Increased long non-coding RNA NORAD reflects serious cardiovascular stenosis, aggravated inflammation status, and higher lipid level in coronary heart disease

Xiaoyun Zhang1 | Xuetong Kan2 | Jingjing Shen3 | Jian Li4

1 General Practice, Tianjin Fifth Central Hospital, Tianjin, China
2 Clinical Laboratory, Tianjin Fifth Central Hospital, Tianjin, China
3 Department of Cardiovascular Medicine, Tianjin Fifth Central Hospital, Tianjin, China
4 Department of Rheumatology and Immunology, Tianjin Fifth Central Hospital, Tianjin, China

Correspondence
Xiaoyun Zhang. General Practice, Tianjin Fifth Central Hospital, No. 41 Zhejiang Road, Tianjin 300450, China.
Email: yunzhuang0469282@163.com

Abstract
Objective: Long non-coding RNA activated by DNA damage (Inc-NORAD) modulates inflammation, lipid level, and atherosclerosis in various cardiovascular diseases. This study intended to investigate the dysregulated expression of Inc-NORAD, and its linkage with clinical characteristics, inflammatory cytokines, and accumulating major adverse cardiovascular events (MACE) in coronary heart disease (CHD) patients.

Methods: Totally, 160 CHD patients, 30 disease controls (DCs), and 30 healthy controls (HCs) were included. The reverse transcription-quantitative polymerase chain reaction was used to detect Inc-NORAD expression in peripheral blood mononuclear cell samples from all participants. Enzyme-linked immunosorbent assay was applied to detect proinflammatory cytokines and adhesion molecules in CHD patients. Then, MACE was recorded during a median follow-up of 12 (range: 1.0–27.0) months.

Results: Inc-NORAD was highest in CHD patients, followed by DCs, and lowest in HCs (p < 0.001). In CHD patients, Inc-NORAD was positively linked with Gensini score (p = 0.001). Meanwhile, Inc-NORAD was positively linked to C-reactive protein (p = 0.023), tumor necrosis factor-alpha (p = 0.016), interleukin (IL)-6 (p = 0.003), IL-8 (p = 0.018), and IL-17A (p = 0.029). No relation of Inc-NORAD with vascular cell adhesion molecule-1 (p = 0.094) and intercellular adhesion molecule-1 (p = 0.060) was found. Furthermore, Inc-NORAD was positively related to total cholesterol (p = 0.014) and low-density lipoprotein cholesterol (p = 0.004), whereas Inc-NORAD was not linked to triglyceride (p = 0.103) and high-density lipoprotein cholesterol (p = 0.533). However, Inc-NORAD (high vs. low), and its higher quartiles were both not linked to accumulating MACE rate (p > 0.05).

Conclusion: Increased Inc-NORAD is linked with aggravated stenosis degree, inflammation status, and blood lipid in CHD patients. However, further validation is required.

Keywords
blood lipid, coronary heart disease, inflammatory cytokines, Inc-NORAD, stenosis degree
1 | INTRODUCTION

Coronary heart disease (CHD), also known as ischemic heart disease, is a common type of heart disease where the arteries struggle to deliver sufficient oxygen-rich blood to the heart. According to 2022 heart disease data, CHD contributes to nearly 42% of cardiovascular disease death in the United States. At present, the treatments for CHD patients have achieved certain progress, including lifestyle changes (such as healthy eating, exercise, smoking cessation, etc.), the administration of medicines (such as antithrombotic drugs, statins, beta-blockers, etc.), and surgery (such as percutaneous coronary intervention and coronary artery bypass grafting, etc.). Nevertheless, the incidence of recurrence achieves nearly 14% to 18%, and the incidence of all-cause cardiovascular death achieves approximately 39%–42%, both of them remain high in CHD patients. Thus, it is crucial to discover potential biomarkers to monitor the disease progression and further improve the stratified management in CHD patients.

