

on a finite difference grid being used (9). The results gave atmospheric temperatures over the winter polar cap region averaging near 140°K and surface wind speeds in that region on the order of 10 meters per second or less; in fact, a polar CO₂ ice cap of about the size of the observed cap did form in the experiment.

The cold polar atmospheric temperatures in the experiment can be explained qualitatively. The atmosphere of Mars is a very efficient radiator, so that any perturbations in temperature from local radiative equilibrium are rapidly damped. Hence, for a given intensity of circulation relatively little heat will be transported (compared with Earth, for example). The result is that horizontal heat transfer into the polar cap region is rather inefficient. The numerical model did not incorporate the possibility of ice cloud formation. However, if ice clouds did form over the polar cap region, more energy would be lost due to upward emission by the clouds at a temperature warmer than the ground, so that more horizontal heat transport would be required to maintain a given air temperature

than would be required without clouds; at the same time, there is no obvious way in which ice cloud formation could bring about the required increase in circulation intensity.

On this basis I conclude that CO₂ condensation in the Mars polar caps is quite likely. However, the deposition rate may be strongly dependent on the formation of extremely tenuous water ice clouds in the winter polar regions.

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6 September 1966

Electrophoretic Variation of Galactose-1-Phosphate Uridyltransferase

Abstract. *A specific method for starch-gel electrophoresis of galactose-1-phosphate uridyltransferase has been developed. Electrophoresis of red-cell hemolyzate from normal subjects and subjects homozygous for the Duarte variant has shown that the Duarte variant has a slightly faster electrophoretic mobility than the normal enzyme under the various conditions used. Molecular-weight estimation on Sephadex G-200 indicates that this observed difference in electrophoretic mobility of the Duarte variant is not due to difference in molecular size. Both enzymes have a molecular weight of approximately 85,000.*

Galactosemia is a recessively inherited error of metabolism characterized by virtual absence of the activity of the enzyme, galactose-1-phosphate uridyltransferase (UDP glucose: α -D-galactose-1-phosphate uridyltransferase, E.C. 2.7.7.12). The red blood cells of heterozygotes for galactosemia have approximately one-half the normal amount of this transferase (1). Recently, however, we have presented genetic evidence that some individuals with approximately one-half normal red-cell transferase activity are actually not heterozygous for galactosemia. Instead, such individuals were homozygous for another gene, the Duarte variant. This gene is much more prevalent than the gene for galactosemia, reaching a fre-

quency of approximately 0.055 in the general population (2). Identification of individuals homozygous for the Duarte variant was based entirely upon the pattern of transmission of quantitative deficiency of the red-cell transferase activity within a family. Biochemical investigation revealed no abnormality in the active site of the enzyme; the pH optimum, thermal stability, and affinity of the enzyme for both of its substrates appeared to be normal (3). We have now developed a sensitive technique for the accurate localization of transferase activity on starch gels and show that the transferase of the Duarte variant has a different electrophoretic mobility in appropriate systems than that of transferase from the

red cells of normal individuals or individuals heterozygous for galactosemia.

Electrophoresis of hemolyzates prepared from saline-washed red cells that have been frozen and thawed and of partially purified enzyme (3) was carried out with the vertical starch-gel method (4). Gels containing 13, 16, and 20 percent starch in 0.005 to 0.010M potassium phosphate buffer, pH 7.0, were used. The gels were chilled for 2 to 3 hours before application of the samples. The buffer compartments contained 0.1 to 0.5M potassium phosphate buffer, pH 7.0. Electrophoresis was carried out for 16 hours at 4°C at a gradient of 4 volt/cm. The transferase was localized on the gel by the use of a reaction mixture comprised of the following: 1.42 mM uridine diphosphoglucose (UDPG); 5.65 mM galactose-1-phosphate (Gal-1-P); 12.4 mM cysteine; 1.2 mM triphosphopyridine nucleotide (TPN); 8 mM MgCl₂; 91.0 mM tris-acetate buffer, pH 8.0; 1 unit of glucose-6-phosphate dehydrogenase (G-6-PD) per milliliter; 0.8 unit of phosphoglucomutase (PGM) per milliliter; and 0.005 unit of 6-phosphogluconic dehydrogenase (6-PGD) per milliliter.

The position of transferase was ascertained by examining the gel under long-wave ultraviolet light. In the presence of the transferase, UDPG and Gal-1-P react to form glucose-1-phosphate, which is converted to glucose-6-phosphate by PGM. In the presence of G-6-PD and 6-PGD, glucose-6-phosphate is oxidized and triphosphopyridine nucleotide (TPN) is reduced. The reduced TPN can readily be observed on the gel because of its fluorescence in ultraviolet light. The specificity of the reaction was confirmed as follows: (i) "galactosemic" hemolyzates resulted in the appearance of no fluorescent bands, and (ii) no fluorescence occurred when either UDPG or Gal-1-P was omitted from the reaction mixture.

