

are strongly absorbed. The removal of these two radiations in this manner is not unique to cerium earth minerals, for most yellow, orange, or red minerals would do likewise. The yellow radiation (578 m μ), however, lies well within the strong absorption band of neodymium. This radiation emerges greatly weakened from a thin section or a grain of a cerium earth mineral, whereas the green radiation (546 m μ) is relatively little affected, because it lies in an adjacent spectral region of relatively high transmittance. The net result is that cerium earth minerals assume the emerald color of the unabsorbed component, whereas other minerals, more or less, retain the color that they have under white light. Non-opaque, nonmetamict cerium earth minerals are the only ones known to us that have the requisite absorption properties to produce this effect, so the test appears to be highly specific. The samples are observed directly with the naked eye without recourse to a spectroscope, thereby greatly simplifying the task of recognizing cerium earth minerals in mineral concentrates.

The principle of the method should be applicable to the identification of any mineral that has a strong, narrow band in its absorption spectrum. A light source emitting only two wavelengths of visible radiation would be devised, one wavelength being selected to lie within the absorption band and the other to lie nearby on either side of the band in a region of low absorption. The mineral would assume the color of the unabsorbed radiation.

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Hemoglobin J

Previous reports have defined nine abnormal human hemoglobins that differ in their physicochemical properties, and several summaries of the literature to date are available (1, 2). Two of the abnormal hemoglobins described, H (3) and I (4) have been found to have a higher electrophoretic mobility at a pH of 8.6 than normal adult hemoglobin (5). The subject of this report is a third such hemoglobin that migrates more rapidly than normal adult hemoglobin at

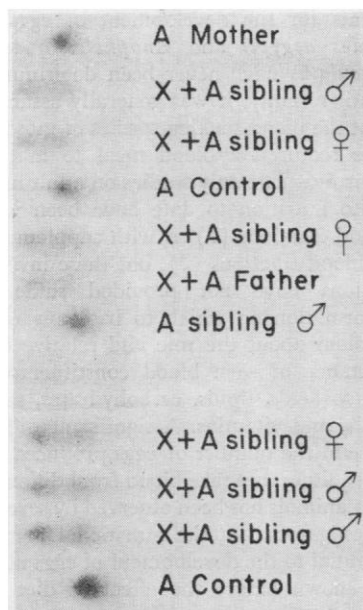


Fig. 1. Comparison by filter-paper electrophoresis of hemoglobin solutions from members of the family under study with normal control in Veronal buffer of pH 8.6 and approximate ionic strength of 0.025. Migration is to the left.

pH 8.6 but has electrophoretic properties differing from hemoglobins H and I. We propose that this hemoglobin be assigned the letter J for purposes of identification (6).

This hemoglobin was found in a young Negro female with bilateral cystosarcoma phylloides. It was first identified by paper electrophoresis using the apparatus described by Smith and Conley (7) with Veronal buffer at pH 8.6 and approximate ionic strength of 0.025 (Fig. 1). Blood from 13 other members of the family has been examined, and the abnormality has been found in seven (Fig. 2). The father and six of nine siblings, including the propositus, had this component in addition to normal adult hemoglobin. The presence of previously described abnormal hemoglobins could not be detected. A search for the presence of the sickling phenomenon and fetal hemoglobin was unrewarding. The scatter of this family over the eastern seaboard curtailed the number of studies that could be made.

In four patients with the abnormal hemoglobin, dry smear morphology could be examined; in these instances it appeared normal. Hematocrits were obtained on all members of the family and were normal. Fragility studies were obtained in six instances of the abnormality and were normal in each case. Bilirubin was normal in the four instances in which it was obtained. None of the family had historical evidence of hematologic difficulties, and physical examination of those who were available to us failed to

reveal abnormalities that were attributable to this abnormal component.

