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MicroRNAs as Potential Biomarkers and Therapeutic Targets in Renal Cell Carcinoma

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Abstract

Optimal management of patients will be guided by ideal biomarkers, which can help clinicians to implement early detection, prognosis, prediction of benefit from therapies, recurrence or progression of human cancers, although biomarkers have not yet become clinically routine, especially in renal cell carcinoma (RCC). microRNAs (miRNAs) are chemically stable and can thus be detected in a broad range of clinical samples and hence, have diagnostic and prognostic value in many human malignancies. The discovery that miRNAs function as key regulators of carcinogenesis and tumor progression has initiated extensive research in the cancer field, leading to the development of newer anti-cancer therapies. Growing studies have identified some miRNAs as promising biomarkers and therapeutic targets of RCC. This review discusses the current status of miRNAs in RCC, focusing on miRNAs as potential diagnostic, prognostic, and predictive biomarkers and their therapeutic potential.

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Introduction.

Renal cell carcinoma (RCC) is the most lethal urological malignancy with the highest mortality rate at over 30%, and the incidence of RCC is increasing by 2~4% each year [1]. In Early stage RCC confined to the kidney can be cured in approximately 90% of patients by surgery, but these rates drop substantially in patients with advanced disease [1]. Unexpectedly, 30% of RCC have advanced disease at diagnosis and 30% of patients with organ-confined RCCs will subsequently develop metastatic recurrence resistant to conventional cytotoxic chemotherapy and die of their disease [2]. Recently, an improved understanding of genetic changes in RCC has produced pharmaceuticals based on specific molecular targets. However, eventually the vast majority of treated patients with mRCC develop progressive disease due to acquired resistance or for other reasons. Hence, biomarkers for early detection, follow-up and predicting treatment efficacy and new strategies to treat this disease are urgently needed. Until now, DNA, RNA and protein profiles of cancer research have not fully elucidated the pathogenesis of RCC. miRNA has been identified as a crucial regulator of human cancer pathogenesis from initiation to metastasis [3-7], which suggests that miRNA has therapeutic potential against RCC. Intriguingly, growing evidence have showed that miRNAs are chemically stable

and can be detected in a broad range of body fluids and hence, have diagnostic and prognostic value in many malignancies [8-11]. This review summarizes miRNAs potentially serving as biomarkers and therapeutic targets for RCC.

Sorting out small RNAs.

Small RNAs are short (approximately 18~30 nts), non-coding RNA molecules that can regulate gene expression in both the cytoplasm and the nucleus via post-transcriptional gene silencing (PTGS), chromatin-dependent gene silencing (CDGS) or RNA activation (RNAa) [12]. Endogenous small RNAs are diverse and comprise miRNAs, small interfering RNAs (siRNAs), and piwi-interacting RNAs (piRNAs) [13]. miRNAs in association with Argonaute (AGO) and GW182 proteins, forming the RNA-induced silencing complex (RISC), mediate fine tuning of gene expression and are involved in various biological key processes. Each miRNA is predicted to target hundreds of mRNAs thus influencing key regulatory mechanisms of cells. Consequently, deregulated miRNA expression has been described to contribute to the initiation and progression of human cancer and other diseases [14]. SiRNAs are oligonucleotides of around 21~23 nts in length. SiRNAs, generated from double-stranded RNAs (dsRNAs), trigger sequence-specific mRNA decay also known as RNA interference (RNAi). The third developmentally vital class of small RNAs is the piwi-interacting (pi) RNAs, which play a role in the formation of the germ line. In mammals, these ~27 nt ssRNAs are expressed in the reproductive organs, mainly the testis [15]. Research has indicated that small RNAs play important roles in cellular processes such as cell differentiation, growth, migration, apoptosis, metabolism and defense. Accordingly, miRNAs as a representative of small RNAs are critical regulators of development, physiology and disease [12].

Differential expression of miRNAs in RCC.

