Clinics of Oncology

Research Article

ISSN: 2640-1037 |Volume 6

IRF5 Promotes the Progression of Hepatocellular Carcinoma and is Regulated by Trim35

Ying F[1]*, Zhihui L[2]*, Bangzhong L[2], Nan L[3], Mingzhen Y[2] and Ping W[2]

[1] Department of Gastroenterology and Hepatology, Zhongshan Hospital, Fudan University, Shanghai 200032, China

[2] Department of Rehabilitation, Zhongshan Hospital, Fudan University, Shanghai 200032, China

[3] Department of Hepatic Surgery I (Ward I), Shanghai Eastern Hepatobiliary Surgery Hospital, Shanghai, 201805, China

***Corresponding author:**

Ying F and Zhihui L
Department of Rehabilitation,
Zhongshan Hospital, Fudan University, 180 Fenglin Rd, Shanghai 200032, PR China.
Tel: +86-021-64041990,
E-mail: wang.ping8@zs-hospital.sh.cn.

Received: 30 May 2022
Accepted: 07 Jun 2022
Published: 13 Jun 2022
J Short Name: COO

Citation:

Ying F and Zhihui L, IRF5 Promotes the Progression of Hepatocellular Carcinoma and is Regulated by Trim35. Clin Onco. 2022; 6(7): 1-8

Keywords:

IRF5; HCC; TRIM35; Cell growth

1. Abstract

The IRF family of proteins involves in the tumor progression. However, but the functions of IRF5 in the tumorigenesis are largely unknown. Here, IRF5 was found to be up-regulated in hepatocellular carcinoma (HCC). Interfering with IRF5 inhibited the growth and tumorigenic ability of HCC cells. When studying the molecular mechanism, it was found that TRIM35 interacted with IRF5, promoting the ubiquitination and degradation of IRF5. In the clinical specimens of HCC, TRIM35 was negatively correlated with the expression of IRF5. These observations reveal the oncogenic function of IRF5 in the progression of HCC, suggesting that IRF5 is a promising target for the therapy of HCC.

2. Introduction

HCC is frequently observed worldwide [1, 2]. The five-year survival rate remains low, even with comprehensive treatment involving chemotherapy, radio-therapy and immunotherapy [3-5]. Further research on the molecular mechanism of hepatocarcinogenesis is of great significance in identifying a new target for HCC treatment. IRF (interferon regulator factor) refers to a group of multifunctional transcription factors which can specifically recognize and interact with the interferon-stimulated responsive element (ISRE) in the IFN promoters and the interferon-stimulated gene (ISG) [6]. This activates the relevant signal and regulates the expression of the target gene [7,8]. The result is that IRF should be able to play a biological role in antiviral infection, congenital immunity, adaptive immunoregulation, cell proliferation, cell apoptosis and cell homeostasis [9-11].

IRF is often abnormally expressed in tumors. The expression of IRF1 is down-regulated in colorectal carcinoma (CRC) [12,13]. The overexpression of IRF1 in CRC cells inhibits cell growth, promotes apoptosis and causes radiotherapeutic sensitivity [12,14,15]. More interestingly, the members of the IRF family regulate each other during the tumorigenesis process. In the esophageal squamous cell carcinoma (ESCC) samples, IRF1 was down-regulated while IRF2 was overexpressed; IRF2 inhibited IRF1 from entering the nucleus, and they cooperated to promote the progress of the esophageal carcinoma [16]. In innate immune, IRF1 promotes innate immune by activating IRF3 [17]. The role of IRF5 in tumors is poorly understood. Previous study shows that IRF5 can promote the growth of thyroid carcinoma cells [18]. IRF5 can inhibit HCV replication and HCV-related hepatocarcinogenesis [19], but in the HPA (Human Protein Atlas) database, the abundance of IRF5 is inversely correlated with the outcome of HCC. Therefore, elucidating the role of IRF5 in HCC from a cell-biological perspective is of great significance.

TRIM35 is an E3 ligase and a suppressor in many types of tumors [20-23]. In HCC, multiple microRNAs promote the proliferation of HCC cells by targeting TRIM35 [21]. TRIM35 can interact with

PKM2 to inhibit the glycolysis and tumorigenic ability of HCC cells [23,24]. The expression of TRIM35 and PKM2 in HCC predicts the prognosis for HCC patients [23,24]. To further elucidating the functions of TRIM35 in HCC, the proteins interacting with TRIM35 were predicted using the STRING database. IRF5 is one of the potential proteins to interact with TRIM35. In the present work, the abundance of IRF5 in HCC was studied. In addition, the role of IRF5 in HCC cells and the regulating effect of TRIM35 on IRF5 were also examined.

3. Materials and Methods

3.1. Cell culture

The HepG2, Huh7 and QGY cells were obtained from the Cell Bank, Chinese Academy of Sciences. The cells were cultured in DMEM medium with 10% fetal bovine serum (FBS) and antibiotics (100 U/mL of penicillin and 100 µg/ml of streptomycin). Cells were in a constant temperature incubator (5% CO_2, 37℃). Lipofectamine 8000 was used for the cell transfection.

