INSIDE LI

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S1P signaling in Hodgkin lymphoma migration

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Sphingosine-1-phosphate receptor 1 (S1PR1) is a G protein-coupled receptor that binds S1P. The binding to S1PR1 of S1P, which is present at high levels in blood and lymph, promotes lymphocyte migration through activation of the G_i -phosphatidylinositol-3-kinase (G_i -PI3K) pathway and the small GTPase Rac. Classic Hodgkin lymphoma (CHL) typically presents with involvement of contiguous lymph nodes. The mechanism of spread is unknown, but it must involve migration of neoplastic cells.

Because the neoplastic cells of CHL are abnormal B lymphocytes (Hodgkin-Reed-Sternberg cells), Kluk *et al* asked whether S1PR1 signaling might be involved in CHL cell migration. They discovered that S1PR1 was highly expressed at both the RNA and protein levels in CHL cell lines. They then demonstrated that S1P stimulated migration in CHL cell lines *in vitro*. Moreover, both VPC44116 and FTY720-P, antagonists of S1PR1 function, inhibited S1P-stimulated CHL cell migration *in vitro*. Finally, because S1PR1 cell migration is dependent on G_i -PI3K signaling, the authors demonstrated that inhibition of PI3K signaling also inhibited S1P-stimulated CHL cell migration. To apply their cell line findings to patients with CHL, the authors used immunostaining to examine S1PR1 expression in 57 cases and found that only 7 (12%) expressed

S1PR1. When they took follow-up into account, S1PR1 positivity was present in 5 of 21 (24%) of CHL cases with multiple recurrences, suggesting that S1PR1 immunoreactivity might be a prognostic biomarker for CHL. An expanded assessment of more CHL patients will be necessary to confirm a statistically significant association between S1PR1 immunoreactivity and recurrence.

Mechanism of TTF1 expression in SCLC

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Thyroid transcription factor 1 (TTF1) plays critical roles in thyroid, lung, and brain development. In cancer, it is expressed exclusively in lung and thyroid cancers. In the adult lung, TTF1 is stably expressed in peripheral lung epithelial cells. Therefore, it is not a surprise that about 70% of pulmonary adenocarcinomas, which tend to arise in the peripheral lung, express TTF1. What is surprising is that more than 90% of small-cell lung cancers (SCLCs), which tend to arise centrally, also express TTF1.

Because TTF1 is important for fetal brain morphogenesis and SCLCs express a neuroendocrine cell lineage-expression pattern, Sakaeda *et al* wondered whether TTF1 expression might be due to a mechanism related to the neuroendocrine expression pattern. They found that

BRN2, a class III POU domain transcription factor reported to play an important role in development of the endocrine hypothalamus, drives expression of TTF1 in SCLC. BRN2 was found to bind the promoter upstream of the *TTF1* gene. Ectopic expression of BRN2 upregulated *TTF1* expression whereas depletion of BRN2 in SCLC cell lines inhibited *TTF1* expression. Furthermore, the combination of BRN2 and TTF1 expression was found in primary SCLC tumor samples. To ascertain whether epigenetic mechanisms play a role in *TTF1* gene expression, the authors performed bisulfite sequencing of the CpG islands upstream of *TTF1*. They found no evidence of the hypomethylation that might have explained the *TTF1* expression seen in SCLC. In summary, Sakaeda *et al* found that a neural-specific transcription factor drives *TTF1* expression in pulmonary SCLC.

Waves of MMP-9 contribute to renal fibrosis

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The zinc-dependent matrix-degrading enzymes called matrix metalloproteinases (MMPs) have diverse biological functions dependent on their expression and the availability of different substrates in various cellular contexts. Although it is obvious that MMPs contribute to

matrix degradation directly through their proteolytic functions, it is less obvious how they contribute to matrix deposition, which has been characterized as a profibrotic effect. MMP-9, also known as gelatinase B, has been implicated in renal fibrosis because it fosters epithelial-to-mesenchymal transition (EMT) of renal tubular epithelial cells (TECs), which produce actin after conversion to myofibroblasts. However, many details about how MMP-9 promotes renal fibrosis remain unknown.