In order to develop further understanding of the molecular mechanism involved in the pathogenesis of RCC, increasing groups investigate miRNA expression profiles in the RCC tissues [16-22] and serum [23, 24]. These studies suggest the miRNA expression profiles or even single miRNA could become useful biomarkers for RCC diagnosis, prognosis, and treatment optimization. The first study in this setting was done by Gottardo et al. They reported 4 human miRNAs (miR-28, miR-185, miR-27, and let-7f-2) significantly up-regulated and no down-regulated through studying the expression of 248 miRNA clusters (161 human, 84 mouse, and 3 Arabidopsis) in a set of 27 kidney specimens [16]. However, the most of latter research showed that dysregulated miRNAs in RCC are reduced. These altered miRNAs in RCC in each study are not consistent. The inconsistencies may be caused by the different discovery microarray platforms, samples themselves, experimental conditions, and so on [16-20, 22]. Clear cell RCC (ccRCC), the most common subtype of RCC, originates from the epithelial cells of the proximal tubulus of the kidney. Recently, through analyzing the miRNA expression profiles of 10 ccRCC-derived cell lines and the renal proximal tubular epithelial cells (PTECs), Gerben et al. [25] reported 15 miRNAs were markedly reduced and 8 miRNAs increased in ccRCC cells. Some of deregulated miRNAs in ccRCC cells were consistently reported in ccRCC tissues, such as the miR-200 family and miR-205 [25, 26].

miRNAs as biomarkers in RCC.

miRNAs as diagnostic biomarkers in RCC.

Identification of stable biomarkers for diagnosis remains a major challenge in cancer research, especially RCC. Following the discovery of the existence of circulating miRNAs in the plasma and serum of cancer patients [8-10], growing evidence show that circulating miRNAs are promising biomarkers for noninvasive diagnosis in various tumor entities. Recent studies have suggested that the deregulated miRNA expression profiles and the identification of their targets in RCC offer the opportunity to develop new diagnostic strategies. The miRNA expression profiles may be helpful 1) to distinguish normal from malignant tissues, 2) to identify the tissue of origin in poorly differentiated tumors or tumors of unknown origin and 3) to differentiate tumors from the same organ with different histology [27, 28]. miRNA profiles offer some important potential advantages over standard mRNA or other protein-based profiles. miRNAs have been shown to be very stable in tissues and biological fluids, including serum, plasma, stool and urine and are protected from endogenous RNase by virtue of their small size and perhaps by packaging within exosomes [9]. Jung et al.[29] observed the significant distinction of miRNA expression profiles between ccRCC and normal kidney tissues. Their further investigation showed that combination of miR-141 and miR-155 had 97% accuracy for identification of ccRCC. miR-141 or miR-200c alone could yield 93% or 94% accuracy in discriminating ccRCC tissues from normal kidney tissues, respectively. This is the first ccRCC/normal classifier constructed with miRNAs. Despite each type of RCC has distinct histological appearance, many cases in which the morphological criteria are not conclusive.

As RCC subtype is a major determinant for their prognosis and the sensitivity to targeted therapies, accurate classification is very important. Some reports have showed the ability of miRNAs to distinguish between RCC subtypes [30-33]. Petillo et al.[30] showed the overexpression of miR-424 and miR-203 in ccRCC relative to pRCC, as well as the lower expression of miR-203 in the benign oncocytomas (where it is underexpressed relative to normal kidney) than the malignant chRCC (where it is overexpressed relative to normal kidney). A high degree of similarity in microRNA expression between ccRCC and pRCC and between chRCC and oncocytoma, and a lower degree of similarity between these pairs were reported [31, 32]. Fridman et al. [32] defined a two-step decision-tree classifier that used expression levels of six microRNAs: the first step uses expression levels of miR-210 and miR-221 to distinguish between the two pairs of subtypes; the second step uses either miR-200c with miR-139-5p to identify oncocytoma from chRCC, or miR-31 with miR-126 to identify ccRCC from pRCC. Identification sensitivity of the classifier was 94% for oncocytoma, 86% for chRCC, 94% for ccRCC, and 100% for pRCC, with overall accuracy of 93% [32]. Another study provided a stepwise decision tree to distinguish between normal and each of the subtypes in a maximum of four steps based on miRNA microarray analysis [31]. The system had a sensitivity of 97% in distinguishing normal from RCC, 100% for ccRCC subtype, 97% for pRCC subtype, and 100% accuracy in distinguishing oncocytoma from chRCC subtype. This system was cross-validated and showed an accuracy of about 90%. The classification system provided accurate classification (an accuracy of about 98%) between any given subtype pair. miR-21 is up-regulated in a variety of cancers, including RCC [33-37]. Expression of miR-21 is higher in both ccRCC and pRCC subtypes than that of healthy kidney and both chRCC and oncocytoma [33, 38]. Furthermore, miR-21 could distinguish ccRCC and pRCC subtypes from chRCC and oncocytoma with the sensitivity of 83% and specificity 90% [33]. Growing evidence are available on circulating miRNAs as biomarkers of RCC. Wulfken et al.[39] reported that miR-1233 was upregulated in RCC tissue and serum, and serum miR-1233 could detect RCC with 77.4% sensitivity and 37.6% specificity. Redova et al. [40] showed that combination of serum miR-378 (overexpression in RCC patients) and miR-451 (downexpression in RCC patients) could yields a ROC curve area of 0.86 with 81% sensitivity and 83% specificity in discriminating RCC patients from healthy controls. Wang et al. [41] demonstrated that the panel of 5 serum miRNAs (upregulated miR-193a-3p, miR-362 and miR-572, and downregulated miR-28-5p and miR-378 in RCC) could serve as an early detection marker for RCC with the accuracy of 0.807. Theoretically, urinary miRNAs may be filtered and excreted by, or directly from, the kidney and/or urinary tract. Therefore, it is possible that urinary miRNAs are promising noninvasive biomarkers of RCC. Melkonyan et al. [23] detected 22 different urinary miRNAs, but none was kidney-specific. Hanke et al. [24] showed that analysis of miRNA species in single urine samples revealed the miRNA ratios miR-126 : miR-152 and miR-182 : miR-152 were significantly elevated in urine of urothelial bladder cancer patients compared with urine of healthy donors and patients with urinary tract infections, enabling a separation of tumor patients from the control groups. The expression of miR-15 and miR-15a in urine from patients with RCC was significantly higher than that of oncocytoma and urinary tract inflammation [42, 43]. More recently, the panel of urinary miR-122, miR-1271 and miR-15b was identified as urinary biomarkers of ccRCC [44]. Thus, these studies have revealed a new possibility in the development of non-invasive investigation of RCC by using specific urinary miRNAs as diagnostic biomarkers.

