Clinical and Experimental Medicine (2025) 25:84
https://doi.org/10.1007/s10238-025-01602-5

RESEARCH

Investigating the pharmacological mechanisms of clopidogrel for carotid stenosis treatment based on network pharmacology and molecular docking techniques

Xu Wang[1] · Haibin Lu[1] · Jing Xie[1] · Chenglei Zhang[2]

Received: 4 January 2025 / Accepted: 14 February 2025

Abstract
Carotid artery stenosis is a manifestation of atherosclerosis and is associated with an increased risk of various cardiovascular diseases. Clopidogrel is an antiplatelet drug widely used for the prevention and treatment of atherosclerosis-related diseases. This study explores the potential molecular mechanisms of clopidogrel in the treatment of carotid artery stenosis through network pharmacology and molecular docking techniques. First, network pharmacology methods were used to construct a clopidogrel target network and identify its possible 127 action targets. Secondly, the gene ontology enrichment analysis indicated that clopidogrel for treating carotid stenosis is closely related to inflammatory responses, platelet activation, and angiogenesis. The Kyoto Encyclopedia of Genes and Genomes analysis revealed associations with lipid metabolism and atherosclerosis. Subsequently, molecular docking technology was employed to screen the binding affinity of clopidogrel to these targets. The results revealed that clopidogrel exhibited binding energies less than −4.20 kcal/mol with multiple targets, including TNF, MMP9, PTGS2, CCL2, TLR4, and IL-10. This indicates that clopidogrel has high binding affinity and stable binding modes with these targets, thereby exerting anti-inflammatory effects. This study reveals the potential molecular mechanisms of clopidogrel in the treatment of carotid artery stenosis through network pharmacology and molecular docking techniques. The experimental results provide a theoretical basis for the application of clopidogrel in the treatment of carotid artery stenosis and offer new ideas for further drug development and personalized treatment.

Keywords Clopidogrel · Carotid artery stenosis · Network pharmacology · Molecular docking · Inflammatory response

Background

Clopidogrel is an antiplatelet drug used to prevent and treat heart, brain, and other arterial circulation disorders caused by high platelet aggregation [1]. The development of clopidogrel began in 1972. Due to its significant therapeutic effects and better tolerance, it replaced ticlopidine, which caused severe hematological adverse reactions, and was marketed globally in 1988 [2].

Clopidogrel is mainly used for patients with acute coronary syndrome (ACS) and percutaneous coronary intervention (PCI), effectively reducing the incidence of stent thrombosis and significantly improving prognosis. Clopidogrel irreversibly binds to the ADP receptor P2Y12 on the platelet surface, blocking the inhibition of adenylate cyclase by ADP, thereby inhibiting platelet aggregation [3, 4]. Clopidogrel can also be used in combination with aspirin to enhance antiplatelet effects and reduce the risk of cardiovascular events [5].

Carotid artery stenosis is a common vascular disease, mainly due to obstruction of blood flow in the carotid artery, leading to insufficient blood supply to the brain [6]. One of the main causes is atherosclerosis, where lipid substances accumulate on the vessel walls, forming plaques that gradually narrow the lumen [7]. Secondly, it is also closely related to vascular lesions associated with development, inflammation, or autoimmune conditions, such as Takayasu arteritis, fibromuscular dysplasia,

✉ Chenglei Zhang
aries140aa@163.com

[1] The First People's Hospital of Yinchuan, Yinchuan 750001, Ningxia, China

[2] General Hospital of Ningxia Medical University, 804 Shengli South Street, Xingqing District, Yinchuan 750003, Ningxia, China

Published online: 14 March 2025

and moyamoya disease. Or, it may be caused by various reasons leading to blood entering between the layers of the vessel wall, causing dissection and forming carotid artery dissection [8]. Although clopidogrel is often used clinically as a first-choice drug for the treatment of carotid artery stenosis, its pharmacological mechanism is still unclear.

Network pharmacology is an emerging pharmacological research method that integrates principles and techniques from systems biology, network science, computational biology, and traditional pharmacology [9]. The core idea of network pharmacology is to study the mechanisms and efficacy of drugs by analyzing the complex molecular interaction networks within biological systems [10]. With the accumulation of biomedical data and advances in computational technology, network pharmacology is becoming increasingly important in drug development and precision medicine [11]. Molecular docking is a computational chemistry method used to predict and analyze the interactions and binding modes between small molecules (such as drug candidates) and biological macromolecules (such as proteins and nucleic acids) [12]. This technique is crucial in exploring drug mechanisms as it helps researchers understand how drug molecules bind to biological targets and predict potential pharmacological mechanisms [13].

This study, based on network pharmacology and molecular docking techniques, aims to identify common targets between clopidogrel and carotid artery stenosis, explore potential core targets and biological pathways, and predict the binding modes of clopidogrel to these core targets. This experiment aims to explore the pharmacological mechanisms of clopidogrel to provide more effective data support for future clinical trials.

Methods

Clopidogrel drug target screening

By searching "Clopidogrel" in the PubChem database, the standard structure and SMILES notation of clopidogrel were determined. Based on the SMILES notation, potential targets of clopidogrel were collected from the SwissTargetPrediction database (http://www.swisstargetprediction.ch/), ChEMBL database (https://www.ebi.ac.uk/chembl/), and GeneCards database (https://www.genecards.org/). The structures of the search results were carefully examined and compared to ensure consistency. Potential targets were integrated and deduplicated, and the obtained target names were standardized using the UniProt database. Finally, these combined targets were used to construct a clopidogrel target library.

Carotid artery stenosis disease target screening

Predict disease-related target genes using the GeneCards database. Search for "Carotid artery stenosis" in the GeneCards database and export the resulting disease genes to an Excel spreadsheet. Then, carefully examine, calibrate, and deduplicate the obtained target genes, and standardize the target gene names using the UniProt database. Finally, use these merged target genes to construct the "Carotid Artery Stenosis" target gene library.

