocardium moves centripetally into the cavity. This complex motion includes shortening, thickening and twisting in multiple directions, so that the endocardial layer contributes significantly more to cardiac contraction than to the epicardial layer [15,20]. (ii) Owing

to the unique characteristics of myocardial ischemia, the subendocardial layer is the first to be affected by myocardial ischemia. In the early stages, a decrease in longitudinal motion of the left ventricle may be evident. Animal studies [22,25] have also revealed that long-term intensive training can cause structural and functional changes in coronary artery resistance. (iii) In addition, it has been determined that LV GLS may be a substitute for LV filling pressure and that LV end-diastolic pressure varies proportionally with LV GLS [26]. The volume load of the heart increases, the left ventricular end-diastolic volume increases, the stress on the left ventricular wall increases [27] and, compared with traditional metrics, stratified strain provides a more accurate and specific assessment of changes in left ventricular systolic function. (iv) The exercise rat group exhibited an increase in volume load after swimming training, which may have caused changes in the extracellular matrix or an increase in reactive fibers, which was reflected in a decrease in LV GLS after LVEF retention. Rushmer's myocardial band theory [22] also suggests that the endocardial layer contributes significantly more to the contraction of the heart than to the epicardial layer. In addition, the motion direction of the heart is mainly in the long-axis direction, which can explain why the longitudinal strain of all three myocardial layers in the rat decreased after exercise, while circumferential strain only decreased the endocardial strain. It also shows that the endocardium is more sensitive to the changes of ventricular injury. Circumferential strain is less sensitive than longitudinal strain in responding to reduced LV systolic function, but can still indicate the severity of ischemic injury to myocardial fibers. The above findings suggested that although LVEF was preserved in the early stages of myocardial damage, LV GLS and GCS were already altered. GLS was more sensitive than GCS in early diagnosis of changes in LV systolic function in exercising rats. On the basis of ROC curve analysis of the strain parameters GLS and GCS, GLSendo (AUC = 0.97) was the best predictor of LV systolic function in exercising rats, as it was more sensitive to minor changes in LV myocardium than GCS when LVEF was maintained at a normal level. The histological factors and specific conduction pathways that caused decreased myocardial strain and myocardial hypertrophy in exercising rats need to be investigated in future studies.

Limitations

There were several limitations in this study that need to be acknowledged. (i) STI-Stratified strain requires a higher frame rate and a clear view of ventricular myocardium endocardium, myocardium and epicardium, which requires higher image quality. However, some images were of poor quality because of the faster heart rate in rats. (ii) The sample size was small and the findings need to be further validated using studies with large samples. (iii) The division of the LV myocardium into three layers did not clarify whether it conformed to the actual anatomical structure. (iv) The impairment of diastolic function often preceded systolic function. However, it was not possible to accurately measure Doppler indices of diastolic function in the mitral valve and annulus because of the rapid heart rate in rats. (v) Suboptimal inter-vendor reproducibility of strain measurements has been reported with STE.

Conclusions

Endurance athlete rats undergo subclinical changes in the heart after prolonged swimming training, and the stratified strain technique is sensitive to these subtle changes. Stratified strain provides a more specific and accurate assessment of LV systolic function changes in exercising rats than traditional echocardiographic indices. The longitudinal strain parameter was more sensitive to the decrease in LV systolic function than the circumferential parameter. GLSendo (AUC = 0.97) was the best parameter for examining LV systolic function in exercising rats.

Conflict of interest

The authors declare no competing interests.

Acknowledgments

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Data availability statement

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

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