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IGF2BP1 Promotes Proliferation and Metastasis of Liver Cancer by Regulating CBX6 mRNA Stability

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Abstract

The aim of our study was to elucidate the mechanism of action of the Insulin-like Growth Factor 2 mRNA-Binding Protein 1 (IGF2BP1) on the phenotype of the Hepatocellular Carcinoma (HCC). To gain insight into the mechanism of action of the IGF2BP1 on HCC, the IGF2BP1 shRNA sequences were transfected into HCC cells. The HCC phenotypes were transduced into HCC cells. The effects of IGF2BP1 on HCC were investigated through assays of cellular proliferation, invasion and migration. Next, we screened for possible molecular mechanisms from the TCGA database, and found a target gene Chromobox 6 (CBX6). The RIP-qPCR was used to detect the binding of the IGF2BP1 and CBX6 mRNA. We adjusted the attenuation of the mRNA of the CBX6 using quantitative Real-Time PCR (qRT-PCR). Furthermore, rescue experiments were used to further demonstrate that CBX6 is a target of IGF2BP1. The IGF2BP1 was upregulated in HCC, and promoted the malignant phenotype of HCC cell lines. Further exploration indicated that the IGF2BP1 binds directly to the CBX6 and inhibits its mRNA attenuation, suggesting that the IGF2BP1 impacts CBX6 mRNA stability. Thus, it can be concluded that the IGF2BP1 facilitated the HCC phenotypes by promoting the CBX6 mRNA stability.

Keywords: IGF2BP1; Hepatocellular carcinoma; RNA binding proteins; CBX6; mRNA stability

Introduction

Hepatocellular Carcinoma (HCC) is the second leading cause of tumor-related deaths in the world, accounting for 90% of all primary liver cancer [1]. There are approximately 850,000 new cases in worldwide each year [2]. It is well known that the risk factors for HCC include infection with hepatitis B and C virus, a large number of alcohol intake, aflatoxin B1, and metabolic liver disease [3]. Due to the high incidence of recurrence and metastases, patients with HCC usually have a poor prognosis, especially in advanced patients. Although the treatment of liver cancer has made great progress in recent decades, from interventional therapy, radical resection, liver transplantation to targeted therapy and immunotherapy, the prognosis of liver cancer patients is still not ideal [4]. The recurrence and metastasis rate are as high as 40% to 70% [5]. Therefore, it is urgent to develop new treatment strategies to reduce the mortality of hepatocellular carcinoma.

The insulin-like growth factor 2 mRNA-Binding Protein 1 (IGF2BP1) is the most significantly up-regulated RNA Binding Proteins (RBPs) in HCC which is a type of post-transcriptional regulator, affecting the stability of the transcript [6,7]. The N6-methyladenosine (m6A) is one of the most chemical modifications, and is highly detected in eukaryotic messenger RNA (mRNA). The m6A modification is mainly catalyzed by m6A methyltransferase, removed by m6A demethylase, and recognized by m6A binding protein. Recent studies have shown that IGF2BP1 could selectively bind to the mRNA modified by m6A methylation, and it has the function of interpreting the information of m6A modification [8]. IGF2BP1 is highly expressed in a wide range of embryonic tissues, is low or not expressed in adult normal tissues, and is re-expressed after malignant transformation, which indicates that IGF2BP1 plays an important role in tumor development [9,10]. As a post-transcriptional driving factor, IGF2BP1 promoted tumor development by inhibiting the attenuation of oncogene mRNA, including MYC and E2F [7,11]. The mechanism of IGF2BP1 in HCC remains to be explored; this article will further explore the effect of IGF2BP1 on HCC and its molecular mechanism on the basis of previous studies.

Chromobox 6 (CBX6) is one of chromobox protein family [12], which is a subunit of Polycomb Repressive Complex 1 (PRC1) mediating epigenetic gene repression and acts as a transcriptional regulator [13]. Current study demonstrated that CBX6 downregulated in breast cancer, CBX6 was silenced epigenetically by EZH2 in a PRC2-dependent manner, and overexpression of CBX6

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resulted in cell proliferation inhibition, induced the cell cycle G0/G1 phase arrest and dramatically suppressed the migration and invasion capacities [14]. In addition to, CBX6 as a major Polycomb-Group (PcG) protein silenced atrix Metalloproteinase 2 (MMP-2) in non-invasive mesothelioma cells, MMP-2 upregulation in invasive mesothelioma cells, which might be caused by CBX6 degradation [15]. Interestingly, CBX6 is highly expressed in HCC and promotes cell proliferation, migration, invasion and G1/S phase transition, furthermore CBX6 can accelerate the Epithelial-Mesenchymal Transition (EMT) process in HCC cells by upregulating the expression of snail and Zeb1 [16,17].

