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Identification of Potential Drug Targets for the Treatment of Severe Burn Wounds from a Multi-Omics Perspective

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Purpose: Severe burns result in significant skin damage, impairing its primary role as an infection barrier and presenting substantial treatment challenges. Despite improvements in the treatment of burn patients due to advancements in materials and techniques, there remains a need for novel therapeutic approaches to enhance burn prognosis further.

Patients and Methods: Several types of genomic methods are used in this study, such as differential gene expression analysis, weighted gene co-expression network analysis (WGCNA), machine learning, and Mendelian randomization (MR), to find genes that are linked to severe burns and create a diagnostic nomogram to see how well these genes can predict severe burns. Drug prediction was conducted using the DsigDB database, and molecular docking was used to validate the pharmacological value of drug targets. The effects of genes and drugs on burn wounds were validated through Western Blot (WB) and cell scratch assays.

Results: In patients with severe burns, multi-omics analysis revealed increased CYP19A1 expression. In severe burn cell models, WB further confirmed the elevated expression of CYP19A1. Drug prediction indicated that mevastatin binds effectively to the CYP19A1 gene expression protein. The healing area of scalded HaCat cells was much bigger after 24 hours of mevastatin treatment compared to the scald-only group, as shown by cell scratch assays after 24 and 48 hours.

Conclusion: This study innovatively integrates multi-omics approaches into burn wound research, uncovering for the first time that mevastatin promotes burn wound healing by downregulating CYP19A1 expression. This discovery may provide a new foundation for developing burn wound therapeutics and potentially reduce drug development costs.

Keywords: burn, WGCNA, machine learning, Mendelian randomization, drug targets, genetics

Introduction

Burns remains a prevalent form of trauma, causing severe physiological and psychological damage to patients, particularly in cases of severe burns (including moderate and critical burns), which are challenging to treat, costly, and associated with high mortality rates.[1–3] Studies indicate that increased levels of inflammatory cytokines such as interleukin-6 (IL-6), interleukin-1β (IL-1β), tumor necrosis factor-α (TNF-α), and prostaglandins, along with elevated apoptotic cells, contribute to multi-organ system failure in severe burns.[4,5] The extensive damage caused by severe burns can lead to significant deformities, disabilities, and even death. Despite substantial improvements in burn treatment due to the introduction of biomaterials and advanced technologies, many patients still have poor prognoses. Healthcare teams and patients are confronted with a substantial challenge, underscoring the necessity of developing novel therapeutic strategies and drug targets.

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Researchers from various disciplines have extensively employed bioinformatics research to extract prospective information and elucidate disease mechanisms. Recent advances in bioinformatics analysis have provided new insights into the molecular mechanisms of diseases. Machine learning is an effective approach to recognizing patterns in data, which is frequently implemented in clinical environments. Today, machine learning models are employed extensively in the medical field for data mining, medical diagnostics, and disease risk prediction, demonstrating predictive solid performance. MR, a tool-based approach utilizing single nucleotide polymorphisms (SNPs) as instruments, helps infer causal relationships between exposures and outcomes. MR minimizes confounding factors, thus reducing bias between exposure and outcomes.

In this study, we present several novel findings in the field of burn research. First, we identified new therapeutic targets for burns using Mendelian randomization (MR) and large-scale genome-wide association study (GWAS) data. This approach integrates cis-expression quantitative trait loci (eQTL) data with burn-related association data to establish causal relationships between exposure and outcomes, thereby improving the prediction of drug efficacy. Second, we validated the pharmacological activity of potential burn drug targets through drug prediction and molecular docking studies. These targets, which are closely associated with immune function, were evaluated for their feasibility and potential efficacy by assessing their binding affinity and interaction patterns with drugs.

For this study, we selected raw microarray datasets from the Gene Expression Omnibus (GEO) database. After identifying differentially expressed genes (DEGs) between severely burned patients and healthy controls, we performed WGCNA followed by Gene Ontology (GO), Disease Ontology (DO), and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway analyses. The identified DEGs were subjected to gene selection using machine learning models and integrated with target genes selected via Mendelian randomization for drug target validation.

Our study offers significant insights for discovering new therapeutic targets for burns. By integrating WGCNA, differential gene expression analysis, machine learning, MR, drug prediction, molecular docking, and experimental validation, we provide essential guidance for developing more effective and targeted treatment methods.

Material and Methods

Study Design

Centre for Biotechnology Information GEO (http://www.ncbi.nlm.nih.gov/geo)[6] includes data from 244 severe burn samples and 35 healthy control samples. We selected samples from severe burn patients who were admitted within 48 hours post-injury and included healthy controls, resulting in a total of 155 samples. Among the patients, 42 were female (27%) and 113 were male (73%). The age of the patients ranged from 0 to 86 years, with a mean age of 27.41 ± 18.84 years and a median age of 25.67 years. Blood samples were collected from all participants, and all patients had a burn area greater than 20% of the total body surface area.

A gene differential analysis was performed on the 157 samples, combined with WGCNA and machine learning models, to identify differentially expressed genes associated with severe burns. Our study does not involve animal or human clinical trials, and there are no ethical concerns. The study utilizes publicly available de-identified data from participant research that has already been approved by an ethics committee for human research. Consequently, no separate ethical approval was required for this study.

Current research focuses on cis-eQTLs, using GTEx eQTL summary data for Summary-data-based Mendelian Randomization (SMR) analysis to study the relationship between gene expression and burns. We identified hub genes by integrating differential gene expression analysis, WGCNA, and SMR results. Drug prediction was conducted for hub genes, and candidate drugs were subjected to molecular docking to evaluate their interaction patterns and binding affinity with target genes. External experimental validation was performed to verify the differential expression of hub genes in severe burns and the regulatory effects of candidate drugs on hub genes and burn wound healing (Figure 1).

Identification of DEGs and Construction of Weighted Co-Expression Networks

DEGs between severe burn samples and healthy controls were analyzed using the Limma package in R software. With an adjusted P-value <0.05 and $|\log FC| \geq 1$, significant genes were filtered out. DEGs were visualized using the ggplot2 package. Co-expression networks were constructed using the WGCNA R package, calculating gene significance (GS)