3.2. Clinical samples

The clinical samples were collected from the Shanghai Eastern Hepatobiliary Surgery Hospital after the study was approved by their Ethics Committee. All of the patients have signed the consent form.

3.3. Immunohistochemistry (IHC)

The array of HCC tissues was provided by the Shanghai Eastern Hepatobiliary Surgery Hospital. Dewaxing, rehydration and antigen recovery in the 100°C EDTA solution were done. Then, the activity of endogenous peroxidase was blocked. Before being incubated with the IRF5 antibody (Sigma, HPA046700, 1:100) at 4°C overnight, the tissue sections were washed with PBS. On the next day, before being incubated with secondary antibody at room temperature for 1h, the tissue sections were washed with PBS. The signal was developed using 3,3,0-diaminobenzidine (DAB), and the nucleus were stained using hematoxylin. Both the staining intensity and protein expression level were automatically scored by the Vectra 2.0 system.

3.4. qPCR

RNA was extracted with TRIzol (Invitrogen) and reversely transcribed into cDNA using the PrimeScript™ RT kit (Takara) according to the instructions. qPCR was performed using The SYBR Green kit and CFX96 real-time fluorescent quantitative PCR detection system (Bio-Rad, Richmond, CA, USA). The levels of the transcripts were calculate using the $2^{-\Delta\Delta}$Ct method. The primer sequences for IRF5 were: Forward primer, 5'-tgtgcccttaacaagagccg-3'; Reverse primer, 5'-ctctgtgaggctcaggcttg-3'.

3.5. Western blot

The cells were cleaned twice with PBS, and lysed on ice with the RIPA Lysis buffer, which contains the protease inhibitor and phosphatase inhibitor. The supernatant was collected after the cell lysis was centrifuged, and the protein concentration was quantified using the BCA protein detection kit. An equal amount of protein was taken for the SDS-PAGE analysis. After separation, the protein was transferred onto the PVDF membrane and incubated with a specific primary antibody at 4°C overnight. Then, the membrane was incubated with the HRP-conjugated secondary antibody for 1-2h. The immune signal was detected with a chemiluminescence reagent (Milliwell, WBKLS0050), and analyzed with Image Lab software. The primary antibodies used in this experiment were as follows: IRF5 (Sigma, HPA046700, 1:100), tubulin (Santa Cruz Biotechnolog, sc-5286, 1:4000), Flag (Sigma, F9291; 1:3,000), HA(Sigma, H3663, 1:2000), TRIM35 (Sigma, HPA019647, 1:100), Ubiquitin (proteintech, 10201-2-AP, 1:1000).

3.6. CCK8

Cells were seeded into a 96-well plate, and each well contained 1000 cells. 18 hours later, cells were incubated with a fresh medium containing 10% CCK8 for 2 hours. Then, OD450 nm was evaluated. The measurement was performed on day 1, 3, 5 and 7, respectively.

3.7. Edu assay

Cells were seeded into a 96-well plate, and each well contained 1000 cells. 18 hours later, the proliferation was evaluated with the Cell-Light EdU Apollo567 In Vitro Kit (RiboBio, C10310-1). Photos were taken by the fluorescent microscope for analysis.

3.8. Soft agar assay

When the confluence reached 60-70%, the cells were digested, and a cell suspension was prepared. Lower-layer gel (20% FBS, 40% 2×RPMI1640 (Basal Medium Eagle) 40% 1.25% Agar) was prepared. 400µL of gel was added to each well in the 24-well plate. The 24-well plate with the gel was placed in an incubator at 37°C. The gel was solidified for later use. Upper-layer gel (25% FBS, 37.5% 2×RPMI1640, 37.5% 1% Agar, 0.8% 2mM L-glutamine) was prepared and mixed evenly with the cell suspension. 400µl (containing 1000 cells) was added to each well and placed in a constant temperature incubator (37°C, 5% CO_2) for two weeks. 5 fields were selected randomly under the microscope for colonies counting.

3.9. Immunoprecipitation

In order to detect the interaction between exogenous IRF5 (Flag-IRF5) and TRIM35 (HA-TRIM35), the Flag-IRF5 and HA-TRIM35 plasmids were transferred into 293T. 48h after transfection, the cells were lysed with an IP lysis buffer (50mM Tris-HCl, pH 8.0, 150mM NaCl, 1 %NP-40, protease and phosphatase inhibitor), with the supernatant collected. The beads coupling anti-Flag antibody (Sigma, A2220) were added to the supernatant for incubation overnight at 4°C. The next day, the beads were washed 3 times in wash buffer (50mM Tris-HCl (pH 8.0), 150mM NaCl, 1%NP-40), with 1×loading buffer added, heated at 100°C for 5min, and then the supernatant was taken for western blot

analysis. To detect whether there was any interaction between the endogenously expressed IRF5 and TRIM35 in HCC cells, an IP lysis buffer containing protease and a phosphatase inhibitor was sued for lysis. An equal amount of protein was taken, and 0.25μg of IRF5 antibody was added for incubation overnight at 4°C. The next day, 40μL of Protein A/G beads (bimake, B23202) was added for another incubation overnight at 4°C. The beads were washed for 3 times with the wash buffer, and then 1×loading buffer was added for western blotting analysis.

