Circulating Tumour Cells and FOXP3 in Regulatory T-Cells as New Modalities in Cancer Diagnosis and Metastasis Location Prediction

*Ikhwan Rinaldi**

Division of Hematology and Medical Oncology, Department of Internal Medicine, Faculty of Medicine Universitas Indonesia - Cipto Mangunkusumo Hospital, Jakarta, Indonesia.

Corresponding Author:
Prof. Ikhwan Rinaldi, MD., PhD. Division of Hematology and Medical Oncology, Department of Internal Medicine, Faculty of Medicine Universitas Indonesia - Cipto Mangunkusumo Hospital. Jl. Diponegoro no. 71, Jakarta 10430, Indonesia. Email: ikhwanrinaldi@gmail.com.

Cancer is a complex group of diseases which arises from uncontrolled growth and spread of abnormal cells in the body. The pathophysiology of cancer involves a sequence of events at the cellular and molecular levels, often initiated by genetic mutations or alterations. These mutations can be acquired due to various factors like environmental exposures such as from carcinogens, lifestyle choices, or inherited genetic conditions. When a cell's DNA is damaged or mutated, it can disrupt the normal regulatory mechanisms that control cell division and apoptosis, leading to uncontrolled proliferation and cancer.

One of the important hallmarks of cancer is angiogenesis.[1,2] Angiogenesis plays a crucial role in cancer progression by providing the tumour cells with essential nutrients and oxygen, allowing it to grow and metastasize.

In cancer, angiogenesis becomes dysregulated. Tumour cells release signalling molecules, such as vascular endothelial growth factor (VEGF), that stimulate nearby blood vessels to sprout new branches toward the tumour, forming a network of abnormal blood vessels.[2,3] These vessels then supply the tumour with a constant flow of blood, facilitating its growth and also, enabling it to enter bloodstream and invade other locations (hematogenic spread).

Hematogenic spread, also known as hematogenous metastasis, is a key mechanism by which cancer cells travel through the bloodstream to establish secondary tumours in distant parts of the body.[4–6] Once inside the bloodstream, these circulating tumour cells (CTCs) travel throughout the body, carried along by the blood flow.[7,8] Although the majority of these cells perish in the bloodstream or fail to survive in distant tissues, some manage to evade the body's defence mechanisms and successfully establish secondary tumours in distant organs.

The shedding of CTC clusters from solid cancers is influenced by various biological factors that are not fully understood. Studies in breast cancer have shown that hypoxia contributes to the formation of CTC clusters, while normoxia is associated with single CTCs.[9–11]

CTCs have emerged as a significant area of study in cancer research, offering insights into cancer biology, prognosis, and treatment response.[12,13] They can be used as non-invasive biomarkers for diagnosis, monitoring disease progression and guiding treatment decisions.

CTCs is a non-invasive alternative to traditional tissue biopsies, which can be invasive and difficult to obtain.[7] This "liquid biopsy" is particularly beneficial for patients with tumours in difficult-to-reach locations or those undergoing frequent monitoring. Thus, the presence of CTCs in blood may be used to diagnose the type of cancers instead traditional biopsies.[14] Furthermore, CTCs can also be used to analyse

DNA mutations, epigenetic alterations, and metabolite profiling of cancer cells.[14–17]

Studies investigating the clinical utility of CTCs have also revealed their prognostic significance in different cancer types.[18–20] Elevated CTC counts have been associated with poor prognosis and increased metastatic potential, serving as an independent predictor of disease progression and survival outcomes. Higher CTC counts also often correlate with a more advanced stage of the disease and poorer prognosis.[21,22] Finally, a decrease in the number of CTCs following treatment often correlates with a positive response, indicating that the therapy is effectively targeting and reducing the tumour burden.[22–24] Conversely, an increase in CTC count during or after treatment might suggest resistance or disease progression.

