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REVIEW ARTICLE

CD147/EMMPRIN: an effective therapeutic target for hepatocellular carcinoma

Xu Tang[1,*], Na Guo[2,*], Lixian Xu[3], Xingchun Gou[2,3,*], and Man Mi[2]

[1]Department of Pathology, Sichuan College of Traditional Chinese Medicine, Mianyang, China, [2]Lab of Cell Biology & Translational Medicine, Xi'an Medical University, Xi'an, China, and [3]Department of Anesthesiology, Stomatological College, the Fourth Military Medical University, Xi'an, China

Abstract

Hepatocellular carcinoma (HCC) is characterized by high resistance to conventional systemic therapies, rapid progression, easy metastasis and frequent recurrence. There is therefore an urgent requirement to develop novel systemic agents which specifically target hepatoma-associated antigen in the tumors of HCC patients. CD147, a transmembrane glycoprotein, is highly expressed by HCC cells and is strongly associated with HCC progression and prognosis. CD147 in HCC cells modulates HCC growth, promotes invasion and metastasis by stimulating adjacent fibroblasts and HCC cells to produce elevated levels of several extracellular matrix metalloproteinases (MMPs) in the HCC microenvironment. It is also involved in HCC angiogenesis and multidrug resistance (MDR). Clinical progress has been made in HCC treatment using CD147-directed monoclonal antibodies. Here, we give an overview of the literature regarding the molecular features and expression of CD147 in human HCC tissues. We specifically focus on the role of CD147 in HCC invasion and metastasis, as well as in angiogenesis and multidrug resistance. In addition, advances in therapeutic strategies targeting HCC CD147 are summarized.

Keywords: CD147, hepatocellular carcinoma, therapeutic target, EMMPRIN, MMPs

Introduction

Hepatocellular carcinoma (HCC) is the fifth most common malignancy worldwide and the third leading cause of death from cancer due to its very poor prognosis (Parkin et al., 2005). More than 1 million cases of HCC occur globally each year. Statistically, HCC was diagnosed in 30% to 40% of all patients at early stages in which only 20% of all patients are amenable to curative therapies, such as resection, liver transplantation and radiofrequency ablation (Llovet et al., 2003; Seong, 2009). In well-selected patients 5-year survival rates of 60–70% have been achieved (Llovet et al., 2003). However, due to lack of effective treatment options, HCC at advanced stages usually carries a dismal prognosis because of liver dysfunction, and most importantly, a high rate of metastasis (Bruix et al., 2001; Bruix & Sherman, 2005). Although a great deal of progress has been made in terms of chemotherapy, which provides significant survival benefits for patients with HCC, it is associated with appreciable side effects, highlighting the need for therapeutic strategies that target tumor cells without compromising normal tissue function (Roberts & Gores, 2006; Wysocki, 2010). Thus, the development of novel systemic agents targeting tumor-associated antigens with low toxicity and few side effects is actively being pursued.

CD147 protein with different origins from human cells and tissues has been discovered by multiple independent laboratories. It has been designated as an extracellular matrix metalloproteinase inducer (EMMPRIN) (DeCastro et al., 1996), HAb18G (Chen et al., 1999) or M6 antigen (Kasinrerk et al., 1992). CD147 is highly expressed on the surface of various malignant tumor cells, including

*These authors contributed equally to this work.

Address for Correspondence: Xingchun Gou, Lab of Cell Biology & Translational Medicine, Xi'an Medical University, Xi'an, China. E-mail: gouxingchun@189.cn; Man Mi, Lab of Cell Biology & Translational Medicine, Xi'an Medical University, Xi'an, China. Tel: +86 29–86177472. Fax: +86 29–86177472. E-mail: mmwpmm@263.com.