3.10. Data statistics

Data was expressed as mean ± SD. The data were analyzed using the *t* test. A survival curve was plotted by the Kaplan-Meier method, while the log-rank test was used for analysis. GraphPad Prism 8 and SPSS 17.0 were used for statistical analysis.

3. Results

3.1. IRF5 is overexpressed in HCC

To study the abundance of IRF5, the transcripts of IRF5 in HCC tissues and matched adjacent non-cancerous tissues were measured. It was found that the levels of IRF5 transcripts in the cancer tissues were higher than that in normal tissues (Figure 1A). When the levels of IRF5 mRNA in the tumors was compared with that of IRF5 in the paired adjacent tissues, higher levels of IRF5 mRNA in the tumor tissues were observed (Figure 1B). After this, the protein levels of IRF5 were measured. In the tested HCC tissues, the level of IRF5 protein was relatively high (Figure 1C). Moreover, the results were verified using immunohistochemistry staining (Figure 1D). In the Human Protein Atlas database, IRF5 expression is inversely correlated with the outcome of HCC (Figure 1E). These observations indicate that IRF5 might be important for the HCC development.

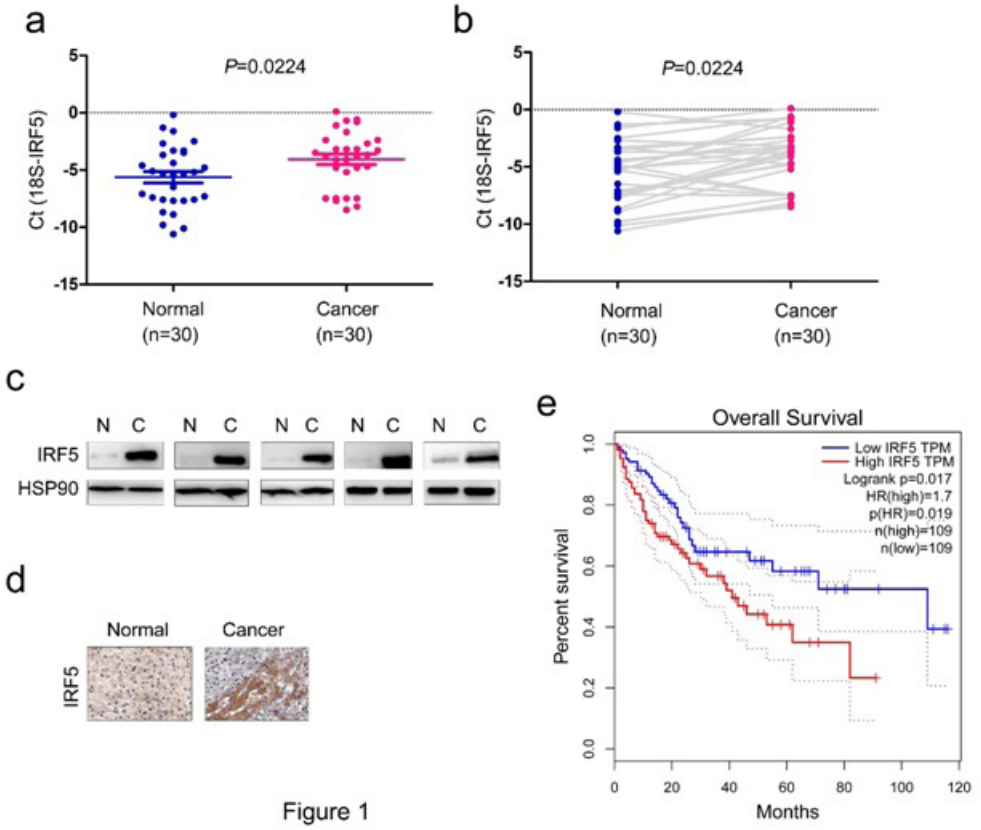

Figure 1

Figure 1: IRF5 expression is upregulated in hepatocellular carcinoma.

(A) The levels of IRF5 mRNA in 30 cancer samples (cancer) and 30 adjacent tissues (normal) were examined by qPCR. 18S, internal control. The Ct values (18S-IRF5) were plotted on the ordinate and analyzed. (B) The mRNA levels of IRF5 in paired cancer tissues (cancer) and non-cancerous tissues (normal) were analyzed. (C) The levels of IRF5 protein in 5 cancerous tissues (C) and paired non-cancerous tissues (N) were examined by Western blotting. (D) The levels of IRF5 protein in the tumor samples and adjacent non-cancerous tissue was examined by IHC. (E) Mining the HPA database and analyzing the correlation between IRF5 transcripts and the HCC outcome.