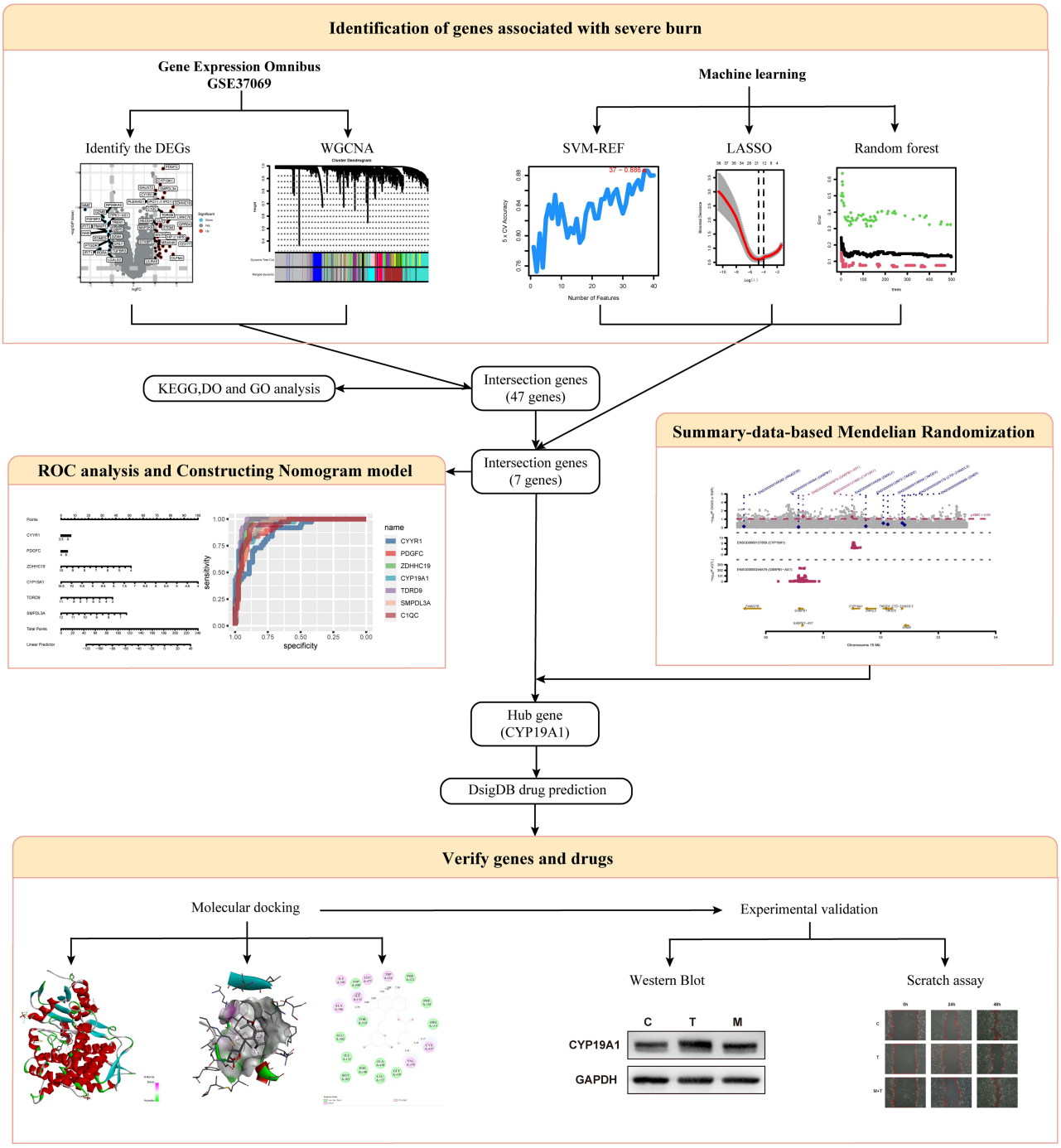

Figure 1 Flowchart.

and module membership (MM) with the WGCNA algorithm and clustering genes into different modules. Relevant gene modules were selected based on $P < 0.05$ and the highest Pearson correlation coefficients.

Functional Enrichment Analysis

The clusterProfiler R package was employed to conduct KEGG, GO, and DO pathway enrichment analyses to clarify the biological functions and signaling pathways of hub genes. Default parameters were used for clusterProfiler. The identification threshold for GO functions and KEGG pathways was $P < 0.05$.

Machine Learning and Nomogram Construction

To achieve optimal selection, three models were constructed: the Support Vector Machine Recursive Feature Elimination (SVM-RFE) model using the "e1071" and "caret" R packages, the Least Absolute Shrinkage and Selection Operator (LASSO) model using the "glmnet" R package and the Random Forest (RF) model using the "randomForest" R package. Cross-validation across these three models identified severe burn-related genes as characteristic genes for severe burns. A nomogram was constructed using the "rms" R package to evaluate the predictive value of various clinical features and risk scores.

Burn GWAS Data and SMR

Burn data were sourced from a comprehensive study in the Finngen database (https://www.finngen.fi/en),[7] which includes 3134 cases and 306020 controls, primarily analyzing the European population, including both males and females. The GTEx Consortium V8 eQTL summary-level data was used in this investigation (https://gtexportal.org/). SMR analysis used eQTLs as instrumental variables for effect estimation, exploring the association between gene expression levels and the outcomes of interest. A heterogeneity test dependent on instruments (HEIDI) was conducted to assess the reliability of the observed associations. P_{HEIDI} values less than 0.05 indicate evidence supporting that observed associations may be due to different genetic variants in high linkage disequilibrium. SMR software version 1.3.1 was used for the analysis.

Single-Sample Gene GSEA and Gene Expression Differences

The GSVA tool from Bioconductor was utilized to conduct single-sample gene set enrichment analysis (ssGSEA),[8] available at http://www.bioconductor.org.[9] This method calculates individual enrichment scores for each sample and gene set, identifying aberrant pathways.[8,10]

Candidate Drug Prediction

Assessing protein-drug interactions is crucial for understanding whether target genes can be used as practical drug targets. This study utilized the Drug Signature Database (DSigDB) for this purpose. There are 22,527 gene sets and 17,389 distinct compounds in DSigDB, a comprehensive database that links drugs and other chemicals with their target genes, encompassing 19,531 genes. Candidate drugs were predicted, and their pharmacological activity was assessed by uploading the target genes identified to DSigDB.

Molecular Docking

To gain deeper insights into candidate drugs' regulatory effects and therapeutic potential, molecular docking was performed at the atomic level to assess the interaction patterns and binding affinity between candidate drugs and target genes. Using molecular docking, we simulated the interaction patterns and binding affinity between targets and ligands. By pinpointing ligands that exhibited strong binding affinity and advantageous interactions, we prioritized the drug targets for subsequent experimental validation and refined the design of potential drug candidates. Molecular docking analysis of critical drugs and their target proteins was conducted using the AutoDock Vina software (http://autodock.scripps.edu/). Structural information for small molecule drugs was retrieved from the PubChem Compound Database[11](https://pubchem.ncbi.nlm.nih.gov/), while protein structures related to gene expression were acquired from the PDB (http://www.rcsb.org/).

