

Decreased Vagal Influence on the Heart After 24-week Carnitine Supplementation

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Abstract

L-carnitine which shuttles fatty acid into the mitochondria is known diet supplement used by athletes to improve physical performance. It is also used in supplementary treatment in cardiac insufficiency. Since effect of L-carnitine on cardiovascular regulation is unclear we investigated whether its prolonged supplementation would affect autonomic nervous system (ANS) control upon the heart in healthy elderly volunteers.

Thirteen apparently healthy women aged 64 to 71 were supplemented in the double-blinded fashion with either 1.5 g of L-carnitine or placebo for 24 weeks. High resolution electrocardiogram was recorded before and after the supplementation. Time- and frequency-domain heart rate variability (HRV) analysis was performed to assess ANS control on the heart.

Following L-carnitine supplementation the overall HRV represented by TSP or SDNN insignificantly decreased with a significant reduction of parasympathetic-related HRV indexes: rMSSD, HF and SD1. Consistently, a tendency towards an increase of indexes of sympathovagal balance: SDNN/rMSSD and SD2/SD1 may reflect vagal withdrawal. Reduction of autonomic control on the heart after L-carnitine chronic supplementation may be regarded as its adverse effect related to reported increased risk of severe cardiac events.

1. Introduction

L-carnitine is a low-molecular amino acid derivative contributing in cellular energy balance. It plays a crucial role in fatty acid transport into mitochondria especially in myocardium [1] and skeletal muscle. Thus L-carnitine has been implemented in treatment of some cardiac pathologies [2] and for improvement of exercise performance. It has also been used for decades as a freely available dietary supplement [3]. Notwithstanding the large number of studies on functional effects of L-carnitine supplementation in skeletal muscle and heart, there is

virtually no data about its regulatory relevance including the autonomic heart rhythm control.

In this study, we investigated the effect of 24-week L-carnitine supplementation on autonomic control of the heart in elderly people with no reported diseases. Heart rate variability (HRV) analysis is commonly used noninvasive method to assess autonomic nervous system (ANS) activity. It is well known that HRV is reduced with age [4–6], which correlates with increased risk of dangerous cardiovascular events. We hypothesized that L-carnitine supplementation might improve the ANS heart control.

2. Materials and Methods

Apparently healthy volunteers aged 64 to 71 responded to the advertisements in the local newspaper at the University of Third Age and at the Senior Activity Center. Subjects with cancer, cardiovascular, gastrointestinal, liver or renal diseases as well as with atrial fibrillation or frequent extrasystolic beats were excluded from the study. Exclusion criteria included prolonged treatment with thyroxine, beta-blockers, ACE inhibitors or calcium channel blockers. Thirteen females were finally enrolled and accomplished study. All individuals signed their consent to participate in the study. The study was conducted in accordance with the Declaration of Helsinki, and the experimental protocol was approved by the Independent Bioethics Commission for Research at Medical University of Gdansk (NKBBN/354-304/2015).

The participants were randomly divided into two groups: placebo (N=7) or L-carnitine (N=6) in the double-blinded fashion. Subjects were receiving orally either 1500 mg of L-carnitine-L-tartrate or an isonitrogenous placebo per day for 24 weeks. Before the start and following the supplementation subjects arrived at the laboratory for tests [7] including electrocardiogram (ECG) recording. After a short initial relaxation (a few minutes) in supine position, high resolution (4kHz) ECG was recorded continuously for 10 to 20 min with the use of PowerLab 26T (AdInstruments, Sydney, Australia).

2.1. Heart Rate Variability measurements

ECG recordings were visually checked to identify artifactual fragments or extrasystolic beats. Five-minute time-series of the best quality were selected from the original recordings. RR intervals (RRi) were identified using automatic R-peak detection of ECG (LabChart 8 Pro software, AdInstruments, Sydney, Australia). Additionally, sporadic false positive R wave detections, missing fragments or outstanding RRi were also manually corrected with use of Kubios Professional 3.2 software (Kubios Professional 3.2, Kuopio, Finland). The smooth priors method with $\lambda = 500$ and interpolation rate = 4 Hz was used for smoothing data set prior to spectral analysis of HRV [8]. After correction of RRi-time-series to obtain normal-to-normal RR intervals (NN-time-series), HRV analysis was performed with use of Kubios Professional 3.2 software.

HR (heart rate), mean RRi, SDNN (the standard deviation of all normal NN intervals), rMSSD (square root of the mean of the squares of differences between adjacent NN intervals) and SDNN/rMSSD ratio were taken as

a representative time-domain parameters. The frequency-domain parameters were assessed with fast Fourier transform (FFT) algorithm with estimation of spectral density using Welch's periodogram [9]. Power spectral densities were assessed from three 150-second windows with 50% overlapping. Standard predefined spectral bands [10] were set at 0.04 ± 0.15 Hz (low frequency, LF) and 0.15 ± 0.4 Hz (high frequency, HF) and were expressed in absolute values (ms^2). Total spectral power (TSP) and LF/HF index were also calculated. Additionally SD1 and SD2 from Poincare plot as well as SD2/SD1 index were assessed as representatives of non-linear HRV parameters.

