Editorial for Cancer Microenvironment and Pharmacological Interventions

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Editorial on the themed issue:

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The instrumental role that the cancer microenvironment plays in the pathogenesis of cancer was highlighted over 130 years ago by Stephen Paget, when he proposed the "seed and soil" hypothesis in 1889. He suggested that cancer metastasis is dependent on the characteristics of cancer cells the *seed* , and also on the composition of the supportive tissue microenvironment the *soil* , in which the cancer cells are able to successfully colonize. Despite advances in our understanding of the tumour microenvironment (TME) and the bone marrow microenvironment (BME), in the context of solid and haematological malignancies respectively, for many years the complexity of the tumour microenvironment has been somewhat overlooked. Most studies have been cancer-cell focussed deciphering the genetic, molecular and biochemical properties of cancer cells. These efforts have led to the generation of cancer cell selective targeting treatments, including immunotherapies, leading to some improvements in cancer patient outcomes. However, the development of therapy resistance and relapse, which are in part driven by the interaction of the cancer cells with their protective cancer microenvironment, are major unmet clinical needs in the field of cancer (Miari et al., 2021). Most FDA approved drugs solely target cancer cells, and thus fail to modulate the cancer-promoting TME.

This themed issue aimed at collating articles critically discussing cancer-supporting features of the tumour microenvironment in haematological and solid malignancies, including the cellular (e.g. mesenchymal stromal/stem cells) and non-cellular elements (e.g. extracellular matrix; ECM components). The issue also captures cutting-edge basic science (e.g. 3D *in vitro* model systems), to more accurately model and therapeutically target cancer cell-to-TME interactions, with the intention of improving the likelihood of identifying approaches that can improve the efficacy of existing anti-cancer interventions, through to the development of novel therapeutic interventions modulating the cancer-promoting TME.

Specifically, this themed issue there are six articles focussed on the following:

Mesenchymal stromal/stem cells (MSCs) and cancer associated fibroblasts (CAFs), along with tumour-associated macrophages (TAMs) (Cassetta and Pollard, 2018), are one of the most prominent cell types present within the TME, and have been the discussed in depth in this issue, as it is included in three articles and thus, it represents the increasing amount of studies investigating MSCs/CAFs as key drivers in: reprogramming the microenvironment towards a cancer-permissive environment; cancer development and progression; cancer therapy resistance, and serving as therapeutic targets in cancer. Although research on MSCs/CAFs is highly encouraging, it is predominantly still at the preclinical phase, with no therapies targeting/modulating MSC/CAF function currently approved. Therapeutic intervention at the level of

MSCs/CAFs has been hindered mainly due to the extensive diversity/heterogeneity of this cell population (**Miari & Williams**). Interestingly, an immunocytokine, Simlukafusp Alfa (FAP-IL2v), a monoclonal antibody (mAb) targeting the CAF marker, fibroblast activation protein α (FAPα), conjugated to an IL-2 variant (IL-2v), has been tested in a recently completed Phase I clinical trial (**NCT02627274**) (Waldhauer et al., 2021). This therapy was tested in combination with the anti-HER2 and anti-EGFR mAbs Trastuzumab and Cetuximab respectively, for treating breast cancer and head and neck cancer patients. This novel therapy exploits the inherent tumour tropism of CAFs (**Borzone & Wheadon**), coupled with the IL-2 variant retaining its propensity for solely binding with high affinity to the IL-2 receptor signalling subunits β and γ (IL-2Rβγ), immunstimulating (via T-cell expansion), and subverting the immunosuppressive effects (via regulatory T cell [Treg] mediated immune tolerance), associated with IL-2Rα engagement/activation. Advantages of this therapy over traditional IL-2 cytokine therapies are two-fold. First, enhanced half-life of IL-2 and efficacy to stimulate anti-tumour immune responses within the TME. Second, circumventing severe adverse effects associated with systemic/untargeted IL-2.

The fourth paper in the series by **Zou and colleagues**extensively reviews the function, regulation, and distribution of the immune checkpoint ligand programmed death-ligand 1 (PD-L1), as to better understand these aspects, to aid development of strategies improving the efficacy of PD-L1 immunotherapies/mAbs. There are 13 approved mAbs targeting programmed cell death protein-1 (PD-1) or PD-L1, and these have been revolutionary in treating patients with solid cancers. However, the clinical efficacy of these immunotherapies has disappointingly been less than predicted, resulting from several cancer cell-inherent and TME-mediated resistance mechanisms.

One of the major immune (and potentially treatment) evasion strategies is exogenous/exosomal PD-L1 (exoPD-L1), that is present on extracellular vesicles (EVs) secreted by cancer cells. This mechanism is particularly exemplified glioblastoma, in which glioblastoma cells release PD-L1$^+$ EVs, driving formation of immunosuppressive monocytes capable of suppressing T-cell proliferation (Himes et al., 2020). Importantly, this would potentially render PD-1/PD-L1 mAbs clinically ineffective, via PD-L1$^+$ EVs acting as a decoy for PD-1/PD-L1 mAbs and preventing the targeting of cancer cells and the surrounding immune microenvironment. ExoPD-L1 could serve as a biomarker for PD-L1 immunotherapy response and has advantages over conventional biopsy. Moreover, it is tempting to speculate that the efficacy of anti-PD-L1 immunotherapy, could be enhanced by providing cancer patients with a first low dose of anti-PD-L1 mAb, to bind to decoy PD-L1$^+$ EVs, with further doses used to target PD-L1 present in cancer cells and immune cells within the TME.

Accurate modelling of cancer cell-to-TME interactions and overcoming TME -driven therapy resistance in haematological and solid cancers, remain as outstanding needs in the cancer field. Current *in vitro* and*in vivo* models for investigating cancer cell-to-TME interactions, predominantly rely on the use of 2D *in vitro* co-culture model systems and xenograft murine models. However, there are caveats associated with these model systems. 2D model systems overestimate therapeutic efficacy (Dainiak et al., 2008), failing to translate into clinical efficacy. Furthermore, xenograft mouse models lack a functional immune system, with findings generated from these models not directly applicable to humans, as human cancer cell-human TME interactions differ from human cancer cell-murine TME interactions at the cellular and molecular level (**Martinez & Guzman**). Three-dimensional model systems, incorporating TME-resident cells (e.g. X,Y,Z) and ECM components hold much promise (**Nyamondo & Wheadon**) as they more accurately reflect TME features, such as hypoxia and reactive oxygen species, not present in most 2D models. Furthermore, 3D models mirror the well-established therapy resistant effects previously reported for the TME. **Ogana et al** discussed targeting integrins to overcome TME-elicited drug resistance in acute myeloid leukaemia (AML). Although no integrin-targeted treatments have been approved to date, this represents a potentially effective therapeutic approach not just in AML, but also for other blood cancers and solid cancers, to circumvent cell adhesion-mediated drug resistance (CAM-DR).

To summarise, conclusions of the articles in this issue provide novel insights into how alterations in cellular and non-cellular components of the microenvironment drive cancer development and progression, and

mediate therapy resistance/failure, promoting relapse and sub-optimal clinical outcomes in cancer patients. Despite the extensive evidence on these highly important issues, these articles show that there are several outstanding features to be determined in the fascinating area of cancer microenvironment and pharmacological intervention. After reading this issue, aspects such as, modelling and targeting complex cancer cell-to-TME interactions to improve the efficacy of existing and new therapies, will become more apparent to the reader. This will reinforce the concept that studies identifying and targeting cancer-cell-inherent and cancer microenvironment driven mechanisms of resistance, are still crucially needed to enhance survival and quality of life in cancer patients.

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