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*Original Article*

# PRDM5 expression correlates with malignant behaviors and indicates favorable prognosis in HCC

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**Abstract:** PR (PRDI-BF1 and RIZ) domain protein 5 (PRDM5) is a newly identified member of the PRDM family and has been suggested to play important roles in tumor development. However, the association between PRDM5 expression and the clinical characteristic of hepatocellular carcinoma (HCC) is not well investigated. In this present study, one-step quantitative reverse transcription-polymerase chain reaction (qPCR) with 10 fresh-frozen HCC and immunohistochemistry (IHC) analyses in 90 HCC cases were performed respectively to explore the relationship between PRDM5 expression and the clinicopathological items of HCC. The results showed that the expressions of PRDM5 were significantly decreased in HCC tissues than in corresponding non-cancerous tissues ($P < 0.05$). The IHC information implied that the PRDM5 protein expression in HCC was statistically correlated with pathological grade ($P = 0.014$), T ($P = 0.001$) and TNM stage ($P = 0.001$). Both survival analysis and Kaplan-Meier curve demonstrated that higher PRDM5 protein level ($P = 0.048$) was substantially associated with the favorable prognosis of patients with HCC. The data suggested that PRDM5 may be identified as a novel prognostic biomarker for HCC.

**Keywords:** PRDM5, HCC, qPCR, IHC, prognosis

## Introduction

Hepatocellular carcinoma (HCC) accounts for 90% of cases of primary liver cancer and represents one of the leading causes of cancer mortality worldwide [1]. In China, HCC ranks third in mortality after gastric and esophageal cancer, and the number of HCC patients accounts for more than half of HCC cases globally [2, 3]. The development of HCC is a complicated and multistep process, which critically associates with a number of factors including chronic alcohol consumption, HBV/HCV infections and cirrhosis [4, 5]. Due to the high rate of recurrence or metastasis after curative resection, the overall survival of HCC patients remains poor despite various progresses in surgical techniques and other therapy strategies [6, 7]. The most common causes of death among HCC patients are tumor progression, metastasis, and recurrence [8]. Therefore, the identification of novel biomarkers which can predict the poor prognosis or the high risk of metastasis is substantially necessary.

PR (PRDI-BF1 and RIZ) domain proteins (PRDM) are a subfamily of the kruppel-like zinc finger transcription factors [9]. PRDM have been reported to play significant roles during cell differentiation and malignant transformation, and several studies indicate that PRDM family members are negative factors of tumorigenesis [10-12]. PRDM5 (or PFM2) is a recently identified member of the PRDM family, which gene locates on human chromosome 4 [13]. The function of PRDM5 derives from the association with chromatin modifying enzymes, specifically G9A and HDAC1, which are recruited to the promoters of its target genes to determine chromatin modification [14]. Two studies showed the decreased expression levels of PRDM5 mRNA and protein in breast and cervical cancers [15, 16]. PRDM5 silencing by promoter methylation was witnessed in various types of human cancer inc-

