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ORIGINAL RESEARCH

Identification of Three Core Secretome Genes Associated with Immune Infiltration in High Tumor Mutation Burden Across 14 Major Solid Tumors

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Background: Secretome genes, encoding proteins secreted from the cell, are involved in the tumor immune response and correlated with levels of tumor mutation burden (TMB) in multiple tumors. This study aimed to identify core secretome genes and their potential association with immunomodulators and immune infiltration in high TMB groups across 14 major solid tumors through bioinformatics analysis.

Methods: Multi-omics data for 14 major solid tumors were downloaded from The Cancer Genome Atlas (TCGA) database. Patients were divided into high TMB (TMB-high) and low TMB (TMB-low) groups using the median TMB values for each of the solid tumors. The CIBERSORT algorithm was conducted to estimate the proportion of 22 tumor-infiltrating immune cells (TIICs). Kaplan–Meier analysis and the log-rank test were utilized to screened prognosis-related genes. The correlations between core secretome genes and TIICs were analyzed using Spearman correlation coefficients.

Results: In TMB-high groups, multi-omics data analysis revealed that secretome genes were strongly associated with clinical characteristics, and 65 prognosis-related secretome genes were screened. Among the prognosis-related genes, 21 core secretome genes were identified, and strongly associated with five types of TIICs, namely activated NK cells, follicular helper T cells, CD8 T cells, and macrophages M0 and M2. Notably, three secretome genes (*ADAMTS12*, *COL12A1*, and *COL5A2*) were significantly related to immunomodulators and TIICs in multiple solid tumors. In addition, 12 core secretome genes were significantly differentially expressed between responding and non-responding patients receiving immunotherapy. Furthermore, core secretome genes may be involved in the PI3K/AKT signaling pathway.

Conclusion: We examined the prognostic significance of secretome genes and their potential association with immunomodulators and immune infiltration across 14 major solid tumors. In summary, three secretome genes (*ADAMTS12*, *COL12A1*, and *COL5A2*) may be pivotal mediators of immune infiltration in TMB-high patients, which may help to identify patients who could benefit from immunotherapy.

Keywords: secretome genes, tumor mutation burden, immune infiltration, prognosis, solid tumors

Introduction

The tumor secretome, a potential treasure trove of biomarkers and pharmaceutical targets for cancers, plays a critical role in oncogenesis and cancer treatment.[1,2] Secretome genes contribute to the hallmarks of cancer, function through the synergistic effects of multiple factors, and share common mechanisms of tumor

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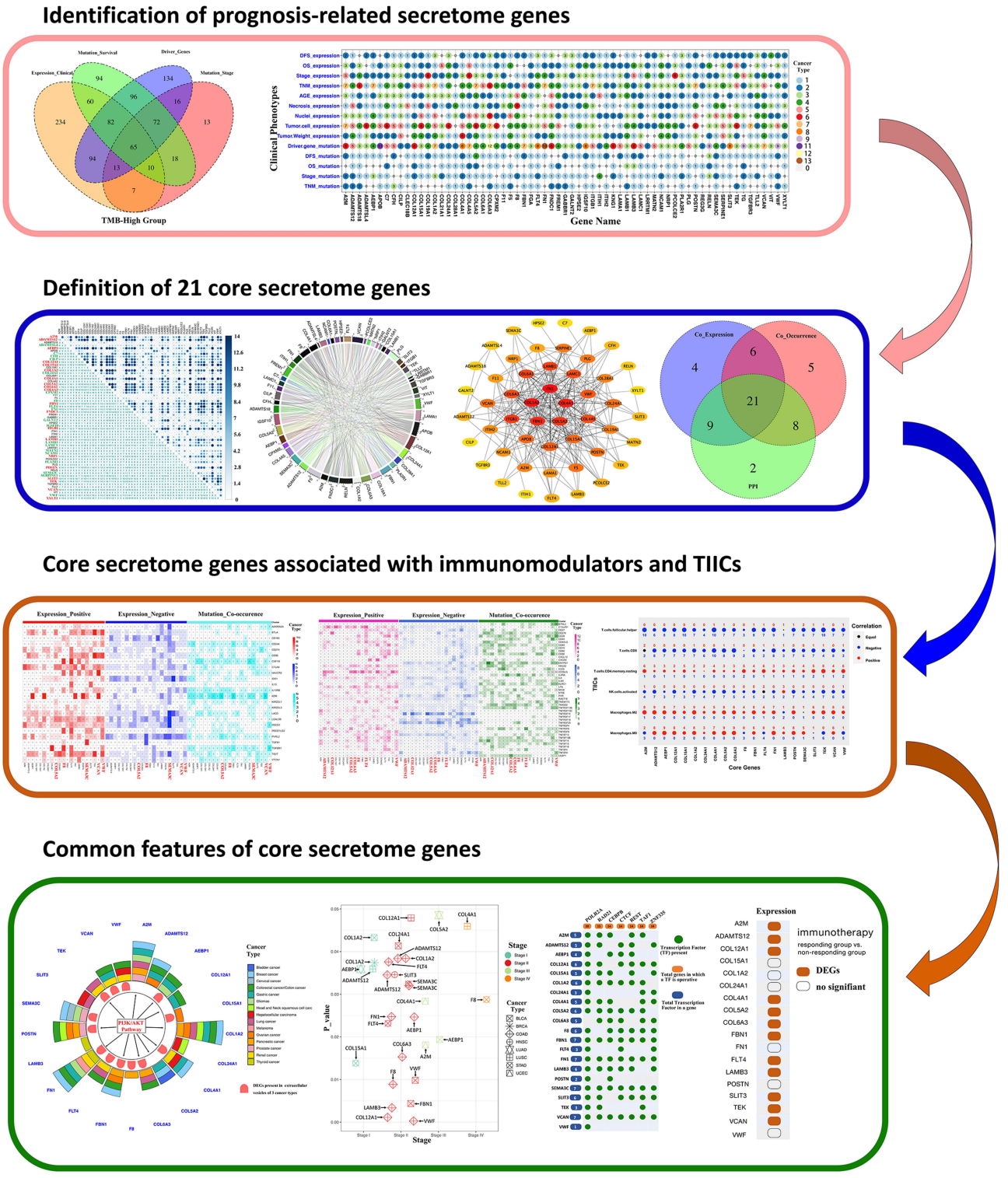
Graphical Abstract



