Cell. Mol. Biol. (ISSN: 1165-158X)

Journal Homepage: www.cellmolbiol.org

Cellular and Molecular Biology

Original Article

Cytoplasmic expression of UTP23 promotes colorectal cancer progression

Fang E[1,2,3,#,*], Lili Qian[4,#], Jinlong Tang[5], Zhaowei Tong[3,*]

[1] *Life Sciences Institute, Zhejiang University, Hangzhou, China*
[2] *Liangzhu Laboratory, Zhejiang University, Hangzhou, China*
[3] *Huzhou Key Laboratory of Precision Medicine Research and Translation for Infectious Diseases, Huzhou Central Hospital, Affiliated Central Hospital, Huzhou University, Huzhou, Zhejiang, China*
[4] *Cancer Center, Department of Pathology, Zhejiang Provincial People's Hospital, Affiliated People's Hospital, Hangzhou Medical College, Hangzhou, Zhejiang, China*
[5] *Department of Pathology, The Second Affiliated Hospital, Zhejiang University School of Medicine, Hangzhou, Zhejiang, China*

Article Info

Article history:

Received: January 31, 2023
Accepted: April 01, 2023
Published: January 31, 2024

Use your device to scan and read the article online

Abstract

UTP23 (UTP23 small subunit processome component) plays a pivotal role in the intricate processing and maturation of the small subunit of ribosomes within the nucleolus. In cases of nucleolar stress, such as those observed in certain tumor cells, the aberrant nucleolar organization and structure can lead to the translocation of nucleolar proteins into the nucleus or cytoplasm, consequently impacting the physiological processes of the tumor cells through non-ribosome-related functions. Our investigation revealed altered localization of UTP23 protein in colorectal cancer clinical tissue samples. Upon analyzing UTP23 expression and its correlation with patient prognosis in a cohort of 143 colorectal cancer patients, the result suggested that high cytoplasmic expression pattern of UTP23 occurred in early-stage metastasis-free colorectal cancer and was significantly associated with poor prognosis. Furthermore, we demonstrated that cytoplasmic expression of UTP23 significantly promoted the metastatic and invasive capabilities of colorectal cancer cells, which was not shown in the nucleolcalised UTP23. Intriguingly, mass spectrometry results suggested that KRT5 binds to UTP23 and showed a regulatory influence on UTP23 metastatic potential in colorectal cancer cells. Conclusively, our study demonstrated that the localization of UTP23 plays a key role in colorectal cancer metastatic progression, which may serve as a novel prognostic indicator.

Keywords: Cytoplasmic, UTP23, Colorectal cancer

1. Introduction

Colorectal cancer (CRC) stands as a prevalent gastrointestinal malignancy, representing approximately 10% of all cancer diagnoses worldwide [1]. Alarmingly, 15-20% of CRC patients present with metastasis at the time of their initial surgery, and post-surgical recurrence of metastatic disease afflicts nearly 50% of patients. These statistics underscore the urgent need for further exploration and advancement in the realms of screening, diagnosis, and treatment of colorectal cancer. Colorectal cancer is characterized by its high heterogeneity and intricate molecular underpinnings, necessitating a deeper understanding of its molecular expression patterns. These molecular signatures hold profound implications for the accurate diagnosis of colorectal cancer and the prognostication of its clinical course [2, 3]. Thus, the identification of novel biomarkers for early detection and targeted treatment of CRC is important in the quest to enhance patient outcomes and survival rates.

UTP23 is a protein associated with the processing and maturation of the small subunit of ribosomes in the nucleolus and is mainly involved in the early assembly of ribosomes within the nucleolus [4]. Recent studies have shown that the expression level alteration of UTP23 in ovarian cancer is associated with paclitaxel treatment resistance [5]. Furthermore, a comprehensive single nucleotide polymorphism (SNP) analysis has uncovered a specific locus for UTP23 in colorectal cancer susceptibility, further underscoring its relevance in the realm of oncology [6]. Early studies suggested that the primary functions of the nucleolus were rRNA transcription and ribosome synthesis. Ribosomes are produced and assembled in the nucleolus and processed into mature ribosomes after entering the cytoplasm, which is responsible for protein translation in the cell [7]. In recent years, groundbreaking discoveries have illuminated the dynamic nature of the nucleolus, unveiling its evolving composition and multifaceted functions. In addition to regulating the processing of cellular ribosomes, the nucleolus can also influence processes such as gene transcription, chromatin structure and spatial recombination of genes through its special structure [8-10]. Certain tumor cells undergo nucleolar stress, leading to

* Corresponding author.
E-mail address: efang@zju.edu.cn (F. E); hztongzhaowei@163.com (Z. Tong).
# These authors contributed equally
Doi: http://dx.doi.org/10.14715/cmb/2024.70.1.33

aberrant organization and structure within the nucleolus. This disturbance causes nucleolar proteins to translocate to the nucleus or cytoplasm, subsequently impacting the physiological processes of tumor cells through mechanisms unrelated to ribosomal function [11]. Changes in the subcellular localization of proteins, as they shift between the nucleolus and the cytoplasm, can significantly influence a diverse range of cellular physiological processes, including DNA damage repair, gene transcription, cellular stress responses, telomere maintenance, and the regulation of apoptosis [12-14].

