

Original Research

TCOF1 is Identified to be an Unfavorable Biomarker and is Associated with Molecular Classification in Endometrial Cancer

Caiqiu Xu^{1,†}, Qinghai Wang^{1,†}, Yilin Zhang¹, Qirun Guo¹, Huiping Jiang^{1,*}, Suiqun Guo^{1,*}

¹Department of Obstetrics and Gynecology, The Third Affiliated Hospital of Southern Medical University, 510630 Guangzhou, Guangdong, China

*Correspondence: guosq2005@126.com (Suiqun Guo); Jianghp2022@163.com (Huiping Jiang)

†These authors contributed equally.

Academic Editor: Michael H. Dahan

Submitted: 9 February 2022 Revised: 9 March 2022 Accepted: 10 March 2022 Published: 11 May 2022

Abstract

Background: Endometrial cancer (EC) is one of the most common gynecologic cancers of the female reproductive system. Its incidence and mortality are currently increasing. Patients with early-stage EC have a much better prognosis than those with late-stage EC. Therefore, early detection, diagnosis, and treatment are critical to improving the outcome for EC patients. The proposition of molecular classification promotes the individualization for diagnosis and treatment of EC. *TCOF1* has been identified as an oncogenic gene in several tumors but has been seldom studied in EC. **Methods:** TCGA and immunohistochemistry (IHC) experiments were performed to verify the protein level of *TCOF1* expressed in endometrial cancer while its prognostic ability in EC patients was assessed by the TCGA database. Linked Omics database, Web Gestalt gene enrichment, and string database were applied to analyze the possible biological functions of *TCOF1* in EC. Mutation types of *TCOF1* in EC and its mutation frequency were explored in c-BIOPORTAL. The Relationship between molecules was detected by utilizing the GEPIA database. **Results:** *TCOF1* is up-regulated in endometrial cancer compared to para cancer and it was positively correlated with poor prognosis of patients. *TCOF1* is mutated in endometrial cancer and is closely associated with microsatellite instability (MSI), this being one type of molecular classification in EC. **Conclusions:** *TCOF1* may function as a potential biomarker and is associated with molecular classification in endometrial cancer.

Keywords: *TCOF1*; endometrial cancer; molecular classification

1. Introduction

Endometrial cancer (EC) is the leading malignancy of the female reproductive system in developed countries. There were 417,000 new cases of endometrial cancer and 97,000 deaths reported in 2020 [1,2]. Hysterectomy and or bilateral salpingo-oophorectomy are the standard treatments for endometrial cancer. However, it is well documented that patients diagnosed at early-stage EC experienced a better prognosis after surgical treatment or combined radiotherapy than those diagnosed at a late stage [3]. It is generally accepted that causative factors of endometrial cancer include obesity, long-term stimulation of estrogen, diabetes mellitus, and delayed menopause [4,5]. However, the pathogenesis of endometrial cancer is still unclear. New biological markers for endometrial cancer are being sought. Molecular classification has been proposed by The Cancer Genome Atlas (TCGA) for the diagnosis of endometrial cancer, which classified endometrial cancer into 4 types, including *POLE* mutation, low copy number, high copy number, and microsatellite instability (MSI). The incorporation of TCGA into diagnostic guidelines suggests that it is of importance for individualized diagnosis and treatment of patients with endometrial cancer [6–9].

Lynch syndrome is a familial genetic disorder and is a well-known risk factor for endometrial cancer [10]. The probability of developing endometrial cancer in patients

with Lynch syndrome is comparable to that of colorectal cancer [11]. As cancer often occurs in Lynch syndrome, this subset makes up 2% of patients with endometrial cancer. Approximately 50% of patients with Lynch syndrome will develop endometrial cancer [12].

Mutations in DNA mismatch repair genes are an important factor in the pathogenesis of Lynch syndrome [10, 13]. Utilized in the detection for Lynch syndrome, assessment of DNA mismatch repair (MMR) proteins by IHC is a part of the diagnostic evaluation for patients with EC [14]. The primary function of MMR is to correct mispairing in individual nucleotides during DNA replication and mutations in MMR cause microsatellite instability. *MLH1*, *MSH2*, *MSH6*, and *PMS2* are the four most commonly mutated mismatch repair genes [15,16].

TCOF1 (Treacle Ribosome Biogenesis Factor 1) is localized on chromosome 5 (5q32-33.3) with 152 KD. It was initially identified as a gene closely associated with the pathogenesis of Treacher Collins syndrome [17]. Further findings demonstrated that *TCOF1* is involved in the development of multiple diseases and has important roles in cell proliferation, apoptosis, and DNA damage repair [18]. Upregulation of *TCOF1* promotes the growth and stemness formation in breast cancer [19]. *TCOF1* activates the *KRAS* gene and EMT signaling pathway, while a negative relevance exists between *TCOF1* and anti-tumor immune cell



infiltration in hepatocellular carcinoma [20]. As a potential oncogene in hepatocellular carcinoma, patients with higher expression of *TCOF1* may experience a worse prognosis. *TCOF1* encodes a cytoplasmic protein that is involved in the transcription of ribosomal DNA which is specifically enriched in telomeres, leading to defective telomere replication and genomic instability [21]. This has given rise to the study of *TCOF1* in endometrial cancer patients.

In this research, we evaluated the expression of *TCOF1* in patients with EC and explored its relationship with clinicopathologic characteristics by public databases and IHC. To detail the value of *TCOF1* in EC, we investigated the relationship between *TCOF1* and MMR (*MLH1*, *MSH2*, *MSH6*, and *PMS2*), which is related to microsatellite instability (MSI) [22], one kind of molecular classification of EC as *TCOF1* is involved in DNA damage repair [23]. To the best of our knowledge, MMR is a response to DNA damage [24].

2. Materials and Methods

2.1 UALCAN Database

The UALCAN (<http://ualcan.path.uab.edu/>) database, which can be used to analyze the differential expression of genes in normal tissues and tumor tissues and explore the relationship between genes and clinicopathologic parameters of patients, is an online database based on the TCGA database. We explored the expression of *TCOF1* in pan-cancer and analyzed the correlation between *TCOF1*, *NOLC1*, *MLH1*, *MSH2*, *MSH6*, and *PMS2* and their clinicopathologic parameters in patients with endometrial cancer. *NOLC1* plays an important role in nucleolar and rRNA synthesis and participates in ribosomal biogenesis [25]. Studies have demonstrated that it can promote tumor proliferation, invasion, and metastasis [25,26]. Also indicated is that *NOLC1* and *TCOF1* are mineralocorticoid receptor-related proteins and can be used as regulatory cofactors of *CKI* [27,28]. *MLH1*, *MSH2*, *MSH6*, and *PMS2* play an essential role in mismatch repair (MMR) [29].

2.2 TCGA Database

TCGA (The Cancer Genome Atlas program) was jointly launched by NCI (National Cancer Institute) and NHGRI (Human Genome Research Institute) in 2006 [30]. TCGA has tested 33 tumors from 20,000 patients, including 10 rare tumors, with a data volume of 2500t. Based on large-scale sequencing technology and through extensive cooperation, we can understand the molecular mechanism of cancer and finally get a complete set of “maps” related to all cancer genome changes, and improve scientific understanding of the molecular mechanism of cancer pathogenesis and improve the ability for diagnosis, treatment, and prevention. The database includes biospecimen, clinical information, sequencing reads, transcriptome profiling, simple nucleoside variation, copy number variation, and DNA methylation of patients. The *TCOF1* mRNA ex-

pression data in the endometrial cancer dataset were downloaded from the TCGA database, which includes 543 endometrial cancer tissue specimens and 35 normal endometrial tissue specimens (23 pairs of cancer and para cancer tissue samples).

2.3 GEPIA

GEPIA is a database providing functionalities according to datasets from TCGA and GTEx [31], which help analyze gene expression correlations. We analyzed the correlation between *TCOF1* and associated genes, including *ARRB1*, *NOLC1*, *SNK2A2*, *KBTBD8*, *ARRB2*, *MSH2*, *MSH6*, *PMS2*, and *MLH1* in GEPIA.

2.4 c-BioPortal

c-Bioportal (<http://www.cbioportal.org/>) database provides the frequency and type of gene mutations. We analyzed that of *TCOF1* in endometrial cancer patients.