progression and metastasis.[3,4] Decoding the secretome genes in the tumor microenvironment, including chemokines, cytokines, and growth factors, could provide useful biomarkers for predicting the immunotherapeutic response, immune infiltration, and immunosuppression.[5,6] Chemokines and cytokines are critical mediators of

immune responses and are potential targets for cancer immunotherapy.[7,8] Furthermore, secretome genes tend to show a decrease in changes in expression, which is related to their involvement in tumor suppression and cell-matrix adhesion functions.[9] Therefore, secretome genes may hold great promise as potential cancer prognostic biomarkers and play roles in tumor immune infiltration across various cancer types.

Tumor mutation burden (TMB), which is an independent predicator for immunotherapy, can be used to determine the efficacy of immune checkpoint inhibitor therapy in various tumors.[10,11] In diverse tumors, patient with high TMB exhibit prognostic correlation with poorer survival, but they can gain a more favorable prognosis after treatment with immunotherapy, representing a relevant prognostic biomarker.[12,13] TMB levels are positively correlated with diverse immune-related genes and immune signatures, and are predisposed to be identified by immune defense mechanisms in patients with high TMB.[14] High TMB level is associated with clinical outcomes and may influence immune cell infiltrations in various tumors.[14] Tumor-infiltrating immune cells (TIICs), which play significant roles in the tumor immune environment, can act as indicators of the immune response and targets of therapeutic strategies against tumors.[15,16] The co-expression of TIICs and inhibitory immune-related genes is involved in resistance to immunotherapy in multiple tumors.[17–20] Increasing evidence shows that secreted factors and secretome genes in combination with TIICs are connected with tumor immunotherapy, which may offer emerging targets and predictors of response to immunotherapy.[21–23] Thus, comprehensive analysis of the prognostic roles of secretome genes in TMB-high patients could help to identify candidate immunotherapy biomarkers and elucidate the underlying molecular mechanisms involved in immune infiltration.

To identify favorable prognostic genes in TMB-high cancer patients, we performed an integrative analysis of the expression and genetic alteration of secretome genes with clinical outcomes across 14 major solid tumors from The Cancer Genome Atlas (TCGA) database. Then, we explored core secretome genes from co-expression, co-mutation, and protein–protein interaction (PPI) networks from prognosis-related genes. Utilizing immunomodulator genes and the abundance of TIICs, we further identified the relationship between core secretome genes and immune infiltration. In addition, we detected the common molecular mechanism of core secretome genes underlying the prognostic capability in the TMB-high group. As secretome genes are strongly related to immune infiltration in TMB-high patients, they could be applied as prognostic biomarkers and immunotherapeutic targets for "high-risk" patients who could benefit from precise immunotherapy, and will be helpful in detecting the tumor immune infiltration process.

Materials and Methods

Generation of Secretome Genes from the Human Protein Atlas (HPA) and UniProt Databases

Coding genes of secretory proteins were screened from the HPA and UniProt databases, according to annotation information. A total of 1708 genes were predicted to have secreted protein products in the HPA database (https://www.proteinatlas.org/), coming from four sources (HPA, SignalP, Phobius, and SPOCTOPUS). In addition, 2723 secretome genes were downloaded from the UniProt website (https://www.uniprot.org) and filtered by two categories, ie, secretory protein and secreted protein. In total, 1507 secretome genes were identified from these two databases.

Data Collection and Preprocessing

Patient cohorts used in this study are listed in Supplementary Table S1. RNA-Seq gene expression data (RSEM algorithm, normalized_count, and scaled_estimate) and relevant clinicopathological data across 14 cancer types were obtained from TCGA database via the Fire Browse website (https://gdac.broadinstitute.org/). Somatic mutations using Mutect2 for variant calling were downloaded from the GDC data portal using the TCGAbiolinks package,[24] and mutation annotation format (MAF) files were analyzed using the maftools R package.[25] We acquired available mutation driver genes from the DriverDBv3 database (http://driverdb.tms.cmu.edu.tw).[26]

Differentially Expressed Genes (DEGs) and Tumor Mutation Burden (TMB) Analysis

We analyzed mRNA expression and somatic mutation data of secretome genes in 14 cancer types. The mRNA-seq data were preprocessed by "limma" in the R package, and significant DEGs were identified with a false discovery rate (FDR) cut-off of 0.05.[27] The MAF files of somatic mutations and TMB were processed using maftools.[25] The total number of somatic mutations (insertion, deletion, and signal nucleotide polymorphism) was summed to yield

a mutation burden for each patient. The TMB-high group (samples with TMB values higher than the median TMB value) and TMB-low group (samples with TMB values lower than the median TMB values) were defined per cancer type based on the median TMB values of the relevant patients.

Evaluation of Association Between Gene Expression and Clinicopathological Features

The expression levels of secretome genes were merged with the corresponding clinicopathological features of each patient, including progression stage (stage); tumor, node, metastasis (TNM); percentage of tumor necrosis (tumor necrosis); percentage of tumor nuclei (tumor nuclei); tumor weight; age at initial pathological diagnosis (age); and percentage of tumor cells (tumor cells). The relationship between secretome gene expression levels and clinicopathological features was assessed using Spearman's correlation in TMB-high groups with a threshold of p-value <0.05.

