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## **Two-Million-Year Record of Deuterium Depletion in**

## **Great Basin Ground Waters**

Abstract. Fluid inclusions in uranium series-dated calcitic veins from the southern Great Basin record a reduction of 40 per mil in the deuterium content of groundwater recharge during the Pleistocene. This variation is tentatively attributed to major uplift of the Sierra Nevada Range and the Transverse Ranges during this epoch with attendant increasing orographic depletion of deuterium from inlandbound Pacific storms.

The mineralogy and fossil assemblages of lake beds currently provide the only tool for deciphering pre-Mid-Wisconsin paleoclimatic conditions in the Great Basin. Calcitic veins, which mark the sites of paleoground-water flow (1) within a major modern regional flow system, provide a supplementary record of paleoclimatologic and paleohydrologic events extending into the Pliocene. The calcitic veins (Fig. 1) occur in near vertical fractures in Pliocene and Pleistocene sediments at Ash Meadows, Nevada, and Furnace Creek Wash, California (Fig. 2). The veins are dense, vary from a millimeter to a meter in thickness, locally occur in swarms, and are commonly finely laminated parallel to fracture walls. They can be traced vertically for tens of meters and horizontally over hundreds of meters. Paleoclimatologic findings derived from fluid inclusions in these veins are presented here.

Veins lining opposing walls of open fractures were selected, out of hundreds of vein-sealed fractures, for both uranium-series dating and deuterium (D) analyses. Each vein was divided into groups of laminae representing the youngest (free face) to oldest (adjacent to fracture wall) portions of the vein (2). The dates of laminae of vein DH2 were calculated from <sup>230</sup>Th/<sup>234</sup>U activity ratios; dates of the older veins were calculated from  $^{234}$ U/ $^{238}$ U ratios (3–5). The veins range in age from about 64,000 years to 1.7 million years. Extrapolation of growth rates indicates that veins 10A and 10B may be as old as 2.6 million years.

We determined the deuterium content of fluid inclusions by using the extractive procedure of Harmon *et al.* and modifications thereof (6). Samples were heated under vacuum for 12 to 14 hours at  $150^{\circ}$ C to remove sorbed water and then were crushed under vacuum in stainless steel tubes. Liberated water was extracted at 200°C and converted, using uranium, into hydrogen for D/H analyses. The deuterium content is expressed in parts per thousand difference (per mil) relative to standard mean ocean water (SMOW) [normalized to the V-SMOW/SLAP scale (7)]. The  $\delta$ D values are plotted against age in Fig. 2.

Fig. 1. Calcitic vein swarm, Furnace Creek Wash, Death Valley, California. Veins, which mark the routes of paleoground-water flow, occur in fanglomerate of the Pliocene Funeral Formation (17). A lowangle fault (upper third of the photo) offsets the near-vertical uranium series-dated veins at this location. Relief is on the order of 50 m at the center line of the photo.

The  $\delta D$  of fluid inclusions of late Pliocene age (laminae 10B-5 through 10B-7 and 10A-5 through 10A-8) average about -60 per mil, or about 40 per mil heavier than that of modern recharge (-99 per)mil) and late (?) Wisconsin age (5) ground water (-102 per mil) discharging today from the major artesian springs in the region (Fig. 2). More significantly, the  $\delta D$  values, in general, become progressively lighter with decreasing age over the last 1 to 2 million years. Do these values represent deuterium of Pliocene and Pleistocene ground water trapped in the veins during the calcite precipitation, or are they artifacts of (i) water-mineral exchange, (ii) remobilization of fluid inclusions during calcite recrystallization, or (iii) selective diffusion of  ${}^{1}\text{H}_{2}\text{O}$  from the fluid inclusions? We cannot attribute the changes in deuterium to water-mineral exchange because the water-bearing fractures in the regional carbonate aquifer, feeding the modern (and fossil) flow system, are typically coated with calcite or dolomite (8). This coating precludes the exchange of hydrogen between water and clay minerals during flow from recharge to discharge areas. In fact, the difference in