2.5 String Database

String (<https://cn.string-db.org/>) is a database for analyzing protein interaction. Possible reciprocal proteins of *TCOF1* were analyzed and the top 5 are shown.

2.6 Linked Omics Database and Web Gestalt Gene Enrichment

Linkedomics database (<http://www.linkedomics.org/login.php>) is a multidimensional online data analysis platform [32], which mainly consists of three analytical modules. The LinkFinder module is used to analyze the gene profiles co-expressed with *TCOF1* in EC, and the LinkInterpreter module is used for GO annotation and KEGG enrichment analysis.

2.7 Immunohistochemical Assay (IHC)

The endometrial cancer tissue microarray was obtained from Shanghai Outdo Biotech Company. IHC test kit (PV-9000) was from Zsbio (Beijing, China) for *TCOF1* protein expression analysis. *TCOF1* was incubated at a concentration of 1:100 (PTG). IHC assay was performed as per our previous research [33]. The final IHC score was determined by two senior clinicopathologists, with a score of ≥ 6 being extremely high expression and < 6 being recorded as low expression.

2.8 Statistical Analysis

t-test or Wilcoxon rank-sum test was performed to analyze the differences between the two groups. Survival analysis was performed by Kaplan-Meier. The diagnostic efficacy of the index was evaluated by the operating characteristic curve (ROC). Spearman correlation analysis was used to assess the correlation between *TCOF1* and other genes in GEPIA database. Univariate and Cox regressions were used to examine relationships between single clinicopathologic parameters and clinical prognosis in patients

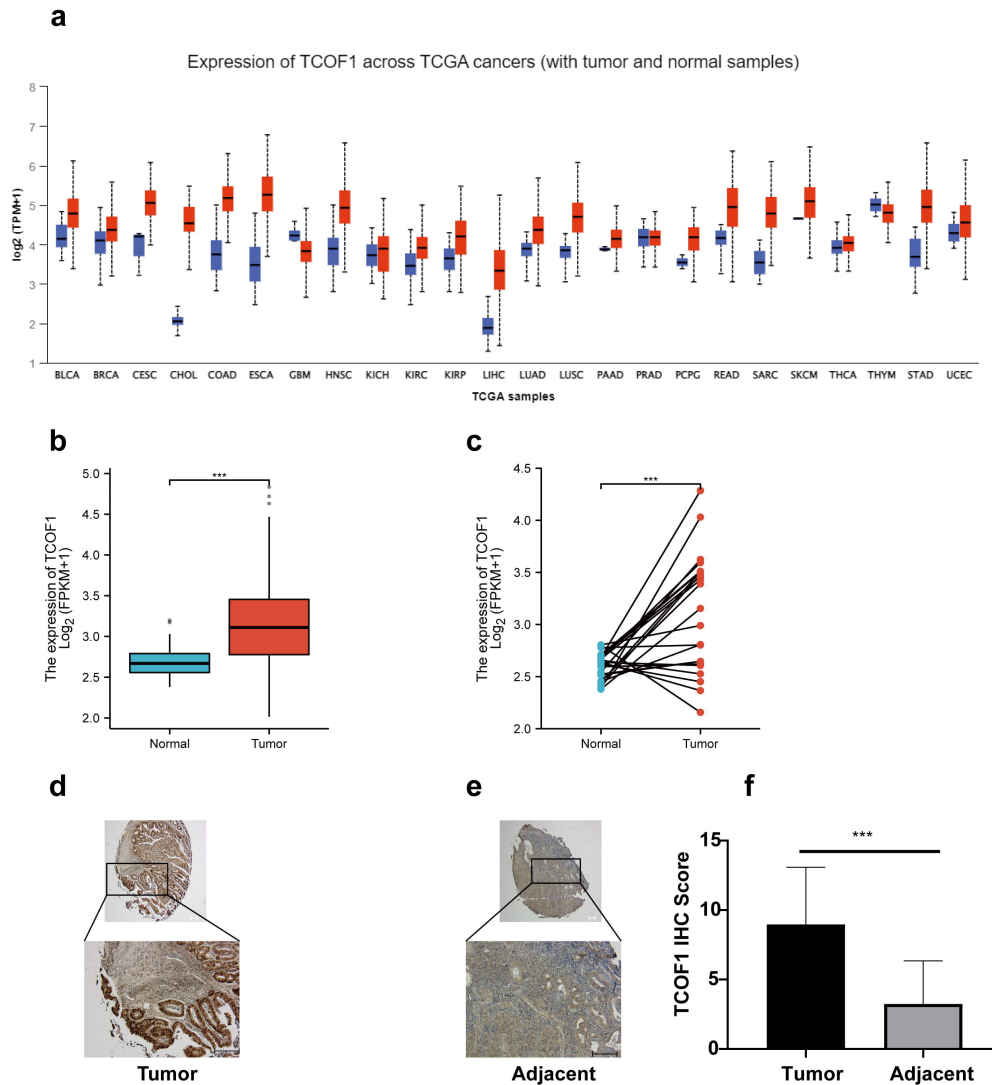


Fig. 1. Expression of *TCOF1*. (a) Expression of *TCOF1* in Pan-cancer in UALCAN. (b) Expression of *TCOF1* in endometrial carcinoma in TCGA database based on unpaired tissue. (c) Expression of *TCOF1* in endometrial carcinoma in TCGA database according to paired tissue. (d) The typical image of *TCOF1* in EC (scale bar: 50 μm). (e) The typical image of *TCOF1* in the Adjacent tissue of EC (scale bar: 50 μm). (f) Statistical analysis of the expression of *TCOF1* between endometrial cancer and adjacent tissue. $***p < 0.001$. BLCA, Bladder urothelial carcinoma; BRCA, acronym for Breast invasive cancer; CESC, acronym for cervical squamous cell carcinoma and endocervical adenocarcinoma; CHOL, acronym for Cholangiocarcinoma; COAD, acronym for Colon adenocarcinoma; ESCA, Esophageal carcinoma; GBM, acronym for glioblastoma multiforme; HNSC, acronym for Head and Neck squamous cell carcinoma; KICH, Kidney Chromophobe; KIRC, acronym for Kidney renal clear cell carcinoma; KIRP, Kidney renal papillary cell carcinoma; LIHC, Liver hepatocellular carcinoma; LUAD, Lung adenocarcinoma; LUSC, acronym for Lung squamous cell carcinoma; PAAD, Pancreatic adenocarcinoma; PRAD, Prostate adenocarcinoma; PCPG, Pheochromocytoma and Paraganglioma; READ, Rectal adenocarcinoma; SARC, Sarcoma; SKCM, Skin Cutaneous Melanoma; THCA, acronym for Thyroid carcinoma; THYM, Thymoma; STAD, Stomach adenocarcinoma; UCEC, Uterine Corpus Endometrial Carcinoma.

with EC. $p < 0.05$ was considered a statistically significant difference.

3. Results

3.1 Expression of *TCOF1* in EC

First, we explored the expression of *TCOF1* in pan-cancer in the UALCAN database which revealed that

TCOF1 is up-regulated in most tumors compared with normal tissues (Fig. 1a). Thus, we downloaded the data for endometrial cancer from the TCGA database, which contained 543 cancer tissues and 35 normal tissues, including 23 pairs of carcinoma and its para cancer tissues. This demonstrated that expression of *TCOF1* mRNA was significantly higher in EC tissues than that in normal groups ($p <$