Long non-coding RNA activated by DNA damage (lnc-NORAD) has received a lot of attention for its capacity to regulate lipid metabolism, atherosclerosis, and inflammation by modulating microRNA (miR)-495-3p, miR-125a-3p, vascular endothelial growth factor (VEGF) gene transcription, etc. For example, lnc-NORAD inhibition suppresses blood lipid level, atherosclerotic plaque area, inflammation, and oxidative stress by regulating miR-495-3p in the aorta of atherosclerosis mice. Meanwhile, lnc-NORAD knockdown inhibits atherosclerosis and vascular endothelial cell injury by elevating VEGF gene transcription in apolipoprotein E (ApoE)−/− mice. Moreover, lnc-NORAD knockdown reduces inflammation, as well as improves cardiac functions and fibrosis via modulating the microRNA (miR)-125a-3p/Fyn pathway in diabetic cardiomyopathy (DCM) mice. Considering lipid metabolism, atherosclerosis, and inflammation play important roles in the pathology and progression of CHD, it could be hypothesized that lnc-NORAD might have high potency to serve as a candidate biomarker for CHD patients. Nevertheless, no relevant study reports that.

Accordingly, this study aimed to explore the dysregulated expression of lnc-NORAD, as well as its linkage with stenosis degree, inflammatory cytokines, lipid level, and major adverse cardiovascular events (MACE) in CHD patients.

2 | METHODS

2.1 | Participants

This study was performed between January 2020 and July 2021. The protocol of our study was permitted by Ethics Committee. The informed consent was acquired from each participant.

A total of 160 CHD patients who had coronary angiography (CAG) for unknown chest pain or suspected CHD symptoms were continuously recruited. The CHD patients were enrolled if they were: (1) diagnosed as CHD by CAG; (2) ≥18 years old; (3) understood and accepted the research protocol. The CHD patients were excluded if they had: (1) active infections; (2) severe liver or kidney disorders; (3) systemic inflammatory or autoimmune disorders; (4) solid cancer or hematological malignancies; (5) women with positive pregnancy test or lactation.

At the same time, another 30 non-CHD patients who had CAG for unknown chest pain or suspected CHD symptoms (angina, etc.) were included as disease controls (DCs). The inclusion criteria for DCs were: (1) diagnosed as non-CHD by CAG; (2) ≥18 years old. Besides, 30 healthy adults were enrolled as healthy controls (HCs). The DCs and HCs were age- and sex-matched with CHD patients. The DCs and HCs were excluded for the same exclusion criteria as CHD patients.

2.2 | Data and sample collection

The CHD patients’ demographics and medical history were collected after enrollment. For evaluating the degree of coronary artery stenosis, the Gensini score of CHD patients was calculated through CAG findings. The CHD patients’ biochemical indexes were obtained via routine blood tests after admission. The peripheral blood (PB) samples of CHD patients and DCs were collected before CAG. The PB samples of HCs were gathered after enrollment. For further lab tests, the peripheral blood mononuclear cells (PBMCs) of all participants and the serum of CHD patients were separated from PB samples.

2.3 | Reverse transcription-quantitative polymerase chain reaction (RT-qPCR) assay

Lnc-NORAD expression from PBMCs was estimated by RT-qPCR. In brief, the extraction of total RNA was performed using Trizol reagent (Beyotime, China). The reverse transcription (42°C, 15 min) was finished by BeyoRT™ M-MuLV Reverse Transcriptase (Beyotime, China). Continuously, qPCR was carried out with BeyoFast™ SYBR Green qPCR Mix (Beyotime, China). The RT-qPCR (the thermocycling condition was 1 cycle of 94°C for 30 sec, 40 cycles of 55°C for 1 min, and 40 cycles of 72°C for 1 min) was performed by SYBR Green (Takara Biotechnology, China). The primer sequence was as follows: NORAD forward: 5′- CATTGGGCAGACCTACCTA-3′; reverse: 5′- ACGTGCCCTGTCATTACC-3′; GAPDH forward: 5′- AAACATTGTGGTCGATTGGG-3′; reverse: 5′- CCTGGAAGATGGTGATGG-3′. The primers were referenced from former literature. Lnc-NORAD was calculated by 2−ΔΔCt method and glyceraldehyde-3-phosphate dehydrogenase (GAPDH) was treated as an internal reference. The detection of lnc-NORAD by RT-qPCR was triplicate.

