The Mass-Independent Fractionation of Oxygen: A Novel Isotope Effect and Its Possible Cosmochemical Implications

Abstract. *Experimental evidence is presented which demonstrates a chemically produced, mass-independent isotopic fractionation of oxygen. The effect is thought to result from self-shielding by the major isotopic species* $^{16}O_2$*, but other possible mechanisms such as molecular symmetry cannot be ruled out. In a three-isotope plot, the experimentally produced fractionation line is essentially equal in slope to the observed carbonaceous chondrite mixing line. The implications for the early history of the solar system are discussed.*

The abundances of stable isotopes are partitioned (fractionated) as a result of three major processes: (i) kinetic processes, (ii) isotopic exchange reactions, and (iii) physicochemical effects such as diffusion, condensation, and evaporation. The theoretical basis for estimating the magnitude of fractionation for each of these processes differs, but the underlying principle for each is that the effects are all dependent on mass. An observation of a mass-independent isotopic fractionation of oxygen in the high-temperature minerals in carbonaceous chondritic (C2 and C3) meteorites was made by Clayton *et al.* (*1*). They demonstrated that the oxygen isotopic composition of carbonaceous chondrites, when plotted on a three-isotope coordinate system [$^{17}O/^{16}O$ versus $^{18}O/^{16}O$, expressed in the conventional delta notation $\delta^{17}O$ versus $\delta^{18}O$ (*1, 2*)], produces a line with a slope near unity, whereas a similar plot of the oxygen isotopic composition of terrestrial or lunar materials produces a mass-dependent fractionation line with slope near 1/2 (0.52). This was interpreted as indicating the admixture to normal oxygen of nearly pure ^{16}O which predated the solar system and which presumably was the result of a nuclear event such as occur in supernovae. Mass-independent isotopic fractionations in other meteoritic elements have been reported (*3*) and are generally attributed to nuclear processes. The possibility, however, that chemical effects may be responsible for the observed fractionations has been suggested, in the face of considerable controversy, by Arrhenius [see (*4*)].

We report here a demonstration of a chemically produced mass-independent isotopic fractionation. The apparatus employed is shown in Fig. 1. Ozone was generated by electrical discharge in molecular oxygen and cryogenically removed after its formation (*5*). Following the reaction we mass spectrometrically analyzed both the product ozone and unreacted molecular oxygen for the oxygen isotopic composition ($\delta^{18}O$ and $\delta^{17}O$) (*6*). For convenience, we report our starting oxygen as having $\delta^{17}O = \delta^{18}O = 0$ so that isotopic partitioning is easily visualized. Regardless of normalization, mass-dependent processes produce fractionations with $\delta^{18}O = 2\delta^{17}O$ (*1*).

The results of our experiments obtained on two different mass spectrometers are listed in Table 1 and shown in Fig. 2 (*7*). The slope of the line is 1.00 (± 0.01) with a correlation coefficient of .999.

In explaining our data, we considered several mass-independent isotopic fractionation mechanisms that have been suggested. (i) Curve crossing of predissociative states with quantum mechanical tunneling from the bound to free repulsive states (*4*) may lead to mass-independent fractionations; however, a curve-crossing geometry that produces an isotopic fractionation line with slope 1 is unlikely. Predissociative effects may not strictly be ruled out, but the observation of equal dissociative rates for the heteronuclear species makes this mechanism less compelling. (ii) Nuclear spin effects may be ruled out since they preferentially effect the isotope that has nuclear spin, in this case ^{17}O ($I = 5/2$), which would not result in a fractionation with slope 1 (*8*). We think the most consistent explanation requires the mass-independent fractionation to occur in the primary step, the dissociation of molecular oxygen. We feel that the mass-independent fractionation is a result of optical shielding by the major isotopic species $^{16}O^{16}O$, which is more abundant than $^{16}O^{18}O$ and $^{16}O^{17}O$ by factors of 245 and 1348, respectively. Selective removal of the well-known $^{16}O_2$ lines in the Schumann-Runge region, which are shifted from the heteronuclear lines, results in preferential dissociation of $^{17}O^{16}O$ and $^{18}O^{16}O$ with subsequent formation of isotopically heavy ozone (with respect to the starting molecular oxygen), as we observe. A similar shielding effect has been demonstrated with selective laser photolysis of the $v' = 4$ band of O_2 (*9*). The dissociation of the heteronuclear species would be isotopically nonselective, resulting in ozone production which scales with the natural abundances of ^{17}O and ^{18}O. This results in ozone enriched in ^{17}O and ^{18}O and with slope 1 in a plot of $\delta^{17}O$ versus $\delta^{18}O$. We may not unequivocally state whether shielding occurs on the electrons from the tesla coil or from ultraviolet radiation generated inside the vessel. Future work with different radiation sources will make it possible to quantitatively resolve the origin of the effect. A shielding effect is a potentially efficient means of isotopic enrichment, particularly for deuterium, ^{235}U, ^{15}N, or any isotopic species with a small natural abundance (< 1 percent). Photoproduction of isotopically heavy ozone in the stratosphere as a result of optical shielding has also been suggested (*10*).