 $\delta D$  between modern recharge and late (?) Wisconsin (5) discharge waters is only 3 per mil (Fig. 2). With regard to recrystallization, individual crystals within laminae may be as large as 0.5 cm or more, a feature commonly associated with recrystallization processes. However, petrographic examination of the samples has shown the following textural features to be common: (i) increasing crystal size from bottom to top within any one lamination, (ii) essentially planar crystal boundaries, and (iii) essentially no poikilotopic or porphyrotopic textures. These three features suggest an origin by primary crystallization into an open space (the fracture) rather than as a result of recrystallization. (Laminae 10A-1/2 and 10B-2 may be an exception; we cannot rule out the possibility that some of the fluid inclusions in these laminae are secondary.) Possible enrichment of the deuterium content of the fluid inclusions, by the preferential outward diffusion of <sup>1</sup>H<sub>2</sub>O over HDO after uplift of some veins above the water table hundreds of thousands of years ago, is not supported by the distribution of  $\delta D$  in the veins or by the very dense nature of the vein calcite. For example, if diffusion were a factor, we would expect the youngest laminae of vein 10B-a vein that occurs hundreds of meters above the modern water table-to be the most enriched in deuterium, with the oldest layers, as much as 10 cm inward from the free face, to be the most depleted. We observe the opposite. We are unable to find any reason why the  $\delta D$  values of the fluid inclusions should be significantly different from that of late Pliocene and Pleistocene ground-water recharge.

Let us consider possible causes for the decrease in  $\delta D$  of southern Great Basin ground water (40 per mil) since the latest Pliocene, as recorded in the calcitic veins. We cannot attribute the overall decrease to astronomically driven (that is, Milankovitch) cycles because it is predominantly an undirectional change.

Some of the "reversals" in our data, for example, between laminae 10A-3 and 10A-1/2 (or between laminae 10B-3 and 10B-2), might indeed be related to climatic cycles. However, the limited accuracy of our ages, the large range in our  $\delta D$  measurements for individual laminae (Fig. 2), and the possibility that laminae 10A-1/2 and 10B-2 contain inclusions of secondary origin preclude meaningful commentary on possible reversals at this time. An undirectional process acting over a period of 2 million years appears necessary to explain the gross variations we see. We suspect that the depletion of deuterium with decreasing age of our veins reflects gradual uplift of the Sierra Nevada Range and the Transverse Ranges during the Pleistocene. This uplift, which amounted to no less than hundreds of meters (see below), would have progressively depleted the moisture and deuterium content of winter-spring Pacific storms entering the southern Great Basin from the northwest, west,



Fig. 2. Deuterium content of fluid inclusions in Pliocene and Pleistocene calcitic veins, southern Great Basin, Nevada and California;  $\blacktriangle$ , calcitic vein; the vertical bar is the range, unless too small to show, and the triangle marks the average of the  $\delta D$  measurements; the number in parentheses is the number of analyses for vein laminae; the horizontal bar is the error in the indicated age, unless too small to show;  $\square$ , Holocene flowstone from Trout Springs cave in the Spring Mountains recharge area;  $\bigcirc$ ,  $\delta D$  of modern ground water from Trout Springs and Cold Creek Spring in the Spring Mountains, a major recharge area;  $\bigcirc$ ,  $\delta D$  of late (?) Wisconsin age ground water emerging from major springs in the Ash Meadows and east-central Death Valley regional discharge areas. The  $\delta D$  values for spring waters are from (5, 10). Data for the veins are available (2). The  $\delta D$  values for laminae 10A-1/2 and 10B-2 are tentative; these laminae may contain fluid inclusions of secondary origin; m.y., million years.

and southwest (9). The fluid inclusions in the calcitic veins presumably record this progressive depletion of deuterium in Pleistocene winter and spring recharge (10)