2.4 | Enzyme-linked immunosorbent assay (ELISA)

The levels of tumor necrosis factor-alpha (TNF-α), interleukin (IL)-6, IL-8, IL-17A, vascular cell adhesion molecule (VCAM)-1,
and intercellular adhesion molecule (ICAM)-1 in CHD patients’ serum were quantified by commercial ELISA kits (TNF-α, cat. no. BMS223-4; IL-6, cat. no. KHC0061; IL-8, cat. no. BMS204-3; IL-17A, cat. no. BMS2017; VCAM-1, cat. no. KHT0601; ICAM-1, cat. no. BMS241); (Thermo Fisher Scientific). The procedures were carried out by manufacturer’s protocols.

### 2.5 Follow-up

All CHD patients were followed up by telephone follow-up survey and periodic reexamination. The last follow-up date for CHD patients was June 29, 2022. The MACE of CHD patients was recorded, including cardiovascular death, any myocardial infarction, any repeat revascularization, and hospital admission for cardiovascular cause. Then the accumulating MACE rate was calculated. The median duration of follow-up was 12.0 months with a range of 1.0–27.0 months.

### 2.6 Statistical analysis

The statistical analyses and graph plotting were performed through SPSS v27.1 (IBM Corp.) and GraphPad Prism v7.01 (GraphPad Software Inc.), respectively. The comparisons were analyzed via Kruskal–Wallis test by ranks. The diagnostic ability of lnc-NORAD was analyzed via Receiver operating characteristic (ROC) curve. The correlation was evaluated through Spearman’s rank correlation test. The accumulating MACE rate was performed via Kaplan–Meier curves, and the differences were analyzed through the log-rank test. A p value < 0.05 was considered statistically significant.

### 3 RESULTS

#### 3.1 Clinical features

The enrolled CHD patients included 38 (23.8%) females and 122 (76.2%) males with a mean age of 62.2 ± 10.1 years. Meanwhile, the median (interquartile range [IQR]) value of total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C), and C-reactive protein (CRP) were 4.7 (3.8–5.3) mmol/L, 3.2 (2.6–3.9) mmol/L, and 6.4 (4.9–9.2) mg/L, respectively. Regarding the stenosis degree, the median (IQR) value of Gensini score was 30.5 (17.5–49.0). Furthermore, the median (IQR) values of TNF-α, IL-6, IL-8, and IL-17A were 39.5 (30.6–48.3) pg/ml, 18.5 (15.4–23.5) pg/ml, and 103.0 (81.2–130.4) pg/ml, separately. Regarding the adhesion molecules, the median (IQR) values of vascular cell adhesion molecule-1 (VCAM-1) and intercellular adhesion molecule-1 (ICAM-1) were 611.0 (473.9–821.3) ng/ml and 117.3 (88.3–164.1) ng/ml in CHD patients. Other clinical information is listed in Table 1.

#### 3.2 Lnc-NORAD expressions

Lnc-NORAD was highest in CHD patients (median [IQR]: 2.71 (2.16–4.29)), followed by DCs (median [IQR]: 1.58 (1.05–2.59), and lowest in HCs (median [IQR]: 1.05 (0.72–1.62)) (p < 0.001) (Figure 1A). Meanwhile, lnc-NORAD exhibited an acceptable ability for

