Integrative Network Pharmacology: Multilayer Omics, Bioinformatics, and Polypharmacological Drug Design

Mehta Vishwa, Mistri Jaymeen, Kanojia Kashvi, Chauhan Yash

L.J Institute of Pharmacy, Gujarat, India.

Corresponding author: Mehta Vishwa
Email: vishwamehta329@gmail.com
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Abstract

Network pharmacology represents a transformative shift from "one drug–one target" paradigm to a systems-level approach that integrates biological networks, omics data, and computational intelligence. This comprehensive review articulates the conceptual foundations, architecture, and practical applications of network pharmacology in drug discovery and translational medicine. By incorporating multilayer omics—genomics, transcriptomics, proteomics, metabolomics, and single-cell data—with bioinformatics tools and graph theory, researchers can delineate disease modules, predict polypharmacological interactions, and repurpose drugs with increased precision. The work discusses cutting-edge computational strategies including graph neural networks, similarity network fusion, and explainable AI for robust modeling and interpretation. Case studies in oncology, neurodegeneration, infectious diseases, autoimmunity, and ethnopharmacology underscore the method's potential in revealing novel mechanisms and multi-target therapeutics. Challenges such as incomplete interactomes, data heterogeneity, and model opacity are acknowledged, with solutions proposed through dynamic modeling, FAIR data practices, and ethical AI frameworks. Ultimately, network pharmacology emerges as a pivotal discipline enabling precision medicine through integrative, predictive, and mechanistically informed drug design.

Keywords: Network pharmacology, Multi-Omics Integration, Bioinformatics, Drug Repurposing,Polypharmacology, Computational Drug Discovery

Abbreviation	Full Form	Abbreviation	Full Form
AI	Artificial Intelligence	**Kd**	Dissociation Constant (binding affinity)
ADRs	Adverse Drug Reactions	**MTDL(s)**	Multi Target Directed Ligand(s)
ATAC-seq	Assay for Transposase-Accessible Chromatin using Sequencing	**ML**	Machine Learning
CMap	Connectivity Map	**MoA**	Mechanism of Action
CPTAC	Clinical Proteomic Tumor Analysis Consortium	**NDEx**	Network Data Exchange
CRISPR–Cas9	Clustered Regularly Interspaced Short Palindromic Repeats–CRISPR associated protein 9	**NMR**	Nuclear Magnetic Resonance
DEGs	Differentially Expressed Genes	**OMIM**	Online Mendelian Inheritance in Man
DTI(s)	Drug-Target Interaction(s)	**PPI**	Protein–Protein Interaction
DTN(s)	Drug–Target Network(s)	**RWR**	Random Walk with Restart
eQTL	Expression Quantitative Trait Loci	**scRNA-seq**	Single Cell RNA Sequencing
FAERS	FDA Adverse Event Reporting System	**SNF**	Similarity Network Fusion
GAN(s)	Generative Adversarial Network(s)	**TCGA**	The Cancer Genome Atlas
GC(s)N	Graph Convolutional Network(s)	**VAE(s)**	Variational Autoencoder(s)
GEO	Gene Expression Omnibus	**WGCNA**	Weighted Gene Co-Expression Network Analysis
GEM(s)	Genome-Scale Metabolic Model(s)	**XAI**	Explainable Artificial Intelligence
GTEx	Genotype-Tissue Expression		
GWAS	Genome-Wide Association Study/Studies	**MTDL(s)**	Multi Target Directed Ligand(s)
GNN(s)	Graph Neural Network(s)	**KEGG**	Kyoto Encyclopedia of Genes and Genomes