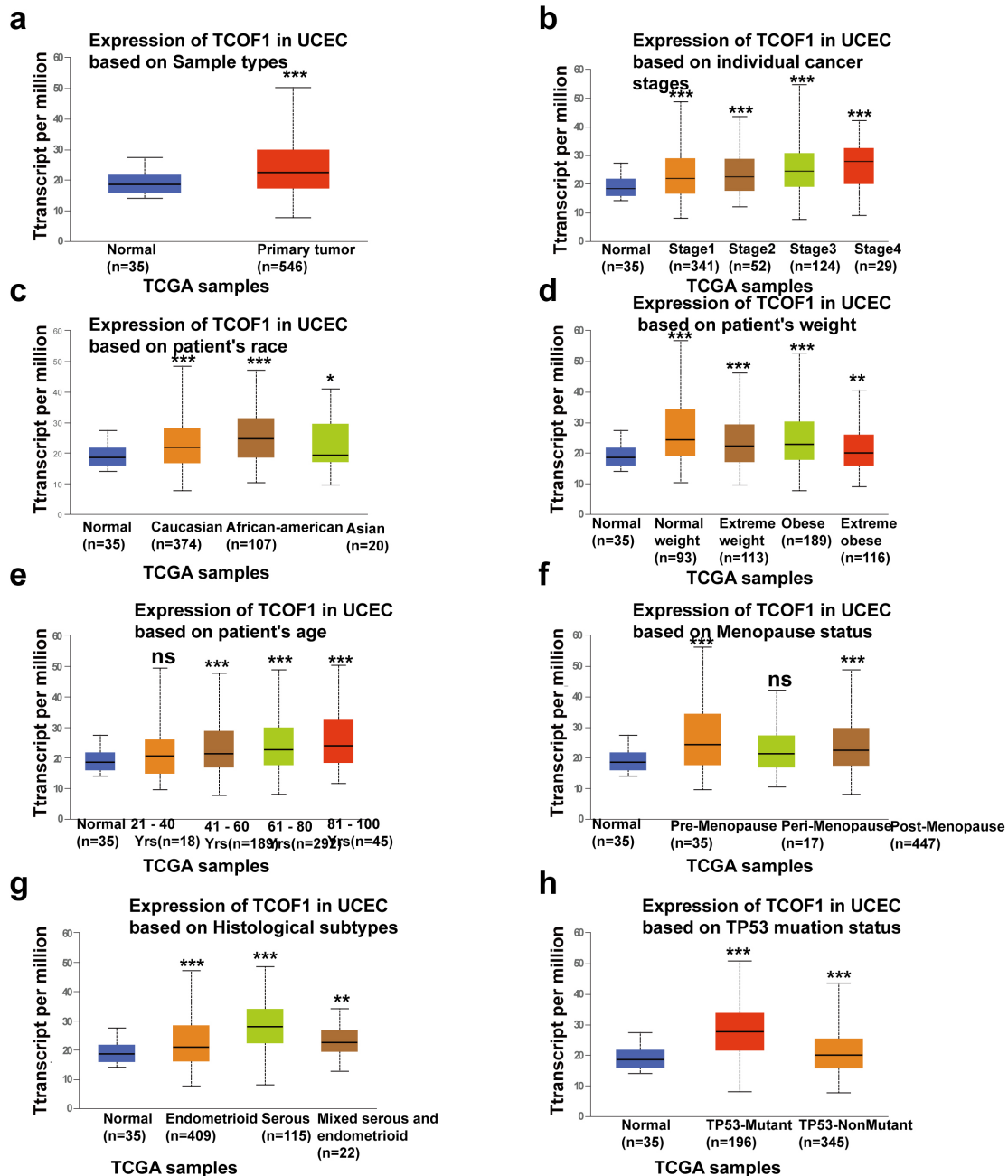


Fig. 2. Relationship between *TCOF1* mRNA expression and clinical features of EC. Analysis is shown in UALCAN database as sample types (a), cancer stages (b), race (c), weight (d) and patient's age (e), menopause status (f), historical subtypes (g), and TP53 mutation status (h) respectively. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, ns, non sense, $p > 0.05$.

0.001) (Fig. 1b) with similar results being obtained in paired samples (Fig. 1c). Further, we performed independent experiments with immunohistochemical assays, which indicated that *TCOF1* was primarily localized in the nucleus. Positive staining for *TCOF1* protein was brownish-yellow granules with the expression of *TCOF1* protein being dramatically higher in EC tissues compared with normal samples ($p < 0.001$) (Fig. 1d–f).

3.2 Relationship Between Expression of *TCOF1* mRNA and Clinicopathological Parameters of Patients with EC

Further subgroup analysis of multiple clinicopathologic characteristics in 546 cases of endometrial cancer and 35 cases of non-cancer via the online UALCAN database was performed. This suggested that *TCOF1* was highly expressed in subgroups, including tumor stage, patient race, patient weight, patient age, menstrual status, historical subtypes, and *P53* mutation status than that of normal tissue (Fig. 2). Considering *TCOF1* may act as a poten-

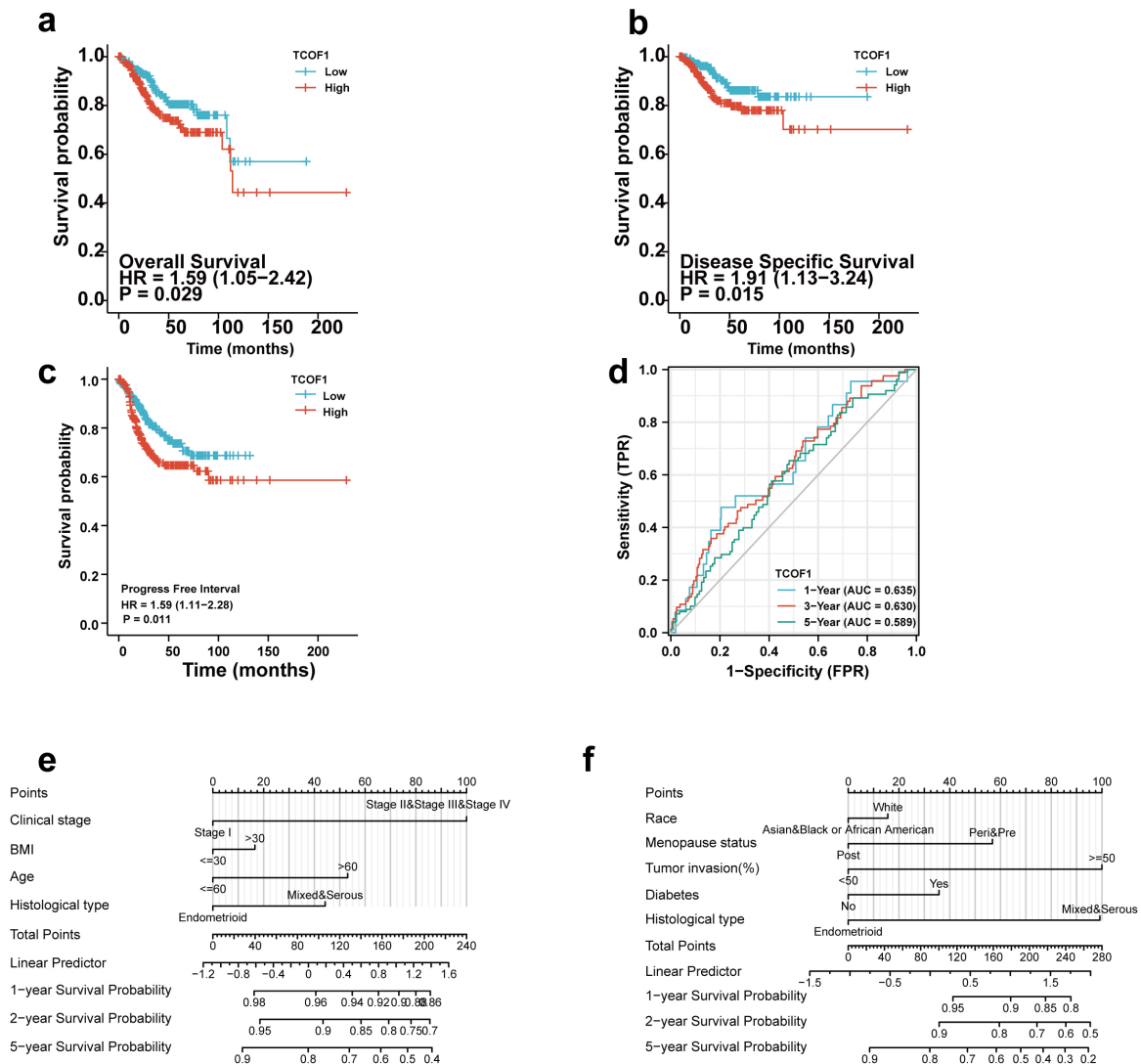


Fig. 3. Survival analysis for patients with endometrial carcinoma depending on *TCOF1* mRNA level. TCGA cohorts are shown for OS (a), DSS (b), and PFI (c). ROC curve of *TCOF1* gene expression in patients with EC (d). Univariate Cox regression results based on TCGA database (e, f).

tial biomarker for the diagnosis of endometrial cancer, we explored the relationship between *TCOF1* and the prognosis of patients with EC, which demonstrated that patients had worse overall survival, disease-free survival, and progression-free survival with higher expression of *TCOF1* (Fig. 3a–c). As shown, AUC at 1-year, 3-year as well as 5-year is larger than 0.5. (Fig. 3d). Univariate Cox regression indicated that *TCOF1* was associated with Age ($p < 0.001$), BMI ($p < 0.001$) and Clinical stage ($p < 0.001$), tumor invasion ($p < 0.001$), and histological type ($p < 0.001$).

