Pathology of circulating tumor cells and the available capture tools (Review)

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Abstract. Circulating tumor cells (CTCs), are tumor cells that diffuse into the circulating blood and serve an important role in the progress of cancer. During the early stages of cancer, CTCs undergo an epithelial-mesenchymal transition and obtain a more invasive phenotype. Subsequently, the tumor cells enter the circulating blood with the aid of immune cells, and enter a dormant state upon reaching distal organs. As the tumor progresses, metastasis may occur under certain conditions. The capture technologies available for CTCs are based on antibody-based capture, or capture based on the physical properties of CTCs, as well as modern technologies that integrate both these methods. Emerging modern technologies have increased the accuracy and efficiency of tumor cell capture, and have thus improved our understanding of tumor cells, and the molecular mechanisms underlying their properties. CTCs serve an important role in disease progression, prediction of patient prognosis and individualized treatment.

Contents

1. Introduction

The metastasis of cancer is a complex procedure, ultimately resulting in patient death (1). During metastasis, malignant tumor cells detach from the primary tumor and enter the circulatory system during the early stages of cancer, and when these circulating tumor cells (CTCs) reach a distal organ, they enter a dormant state or form a metastatic tumor (2-4). Metastasis contributes to ~90% of all cancer-associated deaths (5,6). In recent years, with advances in capture and identification technologies, our understanding of CTCs and the associated mechanisms underlying their action has become more accurate. In 2017, the American Joint Committee on Cancer included the presence of CTCs in the M0(i+) classification stage in breast cancer (7). CTCs represent the metastatic capacity of cancer, and may allow for an earlier diagnosis compared with imaging and clinical evidence. Thus, in the present review the physiological processes, of CTCs including, engendering and spread to distant organs, and the analytical tools available, as well as CTC research methods are described.

2. Physiological processes of CTCs

Tumor cells gain invasive ability and enter the circulatory system. CTCs arise from the tumor cells of primary tumors, metastases, lymph nodes or disseminated tumor cell pools (5,8). Before tumor cells enter the circulating system, a series of phenotypical and genotypical changes occur, including cytoskeletal reorganization, protease secretion, changes to the expression profile of adhesive proteins and receptors, and these changes allow CTCs to acquire an invasive phenotype and detach from the primary tumor (Fig. 1A) (9,10). Epithelial-mesenchymal transition (EMT) serves a key role in the loss of cell polarity and decrease of cell-cell adhesion resulting in an increase in the invasiveness of tumor cells (11,12). Predisposing factors of EMT include transforming growth factor-β (TGF-β), interleukin-35 and interleukin-6 which are secreted by macrophages, activation of the WNT signaling pathway and platelet-derived growth factor, and exposure to nicotine, alcohol and ultraviolet light also exacerbate this process (12-17). Epidermal growth factor (EGF) and hepatocyte growth factor, which are secreted by monocytes and neutrophils, and the presence of inflammation are also strong predisposing factors for EMT (18). Additionally, Twist, SNAIL, Zeb and other genes have also been shown to serve a crucial regulatory role in EMT (19-21). As tumor cells undergo EMT, the number of intercellular junctions reduce

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notably, as well as the expression of epithelial markers, such as keratin, E-cadherin and epithelial cell adhesion molecule (Ep-CAM) (22). Conversely, expression of mesenchymal markers such as vimentin are increased, and subsequently the morphology of the cells are altered allowing for detachment from the primary tumor, and ultimately becoming invasive (14,23). Centrosome amplification and CTC clusters are also hypothesized to confer invasive and metastatic properties to tumor cells (24,25).