### Table 1 Characteristics of CHD patients

| Items                        | CHD patients (N = 160) |
|------------------------------|------------------------|
| Demographics                 |                        |
| Age (years), mean ± SD       | 62.2 ± 10.1            |
| Gender, n (%)                |                        |
| Female                       | 38 (23.8)              |
| Male                         | 122 (76.2)             |
| BMI (kg/m²), mean ± SD       | 24.6 ± 3.0             |
| Smoke, n (%)                 |                        |
| Never                        | 83 (51.9)              |
| Former                       | 44 (27.5)              |
| Current                      | 33 (20.6)              |
| Medical history              |                        |
| Hypertension, n (%)          | 114 (71.3)             |
| Hyperlipidemia, n (%)        | 92 (57.5)              |
| DM, n (%)                    | 45 (28.1)              |
| Family history of CAD, n (%) | 41 (25.6)              |
| Biochemical indexes          |                        |
| FBG (mmol/L), median (IQR)   | 5.5 (4.7–6.5)          |
| Scr (μmol/L), median (IQR)   | 74.2 (66.4–84.5)       |
| SUA (μmol/L), median (IQR)   | 349.1 (302.2–398.6)    |
| TG (mmol/L), median (IQR)    | 1.8 (1.0–2.5)          |
| TC (mmol/L), median (IQR)    | 4.7 (3.8–5.3)          |
| LDL-C (mmol/L), median (IQR) | 3.2 (2.6–3.9)          |
| HDL-C (mmol/L), median (IQR) | 0.9 (0.8–1.1)          |
| CRP (mg/L), median (IQR)     | 6.4 (4.9–9.2)          |
| Stenosis degree              |                        |
| Gensini score, median (IQR)  | 30.5 (17.5–49.0)       |
| Inflammatory cytokines       |                        |
| TNF-α (pg/ml), median (IQR)  | 39.5 (30.6–48.3)       |
| IL-6 (pg/ml), median (IQR)   | 18.5 (15.4–23.5)       |
| IL-8 (pg/ml), median (IQR)   | 103.0 (81.2–130.4)     |
| IL-17A (pg/ml), median (IQR) | 44.6 (33.3–65.8)       |
| Adhesion molecules           |                        |
| VCAM-1 (ng/ml), median (IQR) | 611.0 (473.9–821.3)    |
| ICAM-1 (ng/ml), median (IQR) | 117.3 (88.3–164.1)     |

Abbreviations: BMI, body mass index; CAD, coronary artery disease; CHD, coronary heart disease; CRP, C-reactive protein; DM, diabetes mellitus; FBG, fasting blood glucose; HDL-C, high-density lipoprotein cholesterol; ICAM-1, intercellular adhesion molecule-1; IL-17A, interleukin-17A; IL-6, interleukin-6; IL-8, interleukin-8; IQR, interquartile range; LDL-C, low-density lipoprotein cholesterol; Scr, serum creatinine; SD, standard deviation; SUA, serum uric acid; TC, total cholesterol; TG, triglyceride; TNF-α, tumor necrosis factor-alpha; VCAM-1, vascular cell adhesion molecule-1.
discriminating CHD patients from DCs (area under the curve (AUC): 0.767, 95% confidence interval (CI): 0.680–0.854); besides, the level of Inc-NORAD at the best cut-off point was 1.950ng/ml (Figure 1D). Notably, Inc-NORAD had an impressive capacity for distinguishing CHD patients from HCs (AUC: 0.901, 95% CI: 0.847–0.955); additionally, the level of Inc-NORAD at the best cut-off point was 1.950ng/ml (Figure 1C). However, the ability of Inc-NORAD to discriminate DCs from HCs was weakened (AUC: 0.704, 95% CI: 0.574–0.835); moreover, the level of Inc-NORAD at the best cut-off point was 1.005ng/ml (Figure 1D).

3.3 | Linkage of Inc-NORAD with stenosis degree, inflammation, adhesion molecules, and blood lipid

Lnc-NORAD was positively linked to the Gensini score \((p = 0.001)\) (Figure 2A); meanwhile, a positive linkage was also observed between Inc-NORAD and CRP \((p = 0.023)\) (Figure 2B). Concurrently, positive correlations were observed in Inc-NORAD with TNF-\(\alpha\) \((p = 0.016)\) (Figure 3A), IL-6 \((p = 0.033)\) (Figure 3B), IL-8 \((p = 0.018)\) (Figure 3C), IL-17A \((p = 0.029)\) (Figure 3D). Nevertheless, Inc-NORAD was not related to VCAM-1 \((p = 0.094)\) (Figure 3E) or ICAM-1 \((p = 0.060)\) (Figure 3F). Moreover, Inc-NORAD was positively linked with TC \((p = 0.014)\) and LDL-C \((p = 0.004)\). However, no correlation was found in Inc-NORAD with triglyceride (TG) \((p = 0.103)\) or high-density lipoprotein cholesterol (HDL-C) \((p = 0.533)\) in CHD patients (Table 2).

