Pharmacological Effects of Nardosinone in Anti-Rheumatoid Arthritis, Anti-Tumor, Anti-Arrhythmia, and Myocardial Protection

Huizhen PEI, Jiaru YE, Shuang REN, Linbo JIA, Wenhui ZHAO, Chenghao JIN*

College of Life Science and Biotechnology, Heilongjiang Bayi Agricultural University, Daqing 163319, China

Abstract This paper systematically summarizes the pharmacological effects and molecular mechanisms of nardosinone (Nar) by consulting the literature at home and abroad. The results demonstrate that Nar alleviates the inflammatory response in rheumatoid arthritis by inhibiting the TLR4/MyD88/NF-κB signaling pathway, induces ferroptosis in HT-1080 cells by promoting the accumulation of ROS and lipid peroxides, and exerts antiarrhythmic effects by inhibiting INa, Ito, and ICa-L currents in ventricular myocytes and prolonging their recovery time from inactivation. Moreover, it produces antidepressant effects by reducing the immobility time of mice in behavioral despair models, and protects the myocardium by activating the PI3K/Akt/mTOR pathway to inhibit apoptosis. Nar exhibits multi-target pharmacological activity and has application prospects in the prevention and treatment of autoimmune diseases, tumors, cardiac diseases, and psychiatric disorders.

Key words Nardosinone, Anti-rheumatoid arthritis, Anti-tumor, Anti-arrhythmia, Anti-depression, Myocardial protection

0 Introduction

Nardostachys chinensis is a plant of *Nardostachys* in the family Valerianaceae[1], mainly distributed in Qinghai, Sichuan, Yunnan, Tibet, and other regions. Traditional Chinese medicine theory holds that it has a range of biological activities, including antiarrhythmic, anti-myocardial ischemia, antioxidant, analgesic, and antimalarial effects[2-5]. Nardosinone (Nar) is a sesquiterpenoid compound isolated from *N. chinensis*. Nar has a molecular formula of $C_{15}H_{22}O_3$ and appears as a white powder, which is soluble in organic solvents such as methanol, ethanol, and DMSO. Modern pharmacological studies have found that Nar exhibits various pharmacological effects, such as anti-rheumatoid arthritis, anti-tumor, antiarrhythmic, antidepressant, myocardial protective activities, thus attracting extensive attention from researchers worldwide. This paper reviews the pharmacological effects and molecular mechanisms of Nar, aiming to provide a theoretical basis for further research, development, and utilization of Nar.

1 Anti-rheumatoid arthritis activity

Rheumatoid arthritis (RA) is a common autoimmune disease[6]. Persistent inflammatory stimulation promotes synovial hyperplasia and pannus formation, leading to cartilage and bone destruction in RA patients. Studies have shown that Nar can alleviate the symptoms of RA and exhibits a good anti-RA effect.

Lan *et al.*[7] examined the histopathological changes in the synovial tissue of rat ankle joints in the blank control group, model group, methotrexate (MTX) group, and Nar group using hematoxylin-eosin (HE) staining. The results showed that the synovial cells in the model group were hyperplastic obviously, and the synovial layer was significantly thickened. In contrast, both the MTX group and the Nar group exhibited only mild synovial thickening with a small amount of inflammatory cell infiltration, indicating that Nar can improve the pathological damage of the synovial tissue in the ankle joints of collagen-induced arthritis (CIA) model rats, and inhibit synovial cell hyperplasia and inflammatory cell infiltration. Furthermore, the expression levels of interleukin-1β (IL-1β), tumor necrosis factor-α (TNF-α), and inducible nitric oxide synthase (iNOS) in the serum of rats in each group were detected by enzyme-linked immunosorbent assay (ELISA). The results revealed that compared with the blank control group, the serum levels of IL-1β, TNF-α, and iNOS were significantly elevated in the model group. In contrast, compared with the model group, the serum levels of IL-1β, TNF-α, and iNOS in both the MTX group and the Nar group were significantly reduced. These findings indicate that Nar can significantly inhibit the expression of pro-inflammatory factors in CIA rats, reduce the release of inflammatory mediators, and thereby alleviate the inflammatory response of RA. The protein expression levels of Toll-like receptor 4 (TLR4), myeloid differentiation factor 88 (MyD88), nuclear factor kappa-B p50 subunit (NF-κB p50), and nuclear factor kappa-B p65 subunit (NF-κB p65) were further detected by Western blotting (WB). The results showed that compared with the blank group, the expression levels of TLR4, MyD88, NF-κB p65 and NF-κB p50 in the synovial tissue of rats in the model group were significantly higher, while compared with the model group, the expression levels of TLR4, MyD88, p-NF-κB p65 and NF-κ Bp50 in the synovial tissue of rats in the MTX group and Nar group decreased significantly. These findings indicates that Nar can inhibit the inflammatory response by downregulating the expression levels of proteins and genes related to the TLR4, MyD88, and NF-κB signaling pathways. In summary, Nar alleviates the inflammatory response in RA, and its action is associated with the inhibition of

