WITHDRAWN: Identification of TFE3/TFEB and its Related Genes as Prognostic Biomarkers in Acute Myeloid Leukemia

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Research

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The full text of this preprint has been withdrawn by the authors while they make corrections to the work. Therefore, the authors do not wish this work to be cited as a reference. Questions should be directed to the corresponding author.

Abstract

Background: Acute myeloid leukemia (AML) is one of the most common hematologic malignances with an ever-increasing incidence and high mortality. TFE3 and TFEB, two transcription factors that mediate cellular adaptation to stress by simultaneously promoting lysosomal biogenesis, autophagy induction, as well as expression of critical mitochondrial and metabolic regulators, which are substantial contributors to cell fate and cancer progress. However, the expression and prognostic values of TFE3/TFEB in AML have not been clarified.

Objective: To explore the expression and role of TFE3/TFEB in AML and thus to find potential therapy.
Methods: RNA sequence data from AML patients and healthy donors were obtained from the Gene Expression Omnibus (GEO) database. Differentially expressed genes (DEGs) analysis were performed by GEO2R. TFE3/TFEB related genes were obtained from UALCAN. Gene ontology (GO) and KEGG pathway were analyzed by WEB-based GEne SeT AnaLysis Toolkit (WebGestalt) and DAVID. Protein-protein interactions (PPIs) network construction and module analysis were performed by STRING and Cytoscape. The Kaplan-Meier survival curves were drawn in TCGA portal.

Results: We found TFE3 and TFEB can be used prognostic factors for AML, and most of their positively related genes were worse prognostic factors too. ITGB2, FGR, ITGAM, ITGAX and SELPLG were identified as the most significant genes in survival-related genes contributed by TFE3 and TFEB.**Conclusions:** In this study, we performed a comprehensive analysis of gene expression and gene function to identify key prognostic biomarkers in AML.

Background

Acute myeloid leukemia (AML) is a malignant disorder of the bone marrow which is characterized by the clonal expansion and differentiation arrest of myeloid progenitor cells (1), and the most common type of myeloid malignancy in adults with an incidence of 4.3 per 100,000 persons (2), which is mainly characterized by the block of myeloid differentiation and expansion of immature myeloid progenitors (blasts) in the bone marrow (BM) of patients. The clinical outcome of AML patients was closely related to immune, molecular, and cytogenetic abnormalities (3–5), as well as any other factors, such as age and family history. Over the past few decades, diagnosis and treatment in patients with AML has improved, but the overall survival (OS) rate for AML is still low, less than 50% (6), and the overall five-year survival rate is approximately 24% (2).Of all subtypes of leukemia, AML accounts for the highest percentage (62%) of leukemic deaths (2). Therefore, finding effective prognostic biomarkers has been being one of the most urgent clinical needs and research hotspots.

The most common treatment for AML is intensive chemotherapy, which aims at eradicating the leukemic cell population. Aggressive metabolic changes are key hallmarks of cancer and a variety of research has revealed the strong relation between chemo-resistance and metabolism changes. For example, Zhou et al. (7), pointed the possible relation between the relapse of AML and increased oxidative stress in vivo.

Additionally, Herst et al. (8) have demonstrated a possible relation between the level of glycolytic metabolism of AML blasts and resistance to chemotherapy.

The microphthalmia family (MITF, TFEB, TFE3, and TFEC) of transcription factors is emerging as global regulators of cancer cell survival and energy metabolism, both through the promotion of lysosomal genes as well as newly characterized targets, such as oxidative metabolism and the oxidative stress response (9). The MiT/TFE family of basic helix-loop-helix (bHLH) transcription factors recognizes the transcription initiation or E-box (Ephrussi boxes) sites (CANNTG) in the genome (10), thus activate gene transcription. In addition, MiT/TFE factors can regulate lysosomal signaling, which includes the mTORC1 and Wnt/β-catenin pathways, which are both substantial contributors to oncogenic signaling. Transcription factor E3 (TFE3) and transcription factor EB (TFEB) had been proved to binds to Coordinated Lysosomal Expression and Regulation (CLEAR) elements to induce lysosomal biogenesis and autophagy (9). Lysosomes are organelles found in all eukaryotic cells and are emerging as key regulators of a wide range of cellular functions such as metabolic signaling in addition to their established role in macromolecule degradation and recycling (11, 12). What's more, under conditions of prolonged ER stress, TFEB and TFE3 contribute to cell death, thus revealing an unexpected role for these proteins in controlling cell fate (13) and disease progression.

A number of studies have determined TFE3 and TFEB as being oncogenes, however, there have been few studies on TFE3 /TFEB in acute myeloid leukemia. Here, we presented high TFE3/TFEB as an adverse prognostic biomarker for AML with concrete data, and also explored the prognostic value of correlated genes with TFE3/TFEB in AML.

Results

Identification and functional enrichment analysis of DEGs between AML patients and healthy donors

A total of 13,295 non-repetitive genes and 1111 DEGs were identified from 26 AML patients and 18 healthy donors, including 365 upregulated and 746 downregulated genes in AML based on the cut-off criteria (adjust $P < 0.05$ and $|\log FC| > 1$, Supplementary material 1). The overall aberrantly expressed genes were shown in the Volcano plot (Fig. 1A), and the top 50 DEGs was shown in the heatmap (Fig. 1B), among these DEGs, TYROBP, PRKCD, LGALS1, ITGAM, ITGB2, TIMP1, HOMER3, CCL5 and CD33 were obviously over-expressed in AML and PLOD2, PBX1, PFKM, NAP1L3, MYH10, TSPYL5, FBN1, CRHBP and NUDT11 were down-regulated in AML samples. To further investigate the function of DEGs in AML, the enrichment analysis of GO, including biological processes (BP), cell component (CC), and molecular function (MF) were summarized in Fig. 1C, and the results show that changes in biological process of DEGs were significantly enriched in metabolic process, biological regulation and response to stimulus. As for cellular component, DEGs were enriched in nucleus, membrane-enclosed lumen and membrane. Molecular function of DEGs were mainly participated in protein binding, ion binding and nucleic acid

binding. KEGG pathway analysis (Fig. 1D) revealed that the DEGs were mainly enriched in DNA replication, mismatch repair, and metabolic pathways.

