

A Novel N7-Methylguanosine (M7g) Methylation-Associated Mirna Signature for Prognostic Value in Patient with Carcinoma of Colon

Chongyang Wang^{1*}, Shouji Liu²

¹Department of Anorectal Surgery, the First Affiliated Hospital of Hunan College of TCM, Zhuzhou 412000, China

²Department of Anorectal Surgery, the First Affiliated Hospital of Hunan College of TCM, Zhuzhou 412000, China

*Corresponding author

Chongyang Wang, Department of Anorectal Surgery, The First Affiliated Hospital of Hunan college of TCM, 571 Renmin middle Road, Zhuzhou 412000, and China.

Submitted: 27 Jul 2022; Accepted: 02 Aug 2022; Published: 17 Aug 2022

Citation: Wang C*, Liu S. (2022). A Novel N7-Methylguanosine (M7g) Methylation-Associated Mirna Signature for Prognostic Value in Patient with Carcinoma of Colon. *J Gene Engg Bio Res*, 4(2), 271- 278.

Abstract

Background

M7G methylation-associated genes were selected as the main research perspectives in this study, establishing a novel m7G methylation-associated miRNA risk signature for prognostic value in-patient with carcinoma of colon.

Methods

We identified two genes that are m7G methylation-associated and have been validated by searching in the PubMed database. The m7G methylation-associated miRNAs were screened via estimating the differentially expressed of miRNAs in 457 carcinoma of colon patient samples and 8 matched samples from a database named The Cancer Genome Atlas (TCGA). Selecting Single-factor Cox analyses as well as multi-factor Cox analyses constructed the risk signature of 12 m7G methylation-associated miRNAs. Nomograms was adopted to predict the potential impact of m7G methylation-associated miRNAs on occurrence, development. The functions of DEGs that deserving to be noticed were classified and analyzed by functional enrichment analysis.

Disease ontology analysis was used to explore the potential connection various diseases and carcinoma of colon. The outcome of associated analysis about immunocyte or immunological functioning revealed that what role does immunocyte or immunological functioning play in the development of carcinoma of colon. Finally, the relationship between high or low expression of significantly DEGs and immune cells and immune function was analyzed.

Results

The results of nomogram analysis were as follows that the risk signature might predict the prognosis and the overall cumulative probability of survival (1-,3-,5-year) effectively about patients with carcinoma of colon. Through the analysis of Kaplan-Meier curve outcomes, we can draw a conclusion that depressed expressed of hsa-miR-21-3p might be significantly relevant with decreased survival of patients. What the analyses of Gene ontology (GO), Kyoto Encyclopedia of Genes and Genomes (KEGG) and Disease ontology(DO), taking advantage of the common DEGs about risk score and immune score, demonstrated that these common DEGs play a partial role in the occurrence and development of tumors. Through disease ontology analysis, we found that eating disorder might be associated with carcinoma of colon.

Conclusions

M7G methylation-associated miRNAs were related to the prognosis of patients with carcinoma of colon. A risk signature established based on these miRNAs could effectively predict the survival rate of later period in patients with carcinoma of colon. According to the displayed outcome from Our research that m7G methylation-associated miRNA regulators can serve as reliable prognostic biomarkers of carcinoma of colon, which might Greater probably be applied as potential targets of therapeutic strategies.

Keywords: Carcinoma of Colon, M7g Methylation-Related MiRNAs, Prognosis, Prognostic Model.

Introduction

It is kind of that stage right now: Our Alternative plan of comprehensive treatment for carcinoma of colon is still very scarce. In addition to radiation therapy, chemotherapy, surgical operation, Immune related or targeted drug therapy, we have no better

choice to deal with the related circumstances of body caused by tumors.

The occurrence and development of carcinoma of colon tend to be younger, and it is one of the most common type of malignancy.

nant tumor in gastrointestinal digestive system [1]. Colorectal cancer (CRC) is the third deadliest and fourth most common tumor in the world [2, 3]. The popularization of painless colonoscopy has increased the detection rate and diagnosis rate of colon cancer [4]. The number of deaths from colon cancer is increasing year by year. The related pathological mechanisms of colon cancer cover a wide range. Some scholars believe that the metabolic mechanism of colon cancer can be explored from the perspectives of cell scorch death, iron death and necrotic apoptosis [5-9]. Although these mechanisms are participated in the transformation process of carcinoma of colon and affect the predicted 5-year survival rate of patient's carcinoma of colon from different angles, it is still necessary to continue to explore new mechanisms in order to further study the pathogenesis of colon cancer. The current diagnosis and treatment methods of colon cancer include surgery, chemotherapy, targeted therapy, radiotherapy and so on [10]. However, at this stage, according to the understanding of the relevant mechanisms of colon cancer and the formulation of the corresponding treatment plan, the 5-year survival rate of patients is ideal. Some patients have recurrence of colon tumor and distant metastasis of liver and lung within 5 years [11]. Therefore, it is momentous to search new research mechanisms to clarify the occurrence and development process of carcinoma of colon and the prognosis of patients, to promote and comprehensively evaluate the formation of individualized programs.

DNA methylation, a mode of DNA modification, which can affect hereditary potency without changing the original structure of DNA [12, 13]. DNA methylation can shut down the gene transcription process, while demethylation can induce gene reactivation and expression [14]. A large number of studies have shown that DNA methylation leads to conformational changes in some regions of DNA, destroying the structure stability, which affects the process of effective binding between DNA and protein, and inhibits the binding efficiency of transcription factors to DNA in the promoter region, the mechanism of DNA methylation inhibiting gene transcription, dominating gene transcription and expression process [15-18]. DNA methylation under the action of DNA methyl transferase can be replicated to offspring DNA through the process of gene transcription. Some studies have shown that m7G related genes mediate the transcriptional process of tumors [19]. RNA methyl transferase mettl1 participates in N7 methyl guanosine (m7G) modification of tRNA [20]. Here, we discovered that there may be a significant negative correlation between the low survival rate of cancer patients and the overexpression of Mettl1 in vivo [21]. Mettl1 depletion resulted in the abundance of m7G modified tRNA significantly decreased and cell generation cycle changes and inhibited carcinogenicity. In the opposite direction, mettl1 over-activating likely result in transformation from normal cells to cancer cells, eventually malignant tumor [22].

