Fatal attractions? Correlations of CXCL12–CXCR4–CXCR7 expression with disease progression in melanoma and Kaposi sarcoma

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Commentary

Fatal Attractions? Correlations of CXCL12-CXCR4-CXCR7 expression with disease progression in melanoma and Kaposi's Sarcoma.

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Chemokines are a family of small chemotactic cytokines that regulate tissue homeostasis and immune cell migration and recruitment to sites of inflammation along chemotactic gradients. In recent years, CXCL12 (or stromal derived growth factor-1, SDF-1), has emerged as a cancer-associated factor through the promotion of tumour cell proliferation, migration and metastasis. Numerous tumour cell types over-express the G-protein coupled receptor for CXCL12, C-X-C chemokine receptor type 4 (CXCR4). Signalling through the CXCL12/CXCR4 axis mediates recruitment of tumour cells to secondary tumour sites that express high levels of the ligand, including metastatic niches within lymph nodes, lung, liver and bone [1]. Consistent with this, a previous meta-analysis revealed that overexpression of CXCR4 correlates with poor prognosis in multiple tumour types [2]. Paradoxically though, expression of the ligand and receptor within tumours may also have a protective effect against metastasis (potentially by swamping chemotactic CXCL12 gradients [3]. Despite such nuances of context-dependent chemokine tumour biology, in general, both CXCL12 and its receptor are considered potential novel therapeutic targets for cancer.

While CXCL12/CXCR4 signalling has many important homeostatic functions, CXCR7 (recently renamed Atypical Chemokine Receptor 3, or ACKR3), is recognised as an alternative high affinity receptor for CXCL12 that is more selectively expressed in tumour-associated vasculature and subsets of cancer cells. CXCR7 can function as a decoy receptor affecting CXCL12 gradients and, unlike CXCR4, does not induce intracellular calcium signalling. However, CXCR7 signalling via β-arrestin rather than G-proteins is reported to enhance cell migration, adhesion and proliferation. Thus CXCR7 does not act merely a 'sink' for CXCL12 [4] and it has become increasingly important to expand our understanding of cancer-associated chemokine expression to include the CXCL12/CXCR4/CXCR7 triad.

In this issue, two studies focussing on primary cutaneous melanoma (McConnell *et al*) and Kaposi's sarcoma (Desoyner *et al*) highlight the potential for CXCL12/CXCR4/CXCR7 chemokine ligand/receptors as prognostic biomarkers for skin neoplasias.

Previous informative studies on the correlation between both CXCL12 and CXCR4 expression levels and prognosis in melanoma are scarce and occasionally contradictory. Increased CXCR4 expression correlates with melanoma ulceration, thickness and disease progression, while the role of CXCL12 and CXCR7 remains ill-defined.

To redress this lack of data, McConnell *et al* retrospectively compared expression levels of CXCL12, CXCR4 and CXCR7 with clinical correlates in a cohort of 77 formalin-fixed paraffin embedded (FFPE) tissues from patients with benign melanocytic naevi (n=13) or primary melanoma (n=64) (AJCC Stage 1 – 3 at diagnosis). In the primary tumour panel, immunohistochemical (IHC) analysis revealed higher CXCR4 expression levels in tumours that subsequently metastasized during the seven year follow-up period. These data would be consistent with the idea that CXCR4 expression in melanoma promotes metastasis, and in fact, higher CXCR4 expression in patients with stage II disease at diagnosis predicted significantly decreased disease free survival with these patients having a three-fold higher risk of disease recurrence. Interestingly, although in vitro analysis of CXCL12 expression in melanoma cell

lines and dermal fibroblasts indicated that all cells expressed cytoplasmic CXCL12 (detected by immunofluorescence (IF)), this cytoplasmic expression pattern did not correlate with the ability of the cells to secrete detectable levels of CXCL12 – a potential complication worth noting when interpreting CXCL12 IHC and IF expression studies. Nevertheless, elevated expression of CXCL12 in the tumour microenvironment (epidermis) correlated with an increase in the time taken to metastasize; the implication being that the high CXCL12 gradient in the epidermis would restrict deeper melanoma invasion and promote radial rather than vertical melanoma growth through the dermis. CXCL12 expression was downregulated in BRAF/NRAS mutant tumours compared with wild type tumours revealing a potential mode of regulation of CXCL12 expression by MAPK activation. It would be of interest now to evaluate the extent of the causal relationship between loss of CXCL12 expression and the mutant RAS/RAF melanoma phenotype. Little expression of the CXCR7 receptor was evident in primary melanoma tumour tissue, but CXCR7 expression was detected in tumour-associated endothelial cells, suggesting further potential tumour/microenvironment interactions and adding to the potential complexity of chemokine biology in melanoma. Indeed expression of CXCR7 on tumour-associated endothelial cells in breast cancer restricts metastasis [5] emphasising the importance of the microenvironment in disease progression.

In Desoyner et al, expression of the CXCL12/CXCR4/CXCR7 trio is analysed in Kaposi's sarcoma (KS). KS has a complex aetiology involving a skin microenvironment loaded with growth factors and cytokines, immune suppression and HHV-8 infection. The authors report that CXCL12, CXCR4 and CXCR7 are all upregulated in both AIDS-associated (n=12) and classic (n=12) Kaposi's sarcoma when compared with benign angioma tissue (n=10). This research builds on previous *in vitro* data implicating the trio in KS pathobiology but it extends the number of patient samples analysed for chemokine and chemokine receptor expression, and importantly provides analysis of clinical correlates of KS-type and lesion stage (macules, papules or nodules). The proportions of CXCL12, CXCR4 and CXCR7 positive cells were significantly higher in nodules (70%) compared with the less severe macules and papules. Expression of the trio therefore increases with lesion severity. Unfortunately, plasma CXCL12 levels did not correlate with the levels of detection of CXCL12 in lesions, so would not be a useful clinically as a blood borne biomarker for disease progression, severity or response to treatment.

Both studies highlight the potential for CXCL12/CXCR4/CXCR7 as biomarkers in skin neoplasias. Further study is warranted with larger patient cohorts, coupled with integration of these findings with expression patterns on lymphocytes [6] and investigation of the significance and role of cytoplasmic ligand and cytoplasmic and nuclear receptor expression in tumour cell biology. This should help determine when the CXCL12/CXCR4/CXCR4 axis acts to promote disease progression and thus when it might be usefully targeted. In the future, specific reagents will be needed that can detect the six different CXCL12 isoforms (α, β, γ, δ, ε, and φ). These will be invaluable in deciphering the potentially different roles of the isoforms in cancer development [7].

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