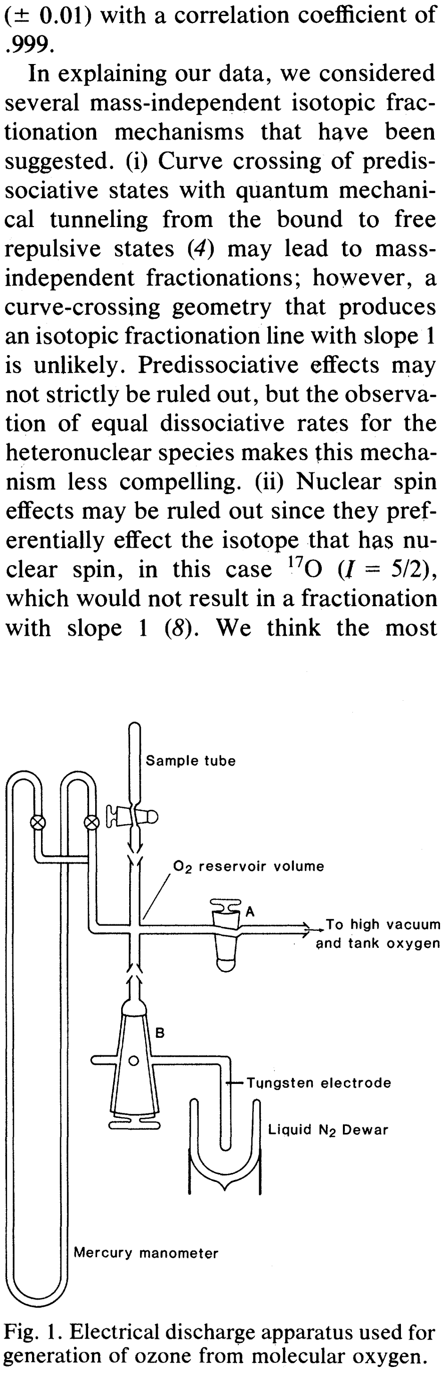

Fig. 1. Electrical discharge apparatus used for generation of ozone from molecular oxygen.

A second, albeit less likely, explanation for our results is that the slope 1 plot results from the equal, and greater (than homonuclear), dissociation rates of the heteronuclear species due to the appearance of alternate rotational lines in the Schumann-Runge band system. The alternate rotational lines, superimposed on the vibrational states, appear for $^{16}O^{17}O$ and $^{16}O^{18}O$ and are absent for $^{16}O_2$ (*11*). Given the nature of our experiments, we cannot rule out this suggestion.

