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Clinical research

Beneficial effects of chronic low-frequency stimulation of thigh muscles in patients with advanced chronic heart failure

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Aims Patients with chronic heart failure (CHF) exhibit detrimental changes in skeletal muscle that contribute to their impaired physical performance. This study investigates the possibility of counteracting these changes by chronic low-frequency electrical stimulation (CLFS) of left and right thigh muscles.
Methods and results (mean±SD) 32 CHF patients (53±10 years) with an LVEF of 22±5%, NYHA II-IV, undergoing optimized drug therapy, were randomized in a CLFS group (CLFSG) or a control group (controls). The groups differed in terms of the intensity of stimulation, which elicited strong muscle contractions only in the CLFSG, whereas the controls received current input up to the sensory threshold without muscle contractions. Functional capacity was assessed by peak VO_2, work capacity, and a 6-min-walk (6-MW). Muscle biopsies were analyzed for myosin heavy chain (MHC) isoforms, citrate synthase (CS) and glyceraldehydephosphate dehydrogenase (GAPDH) activities. Peak VO_2 (ml min^{-1} kg^{-1}) increased from 9.6±3.5 to 11.6±2.8 (P<0.001) in the CLFSG, and decreased from 10.6±2.8 to 9.4±3.2 (P<0.05) in the controls. The increase in the CLFSG was paralleled by increases in maximal workload (P<0.05) and oxygen uptake at the anaerobic threshold (P<0.01). The corresponding values of the controls were unchanged, as also the 6-MW values, the MHC isoform distribution, and both CS and GAPDH activities. In the CLFSG, the 6-MW values increased (P<0.001), CS activity was elevated (P<0.05), GAPDH activity decreased (P<0.01), and the MHC isoforms were shifted in the slow direction with increases in MHCI at the expense of MHCIId/x (P<0.01).
Conclusion Our results suggest that CLFS is a suitable treatment to counteract detrimental changes in skeletal muscle and to increase exercise capacity in patients with severe CHF.

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Introduction

In patients with chronic heart failure (CHF), exercise intolerance is related to several causes. It is now well established that changes in the periphery as a consequence of left ventricular performance itself are important determinants of reduced exercise capacity and enhanced fatigability.[1] The limited physical performance of CHF patients may not be entirely due to an impairment of cardiac and lung functions, but may also result from peripheral hemodynamic variables and/or abnormalities in skeletal muscle metabolism and structure,[2] such as decreases in capillary density, mitochondrial content, reduced enzyme activities of terminal substrate oxidation, and mitochondrial ATP synthesis.[2–7] Skeletal muscles of CHF patients also exhibit atrophy and fiber type transitions[3–5] with changes in myosin heavy chain isoforms in the slow-to-fast direction.[8,9]

According to several randomized trials, exercise can safely increase physical capacity by 15–25%, and improve symptoms and perception of quality of life in patients with stable NYHA class II and III heart failure.[10] Improvements in oxidative muscle metabolism, peak oxygen consumption and respiratory function[11] are accompanied by an increase in exercise tolerance and by a partial relief of symptoms such as dyspnea and muscle weakness.[7,12–14]

Exercise regimens for CHF are recommended as diligently supervised in-hospital training programs,[15] but mainly address stable NYHA class II and III patients. Patients with NYHA IV are generally excluded because exercise training may lead to potentially harmful systemic effects. As such, an alternative method of muscle training would be highly desirable. In this regard, neuromuscular electrical stimulation[16] may be considered as an alternative method of muscle training. Moreover, electrical stimulation seems to be less strenuous for CHF patients than exercise training. Electrical stimulation has indeed been reported to have beneficial effects in CHF patients. Long-term stimulation of thigh muscles with impulse frequencies eliciting tetanic contractions has been shown to increase muscle cross-sectional area and strength, and also to improve resistance to fatigue.[17–19]

In a study on healthy male volunteers, we have previously shown that chronic low-frequency stimulation (CLFS) of thigh muscles improves physical performance as demonstrated by an elevated VO_2 uptake at the anaerobic threshold. In addition, CLFS induced metabolic changes in the stimulated muscles towards a more aerobic-oxidative energy supply, and fast-to-slow transitions in MHC isoform expression.[20]

These findings encouraged us to investigate whether or not CLFS can be applied to more advanced CHF patients for counteracting detrimental changes in muscle properties and physical performance. In addition to measurements at the functional level, activity determinations of marker enzymes of glycolytic and aerobic-oxidative metabolism, and analyses of MHC isoform expression served to verify the effects of CLFS.