Received: November 12, 2025 Accepted: April 20, 2026

Supported by Central Government Funds for Local University Reform and Development (Talent Cultivation Program) (2020GSP16); College Student Innovation and Entrepreneurship Training Program of Heilongjiang Province (202510223075).

Huizhen PEI, undergraduate, research fields: pharmacology of anticancer drugs. * Corresponding author. Chenghao JIN, professor, doctoral supervisor, research fields: pathogenesis of cancer and drug development.

the TLR4, MyD88, and NF-κB signaling pathways as well as the regulation of related metabolic pathways. It indicates that Nar exhibits good anti-RA activity and is expected to be developed into a new drug for treating RA.

2 Antitumor activity

Tumors is generally caused by unlimited clonal proliferation of local cells. They can induce abnormal hyperplasia of local tissues accompanied by malignant progression such as infiltration and metastasis, while also disrupting metabolism, immunity, and organ function. Studies have shown that Nar can induce ferroptosis in human fibrosarcoma cells (HT-1080), thereby exerting antitumor effects.

Chu *et al*.[8] detected the levels of reactive oxygen species (ROS) and lipid peroxides in cells by flow cytometry and fluorescence scanning confocal microscopy. The results showed that after 2 h of Nar treatment, the levels of ROS and lipid peroxides in HT-1080 cells were significantly elevated. However, upon the addition of ferroptosis inhibitors, both ROS and lipid peroxide levels in HT-1080 cells decreased. Among the inhibitors, deferoxamine mesylate (DFO) exhibited a more pronounced inhibitory effect than Lip-1. These findings indicate that Nar induces ferroptosis in HT-1080 cells by promoting the accumulation of intracellular ROS and lipid peroxides. Moreover, cell viability was assessed using the MTS assay after treatment with Nar alone or in combination with cell death inhibitors. It was found that Nar inhibited HT-1080 cell viability in a concentration-dependent manner (0.1, 0.2, 0.3, 0.4, and 0.5 mM), with a half-maximal inhibitory concentration (IC_{50}) of 391 μM at 24 h, indicating that Nar effectively induces HT-1080 cell death. Furthermore, the type of cell death induced by Nar was investigated using the apoptosis inhibitor z-vad-fmk and the necroptosis inhibitors necrostatin-1 (Nec-1). The results showed that neither z-vad-fmk nor Nec-1 had a significant effect on Nar-induced cell death. In contrast, both ferroptosis inhibitors, DFO and liproxstatin-1 (Lip-1), significantly suppressed Nar-induced cell death, indicating that Nar can specifically activate the ferroptosis pathway and induce cell death. In summary, Nar induces ferroptosis in HT-1080 cells by promoting the accumulation of intracellular ROS and lipid peroxides in an Fe-dependent manner, thereby exerting anti-human fibrosarcoma effects. These findings indicate that Nar exhibits good antitumor activity and is expected to be developed into a novel therapeutic agent for the treatment of tumors.