To further understand the pathogenesis of colorectal cancer, We searched the keywords in PubMed, for instance, "miRNA", "microRNA" and "colon cancer", "colorectal cancer", "rectal cancer". Many miRNAs, belonging to a species of noncoding RNAs, have been found to facilitate to the development of colorectal cancer [23]. These transcripts affect TGF- β Wnt/ β - Expression and activity of CRC related pathways such as catenin, MAPK, PI3K / Akt. In the prerequisite of colorectal cancer,

miRNA interacts with long noncoding RNA and affects the CRC process [24-26]. Some researchers even evaluated the feasibility of miRNA in colorectal cancer in animal models, and miRNA levels in feces and serum have been employed to differentiate patients with colorectal cancer or healthy controls, revealing the diagnostic function of these miRNAs in colorectal cancer [27, 28]. MicroRNAs (miRNAs) play an important regulated role in a variety of genes in all aspects of carcinogenesis [29]. Therefore, they have been proposed as the target of treatment, the way and marker of early tumor detection [30]. Therefore, we can predict and analyze the long-term survival of tumor by predicting the upstream miRNA of tumor through m7G related genes.

Methods

Data sources

The TCGA database was employed to acquire the matrix data of miRNA in patients with carcinoma of colon, the matrix data of miRNA including 465 patient samples (457 tumors and 8 control samples). The next research process was based on these matrix data for analysis. In addition, patient data including age, gender, TNM staging, Pathological classification, survival outcome, and survival duration were collated for analysis.

Identification of Differentially Expressed m7G Methylation-Associated miRNAs

By summarizing the previous m7G-related comprehensive literature, we acquired METTL1 and WDR4, which were authenticated to be m7G methylation-related gene, taking advantage of two genes Prediction of upstream regulation related miRNAs. Based on above statistical analysis, 792 miRNAs known to be associated with the m7G methylation-related gene (METTL1 and WDR4) were examined. The expression of these miRNAs in carcinoma of colon tumors and normal tissue samples was compared using the R 'edgeR' and 'limma' packages. The differentially expressed miRNAs and the expression of these miRNAs in the sample tissues were obtained by analysis (Up or down regulation).

Generation of the Prognostic m7G Methylation-Related miRNA Model

Exploring the association between m7G methylation-related miRNA expression and overall survival (OS) in patients with carcinoma of colon used the cohort downloaded from the TCGA. We set $P < 0.05$ as the selection criteria for univariate and multivariate Cox regression analysis. R "survival" and "forest plot" packages were applied to our Analyses. Kaplan-Meier curves were utilized for survival analyses, with log-rank tests and univariate Cox proportional hazard regression models used to generate p-values as well as hazard ratios (HRs) with 95% confidence intervals (CIs). M7G methylation-related MiRNAs that may have significant prognostic significance can be retained and used for subsequent statistical analysis and evaluation. A risk score was defined based on these prognostic miRNAs. Using the R 'survival', 'ROCR', and 'time ROC' packages to generate receiver operating characteristic (ROC) curves ultimately analyzed the 1-, 3- and 5-year OS of patients. The prognostic relevance of m7G methylation-associated miRNAs was further assessed through multivariate Cox regression analysis. Kaplan Meier analyses were conducted as above and prognostic miRNAs were chosen for subsequent evaluation.

Prognostic Analyses

Clinical data, involving age, gender, and Tumor stage, were collected from the TCGA dataset. Univariate and multivariate Cox regression models were carried over into analyze the prognostic value of these variables and the risk scores generated above. Cox regression analysis have been adopted to display prognostic nomogram afterwards.

Evaluation of Differential Genes

The DEGs of carcinoma of colon were identified associated with risk score and immune score. Gene ontology (GO), Kyoto Encyclopedia of Genes and Genomes (KEGG) and Disease ontology enrichment analyses were conducted using the R software.

Immune Correlation Analysis

Based on the single-sample gene set enrichment analysis (ssGSEA), we made further efforts to compare the enrichment performance of 16 types of immune cells and the activity of 13 immune-associated pathways between the low and high-risk groups in TCGA cohort. We explored the correlation analysis between immune cells and immune function. Moreover, we found that the expression of significantly different genes after screening was correlated with the expression of immune cell activity and immune function.

Results

Differentially Expressed m7G Methylation-Related miRNAs in Carcinoma of Colon

Comparing the 457 tumor tissue samples about the expression of 792 m7G methylation-associated miRNAs with the 8 normal healthy samples, we figured out the differential m7G methylation-associated miRNAs gene expression, Including up-regulated miRNA and down-regulated miRNA. Significantly, there are 14 miRNAs(hsa-miR-194-3p, hsa-miR-21-3p, hsa-miR-887-5p, hsa-miR-200c-5p, hsa-miR-221-3p, hsa-miR-193a-5p, hsa-miR-150-3p, hsa-miR-361-3p, hsa-miR-9-5p, hsa-miR-149-3p, hsa-miR-216a-5p, hsa-miR-378d, hsa-miR-628-3p, hsa-miR-31-5p) distinctively expressed $P \leq 0.05$, hsa-miR-216a-5p and hsa-miR-9-5p ($P \leq 0.001$). The expressions of hsa-miR-21-3p, hsa-miR-200c-5p, hsa-miR-221-3p, hsa-miR-9-5p, hsa-miR-216a-5p, hsa-miR-378d, hsa-miR-628-3p and hsa-miR-31-5p were increased in tumor samples compared to normal healthy samples. Univariate regression analyses showed that eleven of these m7G methylation-associated miRNAs were significantly correlated with lower OS ($HR > 1$) (Figure 1A). By performing the least absolute shrinkage and selection operator (LASSO) Cox

regression analysis, a 12- gene signature was constructed according to the optimum λ value (Figure 1B, Figure 1C, Figure 1D). Cox model revealed and constructed 12 m7G methylation-associated miRNAs signature.