In our comprehensive investigation, we have uncovered a distinctive shift in the localization of UTP23 in CRC patient samples and in vitro. Furthermore, our results have revealed that increased expression of UTP23 in the cytoplasm exerted a potent stimulatory effect on the proliferation, migration, and invasion of colorectal cancer cells. Moreover, our study has delineated the intricate interaction between UTP23 and KRT5 in cytoplasm, elucidating its pivotal role in driving the progression of colorectal cancer.

2. Materials and Methods

2.1. Cell culture and cell lines

The human colorectal cancer RKO, HCT116, SW480, HCT8, DLD1, SW620, HT29, LOVO cell line was purchased from the American Type Culture Collection (ATCC, Manassas, VA, USA). 293T was purchased from the cell bank at the Chinese Academy of Sciences (Shanghai, China). The normal human colonic epithelial cell NCM460 was obtained from EK-Bioscience (Shanghai, China). 293T was maintained in DMEM medium (Gibco, Rockville, MD, USA). HCT116, SW480, HCT8, DLD1 and NCM460 were maintained in RPMI 1640 medium supplemented with 10% fetal bovine serum (Gibco, Rockville, MD, USA), 1% penicillin and streptomycin. All cell lines were maintained in a humidified atmosphere with 5% CO_2 at 37°C.

2.2. Clinical materials

Colorectal carcinoma and paired normal tissue samples (n=143) were obtained from patients during operation at the Department of Medicine, Sir Run Run Shaw Hospital, Zhejiang University School of Medicine. All participating patients were informed. Patients or the public were not involved in the design, conduct, reporting, or dissemination plans of our research.

2.3. Cell proliferation, migration and invasion assay

2000 cells were seeded in 96-well plates and incubated with 100 µl culture medium, then were measured by CCK8 reagent (Boster, CAT#AR1160, Wuhan, China). For migration experiments, 1×10^5 - 2×10^5 HCT116, HCT8, RKO and DLD1 cells were plated into the upper chamber of an 8 µm transwell chamber (Corning Costar, CAT# 3422, Corning, NY, USA). For invasion experiments, 50 µl of diluted Matrigel (Corning, BD Matrigel, CAT#356234, Corning, NY, USA) was added before plating cells in a transwell chamber. Then, 600 µl of medium containing 10% FBS was added to the lower chamber. After 24–48 h of culture, the chambers were fixed with 4% paraformaldehyde and stained with crystal violet. After the chambers were air-dried, photographs were taken, and the percentages of positive cell area were quantified using ImageJ software (1.49 v). Three fields were observed per chamber, and the experiment was repeated three times.

2.4. Immunoblotting

Whole cell lysates were extracted with RIPA and a complete protease inhibitor mix (Fdbio science, CAT#FD009) followed by ultrasonication. SDS-PAGE was performed to separate equal amounts of protein lysates followed by transferring onto nitrocellulose membranes (Millipore, Billerica, MA, USA), blocking with 5% skim milk and incubating with antibody overnight at 4°C. Then membranes were incubated with fluorescence secondary antibodies for 1h RT and detected by Odyssey (LI-COR). For the immunoprecipitation assay, Co-IP lysis buffer (20 mM Tris-HCl, 150 mM NaCl2, 1.5 mM EDTA, 0.5 mM NaVO4, 0.5% NP-40, pH=8.0) with complete protease inhibitor were used for harvest whole cell lysate. Cell lysates were incubated with beads overnight at 4°C. The beads were washed by washing buffer (20 mM Tris-HCl, 50 mM NaCl2, 1.5 mM EDTA, 0.5 mM NaVO4, 0.5% NP-40, pH=8.0), and eluted by elution buffer (Glycine, pH=2.0) for the Western blot assay.

2.5. Plasmid Constructions

The full-length or partial UTP23 CDS with a 3 × FLAG tag was cloned into the over-expression vector pCDH-CMV-MCS-EF1-puro. The shRNA was designed and synthesized from the gene sequence of UTP23 and constructed into PLKO.1 vector. The UTP23 mutants were designed and constructed by using the Mut Express II Fast Mutagenesis Kit V2 (Vazyme, Nanjing, China).

2.6. Immunohistochemical

The paraffin-embedded tissue sections were cut into 4 µm slides for the hematoxylin and eosin (H&E) and immunohistochemical (IHC) assays. All the slides were dewaxed in different concentration dimethylbenzene solutions and then treated with citrate buffer (pH 6.0) at high pressure for the appropriate time. After the antigen retrieval, the slides were blocked by 10% fetal bovine serum for 30 mins and incubated with corresponding primary antibody for more than 12 hours. On the next day, all the slides were incubated with secondary antibodies, DAB solutions, and Meyer's hematoxylin successively. Last the slides were dehydrated and ended up covering the coverslips.

2.7. Immunofluorescence Assays

Cells seeded on the confocal dishes were fixed with 4% paraformaldehyde and then treated with 0.1% Triton X-100. The cells were blocked by 10% fetal bovine serum and incubated with the corresponding primary antibody overnight. On the next day, cells were incubated with the corresponding secondary fluorescence antibody and then with DAPI buffer. All the immunofluorescence assays were repeated three times.