Material and Methods

Immunohistochemistry

Liver tumor tissue samples were dewaxed in xylene and hydrated with gradient ethanol, and incubated with primary antibody overnight at 4°C. After incubating with the secondary antibody at room temperature, the tissue was stained with diaminobenzidine. Use Image-Pro Plus 6.0 software to perform semi-quantitative analysis by multiplying the staining intensity by the area of positively stained cells. All patients have signed informed consent and the study was supported by the Ethics Committee of the First Affiliated Hospital of Chongqing Medical University.

Cell culture and transfections

Human liver cancer cell lines SK-Hep1, HepG2, Hepa3B, Huh7 and an immortalized normal liver cell line L02, were purchased from the Shanghai Cell Bank Type Culture Collection Committee (Shanghai, China) and stored at the First Affiliated Hospital of Chongqing Medical University. The HepG2, Hepa3B, Huh7 and L02 cells were cultured in DMEM (Gibco, Gaithersburg, MD, USA) and the SK-Hep1 cells in MEM (Gibco, Gaithersburg, MD, USA) with 10% fetal bovine serum (FBS) (PAN, Adenbach, Germany) and grown at 37°C in 5% CO_2 incubator. The short hairpin RNA in the pLVX-shRNA lentivirus targeting IGF2BP1, the sequences of shRNA oligonucleotides targeting IGF2BP1 were as follows: pLVX-shIGF2BP1-1, 5'-CAGGGAAGAATCTATGGCAAA-3'; pLVX-shIGF2BP1-2, 5'-CCTGAAGAAGGTAGAGCAAGA-3'; pLVX-shIGF2BP1-3, 5'-ACAGTAGAGAACTGTGAGCAA-3', which were bought from Genechem (Genechem, Shanghai, China). In accordance with the manufacturer's protocol, cells in logarithmic phase were transduced with lentivirus. Before measuring the transfected effectiveness by Reverse Transcription-quantitative Polymerase Chain Reaction (RT-qPCR) and Western blot, the transduced cells were selected for one week with puromycin to obtain stable transfection.

Quantitative real-time PCR

TRIzol reagent was employed to collect the total cellular RNA. According to protocol, reverse transcription was performed using PrimeScript RT kit (Takara, Dalian, China) to obtain cDNA. TB Green TM Premix Ex Taq (Takara, Dalian, China) was used to quantitatively detect the mRNA expression level of target genes. The qPCR conditions are as follows: Pre-denaturation at 95°C for 30 s, then 45 cycles at 95°C for 5 s, and then annealing and extension at 60°C for 30 s. GAPDH is used as an internal reference. Measure the relative mRNA expression of target genes by $2^{-\Delta\Delta}$Cq method. The primer sequences were listed as follows: IGF2BP1 forward 5'-CTGAAGATCCTGGCCCATAA-3', reverse 5'- AAGGTCTTGCAACGAGGAGA-3'. GAPDH forward 5'-GGAGCGAGAT CCCTCCAAAAT-3', reverse 5'-GGCTGTTGTCATACTTCTC ATGG-3'.

Western blot analysis

RITA buffer (Beyotime, Shanghai, China) was employed to extract total protein from cells, and the BCA protein assay kit (Beyotime, Shanghai, China) was used to quantitatively determine the extracted protein. The proteins were separated by 10% Sodium Dodecyl Sulfate-Polyacrylamide Gel Electrophoresis (SDS-PAGE). Incubate the protein-converted PVDF with the primary antibody overnight at 4°C, and then incubate them with the corresponding 1:5000 secondary antibody (Abbkine, CA, USA) for 1 h at room temperature. The bands were visually analyzed using the Bio-Rad gel imaging system (Bio-Rad, CA, USA) with western blotting kit (Advansta, Menlo Park, USA).