HMDB	Human Metabolome Database		

1. Introduction

Complex diseases such as cancer, diabetes, and neurodegenerative disorders often arise from dysregulation across multiple genes and pathways. Traditional "one drug–one target" models have proven inadequate, as over 90% of drug candidates fail during development—largely due to insufficient efficacy [2]. This high attrition underscores the limitations of simplified methods, which overlook pathway crosstalk, compensatory mechanisms, and patient variability [1] [2]. By considering medications as modulators of molecular networks, network pharmacology transforms disease as disruptions in these systems. [1] [3]. Originating from Hopkins' polypharmacology concept
[1] and further developed by Barabási's network medicine [3], this approach leverages network topology (e.g., hubs, modules) to reveal drug–target–disease relationships [3] [16]. For instance, analyses of traditional herbal medicines map phytochemicals onto protein–target networks, uncovering multi-target mechanisms [5] [18]. The rise of high-throughput omics has catalyzed practical advances in this field. Genomic, transcriptomic, proteomic, metabolomic, and epigenomic data now support the construction of context-specific interactomes and disease modules [4] [6]. Large-scale resources like TCGA [48] and GTEx [49] enable linkage of genetic variation to molecular and phenotypic outcomes. Integrative studies have, for example, associated somatic mutations with altered signaling networks in cancer [26] [28]. Concurrently, machine learning and graph-based AI enhance network pharmacology. Techniques such as graph convolutional networks (GCNs) can predict drug–target interactions, model polypharmacological effects, and forecast patient-specific responses by leveraging network features [7] [12].

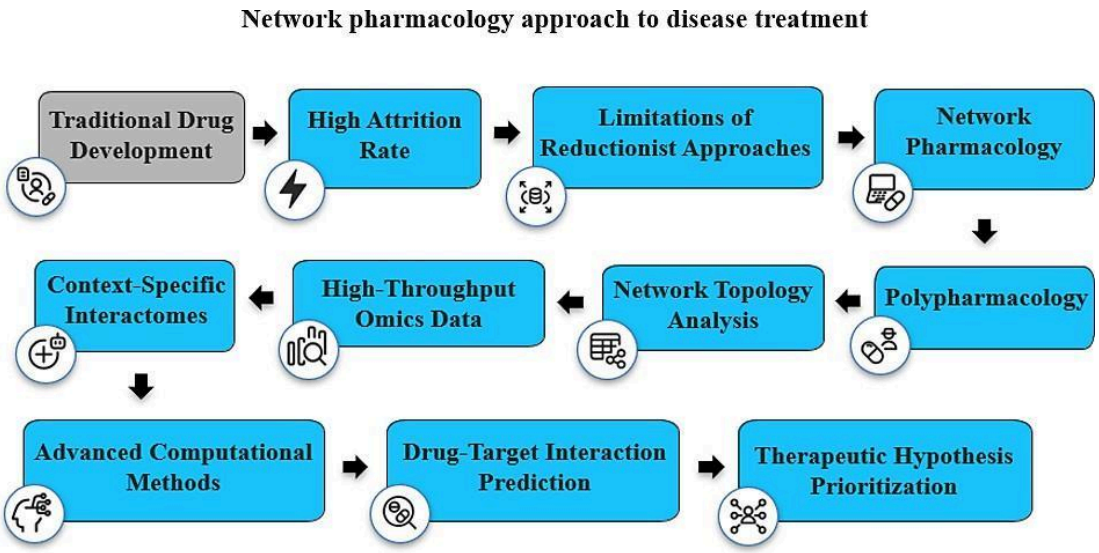

Figure 1 : Illustrates shift from traditional paradigms toward network-based strategies that integrate omics data and computational tools to prioritize therapeutic targets.

This review outlines the conceptual foundations, computational methods, and translational impact of network pharmacology. We begin with key principles of network modeling and polypharmacology, then explore how multi-omics integration and bioinformatics tools enable network construction. Case studies in oncology, neurodegeneration, infectious disease, and traditional medicine demonstrate its utility. Finally, we examine current challenges and future directions, including dynamic network modeling, digital twins, and ethical

considerations [70] [72].

2. Conceptual Foundation of Network Pharmacology

The combination of pharmacology, systems biology, and network science gave origin to network pharmacology. Hopkins (2008) [3] formally introduced it as a response to the limitations of single-target drug discovery, emphasizing that many effective drugs inherently act on multiple targets [3] [16]. Concurrently, Barabási's network medicine framework [1] revealed that diseases correspond to perturbations in specific modules of the interactome. These insights led to the establishment of network pharmacology as an interdisciplinary field modelling drug–target–gene–pathway–phenotype relationships using network structures [3] [1] [16]. Early applications validated its predictive power, identifying drugs through network proximity [8] [13] or mapping active compounds in traditional medicine to biological targets [5] [18]. Today, network pharmacology supports polygenic disease modeling, personalized therapy, and drug repositioning [8] [16].

