

could provide 800 to 1600 mg of calcium daily. We, thus, suggest that since there may be unusual sources of calcium in native diets, the nutritional status of Peruvian natives be re-evaluated. We also wish to reiterate the suggestion made nearly 20 years ago (7) that the results of nutritional studies on non-Western groups be only tentatively accepted until the results have been carefully checked by a thorough analysis of all nutrient sources (8).

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Milk Analysis of the Kangaroo Rat, *Dipodomys merriami*

Abstract: *The milk of Dipodomys merriami has an average water content of 50.42 percent, which is low compared with milk from other mammals. The fat content is about 23.48 percent. The significance of these values is discussed in terms of the animal's water balance.*

Numerous workers have studied the various mechanisms of water conservation in desert mammals. However, there is one particular area of water balance that has been studied very little, yet might yield useful information. This is the effect of the environment upon the composition of the milk, in various species of animals, particularly its water

content, and the evaluation of the results in terms of adaptive significance.

We were therefore interested in making a preliminary analysis of the milk of the kangaroo rat, *Dipodomys merriami*. Rats were collected 35 miles south of Tucson on the Santa Rita Experimental Range at an elevation of approximately 1100 meters. The collections were made during April and May, a period of great reproductive activity in the kangaroo rat from this region and, also a time of scant rainfall and low humidity in the area.

The milk samples were collected from rats which had been anesthetized with ether and injected with pitocin. Then, by manual expression, the drops of milk were collected into a blood-mixing pipette. The milk was then transferred to a sterile syringe, sealed tightly, and stored in a freezer until analyzed. Because of the small amounts in the samples, a modified method of milk analysis was used. This method was suggested by Stull (1).

The results are summarized in Table 1. The milk is very concentrated compared with that from the dairy cow, which contains about 88 percent water. The milk of another desert mammal, the camel, contains 87.7 percent water (2). However, both the camel and her calf are exposed to almost the full effects of the desert heat in the daytime, and water is utilized for temperature regulation (3). Similarly, this might explain the high water content, 83.55 percent (4), of the milk from the collared peccary, *Pecari tajacu*, another large desert mammal which is also exposed to the daytime heat. In contrast, the kangaroo rat avoids the intense, solar radiation by remaining in burrows. This burrowing habit, together with other adaptations described by Schmidt-Nielsen (5), make it probable that the amount of water lost by evaporation in the rat is very small. Furthermore, the newborn rat probably produces a concentrated urine similar to that of its parent; consequently, the young rat may not require a large supply of water in the milk.

The only other mammals known to have milk with such low water content are seals and whales (Table 2). Although the fat content of kangaroo rat milk is high, that of marine mammals is even higher. The functional significance of a high fat content is still questionable. Irving (6) has suggested that the advantages of fat stores in Arctic birds might be that fat, when compared with

Table 1. Summary of some of the milk constituents of *Dipodomys merriami*. All samples represent a pooling from more than one rat. The contributions of each animal to the sample are not equal. The stage of lactation is unknown.

No. of rats	Sample size (g)	Fat (%)	Non-fat solids (%)	Water (%)
3	0.0791	24.65	33.50	41.85
2	.1192	18.79	23.83	57.38
4	.1172	29.61	20.90	49.49
2	.1101	20.89	26.16	52.95
Average	.1064	23.48	26.10	50.42

Table 2. Summary of the milk constituents of some marine mammals.

Fat (%)	Protein (%)	Water (%)
42.82	Harp seal (<i>Phoca groenlandica</i>) 11.98	43.79
40.43	Hooded seal (<i>Cystophora cristata</i>) 6.65	49.85
36.5	California sea lion* (<i>Zalophus californianus</i>) 13.8	47.3
38.13	Blue whale (<i>Balaenoptera musculus</i>) 12.79	47.17
30.60	Fin whale (<i>B. physalus</i>) 13.14	54.10

* From M. E. Q. Pilson and A. L. Kelly, *Science* **135**, 104 (1962). All others from a compilation made by E. Sivertsen, *Hvalradets Skrifter* No. 26 (1942).

carbohydrate and protein, has the highest energy production for its weight; less water is required for its storage, yet when metabolized it yields the most water. Applying these principles to seals and kangaroo rats, a milk containing a high proportion of fat, in contrast to one high in either carbohydrate or protein, might be advantageous to both the mother and the young for water conservation. The mother produces a concentrated milk where little water is lost during nursing, and the young obtains a high-energy food supply that requires little water for its storage or use.

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References and Notes

1. J. W. Stull of the Department of Dairy Science, University of Arizona. After ether extraction of the fat, the solvent is evaporated in a vacuum at 135°C, and the extracted fat is weighed. Then, the remaining non-fat portion, which has been dissolved in an ammonium hydroxide solvent, is desiccated in a vacuum oven at 100°C. Controls with cow's milk showed a close correlation. Dr. Stull's assistance in the milk analysis is appreciated.
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Erythroid Homeostasis in Normal and Genetically Anemic Mice: Reaction to Induced Polycythemia

Abstract. After hypertransfusion, normal *ww* and anemic *WW^v* mice show the same increase in red-cell volume, decrease in reticulocytes, and temporary cessation of blood formation. The time required for the red cell volume to return to the value observed before treatment is the same for both groups of mice. The cell volume at which new erythrocytes are again released into the circulation depends upon the genotype of functioning blood-forming tissue. Injections of erythropoietin stimulate red cell formation in polycythemic *ww* mice, but have much less (if any) effect upon polycythemic *WW^v* mice.