Once cells detach from the primary tumor, the tumor cells near the neovascular site enter blood vessels through the sparse vascular endothelium under increased pressure of tumor tissue growth which facilitates extrusion (26). Additionally, changes in the extracellular matrix increase the synthesis of key components such as collagen and fibronectin, and simultaneously promote the development of abnormal structures in the extracellular matrix, through increased activity of members of the cathepsin and matrix metalloproteinases families of proteins, which are secreted by neutrophils and mast cells (27,28). Macrophages and inflammatory monocytes enhance the migration of tumor cells by increasing the synthesis and cross-linking of collagen and fibronectin (29,30). Natural killer cells reduce the expression of fibronectin by secreting interferon-γ, thereby limiting the migration of tumor cells (31). These changes all contribute to the breakdown of the extracellular matrix and intravasation of tumor cells into blood vessels. Subsequently, tumor cells use chemotaxis to migrate to sites of high vascular permeability to intravasate (Fig. 1A), and macrophages serve an essential role in this process. Macrophages alter vascular structures and increase permeability by secreting a series of growth factors such as the vascular endothelial growth factor (VEGF), fibroblast growth factor, and placenta-derived growth factor, which facilitate migration of tumor cells through the vascular endothelium (32,33). Additionally, macrophages recruit tumor cells via the epidermal growth factor (EGF) paracrine loop to sites where vascular permeability is higher and thus more readily penetrable (34). This paracrine loop is regulated by the secretion of colony-stimulating factor (CSF-1) from tumor cells. CSF-1 regulates macrophages, increasing the production of EGF, and these macrophages are recruited to blood vessels, forming a chemical concentration gradient of EGF to guide the tumor cells towards the blood vessels (35,36). This macrophage-mediated chemotaxis can be antagonized by T cells (37).

CTCs survive in the bloodstream and extravasate from the blood vessels at specific locations. CTCs in the blood face three challenges to survive, blood flow shear, anoikis, and immune cell identification and killing (38). Blood flow shear force is a double-edged sword. Under certain conditions, blood flow shear can increase the invasive and metastatic capacity of CTCs by activating yes-associated protein 1, as well as the ability to adhere to and penetrate blood vessels (39). Conversely, CTCs can be physically destroyed by blood flow shear (40), and the longer CTCs are present in the blood circulation, the lower their proliferative capacity becomes, and eventually the cells enter dormancy (41). Tumor cells undergoing EMT exhibit an improved ability to resist the blood flow shear force (42). When the tumor cells detach from their original microenvironment, the original extracellular matrix is unable to provide the necessary cytokines and signals for growth and survival resulting in death. This programmed death of the tumor cells is called anoikis. In CTCs, activation of the Akt pathway triggers tropomyosin-related kinase B and inhibits caspase-related apoptosis to counter anoikis (43). Furthermore, the Akt signaling pathway is activated in tumor cells and resists apoptosis induced by tumor necrosis factor-related apoptosis-inducing ligand, and this is mediated by vascular cell adhesion factor-1 (VCAM-1) (44). Platelets serve a crucial role in the resistance of CTCs to immune cell-mediated killing (Fig. 1B). Platelets can adhere to CTCs, forming an armor to resist blood flow shear (45), and natural killer (NK) cells recognize and kill the CTCs (46,47). However, platelets induce EMT in CTCs via the NF-κB and TGF-β signaling pathways (48), and as mentioned earlier, CTCs which have undergone EMT are more resistant to blood flow shear (42). Platelets may also prevent CTCs from NK cell recognition by transferring major histocompatibility complex 1 molecules to CTCs and secreting TGF-β to decrease the expression of NK cell NK Group 2D (49,50). Furthermore, platelets act as a link between CTCs and $CD11^+$ macrophages to form cell clusters which help CTCs survive in the bloodstream (51). Additionally, CTCs selectively express Programmed death-ligand 1 (PD-L1), CD47 and Fas/Fasl which reduce the elimination of CTCs by immune cells (52-55).

Extravasation of CTCs from blood vessels is associated with several factors, such as the vascular endothelium linkage state, blood circulation state, capillary structure and adhesion of CTCs to the vessel wall (56-59). When CTCs pass through the capillaries, the flow rate decreases and CTCs adhere to the vascular endothelium with the assistance of platelets and neutrophils (60,61). Mononuclear macrophages are recruited to the vascular endothelium, and CTCs adhere through C-C Motif Chemokine Ligand 2, and subsequently secrete VEGF, increasing the permeability of vascular endothelium and thus assisting extravasation of CTCs (62,63).