Expression of TFE3/TFEB and its positively correlated genes in AML

TFE3 is up-regulated in AML patients than healthy donors (log FC = 1.13, adjust p value = 0.0038), and the expression pattern was shown in Fig. 2A. Similar to TFE3, TFEB expression was also increased in AML (log FC = 1.21, adjust p value = 0.0191, Fig. 2B). TFE3 and TFEB expression is higher in M5 compare to M0, M1, M2, M3, M4 ($P < 0.05$, Fig. 2A-B). The patients aging from 61 to 80 years seems have higher TFE3/TFEB expression than 21–40 years or 41–60 years patients ($p < 0.05$, Fig. 2A-B). Genes positively correlated with TFE3/TFEB in AML patients were shown in supplementary material 2, and the genes were analyzed further at the cut-off of Pearson-CC ≥ 0.70. The common genes between all upregulated genes in AML patients and TFE3/TFEB positively related genes were 38 and 30 respectively (Fig S1A-1B), of which 18 genes were not only correlated with TFE3 but also positively associated with TFEB (Fig S1C). Interestingly, we found EHBP1L1 and PLXNB2 were decreased in AML patients with PML/RAR-fusion compared to the patients without PML/RAR-fusion (Fig. 2C, $P < 0.05$). Additionally, AML patients with RAS activation have higher expression in ITGAX, ITGAM, RNF19B, GPSM3 and RGS19 than the patients without RAS activation (Fig. 2C, $P < 0.05$).

TFE3/TFEB positively correlated genes were associated with the survival of AML patients

There were total of 50 genes positively corelated with TFE3/TFEB and upregulated in AML (Fig S1C), 38 genes related to TFE3 (Fig S1A) and 30 genes related to TFEB (Fig S1B) respectively, in which 29 genes were associated with AML patient's survival (Fig S1D). 11 genes (RGS19, ITGAM, DOK2, S100A6, FGR, MVP, TNFAIP2, PRKCD, ARAP1, EHBP1L1 and LRP10) of both TFE3 and TFEB corelated DEGs (18 in total) were found to indicate poor survival (Fig. 3) in AML. TFE3 and 9 of its correlated DEGs, namely CTSD, PTPN6, PLXNB2, GPSM3, SELPLG, DNASE1L1, RNF19B, GRINA, SH3BGRL3 were also worse prognostic markers in AML patients (Fig. 3). Additionally, patients with high expression in TFEB and its correlated DEGs POLD4, COTL1, ARPC1B, RHBDF2, ITGAX, TNFRSF1B, ITGB2 have poor prognosis too (Fig. 3). 29 survival related genes expression in AML was shown in Fig S2. Most of the genes were over-expressed in M5 than any other type according to the FAB classification. The PPI network of 29 genes associated with survival were constructed (Fig. 4A). Results showed that genes were mainly enriched in leukocyte migration, integrin-mediated signaling pathway, Fc-gamma receptor signaling pathway involved in phagocytosis, autophagy and cell adhesion (Table 1). Most of the molecular were extracellular exosome and integrin complex. The molecular function was related to staphylococcus aureus infection and cell adhesion molecules (CAMs). Finally, 5 hub genes (ITGB2, FGR, ITGAM, ITGAX, SELPLG) were found (Fig. 4B) from the 29 survival related genes and the functional roles of 5 hub genes with score ≥ 5 was shown

in Table 2. The high expression of the 5 hub genes in AML blasts (Fig. 5A) and the expression in AML patients based on FAB classification and age was shown in Fig. 5B-C. Figure 4E shown the hub genes positively correlation expression with TFE3 or TFEB.TFE3 and TFEB are co-expression in AML patients with a Pearson CC of 0.69, and the Pearson-CC of other genes with TFE3 or TFEB were ≥ 0.07.

Table 1
Significantly enriched GO terms and KEGG pathways of prognosis-predictive DEGs in AML patients ($p < 0.05$). Biological process (BP); Molecular function (MF); Cellular component (CC); KEGG pathway analysis (KEGG).

	Term	Pathway description	Genes	P-value
BP	GO:0050900	leukocyte migration	DOK2, SELPLG, ITGAX, PTPN6, ITGAM	1.32E-06
BP	GO:0007229	integrin-mediated signaling pathway	ITGAX, ITGB2, FGR, ITGAM	5.25E-04
BP	GO:0038096	Fc-gamma receptor signaling pathway involved in phagocytosis	ARPC1B, FGR, PRKCD	1.76E-02
BP	GO:0006914	autophagy	TFEB, RGS19, CTSD	1.92E-02
BP	GO:0007155	cell adhesion	SELPLG, ITGAX, ITGB2, ITGAM	3.65E-02
CC	GO:0070062	extracellular exosome	ITGB2, ARPC1B, COTL1, S100A6, PLXNB2, DNASE1L1, MVP, SH3BGRL3, FGR, PTPN6, CTSD, ITGAM, PRKCD	4.77E-04
CC	GO:0008305	integrin complex	ITGAX, ITGB2, ITGAM	7.80E-04
MF	GO:0019901	protein kinase binding	ITGB2, RGS19, MVP, FGR PTPN6, PRKCD	2.88E-04
MF	GO:0001784	phospho-tyrosine binding	FGR, PTPN6	2.06E-02
KEGG	hsa05150	staphylococcus aureus infection	SELPLG, ITGB2, ITGAM	4.46E-03
KEGG	hsa04514	cell adhesion molecules (CAMs)	SELPLG, ITGB2, ITGAM	2.84E-02

Table 2
Functional roles of hub genes in prognosis-predictive DEGs.