3.2. IRF5 promotes the growth and colony formation of HCC cells

To study the IRF5's functions, exogenous IRF5 was forced expressed in Huh7 and HepG2 cells, allowing the functions of IRF5 in cell growth to be detected (Figure 2A). In the CCK8 assay, the promotion of the growth was observed upon the overexpression of IRF5 in the HCC cells (Figure 2B). After this, the anchorage-independent growth of HCC cells was evaluated. It shows that the expression of IRF5 promoted the anchorage-independent growth (Figure 2C-D). IRF5 in Huh7 and QGY cells was then interfered with two shRNA sequences (Figure 3A). It was found that the growth of HCC cells in the liquid medium were inhibited after the expression of IRF5 was down-regulated (Figure 3B). Moreover, down-regulation of IRF5 impaired the growth of HCC cells on the soft agar (Figure 3C-D). Cell growth is the result of apoptosis and proliferation. The effect of the expression of IRF5 on the proliferation was detected by means of an EdU assay. The results showed that silencing IRF5 impaired the proliferation of HCC cells (Figure 3E-F).

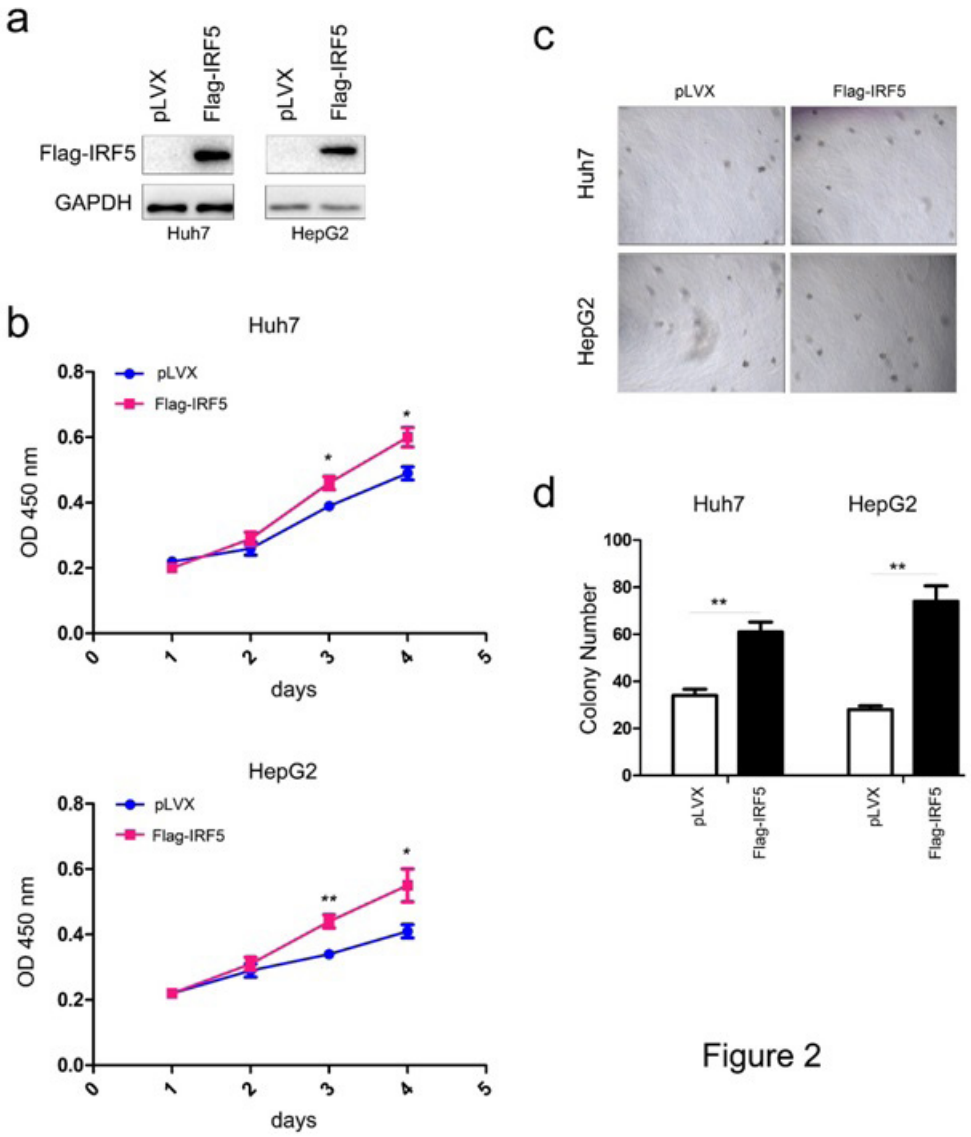

Figure 2: IRF5 promotes the growth of hepatocellular carcinoma cells.

(A) The exogenous Flag-IRF5 was forced expressed in Huh7 and HepG2 cells, and examined using western blot. (B) Cell growth was evaluated with CCK8. (C-D) The anchorage-independent growth of HCC cells was examined after the overexpression of IRF5. The colonies were counted and statistically analyzed.

*, $P<0.05$; **, $P<0.01$.