3.4 | Relationship between Inc-NORAD and accumulating MACE rate

According to the median value of Inc-NORAD in CHD patients; CHD patients were divided into Inc-NORAD high and low groups. It was found that no correlation was discovered between Inc-NORAD and accumulating MACE rate \((p = 0.147)\) in CHD patients; the 1-year and 2-year accumulating MACE rates were 8.2% and 20.0% in patients with Inc-NORAD high; whereas they were 4.3% and 7.5% in patients with Inc-NORAD low (Figure 4A).

Moreover, according to the 1/4 quartile, median, and 3/4 quartile levels of Inc-NORAD in CHD patients, CHD patients were divided into Inc-NORAD quartiles 1 (minimal-1/4 quartile), 2 (1/4 quartile-median), 3 (median-3/4 quartile), and 4 (3/4 quartile-maximal) groups. It was observed that accumulating MACE rate did not differ among patients with Inc-NORAD quartiles 1, 2, 3, and 4 \((p = 0.396)\). Specifically, the 1-year, 2-year accumulating MACE rates in patients with Inc-NORAD quartile 4 were 11.3% and 21.1%; they were 5.3% and 18.5% in patients with Inc-NORAD quartile 3, 5.3% and 11.2% in patients with Inc-NORAD quartile 2, as well as 3.1% and 3.1% in patients with Inc-NORAD quartile 1 (Figure 4B).

4 | DISCUSSION

Lnc-NORAD regulates atherosclerosis in various ways, thereby facilitating the pathogenesis and progression of cardiovascular diseases.8,10 A study reports that Inc-NORAD facilitates...
atherosclerosis by suppressing VEGF gene transcription in ApoE−/− mice.10 Meanwhile, Inc-NORAD inhibition suppresses atherosclerosis by elevating miR-495-3p to attenuate Krüppel-like factor 5 (KLF5).8 Apart from its regulation in atherosclerosis, Inc-NORAD is found to be upregulated in various cardiovascular diseases.8,15 For instance, Inc-NORAD is elevated in myocardial infarction (MI) left ventricle tissues in mice.15 Moreover, an increment of Inc-NORAD is found in atherosclerotic mice.8 The present study observed that Inc-NORAD was highest in CHD patients, followed by DCs, and lowest in HCs; meanwhile, Inc-NORAD was linked to higher CHD risk. The possible reason would be that: Inc-NORAD could facilitate CHD by boosting inflammation, endothelial dysfunction, atherosclerotic plaque area, and collagen fibers.8 Therefore, Inc-NORAD was linked to increased CHD risk.

The current study also estimated the linkage of Inc-NORAD with inflammation status, stenosis degree, and blood lipid. The detailed findings and corresponding explanations were listed as follows: First of all, Inc-NORAD was positively linked to inflammation (reflected by CRP, TNF-α, IL-6, IL-8, and IL-17A). The potential reason would be that: Inc-NORAD might regulate the inflammation in various ways, such as targeting the miR-485/nuclear respiratory factor 1 (NRF1) or miR-495-3p/KLF5 axis.8,16 Therefore, Inc-NORAD was positively related to inflammation. Secondly, a positive linkage was discovered between Inc-NORAD and stenosis degree. The possible explanation could be that: Inc-NORAD could regulate inflammation, cardiac fibrosis, atherosclerosis, etc. in various ways as mentioned above, which directly or indirectly facilitated the cardiovascular stenosis in CHD.
patients. Thus, Inc-NORAD was positively linked to stenosis degree. Thirdly, it was also discovered that raised Inc-NORAD was linked with increased blood lipid levels. It could be argued that Inc-NORAD might mediate blood lipid levels by modulating miR-495-3p to reduce KLF5. Therefore, a positive linkage was also found between Inc-NORAD and blood lipid levels. Notably, VCAM-1 and ICAM-1 could promote leukocyte infiltration, neutrophil recruitment, and endothelial cell proliferation to participate in the progression of atherosclerosis; therefore, they may play important roles in the progression of CHD. As a result, this study also evaluated the linkage of Inc-NORAD with VCAM-1 and ICAM-1; however, no linkages were found, which might due to the small sample size.