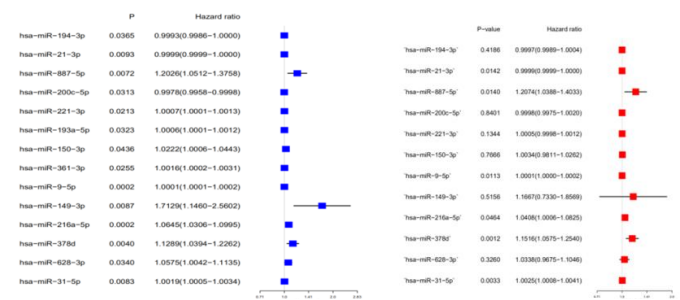


Figure 1A

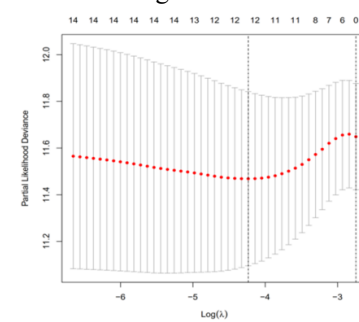


Figure 1C

Figure 1B

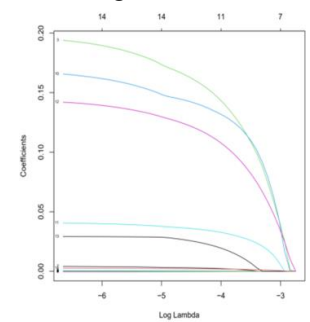


Figure 1D

Risk-Related miRNA Identification

In consideration of the potential prognostic capability of some m7G methylation-associated miRNAs, experiments were developed to identify the risk-related miRNAs that may be capable of predicting outcomes in carcinoma of colon patients. Through the analysis of Kaplan-Meier curve outcomes, we can draw a conclusion (Figure 2A to Figure 2F) that depressed expression of hsa-miR-21-3p might be significantly relevant with decreased survival of patients.

Meanwhile, hsa-miR-31-5p, hsa-miR-216a-5p, hsa-miR-221-3p, hsa-miR-378d and hsa-miR-887-5p may act a comparatively disadvantageous role in the context of carcinoma of colon-related m7G methylation such that patients who express high-caliber of these miRNAs are likely to have a disposition to inferior oncologic outcomes.

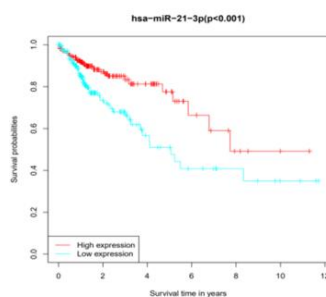


Figure 2A

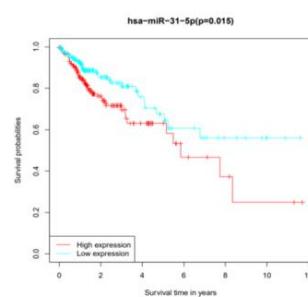


Figure 2B

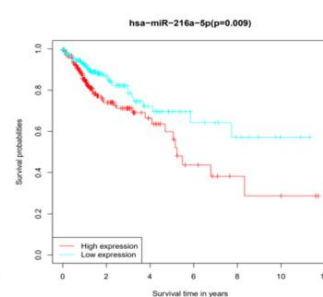


Figure 2C

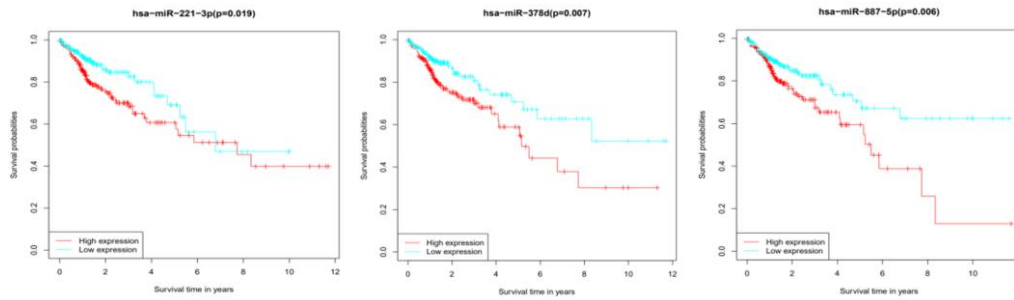


Figure 2D

Figure 2E

Figure 2F

Formation of the Prognostic m7G Methylation-associated miRNA Model

According to the median risk score, we divided carcinoma of colon patients into low and high-risk cohorts. From analysis, we discovered High-risk patients following significantly poorer OS, and the difference between low and high-risk cohorts may be greater probability increased as time goes. The ROC curve revealed that the AUC values for the 1, 3 and 5-year survival rates were 0.733, 0.742 and 0.706 respectively (Figure 3A, Figure 3B).

