Histol Histopathol (2018) 33: 343-355

http://www.hh.um.es

Histology and Histopathology

From Cell Biology to Tissue Engineering

Review

Emerging evidence of molecular biomarkers in hepatocellular carcinoma

Shinichi Umeda, Mitsuro Kanda and Yasuhiro Kodera

Department of Gastroenterological Surgery (Surgery II), Nagoya University Graduate School of Medicine, Nagoya, Japan

Summary. Hepatocellular carcinoma (HCC) is the third most common cause of cancer-related death worldwide. Patients with HCC generally present at an advanced stage resulting in death within 6-20 months. Therefore, novel treatment modalities and sensitive prognostic markers that can decrease the mortality rate of HCC are required. HCC is a complex and heterogeneous tumor with multiple genetic aberrations. It has been well described that accumulation of genetic and epigenetic changes leads to the clonal selection of cancer cells harboring aggressive tumor behavior. Aberrant expression of cancer-related genes is one of the hallmarks of cancer cells and plays a role in hepatocarcinogenesis. Epigenetic alterations, such as the alteration of DNA methylation and histone modification in cancer cells, can also induce the activation and inactivation of cancer-related genes. Studies have shed light on the link between HCC-related genes and molecules, and a better understanding of the mechanisms of HCC pathogenesis could be translated into clinical biomarker tools. Moreover, analyses of genetic and epigenetic alterations have identified potential biomarkers that might be targeted therapeutically. In this review, we update the current knowledge of biomarkers in HCC, examine recently published literature, and introduce some representative molecules in each category.

Offprint requests to: Mitsuro Kanda, Department of Gastroenterological Surgery (Surgery II), Nagoya University Graduate School of Medicine, 65 Tsurumai-cho, Showa-ku, Nagoya 466-8550, Japan. e-mail: m-kanda@med.nagoya-u.ac.jp

DOI: 10.14670/HH-11-936

Key words: Hepatocellular carcinoma, Biomarker, Prognosis, Expression

Introduction

Hepatocellular carcinoma (HCC) is the most common type of primary liver cancer representing 85% of liver cancers (Marra et al., 2011), and ranks as the third most common cause of cancer-related death worldwide (Forner et al., 2012; Siegel et al., 2015). Patients with HCC generally present at an advanced stage due to compensated cirrhosis defined by the absence of pathognomonic symptoms, resulting in death within 6-20 months (El-Serag, 2012). Furthermore, the recurrence rate after curative resection remains high at approximately 70% (Poon et al., 2009). Therefore, complete cure of this disease is quite challenging, suggesting an urgent need for enhanced treatment modalities and sensitive prognostic markers that will dramatically decrease the mortality rate of HCC. Chronic infection with hepatitis virus is a major risk factor in the development of the HCC (Sukowati et al., 2016). Oncogenic properties of these viruses are related with their genotypic characteristics and the ability of viral proteins to interact with host proteins, thus altering the molecular pathway balance of cells (Sukowati et al., 2016).

HCC is a complex and heterogeneous tumor with multiple genetic aberrations (Marra et al., 2011). It has been well described that the accumulation of genetic and epigenetic changes leads to the clonal selection of cancer cells harboring aggressive tumor behavior (Marquardt et

al., 2015). Recently, next generation sequencing technology and bioinformatic analysis have been widely used to screen genetic alterations, which have helped us identify the biomarker carcinogenesis and progression of HCC (Li et al., 2017). Aberrant expression of cancer-related genes is one of the hallmarks of cancer cells and plays a role in hepatocarcinogenesis. However, epigenetic changes, such as the alteration of DNA methylation and histone modification in cancer cells, can also induce the activation and inactivation of cancer-related genes (Nishida and Kudo, 2013). Undertaking studies that link specific molecular classes of tumors or aberrations to therapeutic responses might translate understanding of molecular mechanisms of HCC pathogenesis into clinical tools.

In this article, we review the recent literature on molecular biomarkers related to HCC initiation and progression, which are potential diagnostic tools and therapeutic targets. Additionally, some representative molecules have been selected for this review based on the following criteria: 1) high novelty, 2) data from a large number of patients, and 3) solid data generated from functional analysis or *in vivo* studies.

Oncogenes

Genetic and epigenetic alterations of oncogenes such as point mutations, gene amplifications, changes in the promoter region, or copy number alterations due to viral infection or exposure to hepatotoxic stress alter the phenotypic nature of the hepatocyte, resulting in tumor development (Nishida and Kudo, 2016; Sia et al., 2017). As a consequence, expression of several altered oncogenes are contributory factors in cancer cell proliferation, invasion, and migration. Therefore, these oncogenes will be the novel diagnostic and prognostic factors in HCC resected patients, and these or upstream genes could be potential targets for unprecedented therapy. We have summarized in Table 1 genes upregulated in cancerous tissue or serum of HCC patients. Findings from several studies listed in Table 1 are summarized below (Chen et al., 2015a,b; Gao et al., 2015; Hu et al., 2015; Huang et al., 2015a-c; Hwang et al., 2015; Ji et al., 2015; Jiao et al., 2015; Kang et al., 2015; Lin et al., 2015; Tang et al., 2015; Xu et al., 2015; Zhang et al., 2015a,b; Zheng et al., 2015; Cui et al., 2016; Hou et al., 2016; Jiang et al., 2016; Liu et al., 2016; Shimizu et al., 2016; Tian et al., 2016; Yin et al., 2016; Wang et al., 2016; Xiao et al., 2016; Xu et al., 2016a,b; Zheng et al., 2017).

Ubiquitin-specific protease 22 (USP22)

USP22 is a novel human deubiquitinating enzyme. Recent studies have demonstrated that USP22 can inhibit the transcription of the p21 gene and might be related to clinical prognosis in malignancies (Zhang et al., 2008; Atanassov and Dent, 2011); however, the clinical significance of USP22 expression in patients with HCC has not yet been investigated. Tang et al. found that expression levels of USP22 were significantly higher in HCC than in normal liver tissues (Tang et al., 2015). High USP22 expression in HCC was significantly correlated with clinical stage and tumor grade. Kaplan-Meier analysis showed that elevated USP22 expression predicted poorer overall survival (OS) and recurrence-free survival (RFS) (Tang et al., 2015). High USP22 expression was also associated with shortened survival time in patients at advanced tumor stages and with high-grade HCC. Multivariate analysis revealed that USP22 expression was an independent prognostic parameter in HCC (Tang et al., 2015). These findings provide evidence that high USP22 expression might be important in tumor progression and serves as an independent molecular marker for a poor HCC prognosis. Thus, USP22 overexpression identifies patients at high risk and represents a novel therapeutic molecular target for this tumor.

Chromobox 6 (CBX6)

CBX6 is a member of the chromobox protein family and it may be a reader of different, as yet undetermined, trimethyllysine sites and may be capable of binding to RNA (Kaustov et al., 2011; Klauke et al., 2013). However, the role of CBX6 in HCC development and progression has not been explored. Zheng et al. demonstrated that CBX6 expression was increased in HCC cells and tumor tissues, and that increased expression was associated with poor outcomes (Zheng et al., 2017). They found that CBX6 expression was significantly correlated with larger tumor sizes and multiple tumors. Patients with higher CBX6 expression levels had significantly shorter RFS and OS, and increased CBX6 expression was an independent unfavorable prognostic factor in HCC patients (Zheng et al., 2017). They also conducted *in vitro* and *in vivo* studies with CBX-knockdown cells and demonstrated that CBX6 profoundly promoted HCC cell growth. Their investigation revealed that the S100A9/NF-κB/MAPK pathway was essential for mediating CBX6 function (Zheng et al., 2017). They concluded that CBX6 contributes to tumor progression and may be a novel prognostic biomarker for HCC, and a therapeutic target in the treatment of the disease.

Neurotrophin receptor-interacting melanoma antigen-encoding protein (NRAGE)

NRAGE is involved in the process of apoptosis through interactions with the p75 neurotrophin receptor and apoptosis-antagonizing transcription factor (Salehi et al., 2000). NRAGE functions as an oncogene in esophageal and lung cancers (Yang et al., 2014). However, there are currently no studies concerning NRAGE expression in HCC. Shimizu et al. aimed to assess the clinical significance of NRAGE expression in HCC, as well as its relevance as a novel biomarker of

Table 1. Oncogenes.