miRNAs as prognostic biomarkers in RCC.

Therapeutic decision-making of RCC largely depends on defining the prognosis. Currently, tumor–node–metastasis (TNM) stage, Fuhrman nuclear grade, and RCC subtype are used to evaluate the prognosis of RCC. However, all prognostic models are not yet optimal [45]. Recent studies showed that individual miRNAs have prognostic relevance in RCC. High miR-21 expression levels in RCC are associated with late stages, higher tumor grades, larger tumor size, and shorter disease-free survival (DFS) and overall survival (OR) [33, 36]. Petillo et al.[30] investigated miRNA expression profiles that distinguish ccRCC cases with poor vs. good prognosis. This study showed S-has-miR-32 was over-expressed in poor prognosis cases relative to good prognosis cases. The significantly lower level of miR-106b in RCC patients who developed metastasis has been observed [46]. Wang et al. showed that miR-100 was overexpressed in RCC and associated with poor prognosis in patients with RCC [47]. However, several studies have consistently identified that miR-100 was downregulated in RCC [21, 48]. These miRNAs may be incorporated into existing models to improve their accuracy after validation on a larger group of patients. To date, there are very few indicators that can predict which RCC patients will develop a recurrence. RCC patients who developed a recurrence had a significant increase in hypermethylated miR-9-1, miR-9-3 and miR-124-3 in their primary tumor [49, 50]. What's more, methylation of miR-9-3 and miR-124-3 was significantly associated with an

increased risk of recurrence and high methylation levels of either miR-9-1 or miR-9-3 resulted in a significant, nearly 30-month decrease in recurrence-free survival time (miR-9-1 and miR-9-3) [49, 50]. Tumor metastasis and grade was positively correlated with methylation of miR-124-3 in RCC [50]. These results suggested miR-9 and miR-124-3 function as a tumor suppressor for RCC tumorigenesis, and methylation status of miR-9-1, miR-9-3 and miR-124-3 could be a biomarker for a poor prognosis due to the development of metastatic recurrence [49]. Gleadle and colleagues showed, for the first time, the effect of the VHL tumor suppressor gene on miRNA expression in RCC [51]. A significant increase in miR-210 expression was observed in the tumor tissue by some studies. MiR-210 levels also showed a correlation with a HIF-regulated mRNA, carbonic anhydrase IX (CAIX), and with VHL mutation or promoter methylation. The expression of miR-210 was shown to be a prognostic marker as its expression correlated with sarcomatoid changes, Fuhrman nuclear grade, lymph node metastasis, patient survival [51-53]. Moreover, miR-210 could effectively distinguish malignant from non-malignant tissues with a classification accuracy rate of 88% [54].

miRNAs as predictive biomarkers in RCC.