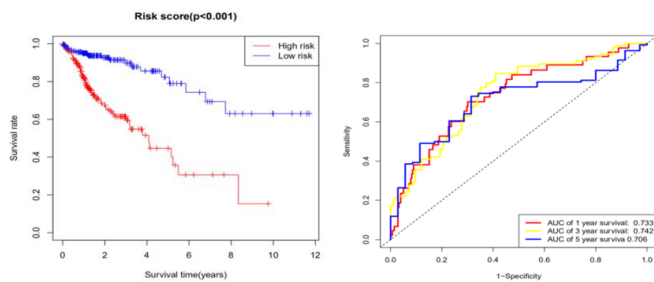


Figure 3A

Figure 3B

Univariate and Multivariate Analyses of the Risk Model

From the results of univariate and multivariate regression analysis, it can be concluded that the risk score has independent predictability in the prognosis of patients. In Cox regression analyses, grade and risk scores were associated with carcinoma of colon patient prognosis. Especially, grade and risk scores were independent predictors of prognosis in carcinoma of colon patients (HR =2.0715, 95% CI: 1.5926 to 2.6944; HR=3.0020, 95% CI: 1.7995 to 5.0080, respectively) (Figure 4A, Figure 4B).

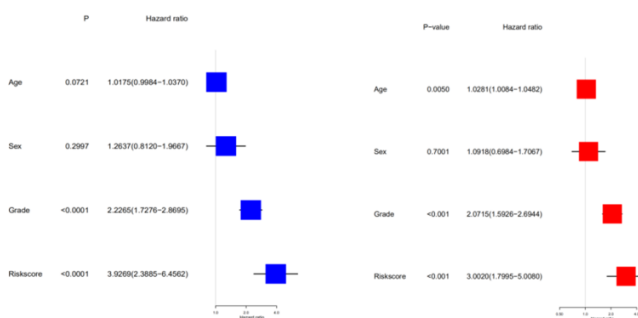


Figure 4A

Figure 4B

Development of the m7G Methylation-Associated miRNA Nomogram Models

Nomograms (Figure 5A) for OS at 1-, 3-, and 5-year were conducted based on age, sex, pathologic grade, and risk scores. The intersection of the vertical line of each scoring item and the corresponding scoring point axis is the score of the item. The total score of the variable is obtained by adding and summarizing the scores of each item. Depending on the total score, we obtained that the 1-, 3- and 5-year survival rates of patients with carcinoma of colon were 0.961, 0.907 and 0.851, respectively. Through the drawing of calibration chart, we explained that the predicted survival time of nomogram is highly consistent with the prognosis of actual patients (Figure 5B).

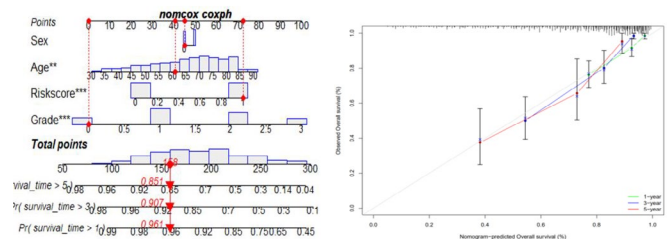


Figure 5A

Figure 5B

Evaluation of differential genes

DEGs of carcinoma of colon were screened separately according to risk score and immune scor (downloaded from <https://bioinformatics.mdanderson.org/estimate/disease.html/>), then we found the common differential genes (295 DEGs) from the two groups of DEGs (Figure 6A). GO enrichment analyses (Figure 6B) of these DEGs revealed that they were enriched in biological processes (BPs) including keratinocyte differentiation, extracellular matrix organization, and extracellular structure organization? They were also enriched for molecular function terms (MFs) including hormone activity, receptor ligand activity and signaling receptor activator activity, as well as cellular component terms (CCs) including collagen-containing extracellular matrix, collagen trimer, certified envelope, myosin filament, and synaptic membrane. KEGG pathway enrichment analyses (Figure 6C) indicated that these DEGs were enriched in the Neuroactive ligand-receptor interaction, Cell adhesion molecules, and Protein digestion and absorption pathways. From the enrichment analysis of diseases according to DEGs (Disease ontology) (Figure 6D), we can know that eating disorder, genetic diabetes, obesity, metabolic syndrome X, amenorrhea and over nutrition are associated with carcinoma of colon.

