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MiRNA-93-5p promotes the biological progression of gastric cancer cells *via* Hippo signaling pathway

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Abstract. – OBJECTIVE: To clarify the influence of microRNA-93-5p (miRNA-93-5p) on biological behaviors of gastric cancer (GC) cells and its regulatory effect on Hippo pathway.

MATERIALS AND METHODS: SGC-7901 and HGC-27 cells were used for establishing miRNA-93-5p overexpression and downregulation model through transfection of miRNA-93-5p mimics or inhibitor, respectively. Relative levels of genes in Hippo pathway were determined in GC cells transfected with miRNA-93-5p mimics or inhibitor by quantitative Real-time polymerase chain reaction (qRT-PCR). Regulatory effects of miRNA-93-5p on proliferative, migratory and invasive abilities of GC cells were evaluated by cell counting kit-8 (CCK-8), colony formation and transwell assay, respectively.

RESULTS: MiRNA-93-5p was markedly upregulated by transfection of miRNA-93-5p mimics into SGC-7901 cells, which was downregulated by transfection of miRNA-93-5p inhibitor into HGC-27 cells. Overexpression of miRNA-93-5p accelerated GC cells to proliferate, migrate and invade. Meanwhile, miRNA-93-5p overexpression in GC cells upregulated downstream genes in Hippo pathway, including CDX2, FOXM1 and CTGF.

CONCLUSIONS: MiRNA-93-5p enhances proliferative, migratory and invasive abilities of GC cells by activating Hippo pathway, which may serve as a diagnostic and therapeutic target for GC.

Key Words

MiRNA-93-5p, Hippo Pathway, Gastric cancer, Biological functions.

Introduction

Globally, gastric cancer (GC) is the third most common malignancy. There are approximately 723,000 people die of GC each year throughout the world[1]. Medical technology and therapeutic approaches for GC have been advanced in recent years. Nevertheless, the mortality of GC still remains high due to hidden symptoms, undefined biological characteristics and genetic heterogeneity[2]. GC is a heterogeneous disease associated with a variety of genetic and epigenetic changes. Genetic mutations, somatic cell copy number changes, structural variations, epigenetic variations, and transcriptional regulatory activities of mRNAs and non-coding RNAs all could influence the tumorigenesis of GC[3-5]. Therefore, explorations on the molecular mechanism of GC contribute to identify novel therapeutic target, which provides theoretical references for clinical treatment of GC. MicroRNAs (miRNAs) are a class of non-coding RNAs that bind to the 3'UTR of the messenger RNA in an incomplete base-pairing manner, and further regulate the expressions of relevant downstream proteins[6]. In recent years, miRNAs have been proved to be crucial in mediating the occurrence and progression of GC[7,8]. MiRNA-93-5p belongs to the miR-106b-25 family and is located on chromosome 11q22.1. It is relatively conserved in mammals[9]. MiR-93 is lowly expressed in colon cancer lesions, which is closely related to metastasis, differentiation and poor prognosis of colon cancer. Upregulation of miR-93 inhibits the invasion and metastasis of colon cancer by inhibiting Wnt/β-catenin pathway[10]. In liver cancer, miR-93 accelerates tumor progression by directly targeting phosphatase and tensin homolog deleted on chromosome ten (PTEN) and CDKN1A *via* activating c-Met/PI3K/Akt pathway[11]. As an oncogene, miR-93 promotes the occurrence and progression of liver cancer and GC by targeting PDCD4[12,13]. It is reported that miRNA-93-5p participates in the carcinogenesis of colorectal cancer, esophageal cancer and lung cancer[14-16]. Regulatory functions of Hippo pathway in biological processes in mammals have been well explored, which are capable of regulating tissue regeneration and tumor progression[17]. As a crit-

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ical regulatory, Hippo pathway is able to inhibit tumor activity by mediating the proliferation and apoptosis tumor cells[18]. Clearly, Hippo pathway is closely related to the tumorigenesis and tumor progression. Therefore, precise control of Hippo pathway activity, maintenance of cellular homeostasis and clarification of inactivated key genes in Hippo pathway are of significance to tumor treatment. A relevant study pointed out that Hippo pathway is related to *H. pylori* infection and mediates the development and metastasis of GC[19]. Due to epigenetic changes, the core components of Hippo pathway, MST1/2 and LATS1/2, are often observed to be downregulated in GC[20]. In this paper, we evaluated the potential function of miRNA-93-5p in regulating cellular behaviors of SGC-7901 and HGC-27 cells. Through a series of functional experiments, we clarified the potential role of Hippo pathway influenced by miRNA-93-5p in the progression of GC, which provided novel directions in clinical treatment of GC.