Methods

Study protocol

In a prospective, randomized, controlled trial investigating the response of thigh muscles in severe CHF patients, a 10-week low frequency stimulation protocol was compared to sham stimulation (control). Randomization was based on a sealed envelope randomization list. The study protocol, which conforms to the principles of the Declaration of Helsinki including current revisions and GCP guidelines of the European Union, had been approved by the local ethics committee of the Medical Faculty of the University of Vienna. All patients gave written informed consent.

Inclusion and exclusion criteria

Patients were eligible for inclusion in the study if they had symptomatic (NYHA class II–IV) systolic left ventricular dysfunction <35% LVEF, measured by radionuclide ventriculography within one month prior to randomization. Medical treatment with respect to neurohumoral antagonists had to be optimized at least three months prior to study entry and maintained throughout the study period. Exclusion criteria were as follows: signs of acute heart failure, unstable angina or severe arrhythmia three months prior to enrolment in the study, or other disorders counteracting exercise testing or low-frequency stimulation of the thigh muscles, such as an implanted cardiodefibrillator. Patients were conescutively recruited for the study according to the inclusion and exclusion criteria.

Stimulation protocol

The knee extensor and hamstring muscles of both legs were exposed to transcutaneous neuromuscular electrical stimulation for a total of 4 h per day, divided into 2-h morning and 2-h evening sessions, for 10 weeks (7 days per week). The same stimulation protocol was previously used in a study on healthy, male subjects.[20] In CHF patients, exercise duration is superior to intensity.[10] We, therefore, set the duration at 4 h per day, but limited contraction intensity to 25–30% of maximal voluntary contraction under baseline conditions. A portable battery-powered, programmable stimulator (STIWELL 1200, MEDEL, Innsbruck, Austria) was used. Patients could not change the individual settings of their stimulators which had been programmed by one of the investigators separately for each patient. The current was applied via self-adhering rectangular surface electrodes (130 cm^2 ValueTrode® AXELGAARD Manufacturing Co., Ltd., Lystrup, Denmark). The electrodes were placed bilaterally on the quadriceps muscles, 5 cm distal to the inguinal fold and 3 cm proximal to the upper border of the patella. Similarly, the hamstring muscles were stimulated by electrodes placed 10 cm distal to the sciatic tuberosity and 5 cm proximal to the knee joint fold. The stimulator allowed separate adjustment of intensity up to 100 mA for each of its four channels. Biphasic symmetric, constant voltage impulses with a pulse width of 0.5 msec and a frequency of 15 Hz were used. Impulse trains were delivered for 2 s and paused for 4 s. Before the onset of stimulation, maximum voluntary force of the knee extensor and hamstring muscles was measured in triplicate using the leg extension apparatus of the Cybex 6000 dynamometer (Cybex, Henley). The current intensity in the treatment group (CLFSG) was adjusted for each patient to produce 25 to 30% of the maximal voluntary contractile force. The control group (controls) was exposed to the same regimen of the CLFSG, except that the intensity of stimulation did not lead to visible or

palpable contractions as judged objectively and subjectively. To avoid differences in the patients' motivation, both groups were told that the study was performed to compare the effects of two different stimulation protocols. Patients were advised to perform stimulation while being seated on a chair and performing activities as usual, such as taking their meals, watching television, reading, etc. To exclude the risk of falling down, the patients were advised not to walk around during stimulation.

After the patients had become familiar with the stimulation procedure and were able to set the stimulator and place the electrodes properly, they continued the stimulation at home. All patients were reviewed once a week for an electrode check and to encourage their adherence to the stimulation program. The patients' compliance was ascertained by controlling the individual stimulation times stored by the stimulator software and reached almost 100%.

