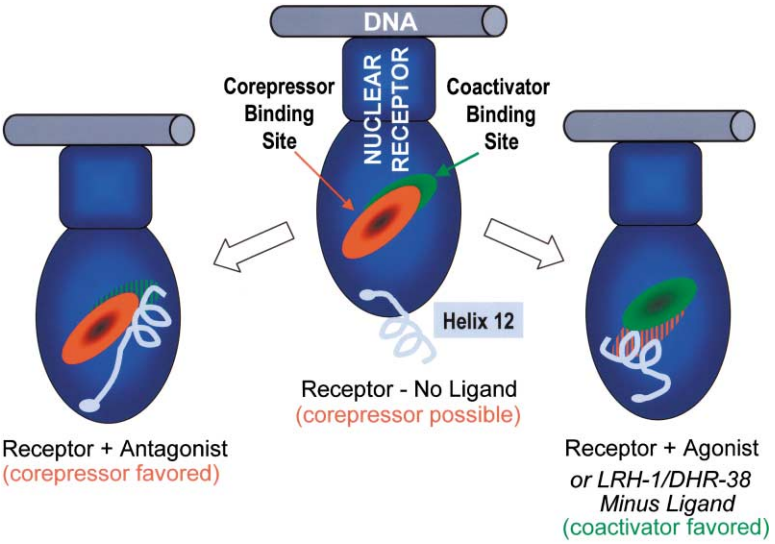

Figure 1. Model of Regulation of Nuclear Receptor Function

A schematic representation of a generic nuclear receptor bound to DNA is presented. Two partially overlapping surfaces within the receptor are highlighted: a corepressor interaction site (red) and a coactivator interaction site (green). Binding of antagonist by the nuclear receptor alters the position of a C-terminal helix 12 (gray), blocking the coactivator interaction site and stabilizing corepressor recruitment (left); binding of agonist has the reverse effects (right). Some nuclear receptors can also recruit corepressor in the absence of any hormone ligand (center). The unliganded LRH-1 and DHR38 orphan receptors mimic the conformation assumed by other receptors in the presence of agonist (right).

example, a nonexchangeable lipid in the hepatic nuclear factor 4-α serves as a hydrophobic core that allows the polypeptide chain to coalesce into an active conformation in the absence of ectopic agonists (Dhe-Paganon et al., 2002; Wisely et al., 2002). A different example is provided by the unliganded retinoic acid receptor β and γ isoforms and by the constitutively activated androstane receptor (CAR), which have been proposed to assume agonist-like conformations as a result of stabilizing contacts between helix 12 and adjacent receptor surfaces (Dussault et al., 2002; Farboud et al., 2003). Conversely, constitutive antagonist-like conformations have been elucidated for the *Ultraspiracle* orphan (Billas et al., 2001; Clayton et al., 2001).

It has been suggested that orphan receptors arose from a ligand-responsive ancestor and only later evolved mechanisms to recruit coactivators in the absence of ligand. However, the studies highlighted here suggest an intriguing alternative: the *Ur* receptor may have been an orphan, much like DHR38 or Nurr1, with nuclear receptors only later acquiring hydrophobic ligands in place of the phenylalanines at the primordial receptor core. Perhaps receptors such as LRH-1 that possess both constitutive activity and ligand binding pockets represent transitional forms in this evolutionary scheme. Whatever their heritage, nuclear receptors continue to provide new insights into metazoan transcription and regulation.

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Selected Reading

Baker, K.D., Shewchuk, L.M., Kozlova, T., Makishima, M., Hassell, A., Wisely, B., Caravella, J.A., Lambert, M.H., Reinking, J.L., Krause, H., et al. (2003). Cell *113*, 731–742.

Billas, I.M., Moulinier, L., Rochel, N., and Moras, D. (2001). J. Biol. Chem. *276*, 7465–7474.

Clayton, G.M., Peak-Chew, S.Y., Evans, R.M., and Schwabe, J.W. (2001). Proc. Natl. Acad. Sci. USA *98*, 1549–1554.

Dhe-Paganon, S., Duda, K., Iwamoto, M., Chi, Y.I., and Shoelson, S.E. (2002). J. Biol. Chem. *277*, 37973–37976.