No.	Gene symbol	Full name	Function
1	ITGB2	integrin beta-2	a receptor for ICAM1, ICAM2, ICAM3 and ICAM4
2	FGR	tyrosine-protein kinase FGR	non-receptor tyrosine-protein kinase that transmits signals from cell surface receptors devoid of kinase activity and contributes to the regulation of immune response,including neutrophil monocyte macrophage and mast cell functions
3	ITGAM	integrin alpha-M	adhesive interactions of monocytes, macrophages and granulocytes
4	ITGAX	integrin alpha-X	recognize the sequence G-P-R in fibrinogen and mediates cell-cell interaction
5	SELPLG	P-selectin glycoprotein ligand 1	critical for initial leukocyte capture

Functional analysis of TFE3/TFEB and possible mechanism

Functional partners of TFE3 (11 nodes, 25 edges, PPI enrichment p-value = 0.0002) and TFEB (11 nodes, 33 edges, PPI enrichment p-value = 2.29E-07) predicted by STRING were shown in Fig. 4C-D respectively and more specific function was shown in Table S1-2. Most of the genes were lysosome and transcription factor complex and they were involved in autophagy, positive regulation of transcription from RNA polymerase II promoter and positive regulation of transcription, DNA-templated. The pathways were involved in cancer, lysosome, cell cycle and transcriptional mis-regulation in cancer (Table 3).

Table 3
GO and KEGG analysis of predictive functional partners of TFE3 and TFEB ($p < 0.05$).

Category	Term	Pathway description	Genes	P-value
BP	GO:0006914	autophagy	TFEB, MTOR, CTSD, LAMP1, FOXO1, SQSTM1	1.66E-07
BP	GO:0045944	positive regulation of transcription from RNA polymerase II promoter	MITF, TFEB, SMAD3, SMAD4, TFDP1, FLCN, TFE3, FOXO1, SQSTM1	2.00E-06
BP	GO:0045893	positive regulation of transcription, DNA-templated	MITF, TFEB, E2F3, SMAD3, SMAD4, TFE3, FOXO1	7.50E-06
BP	GO:0010718	positive regulation of epithelial to mesenchymal transition	SMAD3, SMAD4	3.29E-02
CC	GO:0005764	lysosome	FLCN, MTOR, CTSD, LAMP1, CTSK	4.83E-05
CC	GO:0005667	transcription factor complex	E2F3, SMAD3, SMAD4, TFDP1	7.13E-04
MF	GO:0003700	transcription factor activity, sequence-specific DNA binding	MITF, TFEB, E2F3, SMAD3, SMAD4, TFDP1, TFE3	2.41E-04
MF	GO:0046983	protein dimerization activity	MITF, TFEB, E2F3	4.27E-04
KEGG	hsa05200	pathways in cancer	MITF, E2F3, SMAD3, SMAD4, MTOR, FOXO1	1.53E-03
KEGG	hsa04110	cell cycle	E2F3, SMAD3, SMAD4, TFDP1	2.70E-03
KEGG	hsa05202	transcriptional mis-regulation in cancer	ASPSCR1, PRCC, TFE3, FOXO1	6.24E-03

Biological process (BP); Molecular function (MF); Cellular component (CC); KEGG pathway analysis (KEGG)

Discussion

TFEB (transcription factor EB), TFE3 (transcription factor E3) and MITF (melanogenesis-associated transcription factor) are members of the microphthalmia-transcription factor E (MiT/TFE) subfamily (6, 25, 26). Among them, TFE3 and TFEB play crucial roles in controlling autophagy and lysosomal biogenesis (27, 28). Autophagy lysosomal axis dysregulation has been linked to a variety of diseases such as cancers, neurodegenerative diseases, and inflammatory disorders (29, 30). A number of studies have determined TFE3 and TFEB as being oncogenes. Chromosomal translocations resulting in gene fusions involving TFE3 or TFEB are implicated in the development of sporadic renal cell carcinomas (RCC) and soft tissue sarcomas. These genetic rearrangements cause overexpression of the TFE proteins (31–33), and in the case of TFEB-MALAT1 fusion, places TFEB under the control of a more active promoter resulting in a 60-fold higher expression (32). But, up to now, the of TFE3/TFEB expression remains unclear for AML patients.

GO and KEGG analysis show DEGs between AML patients and healthy donors were enriched in metabolic process, myeloid leukocyte activation, mismatch repair and DNA replication. TFE3 and TFEB had already been reported as the "master regulators" of autophagy and lysosomal biogenesis (34), which is crucial in cell metabolism and cell cycle. So, we pay our attention to the common high-expression genes in AML blasts and TFE3/TFEB positively correlated genes expression. We found these genes mainly enriched in integrin-mediated signaling pathway, cell adhesion and leukocyte migration. And surprisingly, we found most of the genes is related to the prognosis of AML patients.

We then focused on the functional partners of TFE3 and TFEB, GO enrichment analysis and KEGG pathway enrichment analysis demonstrated that the functions of these genes are primarily related to the autophagy, positive regulation of transcription, lysosome and pathways in cancer, which is consistent with our prediction. A study also shows that TFE3 is a functional partner for the E2F3 transcription factor (35),which control transcription of genes involved in DNA replication, cell cycle progression, and cell fate determination, which is consistent with our results.