Symbol (location)	Biological function	Specimen	Detection methods	Pt	Survival	Relevant clinical factors	Functional analyses	Interacting molecules	*In vivo*	Reference
Early detection										
NEU1 (6p21.33)	Lysosomal enzyme	Tissue	QPCR	114	OS	Stage, AFP, differentiation	Proliferation, migration	(-)	(-)	Hou et al., 2016
TGFB1 (19q13.2)	A member of transforming growth factor-beta, superfamily	Circulating	ELISA	91	OS PFS	AFP	Viability	(-)	(-)	Lin et al., 2015
TRIM28 (19q13.43)	Transcription factor	Tissue	QPCR	116	OS	Size, stage	Proliferation	(-)	(-)	Wang et al., 2016a-c
USP22 (17p11.2)	Transcription factor	Tissue	QPCR, IHC	168	OS DFS	Size, stage, HG	Viability, apoptosis	(-)	(-)	Tang et al., 2015
WWP1 (8q21.3)	Regulator of protein degradation, transcription, and RNA splicing	Tissue	QPCR, IHC	149	OS PFS	Size, stage, HG, VI	Migration, invasion, apoptosis	(-)	(-)	Zhang et al., 2015a-e
Monitoring recurrences										
ATAD2 (8q24.13)	Mediator of assembly, operation, or disassembly of protein complexes	Tissue	IHC	182	OS DFS	Size, stage, VI	(-)	(-)	(-)	Hwang et al., 2015
BCORL1 (Xq26.1)	Transcription factor	Tissue	WB	86	OS DFS	Multiplicity, stage, VI	Migration, invasion	E-cadherin	(-)	Yin et al., 2016
CA9 (9p13.3)	Zinc metalloenzyme	Tissue	IHC	838	OS DFS	Size, AFP, VI, HG	(-)	(-)	(-)	Kang et al., 2015
CBX6 (22q13.1)	Transcription factor	Tissue	QPCR, IHC	313	OS DFS	Size, multiplicity	Apoptosis, colony	S100A9	Yes	Zheng et al., 2017
DHX33 (17p13.2)	RNA helicase	Tissue	QPCR, IHC, WB	520	OS DFS	Size, AFP	(-)	(-)	(-)	Tian et al., 2016
MCAM (11q23.3)	Cell adhesive molecule	Tissue	QPCR, IHC, WB	120	OS DFS	Stage, AFP, VI, satellite	Migration, invasion	IL8, STAT1	Yes	Jiang et al., 2016
NFE2L2 (21q21.3)	Transcription factor	Tissue	IHC	65	OS DFS	Size, differentiation	Viability, apoptosis	Bcl-xL, 1MMP-9	(-)	Zhang et al., 2015a-e
NRAGE (Xp11.22)	Tumor-specific antigen	Tissue	QPCR, IHC	151	DFS	Age, AFP	(-)	AATF	(-)	Shimizu et al., 2016
Prediction of survival										
ACTL6A (3q26.33)	Chromatin remodeling and nuclear migration	Tissue	QPCR, WB	100	OS DFS	Multiplicity, stage, AFP, VI, HG	Proliferation, migration, Invasion	OX2, Notch1	Yes	Xiao et al., 2016
CTBP2 (10q26.13)	Transcription factor	Tissue	IHC	100	OS	Stage, VI, HG	Migration, invasion	GLI1, SNAI1	Yes	Zheng et al., 2015
GIT1 (17q11.2)	Maintenance of cell cytoskeleton	Tissue	QPCR	130	OS DFS	Multiplicity, stage, VI, HG	Proliferation, viability, migration, invasion, apoptosis	ERK, MMP9	(-)	Chen et al., 2015a,b
GNA13 (17q24.1)	Transducer in transmembrane signaling	Tissue	QPCR, IHC, WB	246	OS DFS	Multiplicity, stage	Proliferation, invasion	(-)	(-)	Xu et al., 2016a,b
NKX6-1 (4q21.23)	Neural development and pancreatic cell differentiation	Tissue	QPCR, IHC, WB	231	OS DFS	Size, stage, differentiation	Proliferation, invasion	(-)	(-)	Huang et al., 2015a,b
PTOV1 (19q13.33)	Metabolic enzyme	Tissue	IHC	215	OS	Stage, cirrhosis, AFP, VI	(-)	(-)	(-)	Chen et al., 2015a,b
WASF3 (13q12.13)	Maintenance of cell cytoskeleton	Tissue	IHC	120	OS	Stage	Migration, invasion	(-)	(-)	Ji et al., 2015
Prediction of treatment response										
CBX4 (17q25.3)	Transcription factor	Tissue	IHC, WB	727	OS DFS	Stage, HG	TUNEL assay, cell counting kit-8 assay, chemoembolization	(-)	(-)	Jiao et al., 2015
CHMP4B (20q11.22)	Multivesicular body protein	Tissue	IHC, WB	93	OS	Stage, cirrhosis, AFP	Proliferation, apoptosis, cycle, doxorubicin	(-)	(-)	Hu et al., 2015
DLX2 (2q31.1)	Embryonic development	Tissue	IHC	124	OS	Differentiation	Proliferation, apoptosis, sorafenib	PCNA, Cyclin D1, Cyclin A.	(-)	Liu et al., 2016
HOXC8 (12q13.13)	Transcription factor	Tissue	IHC, WB	86	OS	Differentiation	Viability, apoptosis, oxaliplatin	Ki67	(-)	Xu et al., 2015
SLC16A4 (1p13.3)	Monocarboxylate transporter	Tissue	IHC, WB	318	OS DFS	Size, AFP, TACE treatment resistance	Proliferation, migration, invasion	AKT, HIF-1α.	(-)	Gao et al., 2015
SYF2 (1p36.11)	Cell cycle regulation	Tissue	IHC, WB	92	OS	Size, Ki-67, HG	Proliferation, apoptosis, colony, doxorubicin	Cyclin D1	(-)	Zhang et al., 2015a-e
TRIM32 (9q33.1)	Transcription factor	Tissue	QPCR, IHC, WB	116	OS	Size, HG	Proliferation, apoptosis, colony, oxaliplatin	(-)	(-)	Cui et al., 2016

Pt, number of patients; QPCR, quantitative real-time reverse transcription-polymerase chain reaction; IHC, immunohistochemistry; WB, western blotting; OS, overall survival; DFS, disease-free survival; PFS, progression-free survival; stage, UICC stage; AFP. Alfa fetoprotein; HG, histological grade; VI, vascular invasion; TACE, transcatheter arterial chemo-embolization.

tumor progression (Shimizu et al., 2016). They conducted quantitative polymerase chain reaction (qPCR) and immunohistochemistry (IHC) to determine the NRAGE expression level in 151 resected HCC samples and adjacent noncancerous liver tissues. As a result, higher levels of NRAGE was correlated significantly with a shorter disease-specific survival time and was identified as an independent prognostic factor using multivariate analysis (Shimizu et al., 2016). Therefore, they concluded that NRAGE mediates the progression of HCC and may serve as a novel biomarker of the malignant phenotype of the disease, and they suggested that analysis of NRAGE expression may enhance the clinical management of HCC.

Actin-like 6A (ACTL6A)

ACTL6A is a member of adenosine triphosphate-dependent SWI/SNF-like BRG1/brm-associated factor chromatin remodeling complexes and encodes a 53-kDa subunit of the BAF complex in mammals (Zhao et al., 1998). Xiao et al. found that ACTL6A plays an essential role in metastasis and epithelial-mesenchymal transition (EMT) of HCC (Xiao et al., 2016). An elevated level of ACTL6A in HCC was correlated with aggressive clinicopathological features and was an independent poor prognostic factor for OS and disease-free survival (DFS) in HCC patients. Ectopic expression of ACTL6A markedly promoted HCC cells migration, invasion, as well as EMT *in vitro*, and promoted tumor growth and metastasis in a mouse xenograft model (Xiao et al., 2016). Opposite results were observed when ACTL6A was knocked down. Further studies indicate that ACTL6A might manipulate sex-determining region Y (SRY)-box 2 (SOX2) expression and then activate Notch1 signaling. They concluded that ACTL6A promotes metastasis and EMT by SOX2/Notch1 signaling, indicating a prognostic biomarker candidate and a potential therapeutic target for HCC (Xiao et al., 2016).

Charged multivesicular body protein 4B (CHMP4B)

CHMP4B is thought to be one of the core components of ESCRT-III that facilitates the budding and scission of membrane vesicles (Wollert et al., 2009). Hu et al. explored the prognostic significance of CHMP4B in human HCC and its impact on the physiology of HCC cells, and they found that CHMP4B could be a promising prognostic biomarker as well as a potential therapeutic target in HCC (Hu et al., 2015). Their analysis showed that CHMP4B was significantly upregulated in HCC tissues. They found that high CHMP4B expression was correlated with multiple clinicopathological variables, including AFP, cirrhosis, AJCC stage, Ki-67 expression, and poor prognosis. More importantly, CHMP4B served as an independent prognostic factor for survival in HCC patients (Hu et al., 2015). Using HCC cell cultures, they showed that the expression of CHMP4B was progressively upregulated after cells were released from serum starvation. Flow cytometry and CCK-8 assays indicated that interference of CHMP4B led to cell cycle arrest and proliferative impairment of HCC cells. Additionally, depletion of CHMP4B expression could increase the sensitivity of HepG2 and Huh7 cells to doxorubicin (Hu et al., 2015).

Tumor suppressor genes

A few signaling pathways are important for occurrence and growth of HCC and these pathways are negatively suppressed by tumor suppressor genes (Inokawa et al., 2016). Tumor suppressor genes are down regulated by mutations, loss of chromosomal regions, or promoter methylation (Martin, 2008). Thus, downregulation of tumor suppressor genes may be diagnostic and prognostic markers in HCC patients. Here in Table 2 we list recently reported tumor suppressor genes regulated by mechanisms other than promoter hypermethylation and summarize some of the key findings from some of these studies (Hirata et al., 2015; Jiang et al., 2015; Kanda et al., 2015; Liu et al., 2015a,b; Shi et al., 2015; Wan et al., 2015; Wang et al., 2015a-c; Zhang et al., 2015a-e; Shi et al., 2016; Song et al., 2016).

