Volcanic Ash: Terrestrial versus Extraterrestrial

Abstract. *A principal difference between terrestrial and extraterrestrial lavas may consist in the greater ability of terrestrial lavas to form thin films (like those of soap bubbles) and hence foams. It would follow that, in place of the pumice and spiny shards found in terrestrial volcanic ash, an extraterrestrial ash should contain minute spherules. This hypothesis may help to explain lunar microspherules.*

Many terrestrial volcanic deposits contain either pumice (a solidified rock foam) or volcanic shards, which are fragments of pumiceous rock. On the other hand, pumices and the spiny shards derived from them are rare in returned lunar samples. A part of the cause is no doubt connected with the fact that felsic lavas foam more easily than mafic lavas; but this is not the whole story, because there are terrestrial basaltic cinders although the known examples of lunar felsic rocks are not pumiceous or shardy.

To understand why this may be so, let us consider how bubbles are formed. The bubbles that are important here are not merely gas inclusions in a liquid (single-walled bubbles) but are double-walled, like soap bubbles; they consist of a thin film of liquid between two gases.

Double-walled bubbles cannot be formed in pure substances, such as water, because the film at the top of the bubble is being pulled downward under gravity. Surface tension acts equally in all directions and hence does not have any net effect; thus the bubble thins at the top and eventually ruptures.

Bubbles can, however, be made in impure materials such as soapy water. In this case, the primary effect of the soap is to reduce surface tension, which we here think of as surface energy, expressed in joules per square meter. It is then clear that the soap will tend to move into the surface layer, because this process reduces the surface energy. The resulting layer of soap at the surface of the water film has the effect of stabilizing it mechanically, because, when the film is stretched thin, the surface layer is diluted and the surface tension therefore increases. This increase in surface tension pulls the film in the direction of the thin places and resists further thinning. This reasoning explains why bubbles can be blown in soapy water but not in pure water.

Clearly, something of the same kind must happen in terrestrial lavas to permit bubble formation. McBirney and Murase (*1*), quoting measurements by Parikh (*2*), postulated that in terrestrial lavas it is water that reduces the surface tension and so permits foaming. Bikerman (*3*) has noted that low surface tension and volatility are closely correlated both in theory and in fact.

The question then arises whether the very dry and volatile-poor lunar lavas are capable of forming double-walled bubbles. One way to find out is to search for cases where two single-walled bubbles have come into contact. If the single-walled bubbles coalesce, this result suggests that the material cannot support a thin film; but if a thin film—a septum—is observed to survive, separating the bubbles, then the material can evidently support double-walled bubbles. Bell, who has observed bubbles in lunar glasses (*4*), has informed me that he has not thus far observed septa of this kind.

It might be thought that viscosity would promote foaming, and in fact it may do so under special circumstances (*3*, p. 98). But, in general, bubble-wall textures are observed in terrestrial ash of all types, including basalts (of low viscosity) as well as rocks of intermediate and high silica content (and high viscosity) (*5*).

A violent uprush of gas through a liquid that cannot form a foam might very well produce a shower of small liquid droplets, which might cool to glass spherules instead of the pumices, shards, and cinders of terrestrial volcanic eruptions. Perhaps this hypothesis explains why typical volcanic materials are so rarely observed in the lunar samples or in meteorites. Their place may be taken by the microspherules that are observed on the moon. This argument may help us to understand why spherules are so rarely seen in terrestrial volcanic deposits.

I therefore predict that experimental measures will show a marked difference between lunar and terrestrial lavas in the ability to produce foams. When the experiment is done, attention should be paid to Parikh's finding (*2*) that the surface tension of a dry silicate melt is affected by even small quantities of water in the gas with which it is in contact.

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Muscle Crossbridges: Absence of Direct Effect of Calcium on Movement Away from the Thick Filaments

Abstract. *Fluorescence depolarization experiments show that the rotary mobility of myosin heads is hindered by the assembly of the thick filament. Calcium, with or without magnesium adenosine triphosphate, does not alter this hindrance in synthetic filaments. This implies that calcium does not directly move the crossbridges toward thin filaments on activation of muscle.*

It is generally agreed that muscle contraction is caused by the action of ATP (*1*) fueled impellers (crossbridges) operating between two kinds of interdigitating filaments. On neural stimulation, calcium ions are released from the sarcoplasmic reticulum and interact with the contractile apparatus. Some time ago, Ebashi and Endo (*2*) established that a Ca-activated protein "switch" resides on "thin" (actin-containing) filaments of vertebrate muscles. Several workers have since suggested a parallel Ca^{2+}-driven mechanism on the "thick" (myosin-containing) filaments.