Clopidogrel and carotid artery stenosis intersection target screening

Identify the intersecting target genes of clopidogrel and carotid artery stenosis using Venny 2.1.0 (https://bioinfogp.cnb.csic.es/tools/venny). The downloaded Venny image is used for result presentation. The overlapping part is considered the potential pharmacological target genes for clopidogrel in the treatment of carotid artery stenosis.

Construction of Protein–Protein Interaction (PPI) Network and Core Target Screening.

Input the intersecting genes between carotid artery stenosis and clopidogrel potential targets into the STRING database [14]. During the analysis, restrict the species to "Homo sapiens" and set the "minimum required interaction score" to "medium confidence (> 0.4)". These parameters ensure the analysis of active target proteins corresponding to target genes, generating initial protein–protein interaction (PPI) images and raw STRING data. Then, import the raw data generated by STRING into Cytoscape software (version 3.10.2), which is used for the visualization and analysis of network biology, capable of calculating parameters for each node in the network and visualizing molecular connections. To screen for core targets and create visualized network diagrams using Cytoscape, we first selected an appropriate layout algorithm, such as the "Force-directed" or "Hierarchy" layout, to organize the network structure. We then adjusted the node and edge styles through the "Style" menu, customizing attributes like color, size, and shape to better highlight network characteristics. Next, we installed and launched the "NetworkAnalyzer" plugin to calculate topological parameters, including degree centrality. Based on these parameters, we set selection criteria in the "Select" menu to identify highly connected nodes as core targets. Finally, we utilized these settings to generate the visualized network diagram, which effectively illustrates the relationships and key nodes within the network.

Gene function and pathway enrichment analysis of target proteins

Input the core target genes filtered from the protein–protein interaction (PPI) network diagram into the DAVID database (https://david.ncifcrf.gov/) for GO and KEGG enrichment analysis. Set $P < 0.05$ as the significance threshold. GO enrichment analysis includes biological process (BP), cellular component (CC), and molecular function (MF). Use the bioinformatics platform (http://www.bioinformatics.com.cn/) to visualize the potentially relevant enriched pathways.

Molecular docking

Use the free open-source software Avogadro to design the structure of clopidogrel and perform energy minimization and geometry optimization. The final determined structure is saved in.pdb format to facilitate subsequent molecular docking preparation. The crystal structure of the core protein is obtained from the Structural Bioinformatics Protein Database (PDB) (http://www.rcsb.org) and saved in.pdb format, and potential binding sites are screened and determined using the DeepSite database (https://open.playmolecule.org/tools/deepsite).

Use AutoDockTools 1.5.6 software to perform the following processing on the protein: First, add the PDB files of the receptor protein and small molecule ligand. Then, introduce hydrogen atoms and calculate Gasteiger charges, saving the results as.pdbqt files. The size of the docking box was set to 40, and the docking binding position was adjusted accordingly. Specify that all flexible bonds of the small molecule ligand as rotatable and set the receptor protein for rigid docking. Obtain docking results by executing AutoGrid4 and AutoDock4, and export the binding energy. Finally, use PyMOL software to export the protein–ligand complex and store it as a.pdb file for subsequent visualization of the results (Fig. 1).

Results and discussion

Intersection targets of clopidogrel and carotid artery stenosis

After calibration and deduplication in the GeneCards, SwissTargetPrediction, and ChEMBL databases, 250 targets related to clopidogrel were screened. From the GeneCards database, 2500 target genes related to carotid artery stenosis were screened. As shown in the Venn diagram, 127 intersecting genes between clopidogrel and carotid artery stenosis were identified (Fig. 2).

Screening of PPI and core genes of potential targets

The initial PPI network was constructed using the STRING database (Fig. 3a). The downloaded PPI data were imported into Cytoscape 3.10.0 software, generating a new PPI network for visualization and analysis. Using the Centiscape 2.2 module in Cytoscape software, 25 core targets were screened and the corresponding PPI network was constructed. From light yellow to dark purple, the node colors indicate the increase in node degree within the PPI network (Fig. 3b). This network consists of 25 nodes and 208 edges, with node colors indicating the increase in node degree.

The six core targets screened are TNF, MMP9, PTGS2, CCL2, TLR4, and IL-10. The TNF gene (tumor necrosis factor gene) can promote inflammatory responses, increase the expression of adhesion molecules in endothelial cells,

