Review Article

A Systematic Review of Clinical Validated and Potential miRNA Markers Related to the Efficacy of Fluoropyrimidine Drugs

Xiaomeng Sun,1 Jiani Chen,2 Xintao Chen,3 Qianmin Gao,3 Wei Chen,2 Xun Zou,2 Feng Zhang,2 Shouhong Gao,2 Shi Qiu,4 Xiaoqiang Yue,5 Houshan Yao,3 Xuan Liu,5 and Mingming Li2

1Institutes of Biomedical Sciences, Fudan University, No. 131, Dongan Rd., Shanghai 200032, China
2Department of Pharmacy, Changzheng Hospital, Naval Medical University, Shanghai 200003, China
3Department of General Surgery, Changzheng Hospital, Naval Medical University, Shanghai 200003, China
4Traditional Chinese Medicine Resource and Technology Center, Shanghai University of Traditional Chinese Medicine, Shanghai 201203, China
5Department of TCM, Changzheng Hospital, Naval Medical University, Shanghai 200003, China

Correspondence should be addressed to Houshan Yao; 58853993@qq.com, Xuan Liu; 18616630816@163.com, and Mingming Li; limingming@smmu.edu.cn

Received 31 March 2022; Revised 15 July 2022; Accepted 29 July 2022; Published 23 August 2022

Academic Editor: Zhijie Xu

Copyright © 2022 Xiaomeng Sun et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Colorectal cancer (CRC) is becoming increasingly prevalent worldwide. Fluoropyrimidine drugs are the primary chemotherapy regimens in routine clinical practice of CRC. However, the survival rate of patients on fluoropyrimidine-based chemotherapy varies significantly among individuals. Biomarkers of fluoropyrimidine drugs’ efficacy are needed to implement personalized medicine. This review summarized fluoropyrimidine drug-related microRNA (miRNA) by affecting metabolic enzymes or showing the relevance of drug efficacy. We first outlined 42 miRNAs that may affect the metabolism of fluoropyrimidine drugs. Subsequently, we filtered another 41 miRNAs related to the efficacy of fluoropyrimidine drugs based on clinical trials. Bioinformatics analysis showed that most well-established miRNA biomarkers were significantly enriched in the cancer pathways instead of the fluoropyrimidine drug metabolism pathways. The result also suggests that the miRNAs screened from metastasis patients have a more critical role in cancer development than those from non-metastasis patients. There are five miRNAs shared between these two lists. The miR-21, miR-215, and miR-218 can suppress fluoropyrimidine drugs’ catabolism. The miR-326 and miR-328 can reduce the efflux of fluoropyrimidine drugs. These five miRNAs could jointly act by increasing intracellular levels of fluoropyrimidine drugs’ cytotoxic metabolites, leading to better chemotherapy responses. In conclusion, we demonstrated that the dynamic changes in the transcriptional regulation via miRNAs might play significant roles in the efficacy and toxicity of the fluoropyrimidine drug. The reported miRNA biomarkers would help evaluate the efficacy of fluoropyrimidine drug-based chemotherapy and improve the prognosis of colorectal cancer patients.

1. Introduction

Colorectal cancer (CRC) is the third most commonly diagnosed malignancy and the second leading cause of cancer death worldwide [1]. The global burden of CRC is expected to increase to more than 2.5 million new cases in 2035 [1]. The incidence of CRC in developed countries, such as in Europe and North America, has stabilized and declined. In contrast, the incidence of CRC in developing countries is still on the rise [2], especially in China [3]. It is estimated that 4.3 million new cancer cases and 2.9 million new cancer deaths occurred in China in 2018 [2], meaning 30% and 40% higher cancer incidence and mortality than in the UK and USA [2]. Fluoropyrimidine-based (5-fluorouracil/5-FU, Capecitabine, Tegafur) drugs are the most commonly used chemotherapeutic agents in CRC treatment. They significantly improved the survival rates of CRC patients [4, 5]. However, many CRC patients will still experience recurrences or develop...
advanced diseases. The mutations in coding genes [6] and microRNAs (miRNAs) [7] were related to individual heterogeneity of drug efficacy and toxicity. Personalized therapy has been proved to aid clinicians in improving the treatment outcome [6]. The essential requirement is reliable marker systems with solid clinical evidence or precise molecular mechanisms.

The enzyme thymidylate synthase (TS) can methylate deoxyuridine monophosphate (dUMP) to form deoxythymidine monophosphate (dTMP). 5-FU acts as an inhibitor of TS by reducing the dTMP formation and ultimately blocking the formation of thymidine, an essential nucleoside for DNA replication and repair. Administration of 5-FU can rapidly induce cancer cell death via lack of thymidine [8]. The pharmacokinetics or pharmacodynamics of fluoropyrimidine drugs and their reactive metabolites are closely related to their efficacy [9]. These are controlled by enzymes and compounds that participate in drug transportation and metabolism. For instance, the calcium folinate can enhance 5-FU’s cytotoxicity by providing exogenous folinate, stabilizing the 5-FU-TS complex [10].

Much effort has been made to screen for biomarkers that can affect or measure the activity of these effectors to predict chemotherapy response. MiRNAs can act as gene regulators that affect the tumor cell life cycle, such as growth, differentiation, and apoptosis. MiRNAs are small noncoding RNAs containing 21-24 nucleotides that play essential posttranscriptional regulatory roles in diverse organisms, including humans [11]. They affect protein function by combining with semi-complementary target miRNAs, resulting in mRNA destabilization and translation repression [12]. After the discovery of miRNA in the early-90s of twentieth century, the roles of miRNA in the pharmacology of standard chemotherapy drugs such as 5-FU were studied. Lorena Rossi and colleagues published one of the earliest studies in 2007 [13]. They proved that 5-FU could alter miRNA expression in malignant cells profoundly. These miRNAs include two of the most important miRNA markers, miR-21 and miR-200b, whose functions have been validated in many following observational studies since then. In the second decade of the twenty-first century, huge progress has been made to screen miRNA makers related to the efficacy or safety of 5-FU and 5-FU-based drugs. And a significant portion of these works was completed by European researchers such as Torben Frøstrup Hansen, who linked another important miRNA biomarker, miR126, with 5-FU’s efficacy from a randomized phase III study with A total of 230 patients [14]. However, most therapy-associated miRNA biomarkers are deduced from molecular biological mechanisms based on cellular models, which leaves them lacking clinical validation. Besides, a single biomarker also lacks sensitivity and specificity. A miRNA panel could help increase the predictive efficiency [15].

Chen Lab is one of the pioneers in utilizing computational algorithms to discover small molecule drug-miRNA associations in batches. Dual-network collaborative matrix factorization (DCMF) [16], Ensemble of kernel ridge regression-based small molecule-miRNA association prediction (EKRRSMMA) [17], and bounded nuclear norm regularization for small molecule-miRNA associations prediction (BNNRSMMA) [18] methods were developed to fulfill the need, contributing to the miRNA panel discovery and validation efforts. Based on these sophisticated and subtle studies, to acquire a promising miRNA biomarker panel for predicting the efficacy of fluoropyrimidine drugs, each potential miRNA biomarker should have a clear biological function or should have been validated by retrospective or observational clinical trials. In this study, we reviewed miRNAs that may affect fluoropyrimidine drugs’ metabolism and miRNAs that are associated with the response to fluoropyrimidine therapy. Five miRNAs meet both criteria, and their biological roles are further discussed.

2. Materials and Methods

2.1. Searching Strategy. We searched research articles on enzymes affected by miRNAs in the fluoropyrimidine drug metabolic pathway by the following strategy. Online databases including Embase, PubMed, Google Scholar, and Science Direct (updated to Sep 1, 2019) were searched [19]. We explicitly designed the searching strategy with the following terms: (“TS” OR “thymidylate synthase”) AND (“miRNA” OR “microRNA” OR “miR-“ OR “microRNAs” OR “miRNAs”). Table 1 shows a complete list of search keywords used in this article. Each result was recorded with title, date of publication, author list, and abstract for finer screening as described below.

