The effect of oral N-acetylcysteine on galectin-3 and global longitudinal strain in patients with acute myocardial infarction

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Abstract

***Objective*: Galectin-3 (Gal-3) plays a big role in the development of cardiac fibrosis; however, its role in remodeling after acute myocardial infarction (AMI) has not received sufficient attention. Post-AMI measurements of global longitudinal strain (GLS) are beneficial in providing information about infarct area and remodeling. We aimed to determine the effect of N-acetylcysteine (NAC) on Gal-3 and GLS in AMI.**

***Design*: This was a randomized, single-blind study with pre- and post-treatment evaluations performed from May 1 to August 31, 2018.**

***Setting*: Dr. Moewardi Hospital, Indonesia**

***Patients*: ST elevation myocardial infarction (STEMI) patients who received fibrinolytic therapy were randomly allocated to NAC and control groups.**

***Interventions*: A total of 32 STEMI patients were administered fibrinolytic therapy (17 patients were administered standard therapy plus 600 mg NAC orally three times a day for 72 hours and 15 patients were administered standard therapy plus placebo as the control). Gal-3 samples were taken during admission and at 72 hours in both groups, while GLS measurement was only performed 72 hours after admission.**

***Measurements and results*: Gal-3 levels in the NAC and control groups at admission were not significantly different; however, levels were significantly different after 72 hours (p=0.017). After comparing Gal-3 levels during admission and at 72 hours, the NAC group showed significant differences between Gal-3 levels at the time of admission and at 72 hours (p=0.0001); no difference was found in the control group. There were also significant intergroup differences in Gal-3 level changes (p=0.014). In the NAC group, a better and significantly different 72-h GLS value was obtained from that in the control group (p=0.023).**

***Conclusion*: Supplementary therapy with NAC can reduce Gal-3 levels and GLS in AMI patients receiving fibrinolytic therapy.**

Key words: Acute myocardial infarction, N-acetylcysteine, galectin-3, global longitudinal strain.

Background

Coronary heart disease (CHD) is still the leading cause of death. (1) Acute myocardial infarction (AMI) was previously seen more frequently in developed countries; however, the incidence of AMI is currently increasing in developing countries as well. (2) In Indonesia, deaths from cardiovascular disease are increasing every year and are becoming the leading cause of mortality. (3) Inflammation in CHD and AMI are caused by the formation of atheroma plaques in the coronary arteries.

Current evidence suggests that galectin plays an important role in acute and chronic inflammatory responses, as well as other diverse pathological processes. (4) Galectin-3 (Gal-3) is secreted by macrophages, which are activated and modulated by several physiological and pathological processes, such as inflammation and fibrosis. Gal-3 is a lectin that binds β-galactoside and is expressed by almost all types of immune and inflammatory cells, both constitutively and inductively. (5) It has a major role in the development of heart fibrosis under conditions of excessive pressure (pressure overload), neuro-endocrine activation, and hyperten-

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sion; however, its role in remodeling after AMI has not received enough attention. (6) It has been proven that Gal-3 induces cardiac fibroblasts to proliferate by producing type I collagen in the myocardium. (7)

Conventional echocardiography provides a quick overview of the general condition of the myocardium; however, the measurement of a small left ventricular ejection fraction (LVEF) detects initial and minimal pathological changes only. (8) AMI can cause regional wall motion abnormalities, and the specific myocardial region affected is associated with the distribution of the coronary arteries. The strain seen on echocardiography is used to describe shortening, thickening, and lengthening of the local myocardium as a measure of regional left ventricular function. Global longitudinal strain (GLS) decreases in patients with AMI and is associated with the infarct area and LVEF; it also predicts left ventricular changes, possible clinical events, and the heart's response to reperfusion strategies. (9) GLS, a measure of global left ventricular function, is calculated as the average of all segments. (10)

N-acetylcysteine (NAC) is a compound containing thiol with antioxidant, (11) anti-inflammatory, (12) and antiremodeling (13) effects. In patients with ST segment elevation myocardial infarction (STEMI) treated with fibrinolytic, NAC administration reduced the levels of matrix metalloproteinases (MMP) 9 and 2, which contribute to post-infarct cardiac remodeling and increase the incidence of major adverse cardiac events (MACE) in the first year of follow-up. NAC, in combination with streptokinase, can significantly reduce oxidative stress and improve ventricular function in patients with myocardial infarction. (11)

The aim of this research was to determine the effect of 600 mg NAC administered orally three times a day for three consecutive days on Gal-3 and GLS in patients with STEMI who received fibrinolytic therapy.