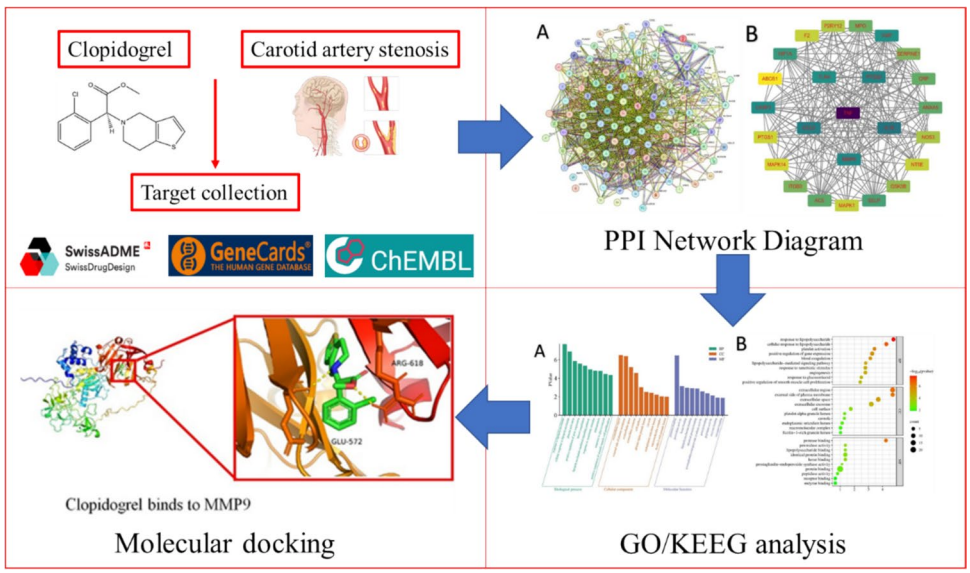

Fig. 1 Integrative workflow for this study

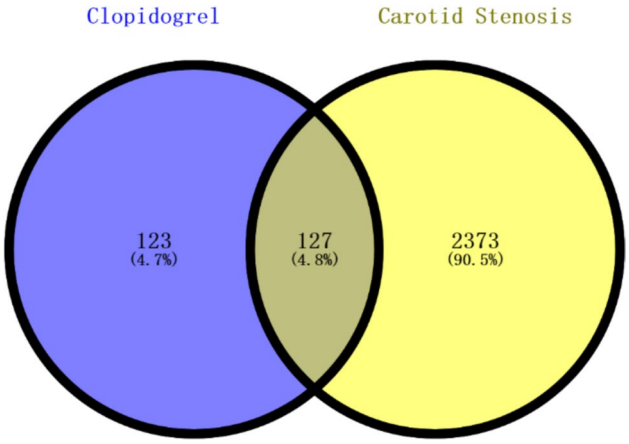

Fig. 2. 127 Potential Genes of Clopidogrel and Carotid Artery Stenosis. The blue section represents the targets associated with clopidogrel, while the yellow section represents the targets related to carotid artery stenosis. The overlapping area in the center indicates the intersecting targets shared between the two

and attract more monocytes into the vessel wall, leading to plaque formation and enlargement [15]. High levels of TNF-α are associated with vulnerable plaques (more likely to rupture, leading to thrombosis and embolic events). Studies show that serum TNF-α levels in patients with carotid artery stenosis may be higher than in those without stenosis, and TNF-α levels may correlate with the severity of carotid artery stenosis [16].

MMP9 is an enzyme that plays a role in extracellular matrix remodeling and can degrade various extracellular matrix components, including collagen and elastin. Studies show that MMP9 can degrade the extracellular matrix in plaques, making them more vulnerable and increasing the risk of rupture and thrombosis [17]. Increased MMP9 expression is associated with vascular inflammation, which is a key factor in the development of atherosclerosis [18]. MMP9 expression is positively correlated with the extent of neovascularization in carotid plaques and negatively correlated with plaque elasticity [19]. Therefore, MMP9 may play an important role in the occurrence, development, and complications of carotid artery stenosis and may serve as a potential therapeutic target or biomarker.

The PTGS2 gene encodes prostaglandin-endoperoxide synthase 2, also known as cyclooxygenase 2 (COX-2). PTGS2 is a key enzyme in the inflammatory process, promoting the synthesis of prostaglandins, which play a role in inflammation, pain, and fever responses [20]. In the development of atherosclerosis, upregulation of PTGS2 may be related to the inflammatory process in carotid artery stenosis [21]. In addition, PTGS2 may play a role in the formation and development of atherosclerotic plaques. PTGS2 is involved in the metabolism of lipids, collagen, and other extracellular matrix components in the vessel wall, which are related to plaque stability and vulnerability [21]. Therefore, inhibiting the activity of PTGS2 may help slow the progression of carotid artery stenosis.