We have demonstrated that a mass-independent isotopic fractionation of oxygen may be chemically produced which strikingly resembles that observed in carbonaceous chondrites. This effect indicates that identification of nuclear events by mass-independent isotopic variations may not be made, at least in the case of oxygen. It is of interest to explore the possible role of this process in the presolar nebula. Large variations have been observed in the abundance ratios of CO isotopes in the surface layers of molecular clouds due to isotope-selective photodestruction of CO (*12*). This isotopically selective process is a result of self-shielding such as apparently occurs in our experiments. At present, we are not certain of the chemical speciation of oxygen in the presolar nebula. However, shielding requires only that oxygen be contained in a gas-phase molecule such as CO, SiO, OH, H_2O, or O_2, all of which are components of molecular clouds (*13*). If molecular symmetry is responsible for the isotopic fractionation, then oxygen must be contained in O_2 or a symmetric molecule such as SiO_2, TiO_2, or CO_2. In light of the present uncertainties about speciation, we can only state that the close resemblance of our isotopic fractionation to that observed in carbonaceous chondrites strongly suggests the importance of photochemical processes in the presolar nebula. Future work on isotopic fractionation of other molecules will allow quantitative calculations on the importance of photochemical processes.

Regarding potential dissociative energy sources, young T-Tauri stars, which are considered precursors to low-mass stars, are commonly associated with molecular clouds (*14*). Many of the T-Tauri stars have been studied with the International Ultraviolet Explorer (IUE) and have been found to emit more ultraviolet radiation than the present sun (*14*, *15*). It has been suggested that at an age of the sun of less than 10^6 years, ultraviolet radiation in the Schumann-Runge band region was enhanced by factors of at least 10^4 (*14*). At present, we cannot be certain whether or when an enhanced ultraviolet source was available, but astrophysical observation suggests that it is at least plausible. In addition, external ultraviolet sources may also be of importance (*16*).

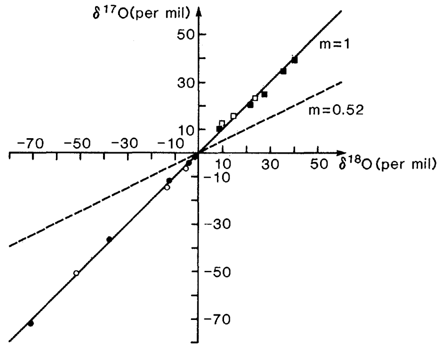

Fig. 2. Experimental data on isotopic composition. The squares represent ozone samples and the circles molecular oxygen reservoirs. Filled symbols represent measurements at the University of California, San Diego, and open symbols represent measurements from the University of Chicago.

An outstanding problem with the existing models for oxygen isotopic anomalies is that an admixture of pure ^{16}O into a nebular reservoir, isotopically like terrestrial oxygen, explains only the C2-C3 mixing line. In fact, most meteorites lie above the terrestrial line. Regardless of what one assumes, at least two components are required to account for the observed meteoritic oxygen isotopic distributions. If, however, a photochemical mechanism were operative on a reservoir which isotopically resembles the intersection of the C2-C3 line with the terrestrial one, two reservoirs would be produced, lying on different sides of the terrestrial line. In addition, the C2-C3 fractionation line would be produced. Although we may not be certain that the reservoir was isotopically like terrestrial oxygen, our suggestion requires only one component, which may be partitioned into the various meteoritic classes without resorting to nuclear processes. By mass balance arguments, the photochemical model cannot be ruled out since the large ^{16}O enrichments contained in the relatively small C2-C3 reservoir are counterbalanced by the relatively small ^{16}O depletions observed in the large H, L, and LL chondrite reservoir. Finally, as first discussed by Clayton *et al.* (*1*), admixture of as much as 5 percent pure ^{16}O from a nuclear event should be accompanied by isotopic effects in elements such as magnesium and silicon. After nearly a decade, no correlations have been observed between oxygen isotopic anomalies and anomalies in other elements, with the exception of three unusual inclusions (*17*). Even though they have been looked for (*3*), no nuclear anomalies have been reported for silicon (except for very small effects in inclusions C1 and EK1-4-1). The lack of a correlation is a major problem in accounting for the suggested nuclear processes. If a photochemical effect similar to ours were responsible, no anomalies would be expected in silicon or magnesium since they have large isotopic abundances of the minor isotopes and no diatomic precursors. The observed deuterium and ^{15}N enrichments in the carbonaceous chondrites (*18*) may be another hint of the presence of a similarly produced photochemical fractionation, since both species have diatomic progenitors as well as large isotopic differences.