2.8. Mass Spectrometry

The FLAG magnetic beads were incubated with DLD1-WT/MU cell lysate, respectively. The precipitated complexes were delivered to PTMBIO for mass spectrometry analysis.

2.9. Statistical Analysis

Data represent mean ± SEM from three independent experiments. P-value <0.05 was considered as statistically

significant. NS denotes not significant.

3. Results

3.1. Altered localization of UTP23 in colorectal cancer tissues and cell lines

To investigate the expression and localization of UTP23 in colorectal cancer, we used immunohistochemistry to examine 143 colorectal and paracancerous tissues in our CRC tissue bank. Interestingly, the localization of UTP23 showed a notable transformation, shifting from the nucleus to the cytoplasm in some colorectal cancer specimens (Figure 1A). The maximum value of the Jordon index was used as a threshold to categorize the samples into UTP23 high and low-expression groups. Pearsonx2 test showed no correlation between UTP23 localization and clinicopathological parameters (Table 1). The patients with elevated cytoplasmic expression of UTP23 exhibited a notably poorer prognosis in the cohort lacking lymph node metastasis when compared with the low cytoplasmic expression patients (Figure 1B). Furthermore, we conducted an analysis of the expression and localization of UTP23 in colorectal cancer cell lines using immunofluorescence. The result revealed that the intestinal epithelial cell line NCM460 exhibited a minimal level of UTP23 expression, with virtually no discernible cytoplasmic presence. However, the expression of UTP23 showed increased in other colorectal cancer cell lines, with a substantial portion loca-

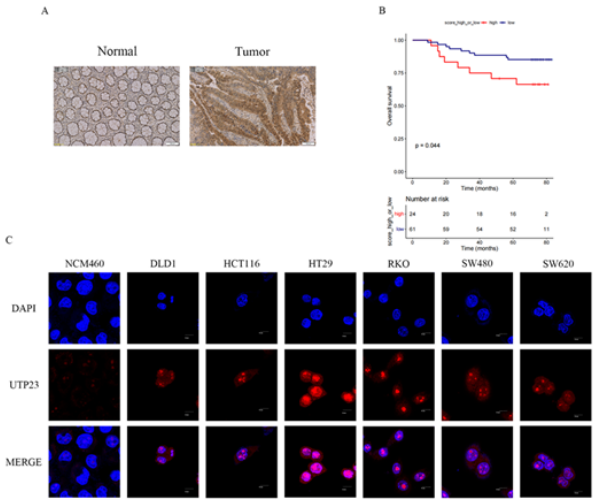

Fig. 1. Characterization of the expression of UTP23 in colorectal cancer clinical samples and cell lines. (A) Immunohistochemical analysis of UTP23 expression in clinical samples of colorectal cancer and adjacent paracancerous tissues (Magnification 200 ×). (B) Correlation between UTP23 expression levels and prognosis in patients diagnosed with colorectal cancer. (C) Immunofluorescence assessment of UTP23 expression and subcellular localization in colorectal cancer cell lines. UTP23 staining is shown in red; blue (DAPI) indicates cell nuclei(scale bar= 10 μm). ns, not significant with P> 0.05, *P<0.05, ***P<0.001.

Table 1. Relationship between UTP23 expression and clinicopathologic parameters.

Parameters	Cytoplasmic expression			Nuclear expression		
	Low(n)	High(n)	*P*	Low(n)	High(n)	*P*
Age						
≤60	52	12	0.018	56	8	0.294
>60	50	29		64	15	
Gender						
Male	56	26	0.352	73	9	0.054
Female	46	15		47	14	
Tumor location						
Colon	58	22	0.637	66	14	0.662
Rectum	42	19		52	9	
Histological grade						
High	78	28	0.267	92	14	0.097
Low	23	13		27	9	
Lymph node metastasis						
Yes	36	17	0.631	41	12	0.137
No	61	24		74	11	
Distant metastases						
Yes	9	6	0.357	13	2	0.703
No	83	33		96	20	
TNM stage						
Early	61	22	0.383	72	11	0.221
Late	38	19		45	12	

lized in the cytoplasm (Figure 1C). These suggested that the translocation of UTP23 may provide an effect on CRC prognosis.

3.2. Knock-down UTP23 inhibits CRC cell proliferation, migration and invasion in vitro

To further investigate the biological function of UTP23 in colorectal cancer, we examined the mRNA and protein levels of UTP23 in CRC cell lines (Figure S1A-B). The results showed that UTP23 had higher expression in RKO, and HCT116 cells and lower expression in HCT8 and DLD1 cells. First, we used shRNA to stablely knock down the expression of UTP23 in HCT116 and RKO cell lines (Figure S1C-D). CCK8 assay and clone formation assay data revealed that down-regulated UTP23 decreased the proliferation ability in both cell lines (Figure 2A-B). In addition, the Transwell assay and Wound healing assay showed that the migration and invasion abilities were also decreased when UTP23 was knocked down (Figure 2C-D). We also used the sphere assay to detect the sphere formation capability in UTP23 knockdown RKO and HCT116 cell lines. As shown in Figure 2E, UTP23 knockdown deregulated the sphere formation. Collectively, down-regulated UTP23 inhibited CRC proliferation and metastasis in vitro.