B cell translocation gene 1 (BTG1)

BTG1 was originally identified as a translocation partner of the cMyc gene in a patient with B cell chronic lymphocytic leukemia (Cho et al., 2004). Experiments showed that BTG1 is a Bcl2-regulated mediator of apoptosis and that it negatively regulates cell proliferation in breast and ovarian cancer. However, the role of BTG1 in gastroenterological malignancies including HCC remains unclear. We have identified several HCC-related genes using expression and epigenetic analyses (Kanda et al., 2015). From the exhaustive expression analysis obtained via our microarray data, BTG1 was identified as a candidate tumor suppressor gene in HCC. Decreased expression of BTG1 mRNA was confirmed in a majority of HCC cell lines (89%) and clinical HCC tissues (85%) compared with noncancerous liver tissues. Mutations or promoter hypermethylation were not identified in HCC cell lines. BTG1 mRNA expression levels were not influenced by background liver status (Kanda et al., 2015). Downregulation of BTG1 mRNA in HCC was significantly associated with shorter disease-specific and RFS rates. Multivariate analysis of disease-specific survival rates identified BTG1 mRNA downregulation as an independent prognostic factor in HCC (Kanda et al., 2015). Our results indicate that altered BTG1 expression might affect hepatocarcinogenesis and may represent a novel biomarker in HCC carcinogenesis and progression.

Forkhead box family 2 (FOXF2)

FOXF2 is a member of the FOX family of transcription factors. FOXF2 plays a key role in several tumors but its expression and role in HCC remains unknown (Nik et al., 2013). Shi et al. surveyed the expression levels of FOXF2 using IHC, western blots, and real-time PCR (Shi et al., 2016). They analyzed FOXF2 expression in 295 clinicopathologically characterized HCC cases. Using RNA interference, they investigated the effects of FOXF2 depletion on tumor cell behavior *in vitro*. FOXF2 downregulation was observed in HCC tissues compared with peritumorous tissues, and its expression levels were closely correlated with OS and RFS in patients with HCC. RNAi-mediated silencing of the FOXF2 gene in the MHCC-97H cell line significantly promoted proliferation and was anti-apoptotic (Shi et al., 2016). They concluded that their results indicated that FOXF2 may serve as a prognostic biomarker in HCC and might be a promising target in the treatment of patients with HCC.

CYP3A5

CYP3A5 is a cytochrome P450 protein that functions in the liver metabolism of many carcinogens and cancer drugs (Windmill et al., 1997; Zhou, 2008). However, it has not been thought to directly affect cancer progression. Jiang et al. performed a perspective study and demonstrated that CYP3A5 was downregulated in multiple cohorts of human HCC examined (Jiang et al., 2015). Lower CYP3A5 levels were associated with more aggressive vascular invasion, poor differentiation, shorter time to disease recurrence after treatment, and worse OS. Mechanistic investigations showed that CYP3A5 overexpression limited MMP2/9 function and suppressed HCC migration and invasion *in vitro* and *in vivo* by inhibiting AKT signaling. Notably, AKT phosphorylation at Ser473 was inhibited in CYP3A5-overexpressing HCC cells, an event requiring mTORC2 but not rictor-mTOR complex formation (Jiang et al., 2015). CYP3A5-induced ROS accumulation was found to be a critical

Table 2. Tumor suppressor genes.

Symbol (location)	Biological function	Specimen	Detection methods	Pt	Survival	Relevant clinical factors	Functional analyses	Interacting molecules	*In vivo*	Reference
Early detection										
BTG1 (12q21.33)	B-cell differentiation	Tissue	QPCR, IHC	151	DFS	Size, stage, PIVKA2, differentiation, VI, serosa infiltration	(-)	PIVKA2	(-)	Kanda et al., 2015
DACH1 (13q21.33)	Transcription factor	Tissue	IHC, WB	95	OS	Stage, Ki-67, HG	Viability, migration, invasion, colony	GSK3β, β-catenin	(-)	Liu et al., 2015a,b
Monitoring recurrences										
BARX2 (11q24.3)	Cell adhesive molecule	Tissue	QPCR, IHC, WB	231	OS	Stage, differentiation	(-)	(-)	(-)	Zhang et al., 2015a-e
CX3CR1 (3p22.2)	Chemokine receptor	Tissue	QPCR, IHC, WB	240	OS DFS	Differentiation, VI	Migration, invasion	CCR7	Yes	Shi et al., 2015
FOXF2 (6p25.3)	Transcription factor	Tissue	IHC	295	OS DFS	Size, ifferentiation, HG	Proliferation, apoptosis	(-)	(-)	Shi et al., 2016
OTUD7B (1q21.2)	Metabolic enzyme	Tissue	IHC	230	OS DFS	Size, stage, satellite nodule, VI	(-)	MMP-9	(-)	Wang et al., 2015a,b
Prediction of survival										
CYP3A5 (7q22.1)	Metabolic enzyme	Tissue	IHC	159	OS DFS	(-)	Migration, invasion	mTORC2/Akt	Yes	Jiang et al., 2015
DLC1 (8p22)	Rho GTPase activating protein	Tissue	IHC	80	OS	Stage, differentiation, VI, dissemination	(-)	Rho A, ROCK2, moesin	(-)	Song et al., 2016
PRRX1 (1q24.2)	Transcription co-activator	Tissue	QPCR	62	OS	(-)	Proliferation, colony, sphere, irradiation	CD13, CD133, EpCAM	(-)	Hirata et al., 2015
TPD52 (8q21.13)	DNA repair and vesicle trafficking	Tissue	QPCR, IHC, WB	154	OS DFS	Stage	(-)	P21, P53, BCL2, P-GSK-3β	(-)	Wang et al., 2016a,b
TRIM26 (6p22.1)	DNA-binding protein	Tissue	QPCR, IHC	242	OS DFS	Stage, AFP	Proliferation, migration, invasion, colony	(-)	(-)	Wang et al., 2015a,b

Pt, number of patients; QPCR, quantitative real-time reverse transcription-polymerase chain reaction; IHC, immunohistochemistry; WB, western blotting; OS, overall survival; DFS, disease-free survival; stage, UICC stage; PIVKA2, protein induced by vitamin K absence 2; VI, vascular invasion; HG, histological grade; AFP, Alfa fetoprotein.

upstream regulator of mTORC2 activity, consistent with evidence of reduced GSH redox activity in most clinical HCC specimens with reduced metastatic capacity (Jiang et al., 2015). They concluded that their results defined CYP3A5 as a suppressor of HCC pathogenesis and metastasis with potential utility as a prognostic biomarker in HCC.

Hypermethylation of tumor supressor genes

DNA methylation is the earliest discovery of epigenetic regulation of gene expression (Gibbs et al., 2010). Hypermethylation of CpG islands is generally associated with tumor suppressor gene silencing under premalignant conditions, such as chronic hepatitis or liver cirrhosis (Lee et al., 2014). DNA demethylation is a key regulator not only in tumor progression, but also in tumorigenesis of HCC from premalignant conditions, such as chronic hepatitis or liver cirrhosis (Lee et al., 2017). Therefore, detecting hypermethylation may predict the occurrence of HCC in patients with the premalignant conditions described above. In Table 3, we have also included candidate methylated tumor suppressor genes in HCC (Ezaka et al., 2015; He et al., 2015; Oya et al., 2015; Ding et al., 2016; Wang et al., 2016a-c).

Forkhead box D3 (FOXD3)

FOXD3, which belongs to the forkhead family of transcription factors, was originally identified in embryonic stem cells (Pan and Thomson, 2007). Recent studies showed that FOXD3 is a regulator in tumor progression (Schmid and Muller, 2013). Aberrant epigenetic regulation of the FOXD3 promoter, such as DNA hypermethylation, is observed in several human cancers. A recent study reported that FOXD3 might be negatively correlated with AKT2 in HCC tissues. However, the function and mechanism of action of FOXD3 in hepatocarcinogenesis and progression still require further investigation. He et al. found that FOXD3 was decreased in HCC tissues and correlated with differentiation, AFP, and poor survival in HCC patients (He et al., 2015). Downregulation of FOXD3 in HCC tissues was mainly due to promoter hypermethylation. In vitro and *in vivo* functional results showed that ectopic FOXD3 inhibited proliferation, migration, EMT, and invasion in HepG2 and SMMC-7721 cells, while FOXD3 depletion in HepG2 and QGY-7701 cells had opposite effects. Moreover, FOXD3 was sufficient to suppress tumor growth and pulmonary metastatic potential in mice. Their findings suggest that downregulation of FOXD3, due to promoter hypermethylation, plays an important role in the progression of HCC and may be a promising prognostic biomarker in HCC patients (He et al., 2015).

Adherens junctions associated protein 1 (AJAP1)

AJAP1 is a component of adherens junctions, which colocalizes with and apparently integrates into E-cadherin-mediated adherens junctions (Lin et al., 2012). Ezaka et al. focused on AJAP1 as a candidate novel HCC-related gene (Ezaka et al., 2015). Their previous microarray study revealed that AJAP1 expression was reduced in HCCs compared with corresponding noncancerous liver tissues. They conducted this study to investigate the expression and regulatory mechanisms of

Table 3. Hypermethylation of tumor suppressor genes.

Symbol (location)	Biological function	Specimen	Detection methods	Pt	Survival	Relevant clinical factors	Functional analyses	Interacting molecules	*In vivo*	Reference
Early detection										
FOXD3 (1p31.3)	Transcription factor	Tissue	QPCR, IHC, WB, MSP	119	OS	Size	Proliferation, migration, invasion, colony	(-)	Yes	He et al., 2015
Monitoring recurrence										
AJAP1 (1p36.32)	Cell adhesion molecule	Tissue	QPCR, IHC, MSP	144	OS DFS	Size, PIVKA-II	(-)	SRC	(-)	Ezaka et al., 2015
DPYSL3 (5q32)	Cell adhesion molecule	Tissue	QPCR, IHC	151	DFS	AFP, PIVKA-II, capsule, serosal infiltration	Proliferation, migration, invasion	VEGF, FAK	(-)	Oya et al., 2015
Prediction of survival										
PDCD4 (10q25.2)	Transcription factor	Tissue	QPCR, Bisulfite sequencing	56	OS	Differentiation	(-)	(-)	(-)	Ding et al., 2016
RASSF10 (11p15.3)	Regulator of vesicular trafficking	Tissue	QPCR, IHC, MSP	48	OS DFS	Stage, cirrhosis, differentiation, tumor thrombus	Proliferation, migration, invasion, colony	(-)	Yes	Wang et al., 2016a-c

Pt, number of patients; QPCR, quantitative real-time reverse transcription-polymerase chain reaction; IHC, immunohistochemistry; WB, western blotting; MSP, methylation specific polymerase chain reaction; OS, overall survival; DFS, disease-free survival; stage, UICC stage; AFP. Alfa fetoprotein; PIVKA2, protein induced by vitamin K absence 2.