Identification of Core Secretome Genes

To define core secretome genes, we analyzed the relations between outcome-associated genes, including gene co-expression, mutational co-occurrence, and PPI. We applied $\log_2$ (normalized count + 1) to calculate the interaction of gene expression using Spearman's correlation coefficients. The mutational co-occurrence for each pair of genes was determined using the SomaticInteraction function in maftools. A p-value <0.05 was used as the cut-off. The Search Tool for the Retrieval of Interacting Genes (STRING) database and Cytoscape software were used to retrieve and reconstruct a PPI network for the prognosis-related secretome genes.[28,29] Hub genes were selected based on topological degree analysis using the CytoHubba plugin.[30]

Assessment of Immune Infiltration

The transcripts per million (TPM) values per sample were obtained from scaled estimates by multiplying by 1e6. We used the CIBERSORT analytical tool to classify and estimate the abundances of 22 immune cell types for each patient based on TPM values.[31] The recommended model parameters were performed using the LM22 signature gene expression file and default signature matrix at 1000 permutations. The relationships between core secretome gene expression levels and the proportions of TIICs were further assessed using Spearman's correlation coefficients in R package. A p-value <0.05 was considered statistically significant.

Survival Analyses and Clinical Enrichment Analysis

We used the univariate Cox proportional hazards regression model (Coxph) and log-rank test in the R "survival" package to examine the influence of gene expression on survival rate, including overall survival (OS) and disease-free survival (DFS). According to the Coxph results, for genes with p<0.05, a better survival rate was found if the values of the regression coefficient and $\log_2$ fold-change in expression changed in the opposite directions. This suggests that higher expression of potential oncogenes was significantly related to poor prognosis. We used the mafSurvival function in maftools to analyze the correlation between mutations and survival based on the mutation status of given genes, with p-values <0.05 considered to be statistically significant. We used clinical enrichment analysis to identify enriched mutations for core secretome genes with tumor stage among various groupwise comparisons using the maftools R package. The significance threshold was p<0.05.

Results

Evaluation of Prognosis-Related Secretome Genes in TMB-High Patients Across 14 Solid Tumors

We identified 1507 genes encoding secretory proteins in the Human Protein Atlas (HPA, https://www.proteinatlas.org/) and UniProt (https://www.uniprot.org/database/) databases for the evaluation of secretome genes. According to the median TMB value of each cancer, we divided cancer patients into TMB-high and TMB-low groups across the 14 cancer types (Supplementary Table S1).

To screen clinically relevant prognosis-related secretome genes, nine clinicopathological features of cancer patients were considered, comprising stage, TNM, tumor necrosis, tumor nuclei, tumor weight, age, tumor cells, and patient survival (OS and DFS). In addition, the total number of cancer types significantly associated with clinical outcomes for each gene was applied to screen potential prognosis-related secretome genes. According to the expression levels of 1507 secretome genes, 660 and 1007 genes were significantly correlated with patient survival rate (OS or DFS) and tumor stage (stage or TNM),

respectively, in two or more cancer types. Among the above genes, 565 were significantly associated with both survival and stage in at least two cancer types (Figure 1A, Supplementary Table S2). These 565 candidate genes showed strong correlation with the other five clinical phenotypes (Supplementary Table S2). We next examined the association between genetic alterations in secretome genes and clinicopathological characteristics in the TMB-high group. Among the 1507 secretome genes, 572 were significantly associated with survival or stage in at least one cancer type, including 214 gene mutations correlated with survival (OS or DFS) and 497 gene alterations associated with stage (stage or TNM) (Figure 1B, Supplementary Table S2). Overlapping these results with mutational analysis, 165 of the 572 secretome genes were significantly associated with survival and stage in the TMB-high group in at least one cancer type (Figure 1C). Notably, 973 of the 1507 secretome genes were identified as mutation drivers from the DriverDBv3 database across multiple cancer types.[26] In conclusion, we screened 65 potential prognosis-related genes whose expression levels and genetic alterations were associated with survival and stage in TMB-high groups (Figure 1D).

In all cancer patients, we analyzed 65 potential prognosis-related gene expression levels in relation to clinical outcome with the above methods. The results showed that 59 genes were significantly correlated with clinical outcomes, and six genes (*CLEC18B*, *COL21A1*, *FGA*, *KNG1*, *REG3G*, and *TG*) showed weak correlations with clinical outcomes in all patients (Supplementary Figure S1). These six genes were then separately explored regarding their role in tumor immune infiltration. In all patients, common down-regulated genes were closely related to tumor nuclei and cells, and common up-regulated genes were strongly associated with tumor weight, necrosis, and patient age (Supplementary Figure S1, Supplementary Table S3).