2.1 Historical Evolution and Definition

Rooted in polypharmacology and systems biology, network pharmacology emerged as a challenge to the "magic bullet" paradigm. Hopkins [3] noted that many approved drugs interact with multiple targets, while Barabási et al. [1] showed disease genes cluster in PPI network modules, suggesting drugs must affect entire modules. This led to explicit modeling of drug–target–disease networks. For instance, Guney et al. [13] demonstrated that the proximity of drug targets to disease genes in the interactome predicts efficacy. Similarly, studies of herbal medicine used network pharmacology to uncover multi-target mechanisms by linking phytochemicals to proteins
[5] [18]. Thus, network pharmacology has matured into a defined discipline integrating graph theory, pharmacology, and computational systems biology [3] [1].

2.2 Principles and Architecture of Network-Based Models

Network pharmacology employs graph-theoretic models, commonly represented as G = (V, E), where V denotes nodes (e.g., genes, proteins, drugs) and E denotes interactions [9] [11]. Edges may be undirected (e.g., PPIs) or directed (e.g., regulatory links), and can be weighted by affinity or confidence scores. Multi-layer networks extend this by incorporating various interaction types across distinct layers (e.g., signaling, metabolic) connected by shared nodes [12]. This structure facilitates the integration of diverse biological data.

Key structural features include hubs (high-degree nodes), bottlenecks (high betweenness), and modules (dense subgraphs reflecting biological functions) [11]. Modules often map to specific pathways or disease mechanisms and are detected using clustering algorithms like Louvain or Markov clustering. Hubs may be critical drug targets, although their essentiality may pose toxicity risks [19]. Topological metrics guide target prioritization. Degree centrality identifies highly connected nodes; closeness measures the speed of node accessibility; betweenness captures nodes acting as network bridges; and eigenvector/PageRank centrality ranks nodes by their influence
[11] [19]. Jeong et al. [19] showed in yeast PPI networks that high-degree genes are often essential, supporting centrality-based target selection. Dynamic models such as random walk with restart (RWR) and heat diffusion simulate how perturbations spread across networks, estimating functional proximity to disease genes [9] [13]. In drug repurposing, drugs targeting nodes near disease modules in the network are considered potential therapies. Modern approaches also incorporate dynamic, context-specific data. As biological networks vary across conditions and time, methods like differential network analysis and dynamic

Bayesian or Boolean networks help capture temporal and conditional interactions [66] [12]. These models offer greater biological fidelity than static PPI maps, especially when integrating time-series or perturbation-driven multi-omics data.

Table 1: Classification and Functional Scope of Key Biological and Pharmacological Networks

Network Type	Structure	Key Components	Data Sources/Tools	Functional Scope/Applications	Ref.
Drug–Target Networks (DTNs)	Bipartite graph: G = (D ∪ T, E)	Drug–target interactions, MoA, binding Kd	DrugBank, BindingDB, ChEMBL	Polypharmacology, off-target prediction, drug repositioning	[14], [15], [21]
Disease–Gene Networks	Unipartite or bipartite: diseases ↔ genes	GWAS, DEGs, eQTL	DisGeNET, OMIM, GWAS Catalog	Disease module detection, biomarkers, comorbidity mapping	[13], [22], [23]
Drug–Disease Networks	Weighted similarity or expression reversal	Drug–disease similarity, signature reversal	LINCS, CMap	In silico repurposing, MoA discovery	[26], [27]
Protein–Protein Interaction	Undirected graph: proteins as nodes	Verified PPI pairs	STRING, BioGRID, IntAct	Basis of interactomes, pathway inference, co-expression	[16], [24]
Pathway-Based Networks	Meta-networks of pathways (e.g., KEGG)	Pathway overlap and crosstalk	KEGG, Reactome	Pathway enrichment, system-level interaction mapping	[28], [29]
Heterogeneous Multi-Layer	G = (V, E, L); layers = genes, drugs, diseases	Multi-entity relationships	NDEx, Hetionet, OmicsNet	Integrative biology, cross-domain insights, personalized care	[30], [31]

1. Network Pharmacology in Drug Discovery

Network pharmacology provides a systems-level framework for drug discovery, moving beyond the reductionist "one-drug-one-target" model by analyzing drug–target–disease interactions within complex biological networks [26]. By integrating graph theory, omics data, and network topology, it addresses challenges in drug repurposing, polypharmacology, target prioritization, and adverse effect prediction with enhanced mechanistic insight [26].