Dormancy and activation of tumor cells. CTCs do not immediately contribute to metastasis, but remain dormant possibly for several years following extravasation and colonization of a distant site (Fig. 1B) (64). However, the processes underlying the ability of tumor cells to survive and enter a dormant state are complex. Myeloid cells, monocytes and macrophages, including neutrophils and the factors secreted by these cells accumulate at the metastatic site, producing an inhibitory environment for immune responses, thus assisting the tumor cells entry into dormancy prior to the arrival of tumor cells at the dissemination site (34,65,66). Additionally, tumor cells resist apoptosis through activation of the Akt signaling pathway and thus improving tumor viability (4,44,67). Tumor cells enter dormancy and are divided into three stages, angiogenesis dormancy, immune dormancy and tumor cell dormancy (64). Tumor growth requires a large number of new blood vessels to ensure a sufficient supply of nutrients (68,69). During the dormant phase, angiogenesis is not activated (64). Proliferating tumor cells are in equilibrium with tumor cells that die due to a lack of oxygen and other nutrients due to the angiogenesis dormancy (68,70). In terms of immune dormancy, it has been confirmed in animal models that the presence of lymphocytes, particularly $CD8^+$ T cells, can maintain tumor dormancy and reduce metastasis (71). The deletion of $CD8^+$ T cells results in a 100% tumor

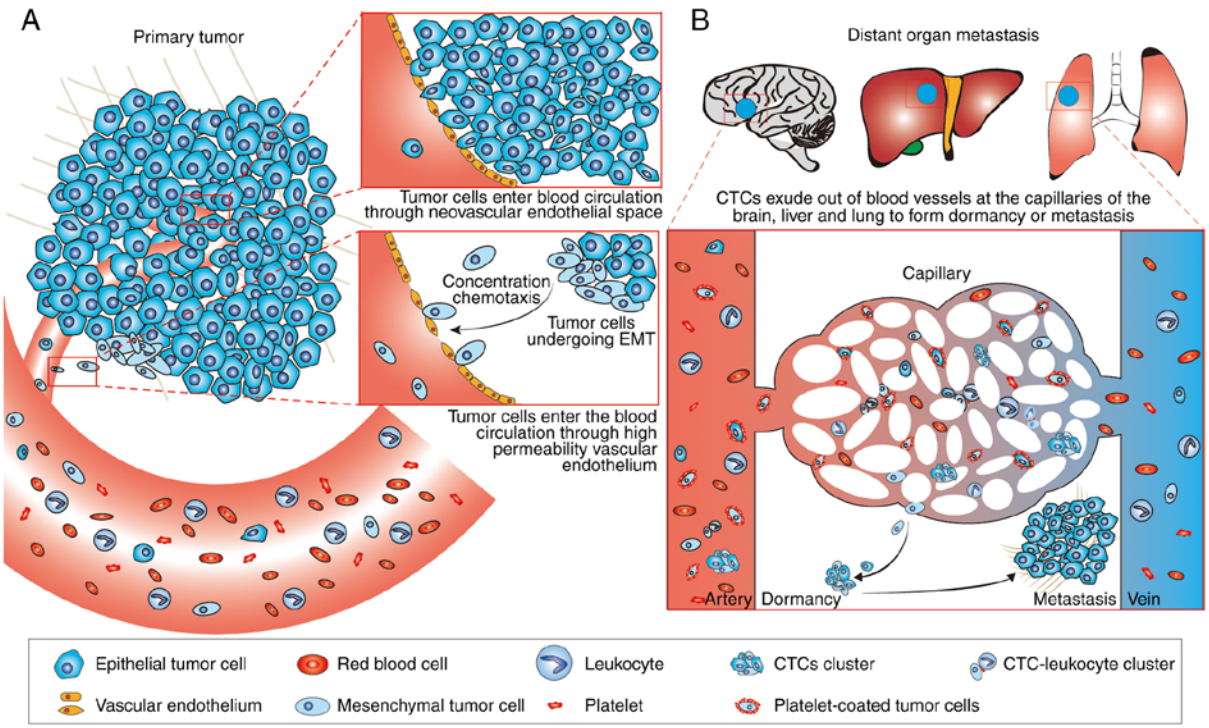

Figure 1. Biology of CTCs. (A) Tumor cells enter blood circulation through gaps between neovascular endothelial cells or vessels with greater permeability. CTCs counteract blood flow shear, anoikis and resist immune-mediated killing during circulation. (B) When CTCs flow through the capillaries of the distal organs, they eventually extravasate and enter dormancy. Under certain conditions, tumor cells exit this dormant state to form a metastasis. CTC, circulating tumor cell; EMT, epithelial-mesenchymal transition.

