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REVIEW ARTICLE

Breast Cancer Metastasis: Role of Tumor Microenvironment and Resident Macropahges

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ABSTRACT

Metastatic breast cancer is a stage of breast cancer wherever the disease has spread to distant parts of the body. Onset of metastasis is one of the biggest obstacles to the successful treatment of cancer. The potential of a tumor cell to metastasise profoundly depends on its microenvironment, or "niche" interactions with local components. Macrophages provide tropic support to tumors. Resident macrophages contribute a set of common functions, including their capability to defend against microbial infections, to maintain normal cell turnover and tissue remodelling, and to help repair sites of injury. Macrophages are recruited into the tumor microenvironment where they differentiate to become Tumor-associated-macrophages (TAMs). TAMs are the most abundant subpopulation of tumor-stroma and actively drive cancer cell invasion and metastasis. Cancer metastasis is not solely regulated by the deregulation of metastasis promoting or suppressing genes in cancer cells. Recently the interaction between the stromal cells and cancer cells has been demonstrated to promote cancer metastasis. TAMs can advocate epithelial-mesenchymal transition of cancer cells. Loss of e-cadherin, a major phenomenon of epithelial to mesenchymal transition, reduces adhesiveness and releases cancer cells to distant (secondary) sites. A positive correlation between tumor progression and the expression of matrix metallo proteinases in tumor tissues has been demonstrated in numerous human and animal studies. The dynamic interactions of cancer-cells with TAMs actively promote invasion-metastasis cascade through intercellular-signaling-networks that need better elucidation.

Keywords, Macrophages, matrix metallo proteinases, extra cellular matrix, tumor microenvironment, cytokine

1. INTRODUCTION

With millions of new cases worldwide per annum, cancer is by far the most common disease in human. With the present rate of incidences of cancer globally, in coming years the burden of cancer is estimated to be around 23.6 million new cases each year[1,2] by 2030. Breast cancer rates highest in the world in the United States among all the countries. Despite such high prevalence, not much is known as to what triggers initiation and progression of breast cancer.

Metastatic breast cancer cells intermittently alter from the preceding primary breast cancer in properties like receptor status and usually develop resistance to many lines of previous treatment. These cancer cells acquired special properties to metastasise to distant sites, making them especially dangerous[3]. Onset of metastasis is one of the biggest barriers to the successful treatment of cancer. Breast cancer mainly metastasises to the bone, lungs, regional lymph nodes, liver and brain. Bone is the most common site of metastasis in breast cancer. Metastatic lesions are rarely feasible in clinical practice as they are often too widespread or too large to be removed by surgery and frequently exhibit increased resistance to chemotherapy.

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The potential of a tumor cell to metastasise depends on its microenvironment, or 'niche' communications with local factors promoting tumor-cell growth, survival, angiogenesis, invasion and metastasis[4]. In order to progress, tumors of epithelial origin need to acquire features which enable them to, (i) loosen cell-cell contact, rupture the basal membrane and detach from the tumor mass, (ii) invade neighbouring tissue, intravasate into blood or lymph vessels and (iii) extravasate from vessels in distinct organs to create secondary tumor(s). Epithelial tumor cells achieve invasiveness and migratory potential in the process of epithelial-mesenchymal transition (EMT), which is essential for successful metastatic spread[5].

The presence of inflammatory cells in tumors was first described in 1863 and this gave the concept that inflammatory microenvironment plays a key role to promote tumor development and progression. It has now been well understood that the majority of malignant tumors contain numerous macrophages as a main component of the host leukocytic infiltrate[6]. Macrophages are important tumor-infiltrating cells and play pivotal roles in tumor growth and metastasis. However, it is not yet clear how TAMs induce tumor invasion. Macrophages engage in immune responses to tumors in a polarised manner. These macrophages are referred to as tumor associated macrophages (TAMs) and mostly derived from peripheral blood monocytes recruited

