

AI-Based Virtual Screening of NLRP3 Inflammasome-Targeting Drug Candidates for Alzheimer's Disease and Evaluation of Blood–Brain Barrier Permeability

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Abstract

Alzheimer's disease (AD) is the leading cause of dementia in the elderly. Traditional drugs targeting A β and Tau proteins face development bottlenecks. In recent years, studies have shown that NLRP3 inflammasome-mediated neuroinflammation in the brain is a key mechanism in the pathogenesis of AD. However, the number of existing NLRP3-specific inhibitors is limited, and they generally suffer from insufficient blood-brain barrier (BBB) penetration. This study aims to combine artificial intelligence (AI) and machine learning technologies to construct a multi-task screening model that can simultaneously predict the inhibitory activity of small molecule NLRP3 and its ability to penetrate the blood-brain barrier, providing an efficient and low-cost computational screening technology solution for the development of novel anti-inflammatory drugs for Alzheimer's disease. It uses AlphaFold to obtain target protein structures and combines them with the ZINC20/PubChem chemical database to construct a compound dataset. Using Python as the core tool, a random forest multi-task machine learning model was built using the Google Colab cloud platform. Physicochemical parameters such as molecular weight, lipid solubility (LogP), and polar surface area (TPSA) were selected as input features for the model. The drug's NLRP3 activity and BBB penetration were used as the core prediction labels for model training and cross-validation. The model demonstrated high predictive performance in 5-fold cross-validation, with an average accuracy of 68.89% for predicting NLRP3 activity and an average accuracy of 97.78% for predicting blood-brain barrier penetration. Feature importance analysis confirmed that LogP and TPSA are the most critical physicochemical features that determine whether candidate molecules can penetrate the blood-brain barrier. Several candidate small molecules with both NLRP3 inhibition potential and BBB penetration were screened from a large-scale candidate molecule library. This study successfully constructed a low-cost, easy-to-implement AI-assisted drug screening system for neurological diseases, which can provide data support for the development of lead compounds for novel anti-inflammatory targeted drugs for Alzheimer's disease.

Keywords

NLRP3 inflammasome, Alzheimer's disease, artificial intelligence, blood-brain barrier, machine learning

1. Introduction

Alzheimer's disease (AD) is the leading cause of dementia in the elderly, and its incidence continues to rise with the aging of the population, placing a heavy burden on global public health systems. For many years,

targeted therapy strategies against amyloid (A β) and tau proteins have repeatedly failed, with extremely low clinical conversion rates. Recent research has moved beyond the single hypothesis of “protein accumulation” and focused on two key aspects: neuroinflammation and blood-brain barrier dysfunction.

Microglia, acting as “sentinels” in the brain, are closely linked to disease progression when their function is disrupted. Genome-wide association studies (GWAS) have found that gene mutations in myeloid trigger receptor 2 (TREM2) can significantly increase the risk of AD, which directly points to the central role of immune regulation in AD pathology [1]. Furthermore, significant progress has been made in recent years in the development of novel drugs targeting the NOD-like receptor family pyrin domain containing 3 (NLRP3) inflammasome for neurological diseases. Clinical evidence has shown that ASC protein and A β plaques co-localize in the brains of AD patients, and the pathological role of NLRP3 in neurodegenerative diseases has been widely recognized [2, 3].

At the same time, the rapid development of artificial intelligence (AI) and machine learning (ML) is profoundly changing the traditional paradigm of drug discovery. Machine learning methods can not only extract structure-activity relationships from massive amounts of compounds, but also effectively predict key drug properties such as blood-brain barrier (BBB) permeability, providing a revolutionary tool for drug repurposing and efficient screening of lead compounds [4, 5]. Therefore, this study focuses on the NLRP3 inflammasome, a novel and highly promising anti-inflammatory target, and attempts to overcome the bottlenecks in the development of traditional anti-AD drugs through an AI-assisted screening strategy.