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cancers of the liver (Tsai et al., 2006), prostate (Madigan et al., 2008), skin (van den Oord et al., 1997), bladder (Muraoka et al., 1993), lung and breast (Polette et al., 1997). It is also expressed in the endomembrane system (EMS) (Zhao et al., 2011). Elevated expression of CD147 on the cancer cell surface is positively related to cancer progression and prognosis. A variety of *in vitro* studies have suggested that CD147 mainly functions as a cellular adhesion molecule, and is involved in the cell-cell and cell-extracellular matrix interaction that induces the secretion of matrix metalloproteinases (MMPs, mainly including MMP-1, MMP-2 and MMP-9) in the tumor local environment and thus promotes tumor invasion and metastasis (Guo et al., 1997; Sun & Hemler, 2001; Xu et al., 2007c). In addition to being an MMP inducer, CD147 has also been reported to directly enhance tumor invasion and metastasis via MMP-independent mechanisms, such as modulating tumor cell growth and promoting tumor angiogenesis. It is also involved in the multidrug resistance phenotype. Targeted therapy directed at CD147 has been demonstrated to be a useful strategy for HCC treatment, in both laboratory and clinical applications. In this paper, we review the molecular features, expression and the putative roles of CD147 in human HCC, and summarize molecular targeting therapy directed against HCC-associated CD147 in HCC treatment.

The molecular features and expression of CD147 in HCC

CD147 has a broad expression pattern in various epithelial cells with some differences between species, e.g. rat, mouse, chicken and human, but is evolutionary conserved as a highly glycosylated member of the immunoglobulin superfamily (IgSF) of proteins that are particularly rich in complex N-Glycans (Biswas et al., 1995; Tang et al., 2004a). The CD147 antigen consists of two extracelluar Ig domains, a single transmembrane domain and a short cytoplasmic domain of 40 residues. The first Ig domain in the N-terminal is required for counter receptor activity (Sun & Hemler, 2001) and is involved in MMPs induction and oligomerization of MMPs (Yoshida et al., 2000) while the second Ig domain is required for association with caveolin-1 (Tang & Hemler, 2004). The extracellular region contains three Asn glycosylation sites which contribute to both a highly glycosylated form, HG-CD147 (~40–60 kDa), and a lowly glycosylated form, LG-CD147 (~32 kDa). Two major forms of CD147 (43–66 and 35 kDa) have been revealed in purified CD147 from human FHCC-98 hepatoma cells, both containing a single deglycosylated core protein with a molecular weight of ~27 kDa (Yu et al., 2006). Caveolin-1 leads to an increased proportion of HG-CD147 relative to LG-CD147, an increased production of MMP-11 and a higher invasive capability of murine hepatocarcinoma cell lines (Jia et al., 2006). The residues (Yoshida et al., 2000) AAGTVFTTVEDLGSKILLTCSLNDSATEV (Gabison et al., 2005) of the extracellular region of HCC-associated CD147 may play a critical role in CD147 functions with regard to MMPs secretion and tumor invasion (Ku et al., 2007). The transmembrane domain of HAb18G/CD147 contains a glutamic acid residue and a leucine zipper motif that is implicated in protein association with the plasma membrane and the dimerization of DNA-binding proteins (Seulberger et al., 1990; Fossum et al., 1991). Both C-terminal and N-terminal domains are required to mediate the effect of CD147 on the secretion and activation of MMPs and metastasis-related processes in human hepatoma cells (Jiang et al., 2004).

The crystal structure of CD147 on HCC cells comprises an N-terminal IgC2 domain and a C-terminal IgG domain, which are connected by a 5-residue flexible linker. This unique C2-I domain organization is distinct from those of other IgSF members. Four homophilic dimers exist in the crystal and adopt C2-C2 and C2-I dimerization rather than V-V dimerization, as commonly found in other IgSF members (Yu et al., 2008). Currently, four basigin alternative splicing (AS) transcript variants (*Homo sapiens* basigin transcript variants 1, 2, 3 and 4) encoding different isoforms (basigin isoforms 1, 2, 3 and 4) are found in the NCBI Entrez Gene database. Of these four variants, the basigin-2 transcript (accession number NM_198589) is the most predominant splice variant, encoding the well-known basigin/CD147/EMMPRIN (Belton et al., 2008).