**Table 1.** Correlation of PRDM5 expression with the clinicopathological items in HCC

| Groups | No. | PRDM5 | | $\chi^2$ | *p* value |
|---|---|---|---|---|---|
| | | + | % | | |
| Gender | | | | | |
| Male | 77 | 33 | 42.9 | 3.108 | 0.078 |
| Female | 13 | 9 | 69.2 | | |
| Age (years) | | | | | |
| ≥ 60 | 22 | 10 | 45.5 | 0.035 | 0.851 |
| < 60 | 67 | 32 | 47.8 | | |
| Insufficient Data | 1 | | | | |
| Tumor size (cm) | | | | | |
| ≥ 5 | 53 | 25 | 47.2 | 0.001 | 0.996 |
| ≥ 5 | 36 | 17 | 47.2 | | |
| Insufficient Data | 1 | | | | |
| Pathological grade | | | | | |
| Grade 1 | 10 | 9 | 90.0 | 8.505 | 8.505 |
| Grade 2 | 66 | 27 | 40.9 | | |
| Grade 3 | 14 | 6 | 42.9 | | |
| Portal vein invasion | | | | | |
| Positive | 4 | 0 | 0.0 | 3.111 | 0.078 |
| Negative | 10 | 5 | 50.0 | | |
| Insufficient Data | 76 | | | | |
| Liver cirrhosis | | | | | |
| Positive | 36 | 17 | 47.2 | 0.001 | 0.996 |
| Negative | 53 | 25 | 47.2 | | |
| Insufficient Data | 1 | | | | |
| T | | | | | |
| T1 | 42 | 12 | 28.6 | 17.397 | 0.001* |
| T2 | 33 | 25 | 75.8 | | |
| T3 | 11 | 4 | 36.4 | | |
| T4 | 3 | 1 | 33.3 | | |
| Insufficient Data | 1 | | | | |
| N | | | | | |
| Positive | 1 | 0 | 0.0 | 0.858 | 0.354 |
| Negative | 84 | 39 | 46.4 | | |
| Insufficient Data | 5 | | | | |
| M | | | | | |
| Positive | 1 | 1 | 100.0 | 1.160 | 0.282 |
| Negative | 87 | 40 | 46.0 | | |
| Insufficient Data | 2 | | | | |
| TNM stage | | | | | |
| Stage I-II | 43 | 28 | 65.1 | 11.965 | 0.001* |
| Stage III-IV | 43 | 12 | 27.9 | | |
| Insufficient Data | 4 | | | | |

*P < 0.05.

luding ovarian, colorectal, gastric, and hepatocellular tumors [15, 17]. As epigenetic silencing by promoter methylation is highly associated with tumor development, PRDM5 is believed to have the potential to be identified as a useful molecular target for cancer diagnosis and therapy [18, 19]. Although the above data proposed certain tumor-suppressive characteristics for PRDM5, the functional relationship between PRDM5 expression and the clinical pathological attributes of HCC, especially the prognostic significance, remains to be delineated.

In this study, the expression of PRDM5 in HCC tissues compared with non-cancerous tissues was detected. Moreover, the correlation of PRDM5 expression with the clinicopathological items was further analyzed in HCC patients.

## Materials and methods

### *Patient samples*

Ten fresh-frozen HCC samples and corresponding non-cancerous tissue samples were collected from the Department of Hepatobiliary Surgery, Affiliated Hospital of Heibei University, to perform one-step quantitative reverse transcription-PCR (qPCR) test. Ninety cases of HCC were enrolled to construct tissue microarrays (TMA) and the TMA were purchased from Outdo Biotech Co., Ltd (Shanghai, China) to execute immunohistochemistry (IHC) analysis. The original clinical data were also provided by Outdo Biotech Co., Ltd, including gender, age, tumor diameter, cirrhosis status, pathological grade, tumor status (T), lymph node metastasis (N), distant metastasis (M) and TNM stage. None of the patients received radiotherapy and chemotherapy before surgery. Written informed consent was acquired from each patient for publication of this study and any related pictures, and the study protocol was approved by the Human Research Ethics Committee of each hospital.

### *One-step qPCR analysis in fresh HCC tissues*

Ten fresh-frozen HCC samples as well as the corresponding non-cancerous tissue samples were subjected to qPCR test. Total RNA extraction was extracted from the tissues using Trizol (Invertrogen, USA) following the manufacturer's manual. The protocol of qPCR analysis was described previously [20]. The primers for PRDM5 were as follows: forward primer 5'-CAG GTT CTC CCT GAA GTC CT-3' and reverse primer 5'-TGA GAT GGT GCC TCA TGA AC-3'. The glycer-

