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Retraction

Retracted: The Nine RNA Methylation Regulatory Gene Signature Is Associated with the Pathogenesis of Atrial Fibrillation by Modulating the Immune Microenvironment in the Atrial Tissues

Disease Markers

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This article has been retracted by Hindawi, as publisher, following an investigation undertaken by the publisher [1]. This investigation has uncovered evidence of systematic manipulation of the publication and peer-review process. We cannot, therefore, vouch for the reliability or integrity of this article.

Please note that this notice is intended solely to alert readers that the peer-review process of this article has been compromised.

Wiley and Hindawi regret that the usual quality checks did not identify these issues before publication and have since put additional measures in place to safeguard research integrity.

We wish to credit our Research Integrity and Research Publishing teams and anonymous and named external researchers and research integrity experts for contributing to this investigation.

The corresponding author, as the representative of all authors, has been given the opportunity to register their agreement or disagreement to this retraction. We have kept a record of any response received.

References

[1] Q. Wang, S. Zhang, X. Xu et al., "The Nine RNA Methylation Regulatory Gene Signature Is Associated with the Pathogenesis of Atrial Fibrillation by Modulating the Immune Microenvironment in the Atrial Tissues," *Disease Markers*, vol. 2023, Article ID 7277369, 16 pages, 2023.

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Research Article

The Nine RNA Methylation Regulatory Gene Signature Is Associated with the Pathogenesis of Atrial Fibrillation by Modulating the Immune Microenvironment in the Atrial Tissues

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Background. Atrial fibrillation (AF) is the most common type of cardiac arrhythmias and a major cause of cardiovascular disease (CVD)-related deaths globally. RNA methylation is the most frequent posttranscriptional modification in the eukaryotic RNAs. Previous studies have demonstrated close associations between the status of RNA methylation and CVD. *Methods*. We comprehensively evaluated the relationship between RNA methylation and AF. Least absolute shrinkage and selection operator (LASSO) logistic regression analysis was used to establish a risk score model in AF. Biological functional analysis was used to explore the relationship between RNA methylation related signatures and immune microenvironment characteristics. Machine learning was used to recognize the outstanding RNA methylation regulators in AF. *Results*. There was a significant variant of the mRNA expression of RNA methylation regulators in AF. RNA methylation related risk score could predict the onset of AF and closely associated with immune microenvironment features. XG-Boost algorithm and SHAP recognized that NSUN3 and DCPS might play a key role in the development of AF. Meanwhile, NSUN3 and DCPS had potential diagnostic value in AF. *Conclusion*. RNA methylation regulatory genes are associated with the onset of AF by modulating the immune microenvironment. The nine AF risk-related RNA methylation regulatory gene signature is a potential diagnostic biomarker and therapeutic target for AF.

1. Background

Atrial fibrillation (AF) is the most common type of cardiac arrhythmias and is associated with increased risk of cardiovascular disease- (CVD-) related deaths worldwide [1]. AF is associated with increased risk of stroke, myocardial infarction, dementia, and heart failure [2]. Currently, catheter ablation and antiarrhythmic drugs are the main treatment modalities for patients diagnosed with AF [3, 4]. However, catheter ablation normalizes the heart rhythm in only a small fraction of AF patients, whereas antiarrhythmic drugs such as amiodarone, sotalol, propafenone, and flecainide show low efficacy and significantly higher adverse effects [5, 6]. Although several studies have investigated the underlying cellular and molecular mechanisms that regulate AF, specific mechanisms that modulate development and progression of atrial fibrillation are unclear [4]. Furthermore, characterizing the underlying pathogenetic mechanisms of AF is necessary for developing novel and effective targeted therapies.

RNA methylation is the most prevalent posttranscriptional modification of the eukaryotic RNAs [7, 8]. N6-methyladenosine (m6A), N1-methyladenosine (m1A), 5-methylcytosine (m5C), and 7-methylguanosine (m7G) are the most common types of RNA methylation [8]. The regulators of RNA methylation modifications are categorized based on the biological functions as "writers" (methyltransferases), "readers" (recognition and binding to the methylated mRNAs), and "erasers" (demethylases) [9]. RNA methylation

status regulates the RNA processing mechanisms such as nuclear export, RNA translation, splicing, and processing of the noncoding RNAs [10]. Genetic studies have demonstrated that RNA methylation is an epigenetic modification that regulates gene expression without altering the coding sequence of the genes [11, 12]. RNA methylation also plays a significant role in several physiological and pathophysiological processes, including embryonic development and carcinogenesis [13, 14]. The changes in RNA methylation are closely associated with the onset and development of CVD [15]. Furthermore, dysregulated m6A methylation is often linked to CVDs [16, 17]. RNA methylation enzymes and their targets are potential diagnostic biomarkers and therapeutic targets in several human diseases including CVDs. However, the roles of specific RNA methylation regulatory mechanisms in the onset and progression of AF are not clear and require further investigation.

Therefore, in this study, we comprehensively analyzed the expression levels of the RNA methylation regulatory genes in the left atrial tissues of patients with AF and sinus rhythm (SR) from the public GEO datasets to identify potential risk or protective RNA methylation regulatory genes in AF. The LASSO Cox regression analysis was used to develop a nine AF risk-related RNA methylation regulatory gene model. The diagnostic performance of the risk score in predicting AF and the underlying changes in the immune microenvironment was analyzed using the ROC curves. The XG-Boost machine learning algorithm was used to identify critical RNA methylation genes that are associated with the onset of AF.

2. Methods

2.1. Publicly Available AF Patient Datasets. The mRNA expression profiling of AF and sinus rhythm (SR) patient tissues and the corresponding clinical information was extracted and analyzed from the GSE115574, GSE79768, GSE41177, and GSE14975 datasets based on the inclusion and exclusion criteria of GEO (http://www.ncbi.nlm.nih.gov/geo/) databases. Inclusion criteria: datasets involving human left atrial appendage (LAA) samples from the AF and SR patients. Exclusion criteria: datasets with a sample size smaller than 10 [18–21] (Table S1). In each dataset, only the LAA samples from the AF and SR patients were selected. Finally, 42 AF and 29 SR tissues were included in this study. Subsequently, the four gene expression matrix files were merged into one merge cohort. Based on previous studies, the combat algorithm in "sva" package is used to remove batch effect [22]. Principal component analysis (PCA) was used to assess the batch effects and the results of the batch effect removal methods (Supplementary Figure 1).

2.2. Functional Enrichment Analysis of Biological Functions and Pathways. The comprehensive functional enrichment analysis was performed using the Metascape database (http://metascape.org/) [23]. The enriched KEGG (Kyoto Encyclopedia of Genes and Genomes) pathway and GO (Gene Ontology) terms associated with biological processes (BP), molecular functions (MF), and cellular components (CC) were analyzed based on GSEA (Gene Set Enrichment Analysis) using the Bioconductor "clusterProfiler" R package [24].

2.3. AF Risk Score Model Building and Validation. Univariate logistic regression analysis was performed to identify the potential AF risk-related genes among the 72 RNA methylation regulatory genes analyzed. Then, 20 potential AF risk-related RNA methylation regulatory genes from the univariate analysis were entered into the least absolute shrinkage and selection operator (LASSO) regression analysis for dimension reduction, selection of features, and calculation of the risk score. The risk score for each sample in the merged cohort and the individual GEO datasets was calculated using the following formula:

$$\text{Risk score} = \sum_{i=1}^{n} \text{Coefi} * \text{xi}. \quad (1)$$

Coefi denotes the coefficient, and xi represents the expression level for each of the selected RNA methylation regulators.

2.4. XG-Boost Machine Learning Algorithm. XG-Boost (eXtreme Gradient Boosting) and SHAP (Shapley additive explanation) algorithms were used to determine the association between the expression levels of the RNA methylation regulatory genes and the onset of AF [25, 26]. According to the literature, XG-boost can also perform a good fit with a sample size of 100 when using a tree model that classifies with a boosting method. An element of regularization is added to the cost function to prevent overfitting and minimize model complexity. SHAP values are used to interpret the machine learning (ML) models and identify ML models, which are black boxes. Lundberg et al. developed the SHAP framework after analyzing several contemporary algorithms to determine the importance of various features that belonged to the same class of measures. Previously, it was difficult to interpret the prediction cases because most machine learning algorithms provided predictors based on the importance of global features. The SHAP technique calculates the contribution of each input variable towards the decision of the machine learning algorithms. The model predictions are interpreted based on the SHAP values, which are obtained from the "shap" package [27].

