

Cutting Edge View

PAX3-FKHR Chimeric Oncoprotein

Hiding Itself from Immune Detection?

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The ability of cancer cells to avoid detection by the immune system has been a subject of debate and controversy since it was initially described in terms of the immune surveillance hypothesis by Burnet and Thomas in the 1950s.^{1,2} Broadly speaking immune surveillance implies the existence of tumor antigens, molecular markers that could serve as a target for the immune system by virtue of their mutation, or overexpression by transcriptional, genetic or epigenetic mechanisms. Naturally occurring tumor antigens have been most frequently and convincingly described in the context of viral infection, and the overexpression of onco-fetal antigens in cancers such as melanoma.

Key evidence in favor of immune surveillance is the immunoinhibitory environment of tumors and of cancer patients as exemplified by high circulating levels of immunoinhibitory cytokines such as IL10 and TGF β ,^{3,4} loss of MHC class I or β 2-microglobulin expression,⁵ increase in regulatory T cell numbers and activity,⁶ and decrease in numbers and activity of dendritic cells.⁷ Arguments against immune surveillance have included the contention that its existence in normal individuals should necessitate an increased incidence of cancer in patients with severe immunodeficiencies. However, within this patient population, apart from specific viral associated malignancies such as EBV driven lymphoproliferative diseases, Hodgkins or Kaposi's sarcoma, there is no significant increase in common cancers.

However, patients with immunodeficiencies are perhaps not an ideal model for assessment of evidence for or against the immune surveillance hypothesis for three reasons. Firstly, patients with the most severe forms of immunodeficiency have significantly reduced life expectancy unless their immunodeficiency is corrected. Therefore there might not be sufficient time for common epithelial tumors, requiring multiple hits, to develop. Secondly, severe immunodeficiency is not usually associated with loss of natural killer (NK) cell function and these may play a key role in the elimination of common cancers at an early stage of development. Thirdly, severe immunodeficiency in humans is rare, making epidemiological studies of incidence of common cancers difficult. Therefore studies in immunodeficient animals have been used to look for subtle differences in cancer incidence. Several studies in athymic mice with reduced T cells have failed to show any increased cancer incidence but this might be because these strains have increased NK function.^{8,9} Perhaps the most compelling studies in favor of immune surveillance are of RAG^{-/-}STAT1^{-/-} or RAG^{-/-} γ IFN γ ^{-/-} double knock out mice. These animals have no T or B cells and have impaired interferon gamma response, and when allowed to develop into their second year show an increased incidence of various epithelial tumors.¹⁰⁻¹²

Despite this, it has been claimed that common cancers in humans are not naturally immunogenic whereas pathogen associated malignancies are; however, it is claimed, it is the presence of the pathogen itself rather than presence of pathogen independent tumor antigens that is the target of immune response. An alternative model that could explain the lack of increased incidence in the immunocompromised host is that normal cancers in the normal process of oncogenesis acquire an inability to hide their tumor antigens from the immune environment. In other words, ability to avoid immune detection is an integral part of cancer development and does not represent additional mutations that occur, and are selected for, after malignant transformation has already occurred. What is the evidence that deregulation of oncogenes or inactivation of tumor suppressors could suppress the immune environment?

The transcription factor STAT3 has emerged in recent years as having properties of both pleiomorphic transforming capacity and profound immunoinhibitory effects.^{13,14} STAT3 is activated by phosphorylation downstream from IL6 or IL6 like cytokines such as LIF, following signal transduction by Janus Kinases (JAK1 and JAK2). However, the exact transcriptional properties of the STAT family of transcription factors are rather complex. STAT1 behaves most like a tumor suppressor downstream from interferon

gamma signaling, whereas STAT3 is oncogenic and can be constitutively activated in cancer; moreover STAT1 and STAT3 can function as homo or heterodimers with opposing effects. Hence defining the actual transcriptional targets of individual components of this system is a complex task. What has emerged clearly in recent years, however, is the importance of STAT3 in numerous cancer types both as an oncogene and an immunoinhibitory factor. The transcriptional targets of STAT3 include Cyclin D1, VEGF, Bclxl, p53; and its transforming properties include cell cycle progression, prosurvival and proangiogenic effects. However, deregulated STAT3 additionally inhibits migration and function of cells of the immune system.

We have recently described how an oncogenic fusion protein, PAX3-FKHR expressed in alveolar rhabdomyosarcoma, has a dual function of malignant transformation and alteration of the tumor microenvironment to inhibit the cells of the host immune system.¹⁵ The PAX3-FKHR immune effect is independent of its ability to bind PAX3 target promoter sequences but dependent on interaction with STAT3. PAX3-FKHR has the intact DNA binding elements of the PAX3 developmental transcription factor and a strong transcriptional activation sequence from FKHR. Previously published models of the transforming effects of PAX3-FKHR have focussed on the fusion protein being a stronger transcriptional activator of PAX3 target promoters. Moreover PAX3-FKHR appears to need to be expressed at very high levels in cancer cells compared with wild type PAX3¹⁶ and recent animal models have suggested that its formation by chromosomal rearrangement could be the initiating event in formation of alveolar rhabdomyosarcomas.¹⁷ Is it likely that this high level expression of PAX3-FKHR could itself alert the immune system in the developing tumor? The high level of expression especially of the PAX3 sequences, the novel sequences joined at the fusion breakpoint, and expression of novel targets some of which could be seen by the immune system as foreign, are all mechanisms that could potentially alert immune surveillance. The new data indicate that PAX3-FKHR at this early stage of tumor development could inhibit immune responses and allow tumor progression, in a STAT3 dependent manner, through downregulation of MHC class I expression and production of immunoinhibitory cytokines.

Is STAT3 the key factor in linking tumor progression with immune escape? Certainly the high incidence of STAT3 activation in cancers suggests it is a common theme, but is it a key initiating event in progression of those cancers linking immune escape and tumor progression? Clues to answer these questions may emerge if other known primary transforming oncogenes are found to activate STAT3 either through direct interaction, interference with upstream signalling, or inhibition of antagonists such as STAT1.

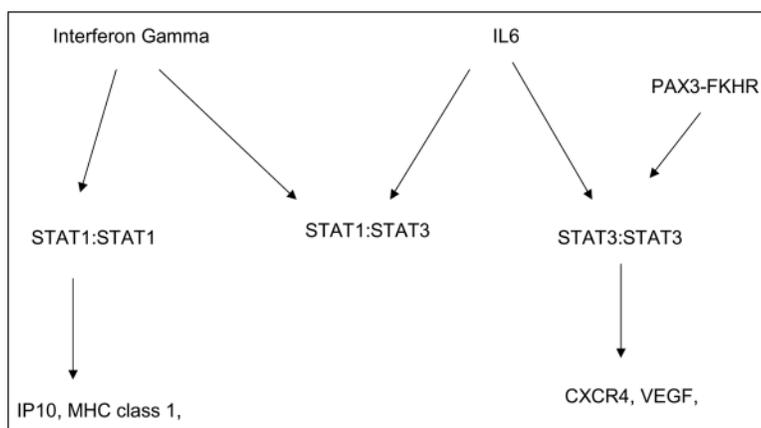


Figure 1. A speculative model of how PAX3-FKHR might interact to activate STAT3. Transcription of pro inflammatory factors downstream from STAT1 and anti-inflammatory factors downstream from STAT3 is seen as a balance between opposing transcriptional effects. Activation of STAT3 by PAX3-FKHR results in loss of expression of danger signals such as IP10 and MHC class 1 by reducing formation of STAT1 transcriptional complexes.

Further Reading

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