Furthermore, we searched clinical trial outcomes related to the efficacy of fluoropyrimidine drugs according to the established paradigm of the Cochrane framework [20]. Online databases including Embase, PubMed, Google Scholar, and Science Direct (updated to Sep 1, 2019) were searched. A predefined searching strategy that outlined and combined the following terms was designed for this review: (a) in the title: (“colorectal” OR “colon” OR “rectum”) AND (“cancer” OR “tumor” OR “tumour” OR “Carcinoma” OR “neoplasia”) AND (“miRNA” OR “microRNA” OR “miR-“ OR “microRNAs” OR “miRNAs”). Relevant reviews or articles on the citation list were independently screened to ensure completeness. Duplicate studies were combined before proceeding to the next round of filtering. b) Considering the most common validation for studies focusing on finding miRNAs to predict better chemotherapy outcomes is whether the potential marker could produce a good ROC curve with reasonable sensitivity and specificity based on regression analysis. We used the following keywords in Mendeley software to locate the publications with detailed efficiency assessments: (“survival” OR “response” OR “os” OR “PFS” OR “side” OR “adverse” OR “toxic” OR “effectiveness” OR “prognosis” OR “diagnosis” OR “diagnostic value” OR “detection” OR “biomarker” OR “sensitivity AND specificity” OR “ROC curve”) AND (“chemotherapy” OR “5-fu” OR “capecitabine” OR “fluoropyrimidine” OR “fluorouracil” OR “FOLFOX” OR “XELOX”) AND (patients). We summarized the search keywords and their representing categories in Table 1.
2.2 Filtering Strategy. The initial searched studies were further filtered based on the following criteria: (a) clinical research should base on humans; (b) all included patients should have had chemotherapy with fluoropyrimidine drugs; and (c) differences in survival should be related to miRNA expressions. Besides, the following studies were discarded: (a) duplicate publication; (b) case reports, letters to the editor, or review articles; and (c) studies with unqualified or insufficient clinical data.

The flow chart of the entire literature filtering process is shown in Figure 1. The initial search returned 107 articles, of which five review articles were excluded. After carefully reviewing their titles and abstracts, 37 unrelated articles were excluded due to the lack of clinical data. The remaining 65 articles were available for further full-text manual check. Another 14 articles were excluded because the survival difference was unrelated to any miRNA expression. And patients from 16 articles did not receive chemotherapy with fluoropyrimidine drugs. In the end, there were 31 articles included in this review.

In the final 31 selected articles, candidate miRNAs and their corresponding target genes were summarized and enriched to the Kyoto Encyclopedia of Genes and Genomes (KEGG) pathways and Gene Ontology (GO) terms. The functional enrichment analysis was performed by the Database for Annotation, Visualization, and Integrated Discovery (DAVID) online tool [21, 22]. A number of matched genes larger than five and P values less than 0.05 (corrected by the two-side Bonferroni test) were considered significant.

3. Results

3.1. miRNAs Affecting the Metabolism of 5-FU. Based on the toxicity-related polymorphisms studies of fluoropyrimidines drugs proposed earlier [23–28], a metabolic pathway of 5-FU was drawn (Figure 2). We then searched for miRNAs that affect these metabolizing enzymes.

The cytotoxic metabolite of fluoropyrimidine drugs is fluorodeoxyuridine monophosphate (FdUMP), inhibiting the TS. 5-FU can be converted to FdUMP through different metabolic pathways (Figure 2). Thymidine phosphorylase (TP, or TYMP) controls one of the main metabolic pathways by converting 5-FU to fluorine de-oxidation pyridine (FUDR). FUDR can subsequently be converted to FdUMP catalyzed by thymidine kinase (TK). Another catalytic pathway converts the 5-FU to FdUMP through a more extended route, with a rate-limiting enzyme DPYD. The primary intermediate metabolites are fluorouridine diphosphate (FUDP) and fluorodeoxyuridine diphosphate (FdUDP), which could be further converted to fluorouridine triphosphate (FUTP) and fluorodeoxyuridine triphosphate (FdUTP). They can incorporate themselves into the newly formed DNA or RNA and suppress the normal replication and repair process. A previous study also showed that FUTP and FdUTP also contributed to the efficacy of fluoropyrimidine drugs [29].

We targeted 42 miRNAs that may affect the expression of enzymes within the fluoropyrimidine drug metabolic pathway (Table 2).
from the studies in Asia. The median number of patients is 84. Six studies derived their conclusion from plasma samples, 24 from tissue, and one from both.

The miRNA related to fluoropyrimidine drug efficacy were listed in two subgroups based on CRC stages. MiRNAs in the first group were screened from patients without metastasis CRC (I, II, or III). MiRNAs in the second group were screened from patients with metastasis CRC (IV). If available, we recorded the number of patients within each stage (I, II, III, or IV) in the bracket. We marked these miRNAs according to the types of tissue (AMT: adjacent mucosal tissues; LR: local recurrences; metastases; plasma; and PT: primary tumors). Each miRNA expression level was tagged by its relationship with adverse clinical outcomes (shorter PFS, OS, or DFS). The statistical significance of each miRNA was annotated as superscript: for log-rank test, *, <0.05; **, <0.01; ***, <0.001; and for COX test, #, <0.05; ##, <0.01; ###, <0.001. The origin of these studies was listed as the two-letter codes for countries and regions: AT, Austria; CN, China; DK, DENMARK; CZ, Czech; ES, Spain; FR, France; DE, Germany; JP, Japan; HK, Hong Kong; NL, Netherlands, NO, Norway; ES, Spain; NL, Netherlands; and PL, Poland.

The top 20 KEGG pathways based on fluoropyrimidine-efficacy-related miRNAs from the two subgroups (with metastasis and without metastasis) are listed in Table 4. Both enrichment results contain pathways directly related to the disease (including types of cancers). Many pathways regulate the normal cellular process, the abnormality of which might be the susceptible factor for cancer development. For miRNAs from the group without metastasis CRC, besides disease or cancer pathways, the FoxO signaling pathway, MAPK signaling pathway, autophagy, and PI3K-Akt signaling pathway are enriched, and their rank is within the top ten of all the enriched pathways. On the other hand, disease (especially cancer) pathways contribute the most to the miRNAs in the metastasis CRC cohort. The ranks of the FoxO signaling pathway and the MAPK signaling pathway were reduced from 2 to 4 and 4 to 17, respectively.

In addition, those fluoropyrimidine-efficacy-related miRNAs from the two subgroups were also subjected to GO term enrichment (Table 5). Based on the results, miRNAs from the two cohorts showed similar cellular component (CC) and molecular functions (MF) characteristics. For CC, they both have intracellular organelle, membrane-bounded organelle, and intracellular membrane-bounded organelle. For MF, they both have enzyme binding, regulatory region nucleic acid binding, and transcription regulatory region sequence-specific DNA binding. However, they showed quite a different biological process (BP). MiRNAs from the cohort without metastasis patients are mainly enriched in metabolic-related functions, such as regulating the cellular metabolic process, the primary metabolic process, and the nitrogen compound metabolic process. On the other hand, miRNA from the cohort with metastasis patients is mainly enriched in cell cycle control mechanisms, such as G1/S transition of mitotic cell cycle, mitotic cell cycle, cell morphogenesis, and cell morphogenesis involved in differentiation.

4. Discussion

This study first summarized a list of 42 miRNAs that may affect fluoropyrimidine drug metabolism based on literature research. Subsequently, we have created another list of 41 miRNAs related to fluoropyrimidine drugs’ efficacy based on clinical trials according to the Cochrane framework. By comparing the two sets, we found that miR-21, miR-215, miR-218, and miR-326, and miR-328 could affect the metabolic pathways of 5-FU and their expressions were associated with CRC survival after fluoropyrimidine adjuvant chemotherapy.

MiR21 is a marker for better efficacy of fluoropyrimidine drugs for CRC patients with and without metastasis. It can suppress the expression of dihydropyrimidine dehydrogenase (DYPD) and thymidine phosphorylase (TP) [30]. DYPD is a crucial enzyme in fluoropyrimidine drugs metabolism [31], which takes charge of the detoxifying process of 5-FU in the liver. A low DPD level can increase internal exposure to 5-FU and its cytotoxicity, resulting in better efficacy. On the other hand, Capecitabine is almost wholly absorbed in the gastrointestinal tract, metabolized to 5-deoxy 5-cytosine nucleoside (5'-DFCR), and finally converted into 5-FU by thymidine phosphorylase (TP) [32, 33]. A low TP level may reduce the catabolism of fluoropyrimidine drug, resulting in extended exposure of 5-FU and its cytotoxic intermediate metabolites. Furthermore, several pieces of literature from Czech [34], German [35], Japanese [36], and Chinese [37] have confirmed that the increased expression of miR-21 was significantly
correlated with good outcomes of adjuvant chemotherapy. Thus, miR-21 is a solid marker for using fluoropyrimidine drugs after CRC surgery.

For miR-215 and miR-218, they are positively related to better chemotherapy response in patients without metastasis. In fluoropyrimidine drug metabolism, they suppress the expression of thymidylate synthetase (TS) [37, 38]. TS is an enzyme that catalyzes the conversion of deoxyxuridine monophosphate (dUMP) to deoxythymidine monophosphate (dTMP) (Figure 2). The dTMP is the fundamental building material for DNA and RNA synthesis. Suppressed TS expression can cause the cells to be more sensitive to genotoxic stress, further activating programmed cell death pathways, resulting in DNA fragmentation [39]. MiR-215 and miR218 could make the tumor cells more sensitive to chemotherapy. Three clinical trials from different regions have indicated that induced expression of miR-215 and miR-218 could lead to a good curative effect and survival [40–42].