2.5. CIBERSORT Analysis. The infiltration scores of the 22 different immune cell types were calculated for each patient included in the study using the "CIBERSORT" R package. CIBERSORT uses gene markers of various immune cells to predict the infiltration levels of 22 types of immune cells in the samples. The immune cells analyzed included naïve B cells, memory B cells, plasma cells, CD8+ T cells, naïve CD4+ T cells, resting, memory, and activated CD4+ T cells, follicular helper T cells, regulatory T cells, gamma-delta T cells, resting and activated NK cells, monocytes, M0, M1, and M2 macrophages, resting and activated dendritic cells, resting and activated mast cells, eosinophils, and neutrophils [28].

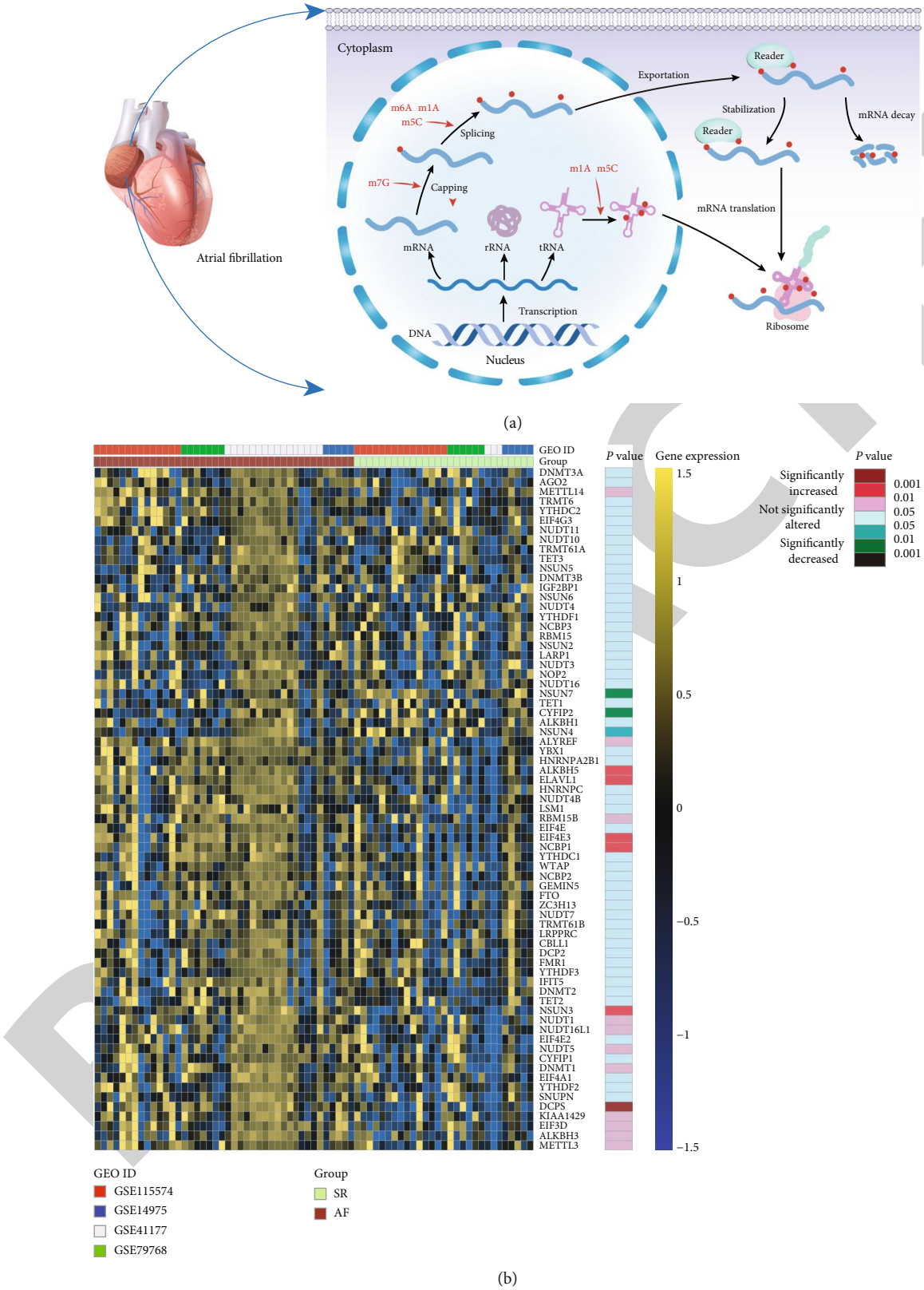
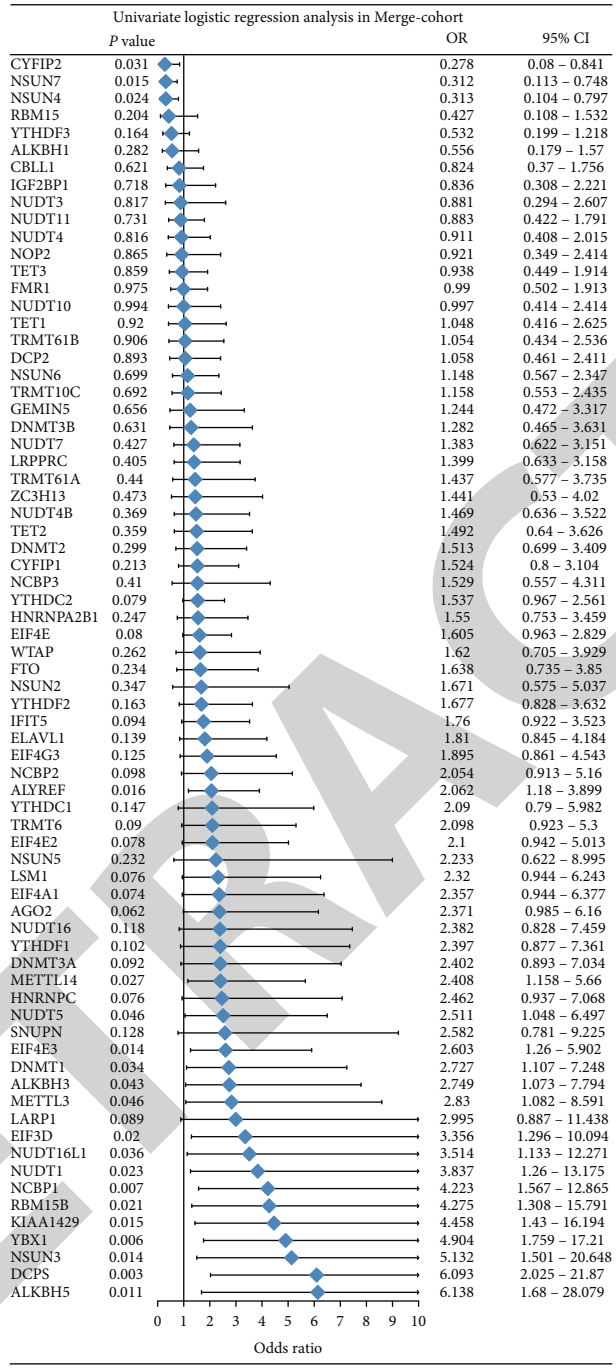


Figure 1: The RNA methylation regulatory mechanism and the expression profiles of RNA methylation regulatory genes in the atrial tissues of patients with atrial fibrillation. (a) Diagrammatic representation shows the various steps of RNA methylation modifications in the AF cells. (b) The mRNA expression levels of 72 RNA methylation regulators in the SR ($n = 29$) and AF ($n = 42$) samples of the merged cohort from the GEO datasets. Note: AF: atrial fibrillation; SR: sinus rhythm.



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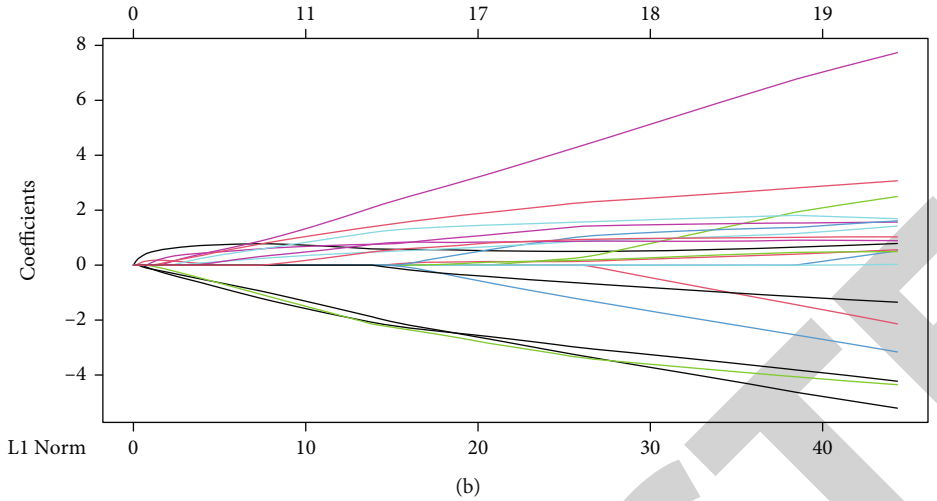