3 Antiarrhythmic activity

Arrhythmia (Arr) is a cardiac ion channel disease caused by abnormal excitation formation or conduction of cardiomyocytes, leading to reduced cardiac output and potentially life-threatening consequences. Abnormal electrical activity of cardiomyocytes, impaired conduction pathways, and autonomic nervous dysfunction can all contribute to arrhythmia. Studies have found that Nar can alleviate the symptoms of arrhythmia and exhibits good antiarrhythmic activity.

Qian[9] acutely isolated rat ventricular myocytes and employed the whole-cell patch-clamp technique to investigate the effects of Nar on INa, Ito, and ICa-L in rat ventricular myocytes. The results showed that with 1 μmol/L Nar, no significant changes were observed in INa, Ito, and ICa-L. However, when the concentration reached 3 μmol/L, Nar exhibited remarkable inhibitory effects on INa, Ito, and ICa-L, and these effects gradually intensified with the Nar concentration increasing, indicating that Nar can inhibit INa, Ito, and ICa-L in rat ventricular myocytes. Furthermore, the effects of Nar on the kinetic characteristics of INa, Ito, and ICa-L in rat ventricular myocytes were evaluated using paired-pulse stimulation combined with the whole-cell patch-clamp technique. The results showed that the recovery time constants (τ) for INa, Ito, and ICa-L after inactivation in the control group were (14.76 ± 1.38), (108.7 ± 3.49), and (19.12 ± 2.29) ms, respectively. Nar significantly prolonged the τ values in a concentrationdependent manner (3, 10, and 30 μmol/L), with the τ values increasing as the concentration rose. These results indicate that Nar can significantly alter the kinetic characteristics of INa, Ito, and ICa-L in rat ventricular myocytes, and concentrationdependently extend the recovery time of INa, Ito, and ICa-L from the inactivated state to the activated state, thereby further stabilizing myocardial electrical activity and exerting antiarrhythmic effects. In summary, Nar exerts concentrationdependent inhibitory effects on the three key ion channels in rat ventricular myocytes: INa, Ito, and ICa-L. Nar can stabilize the electrophysiological activity of cardiomyocytes by modulating the kinetic characteristics of these channels. These findings indicate that Nar possesses good antiarrhythmic activity and is expected to be developed into a novel therapeutic agent for the treatment of arrhythmia.

4 Antidepressant activity

Depression is a common but easily overlooked affective disorder. Its clinical manifestations include low mood, lethargy, reduced speech, pessimism, sleep disturbances, and social phobia[10]. Studies have shown that Nar can alleviate the symptoms of depression and exhibits good antidepressant activity.

Li[11] tested the effect of Nar on the number of spontaneous activities of normal mice through a mouse spontaneous activity experiment. The results showed that while the positive control (diazepam) group significantly reduced the number of spontaneous activities, Nar exhibited a decreasing trend in activity counts at various doses, but the differences were not statistically significant compared with the blank control group, indicating that Nar does not produce significant central inhibition or excitation in normal mice. Furthermore, the effect of Nar on the number of mice falling asleep induced by a sub-threshold dose of pentobarbital sodium was examined in a subthreshold pentobarbital sodium-induced sleep synergy test. The results showed that the positive control groups (diazepam and estazolam) exhibited a significant increase in the positive rate of sleep induction. Although the Nar groups at various doses also showed a rising trend in the positive rate, there were no significant differences compared with the control group,

and the effect of the high-dose Nar group was weaker than that of estazolam. These results indicate that Nar does not have a significant synergistic effect on the hypnotic action of a sub-threshold dose of pentobarbital sodium, although a slight central inhibitory trend was observed. Moreover, a tail suspension test was conducted to evaluate the effect of Nar on the immobility time of suspended mice. The results showed that compared with the control group, all Nar dose groups exhibited significant differences, indicating that Nar significantly reduced the immobility time of mice in the tail suspension test, suggesting that Nar may possess certain antidepressant activity. Furthermore, the effect of Nar on the immobility time during forced swimming was tested by a forced swimming test. The results revealed that Nar at a dose of 0.160 g/kg demonstrated a significant difference compared with the normal control group, and a dose-dependent relationship was observed, confirming that Nar significantly decreased the immobility time in the forced swimming test and further supporting its antidepressant potential. In summary, Nar has no significant effect on spontaneous activities of normal mice or on the hypnotic effect induced by a sub-threshold dose of pentobarbital sodium. However, it significantly shortens the immobility time of mice in behavioral despair models in a dose-dependent manner, exhibiting certain antidepressant activity along with a mild central sedative trend. These findings indicate that Nar possesses potent antidepressant activity and is expected to be developed into a novel therapeutic agent for the treatment of depression.