Basigin-3 is a short isoform comprising only one Ig-like domain (Ig-I domain) in its extracellular portion (Schlegel et al., 2009); it interacts with the internalized basigin receptor-ligand complex (Belton et al., 2008). As one of the most important isoforms of the basigin family in HCC, it inhibits HCC cell proliferation, MMP induction and cell invasion *in vitro* and *in vivo*, probably via hetero-oligomerization with basigin-2 at the interface of the Ig-I domain (Liao et al., 2011).

The particular structural features of CD147 suggest that it is involved in protein-protein interactions. Although the interacting molecules are still not well known, studies have recently suggested that several proteins, including caveolin-1 (Tang & Hemler, 2004), integrins (Curtin et al., 2005), cyclophilins (Yurchenko et al., 2001) and others may interact with CD147, as its ligand or receptor candidates, to mediate a wide range of cellular functions. Annexin II (Zhao et al., 2010), integrin α3β1 (Curtin et al., 2005) and α6β1 (Dai et al. 2009) can interact with CD147, therefore promote invasion and migration of human HCC cells *in vitro*.

The expression of CD147 has been reported to be controlled by the cell survival PI3K/Akt/GSK3β signaling pathway, and is directly regulated by the transcription factor Slug via the signaling cascade TGF-β→PI3K/Akt→GSK3β→Snail→Slug→CD147 (Wu et al., 2011). *In vivo* and *in vitro* analysis has indicated that promoter demethylation increases CD147 expression by enhancing Sp1 binding affinity and is associated with poor prognosis in HCC patients (Kong et al., 2011).

Alongside the cell membrane, CD147 also localizes in the endomembrane system (EMS) and can mediate changes in the internal HCC cell architecture (Zhao et al., 2011).

A high incidence of expression of CD147 in HCC has been reported by several research teams. In a systematic investigation involving the use of tissue microarrays and monoclonal antibodies (mAb) MEM-M6/1 and HIM6, CD147 expression was detected in 112 out of 129 distinct tumor entities with an incidence of 83% in HCCs (Riethdorf et al., 2006). In a later study, HAb18G was used in an investigation of the expression and role of CD147 as a cancer-associated biomarker and the incidence of CD147 expression in liver cancer was reported to be 80% (Li et al., 2009). RNA-based CD147 expression was analyzed in HCC-related tissues in comparison with the corresponding normal tissues based on GEO database expression data, including normal liver, low- and high-grade dysplastic liver tissue, and cirrhotic liver as well as early and advanced hepatocellular carcinoma (Barrett & Edgar, 2006). RNA levels of CD147 in the liver pathologies were found to be significantly higher in comparison with normal liver tissue and correlated with pathogenicity. Based on clinical and pathological analysis, the expression of CD147 was characterized as a significantly unfavorable prognostic factor in HCC (Zhang et al., 2007), and also as a significant predictor of recurrence after liver transplantation in HCC patients (Zhang et al., 2006). Overexpression of CD147 has also been found in HCC cell lines including FHCC-98 and MHCC97-H cells with a higher invasive ability (Lou et al., 2004;). These findings suggest that CD147 can be used as a cancer-associated biomarker for diagnosis and prognostic assessment of HCC. CD147 is involved in tumor growth, invasion and metastasis in HCC, and may be used as a therapeutic target for HCC treatment.

CD147 promotes HCC progression

CD147 modulates HCC growth

CD147 expression was positively correlated with tumor size and pathological tumor-node-metastasis (pTNM) stages of HCC patients (Zhang et al., 2007). Silencing of CD147 by RNAi in mouse hepatocarcinoma Hepa1-6 cells significantly decreased cell growth ability, colony formation in soft agar and tumorigenicity in nude mice (Jia et al., 2008a). CD147-transfected hepatocytes had mesenchymal phenotypes that accelerate tumor formation and tumor metastasis *in vivo*, suggesting that CD147 promotes epithelial-mesenchymal transition and is involved in hepatocarcinogenesis and HCC progression (Wu et al., 2011).

In an orthotopic model of HCC in nude mice, tumor growth was significantly inhibited in groups treated by anti-CD147 mAb HAb18 with an inhibitory rate of 34.3% ($P < 0.001$) as compared with that in the negative control group (Xu et al., 2007a).