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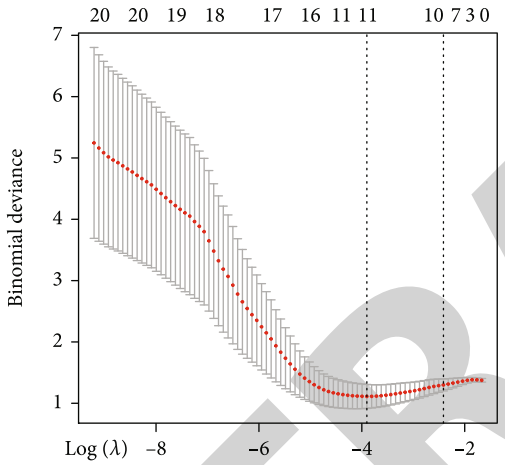

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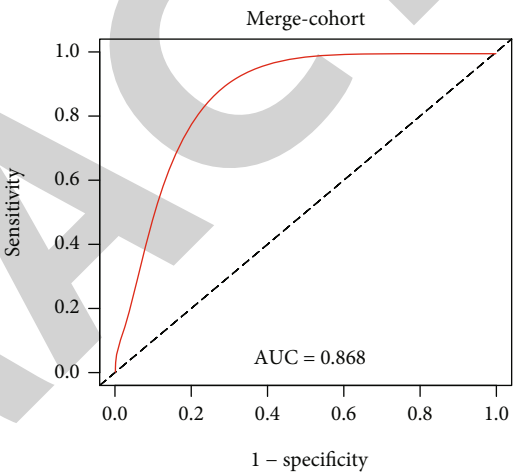

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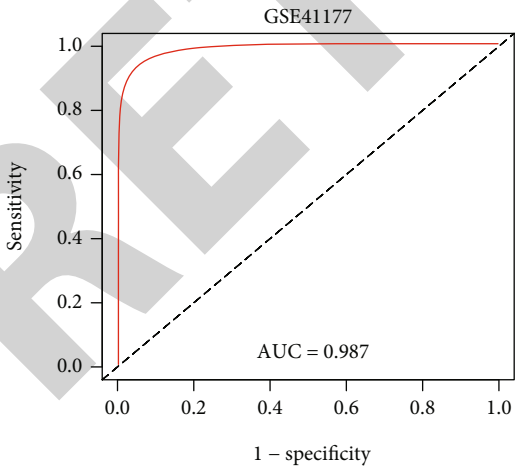

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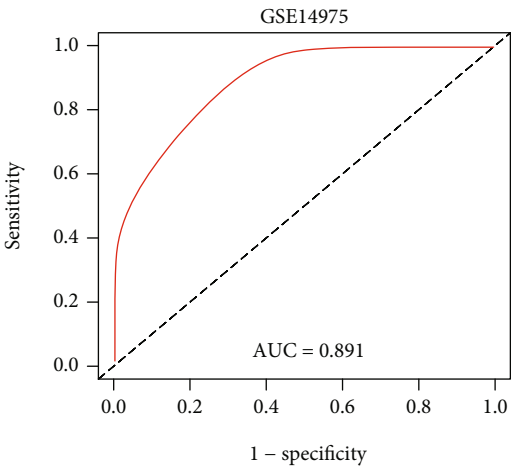

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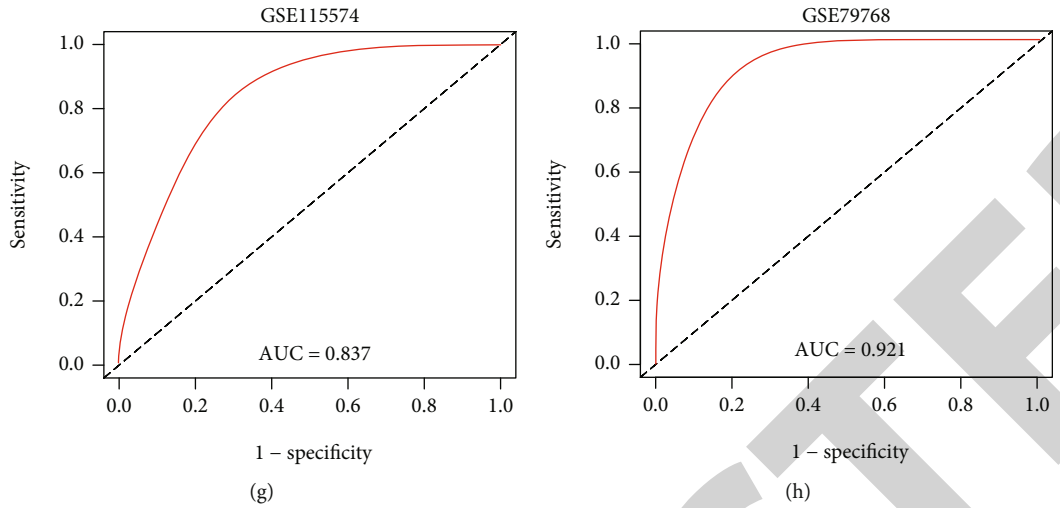


Figure 2: Characterization of the nine AF risk-related RNA methylation regulatory gene signature. (a) Univariate logistic regression analysis shows the expression levels of RNA methylation regulatory genes in the AF samples of the merge cohort. The data is represented as odds ratio (OR) with 95% confidence interval (95% CI). The regulatory genes with P values below 0.05 represent those with significant differences in expression levels between AF and SR samples. (b) LASSO coefficient profiles of 20 RNA methylation regulatory genes. (c) Selection of the tuning parameter (lambda, λ) using 10-fold cross-validation wherein the binomial deviances from the LASSO regression cross-validation model were plotted against log (λ). (d–h) ROC curve analyses show the diagnostic power of the 9-AF-related gene risk score in the (d) merge cohort, (e) GSE41177, (f) GSE14975, (g) GSE115574, and (h) GSE79768 datasets.

2.6. Statistical Analysis. R statistical computing language (version 4.0.4) was used for all statistical analysis unless otherwise mentioned. Student's t-tests (unpaired, two-tailed) followed by chi-square tests and rank-sum tests were used to compare the data between two independent groups for all the quantitative variables. Univariate and multivariate logistic regression analysis was performed to identify the risk factors associated with AF. The statistical significance level was set at $P > 0.05$. ($^{*}P < 0.05$, $^{**}P < 0.01$, and $^{***}P < 0.001$).

3. Results

3.1. RNA Methylation Regulatory Genes Are Dysregulated in the Atrial Tissues of Patients with AF. The pre and poststandardization analysis of the mRNA expression profiling was performed as summarized in Supplementary Figure 1. The gene expression data of 42 AF and 29 SR patients from the GEO datasets was merged and further analyzed after adjusting for the batch effects. Figure 1(a) shows the summary of the known intracellular RNA methylation mechanisms, their localization (nuclear or cytoplasmic), and their effects on RNA metabolism such as capping and splicing of mRNAs in the nucleus, export of mRNAs from the nucleus to the cytoplasm, mRNA translation, ribosomal assembly, and mRNA decay. The analysis of differentially expressed genes (DEGs) in the merged cohort from the GEO datasets demonstrated that genes regulating RNA modifications such as m6A, m5C, m1A, and m7G were frequently dysregulated in the AF patient tissues compared with the SR samples (Figure 1(b)). The left atrial tissues of AF patients showed significantly higher expression levels of the *DNMT1*, *NUDT1*, *NUDT16L1*, *DCPS*, *KIAA1429*, *EIF3D*, *ALKBH3*, *METTL3*, *RBM15B*, *METTL14*, *ALYREF*, *EIF4E3*, *NCBP1*, *ALKBH5*, *YBX1* and *NSUN3* mRNAs compared to the left atrial tissues of the SR patients. Furthermore, the expression levels of *NSUN7*, *CYFIP2*, and *NSUN4* were significantly downregulated in the atrial tissues of AF patients compared to those of the SR patients. Then, we performed integration analysis to investigate the functions of the RNA methylation regulatory genes in AF. PPI network analysis showed significant interactions between proteins associated with RNA methylation (Supplementary Figure 2A). In addition, we conducted Pearson's correlation analysis on the expression profiles of 71 regulators in a merge dataset. These results indicated that most of RNA methylation related regulators were positive correlations (Supplementary Figure 2B). The mRNA expression profiling analysis of the 72 RNA methylation regulatory genes in the merged cohort demonstrated significant association between AF and several RNA methylation regulatory genes except NSUN6, CYFIP2, DNMT3B, TET3, NSUN5, TRMT61A, DNMT3A, NUDT10, and IGF2BP1. Overall, these data demonstrated that dysregulation of RNA methylation regulatory genes was associated with the onset of AF.