into the tumor mass. In the past decade, TAMs have been broadly studied and proposed as a main contributor to tumor progression. Upon activation, the TAMs may secrete a many types of growth factors, cytokines, inflammatory mediators, and proteolytic enzymes those can assist tumor cell metastasis[7]. TAMs are a type of cell belonging to the macrophage lineage. While several types of inflammatory cell participate in cancer development, macrophages specifically play an important key role in breast cancer, where they emerge to be chunk of the pathogenesis of high-grade tumors. They are found in close proximity or within tumor masses. The function of TAMs is controversial as there is growing evidence for their involvement in both pro-tumor (e.g. promoting growth and metastasis by tumor angiogenesis) as well as anti-tumor (tumoricidal and tumorostatic) processes. TAMs interact with a wide range of growth factors, cytokines and chemokines in the tumor microenvironment which educate the TAMs and determine their specific phenotype. In many tumor types TAMs. infiltration level has been shown to be of important prognostic value[8]. TAMs produce factors that promote angiogenesis, tissue remodelling, and bedew the immune response to tumors[9]. Macrophages display a spectrum of phenotypic character, with the tumor microenvironment altering TAMs towards a 'nonclassical' activation state, known as the M2, or wound healing/regulatory state. In high-risk breast cancers these TAMs are present in a good number, making them a crucial therapeutic target to explore[10].

Macrophage secretes cytokines that are small protein molecule which allow cells of the immune system to interact with one another via cytokine receptors expressed at the cell surface. Macrophages secrete the different cytokines under different conditions, TNF-α, IL-1β, IF-α/β, IL-6, IL-10, IL-12, IL-18, MIP-1α, and MIP-1β, ENA-78[11]. They are very important for immune responses towards infection and inflammation. They trigger human granulocytes (neutrophils, eosinophils and basophils) which may lead to acute neutrophilic inflammation[12]. They also potentiate the synthesis and release of other pro-inflammatory cytokines such as IL-1, IL-6 and TNF-α from fibroblasts and macrophages[13].

Owing to the poor knowledge of the biology of metastasis the clinical intervention becomes extremely challenging making metastatic cancer largely untreatable. A better understanding of biology of metastasis becomes well warranted. Knowledge about the molecular pathways that regulate onset and progression of metastasis may lead to identification of not only new prognostic markers but also development of targeted therapeutic regimen against metastatic cancer. So far the majority of the research towards developing an insight about cancer metastasis has remained focused on malignant epithelial cells. However, the more recent studies demonstrate that the surrounding stromal cells can modify primary oncogenic events in tumor cells. Tumor stromal cells have been found to elicit instructive, permissive and inductive effects on the transformed epithelium. Prevailing new rational in the field of drug development is aimed for the development of target selective drugs. Development of such target specific drugs requires molecular understanding of normal and pathologically aberrant molecular pathways.

2. BREAST CANCER METASTASIS

The term "breas" refers to a malignant tumor that is developed by cells in the breast[14]. Metastasis is a multistep process that needs enhanced motility of the tumor cells inside the primary tumor and invasion into surrounding tissues and blood vessels. The tumor microenvironment is crucial for promoting these steps of motility and invasion in tumor cells, either by secretion of chemotactic factors or direct interactions with stromal cells. In rat and mouse mammary tumors, tumor-associated macrophages (TAMs) are necessary for advocating matrix remodelling, angiogenesis, and chemotactic motility of the tumor cells.[10] TAMs derive from circulating blood monocyte, and are directed into the tumor by chemoattractant cytokines called chemokines. Divers tumour cells also produce cytokines called colony-stimulating factors that prolong survival of TAMs. When appropriately activated, TAMs can kill tumor cells or elicit tissue destructive reactions centred on the vascular endothelium. However, TAMs also produce growth and angiogenic factors as well as protease enzymes which degrade the ECM. Thus, TAMs can promote angiogenesis, tumor-cell proliferation, and favour invasion and metastasis[15]. Direct evidence for the importance of protease production by TAMs, neutrophils, and mast cells during experimental carcinogenesis has recently been reported[16]. This dual potential of TAMs. is expressed in the 'macrophage balance' hypothesis[17].