Previously we found that starvation upregulates CD147 expression in HCC cells (data not published) and that CD147 inhibits starvation-induced autophagic cell death in SMMC-7721 HCC cells with an involvement of Beclin 1 down-regulation (Gou et al., 2009). We have also demonstrated that CD147 was downregulated by berberine and probably involved in berberine-induced HCC cell apoptosis and autophagic death *in vitro* (Hou et al., 2011).

CD147 promotes HCC invasion and metastasis

Degradation and remodeling of extracellular matrix by MMPs represent the most critical step in tumor invasion and metastasis (Figure 1). Elevated expression of MMP-2 and MMP-9 has been detected in HCC cancer tissues and cancer cells in culture, accompanied with enhanced expression of CD147 and high metastatic ability (Zhang et al., 2006; Zhang et al., 2007). Most of the MMPs present in the tumor tissue microenvironment are expressed by the stromal cells rather than by the tumor cells themselves (Gabison et al., 2005). The induction of MMP production is mainly mediated by tumor-stromal cell interaction via CD147 in the tumor tissue microenvironment (Toole, 2003; Tang et al., 2004b).

The mitogen-activated protein kinase (MAPK) superfamily which is composed of three major sets of kinases is involved in MMP production and cell motility in HCC cells. Downregulation of CD147 expression by siRNA in HCC cell FHCC-98 (Xu et al., 2007a) or SMMC-7721 (Qian et al., 2008) could inhibit the production of MMP-2 and MMP-9, as well as cell adhesion and invasion potential. The inhibitory effect on MMPs was regulated by MAP kinases ERK (extracellular-receptor kinases) in FHCC-98 cells and by SAPK/JNKs (stress-activated protein kinases/c-Jun N-terminal kinases) in SMMC-7721 cells. The disparity may be related to different siRNA sequences and different cell lines. The enhancing effects of CD147 on the secretion of MMP-2 and MMP-9

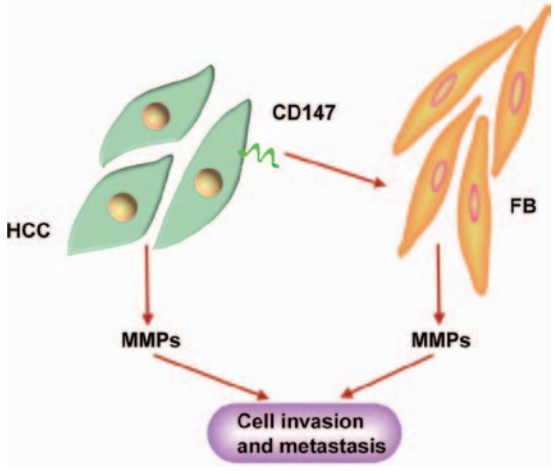

Figure 1. Schematic diagram of the mechanism of CD147 function in HCC invasion and metastasis. The induction of MMP production is mainly mediated by tumor-stromal cell interaction via CD147 in the tumor tissue microenvironment. FB, represents fibroblast; HCC, hepatocellular carcinoma cell; MMPs, extracellular matrix metalloproteinases.

were partially blocked by integrin α6β1 antibodies. Wortmannin, a specific PI3K inhibitor that reverses the effect of CD147 on the regulation of intracellular Ca^{2+} mobilization, significantly reduced the secretion of MMPs in HCC cells. These findings suggest that α6β1 interacts with CD147 thus induce MMPs secretion through the PI3K pathway and MMP secretion is Ca^{2+} dependent (Dai et al., 2009). Similar results were obtained by using α3β1 antibodies (Tang et al., 2008).