Exercise testing

Exercise testing was performed by symptom-limited bicycle ergometry before and after the 10-week-study period, using a ramp pattern exercise protocol individually calculated for each patient according to the formula of Wasserman.[21,22] To achieve an exercise duration of approximately 10 min, the calculated ramp was reduced to 50% of the workload applied to healthy subjects (Ergomedics 900, Sensor Medics Corp., Yorba Linda, CA). Heart rate was recorded continuously with a 12-lead electrocardiograph (Siemens Megacard, Solna, Sweden). Blood pressure was recorded non-invasively every minute during exercise and 5 min of recovery. Patients breathed through a mouthpiece connected to a mass flow meter, measuring minute ventilation by the thermal conductivity technique. Oxygen uptake (VO_2) and carbon dioxide production (VCO_2) were measured breath by breath (Sensor Medics V-MAX, Yorba Linda, CA, USA) to produce 10-s averages for each parameter. The anaerobic threshold was determined with the V-slope method.

A standardized 6-min walk test[23] was conducted before the first and the last stimulation to assess the potential of the patient's daily activity. Quality of life was monitored using the German version of the Minnesota Living with Heart Failure Questionnaire.[24] This instrument is a measure of patient self-assessment, encompassing 21 items that focus on the patient's perception of the impact of congestive heart failure on his/her physical, psychological and socio-economic life.

Tissue analyses

Muscle biopsies were taken 3–4 days before the onset and one day after the last bout of stimulation, from the middle portion of the right vastus lateralis muscle (20 cm above the patella) using a conchotome technique.[25] Two muscle samples were immediately frozen in liquid nitrogen (−196 °C) and stored at −80 °C until they were used for determination of enzyme activity and myosin analysis. The relative concentrations of the two fast myosin heavy chain isoforms (MHCIId/x and MHCIIa) and the slow MHCI isoform were determined by SDS polyacrylamide gel electrophoresis as previously described.[26] Triplicate analyses were performed on each biopsy and each gel was evaluated at least twice. For enzyme activity determination, frozen muscle powder was suspended in a 19-fold volume (mass per volume) of ice-cold 0.1 M KH_2PO_4/Na_2HPO_4 buffer (pH 7.20) containing 2 mM EDTA. The suspension was homogenized under intense cooling for 20 s with a Polytron PT3000 homogenizer (Kinematic, Lucerne, Switzerland). Homogenization was repeated 5 times with 60-s breaks for cooling. The suspension was centrifuged in a refrigerated Eppendorf 5417R centrifuge. The supernatant fraction was used for spectrophotometric measurement of citrate synthase (CS) and glyceraldehydephosphate dehydrogenase (GAPDH) activities at 30 °C, using previously described assays.[27] Enzyme activities were expressed as units per gram wet weight (U/g w wt.).

Statistics

Values are given as mean and standard deviation or as median and interquartile range in case of non-normal distribution. Group comparison was done with Wilcoxon's signed rank test within groups and Mann-Whitney U test between groups. Based on pilot data, sample size was calculated with a type-1 error of 0.05 and a power of 0.8 to detect 20% changes. $P<0.05$ was considered to indicate statistical significance (SPSS for Windows, version 8.0).