Colony formation assay

Inoculate the cells in a 6-well plate, adjusted the cell density to 2,000 in each well and cultured in an incubator with 37°C and 5% CO_2. After 14 days, the cells were fixed by 4% paraformaldehyde for 30 min, and stained with crystal violet for 30 min. Finally, the colonies were pictured and counted by Image 6.0.

Cell proliferation assay

2000 cells were inoculated in each well of a 96-well plate repeating 5 wells in each group, added 10 μl of CCK-8 reagent to each well at 0 h, 24 h, 48 h and 72 h, and measured the absorbance at 450 nm using Bio-Rad (Bio-Rad, CA, USA) microplate reader.

Wound healing assays

The cells in the logarithmic growth phase were seeded in a 6-well plate. The cells were scraped with a 200 μl pipette when the confluency was about 90%. After washing with PBS, the cells were cultured with serum-free medium for 24 h. The cell growth was observed under the microscope (Olympus, Chengdu, China).

Cell migration and invasion assays

10% serum culture medium was added to the lower chamber (Corning, NY, USA) and 100ul cell suspension containing 2×10^4 cells was added to the upper chamber. After 24 h, the cells were fixed with 4% paraformaldehyde for 30 min, stained with 0.1% crystal violet for 30 min. The numbers of migrated cells were observed using the microscope (Olympus, Chengdu, China). For invasion assays, inoculated the cells in the upper chamber with Matrigel (BD Biosciences, NJ, USA), and subsequent steps were similar to the migration assays.

Cell flow assays

For cell cycle assay, cells were digested with trypsin and fixed in 75% ethanol. The cells were incubated with 10 mg/ml RNase A (Sigma-Aldrich, Darmstadt, Germany) for 30 min and stained with propidium iodide (Beyotime, Shanghai, China). Cell-cycle was detected by flow cytometry (BD Biosciences, NJ, USA) and analyzed by FlowJo version 7.6 software (FlowJo LLC, Ashland, USA). For cellular apoptosis, cell culture supernatants were collected and cells were diluted in PBS. Cells were stained with propidium iodide (Beyotime, Shanghai, China) and annexin Vfluorescein isothiocyanate for 20 min in the dark. Apoptosis was examined by flow cytometry (BD Biosciences, NJ, USA). The apoptotic rate was assessed with BD FACS software.

RNA Immunoprecipitation (RIP)-qPCR

The RIP assay was conducted using the RNA immunoprecipitation Kit (P0101, GENESEED, Guangzhou, China) according to the manufacturer's protocol. The mRNA was fragmented with 1000 μl

Buffer A at 4°C for 10 min. Added 100 µl pretreated protein A+G beads to cell supernatant. After blocking the beads, added 5 ug of IGF2BP1 antibody (ab184305, Abcam, Shanghai, China and IgG antibody (CS200621, 17-10499, Merck Millipore, Billerica, MA, USA) to cell supernatant at 4°C for 2 h. After washing twice, added 450 ul cell lysate to each tube beads and react at 4°C for 2 h. The RNA-proteins complex bound to the beads is then eluted. The target RNAs were eluted from the immunoprecipitated complex and purified for the qPCR.

Measurement of mRNA stability

The cells were seeded in each well of 6-well plates, and treated with 5 µM actinomycin D when the confluency was 50%. Then the cells were collected at specified time points. The total RNA was extracted by TRIzol and analyzed by RT-qPCR. Since actinomycin D treatment results in transcription stalling, for each sample, at 0 h the value of the target RNA was 100%, the percentage of target RNA collected at specific time points is the target RNA/internal reference RNA [18]. The mRNA half-life is the linear regression equation, and the log percent ratio is based on the remaining target RNA remaining versus time.

Statistical analysis

All results were analyzed using SPSS 18.0 software and GraphPad Prism 9.0. The t-test and one-way analysis of variance (ANOVA) were using for statistical analysis, the difference was considered significant when values were $p < 0.05$.

Results

IGF2BP1 was overexpressed in HCC

We analyzed the HCC data in the TCGA database and found that compared with normal tissues; the expression of IGF2BP1 was considerably up-regulated in HCC tissues. We performed immunohistochemical analysis of IGF2BP1 expression in 39 HCC patients treated at the First Affiliated Hospital of Chongqing Medical University from 2016 to 2019, and found that IGF2BP1 expression in HCC tissues was significantly higher than that in adjacent normal tissues. The IGF2BP1 expressions in HCC cells line were higher than that in normal liver cell line at the mRNA level and protein level. The expression of IGF2BP1 in SK-Hep1 and Huh7 cells is higher; therefore these cell lines were used in the follow-up of our research experiment.