5 Myocardial protective activity

Ischemic heart disease (IHD) is a leading cause of death and disability worldwide. The clinical manifestations of IHD include myocardial infarction, heart failure, and cardiac dysfunction[12]. Studies have shown that Nar can alleviate the symptoms of IHD and exhibits good protective effects against IHD.

Li *et al.* [13] investigated the effect of Nar on cell viability in a hypoxiainjured H9c2 cardiomyocyte model induced by cobalt chloride ($CoCl_2$) using the Cell Counting Kit8 (CCK8) assay. The results showed that treatment of H9c2 cells with $CoCl_2$ for 24 h decreased cell viability in a concentrationdependent manner (0, 50, 100, 200, and 400 μmol/L). Pretreatment with 50 μmol/L Nar significantly increased cell viability, and this concentration exhibited a more pronounced protective effect, indicating that an appropriate concentration of Nar can effectively mitigate the decline in cell viability caused by hypoxic injury and exert a protective effect against hypoxia-induced damage in cardiomyocytes. Furthermore, the effect of Nar on hypoxiainduced cardiomyocyte apoptosis was evaluated using Hoechst 33342 staining. The results showed that in the 400 μmol/L $CoCl_2$ group, obvious apoptosis was observed, characterized by chromatin condensation and increased nuclear fragments. In contrast, pretreatment with 50 μmol/L Nar markedly improved nuclear morphology and significantly reduced apoptotic fragments, indicating that Nar pretreatment effectively inhibits hypoxiainduced cardiomyocyte apoptosis. Furthermore, the expression levels of proteins related to the phosphoinositide 3kinase (PI3K), protein kinase B (Akt), and mammalian target of rapamycin (mTOR) signaling pathways were examined by Western blotting. The results revealed that $CoCl_2$ treatment significantly decreased the phosphorylation levels of PI3K, Akt, and mTOR, while pretreatment with Nar increased the phosphorylation levels of PI3K, Akt, and mTOR. However, when the PI3K/Akt pathway was blocked by the PI3K inhibitor LY294002, the protective effect of Nar was significantly counteracted. It indicate that Nar alleviates $CoCl_2$-induced hypoxic injury through activation of the PI3K/Akt/mTOR signaling pathway. In summary, Nar exerts a protective effect against $CoCl_2$-induced hypoxic injury in cardiomyocytes by activating the PI3K/Akt/mTOR signaling pathway and inhibiting apoptosis. These findings indicate that Nar possesses good antiIHD activity and is expected to be developed into a novel therapeutic agent for the treatment of ischemic heart disease.

6 Conclusions and prospects

Nar exhibits multiple pharmacological activities, including antirheumatoid arthritis, antitumor, anti-arrhythmia, anti-depression, and myocardial protection. As a key sesquiterpenoid compound isolated from *N. chinensis*, its chemical isolation, purification, and pharmacological activity studies have achieved preliminary results. However, research on its mechanism of action and clinical application is still in the early stages. It is necessary to continuously integrate knowledge and experimental techniques from molecular biology, cell biology, experimental zoology, pharmacology, basic medicine, and related fields to conduct more comprehensive and indepth studies of Nar at the cellular, molecular, and animal levels. Such efforts will provide a theoretical basis and data support for the further development of Narbased traditional Chinese medicine preparations.

References

[1] HU MJ, TANG Y, FENG ZZ, *et al.* Chemical constituents from *Nardostachys chinensis*[J]. Chinese Traditional Patent Medicine, 2019, 41(7): 1597 – 1601. (in Chinese).