Materials and Methods

Cell Culture

Cell recovery: SGC-7901 and HGC-27 cells were taken out from liquid nitrogen, placed in the 37°C water bath for rapid dissolution. Cell suspension was placed in a 15 ml centrifuge tube with 5 mL of Roswell Park Memorial Institute-1640 (RPMI-1640) (HyClone, South Logan, UT, USA) medium containing 10% fetal bovine serum (FBS) (HyClone, South Logan, UT, USA), and centrifuged at 1000 rpm/min for 4 min. Subsequently, the precipitate was re-suspended in 5 mL of RPMI-1640 medium and cultured in a 37°C, 5% CO_2 incubator. Cell passage: cell passage was performed at 80% of confluence. Briefly, cells were washed with phosphate-buffered saline (PBS) for three times, and digested in 2 mL of trypsin containing 0.25% ethylenediaminetetraacetic acid (EDTA) (HyClone, South Logan, UT, USA). During the digestion, cells were maintained in the 37°C, 5% CO_2 incubator for 3-4 min. The same amount of medium was applied to terminate cell digestion after cells became round and floated observed under the microscope. Cells were gently blown to single cell suspension. Cell cryopreservation: until 80-90% of confluence, cells were centrifuged and the precipitate was suspended in 1 mL of cell cryopreservation solution. Cells preserved in cryotubes and placed at 4°C for 30 min, -20°C for 45 min to 2 h and -80°C for long-term preservation.

Transfection

Cells were seeded in the 6-well plate one day prior to transfection. MiRNA-93-5p mimics, inhibitor or NC was diluted in OPTI modified eagle medium (MEM), and subsequently mixed with Lipofectamine™ 3000 (diluted in OPTI MEM as well) (Invitrogen, Carlsbad, CA, USA). The mixture was applied into each well until 40-60% of confluence, and cultured for 4-6 h in OPTI-MEM. Fresh medium containing 10% fetal bovine serum (FBS) was replaced for another 12 h.

RNA Extraction and Quantitative Real-Time Polymerase Chain Reaction (qRT-PCR)

Total ribonucleic acid (RNA) was extracted from cells using TRIzol (Invitrogen, Carlsbad, CA, USA) reagent. Purity and concentration of the extracted RNA were determined using a spectrophotometer. The complementary deoxyribonucleic acid (cDNA) was obtained by reverse transcription of RNA and subsequently amplified by PCR. The mRNA sequences of miRNA-93-5p and glyceraldehyde 3-phosphate dehydrogenase (GAPDH) were searched from GenBank and relevant primers were designed using Primer 5.0 Software. Relative level was calculated by $2^{-\Delta\Delta Ct}$. Primer sequences used in this study were as follows: CDX2, F: 5'-GGGAATTCTTTCCTCTCCTTTGCTCTGCGG-3', R: 5'-GGGAATTCGCGCCCCTGGCAGCCTCCAACG-3'; miRNA-93-5p, F: 5'-CAGCACTCCATATCTCTCTAT-3', R: 5'-CTTCATCAGCGTCAACAG-3'; FOXM1, F: 5'-GAGCACTTGGAATCACAGCA-3', R: 5'-CACCGGGAACTGGATAGGTA-3'; CTGF, F: 5'-CGACTGGAAGACACGTTTGG-3', R: 5'-AGGCTTGGAGATTTTGGGAG-3'; CYR61, F: 5'-ACTTCATGGTCCCAGTGCTC-3', R: 5'-AAATCCGGGTTTCTTTCACA-3'; U6: F: 5'-GCTTCGGCAGCACATATACTAAAAT-3', R: 5'-CGCTTCAGAATTTGCGTGTCAT-3'; GAPDH: F: 5'-CGCTCTCTGCTCCTCCTGTTC-3', R: 5'-ATCCGTTGACTCCGACCTTCAC-3'.

Cell Viability Determination

Cells were seeded in a 96-well plate (Sigma-Aldrich, St. Louis, MO, USA) with 2.5×10^3 cells per well. Each group had 5 replicate wells. At the appointed time points, 10 μL of cell counting kit-8 (CCK-8) solution (Dojindo, Kumamoto, Japan) was applied per well. After incubation for 2 h, the recorded absorbance at 450 nm using a microplate reader was used for plotting the growth curve using GraphPad Prims 5 (GraphPad Prism, Version X; La Jolla, CA, USA).