This hemoglobin component was further studied by moving-boundary electrophoresis, and its uniqueness was established by comparison with hemoglobins H (3) and I (4), the other hemoglobins that have higher mobilities than hemoglobin A at pH 8.6. Moving-boundary electrophoretic analyses were conducted with naturally occurring mixtures of hemoglobin A with H, I, and J (8). Differences were established by comparison of the migration of the abnormal hemoglobin boundaries with that of hemoglobin A. Univalent buffers of ionic strength 0.1 were used. The buffers were as follows: pH 6.5 cacodylate, pH 7.8 barbital, pH 8.6 barbital, and pH 9.8 glycine. Each buffer was 0.08M in sodium chloride and 0.02M in the sodium salt of the buffer. At pH 6.5 hemoglobins A, I, and J migrated as cations, and hemoglobin H as an anion; at pH 7.8 and higher, all four forms migrated as anions.

In each of the buffers, the mobility of hemoglobin J was between those of hemoglobins A and I, the mobility difference between A and I being 0.6 to 0.7×10^{-5} cm² sec⁻¹ v⁻¹ and that between A and J 0.3 to 0.4×10^{-5} cm² sec⁻¹ v⁻¹. The specimen containing hemoglobins A and H was also examined in cacodylate chloride buffer of pH 6.2. The mobility difference between A and H had a large pH dependence (9), increasing from 1.0 to 2.6×10^{-5} cm² sec⁻¹ v⁻¹ between pH 9.8 and 6.2. Thus, of the four hemoglobins, A has the highest net positive charge and J, I, and H follow in order.

The electrophoretic studies showed, in addition to the mobility differences, that the carriers of hemoglobin J possessed more of the abnormal hemoglobin than hemoglobin A. This result may be discerned in the paper electrophoresis experiments shown in Fig. 1. The proportions were found to be 40 percent A and 60 percent J by moving-boundary electrophoresis at pH 6.5. The other asymptomatic carrier states for the abnormal hemoglobins are characterized by the presence of a preponderance of hemoglobin A (1).

The solubility as amorphous ferro-hemoglobin of one of the AJ samples was determined with use of the standard-

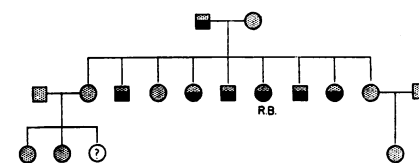


Fig. 2. Incidence of hemoglobin in the R.B. family under study. Hemoglobin J is indicated by black, and normal hemoglobin by gray.

ized procedure described by Itano (10). In this procedure, a 50-mg sample of a hemoglobin or hemoglobin mixture is salted out at 25°C in 10 ml of aqueous solvent that is 2.58M in potassium phosphate buffer of pH 6.8 and contains 100 mg of sodium dithionite (Na₂S₂O₄). The concentration of hemoglobin remaining in solution is then measured. Four determinations on one of the AJ samples yielded a value of 2.00 ± 0.24 g/lit, which is significantly higher than the solubility, 1.39 ± 0.15 g/lit, found for amorphous ferrohemoglobin A under the same experimental conditions (10).

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6. In a recent communication A. C. Allison [*Science* 122, 640 (1955)] assigned the letter J to the hemoglobin reported by J. D. Battle and L. Lewis [*J. Lab. Clin. Med.* 44, 764 (1954)] and the letter K to the hemoglobin reported in the present work. This assignment, which is the reverse of that previously agreed upon by the workers concerned (see Itano, 1) resulted from a misunderstanding in the course of conversations and correspondence between Allison and us. Allison has agreed to our inserting this correction to his communication in the present paper.
7. R. W. Smith and C. L. Conley, *Bull. Johns Hopkins Hosp.* 93, 44 (1953).
8. We are indebted to D. A. Rigas and W. N. Jensen, respectively, for samples containing hemoglobins H and I. Neither of these hemoglobins has been found free of hemoglobin A.
9. H. A. Itano, unpublished work. The moving-boundary studies are being prepared in greater detail.
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Role of Diet in Egg Development by Mosquitoes (*Aedes aegypti*)

Because of their importance as vectors of disease, the yellow fever and malaria mosquitoes have been subjected to intensive investigation for more than 50 years. Nevertheless, the nutritional require-

ments for the development of eggs in *Aedes aegypti* and *Anopheles quadrimaculatus* have never been determined. Until recently, it was generally assumed that the blood-sucking species of mosquitoes required a blood meal to mature their ova. The only studies on adult mosquito nutrition to date have been with blood fractions (1) or with supplements to blood fractions (2), but these investigations have not provided sufficient information on which to base any conclusions about the role and relative importance of such blood constituents as amino acids, lipids, carbohydrates, minerals, and vitamins. A significant difference in the number of eggs produced by mosquitoes that take blood from different host animals has been observed by several investigators, but the nutritional factors essential to the development of eggs must be known before the effect of diet on fecundity can be explained.