For miR-326 and miR-328, their high plasma expressions are positively related to good chemotherapy response in patients with/without metastasis [43]. In fluoropyrimidine drug metabolism, miR-326 and miR-328 can suppress ATP-binding cassette (ABC) subfamily C member 1 (ABCC1) and ATP-binding cassette (ABC) subfamily G member 2 (ABCG2), subsequently. This may lead to the increased intracellular concentration of fluoropyrimidine drugs and their metabolites. And the induced cytotoxicity increases as well (Figure 2). Since the clinical evidence was concluded based on plasma samples, miR-326 and miR-328 may cause an overall suppressed efflux of fluoropyrimidine drugs and their metabolites.

Of the 42 miRNAs that may affect fluoropyrimidine drug metabolism, 37 lack direct clinical evidence on their predictive effect on efficacy of fluoropyrimidine drugs. Detection limits and other experimental factors might limit the discovery of their potential prediction effects. They could be screened as potential biomarkers by future properly designed clinical experiments.

For the 41 miRNAs related to fluoropyrimidine drugs' efficacy based on clinical trials, 36 of them may not affect fluoropyrimidine drug metabolism enzymes. This result suggests that proteins other than those from the fluoropyrimidine drug metabolism pathway may also contribute equally or even more to the efficacy of fluoropyrimidine drugs. Consistent with this finding, we have found that several urine endogenous metabolites can predict fluoropyrimidine drugs' adverse effects [44]. These adverse effects, such as hand-foot syndrome, are predictors of better chemotherapy response alone [45, 46]. Based on the KEGG pathway and GO term enrichment results, miRNAs screened from the patients with and without metastasis showed similar results. These two subgroups of miRNA enrich both the FoxO signaling pathway and the MAPK signaling pathway. The abnormality of these pathways is a susceptible factor for cancer development. The difference is that the latter subgroup of miRNAs enriched more disease or cancer pathways, which may result from the advanced stage of the tumor.

In the future, the studies on miRNA biomarkers could be improved in the following aspects. We think what comes first is that more biological mechanism experiments are needed to reveal the actual function of miRNA markers in cancer development or drug pharmacology. Since a number
Table 2: The list of miRNAs affecting the expression of 5-FU metabolic enzymes.

| Affected protein | MicroRNA | Related cancer (cell lines or patients) |
|------------------|----------|----------------------------------------|
| ABCC5(+)         | miR-101  | HCC                                    |
| ABCC1(+)         | miR-199  | HCC                                    |
| ABCC1(-)         | miR-326  | Breast cancer                          |
| ABCC2(-)         | miR-397  | Hepatoblastoma cell                    |
| ABCC3(-), ABCC6(-) | miR-9  | Glioma cell                            |
| ABCC4(-)         | miR-125  | Hepatocellular carcinoma               |
| ABCCG2(-)        | miR-212  | Myelogenous leukemia                   |
| ABCCG2(-)        | miR-328  | Breast cancer, retinoblastoma, myelogenous leukemia, and CRC |
| ABCCG2(-)        | miR-519  | Colon cancer, breast cancer, and retinoblastoma |
| DPYD(-)          | miR-134  | HCC, lung cancer                       |
| DPYD(-)          | miR-494  | Colon cancer                           |
| DPYD(-)          | miR-582  | HCC                                    |
| DPYD(-), P-gp(-)  | miR-302  | HCC and breast cancer                  |
| P-gp(-)          | miR-103  | Gastric cancer                         |
| P-gp(-)          | miR-107  | Gastric cancer                         |
| P-gp(-)          | miR-129  | Gastric cancer                         |
| P-gp(+)          | miR-130  | Ovarian cancer                         |
| P-gp(-)          | miR-137  | Breast cancer                          |
| P-gp(-)          | miR-138  | Leukemia                               |
| P-gp(-)          | miR-298  | Breast cancer                          |
| P-gp(-)          | miR-30   | Gastric cancer                         |
| P-gp(-)          | miR-331  | Chronic myelogenous leukemia           |
| P-gp(-), ABCB1(-) | miR-451  | Breast cancer, CRC                     |
| P-gp(-)          | miR-506  | CRC                                    |
| P-gp(-), ABCG2(-) | miR-145  | Colon carcinoma                        |
| P-gp(-), ABCG2(-), ABCG5(-) | miR-200 | Breast cancer and melanomas          |
| TP(-), DPYD(-)   | miR-21   | CRC                                    |
| TS(-)            | miR-192  | CRC                                    |
| TS(-)            | miR-196  | Rectal cancer                          |
| TS(-)            | miR-197  | CRC                                    |
| TS(-)            | miR-203  | CRC                                    |
| TS(-)            | miR-215  | CRC, soft tissue sarcoma, renal cancer, and head and neck cancer |
| P-gp(+)          | miR-218  | CRC                                    |
| TS(-)            | miR-24   | Soft tissue sarcoma                    |
| TS(-)            | miR-433  | HCC                                    |
| TS(-)            | miR-450  | Rectal cancer                          |
| TS(-)            | miR-99   | Rectal cancer                          |
| ABCC5(-)TS(-)    | Let-7e    | Rectal cancer, HCC                    |
| TS(-), ABCC3(-)  | miR-192  | CRC and esophageal adenocarcinoma      |
| TS(-), ABCC3(-)  | miR-193  | Esophageal adenocarcinoma              |
| TS(-), ABCC3(-)  | miR-378  | Esophageal adenocarcinoma              |
| TS(-), ABCG2(-)  | miR-520  | HCC, pancreatic cancer, and retinoblastoma |
| TS(-), DPYD(-), P-gp(-), ABCC3(-) | miR-27 | CRC, HCC, lung cancer, gastric cancer, breast cancer, esophageal adenocarcinoma |

The effect of miRNAs on each enzyme’s expression was noted as "+" for inducing and "-" for suppressing. Abbreviations: CRC: colorectal cancer; HCC: hepatocellular carcinoma.
| miRNA     | N   | Region | Sources (N) | Survival | Expression | Stage (N) |
|-----------|-----|--------|-------------|----------|------------|-----------|
| **Without metastasis** | | | | | | |
| miR-1300 [54] | 85 | PL     | PT          | DMFS\(^*\) | —          | I-II      |
| miR-939 [54]  | 85 | PL     | PT          | DMFS\(^*\) | —          | I-II      |
| miR-135b [54] | 85 | PL     | PT          | DMFS\(^{##}\) | +          | I-II      |
| miR-1296 [54] | 85 | PL     | PT          | DMFS\(^{##}\) | +          | I-II      |
| miR-539 [54]  | 85 | PL     | PT          | DMFS\(^{##}\) | +          | I-II      |
| miR-572 [54]  | 85 | PL     | PT          | DMFS\(^{##}\) | —          | I-II      |
| miR-21 [35]   | 145| DE     | PT          | OS\(^*\) | +          | II        |
| miR-215 [40]  | 71 | ES     | PT          | DFS\(^{**,**}\) | +          | II        |
| miR-103a-3p [40] | 71 | ES    | PT          | DFS\(^{*}\), DFS\(^{#}\) | +          | II        |
| miR-143-5p [40] | 71 | ES    | PT          | DFS\(^{#}\) | +          | II        |
| miR-103a-3p [40] | 71 | ES    | PT          | DFS\(^{#}\) | +          | II        |
| miR-143-5p [40] | 71 | ES    | PT          | DFS\(^{#}\) | +          | II        |
| miR-21 [36]   | 125| CN     | PT          | DFS\(^{**}\) | +          | II-III    |
| miR-21 [35]   | 87 | JP     | PT          | OS\(^*\) | +          | II-III    |
| miR-218 [40]  | 63 | CN     | PT          | PFS\(^{**,**}\)/OS\(^{**,**}\) | —          | II-III    |
| miR-17-5p [55] | 240| CN    | PT          | OS\(^{#}\) | +          | II-III    |
| miR-320c [56] | 167| ES    | PT          | OS\(^{##}\)/DFS\(^{##}\) | +          | II-III    |
| miR-625-3p [57] | 77 | DK    | PT          | OS\(^{#}\) | +          | II-III    |
| miR-148a [58] | 201| ES    | PT          | DFS\(^{#}\) | —          | II-III    |
| miR-148a [58] | 201| ES    | Plasma      | DFS\(^{#}\) | —          | II-III    |
| miR-141 [59]  | 56 | ES     | Plasma      | DFS/OS\(^*\) | —          | I-II (35), III(15) |
| miR-200c [59] | 56 | ES     | Plasma      | DFS/OS\(^*\) | —          | I-II (35), III(15) |
| miR-342-3p [60] | 322| CN    | Plasma      | DFS\(^{##}\)/OS\(^{##}\) | +          | I-III     |
| miR-652-3p [60] | 322| CN    | Plasma      | DFS\(^{##}\)/OS\(^{##}\) | +          | I-III     |
| miR-501-3p [60] | 322| CN    | Plasma      | DFS\(^{##}\)/OS\(^{##}\) | +          | I-III     |
| miR-328-3p [60] | 322| CN    | Plasma      | DFS\(^{##}\)/OS\(^{##}\) | +          | I-III     |
| miR-4772-3p [61] | 84 | US    | Plasma      | OS\(^{#}\) | —          | II-III    |
| **With metastasis** | | | | | | |
| miR-126 [62]  | 83 | DK     | PT          | OS\(^{**,**}\)/PFS\(^{**,**}\) | +          | I-III (3), IV(86) |
| miR-199b [63] | 60 | CN     | PT          | OS\(^{#}\) | —          | I-IV      |
| miR-17-5p [64] | 81 | CN     | PT          | OS\(^{##}\) | +          | I-IV      |
| miR-143 [65]  | 52 | AT     | PT          | PFS\(^*\) | —          | II-IV     |
| miR-21 [66]   | 32 | JP     | PT          | PFS\(^*\) | +          | IV        |
| miR-31-3p [67] | 45 | FR     | PT          | PFS\(^*\) | +          | IV        |
| miR-107 [68]  | 78 | ES     | PT          | PFS\(^*\) | +          | IV        |
| miR-889 [68]  | 78 | ES     | PT          | PFS\(^{##}\)/OS\(^{##}\) | —          | IV        |
| miR-337-5p [68] | 78 | ES    | PT          | PFS\(^{#}\) | +          | IV        |
| miR-148a [58] | 71 | ES     | PT          | OS\(^{#}\) | —          | IV        |
| miR-99a-3p [68] | 78 | ES    | PT          | PFS\(^*\) | +          | IV        |
| miR-31 [69]   | 221| CN     | PT, AMT     | DFS/OS\(^*\) | +          | II-IV     |
| miR-365 [70]  | 76 | CN     | PT, AMT     | DFS\(^*\) | —          | I-IV      |
| miR-133a [71] | 125| HK     | PT, AMT     | OS\(^*\) | +          | I-IV      |
| miR-20a-5p [72] | 88 | NZ    | PT (80), LR (3), metastases (5) | PFS\(^*\) | +          | I-IV      |
| miR-92a-3p [72] | 88 | NZ    | PT (80), LR (3), metastases (5) | PFS\(^*\) | +          | I-IV      |
of the miRNA markers were only derived from clinical screening studies, they could be either the actual markers or merely the outcome of cancer development or drug metabolism. This question is complicated because one miRNA may affect many genes, and several miRNAs may regulate one gene. A review by Xing Chen may provide valuable guidance for future research on this aspect. This review summarized and discussed not just the databases of the experimentally validated or potential small molecule-miRNA associations but also four experimental techniques used in the past few years to search for small-molecule inhibitors of miRNAs [47]. Secondly, with the rapid advances in omics techniques, future clinical screening studies could be designed for multi-omics biomarkers, including miRNA, since other biomolecules such as DNA, proteins, and metabolite also contribute to the final phenotype. Last but not least, screening miRNAs from normal tissues other than tumors may provide more informative clues. Accumulating evidence suggests that the efficacy and safety of chemotherapy are not solely dependent on drug metabolism but also on the overall physiological functions [44, 48–53].

