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Exploring the Role of Liver Cancer Stem Cells in Hepatocellular Carcinoma Metastasis

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Abstract: Metastasis refers to disseminating cancerous tumors from their primary site to distant locations inside the body. Cancer cells must go through a sequence of events called the "metastatic cascade" to develop metastases. Each stage necessitates a unique functional alteration. Cancer stem cells (CSCs) play a crucial role in tumor metastasis, but understanding their dynamic behavior and regulating mechanisms remains incomplete. This review explores the influence of liver CSCs on the biological processes that drive the spread and growth of cancer cells, as described by the "metastatic cascade" concept. Liver CSCs can spread to other organs by undergoing epithelial-mesenchymal transition (EMT). This alteration in the microenvironment facilitates cellular dissemination, immune surveillance evasion, dormancy induction, and subsequent reactivation. To effectively prevent and treat advanced hepatocellular carcinoma (HCC) metastases, it is crucial to understand the heterogeneity and features of liver CSCs involved in these processes.

Keywords: Liver cancer; Stem cells; Hepatocellular carcinoma; Signaling pathway; Tumor microenvironment; Metastasis

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1. Introduction

Hepatocellular carcinoma is a cancer of the liver. Hepatocellular carcinoma (HCC) ranks as the sixth most common form of cancer and is the third-highest contributor to cancer-related deaths [1]. For individuals with early-stage solitary tumors, surgical intervention is the primary therapy for their treatment [2]. A study indicates that patients who undergo resection have a 5-year survival rate of over 60% [3]. However, after five years, up to 70% of these patients experience the tumor's return [4]. Liver transplantation is often the optimal surgical treatment option for patients with early-stage tumors of HCC [5] due to the high prevalence of liver disease [6] and cirrhosis in these cases [7]. Globally, approximately 20%–40% of liver transplant recipients receive this form of medication [8]. According to Asrani *et al.*'s study, the occurrence of recurrent HCC after liver transplantation

is between 10% and 20% [9]. This rate may increase if the patient selection criteria are broadened [10]. After liver transplantation, tumor recurrence is frequently associated with the spread of cancer to other organs outside the liver [11,12], especially the lungs and bones [13]. Due to its high metastatic incidence, HCC's poor prognosis continues to be a significant concern [14]. Thus, there is an urgent need to gain greater knowledge of the mechanisms behind HCC metastasis [15].

A range of cellular processes known as the "metastatic cascade" is necessary to develop metastases [16,17]. These processes include being able to spread from where they started, encouraging vascular invasion [18], staying alive while moving through the bloodstream, going into and coming out of dormancy [19], making new cell environments in secondary sites, starting angiogenesis, and avoiding the immune system of the host [20]. The complicated process of tumor metastasis takes years to complete [21]. A tiny percentage of cells can spread to other locations [22]. For these cells to become adept at starting the spread of metastatic illness [22], they must go through a difficult metamorphosis and acquire a range of phenotypic features [23]. This particular subpopulation of cells, which is incurable, still causes the majority of tumor metastasis [24]. Research indicates that cancer cells with stem cell-like phenotypes are the ones that start the metastatic colonization of distant organs [25].

Cancer stem cells (CSCs) are a small group of stem-like tumor cells that can self-renew, differentiate, and initiate tumor formation [26]. In the last two decades, scientific research has significantly increased focus on identifying and understanding CSCs, particularly in the liver [27]. This has brought renewed optimism for the diagnosis and treatment of HCC [28]. Scientists have discovered multiple markers for liver CSCs [29], including OV6, CD24, ICAM-1, CD133, CD90, EpCAM, CD13, CD44, and K19 [30]. Multiple studies have substantiated the correlation between liver CSCs [31] and the spread of HCC despite the precise mechanisms remaining unidentified [32]. Gaining insight into the function of liver CSCs in advancing HCC cell metastasis can potentially enhance the detection and treatment of metastatic HCC [33,34]. Liver CSCs are involved in all stages of liver cancer is spread. This was done to learn more about liver CSCs' critical role in the metastasis of HCC and to develop new ways to treat the disease that work better.