metastasis rate (72). In addition, EMT in tumor cells (26), and disruption of the balance between extracellular signal-regulated kinases and p38 mitogen-activated protein kinases (73), alters the expression of urokinase plasminogen activator receptor and integrin-β1 (74), activation of TGF-β2 signaling pathway (75) and expression changes in Myc gene (64), which all regulate the entry of tumor cells into a dormant state. When tumor cells enter dormancy, immune cells and conventional cytotoxic chemotherapy does not kill these cells; in conditions of post-operative stress, such as ischemia-reperfusion, sympathetic excitation, inflammation and hypercoagulability, these dormant cells may be activated, resulting in recurrence and metastasis in patients who underwent primary tumor resection (34,76). Exit from dormancy is also a complicated process. Tumor cells exit dormancy and gain proliferative capacity, when they undergo mesenchymal-epithelial transition (26), and this process involves tumor necrosis factor-α, interleukin-6 (77), integrin, activation of the Fak signaling pathway (78) and prostaglandin E2 (79). TGF-β receptor antagonists awaken tumor cells from dormancy by blocking the bone morphogenetic protein signaling pathway and thus improving tumor stem cell characteristics (80).

3. Isolation and detection of CTCs

In general, there are 1-10 CTCs per 1 billion blood cells, and these extremely rare CTCs are heterogeneous (81). This combined heterogeneity and rarity poses a great challenge for separating and detecting CTCs, as separation and detection technologies must exhibit specificity, repeatability and in particular sensitivity (2,82). At present, there are various CTC detection and separation techniques which can be crudely divided into two categories (Fig. 2A). The first method relies on specific markers on the surface of CTCs to separate them using antibody capture (83,84). The second method relies on the physical properties of CTCs, such as size, density, and electrophysiological characteristics (2,85). In addition, certain more recent techniques have combined the use of antibody capture with separation based on physical properties, such as preliminary isolation by size of CTCs, and further screening of CTCs using Ep-CAM antibody capture (86). Downstream DNA, RNA and protein levels can be analyzed to confirm that the captured cells are indeed CTCs (8).

The CellSearch system is the only technology approved by the US Food and Drug Administration (FDA) for capture of CTCs, and is a cell-specific marker antibody capture and separation technology. This system uses Ep-CAM antibody-coated immunomagnetic beads for capture of CTCs, similar to AdnaTest (87,88) and IsoFlux (89). In addition to coating of magnetic beads with an Ep-CAM antibody, it is also coated on a microfluidic chip microcolumn (90), herringbone lumen (91) or wavy herringbone lumen (92), and the inner wall of the microfluidic chip. After the blood sample has passed through, CTCs are captured and retained on these devices for additional verification. Furthermore, the Ep-CAM antibody is coated on a functionalized medical line (CellCollector) (93), and the antibody-coated line is left in a patients vein for 30 min to obtain CTCs from the body, which are verified following isolation. Flow cytometry can isolate CTCs from the blood samples using fluorescein-labeled Ep-CAM antibodies (94). However, this method has certain limitations as not all CTCs express Ep-CAM (83). As mentioned above, most of the tumor cells undergo EMT, which results in the downregulation of