Results

Patient characteristics

Thirty-four consecutive patients with severe heart failure were recruited in the study (29 men, 5 women, mean age 53±10 years). Two patients underwent urgent heart transplantation during the study period and, therefore, had to be excluded. A total of 15 patients in the CLFS group and 17 patients in the control group completed the study. The aetiology of cardiomyopathy was ischaemic or idiopathic in 16 patients each, documented by coronary angiography prior to randomization. All patients had a markedly reduced left ventricular ejection fraction (22±5%). Medical therapy was optimized as described in Table 1. Eight patients suffered from refractory advanced heart failure, requiring additional chronic intravenous support. These patients received a chronic ambulatory infusion with vasodilatory prostaglandin E_1 as a pharmacological bridge to heart transplantation or as palliative therapy. Despite this optimized medical treatment, only 7 patients were NYHA class II while 21 patients were in NYHA class III and four in NYHA class IV. Neurohumoral activity, i.e. N-terminal atrial natriuretic peptide and B-type natriuretic peptide plasma levels, were analyzed because it describes the severity of the disease and are presently evaluated as biomarkers in assessment and management of these patients (see Table 1).[28,29] Venous blood samples were obtained after 30 min of supine rest from an indwelling catheter. Test tubes were placed on ice and centrifuged immediately. Plasma samples were stored at −70 °C until analysis. Commercially available assay kits (Biosite Diagnostics, San Diego, USA) were used to determine N-terminal atrial natriuretic peptide (N-ANP) and B-type natriuretic peptide (BNP) by ELISA.

The two groups, CLFSG and controls, were well balanced with regard to all characteristics, including New York Heart Association functional class and left ventricular ejection fraction. Also, there were no differences in medication and analysed neurohormones. (Table 1).

Table 1 Baseline characteristics of patients

	CLFSG (*n*=15)	CG (*n*=17)
m/f (*n*)	14/1	14/3
Age (years)	53±7	53±13
BMI	26.2±3.7	27.5±5.1
Diagnosis of cardiomyopathy (*n*) Non-ischemic/ischemic	7/8	9/8
NYHA II/III/IV (*n*)	5/8/2	2/13/2
LVEF (%)	22±3	21±7
Concomitant drug therapy (*n*):		
Intravenous bridging therapy PGE1 (*n*)	3	5
Intravenous bridging therapy Dobutamine (*n*)	0	1
Digitalis (*n*)	13	11
RAAS antagonists (*n*)	15	17
RAAS antagonists% maximal dose	90±21	87±22
Betablockers (*n*)	14	12
Betablockers % maximal dose	65±42	72±26
Furosemide (*n*)	15	17
Furosemide (mg/d)[a]	65 (42–118)	58 (37–09)
Spironolactone (*n*)	5	6
Spironolactone (mg/d)[a]	50 (50–50)	44 (32–53)
N-ANP (fmol/ml)[a]	4762 (2374–9913)	4983 (2458–9978)
BNP (pg/ml)[a]	221 (57–613)	258 (45–598)

No significant differences were detected for any of the comparisons shown. BMI, body mass index; LVEF, left ventricular ejection fraction assessed by radionuclide ventriculography; N-ANP, N-terminal atrial natriuretic peptide; BNP; B-type natriuretic peptide.

[a]Values expressed as mean ± SD or as median and interquartile range.

Adverse events

Both, the sham stimulation regimen and CLFS were well tolerated by the patients. Muscle soreness and/or electrode-associated skin alterations did not occur. Maximal voluntary strength of the stimulated muscle groups did not differ from baseline data.

Hemodynamic and metabolic parameters

At baseline, no significant differences existed between the two groups (Table 2). The differences in VO_2 at the anaerobic threshold reached the level of significance (P<0.01, Fig. 1) after follow-up between the groups. Peak oxygen uptake increased significantly by 21% (P<0.001) in the CLFSG and decreased by 6% in the CG (P<0.05). The increase in the CLFSG was paralleled by a significant increase in maximal workload (P<0.05) and oxygen uptake at the anaerobic threshold (P<0.01), whereas the corresponding values of the controls remained unchanged.

Quality of life

All patients completed the Minnesota Living with Heart Failure Questionnaire. The score at baseline was 63±23 in the CLFS and 63±19 in the controls. At follow-up, the CLFSG was significantly reduced (53±20, P<0.001) while controls remained unchanged (66±22, n.s.).

Six-minutes walk

The difference between the two groups was significant (P<0.05, Table 2) after follow-up. The walking distance during 6 minutes at baseline was 227±138 meters in the CLFSG and 237±132 meters in the controls. At follow-up, the CLFS increased to 299±137 (P<0.001), whereas the controls remained unaltered at 243±145 m (n.s.).