2. The interplay of liver CSCs and EMT in HCC

Investigating and comprehending the molecular pathways involved in tumor metastasis consists of examinin g the epithelial-mesenchymal transition (EMT) [5]. Epithelial cells undergo a biological process called EMT [35], where they lose their characteristics as epithelial cells and acquire the characteristics of mesenchymal stem cells [36]. Erroneously activating this transformation process in malignancies can have devastating effects as the differentiated epithelial cells gain the ability to migrate and invade, thereby initiating the tumor metastasis process [37]. Researchers have discovered that EMT drives many biological processes, extending beyond the early stages of cancer cell invasion [36]. A growing body of research suggests that giving epithelial tumor cells experimental EMT activation results in the transfer of stemness characteristics, including CSCs [38].

EMT in HCC cells invariably accompanies the acquisition of stemness, according to an increasing body of evidence [39]. There is still some disagreement about the exact connection between EMT and HCC stem cells, but most scientists agree that β1-induced EMT helps tumor cells, including HCC cells, change into CSC phenotypes [40]. Different reports say that turning on Notch1 started the EMT process [41], which turned HCC cells into cells that behaved like CSCs, which were controlled by HIF-1α [41]. It is noteworthy that the process of inducing stemness promotes both stemness and the EMT. MHCC97-L cells' CSC properties by artificially

expressing the *Oct4* and *Nanog* genes, which are linked to stemness [43]. After that, they went through EMT, which made it easier for the tumor to move, invade, and spread, both in living things and in the lab setting [44]. Another study found that culturing CSC spheres in a specialized medium containing neural survival factor-1 enhanced their ability to undergo metastatic development [45], thereby promoting the process of EMT [46]. Nevertheless, specific experimental results question the correlation between stemness and EMT. For example, cells that are negative for vimentin and positive for CD133 (vimentin$^-$CD133$^+$) [36] demonstrate a higher level of aggressive metastasis compared to cells that are positive for both vimentin and CD133 (vimentin$^+$CD133$^+$). On the other hand, HCC cells that are positive for vimentin and negative for CD133 (vimentin$^+$CD133$^-$) show EMT traits but stem cells that are negative for vimentin and positive for CD133 (vimentin$^-$CD133$^+$) do not [47].

2.1. Liver CSC niche required for CSC maintenance and survival

The interaction between tumor cells and the tumor microenvironment (TME) leads to the development, invasion, and spread of many malignancies, such as HCC [48,49]. The structure of the adjacent extracellular matrix (ECM) plays a critical role in tumor advancement [50]. Various cytokines or growth factors, in addition to alterations in tissue oxygen levels, facilitate communication between cancer cells, endothelial cells, stromal fibroblasts, and immune cells [51]. The gathering of additional data reveals the presence of environmental factors causing inflammation, such as TGF-1, chemokine, IL-17 lipopolysaccharide, IL-6 [52], and HGF, in the tumor microenvironment of HCC, which also play a role in regulating stemness [53]. Either CSCs can sustain this stemness or non-CSC cells can acquire it. Normal liver stem cells have the potential to transform into metastatic liver CSCs in reaction to the inflammatory cytokine IL-6 [54].

Specific regions within the microenvironment that harbor CSCs as "CSC niches" [55]. Several malignancies have observed the importance of interactions between CSCs [56] and their surrounding environments, or niches, in facilitating cancer metastasis [57]. One type of extracellular protein complex called laminin-332 helps liver CSCs keep their ability to grow new cells and protect them from chemotherapy when they're not working [58]. Solid tumors, particularly HCC, commonly exhibit a low-oxygen environment [59]. This environment is important in the area where stem cells are located, as it increases HCC stemness by activating HIF-1α [60]. Hypoxia can cause metastases to spread and invade more quickly [61]. This is related to the fast change of tumor cells into liver CSCs, which can create their environment such as a scaffold [62] by releasing proteins interacting with macrophages [63]. Reports suggest that HCC cells in low oxygen conditions generate the protein MYDGF, which could potentially enhance the ability of liver CSCs to self-renew.

Table 1. Signaling pathways activated in hepatic stem cells [64]

Signaling pathway	Role in normal liver development (species)	Role in liver cancer development (source)
BMP	Liver specification from endoderm (mouse)	Differentiation of CD133$^+$ CSC, (PLC/PRL/5, Huh7, MHCC-97L)
FGF	Liver specification from endoderm (mouse)	Cell proliferation and angiogenesis, (HCC-1.2, HepG2, Hep3B)
OSM	Hepatocytic differentiation of hepatoblast (mouse)	Differentiation of EpCAM$^+$ CSC, (Huh1, Huh7, primary HCC)
Wnt	Liver specification from endoderm (mouse), inhibition of hepatic maturation (mouse), biliary differentiation of hepatoblast (mouse), Hepatocytic differentiation of hepatoblast (mouse)	Liver CSC self-renewal (Huh1, Huh7, SMMC7721), cell proliferation (Hep3B)