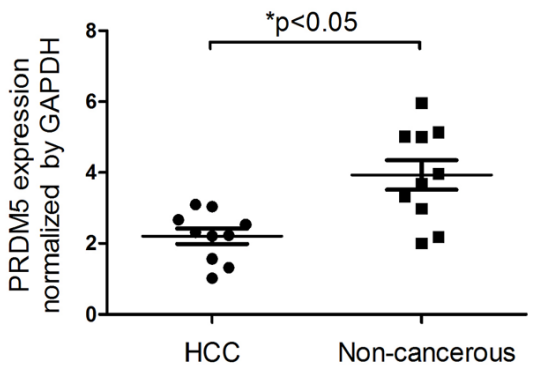


**Figure 1.** One-step quantitative real-time polymerase chain reaction (qPCR) was performed to detect PRDM5 mRNA expression levels in hepatocellular carcinoma (HCC) tissues compared with the corresponding non-cancerous tissues. When normalized to glyceraldehyde 3-phosphate dehydrogenase (GAPDH) mRNA levels, the PRDM5 mRNA level in HCC tissues (2.20 ± 0.222) is significantly decreased than that in corresponding non-cancerous tissues (3.93 ± 0.422).

aldehyde 3-phosphate dehydrogenase (GAPDH) was used as internal control, and the primers for GAPDH were as follows: forward primer 5'-TGC ACC ACC AAC TGC TTA GC-3' and reverse primer 5'-GGC ATG GAC TGT GGT CAT GA-3'. Amplification conditions consisted of 30 min at 42°C for reverse transcription and 2 min at 94°C for Taq activation, followed by 35 cycles at 95°C for 20 s, 56°C for 20 s, and elongation at 72°C for 30 s.

*IHC analysis in HCC TMA*

IHC analysis was performed as previously described [21, 22]. Deparaffinized sections from array blocks were stained using mouse anti-PRDM5 antibody (Abcam). The secondary antibody used was horseradish peroxidase-conjugated anti-rabbit antibody (Dako). Phosphate-buffered saline (PBS) was used as negative control. PRDM5 immunostaining was scored according to intensity and percentage of PRDM5-positive cells simultaneously.

Staining intensity of PRDM5 was scored as follows: 0 (negative), 1 (weakly positive), 2 (moderately positive), and 3 (strongly positive). Staining percentage of PRDM5 was categorized as follows: 1 (0-10%), 2 (11-50%), 3 (51-80%), and 4 (81-100%). The product of the staining intensity and percentage scores was marked as the IHC score. The final result of IHC analysis of PRDM5 was quantified using a two level grading system based on the IHC score: < 4 suggests low expression, while 4-9 suggests high expression.

*Statistical analysis*

The PRDM5 expression in HCC tissues and corresponding non-cancerous tissues was analyzed with the Wilcoxon nonparametric signed-rank test. The associations between PRDM5 expression and clinicopathologic items were evaluated by chi-square tests. Univariate and multivariate Cox regression models were employed to identify prognostic factors that influenced the overall survival. Survival curves were produced using the Kaplan-Meier method. For all tests, the significance level for statistical analysis was set at $P < 0.05$. All data were analyzed by performing SPSS 18.0 (SPSS Inc, Chicago, IL).

## Results

*Summary of clinical information of 90 HCC patients*

The substantial clinical information is summarized in **Table 1**. A total of 90 HCC patients were enrolled from 77 men and 13 women, and the mean age of the patients at the time of surgery was 54.9 years. There were 53 cases with tumor diameter ≥ 5 cm, 36 with tumor diameter < 5 cm. Based on the pathological grade classification, 10 patients were in grade 1, 66 patients were in grade 2 and 14 patients were in grade 3. 4 patients suffered portal vein invasion while 36 patients encountered liver cirrhosis. The number of patients with T1, T2, T3, T4 was 42, 33, 11 and 3 respectively. Positive N was observed in 1 patient, while positive M was also witnessed in 1 patient. According to TNM staging system, 43 patients were in stages I-II, whereas 43 patients were in stages III-IV. Among all the 90 cases, 39 patients survived while 51 patients died.