Moreover, the predictive value of Inc-NORAD for accumulating MACE risk was also analyzed. The finding suggested that Inc-NORAD was slightly related to increased accumulating MACE rate in CHD patients; however, it did not reach statistical significance. The potential reason would be that: the occurrence of MACE was mainly due to the characteristics of the disease itself and the intervention of the treatment modality therefore, the predictive value for individual factors might tend to be weakened. Besides, the follow-up duration was not enough, which might lead to the predictive value of Inc-NORAD for MACE not obvious. As a result, Inc-NORAD lacked predictive value for MACE risk in CHD patients. Clinically, Inc-NORAD plus other traditional prognostic markers, such as CRP, diabetes, etc. might assist in predicting prognosis in CHD patients and improving the management of CHD patients. However, further validation is needed.

Notably, considering that Inc-NORAD was not related to MACE even when CHD patients were divided into Inc-NORAD quartiles 1 (minimal-1/4 quartile), 2 (1/4 quartile-median), 3 (median-3/4 quartile), and 4 (3/4 quartile-maximal) groups; therefore, multivariate regression analysis was not performed in this study. Besides, Inc-NORAD was already linked to other prognostic factors, such as CRP, Gensini score, TNF-α, IL-6, total cholesterol, low-density lipoprotein cholesterol, etc. Therefore, these factors would affect each other and further interfere with the results. Taking these together, this study did not apply multivariate regression analysis to further verify the correlation between them.

Several limitations might exist in this study: (1) this was a single-center study; therefore, selection bias might exist; (2) multiple time point detection of Inc-NORAD was not realized in the current study; however, this could be meaningful for evaluating the change of Inc-NORAD in monitoring the progression of CHD; subsequent studies could take this into account; (3) the detailed mechanism of Inc-NORAD regulated inflammation, atherosclerosis, and lipid level in CHD patients was not explored, which needed to be further considered.

Conclusively, elevated Inc-NORAD is linked with aggravated stenosis degree, inflammation status, and blood lipid in CHD patients. However, further validation is required.

### TABLE 2 Correlation of Inc-NORAD with blood lipid indexes in CHD patients

| Items     | Lnc-NORAD | r | p value |
|-----------|-----------|---|---------|
| TG        | 0.129     | 0.103 |
| TC        | 0.194     | 0.014 |
| LDL-C     | 0.226     | 0.004 |
| HDL-C     | -0.050    | 0.533 |

Abbreviations: CHD, coronary heart disease; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; Lnc-NORAD, IncRNA non-coding RNA activated by DNA damage; TC, total cholesterol; TG, triglyceride.

![Figure 4](image)

**FIGURE 4** No correlation was found between Inc-NORAD and accumulating MACE rate. Association between Inc-NORAD high and accumulating MACE rate (A); association between Inc-NORAD quartile and accumulating MACE rate (B) in CHD patients. CHD, coronary heart disease; Inc-NORAD, long non-coding RNA activated by DNA damage; MACE, major adverse cardiovascular events.
CONFLICT OF INTEREST
None.

DATA AVAILABILITY STATEMENT
The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

INFORMED CONSENT
The informed consent was acquired from each participant.