Table 1 (Continued)

Signaling pathway	Role in normal liver development (species)	Role in liver cancer development (source)
TGF-b	Biliary differentiation of hepatoblast (mouse)	Controversial (mouse, rat, Huh7, primary HCC)
JAG1/Notch	Biliary differentiation of hepatoblast (mouse)	Controversial (mouse, primary HCC)
IL-6/STAT3	Liver regeneration (mouse)	Liver CSC maintenance (mouse)
HGF/c-Met	Liver regeneration (mouse), hepatocyte, trans-differentiation into biliary epithelium (mouse)	Epithelial-mesenchymal transition (mouse)

2.2. Effect of liver CSCs-mediated ECM remodeling on metastasis of HCC

CSCs make lysyl oxidase (LOX), which creates an ECM that is dense and stiff. This stiff ECM is essential for developing and preserving HCC's CSC properties [65]. Changes in the stiffness of the hepatic CSC environment may affect how CSCs renew themselves and what they are like as HCC metastasis spreads [66]. The ECM experiences substantial alterations throughout the advancement of cancer [67] and plays a pivotal role in the metastatic process [68]. The mechanical properties of the ECM [69] surrounding tumor cells undergo significant alterations, particularly in terms of stiffness, as the cancer cells proliferate, infiltrate, and metastasize [70]. Metastatic HCC had tumor tissues that were stiffer than those of people who did not have metastatic HCC [71,72]. However, the correlation between CSC stemness and extracellular matrix stiffness in HCC is still a subject of debate [73]. In HCC cells, enhanced matrix rigidity promotes stem cell growth. CD133+ liver CSCs create a specialized matrix by altering the ECM [74], resulting in a soft spot. This soft spot matrix has the ability to increase stemness maintenance, treatment resistance, and metastatic HCC cell spread. Still, studies have shown that a stiffer matrix improves the features of CSCs [75] and lowers the number of times sorafenib-induced cell death happens. Using a stiffer matrix significantly improved the characteristics of liver cancer stem cells (LCSCs) compared to a softer matrix.

3. Relationship between liver CSCs and circulating tumor cells

Circulating tumor cells (CTCs) are cancerous cells that get out of primary tumors and into the bloodstream, where they start metastases that spread to other parts of the body [76]. Previous studies have demonstrated that CTCs possess the traits of CSCs [77]. However, there is a limited understanding of the characteristics of circulating CSCs in HCC [78]. The CSC marker CD44 revealed the presence of CTCs in 71.4% of patients diagnosed with HCC [45]. As a result, these patients had a significant number of CTCs that exhibited characteristics of CSCs [79]. Most current methods for capturing CTCs depend on the detection of epithelial cell adhesion molecule (EpCAM), which is also applicable to HCC [80]. People commonly use EpCAM as a biomarker to detect liver CSCs. The stem cell-like properties of HCC $EpCAM^+$ CTCs have been proven by studying the presence of stem cell-related markers and their important ability to form tumors [81]. Several studies have shown that monitoring and predicting the outcomes of patients with HCC relies on the quantity of circulating CSCs.

4. Quiescence/dormancy liver CSCs in HCC metastasis

Clinical manifestations of HCC metastases typically occur several years after the removal of the initial tumor

through resection or liver transplantation [6]. The metastasis originates from a specific group of cancer cells that have spread and can remain inactive until becoming active again [21]. Upon immunological examination, LCC cells exhibit characteristics similar to those of stem cells that are critical for their survival, ultimately leading to the development of metastatic expansion under favorable conditions [82]. CSCs can evade immune system recognition and attack through the release of diverse proteins or cytokines that regulate the immune response [83]. This enables them to maintain a condition of quiescence and dormancy. As a result, the immune system is unable to identify CSCs until they have spread and returned due to their transition into a dormant state.

Gaining insight into the molecular properties of dormant CSCs [84] and the mechanisms responsible for their reactivation is crucial for developing effective therapeutic approaches to prevent cancer relapse and metastasis [21]. Researchers have identified CD13 as a partially inactive marker for CSC in human liver cancer cell lines and clinical samples [85]. It protects cells from apoptosis caused by genotoxic chemo/radiation stress by stopping the cell cycle in the G0 phase and lowering the damage that reactive oxygen species (ROS) do to DNA [86]. A subsequent study demonstrated that liver CSCs expressing CD13 could maintain a state of inactivity and resistance to chemotherapy drugs by using tyrosine through aerobic metabolism [87]. A specific CSC niche containing a high concentration of laminin-332 encloses side population (SP) CSCs [74]. This niche plays a crucial role in keeping liver CSCs in a dormant condition.