### Table 3: Continued.

| miRNA         | N  | Region | Sources (N) | Survival | Expression | Stage (N) |
|---------------|----|--------|-------------|----------|------------|-----------|
| miR-92b-3p [72] | 88 | NZ     | PT (80), LR (3), metastases (5) | PFS⁺ | + | I-IV |
| miR-30a-5p [72] | 88 | NZ     | PT (80), LR (3), metastases (5) | PFS⁺ | + | I-IV |
| miR-98-5p [72] | 88 | NZ     | PT (80), LR (3), metastases (5) | PFS⁺ | + | I-IV |
| miR-17-5p [72] | 88 | NZ     | PT (80), LR (3), metastases (5) | PFS⁺ | + | I-IV |
| miR-126 [73] | 68 | DK     | Plasma | PFS⁺ | + | IV |
| miR-148 [43] | 150 | NO     | Plasma | PFS⁺ | + | IV |
| miR-326 [43] | 150 | NO     | Plasma | PFS⁺ | + | IV |
| miR-27b [43] | 150 | NO     | Plasma | PFS⁺ | + | IV |

### Table 4: Top 20 significant KEGG pathways enriched by fluoropyrimidine drug efficacy-related miRNAs in CRC cohorts with/without metastasis.

| KEGG pathway                          | Genes | Ratio (%) | P value | Genes | Ratio (%) | P value |
|----------------------------------------|-------|-----------|---------|-------|-----------|---------|
| **Without metastasis**                 |       |           |         |       |           |         |
| MicroRNAs in cancer                    | 62    | 20.00     | 9.87E-11| Chronic myeloid leukemia | 22    | 28.95     | 6.36E-09 |
| FoxO signaling pathway                 | 36    | 27.48     | 7.13E-10| Cellular senescence    | 30    | 18.75     | 2.46E-07 |
| Cellular senescence                   | 40    | 25.00     | 1.21E-09| TGF-beta signaling pathway | 22    | 23.40     | 5.48E-07 |
| MAPK signaling pathway                | 57    | 19.39     | 3.38E-09| FoxO signaling pathway | 26    | 19.85     | 8.85E-07 |
| Autophagy                             | 33    | 24.09     | 2.47E-07| Non-small cell lung cancer | 18    | 27.27     | 1.09E-06 |
| Proteoglycans in cancer               | 42    | 20.49     | 3.02E-07| MicroRNAs in cancer | 43    | 13.87     | 1.31E-06 |
| PI3K-Akt signaling pathway            | 60    | 16.95     | 3.1E-07 | Pancreatic cancer | 19    | 25.00     | 1.99E-06 |
| Hepatitis B                           | 36    | 22.22     | 4.58E-07| Signaling pathways regulating pluripotency of stem cells | 26    | 18.18     | 5.82E-06 |
| AGE-RAGE signaling pathway in diabetic complications | 26 | 26.00 | 2.53E-06 | Pathways in cancer | 59 | 11.11 | 1.22E-05 |
| Pancreatic cancer                     | 76    | 14.31     | 5.99E-06| Proteoglycans in cancer | 31    | 15.12     | 2.4E-05 |
| Pathways in cancer                    | 23    | 26.74     | 9.64E-06| Hepatocellular carcinoma | 27    | 16.07     | 4.4E-05 |
| Colorectal cancer                     | 23    | 26.74     | 9.64E-06| Glioma           | 17    | 22.67     | 5.26E-05 |
| Kaposi sarcoma-associated herpessviruses infection | 37 | 19.58 | 9.82E-06 | Hepatitis B | 26 | 16.05 | 7.44E-05 |
| Glioma                                | 21    | 28.00     | 1.52E-05| Cell cycle       | 22    | 17.74     | 9.85E-05 |
| Human cytomegalovirus infection       | 41    | 18.22     | 1.78E-05| Prostate cancer  | 19    | 19.59     | 0.000119 |
| Chronic myeloid leukemia              | 21    | 27.63     | 1.94E-05| MAPK signaling pathway | 37    | 12.59     | 0.000177 |
| Bladder cancer                        | 15    | 36.59     | 2.18E-05| Gastric cancer  | 24    | 16.11     | 0.000187 |
| Prostate cancer                       | 24    | 24.74     | 2.41E-05| AGE-RAGE signaling pathway in diabetic complications | 19 | 19.00 | 0.00019 |

Genes related to miRNA from the two CRC cohorts were subjected to KEGG pathway enrichment analysis. The top 20 pathways with gene numbers higher than five and P values less than 0.05 (corrected by the two-side Bonferroni test) were listed here.
Table 5: Top five significant gene ontology (GO) terms enriched by fluoropyrimidine drug efficacy related miRNAs in CRC cohorts with/without metastasis.