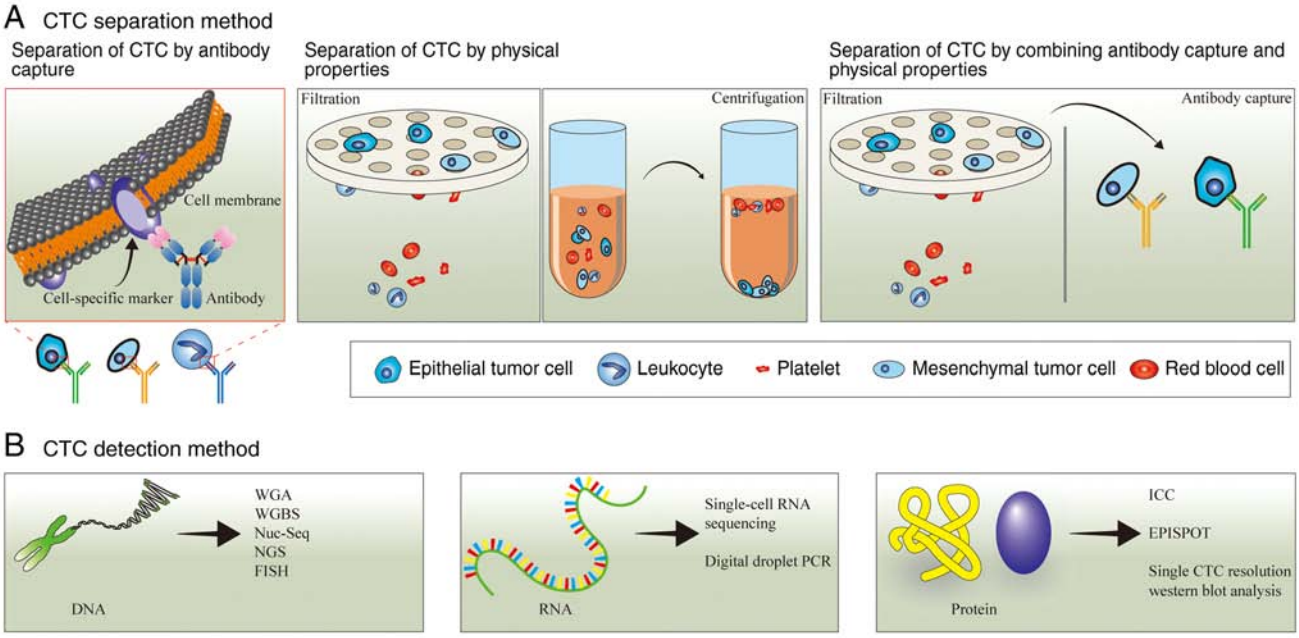

Figure 2. CTC separation and detection methods. (A) Separation methods of CTCs are divided into two types, antibody capture and physical capture. In antibody capture, the most commonly used antibody is Ep-CAM; Vimentin and leukocyte antibody CD45 are used for negative isolation. Physical capture primarily uses the difference in size and density of cells to separate CTC by filtration or centrifugation. One method combines both antibody capture with physical capture. (B) CTCs can be identified and analyzed based on DNA, RNA and protein expression, after obtaining CTCs. FISH, WGA are commonly used for DNA analysis, single-cell RNA sequencing is commonly used for RNA analysis, and ICC is commonly used for protein expression analysis. CTC, circulating tumor cell; Ep-CAM, epithelial cell adhesion molecule; FISH, fluorescence *in situ* hybridization; WGA, whole-genome amplification; ICC, immunocytochemistry; WGBS, whole-genome bisulfite sequencing; Nuc-Seq, whole-genome and exome single cell sequencing approach; NGS, next generation sequencing; EPISPOT, epithelial immunospot.

cell-surface epithelial markers such as Ep-CAM and keratin, and simultaneously, expression of mesenchymal markers, such as vimentin, are increased (12). This results in only 80% of solid tumors expressing Ep-CAM (95). Therefore, other types of antibodies have been adapted for capture of non-epithelial CTCs, such as the mesenchymal antibodies N-cadherin (96), Vimentin (97), and stem cell-specific antibodies CD44 (98). Heterogeneity of CTCs leads to inefficient capture of CTCs in different patients, and even in the same patient when only one antibody is used (82). Therefore, a negative screening technique was introduced. By enriching and discarding white blood cells expressing CD45, the remaining cells were identified to be CTCs, expressing different markers using tools such as Diagnostic Leuk Apheresis (99) and RosetteSep (100).

