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The Prognostic Values and Clinical Implications of m6A Methylation Regulators in Epithelial Ovarian Cancer

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Abstract

N6-methyladenosine (m6A) exists in both DNA and RNA modification. RNA m6A modification drives tumor initiation and metastasis through regulating cancer stem cells. However, the detailed mechanisms and the distinct m6A regulatory gene type underlying ovarian cancer mRNA modification remain unclear. Here, we analyzed Copy Number Variation (CNVs) and mRNA expression of ovarian cancer cases in TCGA dataset to determine the copy number variation patterns of m6A regulatory genes, and the associations between m6A dysregulation or certain regulatory gene and overall survival. We showed the *KIAA1429*, as the writer gene, had highest amplification percentage and were associated with overall survival, or disease-free survival, whereas the associations with prognostic survival were independent of other prognostic factors including stage, grade, and debulking status of the tumour. Besides, *METTL14* and *YTHDC2*, one as the writer gene and the other as reader gene, was also related with clinical outcome. Furthermore, subgroups analysis addressed that m6A upregulation especially writer gain contributed to prognosis in epithelial ovarian cancer. Collectively, our data addressed that m6A upregulation are likely to be critical to the clinical outcome, and *KIAA1429* showed the highest correlation with clinical outcome in ovarian cancer among m6A regulatory genes.

Keywords: RNA modification; Methyltransferase; Demethylases; Epigenetics; Prognostic signature

Abbreviations

m6A: methylation of N6 Adenosine; EOC: Epithelial Ovarian Cancer; TCGA: The Cancer Genome Atlas; CNV: Copy Number Variation; OS: Overall Survival; DFS: Disease-Free Survival; PFS: Progression-Free Survival; HR: Hazard Ratio; K-M plotter: Kaplan-Meier Plotter; GEO: Gene Expression Omnibus; EMT: Epithelial-Mesenchymal Transition; mRNA: messenger RNA; GIST: Genomic Identification of Significant Targets in Cancer; AML: Acute Myelocytic Leukemia; ROS: Reactive Oxygen Species; HIF-α: Hypoxia Induced Factor-α; GSEA: Gene Set Enrichment Analysis

Introduction

Epithelial Ovarian Cancer (EOC) is the most prevalent cause of death in gynecologic malignancies. The patients are usually diagnosed at advanced stages for the lacking of clinically meaningful biomarkers for early screening and understanding of molecular pathogenesis. Debulking surgery followed by chemotherapy can cause 50% to 80% complete clinical response in advanced stage cases. However, most cases will relapse and eventually die for recurrence or metastasis [1,2]. Recent studies focused on target therapy or immune therapy which is proven to prolong survival time in advanced-stage ovarian cancer patients. But the molecular pathogenesis and mechanism is underestimated. Hence, finding the functionally relevant molecular biomarkers and new therapeutic targets are still challenging. N6-methyladenosine (m6A) exists in both DNA and RNA modification, namely m6dA and m6A. Recently, m6A is proved to be the most abundant in mRNA modification, and the most powerful in the regulation of mRNA [3]. m6A regulators consists of many catalysase including methyltransferase "writers" (*WTAP*, *METTL3*, *METTL14*, MT-A70 and *KIAA1429*), binding proteins "readers" (*YTHDF1*, *YTHDF2*, *YTHDF3*, *YTHDC1* and *YTHDC2*), and demethylase "erasers" (*FTO* and *ALKBH5*) [4]. M6A dysregulation exists in various process including stem cell fates, somatic cell reprogramming, and causes impaired cell proliferation, self-renewal capacity, developmental defects and cell death [5-7]. Articles showed that RNA m6A modification drives tumor initiation and metastasis through regulating cancer stem

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Table 1: Mutation of m6A regulatory genes in epithelial ovarian cancer patients from TCGA dataset.

Sample ID	Altered	*ALKBH5*	*FTO*	*METTL14*	*METTL3*	*WTAP*	*VIRMA*	*RBM15B*	*RBM15*	*ZC3H13*	*YTHDF1*	*YTHDF2*	*YTHDF3*	*YTHDC1*	*YTHDC2*	*HNRNPC*
TCGA-10-0938-01	1										H365Y					
TCGA-13-0920-01	1		S246I								V200I					
TCGA-23-1117-01	1											R355W				
TCGA-25-1319-01	1													R575*		
TCGA-61-2008-01	1								G563A							
TCGA-09-2051-01	1									K385N						
TCGA-24-2271-01	1	Q174R														

cells [8]. It is found to be associated with tumorigenesis in different cancers types, including breast cancer, hematologic malignancies [9-11]. Specially, the overall level of m6A regulators is found to be higher in ovaries compared to other organs and tissues, suggesting that m6A modification plays a critical role in reproduction system [12]. One finding suggested that *METTL3* has an oncogenic role in ovarian cancer development and aggressiveness [13]. The other finding showed that *ALKBH5* was a candidate oncogene and a potential target for ovarian cancer therapy [14]. However, due to the heterogeneity and bias, comprehensive analysis is needed to analysis the expression of m6A regulators, the roles of m6A regulators in cancer progression and metastasis, the function in clinical survival, the correlation between m6A regulators and clinicopathological characteristics in ovarian cancer. Hence, we systematically analyzed the ovarian cancer cases from the TCGA database including the clinical and sequencing data of 579 cases, evaluated the expression spectrum of specific m6A gene, and addressed the association between the genetic alteration or mRNA expression and clinicopathological features or clinical outcomes. In this study, we found that the alteration and mRNA expression level of m6A RNA methylation regulators play a critical role in EOC progression and metastasis.