The observed depletion in deuterium is consistent with the estimated late Cenozoic uplift of these ranges and measured deuterium-altitude lapse rates. Geologic evidence indicates that the central Sierras have been uplifted on the order of 600 m in the past 2 million years (11), while the Transverse Ranges apparently have been rapidly uplifted to their present height (2000 to over 3000 m) in the last 1 million years (11). The major orographic effects of the Sierra Nevada on the deuterium content of modern precipitation is well documented (12, 13). If we use deuterium-altitude lapse rates of 25 to 50 per mil per 1000 m (for the 1000to 3000-m range) presented by Smith et al. (13), a 600-m rise of the Sierras during the Pleistocene would be expected to have progressively depleted the deuterium content of storms crossing this range by 15 to 30 per mil. We have emphasized the uplift of the Sierra Nevada and Transverse Ranges because they are the major orographic barriers bounding the southern Great Basin. However, we are aware that portions of the Coast Ranges were also uplifted hundreds of meters during the Pleistocene (11) and suggest that such uplift would also have contributed to the hypothesized progressive depletion of moisture and deuterium from inland-bound Pacific storms during this epoch.

Other partial explanations for the deuterium depletion during the Pleistocene are possible. For example, Pacific storms entering California today are fed by moisture transported by polar, modified polar, and tropical air (14). If these moisture sources have (and had) significantly different isotopic contents, then a systematic variation in their relative contributions to Great Basin recharge during the Pleistocene might account for the variations we see. Alternatively, if summer precipitation during the late Pliocene and early Pleistocene was considerably greater than modern amounts (about 30 percent of annual), then perhaps such deuterium-enriched rain (10), unlike modern summer precipitation, contributed to recharge (10). The difficulty with both alternate hypotheses is that they require a mechanism to explain the progressive depletion of deuterium during the Pleistocene rather than cyclic variations of this isotope expectable from alternating glacial and interglacial climates.

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Further speculation on the causes of the deuterium depletion, as well as on the apparent absence of a depletion during the late Pliocene to early Pleistocene (Fig. 2), must await refinement (15) of the crude record of  $\delta D$  versus time presented here as well as study of the isotope meteorology of the various storm types reaching southern and central California and their modification enroute to the Great Basin. Whatever its cause, the 40 per mil depletion in  $\delta D$  is several times larger than that suggested (16) to have occurred during the entire Tertiary period in the western United States. To the degree that the deuterium depletion reflects Sierra Nevada-Transverse Ranges uplift, it provides a proxy record of increasing aridity in the southern Great Basin during the Pleistocene Epoch.

ISAAC J. WINOGRAD

U.S. Geological Survey, Reston, Virginia 22092

BARNEY J. SZABO

Branch of Isotope Geology, U.S. Geological Survey, Denver Federal Center, Denver, Colorado 80225 TYLER B. COPLEN

U.S. Geological Survey Reston, Virginia 22092

ALAN C. RIGGS

Zoology Department, University of Washington, Seattle 98105

PETER T. KOLESAR Department of Geology,

Utah State University, Logan 84322

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- he sample preparation and analytical methods that we used for uranium and thorium dating were those described by B. J. Szabo, W. J. Carr, and W. C. Gottschall [U.S. Geol. Surv. Openle Rep. 81-119 (1981)]. The activity ratios of Th/<sup>232</sup>Th are greater than 20, within the limits of error, indicating the absence of initial detrital <sup>230</sup>Th contamination. A major assumption in our assignment of approximate  $^{234}U/^{238}U$  dates to the vein calcite is that the initial  $^{234}U/^{238}U$  of the ground water that precipitated the calcite was similar to that in water now issuing from the major artesian springs, namely,  $2.76 \pm 0.09$  for seven Ash Meadows springs and  $2.41 \pm 0.22$  for three major springs in the Furnace Creek-east central Death Valley region. We believe this assumption is reasonable for the purposes of this study for several reasons. First, the artesian springs and the dated veins occur in the dis-charge area of a vast ground-water basin; the Ash Meadows subbasin has a catchment area of no less than  $12,000 \text{ km}^2$  (5). Unlike the case for recharge areas where temporal and geographic variations in water chemistry and isotopic ratios are common (4), in discharge areas relatively constant chemistry is expected and has been recorded at Ash Meadows, albeit over a period of only 50 years (5). During the last 2 million years, neither the mineralogy of the Cenozoic and Paleozoic aquifers traversed by the ground water nor the location of the principal recharge areas is likely to have changed significantly. The water chemistry and the  $^{254}U/^{28}U$  ratios in the discharge area should have remained relatively constant during this time frame. Second, the