Upregulation of IGF2BP1 was associated with the poor prognosis of HCC

The multivariate Cox regression analysis of TCGA database showed that IGF2BP1 was an independent prognostic factor affecting the survival of HCC patients, and the clinical correlation analysis showed that the expression of IGF2BP1 was positively correlated with tumor grade. We further investigated a statistical analysis between the expression of IGF2BP1 and clinical characteristics of HCC patients, and the results showed that the expression of IGF2BP1 was related to TNM stage (P=0.024).

IGF2BP1 promoted the proliferation of HCC cells

We designed 3 different short hairpins RNA in lentivirus targeting IGF2BP1 to reduce the expression of IGF2BP1 in HCC cell lines. The transfected effectiveness was analyzed by RT-qPCR and WB. We selected two sequences with the lowest knockdown efficiency for subsequent experiments in each cell line. Then, Cell Counting Kit-8 (CCK8) assays and colony formation assay showed that the proliferation ability was obviously suppressed when IGF2BP1 was inhibited in SK-Hep1 and Huh7 cells. These data indicated that IGF2BP1 regulated the proliferation of HCC cells.

IGF2BP1 promoted the migration and invasion by upregulating EMT

We performed wound healing assays, transwell and matrigel assays to assess the effect of IGF2BP1 on migration and invasion. Results showed that knockdown of IGF2BP1 significantly inhibited wound healing, migration and invasion assays. Meanwhile, knockdown of IGF2BP1 altered the expression of EMT-related proteins including E-cadherin, N-cadherin and vimentin. These results implicated knockdown of IGF2BP1 inhibited invasion and migration through downregulating EMT proteins.

Downregulation of IGF2BP1 leads to cell cycle arrest and cell apoptosis

In order to explore the relationship between IGF2BP1 and cell cycle and apoptosis, we performed cell cycle and apoptosis experiments. Cell cycle assays showed that knockdown of IGF2BP1 decreased the ratio of cells in G1 phase, increased the ratio of cells in S phase compared with negative control. Cell apoptosis assays showed that knockdown of IGF2BP1 increased cell apoptosis in HCC. These results suggested that inhibition of IGF2BP1 induces cell cycle arrest and apoptosis.

IGF2BP1 regulates CBX6 expression at the posttranscriptional level

To gain further insights into the molecular mechanisms underlying IGF2BP1's cellular functions, we analyzed the RNA-seq expression profiles after the interference with IGF2BP1 expression (GSE116133) in Gene Expression Omnibus (GEO) database. In recent studies, IGF2BPs were reported to enhance the expression of MYC and other target transcripts in a m6A-dependent manner [19]. To elucidate whether the altered gene expression was ascribed to m6A methylation, we analyzed the m6A-seq expression profiles in liver cancer cell (GSE90642). It has been reported that IGF2BP1-CLIP hits were identified in the 3'UTR of target mRNA which suggested conserved regulation *via* this cis-element [9,20]. To clarify these m6A methylation modification were mainly located near the 3' Untranslated Region (3' UTR), we analyzed poly(A) RNA-Seq of liver cancer cells interferenced with IGF2BP1 (GSE146802). In order to further confirm the transcripts directly regulated by IGF2BP1, we analyzed IGF2BP1 RIP-seq expression profiles which was performed to determine the transcripts that bind to IGF2BP1 (GSE90639). At the intersection of RNA-seq, m6A-seq, RIP-seq and poly(A) RNA-Seq, one gene (CBX6) was screened, which indicated that IGF2BP1 may bind to the 3'UTR of this transcript and regulate its m6a level, resulting in changes in the expression of the transcript. In addition to, pan-cancer co-expression analysis showed that IGF2BP1 and CBX6 expression are positively correlated ($p<0.001$). To determine whether IGF2BP1 directly binds to the target mRNA, we performed a RIP-qPCR assay, which showed that the knockdown of the IGF2BP1 in the HCC cells significantly reduced the binding of CBX6 mRNA.