Experimental Validation

Cell Culture and Modeling

The immortalized human keratinocyte cell line (HaCat cells) was purchased from Zhongqiao Xinzhao Biotechnology Co., Ltd. (Shanghai, China). The HaCaT cells in this investigation, were cultured in DMEM media supplemented with 10% fetal bovine serum (FBS) (Gibco, Thermo Fisher Scientific) at 37 °C and 5% CO_2, with a daily medium change. Following the manufacturer's instructions from MedChemExpress, the stock solution was prepared using DMSO (250 mg/mL) and combined with ultrasonic treatment. HaCaT cells were digested with trypsin and distributed into

a 6-well plate at a density of 1.5 × 10^5 cells per well to facilitate the simulation of an in vivo burn model and to assess the impact of mevastatin on burns. The cells were divided into three groups: the control group (C group), where cells were placed in a water bath at room temperature and treated with saline; the burn group (B group), where cells at 80–90% confluence were treated in a 43°C water bath for 50 minutes[12] and treated with saline; and the mevastatin group (M group), where cells were treated with 5 μM mevastatin after burn injury.

Western Blot Analysis

The proteins were extracted and separated using SDS-PAGE at 80V for 120 minutes. Afterward, the proteins were transferred to a 0.45 μm PVDF membrane at 200 mA and blocked with a non-protein blocking solution (Epizyme, Shanghai, China) at 37°C for 15 minutes. The membrane was subjected to overnight incubation at a temperature of 4°C with primary antibodies (CYP19A1, Abclonal, Catalog No. A12238, 1:750 dilution; GAPDH, Proteintech, Catalog No. GB12002-100, 1:200 dilution), then washed three times with 1×TBST for 5 minutes each. The membrane was incubated with HRP-conjugated secondary antibodies (HRP-conjugated Goat Anti-Rabbit IgG, Servicebio, Catalog No. GB23303, 1:3000 dilution; HRP-conjugated Goat Anti-Mouse IgG, Servicebio, Catalog No. GB23301, 1:3000 dilution) at room temperature for 1 hour, followed by three washes with 1×TBST for 5 minutes each. WB analysis was performed using chemiluminescence. The membrane was incubated in an enhanced chemiluminescence reagent (ECL, GlpBio, USA) for 1 minute and exposed to a developer for imaging.

Scratch Assay

Following the burn treatment of HaCat cells as described, a scratch was made on the long axis of a 6-well plate using a 200 μL pipette tip. After two washes with PBS, images were captured and observed using an optical microscope (×20, Leica). The Drug Treatment Group cells were treated with 5 μM mevastatin and then incubated in a cell incubator. Images were taken at 24 and 48 hours post-treatment to assess wound healing. The wound healing area was analyzed using ImageJ software (v1.8.0, NIH) for 24 and 48 hours of data.

Statistical Analysis

The experimental data were analyzed using SPSS software version 26.0, while graphs were generated using GraphPad Prism version 8.0. All data are derived from at least three independent experiments and are expressed as mean ± standard error of the mean (Mean ± SEM). One-way analysis of variance (ANOVA) was used to compare differences between the three groups, with a P value < 0.05 considered statistically significant.

Results

Analysis and Identification of Severe Burn-Related Genes

The gene expression dataset GSE37069 from the GEO database was analyzed, including 120 severe burn patients and 35 healthy controls. Based on $|\log2\ FC| \geq 1.0$ and adj. P-Val ≤ 0.05, 60 DEGs were identified, including 20 down-regulated and 40 upregulated genes (Table S1). The results of the expression level analysis are presented in the volcano plot and heat map (Figure 2A and B). The ridge plot shows that DEGs are primarily enriched in peroxisome, glycolysis, B cell receptor signaling pathway, and complement and coagulation cascades (Figure 2C).

WGCNA was used to cluster samples in the training set, determining the scale-free topology fit index and performing average connectivity analysis (Figure 2D and E), with the optimal soft threshold $\beta = 11$. The scale-free topology fit index R^2 approached the threshold of 0.9 (red line) when average connectivity approached zero. The dynamic tree-cut method was employed to identify modules, with a minimum of 30 genes per module and a deep split level of 2 (Figure 2F). Fourteen modules were correlated with the control and burn groups, resulting in a correlation heatmap (Figure 2G). The red module, which contained 512 genes, was deemed critical due to its significant correlation with severe burns ($r = 0.88$, $P < 0.001$). A high correlation between MM and GS is evident in the scatter plot of MM-GS for the red module. (Figure 2H). A Venn diagram further identified 47 genes associated with severe burns (Figure 2I).