Dussault, I., Lin, M., Hollister, K., Fan, M., Termini, J., Sherman, M.A., and Forman, B.M. (2002). Mol. Cell. Biol. *22*, 5270–5280.

Farboud, B., Hauksdottir, H., Wu, Y., and Privalsky, M.L. (2003). Mol. Cell. Biol. *23*, 2844–2858.

Glass, C.K., and Rosenfeld, M.G. (2000). Genes Dev. *14*, 121–141.

Sablin, E.P., Krylova, I.N., Fletterick, R.J., and Ingraham, H.A. (2003). Mol. Cell *11*, 1575–1585.

Wisely, G.B., Miller, A.B., Davis, R.G., Thornquest, A.D., Jr., Johnson, R., Spitzer, T., Sefler, A., Shearer, B., Moore, J.T., Willson, T.M., and Williams, S.P. (2002). Structure *10*, 1225–1234.

Wang, Z., Benoit, G., Liu, J., Prasad, S., Aarnisalo, P., Liu, X., Xu, H., Walker, N.P.C., and Perlmann, T. (2003). Nature *423*, 555–560.

The Ancestry of Segmentation

Recent studies of segmentation in the spider suggest that the ancestral vertebrate and arthropod segmentation mechanisms utilized the *Notch* signaling pathway and bolster the argument that segmentation is an ancestral feature of all bilaterians.

The ancestry of animal segmentation has been a long-debated topic. Three of the major animal phyla, arthropods, annelids, and chordates, display an obvious pattern of segmentation. It is unclear, however, whether or not segmentation in these groups is independently derived or the consequence of the derivation of these three groups from a common ancestor that was segmented (Davis and Patel, 1999).

Several lines of evidence suggest that segmentation

evolved independently. For example, our current understanding of animal phylogeny suggests that, if the common ancestor to annelids, arthropods, and vertebrates was segmented, then segmentation must have been lost independently in multiple other animal phyla (such as echinoderms and mollusks). Also, segmentation in arthropods is primarily a property of the ectoderm, while mainly a property of the mesoderm in vertebrates.

In more recent years, the debate has been taken to a molecular level. We have a detailed understanding of the molecular basis of segmentation in *Drosophila melanogaster*, and many researchers have been engaged in examining the expression and potential function of homologs of these genes in other animals. In *Drosophila*, the blastoderm embryo is rapidly subdivided into sequentially smaller and smaller units through the action of maternal gradients and zygotic gap, pair-rule, and segment polarity genes. Many of these genes are well conserved throughout animal evolution and are thought to have been already present in the common ancestor of most animal phyla. But this alone does not argue for a common ancestry of segmentation because we also know that these genes play multiple roles in development and that their ancestral role may not have been in segmentation.

In a few cases, arguments have been made that vertebrate homologs of *Drosophila* segmentation genes are involved in vertebrate segmentation. The most striking example is seen for the *hairy* class of transcription factors. In *Drosophila*, *hairy* is a pair-rule gene that is expressed in a seven-stripe pattern in the blastoderm embryo (thus, in a two-segment periodicity), and loss of *hairy* activity causes a pair-rule pattern of deletions in the larval cuticle. Initial studies suggested that a *hairy* class gene in the zebrafish was also expressed in a pair-rule pattern during the process of segmentation, and this striking similarity was used to argue for a common ancestry of segmentation (Muller et al., 1996; Kimmel, 1996). Subsequent analysis, however, revealed that the zebrafish expression pattern was actually a segmental one and raised the possibility that the involvement of *hairy* in segmentation in flies and fish was independently derived.