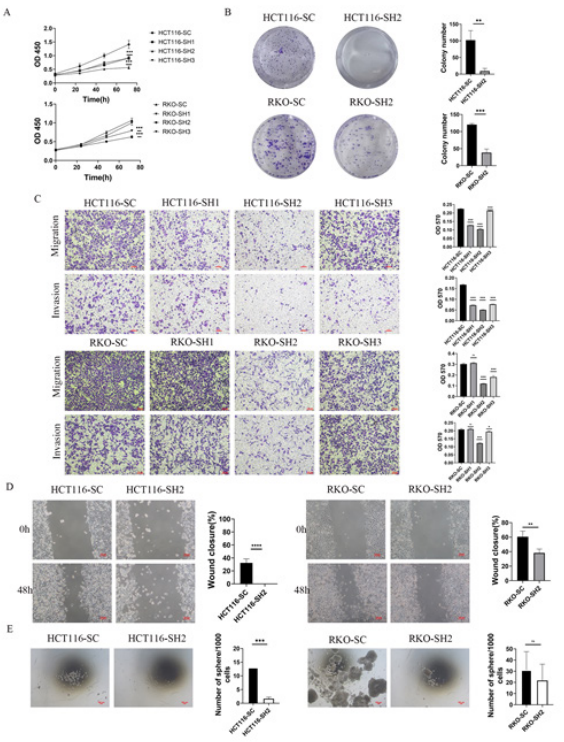

Fig. 2. UTP23 knockdown impairs the proliferation and metastatic potential of colorectal cancer cells. (A, B) Cell proliferation ability was validated using CCK8 (A) and colony-forming assay (B). (C) Transwell assay was conducted to assess the migration and invasion abilities of HCT116 and RKO cells following UTP23 knockdown. The accompanying graph displays the quantitative analysis results (scale bar= 100 μm). (D) Wound healing assay was performed to evaluate the mobility of HCT116 and RKO cells after UTP23 knockdown. The corresponding graph depicts the quantitative analysis results. (scale bar= 100 μm). (E) Sphere assay was conducted to assess the sphere formation capability of HCT116 and RKO cells following UTP23 knockdown. The right graph shows the quantitative analysis results (scale bar= 100 μm). ns, not significant with P>0.05, *P<0.05, ***P<0.001.

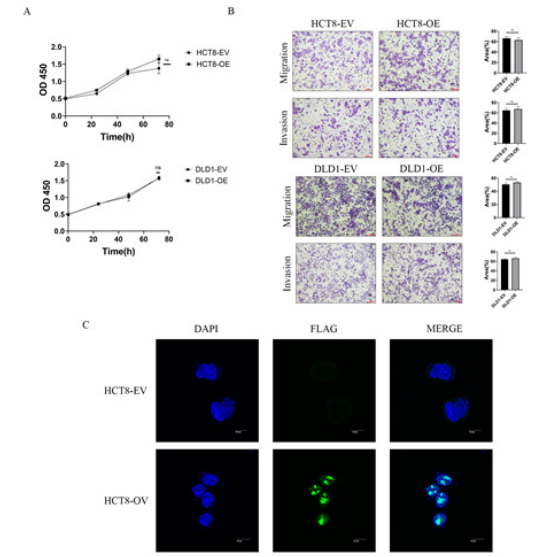

Fig. 3. Overexpression of UTP23 did not impact the malignancy of colorectal cancer cells. (A) The cell proliferation capacity was validated through the use of a CCK8 assay. (B) Transwell assay was employed to assess the migration and invasion capabilities of HCT8 and DLD1 cells following UTP23 overexpression. The accompanying graph illustrates the quantitative analysis findings (scale bar= 100 μm). (C) The subcellular localization of exogenously introduced UTP23 in HCT8 cells was visualized using immunofluorescence (scale bar= 10 μm). ns, not significant with P>0.05, *P<0.05, ***P<0.001.

3.3. The cytoplasmic expressed UTP23 significantly enhance the proliferation and metastatic potential in CRC cells

In order to delve deeper into the impact of UTP23 in CRC, we overexpressed UTP23 in HCT8 and DLD1 cell lines (Figure S1E-F), and examined the proliferative and metastatic capacity of CRC cell lines by using CCK8 and transwell assays. We found that overexpressed UTP23 did not exert any effect on the proliferative and metastatic capacity in HCT8 and DLD1 cell lines (Figure 3A-B). Taking into account the expression pattern of UTP23 in clinical samples, we employed an immunofluorescence assay to examine the localization of exogenously overexpressed UTP23 in cells. The immunofluorescence result suggested that the exogenous UTP23 was predominantly localized in the nucleus, with minimal expression observed in the cytoplasm (Figure 3C). In order to analyze whether the tumor-associated biological function of UTP23 was dependent on its location. We used the cNLS Mapper and Wregex to analyze the amino acid sequence of UTP23. We found there were three distinct nuclear localization sequences in UTP23, spanning amino acid ranges 192-197, 208-213, and 224-229 (Figure 4A). Subsequently, we introduced mutations to the amino acid sequences 192-197, 208-213, and 224-229, substituting them with alanine. Then we overexpressed the mutant UTP23 and verified its expression through western blot and qRT-PCR analyses (Figure 4B). Moreover, the immunofluorescence analysis revealed a diffuse expression pattern of mutant UTP23 in CRC cells (Figure 4C). Upon further investigation of UTP23 localization in CRC cells, we used western blot analysis of nuclear plasma isolation. The result revealed consistent with the immunofluorescence assay that the UTP23 mutant was

translocated into the cytoplasm in comparison to the wild type (Figure 4D).