3.2. RNA Methylation Regulatory Genes Are Associated with Atrial Fibrillation. We performed further bioinformatics analysis to determine the relationship between the RNA methylation regulatory genes and the pathogenesis of atrial fibrillation. Univariate logistic regression analysis demonstrated that 20 RNA methylation regulatory genes were associated with AF (Figure 2(a)). Among these, CYFIP2, NSUN7, and NSUN4 were potential protective factors, whereas KIAA1429, YBX1, NSUN3, DCPS, ALKBH5, and 12 other genes were potential risk factors of AF. Furthermore, LASSO regression analysis was performed using the 20 AF-related RNA methylation regulatory genes to identify the prognostic-associated risk genes (Figures 2(b) and 2(c)).

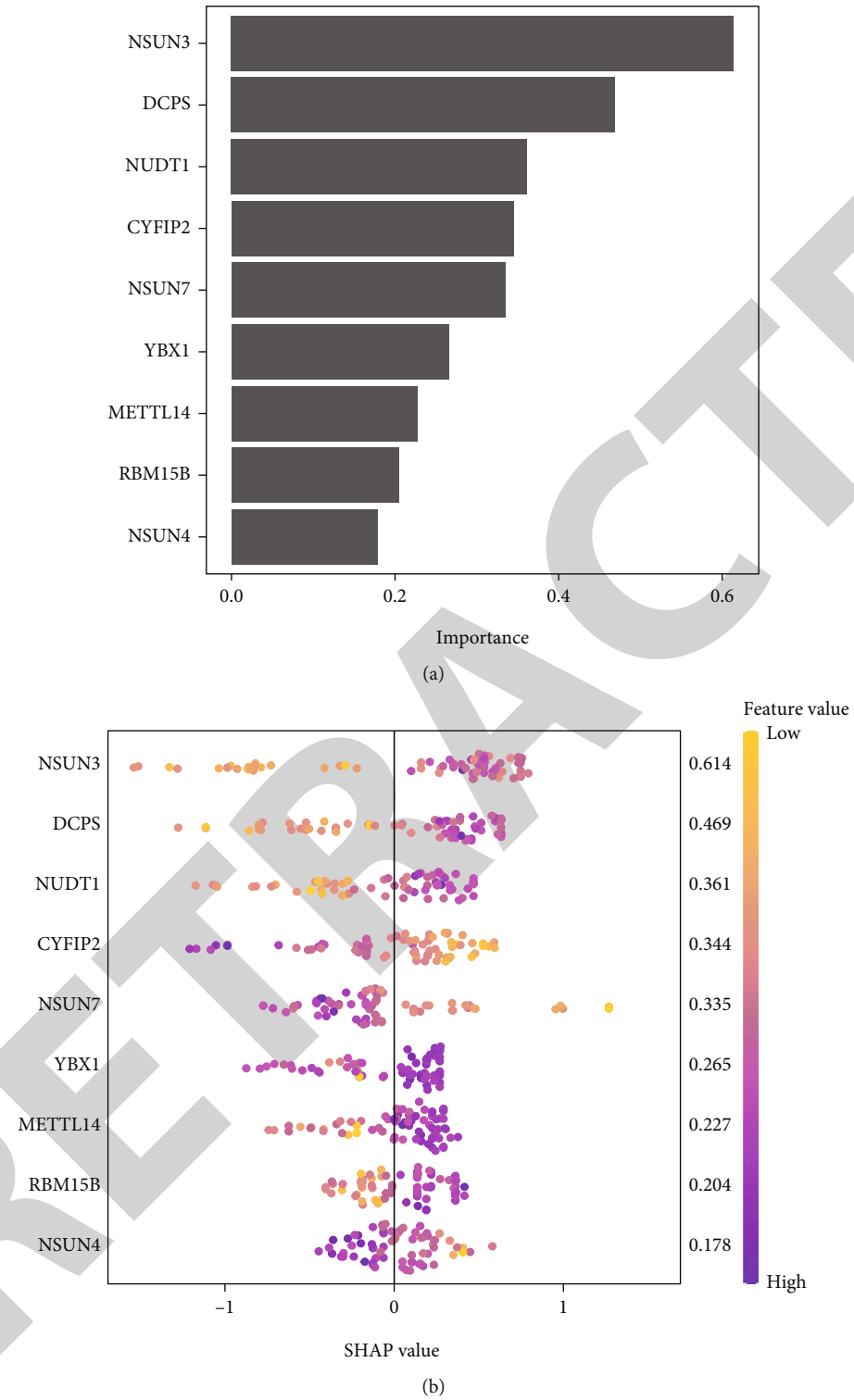
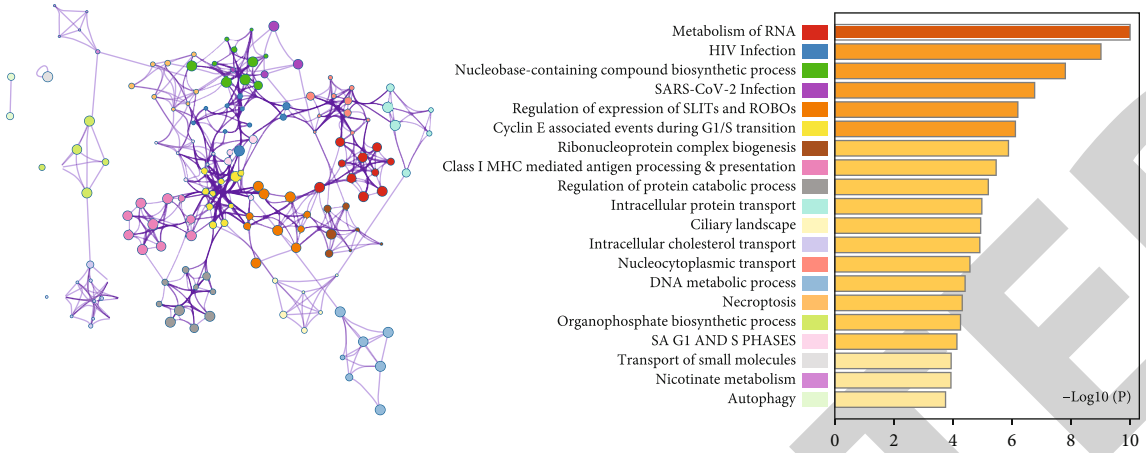


Figure 3: Screening of critical AF-related RNA methylation regulatory genes using the XG-Boost algorithm. (a) Bar chart shows evaluation of the importance of the RNA methylation regulatory genes to AF based on the analysis using the XG-Boost algorithm. (b) SHAP summary plot shows the top nine RNA methylation regulatory genes based on the SHAP values evaluated by the XG-Boost algorithm.

Lasso regression analysis showed that 9 out of the 20 RNA methylation regulatory genes (DCPS, YBX1, NSUN3, NSUN7, RBM15B, NUDT1, NSUN4, METTL14, and CYFIP2) were significant risk factors for AF. Therefore, we constructed a 9 AF-related RNA methylation regulatory gene risk signature model based on the minimum criterion (Figure 3(b)) to accurately distinguish AF patients from non-AF or SR patients based on the RNA methylation modifications in the atrial tissues. The risk score was calculated for the merged cohort and all the individual



