mentation-rate cores from Ocean Drilling Program (ODP) site 983 (*5*) and a global relative paleointensity average spanning 0 to 800,000 years ago (*6*) show low field intensity during geomagnetic excursions (large deviations of the geomagnetic field direction from that due to an axial dipole).

These observations indicate a general connection between directional and intensity variations, and demonstrate the need for simultaneous records of intensity and direction. Guyodot *et al.* (*7*) have suggested that the relative paleointensity at ODP site 983 varies with periodicities characteristic of Earth's orbital eccentricity, obliquity, and precession. However, the robustness and interpretation of these observations remain controversial. Complexities in analyzing these records include the length and sampling interval of the time series, the spectral techniques, and the proxy used in the relative paleointensity estimate. The choice of proxy is critical in determining whether orbital periodicities in the record reflect geomagnetic intensity changes or climatic variations. Stoner *et al.* (*8*) claim that millennial-scale correlations among relative paleointensity records from geographically distant locations are possible, but the high-frequency spectral coherence of such records is uncertain (*9*).

Absolute paleointensity estimates are possible from lava flows, but the measurements are notoriously difficult because of the risk of sample alteration during the experiment. Submarine basaltic glass (SBG) is less prone to such alteration. For the period from 0 to 5 million years ago, new SBG absolute paleointensity data (see the figure) substantially improve lava flow paleointensity data, which are less extensive than their sedimentary counterparts and discontinuous in time. Tauxe and Love (*10*) have reported more than 50 new reliable estimates of paleointensity from SBG, more than doubling the number of similar-quality measurements available from the existing paleointensity database.

The new data are concentrated in the previously undersampled 0.4 to 4.0 million year period. They have led to the assertion that the oft-quoted average dipole moment for the past 0.78 million years is too high because of the preponderance of young (0 to 0.3 million year) data, and that the average field intensity prior to 0.3 million years ago was lower by a factor of ~2. How many data points are needed to define an average remains an open question, given the large geographic (about 20% standard deviation for the present field) and temporal variability in the dipole moment. Thus, despite the superior quality of the new data, their temporal and spatial distribution remain inadequate, and further data are needed to understand the long-term average field intensity.

Advances in understanding Earth's magnetic field behavior require continuing improvements in data distribution, quality, and accessibility. The use of stringent laboratory procedures is critical for mapping regional differences in field behavior and obtaining temporal resolution of a few thousand years. Continuous long-core relative paleointensity measurements and high-quality absolute paleointensity measurements (*11*) have led to a substantial increase in sediment and lava flow data. A promising avenue for future paleointensity work avoids the heating of samples through use of microwaves (*12*).

Until now, limited data sets have led to a somewhat artificial separation of studies of paleodirection and paleointensity. The availability of colocated, contemporaneous records of intensity and direction with better temporal information promote a different approach: that of analyzing the full vector evolution of the geomagnetic field.

Perhaps the most exciting implications of the improved data sets and models are the suggestion of lower mantle influence on the dynamics of the outer core, and the claimed detection of orbital periodicities in geomagnetic records. The arguments in favor of such interactions may be qualitatively appealing but are not yet supported by strong theoretical arguments. Addressing these questions will require improved understanding of geomagnetic field variations and close integration with research in paleoclimate, orbital dynamics, and geophysical studies of deep Earth.

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BIOPHYSICS

Myosin Motors Walk the Walk

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Like its better-known cousin the myosin II of muscle, myosin V is a molecular motor that moves along actin filaments powered by the hydrolysis of ATP. However, unlike muscle myosin, which depends on teamwork for movement, myosin V works alone to move intracellular vesicles around cells. A hotly debated question is whether the two heads of the myosin V motor move along an actin filament in a hand-over-hand manner (akin to human walking), or whether they shuffle along one behind the other like "inchworms." On page 2061 of this issue, Yildiz *et al*. (*1*) report data that are consistent with the "hand-over-hand" model. They used total internal reflection fluorescence light microscopy to track a single fluorophore attached to one of the myosin heads as it moved along an actin filament. They found that the myosin head "swings" through 74 nm for each molecule of ATP hydrolyzed, each time advancing the myosin V by about 37 nm. This discovery provides compelling evidence in favor of the hand-over-hand model.

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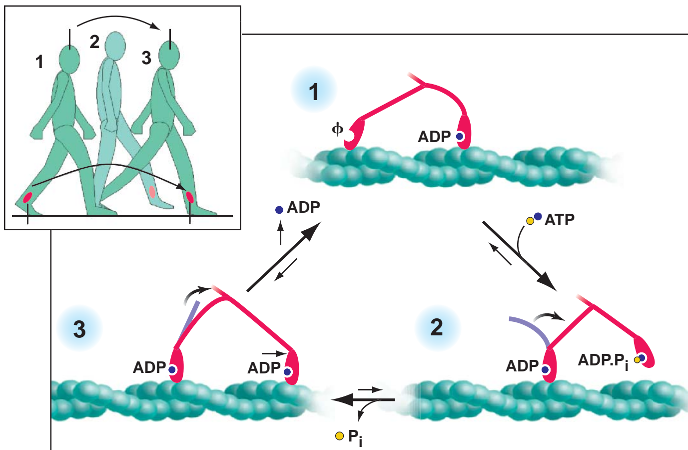
Myosins are a diverse protein family comprising 18 different classes (*2*), of which muscle myosin II is the best characterized. Myosin II works by converting small structural rearrangements at the catalytic site within the motor domain into a large swing or power-stroke of the light-chain binding domain. This serves as a flexible lever arm, transferring force to the object that is being moved. In this model, the presence of nucleotide (ATP or ADP and inorganic phosphate) at the catalytic site is tightly coupled both to the affinity of myosin for actin and to the lever-arm position. The "power-stroke" must occur when myosin is firmly attached to actin, and a "recovery-stroke" when it is detached. If the two heads of a double-headed myosin molecule cycle asynchronously, then they could move along the actin filament processively (that is, in a series of steps) (*3*, *4*). However, for this system to work, at least one of the two myosin heads must be bound to actin at all times. Thus, either the two heads must work in a coordinated fashion or each myosin head must spend most of its cycle time attached to actin (having a high "duty-cycle" ratio).