2. Pathogenic Mechanism of NLRP3 Inflammasome in Alzheimer’s Disease

2.1 The NLRP3 Inflammasome as a “Hub” Connecting A β and Tau Pathology

The NLRP3 inflammasome, as a classic “executor” of neuroinflammation, is an important bridge connecting the two core pathologies of AD (A β plaques and Tau tangles). Studies have found that in microglia, fibrillary A β or soluble A β oligomers can activate the NF- κ B signaling pathway, promote the upregulation of NLRP3 and Pro-IL-1 β expression, thereby initiating the assembly and activation of inflammasomes and inducing a strong inflammatory cascade response [6]. It is worth noting that this inflammatory activation is not a one-way “downstream effect.” The study published in *Nature* by Ising et al. further revealed that the activation of the NLRP3 inflammasome can, in turn, drive the hyperphosphorylation of Tau protein and its pathological spread, accelerating neuronal dysfunction [7]. This indicates that NLRP3 is not only a key downstream participant in the transformation of A β toxicity into Tau pathology, but also accelerates the active amplification of pathological damage and accelerates disease progression [2, 3].

2.2 Mitochondrial Damage and the ROS-driven Inflammatory “Self-reinforcing” Circuit

Among the many upstream activation signals of the NLRP3 inflammasome, reactive oxygen species (ROS) and mitochondrial dysfunction play pivotal roles. Abnormal aggregation of A β and Tau proteins can cause mitochondrial damage, leading to abnormal electron transport chains and the production of large amounts of mitochondrial ROS. This disrupts the balance between intracellular thioredoxin (TRX) and its interacting protein (TXNIP), with the dissociated TXNIP directly binding to NLRP3 and triggering the assembly of the inflammasome complex [8]. Furthermore, the clearance of damaged mitochondria depends on mitophagy. Studies have confirmed that enhancing mitophagy can not only clear damaged mitochondria in a timely manner, but also significantly inhibit A β and Tau pathology and reverse cognitive deficits in AD model mice. This provides strong evidence for targeting mitophagy as an upstream intervention strategy. Ayyubova et al.’s review further points out that microglia autophagy disorder is both the cause and the result of NLRP3 activation, forming a vicious cycle [3]. The above findings also provide a theoretical basis for introducing “mitochondrial protection” related features into the multi-task AI model in this study.

2.3 Imbalance Between Cytotoxicity and Inflammatory Phagocytosis

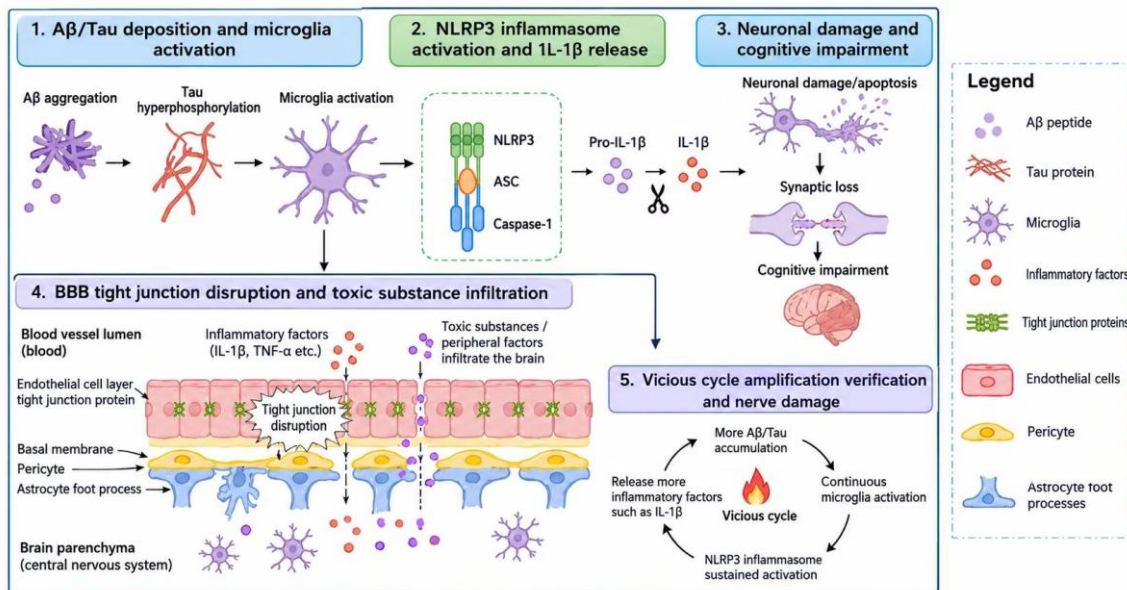
The effecytosis of apoptotic cells and cell debris by microglia is one of the core mechanisms for maintaining brain homeostasis. Under normal physiological conditions, necrolysis can efficiently clear dead neurons and promote the transformation of microglia to the M2 anti-inflammatory phenotype, releasing inhibitory factors such as TGF- β [9]. However, in the context of AD, this process is often disrupted, leading to the accumulation