Breast cancer can spread to any secondary site in the body but metastases appear preferentially in bone, brain, lung and liver[18]. Presumably these sites provide a microenvironment favourable for the growth and development of breast cancer cells[19]. Metastasis is the ultimate step in the multistage process of tumor progression in which tumor cells disperse from the primary tumor and colonize distant organs. Subsets of neoplastic cells are able to form distinct tumor colonies from the primary tumor. Not all cells within a tumor are able to metastasise. Metastatic cells are a subset within a heterogeneous tumorigenic population that are endowed with additional capabilities to those required for uncontrolled growth[20].

Development of metastasis requires tumor cells to complete a complex cascade of events. First, malignant cells invade adjacent tissues and penetrate into the lymphatic and/or circulatory systems[21]. Cells then detach from the primary tumor mass and disseminate to different organs of the body. Dissemination is a complex cell motility anomaly that requires the molecular coordination of the chemotaxis, contractilile activities, protrusion, and invasion of tumor cells to achieve directed cell migration. Invasive carcinoma cells acquire a migratory phenotype correlated with enhanced expression of several genes involved in cell motility[22]. This allows carcinoma cells to counter to hints from the microenvironment that trigger tumor invasion[5].

Therefore, molecules involved in cancer cell migration might be promising targets for anti-metastasis therapy.

During transport, tumor cells can travel alone or as emboli composed of tumor cells (homotypic) or tumor cells and host cells (heterotypic). At the secondary site, tumor cells or emboli either arrest due to physical limitations (e.g. too large to traverse a capillary lumen) or bind to specific molecules in particular organs or tissues[3]. Once there, tumor cells must then proliferate in the vasculature or extravasate into surrounding tissue. To form macroscopic metastasis, cells must then recruit a vascular supply. Less than 0.1 per cent of cells that enter the vasculature survive to form clinically detectable macroscopic metastasis.

2.1 Key Molecules Involved in Metastasis

The completion of all of the steps of metastasis does not always occur and the process of metastasis is highly inefficient, with colonization being the least efficient. In order for the tumor cell to become metastatic, several changes are needed to allow intra and extra-vasation as well as colonisation[23]. These cancer cells activate a cell differentiation circuit called EMT (Epithelial to Mesenchymal Transition) by inducing expression of several EMT-permissive transcription factors. The interplay between stromal cells and the epithelium is altered in cancer, with the stroma playing an important chunk in cancer progression by stimulation of proliferation, invasion and angiogenesis[24-26]. Fibroblasts are a major cellular part of the stroma and in so many types of tumours they are able to promote the proliferation and malignant growth of epithelial cells, due to change in expression of growth and survival factors, extracellular matrix proteins and proteases[24,27,28] . This allows the cancer cell to acquire characteristics of mesenchymal cells such as invasiveness and motility.

These changes in characteristics involve several proteins responsible for tissue invasion and spread, and some of the key players are Cell-cell adhesion molecules (CAMs), Integrins. Another strategy in successful colonisation is enhancing the expression of extracellular proteases (such as matrix metallo proteinases (MMPs)) while diminishing levels of protease inhibitors. Cells in the stroma close to cancer cells secrete active proteases, which facilitate invasion by degrading ingredient of the extracellular matrix[29].

2.2 Macrophage with Two Faces, M1/M2 Polarisation

Macrophages exist in most of the tissues and play very important roles in the maintenance of tissue homeostasis. In mature adults, peripheral blood monocytes differentiate into macrophages, Heterogeneity and plasticity are important features of macrophages[30]. Two specific states of polarised activation for macrophages have been proposed, (a) classically activated (M1) macrophage and (b) the alternatively activated (M2) macrophage subsets[31]. Differential cytokine production is a key feature of polarised macrophages[18] (Fig. 1).