[2] JIAN P, LI QH, FAN LH. Experimental study on the inhibitory effect of nardosinone on myocardial cell in rats with tachyarrhythmia[J]. The Chinese Journal of Clinical Pharmacology, 2015, 31(22): 2240 – 2242. (in Chinese).

[3] LIU ML, DUAN YH, HOU YL, *et al.* Nardoaristolones A and B, two terpenoids with unusual skeletons from *Nardostachys chinensis* Batal[J]. Organic Letters, 2013, 15(5): 1000 – 1003.

[4] JING LL, MA HP, FAN XF, *et al.* Study on the antioxidant activity of different extracts from *Nardostachys chinensis* Batal[J]. Chemical Research and Application, 2014, 26(10): 1591 – 1596. (in Chinese).

[5] ZHANG JB, LIU ML, LI C, *et al.* Nardosinane-type sesquiterpenoids of *Nardostachys chinensis* Batal[J]. Fitoterapia, 2015: 100195 – 100200.

[6] DI MATTEO A, BATHON JM, EMERY P. Rheumatoid arthritis[J]. Lancet, 2023, 402: 2019 – 2033.

[7] LAN WY, YAN JJ, LU QW, *et al.* Exploration on the regulation of TLR4/MyD88/NF-κB signaling pathway by nardosinone in the treatment of rheumatoid arthritis based on untargeted metabolomics[J]. China Journal of Traditional Chinese Medicine and Pharmacy, 2025, 40(12): 5671 – 5677. (in Chinese)

(To page 57)

menting bloom's taxonomy to enhance student learning in biology[J]. CBE—Life Sciences Education, 2008, 7(4): 368 – 381.
[9] BIGGS J. Enhancing teaching through constructive alignment[J]. Higher Education, 1996, 32(3): 347 – 364.
[10] WANG XM, LIU ZJ. Research on instructional design of college english reading and writing course based on bloom's taxonomy of cognitive objectives[J]. Technology Enhanced Foreign Language Education, 2015, (2): 43 – 48, 63. (in Chinese).
[11] SWELLER J, VAN MERRIËNBOER JG, PAAS F. Cognitive architecture and instructional design: 20 years later[J]. Educational Psychology Review, 2019, 31: 261 – 292.
[12] BLACK P, WILIAM D. Assessment and classroom learning[J]. Assessment in Education: Principles, Policy & Practice, 1998, 5(1): 7 – 74.
[13] HATTIE J, TIMPERLEY H. The power of feedback[J]. Review of Educational Research, 2007, 77(1): 81 – 112.
[14] XIA Q, CHIU TKF, LEE M, *et al.* A scoping review on how generative artificial intelligence transforms assessment in higher education[J]. International Journal of Educational Technology in Higher Education, 2024, 21: 40.

(From page 48)

[8] CHU Z. Nardosinone, a natural product, induces ferroptosis in HT1080 tumor cells and its mechanism[D]. Guiyang: Guizhou Medical University, 2023. (in Chinese).
[9] QIAN W. Applied basic research on the antiarrhythmic effect of nardosinone[D]. Yangzhou: Yangzhou University, 2020. (in Chinese).
[10] MALBERG JE, EISCH AJ, NESTLER EJ, *et al.* Chronic antidepressant treatment increases neurogenesis in adult rat hippocampus[J]. Journal of Neuroscience, 2000, 20(24): 9104 – 9110
[11] LI Q. A preliminary study on the antidepressant effect and mechanism of nardosinone[D]. Beijing: Beijing University of Chinese Medicine, 2011. (in Chinese).
[12] NARULA J, PANDEY P, ARBUSTINI E, *et al.* Apoptosis in heart failure: Release of cytochrome c from mitochondria and activation of caspase-3 in human cardiomyopathy[J]. Proceedings of the National Academy of Sciences of the United States of America, 1999, 96(14): 8144 – 8149.
[13] LI HY, MEI XY, YANG HY, *et al.* Protective effect of nardosinone on hypoxic injury of H9c2 cardiomyocytes mediated through the PI3K/Akt/mTOR pathway[J]. Chinese Journal of Comparative Medicine, 2023, 33(7): 41 – 47. (in Chinese).