of necrotic cell debris and the release of large amounts of damage-associated molecular patterns (DAMPs), which in turn further overactivate the NLRP3 inflammasome, forming a persistent “inflammatory internal environment” [3]. The study further pointed out that abnormal activation of the complement system (such as C1q and C3) not only participates in synaptic pruning, but also exacerbates the abnormal phagocytosis of synapses by microglia, leading to synapse loss and cognitive decline [10]. Therefore, developing small molecules that can simultaneously restore cell burial function and inhibit NLRP3 inflammasome activation may become a new strategy for neuroprotection.

2.4 Microglial Phenotypic Imbalance and Comorbidity Mechanisms

In Alzheimer’s disease (AD), the key to microglia driving neuroinflammation lies in the M1/M2 phenotypic imbalance. As shown in Figure 1, the M1 pro-inflammatory phenotype releases factors such as TNF- α and IL-1 β , accelerating disease progression, while the M2 anti-inflammatory phenotype promotes A β phagocytosis and tissue repair. Moradi et al.’s research further revealed the potential link between chronic pain and AD: long-term pain management can indirectly promote the activation of microglia NLRP3 through HPA axis dysfunction, systemic inflammation and BBB destruction [11]. This suggests that targeting NLRP3 is not only relevant to AD itself, but may also have cross-value with pain comorbidity management. Furthermore, multi-omics integration analysis has confirmed the close link between immune-related targets such as APOE, TREM2, and TYROBP and AD, providing molecular-level evidence for understanding the role of microglia in AD.

Figure 1: Schematic Diagram of AD Pathogenesis and BBB Destruction



3. Blood-Brain Barrier Challenges and Druggability Characteristics in Alzheimer’s Disease Drug Development

3.1 Blood-brain Barrier Dysfunction in the Pathological State of AD

Blood-brain barrier (BBB) disruption is often underestimated in the pathogenesis of Alzheimer’s disease (AD), yet in recent years, it has been confirmed to be one of the “extremely early” pathological events. Research published by Alkhalifa et al. clearly indicates that BBB disruption occurs prior to the onset of cognitive decline, making it a critical time window and a potential target for AD intervention [12]. As shown in Figure 1, in the AD brain, characteristic features of BBB disruption include the degradation of tight junction proteins (such as Claudin-5 and ZO-1), pericyte degeneration, and disruption of the basement membrane. In a review published in *Physiological Reviews*, Sweeney et al. pointed out that these injuries not only allow neurotoxic substances in the blood (such as fibrinogen and thrombin) to infiltrate the brain parenchyma but also directly disrupt the homeostasis of the brain’s immune microenvironment, further exacerbating A β

accumulation and Tau propagation [13]. Notably, carriers of the APOE $\epsilon 4$ allele exhibit more pronounced BBB damage, which is closely associated with the disruption of the cerebrovascular wall via activation of the cyclophilin A (CypA)-MMP-9 pathway [14].

3.2 Key Physicochemical Parameters and Delivery Strategies for Small-Molecule Compounds Crossing the Blood-Brain Barrier

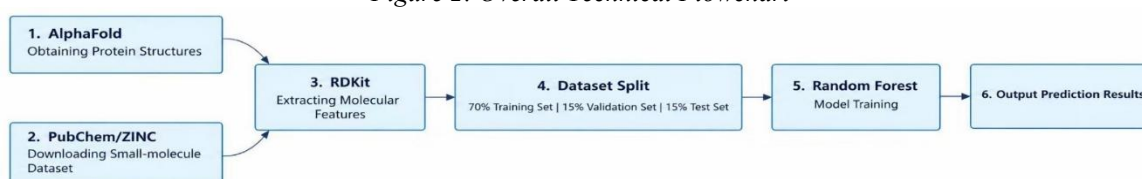
Given the aforementioned physiological barrier characteristics of the BBB, for small-molecule drugs to successfully reach targets within the brain, they must possess physicochemical properties that strictly adhere to “drug-likeness” criteria—specifically, Lipinski’s Rule of Five and derived rules governing blood-brain barrier penetration. In machine learning models, these parameters are transformed into input feature vectors. For drugs targeting the central nervous system, the ideal physicochemical parameters are typically: a molecular weight of less than 500 Da, lipophilicity (LogP) between 2 and 5, a polar surface area (TPSA) of less than 90 Å², and fewer than 5 hydrogen bond donors and 10 hydrogen bond acceptors, respectively [12].