3.3 Protein-Protein Interaction Networks of *TCOF1*

Protein-Protein Interaction Networks (PPI) are defined as protein-protein interaction to participate in biological processes such as cell energy metabolism, cell cycle regulation, and intracellular signal pathway [34]. The co-expressed genes of *TCOF1* were enriched and analyzed by

LinkOmics. It showed that numerous genes were closely associated with *TCOF1* (Fig. 4a). Graphs showed the top 50 significant genes which are associated with *TCOF1* (Fig. 4b,c). String database is a powerful database for protein interactions. The top 5 interactants of *TCOF1*, which includes *CSNK2A2*, *NOLC1*, *KBDBC8*, *ARRB1*, *ARRB2* are shown in Fig. 5a. We examined the correlation between *TCOF1* and *CSNK2A2*, *NOLC1*, *KBDBC8*, *ARRB1*, *ARRB2* expression in endometrial cancer in the GEPIA database. *TCOF1* was positively correlated with *CSNK2A2*, *NOLC1*, *KBDBC8*, *ARRB2* expression (Fig. 5c–f), but not *ARRB1* (Fig. 5b). It was obvious that *TCOF1* was more closely aligned with *NOLC1*, and the study suggested that *TCOF1* and *NOLC1* were paralogous homologs [27]. Therefore, we investigated the clinicopathologic parameters according to the expression of *NOCLI* in endometrial cancer via UALCAN database. Interestingly, *NOLC1* was closely associ-

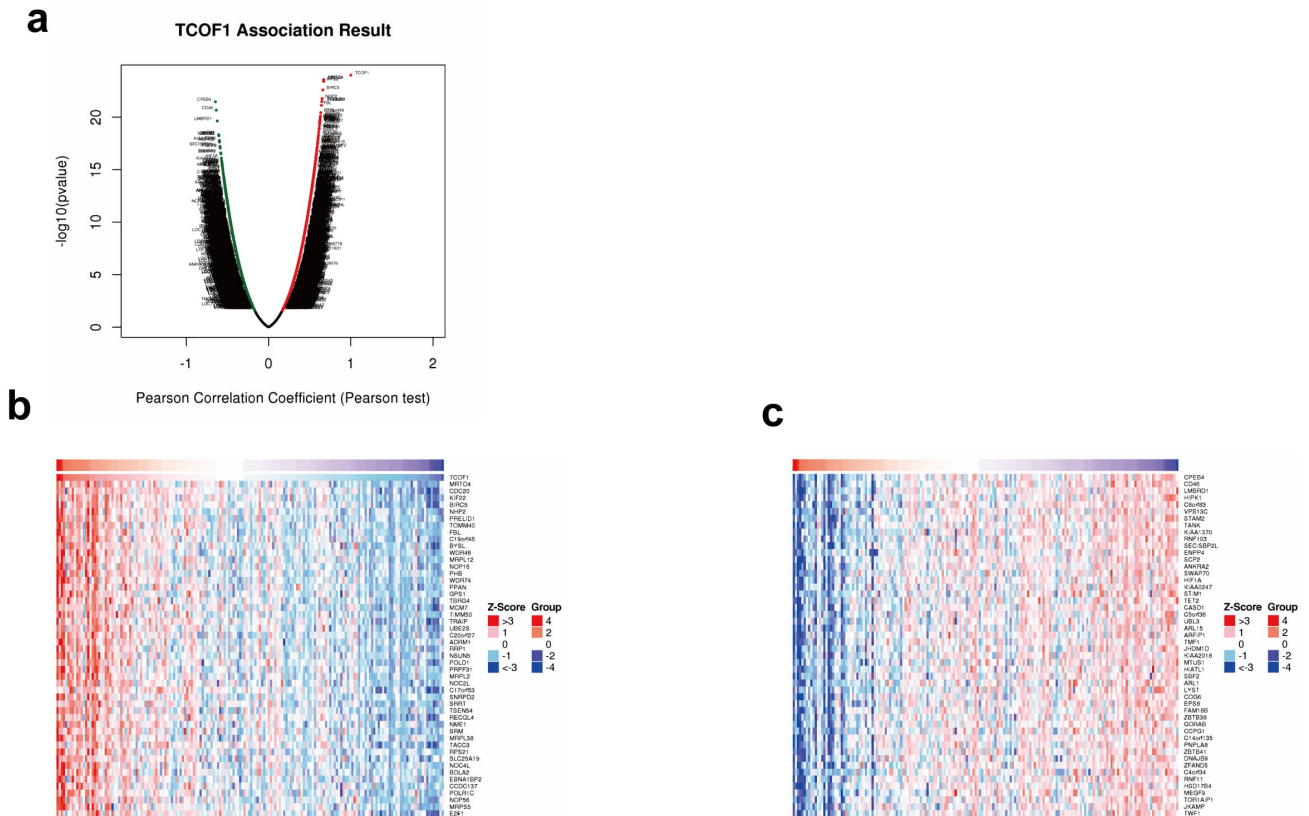


Fig. 4. Co-expression genes of TCOF1. *TCOF1* gene co-expression gene volcano map (a) in LinkOmics. Heat map of top 50 most significantly positively (b) and negatively (c) associated genes co-expressed with *TCOF1*.

ated with tumor stage, patient race, patient weight, patient age, menstrual status, tissue classification, and *P53* mutation status in endometrial cancer patients, which is similar to that of *TCOF1* (Fig. 6).

3.4 Enrichment Analysis of *TCOF1* and its Co-Expressed Genes

To clarify the possible molecular mechanism involved in the process that *TCOF1* promotes endometrial cancer progression, enrichment analysis was applied to determine the co-expressed genes of *TCOF1*. Gene ontology (GO) enrichment analysis was divided into three sections, which contained biological processes, cellular components as well as molecular functions. Genes co-expressed with *TCOF1* were chiefly involved in biological regulation, the process of metabolism, and reaction to stimulus. Cellular component analysis revealed that the genes co-expressed with *TCOF1* were mainly located in the membrane, nucleus, and membrane-enclosed lumen (Fig. 7).

3.5 Mutation in *TCOF1* and its Relationship with Typical Molecular of MMR

Given that mutations in *TCOF1* are an important cause of Treacher Collins Syndrome [35], to obtain knowledge of the mutation of *TCOF1* in EC, c-BioPortal database was searched to investigate the mutation of *TCOF1*. Results

showed that among 242 patients with endometrial cancer, 8 patients had *TCOF1* mutations, including 7 missense mutations and 1 truncating mutation (Fig. 8a,b). Being that *TCOF1* participates in the process of DNA damage repair with DNA mismatch repair is a measure to deal with DNA damage repair [36], and that deletion of DNA MMR causes the gathering of mismatches within the cycle of DNA replication, resulting in the development of microsatellite instability (MSI) [37]. *MLH1*, *MSH2*, *MSH6*, and *PMS2* are the typical genes of DNA mismatch repair. We explored the relationship between *TCOF1* and the genes of DNA mismatch repair in the GEPIA database. The results showed that *MLH1* ($R = 0.12$, $p < 0.001$), *MSH2* ($R = 0.42$, $p < 0.001$), *MSH6* ($R = 0.36$, $p < 0.001$), and *PMS2* ($R = 0.35$, $p < 0.001$) (Fig. 8c–f) were significantly and positively correlated with *TCOF1*. Further, we explored the correlation between *MSH2*, *MSH6*, and *PMS2* with clinicopathologic parameters via the UALCAN database in endometrial carcinoma., which indicated that they were strongly correlated with tumor stage, patient race, patient weight, patient age, menstrual status, tissue staging, and *P53* mutation status in patients with endometrial cancer (Supplementary Figs. 1,2,3).