IGF2BP1 regulates CBX6 expression and stability

Since IGF2BP1 can regulate target RNA after transcription, whether its knockout affects the stability of CBX6 mRNA in liver cancer cells, we specifically blocked RNA Polymerase II transcription with alpha-amanitin as previously shown in other tumor entities [7,21]. As we thought, inhibition of IGF2BP1 led to CBX6 mRNA

expression decrease. For SK-Hep1 in control group the half-life of CBX6 was ~5.30 h, the half-life in IGF2BP1 knockdown groups were ~3.48 h and ~3.02 h. For Huh7 in control group the half-life of CBX6 was ~6.38 h, the half-life in IGF2BP1 knockdown groups were ~3.98 h and ~4.67 h. Thus, our data indicate that IGF2BP1 associated with CBX6 mRNA, promoted CBX6 mRNA stability at the posttranscriptional level.

Inhibit CBX6 can partially rescue the malignant phenotype of IGF2BP1 knockdown

PcG is an important epigenetic regulatory protein that can maintain transcriptional inhibition. CBX6 is an important part of Polycomb protein and can read the key epigenetic mark, H3K27ME3. We found that knockdown of IGF2BP1 decreased CBX6 protein expression. CBX6 has been verified to be overexpressed and contributes to tumor progression in HCC [17]. To further characterize whether expression of CBX6 could partially rescue the proliferation rate of the malignant phenotype in IGF2BP1 knockdown cells, we establish overexpression of CBX6 by small interfering RNA, CBX6 overexpression efficiency was confirmed at both the mRNA and protein levels. The results showed that overexpression of CBX6 can partially rescue the proliferation rate, migration, invasion and cell cycle in IGF2BP1 knockdown cells. Together, these data indicate that CBX6 is a downstream target of IGF2BP1 and serves as a tumor promoter in HCC.

Discussion

The occurrence of liver cancer is a gradual process involving many factors, including heredity, epigenetics and genetic modification. IGF2BP1 is the most expressed RBP in liver cancer, which can promote tumor proliferation by stabilizing c-MYC and Ki67 mRNA [7]. In addition, IGF2BP1 has been reported as a post-transcriptional driver of tumor cell proliferation, migration, metastatic potential and therapeutic resistance [22,23]. IGF2BP1 has been confirmed to increase expression in lung cancer, esophageal cancer, breast cancer and other tumors [24], and can promote tumor cell proliferation, migration and invasion. These suggest that IGF2BP1 is an important protumorigenic factor. In this study, we further explored the role of IGF2BP1 in liver cancer. Analysis the result of a large sample of bioinformatics database shows that IGF2BP1 is significantly up-regulated in HCC. Our verification also meets this conclusion. At a cellular level, knockdown of IGF2BP1 significantly inhibited proliferation, migration, invasion, cell cycle and apoptosis of HCC cells.

Due to the m6A methylation modification function of IGF2BP1, we further explored its molecular mechanism for regulating liver cancer. According to the different array sequencing results in the GEO database, we identified a liver cancer promoter, CBX6, as a direct downstream target of IGF2BP1, which is modified at the 3'UTR in a m6A methylation dependent manner. CBX6 is a subunit of multiple bacteria suppression complex 1 (PRC1), which mediates epigenetic gene suppression. CBX6 is down-regulated in Glioblastoma Multiform (GBM), and its overexpression results in cell growth arrest [25]. In addition, CBX6 is significantly down-regulated in human breast cancer, it can inhibit cell proliferation and induce G0/G1 phase arrest, and overexpression of CXB6 reduces breast cancer cell migration and invasion rate [14]. However, CBX6 expression tends to increase in HCC tissues; its high expression promotes cell proliferation and enhances the invasion and metastasis ability of HCC cells through regulating transcription factors snail/zeb1-mediated EMT mechanism [16]. The mRNA attenuation experiment supported the result; IGF2BP1 promotes the stability of CBX6 mRNA, modified at post-transcriptional level. Previous studies have found that CBX6 has the ability to promote the proliferation and metastasis of liver cancer [17]. We found that after suppressing transcription with a transcription inhibitor, the attenuation of CBX6 was significantly faster when IGF2BP1 was knockdown. The results of rescue experiment showed that overexpression of CBX6 can partially rescue the malignant phenotypes after IGF2BP1 knockdown. Therefore, IGF2BP1 and CBX6 co-expression promotes HCC progression.