*Detection of PRDM5 mRNA expression by qPCR test*

Total RNA was extracted from 10 HCC tissues and the corresponding non-cancerous tissues, and then the one-step qPCR was performed to detect PRDM5 mRNA expression. When normalizing to GAPDH, the means of PRDM5 mRNA in HCC tissues and that of the corresponding non-cancerous tissues were 2.20 ± 0.222 and 3.93 ± 0.422, respectively ($t = 3.626$, $df = 18$,

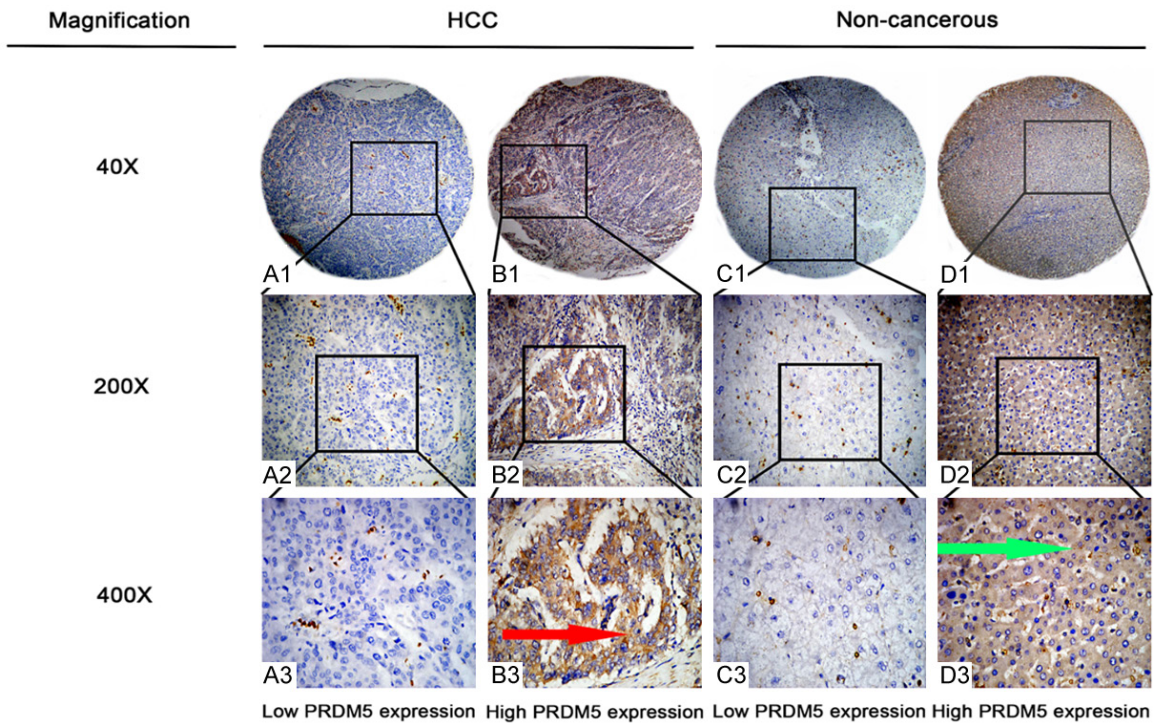


**Figure 2.** Representative images of PRDM5 protein expression in hepatocellular carcinoma (HCC) tissues and corresponding non-cancerous tissues. (A1-3) Low expression of PRDM5 protein in HCC tissue. (B1-3) High expression of PRDM5 protein in HCC tissue. Red arrow shows positive staining in the cytoplasm of cancer cells. (C1-3) Low expression of PRDM5 protein in corresponding non-cancerous tissue. (D1-3) High expression of PRDM5 protein in corresponding non-cancerous tissue. Green arrow shows positive staining of non-cancerous cells. Original magnification × 40 in (A1-D1); × 200 in (A2-D2); × 400 in (A3-D3).

P = 0.002). PRDM5 expression in the HCC tissues was nearly 1.79-fold lower than that in corresponding non-cancerous tissues (**Figure 1**).

*Detection of PRDM5 protein expression by IHC analysis*

High PRDM5 expression was observed in 42 (46.7%) of the 90 HCC tissue samples compared with 57 (63.3%) of 90 corresponding non-cancerous tissue samples, and the difference showed statistically significance ($\chi^2$ = 5.05, P = 0.03). The result of IHC analysis was in accordance with the previous qPCR test in which lower level of PRDM5 mRNA expression was exhibited. High expression of PRDM5 protein was mainly localized in the cytoplasm of HCC cells (**Figure 2**).