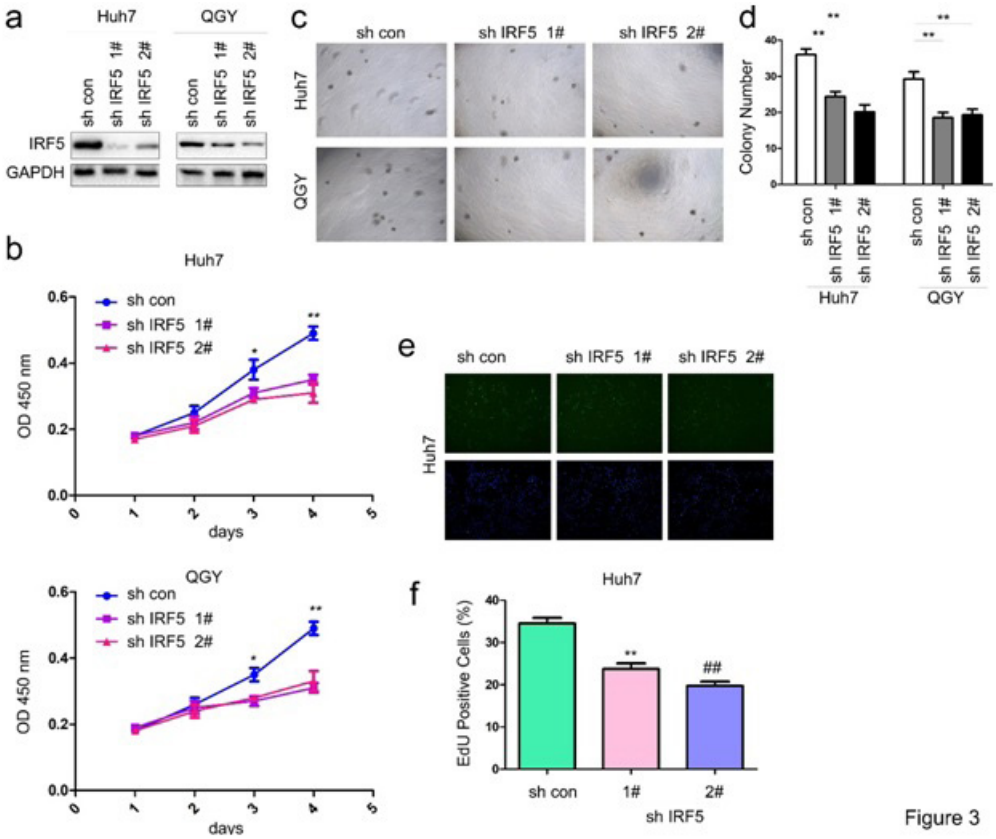

Figure 3: Silencing IRF5 inhibits the proliferation.

(A) The IRF5 knockdown efficiency was examined with Western blot. (B) Cell growth was analyzed by CCK8 assay. (C-D) The anchorage-independent growth of HCC cells was examined after the knockdown of IRF5. The colonies were counted and statistically analyzed. (E-F) Cell proliferation was analyzed with EdU assay. EdU positively stained cells were counted and statistically analyzed. *, $P<0.05$; **, $P<0.01$.

3.3. TRIM35 regulates the stability of the IRF5 protein

In order to study the regulation of IRF5 in HCC, the proteins interacting with IRF5 were searched with the help of STRING database. The results showed that IRF5 interacted with the E3 ubiquitin ligase TRIM35 (Figure 4A). The binding between IRF5 and TRIM35 was evaluated by means of a co-immunoprecipitation assay (CO-IP). In Huh7 and HepG2 cells, the exogenously expressed HA-TRIM35 interacted with the exogenously expressed Flag-IRF5 (Figure 4B). The CO-IP results also showed that the endogenously expressed IRF5 and TRIM35 in Huh7 cells had formed a complex (Figure 4C). Moreover, TRIM35 promoted the ubiquitination and degradation of IRF5 (Figure 4D).

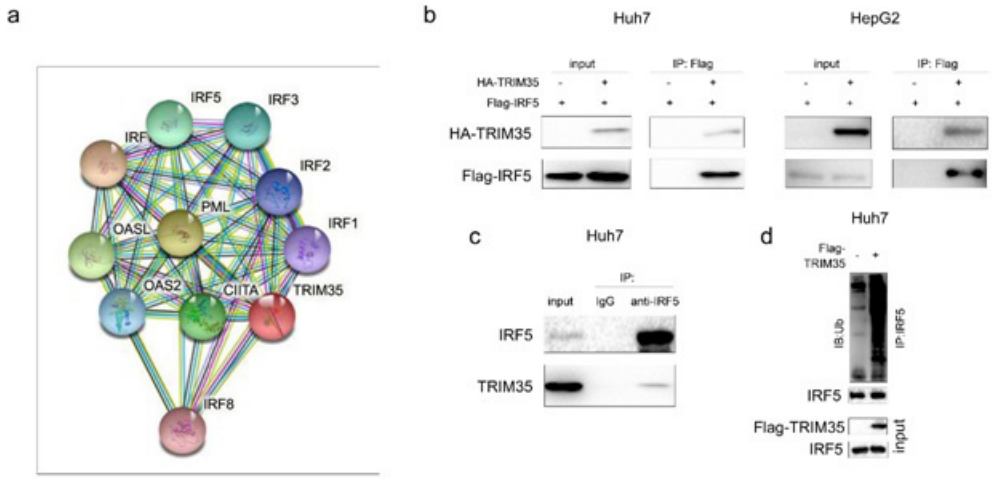

Figure 4: TRIM35 promotes the degradation of IRF5 in HCC cells.

