Cytoplasm and cell motility

Editorial overview

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In taking over the stewardship of this issue of *Current Opinion in Cell Biology* from Marc Kirschner and Klaus Weber, we plan to continue to provide readers with annual overviews of all the most active areas of research on cell motility and the cytoskeleton as well as to cover other topics. Among the topics selected for review this year, we call to your attention several that have not been reviewed previously in this series: mechanical properties of cytoskeletal components (Janmey, pp 4–11), the plant cytoskeleton (Staiger and Lloyd, pp 33–42), intermediate filament protein gene expression (Zehner, pp 67–74), intermediate filament-associated proteins (Foisner and Wiche, pp 75–81), myosin regulation (Sellers, pp 98–104), myosin polymerization (Trybus, pp 105–111), and giant elastic proteins (Trinick, pp 112–119). The reviews are designed to cover five areas: general topics; microtubules; intermediate filaments; actin and myosin; and membrane associations.

We have been particularly pleased to see the emergence of some general principles that apply to two or even all three divisions of the cytoskeleton. Some of these generalizations will be found in the reviews covering general topics and membrane associations. Examples include parallel mechanisms of nucleotide hydrolysis accompanying actin and microtubule polymerization, and the dynamic state of assembly of all three cytoskeleton polymers in live cells.

We will leave the details to the authors of the individual reviews, but would like to highlight here a few of the major events of 1990. As predicted in this issue last year, Kabsch, Holmes and colleagues in Heidelberg have determined the three-dimensional structure of actin at atomic resolution [1], and proposed a model for the structure of the actin filament that is consistent with X-ray fiber diffraction data [2]. Although this work was published late in the year, Marie-France Carlier and Paul Janmey have reviewed some of the implications here. Everyone working on actin is now using this seminal work in the analysis of their own research and we look forward to a review of the impact of this research next year. This work marks the beginning of the analysis of the cytoskeleton and its motors at the atomic level.

The redundancy hypothesis is a major theme that received strong experimental support this year (see Noegel and Schleicher, pp 18–26); and Murphy, pp 43–51). Although there are obvious exceptions such as yeast in general and myosin-II in *Dictyostelium* in particular, most eukaryotic cells appear to have fail-safe cytoskeletons. Not only are there multiple isoforms of many of the proteins such as actins, intermediate filaments, tubulins (see Murphy, pp 43–51), and myosin-Is, but there are frequently groups of molecularly distinct proteins with overlapping functions. It has generally been difficult to assign specific functions to individual isoforms, but this has now been achieved for specialized tubulin isoforms in *Drosophila* and *Aspergillus*. Actin filament crosslinking proteins are a particularly good example covered by Noegel and Schleicher. The family of kinesin variants has also grown impressively (see [3]). The biological sense of this redundancy has not yet become clear. It may simply reflect the lack of sophistication of our assays which do not yet distinguish subtle but important differences in the functions of related proteins. Whatever the reason for these overlapping functions, it is remarkable how healthy a cell can be without one or more of its cytoskeletal proteins.

We are also impressed that the inventory of cytoskeletal proteins continues to grow at a steady rate. In fact, since the first cytoplasmic actin-binding proteins, profilin and macrophage 'actin-binding protein' were discovered in the mid 1970s, new classes of actin-binding proteins have been discovered at a constant rate of two new classes per year. This year, the big surprise has been a totally new class of low-molecular-weight actin-sequestering proteins [4]. It is remarkable that this abundant class of protein has been missed for more than a decade, as it seems to account for much of the unpolymerized actin in non-muscle cells. Even more remarkably, this cytoplasmic protein is related to the thymic hormone, thymosin [5].

The whole field was surprised, not once, but twice, by Vallee and colleagues. First they discovered a new class of microtubule motor that they have called 'dynamin' [6]. Second, they showed that this is the first motor protein of any kind to use GTP rather than ATP as its immediate en-

Abbreviations

IF—intermediate filament; **IFAP**—intermediate filament-associated protein

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ergy source [7]. Another noteworthy event was the publication of evidence for the existence of the microtubule motor, dynein, at the kinetochore (see Reider pp 59–66). The chromosomal motor may have been sighted at last but we are still waiting to see whether dynein actually fills this role.