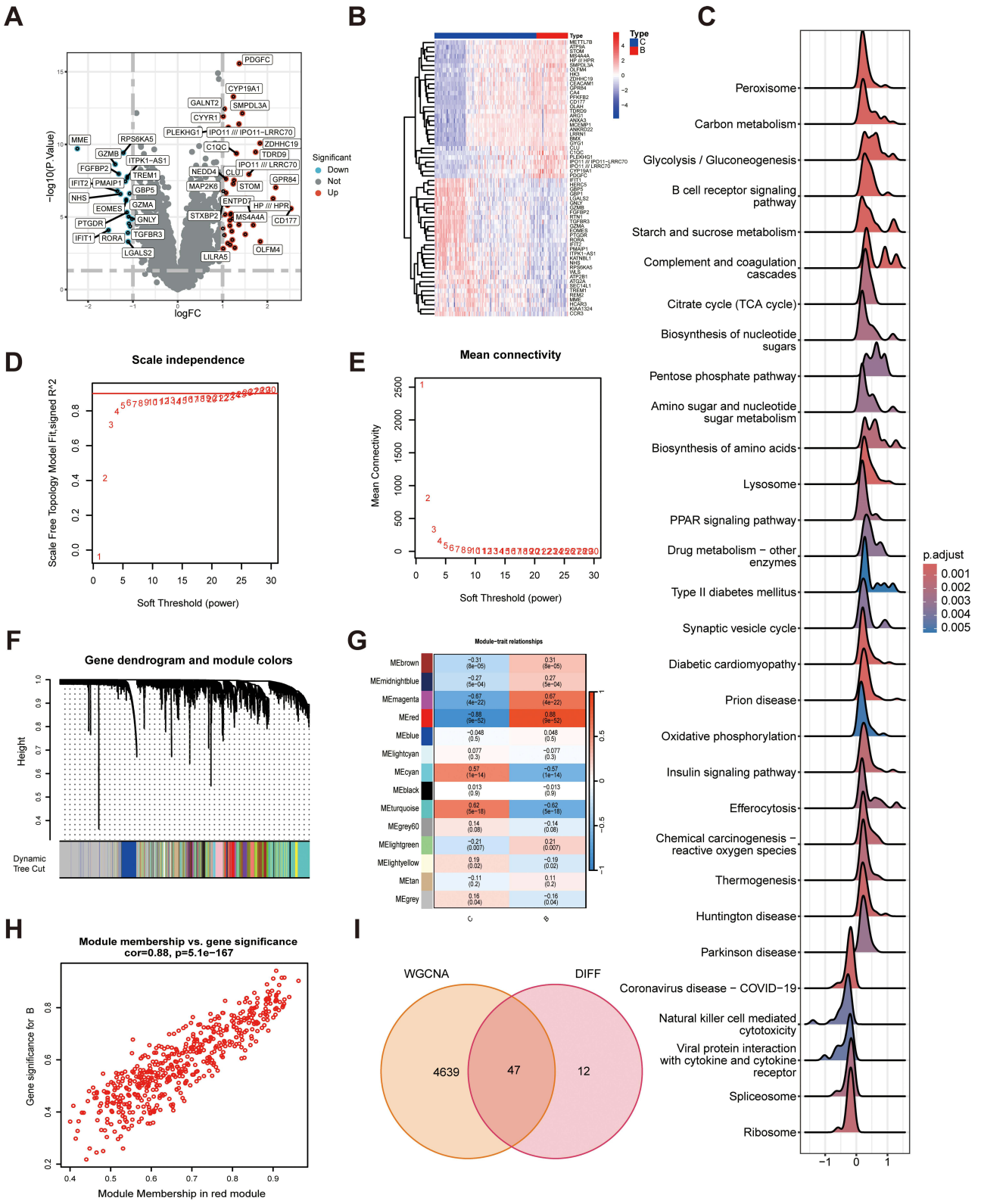

Figure 2 DEGs Identification and WGCNA Analysis. (**A**) Volcano plot of DEGs; (**B**) Heatmap of DEGs; (**C**) Signaling pathways highly correlated with severe burns; (**D** and **E**) Average connectivity analysis; (**F**) Cluster dendrogram of co-expressed genes in severe burns; (**G**) Heatmap of correlations between gene modules and traits (values in parentheses are p.adj, values outside parentheses are correlation coefficients for different modules); (**H**) Scatter plot of the red module with the highest correlation coefficient; (**I**) Venn diagram identifying genes associated with severe burns.

Biological Classification and Pathway Enrichment Analysis of Severe Burn-Related Genes

The 47 identified genes associated with severe burns were subjected to biological classification and pathway enrichment analysis. The TNF signaling pathway, MAPK signaling pathway, and nitrogen metabolism were identified as enrichment in the KEGG pathway enrichment analysis (Figure 3A). The GO analysis indicated that the biological process (BP) terms were primarily related to cellular damage, cell components (CC) were predominantly enriched in granule membrane, and molecular functions (MF) were enriched in protein tyrosine kinase activity (Figure 3B). DO analysis showed that the related genes were primarily associated with cancer (Figure 3C).

Machine Learning Identifies Relevant Genes

The analysis of DEGs and WGCNA identified 47 genes associated with severe burns. To further identify relevant genes, we constructed three machine learning models, SVM-RFE, LASSO, and RF, using these 47 related genes. SVM-RFE identified a total of 47 relevant genes (Figure 4A and B), LASSO identified 18 relevant genes (Figure 4C and D), and RF identified 10 relevant genes (Figure 4E and F) (Table S2). In addition, a ten-fold cross-validation was conducted to assess the stability of the predictive models. Through cross-analysis of the genes identified by the three machine learning models, seven genes associated with severe burns were ultimately identified: CYYR1, PDGFC, ZDHHC19, CYP19A1, TDRD9, SMPDL3A, and C1QC (Figure 4G).

Construction of Diagnostic Nomogram

To provide more targeted diagnostic support for patients with severe burns, this study constructed a diagnostic nomogram based on seven relevant genes (Figure 5A), with the total score predicting the risk of severe burns. We calculated the area under the receiver operating characteristic (ROC) curve (AUC) for these seven genes (Figure 5B) and presented the AUC values for each gene (Figures 5C–I). Each gene's AUC value exceeded 0.85, demonstrating their significant predictive value for patients with severe burns. These results further confirm the critical clinical significance of the seven relevant genes in patients with severe burns.

Smr

We employed the SMR method to screen for burn-related genes. Figure 6A is a scatter plot of the instrumental variables used in the SMR method. Using GTEx Consortium data for SMR analysis, we identified 436 genes associated with burns (Table S3). The genes identified through SMR screening were combined with the seven genes selected by the machine learning model, ultimately resulting in the identification of a single key gene: CYP19A1. The P_{HEIDI} value of the CYP19A1 gene was 0.92, indicating no heterogeneity (Table S3).

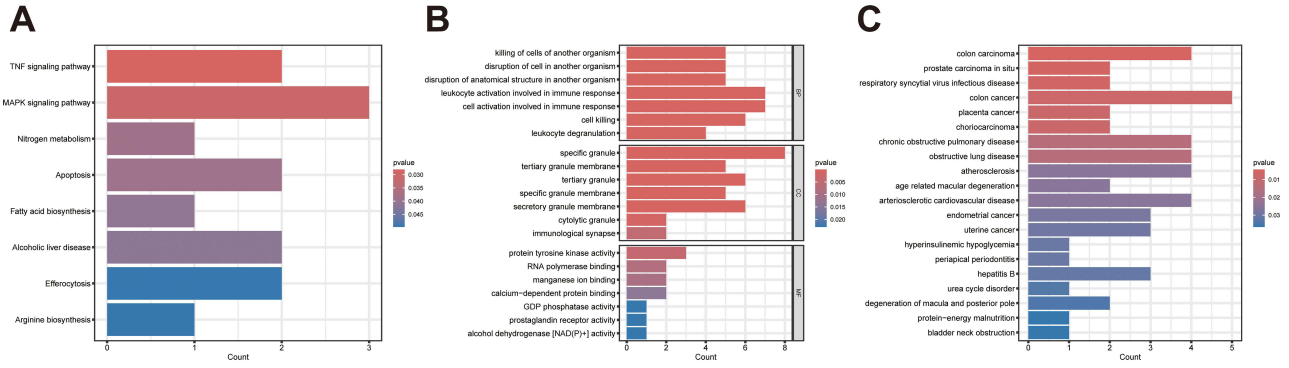

Figure 3 Pathway Enrichment Analysis. (**A**) KEGG enrichment analysis; (**B**) GO enrichment analysis; (**C**) DO enrichment analysis.