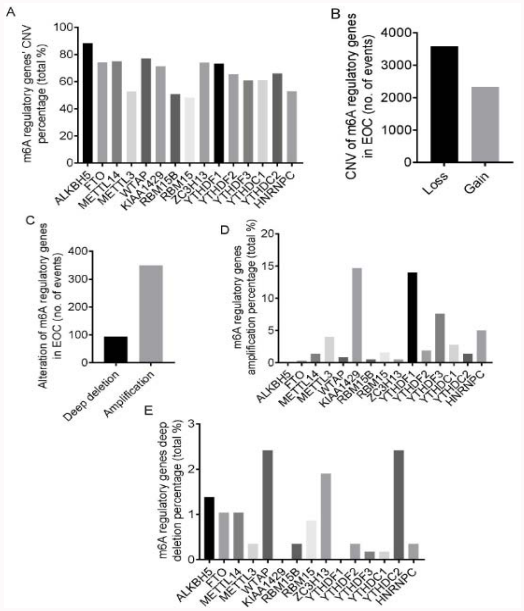

Figure 1: CNVs patterns of m6A regulatory genes in EOC. A) M6A regulatory genes' CNV percentage of EOC samples from TCGA. B) M6A regulatory genes' copy number gain or loss events in EOC patients. C) M6A regulatory genes' amplification or deep deletion. D) M6A regulatory genes' amplification percentage of EOC samples from TCGA. E) M6A regulatory genes' deep deletion percentage of EOC samples from TCGA.

Material and Methods

Data collection

We download the CNV data, mRNA expression, mutation, and clinical data of TCGA dataset by cBioportal platform (http://www.cbioportal.org/) and TCGA-assembler (http://www.compgenome.org/TCGA-Assembler/), which is open for the public.

Data processing

There are 579 ovarian cancer patients with CNV data from the TCGA database. Segmentation analysis and GISTIC algorithm were used to identify the copy-number alteration [15]. There are 316 samples with mutation data and CNVs information. 379 cases were included with mRNA expression data and 3 replicates were removed by patient id. Clinical data and prognostic factors included survival data (overall survival and progression free survival), age, stage, grade, debulking status (optimal and suboptimal), and chemotherapy response (sensitivity and resistant).

Gene set enrichment analysis (GSEA)

Gene set enrichment analysis (http://software.broadinstitute.org/-gsea/index.jsp) is a method to detect the critical biological process in ovarian cancer by genes enrichment analysis comparing two distinct biological status. In our study, two groups were included according the quartile of *KIAA1429* mRNA expression level, with one group are cases with lowest quartile expression and the other group with highest quartile expression. Process with normalized p-value <0.05 and FDR <0.25 were identified to be significantly enriched.

Kaplan-Meier (K-M) plotter database

The Kaplan Meier plotter (http://kmplot.com/analysis) is based on an online database [16] and is capable to assess the association of genes on survival in ovarian cancer. K-M plotter was mainly used in validation for the prognosis value of *KIAA1429* mRNA expression. The correlation of individual *KIAA1429* mRNA expression and survival in ovarian cancer was analyzed online and presented with the hazard ratio, 95% confidence intervals and computed log rank p-value.

Statistical analysis

All the statistical analysis was conducted by R language 3.6.1 (https://www.r-project.org/). We analyzed the association between clinicopathological features or prognostic factors with the CNV or mRNA expression level of m6A regulatory genes with chi-square test. Kaplan-Meier curve and Cox regression analysis were used for univariate or multivariate analysis and for evaluation of the prognosis value of m6A regulatory gene. Cox proportional hazard regression

Table 2: CNV patterns in epithelial ovarian cancer samples (n=576).

		Diploid	Deep deletion	Shallow deletion	Copy number gain	Amplification	CNV Sum	Percentage	sum
Eraser	*ALKBH5*	68	8	484	19	0	511	0.882556131	579
	FTO	149	6	378	44	2	430	0.742659758	579
Writer	*METTL14*	146	6	387	32	8	433	0.747841105	579
	METTL3	274	2	166	114	23	305	0.526770294	579
	WTAP	134	14	359	67	5	445	0.768566494	579
	KIAA1429	167	0	72	255	85	412	0.711571675	579
	RBM15B	286	2	190	98	3	293	0.506044905	579
	RBM15	300	5	104	161	9	279	0.481865285	579
	ZC3H13	150	11	351	64	3	429	0.740932642	579
Reader	*YTHDF1*	155	0	28	315	81	424	0.732297064	579
	YTHDF2	201	2	220	145	11	378	0.652849741	579
	YTHDF3	227	1	64	243	44	352	0.607944732	579
	YTHDC1	225	1	239	98	16	354	0.611398964	579
	YTHDC2	197	14	308	52	8	382	0.659758204	579
	HNRNPC	273	2	160	115	29	306	0.528497409	579

model was performed using R language. Results with a p-value <0.05 were considered to be significant.

Results

Alteration of m6A regulatory genes in EOC patients

With the analysis from sequencing data in TCGA, only 20 independent samples were identified with m6A genes mutations (Table 1). *TP53* was shown with high mutation (88%) in this cohort, which is in line with published literatures [17]. However, Copy Number Variations (CNVs) of m6A regulatory genes that included deep deletion/shallow deletion/ copy number gain/ amplification, had high frequency in EOC cases (Figure 1A), with loss of copy number (3584/5733) in most of the CNV events (Figure 1B) (Table 2). Moreover, for alteration only including amplification and deep deletion as shown in cBioportal database, m6A regulatory genes showed higher frequency of amplification (Figure 1C, S1A). Among them, *KIAA1429* or *VIRMA* (writer) showed the highest frequency of amplification (85/576) in this cohort (Figure 1D, S1B), *WTAP* (14/576) and *YTHDC2* (14/576) had relative high frequency of deep deletion (Figure 1E) (Table 2), which are m6A "writer" and "reader" genes, showing the important of writers in m6A regulation.