The number of characterized microtubule-associated proteins has increased steadily, but emerging sequence homologies among proteins that differ considerably in size promise to simplify relationships and make some sense of the evolution of the system (Olmstead, pp 52–58). There has also been a proliferation of proteins that participate in linking actin filaments to membranes and again, just in time, similarities in primary structure have began to emerge, reducing the apparent complexity of the process at the molecular level (Luna, pp 120–126).

Data continue to accumulate suggesting that the various types of intermediate filaments (IFs) and their associated proteins form an interconnecting network which courses from the nucleus to the cell surface. Within the interphase nucleus, the type V IF proteins (the nuclear lamins) form a polymerized structure known as the nuclear lamina. The nuclear lamins have received considerable attention in several areas over the past year or so. These include the determination of the nuclear location signals responsible for the targeting of the nuclear lamin proteins into the nucleus, as well as the molecular basis of their assembly into polymers only at the inner face of the nuclear envelope. The nuclear lamin network appears to be quite stable during interphase, but undergoes an explosive depolymerization preceding mitosis when the nuclear envelope breaks down. It has been known for many years that this disassembly of the nucleus prior to the formation of the mitotic spindle is accompanied by the hyperphosphorylation and subsequent disassembly of the nuclear lamina. Last May it became apparent that this phosphorylation is due to the action of MPF/Cdc2 with the publication of findings from three laboratories [8–10]. Likewise, at the end of mitosis when daughter cell nuclei reform, there is dephosphorylation and reassembly of the nuclear lamins. This can occur on chromosome surfaces in the absence of membranous components (McKeon, pp 82–86).

At the level of the cell surface, IFs have been known to interact with cytoplasmic plaque structures of desmosomes in regions of cell–cell contact and with hemidesmosomes in regions of cell–basal lamina interactions. It is only recently that significant strides have been made towards understanding the molecular basis of these interactions through the sequencing of various desmosome-associated proteins, especially the desmoplakins in which there are domains capable of binding to IF proteins [11]. Interestingly, the initial insights into the plaque components of hemidesmosomes came from autoantibodies derived from patients with bullous pemphigoid, a blistering disease. Even more surprisingly, one of the hemidesmosomal plaque proteins shows extensive homology to the carboxyl terminus in the same region of desmoplakin. Therefore, it is a likely candidate for IF binding to the hemidesmosomal plaque (Jones and Green, pp 127–132).

Numerous other intermediate filament associated proteins (IFAPs) have been described in the literature. The two best characterized of these are filaggrin and a 300 kD protein frequently termed plectin. It has been suggested that the latter protein has a multitude of functions including that of an IF–IF crosslinker and may be capable of network formation on its own. The intriguing finding that there is extensive homology in the carboxy-terminal domains of plectin and human desmoplakins (unpublished results) is discussed in the review by Foisner and Wiche (pp 75–81). The publication of these data in the near future will certainly bring home the message that there are a large number of *bona fide* IFAPs regulating IF networks in cultured cells. Filaggrin remains the most structurally well characterized IFAP. It appears to be involved in the formation of bundles of keratin containing IF (tonofilaments) in keratinocytes. More recently, another skin cell protein, loricrin, has also been shown to possess IFAP-like properties [12]. It appears that many more IFAPs are waiting to be discovered. Undoubtedly these proteins will play critical roles in the regulation of the supramolecular organization of the various types of IF systems.

