

SHORT COMMUNICATION

SOME PROPERTIES OF RED BLOOD CELL GLUCOSE-6-PHOSPHATE DEHYDROGENASE FROM THREE SPECIES OF KANGAROO*

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Richardson and Czuppon (1969) reported a cline in red blood cell glucose-6-phosphate dehydrogenase levels in a marsupial—the wallaroo *Macropus robustus* Gould, 1841. High dehydrogenase levels were found in animals from the dry inland areas of Australia, while animals from the wetter eastern coastal areas had lower levels. Richardson and Czuppon (1970) presented evidence to show that this phenomenon was inherited and not the result of direct environmental influences. The variation in dehydrogenase levels could be due to altered function of the enzyme, variations in the rate of production of the enzyme, or to a change in the half-life of the enzyme. This communication reports some biochemical properties of the enzyme. These properties were also determined for enzyme preparations from two related species—the grey kangaroo, *Macropus giganteus* Shaw, 1790, and the red kangaroo, *Megaleia rufa* (Desmarest, 1822).

The methods used for purifying the enzyme and for determining K_m values with glucose 6-phosphate and TPN as substrate were those recommended by Kirkman (1962). Utilization of the substrate analogue 2-deoxyglucose 6-phosphate relative to glucose 6-phosphate was determined for each animal. For the pH activity curves the standard assay procedure (Zinkham, Lenhard, and Childs 1958) was used with the pH of the Tris-HCl being altered to give the desired pH in the final solution.

The results are summarized in Tables 1 and 2. The purified enzyme preparation contained less than 1% glucosephosphate isomerase and less than 0.01% 6-phosphogluconate dehydrogenase.

In the three species examined the K_m values with glucose 6-phosphate and TPN are markedly higher than those reported for humans (Kirkman 1962). While the K_m TPN values for the inland and coastal wallaroos are indistinguishable, a slight difference is apparent between the K_m glucose 6-phosphate values. This variation, which would lead to a difference of about 1 unit of activity between inland and coastal animals under the normal assay conditions, is insufficient to explain the difference of 13 units found between the two forms.

The pH activity curves for the dehydrogenases of the inland and coastal wallaroos and of the red kangaroo were similar. The grey kangaroo preparation was active over an even wider range of pH and the curve differed in shape from that of the other kangaroos, being biphasic.

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While the properties of the glucose-6-phosphate dehydrogenase of these three species have much in common, as would be expected from their close phylogenetic relationship, in each case there is some slight, presumably evolutionary, variation. The reported variation in dehydrogenase levels between inland and coastal forms was not due to alteration in any of the properties studied.

TABLE 1
SUMMARY OF THE BIOCHEMICAL PROPERTIES OF THE PURIFIED GLUCOSE-6-PHOSPHATE
DEHYDROGENASE PREPARATIONS

Species*	Glucose-6-Phosphate Dehydrogenase Level in Population ($\mu\text{g/g Hb}$)†	K_m (μM)		Relative 2-Deoxyglucose 6-Phosphate Utilization	Specific Activity
		Glucose 6-Phosphate	TPN		
<i>Macropus robustus</i>					
Inland (2)	25.7 ± 3.2 (21)	110, 125	37.5, 40	<1%	130
Coastal (2)	13.0 ± 1.5 (23)	147, 150	37, 37	<1%	238
<i>Megaleia rufa</i> (1)	4.65 ± 0.4 (55)	140	15.5	<1%	44
<i>Macropus giganteus</i> (1)	25.5 ± 3.7 (8)	125	37	<1%	336
Human	4.22 ± 0.11 (202)	50-78	2.9-4.4	<4%	

* No. of preparations given in parenthesis.

† Mean values $\pm 95\%$ confidence limits given. No. of determinations given in parenthesis.

TABLE 2
ACTIVITY OF GLUCOSE-6-PHOSPHATE DEHYDROGENASE AT VARIOUS pH VALUES
Activities expressed as a percentage of the activity at pH 9.0. Number of
preparations (n) given in parenthesis. When n was > 1 , the average was taken

Species	pH								
	3	6.5	7.05	7.6	8.0	8.4	9.0	9.5	10.4
<i>Macropus robustus</i>									
Inland ($n=2$)	0	39	61	78	93	97	100	82	5
Coastal ($n=1$)	0	29	50	83	94	97	100	69	0
<i>Megaleia rufa</i> ($n=1$)	5	35	60	85	95	100	100	100	15
<i>Macropus giganteus</i> ($n=3$)	27	54	79	92	83	100	100	100	6
Human*	—	63	72	84	91	96	100	87	—

* From Kirkman (1962).

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