*Correlation of PRDM5 expression with the clinicopathological items*

The association between PRDM5 protein expression and representative clinicopathological items was illustrated in **Table 1**. Positive PRDM5 expression was significantly related to pathological grade (P = 0.014), T (P = 0.001) and TNM stage (P = 0.001). In comparison, other clinical items, including gender, age, tumor size, portal vein invasion, liver cirrhosis, N and M barely showed correlation with PRDM5 protein expression (**Table 1**).

*Survival analysis*

The univariate analysis revealed several factors that correlated with overall survival of 90 HCC patients, including PRDM5 expression (P = 0.001), tumor size (P = 0.020), pathological grade (P = 0.038), T (P = 0.017) and TNM stage (P = 0.001). Multivariate analysis further screened that PRDM5 expression (P = 0.048) was the independent predictors for 90 HCC patients (**Table 2**). Kaplan-Meier survival curves also demonstrated that HCC patients with high PRDM5 expression encountered significantly favorable overall survival (**Figure 3**).

## Discussion

Thus far, seventeen human PRDM members have been identified and reported to act tumor

**Table 2.** Identification of prognostic factors by univariate and multivariate analysis in HCC

| | Univariate analysis | | | Multivariate analysis | | |
|---|---|---|---|---|---|---|
| | HR | *p* value | 95% CI | HR | *p* value | 95% CI |
| PRDM5 expression | | | | | | |
| High versus Low | 0.38 | 0.001* | 0.213-0.677 | 0.51 | 0.048* | 0.265-0.998 |
| Gender | | | | | | |
| Male versus Female | 1.51 | 0.950 | 0.643-3.537 | | | |
| Age (years) | | | | | | |
| ≥ 60 versus < 60 | 0.60 | 0.161 | 0.290-1.228 | | | |
| Tumour size (cm) | | | | | | |
| ≥ 5 versus < 5 | 2.06 | 0.020* | 1.119-3.804 | 1.43 | 0.337 | 0.690-2.959 |
| Pathological grade | | | | | | |
| Grade 1-2 versus Grade 3 | 0.50 | 0.038* | 0.038* | 0.67 | 0.246 | 0.340-1.318 |
| Portal vein invasion | | | | | | |
| Positive versus Negative | 1.06 | 0.947 | 0.211-5.278 | | | |
| Liver cirrhosis | | | | | | |
| Positive versus Negative | 0.71 | 0.262 | 0.390-1.292 | | | |
| T | | | | | | |
| T1 versus T2 versus T3 versus T4 | 0.60 | 0.017* | 0.392-0.914 | 1.14 | 0.633 | 0.633 |
| N | | | | | | |
| Positive versus Negative | 2.92 | 0.294 | 0.395-21.659 | | | |
| M | | | | | | |
| Positive versus Negative | 1.22 | 0.842 | 0.168-8.890 | | | |
| TNM stage | | | | | | |
| Stage I-II versus Stage III-IV | 0.31 | 0.001* | 0.001* | 0.42 | 0.085 | 0.156-1.127 |

*P < 0.05.

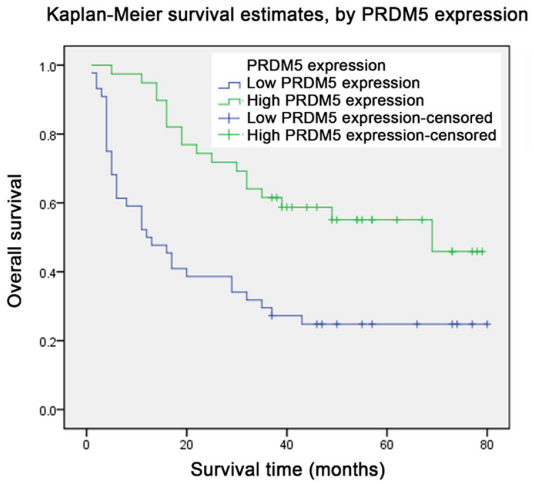


**Figure 3.** Survival analysis of hepatocellular carcinoma (HCC) patients by Kaplan-Meier curve. Overall survival rate in patients with high PRDM5 protein expression (green line) was significantly higher than that in patients with low PRDM5 expression (blue line).