Methods

This study was a randomized, single-blind study with pre- and post-treatment evaluations done in the Intensive Cardiovascular Care Unit (ICVCU) of Dr. Moewardi Hospital, Indonesia, from May 1 to August 31, 2018 in STEMI patients who received fibrinolytic therapy. Patients were randomly allocated to receive NAC 600 mg three times daily for three days (treatment group) or to have no NAC (control group). Galectin-3 samples were taken during admission and at 72 hours from both groups. GLS measurement was performed 72 hours after admission. We evaluated a total of 32 patients (17 patients in the NAC group and 15 patients in the control group).

The definition and management of patients with STEMI was according to the European Society of Cardiology (ESC) Guidelines. (14) The inclusion criteria were as follows: STEMI patients with symptom onset less than 12 hours and following on fibrinolytic administration, age of 18-75 years, and no contraindications for fibrinolysis. Exclusion criteria were patients with the following history: previous AMI or chronic heart failure, valvular heart disease, chronic renal failure, hepatic cirrhosis, chronic inflammatory disease or malignancies, acute infection or sepsis, and acute stroke.

The fibrinolytic used in this study was prepared by diluting 1.5 million units of streptokinase with 100 ml of NaCl. It was infused intravenously for 30-60 minutes.

A 4 ml blood sample was taken at the time of admission and after 72 hours of NAC administration. Prodia Laboratory measured the Gal-3 levels using the ELISA method. Since GLS measurements were not possible at the time of admission, they were obtained only after 72 hours using Vivid S6 Cardiovascular Ultrasound System (GE Healthcare, Wisconsin, USA).

Data were presented in the form of mean and standard deviation and the mean difference test was used. Analysis was done using SPSS 22.0 for Windows. A $p<0.05$ was considered statistically significant.

All study participants provided informed consent, and the study design was approved by the appropriate ethical clearance from the Ethics Committee of Dr. Moewardi Hospital/Medical Faculty Sebelas Maret, Surakarta, Indonesia.

Results

The basic characteristics of the patients are shown in **Table 1**. In the NAC and control groups, no significant differences were found in the following: demographic variables (sex and age), risk factors (hypertension, smoking, and diabetes mellitus), clinical conditions (onset, systolic and diastolic blood pressure, heart rate, Killip class, and AMI location), laboratory parameters (hemoglobin, estimated glomerular filtration rate [eGFR], low density lipoprotein [LDL], and triglycerides), and therapy.

The levels of Gal-3 in the NAC group and the control group at the time of admission did not show any significant differences; however, the Gal-3 levels between the NAC and control group at 72 hours were found to be significant differences ($p=0.017$; **Table 2**).

For NAC group, Gal-3 levels were compared during admission and after 72 hours. They showed a significant difference (p=0.0001). No differences were found in the control group (**Table 3**).
There was a significant difference in the changes (delta) of Gal-3 levels between the NAC group and the control group (p=0.014; **Table 4**).
In the NAC group, the 72-hour GLS value was better and was significantly different from the 72-hour GLS value of the control group (p=0.023).