In addition to capturing CTCs using antibodies, it is possible to isolate CTCs based on their physical properties. Systems to isolate CTCs from blood cells based on size include ISET (101), CanPatrol (102), ScreenCell (103), Parsortix (104), CelSee (105) and microcavity array system (106). However, this method has certain drawbacks as not all CTCs are larger than white blood cells (90). Using the differences in density between CTCs and blood cell, CTCs can be obtained by centrifugation, for example by using Ficoll density gradient separation (107) and OncoQuick (108). Other techniques for capturing CTCs include separation based on electrophysiological features, such as dielectrophoretic field-flow fractionation (109) and dielectrophoretic array platform (110). In addition to the above separation methods, there are several novel detection methods, such as the multifunctional Branched Nanostraw-electroporation platform (111), which can capture CTCs, and also puncture the cell membrane without damaging the cell to assess the intracellular environment and transport drug molecules into the CTCs. Optofluidic flow cytometer can be used for consecutive CTC separation, 3D focusing and single-cell phenotypic counting (112). The Cytophone platform uses an *in vivo* photoacoustic flow cytometry platform with a high pulse rate laser and focused ultrasound transducers for label-free detection of CTCs (113). Furthermore, there are separation methods that combine antibody capture with physical properties, such as CTC-iCHIP (86) and 3D-printed microfluidic devices (114), which capture CTCs more efficiently by combining antibody beads with CTC size filtration.

CTC detection technology. After CTCs have been isolated using one of the above methods, confirmation of their identity needs to be verified, and a series of tests are performed to assess the cells and obtain information regarding the original tumor information from which the CTCs were derived (Fig. 2B). Immunocytochemistry is used to determine the expression of specific markers, thereby defining the identity of the CTCs. For example, in CellSearch, cells ≥4 μm, 4,6-diamino-2-phenylindole ($DAPI^+$), $keratin^+$ and $CD45^-$ are used as criteria for determining CTCs (115), and the expression of molecules on the surface of CTCs may also be analyzed by this technique, such as by staining for EMT-associated molecules (116). The classification of CTCs may also be confirmed using a 4-color fluorescence detection system (112). If cells are isolated and cultured, the proteins secreted by the cells can be detected using EPISPOT, to determine the identity of the CTCs (117). PCR can also be used for the identification of CTCs and for profiling the transcript of tumors, EMT, stem cell characteristics such as Ep-CAM, aldehyde dehydroge-

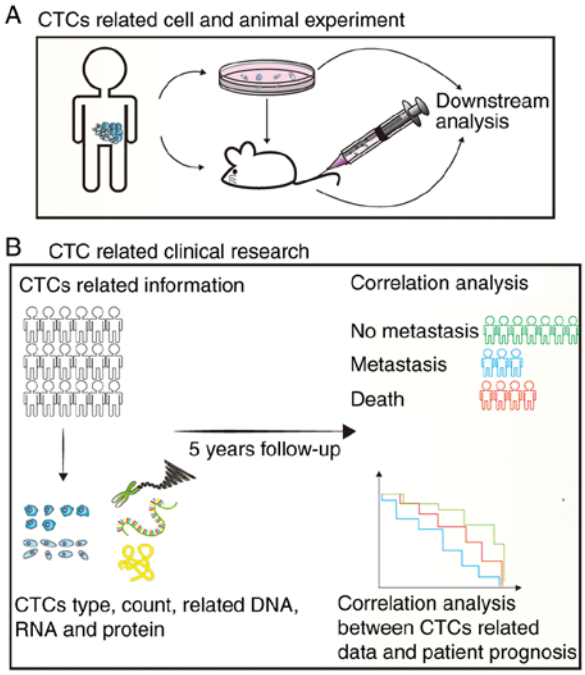

Figure 3. CTC associated research methods. (A) CTC associated cell and animal methods. A large number of CTCs are obtained from patients with high CTC counts, which are directly injected into nude mice for *in vivo* analysis, or cultured into cell lines for *in vivo* or *in vitro* studies. (B) Clinical trials of CTCs. The value of CTCs for prognosis was tested by combining CTC-associated detection and patient follow-up data.