The reasons for this current speculation are diverse. Early ideas of crossbridge size and shape (*3*) would require the globular heads of myosin (subfragment 1 or S-1 moieties) to travel radially for some distance in order to contact actin. The mass shift away from the thick filament axis—inferred from changes in the vertebrate muscle x-ray diffraction pattern on activation—seems to occur even when filament overlap is ostensibly zero (*4*). This effect could arise from an actin-independent mechanism on thick filaments. Indeed, isolated molecules from certain invertebrates do seem to have a myosin-based regulatory system (*5*). But this system has not been found in vertebrate skeletal myosins, nor is it clear that such a system could be re-

sponsible for the mass movement between filaments. As shown below, the inferences based on x-ray observations have plausible, experimentally based alternatives. Recently, however, Morimoto and Harrington (*6*) reported evidence from centrifuge and viscosity experiments indicating that changes in $[Ca^{2+}]$ from 10^{-7} to $10^{-5}M$ cause the S-1 moieties of both synthetic and native thick filaments to fan out radially.

For other purposes we developed a method (time-resolved fluorescence anisotropy decay) for detecting whether S-1 moieties are executing rotational Brownian motion (*7*); it occurred to us that the reported Ca^{2+} effects could thus be examined directly and with much higher sensitivity than is afforded by other methods. We report here that, with myosin in either synthetic or native thick filaments, the rotary mobility of the S-1 moiety is strongly inhibited and that (in synthetic filaments) this inhibition is not removed by Ca^{2+} whether Mg^{2+} ATP is present or not. As well foreshadowed by the work of Kaminer and Bell (*8*), *p*H increments in the range 6.5 to 8.3 do markedly alter the observed rotational mobility, and it is possible that the effects of *p*H increments may have sometimes been mistaken for the effects of Ca^{2+} increments.

The principle and practice of our method have been detailed elsewhere (*5*). A fraction of the S-1 moieties is covalently labeled with the fluorophore 1,5-IAEDANS (*1*) and the directional properties of the emission are studied as a function of time. Rotational mobility is expressed by a parameter, ϕ (reported in nanoseconds), the time it would take a perfectly aligned population of protein-bound fluorophores to randomize to $1/e$ of the original alignment as a result of rotational Brownian movement. In this work the attached dye is located almost exclusively on a particular thiol of the S-1 moiety, whether the system under study is a myosin filament or a myofibril. [This specificity is achieved and verified by methods reported elsewhere (*9*).] We had found (*5*) that on passing from dissolved myosin (in 0.6*M* KCl) to myosin suspended in 0.05*M* KCl, ϕ increases from 450 to > 1200 nsec; that is, under conditions conducive to filament formation the rotational mobility of the S-1 moieties is sharply restricted compared to the high "swivel" mobility of S-1 moieties that are members of individual myosin molecules. We report that immobilization occurs in glycerinated myofibrils (which we take here to be native filaments) and in well-prepared synthetic filaments that resemble physiological filaments. It was shown that in high salt the heads could tumble almost 4π steradians. The observed immobilization in low salt presumably occurs because of limitation of the S-1 trajectory by the core of the thick filament. We reasoned that any condition that caused the cross-bridges to fan out from the filaments would tend to restore the S-1 moieties to the free-swinging ($\phi \simeq 450$ nsec) state of individual molecules by increasing the available solid angle.

Freshly prepared myosin labeled with 1,5-IAEDANS (*10*) was used to generate thick filaments according to the method of Morimoto and Harrington (*6*). The Ca^{2+} concentration was set by adding (at least 1 hour before the fluorescence measurements) 1 m*M* EGTA to give $p\text{Ca} \simeq 8.5$ or by adding $10^{-4}M$ $CaCl_2$ to give $p\text{Ca} \simeq 3.97$. These concentrations were determined by using the effective EGTA binding constants of Hellman and Podolsky (*11*) and atomic absorption spectroscopy to measure (intrinsic) Ca concentrations before addition (3 to 10 μM). The addition of $CaCl_2$ or EGTA (set to *p*H 6.8) diluted the filament solution (1 mg/ml) by 1 percent. This method of Ca addition precluded the possibility of small *p*H changes arising from hydrogen ion production by EGTA reacting with large (millimolar) amounts of Ca. In this respect our procedures differ from those of Morimoto and Harrington. Labeled glycerinated myofibrils were prepared by homogenizing labeled rabbit psoas fibers. Fibers were tested for their ability to relax after labeling by measuring their (static) stiffness. The experimental apparatus used has been described (*12*) except that here a longer excitation wavelength (380 nm) was used.