Correlation between M6A regulatory genes alterations and clinicopathological factors

To address the correlation between alteration (deep deletion or amplification) of m6A regulatory genes and the clinicopathological and molecular characteristics, we assessed the related prognostic factors including age, stage, grade, debulking status, and chemotherapy sensitivity. The results depicts that the amplification or deep deletion of m6A regulatory genes were associated with grade (Table 3, p=0.016), but not with age, stage, debulking status and chemotherapy response. It is reported that missense mutations of *TP53* were found to be the most frequent, while mutations of BRCA*1* and BRCA*2* showed the critical prognostic value in ovarian cancers [18]. *TP53* and BRCA mutation play a critical role in the progression and pathogenesis of ovarian cancer. Here we evaluated the association between the variation of m6A regulatory genes and the mutation of these genes. In fact, the alterations of m6A regulatory genes were not significantly correlated with BRCA and *TP53* mutations (Table S1), which indicated that m6A regulation affects ovarian cancer through other critical genes and molecular pathways.

Table 3: Clinicopathological features of EOC patients with or without m6A regulatory genes' alteration. Alteration, amplification or deep deletion.

		With alteration	Without alteration	p
Age	≤ 60	139	175	0.479
	>60	120	134	
Stage	I	6	11	0.213
	II	11	19	
	III	191	242	
	IV	47	40	
Grade	G1	0	6	0.016
	G2	24	43	
	G3	227	252	
Debulking status	Optimal	161	201	0.495
	Sub-optimal	67	73	
Chemotherapy sensitivity	Sensitivity	88	106	0.259
	Resistant	34	55	

Association between CNVs of m6A regulatory genes and survival of EOC patients

To identify the association between m6A regulatory genes alteration and ovarian cancer patients' survival, we evaluated the clinical value of CNVs on the Overall Survival (OS) and Progression-Free Survival (PFS) among all ovarian cancer patients. Figure 2A-2B addressed that there was no significant difference of OS or PFS between individuals with or without alteration of m6A regulatory genes including gene amplification or deep deletion. Because cases number was limited and small with amplification or deep deletion in individual regulatory gene, here we analyzed the prognostic value of alteration with the CNVs data in individual gene. We found that copy number gain or amplification of *KIAA1429* (writer, Hazard Ratio [HR] =0.70, p=0.0012), while shallow deletion or deep deletion of *YTHDC2* (reader, HR=0.75, p=0.0087) were related with

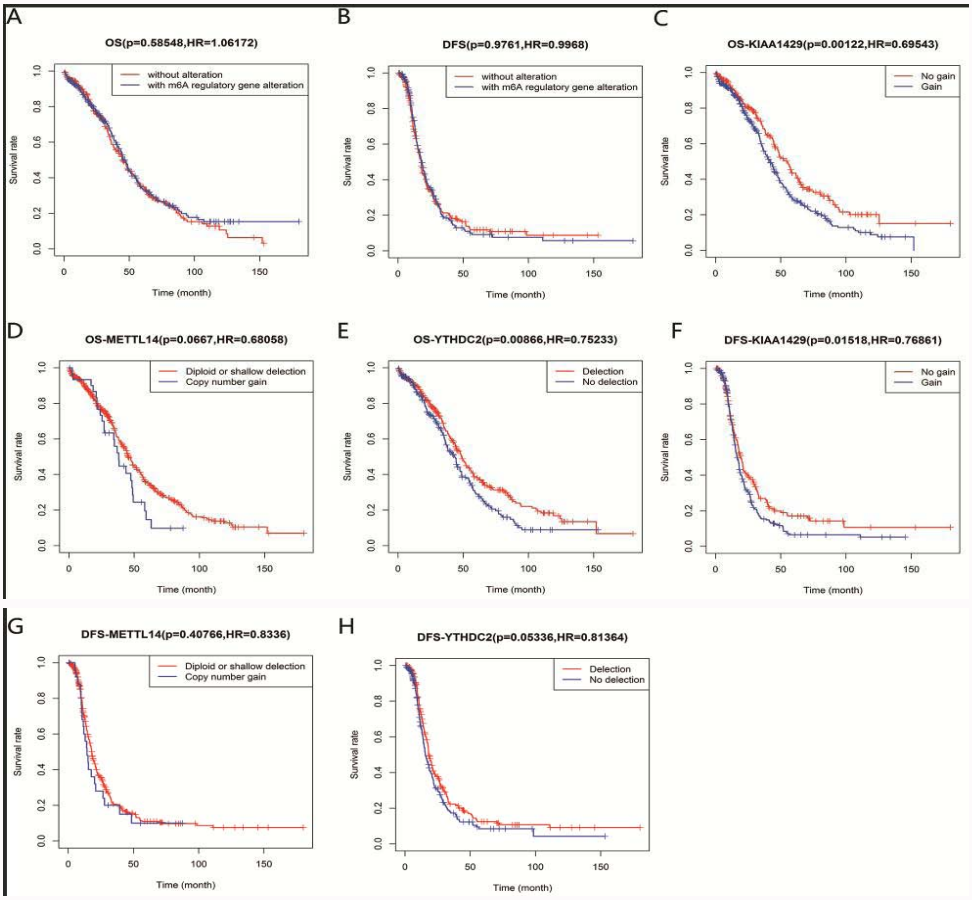

Figure 2: Overall survival of EOC patients with or without m6A regulatory genes' alteration. A,B) OS and DFS of EOC patients with alteration of m6A regulatory genes or without alteration or diploid genes. C,D) OS and DFS of EOC patients with alteration of KIAA1429, *METTL14* and *YTHDC2*.