| GO term                              | Without metastasis | With metastasis |
|--------------------------------------|--------------------|-----------------|
|                                      | Genes | Ratio (%) | P value | Genes | Ratio (%) | P value |
| CC Intracellular organelle           | 791   | 6.01      | 2.60E-23 | 1079  | 8.20      | 2.15E-29 |
| CC Nucleus                           | 534   | 6.88      | 5.02E-21 | 1059  | 8.20      | 1.55E-27 |
| CC Membrane-bounded organelle        | 772   | 5.98      | 2.24E-20 | 959   | 8.43      | 2.23E-25 |
| CC Intracellular membrane-bounded organelle | 705   | 6.20      | 2.78E-20 | 444   | 10.63     | 4.40E-23 |
| CC Nucleoplasm                       | 333   | 7.97      | 4.07E-19 | 505   | 10.16     | 7.21E-23 |
| MF Enzyme binding                    | 219   | 9.23      | 5.89E-18 | 278   | 11.72     | 6.49E-18 |
| MF Regulatory region nucleic acid binding | 113   | 10.82     | 2.79E-12 | 192   | 13.41     | 2.22E-17 |
| MF Sequence-specific DNA binding     | 136   | 9.79      | 8.82E-12 | 218   | 12.20     | 3.41E-15 |
| MF Transcription regulatory region sequence-specific DNA binding | 112   | 10.74     | 9.23E-12 | 148   | 14.18     | 6.59E-15 |
| MF Sequence-specific double-stranded DNA binding | 114   | 10.45     | 2.42E-11 | 147   | 14.09     | 1.88E-14 |
| BP Regulation of cellular metabolic process | 510   | 7.77      | 6.99E-34 | 52    | 19.48     | 9.10E-09 |
| BP Regulation of primary metabolic process | 484   | 7.62      | 1.38E-28 | 132   | 14.12     | 7.85E-13 |
| BP Regulation of nitrogen compound metabolic process | 473   | 7.68      | 1.55E-28 | 146   | 13.21     | 7.20E-12 |
| BP Regulation of metabolic process    | 532   | 7.29      | 4.96E-28 | 135   | 11.93     | 1.70E-07 |
| BP Regulation of macromolecule metabolic process | 496   | 7.38      | 4.72E-26 | 96    | 11.82     | 1.77E-04 |

Genes related to miRNA from the two CRC cohorts were subjected to G.O. enrichment analysis. The top G.O. terms with gene numbers higher than five and P values less than 0.05 (corrected by the two-side Bonferroni test) were listed here. Three ontology sources were analyzed in this step: cellular component (CC), molecular function (MF), and biological process (BP).
5. Conclusions

In conclusion, we have found that 41 miRNAs are related to fluoropyrimidine drugs’ efficacy with solid clinical evidence. They are promising candidate markers for predicting fluoropyrimidine drugs’ efficacy in the future clinical application of personalized medicine. The miRNAs screened from metastasis CRC patients have a more critical role in cancer development based on bioinformatic analysis than those screened from non-metastasis CRC patients. Among the 41 miRNAs, miR-21, miR-215, and miR-218 can suppress fluoropyrimidine drugs’ catabolism; miR-326 and miR-328 can reduce the efflux of fluoropyrimidine drugs. Together, these five miRNAs can increase the intracellular levels of cytotoxic metabolites of fluoropyrimidine drugs, leading to better chemotherapy response.

Abbreviations

5-FU: 5-fluorouracil
CRC: Colorectal cancer
DAVID: Visualization and integrated dDiscovery
dTMP: Deoxythymidine monophosphate
dUMP: Deoxyuridine monophosphate
DYPD: Dihydropyrimidine dehydrogenase
FdUMP: Fluorodeoxyuridine monophosphate
GO: Gene Ontology
KEGG: Kyoto eEncyclopedia of gGenes and gGenomes
miRNA: microRNA
TP: Thymidine phosphorylase
TS: Thymidylate synthase.

Conflicts of Interest

All authors declared that no conflicts of interest exist.

Authors’ Contributions

Houshan Yao, Lei Liu, and Mingming Li contributed to the study design and concept. Xiaomeng Sun, Jiani Chen, Xintao Chen, and Qianmin Gao contributed to the literature searching and manuscript drafting. Wei Chen, Xun Zou, Feng Zhang, Shouhong Gao, and Shi Qiu contributed to the literature screening. All authors read and approved the final manuscript. Xiaomeng Sun, Jiani Chen, and Xintao Chen contributed equally.

Acknowledgments

This study was supported by the Shanghai Science and Technology Committee, China, to Xuan Liu (Grant No. 17ZR1438800) and to Xiaoqiang Yue (Grant No. 19401971700).

References

[1] F. Bray, J. Ferlay, I. Soerjomataram, R. L. Siegel, L. A. Torre, and A. Jemal, “Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries,” CA: a Cancer Journal for Clinicians, vol. 68, no. 6, pp. 394–424, 2018.
[2] R. M. Feng, Y. N. Zong, S. M. Cao, and R. H. Xu, “Current cancer situation in China: good or bad news from the 2018 global cancer statistics?” Cancer communications, vol. 39, no. 1, p. 22, 2019.
[3] J. Yin, Z. Bai, J. Zhang et al., “Burden of colorectal cancer in China, 1990-2017: findings from the global burden of disease study 2017,” Chinese Journal of Cancer Research, vol. 31, no. 3, pp. 489–498, 2019.
[4] Z. Wu and Y. Deng, “Capecitabine versus continuous infusion fluorouracil for the treatment of advanced or metastatic colorectal cancer: a meta-analysis,” Current Treatment Options in Oncology, vol. 19, no. 12, p. 77, 2018.
[5] F. Chionh, D. Lau, Y. Yeung, T. Price, N. Tebbutt, and Cochrane Colorectal Cancer Group, “Oral versus intravenous fluoropyrimidines for colorectal cancer,” Cochrane Database of Systematic Reviews, vol. 2017, no. 8, article CD008398, 2017.
[6] N. S. Ab Mutalib, N. F. Md Yusof, S. N. Abdul, and R. Jamal, “Pharmacogenomics DNA biomarkers in colorectal cancer: current update,” Frontiers in Pharmacology, vol. 8, p. 736, 2017.
[7] A. Izzotti, C. Ceccaroli, M. Geretto, F. G. Ruggieri, S. Schenone, and E. Di Maria, “Predicting response to neoadjuvant therapy in colorectal cancer patients the role of messenger-and micro-RNA profiling,” Cancers, vol. 12, no. 6, p. 1652, 2020.
[8] D. B. Longley, D. P. Harkin, and P. G. Johnston, “5-fluorouracil: mechanisms of action and clinical strategies,” Nature Reviews. Cancer, vol. 3, no. 5, pp. 330–338, 2003.
[9] S. W. Lam, H. J. Guchelaar, and E. Boven, “The role of pharmacogenetics in capecitabine efficacy and toxicity,” Cancer Treatment Reviews, vol. 50, pp. 9–22, 2016.
[10] E. Carrillo, S. A. Navarro, A. Ramírez et al., “5-fluorouracil derivatives: a patent review (2012 – 2014),” Expert Opinion on Therapeutic Patents, vol. 25, no. 10, pp. 1131–1144, 2015.
[11] E. Berezikov, “Evolution of microRNA diversity and regulation in animals,” Nature Reviews. Genetics, vol. 12, no. 12, pp. 846–860, 2011.
[12] D. P. Bartel, “MicroRNAs: target recognition and regulatory functions,” Cell, vol. 136, no. 2, pp. 215–233, 2009.
[13] L. Rossi, E. Bonmassar, and I. Faraoini, “Modification of miR gene expression pattern in human colon cancer cells following exposure to 5-fluorouracil _in vitro_,” Pharmacological Research, vol. 56, no. 3, pp. 248–253, 2007.
[14] T. F. Hansen, R. Christensen, R. F. Andersen, F. B. Sørensen, A. Johnsson, and A. Jakobsen, “MicroRNA-126 and epithelial growth factor-like domain 7-an angiogenic couple of importance in metastatic colorectal cancer. Results from the Nordic ACT trial,” British Journal of Cancer, vol. 109, no. 5, pp. 1243–1251, 2013.
[15] J. Lu, G. Getz, E. A. Miska et al., “MicroRNA expression profiles classify human cancers,” Nature, vol. 435, no. 7043, pp. 834–838, 2005.
[16] S. H. Wang, C. C. Wang, L. Huang, L. Y. Miao, and X. Chen, “Dual-network collaborative matrix factorization for predicting small molecule-miRNA associations,” Brief Bioinform, vol. 23, no. 1, 2022.
[17] C. C. Wang, C. C. Zhu, and X. Chen, “Ensemble of kernel ridge regression-based small molecule-miRNA association prediction in human disease,” Brief Bioinform, vol. 23, no. 1, 2022.
[18] X. Chen, C. Zhou, C. C. Wang, and Y. Zhao, "Predicting potential small molecule-miRNA associations based on bounded nuclear norm regularization," Brief Bioinform, vol. 22, no. 6, 2021.

[19] Y. Sun, S. Lu, P. Zhang, Z. Wang, and J. Chen, "Steroid injection versus physiotherapy for patients with adhesive capsulitis of the shoulder: a PRISMA systematic review and meta-analysis of randomized controlled trials," Medicine (Baltimore), vol. 95, no. 20, article e3469, 2016.

[20] M. Cumpston, T. Li, M. J. Page et al., "Updated guidance for trusted systematic reviews: a new edition of the Cochrane handbook for systematic reviews of interventions," Cochrane Database of Systematic Reviews, vol. 10, article ED000142, 2019.

[21] Y. Sun, X. Sun, S. Liu, L. Liu, and J. Chen, "The overlap between regeneration and fibrosis in injured skeletal muscle is regulated by phosphatidylinositol 3-kinase/Akt signaling pathway - a bioinformatic analysis based on lncRNA microarray," Gene, vol. 672, no. 672, pp. 79–87, 2018.