In a preliminary note from this laboratory, Lea *et al.* (3) reported that both *Ae. aegypti* and *An. quadrimaculatus* would ingest a skim milk and honey solution from a saturated cotton pad and would subsequently develop and lay viable eggs. In the present study (4), numerous substances in sugar solution and on saturated pads were fed to cages of 200 fertile female *Aedes* for 16 days. Although the test food was always available to the mosquitoes, no attempt was made to control the amount of food ingested or the number of insects feeding at any time. Of the foods tested, only certain proteins or their enzymatic hydrolyzates were found to stimulate egg production. Daily counts of the eggs laid over a 16-day period have been totaled for several of these foods (Table 1). Oviposition by *An. quadrimaculatus* followed the feeding of either egg albumin or proteose-peptone, the only foods tested on this species other than milk.

Although both species of mosquitoes will remain alive and vigorous for several months on a sugar solution alone, neither species has ever been known to mature eggs on a sugar diet. Therefore, it was evident that in the protein-sugar mixtures tested, protein was a major nutritional factor required for egg development and consequently for reproduction by the female mosquito.

The feeding tests were extended to include known mixtures of purified amino acids, thus affording a more accurate means of evaluating the importance of each amino acid in the diet. A medium containing 18 amino acids, dextrose and levulose, and a salt mixture was formulated, which, when fed for 14 days to cages of 400 female *Aedes*, resulted in oviposition of viable eggs. Another mixture of 12 acids (Table 2, medium A) was found to be as effective as the mixture of 18 acids. From medium A, each

Table 1. Egg production from test foods. The values represent egg production after 16 days from 200 *Ae. aegypti* females. The liquid foods contained 90 ml plus 10 ml of honey; the dry foods contained 10 g plus 10 ml of honey diluted to 100 ml with water.

Food	Eggs (No.)
Citrated (hemolyzed) beef blood	15,905
Fresh skim milk	3,072
Powdered egg albumin	9,408
Proteose-peptone	2,815
Enzymatic digest of soybean meal	1,092
Enzymatic digest of yeast	2,416
Enzymatic digest of casein	7,409
Enzymatic digest of lactalbumin	2,733

amino acid was omitted singly, and the effect on egg production was observed in the egg counts. Of the 12 acids, there were eight (arginine through valine) which, when omitted, made the medium inadequate for the development of any eggs. The omission of histidine or methionine so limited the rate of ovarian growth that only a few eggs were laid, while the omission of cystine or glutamic acid reduced oviposition to a lesser degree. Although some eggs were laid on a mixture of the eight acids alone, the best results have been obtained with a medium containing all 12 amino acids.

Tests were also made in which the concentration of each of the 12 acids was varied. The optimum quantity of each acid was then used to establish a new medium (Table 2, medium B) that enabled the mosquitoes to lay an average of 14,000 eggs in 14 days. This was twice the number laid on medium A and indicated the importance of proper balance among the amino acids in the mixture. In addition, some preliminary tests of other factors in blood showed that lipids are dispensable but that minerals may be

Table 2. Amino acid composition of test diets. Each medium also contained 5 g dextrose, 5 g levulose, 0.15 g of a salt mixture, and 100 ml of water.

Amino acid	Composition (g/100 ml)	
	Medium A	Medium B
L-Arginine	0.5	0.38
DL-Isoleucine	1.0	0.50
L-Leucine	1.0	0.75
L-Lysine	0.9	0.75
DL-Phenylalanine	0.7	1.20
DL-Threonine	0.8	0.30
L-Tryptophan	0.4	0.30
DL-Valine	1.0	1.00
L-Histidine	0.7	0.15
L-Methionine	0.2	0.15
L-Cystine	0.2	0.15
L-Glutamic acid	1.0	1.00