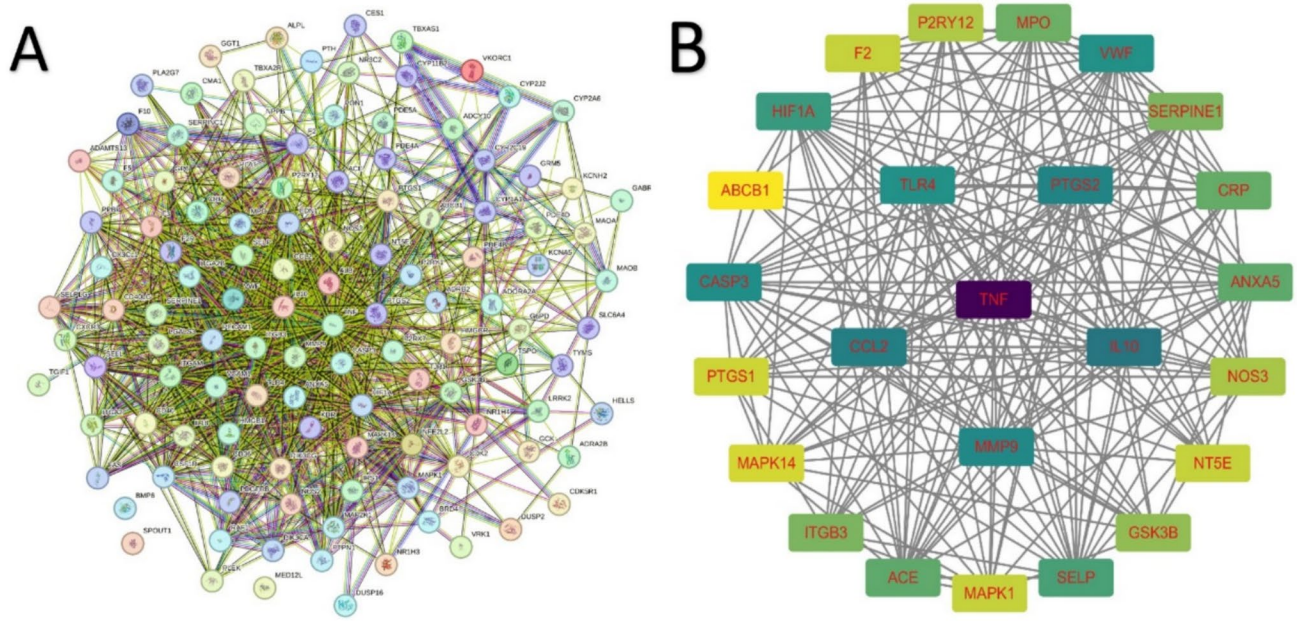

Fig. 3 PPI Network Diagram of Potential and Core Targets. Panel A shows the original PPI (protein–protein interaction) network. Panel B represents the core target network after screening. Each node corresponds to a gene, with darker colors indicating a higher number of interacting proteins. The five genes within the inner circle (TNF, MMP9, PTGS2, CCL2, TLR4, and IL-10) are identified as the most critical targets

CCL2, also known as Monocyte Chemoattractant Protein-1 (MCP-1), is a chemokine that plays a key role in inflammation and immune responses [22]. CCL2 can attract monocytes into the arterial wall, which may then differentiate into foam cells, a critical step in atherosclerotic plaque formation. Meanwhile, CCL2 exacerbates the inflammatory response in the vessel wall by promoting the migration and activation of inflammatory cells. Increased CCL2 expression is also associated with plaque instability and vulnerability, potentially increasing the risk of plaque rupture and thrombosis, thus worsening carotid artery stenosis [23]. Due to its role in the development of carotid artery stenosis, CCL2 may become a potential target for treating carotid artery stenosis. Inhibiting the activity of CCL2 or the signaling of its receptor CCR2 may help slow the progression of carotid artery stenosis.

TLR4, or Toll-Like Receptor 4, is a pattern recognition receptor that plays an important role in the immune system. The activation of TLR4 can promote inflammatory responses, and inflammation is a key factor in atherosclerosis [24]. In atherosclerotic plaques, the activation of TLR4 can increase the expression of inflammatory factors, such as tumor necrosis factor-α (TNF-α) and interleukin-6 (IL-6), which further promote plaque development and instability [25]. TLR4 may also participate in the occurrence and development of atherosclerosis by affecting the autophagy process. Autophagy is an intracellular cleaning mechanism, and the activation of TLR4 may affect this process, thereby influencing plaque stability [26].

IL-10 is an anti-inflammatory cytokine produced by Th2 cells, macrophages, and other immune cells, regulating immune responses and inhibiting inflammatory processes [27]. IL-10 exerts its anti-inflammatory effects by inhibiting the production of pro-inflammatory cytokines (such as TNF-α, IL-1β, and IL-6) and reducing immune cell activation [28, 29]. IL-10 exhibits anti-inflammatory properties that can attenuate endothelial cell damage and enhance the barrier function of endothelial cells. Endothelial cell damage is one of the critical factors in the development and progression of atherosclerosis. The protective effects of IL-10 on endothelial cells contribute to maintaining vascular integrity and reducing lipid infiltration and inflammatory cell adhesion. Changes in IL-10 levels can serve as a biomarker for assessing the inflammatory status and therapeutic efficacy in carotid artery stenosis. By monitoring serum IL-10 levels in patients with carotid artery stenosis, we can promptly evaluate the therapeutic response.

GO enrichment analysis of core targets

According to the GO functional enrichment analysis results of core targets for the effect of clopidogrel in treating carotid artery stenosis, there are 154 biological processes (BP), 17 cellular components (CC), and 23 molecular functions (MF). GO analysis was ranked based on false discovery rate (FDR) values and P-values, selecting the top 10 items with the lowest FDR values in BP, CC, and MF, and displayed through horizontal gradient bar charts and bubble charts (Fig. 4).

In the BP results, the main biological processes are closely related to the response to lipopolysaccharide (LPS). LPS can promote inflammatory responses by activating Toll-like receptors (such as TLR4), and inflammation is a key factor in atherosclerosis. Immune cells activated by LPS release various inflammatory factors, such as tumor necrosis factor-α (TNF-α) and interleukin-6 (IL-6), which further promote plaque development and instability [30]. LPS can also directly act on vascular endothelial cells, causing endothelial dysfunction, which is an important event in the early stages of atherosclerosis. LPS promotes the formation of atherosclerotic plaques by activating vascular endothelial cells and immune cells [31].

Additionally, platelet activation and angiogenesis are biological processes related to clopidogrel for treating carotid stenosis. Platelet activation is involved in multiple stages of atherosclerosis, including platelet adhesion, aggregation, and release reactions after endothelial injury, as well as the migration and proliferation of smooth muscle cells. Platelet parameters such as platelet count (PLT) and mean platelet volume (MPV) can reflect the quantity, size, and activation degree of platelets. These parameters are related to carotid artery stenosis and may serve as indicators for assessing the risk of carotid atherosclerosis [32]. Angiogenesis within atherosclerotic plaques may affect plaque stability. Newly formed vessels may be more prone to rupture, leading to the entry of plaque contents into the bloodstream, increasing the risk of thrombosis and embolic events. Angiogenesis-related factors, such as vascular endothelial growth factor (VEGF), may serve as biomarkers for the progression of carotid artery stenosis and patient prognosis [33].

The CC results indicate that clopidogrel for treating carotid stenosis may act on the extracellular region, plasma membrane external side, extracellular space, and extracellular exosomes. The MF results suggest that the molecular processes of clopidogrel for treating carotid stenosis may include protease binding, peroxidase activity, heme binding, and lipopolysaccharide binding.