Colony Formation Assay

Cells were prepared for suspension and seeded in the 6-well plate with 300 cells per well. Cells were incubated with 4 μM cisplatin or 5 μM 5-FU for 15 days. By fixation in methanol for 15 min and dye with 0.4% violet crystal for 20 min, colonies were captured for counting under a microscope (Leica, Wetzlar, Germany).

Transwell Migration and Invasion Assays

2×10^4 cells suspended in 400 μL of serum-free medium and 800 μL of complete medium with 15% fetal bovine serum (FBS) were supplied on the basolateral and apical chamber, respectively. 12 h later, transwell chamber (Invitrogen, Carlsbad, CA, USA) with 8.0 μm pore size was taken out and the medium was removed. Cell fixation using methanol for 10 min and 0.4% crystal violet dye was performed. Images were taken under a microscope for counting migratory cells. Transwell invasion assay was conducted in the same way as the above mentioned except for 100 μL of matrigel pre-coating in transwell chamber.

Statistical Analysis

Statistical Product and Service Solutions (SPSS) 18.0 (SPSS Inc., Chicago, IL, USA) was used for all statistical analysis. Data were represented as mean ± SD (Standard Deviation). The *t*-test or Fisher exact test was used for analyzing intergroup differences. $p<0.05$ indicated the significant difference.

Results

MiRNA-93-5p Influenced Proliferative Ability of GC

To examine the potential role of miRNA-93-5p in biological behaviors of GC, miRNA-93-5p mimics and inhibitor were constructed. MiRNA-93-5p was upregulated in SGC-7901 cells transfected with miRNA-93-5p mimics, which was downregulated in HGC-27 cells transfected with miRNA-93-5p inhibitor (Figure 1A, 1B). As CCK-8 assay indicated, SGC-7901 cells overexpressing miRNA-93-5p showed the accelerated viability (Figure 1C). On the contrary, HGC-27 cells with

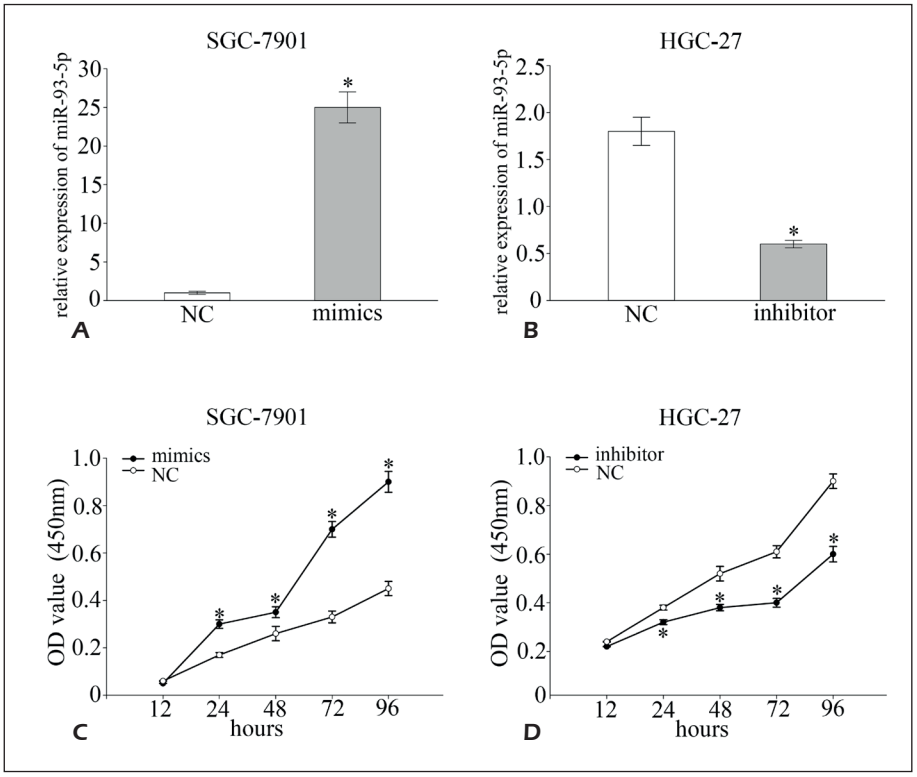

Figure 1. MiR-93-5p influenced proliferative ability of GC. **A**, Transfection efficacy of miR-93-5p mimics in SGC-7901 cells. **B**, Transfection efficacy of miR-93-5p inhibitor in HGC-27 cells. **C**, CCK-8 assay depicted viability of SGC-7901 cells transfected with miR-93-5p mimics or NC. **D**, CCK-8 assay depicted viability of HGC-27 cells transfected with miR-93-5p inhibitor or NC. *$p<0.05$, compared with control.