In our research, hub genes, namely ITGB2, FGR, ITGAM, ITGAX and SELPLG were identified in analyzing the PPIs network of survival-related DEGs, indicating these genes may be vital in prognosis of AML patients. ITGB2, ITGAM, ITGAX belong to integrin complex for leukocyte migration, cell adhesion and integrin mediated signaling pathway. HUANG et al. reported ITGAM is one of the most significant modules and correlated with poor overall survival in AML, which is the same as our results. Clinically, ITGAX is considered to be useful in the identification of hairy cell leukemia and in an analysis, CD11c was expressed in 49% of CLL, 57% of B-NHL, and 100% of Hairy cell leukemia and B-PLL (36). Umit E.G., et al. reported ITGAX expression in chronic lymphocytic leukemia is related with complications and survival (37). And in our study, we found ITGAX was up-regulated in AML blast and it predicted a worse prognosis in AML too. What’s more, we found ITGAX and ITGAM expressed higher in RAS-activation patients compared to without activation patients, this revealed ITGAM and ITGAX might have functional roles in RAS-activation signaling. ITGB2 (CD18) was concerned as a chronic lymphocytic leukemia (CLL) susceptibility variant, the relationship of ITGB2 with AML is still unknown, our results showed it also can be defined as a poor prognostic biomarker. AML is often associated with constitutive tyrosine kinase

signaling, these pathways involve the non-receptor tyrosine kinases Fes, Syk and the three Src-family kinases expressed in myeloid cells (Fgr, Hck, and Lyn), in our research, FGR, HCK, LYN is overexpressed in AML blasts as predicted, and FGR is one significant survival marker. One study had found selective inhibition of the myeloid Src-family kinase Fgr potently suppresses AML cell growth in vitro and in vivo(38), which indicate that FGR might be a therapeutic target for AML. SELPLG encodes a membrane-associated glycoprotein that binds to not only P-selectin, but also E- and L- selectin, and is important for the recruitment of leukocytes to sites of inflammation. SELPLG amplification have been proved in Primary effusion lymphoma (39), and the protein encoded by SELPLG is important for cell migration. What's more, in a murine lymphoma metastasis model, SELPLG expression was critical for lymphoma cell colonization and tumor formation in the liver and spleen (40) .However, the roles of SELPLG in AML haven't been reported.

The autophagy–lysosome system is a catabolic cellular process for whole organelles, protein aggregates, and other macromolecules (41). During carcinogenesis, autophagy exerts an antitumorigenic effect by degrading and/or recycling damaged cellular organelles. However, following tumor induction, cancer cells coopt autophagy as a cell survival mechanism to promote nutrient reallocation for diverse cellular needs. Therefore, autophagy can suppress cancer development through its cytoprotective properties; however, once cancer has developed, these same properties sustain survival of the tumor (42). TFE3/TFEB are regulators of the autophagy–lysosome system, and increasing evidence suggests that they also directly regulate metabolic and growth signaling pathways, and as such, represent an attractive therapeutic target with wide potential cancer. But with respect to cancer, impacts on immune system function merit further investigation.

Conclusions

In this study, we used comprehensive bioinformatics analyses to identify prognosis-predictive genes related to TFE3/TFEB, the expression of which was significantly related to survival in AML patients, which may help us further investigate the role that the TFE3/TFEB play in AML.

Methods

Data source

We obtained gene expression profiling data (GSE 9476) from the Gene Expression Omnibus (GEO) database at the National Center for Biotechnology Information. It was submitted by Stirewalt DL based on the GPL96 [HG-U133A] Affymetrix Human Genome U133A Array platform. We extracted gene expression profiling data of leukemic blasts from 26 AML patients and normal CD34 + hematopoietic cells from 18 healthy donors. The genes expression and correlated genes in AML patients were obtained from UALCAN (14) (http://ualcan.path.uab.edu/analysis.html), a comprehensive web resource, provides analyses based on The Cancer Genome Atlas (TCGA) and MET500 cohort data.

Identification of DEGs

GEO provides users with a useful tool called GEO2R that can be used to analyze microarray data. GRO2R (https://www.ncbi.nlm.nih.gov/geo/geo2r/) were used to analyze gene expression in GSE 9476. Genes were considered statistically significant as |Log FC| ≥1.0 and adj P < 0.05.

GO and KEGG analysis

GO enrichment analysis, including Biological Process, Cellular Component, Molecular Function, and GO functional pathway for the DEGs were performed by using Over-Representation Analysis (ORA) based on WebGestalt (15–18) (http://www.webgestalt.org/). We used DAVID (19, 20) (https://david.ncifcrf.gov/) to make KEGG pathway analysis.

Venn diagram of common genes

Through the GO analysis, we identified the expression of aimed gene and its correlation expression genes in AML. Then, we use the Venn diagram network tool to draw the Venn diagram. (http://bioinformatics.psb.ugent.be/webtools/Venn/).

Survival analysis

Survival analysis was performed by Kaplan-Meier survival plots from TCGA portal (21) (http://www.tcgaportal.org/), log-rank P value 0.05 was concerned to be significant.

Protein–protein interactions network construction and hub genes identification

Protein–protein interactions (PPIs) network is important to reveal and annotate all functional interactions among cell proteins. For this study, the online database resource STRING (22, 23) (Search Tool for the Retrieval of Interacting Genes/Proteins) was used (http://string-db.org). The hub genes associated with survival were identified by Cytoscape (24). In this study, the PPIs network analysis of key genes and predicted functional partners were also found in STRING.

Abbreviations

AML: Acute myeloid leukemia; DEGs: Differentially expressed genes; BM: bone marrow; OS: overall survival; BP: biological processes; CC: cell component; MF: molecular function; log FC: log2 fold change; GEO: Gene Expression Omnibus; PPIs: Protein–protein interactions; TCGA: The Cancer Genome Atlas.