A major application is drug repurposing; wherein approved drugs are computationally screened for new indications. The network proximity approach calculates shortest paths between drug targets and disease genes within the human interactome, with closer proximity indicating greater therapeutic potential [13]. During the COVID-19 pandemic, this strategy helped identify *Baricitinib*, a JAK inhibitor, as a candidate due to its proximity to SARS-CoV-2-related host pathways [3].

Signature-based methods like the Connectivity Map (CMap) compare disease-specific gene expression profiles with those induced by drugs. Metformin, for instance, was shown to reverse the AML transcriptional signature by modulating AMPK/mTOR signalling [15] [14]. These approaches leverage existing molecular data for rapid repurposing, though most rely on static networks. Future efforts aim to incorporate dynamic, condition-specific models to enhance precision. Network pharmacology also advances polypharmacology and multi-target drug design, which are crucial for treating complex diseases involving redundant or compensatory pathways. Multitarget-directed ligands (MTDLs)—single compounds acting on multiple targets—can improve efficacy and reduce resistance [16]. Network models help identify synergistic target sets for MTDL development, supported by computational methods like pharmacophore merging and multi-target docking [17]. Natural compounds such as *curcumin* exemplify this, interacting with diverse proteins linked to inflammation and stress responses [18]. Additionally, network analysis of herbal medicines enables deconvolution of complex mixtures and identification of active phytochemicals [5] [18]. However, ensuring safety and specificity remains a challenge, necessitating robust ADMET and off-target risk assessments [26].

1.1 Target Prioritization and Validation

Network pharmacology transforms target prioritization by integrating topology with functional data. Nodes with high degree or betweenness centrality are often pivotal in disease networks but may pose toxicity risks [19]. To improve selectivity, functional genomics datasets (e.g., CRISPR–Cas9, RNAi screens) are mapped onto PPI networks, identifying targets that are both central and essential [20]. For example, CRISPR data integrated with cancer-specific interactomes has revealed subtype-specific vulnerabilities not evident from genomics alone [20]. Incorporating time-course and perturbation-based multi-omics will further refine causal target identification.

1.2 Adverse Drug Reaction (ADR) Prediction

ADRs are a leading cause of late-stage drug failure. Network pharmacology predicts off-target effects by extending drug interaction maps into PPI and protein–metabolite networks, highlighting connections to critical physiological nodes [21]. Side-effect databases like SIDER and FAERS provide benchmarks for validating these predictions [22] [23].

Machine learning models now combine chemical and network features—such as clustering coefficients and centrality measures—to predict compound toxicity [7]. Graph-based deep learning models (e.g., GCNs) have shown promise in modeling polypharmacy side effects [7]. However, these models are often opaque and population-based. Enhancing ADR prediction will require integrating patient-specific omics and pharmacogenomics, along with developing explainable AI systems [59] [67].

2. Integration of Multi-Omics Data

Integrating diverse omics datasets is central to modern network pharmacology, enabling the construction of

context-specific interactomes that better capture cellular complexity [4] [6]. Layering genomics, transcriptomics, proteomics, metabolomics, and other modalities enhances predictive power by providing complementary insights.

2.1 Genomics and Epigenomics

Genomic data (e.g., GWAS variants, somatic mutations) offer upstream insights into disease predisposition. Variants are mapped to genes via eQTLs and then to interaction networks [29]. Epigenomic profiles—DNA methylation, histone modifications, and chromatin accessibility—identify regulatory elements and reveal how genetic risk influences gene expression [30]. Incorporating 3D genome data (e.g., Hi-C) aids in linking non- coding variants to distal targets. While much of the non-coding genome remains poorly characterized, multi- omics correlations can illuminate functional relevance.