Discussion

The effect of N-acetylcysteine on galectin-3

Administration of oral NAC as additional therapy for AMI showed a significant difference in lowering Gal-3 levels in STEMI patients (p=0.017). The results of this study were in line with those of the study by Talasaz, et al in 2014, (13) which aimed to evaluate the effect of NAC on cardiac remodeling by measuring MMP 9 and MMP 2 levels in 98 AMI patients. MMP 9 and MMP 2 levels were significantly lower in the group of patients who received oral NAC supplementary therapy as compared to the placebo group (p=0.014 and p=0.045, respectively). In 2013, Talasaz, et al (15) conducted a study to determine the effect of NAC on inflammatory factors such as pro-fibrotic cytokines, transforming growth factor-β (TGF-β), and tumor necrosis factor-α (TNF-α) in AMI patients. It concluded that administration of NAC could prevent TGF-β. TGF-β has a strong association with left ventricular ejection fraction, therefore it might be important in preventing remodeling. Gal-3 plays a role in this process where macrophages and TGF-β induce the activation of the myofibroblast through Gal-3; however, recruitment of macrophages and expression of TGF-β does not depend on Gal-3. (6)
Since Gal-3 has an important role in the pathophysiology of adverse cardiac remodeling and is an independent predictor of heart failure after AMI, it can be hypothesized that therapy inhibiting Gal-3 can prevent the development of heart failure in patients with AMI.
Decreasing Gal-3 levels through NAC administration can be explained by Gal-3's role as an antioxidant and anti-inflammatory agent that has an indirect inhibiting mechanism. NAC is a reactive oxygen species (ROS) scavenger, but its main role as a therapeutic antioxidant comes from its role as a precursor of cysteine in the synthesis of glutathione (GSH). Under the conditions of oxidative stress, the amount of GSH is reduced and this can be restored with NAC supplementation. (16) Inhibition of inflammation by NAC is through inhibiting the induction of the activator protein 1 (AP-1) and nuclear factor-kappa B (NF-κB) proinflammatory transcription factors, which are induced in response to oxidative stress. (17)

The effect of N-acetylcysteine on global longitudinal strain

Administration of oral NAC as additional therapy showed significant difference in lowering GLS in STEMI patients (p=0.023). This result is in line with that of a study by Joyce, et al in 2013. (18) GLS measurements in post-AMI patients showed specific benefits in the evaluation of left ventricular ejection fraction (LVEF) with regards to expansion of the infarct area. (18) GLS decrease in patients with AMI and is associated with the infarct area and LVEF; it also predicts left ventricular changes, possible clinical events, and the heart's response to reperfusion strategies. (9) It has been proven that NAC can reduce the effects of thrombolytic agents, such as reperfusion injury with stunning myocardial manifestations, arrhythmias, myocardial damage, and expansion of infarct size, in post-AMI patients. NAC in combination with streptokinase significantly reduces oxidative stress and improves ventricular function in patients with myocardial infarction (11) by decreasing plasma hydroperoxide concentration. (19)
Oxidative stress is implicated in the remodeling process after AMI. NAC is a potent antioxidant that can directly eliminate hydroxyl radicals, which reduces the detrimental effects of oxidative stress. Reduction of oxidative lesions in the target tissue allows adequate oxygen consumption and increases overall oxygenation. Through its action on vascular cell adhesion molecule 1 and adhesivity mediated by endothelium, NAC is beneficial in preventing early phase remodeling. (13) MMP is an important mediator that contributes significantly to left ventricular remodeling and its level is enhanced by TNF-α. TNF-α is a strong proinflammatory cytokine, and its role has been described in modulating MMP expression in vitro. The increase in MMP levels is associated with high collagen matrix concentrations, which can accelerate the development of progressive heart failure. By increasing glutathione content in cardiac tissue, NAC inhibits neutral sphingomyelinase, which mediates the harmful effects of TNF-α and it also prevents activation of NF-κB. (13)
Ischemia that occurs after AMI triggers an increase in levels of pro-fibrotic cytokines, such as TGF-β, which induces fibrotic deposition in cardiomyocytes. TGF-β plays a significant role in the pathogenesis of the remodeling process. Inhibiting its activity in the proliferative phase of remodeling

can prevent the occurrence of left ventricular hypertrophy and reduce the expansion of fibrosis in infarcted segments of myocardium, thus improving left ventricular geometry. NAC can convert this cytokine into its inactive form and inhibit the binding of these cytokines to its receptors. On the other hand, fibronectin, a glycoprotein involved in tissue remodeling, can be released in response to various cytokines, including TGF-β as the strongest stimulator. Thus, NAC can be effective in blocking tissue remodeling through inhibition of fibronectin production induced by TGF-β. (20)

Conclusion

Supplementary therapy with NAC can reduce Gal-3 levels and GLS in STEMI patients receiving fibrinolytic therapy as compared to STEMI patients who do not get additional NAC therapy. The findings of this study will provide a therapeutic option for the successful management of patients with AMI.