Furthermore, we used the CCK8 assay and clone formation assay to detect the proliferative capacity of mutant UTP23 overexpressed DLD1 and HCT8 cells. As shown in Figure 5A-B, mutant UTP23 promoted proliferation in both cell lines. The conducted transwell migration, invasion, and wound healing assays revealed that the overexpression of mutant UTP23 significantly promotes the metastatic capacity of CRC cells (Figure 5C and 5E). In addition, the expression of cell junction proteins Occludin and E-cadherin was found to be diminished in UTP23 mutant CRC cells (Figure 5D). Moreover, the overexpression of mutant UTP23 was found to augment the spheroid-forming capacity of CRC cells (Figure 5F). In conclusion, the cytoplasmic UTP23 showed a carcinogenesis ability both in proliferation and metastasis.

3.4. KRT5 interacts with cytoplasmic UTP23

To validate the molecular pathway through which mutant UTP23 drives CRC progression, we used immunoprecipitation-mass spectrometry to identify the protein partners of wild-type UTP23 and mutant UTP23. As showed in Figure 6A, the mass spectrometry predicted the binding ability between mutant UTP23 and KRT5, but not wild-type UTP23. KRT5 has been reported to play a role in tumorigenesis and progression in a variety of tumor types

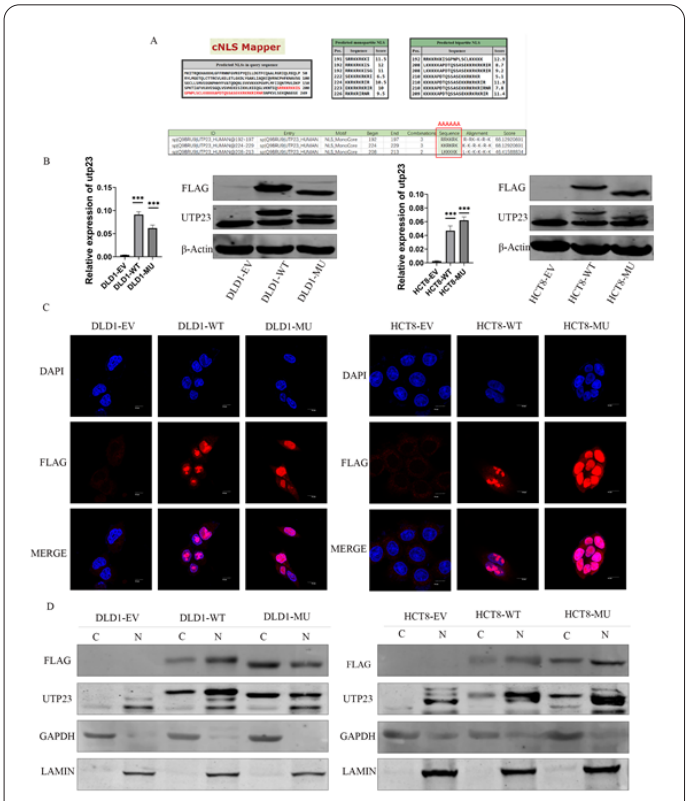

Fig. 4. Overexpression of mutant UTP23 in CRC cell lines. (A) The cNLS (http://nls-mapper.iab.keio.ac.jp/cgi-bin/NLS_Mapper_form.cgi) and Wregex (http://ehubio.ehu.eus/wregex/) sites predict the NLS sequence for UTP23. (B) Western blot and qPCR analyses were utilized to identify the overexpression of wild-type and mutant UTP23 in DLD1 and HCT8 cells. (C) Immunofluorescence was employed to ascertain the subcellular localization of wild-type and mutant UTP23 in DLD1 and HCT8 cells (scale bar= 10 μm). (D) Western blot analysis was conducted to identify the expression of UTP23 and mutant UTP23 in the nucleus and cytoplasm of DLD1 and HCT8 cells, respectively. ns, not significant with P>0.05, *P<0.05, ***P<0.001.