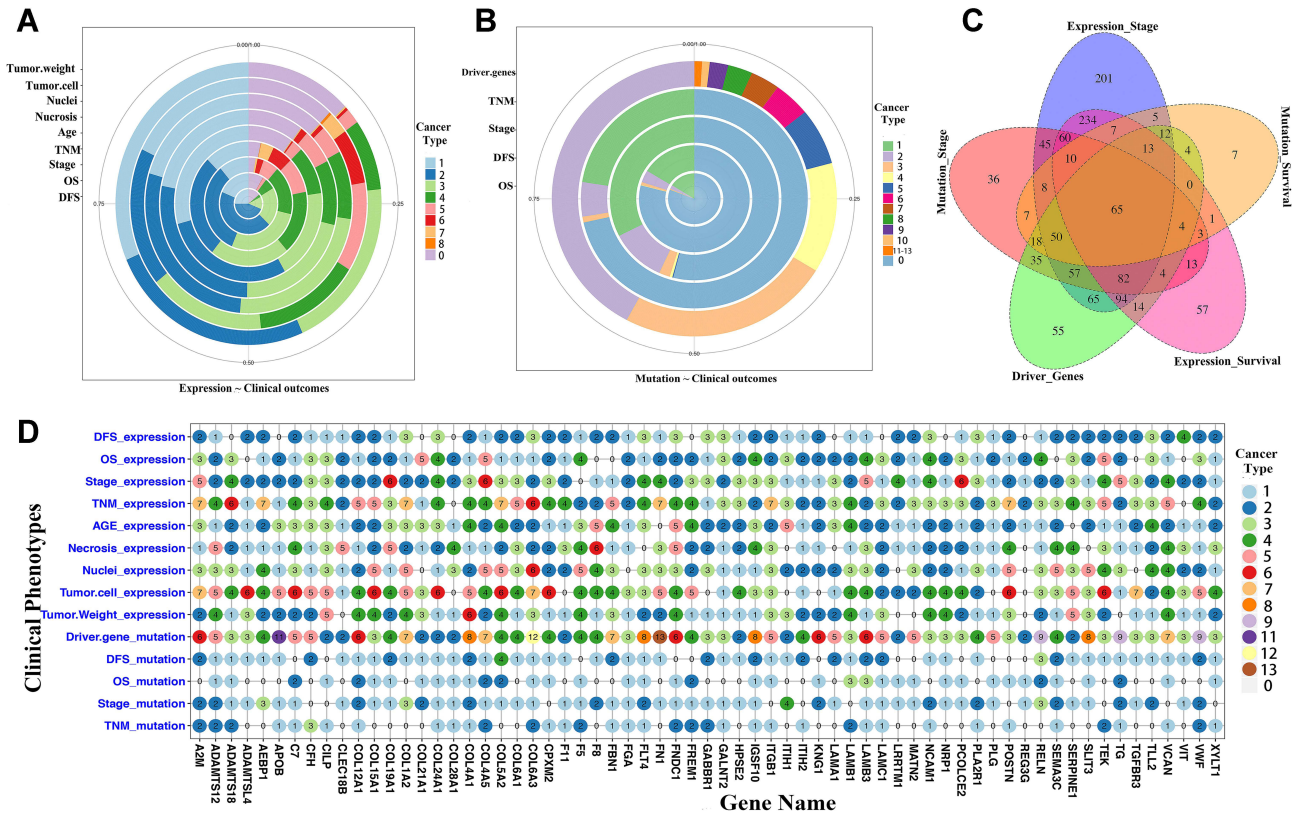


Figure 1 Prognosis-related biomarkers of secretome genes in TMB-high group across 14 cancer types. (**A**) Proportion of 565 secretome genes with significant association between gene levels and clinical outcomes across 14 solid tumors. (**B**) Percentage of 572 secretome genes in which genetic alterations were significantly associated with clinical outcomes for each clinical phenotype. Circles represent the fraction of secretome genes in which gene expression levels (**A**) and genetic alterations (**B**) were significantly related to clinical outcomes in different cancer types. (**C**) Venn diagram of 65 prognosis-related secretome genes showing associations between clinical outcome and expression levels and genetic alterations in TMB-high group. (**D**) Distribution of 65 prognosis-related secretome genes with clinical outcomes in different cancer types. Numbers in circles represent cancer types in which secretome genes were significantly associated with clinical outcomes. Rows represent genes, columns represent clinical outcomes.

Definition of 21 Core Secretome Genes Based on Mutation Co-Occurrence, Gene Expression Correlation, and PPI in TMB-High Group

To explore the core roles of the above 59 prognosis-related genes, we analyzed their expression levels, genetic alterations, and PPIs. First, we determined the correlations in expression levels of each gene pair and found 9171 gene expression pairs with significant correlation across 14 cancer types ($p<0.05$). We counted the total number of significant pairs in all cancer types for each gene and characterized the top 40 as key genes of the co-expression module (Figure 2A). Second, we identified 3759 gene pairs with significant mutational co-occurrence in 14 cancer types and screened the top 40 as key genes of the co-mutation module based on total occurrences (Figure 2B). Among them, we found four gene pairs with significant co-occurring mutations in seven cancer types, including *LAMA1+RELN*, *APOB+COL19A1*, *COL12A1+IGSF10*, and *APOB+VCAN*. Third, we constructed a PPI network for 59 potential prognosis-related genes using the STRING database v11.0 and calculated the degree value using CytoHubba in Cytoscape. The high degree of core secretome genes indicates that there would be potential biological effects in the network. Among these genes, 51 genes had a complex interaction network with more than one degree, and the top 40 genes with the highest degrees were identified as hub genes (Figure 2C). Finally, we defined 21 core secretome genes from the key genes in the above three interaction analyses, indicating that they may play important roles in the regulation of tumor prognosis (Figure 2D).

Core Secretome Genes are Strongly Associated with Immunomodulators

Immune checkpoint genes were identified as prognostic biomarkers in multiple tumors, and their expression levels were associated with clinical parameters and immunotherapeutic response.[32] So, we detected the role of the 21 core genes in the tumor immune response by detecting the association of their expression levels and genetic alterations with immunomodulatory genes (24 immune-inhibitor genes and 46 immune-stimulator genes). According to the frequency of secretome genes in significant pairwise interactions, key immune-related genes were defined as the top 12 and 11 genes with the highest frequency in the correlations of immune inhibitors and immune stimulators, respectively. Among the core secretome genes, the top five genes (*COL5A2*, *F8*, *SEMA3C*, *VCAN*, and *VWF*) were associated with immune-inhibitor genes (Figure 3A) and the top seven genes (*ADAMTS12*, *COL12A1*, *COL5A2*, *COL6A3*, *F8*, *FLT4*, and *VWF*) were related to immune-stimulator genes (Figure 3B) with relevant research on gene expression and mutation. Taken together, we screened nine immune-related secretome genes (*ADAMTS12*, *COL12A1*, *COL5A2*, *COL6A3*, *F8*, *FLT4*, *SEMA3C*, *VCAN*, and *VWF*) that may play prominent roles in immune infiltration, depending on the specific immune cells. In particular, three secretome genes (*COL5A2*, *F8*, and *VWF*) were found in both sets, suggesting that they may play a critical role in the immune response by combining with immunomodulator genes. At the same time, we assessed the performance correlations between the six genes mentioned above (in 'Evaluation of Prognosis-Related Secretome Genes in TMB-High Patients Across 14 Solid Tumors') (*CLEC18B*, *COL21A1*, *FGA*, *KNG1*, *REG3G*, and *TG*) and immunomodulators, but found no strong correlations among them (Supplementary Figure S2A and B).