Now, however, the approach of comparing arthropod and vertebrate segmentation can be turned on its head. We understand enough about vertebrate segmentation that we can now look for evidence of vertebrate segmentation mechanisms in arthropods. Studies of vertebrate segmentation suggest a so-called clock and wavefront model of segmentation. *FGF* signaling appears to be part of the wavefront (Dubrulle et al., 2001), and vertebrate *hairy* homologs appear to be part of the clock mechanism, showing cyclic patterns of expression in the presomitic mesoderm (Palmeirim et al., 1997). An additional component of the clock mechanism is the *Notch*/*Delta* cell-cell signaling pathway. In various vertebrates, *Notch*, *Delta*, and *fringe* genes show early segmental patterns of expression within somites and cyclical expression and activity in the presomitic mesoderm (Dale et al., 2003). In *Drosophila*, however, *Notch*/*Delta* signaling does not appear to play a role in establishing segmentation, although *Serrate*, another ligand for the Notch receptor, does play a role in setting up some of the fine details of cuticle pattern after segment polarity gene expression has already been initiated. It is important to remember, however, that *Drosophila* segmentation is not necessarily a good representative of general arthropod or even insect segmentation. In *Drosophila*, most of the steps of segmentation occur in the early syncytial environment of blastoderm (and require the syncytium for the diffusion of transcription factors), and all segments appear more or less simultaneously. In most insects and other arthropods, however, segments appear one at a time as the embryo elongates, and these segments are patterned in a cellular environment, suggesting that a cell-cell signaling system might be necessary (Davis and Patel, 1999).

In a recent paper, Stollewerk, Schoppmeier, and Damen (2003) report that the *Notch*/*Delta* pathway is utilized in the segmentation of spiders. For both *Notch* and *Delta* homologs (there are two forms of *Delta* in the spider), they observe a dynamic pattern of expression in the posterior end of the developing spider embryo that includes stripes of a segmental register, a pattern bearing similarity to that seen for these genes in vertebrates. Previous studies had also shown a dynamic pattern of *hairy* expression in the posterior of spiders that also resolved into a pattern of stripes in the developing embryo, although it is argued that this expression is pair-rule (Damen et al., 2000). Using an RNAi approach, they go on to show that disruption of any one of the *Notch* and *Delta* genes causes a defect in segmentation. In all cases, segmentation is not abolished; rather, it becomes disorganized. The segment polarity gene, *engrailed*, continues to be expressed in segmental stripes, but the spacing between stripes becomes irregular, and the normally straight boundaries of each stripe are disrupted. It is possible that the RNAi effect produces only a partial loss-of-function phenotype, but, alternatively, it could be that there is some redundancy in the segmentation system used by this arthropod. It may be that *Notch*/*Delta* in the spiders is not part of any clock mechanism but is instead part of a system to coordinate dynamic transcription factor expression in the growth zone of the spider. It should be remembered that the precise role of the *Notch*/*Delta* system is still the subject of some debate in vertebrates as well (Jiang et al., 2000).

These results clearly support the idea that segmentation may have been ancestral to vertebrates and arthropods and that the common ancestor utilized both *Notch*/*Delta* signaling and repetitive expression of *hairy* class transcription factors. Both these components were retained in the lineage leading to vertebrates. Somewhere in the lineage leading to *Drosophila*, however, the involvement of the *Notch*/*Delta* system in segmentation was lost, but *hairy* expression was retained and the striped expression of other transcription factors was added. Spiders, representing a more phylogentically primitive arthropod, display a retention of the *Notch*/*Delta* system, suggesting that the common ancestor of all arthropods used a segmentation mechanism more like that seen in vertebrates than in *Drosophila*. It is important to note that the use of *Notch*/*Delta* signaling in spider segmentation could still represent an independent cooption of this system. Even within *Drosophila*, *Notch* signaling is used to make other boundaries within the developing legs, eyes, and wings, and, thus, there is some precedence for the cooption of this pathway

into numerous developmental programs requiring the formation of distinct boundaries within a tissue. Furthermore, *Notch* signaling also regulates the expression of *hairy* class genes during *Drosophila* neural development, so the link between these two genes could still predate the evolution of segmentation.

To further support the notion of shared ancestry of segmentation, it will also be important to examine the issue of mesodermal versus ectodermal segmentation. Why are the patterns of *Notch*, *Delta*, and *hairy* mesodermal in vertebrates, but apparently ectodermal in spiders? Even more significantly, data from additional species will be crucial to resolving how the segmentation system has evolved. In addition to chelicerates (spiders) and insects, the other major arthropod groups are crustaceans and myriapods (centipedes and millipedes), and a closer look at all these groups may reveal where in the arthropod lineage the involvement of the *Notch* signaling pathway in segmentation was either lost or gained. Also, arthropods and annelids share a more recent common ancestor than arthropods and vertebrates, so, if segmentation is ancestral, we might expect to find evidence for *Notch* signaling in annelid segmentation and, possibly, remnants of segmentation at the level of *Notch*/*Delta* expression in some of the phyla we consider to be unsegmented. While the issue of the ancestry of segmentation will continue to be debated, these recent results from the spider certainly bolster the argument for a common bilaterian ancestor, nicknamed Urbilateria (DeRobertis, 1997), that was segmented.