AJAP1 to establish whether AJAP1 might serve as novel biomarker for progression and recurrence of HCC. AJAP1 expression was reactivated after demethylation of its promoter. AJAP1 mRNA levels correlated inversely with those of SRC in HCC cell lines and tissues, and AJAP1 mRNA levels were suppressed in HCC tissues. Low levels of AJAP1 mRNA in patients were associated significantly with elevated levels of markers, larger tumor size, serosal infiltration, vascular invasion, hypermethylation of the AJAP1 promoter, and copy number loss at the AJAP1 locus (Ezaka et al., 2015). Multivariate analysis identified low AJAP1 expression as an independent factor for predicting DFS. They concluded that AJAP1 is a key molecule associated with recurrence of HCC and that hypermethylation of the AJAP1 promoter is a key regulatory mechanism controlling AJAP1 expression (Ezaka et al., 2015).

RAS-association domain family 10 (RASSF10)

RASSF10 gene is a candidate tumor suppressor gene and the most recently discovered member of the RASSF family (Richter et al., 2009). Hypermethylation of the RASSF10 promoter region results in inactivation of the gene in several cancers (Li et al., 2014). RASSF10 activates the P53 signaling pathway and inhibits the Wnt/β-catenin signaling pathway, two major signaling cascades involved in HCC initiation and progression. RASSF10 overexpression also potentiates docetaxel-induced tumor cell apoptosis (Huang et al., 2015c). However, understanding of the function of RASSF10 in cancer is incomplete, and its role in hepatocarcinogenesis is unknown. Wang et al. examined RASSF10 expression in HCC and its role in hepatocarcinogenesis (Wang et al., 2016a). They found that hypermethylation of the RASSF10 promoter region downregulated its expression in HCC, and that RASSF10 expression was an independent prognostic factor for patient survival and tumor recurrence (Wang et al., 2016a). RASSF10 hypermethylation was associated with polycyclic aromatic hydrocarbon and aflatoxin B1 exposure in HCC tissues, and RASSF10 overexpression suppressed the growth of HCC *in vitro* and *in vivo*. These results indicate that RASSF10 is a potential therapeutic target and may be a useful biomarker in HCC prognosis (Wang et al., 2016a-c).

MicroRNA

MicroRNAs (miRNAs) are short noncoding RNAs, about 22 nucleotides long, that have been found to suppress oncogenes and/or tumor suppressor genes in HCC (Shen et al., 2016; Xu et al., 2016a,b). Therefore, miRNAs are as important as tumor suppressors or oncogenes in cancer. In addition, the expression level of miRNAs is unique in HBV and HCV infections, and it is associated with liver disease progression. Hence, miRNA expression levels in HCC can predict not only the prognosis in HCC patients but also the probability of HCC occurrence in viral hepatitis patients (Murakami and Kawada, 2017). MiRNAs are also expected to be potential novel therapeutic targets in HCC (Huan et al., 2016). In Table 4 we summarize miRNAs that have been recently published and that are associated with HCC progression or suppression. Some of these miRNAs are presented below regarding function and relation to prognosis (Yao et al., 2015; Yu et al., 2015; Wang et al., 2015a-c; Zhuang et al., 2015; Zhang et al., 2015a-e; Huang et al., 2015a,b; Zhao et al., 2015; Feng et al., 2015; Song et al., 2015; Li et al., 2015b; Wu et al., 2016).

MIR150

Studies have demonstrated that miR-150 is predictive of a favorable prognosis in patients with epithelial ovarian cancer, and that it inhibits cell invasion and metastasis by suppressing the transcriptional repressor ZEB1 (Jin et al., 2014). Yu et al. previously reported that overexpression of miR-150 contributed to the suppression of activated hepatic stellate cells (Yu et al., 2015). They evaluated whether serum miR-150 could serve as a new biomarker for the diagnosis and prognosis of HBV-related HCC in patients. Serum miR-150 levels were significantly reduced in HCC patients compared with healthy controls and chronic hepatitis B patients. Serum miR-150 levels were increased after surgery and decreased after tumor recurrence. Receiver operating characteristic curve analysis suggested that serum miR-150 had significant diagnostic value in HBV-related HCC (Yu et al., 2015). Moreover, Kaplan-Meier curve analysis revealed that HCC patients with lower serum miR-150 had a significantly shortened OS. Multivariable analysis indicated that the serum miR-150 level was an independent risk factor for OS. They concluded that serum miR-150 can serve as a noninvasive biomarker in the diagnosis and prognosis of HCC patients (Yu et al., 2015).

MIR451

Previously, Wang et al. have shown that miR-451 upregulation inhibits growth and induces apoptosis in non-small cell lung cancer cells (Wang et al., 2011). However, the clinicopathological and prognostic value of miR-451 and its role in EMT and metastasis of HCC cells remains largely unclear. They showed that reduced miR-451 levels in HCC tissues was observed to be significantly correlated with advanced clinical stage, metastasis, and worse DFS or OS (Huang et al., 2015a). Importantly, miR-451 could inhibit migration and invasion *in vitro*, as well as *in vivo* metastasis of HCC cells through regulating the EMT process. Moreover, the oncogene c-Myc was identified as a direct and functional target of miR-451 in HCC cells. Furthermore, miR-451 downregulation-induced c-Myc overexpression led to

the activation of Erk1/2 signaling, which induced acquisition of the EMT phenotype (Huang et al., 2015a). Collectively, these data demonstrate that miR-451 is a novel prognostic biomarker in HCC patients that functions as a potential metastasis inhibitor in HCC cells through activation of the Erk1/2 signaling, at least partially by targeting c-Myc (Huang et al., 2015a). Thus, targeting the miR-451/c-Myc/Erk1/2 axis may be a potential strategy for the treatment of metastatic HCC.

MiR137

Wu et al. demonstrated that HEY2 expression was inhibited by miR-137 (Wu et al., 2016). HEY2 exerts biological effects on mammalian organ development. Overexpression of HEY2 has been reported in prostate cancer (Tradonsky et al., 2012) and pancreatic ductal adenocarcinomas (Cavard et al., 2009). hemangiomas. However, the role of HEY2 and its clinical significance in HCC remains elusive. They investigated the expression of HEY2 in HCC at both mRNA and protein levels. High HEY2 expression was correlated with tumor multiplicity, tumor differentiation, and TNM stage. HEY2 expression was significantly associated with poor OS and DFS in a training cohort of 361 patients with HCC (Wu et al., 2016). The prognostic implication of HEY2 was validated in another cohort of 169 HCC patients. Multivariate analysis indicated that HEY2 was an independent factor of OS in HCC. In clinical samples, HEY2 expression was inversely associated with miR-137 expression. Furthermore, overexpression of HEY2 increased cell viability, colony formation, and cell migration, whereas knockdown of HEY2 resulted in the opposite phenotype (Wu et al., 2016). Collectively, their data suggest that HEY2 is a promising biomarker for unfavorable outcomes and a novel therapeutic target for the clinical management of HCC.

Long non-coding RNAs

Long non-coding RNAs (lncRNAs) are non-coding RNAs which are >200 nucleotides in length and play a vital role in the regulation of gene expression through diverse mechanisms (Zhang and Zhu, 2014). Recently,

Table 4. MicroRNAs.

Symbol (location)	Specimen	Detection methods	Pt	Survival	Relevant clinical factors	Functional analyses	Interacting molecules	*In vivo*	Reference
Early detection									
MIR21 (17q23.1)	Tissue Circulating	QPCR, IHC, ELISA	97	OS	Hepatitis B, stage, differentiation, VI	(-)	PDCD4, PTEN	(-)	Wang et al., 2015a-c
MIR128-2 (3p22.3)	Circulating	QPCR	182	OS	AFP	(-)	(-)	(-)	Zhuang et al., 2015
MIR150 (19q13.33)	Circulating	QPCR	350	OS	Stage	(-)	(-)	(-)	Yu et al., 2015
Monitoring recurrence									
MIR146a (5q33.3)	Tissue	QPCR, IHC	53	DFS	(-)	Proliferation, migration, invasion, apoptosis, cycle	HAb18G, VEGF	Yes	Zhang et al., 2015a-e
MIR424 (Xq26.3)	Circulating	QPCR	95	OS, DFS	Stage, AFP, VI	(-)	(-)	(-)	Yao et al., 2015
MIR451 (17q11.2)	Tissue	QPCR	88	OS DFS	Stage, VI, HG	Migration, invasion, colony	c-Myc	Yes	Huang et al., 2015a,b
Prediction of survival									
MIR-128-2 (3p22.3)	Circulating	QPCR	182	OS	AFP	(-)	(-)	(-)	Zhuang et al., 2015
MIR137 (1p21.3)	Tissue	QPCR	351	OS DFS	Multiplicity, differentiation	Colony, trans-well assay	HEY2	Yes	Wu et al., 2016
MIR194 (1q41)	Tissue	QPCR	56	OS DFS	Stage, HG	Viability, apoptosis, cycle	MAP4K4	Yes	Zhao et al., 2015
MIR200a (1p36.33)	Tissue	QPCR	115	OS DFS	(-)	Viability, migration, invasion, colony	MACCI	(-)	Feng et al., 2015
MIR-622 (13q31.3)	Tissue	QPCR	56	OS	Hepatitis B, cirrhosis, VI	Proliferation, apoptosis, colony	JNK, NF-kB, MAP4K4	Yes	Song et al., 2015
Prediction of treatment response									
MIR34a-p (1p36.22)	Tissue	In situ hybridization	114	OS	(-)	Proliferation, apoptosis, cisplatin	AXL	(-)	Li et al., 2015a,b

