

## NEWS IN BRIEF

tially, allowing multilabel imaging at a single wavelength and thus avoiding chromatic aberrations.

To demonstrate this, the researchers targeted rsFastLime to mitochondria in live budding yeast and labeled the actin-binding protein Abp1, which localizes to cortical actin patches, with Padron. Using an iterative switching protocol, they monitored the dynamics of these cellular features in three dimensions over several hours. Although the two labels are both green fluorescent proteins, they could be distinguished based on their distinct switching behavior.

Second, the switching of RFSPs can be exploited for subdiffraction-resolution microscopy. Using another new version of Dronpa, so-called broad-spectrum (bs)Dronpa, Jakobs and colleagues demonstrate the utility of these proteins for dual-color superresolution fluorescence microscopy.

“I think we are just at the beginning, with these switchable proteins,” says Jakobs. “I would expect that we will see many further developments, more colors and new properties, and these will make applications possible that we haven’t even thought about yet.”

**Natalie de Souza**

## RESEARCH PAPERS

Andresen, M. *et al.* Photoswitchable fluorescent proteins enable monochromatic multilabel imaging and dual color fluorescence nanoscopy. *Nat. Biotechnol.* **26**, 1035–1040 (2008).

Stiel, A.C. *et al.* Generation of monomeric reversibly switchable red fluorescent proteins for far-field fluorescence nanoscopy. *Biophys. J.* **95**, 2989–2997 (2008).

proteomics we cannot completely cover the whole proteome yet,” says Jesper Olsen, second author on the paper.

Daub’s group also observed substantial upregulation of phosphorylation in mitotic cells. Phosphorylation of about 75% of the protein kinases they identified was upregulated more than twofold in M phase, suggesting that cell-cycle progression involves extensive rearrangements of signaling networks. In total, they identified 219 kinases containing 1,182 phosphorylation sites, including several new phosphorylation sites not previously implicated in mitosis. However, Gygi’s study identified 214 kinases containing 897 phosphorylation sites, demonstrating that in-depth coverage could be achieved without applying specific kinase enrichment approaches.

Both groups attribute their success to careful phosphopeptide enrichment strategies, but also to simply starting with a large sample. “The number one obstacle we overcame was just by using enough protein to start out,” says Gygi. “I think that most people know that secret now.” A burning question in the phosphoproteomics field, however, is how can we identify every phosphopeptide, and how do we know when this has been achieved? This is extremely difficult to answer, but Olsen anticipates that advances in mass spectrometry will push the detection limits. He says: “Improved dynamic range and faster sequencing speed—I think that’s really what we need to have better coverage.”

**Allison Doerr**

## RESEARCH PAPERS

Dephore, N. *et al.* A quantitative atlas of mitotic phosphorylation. *Proc. Natl. Acad. Sci. USA* **105**, 10762–10767 (2008).

Daub, H. *et al.* Kinase-selective enrichment enables quantitative phosphoproteomics of the kinome across the cell cycle. *Mol. Cell* **31**, 438–448 (2008).

## PROTEOMICS

## Mapping proteolytic events in apoptosis

Current platforms for studying proteolysis have not generally allowed researchers to obtain a complete picture of the cleaved fragments of protease substrates on a large scale. Dix *et al.* now present an algorithm that integrates data from electrophoresis and quantitative liquid chromatography–tandem mass spectrometry analyses of fragment peptides into a ‘peptograph’, which plots a complete sequence map. They used this method to map caspase-mediated proteolytic events in apoptosis.

Dix, M.M. *et al.* *Cell* **134**, 679–691 (2008).

## BIOINFORMATICS

## An atlas of mouse embryo morphology

Petiet *et al.* describe a high-resolution, magnetic resonance microscopy (MRM)-based atlas of normal, transgenic and mutant mouse model morphology at the embryonic and neonatal stages. The database of three-dimensional MRM images with more than 200 annotated structures is freely available at <http://www.civm.duhs.duke.edu/devatlas/index.html> and should be a useful resource for developmental studies.

Petiet, A.E. *et al.* *Proc. Natl. Acad. Sci. USA* **105**, 12331–12336 (2008).

## GENOMICS

## Quality control in next-generation sequencing

It is a challenge to assemble the millions of short sequence reads generated by next-generation sequencers such as the Applied Biosystems SOLiD and the Illumina Genetic Analyzer. Li *et al.* now present ‘mapping and assembling with quality’ (MAQ), a freely available software tool that uses a mapping quality score to improve alignment accuracy. The tool should be especially useful for detecting single-nucleotide polymorphisms and insertion-deletions.

Li, H. *et al.* *Genome Res.*, published online 19 August 2008.

## IMMUNOCHEMISTRY

## Polyubiquitin linkage-specific antibodies

The fate of a polyubiquitinated protein is thought to depend on the lysine residue of ubiquitin through which the chains are linked. To investigate this question more thoroughly, Newton *et al.* developed polyubiquitin linkage-specific antibodies, which recognize two of the best-characterized linkages, via Lys48 or Lys63. These antibodies can be used in different applications, including immunoprecipitation, immunofluorescence, immunohistochemistry and western blotting.

Newton, K. *et al.* *Cell* **134**, 668–678 (2008).

## BIOINFORMATICS

## A NetPhorest of phosphorylation motifs

Miller *et al.* present NetPhorest, a valuable database of protein phosphorylation consensus sequence motifs that are recognized by protein kinases and phosphorylation-dependent binding domains. This publicly available atlas (<http://netphorest.info/>) is fed by an automated data pipeline: these data are mapped to phylogenetic trees to derive linear motif models.

Miller, M.L. *et al.* *Sci. Signal.* **1**, ra2 (2008).