Meeting report

Cancer, oncogenes and signal transduction

Edward J McManus and Dario R Alessi

Address: Medical Research Council Protein Phosphorylation Unit, School of Life Sciences, University of Dundee, Dow Street, Dundee DD1 5EH, UK.

Correspondence: Edward J McManus. E-mail: e.j.mcmanus@dundee.ac.uk

Published: 24 June 2004

Genome ***Biology*** 2004, **5:**332

The electronic version of this article is the complete one and can be found online at http://genomebiology.com/2004/5/7/332

A report on the European Molecular Biology Laboratory (EMBL) 'Oncogenes and Growth Control' meeting, Heidelberg, Germany, 17-20 April 2004.

The four-day meeting at the European Molecular Biology Laboratory (EMBL) brought together many of the specialists, mainly from Europe and the USA, working on cancer and signal transduction. It was the 20th meeting in this series, and celebrated 50 years since the discovery of protein kinases by Burnett and Kennedy (*J Biol Chem* 1954, **211:**969-980) and 25 years since the discovery of tyrosine phosphorylation by Hunter and colleagues (Eckhart *et al.*, *Cell* 1979, **18:**925-933). Much of the meeting focused on advances obtained using murine models of oncogenesis, and this aspect was nicely complemented by more mechanistic talks.

Oncogenic signaling

One of the most important recent advances in the field of kinase research was the characterization by Tony Hunter (Salk Institute for Biological Sciences, La Jolla, USA) and his colleagues of the evolutionary relationships between the 518 mammalian protein kinases encoded in the human genome. The kinases represent the largest family of human enzymes, collectively termed the kinome, and knowledge about their evolution has greatly facilitated research into these important enzymes. The value of the kinome data was exemplified by the many talks throughout the meeting describing work in which information from Hunter's study was used. In his talk, Hunter described the families of human kinases and the domains that they contain. Interestingly, 40% of all kinases have multiple splice variants and 10% of the total encode catalytically deficient enzymes that have been termed pseudokinases. The roles of these inactive enzymes are poorly defined, but recent examples indicate that a number of pseudokinases, such as ErbB3 and STRAD, play roles in activating conventional protein kinases, namely the epidermal growth factor (EGF) receptor and the serine-threonine kinase LKB1, respectively.

Julian Downward (Cancer Research UK, London, UK), made use of Hunter's kinome database in an RNA interference (RNAi) screen of all human kinases, to search for enzymes that regulate senescence induced by Ras. One of the novel targets identified encodes MINK, a kinase that resembles the yeast Ste20 kinase; MINK was shown to be stimulated by Ras through the extracellular signal-regulated kinase (ERK) or mitogen-activated protein (MAP) kinase pathway. Activation of MINK by Ras leads to the activation of p38 MAP kinase through the upstream TAK/MKK6 signaling pathway, and activation of p38 was shown to be required for the senescence pathway. This pathway represents one of the longest and most complicated kinase cascades described to date, involving at least eight or nine different protein kinases.

Increasing numbers of protein-kinase inhibitors are being developed and employed to define the physiological roles of the kinases. But a major concern relating to the interpretation of these studies is the fact that most of the drugs that have been developed thus far target the ATP-binding pocket of kinases and are therefore unlikely to be completely specific, as this site is relatively conserved between different kinases. Giulio Superti-Furga (Cellzome Inc. and EMBL, Heidelberg, Germany) presented an elegant approach to identifying other kinases that might be targeted by a kinase inhibitor; in this approach, the inhibitor is immobilized onto an insoluble resin and used to affinity-purify from cell extracts protein kinases that bind the drug. The enzymes that have bound to the drug are then detected by mass spectrometry. Using Gleevec/STI-571 as an example, Superti-Furga and colleagues purified a kinase involved in Alzheimer's disease, which was not previously known to be

targeted by this drug. Superti-Furga speculated that Gleevec might therefore potentially be used to treat Alzheimer's disease. Thus, his approach to identifying targets of kinase inhibitors may not only lead to knowledge concerning the overall specificity of inhibitors, but, when used with clinically approved drugs, could potentially lead to the development of new therapeutic uses for these compounds.

A couple of years ago much excitement was caused by the finding that most melanomas are caused by mutations in the B-Raf kinase that result in activation of the ERK pathway. Remarkably, both activating and inactivating mutations in B-Raf were found in melanomas as well as in other cancers. David Barford (Institute of Cancer Research, London, UK) described studies that he has done with Richard Marais (also at the Institute of Cancer Research) to elucidate the three-dimensional structure of the catalytic domain of B-Raf. The results of this work demonstrate the structural basis by which cancer-associated mutations regulate B-Raf activity. Most interestingly, although most mutations lead to a higher B-Raf activity, a subset of mutants found in human cancers actually inactivate B-Raf; the structural analysis revealed that although these mutations abolish B-Raf activity, they stabilize B-Raf in a conformation similar to that of the active enzyme. How these mutant forms of B-Raf then go on to activate ERK is not known, but one possibility is that the active conformation of B-Raf can stimulate the activation of c-Raf, by an as yet unidentified mechanism.