(A) Mining the STRING database for the binding proteins of TRIM35. (B) The binding of ectopically expressed HA-TRIM35 and Flag-IRF5 was examined using immunoprecipitation assay in Huh7 cells. (C) The binding of endogenously expressed TRIM35 and IRF5 was examined using immunoprecipitation assay in Huh7 cells. (D) The levels of the ubiquitinated IRF5 by TRIM35 were elevated.

3.4. TRIM35 is down-regulated in HCC and negatively correlated with the expression of IRF5

The expression of TRIM35 in HCC was analyzed using a publicly available database. The cBioportal database shows that TRIM35 is deleted in HCC (Figure 5A). The HPA database shows that the expression of TRIM35 is positively correlated with the HCC outcome (Figure 5B). The levels of TRIM35 mRNA remained lower in cancerous tissues (Figure 5C). The level of IRF5 protein and TRIM35 protein in 20 cases of HCC was then measured by means of immunocytochemistry (IHC). The negative correlation between their expression was observed (Figure 5D). The level of IRF5 protein was high in the HCC tissue with low-expressed TRIM35 (Figure 5D).

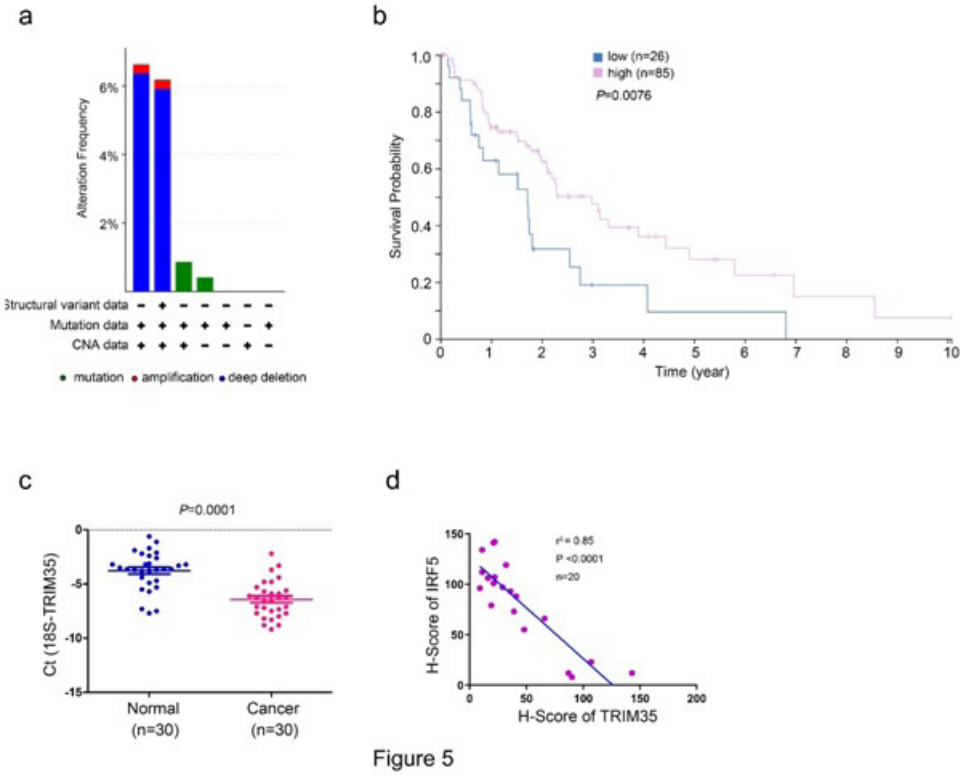

Figure 5: The expression of TRIM35 and IRF5 was negatively correlated in HCC.

(A) Mining the cBioportal database for the mutation, amplification and deletion of TRIM35 in HCC. (B) Mining the HPA database to analyze the correlation between TRIM35 mRNA level and survival. (C) The levels of TRIM35 mRNA were evaluated by qPCR. 18S, internal control. The Ct values (18S-IRF5) were plotted on the ordinate and analyzed. (D) The correlation between the expression of IRF5 and TRIM35 in the clinical HCC tissues was analyzed. The protein levels of IRF5 and TRIM35 in the 20 HCC tissues were examined using IHC and scored.

