Curcumin Prevents Cardiac Remodeling in L-NAME-Induced Hypertensive Rats

Parichat Prachaney1,2,*, Wilaiwan Mothong1, Rarinthorn Samrid1,3, Poungrat Pakdeechote1,3, Pattanapong Boonprom1,3, Benyapha Sripanya4, Thewarid Berkban2

1Department of Anatomy, 2Physiology, Faculty of Medicine, 3Cardiovascular Research Group, Khon Kaen University, Khon Kaen, 40002, Thailand.
2Faculty of Public Health, Kasetsart University, Chalermprakiat Sakon Nakhon Province Campus, Sakon Nakhon, 47000, Thailand.
3Faculty of Medicine, Mahasarakham University, Maha Sarakham, 44000, Thailand.

Background and Objectives: Curcumin (CUR), a yellow-orange dye obtained from turmeric, has anti-hypertensive, antioxidant, and anti-inflammatory effects. The present study investigated the effect of curcumin on cardiac remodeling in L-NAME treated rats.

Methods: Male Sprague-Dawley rats weighing 220-240 g were treated with L-NAME (50 mg/kg/day) in drinking water and orally treated with CUR (100 mg/kg per day) for five weeks (n=8/group). Cardiac remodeling indicators, including wall thickness, cross sectional area (CSA), luminal area and fibrosis of the left ventricle (LV) were measured.

Results: L-NAME-treated rats showed increases in wall thickness, CSA, fibrosis of the LV and a reduction of ventricular luminal area. CUR significantly prevented the development of cardiac remodeling induced by L-NAME in rats (p<0.05).

Conclusion: These data suggested that CUR had cardioprotective effects in L-NAME hypertensive rats.

Keywords: curcumin, cardiac remodeling, L-NAME
Introduction

N-nitro l-arginine methyl ester (L-NAME), L-arginine analog, is a nitric oxide synthase inhibitor. It is widely used to induce hypertension in animals to mimic pathophysiology of human hypertension. It is well established that rats treated with L-NAME had high blood pressure associated with cardiac remodeling including, thickening of ventricular wall, increased cross sectional area, decreased ventricular luminal area, collagen deposition. Recently, oxidative stress that is contributed to L-NAME induced cardiac alterations has been revealed.

Curcumin (CUR) is a component of turmeric and responsible for its yellow color. It has been used for centuries in traditional medicines. The biological effects of CUR including anti-inflammatory, anti-oxidant, antihypertensive and anti-cancer effects have been reported. Although a wide range of potentially therapeutic effects of CUR have been reported, the effect of CUR on cardiac remodeling especially in hypertensive condition induced by L-NAME has not been clarified. Therefore, the aim of the present study was to investigate whether CUR could inhibit the cardiac remodeling process in this hypertensive rat model.

Methods

Animal

Male Sprague-Dawley rats weighing 220-240 g were obtained from Animal Care Unit, Faculty of Medicine, Khon Kaen University, Thailand. All study animals were housed in stainless steel cages and maintained in an air-conditioned room (25.1±1 °C) with 12:12 h light/dark cycle. They were fed with a standard chow diet (Chareon Pokapan Co. Ltd., Thailand) at Animal Care Unit, Faculty of Medicine, Khon Kaen University. This study was approved by the Institutional Animal Ethics Committee of Khon Kaen University, Khon Kaen, Thailand (AEKKU 30/2552).

Experimental designs

After a week of an acclimatization period, rats were randomly assigned to 4 groups (n=8/group) as following.

1. Control group; rats received drinking water and were orally administrated propylene glycol (PG, 1.5 ml/kg) as a vehicle.
2. Control + CUR group; rats received drinking water and were orally administrated CUR at a dose of 100 mg/kg.
3. L-NAME group; rats treated with L-NAME (50 mg/kg/day) in their drinking water were orally administrated PG 1.5 ml/kg as a vehicle.
4. L-NAME + CUR group; rats treated with L-NAME (50 mg/kg/day) in their drinking water were orally administrated CUR at dose of 100 mg/kg.

Morphometric analysis of heart tissue.