Signaling, growth and polarity

An interesting feature of the meeting was the use of spectacular movies of live cells, which kept the audience interested and explained the physiological processes better than words or fixed images could ever do. Seeing is definitely believing! This was exemplified in the talk by Jürgen Knoblich (Research Institute of Molecular Pathology, Vienna, Austria) who established that the Aurora kinase forms a complex with a novel protein named Bora that it phosphorylates. This phosphorylation event was shown to play an important role in enabling proteins, such as the cell-fate determinant Numb, to be asymmetrically segregated into specific daughter cells after cell division. Knoblich proposed that asymmetric localization of Numb requires both a temporal signal mediated by the activation of Aurora by the Cdc2 cell-cycle-regulatory kinase as well as a spatial signal provided by a complex made up of the spatially localized Par3 and Par6 proteins with atypical protein kinase C (PKC).

Many talks discussed the roles of the signal transducer β-catenin/Armadillo in the regulation of transcriptional responses in various organisms. The β-catenin protein is known to play a critical role in the canonical Wnt signaling pathway, where it lies between the cell-surface Wnt signal receptor and the nuclear transcription factor LEF1. Boudewijn Burgering (University Medical Center, Utrecht, The Netherlands) described the discovery of the forkhead transcription factor Foxo as a novel binding partner of β-catenin, and provided biochemical and genetic evidence that β-catenin enhances transcriptional responses mediated by the Foxo transcription factors in response to oxidative stress.

In the past few years there has been a great deal of research into understanding the roles of the serine-threonine kinase LKB1, which is mutated in the rare inherited Peutz-Jeghers cancer syndrome. Hans Clevers (University Medical Center, Utrecht, The Netherlands) displayed striking movies and images showing that the activation of LKB1 in unpolarized intestinal cell lines induces the complete polarization of these cells. The changes included the formation of microvilli at the apical edge, cytoskeletal rearrangements that are characteristic of polarized colonic cells, and the formation of gap junctions. It was previously thought that these events required cell-cell contacts, but this may not be the case, as LKB1 activation induced cell polarization even in isolated cells. It will be interesting to establish the downstream signaling pathway by which LKB1 regulates these cellular events and whether it involves the recently identified LKB1 substrates belonging to the AMP-activated protein kinase (AMPK) family. Tomi Mäkelä (University of Helsinki, Finland) demonstrated that colon cancer cells from patients with mutations in LKB1 had elevated levels of cyclooxygenase-2 (COX-2) activity. Employing the COX-2 inhibitor celecoxib, he demonstrated a remarkable decrease in the size of hamartoma tumors in the intestine of $LKB1^{-/+}$ heterozygous mice as well as in several human patients with Peutz-Jeghers syndrome. This work indicates that celecoxib may be useful for the treatment of cancers that have mutations in LKB1.

A major hope is that knowledge gained from understanding the role of signaling pathways in regulating cell growth and proliferation can be exploited to develop more effective anticancer therapies. Scott Lowe (Cold Spring Harbor Laboratory, USA) described work showing that cancers in mice caused by the activation of protein kinase B (PKB; also known as Akt) were effectively treated by a combination of rapamycin, which inhibits the molecular target of rapamycin (mTOR), and conventional chemotherapeutic agents. But these potent effects were not observed in cancers with other genotypes, such as those caused by the overexpression of the cell-death inhibitor Bcl-2. Lowe also demonstrated that the eIF4E translation factor, which is regulated by Akt and mTOR, is highly oncogenic, and cancers overexpressing this factor failed to respond to rapamycin. These results provide a clear example of how critical it will be to understand the genotype of cancer cells and which signaling pathways are activated in them if we are to be able to treat these malignancies efficiently.

Invasion and metastasis

Recent work by Richard Treisman (Cancer Research UK, London, UK) has found that transcription dependent on the

serum response factor (SRF) is regulated by the Rho GTPase through actin polymerization, but the mechanism by which this is achieved was not understood. This question now seems to have been resolved, as Treisman described a novel SRF binding partner and co-activator named MAL, which is anchored onto polymerized actin in the cytosol and released into the nucleus after activation of the Rho GTPase. In this way, MAL is thought to play a role in SRF-dependent transcription. Interestingly, MAL was also phosphorylated by ERK, another key regulator of immediate-early gene transcription by SRF. The next advance in this area of work will be to elucidate the role that the phosphorylation of MAL plays in the regulation of SRF function. In the subsequent talk, Pernille Rørth (EMBL, Heidelberg, Germany) showed that MAL complexed to SRF plays a critical role in border-cell migration during *Drosophila* oogenesis. Rørth also proposed that accumulation of nuclear MAL is induced by cell deformation during migration, and that invasive cells go through multiple MAL/SRF activation cycles as they migrate.

Another GTPase that received attention at the meeting was Ral. Chris Marshall (Institute of Cancer Research, London, UK) has studied the function of Ral-GDS, a Ras-dependent activator of Ral, using knockout techniques, and he described work that demonstrates that the Ral pathway is not oncogenic alone but cooperates with Ras or Raf to induce cellular transformation. Moreover, he described the discovery of a novel Ral effector protein named ZONAB, which, like β-catenin, has functions in the nucleus regulating transcription and at the plasma membrane controlling cell-cell contacts. Overall, the meeting demonstrated the wealth of research into important signaling pathways that regulate cell growth and cancer. We can look forward to further exciting advances in the field over the coming months.