Pt, number of patients; QPCR, quantitative real-time reverse transcription-polymerase chain reaction; IHC, immunohistochemistry; ELISA, Enzyme-Linked Immuno-Sorbent Assay; OS, overall survival; DFS, disease-free survival; UICC stage; VI, vascular invasion; AFP. Alfa fetoprotein; HG, histological grade.

many studies revealed that lncRNAs play a crucial role in various biological processes in HCC, such as initiation, progression, metastasis, treatment, and prognosis (Liu et al., 2015a,b). In Table 5, we have listed the lncRNAs that are related to HCC progression and prognosis, and summarized them below (Li et al., 2015a,b; Guo et al., 2015, 2016; Hua et al., 2015; Peng and Fan, 2015; Huang et al., 2016; Li et al., 2016a,b).

ZEB1-AS1

ZEB1 has the consistent inverse correlation with E-cadherin across various types of cancers (Valenta et al., 2012). ZEB1 not only suppresses the expression of E-cadherin but also activates mesenchymal genes in the EMT process (Wellner et al., 2009). However, factors that regulate ZEB1 expression are poorly defined. The effects of a specific differentially expressed lncRNA (termed ZEB1-AS1) on tumor progression were investigated *in vitro* and *in vivo*. Li et al. found that ZEB1-AS1 was frequently upregulated in HCC samples, especially in metastatic tumor tissues (Li et al., 2016b). DNA methylation analysis showed tumor-specific ZEB1-AS1 promoter hypomethylation. Aberrant methylation is tightly correlated with overexpression of ZEB1-AS1 in HCC. Patients with ZEB1-AS1 hypomethylation or with high ZEB1-AS1 expression have poor RFS. Functionally, ZEB1-AS1 promotes tumor growth and metastasis, and acts as an oncogene in HCC. ZEB1 inhibition partially abrogates ZEB1-AS1-induced EMT and cancer metastasis (Li et al., 2016a,b). Their results provide novel insights into the function of lncRNA-driven hepatocarcinogenesis, highlight the important role of ZEB1-AS1 in HCC progression, and indicate that ZEB1-AS1 might serve as a valuable prognostic biomarker in HCC.

ANRIL

A recently identified lncRNA, named CDKN2B antisense RNA 1 (ANRIL), is transcribed in the opposite direction from the INK4B-ARF-INK4A gene cluster (Yap et al., 2010). Zhang et al. found that ANRIL was upregulated in gastric cancer tissues and was associated with a poor prognosis (Zhang et al., 2014). However, the prognostic role and underlying mechanism of ANRIL in HCC is still unknown. Hua et al. explored ANRIL expression patterns and its correlation with clinicopathological features in HCC (Hua et al., 2015). ANRIL expression in HCC tissues was significantly higher than in adjacent nontumor tissues. Expression of ANRIL was remarkably associated with histologic grade and TNM stage in HCC patients. In addition, HCC patients with higher ANRIL expression had significantly poorer OS. Multivariate analysis suggested that high ANRIL expression was an independent predictor of poor prognosis. Moreover, *in vitro* assays revealed that decreased expression of ANRIL could suppress cell proliferation, migration, and invasion of HCC cells. They concluded that their results suggest that ANRIL may serve as an efficient clinical biomarker and as a therapeutic target in HCC patients.

Table 5. Long non-coding RNAs.

Symbol (location)	Specimen	Detection methods	Pt	Survival	Relevant clinical factors	Functional analyses	Interacting molecules	*In vivo*	Reference
Early detection									
ZFAS1 (20q13.13)	Tissue	QPCR	113	OS DFS	VI	Invasion	MIR-150, MMP14, MMP16.	Yes	Li et al., 2015a,b
ZEB1-AS1 (10p11.22)	Tissue	QPCR, MSP	120	OS DFS	VI	Proliferation, migration, invasion	ZEB1	Yes	Li et al., 2016a,b
Monitoring recurrence									
ICAM-1 (19p13.2)	Tissue	QPCR	617	OS, DFS	(-)	(-)	(-)	(-)	Guo et al., 2016
NEAT1 (11q13.1)	Tissue	QPCR	95	(-)	Stage, multiplicity, VI, portal vein embolus, capsular infiltration	(-)	MDTH, NM23, MALAT1	(-)	Guo et al., 2015
Prediction of survival									
ANRIL (9p21.3)	Tissue	QPCR	130	OS	Stage, HG	Proliferation, migration, invasion	(-)	(-)	Hua et al., 2015
DGCR5 (22q11.21)	Tissue Circulating	QPCR	120	OS	VI	(-)	(-)	(-)	Huang et al., 2016
GAS5 (1q25.1)	Tissue	QPCR, In situ hybridization	50	OS	Differentiation	Proliferation, invasion	Vimentin	(-)	Li et al., 2016a,b
PANDAR (6p21.2)	Tissue	QPCR	482	OS DFS	Stage, cirrhosis, AFP, multiplicity, VI	Proliferation	(-)	(-)	Peng and Fan, 2015

Pt, number of patients; QPCR, quantitative real-time reverse transcription-polymerase chain reaction; MSP, methylation specific polymerase chain reaction; IHC, immunohistochemistry; WB, western blotting; OS, overall survival; DFS, disease-free survival; UICC stage; AFP. Alfa fetoprotein, HG, histological grade; VI, vascular invasion.

Conclusion

We reviewed emerging evidence on molecular biomarkers of HCC according to the following categories; oncogenes, tumor suppressor genes, methylated tumor suppressor genes, miRNAs, and lncRNAs. The investigation of biomarker combinations might provide more accurate and valuable information for the future personal HCC diagnosis and/or prognosis. Although, we are still a long way from reaching our goal, the accumulation of knowledge on genetic and epigenetic factors is of key importance in making breakthroughs in the management of patients with HCC.

References

Atanassov B.S. and Dent S.Y. (2011). Usp22 regulates cell proliferation by deubiquitinating the transcriptional regulator fbp1. EMBO Rep. 12, 924-930.

Cavard C., Audebourg A., Letourneur F., Audard V., Beuvon F., Cagnard N., Radenen B., Varlet P., Vacher-Lavenu M.C., Perret C. and Terris B. (2009). Gene expression profiling provides insights into the pathways involved in solid pseudopapillary neoplasm of the pancreas. J. Pathol. 218, 201-209.

Chen J., Yang P., Yang J., Wen Z., Zhang B. and Zheng X. (2015a). Git1 is a novel prognostic biomarker and facilitates tumor progression via activating erk/mmp9 signaling in hepatocellular carcinoma. Onco. Targets Ther. 8, 3731-3742.

Chen S.P., Zhang L.S., Fu B.S., Zeng X.C., Yi H.M. and Jiang N. (2015b). Prostate tumor overexpressed 1 is a novel prognostic marker for hepatocellular carcinoma progression and overall patient survival. Medicine (Baltimore) 94, e423.

Cho J.W., Kim J.J., Park S.G., Lee D.H., Lee S.C., Kim H.J., Park B.C. and Cho S. (2004). Identification of b-cell translocation gene 1 as a biomarker for monitoring the remission of acute myeloid leukemia. Proteomics 4, 3456-3463.

Cui X., Lin Z., Chen Y., Mao X., Ni W., Liu J., Zhou H., Shan X., Chen L., Lv J., Shen Z., Duan C., Hu B. and Ni R. (2016). Upregulated trim32 correlates with enhanced cell proliferation and poor prognosis in hepatocellular carcinoma. Mol. Cell Biochem. 421, 127-137.

Ding X., Cheng X., Gong M., Chen X., Yin F. and Lai K. (2016). Hypermethylation and expression silencing of pdcd4 gene in hepatocellular carcinoma: A consort study. Medicine (Baltimore) 95, e2729.

El-Serag H.B. (2012). Epidemiology of viral hepatitis and hepatocellular carcinoma. Gastroenterology 142, 1264-1273 e1261.

Ezaka K., Kanda M., Sugimoto H., Shimizu D., Oya H., Nomoto S., Sueoka S., Tanaka Y., Takami H., Hashimoto R., Okamura Y., Yamada S., Fujii T., Nakayama G., Koike M., Fujiwara M. and Kodera Y. (2015). Reduced expression of adherens junctions associated protein 1 predicts recurrence of hepatocellular carcinoma after curative hepatectomy. Ann. Surg. Oncol. 22 (Suppl 3), S1499-1507.

Feng J., Wang J., Chen M., Chen G., Wu Z., Ying L., Zhuo Q., Zhang J. and Wang W. (2015). Mir-200a suppresses cell growth and migration by targeting macc1 and predicts prognosis in hepatocellular carcinoma. Oncol. Rep. 33, 713-720.