Full-length soluble CD147 is probably released from the surface of tumor cells via microvesicle shedding, which could diffuse away from the local tumor invasion site and stimulate stromal cells at distant sites (MacKenzie et al., 2001; Sidhu et al., 2004). Purified native CD147 from HCC cells had the bioactivity to stimulate human fibroblasts to produce elevated levels of MMP-2 and MMP-9 (Wysocki, 2010). In a coculture model, the invasion ability of HCC cells with cosilenced MMP-2 and MMP-9 genes cocultured with fibroblasts was significantly weaker than that of HCC cells cocultured with fibroblasts that had cosilenced MMP-2 and MMP-9 genes. The inhibitory effects on MMPs secretion showed a similar trend as demonstrated using gelatin zymography (Xu et al., 2007c). As compared with HCC cells with silenced CD147, HCC cells with silenced MMPs exhibited a significantly weaker suppression of MMP secretion and cell invasion in the coculture system (Xu et al., 2007c). These findings suggest that CD147 regulates HCC cell invasion via the modulation of MMP production, and that the modulation of fibroblasts plays a more important role. However, very few studies have focused on the mechanisms by which HCC cells stimulate adjacent fibroblasts to produce MMPs. In mouse fibroblast cells store-operated Ca^{2+} channels existed and were involved in MMPs secretion (Huang et al., 2006). Transfection of CD147 cDNA into fibroblast 3T3 cells enhanced the secretion of MMP-2 and MMP-9 via cGMP/NO-sensitive capacitative calcium entry (CCE) and accordingly attenuated the adhesion ability of fibroblasts (Huang et al., 2005).

Based on an investigation of tissue samples from 111 HCC patients, CD147 was found to be significantly associated with the presence of venous invasion, tumor size and pTNM tumor stages (Zhang et al., 2007). The invasion and metastasis potential of HCC cells were inhibited by downregulation of CD147 expression using siRNA or blocking the CD147 molecule with specific monoclonal antibody (mAb) (Xu et al., 2007c; Qian et al., 2008), whereas the opposite effects were observed after overexpression of CD147 in HCC cells (Tang et al., 2008), indicating that CD147 directly modulates HCC invasion and metastasis.

Tumor cell motility is a critical step in tumor invasion and metastasis. It is regulated by multiple signaling pathways and many factors especially membrane-associated proteins such as the actin cytoskeleton, vinculin, integrins and Annexin II. Downregulation of the CD147 gene in HCC SMMC-7721 cells had been shown to suppress their invasion and adhesive abilities due to alteration of the cytoskeleton including actin, microtubule and vimentin filaments, and inhibition of focal adhesion kinase (FAK) and vinculin expression, and the phosphorylation level of the SAPK/JNK Pathway (Qian et al., 2008). Analogous findings were presented in HCC FHCC-98 cell line via the ERK1/2 pathway (Xu et al., 2007a). CD147 co-localized and interacted with integrin α3β1 and α6β1 in HCC cells. It increased integrin α3β1 and α6β1 activity, enhanced the invasion and metastatic potential of HCC cells via integrin-mediated FAK-paxillin and FAK-PI3K-Ca^{2+} signal pathways leading to cytoskeletal rearrangement (Tang et al., 2008; Dai et al., 2009). Tumor cells can move as individual cells in two interconvertible modes: the mesenchymal mode and the amoeboid mode. CD147-transfected hepatocytes had mesenchymal phenotypes that accelerate tumor formation and tumor metastasis *in vivo* (Wu et al., 2011). Annexin II is a 36-kDa Ca^{2+}- and phospholipid-binding protein. Tyrosine phosphorylation of annexin II was involved in Rho/ROCK-mediated actin rearrangement and cell adhesion (Rescher et al., 2008). The interaction of the extracellular portion of CD147 with annexin II was involved in the interconversion between mesenchymal and amoeboid movement of HCC cells.

CD147 inhibited RhoA/ROCK signaling pathways and amoeboid movement in HCC cells by attenuating annexin II phosphorylation. It could also promote the membrane localization of WAVE2 and Rac1 activation in HCC cells via the integrin-FAK-PI3K/PIP3 signaling pathway, thus contributing to the formation of lamellipodia and mesenchymal movement (Figure 2) (Zhao et al., 2011).