2.2 Transcriptomics

Transcriptomics, particularly RNA-seq, identifies differentially expressed genes (DEGs) in disease or drug-treated states. DEGs mapped to PPI networks define disease or drug-response modules. Tools like WGCNA group genes into co-expression modules associated with clinical phenotypes [24] [25]. In breast cancer, WGCNA identified modules predictive of chemotherapy response [26]. However, transcriptomics lacks information on post-transcriptional regulation, necessitating integration with proteomic data for network accuracy.

2.3 Proteomics

Proteomics quantifies proteins—the functional effectors—along with post-translational modifications (PTMs) using mass spectrometry [27]. Phosphoproteomics highlights active signaling (e.g., kinase–substrate interactions) under different conditions [28]. These data validate predicted PPIs and uncover context-specific protein

complexes. For instance, CPTAC projects have linked mutations to signaling pathways through proteogenomics [28]. Despite challenges like limited dynamic range and incomplete PTM annotation, integrating proteomics with transcriptomics improves network resolution and target confidence.

2.4 Metabolomics

Metabolomics captures the end products of cellular processes, revealing phenotypic states. Techniques like NMR and mass spectrometry profile metabolite levels and fluxes, supporting the construction of metabolic networks and genome-scale metabolic models (GEMs) [31]. These models help identify druggable enzymes and metabolic vulnerabilities. Pharmacometabolomics tracks drug-induced changes, linking metabolic shifts to efficacy or resistance [32]. Though hindered by standardization and complexity, integrating metabolomics with proteomic and transcriptomic networks clarifies drug mechanisms at the pathway level.

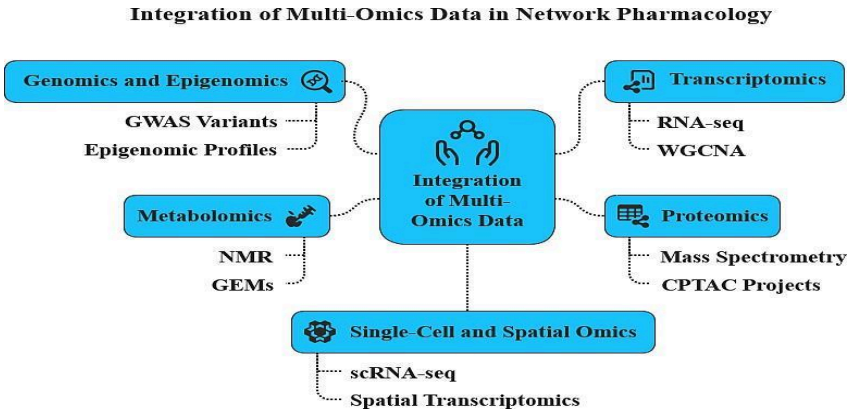

Fig 2 : The figure highlights the integration of genomics, transcriptomics, proteomics, metabolomics, and single-cell/spatial omics in network pharmacology to support drug discovery. It emphasizes key tools like RNA-seq, mass spectrometry, and NMR for analysing complex biological data. GWAS: Genome-Wide Association Studies; WGCNA: Weighted Gene Co-expression Network Analysis; NMR: Nuclear Magnetic Resonance; GEMs: Genome-Scale Metabolic Models; CPTAC: Clinical Proteomic Tumor Analysis Consortium; scRNA-seq: Single-Cell RNA Sequencing.

2.5 Single-Cell and Spatial Omics

Single-cell technologies (e.g., scRNA-seq, scATAC-seq) allow reconstruction of cell-type-specific networks, essential for understanding heterogeneity in cancer and immune responses [33]. Methods like trajectory inference and pseudotime analysis capture dynamic cellular processes [34]. For example, single-cell networks revealed T- cell exclusion pathways in melanoma influencing immunotherapy response [69]. While single-cell data are sparse and noisy, integration with spatial and multi-modal methods (e.g., CITE-seq, spatial transcriptomics) enriches network context.