In summary, we have shown that a chemically produced, mass-independent fractionation of oxygen is possible. Since the effect essentially duplicates the isotopic distribution in carbonaceous chondritic meteorites, we suggest that photochemical processes may have been of importance in the presolar nebula. If this is the case, it establishes a firm link between meteoritic analysis and astrophysical observation.

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Table 1. Experimental results of discharge experiments. The extent of reaction is determined as 100 × (initial pressure minus final pressure)/(initial pressure). Errors for the isotopic measurements are reported at the head of the individual column.

Extent of reaction (%)	Reservoir isotopic composition (per mil)		Ozone isotopic composition (per mil)	
	$\delta^{18}O$	$\delta^{17}O$	$\delta^{18}O$	$\delta^{17}O$
6.51	−1.79 ± 0.10	−1.47 ± 0.29	27.38 ± 0.10	24.91 ± 0.29
12.06	−4.26	−4.11	35.25	34.82
26.01	−12.37	−11.80	39.96	39.03
36.13	−5.31*	−6.50*	9.58*	12.68*
50.40	−13.25*	−14.78*	14.34*	15.74*
66.31	−37.56	−36.41	21.44	20.39
70.38	−51.55*	−50.73*	23.16*	23.03*
89.44	−70.73	−71.90	8.37	10.16

*Analyzed at the University of Chicago.

References and Notes

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5. The reaction system is filled with lecture-bottle oxygen through stopcock B to the desired pressure and shut off. The reaction vessel is immersed in liquid nitrogen to a level 1 cm below the tungsten electrode. The reaction is initiated when a tesla coil is brought in contact with the electrode and halted when the coil is removed. Reaction extent is followed manometrically.
6. The unreacted molecular oxygen is removed by pipetting an aliquot into the sample tube while the ozone is condensed. After the excess oxygen has been pumped away, the liquid nitrogen trap is removed and the ozone transferred to a molecular sieve 13X trap at liquid nitrogen temperature. Warming the trap to 300°C catalytically converts the ozone to molecular oxygen, which is transferred to a sample tube for mass spectrometric analysis. Isotopic measurements were performed on a dual-collector, isotope ratio mass spectrometer (Nier type) with a 6-

inch radius of curvature. The $\delta^{18}O$ and $\delta^{17}O$ measurements were corrected for tailing, valve leak rate, and background (*2*).
7. As an analytical check, a sample set was isotopically analyzed at the laboratory of R. N. Clayton at the University of Chicago. Figure 1 shows that the measurements at Chicago lie along the slope 1 line defined at La Jolla.
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19. We are grateful to R. N. Clayton and T. K. Mayeda for mass spectrometric analysis of the samples. We are particularly indebted to G. Lugmair, who offered valuable advice throughout the project and who patiently read and corrected several manuscripts. Thanks to T. Vander Wood and J. H. Reynolds for their scholarly comments. Support for the project was provided to M.H.T. from the Department of Chemistry, University of California, San Diego.

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Hysteresis in the Force-Calcium Relation in Muscle

Abstract. *Calcium ions activate muscle contraction. The mechanism depends on the calcium sensitivity of the proteins that regulate contraction. Evidence is presented for the reverse phenomenon, where contraction modulates calcium sensitivity. Increasing the force level increased calcium sensitivity in intact fibers, showing that the relation between force and calcium is not unique. A particular calcium concentration can maintain a higher force level than it can create. The results were confirmed in skinned fiber experiments. Transient reduction of the force led to a transient reduction in calcium binding, suggesting a simple mechanism for the hysteresis.*

It is now well established that muscle contraction is activated through the binding of calcium to calcium-sensitive myofibrillar (*1*) or sarcoplasmic proteins (*2*). The details vary among muscle types, but in all cases calcium binding allows actin and myosin to interact and leads eventually to the production of force and shortening through cross-bridge interaction. In this report we examine the question of whether contraction influences calcium sensitivity and calcium binding.