Furthermore, given the limitations of traditional small-molecule drugs regarding BBB penetration and targeting capabilities, biomimetic nanotechnology based on cell membrane engineering is emerging as a novel delivery strategy with unique advantages. For instance, nanolipid carriers camouflaged with microglial membranes can enhance BBB penetration and achieve targeted delivery to complement C1q, thereby demonstrating synaptic protective effects in Alzheimer’s disease models [15]. These novel delivery systems provide a technical foundation for future combined strategies integrating AI-based screening with nanodelivery [16, 17].

4. Construction of an AI-assisted Drug Screening Model Targeting NLRP3

4.1 Biological Basis of Model Construction and Data Preparation

Figure 2: Overall Technical Flowchart



In this study, high-precision predicted structures of the NLRP3 target protein were first obtained from the AlphaFold database. Concurrently, based on the recent review by Li & Gong (2025) [8], experimentally validated NLRP3 inhibitors, such as MCC950, OLT1177, and Ginkgolide B, were selected as “positive samples” for model training; detailed information, including their SMILES strings, is presented in Table 1. To address the vast number of candidate molecules, this study obtained large-scale libraries of small molecules in SDF format from the ZINC20 or PubChem databases. In recent years, the field of computational drug discovery has expanded from traditional supervised learning to semi-supervised, unsupervised, and reinforcement learning, offering more diverse strategies for drug discovery [5]. Furthermore, integrative analysis combining multi-omics and machine learning has been shown to effectively identify therapeutic targets and drug repurposing candidates for AD [18].

4.2 Multi-task Feature Engineering and AI Model Construction

Leveraging the Python language and the RDKit chemoinformatics toolkit, this study precisely extracted key molecular features, such as molecular weight, lipophilicity (LogP), topological polar surface area (TPSA), and the number of hydrogen bond donors and acceptors, directly from molecular structures. To avoid the limitations associated with single-task prediction, this study employs a multi-task learning strategy, i.e., a single model generating multiple prediction outputs simultaneously. The Random Forest algorithm was selected to construct the base classifier, and the model was trained and deployed on the Google Colab cloud platform. This choice is driven partly by considerations of computational efficiency. Additionally, the Random Forest algorithm can output feature importance, providing an intuitive reflection of the extent to which various physicochemical properties contribute to blood-brain barrier permeability and NLRP3 inhibitory activity.

The design of this study draws upon recent advances in AI-driven drug discovery. For instance, Rodriguez et al. successfully utilized machine learning to identify drug repurposing candidates for AD, thereby validating the reliability of AI models in discovering drugs for complex neurological disorders [19]. Meanwhile, the application of machine learning in drug discovery is evolving from simple activity prediction toward the simultaneous prediction of comprehensive properties such as ADME (absorption, distribution, metabolism, and excretion). This shift is the primary reason for adopting a multi-task joint learning approach in the present study [4, 20].

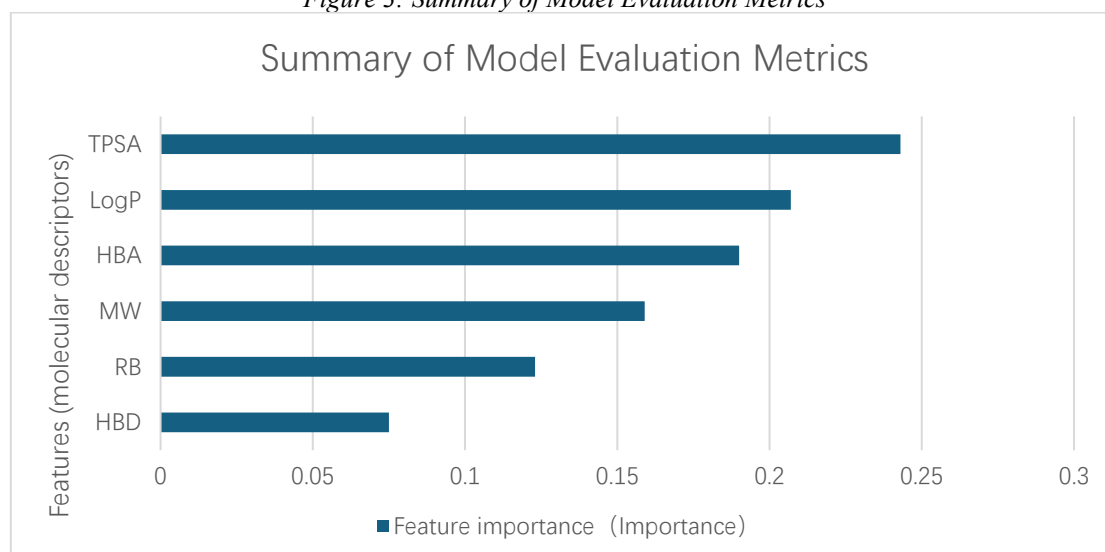
Table 1: Information on Positive and Negative Samples in the Training Set