superimposed laminae, in general, have  $^{234}U/$  ratios that usually decrease (that is, approach secular equilibrium) with increasing rela-tive age (2) determined from field relations. Third, the assumption of relatively constant initial <sup>234</sup>U/<sup>238</sup>U in these ground-water systems was ascertained from calculations of the initial  $^{236}$ U/ $^{238}$ U values from the measured  $^{230}$ Th/ $^{234}$ U ages for laminae of vein DH2 (2). Between about 64,000 and 250,000 years ago, the calculated initial ratios range between 2.53 and 2.85. The average initial uranium ratio in vein DH2  $(2.67 \pm 0.11)$  is concordant (within the limits of (2.07  $\pm$  0.11) is concordant (within the limits of experimental errors) with the values of the ura-nium ratio measured in modern major artesian springs. We used an initial value of 2.67  $\pm$  0.22 to calculate the <sup>234</sup>U/<sup>238</sup>U ages of laminae of the older veins (2). Actually even errors of a few hundred thousand years would not change the finding of a 40 per mil decrease in  $\delta D$  since the early Pleistocene.

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- 10. snow, snowmelt, rain, and ground water in the Spring Mountains (Fig. 2) indicate the absence of modern recharge from deuterium-enriched summer precipitation; winter-spring precipita-tion, 45 percent lighter in deuterium than summer rains, comprises the principal source of recharge [I. J. Winograd and A. C. Riggs, Geol. Soc. Am. Abstr. Programs 16 (No. 6), 698 (1984)
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15. We are continuing efforts to improve the precision of deuterium measurements of fluid inclusions, beginning to examine the <sup>18</sup>O content of the vein calcite, and expanding petrologic studies in an attempt to understand the nature of the vein laminae.

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## Isolation, Experimental Transmission, and Characterization of Causative Agent of Potomac Horse Fever

Abstract. Potomac horse fever, a disease characterized by fever, anorexia, leukopenia, and occasional diarrhea, is fatal in approximately 30 percent of affected animals. The seasonal occurrence of the disease (June to October) and evidence of antibodies to the rickettsia Ehrlichia sennetsu in the serum of convalescing horses suggested that a related rickettsia might be the causative agent. Such an agent was isolated in cultured blood monocytes from an experimentally infected pony. This intracytoplasmic organism was adapted to growth in primary cultures of canine blood monocytes. A healthy pony inoculated with these infected monocytes also developed the disease. The organism was reisolated from this animal which, at autopsy, had pathological manifestations typical of Potomac horse fever. Cross serologic reactions between the newly isolated agent and antisera to 15 rickettsiae revealed that it is related to certain members of the genus Ehrlichia, particularly to Ehrlichia sennetsu. Since the disease occurs in other parts of the United States as well as in the vicinity of the Potomac River, and since it has also been reported in Europe, the name equine monocytic ehrlichiosis is proposed as being more descriptive.