Declarations

Ethics approval and consent to participate:

Not applicable.

Consent for publication:

Not applicable.

Availability of data and materials:

The datasets generated and/or analysed during the current study are available from the Gene Expression Omnibus (GEO) database (GSE 9476) at the National Center for Biotechnology Information. The genes expression and correlated genes in AML patients were obtained from a comprehensive web resource UALCAN (http://ualcan.path.uab.edu/analysis.html), which provides analyses based on The Cancer Genome Atlas (TCGA) and MET500 cohort data.

Competing interests:

The authors declare that they have no competing interests.

Authors' contributions:

Q. L. conceived the study, collected and analyzed the data and wrote the manuscript. Q. L. approved the final manuscript.

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Figures

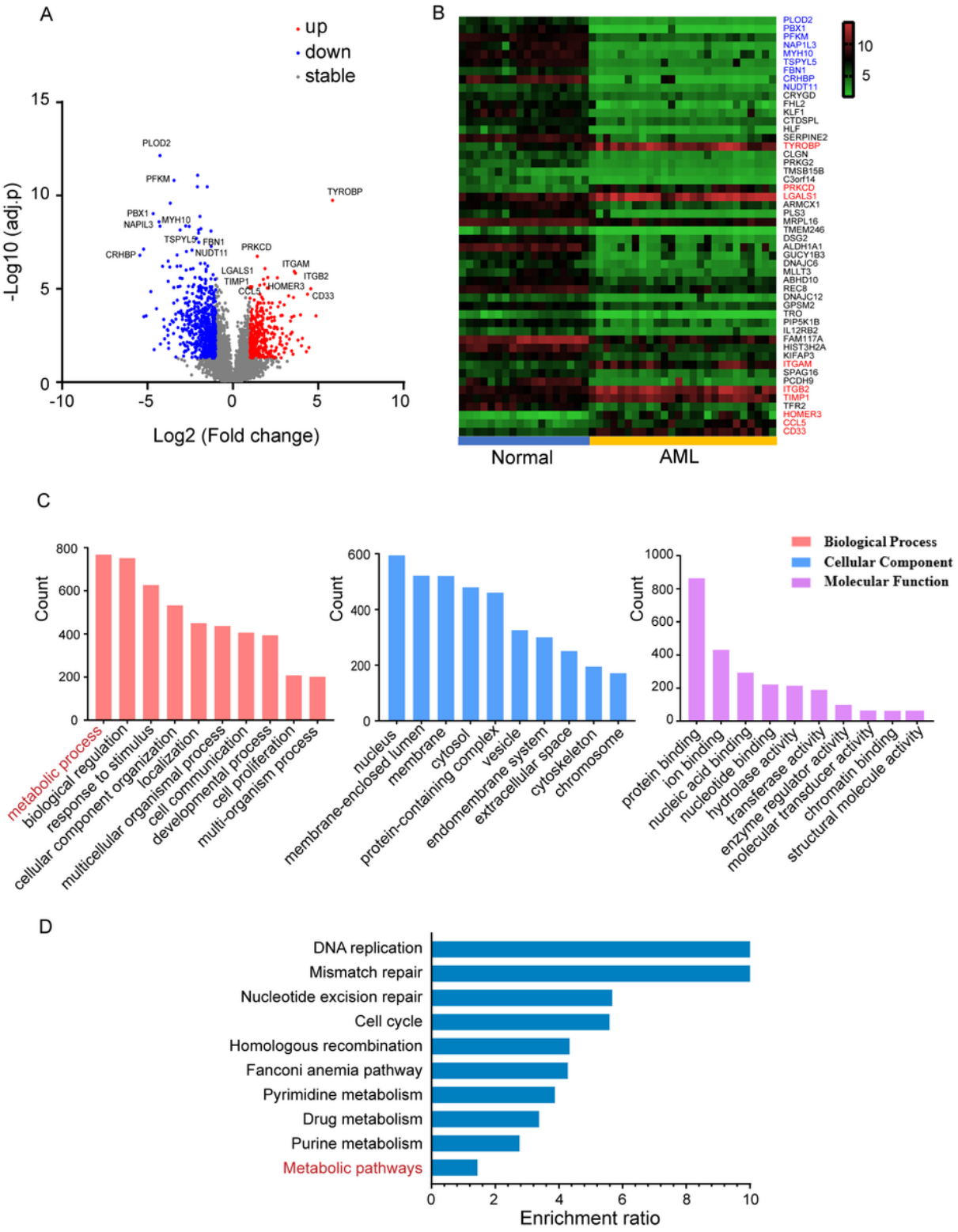

Figure 1

Different expression genes (DEGs) analysis and pathway enrichment of DEGs. The cut-off criteria for DEGs: |log FC| ≥1.0 and adjusted P-value < 0.05, logFC: log fold change, adj.p: adjusted P value. (A) Volcano blot of the genes expression. up means up-regulated genes in red dots, down: down-regulated genes in blue dots, stable: not changed genes, which were grey dots; (B) Heatmap of the top 50 DEGs. 26 AML patients compared to 18 healthy donors. The color in heat maps from green to red shows the

progression from low expression to high expression; (C) GO functional pathway enrichment of DEGs. BP: biological process (in pink), CC: cellular component (in blue) and MF: molecular function (in purple); (D) KEGG pathways of DEGs (FDR≤0.05).