3. TUMOR MICROENVIRONMENT

Resident macrophages contribute a set of common functions, including their capability to defend against microbial infections, to regulate normal cell turnover and tissue remodelling, and to help repair sites of injury[33]. The classically activated M1-macrophages composed of immune effect or cells with a subtle inflammatory phenotype. These are eminently aggressive against bacteria and produce large amounts of lymphokines[34]. The alternatively activated, anti-inflammatory M2-macrophages can be separated into at least three subgroups. These subtypes have many different functions such as regulation of immunity, maintenance of tolerance and tissue repair[34,35]. This difference (M1/M2) is reflected in their metabolism, where macrophages have the exclusive capability to metabolize one amino acid, arginine, to either a 'killer' molecule (nitric oxide) or a 'repair' molecule (ornithine).

Macrophages provide tropic support to tumors and the genetic experiments showed that if you abolish this support, malignancy is restrain strongly and contend that these cells or their particular signaling pathways are potent therapeutic targets. Macrophages have been shown to infiltrate a number of tumors. Poor prognosis is correlated with their number in some cancers including cancers of breast, cervix, bladder and brain (36). Macrophages also form an important component of the inflammatory infiltrate seen in both primary and secondary tumors[36]. where they also exhibit a distinct phenotype and are termed tumor-associated macrophages (TAM). Unlike tumor cells, the genomes of macrophages are substantial, suggesting that they may not as readily become drug resistant. Significant progress has been made in determining the molecular

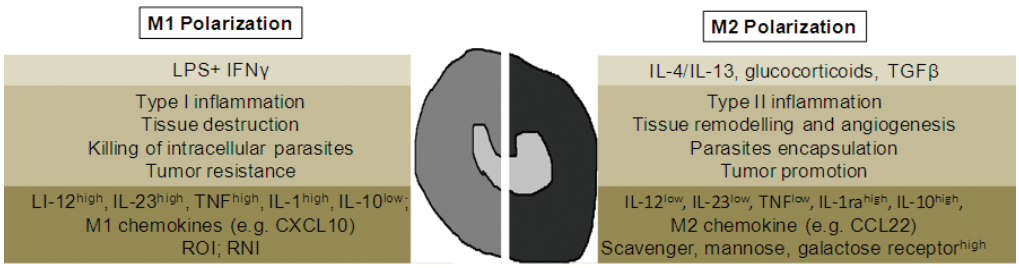

Figure 1. Macrophage polarised activation. Immune and tissue-derived signals view inducing classical (M1) and alternative (M2) macrophage polarised activation. The important functional effects on macrophage functions, molecular markers and effector molecules are schematically represented in both of the cases. RNI, reactive nitrogen intermediate, ROI, reactive oxygen intermediate[32].

basis for both macrophage phenotypes and their actions in promoting specific aspects of tumor behaviour[37].

Macrophages in the tumor microenvironment, referred to as TAMs, can influence tumor behaviour. Simply the presence of TAMs is correlated with poor prognosis in cancer survivors[38]. Clinical studies showed that macrophages promote tumorigenesis and also reported that over 80 per cent of studies show a correlation between macrophage density and poor patient prognosis (36). The transcriptome of TAMs analysed in mouse models of breast cancer has also furnished evidence that an enhancement in macrophage transcripts is predictive of poor prognosis and decreased survival in human breast cancer[39,40].

Tumor-Associated Macrophages (TAMs) represents the major inflammatory component of the stroma of many tumors, and are able to affect different aspects of the neoplastic tissue. TAMs enhance tumor cancer metastasis by several mechanisms that include promotion of angiogenesis, induction of tumor growth, and enhancement of tumor cell migration and invasion. TAMs infiltration has been shown to correlate with cancer metastasis. Consequently TAMs have been suggested as potential targets for novel therapeutic interventions. TAMs are short lived and do not proliferate *in situ*, therefore, their repertoire should be regularly replenished throughout the cancer progression[38,41-44]. Macrophage infiltration is thus a critical event for TAMs assisted tumor progression and targeting this critical event might retard tumor progression. In accordance with this, hindering macrophage infiltration into tumors of colony stimulating factor 1 knockout mice bearing the polyoma middle T oncoprotein considerably decreased the progression to malignancy[45].