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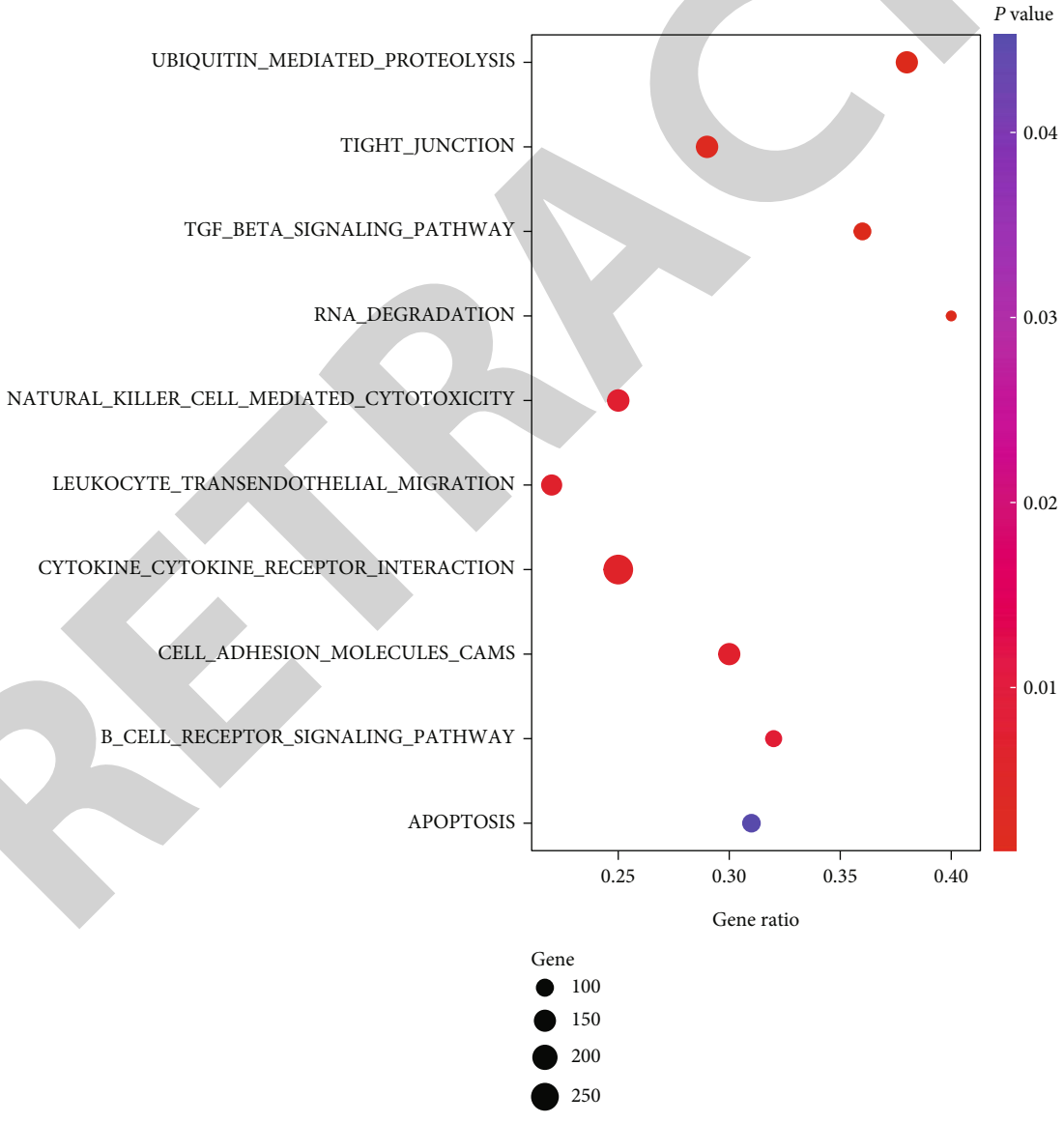

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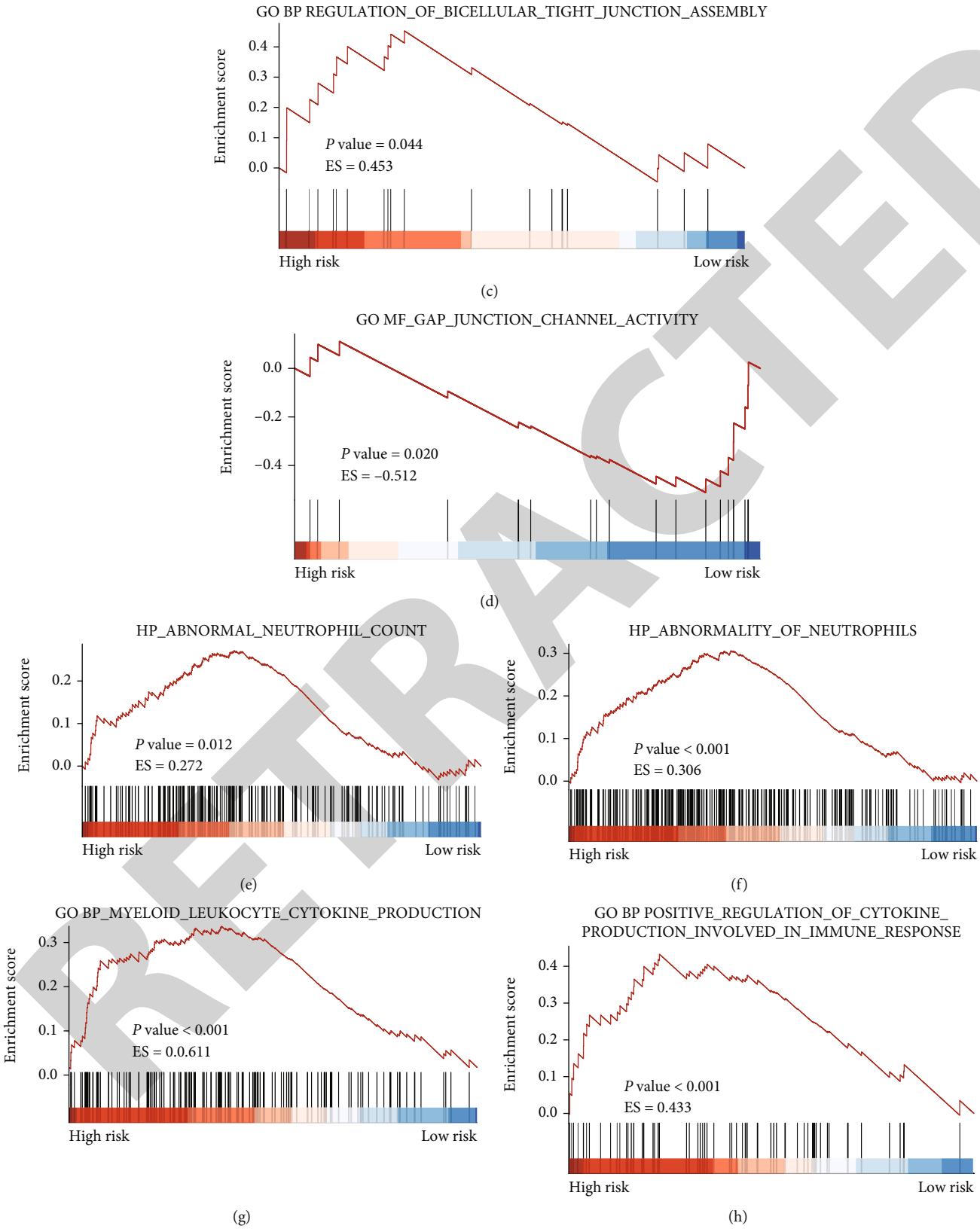

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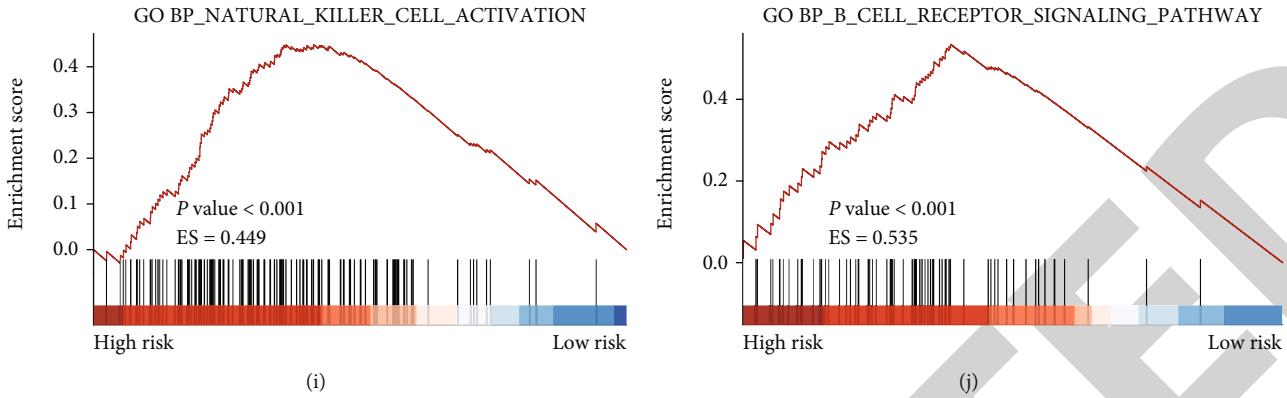