It was found that the polarization decay curves depend strongly on *p*H and

Table 1. Effect of Ca, *p*H, and KCl on S-1 moiety mobility. The *p*H 6.8 solution had a protein content of 1 mg/ml, 90 m*M* KCl, 0.3 m*M* $MgCl_2$, and 40 m*M* imidazole (μ = 0.12). The *p*H 8.3 solution had a protein content of 1 mg/ml, 135 m*M* KCl, and 2 m*M* tris (μ = 0.137). Calcium addition trials spanned five preparations; each half of the difference trial lasted 1 hour. Experiments were done with unchromatographed myosin unless otherwise noted. All experiments were at 5°C. All errors are standard errors of the mean.

Change in conditions	$100\Delta\phi/\phi$ (%)	Trials	Preparations
In *p*H 6.8 solution, *p*Ca 8.5 → *p*Ca 4			
Filaments made from unchromatographed myosin	0.1 ± 2	25	
Filaments made from myosin chromatographed on DEAE-50	−2 ± 3	9	
Filaments made from unchromatographed myosin + 5 m*M* MgATP	3 ± 5	11	
Solution *p*H 8.3 → *p*H 6.8	50 to 70		5
Solution *p*H 8.3 → *p*H 6.5	250		1
In *p*H 6.8 solution, 90 m*M* KCl → 110 m*M* KCl	−15		2
Solution with 0.6*M* KCl (free myosin) to *p*H 6.8 solution	80 to 160		5

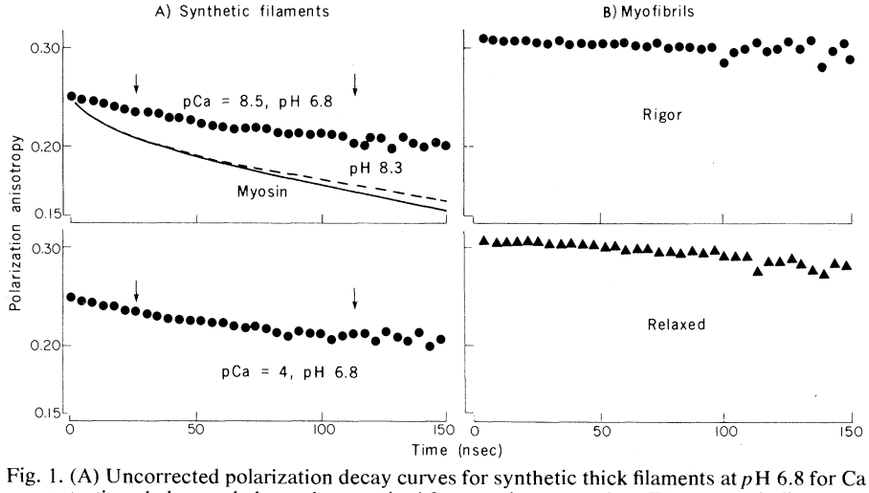

Fig. 1. (A) Uncorrected polarization decay curves for synthetic thick filaments at *p*H 6.8 for Ca concentrations below and above that required for muscle contraction. The arrows indicate the region used to determine ϕ. For reference, curves for myosin (solid line) and thick filaments at *p*H 8.3 (dashed line) are shown (with initial polarization anisotropy normalized to that of *p*H 6.8 filaments). (B) Depolarization of directly labeled myofibrils in rigor and relaxation solutions. These data include first-order corrections for the effect of turbidity. The ratio of ϕ values in rigor and relaxation solutions is unchanged by this correction.

KCl concentration (see Table 1 and Fig. 1). At pH 8.3 and ionic strength (μ) = 0.137, ϕ rose from the free value (in high salt) of 400 nsec to only about 500 nsec, even though electron microscopy and sedimentation indicated that filaments had formed. At pH 6.8 and μ = 0.12, on the other hand, ϕ increased to 750 to 1200 nsec, while at pH 6.5 it was approximately 2000 nsec. In the data reported here no corrections were made for turbidity; experiments at low concentrations showed that turbidity decreases the (low-salt) ϕ value by 10 to 30 percent. Thomas *et al.* (*13*), using saturation transfer electron paramagnetic resonance spectroscopy, also found that the rotational mobility of myosin at low ionic strength decreased with decreasing pH.