Most studies have targeted upon events going on in the primary tumor, generally with metastasis as an end point. Metastasis requires not only the clemency of cells from the primary site but also their transit by the circulation or lymphatic to arrive at a distant site where the cells demand to extravasate, survive, and prosper. This process of metastasis is very ineffective. In humans, there are so many of circulating cells released by tumors every day, but only a few make metastases. In fact, the most likely destiny for these cells is death, with extravasations and establishment of micrometastases being major rate-limiting events[30]. Although it is already known that macrophages inhabitate metastatic lesions[30] only recently has their role in metastasis been appreciated. For tumors to progress and become malignant they must shape their microenvironment that is at least permissive if not promoting. It is possibly due to selection of oncogenic mutations which promote the secretion of molecules that change the cellular composition and behaviour of the microenvironment. Among these changes, one is the recruitment of bone marrow-derived cells, of which macrophages are particularly abundant. Macrophages in primary and secondary tumors brainstorm many properties that increase progression and metastasis[45].

Cancer metastasis is not particularly regulated by the deregulation of metastasis promoting or suppressing genes in cancer cells. Recently the interaction between the stromal cells and cancer cells has been demonstrated to promote cancer metastasis. Solid tumors are composed not only of malignant cells, but also extracellular matrix and many other non-malignant cell types, including endothelial cells, fibroblast and inflammatory cells like macrophages, mast cells, neutrophils, and lymphocytes[8].

Monocytes basically derive from $CD34^+$ myeloid progenitor cells in bone marrow, circulate in the bloodstream, and enter peripheral tissues where they mature into many types of resident macrophages, characterised by low protein synthesis rate, low oxygen consumption and cytokine production. Inflammation due to tissue damage or infection results in resident macrophage turn on, which enhances the production of cytokines, chemokines, and other inflammatory mediators, likewise monocyte recruitment[46]. (Fig. 2).

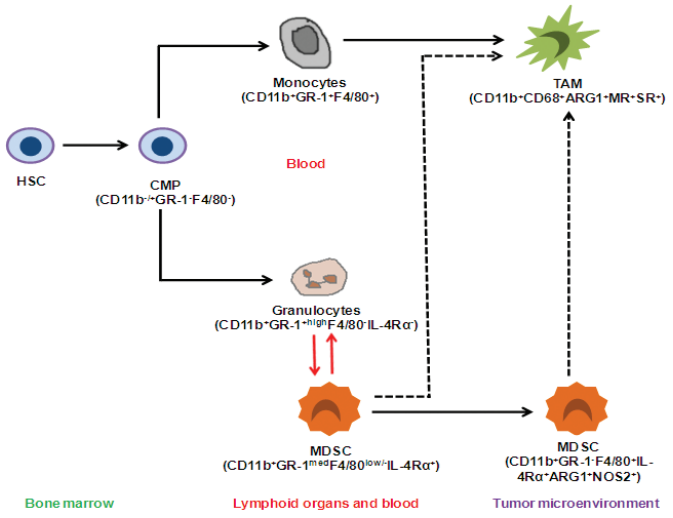

Figure 2. Current view of TAMs. and MDSC differentiation.

HSCs bring about common myeloid precursors (CMPs), which afterwards originate at least three subsets of cells circulating in tumor-bearing hosts. It can be found out by specific markers, monocytes (CD11b+Gr-1+F4/80+), granulocytes (CD11b+Gr-1highF4/80–IL-4Rα–), and MDSCs (CD11b+Gr-1medF4/80low/–IL-4Rα+). Circulating monocytes are recruited by tumors and differentiate into TAMs, gaining protumoral functions. In the process of tumor progression, MDSCs accumulating in blood and in lymphoid organs such as the spleen may also be recruited to the tumor microenvironment, where they become F4/80+ (Fig. 2). This latter pathway of MDSC-T*Am.* phenotype transition (dashed arrow) was recently proposed. Finally, it has been hypothesised that immature forms of granulocytes might differentiate into MDSCs or condition their function and/or further differentiation (red arrows), as proposed by some studies.