4. Discussion

It has been reported that IRF5 inhibits the replication of HCV, suggesting that it may also inhibit HCV-related HCC progression [19]. But in the HPA database, IRF5 is inversely correlated with the outcome of HCC patients. Therefore, further evidence is needed to clearly support the oncogenic roles of IRF5 in HCC. It was seen that the levels of IRF5 mRNA and protein were up-regulated in HCC, and that IRF5 expression accelerated the growth and colony formation of HCC cells. TRIM35 was low-expressed in the HCC specimen, and it promoted the degradation of IRF5 by binding to IRF5; in the clinical tissues, IRF5 expression was reversely correlated with TRIM35 expression. These data indicate that IRF5 is essential for the HCC progression. The most important finding from this study is that IRF5 promotes cell growth. The IRF family of proteins, such as IRF1 and IRF2, have been proven to have completely opposite effects on the growth of ESCC [16]. IRF8 promotes the proliferation of acute myelocytic leukemia (AML) cells [25]. This shows that the IRF protein family have a regulating effect on the growth of tumor cells. In addition, the IRF family interferes with tumor progression by reshaping the tumor immunity microenvironment. For example, the GM/CSF/IRF5 pathway enhances antitumor immunity by activating the Type 1 T-cell response [26]. This indicates that IRF proteins can regulate tumorigenesis via different mechanisms. This study reveals the ubiquitination of IRF5 by TRIM35. Up to date, IRF5 is rarely to be post-transcriptionally modified. IRF5 can be acetylated by histone acetyltransferase (HAT)[27]. The ubiquitination of IRF5 can be catalyzed by pellino-1, which helps M1 macrophages regulate obesity [28]. The results of this study show that TRIM35 mediates the degradation of IRF5. Coincidentally, many of the existing studies show that TRIM35 can also regulate the interferon signaling pathways. The findings of this study further support the previous

conclusions. In conclusion, this work reveals the promoting function of IRF5 in hepatocarcinogenesis, and the regulation of IRF5 by TRIM35, suggesting IRF5 as a promising targe.

5. Acknowledgement

Not applicable.

6. Disclosure of Conflict of Interests

No potential conflict of interest was reported by the author(s).

7. Funding

This work was supported by foundation of Shanghai Institute of Rehabilitation with Integrated Western and Chinese Traditional Medicine (082)

8. Data Availability

The data supporting the results reported in the manuscript can be acquired by the corresponding author.

9. Ethics Statement

All patients participated in this study have signed informed consent, and this research was approved by the Ethics Committee of Shanghai Eastern Hepatobiliary Surgery Hospital.

10. Author Contribution

Fang Ying and Lu Zhihui collected the clinical tissues, performed the IHC staining, and studied the molecular mechanisms; Liu Bangzhong performed the CCK8 assay; Yang Mingzhen performed the soft agar assay; Li Nan provided the clinical tissues; Wang Ping conceived and designed the experiments, analyzed the data, reviewed drafts of the paper, and approved the final draft.

11. Consent for Publication

All authors agreed to the publication of the manuscript.