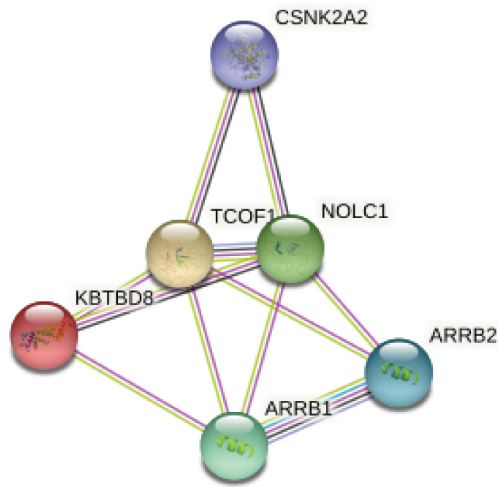
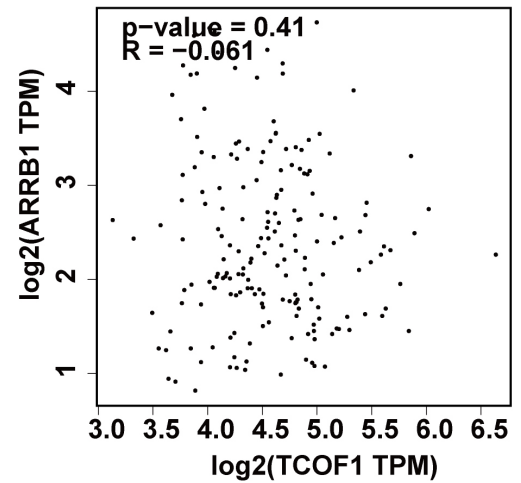
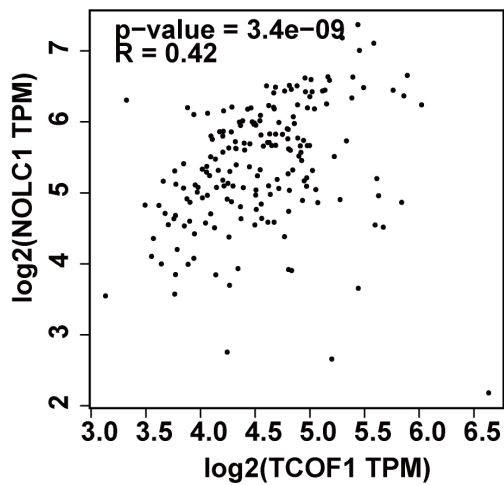
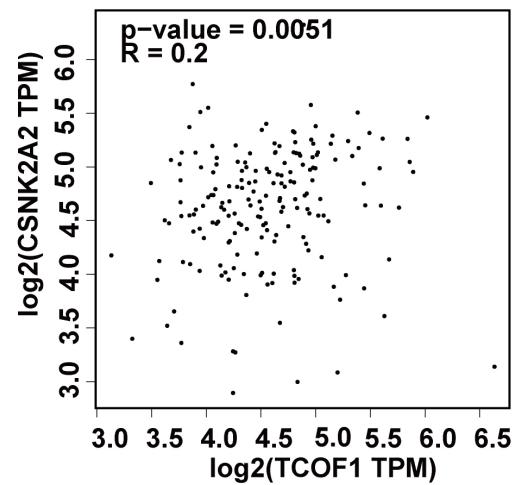
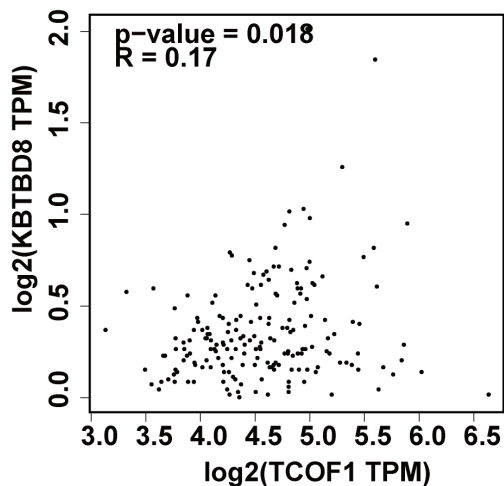
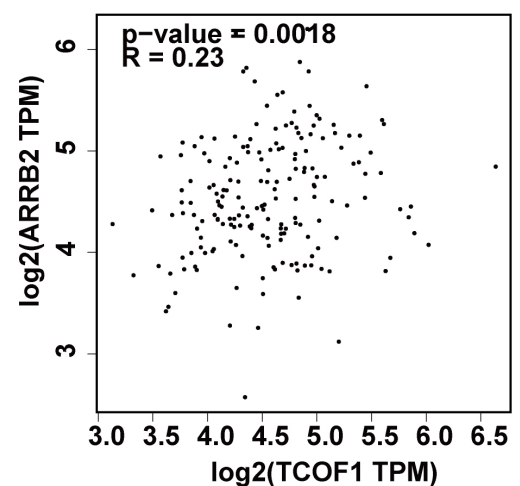
a**b****c****d****e****f**

Fig. 5. Protein-Protein Interaction Networks of *TCOF1*. The top 5 interactants of *TCOF1* in String database (a). Scatter diagram evaluating co-relationship between *TCOF1* and *ARRB1* (b), *NOLC1* (c), *SNK2A2* (d), *KBTBD8* (e), *ARRB2* (f) in GEPIA database.