The macrophages within the tumor, referred to as tumor-associated macrophages (TAMs), are major component of the host leukocytic infiltrate in majority of malignant tumors[47]. These highly versatile cells respond to the presence of stimuli in different parts of tumors

with the release of a distinct repertoire of growth factors, inflammatory mediators, cytokines, chemokines, and enzymes. Many of these factors are key agents in cancer metastasis (Table 1). There are complex paracrine-signaling networks between TAMs and cancer cells to activate each other. TAMs-derived proteases, such as matrix metalloproteinase, urokinase-type plasminogen activator, and cathepsin B can promote cancer cells metastasis[48].

3.1 Tams may Promote Epithelial-mesenchymal Transition of Cancer Cells

Epithelial–mesenchymal transition (EMT) is a process that allows epithelial cells to separate from their neighbours and migrate to distal regions during embryonic development. The EMT confers migratory and invasive properties to epithelial cells and has also been suggested to play a fundamental role during invasion and metastasis of carcinoma cells[49]. Loss of E-cadherin, a major phenomenon of EMT, decreases adhesiveness and releases cancer cells from the primary locus into distant sites. Some of the Cellular Changes Associated with the Epithelial-Mesenchymal Transition are loss of cytokeratin (intermediate filament) expression, Epithelial adherens junction protein (E-cadherin), Epithelial cell polarity and acquisition of fibroblast-like shape, motility, invasiveness, mesenchymal gene expression program, mesenchymal adherens junction protein (N-cadherin), protease secretion (MMP-2, MMP-9), Vimentin (intermediate filament) expression, fibronectin secretion, PDGF receptor expression, αvβ6 integrin expression.

3.2 The Role of Tams in Tumor Progression: Tams are Recruited to Tumor and Re-educated by Microenvironment

Solid tumors recruit an abundance of immune cells into their microenvironment and macrophages can account for up to 50 per cent of the tumor mass[50]. Tumor and stromal cells secrete a number of diverse chemo attractants that recruit blood circulating monocytes[48]. Once in tumors, monocytes differentiate to macrophages primarily because of the presence of macrophage colony-stimulating factor (M-CSF), produced by tumor cells.

Conditioned by the tumor milieu, monocytes differentiate as tumor-educated macrophages and acquire properties of immune-suppressive and pro-tumoural effectors[47]. A number of monocyte chemoattractant derived from tumors have been shown to correlate with increased TAMs. numbers in many human tumors. Such monocyte chemoattractant include CSF-1, the CC chemokines, CCL2 (formally monocyte chemoattractant protein-1), CCL3, CCL4, CCL5 and CCL8, macrophage inflammatory protein-1 alpha (MIP-1α) and macrophage migration inhibition factor (MIF), vascular endothelial growth factor (VEGF)[50]. Chemokine recruited TAMs. also produce a rich milieu of cytokines/ chemokines. In addition, there is confirmation that chemokines direct endothelial progenitor cells in to tumor vasculature[8].

3.3 Paracrine Signaling Networks Between Cancer Cells and Tams

Interaction of TAMs and cancer cells enhances invasiveness of cancer cells and metastasis through several mechanisms, including (i) promotion of angiogenesis, (ii) induction of tumor growth, and (iii) enhancement of tumor cell migration and invasion (31). TAMs can promote solid tumor development by providing factors that enhance invasion of malignant cells into ectopic tissue. This activity centres on a paracrine interaction loop involving macrophage expressed epidermal growth factor (EGF) and epithelial cell-expressed colony-stimulating factor (CSF) 1. Increased expression of CSF-1 is a significant mechanism underlying macrophage recruitment into tissues after CSF-1 binding to its high affinity receptor (CSF-1R) expressed on resident macrophages and some macrophage precursors (Fig. 3). This interaction advocate macrophage proliferation, survival, and tissue recruitment all along development (e.g., branching morphogenesis in the mammary gland), homeostasis, and pathological tissue

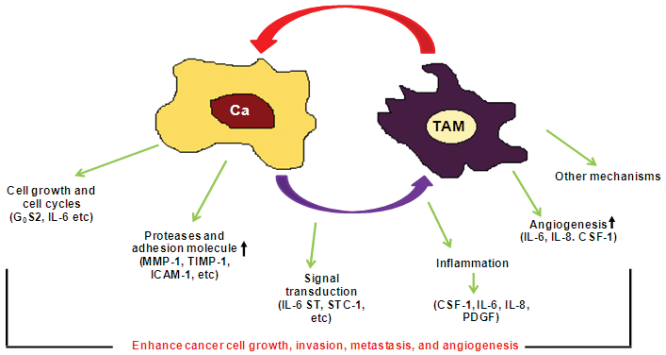

Figure 3. Paracrine signaling networks between TAMs and cancer Cells[51].