References

1. Llovet JM, Kelley RK, Villanueva A, Singal AG, Pikarsky E, Roayaie S, et al. Hepatocellular carcinoma. Nature reviews Disease primers. 2021; 7(1): 6.
2. Craig AJ, Von Felden J, Garcia-Lezana T, Sarcognato S, Villanueva A. Tumour evolution in hepatocellular carcinoma. Nature reviews Gastroenterology & hepatology. 2020; 17(3): 139-152.
3. Dickson I. Liver cancer: A new drug target for hepatocellular carcinoma. Nature reviews Gastroenterology & hepatology. 2016; 13: 375.
4. Duffy AG, Greten TF. Liver cancer: Regorafenib as second-line therapy in hepatocellular carcinoma. Nature reviews Gastroenterology & hepatology 2017; 14(3): 141-142.
5. Fan ST. Hepatocellular carcinoma--resection or transplant? Nature reviews Gastroenterology & hepatology. 2012; 9: 732-737.
6. Hacker H, Tseng PH, Karin M. Expanding TRAF function: TRAF3 as a tri-faced immune regulator. Nature reviews Immunology 2011; 11(7): 457-468.
7. Morales DJ, Lenschow DJ. The antiviral activities of ISG15. Journal of molecular biology. 2013; 425: 4995-5008.
8. Potu H, Sgorbissa A, Brancolini C. Identification of USP18 as an important regulator of the susceptibility to IFN-alpha and drug-induced apoptosis. Cancer research. 2010; 70(2): 655-665.
9. Dhawan T, Zahoor MA, Heryani N, Workenhe ST, Nazli A, Kaushic C. TRIM26 Facilitates HSV-2 Infection by Downregulating Antiviral Responses through the IRF3 Pathway. Viruses. 2021; 13(1): 70.
10. Du Z, Whitt MA, Baumann J, Garner JM, Morton CL, Davidoff AM, et al. Inhibition of type I interferon-mediated antiviral action in human glioma cells by the IKK inhibitors BMS-345541 and TPCA-1. Journal of interferon & cytokine research : the official journal of the International Society for Interferon and Cytokine Research 2012; 32(8): 368-377.
11. Kroetz DN, Allen RM, Schaller MA, Cavallaro C, Ito T, Kunkel SL, et al. Type I Interferon Induced Epigenetic Regulation of Macrophages Suppresses Innate and Adaptive Immunity in Acute Respiratory Viral Infection. PLoS pathogens. 2015; 11: e1005338.
12. Xu X, Wu Y, Yi K, Hu Y, Ding W, Xing C, et al. IRF1 regulates the progression of colorectal cancer via interferoninduced proteins. International journal of molecular medicine. 2021; 47(6): 104.
13. Hong M, Zhang Z, Chen Q, Lu Y, Zhang J, Lin C, et al. IRF1 inhibits the proliferation and metastasis of colorectal cancer by suppressing the RAS-RAC1 pathway. Cancer management and research. 2019; 11: 369-378.
14. Karki R, Sharma BR, Lee E, Banoth B, Malireddi RKS, Samir P, et al. Interferon regulatory factor 1 regulates PANoptosis to prevent colorectal cancer. JCI insight. 2020; 5(12): e136720.
15. Ohsugi T, Yamaguchi K, Zhu C, Ikenoue T, Takane K, Shinozaki M, et al. Anti-apoptotic effect by the suppression of IRF1 as a downstream of Wnt/beta-catenin signaling in colorectal cancer cells. Oncogene. 2019; 38: 6051-6064.
16. Wang Y, Liu DP, Chen PP, Koeffler HP, Tong XJ, Xie D, et al. Involvement of IFN regulatory factor (IRF)-1 and IRF-2 in the formation and progression of human esophageal cancers. Cancer research. 2007; 67(6): 2535-2543.
17. Wang J, Li H, Xue B, Deng R, Huang X, Xu Y, et al. IRF1 Promotes the Innate Immune Response to Viral Infection by Enhancing the Activation of IRF3. Journal of virology. 2020; 94(22): e01231-20.
18. Massimino M, Vigneri P, Fallica M, Fidilio A, Aloisi A, Frasca F, et al. IRF5 promotes the proliferation of human thyroid cancer cells. Molecular cancer. 2012; 11: 21.
19. Cevik O, Li D, Baljinnyam E, Manvar D, Pimenta EM, Waris G, et al. Interferon regulatory factor 5 (IRF5) suppresses hepatitis C virus (HCV) replication and HCV-associated hepatocellular carcinoma. The Journal of biological chemistry. 2017; 292: 21676-21689.
20. Zhang J, Xu Z, Yu B, Xu J, Yu B. Tripartite motif containing 35 contributes to the proliferation, migration, and invasion of lung cancer cells in vitro and in vivo. Bioscience reports. 2020; 40(4): BSR20200065.
21. Yao L, Zhou Y, Sui Z, Zhang Y, Liu Y, Xie H, et al. HBV-encoded miR-2 functions as an oncogene by downregulating TRIM35 but upregulating RAN in liver cancer cells. EBioMedicine. 2019; 48: 117-129.
22. Chen Z, Lu X, Wang Z, Jin G, Wang Q, Chen D, et al. Co-expression of PKM2 and TRIM35 predicts survival and recurrence in hepatocellular carcinoma. Oncotarget. 2015; 6: 2538-2548.

23. Chen Z, Wang Z, Guo W, Zhang Z, Zhao F, Zhao Y, et al. TRIM35 Interacts with pyruvate kinase isoform M2 to suppress the Warburg effect and tumorigenicity in hepatocellular carcinoma. Oncogene. 2015; 34(30): 3946-3956.
24. Song L, Zhang W, Chang Z, Pan Y, Zong H, Fan Q, et al. miR-4417 Targets Tripartite Motif-Containing 35 (TRIM35) and Regulates Pyruvate Kinase Muscle 2 (PKM2) Phosphorylation to Promote Proliferation and Suppress Apoptosis in Hepatocellular Carcinoma Cells. Medical science monitor: international medical journal of experimental and clinical research. 2017; 23: 1741-1750.
25. Liss F, Frech M, Wang Y, Giel G, Fischer S, Simon C, et al. IRF8 Is an AML-Specific Susceptibility Factor That Regulates Signaling Pathways and Proliferation of AML Cells. Cancers. 2021; 13(4): 764.
26. Arnold IC, Artola-Boran M, Gurtner A, Bertram K, Bauer M, Frangez Z, et al. The GM-CSF-IRF5 signaling axis in eosinophils promotes antitumor immunity through activation of type 1 T cell responses. The Journal of experimental medicine. 2020; 217(12): e20190706.
27. Feng D, Sangster-Guity N, Stone R, Korczeniewska J, Mancl ME, Fitzgerald-Bocarsly P, et al. Differential requirement of histone acetylase and deacetylase activities for IRF5-mediated proinflammatory cytokine expression. Journal of immunology. 2010; 185: 6003-6012.
28. Kim D, Lee H, Koh J, Ko JS, Yoon BR, Jeon YK, et al. Cytosolic Pellino-1-Mediated K63-Linked Ubiquitination of IRF5 in M1 Macrophages Regulates Glucose Intolerance in Obesity. Cell reports. 2017; 20: 832-845.