Metastatic dissemination is the most significant prognostic factor for patients with RCC. Moreover, predicting recurrences early can improve patient outcome. Preliminary evidence suggests that miRNAs can serve as predictive markers and indicators of cancer metastasis and relapse. Recent studies suggest that miRNAs dysregulated in metastatic RCC may be decreased [55-57]. Heinzelmann et al. found that 33 miRNAs altered in metastatic RCC compared with non-metastatic RCC are all downregulated, 20 out of which were associated with progression-free survival [55]. In another research, 56 miRNAs downregulated and only 9 miRNAs upregulated in metastatic RCC were reported [56]. Notably, miR-10b, miR-27b, miR-26a/b, miR-29a, miR-181a, miR-151-5p, miR-130a, let-7 family and miR-30 family were consistently decreased. Moreover, many downregulated miRNAs (miR-204, miR-200a/b, miR-215, miR-26a, miR-10b, miR-196a and miR-194) in RCC were further downregulated in metastatic RCC. Even some overexpression miRNAs (miR-16, miR-155, miR-122 and let-7f/g) in RCC were decreased in metastatic RCC. In addition, miRNAs (miR-10b, miR-126, miR-196a, miR-204, miR-215, miR-192 and miR-194) were decreased in metastatic RCC compared with matched primary tumors [57]. Lower expression of miR-215 is related to RCC patients with short disease-free survival time [58]. Upregulation of miR-106b and miR-122 and downregulation of miR-514 in RCC were significantly lower in tumors of patients who developed metastasis compared with non-metastatic tumors [46, 54]. In relation to the TNM stage of RCCs, a general tendency for miR-106b, miR-122 and miR-708 levels to decrease from earlier stages towards advanced was observed [5, 46, 54]. Further investigation showed that miR-122 and miR-514 were related to RCC relapse [54]. To identify miRNA signature of tumor relapse in RCC patients after nephrectomy, Slaby et al. [59] analyzed miRNA expression profiles in tumor tissues of RCC patients with relapse or relapse-free and found 20 miRNAs were upregulated and 44 miRNAs were downregulated in RCC patients developing early recurrence. Many of these dysregulated miRNAs, including miR-143, miR-10b, miR-26a, miR-195, miR-145 and miR-126, are also associated with metastasis [55-57]. Higher miR-127-3p, miR-145 and miR-126 were significantly correlated with relapse-free survival [59]. Accordingly, miRNAs in RCC were gradually regressed during tumor progression. These results suggest miRNAs may be potential predictive marker of early metastasis and relapse after nephrectomy in RCC patients. Therapy targeted at the vascular endothelial growth factor (VEGF) and mammalian target of rapamycin (mTOR) pathways has been the standard of care in metastatic renal cell carcinoma (mRCC). At present, seven such agents have been approved for the treatment of mRCC, including sunitinib, sorafenib, pazopanib, axitinib, temsirolimus, everolimus, and bevacizumab in combination with IFNa. Existing investigation shows that most patients with mRCC gain significant benefit from these drugs. However, 11-29% of patients exhibit progressive disease during treatment with VEGF target therapy [60]. Furthermore, eventually the vast majority of treated patients acquire resistance. Thus, biomarkers are urgently needed to predict drug response and monitor therapeutic response. Examining miRNA expression in the peripheral leukocytes of 38 patients with advanced RCC receiving sunitinib, Gamez-Pozo et al. recently identified 28 miRNAs associated with poor response and 23 miRNAs associated with prolonged response to sunitinib [60]. The predictive models established according to miRNA expression could effectively distinguish the prolonged response group from the poor response group [60].The prolonged response group's median time to progression and overall survival (OS) is significantly longer (14 and 24 months) than the poor response group's (3.5 and 8.5 months) [60]. More recently, miRNA expression in cytoreductive nephrectomy specimens of 20 patients with metastatic ccRCC receiving sunitinib treatment was assessed using a quantitative miRNA PCR-primer array platform [61].