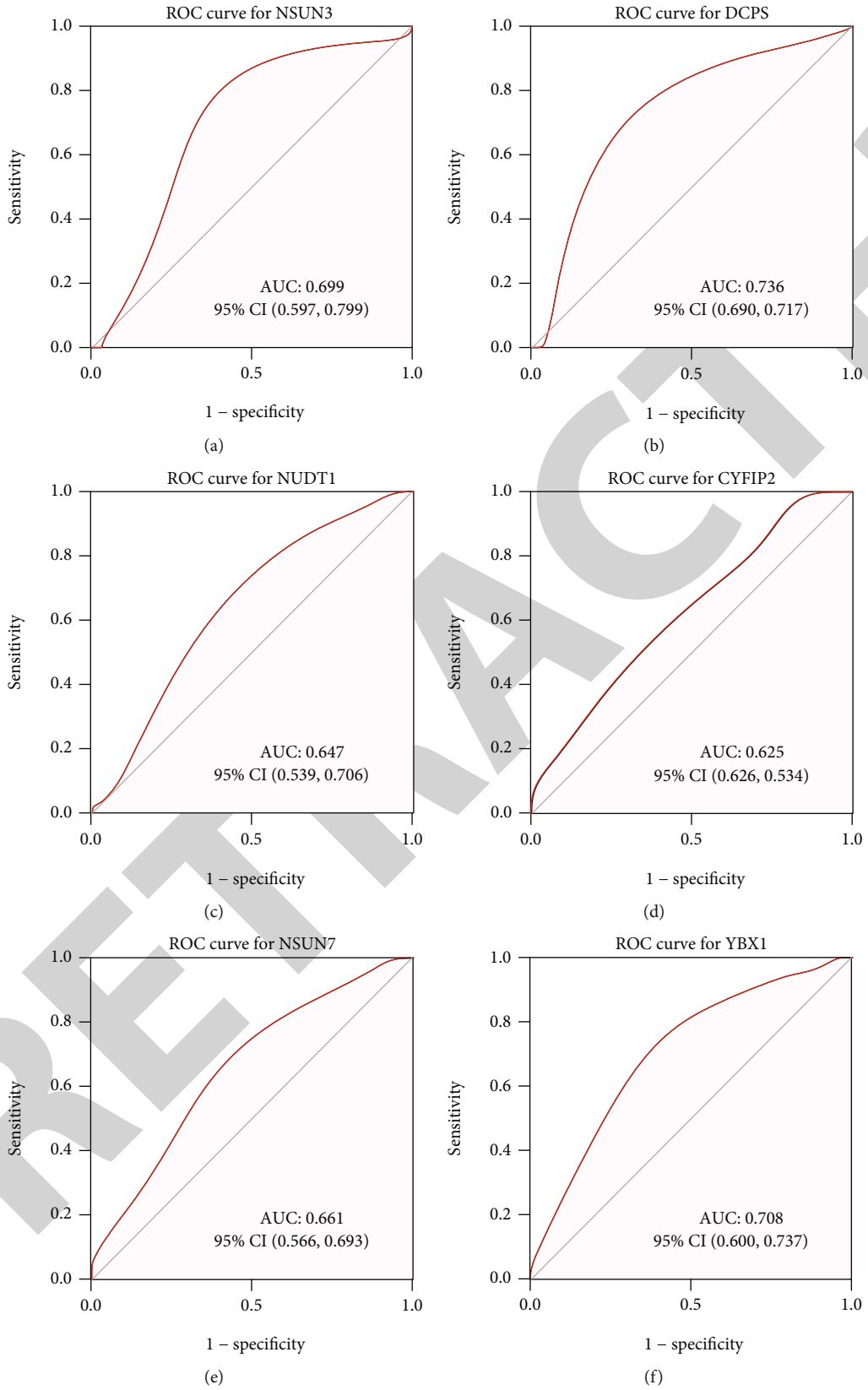
Figure 4: Functional annotation of the AF risk-related RNA methylation regulatory genes. (a) Protein-protein interaction network analysis and the bar chart showing the top 20 significantly enriched biological processes related with the AF risk-related RNA methylation regulatory genes. (b) KEGG pathway enrichment analysis of the differentially expressed genes (DEGs) between the low- and high-risk AF patients. (c–j) GSEA enrichment analysis of the DEGs between low- and high-risk AF patients in merge cohort shows significantly enriched biological processes (BP) and molecular functions (MF).

GEO datasets using the following equation: Risk score = (0.631 × expression of DCPS) + (0.149 × expression of YBX1) + (0.340 × expression of NSUN3) + (−0.314 × expression of NSUN7) + (0.022 × expression of RBM15B) + (0.162 × expression of NUDT1) + (−0.394 × expression of NSUN4) + (0.169 × expression of METTL14) + (−0.226 × expression of CYFIP2). ROC curve analysis was then performed to determine the diagnostic performance of the 9-gene risk score. The risk score showed good performance in distinguishing SR and AF samples in the merged cohort (AUC = 0.868), GSE41177 (AUC = 0.987), GSE14975 (AUC = 0.891), GSE115574 (AUC = 0.837), and GSE79768 (AUC = 0.921) datasets (Figures 2(d)–2(h)). In summary, the 9-gene risk score based on the expression levels of 9 RNA methylation regulatory genes is a potential predictive factor of AF. This risk gene model may be used to accurately stratify AF patients, develop novel targeted therapy for AF, and estimate the therapeutic responses of AF patients.

3.3. AF Risk-Related RNA Methylation Regulatory Genes Are Associated with the Infiltration of Inflammatory Immune Cells in the Atrial Tissues of AF Patients. Next, the top 500 dysregulated genes or differentially expressed genes (DEGs) in the AF samples with high-risk scores were selected, and their biological functions were annotated. The DEGs in the high-risk score AF samples were significantly enriched in pathways related to DNA, RNA and protein metabolism, HIV and SARS-CoV-2 viral infections, MHC class I-mediated antigen processing and presentation, cyclin E-associated events during G1/S transition, and necroptosis (Figure 4(a)). The top 20 significantly enriched biological processes are shown in Figure 4(a). Furthermore, KEGG pathway analysis showed that the high-risk score AF samples were significantly enriched in ubiquitin-mediated proteolysis, tight junctions, TGF-beta signaling pathway, RNA degradation pathways, and immune related pathways (Figure 4(b)). GSEA analysis showed that "regulation of bicellular tight junction assembly" was significantly enriched in the high-risk AF samples, and "gap junction channel activity" was significantly enriched in the low-risk AF samples (Figures 4(c) and 4(d)). Moreover, GSEA data showed that high-risk AF samples were significantly enriched in the pathways related to "abnormality of neutrophils", "cytokine-cytokine receptor interactions", "leukocyte transendothelial migration", and "natural killer cell mediated cytotoxicity" (Figures 4(e)–4(j)). These findings suggested that the RNA methylation modulatory genes were associated with the infiltration of immune cells in the atrial tissues of AF patients.

3.4. XG-Booster ML Analysis Shows That NSUN3 and DCPS Play a Key Role in AF Pathogenesis. The main AF-related RNA methylation regulatory genes were identified by analyzing the RNA methylation regulatory genes using the XG-Boost algorithm and the SHAP values. XG-Boost analysis showed that NSUN3, DCPS, NUDT1, CYFIP2, and DNMT2 were the top five regulators of AF (Figure 3(a)). Figure 3(b) shows the ranking of risk-related RNA methylation regulatory genes based on the feature importance, SHAP values, and the XG-Boost method. Each point in Figure 3(b) represents a feature value for individual AF patients with the x-axis denoting the SHAP value, and the color depth representing the feature value. The variables (AF risk-related RNA methylation regulatory genes) were ranked according to the sum of the SHAP values for all the samples. *NSUN3* and *DCPS* were the most important AF risk-related RNA methylation regulatory genes according to the XG-Boost analysis and the SHAP values. Furthermore, the nine key RNA methylation regulatory genes were validated in the merged cohort using the ROC curve analysis (Figure 5). The AUC values for *DCPS* (AUC = 0.736), *YBX1* (AUC = 0.708), and *NSUN3* (AUC = 0.699) showed good discriminative ability. Combining the results above, we found that NSUN3, DCPS, and YBX1 might play critical roles in AF pathogenesis.