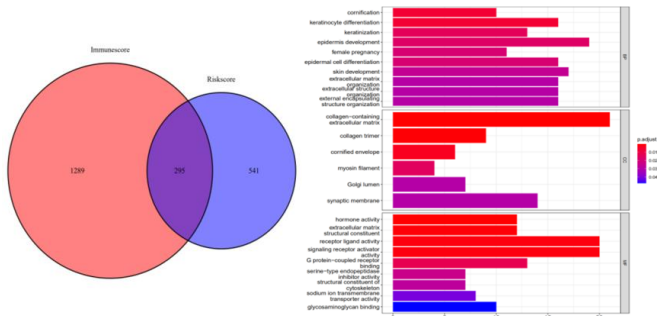


Figure 6A

Figure 6B

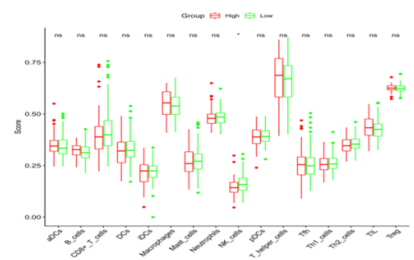


Figure 7A

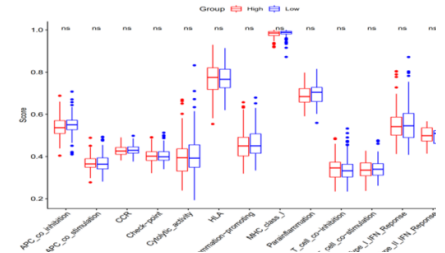


Figure 7B

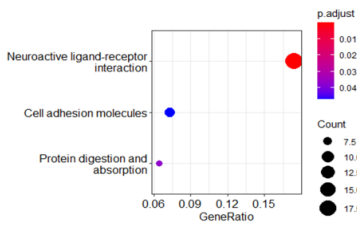


Figure 6C

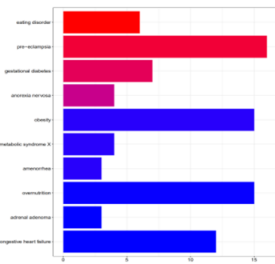


Figure 6D

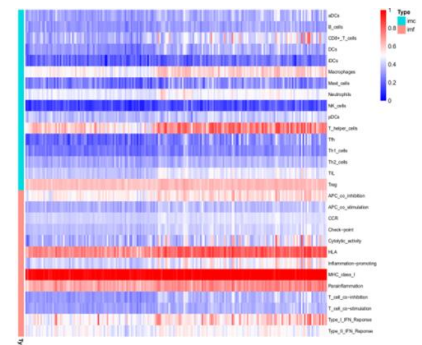


Figure 7C

Correlation Analysis of Immune Activity

Based on the single-sample gene set enrichment analysis (ssGSEA), we made further efforts to compare the enrichment performance of 16 types of immune cells and the activity of 13 immune-associated pathways between the low and high-risk groups in TCGA cohort (Figure7A, Figure7B). Taking it by and large, the low-risk subgroup had higher levels of infiltration of immune cells, particularly natural killer (NK) cells, than the high-risk subgroup. Through analysis, the Heat map of immunocyte and immunological functioning were drawn (Figure7C). Meanwhile, the correlation analysis of immune cells and immune function was further visualized (Figure7D, Figure7E). When assessing the Correlation analysis of immune cells, some conclusions were drawn. For example, T_helper_cells and Macrophages are highly correlated. The stronger the correlation between the two immune cells, it means that there is an obvious interaction between them, which may play a synergistic role in the occurrence and development of colon cancer. In addition, we discovered that Tfh/CD8+ T_cells, TIL/DCs, TIL/Macrophages, Treg/Neutrophils have a high correlation second only to T_helper_cells and Macrophages. When assessing the Correlation analysis of immunological functioning in the TCGA cohort, T_cell_co_inhibition and Check-point represented a high-correlation relationship.

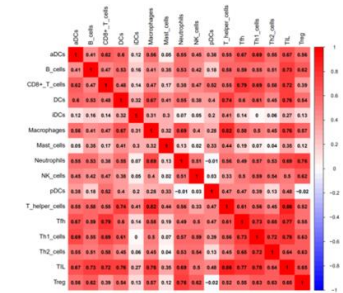


Figure 7D

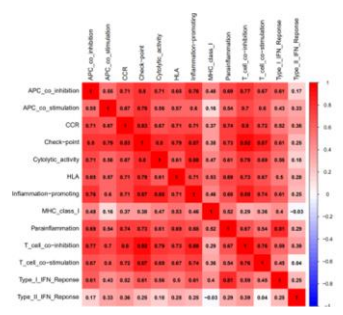


Figure 7E

Association between DEGs and Immune Cells with Immune Function

Analysis according to risk score and immune score, we acquired two group of DEGs, and then we found the significant common differential genes from the two groups of differential genes. We screened the DEGs related to prognosis by univariate Cox regression analysis. By utilizing the differential expressed genes, which we acquired from the above analysis, we further analyze their internal relationship with immune cells and immune function (Figure8). By analyzing the results in the diagram, we conclude that there is a significant positive correlation between MUCL3 and Type_II_IFN_Response and a significant negative correlation between CALML5 and Treg.

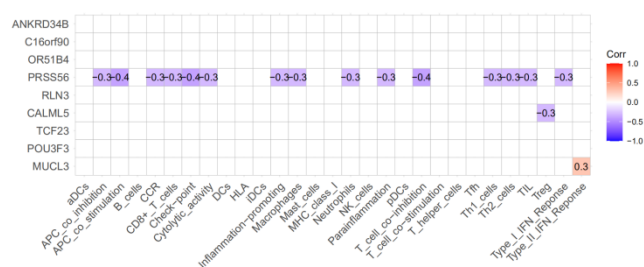


Figure 8

Discussion

As people's dietary structure changes. The incidence of digestive system, diseases increased significantly [31]. Carcinoma of colon is one of the most common type of malignant tumor in gastrointestinal digestive system [32]. The clinical characteristic of carcinoma of colon is recurrence and distant metastasis [33]. Scientists have verified that tumors have obvious genetic correlation [34, 35]. Hence, one can see that our clinical research focuses on the prognostic analysis from the perspective of gene transcription and visualizes it. As we all knows, RNA modification have significant impact in the biological processes of gene transcription and expression regulation [36]. It is predicted that the up-regulation and down-regulation of upstream miRNA may play a functional affection in the occurrence and development of carcinoma of colon. Therefore, m7G methylation-associated genes were selected as the main research perspectives in this study, establishing a novel m7G methylation-associated miRNA risk signature for prognostic value in-patient with carcinoma of colon.

Although it has been more than 60 years since RNA modification was first discovered, N7 methyl guanosine (m7G) remains one of the most common RNA modifications [37]. Some researchers have proved that RNA methyl transferase compound METTL1/WDR4 give impetus to the expression of m7G modification form in some cancers perform highly upward tendency [38]. Dai et al. Discovered that regardless of how degree of overexpression about METTL1/WDR4 and m7G tRNA modifications were associated with poor survival in human intrahepatic cholangiocarcinomas (ICCs).on the other hand, Orellana et al [39, 40]. Identified that merely overexpressing m7G-tRNA Arg-TCT-4-1 under the circumstances that there was no change in the

expression level of METTL1/WDR4, still achieve the process of gradually transforming normal cells into malignant transformation. These results demonstrate convincingly that translational regulation was not just as a starting factor or participant in ontogenesis which contraried to popular belief. We come out a refreshing molecular perspective that m7G-tRNA modifications can accelerate the transformation of normal cell-model into cancer cell-model.