The enzyme in the red cells of homozygotes for the Duarte variant invariably had more rapid electrophoretic mobility than the transferase from normal erythrocytes (Fig. 1). This was true in all 26 experiments with enzyme from the two available homozygotes. The separation of the two types of enzyme was approximately the same at pH values of 8.0, 7.5, 7.0, and 6.5. Incorporation of 1.0M urea into the gel did not influence enzyme activity or the relative mobility of the two enzymes. The separation of the two

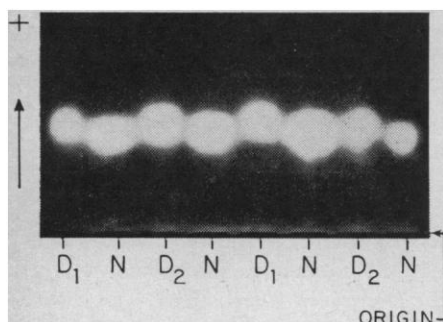


Fig. 1. Starch-gel electrophoretic patterns of galactose-1-phosphate uridylyltransferase from a normal subject and two homozygotes from the Duarte variant. The gel was prepared in 0.01M phosphate buffer, with 0.25M phosphate buffer in the buffer compartments. Each gel was photographed under ultraviolet light after application of the reaction mixture. The slots labeled N were filled with hemolyzate from cells of normal individuals. Those labeled D₁ and D₂ were filled with hemolyzate from the cells of the MO propositus and the BA proposita as described (2). Each preparation was diluted so that the total enzyme activity in each slot was the same, approximately equal to a 1:8 dilution of normal hemolyzate.

enzymes could be demonstrated either when the hemolyzates were diluted to the same enzyme activity (Fig. 1), or when diluted to the same protein concentration. An artificial mixture of normal and Duarte variant enzyme resulted in a broad band, the leading edge of which coincided with that of the Duarte variant and the trailing edge with that of the normal enzyme. Because of the relatively small quantity of mutant enzyme activity in heterozygotes, it is technically quite difficult to demonstrate clearly the presence of the variant in their hemolyzates. However, on careful examination such hemolyzates showed, in addition to normal enzyme, relatively faint fluorescence in a region just ahead of the normal band, corresponding to the Duarte variant.

To determine whether the difference in electrophoretic mobility between the two types of enzyme was due to differences in molecular size, electrophoresis was carried out not only in 16 percent, but also in 13 and 20 percent starch gel. It has been shown that retardation of larger molecules is greater when higher starch concentrations are used (5). The relative migration of the two enzymes was the same at all starch concentrations studied. The molecular weight of the two enzymes, partially purified on DEAE (diethylaminoethyl cellulose) (3), was also

estimated by gel filtration on Sephadex G-200. The material on the column was eluted with 0.15M sodium acetate solution containing 7 mM dithioerythritol and 0.27 mM EDTA (ethylenediaminetetraacetate). Alcohol dehydrogenase (molecular weight, 150,000, Boehringer) from yeast, serum albumin (molecular weight, 68,000, Calbiochem), horseradish peroxidase (molecular weight, 40,000, Sigma), and cytochrome c (molecular weight, 12,000, Nutritional Biochemical Corporation), served as markers. Transferase in effluent from the column was estimated by a modification of the method of Maxwell *et al.* (3, 6). Under these circumstances 95 to 100 percent of enzyme was recovered. The elution pattern was highly reproducible; the molecular weight of normal enzyme was estimated in two filtration experiments at 87,000 and 83,500 and that of Duarte variant enzyme at 81,500 and 85,000.

Our studies indicate that individuals homozygous for the Duarte variant produce a structurally different galactose-1-phosphate uridylyltransferase from that produced by normal individuals. Since the molecular weight of the normal enzyme and the Duarte variant enzyme appear to be approximately the same, the difference in migration in starch gel would appear to be due

to differences in molecular charge or axial ratio. The charge difference appears much more likely, especially since changing the concentration of starch failed to influence the degree of separation of the two enzymes. Presumably the charge difference is due to an amino acid substitution. Since the kinetic properties of the enzyme do not appear to be abnormal, it is reasonable to suppose that the substitution does not affect the active site of the enzyme, but may well influence its rate of synthesis.

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7. Investigation supported in part by NIH grants HE-07449 and HD-01974. Blood samples from one homozygote for the Duarte variant were made available to us by S. Wright and R. Sparkes.

19 September 1966

Magnesium-28 in Rain: Produced by Cosmic Rays

Abstract. Existence of magnesium-28 (half-life, 21.3 hours) produced by cosmic rays in rain at concentrations of 1.7 and 6.1×10^{-1} atoms per milliliter was established radiochemically by isolating this nuclide from several hundred liters of rain samples collected at Fayetteville, Arkansas.

The following radionuclides, with indicated half-lives, produced by cosmic rays were known to occur in the atmosphere, or in atmospheric precipitation, or both, about 10 years ago (1): 2.7-million-year Be¹⁰, 5600-year C¹⁴, 12.5-year H³, 2.6-year Na²², 87-day S³⁵, 53-day Be⁷, 25-day P³³, 14-day P³², and 1-hour Cl³⁹. These radionuclides have since been added to the list of nuclides produced by cosmic rays: 700-year Si³² (2), 15.0-hour Na²⁴ (3), 37-minute Cl³⁸, and 2.8-hour S³⁸ (4).

Many nuclides with masses ranging from 25 to 31 are also expected to be produced by the cosmic-ray spallation reactions on atmospheric argon, but most of them are short-lived. Among these nuclides, 21.3-hour Mg²⁸ and 2.62-hour Si³¹ have half-lives long

enough to enable us to measure their concentrations in rain by the use of present-day, low-level counting and radiochemical techniques. In this work we have measured the concentrations of Mg²⁸ in rain by isolating this nuclide from large volumes of samples collected at Fayetteville (94°W, 36°N), Arkansas.

Samples were collected on the roof

Table 1. Magnesium-28 in rain at Fayetteville (94°W, 36°N), Arkansas.

Date (1966)	Rain-fall (mm)	Sample size (liter)	Conc. of Mg ²⁸ (10 ⁻¹ atom/ml)
16 July	31.5	685	1.7 ± 0.2
20 July	9.1	245	6.1 ± 0.7