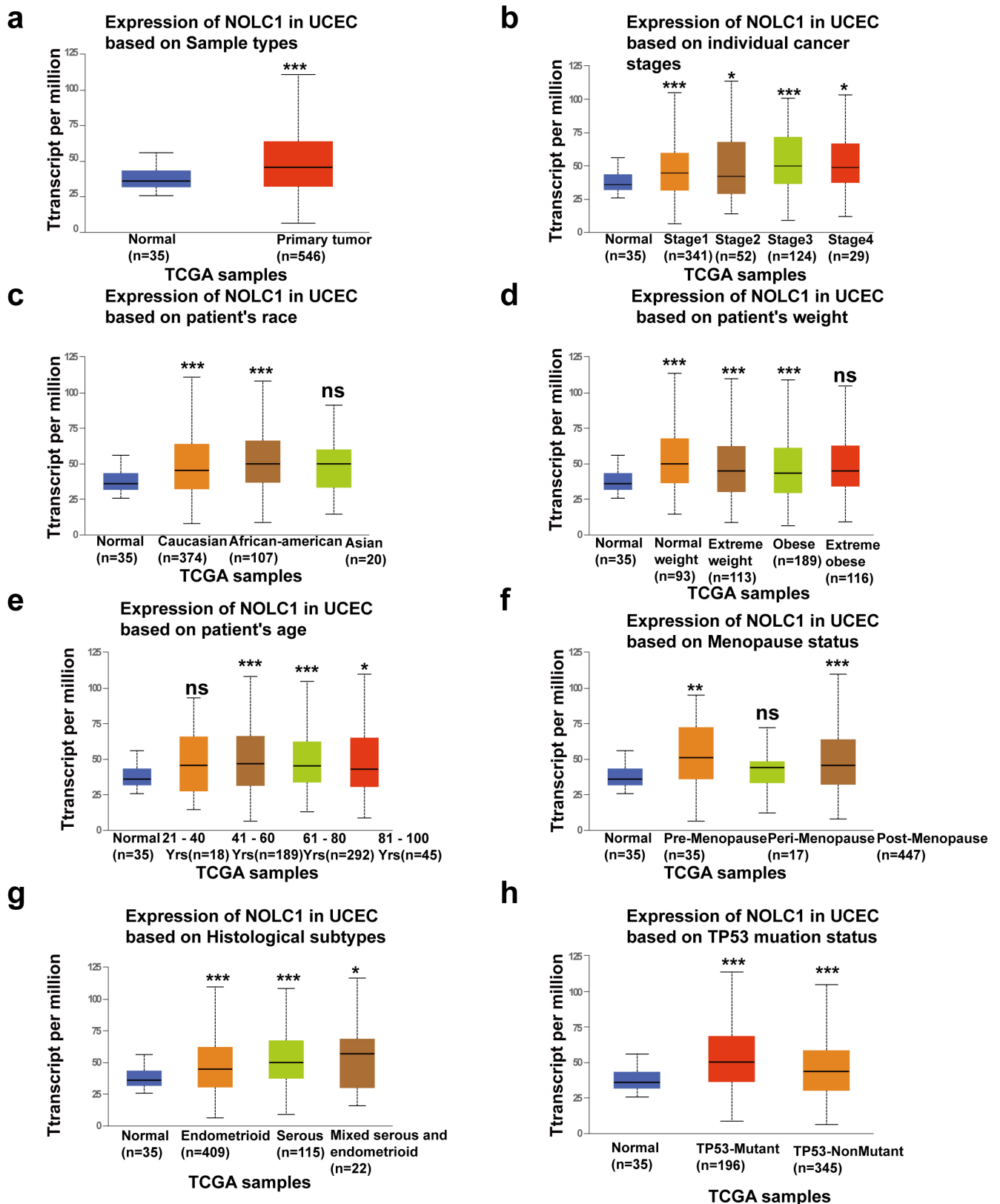


Fig. 6. Box diagram shows the expression of *NOLC1* among different subgroups. Sample types (a), individual cancer stages (b), race (c), weight (d), age (e), menopause status (f) historical subtypes (g) and *TP53* mutation (h). respectively. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$. ns, non sense $p > 0.05$.

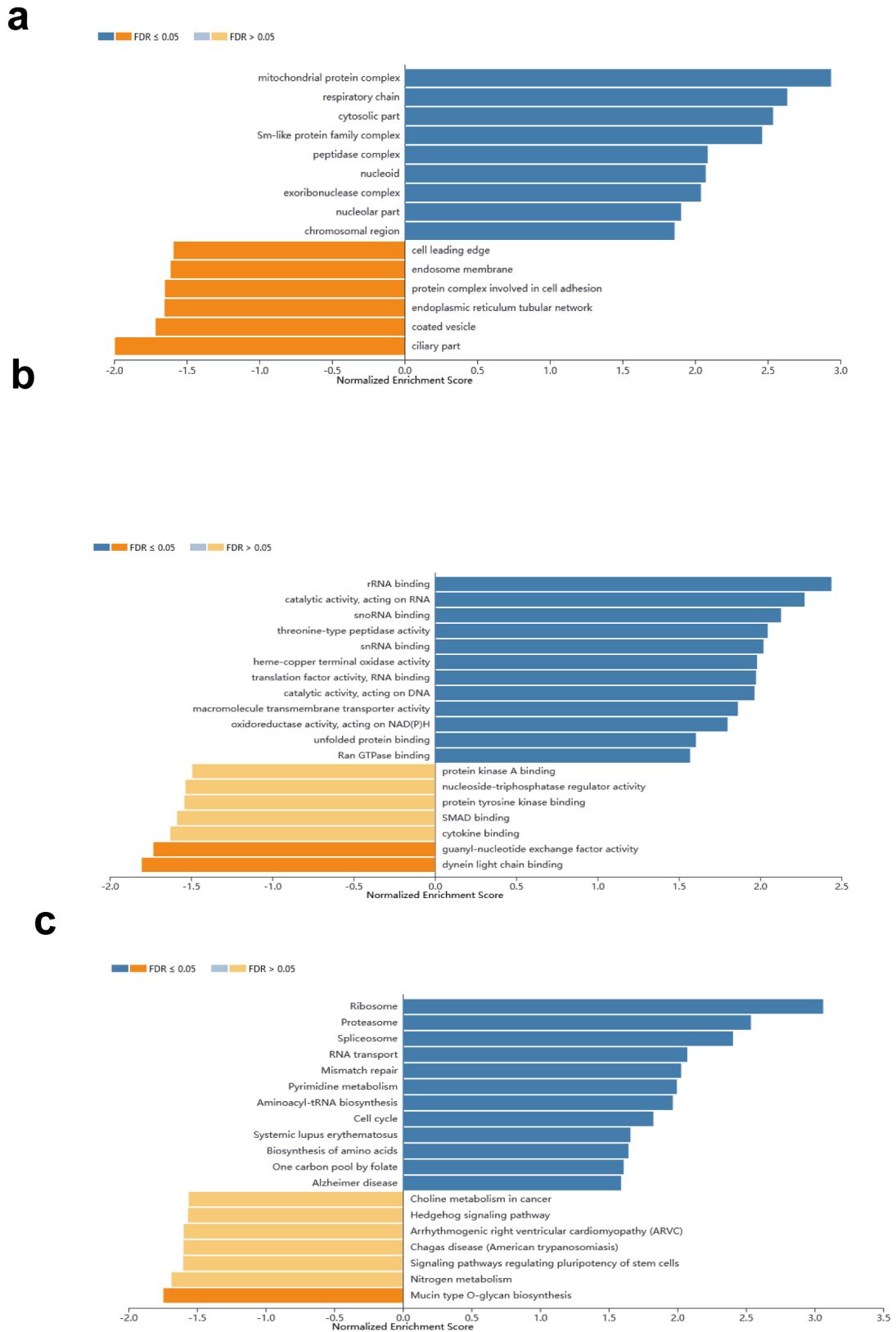


Fig. 7. Functional enrichment analysis of *TCOF1* co-expressed genes by gene ontology (GO) enrichment. (a) Biological process. (b) Cellular component. (c) Molecular function.