To further investigate how MMP-9 promotes renal fibrosis, Tan *et al* undertook a detailed *in vitro* and *in vivo* analysis. They demonstrated that there were two waves of MMP-9 expression in a mouse model of renal fibrosis. The first wave of MMP-9 was expressed by TECs, the second by a combination of TECs, macrophages, and myofibroblasts. Mechanistically, they demonstrated that the initial wave of MMP-9 cleaved osteopontin, and that cleaved osteopontin stimulated an influx of macrophages that essentially amplified the production of MMP-9. Inhibition of MMP-9 decreased EMT and renal fibrosis. The investigators also report evidence that macrophages may contribute to renal fibrosis by participating in TEC EMT induction. Finally, their results suggest that inhibition of MMP-9 early or late—but not at the midstage—in renal fibrosis could result in improved renal function.

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nature.com/pathology

Using a pill to image the GI tract Traditional gastrointestinal (GI) endoscopy is time-consuming and costly. To improve the cost-effectiveness and optical resolution of endoscopy, Gora *et al* developed a tethered optomechanical capsule, the size of a large pill, that is swallowed and propelled along the GI tract via peristalsis. The capsule, which the authors describe in a recent technical report in *Nature Medicine*, captures circumferential, high-resolution cross-sectional images (7 μ m by 30 μ m) and is retrieved by pulling on the tether. Imaging 15 centimeters of esophagus took 6 minutes, on average, for two passes and retrievals. Other advantages are that patients do not require sedation and, because use of the device does not require extensive training, it can be used as a screening tool by primary-care physicians. Once the capsule is retrieved, it is disinfected for reuse.

Nature Medicine 2013;19:238–240; doi:10.1038/nm.3052

A novel source of mutation in breast cancer Breast cancers have somatic mutation spectra dominated by C-to-T transitions. To identify the source of these transitions, Burns *et al* measured the mRNA levels for each of the 11 polynucleotide cytosine deaminase family members in breast cancer cell lines. As described in a recent letter in *Nature*, they found that *APOBEC3B* levels were upregulated by ≥ 3 s.d. relative to controls in most of the cell lines. Furthermore, they demonstrated that upregulation of *APOBEC3B* led to increased levels of genomic uracil and C-T transitions whereas knockdowns had the opposite effect. These results were confirmed in primary breast tumors. Interestingly, increased *APOBEC3B* levels correlated with inactivated *TP53* in primary tumors and cell lines, suggesting that *TP53* inactivation may be required to allow cells to bypass DNA-damage checkpoints triggered by A3B (encoded by *APOBEC3B*).

Nature, published online 6 February 2013; doi:10.1038/nature11881

Autophagy protects hematopoietic stem cells Autophagy is able to help cells survive by maintaining nutrient and energy levels during periods of metabolic starvation or stress. In a study recently reported in *Nature*, Warr *et al* sought to determine whether autophagy could protect hematopoietic stem cells (HSCs) from stress. They discovered that autophagy protected HSCs when deprived of cytokines or nutrients through a FOXO3A-mediated pro-autophagy expression program that enabled rapid activation of autophagy in response to stress. The authors also discovered that autophagy played a key role in helping aged HSCs survive. Their results challenge the prevailing view in the aging field that impaired autophagy is one of the principal causes of cellular aging. Indeed, it appears that autophagy keeps various age-related stresses at bay in HSCs, enabling them to continue to replenish the bone marrow.

Nature 2013;494:323–327; doi:10.1038/nature11895

RNA editing promotes hepatocellular carcinoma Chen *et al* recently reported in *Nature Medicine* their application of high-throughput RNA sequencing to three pairs of hepatocellular carcinoma tumors. In validating the DNA samples, they found that *AZIN1* (encoding antizyme inhibitor 1) had a high frequency of nonsynonymous A-to-I (G) transcript editing, leading to a Ser-to-Gly amino acid substitution. Mechanistically, the Ser-to-Gly substitution at amino acid residue 367 resulted in shifting *AZIN1* from the cytoplasm to the nucleus, where it bound with increased avidity to antizyme, a protein that normally acts as a tumor suppressor by binding to and promoting the degradation of ornithine decarboxylase and cyclin D1, which promote cell proliferation. Thus, mutant *AZIN1* acts as an oncogene by inhibiting the tumor-suppressing effects of antizyme.

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