Robust multi-omics modeling relies on effective integration strategies. Vertical integration merges different omics layers from the same samples; horizontal integration combines datasets of the same type across cohorts. Methods like iClusterPlus [36], joint-NMF, and MOFA [37] extract shared latent features. Similarity Network Fusion (SNF) aggregates similarity graphs into unified networks, improving patient stratification [54]. Tools like PANDA and LIONESS build context-specific regulatory networks from gene expression and motif data [55]. Emerging graph neural networks (GNNs) model nonlinear relationships in multi-omics to predict drug responses [38]. However, integration methods require careful parameter tuning and expert interpretation. Standardized preprocessing (e.g., batch correction [56]), feature scaling, and experimental validation are critical for reproducibility.

Table 2: Representative Tools for Network-Based Multi-Omics Integration in Drug Discovery

Network Type	Structure & Components	Data Sources/Tools	Applications	Ref.
Drug–Target Networks (DTNs)	Bipartite: Drugs ↔ Targets	DrugBank, BindingDB, ChEMBL	Polypharmacology; repurposing; off-target analysis	[14], [15], [21]

Disease–Gene Networks	Unipartite/Bipartite: Diseases ↔ Genes	DisGeNET, OMIM, GWAS Catalog	Disease module discovery; biomarker ID	[13], [22], [23]
Drug–Disease Networks	Similarity/reversal-based graphs	LINCS, CMap	Expression reversal; MoA discovery; repurposing	[26], [27]
Protein–Protein Interaction (PPI)	Undirected graph: proteins ↔ interactions	STRING, BioGRID, IntAct	Network construction; pathway mapping	[16], [24]
Pathway-Based Networks	Meta-networks of interacting pathways	KEGG, Reactome	Crosstalk analysis; pathway-level systems modeling	[28], [29]
Heterogeneous Multi-Layer Networks	Layered graphs: drugs, genes, diseases, pathways	NDEx, Hetionet, OmicsNet	Integrative biology; personalized medicine	[30], [31]

3. Role of Bioinformatics Tools and Databases

Bioinformatics is central to network pharmacology, supporting data acquisition, integration, analysis, and visualization through a diverse ecosystem of platforms and databases.

3.1 Network Construction and Visualization

Cytoscape [39] remains a leading tool for molecular network analysis, extendable via plugins such as ClueGO (gene ontology) and StringApp (STRING integration) [39] [41]. For large-scale or complex networks, Gephi provides advanced visualization [40], while NetworkX (Python) allows flexible analyses. STRING [41] and STITCH [42] offer high-confidence protein–protein and protein–chemical interaction data, respectively. GeneMANIA [43] builds functional association networks using co-expression and pathway data. While these tools primarily support static modeling, demand is growing for dynamic, multi-layered visualization.

3.2 Databases of Interactions and Annotations

Curated databases supply critical interaction and annotation data. DrugBank [44] and ChEMBL [45] provide drug–target profiles, including mechanisms and binding affinities. TTD [46] links targets to disease and clinical contexts, and PharmGKB [47] compiles pharmacogenomic associations. Multi-omics data from TCGA [48], GTEx [49], GEO [50], and PRIDE [51] enable robust network modeling. HMDB [52] supports metabolic network construction. Effective integration requires standardized identifiers (e.g., Entrez, UniProt) and harmonized ontologies.

3.3 Analytical and Integration Tools

Numerous tools support network-based multi-omics analysis. WGCNA [24] [25] enables co-expression network construction, mixOmics [53] applies multivariate methods, and SNF [54] fuses diverse omics data. Gene regulatory networks can be inferred with ARACNe and GENIE3 [55]. To address batch effects, ComBat [56] offers correction before analysis. Single-cell tools like RNA velocity [57] capture temporal dynamics, while node2vec [58] generates graph embeddings. SHAP [59] aids interpretability by attributing predictions to input features.

4. Applications and Case Studies

Network pharmacology has advanced understanding and treatment across oncology, neurodegeneration, infectious diseases, autoimmune conditions, and traditional medicine. These examples demonstrate its utility in elucidating disease mechanisms, repurposing drugs, and designing combination therapies.