5. The effect of liver CSCs on immune evasion

Immune evasion in HCC begins early and progresses gradually and consistently, peaking in middle-stage II tumors [88]. Cancer cells, once isolated from the immunosuppressive milieu of the primary tumor, become vulnerable to immune surveillance. In order to produce metastases, they must successfully avoid being destroyed by the immune system [83]. CSCs elude the immune system, a crucial factor in preserving their capacity to generate tumors [89]. This is accomplished by modifying the molecular expression of CSCs and reprogramming the immune response. Research has shown the essential roles of natural killer (NK) cells in the primary immune response to the development of cancer, specifically HCC [90]. Scientists have demonstrated that liver CSCs that express EpCAM are resistant to the harmful effects of NK cells by increasing the expression of the *CEACAM1* gene [91]. $CD133^+$ liver CSCs possess a high proficiency in evading immune surveillance [79]. They achieve this by establishing a connection with lymphatic endothelial cells and initiating IL-17A signaling [92]. Scientists have found that liver CSCs can evade the body's adaptive immune response by interacting with activated regulatory $CD4^+$, $CD25^+$, and $FoxP3^+$ T cells (Tregs) through a process known as paracrine signaling [93]. Additional investigation is necessary to understand the immune evasion mechanism of liver CSC.

Table 2. Presents the surface markers that impact the signaling pathways, characteristics, and ability to resist therapeutic medicines in LCSCs [85]

LCSs	Phenotype of LCSCs	Signaling involved in LCSCs	Resistance to clinical drug
EpCAM	Cell signaling, metabolism, differentiation, cell adhesion, metastasis, tumorigenesis, chemoresistance, organogenesis, regeneration, self-renewal of Hep3B, HepG2, Huh7, Huh1, and Dt1, Hepa1-6 cells	Activation of the Wnt signaling pathway	Sorafenib
CD47	Self-renewal, tumor-initiating, tumorigenicity, and chemoresistance (MHCC97L, PLC, and Huh7 cells)	Activation of the IL-6/STAT3 signaling pathway, and NF	Doxorubicin, Sorafenib

Table 2 (Continued)

LCSs	Phenotype of LCSCs	Signaling involved in LCSCs	Resistance to clinical drug
$CD13^{+}CD133^{+}$	Tumor initiation, chemoresistance, and anti-apoptosis (Huh7 and PLC cells)	Reduction of ROS-induced DNA damage and inhibition of apoptosis	Doxorubicin, Fluorouracil (5-FU)
$CD13^{+}CD90^{+}$	Tumor initiation, chemoresistance, and anti-apoptosis (Huh7 and PLC cells)	Reduction of ROS-induced DNA damage and inhibition of apoptosis	Doxorubicin, Fluorouracil (5-FU)
CD133	Tumorigenic, cell cycle progression, differentiation, chemoresistance, and self-renewal (Huh7, SMMC7721, PLC8024, PLC8024, HepG2, and HCCLM3 cells)	Activation of AKT/PKB	Doxorubicin, Fluorouracil (5-FU) and Sorafenib
CD24	Metastasis, differentiation, self-renewal and chemoresistance (MHCC97H, HCCLM3, PLC/PRF/5, Huh7, and Hep3B cells)	Autophagy activation, activation of AKT/mTOR signaling pathway, and Notch1 signaling pathway	Cisplatin, Sorafenib
CD90	Tumorigenesis, metastasis, self-renewal and chemoresistance (MHCC97L, PLC, HepG2, Hep3B, primary HCC, and JHH-6 cells)	Activation of the mTOR signaling pathway	Doxorubicin
CD13	Chemoresistance, tumorigenesis and self-renewal (Huh7, PLC, and HepG2 cells)	Activation of ERK1/2 signaling pathway	Sorafenib, Doxorubicin, and Fluorouracil (5-FU)

6. Metabolism reprogramming is crucial to maintain liver CSC stemness

Tumor development widely recognizes metabolic reprogramming as a significant characteristic [94]. This reprogramming provides additional energy and essential components for cellular growth and compensates for redox imbalances to ensure their eventual survival and spread to other parts of the body. CSCs change their metabolism and energy regulation to adapt to a hostile tumor microenvironment (TME) [95] and ensure their survival and maintenance of stem cell properties. Researchers have demonstrated that HCC cells can undergo retro-differentiation into CSCs [27]. Metabolic reprogramming follows this process, involving alterations in mitochondrial activity that lead to decreased membrane potential, reduced ATP generation [96], and increased lactate synthesis. Liver CSCs control the characteristics of stemness by controlling the function of the mitochondria involved in respiration [97].