inhibitory effectiveness. For instance, PRDM1 plays as a transcriptional repressor of c-MYC and induces cell differentiation [23, 24]. PRDM2 is believed to possess tumor suppressive activity mediated by PR domain [25, 26]. PRDM3 is associated with translocation in hematopoietic malignancies and PRDM4 deletion is often witnessed in ovarian, gastric and pancreatic cancers [27, 28]. As for PRDM5, large number of studies have shown that PRDM5 absence is a common molecular event during tumorigenesis and the epigenetic inactivation of PRDM5 is witnessed in a broad spectrum of solid tumor types including digestive, respiratory and gynecologic tumors [29]. A latest study illustrated the decreased expression of PRDM5 as well as revealed the potential diagnostic and prognostic value of PRDM5 methylation for lung cancer patients [30]. Moreover, Galli et al. constructed a remarkable mouse model and proved that knockout of PRDM5 critically contributed intestinal carcinogenesis [31]. To better understand how PRDM5 exerts its function, several mechanical researches have been ongoing. Deng et al. demonstrated that PRDM5 overexpression could in-

duce cell cycle arrest and apoptosis of cancer cells [15]. Shu et al. and Meani et al. explored that Wnt/β-catenin pathway might be a substantial part of PRDM5 tumor-suppressive activity [32, 33]. A recent genomic study also elucidated that PRDM5 could interact with insulator factors to modulate chromatin organization of target genome sites [34]. Based on the above characteristics of PRDM5, we attempted to verify the relationship between PRDM5 expression and various clinicopathological attributes of HCC, especially the prognosis significance of PRDM5.

The qPCR test with small HCC samples illustrated a substantially lower level of PRDM5 mRNA expression in HCC tissues than that in corresponding non-cancerous tissues. Subsequently, the results IHC analysis further demonstrated a statistically decreasing PRDM5 protein expression in HCC tissue comparing with that in non-cancerous tissues. In addition, the expression of PRDM5 was significantly correlated with several clinical items of HCC, including pathological grade, T and TNM stage. The above data, in agreement with results of the previous study which verified that PRDM5 expression at both mRNA and protein levels was reduced in lung cancer tissues and PRDM5 promoter methylation was significantly associated with certain tumor phenotypes [30].

With regard to the prognostic characteristic of PRDM5, both univariate and multivariate analysis exhibited that HCC patients with reduced PRDM5 expression suffered worse survival outcomes than those expressing elevated PRDM5, indicating the potential value of PRDM5 in predicting the prognosis of HCC patients. Kaplan-Meier curve also displayed that the life span of HCC patients with high PRDM5 expression was crucially favorable.

There were some limitations to the present study. For one thing, we failed to collect some other important clinical information of HBV/HCV infection status and the value of α-Fetoprotein (AFP), factors that are considered to be significant components of HCC etiology. For another, the number of HCC patients with positive N and M in this present study was too small hence some negative statistical results might need to be further confirmed. Future researches that enroll larger samples and exhaustive clinical data are necessary.

In summary, this study was the first to detect on the differential expression of PRDM5 in HCC, in both mRNA and protein level. The results conclude that reduced expression of PRDM5 may play important roles in the tumorigenesis of HCC and PRDM5 may be identified as a novel prognostic biomarker in HCC patients.

## Disclosure of conflict of interest

None.

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## References

[1] Siegel R, Naishadham D, Jemal A. Cancer statistics, 2013. CA Cancer J Clin 2013; 63: 11-30.

[2] Fu M, Gu X, Ni H, Zhang W, Chang F, Gong L, Chen X, Li J, Qiu L, Shi C, Bao J. High expression of inositol polyphosphate phosphatase-like 1 associates with unfavorable survival in hepatocellular carcinoma. Int J Clin Exp Pathol 2013; 6: 2515-22.