Erythroid homeostasis of *WW^v* mice, afflicted with a genetically induced macrocytic anemia, differs from that of normal mice. Under normal atmospheric conditions, 1 mm³ of blood from an untreated adult *WW^v* mouse contains 6 or 7 million erythrocytes, that from a normal mouse 11 or 12 million cells (1). These red blood cells form a smaller proportion of the total blood volume in *WW^v* mice (38 to 40 percent) than in *ww* mice (46 to 49 percent). The differing ratios of cell volume to total blood volume (hereafter called hematocrit levels from their method of determination) depend upon the genotype of functioning blood-forming tissue, since *WW^v* mice implanted with isologous *ww* blood-forming tissue acquire and maintain a hematocrit percentage of 46 to 49 percent (2). The anemic mice respond normally to oxygen deprivation, but injections of purified erythropoietin have

little or no influence upon their blood picture (3). In studies of the mechanism of erythroid homeostasis, polycythemic normal mice have frequently been used, since they cease forming new erythrocytes until their hematocrit levels have dropped markedly, or until they have been stimulated by the hormone erythropoietin. They provide both a sensitive tool for assay of erythropoietin activity (4) and a favorable site for study of the kinetics of differentiation and multiplication of red cell precursors (5).

We have used *ww* and *WW^v* mice, highly congenic with the WB × C57BL/6 F₁ hybrid genetic background (6), in a series of experiments to gain further evidence concerning the basis for observed differences in erythroid homeostasis. In each experiment, normal (*ww*) and anemic (*WW^v*) adult littermate pairs were treated identically; they were plethorized to a hematocrit

level of at least 70 percent and observed after the same intervals to determine the duration of polycythemia and the absence of new blood formation.

Approximately 2 percent of the red cells of untreated *ww* mice, and 3 to 4 percent of those of untreated *WW^v* mice, are young incompletely hemoglobinized erythrocytes with a recognizable reticulum; following plethorization, these reticulocytes disappear from the circulation. Table 1 shows results of four experiments, each involving a specified difference in conditions. All mice, normal and anemic, were injected on each of 4 consecutive days with 0.5 ml of a red cell suspension (70 to 90 percent vol/vol in physiological saline). Determinations of the ratio of cell volume to total blood volume were made by the microhematocrit method (7). Each reticulocyte count was based on at least 500 cells, stained by the method of Brecher (7). In experiments 1, 3, and 4, the donors were retired C57BL/6J breeders and in experiment 2, they were 4- to 7-month old WB-B6F₁ hybrid *WW^v* mice. The *ww*-implanted *WW^v* mice in experiment 3 all had typical *ww* blood pictures before and after the period of polycythemia. Erythropoietin (8) was injected intraperitoneally on days 1 to 8 after completion of plethorization.

In each of these experiments, *ww* and *WW^v* mice responded similarly to plethorization, although the extent of cell-volume increase, resulting from equal volumes of added red cells, tended to be greater in nonimplanted *WW^v* recipients (mean increase, 36 percent) than in *ww* recipients (mean increase, 30 percent). In experiments 1 to 3, there was almost complete suppression of erythropoiesis (absence of reticulocytes) throughout the period of polycythemia (17 to 22 days). For a brief period after the cell volume returned to normal level, elevated reticulocyte percentages (8 to 10 percent, sometimes higher in *WW^v* mice) were always observed.

Experiment 2 demonstrates that plethora induced by addition of macrocytic *WW^v* erythrocytes persists at least as long as that induced by addition of an equal volume of normocytic *ww* erythrocytes, indicating that *WW^v* erythrocytes have a normal lifespan.

In experiment 3, *ww* and *WW^v* mice implanted with *ww* blood-forming tissue gave identical responses to plethorization, including the reappearance of reticulocytes when the hematocrit level

Table 1. Responses of normal *ww* and anemic *WW^v* mice to plethorization with or without other treatment. Ht., hematocrit; Ret., reticulocyte; Geno. (No.), genotype and number of plethorized mice. Results in mean ± S.E.

Geno. (No.)	Before transfusion		Peak (day 0)		Interm. (day 8)		Polyc. days	
	Ht. (%)	Ret. (%)	Ht. (%)	Ret. (%)	Ht. (%)	Ret. (%)		
<i>Expt. 1. Plethorization only with C57BL/6J ww RBC</i>								
ww (10)	46 ± 0.3	2 ± 0.3	76 ± 1.0	0.2 ± 0.1	63 ± 1.6	0.2 ± 0.1	18	
WW ^v (10)	36 ± 0.9	4 ± 0.3	73 ± 1.1	0.6 ± 0.1	59 ± 1.3	0.2 ± 0.1	19	
<i>Expt. 2. Plethorization only with isologous WW^v macrocytic RBC</i>								
ww (3)	49 ± 1.0	3.5 ± 0.3	76 ± 1.2	0.2 ± 0.1	69 ± 1.0	0.2 ± 0.1	21	
WW ^v (3)	39 ± 1.0	4.5 ± 0.5	78 ± 1.2	0.2 ± 0.2	66 ± 2.0	0.2 ± 0.1	22	
<i>Expt. 3. Established implant ww cells, plethorization with ww RBC</i>								
ww (4)	48 ± 1.0	3 ± 1.0	72 ± 1.5	0.4 ± 0.2	61 ± 1.0	0.2 ± 0.1	17	
WW ^v (4)	48 ± 1.1	3 ± 0.9	74 ± 0.8	0.2 ± 0.1	64 ± 0.7	0.2 ± 0.2	17	
<i>Expt. 4. Plethorization with ww RBC, plus injection of erythropoietin</i>								
ww (5)	47 ± 0.6	2.7 ± 0.7	80 ± 1.5	0.8 ± 0.4	73 ± 1.4	3.2 ± 0.5	24	
WW ^v (5)	41 ± 1.2	4.0 ± 0.6	77 ± 0.6	1.1 ± 0.8	65 ± 0.6	0.6 ± 0.1	23	