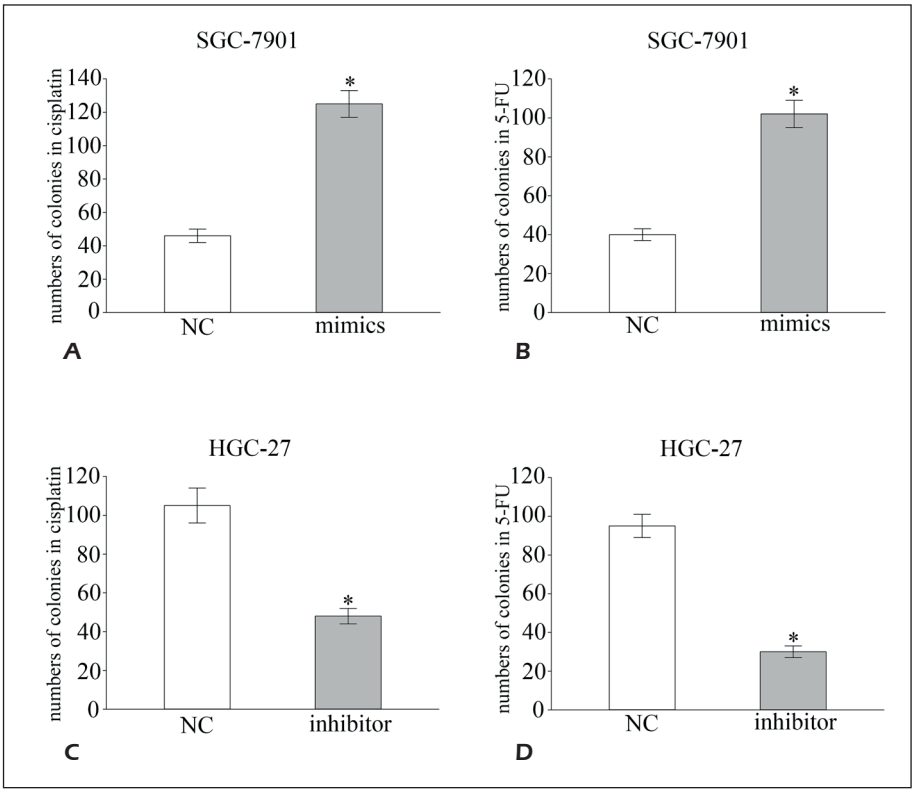

Figure 2. MiR-93-5p influenced clonality of GC. **A**, Numbers of colonies in cisplatin-induced SGC-7901 cells transfected with miR-93-5p mimics or NC. **B**, Numbers of colonies in 5-FU-induced SGC-7901 cells transfected with miR-93-5p mimics or NC. **C**, Numbers of colonies in cisplatin-induced HGC-27 cells transfected with miR-93-5p inhibitor or NC. **D**, Numbers of colonies in 5-FU-induced HGC-27 cells transfected with miR-93-5p inhibitor or NC. *$p<0.05$, compared with control.

miRNA-93-5p knockdown presented decreased viability relative to controls (Figure 1D).

MiRNA-93-5p Influenced Clonality of GC

Colony formation assay demonstrated that after cisplatin induction, more colonies were observed in SGC-7901 cells overexpressing miRNA-93-5p relative to controls (Figure 2A). Similarly, transfection of miRNA-93-5p mimics in 5-FU-induced SGC-7901 cells increased clonality than those transfected with NC (Figure 2B). In HGC-27 cells induced with cisplatin or 5-FU, the number of colonies decreased by miRNA-93-5p knockdown (Figure 2C, 2D).

MiRNA-93-5p Influenced GC Cells to Migrate and Invade

Potential effects of miRNA-93-5p on regulating migratory and invasive abilities of GC were examined by transwell assay. In SGC-7901 cells, miRNA-93-5p overexpression accelerated cells to migrate and invade (Figure 3A). Conversely, downregulation of miRNA-93-5p in HGC-27 cells inhibited migratory and invasive abilities (Figure 3B).