A plausible explanation for the difference in mobility with pH is found from the work of Kaminer and Bell (*8*). Their electron microscopic work shows that at higher pH the filaments are shorter and more disordered, with the "rod" attachments to S-1 having significant angular spread as they emerge from the central zone along the filament axis. At lower pH longer and more regular filaments are formed, with the rod portions more nearly parallel. We confirmed these results by electron microscopy and found a mean length of 0.8 μm at pH 6.8. At higher pH this length decreases, although above pH 8 a significant amount of free myosin may be present. Thus, at lower pH values the S-1's are held more closely against the filament backbone with presumably greater steric hindrance, which is consistent with our results showing decreased mobility at lower pH.

Figure 1 shows results of S-1 mobility studies in myofibrils. Only a small difference is seen between rigor (no Ca and no ATP) and relaxing (+ ATP and no Ca) solutions, even ATP causes actin to dissociate from acto– S-1 and actomyosin labeled with 1,5-IAEDANS (*12*). Sodium dodecyl sulfate gels of myofibrils labeled with radioactive 1,5-IAEDANS showed that the myosin/actin labeling ratio was approximately 6 (and that labeling of other proteins was not significant). Thus it appears that in muscle the crossbridges are almost immobile on this time scale (10^{-7} second). Measurements at both 4° and 18°C show that ϕ for relaxed myofibrils is at least four times longer than for free myosin; so we can conclude that in intact muscle the myosin heads are sterically hindered (perhaps by a notch). This agrees with the interpretation of the prominent thick filament "layer lines" seen in x-ray diffraction studies (*14*) in which the filament is considered to have crossbridges projecting radially in a well-defined orientation.

The results of Ca^{2+} difference experiments on synthetic filaments at pH 6.8 are shown in Table 1. No significant effects of Ca addition were found in the presence or absence of Mg^{2+}ATP. Some experiments were done at pH 6.8 with magnesium adenyl imidodiphosphate and others were done under the conditions of Huxley (*15*) (pH 7, 100 m*M* KCl, and 6 m*M* phosphate buffer). Again, no Ca effects were found in either of these cases. Since the mobility is such a sensitive measure of S-1 order (as witnessed by the effect of small changes in pH or KCl concentration), we conclude that there is no significant radial movement of heads from thick filaments when physiologically activating amounts of Ca^{2+} are added (*16*). Tests with the EGTA-Ca system used by Morimoto and Harrington showed a drop in pH of about 0.13 unit when millimolar Ca was added to 1 m*M* EGTA. Subtle pH effects or changes other than S-1 moving away from the filament backbone could be responsible for the changes in sedimentation coefficient and viscosity seen by Morimoto and Harrington.

If our conclusion is correct, there is no Ca^{2+}-driven "micromuscle" radially fanning out the crossbridges from the thick filaments. Then there must be alternative explanations for the observations that other workers have taken as indicative of Ca^{2+} effects on the thick filament. First, the S-1 moiety is not spherical but elongate, with an axial ratio in the range 2.8 to 3.5 (*7, 17*); thought of as a prolate ellipsoid, S-1 has to have a length of at least 130 Å—that is, in relaxation, at rest length, its distal tip is not more than about 40 Å from actin so that only a small amount of radial Brownian motion against compliance in the rod portion of myosin is necessary. The transition to the rigor state could then be accomplished by the crossbridges changing their angle of azimuthal tilt (around the fiber axis) as well as changing their declination on actin. Recently it has been shown (*18*) that this scheme could readily furnish the mass transfer required to account for changes in equatorial x-ray patterns. After the crossbridge power stroke the elasticity of the rod portion would draw the crossbridge near the thick filament and in so doing position it above a new actin binding site (in shortening muscle). Finally, the discrepancies between earlier fiber work of Haselgrove (*4*) (whole muscle) and of Nihei *et al.* (*19*) (glycerinated single fiber) might reside in pH effects, or in nonuniformities in sarcomere lengths of stretched fibrils in the former case. The work reported here agrees with the results and conclusions of Nihei *et al.*

In summary, although we have searched for thick filament–based Ca^{2+} effects by a method ideally suited to detect them, we have not found them, and we believe that the previous indications of such effects have alternate explanations.

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References and Notes

1. The following abbreviations are used: ATP, adenosine triphosphate; 1,5-IAEDANS, *N*-(iodoacetylaminoethyl)-5-naphthylamine-1-sulfonic acid; EGTA, ethylene glycol bis(β-aminoethyl ether)-*N,N,N',N'*-tetraacetic acid.
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