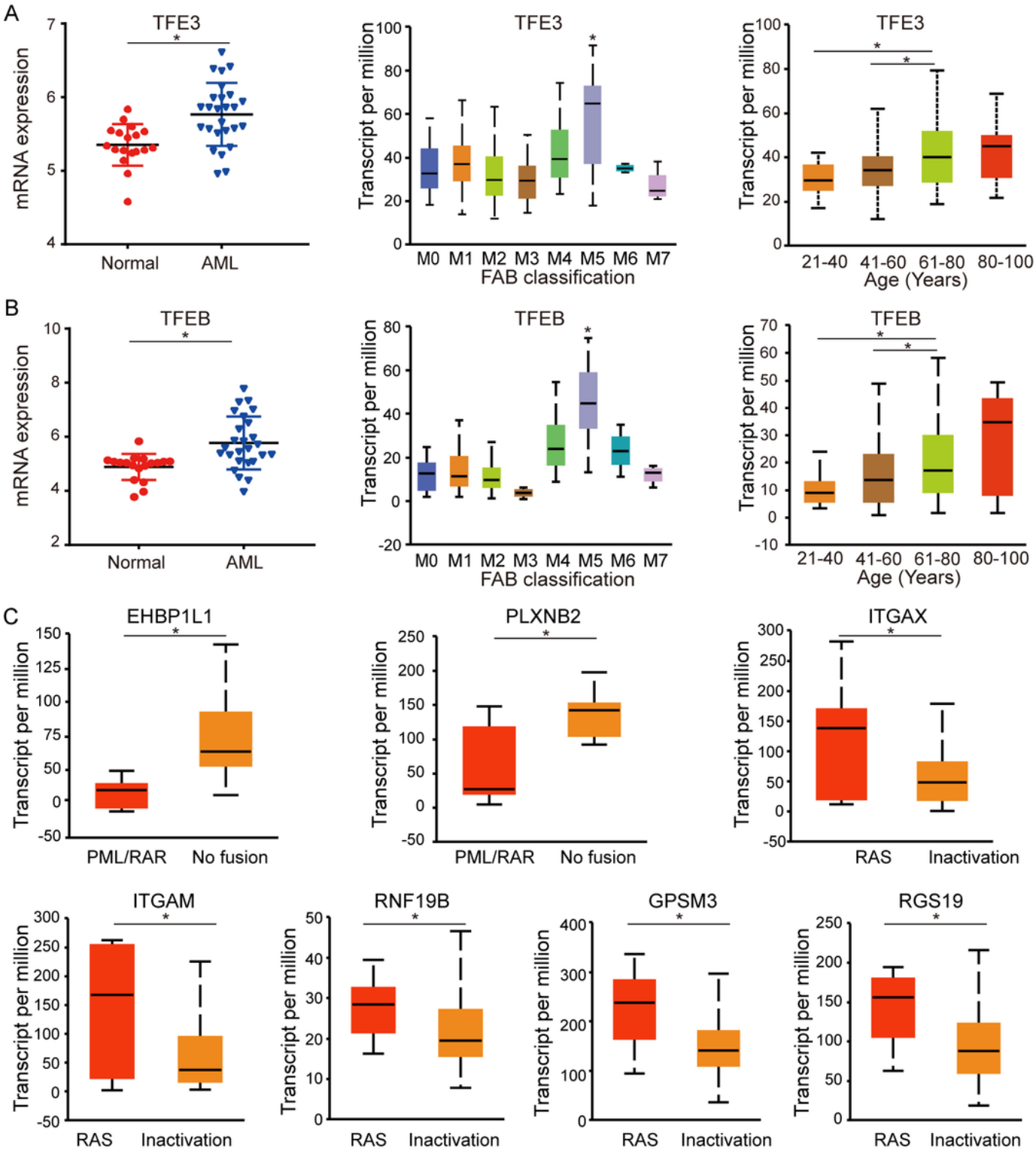

Figure 2

Gene expression in AML. (A-B) TFE3/TFEB expression in AML. Different TFE3/TFEB expression between AML patients (blue) and healthy donors (red) from GSE9476 (left); TFE3/TFEB expression in AML based

on French American British classification from TCGA samples (medium), M0: n=16, M1: n=42, M2: n=39, M3: n=16, M4: n=35, M5: n=18, M6: n=2, M7: n=3; TFE3/TFEB expression in AML based on age from TCGA (right), 21-40 years: n=34, 41-60 years: n=61, 61-80 years: n=72, 81-100 years: n=5; (C) Aberrantly expressed genes in AML patients with PML/RAR-fusion (EHBP1L1, PLXNB2) or with Ras activation (ITGAX, ITGAM, RNF19B, GPSM3, RGS19).