We used aequorin-injected voltage-clamped barnacle single muscle fibers (*3*) to investigate the "quasi-steady state" relations between calcium concentration and force level. In an experiment with a long stimulation pulse (note the slow horizontal sweep rate in Fig. 1A) a constant depolarization leads to a calcium transient that goes through an initial peak and then settles down to a fairly steady level maintained for several seconds. Force behaves similarly. Near the end of the stimulation (marked by arrows) we can measure a quasi-steady-state relation between calcium concentration and force. If the depolarizing pulse is initially stepped briefly to a higher level and then returned to the same level as before, we obtain the result shown in Fig. 1B. Here, despite the initial overshoot, both the membrane depolarization and the calcium transient eventually (see arrow) arrive at levels indistinguishable from those achieved in Fig. 1A, but the force trace does not. It remains at more than twice its previous value. Thus the relation between calcium concentration and force is not unique, but depends on the immediate history of the muscle. This is clear in Fig. 1C, which shows the force-calcium relation after a 10-second depolarization with three different force wave forms produced by a manual control of the stimulation pulse: 1, initial overshoot returning to a steady level (as in Fig. 1B); 2, rapid rise (2 seconds) to a steady level; or 3, slow rise (4 seconds) to a steady level. In each case, the "steady" calcium level produces more force when the fiber previously experienced a higher force (or calcium) level; that is, the steady force for time course 1 > 2 > 3. An obvious interpretation is that increased force and increased cross-bridge interaction occurring early in the contraction heightened the calcium sensitivity. In other words, in this new condition a much higher force can be maintained with a lower calcium concentration (*4*).

Our interpretation of the experiment illustrated in Fig. 1, A to C, can be criticized in several ways. For example, there might be a long-term change in the intracellular magnesium concentration during the pulse which in turn could change the calcium sensitivity of the myofilaments or the sensitivity of the aequorin. Or there could be adenosine triphosphate (ATP) depletion, or a change in *p*H. To explore these possibilities, we turned to the "skinned" fiber preparation (*5*), where there is complete control over the solutions bathing the myofilaments.

To prepare a barnacle muscle skinned fiber (*6*), we first soak an intact fiber for 1 hour in a relaxing solution (*7*) with a detergent (1 percent Triton X-100) added to disrupt the sarcolemma and the sarcoplasmic reticulum. The fibers are next split longitudinally down to bundles of myofibrils of about 50 to 100 μm in diameter (1 to 1.5 mm long) to reduce the diffusion path lengths. The bundle is then attached to a force transducer (*7*) and immersed (stepped) into solutions (*8*) that can induce contraction or relaxation depending on the calcium concentration. These solutions (*5*, *9*) are well buffered against changes in *p*H, in calcium concentration, and in Mg-ATP concentration, which obviates the criticisms mentioned above. Figure 2A illustrates the results of transferring the fiber consecutively from a relaxing solution, to a submaximal calcium activating solution, to a maximal one, and then back to the same submaximal calcium activating solution before returning to a relaxing solution. It can be seen that the same (submaximal) calcium concentration produces more force when the calcium concentration is stepped down (decreasing Ca) from a maximum activation than when it is stepped up (increasing Ca) from the relaxing solution. This difference in force appears to represent a true hysteresis, since leaving the fiber in the submaximal calcium activating solutions for a longer period during either stepping up or stepping down in calcium does not result in the two forces tending to approach one another. If the solution used between the two identical submaximal calcium activating solutions is not maximal, the hysteresis becomes less as the intervening calcium activation is lower. Also, once the fiber relaxes completely in the relaxing solution, the contraction series shown in Fig. 2A can be repeated with the same results, showing that a few minutes of relaxation wipes out the "memory" of the maximal contraction. Data similar to those in Fig. 2A can be obtained for a variety of submaximal calcium concentrations, so that we can plot the relation between calcium concentration and force for the two condi-