For CSC stemness preservation, reprogramming amino acid metabolism is essential [98]. The research found that HCC cells that were not responding to chemotherapy had a CSC phenotype [99], which means they had a changed metabolism and were not metabolically active [100]. Unlike chemo-sensitive HCC cells, these cells demonstrated independence from glucose and reliance on glutamine metabolism. Furthermore, maintaining liver CSCs heavily relies on glutamine metabolism [101]. Specifically, targeting glutamine metabolism or the enzyme glutaminase 1 (GLS1) can decrease the characteristics of CSCs in HCC [39]. These investigations demonstrated that the viability and ability of liver CSCs to regenerate themselves relied on glutamine.

7. Liver CSCs promote angiogenesis

Solid tumors necessitate the formation of novel blood vessels to facilitate their growth and spread to other body parts [102]. There is strong evidence that CSCs are linked to the growth of new blood vessels (tumor

angiogenesis) in the area around the tumor [103]. The development and spread of cancer directly correlate with these connections. Research has demonstrated that CSCs from various types of cancer, including glioblastoma, ovarian cancer, lung cancer, and breast cancer [104], can transform into endothelial cells and subsequently create fully functional new blood vessels. Researchers found an unfavorable prognosis for HCC and elevated expression levels of liver CSC biomarkers associated with tumor angiogenesis [85]. Previous studies have shown that cancer stem-like sphere cells derived from HCC cells can transform into endothelial cells. This transformation occurs regardless of VEGF and NOTCH signaling but relies on Akt and IKK activation [105]. Liver CSCs that are positive for CD90 can affect endothelial cells and help make new blood vessels (angiogenesis) in a way that allows the cancer to spread (pro-metastatic) [79]. Researchers have demonstrated that CD133+ liver CSCs stimulate tumor angiogenesis through the signaling pathway involving neurotensin, interleukin-8, and CXCL1 [53]. In liver cancer, new research has shown that Oct4, a transcription factor linked to stem cells, helps control how CSCs change into cells that look like endothelial cells [106]. The results of this study indicate that liver CSCs primarily stimulate the formation of new blood vessels (angiogenesis) by releasing signaling molecules to neighboring cells or transforming them into endothelial cells. This process is believed to contribute to recurrence, spread to other organs (metastasis), and resistance to treatments that target angiogenesis.

8. Current approaches for dissecting HCC heterogeneity

Prior methodologies for investigating tumor heterogeneity included microscopic analysis of tumor tissues [107], utilization of cancer cell lines with diverse genetic and pathological profiles [108], immunohistochemistry staining, and bulk RNA sequencing to scrutinize distinct cell populations within the tumor. By combining cutting-edge technologies with clinically relevant translational models [109], it is important to effectively study how different types of cells in the tumor bulk interact and behave [110]. These technologies include scRNA-seq, spatial transcriptomic, pathway enrichment analysis, and whole genome sequencing [111]. This approach has the potential to replicate the actual pathogenesis of HCC accurately.

8.1. Organoids

Organoids are three-dimensional tissue models created in a laboratory using stem cells [112]. These models resemble the natural complexity of organs, including their biological, structural, and genetic characteristics [113]. Researchers have used organoids in recent decades to explore the intricate nature of tumors [114], assess the efficacy of drugs, and explore the mechanisms behind tumor formation [115]. This 3D model can be used to explore the cellular heterogeneity within tumors, leading to a comprehensive understanding of the interactions among different subpopulations in tumors [116]. The diverse population of LCSCs poses a significant challenge to achieving a successful therapeutic outcome and contributes to the development of medication resistance [85]. Multiple differentiation factors can be employed, such as dexamethasone, a Notch signaling inhibitor [117], and BMP without Rspo1, to create healthy liver organoids from liver stem cells [118]. These factors are used to guide the stem cells toward growing into organoids based on hepatocytes [119]. These organoids can be used to investigate the transfer of stemness to non-stem-like, highly specialized hepatocytes. This enables us to gain insights into the functions of stem cells and CSCs in the development of diverse tumors [120], as well as the mechanisms underlying drug resistance in populations of diverse cells [121]. Recent research demonstrates the significant

capacity of using organoids to investigate tumor heterogeneity [45], examining organoids derived from primary and metastatic colorectal cancer (CRC) through transcriptome analysis and histology [122]. This investigation showed the presence of both intra- and inter-tumoral heterogeneity in CRC [123]. These organoids made from HCC patient tissues with different histories will be a useful way to study the processes that cause tumor heterogeneity, especially the role of LCSCs in HCC.