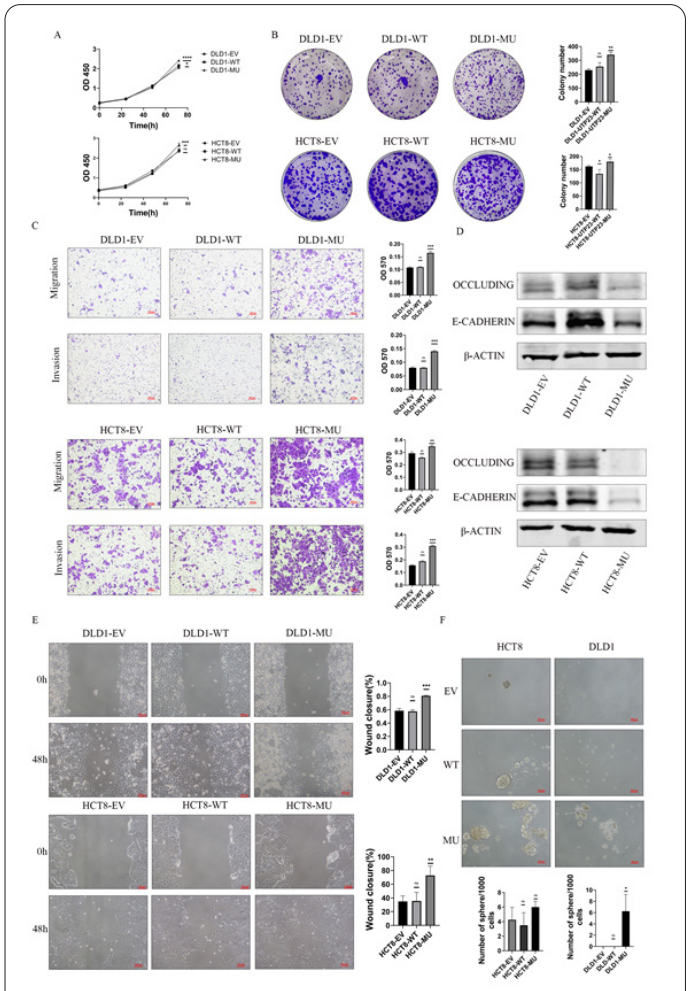

Fig. 5. Overexpression of mutant UTP23 promotes the malignancy of CRC cells. (A, B) Cell proliferation ability was validated using the CCK8 assay (A) and the colony-forming assay (B). The right graph shows the quantitative analysis results. (C) Transwell assay was employed to investigate the migration and invasion capabilities of HCT8 and DLD1 cells following overexpression of wild-type UTP23 or mutant UTP23. The accompanying graph on the right demonstrates the quantitative analysis results (scale bar= 100 μm). (D) The expression of cell junction proteins Occludin and E-cadherin was assessed using western blot analysis. (E) The wound healing assay was conducted to assess the mobility of HCT8 and DLD1 cells following the overexpression of wild-type UTP23 or mutant UTP23. The graph on the right displays the quantitative analysis results (scale bar= 100 μm). (F) The sphere assay was utilized to evaluate the sphere formation capability of HCT116 and RKO cells subsequent to the overexpression of wild-type UTP23 or mutant UTP23. The right graph shows the quantitative analysis results (scale bar= 100 μm). ns, not significant with P>0.05, *P<0.05, ***P<0.001.

[15, 16]. Then, we transfected FLAG-tagged wild-type UTP23 or FLAG-tagged mutant UTP23 together with HA-tagged KRT5 into 293T cells and performed co-immunoprecipitation experiment by anti-HA antibody. The result indicated a weak interaction between wild-type UTP23 and KRT5, but mutant UTP23 exhibited a strong interaction with KRT5 (Figure 6B-C). In addition, it showed the same result when we performed co-immunoprecipitation experiment with anti-Flag antibody to verify the combination with UTP23 and KRT5 (Figure 6D). In order to identify the specific amino acid site at which UTP23 binds to KRT5, we constructed several truncated UTP23 according to the structural domain (Figure 6E). We transfected

FLAG-tagged truncated UTP23 and HA-tagged KRT5 into 293T cells and performed co-immunoprecipitation experiment by anti-FLAG antibody. The result suggested that the sequence 148-181 of UTP23 is a critical region for KRT5 binding (Figure 6F). Moreover, the immunofluorescence experiment showed that the 1-148 and 21-181 UTP23 truncated proteins were localized in the cytoplasm, while the 149-249 truncated protein was localized in the nucleus (Figure 6G). These suggested that nuclear-located UTP23 lost its binding ability with KRT5 even through it had the KRT5 binding domain.

4. Discussion

In our study, we observed altered localization of UTP23 in clinical CRC tissue samples. In addition, the cytoplasmic localization of UTP23 indicated the malignant development in patients. Knockdown UTP23 resulted in significant inhibition of proliferation and metastasis in CRC cells. Furthermore, our findings indicated that overexpression of mutant UTP23, but not wild-type UTP23, facilitated the progression of CRC cells. Additionally, mutant UTP23 interacted with KRT5 in cytoplasm, an intermediate filament protein associated with the progression of multiple tumors.