This current study evaluated the expression of m7G methylation-associated miRNAs and demonstrated that 63 miRNAs were upregulated in tissues (carcinoma of colon) compared with controlled samples and 45 miRNAs were downregulated. In addition, through univariate regression analysis, there was a significant positive correlation among the 14 miRNAs, suggesting that these may work together to regulate m7G methylation in carcinoma of colon. A risk score and prognostic model based on 12 miRNAs was constructed and shown to accurately predict the 1-, three-and 5-year OS in patients with carcinoma of colon. These risk scores, together with tumor stage, were found to be independently associated with patient prognosis. Our novel m7G methylation-associated miRNA model may represent a valuable tool for predicting the survival rate and neoplasm staging in patients with carcinoma of colon.

For the sake of evaluating the prognostic value of m7G methylation-associated miRNAs, univariate regression analyses were performed. The 14 miRNAs were correlated with the prognosis of patients with carcinoma of colon. However, other researchers have reported that upregulation of hsa-miR-194-3p was associated with the early T-stages [41]. Genes Hen G, etl found that hsa-miR-200c-5p was key regulator in HCV-HCC development by analyzing transcriptome-sequencing data [42, 43]. Up-regulation of hsa-miR-221-3p affects proliferation and apoptosis of cells via Bcl-xL/Bax pathway. In line with our findings, some reaches determined that the dysregulation of lncRNAs, seclud-ed altered the expression of several miRNAs, such as hsa-miR-193a-5p and affected the occurrence and development of colon cancer.MiR-361-3p, low expression in CRC, was validated to be targeted and negatively modified by BBOX1-AS1 [44, 45]. A research revealed that some miRNAs may be new markers for the diagnosis of RCC, including hsa-mir-92a-1-5p, hsa-mir-149-3p and hsa-mir-424-3p, because of these miRNAs significantly abnormal expressed in RCC patients [46]. In patients with early stage carcinoma of colon, miR-378 can be examined as a biomarker of colorectal cancer, thus, miR-378 shows advantages in early diagnosis and treatment [47].The expression of hsa-mir-31-5p increased with tumor progression [48].

Our study confirmed that risk signatures could effectively classify colon cancer patients into subgroups with different risk levels. The risk signatures were combined with clinical factors to construct a new nomogram to predict the prognosis of patients with carcinoma of colon. Applying nomograms to predict clinical outcomes was effective, confirming by calibration curve analysis. Thus, this current study constructed models, which can significantly predict the clinical outcomes, such as OS in patients with carcinoma of colon.

We screened the differential genes of carcinoma of colon according to the immune score and risk score of colon cancer, and searched for the common DEGs in the results of score analysis of the two groups. The common differentially expressed genes were utilized for functional analyses and KEGG pathway analyses.

In KEGG pathway analyses, these common DEGs were found to be enriched in the Neuroactive ligand-receptor interaction, cell adhesion molecules and protein digestion and absorption pathways. There is a strong possibility that the Neuroactive ligand-receptor interaction pathway is a key regulator of metastatic progression and chemo resistance in colorectal cancer [49]. The neuroactive ligand receptor interaction pathway may be participated in the regulation of tumor mutational burden (TMB) characterized by microsatellite instability. Since TMB has become a impactful predictor of tumor transformation and immunotherapeutic response, the regulatory pathway of neuroactive ligand receptor interaction may exert an meaningful affection.

In conclusion, through the analysis of the research results, we infer that there is a correlation between the occurrence and development of colon cancer and m7G methylation. There are some limitations in the overall design and result analysis of the research, because of lacking other data sets similarly analyzed to validate our findings. Moreover, if possible in the future, relevant in experimental research can also be used to carry out and support theoretical analysis and clinical experimental results.

This investigation identified new m7G methylated associated miRNA markers capable of forecasting the survival rate of patients with carcinoma of colon. The results obtained from this study may have an impact on the treatment plan of colon cancer patients, and may even guide the chemotherapy plan of colon cancer patients in the future.