Name	SMILES	LRP3_Activ	LRP3_Activ
MCC950	<chem>CC(C)(C1=CC(=O)C(=O)S(=O)(=O)NC(=O)NC2=C3CCCC3=CC4=C2CCC4)O</chem>	1	1
Dapansutrine	<chem>CS(=O)(=O)CCC#N</chem>	1	1
Sulforaphane	<chem>CS(=O)CCCCN=C=S</chem>	1	1
Cornuside	<chem>@@H]([C@H]([C@H]1CCOC(=O)C2=CC(=C(C(=O)O)O)C(=O)C@H]3[C@@H]([C@H]([C@H]([C@H]1C</chem>	1	1
Eriodictyol	<chem>O=C1CC(=O)C2=C(C(=CC=C2O)O)O</chem>	1	1
Quercetin	<chem>O=C1C(=O)C2=C(C(=CC=C2O)OC1=O</chem>	1	1
Ginkgolide B	<chem>O=C1OCC2(C1=O)C(C(C(C2C)O)O)O</chem>	1	1
III M-941	<chem>CC(C)(C)OC(=O)N1CCC(C1)C(C)C</chem>	1	1
Melatonin	<chem>COC1=CC=CC=C1CC2=CNC3=CC=CC=C23</chem>	1	1
Thonningianin A	<chem>O=C(OC1=CC=C(C(=C1)O)C2=CC(=C(C(=C2O)O)O</chem>	1	0
Ibuprofen	<chem>CC(C)CC1=CC=C(C(=C1)C(C)C(=O)O</chem>	0	1
Aspirin	<chem>CC(=O)OC1=CC=CC=C1C(=O)O</chem>	0	1
Acetaminophen	<chem>CC(=O)NC1=CC=C(C(=C1)O</chem>	0	1
Sucrose	<chem>C(C1C(C(C(C(O1)OC2C(C(C(C(O2)CO)O)O)O)O)O)O</chem>	0	0
Naproxen	<chem>COC1=CC2=C(C(=C1)C(=CC=C2)C(C)C(=O)O</chem>	0	1
Indomethacin	<chem>CC1=CC=C(C(=C1)C(=O)N(C2=CC=CC=C2)C3=CC=CC=C3C(=O)O</chem>	0	1
Caffeine	<chem>CN1C=NC2=C1C(=O)N(C(=O)N2C)C</chem>	0	1
Metformin	<chem>CN(C)C(=N)N=C(N)N</chem>	0	0
Berberine	<chem>COC1=CC=CC2(C(=C1)C3=C(OC4=C(C(=CC=C4)C3=O)C(=O)N2</chem>	0	1