nase 1 and Twist1 (118). With the emergence of whole-genome amplification (119), whole-genome bisulfite sequencing (120), whole-genome and exome single-cell sequencing (121), next-generation sequencing (117), third-generation sequencing (120) and low-pass whole genome and targeted next-generation sequencing (121), our understanding of CTC gene mutations has increased notably. In addition to the single-cell level, the association between the CTC phenotype and gene mutations can be visualized using fluorescence *in situ* hybridization (FISH) (122), without the need to separate individual cells by micromanipulation following CTC enrichment. For RNA, the emergence of technologies such as single-cell RNA sequencing (8) and digital droplet PCR (123), have allowed for a deeper understanding of the biological changes of CTCs at the transcriptional level. Furthermore, other novel technologies allow for assessment of the changes in protein expression levels from an individual CTC. For example, single CTC resolution western blot analysis can quantify 12 proteins simultaneously, this technology, when combined with flow cytometry, can quantify >40 proteins, significantly improving screening efficiency (124,125). Fluctuations in pH or detection of lactate concentrations have been employed in a microfluidic device to identify the metabolic status of CTCs (126).

4. CTC associated research methods

Research on CTCs can be divided into two categories, research at the cell and animal level, and clinical research focusing on the clinical prognosis of patients. Examination of phenotypes, characteristics, gene mutation sites and drug susceptibility of CTCs are primarily studied by culturing CTCs-associated cell lines and using cell biology and molecular biology techniques, and subsequently verified using *in vivo* experiments (127-129). In clinical trials, evaluation is performed by combining the number, type, and protein expression profiles of CTCs in patients' blood with clinical prognosis and personalized treatment (123,130). At present, the primary clinical value of CTCs includes the assessment of disease-free survival and overall survival prognosis (131), surveillance of minimal residual lesions, recurrence and metastasis (8,132), and to personalize treatment and determine the effects of the treatment (133,134).

Cell and animal research. Due to the rarity of CTCs, and the difficulty in isolating CTCs whilst simultaneously maintaining their survival outside of the body, certain studies are performed at the cellular or animal level compared with clinical studies of CTCs. In the case of patients with relatively high concentrations of CTCs, cell and animal research of their CTCs is possible (Fig. 3A). In a patient with colorectal cancer, the CTC concentration was >300/7.5 ml, thus blood was enriched and cultured, and a cell line based on their CTCs was successfully obtained (127). Studies on gene transcription, protein expression and secretion levels demonstrated that CTCs have primitive tumor cell and stem cell characteristics, mixed epithelial and mesenchymal phenotypes, and strong tumorigenic ability in animal experiments. In breast cancer, CTCs obtained from patients have mutations in the gene of PIK3CA, estrogen receptor 1, fibroblast growth factor receptor 2, and drug sensitivity to the mutations (128). Another study showed that CTCs with human epidermal growth factor receptor 2/epidermal growth factor receptor/Heparanase/Notch1^{+} have a tendency to metastasize to the brain in animal experiments (129). Similar studies have also been performed in prostate cancer (135), non-small cell lung cancer (136), and small cell lung cancer (137,138), and the tumorigenicity, drug resistance, and mutation sites of CTCs having been confirmed. Through these studies, a more comprehensive understanding of the relationship between CTCs and the metastatic tendency of different organs can be determined (139), and this individualized treatment (128) and the mechanisms underlying increased invasion and metastasis may be elucidated (140).

Clinical research. In clinical studies, research can be broadly divided into two categories, bivariate studies, and multivariate studies. In bivariate studies, the two variables are CTC-related data and patient clinical follow-up data (Fig. 3B). CTC-related data includes the number of CTCs, and protein, DNA and RNA expression profiles. Patient follow-up data primarily include disease-free survival, overall survival, chemotherapy medication and methods, and recurrence. CTCs are used as a predictor of clinical prognosis, chemoresistance, and for detection of tumor metastasis and recurrence. For example, in advanced pancreatic ductal adenocarcinoma, the number of CTCs is an important predictor of disease-free survival and overall survival prior to, and following first-line chemotherapy; the higher the number of CTCs, the shorter the disease-free survival and overall survival of patients (123). However, the mRNA expression levels of activated leukocyte cell adhesion molecule, POU Class 5 Homeobox 1B and smoothened are increased in CTCs, and this is associated with less favorable overall and disease-free survival (123). In metastatic