4.1 Oncology

Cancer's heterogeneity involves disrupted signaling and genomic alterations. Integrating multi-omics data (e.g., TCGA) enables tumor stratification and drug response modeling [26] [28]. For instance, a network combining transcriptomic and mutational profiles predicted paclitaxel response in breast cancer, highlighting immune pathways alongside known microtubule targets [26]. Analyses have also revealed compensatory circuits (e.g., PI3K–AKT and MAPK), guiding rational combination therapies [60]. Synthetic lethality studies, integrating CRISPR screens with PPI networks, uncover subtype-specific vulnerabilities missed by genomics alone [20]. Incorporating single-cell and spatial omics will further support personalized tumor network models.

4.2 Neurodegenerative Diseases

Alzheimer's and Parkinson's involve multifactorial processes like protein aggregation, inflammation, and metabolic dysfunction. Network pharmacology supports multi-target strategies, such as mapping phytochemicals onto PPI networks containing amyloid-beta, tau, and inflammatory proteins. High-centrality compounds show neuroprotective potential [61]. Network proximity analyses have linked antidiabetic drugs (e.g., metformin, GLP- 1 analogs) to Alzheimer's pathways, prompting clinical trials [61] [62]. Challenges remain in building brain- specific interactomes and integrating longitudinal neural data.

4.3 Infectious Diseases

Network approaches rapidly identify therapeutics for emerging infections. During COVID-19, virus–host protein networks helped repurpose approved drugs, with Baricitinib identified via proximity to viral entry and immune pathways [3]. Similar strategies in influenza predicted synergistic antivirals by modeling host–pathogen and cytokine networks [63]. Future work aims to incorporate viral mutation profiles for adaptive host–pathogen models.

4.4 Autoimmune Disorders

Autoimmunity involves complex immune signaling and genetic factors. Network analyses, such as WGCNA of blood transcriptomes, have identified immune gene modules in lupus and rheumatoid arthritis, revealing interferon and Toll-like receptor targets [24]. Modeling immune cell interactions supports rational design of combination immunotherapies. Future efforts will benefit from integrating HLA genotypes and single-cell immune data for personalized maps.

4.5 Traditional and Ethnopharmacology

Network pharmacology modernizes traditional medicine by linking herbs, compounds, targets, and diseases [5]. For example, TCM's QingFeiPaiDu was mapped to host proteins involved in viral entry and

inflammation during COVID-19, providing mechanistic validation [5]. Similarly, analysis of Triphala (Ayurveda) identified gallic acid as an active agent in gut and inflammatory pathways [64]. These studies highlight bioactive components and potential synergies [18] [64], though progress depends on richer phytochemical databases and high-throughput validation.

Table 3: Representative Disease Domains and Network Pharmacological Applications

Disease Domain	Network Approach	Translational Insight	Example/Study	Clinical Impact	Ref.
Cancer	Mutation, expression, PPI integration	Tumor stratification; combo therapies	TCGA-guided kinase inhibitor pairing	Precision treatment; reduced resistance	[26]
Neurodegeneration	Target mapping to neuroinflammation	Drug repurposing; phytochemical synergy	Alzheimer's–Metformin study	Multi-pathway targeting; neuroprotective candidates	[18], [61], [62]
Infectious Disease	Viral-host interactome; diffusion analysis	Rapid drug repurposing (e.g., COVID- 19)	Baricitinib proximity prediction	Fast therapeutic discovery; antiviral/immuno combos	[3], [63]
Autoimmunity	Co-expression and pathway network analysis	Subtype- specific immune targets	WGCNA in lupus stratification	Biomarker-driven therapies	[24]
Traditional Medicine	Herb-compound-target-disease networks	Mechanistic insights into complex formulas	Triphala for GI inflammation	Modernization; lead compound identification	[5], [18], [64]

1. Limitations and Future Perspectives

Despite its promise, network pharmacology faces challenges related to data quality, computational demands, interpretability, and clinical translation. This section outlines key limitations and emerging solutions.

1.1 Data and Methodological Challenges

A major limitation is the incompleteness and bias of current PPI networks, which overrepresent well-studied proteins and omit many tissue- or condition-specific interactions [65]. False positives from high-throughput assays further distort topology, risking misleading conclusions such as incorrect disease modules or hub identification [65]. While initiatives like the Human Interactome Project and text-mining are improving coverage, data sparsity remains a constraint.