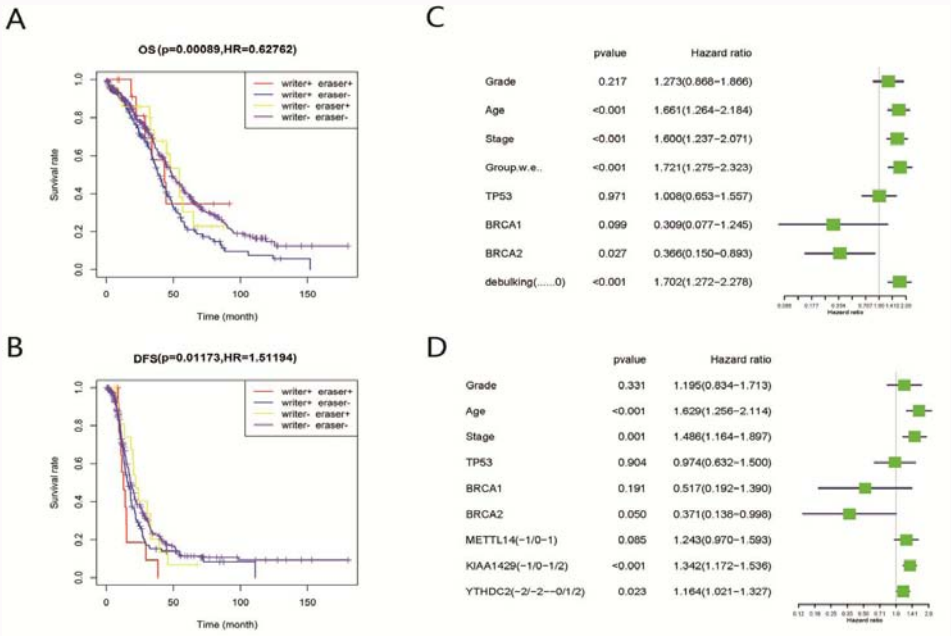

Figure 3: Overall survival of EOC patients with simultaneous alterations of writer genes and eraser genes. A,B) OS and DFS of EOC patients with simultaneous alterations of writer genes and eraser genes. C) Univariate COX regression analysis of simultaneous alterations of writers and erasers for EOC patients' overall survival. D) Univariate COX regression analysis of alteration of KIAA1429, *METTL14* and *YTHDC2* for EOC patients' overall survival.

improved OS and PFS, and copy number gain of *METTL14* (writer) was bordline significantly related with poor OS but not PFS (Figure 2C and 2D for OS and PFS, respectively). The prognostic value of other ten genes was shown in Figure S2 with no significant difference

Table 4: Gene sets enrichment with subgroups of *KIAA1429* expression level in quartile.

GS DETAILS	SIZE	ES	NES	NOM p-val	FDR q-val
NUCLEAR_HORMONE_RECEPTOR_BINDING	28	0.67	2.13	0	0.038
SMALL_CONJUGATING_PROTEIN_LIGASE_ACTIVITY	50	0.57	2.12	0	0.019
ACID_AMINO_ACID_LIGASE_ACTIVITY	56	0.56	2.12	0	0.013
TRANSCRIPTION_FROM_RNA_POLYMERASE_II_PROMOTER	64	0.58	2.11	0	0.011
RNA_METABOLIC_PROCESS	118	0.53	2.01	0	0.020

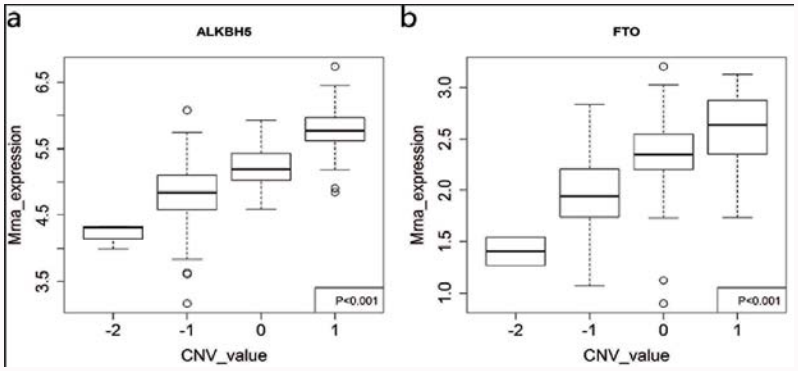

Figure 4A: Different CNV patterns correlated with mRNA expression level of m6A regulatory genes for eraser genes.