Potomac horse fever, a disease characterized by fever, anorexia, leukopenia, and occasional diarrhea, is fatal in approximately 30 percent of affected animals. The disease was first reported in the vicinity of the Potomac River in the state of Maryland in 1979 (1). Since then, the disease has become well established in the eastern part of the United States and has been observed sporadically in

Table 1. Serological cross reactivities among the Potomac horse fever agent (PHFA), *Ehrlichia sennetsu, E. canis*, and *E. equi* as determined by the IFA test. N, no reaction at a serum dilution of 1:10.

| Source of serum                                | Sam-<br>ples<br>(No.) | Average titer |             |          |         |
|--|-----------------------|---------------|-------------|----------|---------|
|  |                       | PHFA          | E. sennetsu | E. canis | E. equi |
| PHF (equine)                                   | 5                     | 1:160         | 1:20        | N        | N       |
| experimental infection                         | 2                     | 1:320         | 1:40        | 1:10     | Ν       |
|  | 4                     | 1:1280        | 1:80        | 1:10     | Ν       |
|  | 6                     | 1:2560        | 1:160       | 1:10     | Ν       |
|  | 2                     | 1:2560        | 1:320       | 1:20     | Ν       |
| PHF (equine) natural                           | 8                     | 1:160         | 1:20        | Ν        | Ν       |
| infection                                      | 6                     | 1:640         | 1:40        | 1:10     | Ν       |
|  | 3                     | 1:1280        | 1:80        | 1:20     | Ν       |
|  | 2                     | 1:2560        | 1:160       | 1:10     | Ν       |
|  | 3                     | 1:2560        | 1:320       | 1:20     | Ν       |
|  | 2                     | 1:5120        | 1:640       | 1:20     | Ν       |
| E. sennetsu (canine)<br>experimental infection | 3                     | 1:40          | 1:640       | 1:80     | 1:10    |
| E. canis (canine)<br>experimental infection    | 5                     | 1:10          | 1:80        | 1:640    | 1:80    |
| E. equi (equine)                               |                       |               |             |          |         |
| experimental infection                         | 3                     | Ν             | 1:20        | 1:80     | 1:640   |
| Normal horse serum                             | 10                    | Ν             | Ν           | Ν        | Ν       |
| Normal canine serum                            | 10                    | Ν             | Ν           | Ν        | Ν       |

other parts of the country and abroad (2).

Over the last 5 years, many state and government laboratories have been investigating the cause of the disease. A number of viruses and bacterial agents and their by-products have been demonstrated in the blood and tissues of affected animals, but none of these have been proved to be etiologically related to the disease (3).

This study was prompted by three research leads regarding the possible nature of the causative agent. First, the disease was found to have a seasonal occurrence, with most cases being observed from June through October. Second, it was demonstrated that the infection could be experimentally transmitted by blood transfusion from acutely infected to susceptible horses (4). Finally, studies in our laboratory revealed antibodies in the serum of convalescing animals that reacted with Ehrlichia sennetsu in the indirect fluorescent antibody (IFA) test (5). Ehrlichia sennetsu is the causative agent of human sennetsu rickettsiosis, a disease that clinically resembles infectious mononucleosis and is prevalent in Japan and other regions of Southeast Asia (6). The most distinguishing characteristic of members of the genus Ehrlichia is their development within a membrane-lined cytoplasmic vacuole of leucocytes. The prototype for this genus, E. canis, is the etiologic agent of canine ehrlichiosis, a worldwide and frequently fatal disease of dogs. On the basis of these findings we directed our efforts toward isolation of the apparent causative rickettsia from the blood of experimentally infected horses and ponies.

A small volume (50 ml) of whole citrated blood from an experimentally infected animal during the acute stage of the disease was inoculated intravenously into a susceptible pony (7). The inoculated animal was monitored daily for evidence of clinical and hematological abnormalities. The animal developed a fever (104°F; 40°C) on day 13 after inoculation that was followed by anorexia, general depression, and diarrhea. Beginning on day 0, blood monocyte cultures were prepared from the inoculated animal at 72-hour intervals according to a method developed in our laboratory for the isolation of E. canis (8). The cells were propagated in 25-cm<sup>2</sup> tissue culture flasks with Medium 199 supplemented with 1 percent L-glutamine and 20 percent heat-inactivated normal horse serum. The cultures were maintained at 37°C in air, and were monitored by the