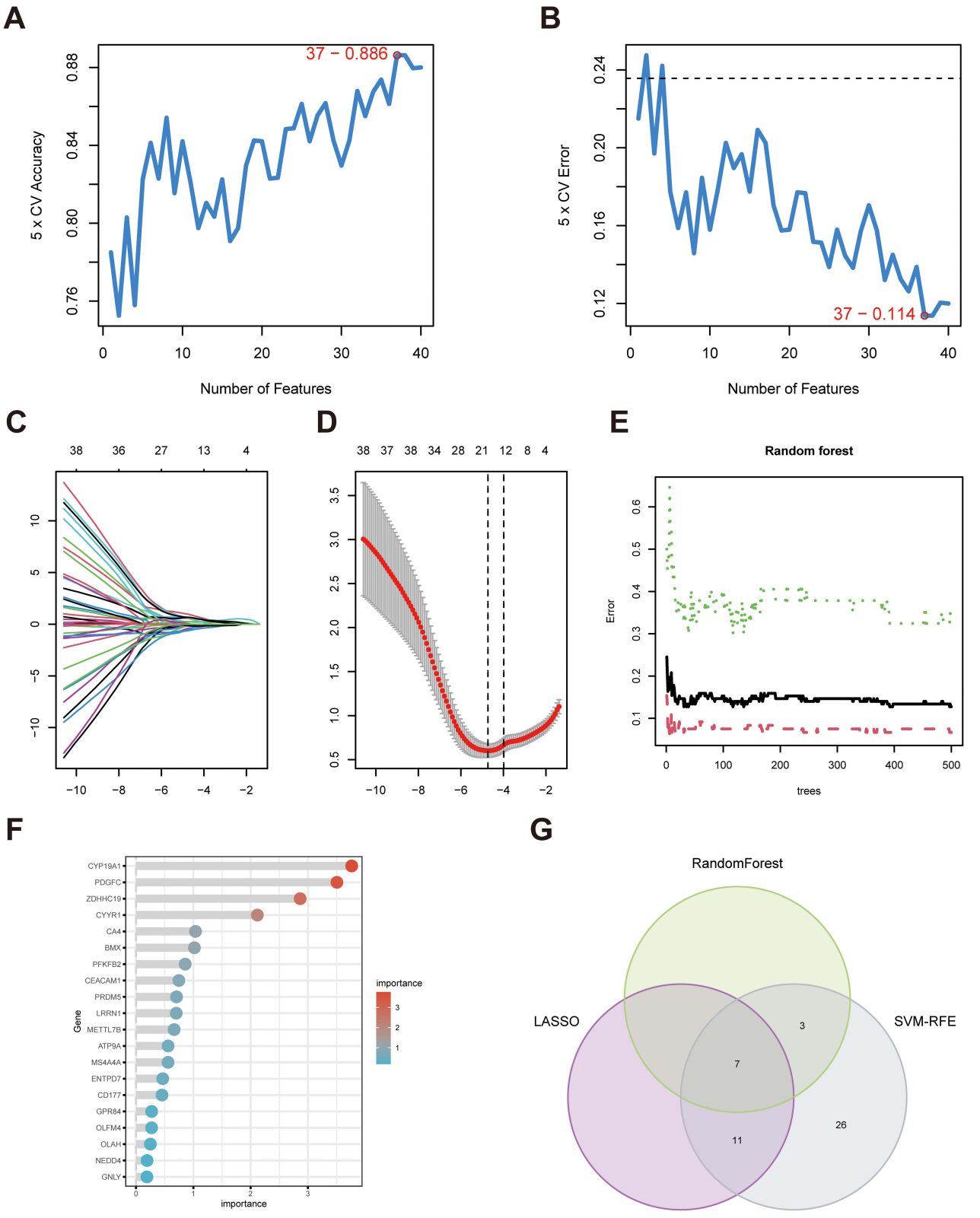

Figure 4 Machine Learning Analysis Identifying Disease Key Genes. (**A** and **B**) SVM-RFE algorithm screening for potential characteristic genes in severe burns; (**C**) Gene coefficient maps of 47 severe burn-related genes; (**D**) Model constructed with 47 genes using LASSO regression; (**E**) Number of optimal classification trees in the RF model; (**F**) Gene importance scores in the RF model; (**G**) Cross-genes selected by three machine learning models.

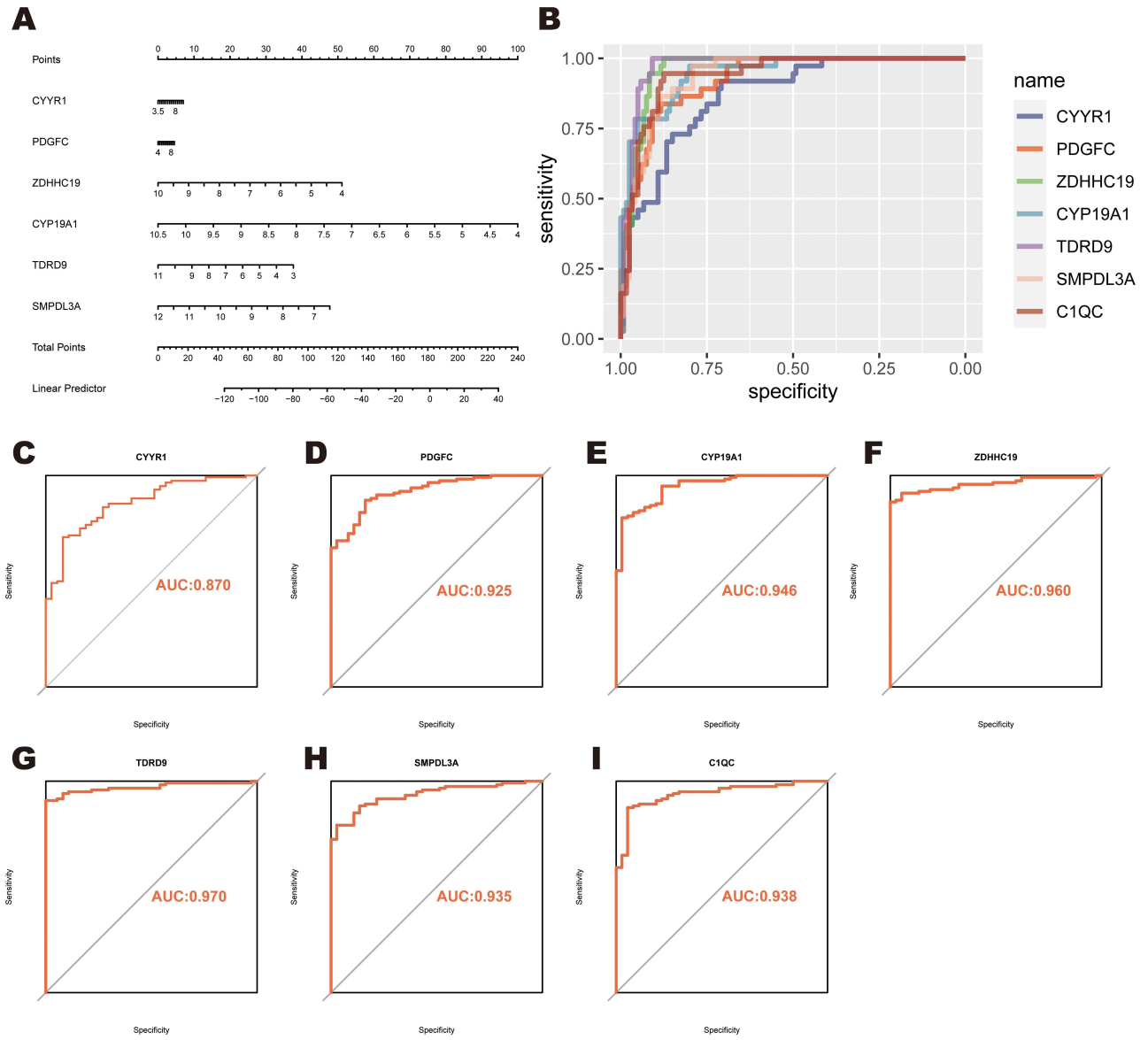

Figure 5 Construction and Validation of the Diagnostic Nomogram. (**A**) Nomogram predicting severe burns; (**B**) ROC analysis of the seven relevant genes; (**C-I**) ROC analysis of relevant genes.