Forner A., Llovet J.M. and Bruix J. (2012). Hepatocellular carcinoma. Lancet 379, 1245-1255.

Gao H.J., Zhao M.C., Zhang Y.J., Zhou D.S., Xu L., Li G.B., Chen M.S. and Liu J. (2015). Monocarboxylate transporter 4 predicts poor prognosis in hepatocellular carcinoma and is associated with cell proliferation and migration. J. Cancer Res. Clin. Oncol. 141, 1151-1162.

Gibbs J.R., van der Brug M.P., Hernandez D.G., Traynor B.J., Nalls M.A., Lai S.L., Arepalli S., Dillman A., Rafferty I.P., Troncoso J., Johnson R., Zielke H.R., Ferrucci L., Longo D.L., Cookson M.R. and Singleton A.B. (2010). Abundant quantitative trait loci exist for DNA methylation and gene expression in human brain. PLoS Genet. 6, e1000952.

Guo S., Chen W., Luo Y., Ren F., Zhong T., Rong M., Dang Y., Feng Z. and Chen G. (2015). Clinical implication of long non-coding rna neat1 expression in hepatocellular carcinoma patients. Int. J. Clin. Exp. Pathol. 8, 5395-5402.

Guo W., Liu S., Cheng Y., Lu L., Shi J., Xu G., Li N., Cheng K., Wu M., Cheng S. and Liu S. (2016). Icam-1-related noncoding rna in cancer stem cells maintains icam-1 expression in hepatocellular carcinoma. Clin. Cancer Res. 22, 2041-2050.

He G., Hu S., Zhang D., Wu P., Zhu X., Xin S., Lu G., Ding Y. and Liang L. (2015). Hypermethylation of foxd3 suppresses cell proliferation, invasion and metastasis in hepatocellular carcinoma. Exp. Mol. Pathol. 99, 374-382.

Hirata H., Sugimachi K., Takahashi Y., Ueda M., Sakimura S., Uchi R., Kurashige J., Takano Y., Nanbara S., Komatsu H., Saito T., Shinden Y., Iguchi T., Eguchi H., Atsumi K., Sakamoto K., Doi T., Hirakawa M., Honda H. and Mimori K. (2015). Downregulation of prrx1 confers cancer stem cell-like properties and predicts poor prognosis in hepatocellular carcinoma. Ann. Surg. Oncol. 22 (Suppl. 3), S1402-1409.

Hou G., Liu G., Yang Y., Li Y., Yuan S., Zhao L., Wu M., Liu L. and Zhou W. (2016). Neuraminidase 1 (NEU1) promotes proliferation and migration as a diagnostic and prognostic biomarker of hepatocellular carcinoma. Oncotarget 7, 64957-64966.

Hu B., Jiang D., Chen Y., Wei L., Zhang S., Zhao F., Ni R., Lu C. and Wan C. (2015). High CHMP4B expression is associated with accelerated cell proliferation and resistance to doxorubicin in hepatocellular carcinoma. Tumour Biol. 36, 2569-2581.

Hua L., Wang C.Y., Yao K.H., Chen J.T., Zhang J.J. and Ma W.L. (2015). High expression of long non-coding rna anril is associated with poor prognosis in hepatocellular carcinoma. Int. J. Clin. Exp. Pathol. 8, 3076-3082.

Huan L., Liang L.H. and He X.H. (2016). Role of micrornas in inflammation-associated liver cancer. Cancer Biol. Med. 13, 407-425.

Huang J.Y., Zhang K., Chen D.Q., Chen J., Feng B., Song H., Chen Y., Zhu Z., Lu L., De W., Wang R. and Chen L.B. (2015a). Microrna-451: Epithelial-mesenchymal transition inhibitor and prognostic biomarker of hepatocelluar carcinoma. Oncotarget 6, 18613-18630.

Huang L.L., Zhang Y., Zhang J.X., He L.J., Lai Y.R., Liao Y.J., Tian X.P., Deng H.X., Liang Y.J., Kung H.F., Xie D. and Zhu S.L. (2015b). Overexpression of nkx6.1 is closely associated with progressive features and predicts unfavorable prognosis in human primary hepatocellular carcinoma. Tumour Biol. 36, 4405-4415.

Huang X., Ma J., Xu J., Su Q. and Zhao J. (2015c). Simvastatin induces growth inhibition and apoptosis in HepG2 and Huh7 hepatocellular carcinoma cells via upregulation of notch1 expression. Mol. Med. Rep. 11, 2334-2340.

Huang R., Wang X., Zhang W., Zhangyuan G., Jin K., Yu W., Xie Y., Xu

X., Wang H. and Sun B. (2016). Down-regulation of lncrna dgcr5 correlates with poor prognosis in hepatocellular carcinoma. Cell Physiol. Biochem. 40, 707-715.

Hwang H.W., Ha S.Y., Bang H. and Park C.K. (2015). ATAD2 as a poor prognostic marker for hepatocellular carcinoma after curative resection. Cancer Res. Treat 47, 853-861.

Inokawa Y., Inaoka K., Sonohara F., Hayashi M., Kanda M. and Nomoto S. (2016). Molecular alterations in the carcinogenesis and progression of hepatocellular carcinoma: Tumor factors and background liver factors. Oncol. Lett. 12, 3662-3668.

Ji Y., Li B., Zhu Z., Guo X., He W., Fan Z. and Zhang W. (2015). Overexpression of WAVE3 promotes tumor invasiveness and confers an unfavorable prognosis in human hepatocellular carcinoma. Biomed. Pharmacother. 69, 409-415.

Jiang F., Chen L., Yang Y.C., Wang X.M., Wang R.Y., Li L., Wen W., Chang Y.X., Chen C.Y., Tang J., Liu G.M., Huang W.T., Xu L. and Wang H.Y. (2015). Cyp3a5 functions as a tumor suppressor in hepatocellular carcinoma by regulating mtorc2/akt signaling. Cancer Res. 75, 1470-1481.

Jiang G., Zhang L., Zhu Q., Bai D., Zhang C. and Wang X. (2016). Cd146 promotes metastasis and predicts poor prognosis of hepatocellular carcinoma. J. Exp. Clin. Cancer Res. 35, 38.

Jiao H.K., Xu Y., Li J., Wang W., Mei Z., Long X.D. and Chen G.Q. (2015). Prognostic significance of cbx4 expression and its beneficial effect for transarterial chemoembolization in hepatocellular carcinoma. Cell Death Dis. 6, e1689.

Jin M., Yang Z., Ye W., Xu H. and Hua X. (2014). Microrna-150 predicts a favorable prognosis in patients with epithelial ovarian cancer, and inhibits cell invasion and metastasis by suppressing transcriptional repressor zeb1. PLoS One 9, e103965.

Kanda M., Sugimoto H., Nomoto S., Oya H., Hibino S., Shimizu D., Takami H., Hashimoto R., Okamura Y., Yamada S., Fujii T., Nakayama G., Koike M., Fujiwara M. and Kodera Y. (2015). Bcell translocation gene 1 serves as a novel prognostic indicator of hepatocellular carcinoma. Int. J. Oncol. 46, 641-648.

Kang H.J., Kim I.H., Sung C.O., Shim J.H. and Yu E. (2015). Expression of carbonic anhydrase 9 is a novel prognostic marker in resectable hepatocellular carcinoma. Virchows Arch. 466, 403-413.

Kaustov L., Ouyang H., Amaya M., Lemak A., Nady N., Duan S., Wasney G.A., Li Z., Vedadi M., Schapira M., Min J. and Arrowsmith C.H. (2011). Recognition and specificity determinants of the human cbx chromodomains. J. Biol. Chem. 286, 521-529.

Klauke K., Radulovic V., Broekhuis M., Weersing E., Zwart E., Olthof S., Ritsema M., Bruggeman S., Wu X., Helin K., Bystrykh L. and de Haan G. (2013). Polycomb cbx family members mediate the balance between haematopoietic stem cell self-renewal and differentiation. Nat. Cell Biol. 15, 353-362.

Lee J.H., Friso S. and Choi S.W. (2014). Epigenetic mechanisms underlying the link between non-alcoholic fatty liver diseases and nutrition. Nutrients 6, 3303-3325.

Lee J., Kim Y., Friso S. and Choi S.W. (2017). Epigenetics in non-alcoholic fatty liver disease. Mol. Aspects Med. 54, 78-88.

Li Z., Chang X., Dai D., Deng P. and Sun Q. (2014). RASSF10 is an epigenetically silenced tumor suppressor in gastric cancer. Oncol. Rep. 31, 1661-1668.

Li T., Xie J., Shen C., Cheng D., Shi Y., Wu Z., Deng X., Chen H., Shen B., Peng C., Li H., Zhan Q. and Zhu Z. (2015a). Amplification of long noncoding rna zfas1 promotes metastasis in hepatocellular carcinoma. Cancer Res. 75, 3181-3191.

Li X.Y., Wen J.Y., Jia C.C., Wang T.T., Li X., Dong M., Lin Q.U., Chen Z.H., Ma X.K., Wei L.I., Lin Z.X., Ruan D.Y., Chen J., Wu D.H., Liu W., Tai Y., Xiong Z.Y., Wu X.Y. and Zhang Q.I. (2015b). MicroRNA-34a-5p enhances sensitivity to chemotherapy by targeting AXL in hepatocellular carcinoma MHCC-97l cells. Oncol. Lett 10, 2691-2698.