MiRNA-93-5p Influenced Hippo Pathway in GC

Here, we examined expressions of key genes in Hippo pathway by qRT-PCR. Transfection of miRNA-93-5p mimics upregulated levels of CDX2, FOXM1 and CTGF, but had no influence on CYR61 level (Figure 4A, 4B). By transfection of miRNA-93-5p inhibitor in HGC-27 cells, relative levels of CDX2, FOXM1 and CTGF were downregulated, while CYR61 level was upregulated (Figure 4C, 4D).

Discussion

Diagnostic rate of early-stage GC is far away from satisfy. Most of GC patients are in advanced stage at the first time of diagnosis, presenting rapid progression and poor prognosis. So far, the comprehensive etiology of GC has not been fully elucidated, and the effective prevention and treatment of GC are still lacked. Hence, it is necessary to develop novel biological hallmarks that contribute to diagnosis, treatment and prognosis of GC. As a type of endogenous, non-coding RNAs, miRNAs regulate gene expressions

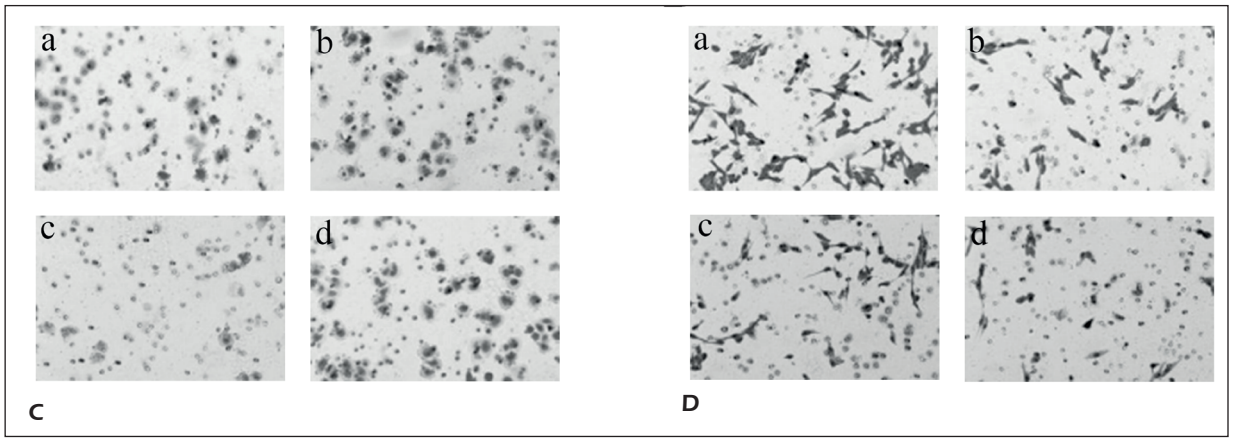

Figure 3. MiR-93-5p influenced migratory and invasive abilities of GC (Magnification, 100×). **A**, a. Migration of SGC-7901 cells transfected with NC. b. Migration of SGC-7901 cells transfected with miR-93-5p mimics. c. Invasion of SGC-7901 cells transfected with NC. d. Invasion of SGC-7901 cells transfected with miR-93-5p mimics. **B**, a. Migration of HGC-27 cells transfected with NC. b. Migration of HGC-27 cells transfected with miR-93-5p inhibitor. c. Invasion of HGC-27 cells transfected with NC. d. Invasion of HGC-27 cells transfected with miR-93-5p inhibitor.

and are closely related to the occurrence and development of tumors[21,22]. They may be potential markers for intervening the progression and prognosis of GC. For example, upregulation of miR-520c is significantly associated with adverse clinical response and prognosis in GC patients[23]. Downregulation of miR-34a could reduce tumor mass of GC[24]. MiR-34a is capable of inhibiting tumor metastasis and enhancing drug-resistance, and thereafter suppresses tumor progression. MRX34 synthesized based on miR-34a was the first miRNA synthetic drug tested in clinical tri-