Myosin heavy chains

At follow-up, there was a significant difference between groups for MHCI and MHCIId/x (P<0.05). There was a

Table 2 Haemodynamic and metabolic variables at baseline and during follow-up

	CLFSG (*n*=15)		CG (*n*=17)	
	Baseline	Follow-up	Baseline	Follow-up
HR rest	72±13	75±17	78±20	75±17
HR peak	96±17	106±21[b]	113±29	113±27
BP systolic rest	110±17	119±18[a]	108±21	105±16[d]
BP systolic peak	128±20	142±18[a]	129±33	122±21[c]
BP diastolic rest	73±8	81±10	70±9	69±12
BP diastolic peak	75±9	81±10[a]	77±13	73±10[a]
Peak VO_2	9.6±3.5	11.6±2.8[a]	10.6±2.8	9.4±3.2[a]
AT VO_2	5.7±2.0	7.5±1.8[b]	6.4±1.9	5.8±2.8[d]
RER	1.1±0.1	1.1±0.1	1.1±0.1	1.1±0.1
6-MW	227±138	299±137[b]	237±132	243±145[c]
Watt	63±30	83±28[b]	75±32	73±31

Before–after values expressed as mean±SD.BP, blood pressure (mmHg); VO_2, oxygen uptake (ml/kg/min); AT, anaerobic threshold (ml/kg/min); RER, respiratory exchange ratio; 6-MW, 6-min walk (meters); HR, heart rate (bpm).

[a]P<0.05 baseline vs follow-up.
[b]P<0.01 baseline vs follow-up.
[c]P<0.05 between groups at follow-up.
[d]P<0.01 between groups at follow-up.

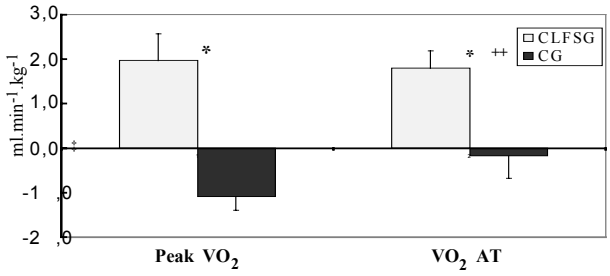

Fig. 1 Differences between baseline and follow-up in oxygen uptake at peak workload and at anaerobic threshold. Mean (SD); *P<0.01.; ++P <0.01 for difference between groups. CLFSG chronic low frequency stimulation group; CG control group.

Table 3 Percentage distribution of myosin heavy chain isoforms at baseline and during follow-up

	CLFSG (n=15)		CG (n=17)	
	Baseline	Follow-up	Baseline	Follow-up
MHC I (%)	28.3±7.8	33.8±5.8[a]	30.2±7.4	28.7±7.3[b]
MHC IIa (%)	40.4±4	40.4±5.7	37.7±5.3	38.4±5.0
MHC IId/x (%)	31.2±8.3	25.1±6.2[a]	32.1±10.2	33.2±9.8[b]

Before–after values expressed as mean (SD). CLFSG, stimulation group; CG control group.

[a] P<0.01 baseline vs follow-up.
[b] P<0.05 between groups at follow up.

significant increase in the relative concentration of MHCI (from 28.3±7.7% to 33.8±5.8%, P<0.01) and a significant decrease of MHCIId/x (from 31.2±8.3% to 25.1±6.2%, P<0.01) in the CLFSG at follow-up (Table 3). The relative concentration of MHCIIa was unchanged (from 40.4±4% to 40.4±5.7%). No differences were registered between the MHC isoform pattern of the controls before and after treatment (Fig. 2).

Enzyme activities

The activity levels of citrate synthase (CS) and glyceraldehydephosphate dehydrogenase (GAPDH), marker enzymes of the citric acid cycle and glycolysis, respectively, displayed significant alterations in the CLFSG. CS activity increased (from 3.3±0.9 to 4.3±0.8 U/g w wt, P<0.05), whereas GAPDH activity decreased (from 277±56 to 236±51 U/g w wt, P<0.05, Fig. 3). In the controls, both CS (3.4±0.7 vs 3.1±0.7) and GAPDH (277±51 vs 289±47) were not significantly changed by the sham stimulation. At follow-up, the groups differed significantly in terms of CS (P<0.01) and GAPDH (P<0.05) (Table 4).