[3] Nordenstedt H, White DL, El-Serag HB. The changing pattern of epidemiology in hepatocellular carcinoma. Dig Liver Dis 2010; 42 Suppl 3: S206-14.

[4] Ding J, Wang H. Multiple interactive factors in hepatocarcinogenesis. Cancer Lett 2014; 346: 17-23.

[5] Wang W, Li GY, Zhu JY, Huang DB, Zhou HC, Zhong W, Ji CS. Overexpression of AGGF1 is correlated with angiogenesis and poor prognosis of hepatocellular carcinoma. Med Oncol 2015; 32: 131.

[6] Bruix J, Gores GJ, Mazzaferro V. Hepatocellular carcinoma: clinical frontiers and perspectives. Gut 2014; 63: 844-55.

[7] Meng FD, Wei JC, Qu K, Wang ZX, Wu QF, Tai MH, Liu HC, Zhang RY, Liu C. FoxM1 overexpression promotes epithelial-mesenchymal transition and metastasis of hepatocellular carcinoma. World J Gastroenterol 2015; 21: 196-213.

[8] Lee SC, Tan HT, Chung MC. Prognostic biomarkers for prediction of recurrence of hepatocellular carcinoma: current status and future prospects. World J Gastroenterology 2014; 20: 3112-24.

[9] Wu H, Min J, Lunin VV, Antoshenko T, Dombrovski L, Zeng H, Allali-Hassani A, Campagna-Slater V, Vedadi M, Arrowsmith CH, Plotnikov