ORCID
Xiaoyun Zhang  https://orcid.org/0000-0002-2631-7706

REFERENCES
1. Jensen RV, Hjortbak MV, Botker HE. Ischemic heart disease: an update. Semin Nucl Med. 2020;50(3):195-207.
2. Tsao CW, Aday AW, Almarzooq ZI, et al. Heart disease and stroke Statistics-2022 update: a report from the American Heart Association. Circulation. 2022;145(8):e153-e639.
3. Akuz A. Exercise and coronary heart disease. Adv Exp Med Biol. 2020;1228:169-179.
4. Jia S, Liu Y, Yuan J. Evidence in guidelines for treatment of coronary artery disease. Adv Exp Med Biol. 2020;1177:37-73.
5. Peters SAE, Colantonio LD, Dai Y, et al. Trends in recurrent coronary heart disease after myocardial infarction among US women and men between 2008 and 2017. Circulation. 2021;143(7):e51-55.
6. Fu DN, Wang Y, Yu L, Liu MJ, Zhen D. Silenced long non-coding RNA activated by DNA damage elevates microRNA-495-3p to suppress atherosclerotic plaque formation via reducing Kruppel-like factor 5. Exp Cell Res. 2021;401(2):112519.
7. Peters SAE, Colantonio LD, Dai Y, et al. Trends in recurrent coronary heart disease after myocardial infarction among US women and men between 2008 and 2017. Circulation. 2021;143(7):e51-55.
8. Liu Y, Zhu Y, Liu S, Liu J, Li X. NORAD lentivirus shRNA mitigates fibrosis and inflammatory responses in diabetic cardiomyopathy via the cerRNA network of NORAD/miR-125a-3p/Fyn. Inflamm Res. 2021;70(10–12):1113-1127.
9. Geldini GG. A more meaningful scoring system for determining the severity of coronary heart disease. Am J Cardiol. 1983;51(3):606.
10. Livak KJ, Schmittgen TD. Analysis of relative gene expression data using real-time quantitative PCR and the 2⁰(Delta Delta C[T]) method. Methods. 2001;25(4):402-408.
11. Greenwood JP, Ripley DP, Berry C, et al. Effect of care guided by cardiovascular magnetic resonance, myocardial perfusion scintigraphy, or NICE guidelines on subsequent unnecessary angiography rates: the CE-MARC 2 randomized clinical trial. JAMA. 2016;316(10):1051-1060.
12. Zhao X, Wei X, Wang X, Qi G. Long noncoding RNA NORAD regulates angiogenesis of human umbilical vein endothelial cells via miR5903p under hypoxic conditions. Mol Med Rep. 2020;21(6):2560-2570.
13. Wang L, Yuan X, Lian L, Guo H, Zhang H, Zhang M. Knockdown of LncRNA NORAD inhibits the proliferation, inflammation and fibrosis of human mesangial cells under high-glucose conditions by regulating the miR-485/NRF1 axis. Exp Ther Med. 2021;22(2):874.
14. Jiang L, Yang A, Li X, Liu K, Tan J. Down-regulation of VCAM-1 in bone mesenchymal stem cells reduces inflammatory responses and apoptosis to improve cardiac function in rat with myocardial infarction. Int Immunopharmacol. 2021;101(Pt A):108180.
15. Gonzalez-Ramos S, Paz-Garcia M, Rius C, et al. Endothelial NOD1 directly mediates cell recruitment in atherosclerosis through VCAM-1. FASEB J. 2019;33(3):3912-3921.
16. Salvador AM, Nevers T, Velazquez F, et al. Intercellular adhesion molecule 1 regulates leaflet ventricular leukocyte infiltration, cardiac remodeling, and function in pressure overload-induced heart failure. J Am Heart Assoc. 2016;5(3):e003126.
17. Zeitouni M, Clare RM, Chiswell K, et al. Risk factor burden and long-term prognosis of patients with premature coronary artery disease. J Am Heart Assoc. 2020;9(24):e017712.
18. Hageman SHJ, de Borst GJ, Dorresteijn JAN, et al. Cardiovascular risk factors and the risk of major adverse limb events in patients with symptomatic cardiovascular disease. Heart. 2020;106(21):1686-1692.
19. Okkonen M, Havulina AS, Ukkola O, et al. Risk factors for major adverse cardiovascular events after the first acute coronary syndrome. Ann Med. 2021;53(1):817-823.
20. Basavarajaiah S, Mitomo S, Nakamura S, et al. Long-term outcome following percutaneous intervention of intra-stent coronary occlusion and evaluating the different treatment modalities. Int J Cardiol Heart Vas. 2021;34:100803.
21. Zheng R, Liu Y, Hao Z, Liao H, Xiao C. Clinical characteristics and prognosis of young patients with coronary heart disease. Med Sci Monit. 2020;26:e922957.
22. Zhao J, Wang X, Wang H, Zhao Y, Fu X. Occurrence and predictive factors of restenosis in coronary heart disease patients underwent sirolimus-eluting stent implantation. Ir J Med Sci. 2020;189(3):907-915.

How to cite this article: Zhang X, Kan X, Shen J, Li J. Increased long non-coding RNA NORAD reflects serious cardiovascular stenosis, aggravated inflammation status, and higher lipid level in coronary heart disease. J Clin Lab Anal. 2022;36:e24717. doi: 10.1002/jcla.24717