KEGG pathway enrichment analysis of core targets

To explore the potential signaling pathways of clopidogrel in the treatment of carotid artery stenosis, core target genes were input into the DAVID database for KEGG pathway analysis. A total of 86 typical signaling pathways were enriched, and the top 20 pathways with the lowest false discovery rate (FDR) values were selected, with biological

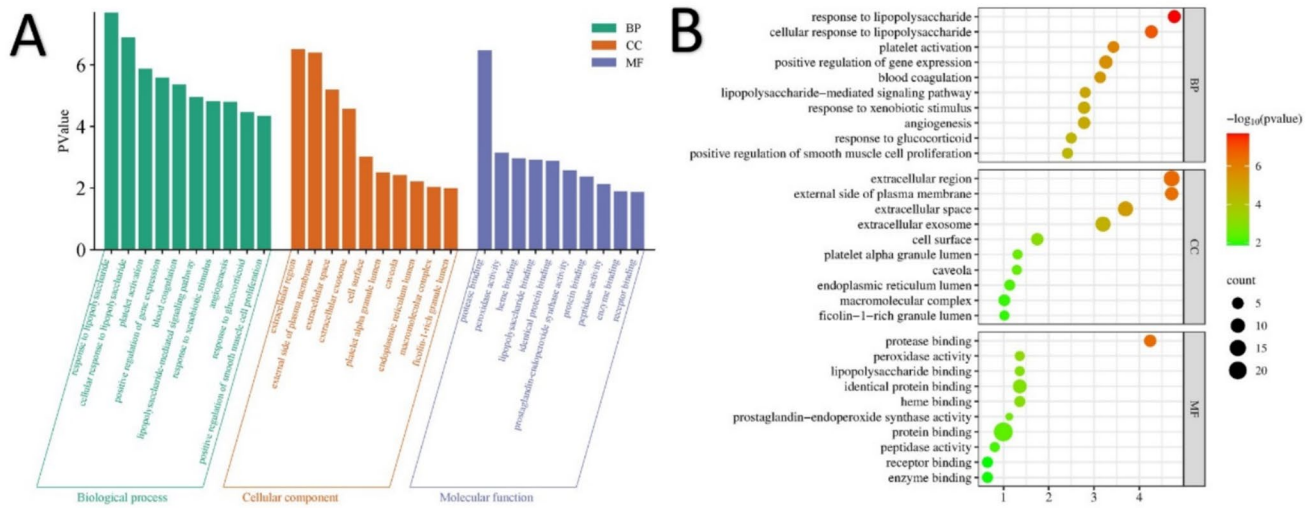

Fig. 4 GO Enrichment Analysis Results of Core Targets: Horizontal Gradient Bar Chart and Bubble Chart. **a** This histogram illustrated the top 10 enriched entries for each GO category (BP, CC, and MF) with smaller FDR values on the 127 potential targets. The FDR values reflected the statistical significance of the enrichment, with smaller values indicating higher significance. The height of each bar corresponds to the gene counts, reflecting the degree of enrichment within the respective category. These enriched entries highlighted key biological processes, cellular components, and molecular functions that are potentially influenced by clopidogrel exposure. **b** The size of each bubble corresponded gene expressions in a particular pathway. The enrichment significance was shown by the color saturation of the bubble

pathways, horizontal gradient bar charts, and bubble charts drawn (Fig. 5).

KEGG analysis results show that the signaling pathways of clopidogrel for treating carotid stenosis are most likely closely related to lipid and atherosclerosis. This result is consistent with previous studies. Abnormal lipid metabolism is a major risk factor for atherosclerosis. Atherosclerosis is characterized by intimal lesions of the affected arteries, accumulation of lipids and complex carbohydrates, fibrous tissue proliferation, and calcium deposition forming plaques [34]. Accumulated low-density lipoprotein (LDL) and very low-density lipoprotein (VLDL) are oxidized or chemically modified to form oxidized low-density lipoprotein (ox-LDL). These modified lipoproteins induce inflammatory responses, recruit macrophages to engulf lipids, form foam cells, and promote the development of atherosclerosis [35, 36].

The main cause of carotid artery stenosis is atherosclerosis, accounting for more than 90% [37]. Atherosclerotic plaques involve the carotid artery, leading to arterial stenosis or even occlusion, causing cerebral ischemia and stroke symptoms. Studies show that pericarotid fat density is positively correlated with the degree of carotid artery stenosis; patients with symptomatic carotid stenosis and recurrent stenosis-related ischemic cerebrovascular events have higher pericarotid fat density [38].

In summary, abnormal lipid metabolism plays a central role in the occurrence and development of atherosclerosis, which is the main cause of carotid artery stenosis. Controlling blood lipid levels and improving lipid metabolism are crucial for preventing and treating atherosclerosis and carotid artery stenosis.

Molecular docking results

The interactions between clopidogrel and six core target genes (TNF, MMP9, PTGS2, CCL2, TLR4, IL-10) were studied through molecular docking analysis. Molecular docking was completed using AutoDock software, and the results showed that the binding energies were all less than −4.20 kcal/mol (see Table 1), indicating that clopidogrel can tightly bind to the core target genes. This result demonstrates the strong affinity between clopidogrel and these target genes, further confirming the important role of clopidogrel in the molecular mechanism of treating carotid artery stenosis [39]. Finally, PyMOL software was used to visually display the binding details of clopidogrel with the core targets (Fig. 6).