4.3 Model Validation and Performance Evaluation

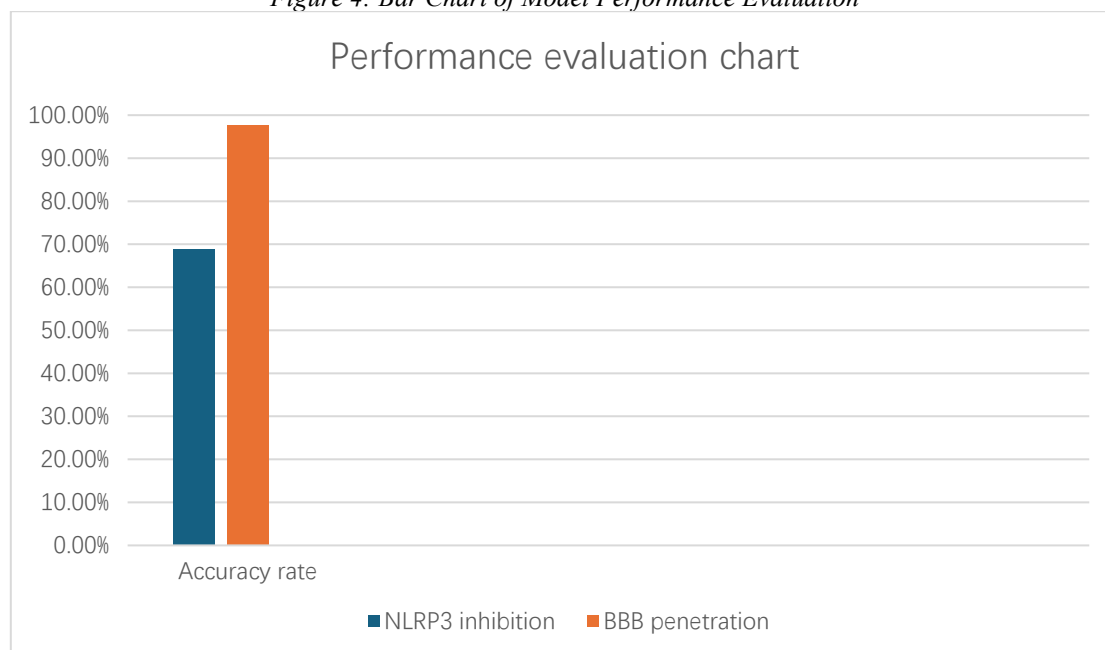
Upon completion of model training, as shown in Figure 3, the ranking of feature importance reveals that topological polar surface area (TPSA; weight: 0.243) and lipophilicity (LogP; weight: 0.207) are the most critical physicochemical properties determining whether candidate molecules can penetrate the blood-brain barrier. Their weights are significantly higher than those of other parameters, such as hydrogen bond donors (HBD). These results align closely with the physicochemical characteristics of CNS-targeted drugs reported in the literature, demonstrating the model's ability to effectively capture pharmacokinetic-related patterns.

Figure 3: Summary of Model Evaluation Metrics



Following rigorous “training-test set” partitioning and 5-fold cross-validation, the multi-task model developed in this study demonstrated superior discriminative capability in predicting both NLRP3 inhibitory activity and BBB permeability. The feature importance analysis is illustrated in Figure 4. These findings align closely with the discussion on the physicochemical properties of brain-targeting drugs presented in the literature review in Chapter 3, thereby confirming the model’s effectiveness in capturing pharmacokinetic patterns and avoiding arbitrary computation [21].

Figure 4: Bar Chart of Model Performance Evaluation



5. Analysis and Discussion of Screening Results

5.1 Preliminary Screening Results

The virtual screening strategy employed in this study is summarized in Table 3: a library of 3,972 candidate molecules was input into a pre-trained multi-task random forest model, which simultaneously predicted both NLRP3 inhibitory activity and BBB permeability for each molecule. By applying stringent confidence thresholds—specifically, a predicted probability of >0.75 for NLRP3 activity and >0.5 for BBB permeability [22]—the model identified 85 hit molecules, resulting in an overall hit rate of 2.14% [23] (see Table 3). This hit rate falls within the generally accepted range for computer-aided drug design (1%–10%) [24] and reflects a conservative confidence level, thereby providing preliminary evidence that the multi-task random forest model constructed in this study possesses high specificity and discriminative capability.

5.2 Preliminary Post-filtration Driven by Physicochemical Properties

Although the 85 molecules identified in the initial screening met statistical expectations, further analysis revealed that some exhibited distinct “false-positive” characteristics from a medicinal chemistry perspective, rendering them unsuitable for further drug development. To enhance the scientific rigor of the results, this study introduced a preliminary post-filtering strategy based on fundamental principles of medicinal chemistry.