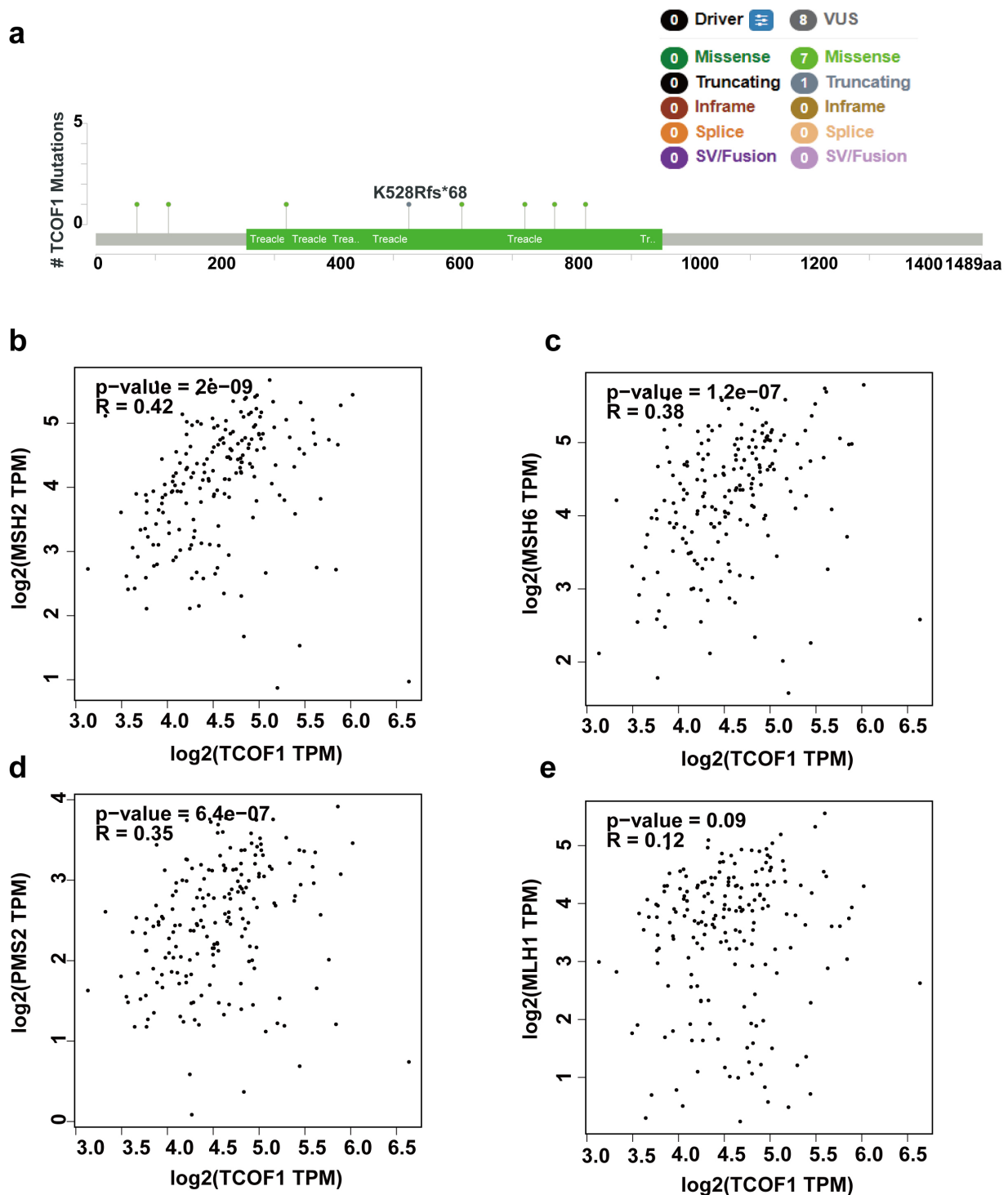


Fig. 8. Mutation of *TCOF1* and its relationship with typical molecular of MMR. *TCOF1* specific mutation site in the c-BioPortal database (a). Scatter diagram evaluating correlation ship between *TCOF1* and *MSH2* (b), *MSH6* (c), *PMS2* (d), *MLH1* (e) in GEPIA database.

4. Discussion

Endometrial cancer was classified into 4 types according to histological types, including endometrioid adenocarcinoma, plasmacytoma, clear cell-like adenocarcinoma of the uterus, and mixed type [38]. The risk factors that pre-

dispose to endometrial cancer and the clinical outcomes of endometrial cancer patients are also diverse [4], which suggests that endometrial cancer is a heterogeneous disease that requires more individualized diagnosis and treatment. The study clearly demonstrated that patients with early-stage endometrial cancer had a better prognosis than those with ad-

vanced recurrent metastatic disease after aggressive surgical treatment [3]. Therefore, it is extremely important to develop new therapeutic regimens and screen new biological markers to facilitate early diagnosis and personalized treatment for patients. A large set of new biomarkers was found with the advent of bioinformatics, which opened an era of big data screening for biomarkers in the field of oncology. *TCOF1* gene mutation is closely associated with the pathogenesis of Treacher Collins syndrome [39], which is involved in cell proliferation, apoptosis, and DNA damage repair processes [18].

In this study, using the TCGA database, we found that TCGA mRNA showed high expression in endometrial cancer and that high expression of *TCOF1* indicated worse OS, DFS, and DFP. To further validate this result, we performed an IHC assay to confirm the protein level of *TCOF1* in endometrial cancer tissue. Results were consistent with the TCGA database. Subgroup analysis showed that the expression of *TCOF1* in various clinicopathologic parameters was higher than that in the normal group. This suggests that *TCOF1* may act as a potential diagnostic indicator for endometrial cancer. To explore the mechanism of *TCOF1* involvement in endometrial carcinogenesis, gene co-expression was used to analyze the possible enrichment of *TCOF1*. PPI analysis revealed that *TCOF1* may interact with *NOLC1* which was reported to be a paralog of *TCOF1*, but it has not been studied in endometrial cancer. Thus we evaluated the relationship between *NOLC1* and the clinical prognosis of patients with endometrial carcinoma with results being similar to *TCOF1*. It was revealed that the mutation of *TCOF1* plays an important role in the diagnosis of Treacher Collins syndrome [17]. Thus, we evaluated this mutation in endometrial cancer. Considering that *TCOF1* improves resistance to DNA damage response [23] and MMR belongs to the styles of DNA damage repair, we discussed the co-expression relationship with *TCOF1* and *MLH1*, *MSH2*, *MSH6*, and *PMS2*, which are the typical genes in MMR. It was revealed that *TCOF1* is positively associated with all of the MMR listed above. More importantly, the expression of *MSH2*, *MSH6*, and *PMS2* was closely related to clinicopathologic parameters in endometrial cancer patients. Interestingly, microsatellite instability (MSI), which is one type of molecular classification of EC, is a signature feature of MMR [40].

We speculate that *TCOF1* is of great significance in the progression of endometrial cancer through DNA damage repair mechanism and it may serve as a potential diagnostic criterion for microsatellite instability (MSI) endometrial cancer.

In conclusion, this study revealed the expression of *TCOF1* in endometrial carcinoma. It is significantly up-regulated compared with normal tissues, which can be considered as a potential biomarker for the diagnosis and prognosis of endometrial cancer. In addition, through the analysis of the mutation and molecular expression correlation of

TCOF1, it was found that *TCOF1* is closely related to the typical molecules of MMR, one of the molecular classifications in EC. However, more in-depth cell experiments and clinical trials are needed to verify the value of *TCOF1* in endometrial cancer.

5. Conclusions

TCOF1 may function as a potential biomarker in endometrial cancer and may be related to microsatellite instability (MSI), one of the molecular classifications in endometrial cancer.

Author Contributions

SQG and HPJ design the study and were responsible for the conception of the present study. CQX and QHW analyzed the data and wrote the manuscript. CQX and QHW contribute the same efforts to this research. YLZ and QRG performed IHC assay and statistical analysis. All authors have read and approved the final manuscript.

Ethics Approval and Consent to Participate

All subjects gave their informed consent for inclusion before they participated in the study. The study was conducted in accordance with the Declaration of Helsinki, and the protocol was approved by the Ethics Committee of Shanghai Outdo Biotech Company (approval number: No.YB M-05-02).

Acknowledgment

Thanks to the team that established the public database, it provided free and convenient conditions for experimental exploration.

Funding

The present study was funded by The Nature Science Fund of Guangdong Province (grant no. 2020A15150).

Conflict of Interest

The authors declare no conflict of interest.

Supplementary Material

Supplementary material associated with this article can be found, in the online version, at <https://doi.org/10.31083/j.ceog4905112>.

References

- [1] Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A. Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA: A Cancer Journal for Clinicians*. 2018; 68: 394–424.
- [2] Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A, *et al.* Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. *CA: A Cancer Journal for Clinicians*. 2021; 71: 209–249.