Similarly, ctDNA, a type of DNA that is released into the bloodstream by cancer cells can also fulfill similar role of CTCs.[25,26] Nevertheless, detection and isolation of CTCs from blood samples represent a technical challenge due to their rarity amidst a vast number of blood cells.[12] It is very difficult to isolate CTCs but various innovative technologies have been developed to isolate and analyse CTCs such as immunomagnetic separation, microfluidics, and advanced imaging modalities.[12,27–31] These technologies continue to be improved with the aim of enhancing the sensitivity, specificity, and cost-effectiveness of CTCs detection.

Other than CTCs, Immune cells can also be used in cancer prognosis as immune cells have been shown to have influence on tumour development, progression, and response to treatment.[32,33] It is important to note that the immune system can either promote tumour growth or actively suppress it.

Certain immune cells, such as T cells, B cells, natural killer (NK) cells, and macrophages, infiltrate the tumour microenvironment.[34,35] Their presence and activity within the tumour hold prognostic value. In many cases, a higher density of certain immune cells within the tumour correlates with better clinical outcomes. For example, the presence of cytotoxic T cells (CD8+ T-cell) within the tumour has been associated with improved survival rates in several cancer types.[36–39]

Conversely, the presence of immune-suppressive cells like regulatory T cells (Tregs) or myeloid-derived suppressor cells (MDSCs) within the tumour microenvironment often indicates a poorer prognosis.[40,41] These cells can dampen the immune response against cancer cells, allowing tumours to evade destruction by the immune system and progress more aggressively.

Treg cells express FOXP3 biomarker. FOXP3, a transcription factor encoded by the FOXP3 gene, is considered the master regulator of Tregs.[42] It plays a fundamental role in the development, function, and stability of these specialized immune cells. Expression of FOXP3 is crucial for conferring suppressive function to Tregs, enabling them to regulate and modulate immune responses effectively.[42]

In cancer, FOXP3 has been shown to promote tumour growth and metastasis.[43–46] This is likely due to increased infiltration of FOXP3-expressing Tregs within the tumour microenvironment causes immunosuppression of immune cells. Thus, it can also be inferred that higher FOXP3 expression may relates to more aggressive cancer. All in all, many literatures have shown the role of immune cells as prognostic marker of many cancers, including in breast cancer.[47–52] Further researches in this area is needed to elucidate the complex roles of immune system in cancer.

Based on pathophysiology of cancer development which has been described, Acta Medica Journal publishes 2 articles. The first article is about CTCs assessment as a new diagnostic modality for colorectal cancer.[53] With new research such as this, it is hoped that in the future, analysis of cancer cells can be conducted through CTCs (liquid biopsy) instead of traditional biopsy to bring more convenience to patients, which may also indirectly increase the number of patients willing to take biopsy. Meanwhile, the second article is regarding the role of FOXP3 and T-cell in predicting metastasis location of triple negative breast cancer.[54] As described above, immune system has been shown to play a crucial role in cancer prognosis and management. We therefore can expect more

groundbreaking results from immuno-oncology field in the future.

Another article that is published by Acta Medica is about the role of unrefined extra virgin olive oil (EVOO) supplementation during capecitabine chemotherapy to reduce incidences of hand-foot syndrome (HFS).[55] Currently, HFS is one of the causes of treatment cessation in capecitabine chemotherapy and studies analysing the role of EVOO may help in reducing side effects of chemotherapy for patients.

In summary, the intricate interplay between genetic mutations, angiogenesis, hematogenic spread, CTCs, immune cells, and systemic cancer therapy defines the complex landscape of cancer progression and treatment. Understanding the role of immune cells, particularly Tregs marked by FOXP3, as prognostic markers in various cancers, alongside advancements in cancer diagnosis involving CTCs, holds promise in understanding cancer prognosis and improving cancer management. Moreover, ongoing research into alleviating chemotherapy-induced side effects, like HFS offer avenues for improving patient care and treatment outcomes in cancer management.

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