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Selected Reading

Dale, J.K., Maroto, M., Dequeant, M.L., Malapert, P., McGrew, M., and Pourquie, O. (2003). Nature *421*, 275–278.

Damen, G.M., Weller, M., and Tautz, D. (2000). Proc. Natl. Acad. Sci. USA *97*, 4515–4519.

Davis, G.K., and Patel, N.H. (1999). Trends Genet. *15*, M68–M72.

De Robertis, E.M. (1997). Nature *387*, 25–26.

Dubrulle, J., McGrew, M.J., and Pourquie, O. (2001). Cell *106*, 219–232.

Jiang, Y.J., Aerne, B.L., Smithers, L., Haddon, C., Ish-Horowicz, D., and Lewis, J. (2000). Nature *408*, 475–479.

Kimmel, C.B. (1996). Trends Genet. *12*, 329–331.

Muller, M., v. Weizsacker, E., and Campos-Ortega, J.A. (1996). Development *122*, 2071–2078.

Palmeirim, I., Henrique, D., Ish-Horowicz, D., and Pourquie, O. (1997). Cell *91*, 639–648.

Stollewerk, A., Schoppmeier, M., and Damen, G.M. (2003). Nature, *423*, 863–865.

Cell Walls, Cell Shape, and Bacterial Actin Homologs

The synthesis of the peptidoglycan layer, one of the key determinants of cell shape in *B. subtilis*, has been shown by Daniel and Errington to occur in a helical pattern. This pattern is generated by the actin homolog MbI.

Bacteria have been model systems in biology for a simple reason: they are far less complicated than eukaryotic cells. Students have been taught for years that among the many differences between bacteria and eukaryotic cells, bacteria lack a nucleus and a cytoskeleton. Ironically, we have been learning over the last few years that many bacteria do indeed have a cytoskeleton, and that it is composed in part of proteins that are true homologs of the key eukaryotic cytoskeletal proteins. Within the past five years, we have seen the demonstration that bacterial FtsZ is a structural homolog of tubulin (Lowe and Amos, 1998), and that bacterial MreB (van den Ent et al., 2001) and ParM (Moller-Jensen et al., 2002) are structural homologs of actin. These observations mark a fundamental turning point in our understanding about the existence of a bacterial cytoskeleton, as well as in our comprehension of the origins of the eukaryotic cytoskeletal system. So it is indeed ironic that our current knowledge of the quite complex eukaryotic cytoskeleton greatly exceeds what we know about the much simpler bacterial one.

It is equally ironic that 20 years ago, studying the eukaryotic cytoskeleton was considered as interesting as, say, studying histones. The interest in the eukaryotic cytoskeleton has grown tremendously, in large part due to the realization that the cytoskeleton is a highly dynamic, and highly regulated, component of cells (Pollard et al., 2000). Similarly, histones, which were once considered to be among the most boring of eukaryotic proteins, have changed roles with the understanding that they are also highly dynamic and regulated (Jenuwein and Allis, 2001).

New insights into the bacterial cytoskeleton come from the work of Daniel and Errington, presented in the June 13 issue of *Cell* (Daniel and Errington, 2003). They explore the synthesis of the external peptidoglycan (PG) cell wall, which is a major determinant of cell morphology in eubacteria. It has been shown that if this meshwork structure of long glycan strands crosslinked by short peptides is removed from a cell, it retains the shape of the cell from which it came, much as an exoskeleton will retain the shape of the invertebrate that generated it. The technical advance that is the basis of their work is the use of a fluorescent reporter to illuminate the sites of PG synthesis in living cells. It has been previously believed that PG synthesis occurred uniformly over the cylindrical surface of *B. subtilis* (Archibald, 1985). With the temporal and spatial resolution provided by the fluorescent technique, Daniel and Errington have been able