ssGSEA and Gene Expression Differences

ssGSEA of the CYP19A1 gene revealed significant enrichment of arginine biosynthesis and fatty acid biosynthesis in the severe burn group (Figure 6C). Differential analysis of samples from the severe burn gene expression dataset GSE37069 showed that within 48 hours post-injury, the expression level of the CYP19A1 gene in severe burn patients exhibited a statistically significant increase compared to the control group (Figure 6B). In addition, we performed the same analysis using another GEO dataset (GSE19743). The results showed that the expression of the CYP19A1 gene was also significantly increased in the burn group (Figure 6D), further validating our findings.

Drug Prediction

Candidate drugs targeting CYP19A1 gene intervention were predicted using the DSigDB database. The top 10 potential small molecule drugs were listed based on adjusted P-values (Table 1).

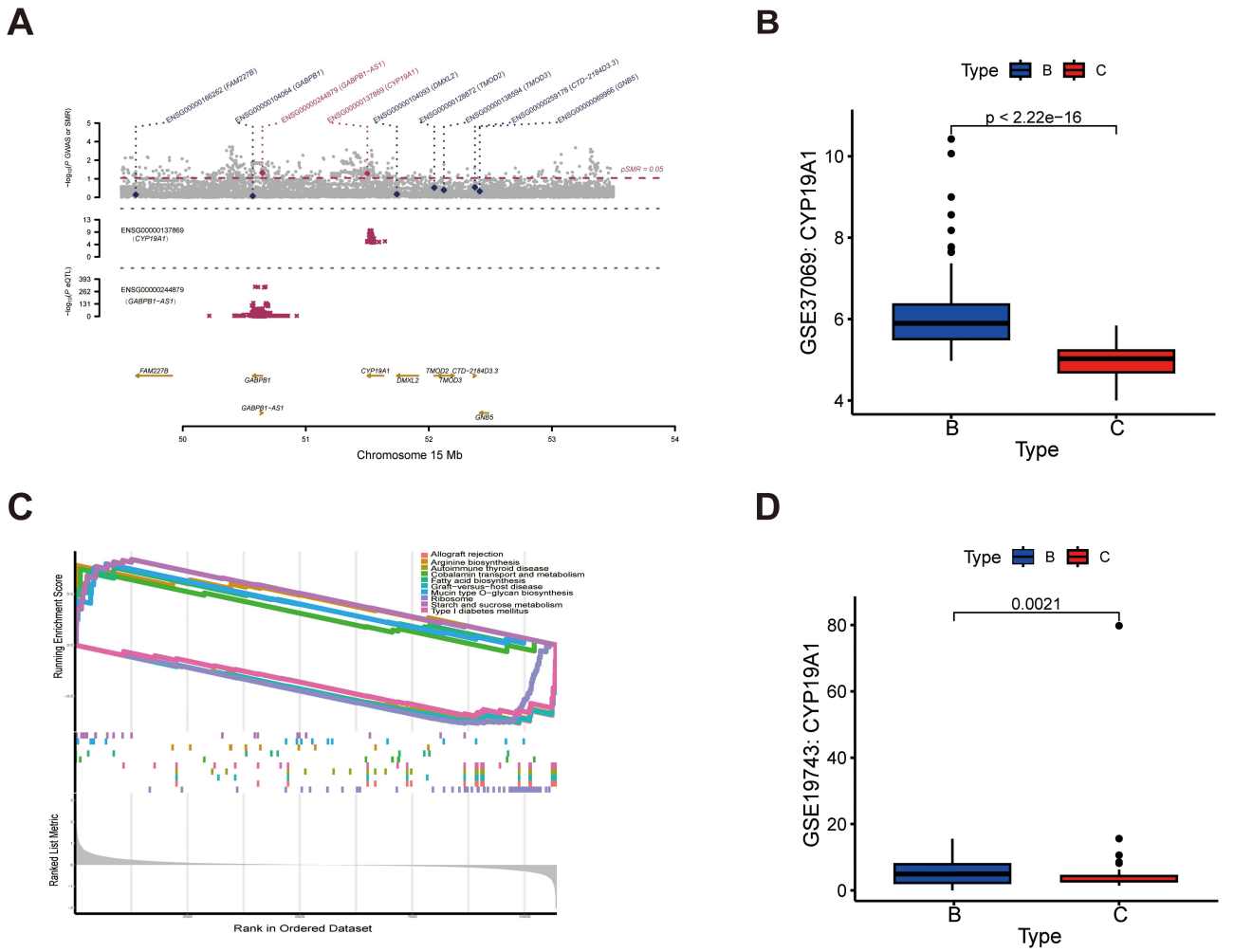

Figure 6 SMR, ssGSEA, and CYP19A1 Gene Expression Differences in Severe Burns. (**A**) Instrumental variable scatter plot for CYP19A1; (**B**) Differential expression of the CYP19A1 gene in the GSE37069 dataset; (**C**) ssGSEA; (**D**) Differential expression of the CYP19A1 gene in the GSE19743 dataset.

Molecular Docking

Molecular docking analysis was conducted to assess the binding affinity of the top 10 candidate drugs with the CYP19A1 gene and understand their pharmacological potential. Structural data for the CYP19A1 gene were obtained from the PDB database (PDB ID: 3S79). Autodock Vina v.1.2.2 was utilized to analyze the binding sites and interactions between the top 10 candidate drugs and the protein encoded by CYP19A1 and to calculate the binding energy for each drug-protein complex. Ultimately, five effective drugs were identified, and their docking results with the protein are presented (Figure 7 and Table 2). Each candidate drug is bound to the protein through electrostatic solid interactions and hydrogen bonds, with all drugs located within the protein's binding pocket (Figure 7). The results indicated that mevastatin exhibited the lowest binding energy with the CYP19A1 protein (−9.0 kcal/mol), demonstrating the most stable binding.