Most models use static, averaged networks that overlook biological dynamics such as stage-specific signaling or transient complexes [66]. New approaches—leveraging time-series data, time-varying networks, and RNA velocity—aim to capture these dynamics [57], though integration into pharmacological models is still nascent.

Multi-omics integration is hindered by inconsistent data scales, batch effects, and sample mismatches [35]. Without robust harmonization (e.g., ComBat [56]), false correlations may arise. Cross-modal links (e.g.,

SNPs to metabolites) rely on incomplete resources like eQTL databases and metabolic maps [29] [31]. Adoption of standardized, FAIR-compliant pipelines is essential for reproducibility [71] [72].

1.2 AI and Machine Learning Integration

AI offers powerful tools for network pharmacology, but also introduces challenges. Deep learning models (e.g., GNNs, VAEs) capture complex patterns [58], yet often function as opaque "black boxes." This lack of interpretability hampers mechanistic insight—crucial in drug discovery. Tools like SHAP [59], attention layers, and GNNExplainer [67] offer partial transparency, but comprehensive biological explainability remains limited.

Generative models (e.g., GANs, VAEs) support de novo molecule design [68], yet often ignore key drug development factors like synthesis feasibility, toxicity, and ADMET profiles. Integrating pharmacokinetic constraints and medicinal chemistry rules is ongoing. Model bias is also a concern, as training datasets often lack diversity in drug types, cell lines, and populations. Mitigation strategies include federated learning, domain adaptation, and more inclusive data collection.

1.3 Personalized and Ethical Considerations

While promising for precision medicine, patient-specific applications are still emerging. Advances in single-cell and spatial omics allow individualized network models capturing tumor and tissue heterogeneity [33]. A future goal is the creation of digital twins—virtual patients that integrate multi-omics and clinical data to simulate disease and treatment responses [70]. However, this requires high-quality, costly data and robust physiological models. Ethical and regulatory challenges—including data privacy, clinical validation, and transparency—must be resolved before such models can be deployed clinically [71] [72].

Table 4: Major Challenges and Strategic Solutions in Network Pharmacology Research

Challenge	Description	Strategic Solution	Example Tools/Initiatives	Ref.
Incomplete Interactome	Missing or generic PPI/gene-disease interactions	Context-specific networks; multi-source integration	STRING [41], OmniPath, Rolland map	[41], [65]
Omics Heterogeneity	Scale, noise, and sparsity across omics layers	Multi-view learning; batch correction; data harmonization	SNF, mixOmics, Leek normalization	[53], [54],[56]
AI Model Opacity	Deep models lack interpretability	Explainable AI (e.g., SHAP, attention layers, subgraph visualization)	GNNExplainer, SHAP	[59], [67]
Ethical & Regulatory	Data privacy, bias, and limited oversight	Validation frameworks; FAIR data; ethical AI guidelines	FDA guidance, EU initiatives	[71], [72]
Personalization & Equity	Homogeneous datasets limit generalizability	Diverse cohort inclusion; federated learning; real-world data use	TCGA+GTEx harmonization, eQTL meta-analyses	[48], [29]

Conclusion

Network pharmacology marks a paradigm shift in drug discovery and systems medicine by reframing diseases and therapeutics as components of interconnected networks. This systems-level perspective aligns with the polygenic basis of most disorders and supports drug repurposing, multi-target drug design, biomarker discovery, and personalized therapy through integrative modeling. Practical successes—such as identifying anti-cancer combinations and repurposing diabetes drugs for Alzheimer's—demonstrate its impact [60] [62].

Despite its promise, the field remains nascent. Major challenges include incomplete interactomes, data heterogeneity, and the opacity of complex models. Progress depends on advances in multi-omics data (e.g., single- cell, temporal), dynamic network modeling, and ethically sound AI applications [65] [72]. As computational and experimental methods continue to converge, network pharmacology is poised to become central to precision medicine—enabling deeper insights into disease mechanisms and the development of safer, more effective therapies.

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