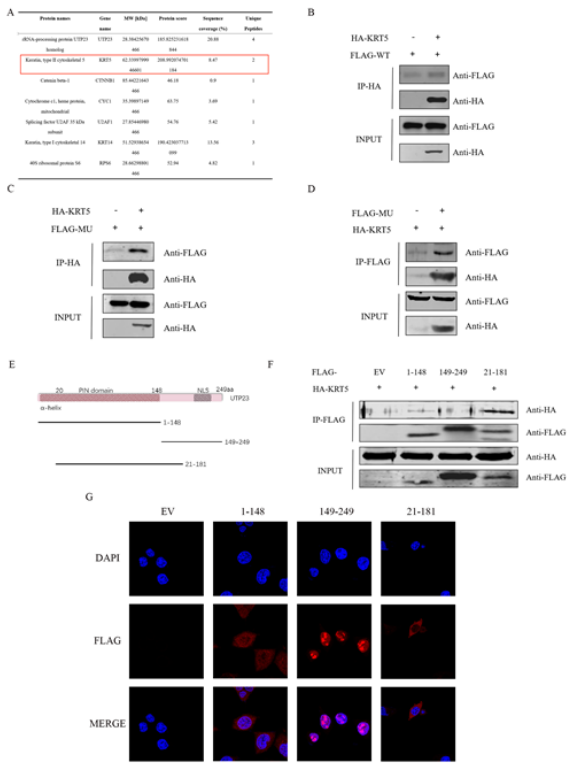

Fig. 6. The interaction between mutant UTP23 and KRT5 enhances the malignancy of colorectal cancer cells. (A) The amino acid sequence of KRT5 was identified through Co-IP (co-immunoprecipitation) and MS (mass spectrometry) analyses. (B) Co-IP analysis confirmed a weak binding between wild-type UTP23 and KRT5 in 293T cells. (C,D) Co-IP analysis confirmed a strong binding of mutant UTP23 to KRT5 in 293T cells. (E) Schematic diagram of truncated UTP23 plasmid construction. (F) The Co-IP assay unveiled that the amino acid sequence 148-181 of UTP23 constitutes a critical region for KRT5 binding. (G) Immunofluorescence was utilized to visualize the localization of truncated UTP23 in 293T cells. FLAG staining is shown in red; blue (DAPI) indicates cell nuclei (scale bar= 10 μm).

The regulatory role of cell junctions in CRC neoplasia is well-documented [17, 18]. In our study, we observed that cytoplasmic expression of UTP23 led to a decrease in the expression of cell junction proteins, including Occludin and E-cadherin. This disruption of cell junctions has the potential to contribute to intestinal inflammation, ultimately progressing to CRC.

UTP23 is a protein primarily localized in the nucleolus. In our study, we observed that the knockdown of UTP23 expression significantly inhibited the progression of CRC cells, whereas overexpression of wild-type UTP23 did not yield the same effect. This observation could be attributed to the fact that UTP23 plays a crucial role in ribosome processing, and the knockdown of UTP23 directly inhibited protein synthesis in CRC cells. The mutant UTP23 is observed to be diffusely expressed in CRC cells and is capable of promoting CRC progression through non-ribosomal processing-related functions. Additionally, there have been reports indicating altered localization of nucleolus protein, which consequently impacts tumor progression through non-ribosomal processing-related functions [19, 20]. Furthermore, our investigation into UTP23 mutations in CRC patients within the TCGA database unveiled a missense mutation that was exclusive to a single patient. This mutation occurred within the nuclear localization sequence (NLS), where lysine at position 195 was altered to glutamine. This substitution of a basic amino acid with a neutral amino acid may potentially disrupt the disordered structure of UTP23, subsequently impacting the localization of UTP23 within the nucleus.

KRT5 is an intermediate filament protein that is distributed in the cytoplasm for the formation of fibers network [21, 22]. KRT5 has been utilized as a molecular marker associated with promoting tumor progression in a variety of cancer types [23, 24]. Our findings indicate that UTP23 has the capability to bind to KRT5 in the cytoplasm, thereby contributing to the promotion of CRC progression. However, the exact molecular mechanism remains elusive.

Declaration of Competing Interest

The authors declare no competing interests.

Ethics approval and consent to participate

Ethical approval for patients was obtained from the Ethics Committee of Department of Medicine, Zhejiang University (Ethics Committee number: 2021–021).

Informed Consent

All participating patients were informed. Signed written informed consents were obtained from the patients.

Availability of data and material

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Authors' contributions

Conceptualization: FE; methodology: FE, JLT and LLQ; investigation: FE, LLQ,; writing—original draft: FE; writing—review and editing: FE and ZWT; supervision: FE and ZWT.

Funding

No funding.

Acknowledgements
We Thank Qiong Huang and Yanwei Li from Zhejiang University Public Technology Platform for their technical guidance.