CD147 induces HCC angiogenesis

Angiogenesis also plays a critical role in tumor growth, invasion and metastasis. Vascular endothelial growth factor (VEGF) and extracellular matrix remodeling by MMPs in the tumor microenvironment are crucial for angiogenesis (Figure 3). CD147 expression was positively correlated with the expression of VEGF, MMP-2, MMP-9 and microvessel density CD34 (MVD-CD34) in HCC tissues (Zhang et al., 2006; Zhang et al., 2007). CD147 depletion by RNAi in mouse hepatocarcinoma Hca-F cells which had highly metastatic potential in the lymph nodes resulted in significantly decreased expression of MMP-11, VEGF-A at both mRNA and protein levels, and attenuated the invasive, adhesive and metastatic ability of Hca-F cells to lymph nodes both *in vitro* and *in vivo* (Jia et al., 2007). Silencing of CD147 in mouse hepatocarcinoma Hepa1-6 cells also significantly impeded the expression of VEGF-A at both mRNA and protein levels (Jia et al., 2008a).

Recently, it was reported that some oncoviruses such as Kaposi's sarcoma-associated herpes virus (KSHV) could promote fibroblast and endothelial cell invasiveness following de novo infection through the upregulation of CD147 (Qin et al., 2010). Also, it was

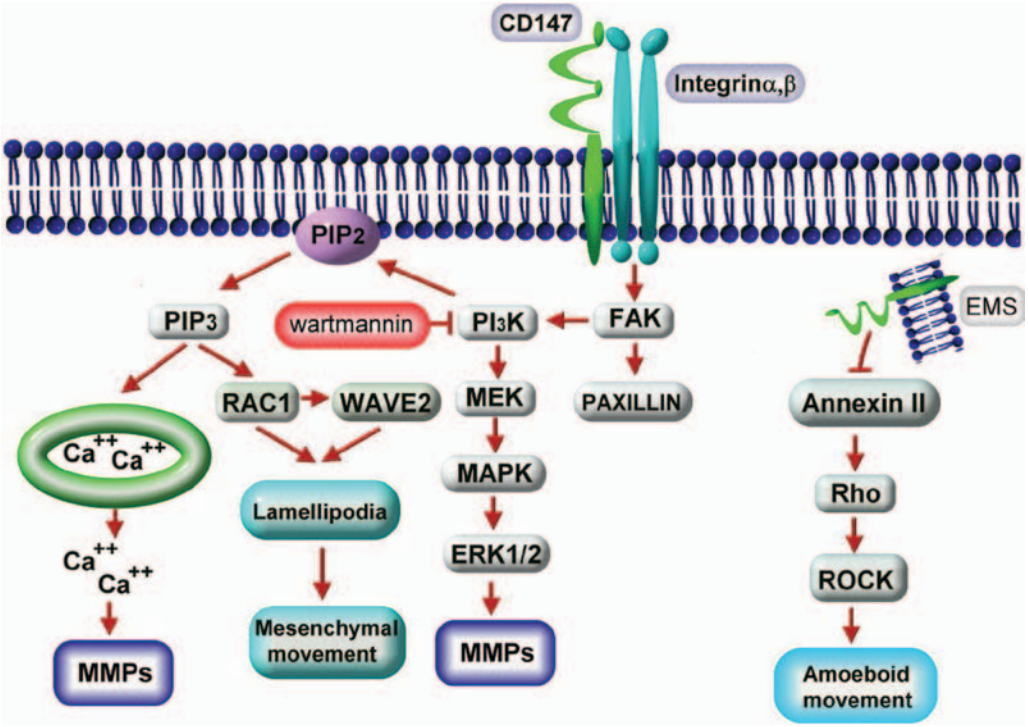

Figure 2. Schematic representation of the major molecular mechanisms of CD147 in the MMPs secretion and the interconversion between amoeboid and mesenchymal movement in the process of cell invasion and metastasis. CD147 plays an important role in HCC invasion and metastasis via several different pathways including integrin-mediated FAK-paxillin, FAK-PI3K-Ca^{2+}, RhoA/ROCK, WAVE2 and Rac1 signaling pathways. FAK, focal adhesion kinase; EMS, endomembrane system; ROCK, Rho-kinase.

