selective evolutionary events. The probability that a specific gene will be successfully transferred to a new host depends on the exact mechanism of gene transfer (transformation, transduction, or conjugation) (*13*), on the relationships of these mechanisms to the types of nucleic acids that are being transferred (single-stranded, double-stranded, linear, or circular) (*14*), and even on such factors as the intracellular distribution of integrases, the enzymes that integrate DNA into the genome (*15*). Furthermore, horizontal gene transfer does not seem to be a minor, obscure element of prokaryotic evolution—evidence suggests that it still continues today (*16*).

The transfer of DNA among bacteria plays a major part in continuous prokaryotic horizontal gene transfer. In a recent study, Lawrence and Ochman demonstrated that in the 100 million years following the divergence of *E. coli* from *Salmonella*, 755 genes from many sources have been introduced into the *E. coli* chromosome by more than 230 lateral transfer events and these insertions often occur close to transfer RNA (tRNA) loci (*17*). As lysogenic coliphages (viruses that infect bacteria) are known to insert nucleic acid preferentially at tRNA loci, the *E. coli* insertions are likely to have been mediated by this group of bacteriophages. Moreover, the genome of *Bacillus subtilis*, a well-characterized Gram Positive bacterium, also contains a large number of genes that resemble bacteriophage-like inserts (*18*), lending further credence to the view that the transfer of genes between bacteria by bacteriophages has been an important mechanism of horizontal gene transfer.

Completion of genome sequencing projects for prokaryotes such as *Deinococcus radiodurans* (a radiation-resistant bacterium) and for eukaryotes such as *Drosophila* are sure to spur the dissolution of old paradigms and yield a new wave of revelations about the evolutionary tree of life.

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PERSPECTIVES: SIGNAL TRANSDUCTION

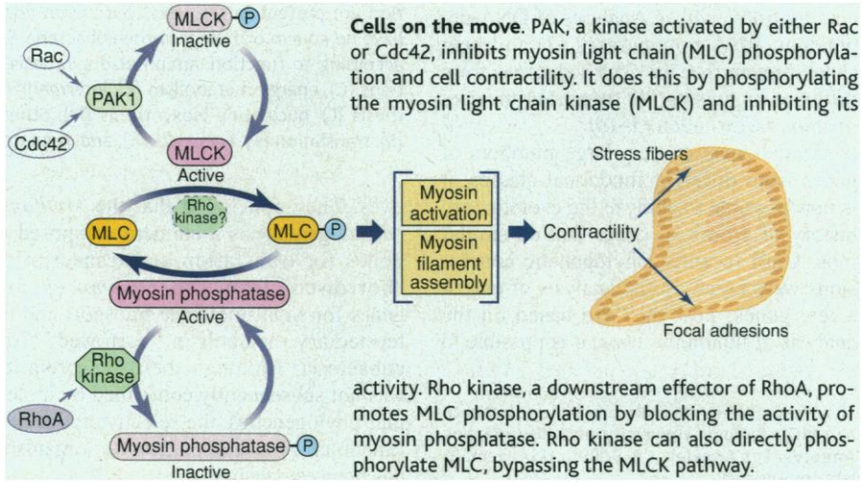
Crosstalk Between Rac and Rho

Keith Burridge

The cytoskeletal changes that alter the adhesion, spreading, and motility of cells depend on a complex interplay among molecules that regulate actin, myosin, and other cytoskeletal components. Members of the Rho family of guanine nucleotide triphosphate (GTP)–binding proteins—including RhoA, Rac, and Cdc42—are important regulators of cytoskeletal organization (*1*). RhoA controls the assembly of stress fibers (bundles of actin and myosin filaments that attach to the plasma membrane at points called focal adhesions), Rac regulates the formation of membrane ruffles, and Cdc42 governs the extension of slender membrane protrusions (filopodia or microspikes) (*1*). Although a hierarchy has been established in which activated Cdc42 stimulates Rac activity and Rac stimulates RhoA activation (*1*), during cell movement these proteins work antagonistically. Rac and Cdc42 promote protrusive events at the leading edge of cells, whereas RhoA induces retraction of the leading edge. Little is known about how Rac or Cdc42 oppose the action of RhoA. On page 2083 of this issue, Sanders *et al.* identify a possible pathway through which Rac may counteract RhoA (*2*). The authors demonstrate that p21-activated kinase (PAK), which is activated by either Rac or Cdc42, blocks the phosphorylation of myosin light chains induced by RhoA. This results in decreased myosin activity, a reduction in contractility, and the disassembly of stress fibers (*3*).

In nonmuscle cells, the activity of myosin II (the myosin found in all cell types that is composed of two heavy chains and four light chains) is regulated by phosphorylation of myosin light chains. This stimulates myosin adenosine triphosphatase activity and induces a conformational change in myosin that promotes its assembly into bipolar filaments. The activated myosin generates tension on the actin filaments and bundles them into stress fibers. Myosin light chain phosphorylation is regulated both by kinases (which add a phosphate group) and phosphatases (which remove a phosphate group).

Historically, most attention has been paid to the myosin light chain kinase (MLCK), an enzyme regulated by calcium and calmodulin. But a new player arrived on the scene with the discovery that Rho kinase (a downstream effector of RhoA) inhibits a myosin phosphatase that removes phosphate groups from myosin light chains and blocks myosin activity (see the figure) (*4*). Thus, a pathway emerged in which RhoA elevates myosin light chain phosphorylation by inhibiting its dephosphorylation. This scheme became even more



Cells on the move. PAK, a kinase activated by either Rac or Cdc42, inhibits myosin light chain (MLC) phosphorylation and cell contractility. It does this by phosphorylating the myosin light chain kinase (MLCK) and inhibiting its activity. Rho kinase, a downstream effector of RhoA, promotes MLC phosphorylation by blocking the activity of myosin phosphatase. Rho kinase can also directly phosphorylate MLC, bypassing the MLCK pathway.