AN, Schapira M. Structural biology of human H3K9 methyltransferases. PLoS One 2010; 5: e8570.
[10] Jiang G, Liu L, Buyse IM, Simon D, Huang S. Decreased RIZ1 expression but not RIZ2 in hepatoma and suppression of hepatoma tumorigenicity by RIZ1. Int J Cancer 1999; 83: 541-6.
[11] Huang S. Histone methyltransferases, diet nutrients and tumour suppressors. Nat Rev Cancer 2002; 2: 469-76.
[12] Wu Y, Ferguson JE 3rd, Wang H, Kelley R, Ren R, McDonough H, Meeker J, Charles PC, Wang H, Patterson C. PRDM6 is enriched in vascular precursors during development and inhibits endothelial cell proliferation, survival, and differentiation. J Mol Cell Cardiol 2008; 44: 47-58.
[13] Chou YH, Chung KC, Jeng LB, Chen TC, Liaw YF. Frequent allelic loss on chromosomes 4q and 16q associated with human hepatocellular carcinoma in Taiwan. Cancer Lett 1998; 123: 1-6.
[14] Duan Z, Person RE, Lee HH, Huang S, Donadieu J, Badolato R, Grimes HL, Papayannopoulou T, Horwitz MS. Epigenetic regulation of protein-coding and microRNA genes by the Gfi1-interacting tumor suppressor PRDM5. Mol Cell Biol 2007; 27: 6889-902.
[15] Deng Q, Huang S. PRDM5 is silenced in human cancers and has growth suppressive activities. Oncogene 2004; 23: 4903-10.
[16] Cheng HY, Chen XW, Cheng L, Liu YD, Lou G. DNA methylation and carcinogenesis of PRDM5 in cervical cancer. J Cancer Res Clin Oncol 2010; 136: 1821-5.
[17] Watanabe Y, Toyota M, Kondo Y, Suzuki H, Imai T, Ohe-Toyota M, Maruyama R, Nojima M, Sasaki Y, Sekido Y, Hiratsuka H, Shinomura Y, Imai K, Itoh F, Tokino T. PRDM5 identified as a target of epigenetic silencing in colorectal and gastric cancer. Clin Cancer Res 2007; 13: 4786-94.
[18] Laird PW. The power and the promise of DNA methylation markers. Nat Rev Cancer 2003; 3: 253-66.
[19] Baylin SB, Ohm JE. Epigenetic gene silencing in cancer-a mechanism for early oncogenic pathway addiction? Nat Rev Cancer 2006; 6: 107-16.
[20] Gu X, Fu M, Ding Y, Ni H, Zhang W, Zhu Y, Tang X, Xiong L, Li J, Qiu L, Xu J, Zhu J. TIMP-3 expression associates with malignant behaviors and predicts favorable survival in HCC. PLoS One 2014; 9: e106161.
[21] Chen R, Lu M, Wang J, Zhang D, Lin H, Zhu H, Zhang W, Xiong L, Ma J, Mao Y, Zhu J, Xu J. Increased expression of Trop2 correlates with poor survival in extranodal NK/T cell lymphoma, nasal type. Virchows Arch 2013; 463: 713-9.
[22] Mao Y, Zhang DW, Zhu H, Lin H, Xiong L, Cao Q, Liu Y, Li QD, Xu JR, Xu LF, Chen RJ. LMP1 and LMP2A are potential prognostic markers of extranodal NK/T-cell lymphoma, nasal type (ENKTL). Diagn Pathol 2012; 7: 178.
[23] Yu J, Angelin-Duclos C, Greenwood J, Liao J, Calame K. Transcriptional repression by blimp-1 (PRDI-BF1) involves recruitment of histone deacetylase. Mol Cell Biol 2000; 20: 2592-603.
[24] Lin Y, Wong K, Calame K. Repression of c-myc transcription by Blimp-1, an inducer of terminal B cell differentiation. Science 1997; 276: 596-9.
[25] Chadwick RB, Jiang GL, Bennington GA, Yuan B, Johnson CK, Stevens MW, Niemann TH, Peltomaki P, Huang S, de la Chapelle A. Candidate tumor suppressor RIZ is frequently involved in colorectal carcinogenesis. Proc Natl Acad Sci U S A 2000; 97: 2662-7.
[26] Du Y, Carling T, Fang W, Piao Z, Sheu JC, Huang S. Hypermethylation in human cancers of the RIZ1 tumor suppressor gene, a member of a histone/protein methyltransferase superfamily. Cancer Res 2001; 61: 8094-9.
[27] Mitani K. Molecular mechanisms of leukemogenesis by AML1/EVI-1. Oncogene 2004; 23: 4263-9.
[28] Yang XH, Huang S. PFM1 (PRDM4), a new member of the PR-domain family, maps to a tumor suppressor locus on human chromosome 12q23-q24.1. Genomics 1999; 61: 319-25.
[29] Shu XS. Emerging role of PR domain containing 5 (PRDM5) as a broad tumor suppressor in human cancers. Tumour Biol 2015; 36: 1-3.
[30] Tan SX, Hu RC, Tan YL, Liu JJ, Liu WE. Promoter methylation-mediated downregulation of PRDM5 contributes to the development of lung squamous cell carcinoma. Tumour Biol 2014; 35: 4509-16.
[31] Galli GG, Multhaupt HA, Carrara M, de Lichtenberg KH, Christensen IB, Linnemann D, Santoni-Rugiu E, Calogero RA, Lund AH. Prdm5 suppresses Apc(Min)-driven intestinal adenomas and regulates monoacylglycerol lipase expression. Oncogene 2014; 33: 3342-50.
[32] Shu XS, Geng H, Li L, Ying J, Ma C, Wang Y, Poon FF, Wang X, Ying Y, Yeo W, Srivastava G, Tsao SW, Yu J, Sung JJ, Huang S, Chan AT, Tao Q. The epigenetic modifier PRDM5 functions as a tumor suppressor through modulating WNT/beta-catenin signaling and is frequently silenced in multiple tumors. PLoS One 2011; 6: e27346.
[33] Meani N, Pezzimenti F, Deflorian G, Mione M, Alcalay M. The tumor suppressor PRDM5 regu-

lates Wnt signaling at early stages of zebrafish development. PLoS One 2009; 4: e4273.
[34] Galli GG, Carrara M, Francavilla C, de Lichtenberg KH, Olsen JV, Calogero RA, Lund AH. Genomic and proteomic analyses of Prdm5 reveal interactions with insulator binding proteins in embryonic stem cells. Mol Cell Biol 2013; 33: 4504-16.