Experimental Validation

Scratch assays and WB were performed to validate the expression of CYP19A1 in severe burns and investigate the effects of mevastatin on CYP19A1. The results of the cell scratch assay demonstrated that, compared to the burn-only group, the 5μM mevastatin intervention group exhibited a more rapid reduction in wound area at both 24 and 48 hours (Figure 8A–C). WB results showed increased expression of the CYP19A1 protein in the burn group, which significantly decreased following intervention with 5 μM mevastatin (Figure 8D and E).

Table 1 Predicted Candidate Drugs from the DSigDB Database

Term	P-value	Adjusted P-value	Genes
Phosalone CTD 00000106	5.50E-04	0.003615	CYP19A1
MALEIMIDE CTD 00001979	5.50E-04	0.003615	CYP19A1
Amoxicillin CTD 00005390	5.50E-04	0.003615	CYP19A1
IPRODIONE CTD 00001594	5.50E-04	0.003615	CYP19A1
DAZOMET CTD 00000672	5.50E-04	0.003615	CYP19A1
Triciribine CTD 00001109	5.50E-04	0.003615	CYP19A1
Mevastatin TTD 00009287	5.50E-04	0.003615	CYP19A1
Vatalanib succinate TTD 00011775	5.50E-04	0.003615	CYP19A1
Bisphenol AF CTD 00005240	5.50E-04	0.003615	CYP19A1
IMAZODAN TTD 00008552	5.50E-04	0.003615	CYP19A1

Discussion

This study first integrates gene differential expression analysis with WGCNA to identify co-expressed gene modules, thereby denoising and capturing gene relationships at various levels. Subsequently, machine learning models were developed to handle large-scale data efficiently, automate feature selection, and capture complex nonlinear relationships. Mendelian randomization was employed to effectively eliminate confounding factors and explore causal relationships between genes and diseases. Additionally, enrichment analysis and the construction of diagnostic nomograms were performed to investigate the biological significance of these drug targets. Ultimately, a hub gene, CYP19A1, was identified. Drug prediction and molecular docking revealed mevastatin as a chemical compound capable of stably binding to the CYP19A1 protein. This provided reliable theoretical support at the molecular level. Furthermore, the clinical therapeutic value of CYP19A1 as a target gene and mevastatin was validated using Western blot and cell scratch assays. We first discovered that mevastatin can promote burn wound healing by downregulating CYP19A1 expression.

The CYP19A1 gene belongs to the cytochrome P450 family and performs an essential part in drug metabolism and synthesizing cholesterol, steroids, and other lipids, and is localized to the endoplasmic reticulum. It is essential for the final stages of converting androstenedione and testosterone into estrone and estradiol (E2). Patients with severe burns experience a rapid increase in various pro-inflammatory cytokines, with levels correlated to the severity of the injury and prognosis. E2 is widely recognized for its ability to improve both cell-mediated and humoral immune responses, increase the cytotoxicity of NK cells, and stimulate the production of pro-inflammatory cytokines such as TNF-α, IL-1β, and IL-6[13,14] while inhibiting the synthesis of anti-inflammatory cytokines like IL-10.[15]

Mevastatin, a statin drug and competitive HMG-CoA reductase inhibitor, is commonly used to lower serum cholesterol. Research indicates that statins not only possess antimicrobial properties and anti-inflammatory but also reduce oxidative stress and improve endothelial cell function, thereby promoting wound healing.[16,17] In chronic refractory wounds, systemic statin use has been shown to enhance the healing of various chronic wounds.[18,19] Studies have noted that topical application of mevastatin can accelerate wound epithelialization by modulating glucocorticoid receptor ligands and inducing long non-coding RNA Gas5, which inhibits the overexpression of c-Myc.[20] Additionally, research has demonstrated that mevastatin facilitates wound healing by mediating EGF-induced Rac1 activation, leading to the migration phenotype of primary human keratinocytes, actin cytoskeleton reorganization, and lamellipodia formation.[21] Our study results indicate that mevastatin promotes the healing of severe burn wounds by regulating the expression of the protein encoded by the CYP19A1 gene, demonstrating significant clinical therapeutic value.

In this study, we presented several novel discoveries in the field of burn research. First, we identified new therapeutic targets for burns by applying Mendelian randomization methods and large-scale genome-wide association study data.