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complex with the finding that Rho kinase could also directly phosphorylate myosin light chains, potentially usurping the role of MLCK (*5*). It is clear that myosin light chain phosphorylation is elevated in vivo in response to RhoA activation, but whether this is due primarily to inhibition of the myosin phosphatase or to direct phosphorylation of light chain, or to a combination of both, has not yet been established.

In previous work, PAK was shown to promote the disassembly of stress fibers and focal adhesions (*5–7*). Sanders *et al.* now demonstrate that MLCK is a substrate for PAK. Phosphorylation of MLCK by PAK decreases its activity, which in turn results in decreased myosin light chain phosphorylation and a decrease in actin-myosin filament assembly (see the figure). Just as elevated myosin activity promotes the assembly of stress fibers, it has been shown that inhibiting actin-myosin interactions with pharmacological reagents causes the disassembly of these structures (*3*). Consequently, PAK's ability to inhibit myosin light chain phosphorylation accounts for the disassembly of stress fibers and focal adhesions observed in cells overexpressing activated PAK.

Most of the kinases stimulated by Rac, Cdc42, or RhoA have multiple targets, and so it is likely that there are additional ways in which PAK opposes or modifies the actions of RhoA. Indeed, the cytoskeletal rearrangements induced by activated PAK are dramatic (*6–8*)—reminiscent of those seen in cells treated with the actin filament–disrupting drug cytochalasin D—suggesting that cytoskeletal proteins as well as MLCK are targets for PAK.

The observations of Sanders *et al.* are important for understanding cell motility. During this complex process, protrusive and contractile forces must be coordinated. Prominent focal adhesions and stress fibers are associated with cells that do not move. Rac and Cdc42 stimulate cell movement, and to be effective these proteins must not only stimulate protrusion, but must also promote disassembly and turnover of focal adhesions and stress fibers. The current work is important because it suggests how contractile forces in the cell can be restrained by Rac and Cdc42, and how stress fibers and focal adhesions may be disassembled through the action of PAK on MLCK.

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Highly Visible, Curiously Intangible

G. A. Clark

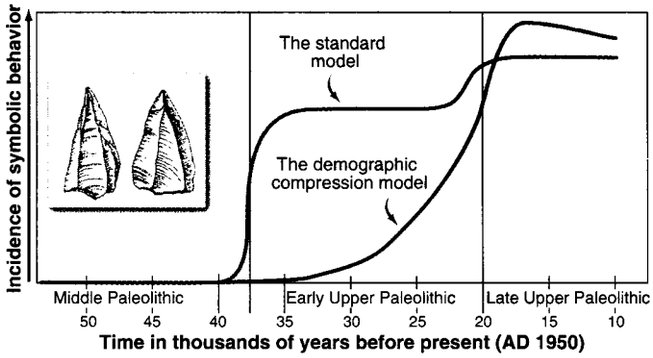
Scientists have been trying to arrive at a consensus about modern human origins (MHO) for more than a century. How is it then that key questions such as whether modern humans evolved only in Africa and migrated from there or evolved in other regions across the world from local archaic ancestors remain unanswered? Many would say that we simply do not have enough data to answer the question of our origins and that with the eventual accumulation of more data, many MHO issues will be resolved. Insufficient data is only part of the answer, however. MHO researchers come from various research traditions. In each of these traditions, different assumptions about the remote human past determine what is considered relevant data, which questions are asked of the data, and how the data are interpreted. More data do not remove the paradigmatic bias implicit within each research tradition, and in consequence people from different fields fail to communicate effectively.

The geneticist Henry Harpending once expressed this problem succinctly when he described MHO research as "a highly visible, yet intangible field" (*1*). The disciplines that contribute to the field (archaeology, human paleontology, and molecular biology) tend to be discovery-driven and focused on methodology. Following a strictly empirical approach ("the facts speak for themselves"), they often have little concern for the logic of inference underlying knowledge claims.

Although these observations apply to all aspects of MHO research, MHO archaeology in Europe is a particularly good example of such epistemological naïvete. Like the larger debate of which it is a part, it can be summarized in terms of two competing models: the continuity model, which contends that modern humans in Europe and elsewhere evolved from their local archaic predecessors, and the replacement model, in which modern humans evolved only in Africa, migrated out of Africa, and replaced other hominids that were the products of earlier, similar radiations (*2, 3*).

Each of these models is based on a set of assumptions that favors some groups of variables at the expense of others, and both define and weight variables thought to be held in common differently (*4*). By making the tenets of the replacement and continuity paradigms explicit, it should in principle be possible to develop tests for their validity—patterns in the archaeological and paleontological records that should hold if in fact the paradigm is an accurate descriptor of reality (*5*). However, such a critically self-conscious approach is often lacking.

According to conventional archaeological systematics, the transition between neandertals and modern humans in Europe coincides with the Middle-Upper Paleolithic transition, 35,000 to 45,000 years ago (see the figure). Although consensus has remained elusive, a dominant "re-



A matter of timing. Comparison of the standard and the demographic compression models for the appearance of symbolic behavior in Europe between 50,000 and 10,000 years before present (yr B.P.). The standard model argues for an "explosion" of evidence for symbolism coincident with the Middle-Upper Paleolithic transition, 40,000 to 35,000 yr B.P. The demographic compression model sees change as much more gradual, with the sharp increase in evidence for symbolism occurring only after 20,000 yr B.P., caused by demographic changes and mainly confined to southern France and northern coastal Spain (*12*).

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