The EMT process results in epithelial cells lose cell polarity, lose their connection with basement membrane and other epithelial phenotypes, and obtain mesenchymal phenotypes such as high migration and invasion, anti-apoptosis and degradation of extracellular matrix [26,27]. However, during the development of cancer, this process is hijacked by tumor cells [28], which eventually lead to cancer cell invasion and distant metastasis. Our experiment proved for the first time that IGF2BP1 can affect the invasion of HCC through EMT. Furthermore, study has shown that IGF2BP1 leads to the decrease of E-cadherin mRNA expression in colon cancer [29]. The above results indicated that IGF2BP1 can promote tumor invasion through EMT phenotype. Coincidentally, CBX6 as the target of IGF2BP1 could regulate the expression of the transcription factor snail/Zeb1B to mediate EMT in HCC [16]. In addition, IGF2BP1 can also decrease the miRNA-directed inhibition of SRF mRNA expression in an n6-methyladenosine m6A-dependent manner, resulting in enhanced SRF-dependent transcriptional activity, thereby promoting tumor cell invasion [9]. Taken together, IGF2BP1 participates in the EMT of liver cancer cells and plays an important role.

Conclusion

We confirmed for that the expression of IGF2BP1 is upregulated in HCC tissues and cell lines, and the expression of IGF2BP1 is related to the survival of patients. In addition, we found that IGF2BP1 gene knockout can inhibit the proliferation and enhance invasion of HCC cells. Most importantly, our results gave new insights into the role of IGF2BP1 in epithelial-mesenchymal transition of HCC, as a binding protein of m6A RNA methylation modification; IGF2BP1 participates in a variety of mRNA biological functions, and has an important regulatory effect on m6A site recognition. In this experiment, we did not elaborate on the mechanism of IGFBP1 m6A RNA methylation to modify mRNA, and we will focus on this part of the content in our next research.

References

1. Forner A, Reig M, Bruix J. Hepatocellular carcinoma. Lancet. 2018;391(10127):1301-14.
2. Craig AJ, Felden JV. Tumour evolution in hepatocellular carcinoma. Nat Rev Gastroenterol Hepatol. 2020;17(3):139-52.
3. Kulik L, El-Serag HB. Epidemiology and management of hepatocellular carcinoma. Gastroenterology. 2019;156(2):477-91.
4. Llovet JM, Montal R, Sia D, Finn RS. Molecular therapies and precision medicine for hepatocellular carcinoma. Nat Rev Clin Oncol. 2018;15(10):599-616.
5. Khemlina G, Ikeda S, Kurzrock R. The biology of Hepatocellular carcinoma: Implications for genomic and immune therapies. Mol Cancer. 2017;16:149.