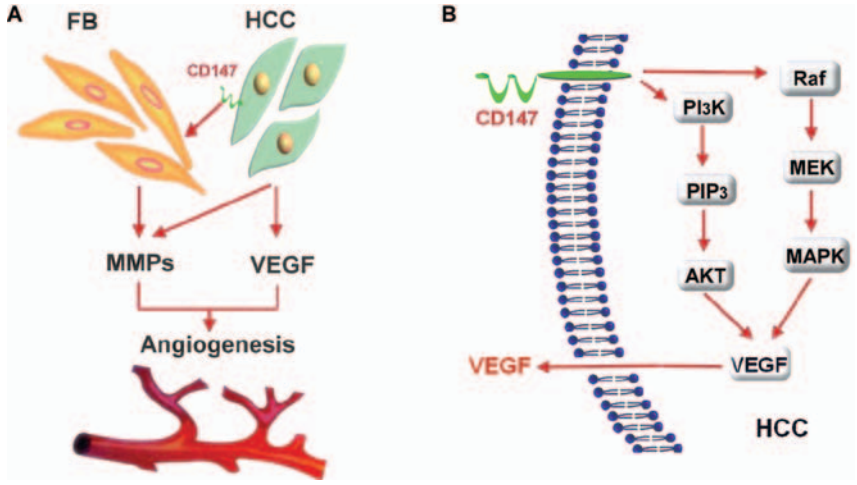

Figure 3. Schematic representation of the major molecular mechanisms of CD147 in the angiogenesis. (A) CD147 regulates angiogenesis via MMP and VEGF secretion. (B) HCC invasion was induced by CD147-dependent PI3K/Akt and MAPK activation of VEGF. FB, represents fibroblast; HCC, hepatocellular carcinoma cell; VEGF, vascular endothelial growth factor.

found that endothelial cell invasion for KSHV infected cells was induced by CD147-dependent PI3K/Akt and MAPK activation of VEGF (Dai et al., 2011), suggesting that CD147 may play an important role for angiogenesis in the virus-related tumor migration.

We found recently that CD147 expression was significantly upregulated in activated human umbilical venule endothelial cells (HUVEC's) (Chen et al., 2009). Inhibition of CD147 expression by specific siRNA led to significantly decreased angiogenesis *in vitro*. CD147 may regulate angiogenesis via several mechanisms including proliferation, survival, migration, MMPs secretion and PI3K/Akt activation. These findings suggest that CD147 is involved in angiogenesis in HCC progression.

CD147 involves HCC multidrug resistance

Multidrug resistance (MDR) is a main cause of treatment failure and mortality in HCC patients and has functional

linkage with tumor metastasis. P-glycoprotein (P-gp) is an adenosine triphosphate (ATP)-dependent transmembrane protein encoded by the MDR1 gene and is often attributed to MDR. It was found that downregulation of CD147 expression in mouse hepatocarcinoma Hepa1-6 cells by RNAi sensitized cells to curcumin treatment (Jia et al., 2008a). In drug resistant subline HepG2/Adr cells, CD147 expression was increased as compared with HepG2 cells (Jia et al., 2008b). The MDR cells produced more MMP-11 and MDR1, which promoted HepG2/Adr cell invasion and increased resistance to chemotherapeutic drugs. On the other hand, CD147 silencing in HepG2/Adr cells by RNAi led to the opposite effect, suggesting that CD147 was responsible for the altered MDR to chemotherapeutic drugs through the regulation of MDR1 expression in human hepatocarcinoma HepG2 cells. These results indicate that CD147 plays an important role in HCC MDR phenotype.