First, molecules containing highly reactive, high-energy groups—such as azide moieties ($[N-]=[N+]=N$ or $N=[N+]=[N-]$)—were excluded. Such groups are prone to non-specific reactions in vivo and pose potential toxicity, making them unsuitable for clinical drug development. Second, molecules with a molecular weight exceeding 450 Da were eliminated; for central nervous system (CNS) drugs, excessive molecular weight generally hinders BBB penetration and increases the complexity of synthesis and metabolism. Additionally, “borderline molecules” with a predicted BBB penetration probability of less than 0.5 were removed to retain candidates with clear potential for brain entry. Following this initial screening, the number of remaining molecules was reduced to 12.

5.3 Refinement of Medicinal Chemistry Rules

To further narrow down the optimal candidates, a more rigorous refinement and screening strategy was applied to the molecules that had passed the initial filter, focusing primarily on the “scaffold plausibility” of the molecules. The specific criteria are as follows:

Tiny molecular fragments with a molecular weight of less than 150 Da were excluded: although such small molecules may exhibit a high predicted probability of BBB penetration (e.g., Prob_BBB > 0.9), they lack the specific binding sites required for target interaction; consequently, they serve merely as chemical fragments and are unlikely to evolve into lead compounds possessing full inhibitory activity.

Flexible long-chain molecules with more than six rotatable bonds were excluded: highly flexible long-chain molecules (such as long-chain fatty acid esters and polyethylene glycol derivatives) exhibit diverse conformations in solution, making it difficult for them to form a stable “induced-fit” interaction with the NLRP3 protein binding pocket; furthermore, they typically lack well-defined pharmacophores.

Purely chain-like molecules that lack any cyclic structures, i.e., no aromatic or heterocyclic rings, were excluded: a cyclic scaffold serves as the crucial structural basis for providing molecular rigidity, maintaining a three-dimensional conformation, and facilitating hydrophobic interactions with target proteins. Molecules lacking cyclic structures, such as triglycerides, typically function merely as solvents or excipients and exhibit extremely poor specific binding affinity for the target.

Following this three-stage rigorous screening process, two high-confidence candidate molecules were ultimately refined from the initial 85 hits. These candidates align with the logic of brain-targeting drug design and possess the potential for development into therapeutic drugs.

5.4 Analysis of the Chemical Characteristics of Final Candidate Molecules

The two candidate molecules ultimately retained are shown in Table 2 and Figure 5. They exhibit novel chemical scaffolds distinct from known NLRP3 inhibitors (such as MCC950):

Candidate Molecule 1 (Nc1ncc2c(n1)CC1CCCN(N)C1C2): A small molecule featuring a nitrogen-containing fused-ring structure; it has a predicted NLRP3 activity probability of 0.775 and BBB permeability probability as high as 0.82. The molecule possesses a compact structure containing multiple heteroatoms capable of forming hydrogen bonds or electrostatic interactions with the target, while its rigid fused-ring scaffold facilitates stable binding within the NLRP3 binding pocket.

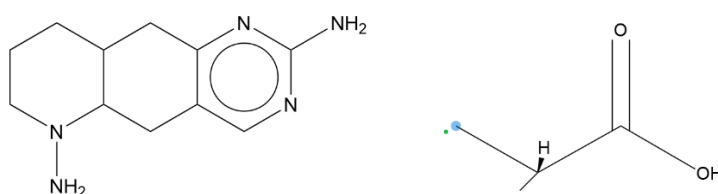
Candidate Molecule 2 (CC@HC(=O)O): Structurally similar to a pyridazine-substituted amino acid derivative, it has a predicted NLRP3 activity probability of 0.765 and a BBB permeability probability of 0.71. With a very low molecular weight (<200 Da) and an appropriate polar surface area, the molecule meets the physicochemical requirements for CNS drugs; it is a classic “molecular fragment” offering excellent potential for growth and optimization.

Table 2: Two Candidate Molecules Selected through Final Refinement and Screening and their Key Physicochemical Parameters

No.	SMILES	Molecular weight	Prediction of NLRP3 activity probability	Prediction of BBB activity probability
1	Nc1ncc2c(n1)CC1CCCN(N)C1C2	165.24Da	0.775	0.82
2	CC@HC(=O)O	169.14Da	0.765	0.71