Discussion

The aim of the present study was to test whether or not CLFS is able to safely improve exercise capacity in more advanced heart failure patients and may be used as treatment alternative to exercise. Taking the limitations of our study into account, namely its relatively small and heterogeneous study population, our results nevertheless show that CLFS leads to considerable improvements in peak exercise capacity and to a greater ability to tolerate submaximal exercise. These are clinically most important results emphasizing the feasibility of this novel treatment.

We also show that CLFS is applicable not only in stable and mild to moderate heart failure patients but also in more advanced stages of the disease. This is of special interest, because, for safety reasons, conventional exercise training is recommended only for stable NYHA class II to III patients.[10] As shown in the present study, CLFS can

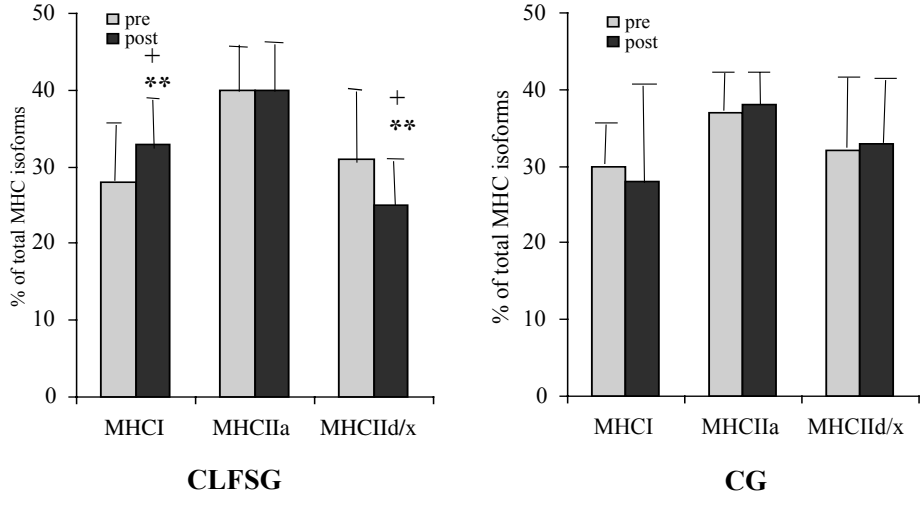

Fig. 2 Percentage of MHC isoforms at baseline and at follow-up expressed as mean (SD). +P<0,01; **P<0.01 for difference between groups. CLFSG chronic low frequency stimulation group; CG control group.

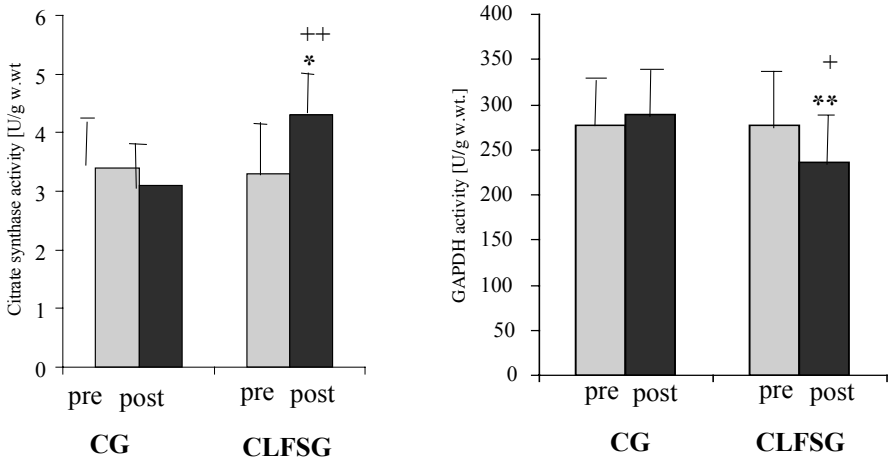

Fig. 3 Before-after values expresses as mean (SD). Enzyme activities were expressed as Units Per gram wet weight (U/g w wt.) *P<0.05; ** P<0.01 for differences baseline-follow-up; +P<0.05; ++P<0.01 for difference between groups. CLFSG chronic low frequency stimulation group; CG control group.