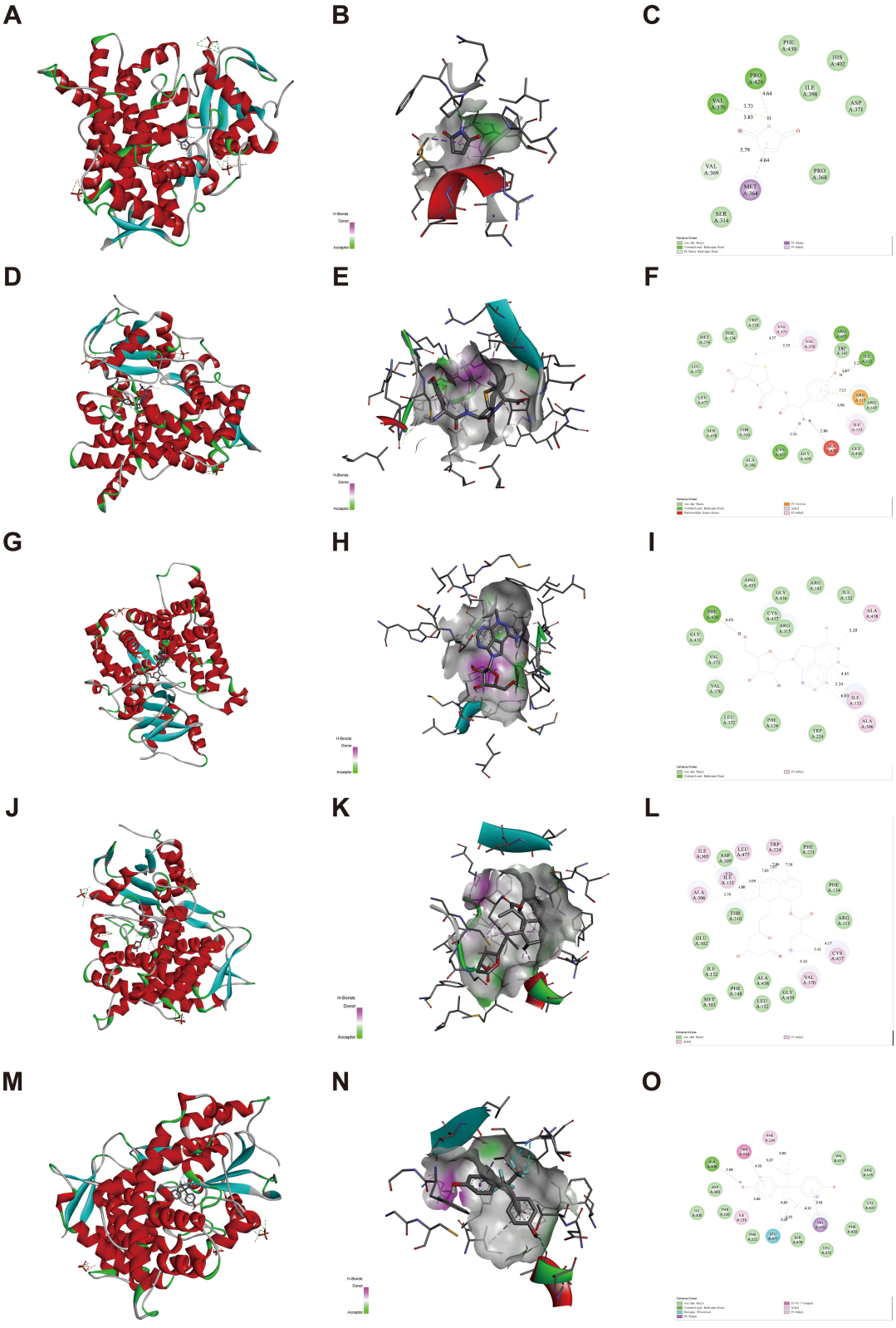

Figure 7 Docking Results of Predictive Drugs with CYP19A1 Protein. (**A-C**) Maleimide; (**D-F**) Amoxicillin; (**G-I**) Triciribine; (**J-L**) Mevastatin; (**M-O**) Bisphenol AF.

Table 2 Binding Energies of Predicted Drugs with CYP19A1 Gene Expression Protein

Drug	PubChem ID	rmsd l.b	Binding energy (kcal/mol)
Maleimide	10935	0.110	−4.1
Amoxicillin	33613	1.940	−7.5
Triciribine	65399	0.948	−6.8
Mevastatin	64715	1.694	−9.0
Bisphenol AF	73864	0.139	−8.2

This method combines cis-eQTL data with burn-related data to establish causal links between exposures and outcomes, thereby enhancing the accuracy of drug efficacy predictions. Second, we combined gene differential expression analysis, WGCNA, machine learning, and Mendelian randomization across multiple omics methods to ensure the consistent significance of Hub genes across these approaches, thus increasing result robustness and reducing the likelihood of false positives. Finally, we validated the pharmacological activity of potential burn drug targets by utilizing drug prediction and molecular docking, assessing the interaction patterns and binding affinity between proteins and drugs, further confirming the feasibility and potential efficacy of the drugs. This study involves a thorough assessment method, including identifying genes and examining their binding characteristics with drugs, proposing a hub gene for severe burns, and providing strong evidence.

However, this study has several limitations. First, the severe burn datasets obtained from the GEO database lack comprehensive clinical information about the patients, preventing us from assessing the associations between target genes, drugs, and clinical outcomes. Second, The Mendelian randomization analysis utilizing the GTEx dataset is constrained by a very modest sample size, potentially impacting the statistical power of the associated genes. Additionally, molecular docking can only simulate binding affinities and cannot model the actual molecular dynamics. Although we identified satisfactory hub genes and drugs through multi-omics approaches and validated them in cellular experiments, further in vitro studies and clinical trials are needed to confirm these findings and elucidate the specific molecular mechanisms.

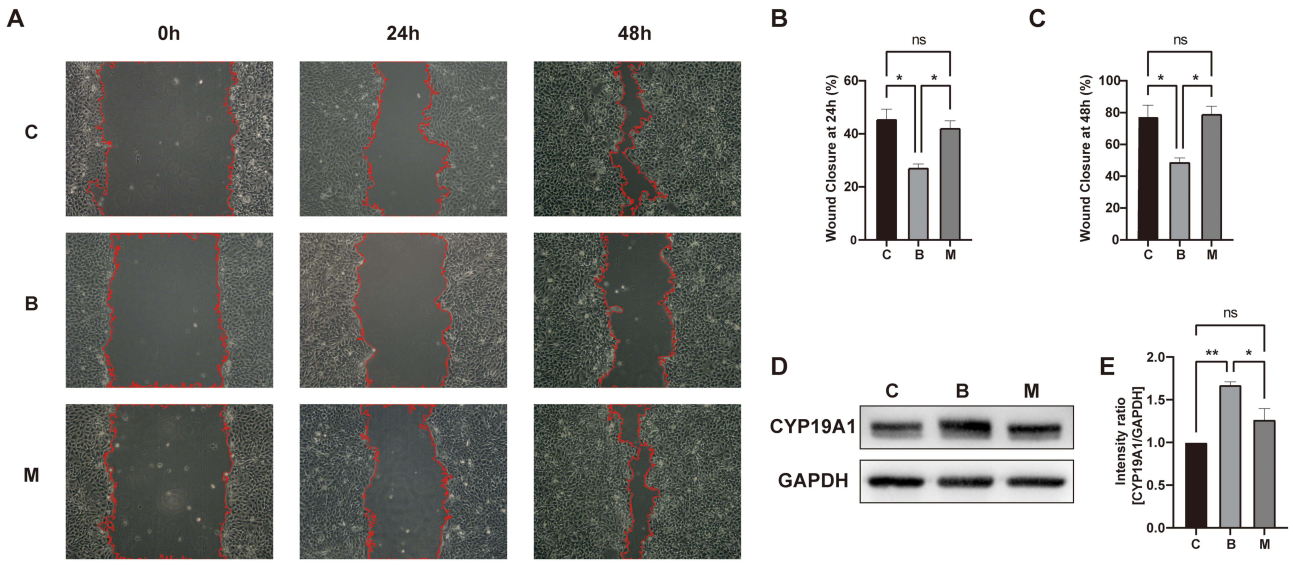

Figure 8 WB and scratch assays. (**A**) Images of cell scratch assay at 0 hours, 24 hours, and 48 hours for the control group, burn group, and mevastatin group; (**B, C**) Scoring of scratch area based on the time after the cell scratch assay; (**D**) Western blot analysis of CYP19A1 protein in samples from the control group, burn group, and mevastatin group; (**E**) Western blot analysis scoring of CYP19A1 protein. Data are presented as mean ± standard error (SE), and statistical analysis was performed using the *t*-test. Representative results of three independent experiments are shown. Scale bar, 100 μm. Significance: * indicates $p < 0.05$; ** indicates $p < 0.005$; 'ns' indicates not significant.

Conclusion

These findings provide new insights and potential methods for more effective severe burn treatment. This study successfully identified the hub gene CYP19A1 for severe burns through the integration of multi-omics approaches, suggesting its potential as an effective target for treating severe burn wounds. Additionally, we confirmed mevastatin as a stable binding drug for CYP19A1 through drug prediction, molecular docking, and experimental validation. We found that mevastatin promotes the healing of severe burn wounds by reducing CYP19A1 expression.

Acknowledgments

We analyzed publicly available datasets from the GEO Research Network (http://www.ncbi.nlm.nih.gov/geo).

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Disclosure

The authors declare that they have no competing interests.

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