Li J., Huang H., Li Y., Li L., Hou W. and You Z. (2016a). Decreased expression of long non-coding rna gas5 promotes cell proliferation, migration and invasion, and indicates a poor prognosis in ovarian cancer. Oncol. Rep. 36, 3241-3250.

Li T., Xie J., Shen C., Cheng D., Shi Y., Wu Z., Deng X., Chen H., Shen B., Peng C., Li H., Zhan Q. and Zhu Z. (2016b). Upregulation of long noncoding rna zeb1-as1 promotes tumor metastasis and predicts poor prognosis in hepatocellular carcinoma. Oncogene 35, 1575-1584.

Li L., Lei Q., Zhang S., Kong L. and Qin B. (2017). Screening and identification of key biomarkers in hepatocellular carcinoma: Evidence from bioinformatic analysis. Oncol. Rep. 38, 2607-2618.

Lin N., Di C., Bortoff K., Fu J., Truszkowski P., Killela P., Duncan C., McLendon R., Bigner D., Gregory S. and Adamson D.C. (2012). Deletion or epigenetic silencing of AJAP1 on 1p36 in glioblastoma. Mol. Cancer Res. 10, 208-217.

Lin T.H., Shao Y.Y., Chan S.Y., Huang C.Y., Hsu C.H. and Cheng A.L. (2015). High serum transforming growth factor-beta1 levels predict outcome in hepatocellular carcinoma patients treated with sorafenib. Clin Cancer Res 21, 3678-3684.

Liu Y., Zhou R., Yuan X., Han N., Zhou S., Xu H., Guo M., Yu S., Zhang C., Yin T. and Wu K. (2015a). DACH1 is a novel predictive and prognostic biomarker in hepatocellular carcinoma as a negative regulator of wnt/beta-catenin signaling. Oncotarget 6, 8621-8634.

Liu Y.R., Tang R.X., Huang W.T., Ren F.H., He R.Q., Yang L.H., Luo D.Z., Dang Y.W. and Chen G. (2015b). Long noncoding rnas in hepatocellular carcinoma: Novel insights into their mechanism. World J. Hepatol. 7, 2781-2791.

Liu J., Cui X., Qu L., Hua L., Wu M., Shen Z., Lu C. and Ni R. (2016). Overexpression of DLX2 is associated with poor prognosis and sorafenib resistance in hepatocellular carcinoma. Exp. Mol. Pathol. 101, 58-65.

Marquardt J.U., Andersen J.B. and Thorgeirsson S.S. (2015). Functional and genetic deconstruction of the cellular origin in liver cancer. Nat. Rev. Cancer 15, 653-667.

Marra M., Sordelli I.M., Lombardi A., Lamberti M., Tarantino L., Giudice A., Stiuso P., Abbruzzese A., Sperlongano R., Accardo M., Agresti M., Caraglia M. and Sperlongano P. (2011). Molecular targets and oxidative stress biomarkers in hepatocellular carcinoma: An overview. J. Transl. Med. 9, 171.

Martin J. (2008). Tumor suppressor and hepatocellular carcinoma. World J. Gastroenterol. 14, 1720.

Murakami Y. and Kawada N. (2017). MicroRNAs in hepatic pathophysiology. Hepatol. Res. 47, 60-69.

Nik A.M., Reyahi A., Ponten F. and Carlsson P. (2013). Foxf2 in intestinal fibroblasts reduces numbers of Lgr5(+) stem cells and adenoma formation by inhibiting wnt signaling. Gastroenterology 144, 1001-1011.

Nishida N. and Kudo M. (2013). Recent advancements in comprehensive genetic analyses for human hepatocellular carcinoma. Oncology 84, (Suppl. 1), 93-97.

Nishida N. and Kudo M. (2016). Clinical significance of epigenetic alterations in human hepatocellular carcinoma and its association

with genetic mutations. Dig. Dis. 34, 708-713.

Oya H., Kanda M., Sugimoto H., Shimizu D., Takami H., Hibino S., Hashimoto R., Okamura Y., Yamada S., Fujii T., Nakayama G., Koike M., Nomoto S., Fujiwara M. and Kodera Y. (2015). Dihydropyrimidinase-like 3 is a putative hepatocellular carcinoma tumor suppressor. J. Gastroenterol. 50, 590-600.

Pan G. and Thomson J.A. (2007). Nanog and transcriptional networks in embryonic stem cell pluripotency. Cell Res. 17, 42-49.

Peng W. and Fan H. (2015). Long non-coding rna pandar correlates with poor prognosis and promotes tumorigenesis in hepatocellular carcinoma. Biomed. Pharmacother. 72, 113-118.

Poon D., Anderson B.O., Chen L.T., Tanaka K., Lau W.Y., Van Cutsem E., Singh H., Chow W.C., Ooi L.L., Chow P., Khin M.W., Koo W.H. and Asian Oncology S. (2009). Management of hepatocellular carcinoma in asia: Consensus statement from the asian oncology summit 2009. Lancet Oncol. 10, 1111-1118.

Richter A.M., Pfeifer G.P. and Dammann R.H. (2009). The RASSF proteins in cancer; from epigenetic silencing to functional characterization. Biochim. Biophys. Acta 1796, 114-128.

Salehi A.H., Roux P.P., Kubu C.J., Zeindler C., Bhakar A., Tannis L.L., Verdi J.M. and Barker P.A. (2000). NRAGE, a novel MAGE protein, interacts with the p75 neurotrophin receptor and facilitates nerve growth factor-dependent apoptosis. Neuron 27, 279-288.

Schmid C.A. and Muller A. (2013). FOXD3 is a novel, epigenetically regulated tumor suppressor in gastric carcinogenesis. Gastroenterol. 144, 22-25.

Shen S., Lin Y., Yuan X., Shen L., Chen J., Chen L., Qin L. and Shen B. (2016). Biomarker microRNAs for diagnosis, prognosis and treatment of hepatocellular carcinoma: A functional survey and comparison. Sci. Rep. 6, 38311.

Shi J.Y., Yang L.X., Wang Z.C., Wang L.Y., Zhou J., Wang X.Y., Shi G.M., Ding Z.B., Ke A.W., Dai Z., Qiu S.J., Tang Q.Q., Gao Q. and Fan J. (2015). CC chemokine receptor-like 1 functions as a tumour suppressor by impairing CCR7-related chemotaxis in hepatocellular carcinoma. J. Pathol. 235, 546-558.

Shi Z., Liu J., Yu X., Huang J., Shen S., Zhang Y., Han R., Ge N. and Yang Y. (2016). Loss of FOXF2 expression predicts poor prognosis in hepatocellular carcinoma patients. Ann. Surg. Oncol. 23, 211-217.

Shimizu D., Kanda M., Sugimoto H., Sueoka S., Takami H., Ezaka K., Tanaka Y., Hashimoto R., Okamura Y., Iwata N., Tanaka C., Yamada S., Fujii T., Nakayama G., Koike M., Nomoto S., Fujiwara M. and Kodera Y. (2016). Nrage promotes the malignant phenotype of hepatocellular carcinoma. Oncol. Lett. 11, 1847-1854.

Sia D., Villanueva A., Friedman S.L. and Llovet J.M. (2017). Liver cancer cell of origin, molecular class, and effects on patient prognosis. Gastroenterology 152, 745-761.

Siegel R.L., Miller K.D. and Jemal A. (2015). Cancer statistics, 2015. CA Cancer J. Clin. 65, 5-29.

Song W.H., Feng X.J., Gong S.J., Chen J.M., Wang S.M., Xing D.J., Zhu M.H., Zhang S.H. and Xu A.M. (2015). MicroRNA-622 acts as a tumor suppressor in hepatocellular carcinoma. Cancer Biol. Ther. 16, 1754-1763.

Song L.J., Liu Q., Meng X.R., Li Sh.L., Wang L.X., Fan Q.X. and Xuan X.Y. (2016). DLC-1 is an independent prognostic marker and potential therapeutic target in hepatocellular cancer. Diagn. Pathol. 11, 19.

Sukowati C.H., El-Khobar K.E., Ie S.I., Anfuso B., Muljono D.H. and Tiribelli C. (2016). Significance of hepatitis virus infection in the oncogenic initiation of hepatocellular carcinoma. World J. Gastroenterol. 22, 1497-1512.

Tang B., Tang F., Li B., Yuan S., Xu Q., Tomlinson S., Jin J., Hu W. and He S. (2015). High USP22 expression indicates poor prognosis in hepatocellular carcinoma. Oncotarget 6, 12654-12667.

Tian Q.H., Zhang M.F., Luo R.G., Fu J., He C., Hu G. and Zeng J.S. (2016). DHX33 expression is increased in hepatocellular carcinoma and indicates poor prognosis. Biochem. Biophys. Res. Commun. 473, 1163-1169.

Tradonsky A., Rubin T., Beck R., Ring B., Seitz R. and Mair S. (2012). A search for reliable molecular markers of prognosis in prostate cancer: A study of 240 cases. Am. J. Clin. Pathol. 137, 918-930.

Valenta T., Hausmann G. and Basler K. (2012). The many faces and functions of beta-catenin. EMBO J. 31, 2714-2736.

Wang R., Wang Z.X., Yang J.S., Pan X., De W. and Chen L.B. (2011). MicroRNA-451 functions as a tumor suppressor in human non-small cell lung cancer by targeting ras-related protein 14 (RAB14). Oncogene 30, 2644-2658.

Wang J.H., Wei W., Guo Z.X., Shi M. and Guo R.P. (2015a). Decreased cezanne expression is associated with the progression and poor prognosis in hepatocellular carcinoma. J. Transl. Med. 13, 41.