References

- Huang, Y., Zou, Y., Xiong, Q., Zhang, C., Sayagués, J. M., Shelat, V. G., & Wang, X. (2021). Development of a novel necroptosis-associated miRNA risk signature to evaluate the prognosis of colon cancer patients. *Annals of translational medicine*, 9 (24).
- Araghi, M., Soerjomataram, I., Jenkins, M., Brierley, J., Morris, E., Bray, F., & Arnold, M. (2019). Global trends in colorectal cancer mortality: projections to the year 2035. *International journal of cancer*, 144(12), 2992-3000.
- Pan, P., Yu, J., & Wang, L. S. (2019). Diet and colon: what matters?. *Current opinion in gastroenterology*, 35(2), 101.
- Wang, P., Berzin, T. M., Brown, J. R. G., Bharadwaj, S., Becq, A., Xiao, X., & Liu, X. (2019). Real-time automatic detection system increases colonoscopic polyp and adenoma detection rates: a prospective randomised controlled study. *Gut*, 68(10), 1813-1819.
- Singhal, R., Mitta, S. R., Das, N. K., Kerk, S. A., Sajjakulnukit, P., Solanki, S., & Shah, Y. M. (2021). HIF-2 α activation potentiates oxidative cell death in colorectal cancers by increasing cellular iron. *The Journal of clinical investigation*, 131(12).
- Ye, S., Xu, M., Zhu, T., Chen, J., Shi, S., Jiang, H., & Xi, Y. (2021). Cytoglobin promotes sensitivity to ferroptosis by regulating p53-YAP1 axis in colon cancer cells. *Journal of cellular and molecular medicine*, 25(7), 3300-3311.
- Yu, S., Hou, D., Chen, P., Zhang, Q., Lv, B., Ma, Y., & Liu, J. (2015). Adenosine induces apoptosis through TNFR1/RIPK1/P38 axis in colon cancer cells. *Biochemical and biophysical research communications*, 460(3), 759-765.
- Yu, J., Li, S., Qi, J., Chen, Z., Wu, Y., Guo, J., & Zheng, J. (2019). Cleavage of GSDME by caspase-3 determines lobaplatin-induced pyroptosis in colon cancer cells. *Cell death & disease*, 10(3), 1-20.
- Xiao, Y., Zhang, T., Ma, X., Yang, Q. C., Yang, L. L., Yang, S. C., & Sun, Z. J. (2021). Microenvironment-Responsive Prodrug-Induced Pyroptosis Boosts Cancer Immunotherapy. *Advanced Science*, 8(24), 2101840.
- Sharma, A., Kim, E. J., Shi, H., Lee, J. Y., Chung, B. G., & Kim, J. S. (2018). Development of a theranostic prodrug for colon cancer therapy by combining ligand-targeted delivery and enzyme-stimulated activation. *Biomaterials*, 155, 145-151.
- Stewart, C. L., Warner, S., Ito, K., Raoof, M., Wu, G. X., Lu, W. P., & Fong, Y. (2018). Cyto-reduction for colorectal metastases: liver, lung, peritoneum, lymph nodes, bone, brain. When does it palliate, prolong survival, and potentially cure?. *Current problems in surgery*, 55(9), 330.
- Law, P. P., & Holland, M. L. (2019). DNA methylation at the crossroads of gene and environment interactions. *Essays in biochemistry*, 63(6), 717-726.
- Nishiyama, A., & Nakanishi, M. (2021). Navigating the DNA methylation landscape of cancer. *Trends in Genetics*, 37(11), 1012-1027.
- Bogdanović, O., & Lister, R. (2017). DNA methylation and the preservation of cell identity. *Current opinion in genetics & development*, 46, 9-14.
- Xie, S., Chen, W., Chen, K., Chang, Y., Yang, F., Lin, A., & Yan, X. (2020). Emerging roles of RNA methylation in gastrointestinal cancers. *Cancer cell international*, 20(1), 1-11.
- Héberlé, É., & Bardet, A. F. (2019). Sensitivity of transcription factors to DNA methylation. *Essays in biochemistry*, 63(6), 727-741.
- Park JW, Han JW. Targeting epigenetics for cancer therapy. *Arch Pharm Res*. 2019 Feb; 42(2):159-170.
- Zappe, K., & Cichna-Markl, M. (2020). Aberrant DNA methylation of ABC transporters in cancer. *Cells*, 9(10), 2281.
- Chen, J., Li, K., Chen, J., Wang, X., Ling, R., Cheng, M., & Chen, D. (2022). Aberrant translation regulated by METTL1/WDR4-mediated tRNA N7-methylguanosine modification drives head and neck squamous cell carcinoma progression. *Cancer Communications*, 42(3), 223-244.
- Pandolfini, L., Barbieri, I., Bannister, A. J., Hendrick, A., Andrews, B., Webster, N., & Kouzarides, T. (2019). METTL1 promotes let-7 MicroRNA processing via m7G methylation. *Molecular cell*, 74(6), 1278-1290.
- Liu, Y., Yang, C., Zhao, Y., Chi, Q., Wang, Z., & Sun, B. (2019). Overexpressed methyltransferase-like 1 (METTL1) increased chemosensitivity of colon cancer cells to cisplatin by regulating miR-149-3p/S100A4/p53 axis. *Aging (Albany NY)*, 11(24), 12328.
- Ma, J., Han, H., Huang, Y., Yang, C., Zheng, S., Cai, T., & Lin, S. (2021). METTL1/WDR4-mediated m7G tRNA modifications and m7G codon usage promote mRNA translation and lung cancer progression. *Molecular Therapy*, 29(12), 3422-3435.