Table 1. Summary of the functions and factors secreted by activated and suppressed innate immune cells found within the tumor

Cell Type	Secreted Factors	Function
Classically Activated macrophages (M1)	IL-12, IL-15, TNF-α, IL-1β, IL-6, ROI	Immune activation, tumor destruction
Tumor Associated macrophages(TAM)	IL-10, TGF-β, arginase, PGE2, IDO	Immune suppression, tumor promotion
Activated NK cell	High IFN-γ, perforin, granzyme	Shape immune response towards Th1 (DC maturation) Cytotoxicity, tumor destruction
Tumor Associated NK cell Low	IFN-γ, IL-10?,VEGF	Shape immune response towards Th2 (Low IFN-γ, immature DC, immature NK cell), tumor promotion

remodelling processes such as those associated with acute tissue injury and during solid tumor development[47].

Although TAMs are important components of tumor stroma, and have an established role in promoting metastasis[52], the intercellular paracrine signals that mediate direct crosstalk between TAMs and tumor cells during metastasis need better elucidation. Furthermore the ensuing molecular events with in tumor cells that eventually impart them ability to invade surrounding tissue and disseminate from primary site during metastasis are poorly understood.

3.4 The Role of MMPS in Cancer Dissemination

MMPs appear to have an important role in cancer as well as in numerous other disease states[53]. A positive correlation between tumor progression and the expression of multiple MMP family members (MMP-1, MMP-2, MMP-7, MMP-9, MMP-11 and MT1-MMP) in tumor tissues has been demonstrated in numerous human and animal studies[54,55]. The ratio of activated to total MMP-2 levels has also been correlated with tumor aggressiveness[56]. The MMP expression has been implicated in tumor progression through enhancing angiogenesis, tumor invasion and metastasis (Table 2). MMPs act primarily to alter the extracellular environment to allow sustained cancer cell growth in an ectopic site, as opposed to having a specific role of allowing the cells to extravasate from the blood stream[57-59].

The 72 kDa MMP-2 (gelatinase A) is the most widely distributed of the MMPs and is expressed constitutively by most cells including endothelial and epithelial cells[60]. The 92 kDa MMP-9 (gelatinase B) is produced by inflammatory cells, including blood neutrophils and tissue macrophages, as well as by stimulated connective tissue cells[61-63]. Cleavage of matrix components by MMPs releases polypeptide fragments with new biological characteristics and signaling components embedded within the matrix. For example, ECM component laminin-5 is cleaved by MT1-MMP and MMP-2 promotes migration of cells[64-65]. Several soluble growth factors are secreted and stored in an inactive form bound to extracellular matrix molecules. In the time of enhanced proteolysis, these factors are then released to act on their target receptors[53].

In addition, urokinase-type plasminogen activator is a serine protease which is synthesised by TAMs in many human tumor types. The levels of urokinase-type plasminogen activator have been exhibited to associate with reduced relapse-free and overall survival in cancer[66]. TAMs can also release cysteine-type lysosomal proteases. Generally, lysosomal cysteine proteases are considered to execute nonspecific bulk proteolysis within the lysosomes.