- [3] Nagase S, Ohta T, Takahashi F, Enomoto T. Annual report of the committee on gynecologic oncology, the Japan Society of Obstetrics and Gynecology: Annual patients report for 2015 and annual treatment report for 2010. *The Journal of Obstetrics and Gynaecology Research*. 2019; 45: 289–298.
- [4] Derbyshire AE, MacKintosh ML, Pritchard CM, Pontula A, Ammori BJ, Syed AA, *et al*. Women's Risk Perceptions and Willingness to Engage in Risk-Reducing Interventions for the Prevention of Obesity-Related Endometrial Cancer. *International Journal of Women's Health*. 2022; 14: 57–66.
- [5] Dalmartello M, Vermunt J, Negri E, Levi F, La Vecchia C. Adult lifetime body mass index trajectories and endometrial cancer risk. *BJOG: An International Journal of Obstetrics & Gynaecology*. 2021. (in press)
- [6] Fung-Kee-Fung M, Dodge J, Elit L, Lukka H, Chambers A, Oliver T. Follow-up after primary therapy for endometrial cancer: a systematic review. *Gynecologic Oncology*. 2006; 101: 520–529.
- [7] Stelloo E, Nout RA, Osse EM, Jürgenliemk-Schulz IJ, Jobsen JJ, Lutgens LC, *et al*. Improved Risk Assessment by Integrating Molecular and Clinicopathological Factors in Early-stage Endometrial Cancer-Combined Analysis of the PORTEC Cohorts. *Clinical Cancer Research*. 2016; 22: 4215–4224.
- [8] Kandath C, Schultz N, Cherniack AD, Akbani R, Liu Y, Shen H, *et al*. Integrated genomic characterization of endometrial carcinoma. *Nature*. 2013; 497: 67–73.
- [9] Wang MH, Hui P. Immunohistochemical and molecular pathological classification in the differential diagnosis and prognosis risk assessment of endometrial carcinoma. *Zhonghua Bing Li Xue Za Zhi*. 2021; 50: 1078–1082. (In Chinese)
- [10] Yang Z, Liu X, Yang X, Liao Q. Screening and identification of Lynch syndrome: a systematic review of the frequency of Lynch syndrome-associated clinicopathologic and molecular characteristics in Lynch syndrome gynecologic cancers. *Translational Cancer Research*. 2021; 10: 4523–4531.
- [11] Maratt JK, Stoffel E. Identification of Lynch Syndrome. *Gastrointestinal Endoscopy Clinics of North America*. 2022; 32: 45–58.
- [12] Anyla M, Lefevre JH, Creavin B, Colas C, Svrcek M, Lascols O, *et al*. Metachronous colorectal cancer risk in Lynch syndrome patients-should the endoscopic surveillance be more intensive? *International Journal of Colorectal Disease*. 2018; 33: 703–708.
- [13] Ijsselstein R, van Hees S, Drost M, Jansen JG, de Wind N. Induction of mismatch repair deficiency, compromised DNA damage signaling and compound hypermutagenesis by a dietary mutagen in a cell-based model for Lynch Syndrome. *Carcinogenesis*. 2021. (in press)
- [14] Mrkonjic M, Turashvili G. EPM2AIP1 Immunohistochemistry can be used as Surrogate Testing for MLH1 Promoter Methylation in Endometrial Cancer. *American Journal of Surgical Pathology*. 2021; 46: 376–382.
- [15] Fu Y, Ye Y, Liu X, Zhu G, Xu Y, Sun J, *et al*. Analyzing microsatellite instability and gene mutation in circulating cell-free DNA to monitor colorectal cancer progression. *Translational Cancer Research*. 2021; 10: 2812–2821.
- [16] Han Q, Liu S, Cui Z, Wang Q, Ma T, Jiang L, *et al*. Case Report and Literature Review: Diagnosis, Tailored Genetic Counseling and Cancer Prevention for a Locally Advanced dMMR/MSI-H/TMB-H Lung Cancer Patient With Concurrent Lynch Syndrome Mediated by a Rare PMS2 Splicing Variant (c.1144+1G>A). *Frontiers in Genetics*. 2021; 12: 799807.
- [17] Marszałek-Kruk BA, Wójcicki P. Identification of three novel TCOF1 mutations in patients with Treacher Collins Syndrome. *Human Genome Variation*. 2021; 8: 36.
- [18] Grzanka M, Piekietko-Witkowska A. The Role of TCOF1 Gene in Health and Disease: Beyond Treacher Collins Syndrome. *International Journal of Molecular Sciences*. 2021; 22.
- [19] Hu J, Lai Y, Huang H, Ramakrishnan S, Pan Y, Ma VWS, *et al*. TCOF1 upregulation in triple-negative breast cancer promotes stemness and tumour growth and correlates with poor prognosis. *British Journal of Cancer*. 2022; 126: 57–71.
- [20] Zhu G, Xia H, Tang Q, Bi F. An epithelial-mesenchymal transition-related 5-gene signature predicting the prognosis of hepatocellular carcinoma patients. *Cancer Cell International*. 2021; 21: 166.
- [21] Wu C, Xia D, Wang D, Wang S, Sun Z, Xu B, *et al*. TCOF1 coordinates oncogenic activation and rRNA production and promotes tumorigenesis in HCC. *Cancer Science*. 2022; 113: 553–564.
- [22] Parente P, Malapelle U, Angerilli V, Balistreri M, Lonardi S, Pucciarelli S, *et al*. MMR profile and microsatellite instability status in colorectal mucinous adenocarcinoma with synchronous metastasis: a new clue for the clinical practice. *Journal of Clinical Pathology*. 2022. (in press)
- [23] Ciccia A, Huang J, Izhar L, Sowa ME, Harper JW, Elledge SJ. Treacher Collins syndrome TCOF1 protein cooperates with NBS1 in the DNA damage response. *Proceedings of the National Academy of Sciences of the United States of America*. 2014; 111: 18631–18636.
- [24] Suganuma T, Workman JL. MPTAC links alkylation damage signaling to sterol biosynthesis. *Redox Biology*. 2022; 51: 102270.
- [25] Chen S, Li Y, Wu M, Xue L, Zhu J, Wu M, *et al*. Nucleolar and Coiled-Body Phosphoprotein 1 Is Associated With Stemness and Represents a Potential Therapeutic Target in Triple-Negative Breast Cancer. *Frontiers in Oncology*. 2022; 12: 731528.
- [26] Wu C, Li L, Chen S, Yang X, Zhang CZ, Cao Y. A Zic2/Runx2/NOLC1 signaling axis mediates tumor growth and metastasis in clear cell renal cell carcinoma. *Cell Death & Disease*. 2021; 12: 319.
- [27] Werner A, Iwasaki S, McGourty CA, Medina-Ruiz S, Teerikorpi N, Fedrigo I, *et al*. Cell-fate determination by ubiquitin-dependent regulation of translation. *Nature*. 2015; 525: 523–527.
- [28] Ruhs S, Griesler B, Huebschmann R, Stroedecke K, Straetz N, Ihling C, *et al*. Modulation of transcriptional mineralocorticoid receptor activity by casein kinase I. *The FASEB Journal*. 2022; 36.
- [29] D'Arcy BM, Arrington J, Weisman J, McClellan SB, Vandana, Yang Z, *et al*. PMS2 variant results in loss of ATPase activity without compromising mismatch repair. *Molecular Genetics & Genomic Medicine*. 2022; e1908.
- [30] Heng HHQ. Cancer genome sequencing: the challenges ahead. *BioEssays*. 2007; 29: 783–794.
- [31] Tang Z, Li C, Kang B, Gao G, Li C, Zhang Z. GEPIA: a web server for cancer and normal gene expression profiling and interactive analyses. *Nucleic Acids Research*. 2017; 45: W98–W102.
- [32] Vasaikar SV, Straub P, Wang J, Zhang B. LinkedOmics: analyzing multi-omics data within and across 32 cancer types. *Nucleic Acids Research*. 2018; 46: D956–D963.
- [33] Guo S, Xiao Y, Li D, Jiang Q, Zhu L, Lin D, *et al*. PGK1 and GRP78 overexpression correlates with clinical significance and poor prognosis in Chinese endometrial cancer patients. *Oncotarget*. 2018; 9: 680–690.
- [34] Martinez E. Multi-protein complexes in eukaryotic gene transcription. *Plant Molecular Biology*. 2002; 50: 925–947.
- [35] Wang P, Fan X, Fan Y. The research progress of Treacher Collins syndrome. *Lin Chung Er Bi Yan Hou Tou Jing Wai Ke Za Zhi*. 2016; 30: 333–338. (In Chinese)
- [36] Gu W, Sun L, Wang J, Chen X. The oncogenic role of treacle ribosome biogenesis factor 1 (TCOF1) in human tumors: a pan-cancer analysis. *Aging*. 2022; 14: 943–960.

- [37] McConechy MK, Talhouk A, Li-Chang HH, Leung S, Huntsman DG, Gilks CB, *et al.* Detection of DNA mismatch repair (MMR) deficiencies by immunohistochemistry can effectively diagnose the microsatellite instability (MSI) phenotype in endometrial carcinomas. *Gynecologic Oncology*. 2015; 137: 306–310.
- [38] Shahin, NA, Aladily T, Alhaj NA, Al-Khader A, Alqaqa S, Aljaberi R, *et al.* Differential Expression of Androgen Receptor in Type I and Type II Endometrial Carcinomas: a Clinicopathological Analysis and Correlation with Outcome. *Oman Medical Journal*. 2021; 36: e245–e245.
- [39] Tse WKF. Treacher Collins syndrome: New insights from animal models. *The International Journal of Biochemistry & Cell Biology*. 2016; 81: 44–47.
- [40] Dal Buono A, Gaiani F, Poliani L, Correale C, Laghi L. Defects in MMR Genes as a Seminal Example of Personalized Medicine: From Diagnosis to Therapy. *Journal of Personalized Medicine*. 2021; 11: 1333.