References

1. Siegel RL, Miller KD, Wagle NS, Jemal A (2023) Cancer statistics, 2023. Ca-a Cancer Journal for Clinicians 73:17-48. doi: 10.3322/caac.21763
2. Fearon ER (2011) Molecular genetics of colorectal cancer. Annual Review of Pathology-Mechanisms of Disease 6:479-507. doi: 10.1146/annurev-pathol-011110-130235
3. YH, Chen YX, Fang JY (2020) Comprehensive review of targeted therapy for colorectal cancer. Signal Transduction and Targeted Therapy 5:22. doi: 10.1038/s41392-020-0116-z
4. Hoareau-Aveilla C, Fayet-Lebaron E, Jady BE, Henras AK, Kiss T (2012) Utp23p is required for dissociation of snR30 small nucleolar RNP from preribosomal particles. Nucleic Acids Research 40:3641-3652. doi: 10.1093/nar/gkr1213
5. Fu Z, Wang C, Chen Y, Zhang X, Wang X, Xie X (2019) Downregulation of UTP23 promotes paclitaxel resistance and predicts poorer prognosis in ovarian cancer. Pathology Research and Practice 215:152625. doi: 10.1016/j.prp.2019.152625
6. Carvajal-Carmona LG, Cazier JB, Jones AM, Howarth K, Broderick P, Pittman A et al (2011) Fine-mapping of colorectal cancer susceptibility loci at 8q23.3, 16q22.1 and 19q13.11: refinement of association signals and use of in silico analysis to suggest functional variation and unexpected candidate target genes. Human Molecular Genetics 20:2879-2888. doi: 10.1093/hmg/ddr190
7. Iarovaia OV, Minina EP, Sheval EV, Onichtchouk D, Dokudovskaya S, Razin SV et al (2019) Nucleolus: A Central Hub for Nuclear Functions. Trends in Cell Biology 29:647-659. doi: 10.1016/j.tcb.2019.04.003
8. Allinne J, Pichugin A, Iarovaia O, Klibi M, Barat A, Zlotek-Zlotkiewicz E et al (2014) Perinucleolar relocalization and nucleolin as crucial events in the transcriptional activation of key genes in mantle cell lymphoma. Blood 123:2044-2053. doi: 10.1182/blood-2013-06-510511
9. Matheson TD, Kaufman PD (2016) Grabbing the genome by the NADs. Chromosoma 125:361-371. doi: 10.1007/s00412-015-0527-8
10. Pichugin A, Iarovaia OV, Gavrilov A, Sklyar I, Barinova N, Barinov A et al (2017) The IGH locus relocalizes to a "recombination compartment" in the perinucleolar region of differentiating B-lymphocytes. Oncotarget 8:40079-40089. doi: 10.18632/oncotarget.16941
11. Ogawa LM, Baserga SJ (2017) Crosstalk between the nucleolus and the DNA damage response. Mol Biosyst 13:443-455. doi: 10.1039/c6mb00740f
12. Boulon S, Westman BJ, Hutten S, Boisvert FM, Lamond AI (2010) The nucleolus under stress. Molecular Cell 40:216-227. doi: 10.1016/j.molcel.2010.09.024
13. Penzo M, Montanaro L, Trere D, Derenzini M (2019) The Ribosome Biogenesis-Cancer Connection. Cells 8:55. doi: 10.3390/cells8010055
14. Russo A, Russo G (2017) Ribosomal Proteins Control or Bypass p53 during Nucleolar Stress. International Journal of Molecular Sciences 18:140. doi: 10.3390/ijms18010140
15. Zhang Z, Tu K, Liu F, Liang M, Yu K, Wang Y et al (2020) FoxM1 promotes the migration of ovarian cancer cell through KRT5 and KRT7. Gene 757:144947. doi: 10.1016/j.gene.2020.144947
16. Ricciardelli C, Lokman NA, Pyragius CE, Ween MP, Macpherson AM, Ruszkiewicz A et al (2017) Keratin 5 overexpression is associated with serous ovarian cancer recurrence and chemotherapy resistance. Oncotarget 8:17819-17832. doi: 10.18632/oncotarget.14867
17. Han F, Yang B, Zhou M, Huang Q, Mai M, Huang Z et al (2022) GLTSCR1 coordinates alternative splicing and transcription elongation of ZO1 to regulate colorectal cancer progression. Journal of Molecular Cell Biology 14:mjac009. doi: 10.1093/jmcb/mjac009
18. Wang T, Wang P, Ge W, Shi C, Xiao G, Wang X et al (2021) The probiotic Companilactobacillus crustorum MN047 alleviates colitis-associated tumorigenesis via modulating the intestinal microenvironment. Food & Function 12:11331-11342. doi: 10.1039/d1fo01531a
19. Yang HW, Kim TM, Song SS, Menon L, Jiang X, Huang W et al (2015) A small subunit processome protein promotes cancer by altering translation. Oncogene 34:4471-4481. doi: 10.1038/onc.2014.376
20. Brunetti L, Gundry MC, Sorcini D, Guzman AG, Huang YH, Ramabadran R et al (2018) Mutant NPM1 Maintains the Leukemic State through HOX Expression. Cancer Cell 34:499-512. doi: 10.1016/j.ccell.2018.08.005
21. Toivola DM, Boor P, Alam C, Strnad P (2015) Keratins in health and disease. Current Opinion in Cell Biology 32:73-81. doi: 10.1016/j.ceb.2014.12.008
22. Etienne-Manneville S (2018) Cytoplasmic Intermediate Filaments in Cell Biology. Annual Review of Cell and Developmental Biology 34:1-28. doi: 10.1146/annurev-cellbio-100617-062534
23. Sizemore GM, Sizemore ST, Seachrist DD, Keri RA (2014) GABA(A) receptor pi (GABRP) stimulates basal-like breast cancer cell migration through activation of extracellular-regulated kinase 1/2 (ERK1/2). Journal of Biological Chemistry 289:24102-24113. doi: 10.1074/jbc.M114.593582
24. van de Rijn M, Perou CM, Tibshirani R, Haas P, Kallioniemi O, Kononen J et al (2002) Expression of cytokeratins 17 and 5 identifies a group of breast carcinomas with poor clinical outcome. American Journal of Pathology 161:1991-1996. doi: 10.1016/S0002-9440(10)64476-8