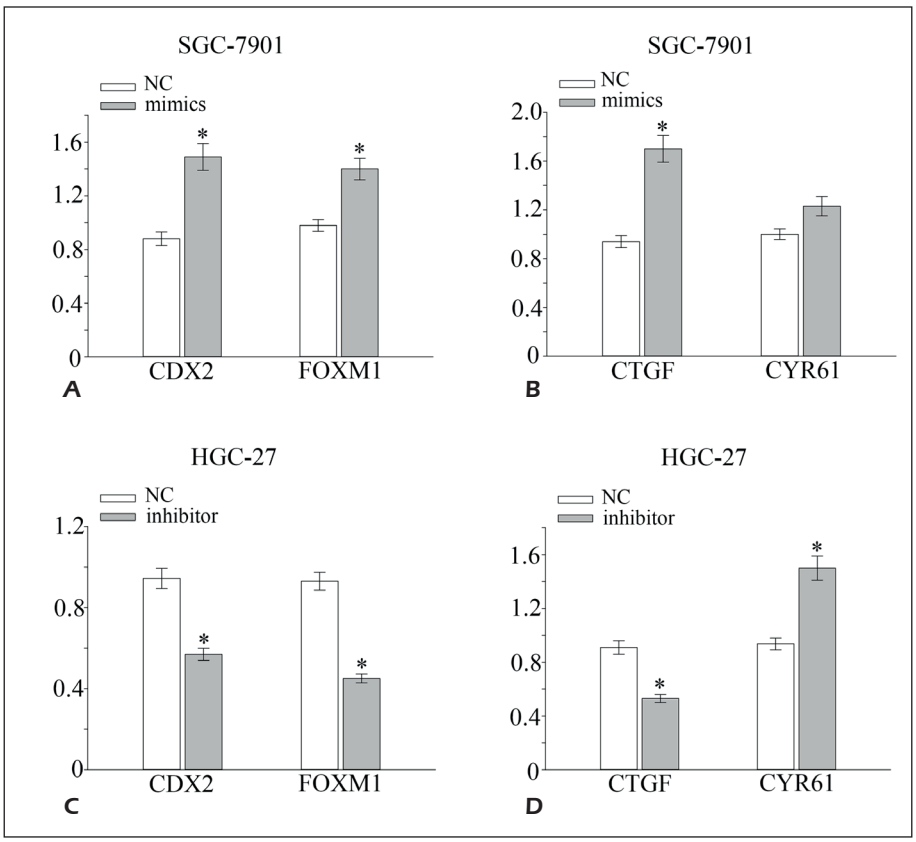

Figure 4. MiR-93-5p influenced Hippo pathway in GC. **A**, The mRNA levels of CDX2 and FOXM1 in SGC-7901 cells transfected with miR-93-5p mimics or NC. **B**, The mRNA levels of CTGF and CYR61 in SGC-7901 cells transfected with miR-93-5p mimics or NC. **C**, The mRNA levels of CDX2 and FOXM1 in HGC-27 cells transfected with miR-93-5p inhibitor or NC. **D**, The mRNA levels of CTGF and CYR61 in HGC-27 cells transfected with miR-93-5p inhibitor or NC. *p<0.05, compared with control.

als[25]. Therefore, miRNA intervention is expected to be a potential therapeutic approach for tumors. MiRNA-93-5p is upregulated in GC, serving as an oncogene that mediates tumor progression[26]. However, its biological function in GC has not been fully explored. In this study, miRNA-93-5p overexpression markedly accelerated GC cells to proliferate, migrate and invade. We suggested that miRNA-93-5p could be utilized as a potential target for clinical treatment of GC. Hippo pathway is reported to be able to inhibit tumorigenesis, tumor development and proliferation, as well as induce apoptosis[27]. This pathway mainly consists of a core inhibitory kinase cascade (MST1/2 and LATS1/2), which phosphorylates the transcriptional coactivator YAP[28]. Activated Hippo pathway induces YAP to translocate into the nucleus and interact with TEAD1-4, further accelerating growth and metastasis by activating downstream genes[29,30]. Many studies[31,32] have already reported the relationship between Hippo pathway and tumor progression. Inactivation of Hippo pathway would lead to upregulation of downstream genes CTGF, CYR61, FOXM1 and CDX2[33]. These key genes are considered to be potential therapeutic targets for GC[34]. Our results illustrated that miRNA-93-5p overexpression markedly upregulated CDX2, FOXM1 and CTGF in SGC-7901 cells. Conversely, knockdown of miRNA-93-5p in HGC-27 cells downregulated their levels. We believed that miRNA-93-5p inactivated Hippo pathway, thus promoting proliferative, migratory and invasive abilities of GC.