The miR-141 expression in poor responders was markedly lower than that in good responders, and miR-141 down-regulation was related to the presence of sarcomatoid features associated with a poor prognosis [61]. Through enhancing ZEB1/2 protein and reducing E-cadherin, downregulation of miR-141 regulated the epithelial-to-mesenchymal transition (EMT), which contributes to treatment resistance and metastases [61-63]. Moreover, miR-141 inhibited the p38α pathway and subsequently increased the sensitivity to reactive oxygen species (ROS)-producing therapeutic agents in ovarian cancer [64]. Collectively, these results indicate miRNAs may be used as predictive biomarkers for therapy response in the future.

miRNAs as therapeutic targets in RCC.

miRNAs are important biological regulators and therefore can serve as novel therapeutic targets for cancer. For miRNAs with oncogenic capabilities, potential therapies include anti-miRNA oligonucleotides, miRNA sponges, miRNA masking, and small molecule inhibitor. Inhibition of miR-122, a liver-specific miRNA contributing to the replication of HCV and hepatocellular carcinoma, significantly repressed HCV replication [65]. For tumor-suppressor miRNAs, restoring suppressor miRNAs by forced expression of those miRNAs may be a useful strategy [66]. Furthermore, miRNAs could enhance response to standard cancer therapies. There are several promising potential therapeutic miRNA candidates in RCC.

Inhibition of oncogenic miRNAs.

miR-21 is up-regulated in a variety of cancers, including RCC [33-37]. Creating a transgenic mouse carrying miR-21 was reported by Frank Slack and co-workers [67]. Genetic machinery, inserted along with miR-21, allowed them to turn miR-21expression on or off by changing the animals' diet. In Slack's experimental model, when they turned on miR-21 expression, the animals quickly developed a pre–B-cell lymphoid malignancy. When the expression of miR-21 was turned off, the animals' tumors completely regressed within a few days. miR-155 is regarded as an oncogenic miRNA on the basis of the finding that B cell malignancies occurred in miR-155-overexpressing transgenic mice [68]. miR-21 and miR-155 have repeatedly been identified through microarray profiling as up-regulated in RCC relative to normal kidney tissue. Knockdown of miR-21 in ccRCC suppressed cell proliferation, migration and invasion, and promoted cell apoptosis [34-37]. Further investigation showed that miR-21 regulated multiply genes, including PTEN/Akt/TORC1, fas ligand (FASL), metalloproteinase inhibitor 3 (TIMP3), p21 and p38 MAP kinases, cyclin E2, transcription factor TCF21 [34-37]. These studies supported an interesting hypothesis that miR-21 and miR-155 may have oncogenic function in RCC. Abrogated ubiquitin-mediated degradation due to VHL inactivation in more than 70% of ccRCC causes accumulation of HIF1α and HIF2α, which increases expression of VEGF and glucose transporter type 1 (GLUT1), consequently promoting angiogenesis and metabolic disorders [6]. It has been well documented that miR-210 could be induced under hypoxic conditions and modulated by HIFs in various solid cancers, including ccRCC [6]. However, the biological significance of miR-210 in RCC is controversial. miR-210 overexpression caused multipolar spindle formation and aneuploidy through centrosome amplification, and reduced cell viability via G2/M-phase arrest in RCC cells [6]. Inhibition of miR-210 did not alter RCC cell viability and cycle [6]. In contrast, Redova et al. reported that inhibition of miR-210 leaded to the decrease of cell viability via G2/M-phase arrest, cell migration and invasion [69]. Thus, further studies regarding the role of miR-210 in RCC in vitro and in vivo is needed. Despite several studies have consistently identified that miR-34a was upregulated in RCC [19, 21, 48], Vogt et al. found inactivation of miR-34a by CpG methylation in 58% RCC [70]. An increase of miR-34a expression during rat oxidative stress-induced renal carcinogenesis and a significant decrease of cellular proliferation in RCC cells after knockdown of miR-34a were observed [71]. In contrast, Yamamura and coworkers showed that miR-34a suppressed cell growth and invasion in RCC cells by indirectly modulating RhoA via targeting c-Myc and downregulating c-Myc–P-TEFb complex [7].The effects of miR-17-92 over expression have been examined in multiple animal models, human cancers, and cell culture systems for its ability to regulate a number of cellular processes that favor malignant transformation [72, 73]. Chow et al.[74] showed that the miR-17-92 cluster was also over expressed in RCC and had an important role in promoting tumor cell proliferation. If the levels of these miRNAs could be manipulated in vivo, effects relevant for RCC progression and treatment efficacy could be obtained.

Restoration of tumor suppressor miRNAs.