Conclusion

Carotid artery stenosis is a common vascular disease in clinical practice, and although clopidogrel is commonly used for treatment, its pharmacological mechanism is not yet clear. Based on network pharmacology, this study screened 127 intersecting targets and identified 25 core targets. GO and KEGG enrichment analyses of these 25 core targets revealed that clopidogrel may improve atherosclerosis symptoms and

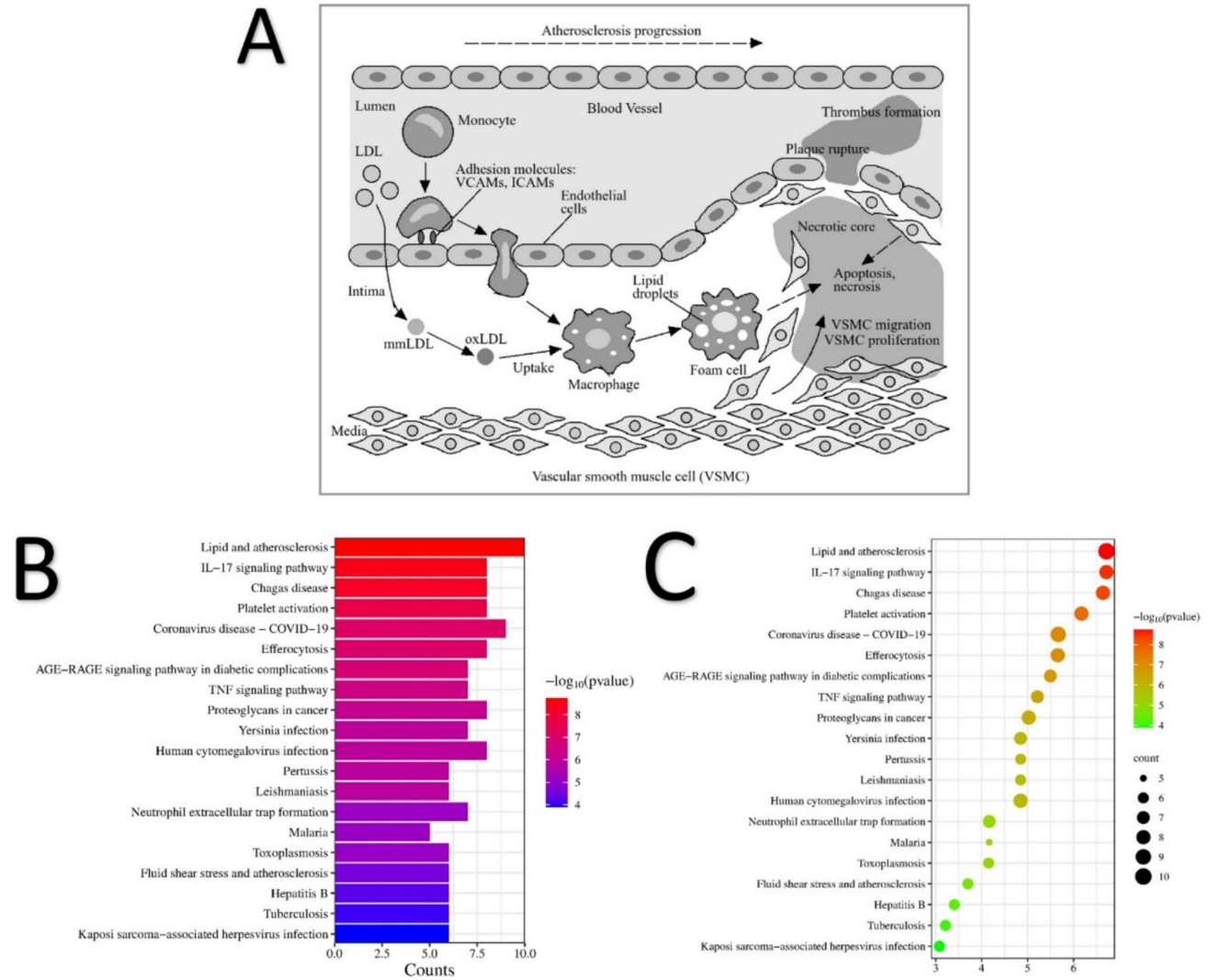

Fig. 5 KEGG Enrichment Analysis Results of Core Targets: Biological Pathways, Horizontal Gradient Bar Chart, and Bubble Chart. **a** Biological schematic diagram of atherosclerosis progression. **b** The bubble diagram visualized the top 20 enriched KEGG signal pathways in reverse order of FDR values. Each bubble represented a specific pathway, where the bubble area indicating the number of enriched genes in the pathway. The intensity of the bubble's color indicated the significance of the enrichment, with darker shades of red representing higher significance. The 127 intersection genes were mainly involved in KEGG pathways, with a notable emphasis on neuroactive ligand–receptor interactions and signaling pathways. **c** The histogram illustrated the frequency and significance of enrichment for each pathway. The length of each bar corresponded to the gene counts, indicating enrichment score and the level of significance, with taller bars representing larger counts and higher enrichment. It presented a concise and visually informative depiction of the top 20 enriched KEGG signal pathways, emphasizing the pathways that were particularly relevant to clopidogrel for treating carotid stenosis

Table 1 Binding energy of clopidogrel with six core target genes

UniProt ID	Gene name	Ligand	Binding energy (kcal/mol)
P13500	CCL2	Clopidogrel	−4.20
P14780	MMP9	Clopidogrel	−4.94
O00206	TLR4	Clopidogrel	−5.09
P35354	PTGS2	Clopidogrel	−5.32
P18893	IL-10	Clopidogrel	−5.72
P01375	TNF	Clopidogrel	−6.40

thus treat carotid artery stenosis by regulating inflammatory responses. Molecular docking technology showed that clopidogrel can tightly bind to the top 6 core targets, further proving that clopidogrel achieves therapeutic effects by regulating these targets. Although our study lacks clinical data validation, which to some extent limits further in-depth investigation, the findings regarding inflammation regulation are rarely reported in the current literature and can provide novel therapeutic strategies for future research. This study aims to provide a theoretical basis for the treatment of carotid artery stenosis with clopidogrel and to aid in