Wang X., Zhang J., Zhou L., Lu P., Zheng Z.G., Sun W., Wang J.L., Yang X.S., Li X.L., Xia N., Zhang N. and Dou K.F. (2015b). Significance of serum microRNA-21 in diagnosis of hepatocellular carcinoma (HCC): Clinical analyses of patients and an HCC rat model. Int. J. Clin. Exp. Pathol. 8, 1466-1478.

Wang Y., He D., Yang L., Wen B., Dai J., Zhang Q., Kang J., He W., Ding Q. and He D. (2015c). TRIM26 functions as a novel tumor suppressor of hepatocellular carcinoma and its downregulation contributes to worse prognosis. Biochem. Biophys. Res. Commun. 463, 458-465.

Wang F., Feng Y., Li P., Wang K., Feng L., Liu Y.F., Huang H., Guo Y.B., Mao Q.S. and Xue W.J. (2016a). RASSF10 is an epigenetically inactivated tumor suppressor and independent prognostic factor in hepatocellular carcinoma. Oncotarget 7, 4279-4297.

Wang Y., Jiang J., Li Q., Ma H., Xu Z. and Gao Y. (2016b). KAP1 is overexpressed in hepatocellular carcinoma and its clinical significance. Int. J. Clin. Oncol. 21, 927-933.

Wang Y., Chen C.L., Pan Q.Z., Wu Y.Y., Zhao J.J., Jiang S.S., Chao J., Zhang X.F., Zhang H.X., Zhou Z.Q., Tang Y., Huang X.Q., Zhang J.H. and Xia J.C. (2016c). Decreased TDP52 expression is associated with poor prognosis in primary hepatocellular carcinoma. Oncotarget 7, 6323-6334.

Wellner U., Schubert J., Burk U.C., Schmalhofer O., Zhu F., Sonntag A., Waldvogel B., Vannier C., Darling D., zur Hausen A., Brunton V.G., Morton J., Sansom O., Schuler J., Stemmler M.P., Herzberger C., Hopt U., Keck T., Brabletz S. and Brabletz T. (2009). The EMT-activator ZEB1 promotes tumorigenicity by repressing stemness-inhibiting microrna. Nat. Cell Biol. 11, 1487-1495.

Windmill K.F., McKinnon R.A., Zhu X., Gaedigk A., Grant D.M. and McManus M.E. (1997). The role of xenobiotic metabolizing enzymes in arylamine toxicity and carcinogenesis: Functional and localization studies. Mutat. Res. 376, 153-160.

Wollert T., Wunder C., Lippincott-Schwartz J. and Hurley J.H. (2009). Membrane scission by the ESCRT-III complex. Nature 458, 172-177.

Wu D.C., Zhang M.F., Su S.G., Fang H.Y., Wang X.H., He D., Xie Y.Y. and Liu X.H. (2016). HEY2, a target of miR-137, indicates poor outcomes and promotes cell proliferation and migration in

hepatocellular carcinoma. Oncotarget 7, 38052-38063.

Xiao S., Chang R.M., Yang M.Y., Lei X., Liu X., Gao W.B., Xiao J.L. and Yang L.Y. (2016). Actin-like 6a predicts poor prognosis of hepatocellular carcinoma and promotes metastasis and epithelial-mesenchymal transition. Hepatology 63, 1256-1271.

Xu P., Zhang X., Ni W., Fan H., Xu J., Chen Y., Zhu J., Gu X., Yang L., Ni R., Chen B. and Shi W. (2015). Upregulated HOXC8 expression is associated with poor prognosis and oxaliplatin resistance in hepatocellular carcinoma. Dig. Dis. Sci. 60, 3351-3363.

Xu J., Li J., Zheng T.H., Bai L. and Liu Z.J. (2016a). MicroRNAs in the occurrence and development of primary hepatocellular carcinoma. Adv. Clin. Exp. Med. 25, 971-975.

Xu Y., Rong J., Duan S., Chen C., Li Y., Peng B., Yi B., Zheng Z., Gao Y., Wang K., Yun M., Weng H., Zhang J. and Ye S. (2016b). High expression of GNA13 is associated with poor prognosis in hepatocellular carcinoma. Sci. Rep. 6, 35948.

Yang Q., Ou C., Liu M., Xiao W., Wen C. and Sun F. (2014). NRAGE promotes cell proliferation by stabilizing PCNA in a ubiquitin-proteasome pathway in esophageal carcinomas. Carcinogenesis 35, 1643-1651.

Yao H., Liu X., Chen S., Xia W. and Chen X. (2015). Decreased expression of serum miR-424 correlates with poor prognosis of patients with hepatocellular carcinoma. Int. J. Clin. Exp. Pathol. 8, 14830-14835.

Yap K.L., Li S., Munoz-Cabello A.M., Raguz S., Zeng L., Mujtaba S., Gil J., Walsh M.J. and Zhou M.M. (2010). Molecular interplay of the noncoding rna anril and methylated histone H3 lysine 27 by polycomb CBX7 in transcriptional silencing of INK4A. Mol. Cell 38, 662-674.

Yin G., Liu Z., Wang Y., Dou C., Li C., Yang W., Yao Y., Liu Q. and Tu K. (2016). BCORL1 is an independent prognostic marker and contributes to cell migration and invasion in human hepatocellular carcinoma. BMC Cancer 16, 103.

Yu F., Lu Z., Chen B., Dong P. and Zheng J. (2015). MicroRNA-150: A promising novel biomarker for hepatitis b virus-related hepatocellular carcinoma. Diagn. Pathol. 10, 129.

Zhang H. and Zhu J.K. (2014). Emerging roles of RNA processing factors in regulating long non-coding rnas. RNA Biol 11, 793-797.

Zhang X.Y., Varthi M., Sykes S.M., Phillips C., Warzecha C., Zhu W., Wyce A., Thorne A.W., Berger S.L. and McMahon S.B. (2008). The putative cancer stem cell marker USP22 is a subunit of the human SAGA complex required for activated transcription and cell-cycle progression. Mol. Cell 29, 102-111.

Zhang E.B., Kong R., Yin D.D., You L.H., Sun M., Han L., Xu T.P., Xia R., Yang J.S., De W. and Chen J. (2014). Long noncoding rna anril indicates a poor prognosis of gastric cancer and promotes tumor growth by epigenetically silencing of miR-99a/miR-449a. Oncotarget 5, 2276-2292.

Zhang M., Zhang C., Zhang L., Yang Q., Zhou S., Wen Q. and Wang J. (2015a). Nrf2 is a potential prognostic marker and promotes proliferation and invasion in human hepatocellular carcinoma. BMC Cancer 15, 531.

Zhang S., Shi W., Chen Y., Xu Z., Zhu J., Zhang T., Huang W., Ni R., Lu C. and Zhang X. (2015b). Overexpression of SYF2 correlates with enhanced cell growth and poor prognosis in human hepatocellular carcinoma. Mol. Cell. Biochem. 410, 1-9.

Zhang X.F., Chao J., Pan Q.Z., Pan K., Weng D.S., Wang Q.J., Zhao J.J., He J., Liu Q., Jiang S.S., Chen C.L., Zhang H.X. and Xia J.C. (2015c). Overexpression of wwp1 promotes tumorigenesis and predicts unfavorable prognosis in patients with hepatocellular carcinoma. Oncotarget 6, 40920-40933.

Zhang Y., Zhang J.X., Huang L.L., He L.J., Liao Y.J., Lai Y.R., Deng H.X., Tian X.P., Kung H.F., Xie D. and Zhu S.L. (2015d). Low expression of barx2 in human primary hepatocellular carcinoma correlates with metastasis and predicts poor prognosis. Hepatol. Res. 45, 228-237.

Zhang Z., Zhang Y., Sun X.X., Ma X. and Chen Z.N. (2015e). MicroRNA-146a inhibits cancer metastasis by downregulating VEGF through dual pathways in hepatocellular carcinoma. Mol. Cancer 14, 5.

Zhao K., Wang W., Rando O.J., Xue Y., Swiderek K., Kuo A. and Crabtree G.R. (1998). Rapid and phosphoinositol-dependent binding of the SWI/SNF-like baf complex to chromatin after T lymphocyte receptor signaling. Cell 95, 625-636.

Zhao Y., Li F., Zhang X., Liu A., Qi J., Cui H. and Zhao P. (2015). MicroRNA-194 acts as a prognostic marker and inhibits proliferation in hepatocellular carcinoma by targeting MAP4k4. Int. J. Clin. Exp. Pathol. 8, 12446-12454.

Zheng X., Song T., Dou C., Jia Y. and Liu Q. (2015). CtBP2 is an independent prognostic marker that promotes GLI1 induced epithelial-mesenchymal transition in hepatocellular carcinoma. Oncotarget 6, 3752-3769.

Zheng H., Jiang W.H., Tian T., Tan H.S., Chen Y., Qiao G.L., Han J., Huang S.Y., Yang Y., Li S., Wang Z.G., Gao R., Ren H., Xing H., Ni J.S., Wang L.H., Ma L.J. and Zhou W.P. (2017). CBX6 overexpression contributes to tumor progression and is predictive of a poor prognosis in hepatocellular carcinoma. Oncotarget 8, 18872-18884.

Zhou S.F. (2008). Drugs behave as substrates, inhibitors and inducers of human cytochrome p450 3a4. Curr. Drug Metab. 9, 310-322.

Zhuang L., Xu L., Wang P. and Meng Z. (2015). Serum miR-128-2 serves as a prognostic marker for patients with hepatocellular carcinoma. PLoS One 10, e0117274.

Accepted September 29, 2017