The miRNA microarray analysis has revealed that most of miRNAs deregulated in RCC tissues and cell lines were reduced. Notably, the miR-200 family consisting of five members, miR-200a/miR-200b/ miR-429 and miR-200c/miR-141, localized in 1p36.33 and 12p13.31 respectively, were all significantly downregulated in RCC [19, 25, 26]. Furthermore, among the miRNAs differentially expressed in

ccRCC, miR-141 and miR-200c are the most significantly down-regulated [20, 22, 25, 26, 29, 46]. Both miR-200c and miR-141 are mechanistically associated with the process of epithelial-mesenchymal transition (EMT). EMT is characterized by a decrease of E-cadherin, loss of cell adhesion, and increased cell motility leading to promotion of metastatic behavior of cancer cells (including RCC) [75]. Zinc-finger E-box binding homeobox 1(ZEB1) is a crucial inducer of EMT in various human tumors directly suppressing transcription of miR-141 and miR-200c, which strongly activate epithelial differentiation in pancreatic, colorectal and breast cancer cells [76]. Nakada et al. found that over-expression of miR-141 and miR-200c caused down-regulation of ZEB2 and up-regulation of E-cadherin in two renal carcinoma cell lines, ACHN and 786-O [20]. ZEB2 was identified as the common target of miR-141 and miR-200c. It already has been reported that ZEB2 up-regulated in a variety of human carcinomas may function as a transcriptional repressor for E-cadherin [75]. More recently, Berkers et al. and Yu et al. [61, 77] showed that overexpression of miR-141 in RCC cells reversed EMT and inhibited cell growth in normoxic and hypoxic conditions by targeting CDC25B. BY using xenograft orthotopic implantations, Chen et al.[78] demonstrated that miR-141 effectively suppressed tumor growth, local invasion, and metastatic colonization by targeting EphA2. On the basis of these data, miR-141 and miR-200c may have a tumor suppressor function in cancer cells and could be a promising treatment in anticancer therapy [22]. Other downregulated miRNAs in RCC also contribute to the development of RCC. Through inhibiting the proto-oncogenic SFKs, miR-205 overexpression in RCC cells markedly suppressed cell growth, migration and invasion, and induced cell apoptosis in vitro and in vivo [4]. miR-708 also had similar role in RCC by targeting survivin, ZEB2 and BMI1 [5]. Moreover, intratumoral delivery of synthetic miR-708 oligonucleotides successfully inhibited tumorigenicity in established renal tumor xenografts [5]. Similar results were also seen in miR-508-3p, miR-509-3p [79], miR-99a [80], miR-145 [81], miR-1826 [82], miR-584 [83], miR-30d [84], miR-138 [85], miR-1285 [26], miR-135a [86]. In addition to angiogenesis related to the loss of VHL gene in ccRCC as a source of nutrients for tumor growth, autophagy that eliminates cellular defective organelles and proteins and recycles nutrients for survival also provides nutrients necessary. Mikhaylova and co-workers [3] demonstrated that RCC cell growth depended on LC3B-mediated autophagy and VHL gene induced the expression of miR-204 in RCC cells, which suppressed tumor growth through inhibition of LC3B-mediated autophagy.

Enhancing of responsiveness of tumors to standard therapy.

Schickel et al. [87] showed that miR-200c affected sensitivity to CD95-mediated apoptosis, and validated FAP-1 as a miR-200 target that regulates CD95-mediated apoptotic signaling in tumor cells, including RCC cells CAKI-1 and ACHN. When miR-200c was highly over-expressed, CAKI-1 and ACHN cells were found to be more sensitive to TRAIL-induced apoptosis, and CAKI-1 cells were also more sensitive to TNFa-induced apoptosis [87]. miR-145 did not directly induce cell apoptosis but enhance the sensitivity to cisplatin of RCC cells [81].

Conclusions and further aspects.

Although we are in the early phases of miRNA research in RCC, it is anticipated that miRNAs will have a significant impact on improving the patient's diagnosis and management. Based miRNA profiles in RCC, we may identify biomarkers for early detection, follow-up of the disease and predicting treatment efficacy, which will improve patient outcome. Following the discovery of the existence of circulating miRNAs in the plasma and serum of cancer patients, growing evidence shows that circulating miRNAs are promising biomarkers for noninvasive diagnosis and prognosis in various tumor entities. The global expression patterns of miRNAs in patient - matched RCC and normal kidney tissues contribute to the growing understanding of the role that miRNAs play in RCC. Therefore, miRNAs can serve as novel therapeutic targets for RCC. Further studies are needed in larger samples of RCC and further investigation is needed to clarify the roles of identified miRNAs in the pathogenesis of RCC. Moreover, further studies of in vitro and in vivo models are necessary to elucidate miRNAs' targets and role in RCC. The study of miRNAs in RCC will play an important role in prospective clinical medicine as diagnostic, prognostic and predictive biomarkers, and therapeutic strategies.

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