Biochemical studies have shown that myosin V is a motor with a high duty-cycle ratio (*5*). Furthermore, the light-chain binding domains of myosin V (each carrying six

calmodulin light chains compared to two for muscle myosin II) are of sufficient length to span the F-actin (filamentous actin) helical repeat, enabling its two heads to walk along actin with minimal distortion (*6*). But it took an optical trapping study (*7*) to provide unequivocal evidence that myosin V does indeed move processively along actin filaments in a series of successive 36-nm steps. Because molecular motors work at a very low Reynolds number (*8*) and because biochemical processes are stochastic, they exhibit jerky stepwise movements. Detailed kinetic analyses of single molecules of myosin V have shown that the dwell periods between steps are due to the rate-limiting release of the product ADP (*9*), such that the motor must pause at this point during each cycle of ATP hydrolysis. Further studies using single-headed myosin V molecules that move nonprocessively have shown that the power-stroke is smaller (25 nm) than the step size of the intact molecule (36 nm). Together these data lead to a minimal three-state model (*3*, *10*) (see the figure). A recent study using a doublet of micrometer-sized beads bound to a single myosin V molecule showed that the complex takes a gentle helical path as it moves forward, making one complete turn around the actin filament for each 2 μm advanced (*11*). If we assume a 28/13 helical geometry for F-actin (that is, 167° rotation per monomer) (*12*), this would introduce the observed left-handed bias to the movement.

Notwithstanding the stunning insights provided by such single-molecule mechanical studies, the crucial question of whether myosin V moves by a hand-over-hand or "inchworm" mechanism remains unanswered. Negatively stained electron micrographs (*13*) favor the hand-over-hand mechanism, whereas a study using a mutant myosin V with an artificially shortened light-chain binding domain (*14*) leaves room for other possibilities. The Yildiz *et al.* paper and another recent optical study (*1*, *15*) now provide convincing evidence in favor of the hand-over-hand model. Both studies exploit the high signal-to-noise ratio of total internal reflection fluorescence light microscopy to make measurements from individual fluorophores (*16*) attached to the light-chain region of the myosin V head. The studies provide complementary information about the angular disposition and spatial location of a single myosin head as the intact motor moves along an actin filament. A single fluorophore, attached to two cysteine residues in a myosin V light chain, reported the location and orientation of the light-chain binding domain. Forkey *et al.* (*15*) found that the polarization axis and hence tilt of the light-chain binding domain changed abruptly by 70° for each alternate step taken by the myosin motor. Yildiz *et al.* discovered that the fluorophore moves stepwise by 74 nm, and that this motion recurs for each alternate step taken by the myosin head (see the figure). Both of these studies are remarkable because they extract a wealth of information from just one fluorophore over a relatively long time scale. Polarization ratios were determined within 30 ms (*15*), and the spatial location was measured with 1-nm precision within 500 ms over an observation period of 100 s or more (*1*). The ability to localize an individual fluorophore with a resolution well below that of the light microscope depends on determining the central position of the diffraction-limited spot of collected light (*17*).



The myosin three-step. A three-state model for how the two heads of myosin walk along an actin filament, consuming one molecule of ATP per step. The "power stroke" of the leading head drives the transition from state 1 to state 2 with the hydrolysis of ATP and the production of ADP (blue) and inorganic phosphate (yellow); the trailing head now becomes the leading head. A tendency for the leading head to "fall" forward (by biased diffusion) into state 2 might produce extra movement and complete the step (state 3). The molecule dwells in state 1 at low ATP concentrations (because the empty site, ϕ, requires ATP to bind before it can be released from actin), in state 2 under conditions of high load and in state 3 under physiological conditions. In the hand-over-hand model, each myosin head moves 74 nm per molecule of ATP hydrolyzed. The body of the molecule moves by half that distance (36 nm). The human walking gait is somewhat similar (although the movement is smooth not jerky): Muscles of the leading leg contract to raise the body to state 2, the body then falls forward (under gravity) to state 3. The moving foot travels twice the distance of the body, whereas the other remains fixed to the ground.

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In both studies, the stepping rate of myosin V was slowed artificially by working at very low ATP concentrations, because then ATP binding becomes rate-limiting and the stepping rate is much slower. Furthermore, both studies relied on biochemical kinetic arguments to establish that the head movements occurred once for each alternate step taken by the whole motor. It is unlikely, but nonetheless possible, that myosin V adopts a different stepping pattern at saturating ATP concentrations or when an external load is applied. However, when the results of both studies are combined, they provide compelling evidence for the hand-over-hand mechanism of myosin movement.

Many questions remain to be answered, such as the order and timing of chemical events at the catalytic site and the resulting movement of the light-chain domain and whole molecule. Simultaneously visualizing labeled nucleotide and the position of the light-chain domain with either optical tweezers or a combination of optical probes might help to address this issue. The question of whether myosin V works through a combination of a power-stroke and a thermally driven process could be answered by optical studies performed under very high viscosity conditions.

Details of how myosins drive muscle contraction or move vesicles around the cell will be of great interest to nanotechnologists seeking to build synthetic nanometer-sized motors. Moreover, many of the single-molecule methods devised to probe how molecular motors work hold great promise for broader application in other fields.

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