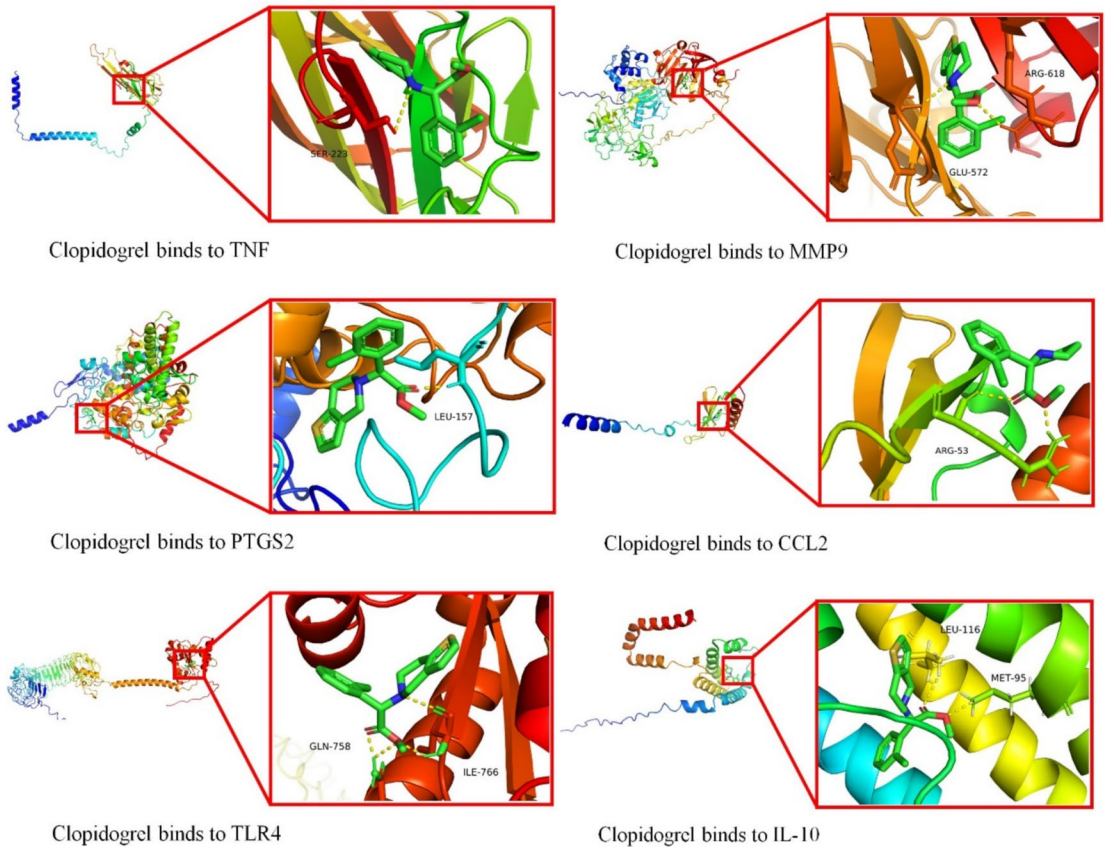

Fig. 6 Molecular docking results of clopidogrel with core targets

subsequent drug combination, new drug development, and exploration of pharmacological mechanisms.

Authors' contribution XW contributed to conceptualization, validation, formal analysis, methodology, data curation, software, visualization, writing–original draft, writing–review and editing. HL helped in supervision, methodology, writing–review and editing. JX contributed to conceptualization, validation, methodology, visualization, writing–review and editing. CZ contributed to conceptualization, supervision, resources, writing–review and editing.

Funding No external funding was used.

Data availability statement No datasets were generated or analyzed during the current study.

Declarations

Conflict of interest The authors declare no competing interests.

References

1. Ford MK, Cohn JR. Clopidogrel hypersensitivity: pathogenesis, presentation and diagnosis. Curr Vasc Pharmacol. 2019;17(2):110–2.
2. Paciaroni M, Bogousslavsky J. Clopidogrel for cerebrovascular prevention. Cerebrovasc Dis. 1999;9(5):253–60.
3. Pereira NL, Rihal CS, So DYF, et al. Clopidogrel pharmacogenetics. Circul Cardiovasc Intervent. 2019;12(4):e007811.
4. Gelbenegger G, Jilma B. Clinical pharmacology of antiplatelet drugs. Expert Rev Clin Pharmacol. 2022;15(10):1177–97.
5. Bakhru MR, Bhatt DL. Interpreting the charisma study: What is the role of dual antiplatelet therapy with clopidogrel and aspirin? Clevel Clin J Med. 2008;75(4):289–95.
6. Arasu R, Arasu A, Muller J. Carotid artery stenosis: an approach to its diagnosis and management. Aust J General Pract. 2021;50(11):821–5.
7. Heck D, Jost A. Carotid stenosis, stroke, and carotid artery revascularization. Prog Cardiovasc Dis. 2021;65:49–54.