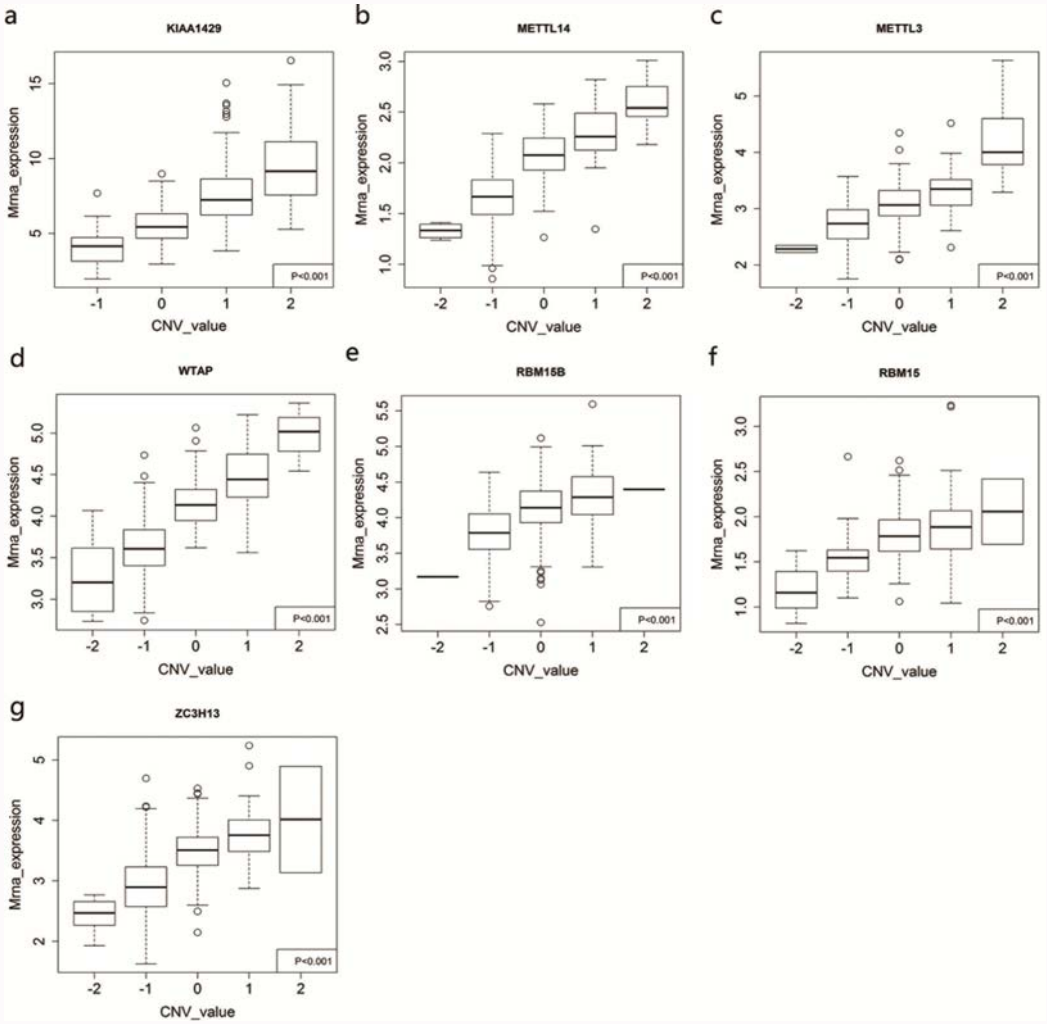

Figure 4B: Different CNV patterns correlated with mRNA expression level of m6A regulatory genes for writer genes.

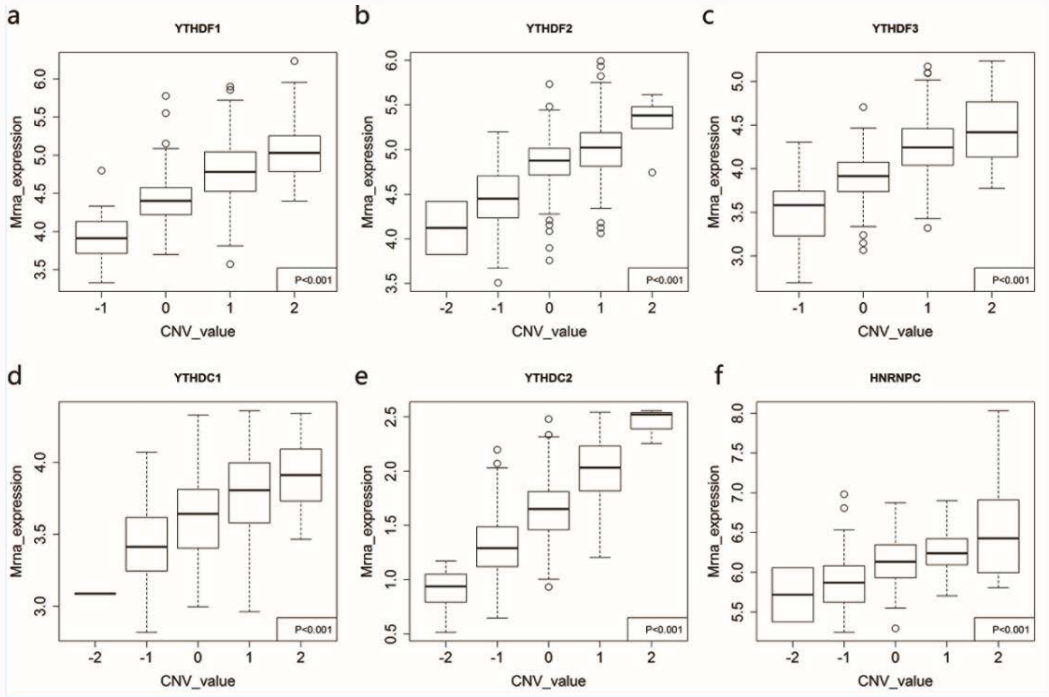

Figure 4C: Different CNV patterns correlated with mRNA expression level of m6A regulatory genes for reader genes.

between different subgroups based on CNVs. The results also showed that writer genes, a group of methyltransferase enzymes, might be important for patient survival. Next, we conducted survival analysis with subgroups of CNVs (copy number gain of writer genes and deletion of eraser genes) to test the prognostic role of m6A regulatory genes especially the writers. Figure 3A and 3B depicts that in eraser deletion patients, the group combination with writer genes gain had worse OS and DFS than those without writer genes gain. This evidence supported the correlation between up-regulated m6A level (especially writer genes gain) and poor survival. By univariable Cox regression analysis adjusted for prognostic factors including stage, grade, BRCA mutation, age, debulking status, and subgroups, we addressed that alteration of m6A regulator genes was an important risk factor independent of other factors (Figure 3C, Table S2, HR=1.51, 95% CI (1.12 to 2.1), p=0.008). Furthermore, *KIAA1429* was independent risk factor for overall survival when conducting multivariate analysis adjusted for prognostic factors (Figure 3D, Table S3, HR=1.30, 95% CI (1.13 to 1.51), p<0.001).