3.5. Correlation Analysis between RNA Methylation Related Regulatory Genes and Infiltration of Immune Cell Types. CIBERSORT analysis showed positive correlation between the risk score of patients with AF and the infiltration levels



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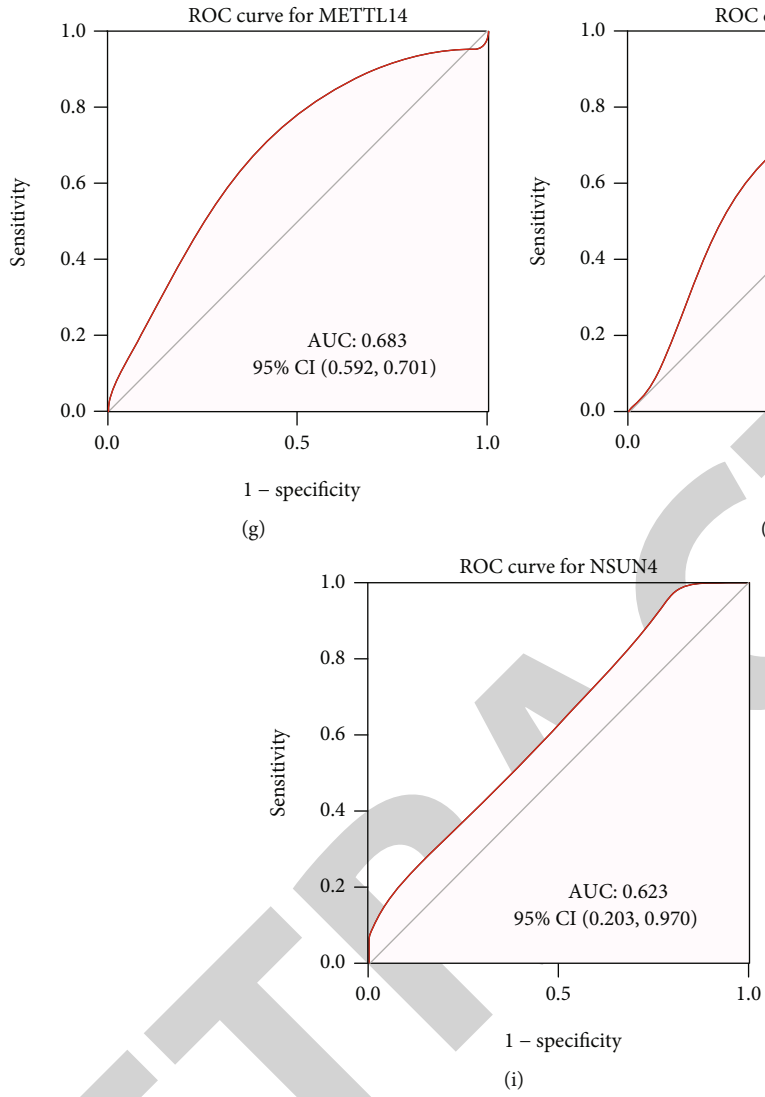


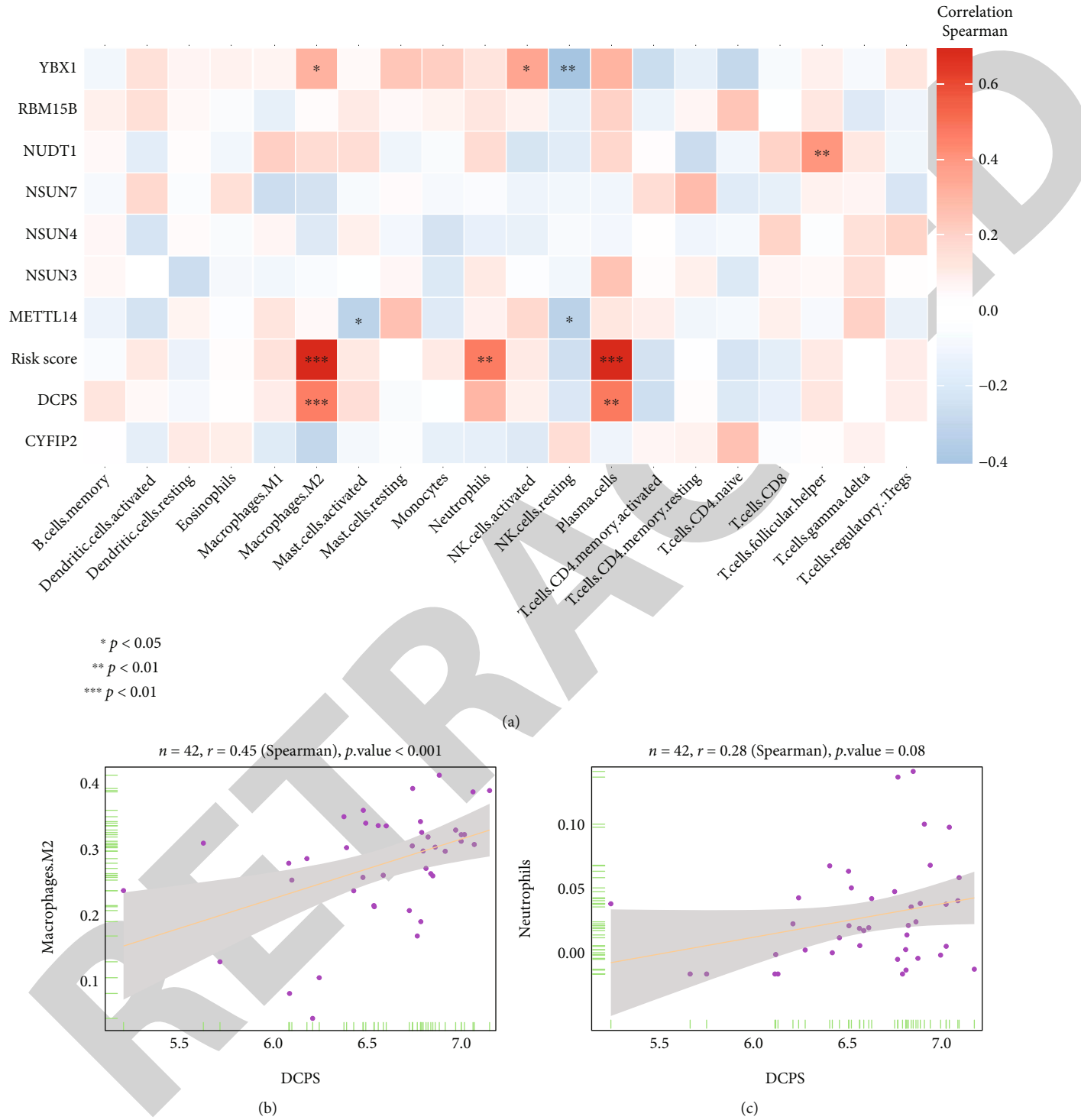
Figure 5: ROC curve analysis of the 9 AF risk-related RNA methylation regulatory genes.(a–f) ROC curves for the analysis of diagnostic power based on the expression levels of (a) NSUN3, (b) DCPS, (c) NUDT1, (d) CYFIP2, (e) NSUN7, (f) YBX1, (g) METTL14, (h) RBM15B, and (i) NSUN4 genes in the AF and SR samples of the merge cohort. The area under the ROC curve (AUC) values for each AF risk-related RNA methylation regulatory gene is indicated in each ROC curve plot.

of plasma cells, neutrophils, and M2 macrophages in the atrial tissues (Figure 6). The results of the correlation analysis were consistent with the findings of the functional enrichment pathway analysis. Furthermore, DCPS expression levels showed significant positive correlation with the infiltration levels of plasma cells, M2 macrophages, and neutrophils in the AF patient samples (Figures 6(b)–6(d)). However, we did not observe any statistically significant correlation between NSUN3 expression levels and the infiltration levels of the immune cells (Figure 6(a)). These results suggested that DCPS regulated AF development and progression by influencing the lesion immune microenvironment.

4. Discussion

RNA methylation including m6A, m1A, m5C, and m7G is the most prevalent posttranscriptional modifications of eukaryotic RNAs [14]. RNA modifications are regulated by the activities of RNA methylation writers, readers, and erasers [10]. RNA methylation modifications modulate gene expression levels without affecting the coding gene sequences. Several studies have confirmed that m6A RNA modifications play a significant role in embryonic development and carcinogenesis [13]. Furthermore, dysregulation of RNA methylation is associated with heart failure, cardiac hypertrophy, aneurysms, vascular calcification, and pulmonary hypertension [17]. RNA methylation enzymes and their targets are potential diagnostic markers of human diseases and therapeutic targets. However, the role of specific RNA methylation modifications and the corresponding regulatory genes in the onset and progression of AF remains unclear.