Core Secretome Genes are Significantly Correlated with TIICs

TIICs indicated the prognostic relevance and reflected the mechanisms underlying the anti-tumor immune response in different tumor types.[33] To clarify the role of core genes in immune infiltration in the TMB-high group, we examined the correlation between the infiltration levels of 22 immune cells and core gene expression levels across 14 solid tumors. We found that their expression levels were mainly positively correlated with the fractions of two TIICs (ie, macrophages M0 and M2) and negatively correlated with the fractions of three TIICs (ie, follicular helper T cells, CD8 T cells, and activated NK cells) (Figure 4). Meanwhile, five core genes (*ADAMTS12*, *COL12A1*, *COL1A2*, *COL5A2*, and *POSTN*) were significantly correlated with the above TIICs in more than six cancer types. A high infiltration level of CD4 memory resting T cells was associated with high expression levels of

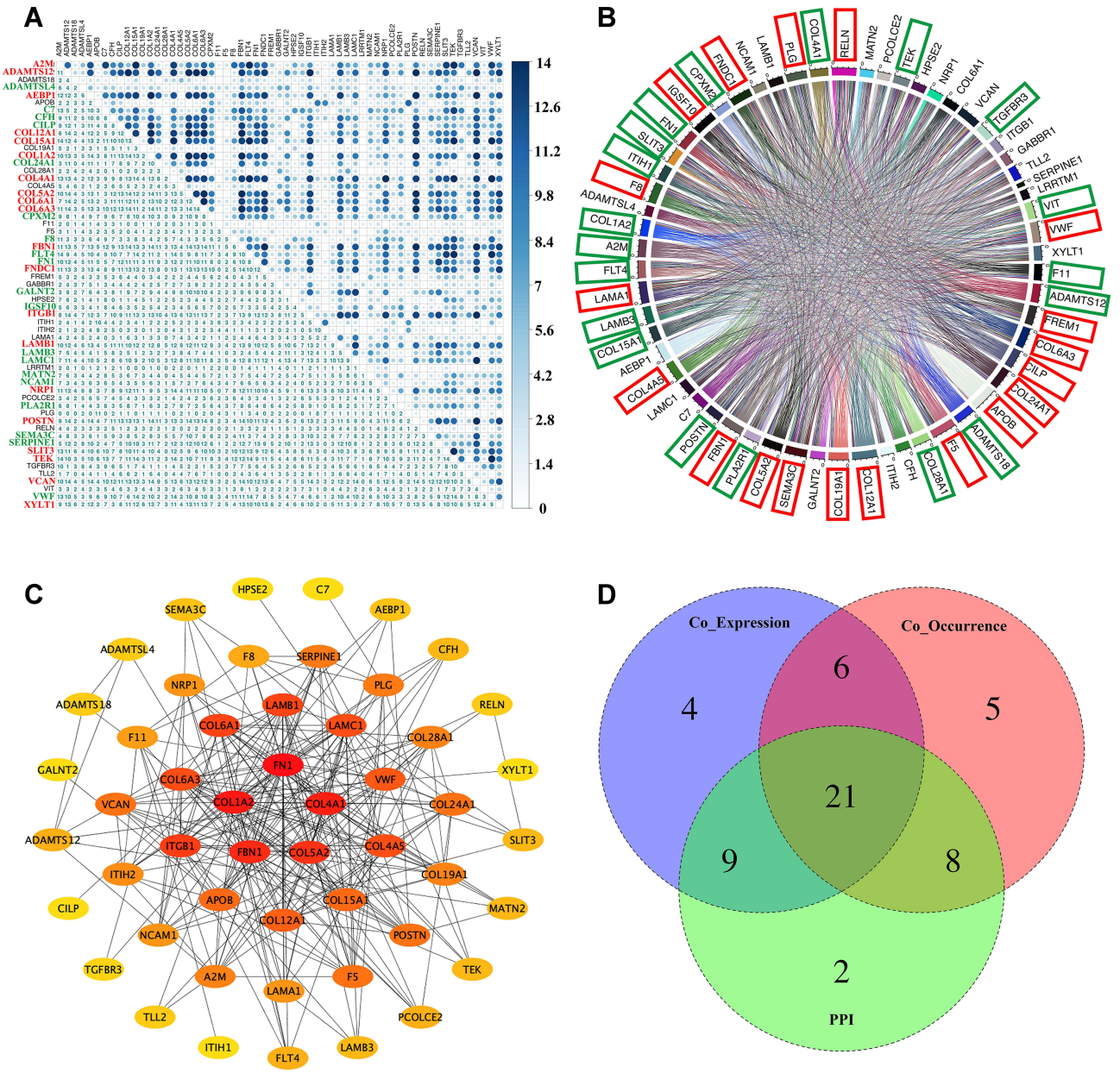


Figure 2 Identification of core genes through co-expression, mutation co-occurrence, and protein–protein interactions across 14 solid tumors in TMB-high group. (**A**) Gene co-expression analysis of 59 prognosis-related secretome genes among 14 solid tumors. Numbers represent cancer types with significant gene pairs. These genes were divided into three groups according to the frequency of secretome genes in significant gene pairs (red represents high group, green represents median group, and black represents low group). (**B**) Mutation co-occurrence analysis of 59 prognosis-related secretome genes across 14 cancer types. Genes were divided into three groups according to the frequency of secretome genes in significant gene pairs, ie, red squares indicate high group and green squares indicate median group. (**C**) PPI network of 52 secretome genes constructed via STRING and Cytoscape. (**D**) Venn diagram of 21 core secretome genes from key genes based on co-expression modules, co-occurrence of mutation, and PPI analysis.

core genes in fewer cancer types (Figure 4), and had a moderate correlation with the remaining TIICs (Supplementary Figure S3A). Consistent with the above results, six genes (*CLEC18B*, *COL21A1*, *FGA*, *KNG1*, *REG3G*, and *TG*) showed a poor correlation with the abundance of 22 immune cells (Supplementary Figure S3B).