Conclusions

We found that miRNA-93-5p enhances proliferative, migratory and invasive abilities of GC cells by activating Hippo pathway, which may serve as a diagnostic and therapeutic target for GC.

Conflict of Interests

The authors declared no conflict of interest.

References

1) Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A. Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin 2018; 68: 394-424.
2) Ren J, Liu J, Sui X. Correlation of COX-2 and MMP-13 expressions with gastric cancer and their effects on prognosis. J BUON 2018; 23: 665-671.
3) Katoh H, Ishikawa S. Genomic pathobiology of gastric carcinoma. Pathol Int 2017; 67: 63-71.
4) Qi R, Wang DT, Xing LF, Wu ZJ. miRNA-21 promotes gastric cancer growth by adjusting prostaglandin E2. Eur Rev Med Pharmacol Sci 2018; 22: 1929-1936.
5) Cui W, Gu Z, Liu H, Zhang C, Liu J. Identification modules of gastric cancer based on protein-protein interaction networks and gene expression data. J BUON 2018; 23: 1013-1019.
6) Rupaimoole R, Slack FJ. MicroRNA therapeutics: towards a new era for the management of cancer and other diseases. Nat Rev Drug Discov 2017; 16: 203-222.
7) Shiotani A, Murao T, Kimura Y, Matsumoto H, Kamada T, Kusunoki H, Inoue K, Uedo N, Iishi H, Haruma K. Identification of serum miRNAs as novel non-invasive biomarkers for detection of high risk for early gastric cancer. Br J Cancer 2013; 109: 2323-2330.
8) Huang Z, Zhu D, Wu L, He M, Zhou X, Zhang L, Wang H. Six serum-based miRNAs as potential diagnostic biomarkers for gastric cancer. Cancer Epidemiol Biomarkers Prev 2017; 26: 188-196.
9) Ma DH, Li BS, Liu JJ, Xiao YF, Yong X, Wang SM, Wu YY, Zhu HB, Wang DX, Yang SM. miR-93-5p/IFNAR1 axis promotes gastric cancer metastasis through activating the STAT3 signaling pathway. Cancer Lett 2017; 408: 23-32.
10) Tang Q, Zou Z, Zou C, Zhang Q, Huang R, Guan X, Li Q, Han Z, Wang D, Wei H, Gao X, Wang X. MicroRNA-93 suppress colorectal cancer development via Wnt/beta-catenin pathway downregulating. Tumour Biol 2015; 36: 1701-1710.
11) Xue X, Wang X, Zhao Y, Hu R, Qin L. Exosomal miR-93 promotes proliferation and invasion in hepatocellular carcinoma by directly inhibiting TIMP2/TP53INP1/CDKN1A. Biochem Biophys Res Commun 2018; 502: 515-521.
12) Huang H, Wang X, Wang C, Zhuo L, Luo S, Han S. The miR-93 promotes proliferation by directly targeting PDCD4 in hepatocellular carcinoma. Neoplasma 2017; 64: 770-777.
13) Ji C, Liu H, Yin Q, Li H, Gao H. miR-93 enhances hepatocellular carcinoma invasion and metastasis by EMT via targeting PDCD4. Biotechnol Lett 2017; 39: 1621-1629.
14) Ohta K, Hoshino H, Wang J, Ono S, Iida Y, Hata K, Huang SK, Colquhoun S, Hoon DS. MicroRNA-93 activates c-Met/PI3K/Akt pathway activity in hepatocellular carcinoma by directly inhibiting PTEN and CDKN1A. Oncotarget 2015; 6: 3211-3224.
15) Ansari MH, Irani S, Edalat H, Amin R, Mohammadi Roushandeh A. Deregulation of miR-93 and miR-143 in human esophageal cancer. Tumour Biol 2016; 37: 3097-3103.
16) Qu MH, Han C, Srivastava AK, Cui T, Zou N, Gao ZQ, Wang QE. miR-93 promotes TGF-beta-induced epithelial-to-mesenchymal transition through downregulation of NEDD4L in lung cancer cells. Tumour Biol 2016; 37: 5645-5651.