[22] T. Zheng, X. Zhang, Y. Wang, and X. Yu, "Predicting associations between microRNAs and target genes in breast cancer by bioinformatics analyses," Oncology Letters, vol. 12, no. 2, pp. 1067–1073, 2016.

[23] D. Rosmarin, C. Palles, A. Pagnamenta et al., "A candidate gene study of capecitabine-related toxicity in colorectal cancer identifies new toxicity variants at DPYD and a putative role for ENOSF1 rather than TYMS," Gut, vol. 64, no. 1, pp. 111–120, 2015.

[24] H. Wang, T. Bian, D. Liu et al., "Association analysis of CYP2A6 genotypes and haplotypes with 5-fluorouracil formation from tegafur in human liver microsomes," Pharmacogenomics, vol. 12, no. 4, pp. 481–492, 2011.

[25] W. J. Fang, H. B. Mou, D. Z. Jin et al., "Characteristic CYP2A6 genetic polymorphisms detected by TA cloning-based sequencing in Chinese digestive system cancer patients with S-1 based chemotherapy," Oncology Reports, vol. 27, no. 5, pp. 1606–1610, 2012.

[26] I. Yamamiya, K. Yoshise, Y. Ishii, H. Yamada, and M. Chiba, "Effect of CYP2A6 genetic polymorphism on the metabolic conversion of tegafur to 5-fluorouracil and its enantioselectivity," Drug Metabolism and Disposition, vol. 42, no. 9, pp. 1485–1492, 2014.

[27] P. Comella, D. Natale, A. Ferrari et al., "Capecitabine plus oxaliplatin for the first-line treatment of elderly patients with metastatic colorectal carcinoma," Cancer: Interdisciplinary International Journal of the American Cancer Society, vol. 104, no. 2, pp. 282–289, 2005.

[28] D. Meulendijks, L. M. Henriks, G. S. Sonke et al., "Clinical relevance of _DPYD_ variants c.1679T>G, c.1236G>A/HapB3, and c.1601G>A as predictors of severe fluoropyrimidine-associated toxicity: a systematic review and meta-analysis of individual patient data," The Lancet Oncology, vol. 16, no. 16, pp. 1639–1650, 2015.

[29] M. Y. Huang, C. H. Wu, C. M. Huang et al., "DPYD, TYMS, TYMP, TK1, and TK2 genetic expressions as response markers in locally advanced rectal cancer patients treated with fluoropyrimidine-based chemoradiotherapy," BioMed Research International, vol. 2013, Article ID 931028, 10 pages, 2013.

[30] J. Deng, W. Lei, J. C. Fu, L. Zhang, J. H. Li, and J. P. Xiong, "Targeting miR-21 enhances the sensitivity of human colon cancer HT-29 cells to chemoradiotherapy in vitro," Biochemical and Biophysical Research Communications, vol. 443, no. 3, pp. 789–795, 2014.

[31] K. Hasegawa, H. Okamoto, K. Kawamura et al., "The effect of chemoradiotherapy or radiotherapy on thymidine phosphorylase and dihydroxyuridine dehydrogenase expression in cancer of the uterine cervix," European Journal of Obstetrics, Gynecology, and Reproductive Biology, vol. 163, no. 1, pp. 67–70, 2012.

[32] M. Miwa, M. Ura, M. Nishida et al., "Design of a novel oral fluoropyrimidine carbamate, capecitabine, which generates 5-fluorouracil selectively in tumours by enzymes concentrated in human liver and cancer tissue," European journal of cancer, vol. 34, no. 8, pp. 1274–1281, 1998.

[33] P. G. Johnston and S. Kaye, "Capecitabine: a novel agent for the treatment of solid tumors," Anti-Cancer Drugs, vol. 12, no. 8, pp. 639–646, 2001.

[34] P. Faltejskova, A. Besse, S. Sevcikova et al., "Clinical correlations of miR-21 expression in colorectal cancer patients and effects of its inhibition on DLD1 colon cancer cells," International Journal of Colorectal Disease, vol. 27, no. 11, pp. 1401–1408, 2012.

[35] N. Oue, K. Anami, A. J. Schetter et al., "High miR-21 expression from FFPE tissues is associated with poor survival and response to adjuvant chemotherapy in colon cancer," International Journal of Cancer, vol. 134, no. 8, pp. 1926–1934, 2014.

[36] S. Li, J. Gao, J. Gu, J. Yuan, D. Hua, and L. Shen, "MicroRNA-215 inhibits relapse of colorectal cancer patients following radical surgery," Medical oncology, vol. 30, no. 2, p. 549, 2013.

[37] M. Svoboda, J. Sana, P. Fabian et al., "MicroRNA expression profile associated with response to neoadjuvant chemoradiotherapy in locally advanced rectal cancer patients," Radiation oncology, vol. 7, no. 1, p. 195, 2012.

[38] X. Zhao, L. Yang, and J. Hu, "Down-regulation of miR-27a might inhibit proliferation and drug resistance of gastric cancer cells," Journal of experimental & clinical cancer research, vol. 30, no. 1, p. 55, 2011.

[39] J. A. Marchal, H. Boulai, I. Suarez et al., "Growth inhibition, G (1)-arrest, and apoptosis in MCF-7 human breast cancer cells by novel highly lipophilic 5-fluorouracil derivatives," Investigational New Drugs, vol. 22, no. 4, pp. 379–389, 2004.

[40] O. Caritg, A. Navarro, I. Moreno et al., "Identifying high-risk stage II colon cancer patients: a three-MicroRNA-based score as a prognostic biomarker," Clinical Colorectal Cancer, vol. 15, no. 4, pp. e175–e182, 2016.

[41] P. L. Li, X. Zhang, L. L. Wang et al., "MicroRNA-218 is a prognostic indicator in colorectal cancer and enhances 5-fluorouracil-induced apoptosis by targeting BIRC5," Carcinogenesis, vol. 36, no. 12, pp. 1484–1493, 2015.

[42] M. K. Boisen, C. Dehlendorff, D. Linnemann et al., "Tissue microRNAs as predictors of outcome in patients with metastatic colorectal cancer treated with first line Capecitabine and Oxaliplatin with or without Bevacizumab," Plos one, vol. 9, no. 10, article e109430, 2014.

[43] J. B. Kjersem, T. Ik Dahl, O. C. Lingjæedere, T. Guren, K. M. Tveit, and E. H. Kure, "Plasma microRNAs predicting clinical outcome in metastatic colorectal cancer patients receiving first-line oxaliplatin-based treatment," Molecular Oncology, vol. 8, no. 1, pp. 59–67, 2014.

[44] Y. Deng, H. Yao, W. Chen et al., "Profiling of polar urine metabolite extracts from Chinese colorectal cancer patients..."
to screen for potential diagnostic and adverse-effect biomarkers,” *Journal of Cancer*, vol. 11, no. 23, pp. 6925–6938, 2020.

[45] C. Zielinski, I. Lang, S. Beslija et al., “Predictive role of hand-foot syndrome in patients receiving first-line capcitabine plus bevacizumab for HER2-negative metastatic breast cancer,” *British Journal of Cancer*, vol. 114, no. 2, pp. 163–170, 2016.

[46] R. D. Hoheinz, V. Heinemann, L. F. von Weikersthal et al., “Capcitabine-associated hand-foot-skin reaction is an independent clinical predictor of improved survival in patients with colorectal cancer,” *British Journal of Cancer*, vol. 107, no. 10, pp. 1678–1683, 2012.

[47] X. Chen, N. N. Guan, Y. Z. Sun, J. Q. Li, and J. Qu, “Micro-RNA-small molecule association identification: from experimental results to computational models,” *Briefings in Bioinformatics*, vol. 21, no. 1, pp. 47–61, 2020.

[48] X. Sun, H. Xu, G. Liu et al., “A Robust Immuno-Prognostic Model of Non-Muscle-Invasive Bladder Cancer Indicates Dynamic Interaction in Tumor Immune Microenvironment Contributes to Cancer Progression,” *Frontiers in Genetics*, vol. 13, 2022.

[49] H. Yao, H. Xu, S. Qiu et al., “Choline deficiency-related multimor factors are susceptible factors for chemotherapy-induced thrombocytopenia,” *Pharmacological Research*, vol. 178, no. 178, article 106155, 2022.

[50] M. Li, X. Sun, H. Yao et al., “Genomic methylation variations predict the susceptibility of six chemotherapy-related adverse effects and cancer development for Chinese colorectal cancer patients,” *Toxicology and Applied Pharmacology*, vol. 427, no. 427, article 115657, 2021.

[51] J. Xu, Z. Lin, J. Chen et al., “Milk and egg are risk factors for adverse effects of capcitabine-based chemotherapy in Chinese colorectal cancer patients,” *Integrative Cancer Therapies*, vol. 21, p. 15347342211054, 2022.