Identification of Common Features in Core Secretome Genes

To further explore the potential role of core secretome genes in immune infiltration, we detected several common features and potential molecular mechanisms of core secretome genes. Evidence from the current literature suggests that the expression levels of the core secretome

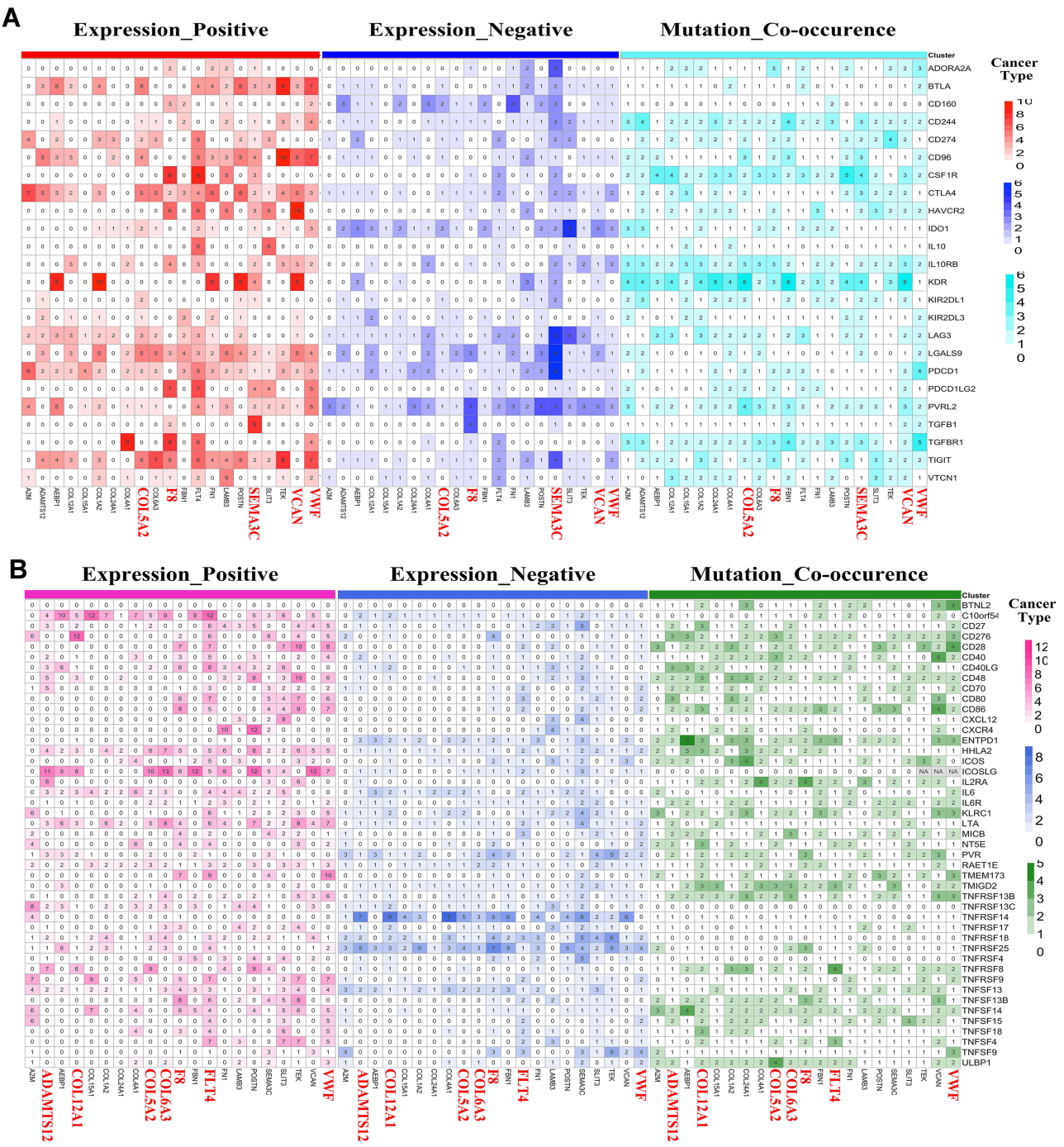


Figure 3 Correlations of 21 core secretome genes with immunomodulator genes in TMB-high group across 14 cancer types. (**A**) Heatmap showing correlations among 21 core secretome genes (bottom) and 24 immune-inhibitor genes (right). (**B**) Heatmap showing correlations among 21 core genes (bottom) and 46 immune-stimulator genes (right). Numbers in (**A** and **B**) show cancer types with significant gene pairs.

genes may be important prognostic biomarkers in various cancer types (Figure 5A, Supplementary Table S4). Furthermore, they may be involved in the PI3K/AKT signaling pathway, which recruits cell infiltration and improves the outcome of immunotherapy.[34] Based on the above results, we speculate that core secretome genes may regulate immune infiltration via the PI3K/AKT signaling pathway. In addition, 13 core secretome genes were significantly differentially expressed in extracellular vesicles of three cancer types (ie, colorectal, liver, and pancreatic cancer) between tumor patients and healthy controls from the BBCancer database (http://bbcancer.renlab.org).