AF-related arrhythmias are associated with significantly high mortality and morbidity rates worldwide [6]. However, mechanisms involved in the development and progression of AF are not clear. The understanding of mechanisms underlying AF is required for the development of novel and more



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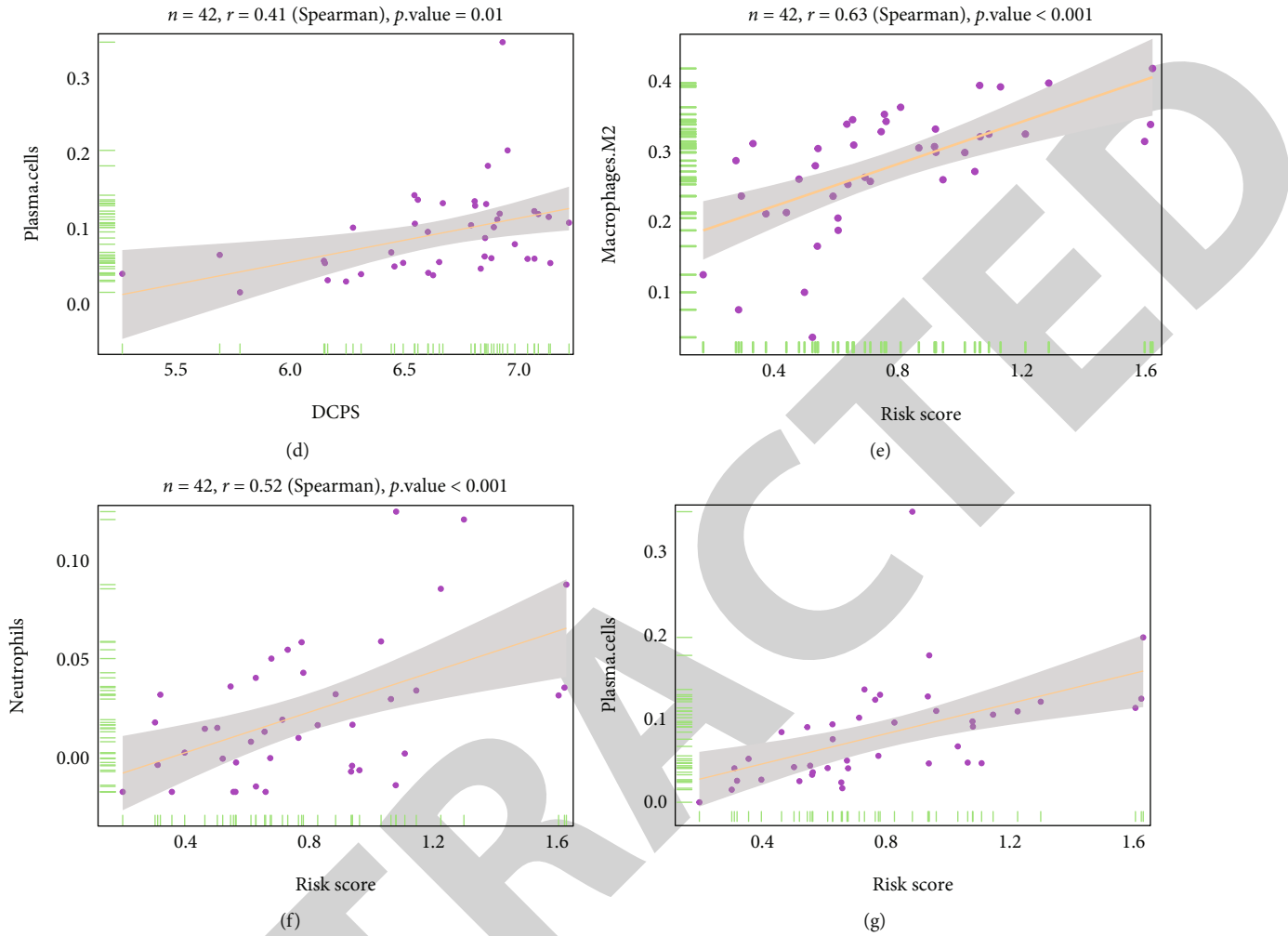

Figure 6: The relationship between infiltration of immune cells and the AF risk-related RNA methylation regulatory gene signatures. (a) The heatmap shows the correlation between the infiltration status of 22 immune cell types and the AF risk-related RNA methylation regulatory gene signature. Red denotes higher correlation, and blue denotes lower correlation. (b–g) Spearman's correlation analysis shows the relationship between the infiltration levels of immune cells such as the plasma cells, macrophages M2, and neutrophils in the atrial tissues of the AF patients and the expression levels of DCPS or the 9-gene risk score (LASSO).

effective diagnostic predictors and therapeutic targets. RNA methylation has emerged as a new research area in the field of medical sciences, and the roles of RNA methylation modifications in cardiac diseases such as CVD are beginning to be understood. Therefore, the present study investigated the expression patterns of RNA methylation regulatory genes that modulate the levels of m6A, m5C, m1A, and m7G RNA modifications in the atrial tissues of AF patients. This study also used bioinformatics analyses to understand the underlying molecular mechanisms, biological functions, and the prognostic potential of AF-related RNA methylation regulatory genes.

The mRNA expression data of the RNA methylation regulatory genes from publicly available AF patient GEO datasets demonstrated that the expression levels of DNMT1, NUDT1, NUDT16L1, DCPS, KIAA1429, EIF3D, ALKBH3, METTL3, RBM15B, METTL14, ALYREF, EIF4E3, NCBP1, ALKBH5, YBX1, and NSUN3 mRNAs were significantly upregulated, and the expression levels of NSUN7, CYFIP2, and NSUN4 were significantly downregulated in the atrial tissues of AF patients compared to those from the SR patients. Furthermore, univariate logistic regression analysis demonstrated that CYFIP2, NSUN7, and NSUN4 were potential protective factors, and KIAA1429, YBX1, NSUN3, DCPS, ALKBH5, and 12 other RNA methylation regulatory genes were potential risk factors in AF. Then, a risk score model was constructed based on 9 critical AF risk-related RNA methylation regulatory genes. The AF patients in the study cohort were classified into high- and low- risk groups based on the median risk score. Functional enrichment analysis of differentially expressed genes related to the AF risk-associated RNA methylation regulatory genes showed significant association between dysregulated bicellular junctions and the risk score. Furthermore, KEGG pathways such as "abnormality of neutrophils", "cytokine-cytokine receptor interaction", "leukocyte trans-endothelial migration", and "natural killer cell mediated cytotoxicity" were significantly enriched in the high-risk AF patient specimens. Previous studies have demonstrated that variants of tight junction proteins such as CAR are associated with increased

incidence of arrhythmia-related diseases. And atrial electrophysiology and structural substrates can be altered by mediators of the inflammatory response, thus increasing the risk of atrial fibrillation [29]. Furthermore, several clinical studies have demonstrated increased infiltration of proinflammatory immune cells in the atrial myocardium of AF patients [30]. Bhat et al. demonstrated that the neutrophil-to-lymphocyte (NLR) ratio was an independent predictor of outcomes in patients with stable coronary artery disease and an independent predictor of prognosis in patients with acute coronary syndromes [31]. Furthermore, infiltration levels of immune cells and the secretion levels of chemokines and cytokines regulate the heart microenvironment in AF patient [32]. Thus, an understanding of AF-associated inflammation and its complex pathophysiology may help to identify specific anti-inflammatory strategies. Previous studies have also shown that RNA methylation plays a key role in shaping the cardiac immune microenvironment [33, 34]. Our data suggested that RNA methylation regulatory proteins modulated the development and progression of AF by regulating the immune microenvironment of the atrial tissues.

We further used the XG-Boost algorithm and the SHAP values to evaluate the RNA methylation regulatory variables and identify the key RNA methylation regulators in AF. This analysis demonstrated that among the 9 AF risk-related RNA regulatory genes, especially DCPS, played a significant role in AF. Furthermore, the expression levels of DCPS showed positive correlation with the infiltration levels of the plasma cells, M2 macrophages, and neutrophils in the atrial tissues. Moreover, DCPS showed good discriminative ability in predicting AF. These results suggested that DCPS modulated AF development and progression by regulating the immune microenvironment in the atrial tissues. DCPS protein is a member of the histidine triad family and plays a key role as an mRNA decapping enzyme in the last step of the $3'$ end mRNA decay [35, 36]. Previous studies have reported that variants of DCPS are associated with neuromuscular disorders [37]. However, the mechanisms by which DCPS regulates AF are not known and require further investigations in future studies.

5. Conclusions

Our study confirmed a close relationship between dysregulation of several RNA methylation regulatory genes and the onset of AF. Furthermore, we constructed and validated a nine AF risk-associated RNA methylation regulatory gene signature for predicting AF. The risk score based on the risk signature showed significant discriminative ability to accurately distinguish AF patient samples from SR samples. The XG-Boost machine learning algorithm demonstrated that DCPS played a key role in AF development by modulating the immune microenvironment in the atrial tissues of AF patients. These results provide considerable evidence supporting the potential of RNA methylation regulatory genes as novel diagnostic biomarkers and therapeutic targets for AF. RNA methylation regulatory genes, especially DCPS, may be useful daily monitoring auxiliary indicators of AF development and recurrence.

Data Availability

All the data in this manuscript can be acquired by request.

Conflicts of Interest

The authors declare that there is no conflicts of interest.

Supplementary Materials

Table S1: the characteristics of GEO databases used in this study. Figure S1: principal components analysis (PCA) of four independent datasets before and after removing batch effect. (A) PCA before removing batch effects. (B) PCA after removing batch effects. Figure S2: integration analysis of RNA methylation regulatory regulators. (A) PPI (protein-protein interaction) analysis. (B) Pearson's correlation analysis based on merge cohort. *(Supplementary Materials)*

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