[52] M. Li, J. Chen, Y. Deng et al., “Risk prediction models based on hematological/body parameters for chemotherapy-induced adverse effects in Chinese colorectal cancer patients,” *Supportive Care in Cancer*, vol. 29, no. 12, pp. 7931–7947, 2021.

[53] M. Li, J. Chen, S. Liu et al., “Spermine-related DNA hypermethylation and elevated expression of genes for collagen formation are susceptible factors for chemotherapy-induced hand-foot syndrome in Chinese colorectal cancer patients,” *Frontiers in Pharmacology*, vol. 12, article 746910, 2021.

[54] M. Bobowicz, M. Skrzypski, P. Czapiwski et al., “107 Prognostic value of 5-microRNA based signature in T2-T3N0 colon cancer,” *Clinical & Experimental Metastasis*, vol. 33, no. 8, pp. 765–773, 2016.

[55] Y. Ma, P. Zhang, F. Wang et al., “Elevated oncofoetal miR-17-5p expression regulates colorectal cancer progression by repressing its target gene _P130_,” *Nature Communications*, vol. 3, no. 1, p. 1291, 2012.

[56] L. Perez-Carbonell, F. A. Sinicrope, S. R. Alberts et al., “MiR-320e is a novel prognostic biomarker in colorectal cancer,” *British Journal of Cancer*, vol. 113, no. 1, pp. 83–90, 2015.

[57] M. H. Rasmussen, N. F. Jensen, L. S. Tarpgaard et al., “High expression of microRNA-625-3p is associated with poor response to first-line oxaliplatin based treatment of metastatic colorectal cancer,” *Molecular Oncology*, vol. 7, no. 3, pp. 637–646, 2013.

[58] M. Takahashi, M. Cuatrececasas, F. Balagué et al., “The clinical significance of MiR-148a as a predictive biomarker in patients with advanced colorectal cancer,” *PloS One*, vol. 7, no. 10, article e46684, 2012.

[59] S. Santausagana, I. Moreno, A. Navarro et al., “Prognostic impact of miR-200 family members in plasma and exosomes from tumor-draining versus peripheral veins of colon cancer patients,” *Oncology*, vol. 95, no. 5, pp. 309–318, 2018.

[60] D. Ji, M. Qiao, Y. Yao et al., “Serum-based microRNA signature predicts relapse and therapeutic outcome of adjuvant chemotherapy in colorectal cancer patients,” *eBioMedicine*, vol. 35, pp. 189–197, 2018.

[61] C. Liu, C. Eng, J. Shen et al., “Serum exosomal miR-4772-3p is a predictor of tumor recurrence in stage II and III colon cancer,” *Oncotarget*, vol. 7, no. 46, pp. 76250–76260, 2016.

[62] T. F. Hansen, F. B. Sørensen, J. Lindebjerg, and A. Jakobsen, “The predictive value of microRNA-126 in relation to first line treatment with capecitabine and oxaliplatin in patients with metastatic colorectal cancer,” *BMC Cancer*, vol. 12, no. 1, p. 83, 2012.

[63] Z. L. Shen, B. Wang, K. W. Jiang et al., “Downregulation of miR-199b is associated with distant metastasis in colorectal cancer via activation of SIRT1 and inhibition of CREB/KISS1 signaling,” *Oncotarget*, vol. 7, no. 23, pp. 35092–35105, 2016.

[64] L. Fang, H. Li, L. Wang et al., “MicroRNA-17-5p promotes chemotherapeutic drug resistance and tumour metastasis of colorectal cancer by repressing PTEN expression,” *Oncotarget*, vol. 5, no. 10, pp. 2974–2987, 2014.

[65] M. Pichler, E. Winter, M. Stotz et al., “Down-regulation of KRAS-interacting miRNA-143 predicts poor prognosis but not response to EGFR-targeted agents in colorectal cancer,” *British Journal of Cancer*, vol. 106, no. 11, pp. 1826–1832, 2012.

[66] Y. Iseki, M. Shibutani, K. Maeda et al., “Prognostic significi-cance of MicroRNA-21 expression in patients with Unresect-able metastatic colon cancer,” *Anticancer Research*, vol. 36, no. 10, pp. 5145–5152, 2016.

[67] G. Manceau, S. Imbeaud, R. Thiébaut et al., “Hsa-miR-31-3p expression is linked to progression-free survival in patients with KRAS wild-type metastatic colorectal cancer treated with anti-EGFR therapy,” *Clinical Cancer Research*, vol. 20, no. 12, pp. 3338–3347, 2014.

[68] S. Molina-Pinelo, A. Carnero, F. Rivera et al., “MiR-107 and miR-99a-3p predict chemotherapy response in patients with advanced colorectal cancer,” *BMC Cancer*, vol. 14, no. 1, p. 656, 2014.

[69] Y. Li, S. Xin, H. Wu et al., “High expression of microRNA-31 and its host gene LOC554202 predict favorable outcomes in patients with colorectal cancer treated with oxaliplatin,” *Oncology Reports*, vol. 40, no. 3, pp. 1706–1724, 2018.

[70] J. Nie, L. Liu, W. Zheng et al., “microRNA-365, downregulated in colon cancer, inhibits cell cycle progression and promotes apoptosis of colon cancer cells by probably targeting cyclin D1 and Bcl-2,” *Carcinogenesis*, vol. 33, no. 1, pp. 220–225, 2012.

[71] T. M. H. Wan, C. S. C. Lam, L. Ng et al., “The Clinicopathological Significance of miR-133a in Colorectal Cancer,” *Disease Markers*, vol. 2014, Article ID 919283, 8 pages, 2014.

[72] M. Neerinckx, D. Poel, D. L. S. Sie et al., “Combination of a six microRNA expression profile with four clinicopathological factors for response prediction of systemic treatment in patients with advanced colorectal cancer,” *PLoS One*, vol. 13, no. 8, article e0201809, 2018.
[73] T. F. Hansen, A. L. Carlsen, N. H. H. Heegaard, F. B. Sørensen, and A. Jakobsen, “Changes in circulating microRNA-126 during treatment with chemotherapy and bevacizumab predicts treatment response in patients with metastatic colorectal cancer,” British Journal of Cancer, vol. 112, no. 4, pp. 624–629, 2015.

[74] F. Borel, R. Han, A. Visser et al., “Adenosine triphosphate-binding cassette transporter genes up-regulation in untreated hepatocellular carcinoma is mediated by cellular micro-RNAs,” Hepatology, vol. 55, no. 3, pp. 821–832, 2012.

[75] Z. Liang, H. Wu, J. Xia et al., “Involvement of miR-326 in chemotherapy resistance of breast cancer through modulating expression of multidrug resistance-associated protein 1,” Biochemical Pharmacology, vol. 79, no. 6, pp. 817–824, 2010.

[76] S. Haenisch, S. Laechelt, H. Bruckmueller et al., “Down-regulation of ATP-binding cassette C2 protein expression in HepG2 cells after rifampicin treatment is mediated by microRNA-379,” Molecular Pharmacology, vol. 80, no. 2, pp. 314–320, 2011.

[77] H. M. Jeon, Y. W. Sohn, S. Y. Oh et al., “ID4 imparts chemoresistance and cancer stemness to glioma cells by derepressing miR-9a-mediated suppression of SOX2,” Cancer Research, vol. 71, no. 9, pp. 3410–3421, 2011.

[78] E. Turrini, S. Haenisch, S. Laechelt, T. Diewock, O. Bruhn, and I. Casborci, “MicroRNA profiling in K-562 cells under imatinib treatment: influence of miR-212 and miR-328 on ABCG2 expression,” Pharmcogenetics and Genomics, vol. 22, no. 3, pp. 198–205, 2012.

[79] X. Li, Y. Z. Pan, G. M. Seigle, Z. H. Hu, M. Huang, and A. M. Yu, “Breast cancer resistance protein BCRP/ABCG2 regulatory microRNAs (hsa-miR-328, -519c and -520h) and their differential expression in stem-like ABCG2+ cancer cells,” Biochemical Pharmacology, vol. 81, no. 6, pp. 783–792, 2011.

[80] Y. Z. Pan, M. E. Morris, and A. M. Yu, “MicroRNA-328 negatively regulates the expression of breast cancer resistance protein (BCRP/ABCG2) in human cancer cells,” Molecular Pharmacology, vol. 75, no. 6, pp. 1374–1379, 2009.

[81] X. T. Xu, Q. Xu, J. L. Tong et al., “MicroRNA expression profiling identifies miR-328 regulates cancer stem cell-like SP cells in colorectal cancer,” British Journal of Cancer, vol. 106, no. 7, pp. 1320–1330, 2012.

[82] K. K. W. To, R. W. Robey, T. Knutsen, Z. Zhan, T. Ried, and S. E. Bates, “Escape from hsa-miR-519c enables drug-resistant cells to maintain high expression of ABCG2,” Molecular Cancer Therapeutics, vol. 8, no. 10, pp. 2959–2968, 2009.