6. Müller-McNicoll M, Neugebauer KM. How cells get the message: Dynamic assembly and function of mRNA-protein complexes. Nat Rev Genet. 2013;14(4):75-87.
7. Gutschner T, Hämmerle M, Pazaitis N, Bley N, Fiskin E, Uckelmann H, et al. Insulin-like growth factor 2 mRNA-binding protein 1 IGF2BP1. Is an important protumorigenic factor in hepatocellular carcinoma. Hepatology. 2014;59(5):1900-11.
8. Huang H, Weng H, Wnag S, Qin X, Shi H, Wu H, et al. Recognition of RNA N6.-methyladenosine by IGF2BP proteins enhances mRNA stability and translation. Nat Cell Biol. 2018;20(3):285-95.
9. Müller S, Bley N, Glaß M, Busch B, Rousseau V, Misiak D, et al. IGF2BP1 enhances an aggressive tumor cell phenotype by impairing miRNA-directed downregulation of oncogenic factors. Nucleic Acids Res. 2018;46(12):6285-303.
10. Zirkel A, Lederer M, Stöhr N, Pazaitis N, Hüttelmaier S. IGF2BP1 promotes mesenchymal cell properties and migration of tumor-derived cells by enhancing the expression of LEF1 and SNAI2 (SLUG). Nucleic Acids Res. 2013;41(13):6618-36.
11. Müller S, Bley N, Busch B, Glaß M, Lederer M, Misiak C, et al. The oncofetal RNA-binding protein IGF2BP1 is a druggable, post-transcriptional super-enhancer of E2F-driven gene expression in cancer. Nucleic Acids Res. 2020;48(15):8576-90.
12. Chan HL, Morey L. Emerging roles for polycomb-group proteins in stem cells and cancer. Trends Biochem Sci. 2019;44(8):688-700.
13. Ma RG, Zhang Y, Sun TT, Cheng B. Epigenetic regulation by polycomb group complexes: Focus on roles of CBX proteins. J Zhejiang Univ Sci B. 2014;15(5):412-28.
14. Deng H, Guan X, Gong L, Zeng J. CBX6 is negatively regulated by EZH2 and plays a potential tumor suppressor role in breast cancer. Sci Rep. 2019;9:197.
15. Sakai K, Nishiuchi T. Proteasomal degradation of polycomb-group protein CBX6 confers MMP-2 expression essential for mesothelioma invasion. Sci Rep. 2020;10:16678.
16. Wang J, He H, Jiang Q, Wang Y, Jia S. CBX6 promotes HCC metastasis *via* transcription factors snail/zeb1-mediated EMT mechanism. Onco Targets Ther. 2020;13:12489-500.
17. Zheng H, Jiang WH, Tao T, Tan HS, Chen Y, Qiao GL, et al. CBX6 overexpression contributes to tumor progression and is predictive of a poor prognosis in hepatocellular carcinoma. Oncotarget. 2017;8(12):18872-84.
18. Essafi-Benkhadir K, Pouysségur J, Pagès G. Implication of the ERK pathway on the post-transcriptional regulation of VEGF mRNA stability. Methods Mol Biol. 2010;661:451-69.
19. Zhu S, Wang JZ, Chen D, He YT, Meng N, Chen M, et al. An oncopeptide regulates m6. A recognition by the m6. A reader IGF2BP1 and tumorigenesis. Nat Commun. 2020;11(1):1685.
20. Lan T, Li H, Zhang D, Liu HL, Hao XY, Yan XK, et al. KIAA1429 contributes to liver cancer progression through N6-methyladenosine-dependent post-transcriptional modification of GATA3. Mol Cancer. 2019;18:186.
21. Weidensdorfer D, Stöhr N, Baude A, Lederer M, Köhn M, Schierhorn A, et al. Control of c-myc mRNA stability by IGF2BP1-associated cytoplasmic RNPs. RNA. 2009;15(1):104-15.
22. Köbel M, Weidensdorfer D, Reinke C, Lederer M, Schmitt WD, Zeng K, et al. Expression of the RNA-binding protein IMP1 correlates with poor prognosis in ovarian carcinoma. Oncogene. 2007;26(54):7584-9.
23. Bell JL, Wächter K, Mühleck B, Pazaitis N, Köhn M, Lederer M, et al. Insulin-like growth factor 2 mRNA-binding proteins IGF2BPs: post-transcriptional drivers of cancer progression? Cell Mol Life Sci. 2013;70(15): 2657-75.
24. Huang X, Zhang H, Guo X, Zhu Z, Cai H, Kong X. Insulin-like growth factor 2 mRNA-binding protein 1 IGF2BP1 in cancer. J Hematol Oncol. 2018;11(1):88.
25. Li G, Warden C, Zou Z, Neman J, Krueger JS, Jain A, et al. Altered expression of polycomb group genes in glioblastoma multiform. PLoS One. 2013;8(11):e80970.
26. Ramesh V, Brabletz T, Ceppi P. Targeting EMT in cancer with repurposed metabolic inhibitors. Trends Cancer. 2020;6(11):942-50.
27. Giannelli G, Koudelkova P, Dituri F, Mikulits W. Role of epithelial to mesenchymal transition in hepatocellular carcinoma. J Hepatol. 2016;65(4):798-808.
28. Derynck R, Weinberg RA. EMT and cancer: More than meets the eye. Dev Cell. 2019;49(3):313-6.
29. Hamilton KE, Noubissi FK, Katti PS, Hahn CM, Davey SR, Lundsmith ET, et al. IMP1 promotes tumor growth, dissemination and a tumor-initiating cell phenotype in colorectal cancer cell xenografts. Carcinogenesis. 2013;34(11):2647-54.