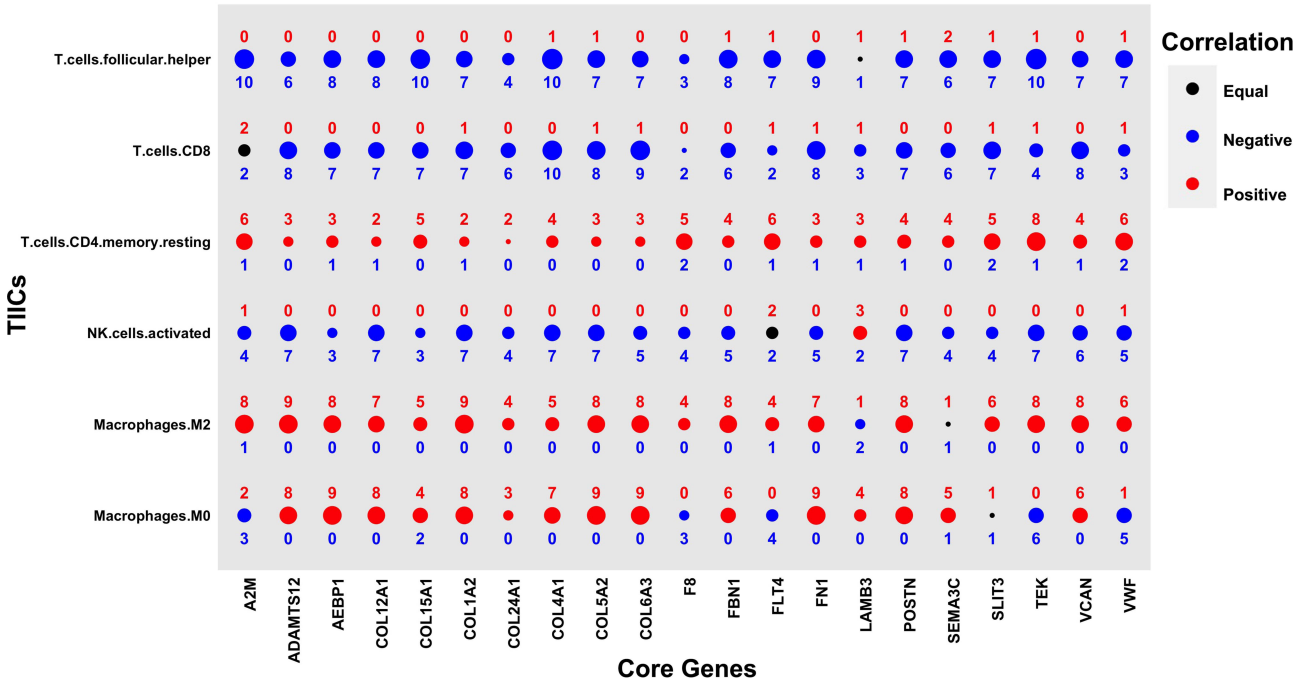


Figure 4 Correlations of 21 core secretome genes with TIICs in TMB-high group across 14 cancer types. Relationships among 21 core secretome gene expression levels and fractions of nine immune infiltration cells. Red and blue numbers indicate positive and negative correlations, respectively. Numbers show cancer types with significant gene pairs.

Notably, 12 core secretome genes showed significant differential expression ($p<0.05$) between responding and non-responding patients receiving immunotherapy, suggesting that they may sensitive to immune checkpoint blockade (Figure 5B).[35] According to the GeneCards database, one transcription factor (*POLA2R*) was present in the core secretome genes, suggesting that a common molecular mechanism for these genes may be involved in the immune response of solid tumors (Figure 5C). Using the clinicalEnrichment function of the maftools package, our results showed that 15 secretome genes exhibited significant enrichment for somatic mutations in stage II, demonstrating that the mutation frequencies of these genes were significantly higher in stage II compared with the other three stages (Figure 5D).

Discussion

In this study, we provided a comprehensive description of the clinical outcome of secretome genes and revealed that core genes may play important roles in immune infiltration in TMB-high patients across 14 solid tumors. Utilizing the expression levels and genetic alterations of secretome genes, we identified 65 prognosis-related genes that were significantly associated with clinical outcomes across various types of cancer in TMB-high patients. Among them, 21 core secretome genes were observed by performing dimensional analysis, and significantly related with immune modulator genes and TIICs in multiple solid tumors. Furthermore, we found differentially expressed core genes for immunotherapy. The results of functional enrichment analysis suggested that core genes were related to the PI3K/AKT signaling pathway.

Immunomodulator genes may reflect the status of immune topographies and act as potential targets for immune checkpoints.[36,37] The expression levels of immunomodulator genes are associated with clinical outcomes and infiltration levels of immune cells.[38] By combining expression data and genetic alteration profiling, we inferred essential immunomodulator genes and showed that core genes may be better predictors of immune infiltration and immunotherapy. Our findings showed that the expression levels of 21 core genes were closely related to the most widely studied immune inhibitors (*CTLA4* and *PDCD1*) and novel immune inhibitors (*LAG3* and *TIGIT*), which are therapeutic targets in clinical and preclinical studies.[39,40] From the core secretome genes (Figure 3A and B), we identified five immune-inhibitor genes (*CD96*, *IL10RB*, *PDCD1*, *PVRL2*, and *TIGIT*) and two immune-stimulator genes (*CD276* and *IL2RA*) as the highest frequency immune-modulator genes in significant gene pairs. Identifying core secretome genes