castration-resistant prostate cancer patients, patients with CTCs expressing nuclear localization androgen receptor splice variant 7 (ARV7) exhibit improved responses to taxanes and androgen receptor inhibitors; conversely androgen receptor inhibitors result in improved outcomes compared with taxanes in patients whose CTCs do not express nuclear localization of ARV7 (133). In breast cancer, CTCs expressing Notch signaling pathway markers are closely associated with brain metastasis (132). Related bivariate studies have also been performed on non-small cell lung cancer (131), colorectal cancer (141), liver cancer (130) and kidney cancer (116). However, studies on hepatic carcinoma have rarely yielded a negative result, counts of CTCs and types of EMT are not associated with clinical stage and predictive recurrence of hepatic carcinoma (130). In addition, CTCs and solid tumor DNA (142) or protein analysis (143) were used as two elements for correlation analysis to compare the variation of primary tumors and CTCs. However, in patients with lung cancer, there was no correlation between PD-L1 on CTCs and in tumor tissues (106).

In multivariate studies, in addition to the aforementioned CTC data and patient follow-up data, TNM staging (144) or other liquid biopsy markers, such as circulating tumor DNA (145) and extracellular vesicles (146), can be used together as predictors of prognosis to improve prediction accuracy. For example, in colorectal cancer, postoperative CTC counts are more valuable than preoperative counts, and TNM staging, and postoperative CA724 and CTCs counts are more accurate for predicting disease-free survival of patients, and there is a model to predict early recurrence and postoperative survival rate using postoperative CA724, CTCs counts, which can include or exclude TNM staging (141). As mentioned above, CTCs are closely associated with platelets and immune cells in the circulatory system; therefore, by combining CTC data, immune-inflammatory cell counts in circulating blood, coagulation status and other factors with patient follow-up data, the accuracy of predicting prognosis and assessing risk can be improved. In metastatic breast cancer, the ratio of CTCs to blood inflammatory cells, such as the ratio of CTCs to monocytes and lymphocytes, can be used as a predictor of prognosis (147). Also in metastatic breast cancer, by combining CTC data with thrombin-anti-thrombin III, fibrinogen, D-dimer and patient follow-up data, it has been shown that the hypercoagulable state contributes to tumor cell metastasis (148).

5. Conclusion

Tumor metastasis and recurrence is a major cause of cancer-associated death, and CTCs serve an important role in this process. During tumor cell detachment from solid tumors, cells enter the blood stream form distant metastases, and acquire an invasive phenotype, resisting the killing effect of the immune-inflammatory cells through EMT and various other mechanisms. During these complex and varied mechanisms, CTCs become heterogenous and unique. Therefore, the requirements for accurate separation and isolation of CTCs should be stringent. Traditional antibody capture technologies serve as the standard operating procedure, and these methods are relatively simple. Emerging capture technologies have greater sensitivities and specificities by combining antibody capture with CTC physics. However, these methods are time-consuming and expensive. Thus, developing a method that is simple, has a high capture rate and is accurate is required. Initially, CTCs were primarily used to predict the prognosis of patients based on their counts. CTC research is now focused on the molecular characteristics and the functions of the various molecular features of CTCs. With the use of CTC animal experiments, our understanding of biological behavior and mechanisms of CTCs has improved vastly and the development of tumor progression surveillance, prognosis assessment and individualized treatment has improved as a result.

In conclusion, CTCs possess value for prognosis assessment, metastasis surveillance and personalized treatment. They may be used to assess the presence of tumor metastases and recurrence, thus improving the prognosis of cancer patients, and reduce tumor-related mortality.

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Availability of data and materials

The datasets used and/or analyzed during the present study are available from the corresponding author on reasonable request.

Authors' contributions

CT designed the study and revised the article. YBC, FSX, JHT collected and accessed relevant literature. CCY, SLH, HWC organized the content and structure of the literature. KG, ZYW, MXH, MRL corrected the content of the article and produced the figures. All authors read and approved the final version of the manuscript.

Ethical approval and consent to participate

Not applicable.

Patient consent for publication

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Competing interest

The authors declare that they have no competing interests.

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