Table 4 Skeletal muscle enzyme activities at baseline and during follow-up

	CLFSG (n=15)		CG (n=17)	
	Baseline	Follow-up	Baseline	Follow-up
CS (U/g w wt.)	3.3±0.9	4.3±0.8[a]	3.4±0.7	3.1±0.7[a,d]
GAPDH (U/g w wt.)	277±56	236±51[b]	277±51	289±47[a,c]

[a] P<0.05 baseline vs follow-up.
[b] P<0.01 baseline vs follow-up.

be used as a suitable alternative, i.e., a home-based treatment for patients with severe CHF. Most importantly, it can be easily administered by the patients themselves and puts less stress on them as compared to conventional exercise training.

It is noteworthy that the beneficial effects were achieved in patients already treated with antagonists of the renin-angiotensin system and additional beta-blockers. Thus, the 20% increase in exercise performance (peak oxygen uptake) resulting from CLFS is over and above the anticipated improvement from standard medical therapy. Additionally, the increase in oxygen uptake in the CLFSG resembles values achieved by active endurance training in patients with less severe disease.[15,30–32] The CLFS-induced increase in peak heart rate and systolic blood pressure is suggestive of a parallel increase in aerobic capacity. As previously shown, rate pressure product at peak exercise and systolic blood pressure are significantly correlated, among other factors, with peak VO_2.[33]

Data concerning the effects of CLFS on muscle function in CHF patients are scarce. In patients with moderate CHF and in heart transplant recipients, a similar increase in peak oxygen uptake was observed under open[18] and randomized[19] conditions. In a recently published study comparing conventional bicycle exercise with functional electrical stimulation of quadriceps and gastrocnemius muscles, Harris and coworkers reported on beneficial effects of electrical stimulation in CHF patients similar to the present results.[34] Using a shorter stimulation regimen (25 Hz, 5 s on, 5 s off, 30 min daily, 5 days per week during 6 weeks), they observed improvements in the 6-min walk test, treadmill exercise time, maximum leg strength, and quadriceps fatigue index. Contrary to the present study, peak VO_2 was unaltered, probably due to the lower intensity of the stimulation regimen used in that study.[34]

The biochemical analyses of the present study were performed to assess additional effects of CLFS on metabolic and molecular properties of the target muscles. Here we show that CLFS evokes significant increases in citrate synthase activity, a mitochondrial marker, concomitant with decreases in glyceraldehydephosphate dehydrogenase activity, a reference enzyme of anaerobic glycolysis. These findings agree with results from numerous animal studies[35] and also confirm our observations on CLFS-induced changes in skeletal muscles of normal human subjects.[20] Moreover, the changes in the enzyme profile of the present study resemble previous observations on exercise-trained CHF patients,[32,33,36] suggesting an improved capacity of aerobic-oxidative energy metabolism. Nevertheless, the mechanisms that mediate increases in peak oxygen uptake may not only relate to long-term changes in mitochondrial content but probably also imply altered short-term regulation of respiratory control.[37]

The alterations in MHC isoform expression, i.e., increases in slow MHCI at the expense of fast MHCIId/x,

represent an additional finding objectifying the effect of CLFS on the target muscles. The observed shifts in the pattern of MHC isoforms resemble our previous results on CLFS-induced changes in normal human muscle.[20] Based on numerous animal studies,[38] these changes are interpreted as fiber type transitions in the fast-to-slow direction.

Taken together, the clinical, physiological and biochemical results of our pilot study point to the suitability of CLFS as a treatment for counteracting detrimental changes of skeletal muscle in advanced CHF patients and, thus, to improve their physical condition, their exercise tolerance and their quality of life.

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