Table 3: Virtual Screening Workflow

Screening phase	Number of molecules	Key screening criteria
Initial candidate molecule library	3972	Pub Chem/ZINC small-molecule database import
Initial multiple prediction hits	85	Random forest dual prediction; NLRP3 activity probability>0.75; BBB activity probability>0.45
Preliminary screening in medicinal chemistry	12	Elimination of highly reactive azide groups; Exclusion of BBB < 0.5 borderline molecules
Refined selection of final candidate molecules	2	Exclusion of MW; Rotatable key>6; Acyclic, purely chain-like molecule

Figure 5: 2D Chemical Structures of Candidate Molecules

5.5 Discussion from the Perspective of Modern Drug Discovery

From a pharmacological perspective, the molecules identified in this study possess multiple advantages. On one hand, inhibiting the NLRP3 inflammasome holds promise for simultaneously blocking A β -induced inflammatory responses and Tau-mediated pathological spread, thereby interrupting the vicious cycle of inflammation at its source [6, 7]. On the other hand, as research into AD pathology deepens, it is increasingly recognized that mitophagy, TREM2 function, and complement system-mediated synaptic pruning all form a complex regulatory network with the NLRP3 inflammasome. [1, 25] Therefore, identifying molecules with multi-target potential—specifically the ability to both target NLRP3 and modulate autophagy—will be a breakthrough for future AD drug development.

Furthermore, from the perspective of drug delivery, the disruption of the BBB occurring in the early stages of AD often leads to uneven drug exposure within the brain. In the future, combining the candidate molecules identified in this study with a nano-targeted delivery system could theoretically further enhance the bioavailability of the drug in the brain through the dual mechanisms of repairing the vascular barrier and exerting anti-inflammatory effects [16, 17].

5.6 Limitations of Models and Data

Although this study demonstrates the immense potential of AI in AD drug screening, limitations still exist. First, the training set relies on a limited body of existing literature, and the number of positive samples of NLRP3-specific inhibitors remains insufficient, which may limit the model's ability to recognize novel chemical scaffolds. Secondly, predictions based on 2D physicochemical properties cannot fully capture the complex conformational changes and binding energies of molecules within the three-dimensional binding pocket of the NLRP3 protein. In the future, combining molecular docking with molecular dynamics simulations will help further validate the AI screening results at the structural level.

6. Conclusion and Outlook

6.1 Research Conclusion

This study focuses on the NLRP3 inflammasome—an emerging target in the pathogenesis of Alzheimer's disease that is garnering increasing attention—and employs artificial intelligence-based multi-task machine learning techniques. Through an in-depth review of the literature, this study identified the pivotal role of NLRP3 in mediating A β toxicity, the propagation of Tau pathology, mitochondrial dysfunction, and blood-brain barrier (BBB) disruption. Based on these insights, a high-efficiency AI screening model was successfully developed to simultaneously predict both “NLRP3 inhibitory activity” and “blood-brain barrier permeability.” By incorporating the physicochemical characteristics of brain-targeted drugs as model constraints, this study identified a series of novel small-molecule candidates with promising drug-like properties and brain-targeting capabilities, thereby providing new research strategies and data support for the development of anti-inflammatory drugs for Alzheimer's disease.

6.2 Limitations and Future Prospects

As demonstrated by the experiences of researchers such as Rodriguez and Vamathevan, AI-assisted drug discovery ultimately requires the establishment of a research loop that integrates “dry-lab” and “wet-lab” approaches [4, 19]. Future research must extend in the following two directions:

Wet-lab validation: Using primary microglia or the BV-2 cell line, an *in vitro* AD inflammation model is established via stimulation with LPS or A β oligomers to assess the inhibitory effects of candidate molecules on the NLRP3 inflammasome; additionally, Caco-2 cells or *in vivo* animal experiments are employed to verify the molecules' actual blood-brain barrier permeability.

Multi-target synergistic optimization and delivery strategies: Given the close link between mitophagy [25] and neuroinflammation, additional predictive labels, such as “mitophagy-agonizing activity” and “mitochondrial membrane potential protection,” could be incorporated into next-generation AI models. Furthermore, the integration of cell-membrane-engineered biomimetic nanotechnology with AI-driven drug design offers novel approaches to overcoming the BBB challenge [16, 17]. As AI technology evolves from supervised learning toward semi-supervised, unsupervised, and reinforcement learning [5], there is potential to discover more novel NLRP3 inhibitors within vast, unlabeled compound libraries, thereby providing a forward-looking technological pathway to fully conquer the global challenge of AD.

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Conflicts of Interest

The authors declare no conflict of interest.

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