Targeted therapy against CD147

A series of *in vitro* studies have shown that the knockdown of CD147 expression by RNAi in HCC cells decreased cell invasion and reduced MMP-2 and MMP-9 secretion (Xu et al., 2007c; Qian et al., 2008). A CD147 mAb (mAb HAb18), specially established for mapping the epitope of residues (39) LTCSLNDSATEV (50) on the extracellular region of the HCC cell CD147 (Ku et al., 2007), and LICARTIN, a ^{131}I-labeled F(ab′)$_2$ fragment of mAb HAb18, were evaluated as agents for HCC treatment. It was found that both mAb HAb18 and LICARTIN suppressed MMP secretion by HCC cells and cocultured fibroblasts, and inhibited HCC cell invasion, and that LICARTIN significantly inhibited the *in vitro* growth of HCC cells (Xu et al., 2007c). In an orthotopic model of HCC in nude mice, HAb18 and LICARTIN treatment effectively reduced tumor growth and metastasis, as well as the expression of three major factors in the HCC microenvironment (MMPs, vascular endothelial growth factor and fibroblast surface protein) in the paracancer tissues (Xu et al., 2007c). In clinical phase I/II trials carried out to evaluate its safety and clinical efficacy in HCC patients, the injection of LICARTIN was found to be targeted and concentrated to tumor tissues and no life-threatening toxic effects were found (Chen et al., 2006). Of the 73 patients completing two cycles, six (8.22%) had a partial response, 14 (19.18%) a minor response and 43 (58.90%) stable disease. The survival rate of progression-free patients was significantly higher than that of patients with progressive disease after either one or two cycles. The main toxicity of LICARTIN in HCC is hematologic and liver enzyme damage. However, many patients in the study had abnormal hematologic or liver function indexes before treatment, and the indexes were actually improved in more than 30% of patients after the treatment. In another randomized controlled trial of LICARTIN for preventing hepatoma recurrence after liver transplantation, sixty post transplantation patients were included (Xu et al., 2007b). At 1-year follow-up, the recurrence rate had significantly decreased by 30.4% (P = 0.0174) and the survival rate had increased by 20.6% (P = 0.0289) in the treatment group, as compared with those in the control group. For the control group versus the treatment group, the hazard ratio for recurrence was 3.60 and that for death was 3.87. LICARTIN treatment also resulted in an earlier decreased AFP level and a longer time period over which there was a normal AFP level than was the case for the placebo (P = 0.0016). The results obtained in these clinical trials suggest that CD147 is a good therapeutic target and that LICARTIN is a safe and effective drug for HCC treatment. Furthermore, the combination of LICARTIN and chemoembolization appeared to extend survival in patients with unresectable HCC compared with historical controls, as well as being well tolerated by patients with Child-Pugh A and B (Wu et al., 2010). This combination may represent a promising treatment modality for patients with intermediate HCC. Now the clinical trial of LICARTIN combined with other therapies are going on (Villanueva & Llovet, 2011). (HAb18-huscFv)$_2$-Fc, a humanized HAb18 established by fabricating a humanized version of HAb18scFv, HAb18-huscFv, to the human IgG1Fc fragment, has been documented to be a more efficient antibody fragment with less immunogenicity and additional cytotoxicity function *in vitro* (Zhu et al., 2009). Transgenic mice generated by co-microinjection of two cassettes encoding the heavy and light chain genes of chHAb18 could highly express functional chHAb18 in their mammary glands, constituting an important step towards high-yield and scaled-up production of this antibody (Wei et al., 2011).

Conclusions and future directions

Although the precise molecular mechanisms and structural basis whereby CD147 modulates numerous phenomena in hepatocarcinogenesis and HCC progression are still not fully understood, it is clear that CD147 is an important tumor biomarker of HCC for diagnosis and prognosis. Targeting therapy directed against CD147 may provide an effective tool in HCC treatment, especially in controlling metastasis and recurrence. Characterization of molecules modulating CD147 expression, and the signal-transduction pathways regulated by CD147 in HCC invasion and metastasis, will be helpful in understanding the mechanisms of CD147 in HCC progress and in finding new target molecules for future drug development. Although mAbs against CD147, such as HAb18 and LICARTIN, have been proved to be safe and effective drugs for HCC treatment, the synergistic anti-cancer efficacy of anti-CD147 in combination with other chemodrugs, resection or radiation therapy needs to be further determined. Besides mAbs targeting CD147, it is necessary to establish systems for the screening and development of more novel therapeutic reagents that directly block CD147 function with relatively minor side effects, especially those derived from natural products such as berberine (Hou et al., 2011).

Declaration of interest

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