Statistical analysis was performed using GraphPad Prism 7 software (La Jolla, CA, USA). All data sets were tested for normality with Shapiro-Wilk test. Statistical analysis was based on Mann-Whitney, Wilcoxon and paired or unpaired Student t-tests, depending whether data have normal or non-normal distribution. P values <0.05 were considered statistically significant. All data are shown as mean values \pm standard deviation of the mean (\pm SD).

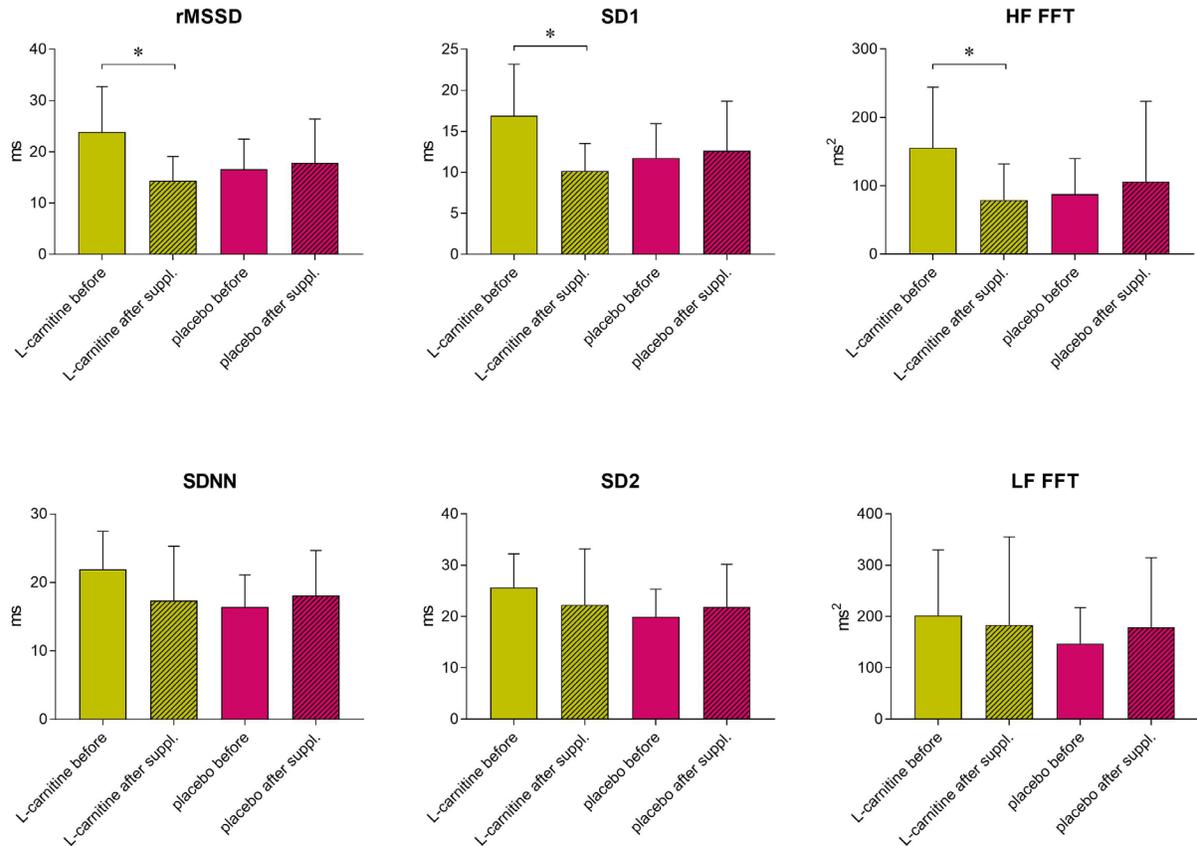


Figure 1. Selected time-domain and spectral HRV parameters before and after L-carnitine (green) or placebo (red) supplementation. * - $p < 0.05$; data are shown as mean \pm SD. Abbreviations are explained in text.

3. Results

3.1. Group characteristic

As shown in Table 1 all participants did not differ in their mean age, body weight (BW) and body mass index (BMI). Either the body weight and the BMI were insignificantly higher in placebo group.

Table 1. Basic characteristic of the participants. Data are shown as mean \pm SD. Abbreviations are explained in text.

Parameter	Placebo	L-carnitine	p
Age (years)	67 \pm 2	67 \pm 3	0.794
BW (kg)	73.9 \pm 18.5	7.2 \pm 6.6	0.534
BMI (kg/m ²)	29.8 \pm 6.8	26.8 \pm 2.5	0.534

3.2. Heart Rate Variability

In L-carnitine group HR ranged between 60 and 85 beats per minute (bpm) before and from 65 to 86 bpm after the L-carnitine supplementation (NS; $p = 0.063$). In placebo group HR remained almost unchanged ranging from 58 to 81 bpm before and from 56 to 79 bpm after 24 weeks. Total HRV represented by TSP or SDNN showed insignificant decrease after L-carnitine supplementation. TSP changed from 405.3 \pm 211.1 to 321 \pm 297 (NS, $p = 0.313$) and SDNN from 21.91 \pm 5.59 to 17.34 \pm 7.98 ms (NS; $p = 0.16$). In placebo group we did not observed alike difference.