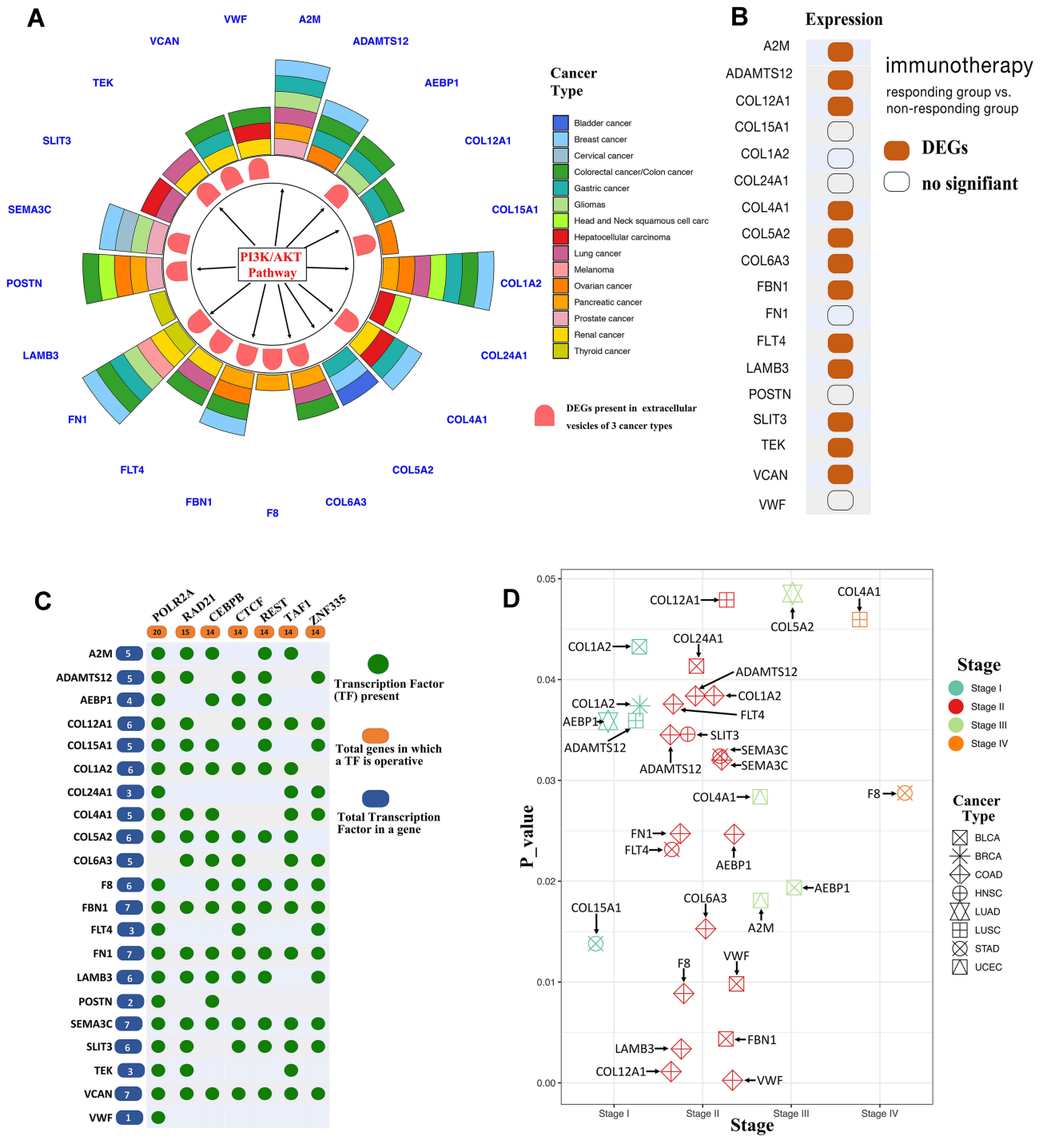


Figure 5 Functional analysis of 21 core genes. (**A**) Circos plot of 21 core genes based on function and differential expression in extracellular vesicles of three cancer types. Outer circle represents 21 core genes identified as potential biomarkers of different cancer types in previous studies. Middle circle represents 21 secretome genes showing differential expression in extracellular vesicles of three cancer types from the BBCancer database (http://bbcancer.renlab.org/). Inner circle represents 11 core secretome genes that may function in tumorigenesis through the PI3K/AKT signaling pathway. (**B**) Twelve core secretome genes sensitive to immunotherapy. (**C**) Predicted transcription factors of 21 core secretome genes from the GeneCards database. (**D**) Fifteen core secretome gene mutations were significantly enriched in stage II.

associated with immune checkpoint genes may help to elucidate the resistance mechanism to immune checkpoint inhibition in the tumor microenvironment. These results suggest that the above immunomodulators may be targets for immune checkpoint blockade and that core secretome genes may facilitate the tumor immune response.

The prognostic outcomes of immunotherapy are linked to TIICs in various types of cancer, and these

could be used as potential predictive biomarkers for cancer immunotherapy.[41,42] Our results demonstrated that there is a high correlation between core gene expression levels and infiltration of five TIICs, which indicated that five core genes (*ADAMTS12*, *COL12A1*, *COL1A2*, *COL5A2*, and *POSTN*) may be involved in regulating tumor immune infiltration in TMB-high patients. M2 macrophages correlated with an increase in the expression levels of core secretome genes, suggesting that they were poor prognostic factors in most cancer types.[43,44] The core gene expression levels were negatively correlated with the infiltration of follicular helper T cells, CD8 T cells, and activated NK cells. These infiltrating immune cells play a pivotal role in the anti-tumor immune response and have implications for immunotherapy.[45,46] TIICs can secrete proinflammatory cytokines and chemokines, and secretome factors have a function in anti-tumor effects, which may largely be due to TIICs.[47] Cytokines are effective in cancer immunotherapy and modulate various populations of immune cells.[48–50] Chemokines can affect the phenotype of immune cell infiltration and recruitment, and their expression levels are correlated with tumor immune responses.[51] Our data raise the possibility that the infiltration-related secretome genes may function as potential mediators of immune cell infiltration in multiple solid tumors.

Consistent with our results, these three genes (*ADAMTS12*, *COL12A1*, and *COL5A2*) were closely related to immune infiltration and may have potential value for immunotherapy in multiple tumors. *ADAMTS12* is a favorable prognostic factor for immune infiltration in gastric cancer and pancreatic adenocarcinoma.[52,53] *COL12A1* may act as a potential prognostic biomarker and an immune-associated therapeutic target in gastric cancer and pancreatic adenocarcinoma.[54–56] *COL5A2* has a strong correlation with immune infiltration and could be an immunotherapeutic target for multiple malignant tumors, including prostate cancer, glioma, lung adenocarcinoma, and pancreatic ductal adenocarcinoma.[57–60] These results demonstrate that the above three genes may act as potential predictors for prognosis, and play important roles in immune infiltration in TMB-high groups. Nevertheless, limitations exist in this study. First, it is a retrospective study based on public databases, without our own data. Second, there is a lack of basic experiments to verify the impact of these secretome genes on immune infiltration.

Conclusion

In summary, we provided a comprehensive description of clinical outcomes of secretome genes in TMB-high patients, and revealed that core secretome genes were strongly associated with immune infiltration across 14 major solid tumors. Three secretome genes (*ADAMTS12*, *COL12A1*, and *COL5A2*) were strongly associated with immunomodulator genes and five subtypes of TIICs. Our study highlights the important role of secretome genes in immune infiltration in TMB-high patients.

Data Sharing Statement

Data sharing is not applicable to this article, as no datasets were generated or analyzed during the current study.

Acknowledgments

We thank all the participants.

Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation, or in all these areas; took part in drafting, revising or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

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Disclosure

The authors report no conflicts of interest in this work.

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