Association between m6A genes level and survival in EOC patients

The effects of alterations in m6A regulatory genes on the mRNA expression were evaluated. Figure 4A-4C addressed that the mRNA expression were significantly correlated with the diverse CNV patterns. Higher mRNA level was shown to be related with amplification or copy number gains for all the m6A regulator genes, while decreased mRNA expression was shown in shallow deletion or deep deletions for the m6A regulator genes. Considering the important role of m6A regulator genes in pathological feature and survival above with the CNV data, we investigated the relationship between individual m6A regulator genes mRNA level and clinical survival. K-M analysis was conducted with the expression data in the TCGA dataset. Figure 5A and 5B depicts the unadjusted HRs and 95% confidence intervals for medium of gene expression levels with OS and PFS. Three seventy six patients with restriction to samples with mRNA data were included for analysis. *KIAA1429* (HR=1.27, p=0.063, Figure 5C), *YTHDC2* (HR=1.31, p=0.039, Figure 5D) were most significantly associated with poor overall survival, and *YTHDC2* (HR=1.39, p=0.011, Figure 5E) showed significant correlation with poor PFS. However, *KIAA1429* showed no significant association with the poor PFS but with the tendency (Figure 5F). In general, the results were in consistent with the prognostic value of m6A genes alteration above that copy number gain or amplification of writers was associated with poor prognosis, illustrating that high m6A expression correlated the poorer outcome.

Validation of prognostic role of *KIAA1429* in EOC patients

To validate the prognostic significance of *KIAA1429* expression in ovarian cancer patients, we integrated the expression profiling in Kaplan-Meier plotter online database (K-M plotter). We evaluated the prognostic value of *KIAA1429* at mRNA level by Kaplan-Meier plotter analysis with cases enrolled from multiple GEO (Gene Expression Omnibus) datasets. Figure 6A, 6B showed that higher *KIAA1429* expression was correlated with shorter overall survival by detecting with different probes (223110_at, HR=1.36 (1.1-1.67), p=0.0038 and 238818_at, HR=1.31 (1.06-1.64), p=0.014), with the PFS shown in Figure 6C, 6D (HR=1.31 (1.08-1.6), p=0.0055; HR=1.37 (1.13-1.66), p=0.0015, respectively).

Gene set enrichment analysis of *KIAA1429* expression level

As mentioned above, m6A upregulation, especially *KIAA1429* gain, is important to promote cancer pathogenesis and progression. Herein we evaluated the molecular mechanism of m6A upregulation in EOC by GSEA analysis (Gene set enrichment analysis). We explored the gene set enrichment between samples with different *KIAA1429* mRNA expression. The GSEA results showed that high *KIAA1429* expression was associated with some critical pathway. Nuclear hormone receptor binding, small conjugating protein ligase activity,

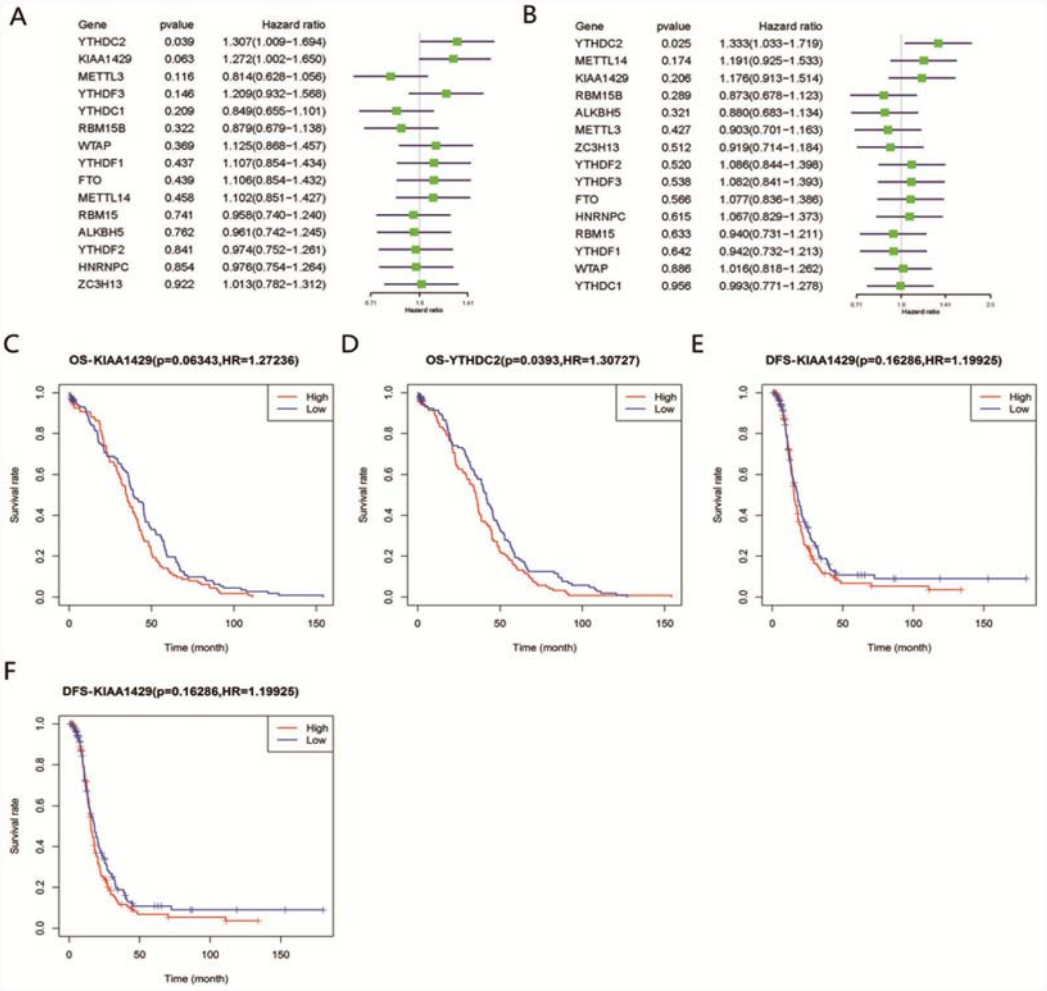

Figure 5: Overall survival of EOC patients with mRNA expression of m6A regulatory genes. A,B) OS and DFS of EOC patients with mRNA expression of m6A regulatory genes. C-F) Kaplan–Meier overall survival and disease-free survival curves of EOC patients with assigned to high- and low-expression of KIAA1429 and *YTHDC2* mRNA expression.