Before the L-carnitine supplementation we did not find any significant differences in HRV parameters related to vagal drive (rMSSD, SD1, HF) between placebo and L-carnitine group (Figure 1). Following L-carnitine supplementation, but not in the placebo group, the vagal HRV indexes were suppressed: rMSSD (from 23.87 \pm 8.84 ms to 14.35 \pm 4.72 ms; $p = 0.034$), SD1 (from 16.91 \pm 6.26 ms to 10.16 \pm 3.34 ms, $p = 0.034$) and HF (from 155.6 \pm 88.53 ms² to 79.38 \pm 52.66 ms², $p = 0.049$; Figure 1). In contrast to decreased short-term HRV parameters only irrelevant tendency towards a decrease of long-term HRV indexes were observed after L-carnitine supplementation: LF 202.3 \pm 127.6 vs. 183.7 \pm 171.9 ms², (NS, $p = 0.71$) and SD2 25.68 \pm 6.52 vs. 22.29 \pm 10.91 ms, (NS, $p = 0.31$).

The tested indexes of sympathovagal balance: SDNN/rMSSD and SD2/SD1 showed an insignificant increasing tendency after L-carnitine supplementation (respectively $p = 0.078$ and $p = 0.067$). Interestingly LF/HF index increased insignificantly in both groups (Table 2).

4. Discussion

Despite common use of L-carnitine as diet supplement routinely used for building of muscle mass and muscular strength and also as fat tissue burning [3] its regulatory effects are poorly understood. L-carnitine was reported to improve systolic function of the left ventricle in patients with heart failure [11], after acute myocardial infarction [12], reduce inflammation and fibrosis in coronary heart disease [13], improve cardiac electrophysiological parameters, ameliorate peripheral neuropathy in diabetes [14], improve body weight reduction during obesity treatment [15], and many more. L-carnitine, by its contribution in the transport of fatty acids through mitochondrial membrane, was thought not only to improve skeletal muscle and cardiac function in older people but also to support ANS regulatory control.

Surprisingly our results show that 24-week L-carnitine supplementation in elderly women evoked slight decline in total HRV and significant reduction in parasympathetic parameters such as: rMSSD, HF or SD1 (Figure 1). Observed changes accompanied by a minor HR increase and shifts of sympathovagal balance indexes towards sympathetic predominance (Table 2) might suggest withdrawal of vagal activity. A decrease in the autonomic function with aging, with shifting the sympathovagal balance towards sympathetic predominance is well-known phenomenon. Our data show that L-carnitine neither did not reduce the age-related sympathetic prevalence nor improve the overall ANS drive on the heart in healthy elderly people. Observed reduction of short-term HRV which is indicative for vagal suppression may be related to reported increased risk of sudden cardiac incidents [16]. The mechanism of such dysregulation is unclear. It is well-known that nitric oxide (NO) affects ANS and can increase the vagal control of the heart [17]. On the other hand, L-carnitine was reported to suppress NO production [18] despite its oxygen radical scavenging properties [19]. Perhaps antioxidant effects of L-carnitine, which like other antioxidants might cause increase in HRV [20], was attenuated by a suppression of NO.

Table 2. RR interval and all tested sympathovagal balance indexes of HRV before and after supplementation of L-carnitine or placebo. P close to significance is bolded. Abbreviations are explained in text.

Parameter	Placebo before (1)	Placebo after suppl. (2)	L-carnitine before (3)	L-carnitine after suppl.(4)	p 1 vs 2	p 3 vs 4	p 1 vs 3	p 2 vs 4
RRi (ms)	905 \pm 102	935 \pm 117	901 \pm 108	863 \pm 89.9	0.313	0.057	0.954	0.250
SDNN/rMSSD	1.01 \pm 0.12	1.06 \pm 0.26	0.99 \pm 0.26	1.21 \pm 0.21	0.548	0.078	0.838	0.628
SD2/SD1	1.76 \pm 0.28	1.84 \pm 0.61	1.69 \pm 0.59	2.14 \pm 0.34	0.770	0.067	0.784	0.311
LF/HF	1.86 \pm 0.68	2.53 \pm 2.01	1.85 \pm 1.63	2.46 \pm 1.06	0.447	0.400	0.987	0.944

Regrettably *in vivo* activity of NO was not assessed in this study. As older population exhibits low parasympathetic activity, any further reduction of vagal drive may likely result in significant suppressive effect on short-term HRV parameters.

5. Summary

L-carnitine supplementation in healthy elder people did not improve ANS control of the heart. Moreover, it caused a decrease in vagal nerve activity which reportedly is an unfavorable effect.

Acknowledgments

The study was supported by National Science Center 2014/15/B/NZ7/00893 and by Ministry of Science and Higher Education for young scientists 01-0336/08/261.

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