On the gene regulation front, much is being learned about the regulation of the type I and type II IF proteins, termed the keratins. Over 20 proteins in this category have been described to date. Of the numerous mechanisms thought to be involved in the regulation of simple epithelial cell keratins, methylation has attracted the most interest (see Zehner, pp 67–74). Another interesting finding is related to the transfection of simple epithelial type I keratin into fibroblasts, which subsequently manage to express this keratin. However, without a partner type II keratin, the type I species is rapidly degraded. If the transfected cell contains a type II keratin gene, then co-expression of both is stabilized. This emphasizes the fact that epithelial cells must possess mechanisms for maintaining equal quantities of the type I and type II keratins which form the heterodimers necessary to support IF polymerization. Keratin expression in stratified squamous epithelial tissues is much more complex as different keratin genes are expressed in the different cell layers. This is intriguing as it is totally unclear why keratin-IF composition changes as a function of the state of differentiation. There is a great deal of activity on keratin gene regulation which should lead to new insights into the functional significance of the differential expression of so many IF proteins. With regard to transcriptional control of type III IF proteins, the vimentin gene has been the most extensively studied. To date, it appears that sequences responsible for cell type-specific expression of vimentin are located in the 5′ end of the vimentin gene. These data have resulted in the definition of four regions regulating chicken and human vimentin gene expression (see Zehner, pp 67–74).

Overall, it has been a very exciting year for the three major cytoskeletal systems of eukaryotic cells. We can only guess at what will happen over the next 12 months.

References

1. KABSCH W, MANNHERZ HG, SUCK D, PAI EF, HOLMES KC: **Atomic Structure of Actin: DNase I Complex.** *Nature* 1990, 347:37–44.
2. HOLMES KC, POPP D, GEBHARD W, KABSCH W: **Atomic Model of the Actin Filament.** *Nature* 1990, 347:44–49.
3. VALE RD, GOLDSTEIN LS: **One Motor, Many Tails: an Expanding Repertoire of Force-Generating Enzymes.** *Cell* 1990, 60:883–885.
4. SAFER D, GOLLA R, NACHMIAS VT: **Isolation of a 5-Kilodalton Actin-Sequestering Peptide from Human Blood Platelets.** *Proc Natl Acad Sci USA* 1990, 87:2536–2540.
5. SAFER D, *ET AL*: *J Biol Chem* 1991, in press.
6. SCHPETNER HS, VALLEE RB: **Identification of Dynamin, a Novel Mechanochemical Enzyme that Mediates Interactions Between Microtubules.** *Cell* 1989, 59:421–432.
7. OBAR RA, COLLINS CA, HAMMARBACK JA, SHPETNER HS, VALLEE RB: **Molecular Cloning of the Microtubule-Associated Mechanochemical Enzyme Dynamin Reveals Homology with a New Family of GTP-binding Proteins.** *Nature* 1990, 347:256–261.
8. HEALD R, MCKEON F: **Mutations of Phosphorylation Sites in Lamin A that Prevent Nuclear Lamina Disassembly in Mitosis.** *Cell* 1990, 61:579–589.
9. WARD GE, KIRSCHNER MW: **Identification of Cell Cycle-Regulated Phosphorylated Sites on Nuclear Lamin C.** *Cell* 1990, 61:561–577.
10. PETER M, NAKAGAWA J, DOREE M, LABBE JC, NIGG EA: ***In Vitro* Disassembly of the Nuclear Lamina and M Phase-Specific Phosphorylation of Lamins by cdc2 Kinase.** *Cell* 1990, 61:591–602.
11. GREEN KJ, PARRY DAD, STEINERT PM, VIRATA MLA, WAGNER RM, ANGST BD, NILLES LA: **Structure of the Human Desmoplakins: Implications for Function in the Desmosomal Plaque.** *J Biol Chem* 1990, 265:2603–2612.
12. MEHRAL T, HOHL D, ROTHNAGL JA, LONGLEY MA, BUNDMAN D, CHENG C, LICHTI U, BISHER ME, STEVEN AC, STEINERT PM, YUSPA SH, ROOP DR: **Identification of a Major Keratinocyte Cell Envelope Protein, Loricrin.** *Cell* 1990, 61:1103–1112.

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