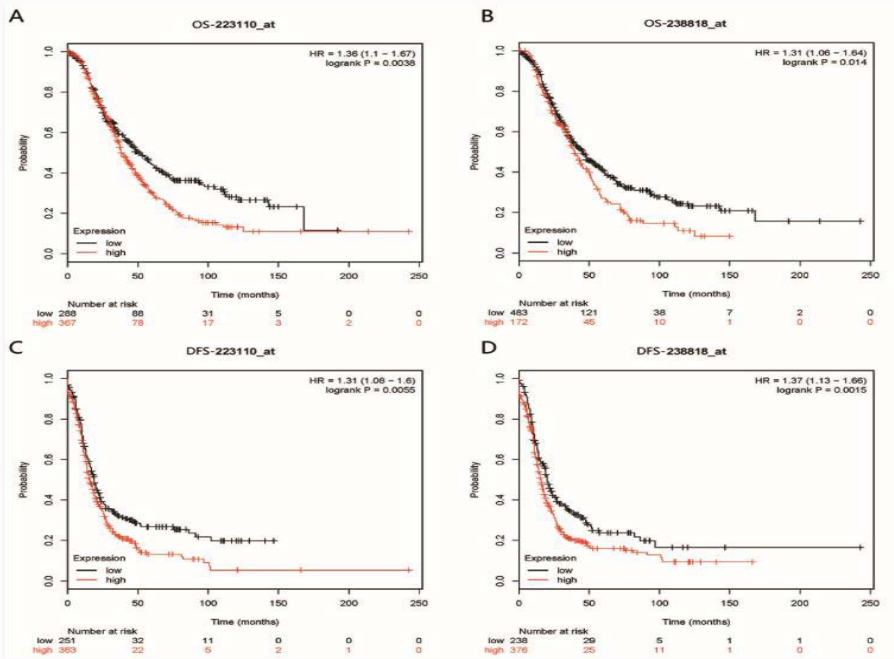

Figure 6: Validation the prognostic role of KIAA1429 expression in epithelial ovarian cancer. A,C) The OS and DFS survival curves of KIAA1429 expression in ovarian cancer with probe 223110_at. B,D) The OS and DFS survival curves of KIAA1429 expression in ovarian cancer with probe 238818_at.

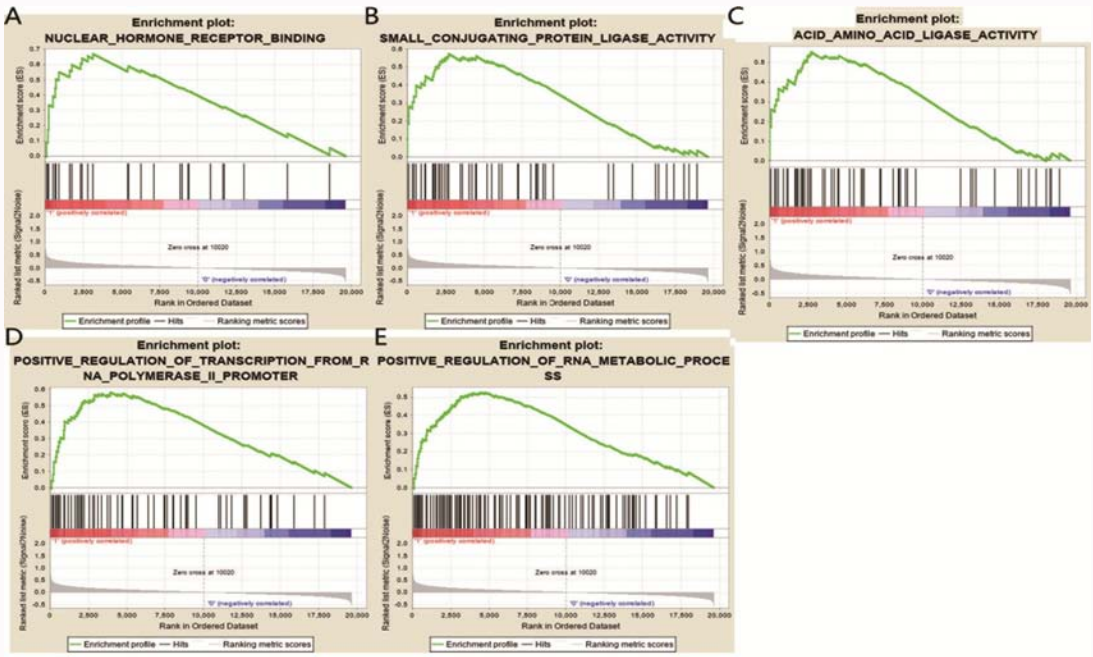

Figure 7: GSEA results with subgroups of KIAA1429 expression level in quartile. Gene set enrichment plots of A) nuclear hormone receptor binding, B) small conjugating protein ligase activity, C) acid amino acid ligase activity, D) positive regulation of transcription from RNA polymerase II promoter, and E) positive regulation of RNA metabolic process related to high KIAA1429 mRNA expression level in the EOC patients.

acid amino acid ligase activity, positive regulation of transcription from RNA polymerase II promoter, and positive regulation of RNA metabolic process were related with *KIAA1429* upregulation in EOC patients, as Table 4 and Figure 7A-7E shown. However, the further mechanism and regulation are needed to be illustrated.