The left ventricles were removed and fixed for 24 h in 4% paraformaldehyde then processed routinely in paraffin and serial 5-µm thick sections were stained with hematoxylin and eosin (Bio-Optica Milano SpA., Milano, Italy) and the sections image were obtained DS-2Mv light microscope, a stereoscope (Nikon SMZ745T with NIS-elements D 3.2, Tokyo, Japan) for morphometric evaluation. The sections which stained by picro-sirius red (Polysciences, Warrington, PA, USA) were captured by Eclipse LV100 POL polarized light microscope (Nikon, Tokyo, Japan) for cardiac fibrosis evaluation. Morphometric and fibrosis evaluations were analyzed with the ImageJ morphometric software (National Institutes of Health, Bethesda, MD, USA). All data were expressed as mean±SEM. Comparison between groups were performed with one-way analysis of variance (ANOVA) and followed by Bonferroni post hoc test. A value of p <0.05 was considered to indicate statistically significant differences.

Results

Effect of CUR on wall thickness, cross sectional area and luminal area

L-NAME administration produced the development of ventricular structural change as indicated by a significant increase in wall thickness of left ventricle (3.28±0.05 mm) when compared to the control group (2.61±0.05 mm) (p<0.05). Interestingly, treatment with CUR showed a significant decrease in left ventricular wall thickness (2.64±0.10 mm) when compared to the L-NAME-induced hypertensive rats (p<0.05) (Figure 1A). The cross sectional area (CSA) in L-NAME-induced hypertensive rats showed a significant increase (65.22±1.59 mm²) when compared to the L-NAME-induced hypertensive rats (p<0.05) (Figure 1A). The cross sectional area (CSA) in L-NAME-induced hypertensive rats showed a significant increase (65.22±1.59 mm²) when compared to the control rats (2.64±0.10 mm) (p<0.05). Furthermore, L-NAME administration and treated with CUR showed to be effectively reduce in CSA of left ventricle (53.07±2.30 mm²) when compared to the hypertensive rats (p<0.05) (Figure 1B). The values of
ventricular luminal area were not significantly different among experimental groups. However, the ventricular luminal area of L-NAME-induced hypertensive rats was lower than the normotensive control rats. In contrast, administration of L-NAME and treated with CUR trended to increase the ventricular luminal area when compared to L-NAME group (Figure 1C).

Effect of CUR on the cardiac fibrosis

Chronic administration of L-NAME produced the development of myocardial fibrosis in left ventricle as indicated by a significant enhance interstitial fibrosis (5.90±0.73 %) when compared to the normotensive control rats (1.46±0.23 %) (p<0.05). Interestingly, treatment with CUR showed to be an effective for decrease in myocardial fibrosis of left ventricle (1.93±0.30 %) when compared to the L-NAME-induced hypertensive rats (p<0.05) (Figure 2).

Figure 1 Effect of 5-week L-NAME and CUR on left ventricular wall thickness (A), cross sectional area (CSA) (B), and ventricular luminal area (C). The image on the top of the panel is the representative image of the left ventricular cross sections from each group.
Discussion

The findings of this study show that rats treated with L-NAME for 5 weeks developed cardiac remodeling. Our results were consistent with several previous studies that nitric oxide deficiency caused cardiac alterations including, cardiac morphology and functions. Cardiac remodeling observed in L-NAME in this study was supported by thickening of ventricular wall, increased CSA, decreased ventricular luminal area, collagen deposition. The underlying mechanisms involved in cardiac remodeling in L-NAME treated rats is still doubtful, however, two possible mechanisms related to hemodynamics and non-hemodynamic aspects have been described.

Firstly, hemodynamic overload or high pressure load in hypertension can stimulate left ventricular remodeling since it is the adaptive response to conserve cardiac output. Secondly, NO deficiency is one of several non-hemodynamic factors contributed in cardiac remodeling.

This study found that CUR prevented the development of cardiac remodeling in L-NAME rats. The possible mechanism might involve the biological effects of CUR since CUR and its derivatives can reduce blood pressure or pressure load on heart work in L-NAME hypertensive rats. Furthermore, CUR has antioxidation that can increase NO bioavailability in this animal model.

![Figure 2](image-url)

**Figure 2** Effect of 5-week L-NAME and CUR on myocardial fibrosis in interstitial regions of the left ventricle. The image histological image is the representative area of the fibrosis from each group. The upper row images are the section stained by picro-sirius red whereas the lower row images are the corresponding area captured under polarized light microscopy (10x). Results are expressed as mean ± SEM. *p<0.05 vs. control group, #p<0.05 vs. L-NAME group.
**Conclusion**

The results of this study indicated that CUR has cardioprotective effect in L-NAME hypertensive rats. These findings provide the additional beneficial effect of CUR on heart in animal model of hypertension.

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**References**