[83] K. K. W. To, Z. Zhan, T. Littman, and S. E. Bates, “Regulation of ABCG2 expression at the 3’ untranslated region of its mRNA through modulation of transcript stability and protein translation by a putative microRNA in the S1 colon cancer cell line,” Molecular and Cellular Biology, vol. 28, no. 17, pp. 5147–5161, 2008.

[84] T. Hirota, Y. Date, Y. Nishibatake et al., “Dihydro tryptamide dehydrogenase (DPD) expression is negatively regulated by certain microRNAs in human lung tissues,” Lung cancer, vol. 77, no. 1, pp. 16–23, 2012.

[85] J. Chai, W. Dong, C. Xie et al., “MicroRNA-494 sensitizes colon cancer cells to fluorouracil through regulation of DYPD,” IUBMB Life, vol. 67, no. 3, pp. 191–201, 2015.

[86] D. Cai, K. He, S. Chang, D. Tong, and C. Huang, “MicroRNA-302b enhances the sensitivity of hepatocellular carcinoma cell lines to 5-FU via targeting Mcl-1 and DYPD,” International Journal of Molecular Sciences, vol. 16, no. 10, pp. 23668–23682, 2015.

[87] L. Zhao, Y. Wang, L. Jiang et al., “MiR-302a/b/c/d cooperatively sensitizes breast cancer cells to adriamycin via suppressing P-glycoprotein (P-gp) by targeting MAP/ERK kinase kinase 1 (MEKK1),” Journal of Experimental & Clinical Cancer Research, vol. 35, no. 1, p. 25, 2016.

[88] Y. Zhang, X. Qu, C. Li et al., “miR-103/107 modulates multidrug resistance in human gastric carcinoma by downregulating Cav-1,” Tumor Biology, vol. 36, no. 4, pp. 2277–2285, 2015.

[89] C. Lu, Z. Shan, C. Li, and L. Yang, “MiR-129 regulates cisplatin-resistance in human gastric cancer cells by targeting P-gp,” Biomedicine & Pharmacotherapy, vol. 86, pp. 450–456, 2017.

[90] N. Li, L. Yang, H. Wang et al., “MiR-130a and MiR-374a function as novel regulators of cisplatin resistance in human ovarian cancer A2780 cells,” Plo S One, vol. 10, no. 6, article e0128886, 2015.

[91] X. Zuo, Y. Li, H. Shen et al., “miR-137 restoration sensitizes multidrug-resistant MCF-7/ADM cells to anticancer agents by targeting YB-1,” Acta Biochimica et Biophysica Sinica, vol. 45, no. 2, pp. 80–86, 2013.

[92] X. Zhao, L. Yang, J. Hu, and J. Ruan, “miR-138 might reverse multidrug resistance of leukemia cells,” Leukemia Research, vol. 34, no. 8, pp. 1078–1082, 2010.

[93] L. Bao, S. Hazari, S. Mehra, D. Kaushal, K. Moroz, and S. Dash, “Increased expression of P-glycoprotein and doxorubicin chemoresistance of metastatic breast cancer is regulated by miR-29B,” The American Journal of Pathology, vol. 180, no. 6, pp. 2490–2503, 2012.

[94] C. Li, J. Zou, G. Zheng, and J. Chu, “MiR-30a decreases multidrug resistance (MDR) of gastric cancer cells,” Medical science monitor: international medical journal of experimental and clinical research, vol. 22, pp. 4509–4515, 2016.

[95] D. D. Feng, H. Zhang, P. Zhang et al., “Down-regulated miR-331-5p and miR-27a are associated with chemotherapy resistance and relapse in leukaemia,” Journal of Cellular and Molecular Medicine, vol. 15, no. 10, pp. 2164–2175, 2011.

[96] H. Zhu, H. Wu, X. Liu et al., “Role of MicroRNA miR-27a and miR-451 in the regulation of MDR1/P-glycoprotein expression in human cancer cells,” Biochemical Pharmacology, vol. 76, no. 5, pp. 582–588, 2008.

[97] O. Kovalchuk, J. Filkowski, J. Meseryv et al., “Involvement of microRNA-451 in resistance of the MCF-7 breast cancer cells to chemotherapeutic drug doxorubicin,” Molecular Cancer Therapeutics, vol. 7, no. 7, pp. 2152–2159, 2008.

[98] N. Bitarte, E. Bandres, V. Boni et al., “MicroRNA-451 is involved in the self-renewal, tumorigenicity, and chemoresistance of colorectal cancer stem cells,” Stem Cells Dayt Ohio, vol. 29, no. 11, pp. 1661–1671, 2011.

[99] H. Zhou, C. Lin, Y. Zhang et al., “MiR-506 enhances the sensitivity of human colorectal cancer cells to oxaliplatin by suppressing MDR1/P-gp expression,” Cell Proliferation, vol. 50, no. 3, p. e12341, 2017.

[100] L. Shi, Z. Wang, G. Sun, Y. Wan, J. Guo, and X. Fu, “miR-145 inhibits migration and invasion of glioma stem cells by targeting ABCG2,” Neuromolecular Medicine, vol. 16, no. 2, pp. 517–528, 2014.

[101] K. Uchino, T. Ochiya, and F. Takeshita, “RNAi therapeutics and applications of microRNAs in cancer treatment,”
Japanese Journal of Clinical Oncology, vol. 43, no. 6, pp. 596–607, 2013.

[102] J. Chen, W. Tian, H. Cai, H. He, and Y. Deng, “Down-regulation of microRNA-200c is associated with drug resistance in human breast cancer,” Medical oncology, vol. 29, no. 4, pp. 2527–2534, 2012.

[103] S. Liu, M. T. Tetzlaff, R. Cui, and X. Xu, “miR-200c inhibits melanoma progression and drug resistance through down-regulation of BMI-1,” The American Journal of Pathology, vol. 181, no. 5, pp. 1823–1835, 2012.

[104] V. Boni, N. Bitarte, I. Cristobal et al., “miR-192/miR-215 influence 5-fluorouracil resistance through cell cycle-mediated mechanisms complementary to its post-transcriptional thymidilate synthase regulation,” Molecular Cancer Therapeutics, vol. 9, no. 8, pp. 2265–2275, 2010.

[105] Z. Sun, N. Zhou, Q. Han et al., “MicroRNA-197 influences 5-fluorouracil resistance via thymidylate synthase in colorectal cancer,” Clinical and Translational Oncology, vol. 17, no. 11, pp. 876–883, 2015.

[106] T. Li, F. Gao, and X. P. Zhang, “miR-203 enhances chemosensitivity to 5-fluorouracil by targeting thymidylate synthase in colorectal cancer,” Oncology Reports, vol. 33, no. 2, pp. 607–614, 2015.

[107] D. Samantarrai, M. Sahu, J. Roy et al., “Erratum: unraveling novel TF-miRNA regulatory crosstalk in metastasis of soft tissue sarcoma,” Scientific Reports, vol. 5, no. 1, article 12254, 2015.

[108] S. Mitra, N. Mukherjee, S. Das, P. Das, C. K. Panda, and J. Chakrabarti, “Anomalous altered expressions of downstream gene-targets in TP53-miRNA pathways in head and neck cancer,” Scientific Reports, vol. 4, no. 4, p. 6280, 2014.

[109] H. W. Z. Khella, M. Bakhet, G. Allo et al., “miR-192, miR-194 and miR-215: a convergent microRNA network suppressing tumor progression in renal cell carcinoma,” Carcinogenesis, vol. 34, no. 10, pp. 2231–2239, 2013.

[110] B. Song, Y. Wang, M. A. Titmus et al., “Molecular mechanism of chemoresistance by miR-215 in osteosarcoma and colon cancer cells,” Molecular Cancer, vol. 9, no. 1, p. 96, 2010.

[111] K. Gotanda, T. Hirota, N. Matsumoto, and I. Ieiri, “MicroRNA-433 negatively regulates the expression of thymidylate synthase (TYMS) responsible for 5-fluorouracil sensitivity in HeLa cells,” BMC Cancer, vol. 13, no. 1, p. 369, 2013.

[112] R. Hummel, C. Sie, D. I. Watson et al., “MicroRNA signatures in chemotherapy resistant esophageal cancer cell lines,” World Journal of Gastroenterology, vol. 20, no. 40, pp. 14904–14912, 2014.

[113] H. Jiang, Q. Dong, X. Luo et al., “The monoclonal antibody CH12 augments 5-fluorouracil-induced growth suppression of hepatocellular carcinoma xenografts expressing epidermal growth factor receptor variant III,” Cancer Letters, vol. 342, no. 1, pp. 113–120, 2014.

[114] F. Wang, X. Xue, J. Wei et al., “Hsa-miR-520h downregulates ABCG2 in pancreatic cancer cells to inhibit migration, invasion, and side populations,” British Journal of Cancer, vol. 103, no. 4, pp. 567–574, 2010.