Discussion

N6-methyladenosine (m6A) plays important roles in cancer progression and metastasis by controlling cell differentiation and cell pluripotency especially cancer stem cells [19]. It has been shown that m6A modification affect the breast cancer stem cells enrichment through changing the *NANOG* mRNA stability, relating with a shorter survival of breast cancer cases [20,21]. In human lung cancer, *METTL3* can promote oncogene translation by decreasing Epidermal Growth Factor Receptor (EGFR) protein expression [22]. Besides, m6A regulatory genes also show an critical oncogenic role in acute myeloid leukemia [23], glioblastoma [8,24], and hepatocellular carcinoma [20].

Especially, Xiaoyao et al. [25] revealed that m6A modification of mRNAs regulates Epithelial-Mesenchymal Transition (EMT) which is an important process for cancer cell metastasis. Upregulation of *METTL3*, one of the writers in m6A regulatory genes, was proved to be shown in ovarian carcinoma, and significantly related with some prognostic clinicopathological factors including grade, lymph-node metastasis, and stage. Interestingly, *METTL3* can promote EMT by upregulating the receptor tyrosine kinase AXL to promote ovarian cancer progression [13]. Moreover, other m6A modification genes, such as m6A demethylases *FTO* and *ALKBH5*, can regulate the Wnt/β-catenin pathway, contributing to PARP inhibitor resistance in BRCA-mutated EOC cells [26]. The published data showed that m6A regulation play an important role in ovarian cancer progression. In this article, we integratively analyzed the CNVs and mRNA expression data from m6A regulator genes in TCGA dataset. We explored the prognostic value of distinct writers, readers, and erasers, the association of copy number alteration and prognosis, and the specific gene regulator for the ovarian cancer prognosis. Surprisely, we found that *KIAA1429*, *METTL14* and *YTHDC2* were associated with cliniopathological features and clinical outcomes. Firstly, we addressed that amplification of *KIAA1429* which showed highest among the thirteen genes' amplification, and *METTL14* had correlation with clinicopathological grade and poor prognosis, which is consistent with the data shown in previous published studies. Besides, we elucidated the *KIAA1429* mRNA expression and its potential clinical significance in epithelial ovarian cancer. The results depicted that *KIAA1429* upregulation was related with poor prognosis, which is in accordance with the relationship between CNV and clinical prognosis. This was proved in breast cancer that *KIAA1429* was highly expressed in breast cancer tissues and down-regulated in non-cancerous breast tissues. The prognosis, especially overall survival of breast cancer was associated with *KIAA1429* expression. *METTL14*, one of the writers in m6A regulatory genes and one important component of the methyltransferase complex, plays a critical oncogenic role in human AMLs and liver cancer by positioning RNA substrates for methylation medicated by a complex comprising *RBM15-WTAP-METTL3-METTL14* [8,18,27-29]. Further- more, our findings also found that amplification of *YTHDC2*, as an m6A modification reader, was correlated with poor clinical outcome in ovarian cancer, which is shown in studies that *YTHDC2* promotes cancer metastasis [30].

We identified the mechanism of *KIAA1429* mRNA expression on clinical prognosis. Finally, 19,624 genes were enrolled into the GSEA process. We addressed that positive regulation of transcription from RNA polymerase II promoter and positive regulation of RNA metabolic process were critical pathways, while articles showed that N6-methyladenosine promote cancer progression through regulating RNA metabolism [19]. As shown by the subgroups analysis, writers

gain plus eraser loss showed association with poor prognosis in EOC. As methyltransferase enzymes, writer genes play an important role and the most part in m6A regulation process. The results implied that the regulation of m6A level might be associated with writers' expression and function.

Our articles showed high *ALKBH5* loss in EOC but lower alteration including deep deletion and amplification. It is ambiguous about the role of *ALKBH5* or the m6A regulatory genes loss in ovarian cancer. *ALKBH5* can induce the stem cell phenotype in breast by mediating the m6A-demethylation of *NANOG* mRNA [21], and further studies proved that *ALKBH5* regulated the malignant behavior of glioblastoma [31]. In ovarian cancer, *ALKBH5* may be a potential oncogene in EOC by inhibiting autophagy of epithelial ovarian cancer through regulating *miR-7* and *BCL2* [14]. Our article showed no relationship between *ALKBH5* CNVs or mRNA expression level with prognostic value including overall survival and prognostic free survival, which is addressed with TCGA database and validated with K-M plotter database. The reason for the inconsistent may be for the limited clinical samples and cell lines which can be solved. In the current study, we used a vast spectrum of tumour samples from the publicity online studies and datasets. This indicated that our conclusions are general. But there were still some shortcomings in this article. We used the CNVs and expression datasets from TCGA dataset in which the sample size may be limited. Then we used GEO as the validation set, which includes a large sample size to improve repeatability. This approach may increase statistical power, but limitation exists insofar as some patients' information will be lost and data integrity will be impaired. Also, GEO samples have the possibility that patients are likely to have differences in clinical characteristics, and treatment. These differences affected the association between the m6A regulator genes and clinical outcome, with the possibility that we increased the false negative ratio. In summary, this study addressed the critical role of m6A upregulation, especially writers' gain or amplification or mRNA upregulation, in clinical outcome of ovarian cancer, providing the prognostic role and possible targets for tumor diagnosis and prognosis. Analysis with large-scale genomic data from databases can reveal the role of m6A methylation regulator genes that is an important part of RNA stability, methylation and functions.

Availability of Data and Material

The datasets generated and analyzed during the current study are available from TCGA, GEO, and Kaplan-Meier plotter that provide free online tools and resources.

Authors' Contributions

CT and JL searched the database and analysis data. CT and SX wrote the main manuscript text and prepared the table. SH critically reviewed the manuscript. All authors read and approved the final manuscript.

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