The effects of copper on erythrocytic oxidant defence in the black rhinoceros *(Diceros bicornis)*

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The most common cause of death among captive black rhinoceros *(Diceros bicornis)* is acute intravascular haemolytic anaemia (Paglia 1993). Autopsy of one animal showed hepatic changes similar to Wilson's disease in humans, and so it was decided to investigate some effects of copper on the red blood cells (RBC) of these animals. Cu (II) is a well known oxidant of both oxy– and deoxy–haemoglobin leading to the formation of methaemoglobin. The RBC of most species are well protected from oxidative damage by their prime cargo, oxygen. This protection is driven by a series of antioxidant mechanisms mostly tied to the pentose phosphate pathway of glucose metabolism. We compared the susceptibility to and recovery from oxidative insult of RBC obtained from the rhinoceros (n=3), a related species the horse (n=4), and humans (n=4).

Glycolytic enzymes and methaemoglobin formation were measured according to Beutler (1984). RBC were incubated with either 1–acetyl–2–phenylhydrazine (APH) or phenazine methosulphate (PMS) or copper sulphate. These three chemicals provide an oxidant stress that becomes manifest as an increase in methaemoglobin formation. The results are shown in Figure 1. There was a very significant effect of copper sulphate in the rhinoceros RBC, leading to a rapid and large increase in methaemoglobin formation. The response to APH and PMS were similar in all three species.

The RBC activity (IU/gHb) of 6 phosphogluconate dehydrogenase was about 10 times higher in the rhinoceros (30.53 ± 3.09) than the horse (3.02 ± 0.28) and 4 times higher than in humans (6.93 ± 0.56). This suggests a high capacity for the pentose phosphate pathway to protect the RBC from oxidant damage. However, copper is a well known inhibitor of this enzyme, exacerbating its oxidant effects on haemoglobin.

Most captive rhinoceros are inadvertently fed a high copper ration (lucerne hay) and often graze on weed species known to cause accumulation of copper (e.g. *Echium* and *Heliotropium*). Their extreme sensitivity to copper toxicosis may explain the high incidence of haemolytic anaemia found in these animals.

Beutler, E. (1984). *Red Cell Metabolism. A Manual of Biochemical Methods*, 3rd edition. Grune and Stratton, New York.

Paglia, D.E. (1993). Acute episodic hemolysis in the African Black rhinoceros as an analogue of human glucose–6–phosphate dehydrogenase deficiency. *American Journal of Hematology* 42, 36–45.

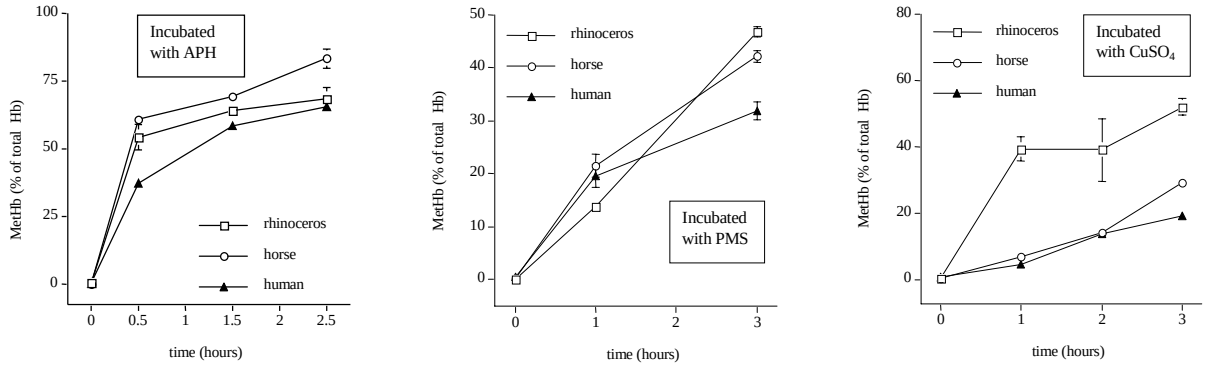

Figure 1 The effects of APH, PMS and $CuSO_4$ on the rates of methaemoglobin formation in the erythrocytes of the rhinoceros, horse and human.