8.2. Precision-cut liver slice

The Precision-Cut Liver Slice (PCLS) [124] is an *ex vivo* model created by slicing human liver tissues. PCLS stands out due to its ability to preserve and maintain the multicellular histoarchitecture, spatial structural relations [125], and genetic characteristics of the original cell populations and organs for a certain period in a laboratory setting [126]. This makes it an excellent model for studying diverse subpopulations of HCC [127]. Researchers have widely employed phenotypic cell-based profiling to investigate medication response and toxicity [128], elucidate fibrosis stages, and assess the effectiveness of anti-fibrotic medicines [129]. The PCLS comprising both HCC and nearby normal liver tissues is the best way to test how well and selectively anti-cancer drugs work on a group of cells with different histological features [130]. Intratumoral heterogeneity (ITH) can be studied on its effects on treatment outcomes by looking at primary circulating tumor cells [131] from other parts of the same tumor. Similarly, pre- and post-chemotherapy primary circulating tumor cells can serve as a valuable model for investigating spatial and temporal variations and inferring treatment effectiveness [132].

8.3. Liquid biopsy

Liquid biopsy is becoming a viable substitute for tumor tissue biopsy [133]. The technology enables the examination of CTCs [134], as well as nucleic acids such as circulating tumor RNA (ctRNA) and circulating tumor DNA (ctDNA) [135], together with other tumor biomarkers present in the bloodstream and other bodily fluids [136]. The main and/or secondary tumors release these materials, displaying their diversity [137]. Specifically, the existence of a diverse population of CTCs [138] in the bloodstream suggests that there is variation in both the physical characteristics and genetic makeup of the tumor [139]. Indeed, the analysis of the physical traits of CTCs can provide valuable information for selecting appropriate treatments [140]. Furthermore, the number of CTCs [141] and their observable traits can contribute to our understanding of the biology of metastasis and the reasons behind medication resistance [142]. Patients with HCC can use spatial examination of CTCs [143] from various blood vessels to predict the occurrence of metastases [144]. The result can provide important information about the differences within a tumor and can potentially anticipate the spread of cancer to other parts of the body by analyzing circulating tumor cells in their specific locations.

9. Conclusion

Metastasis is a complex tumor process that is cancer's most lethal characteristic. Scientists have found that a small group of treatment-resistant cancer cells called phenotypically CSCs, is responsible for the tumor spreading to other organs, even though primary cancer has been painless for years. Therefore, it is imperative to develop more effective treatments that can entirely eradicate these residual cells. Understanding the principles that underpin the entire metastatic process, however, is critical for the development of effective metastase therapeutics [145]. Metastasis is the process by which only a small number of cells inside a tumor can migrate

from their initial position and disseminate to different areas of the body. Researchers have discovered a small number of blood cells, known as CSCs, involved in this process. Various studies have demonstrated that liver CSCs have a substantial influence on various aspects of HCC metastasis. These factors encompass the dissemination of cancer cells through the circulatory system, the capacity to endure adverse circumstances, and the establishment of metastatic locations. Liver CSCs can change into metastasis-initiating cells (MICs) through various biological processes, such as EMT, interactions within the CSC niche, and changes to the ECM. Subsequently, they can infiltrate the circulatory system while retaining their stem cell characteristics. Liver CSCs can survive in the bloodstream or other challenging environments by entering a dormant state, remaining inactive, and undergoing metabolic reprogramming. Liver CSCs exert a substantial influence on angiogenesis, the biological mechanism responsible for the formation of metastatic lesions. This article presents a detailed summary of the role of liver CSCs in different stages of the metastatic process. Understanding the role of liver CSCs in regulating the dissemination of HCC lays the groundwork for developing innovative approaches and therapies to prevent or control metastatic HCC.

Author contribution

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Disclosure statement

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