23. Ghafouri-Fard, S., Hussien, B. M., Badrlou, E., Abak, A., & Taheri, M. (2021). MicroRNAs as important contributors in the pathogenesis of colorectal cancer. *Biomedicine & Pharmacotherapy*, 140, 111759.
24. Wang, H. (2020). MicroRNAs and apoptosis in colorectal cancer. *International journal of molecular sciences*, 21(15), 5353.
25. Mei, H., & Wen, Y. (2021). MicroRNAs for diagnosis and treatment of colorectal cancer. *Endocrine, Metabolic & Immune Disorders-Drug Targets (Formerly Current Drug Targets-Immune, Endocrine & Metabolic Disorders)*, 21(1), 47-55.
26. Xu, J., Meng, Q., Li, X., Yang, H., Xu, J., Gao, N., & Chen, R. (2019). Long Noncoding RNA MIR17HG Promotes Colorectal Cancer Progression via miR-17-5pMIR17HG Role in Colorectal Cancer. *Cancer research*, 79(19), 4882-4895.
27. Shirafkan, N., Mansoori, B., Mohammadi, A., Shomali, N., Ghasbi, M., & Baradaran, B. (2018). MicroRNAs as novel biomarkers for colorectal cancer: New outlooks. *Biomedicine & Pharmacotherapy*, 97, 1319-1330.
28. Moridikia, A., Mirzaei, H., Sahebkar, A., & Salimian, J. (2018). MicroRNAs: Potential candidates for diagnosis and treatment of colorectal cancer. *Journal of cellular physiology*, 233(2), 901-913.
29. Hernandez, R., Sanchez-Jimenez, E., Melguizo, C., Prados, J., & Rama, A. R. (2018). Downregulated microRNAs in the colorectal cancer: diagnostic and therapeutic perspectives. *BMB reports*, 51(11), 563.
30. Zhang, Y., & Wang, J. (2017). MicroRNAs are important regulators of drug resistance in colorectal cancer. *Biological chemistry*, 398(8), 929-938.
31. Cao, W., Chen, H. D., Yu, Y. W., Li, N., & Chen, W. Q. (2021). Changing profiles of cancer burden worldwide and in China: a secondary analysis of the global cancer statistics 2020. *Chinese Medical Journal*, 134(07), 783-791.
32. Zhao, Z., & Qin, X. (2020). MicroRNA-708 targeting ZNF549 regulates colon adenocarcinoma development through PI3K/Akt pathway. *Scientific reports*, 10(1), 1-9.
33. Zhang, Y., Zhou, Y. M., Zhang, Z. J., & Li, X. (2020). miR-210 is a serological biomarker for predicting recurrence and prognosis of colon carcinoma patients with liver metastases after radiofrequency ablation treatment. *Cancer Management and Research*, 12, 9077.
34. Dhooge, M., Baert-Desurmont, S., Corsini, C., Caron, O., Andrieu, N., Berthet, P., & Nagues, C. (2020). National recommendations of the French Genetics and Cancer Group-Unicancer on the modalities of multi-genes panel analyses in hereditary predispositions to tumors of the digestive tract. *European Journal of Medical Genetics*, 63(12), 104080.
35. Turpin, A., Cattan, S., Leclerc, J., Wacrenier, A., Manouvrier-Hanu, S., Buisine, M. P., & Lejeune-Dumoulin, S. (2014). Prédilection héréditaire aux cancers digestifs, mammaires, gynécologiques et gonadiques: état des lieux du syndrome de Peutz-Jeghers. *Bulletin du cancer*, 101(9), 813-822.
36. Zhang, X., Zhang, S., Yan, X., Shan, Y., Liu, L., Zhou, J., & Lai, W. (2021). m6A regulator-mediated RNA methylation modification patterns are involved in immune microenvironment regulation of periodontitis. *Journal of cellular and molecular medicine*, 25(7), 3634-3645.
37. Zou, H., Yang, F., & Yin, Z. (2022). Identifying N7-methylguanosine sites by integrating multiple features. *Biopolymers*, 113(2), e23480.
38. Ma, J., Han, H., Huang, Y., Yang, C., Zheng, S., Cai, T., & Lin, S. (2021). METTL1/WDR4-mediated m7G tRNA modifications and m7G codon usage promote mRNA translation and lung cancer progression. *Molecular Therapy*, 29(12), 3422-3435.
39. Dai, Z., Liu, H., Liao, J., Huang, C., Ren, X., Zhu, W., & Kuang, M. (2021). N7-Methylguanosine tRNA modification enhances oncogenic mRNA translation and promotes intrahepatic cholangiocarcinoma progression. *Molecular cell*, 81(16), 3339-3355.
40. Orellana, E. A., Liu, Q., Yankova, E., Pirouz, M., De Braekeleer, E., Zhang, W., & Gregory, R. I. (2021). METTL1-mediated m7G modification of Arg-TCT tRNA drives oncogenic transformation. *Molecular Cell*, 81(16), 3323-3338.
41. Wang, Y., Huang, L., Shan, N., Ma, H., Lu, S., Chen, X., & Long, H. (2021). Establishing a three-miRNA signature as a prognostic model for colorectal cancer through bioinformatics analysis. *Aging (Albany NY)*, 13(15), 19894.
42. Amirfallah, A., Knutsdottir, H., Arason, A., Hilmarsdottir, B., Johannsson, O. T., Agnarsson, B. A., & Reynisdottir, I. (2021). Hsa-miR-21-3p associates with breast cancer patient survival and targets genes in tumor suppressive pathways. *PloS one*, 16(11), e0260327.
43. Chen, G., Zhang, W., & Ben, Y. (2021). Identification of Key Regulators of Hepatitis C Virus-Induced Hepatocellular Carcinoma by Integrating Whole-Genome and Transcriptome Sequencing Data. *Frontiers in genetics*, 12.
44. Tu, Y., Wu, W., Guo, Y., Lu, F., Li, X., Xu, D., & He, L. (2021). Up-regulation of hsa-miR-221-3p induced by UVB affects proliferation and apoptosis of keratinocytes via Bcl-xL/Bax pathway. *Photodermatology, Photoimmunology & Photomedicine*, 37(4), 269-277.
45. Liu, J., Zhu, J., Xiao, Z., Wang, X., & Luo, J. (2022). BBOX1-AS1 contributes to colorectal cancer progression by sponging hsa-miR-361-3p and targeting SH2B1. *FEBS Open Bio*, 12(5), 983-992.
46. Xiao, C. T., Lai, W. J., Zhu, W. A., & Wang, H. (2020). MicroRNA derived from circulating exosomes as noninvasive biomarkers for diagnosing renal cell carcinoma. *OncoTargets and therapy*, 13, 10765.
47. Gungormez, C., Gumushan Aktas, H., Dilsiz, N., & Borazan, E. (2019). Novel miRNAs as potential biomarkers in stage II colon cancer: microarray analysis. *Molecular biology reports*, 46(4), 4175-4183.
48. Slattery, M. L., Herrick, J. S., Mullany, L. E., Valeri, N., Stevens, J., Caan, B. J., & Wolff, R. K. (2015). An evaluation and replication of miRNA s with disease stage and colorectal cancer-specific mortality. *International journal of cancer*, 137(2), 428-438.
49. Chen, J., Apizi, A., Wang, L., Wu, G., Zhu, Z., Yao, H., & He, S. (2021). TCGA database analysis of the tumor mutation burden and its clinical significance in colon cancer. *Journal of Gastrointestinal Oncology*, 12(5), 2244.

Copyright: ©2022 Chongyang Wang. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.