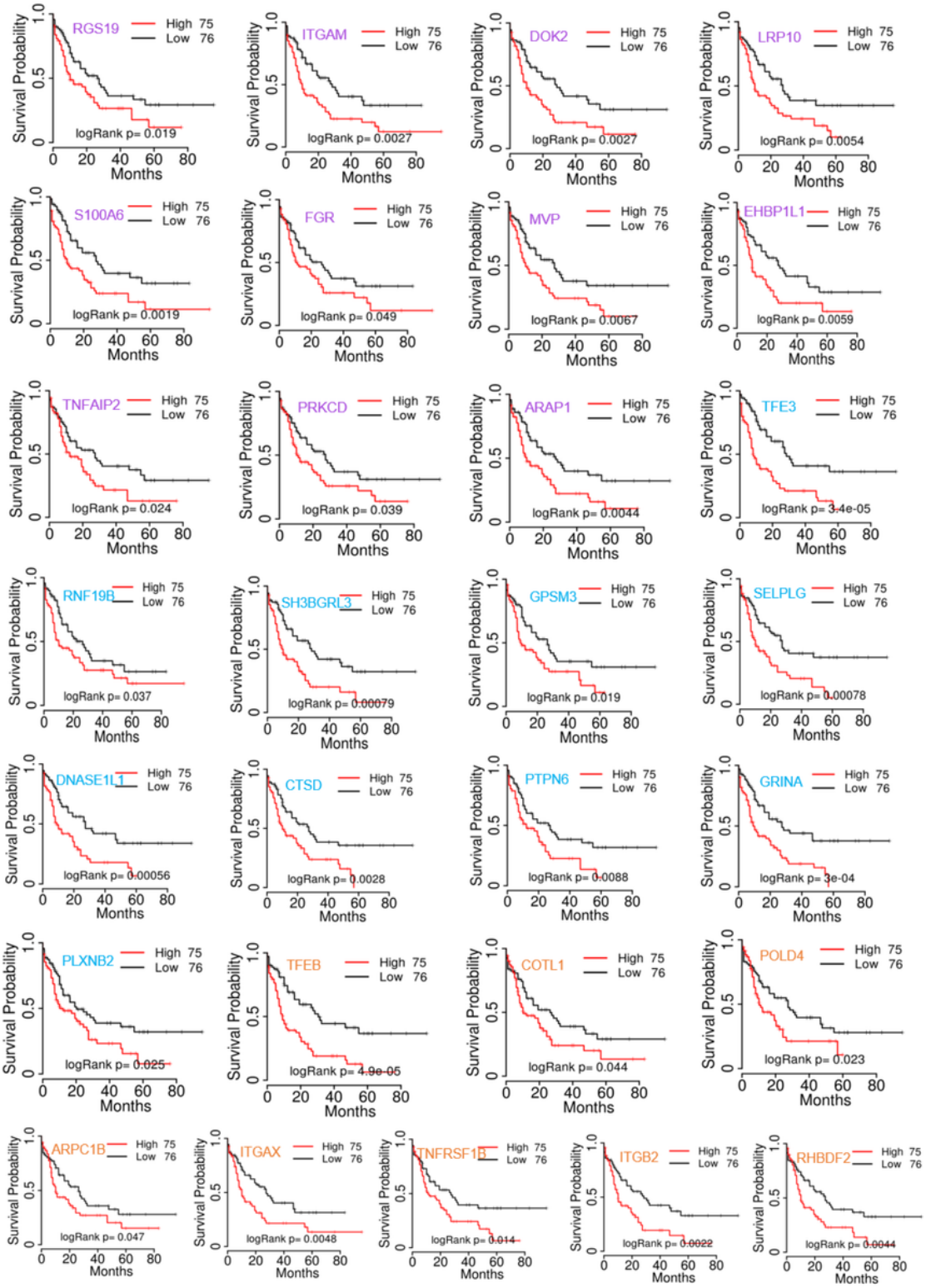

Figure 3

Prognosis of TFE3/TFEB positively related DEGs in AML. Purple means the DEGs are positively corelated to both TFE3 and TFEB, blue means the DEGs are just positively corelated to TFE3, and orange represents the DEGs are positively corelated to TFEB.