Competing interests

The authors report no conflict of interest associated with this work.

Table 1. Baseline characteristics of the patients

Variable	NAC group (n=17)	Control group (n=15)	p value
Demographic			
- Sex (n, %)			0.228
Male	16 (94.1)	12 (80.0)	
Female	1 (5.9)	3 (20.0)	
- Age (years)	55.24±10.19	58.27±8.07	0.363
Risk factors (n, %)			
- Hypertension	12 (70.6)	10 (66.7)	0.811
- Smoker	13 (76.5)	9 (60.0)	0.316
- Diabetes mellitus	5 (29.4)	1 (6.7)	0.100
Clinical conditions			
- Onset (hours)	4.82±2.63	4.80±2.65	0.980
- Systolic BP (mmHg)	136.71±24.39	132.20±28.39	0.626
- Diastolic BP (mmHg)	84.47±17.57	81.00±18.68	0.592
- Heart rate (beat/min)	75.33±19.09	78.00±11.93	0.669
- Killip class (n, %)			0.538
Class I	13 (76.5)	10 (66.7)	
Class II-IV	4 (23.5)	5 (33.3)	
- Infarct type (n, %)			0.421
Anterior STEMI	11 (64.7)	8 (53.3)	
Non-anterior STEMI	6 (35.3)	7 (46.7)	
Laboratory parameters			
- Hemoglobin (g/dl)	13.75±1.80	13.59±1.81	0.796
- eGFR (ml/min/1.73 m^2)	64.40±26.09	72.13±29.97	0.441
- LDL (mg/dl)	125.59±33.41	137.80±83.90	0.737
- Triglyceride (mg/dl)	193.88±149.46	113.47±46.69	0.055
Therapy (n, %)			
- Status of fibrinolytic			0.283
Successful	2 (11.8)	4 (26.7)	
Failed	15 (88.2)	11 (73.3)	
- ACE-I/ARB	15 (88.2)	14 (93.3)	0.927
- Beta blocker	12 (70.6)	14 (93.3)	0.100

Legend: NAC=N-acetylcysteine; STEMI=ST elevation myocardial infarction; eGFR=estimatad glomerular filtration rate; LDL=low density lipoprotein; ACE-I=angiotensin converting enzyme inhibitor; ARB=angiotensin receptor blocker.

Table 2. Changes in Gal-3 levels between groups

Variable	NAC group	Control group	p value
Admission	13.92±4.49	12.70±3.94	0.427
72 hours	8.40±2.55	11.21±3.70	0.017

Legend: NAC=N-acetylcysteine.

Table 3. Changes of Gal-3 levels in NAC and control groups

Variable	NAC group		
	Admission	72 hours	p value
Gal-3	13.92±4.49	8.41±2.55	0.0001
Variable	Control group		
	Admission	72 hours	p value
Gal-3	12.71±3.95	11.22±3.71	0.310

Legend: Gal-3=galectin-3; NAC=N-acetylcysteine.

Table 4. Comparison of changes (delta) in levels of Gal-3 between groups

Variable	NAC group	Control group	p value
Gal-3	-5.51±3.06	-1.49±5.49	0.014

Legend: Gal-3=galectin-3; NAC=N-acetylcysteine.

Table 5. Comparison of 72 hours GLS values between groups

Variable	NAC group	Control group	p value
GLS	-13.62±3.60	-10.61±3.49	0.023

Legend: GLS=global longitudinal strain; NAC=N-acetylcysteine.

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