8. Kremer C, Lorenzano S, Bejot Y, et al. Sex differences in outcome after carotid revascularization in symptomatic and asymptomatic carotid artery stenosis. J Vasc Surg. 2023;78(3):817–27.
9. Nogales C, Mamdouh ZM, List M, et al. Network pharmacology: curing causal mechanisms instead of treating symptoms. Trends Pharmacol Sci. 2022;43(2):136–50.
10. Noor F, Asif M, Ashfaq UA, et al. Machine learning for synergistic network pharmacology: a comprehensive overview. Brief Bioinform. 2023;24(3):1–27.
11. Zhang P, Zhang D, Zhou W, et al. Network pharmacology: towards the artificial intelligence-based precision traditional Chinese medicine. Brief Bioinform. 2024;25(1):1–12.
12. Pinzi L, Rastelli G. Molecular docking: shifting paradigms in drug discovery. Int J Mol Sci. 2019;20(18):4331.
13. Crampon K, Giorkallos A, Deldossi M, et al. Machine-learning methods for ligand–protein molecular docking. Drug Discov Today. 2022;27(1):151–64.
14. Szklarczyk D, Kirsch R, Koutrouli M, et al. The STRING database in 2023: protein–protein association networks and functional enrichment analyses for any sequenced genome of interest. Nucl Acids Res. 2023;51(1):638–46.
15. Linghui W, Lei X. Combined value of serum miR-124, TNF-α and IL-1β for vulnerable carotid plaque in acute cerebral infarction. J College Phys Surg Pak. 2020;30(4):385–8.
16. Yi L, Tang J, Shi C, et al. Pentraxin 3, TNF-α, and LDL-C are associated with carotid artery stenosis in patients with ischemic stroke. Front Neurol. 2020;10:1365.
17. Meteva D, Vinci R, Seppelt C, et al. Toll-like receptor 2, hyaluronan, and neutrophils play a key role in plaque erosion: the OPTICO-ACS study. Eur Heart J. 2023;44(38):3892–907.
18. Shih Y-C, Chen P-Y, Ko T-M, et al. MMP-9 deletion attenuates arteriovenous fistula neointima through reduced perioperative vascular inflammation. Int J Mol Sci. 2021;22(11):1–21.
19. Jiang X-B, Yuan W-S, Wang J-S, et al. Matrix metalloproteinase-9 expression in carotid atherosclerotic plaque and contrast-enhanced MRI in a swine model. J NeuroIntervent Surg. 2014;6(1):24–8.
20. Tanaka M, Shirakura K, Takayama Y, et al. Endothelial ROBO4 suppresses PTGS2/COX-2 expression and inflammatory diseases. Commun Biol. 2024;7(1):1–11.
21. Zhou Y, Zhou H, Hua L, et al. Verification of ferroptosis and pyroptosis and identification of PTGS2 as the hub gene in human coronary artery atherosclerosis. Free Radical Biol Med. 2021;171:55–68.
22. De Souza S, Rosario Claudio J, Sim J, et al. Interleukin-10 signaling in somatosensory neurons controls CCL2 release and inflammatory response. Brain Behav Immun. 2024;116:193–202.
23. Georgakis MK, Bernhagen J, Heitman LH, et al. Targeting the CCL2–CCR2 axis for atheroprotection. Eur Heart J. 2022;43(19):1799–808.
24. Zusso M, Lunardi V, Franceschini D, et al. Ciprofloxacin and levofloxacin attenuate microglia inflammatory response via TLR4/NF-kB pathway. J Neuroinflammat. 2019;16(1):1–12.
25. Li Z, Zou X, Lu R, et al. Arsenic trioxide alleviates atherosclerosis by inhibiting CD36-induced endocytosis and TLR4/NF-κB-induced inflammation in macrophage and ApoE-/- mice. Int Immunopharmacol. 2024;128: 111452.
26. Wu J, Liu S, Banerjee O, et al. Disturbed flow impairs MerTK-mediated efferocytosis in aortic endothelial cells during atherosclerosis. Theranostics. 2024;14(6):2427–41.
27. Nagata K, Nishiyama C. IL-10 in mast cell-mediated immune responses: anti-inflammatory and proinflammatory roles. Int J Mol Sci. 2021;22(9):4972.
28. Xuan Y, Wang L, Zhi H, et al. Association between 3 IL-10 gene polymorphisms and cardiovascular disease risk. Medicine. 2016;95(6): e2846.
29. Xu S, Zhang J, Liu J, et al. The role of interleukin-10 family members in cardiovascular diseases. Int Immunopharmacol. 2021;94: 107475.
30. Ruder AV, Wetzels SMW, Temmerman L, et al. Monocyte heterogeneity in cardiovascular disease. Cardiovasc Res. 2023;119(11):2033–45.
31. Viafara-Garcia SM, Gualtero DF, Avila-Ceballos D, et al. Eikenella corrodens lipopolysaccharide stimulates the pro-atherosclerotic response in human coronary artery endothelial cells and monocyte adhesion. Eur J Oral Sci. 2018;126(6):476–84.
32. Zhu N, Shu H, Jiang W, et al. Mean platelet volume and mean platelet volume/platelet count ratio in nonvalvular atrial fibrillation stroke and large artery atherosclerosis stroke. Medicine. 2020;99(28): e21044.
33. Binod Kumar Y, Renu Y, Hyuk C, et al. Genetic polymorphisms rs699947, rs1570360, and rs3025039 on the VEGF gene are correlated with extracranial internal carotid artery stenosis and ischemic stroke. Ann Clin Lab Sci. 2017;47(2):144–55.
34. Bäck M, Yurdagul A, Tabas I, et al. Inflammation and its resolution in atherosclerosis: mediators and therapeutic opportunities. Nat Rev Cardiol. 2019;16(7):1–38.
35. Neels JG, Leftheriotis G, Chinetti G. Atherosclerosis calcification: focus on lipoproteins. Metabolites. 2023;13:3.
36. Baratta F, Cocomello N, Coronati M, et al. Cholesterol remnants, triglyceride-rich lipoproteins and cardiovascular risk. Int J Mol Sci. 2023;24(5):4268.
37. Bonati LH, Jansen O, de Borst GJ, et al. Management of atherosclerotic extracranial carotid artery stenosis. Lancet Neurol. 2022;21(3):273–83.
38. Yu M, Meng Y, Zhang H, et al. Associations between pericarotid fat density and image-based risk characteristics of carotid plaque. Eur J Radiol. 2022;153:110364.
39. Agu PC, Afiukwa CA, Orji OU, et al. Molecular docking as a tool for the discovery of molecular targets of nutraceuticals in diseases management. Sci Rep. 2023;13(1):13398.