17) Plouffe SW, Hong AW, Guan KL. Disease implications of the Hippo/YAP pathway. Trends Mol Med 2015; 21: 212-222.
18) Ehmer U, Sage J. Control of proliferation and cancer growth by the hippo signaling pathway. Mol Cancer Res 2016; 14: 127-140.
19) Jiao S, Li C, Hao Q, Miao H, Zhang L, Li L, Zhou Z. VGLL4 targets a TCF4-TEAD4 complex to coregulate Wnt and Hippo signalling in colorectal cancer. Nat Commun 2017; 8: 14058.
20) Kang W, Cheng AS, Yu J, To KF. Emerging role of Hippo pathway in gastric and other gastrointestinal cancers. World J Gastroenterol 2016; 22: 1279-1288.
21) Rupaimoole R, Calin GA, Lopez-Berestein G, Sood AK. miRNA Deregulation in cancer cells and the tumor microenvironment. Cancer Discov 2016; 6: 235-246.
22) Wan LY, Deng J, Xiang XJ, Zhang L, Yu F, Chen J, Sun Z, Feng M, Xiong JP. miR-320 enhances the sensitivity of human colon cancer cells to chemoradiotherapy in vitro by targeting FOXM1. Biochem Biophys Res Commun 2015; 457: 125-132.
23) Li YR, Wen LQ, Wang Y, Zhou TC, Ma N, Hou ZH, Jiang ZP. MicroRNA-520c enhances cell proliferation, migration, and invasion by suppressing IRF2 in gastric cancer. FEBS Open Bio 2016; 6: 1257-1266.
24) Liu Y, Sun Y, Zhao A. MicroRNA-134 suppresses cell proliferation in gastric cancer cells via targeting of GOLPH3. Oncol Rep 2017; 37: 2441-2448.
25) Beg MS, Brenner AJ, Sachdev J, Borad M, Kang YK, Stoudemire J, Smith S, Bader AG, Kim S, Hong DS. Phase I study of MRX34, a liposomal miR-34a mimic, administered twice weekly in patients with advanced solid tumors. Invest New Drugs 2017; 35: 180-188.
26) Zhang R, Guo Y, Ma Z, Ma G, Xue Q, Li F, Liu L. Long non-coding RNA PTENP1 functions as a ceRNA to modulate PTEN level by decoying miR-106b and miR-93 in gastric cancer. Oncotarget 2017; 8: 26079-26089.
27) Gregorieff A, Wrana JL. Hippo signalling in intestinal regeneration and cancer. Curr Opin Cell Biol 2017; 48: 17-25.
28) Zygulska AL, Krzemieniecki K, Pierzchalski P. Hippo pathway - brief overview of its relevance in cancer. J Physiol Pharmacol 2017; 68: 311-335.
29) Wang G, Lu X, Dey P, Deng P, Wu CC, Jiang S, Fang Z, Zhao K, Konaparthi R, Hua S, Zhang J, Li-Ning-Tapia EM, Kapoor A, Wu CJ, Patel NB, Guo Z, Ramamoorthy V, Tieu TN, Heffernan,T, Zhao D, Shang X, Khadka S, Hou P, Hu B, Jin EJ, Yao W, Pan X, Ding Z, Shi Y, Li L, Chang Q, Troncoso P, Logothetis CJ, McArthur MJ, Chin L, Wang YA, DePinho RA. Targeting YAP-dependent MDSC infiltration impairs tumor progression. Cancer Discov 2016; 6: 80-95.
30) Guo L, Teng L. YAP/TAZ for cancer therapy: opportunities and challenges (review). Int J Oncol 2015; 46: 1444-1452.
31) Li L, Zhao J, Huang S, Wang Y, Zhu L, Cao Y, Xiong J, Deng J. MiR-93-5p promotes gastric cancer-cell progression via inactivation of the Hippo signaling pathway. Gene 2018; 641: 240-247.
32) Qiao Y, Li T, Zheng S, Wang H. The Hippo pathway as a drug target in gastric cancer. Cancer Lett 2018; 420: 14-25.
33) Deng J, Lei W, Xiang X, Zhang L, Lei J, Gong Y, Song M, Wang Y, Fang Z, Yu F, Feng M, Sun Z, Chen J, Zhan Z, Xiong J. Cullin 4A (CUL4A), a direct target of miR-9 and miR-137, promotes gastric cancer proliferation and invasion by regulating the Hippo signaling pathway. Oncotarget 2016; 7: 10037-10050.
34) Ma Y, Yang Y, Wang F, Wei Q, Qin H. Hippo-YAP signaling pathway: a new paradigm for cancer therapy. Int J Cancer 2015; 137: 2275-2286.