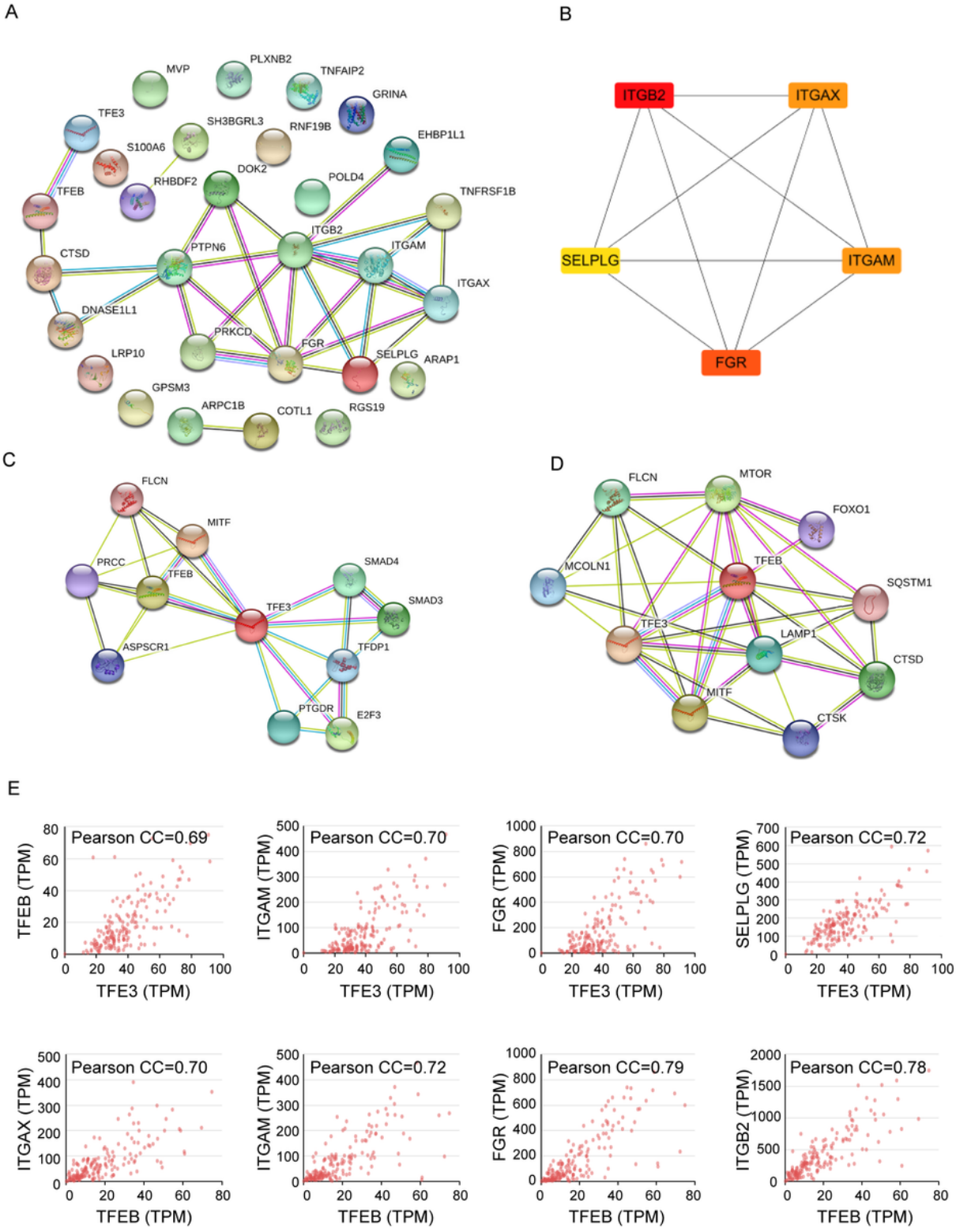

Figure 4

Interactions between TFE3/TFEB and its related genes. (A) Protein-protein interactions network of 29 survival-associated genes which positively corelated with TFE3/TFEB and high expressed in AML; (B)

Hub genes of survival-related genes; (C) Functional analysis of TFE3 partners depicted by STRING; (D) Functional analysis of TFEB partners depicted by STRING; (E) Gene expression correlation between TFEB/TFE3 and the hub genes in AML.

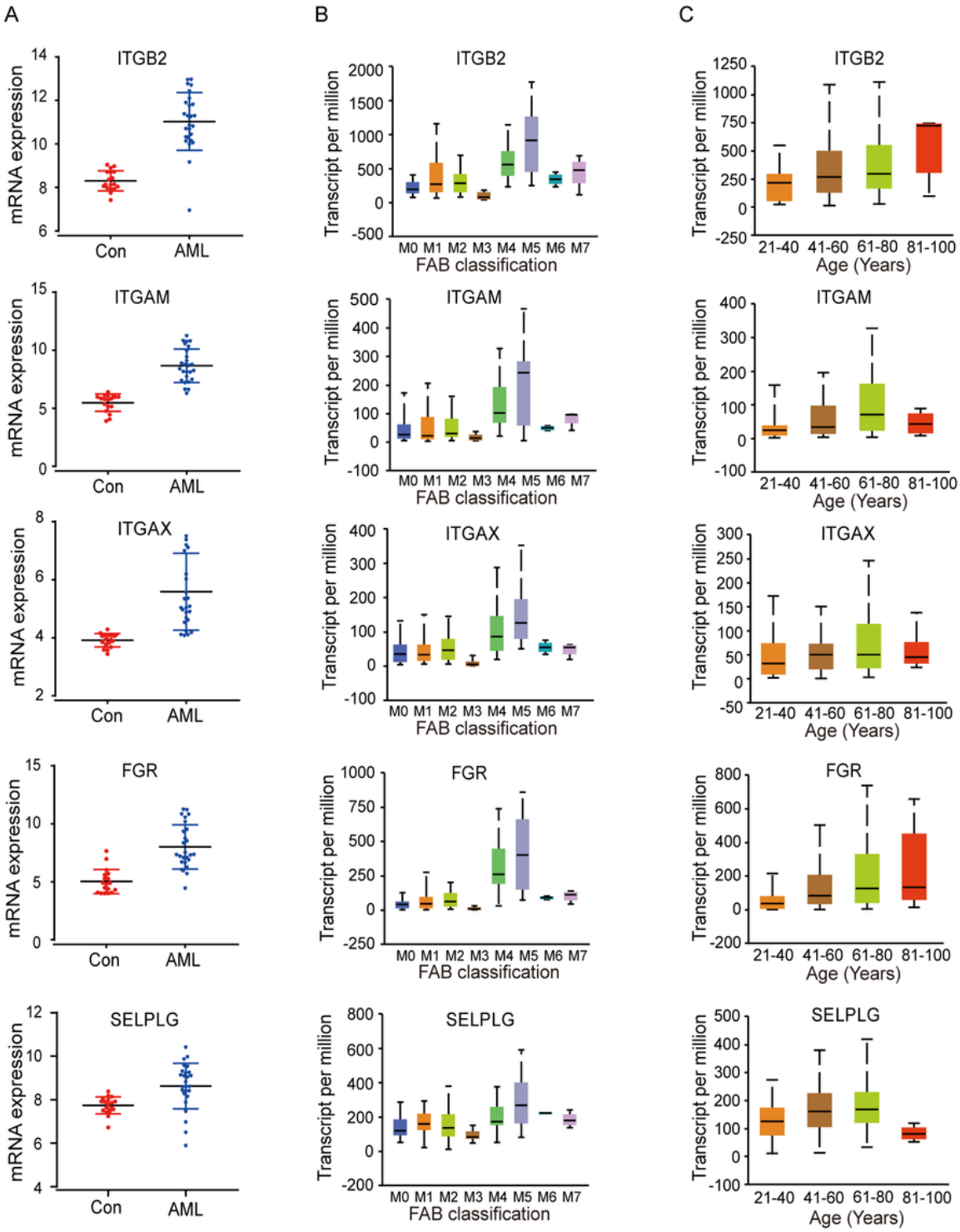

Figure 5

Expression of survival-associated hub genes in AML. (A) Different expression of hub genes in AML patients compared to healthy donors (left penal); (B) Hub genes expression in AML based on French

American British classification from TCGA samples (medium penal), M0: n=16, M1: n=42, M2: n=39, M3: n=16, M4: n=35, M5: n=18, M6: n=2, M7: n=3; (C) Hub genes expression in AML based on age from TCGA samples (right penal), 21-40 years: n=34, 41-60 years: n=61, 61-80 years: n=72, 81-100 years: n=5.

Supplementary Files

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- Supplementaryfiguresandtables.docx