Table 2. Some matrix metalloproteinase and their extracellular matrix substrates[67]

Name of MMP	Alternative name of MMP	ECM substrates
MMP-1	Collagenase-1	Various collagens, gelatin, entactin, aggrecan, tenascin
MMP-2	Gelatinase A	Elastin, fibronectin, various collagens, laminin, aggrecan, vitronectin
MMP-3	Stromelysin-1	Proteoglycans, laminin, fibronectin, gelatin, variouscollagens, fibrinogen, entactin, tenascin, vitronectin
MMP-7	Matrilysin	Same as MMP-3
MMP-9	Galatinase B	Same as MMP-2
MMP-11	Stromelysine-3	Inactive serpin

4. CONCLUSION

The composition of tumor microenvironment is extremely complex; wherein apart from expanding population of transformed cells, a variety of other non-cancer cells are also present such as smooth muscle cells, fibroblasts and macrophages. Collectively these cells are termed as stromal cells and they act as prominent modifiers of tumor growth and progression. The dynamic interaction between tumor cells and the surrounding stromal cells actively govern tumor progression as well as response to therapy. The most prominent subpopulation of these non-cancer cells are macrophages, which are often called tumor-associated macrophages (TAMs). Cytotoxicity of macrophages during the early immune response contributes to tumor killing, however in most solid tumors, macrophages inversely affect prognosis by potentiating cancer progression. In agreement with this, numerous clinical studies showed that macrophages promote tumorigenesis and also reported that over 80 per cent of studies show a correlation between macrophage density and poor patient prognosis. For example, there is a firm correlation between poor survival and increased macrophage quantity in thyroid, lung, and hepatocellular cancers. An equitable transcriptome analysis of follicular lymphoma shows that a macrophage transcriptional mark is a predictor of a poor prognosis.

The resident macrophages of tumor stroma exhibiting pro-tumor functions are often termed as tumor associated macrophages (TAMs). It is documented that TAMs are instrumental in moderating inflammatory responses, promoting angiogenesis and contributing to tissue remodelling, all of which apparently tend to promote tumor progression. The transcriptome of TAMs analysed in mouse models of breast cancer has also provided evidence that enrichment in macrophage transcripts is predictive of poor prognosis and decreased survival in human breast cancer.

Most studies have targeted upon events occurring in the primary tumor, often with metastasis as an end point. Metastasis requires not only the clemency of cells from the primary site but also their transit by the circulation or lymphatic to arrive at a distant site where the cells need to extravasate, survive, and prosper. This process of metastasis is very ineffective. In humans, there are so many of circulating cells released by tumors every day, but only a few make metastases. In fact, the most likely destiny for these cells is death, with extravasations and establishment

of micrometastases being major rate-limiting events. Although it is already known that macrophages inhabitate metastatic lesions, only recently has their role in metastasis been appreciated. For tumors to progress and become malignant they must shape their microenvironment to one that is at least permissive if not promoting. This is possibly due to selection of oncogenic mutations that edge to secretion of molecules that change the cellular composition and behaviour of the microenvironment. Among these changes, one is the recruitment of bone marrow-derived cells, of which macrophages are particularly abundant. Macrophages in primary and secondary tumors posses specific functional attributes that enable them to promote cancer progression and metastasis.

Macrophages provide tropic support to tumors and the genetic experiments that show that if you remove this support malignancy is suppressed strongly and contend that these cells or their particular signaling pathways are therapeutic targets. Unlike tumor cells, the genomes of macrophages are substantial, suggesting that they may not as readily become drug resistant. Significant progress has been made in identifying the molecular basis for both macrophage phenotypes and their actions in promoting specific aspects of tumor behaviour. Macrophages are important tumor-infiltrating cells and play pivotal roles in tumor growth and metastasis. Macrophages participate in immune responses to tumors in a polarised manner. However, it is not yet clear how TAMs induce tumor invasion. Prevailing new rational in the field of drug development is aimed at the development of target selective drugs. Development of such target specific drugs requires molecular understanding of normal and pathologically aberrant molecular pathways. In light of this the present study will provide valuable mechanistic information about hitherto obscure paracrine signaling networks emanating from TAMs that may impart invasive potential to cancer cells. In the quest of anti-metastatic agents we aim to target these pathways using specific inhibitors/neutralizing antibodies and thereby evaluate their suitability/efficacy as potent anti-metastatic agent.

Conflict of Interest : None

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