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EFFECTS OF TRANSLOCATION ON THE BLACK RHINOCEROS (*DICEROS BICORNIS*) - RED AND WHITE CELL PARAMETERS

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Introduction

Translocation of the black rhino has become an important part of its management and conservation in Africa. There are now very few animals living in their original range which have not been moved at one time or another for reasons of security or placement in sanctuaries. This is the case in Kenya and efforts have been made to reduce risks associated with these procedures as the future survival of this species depends on low mortality. Techniques for capture and translocation have improved in recent years (KOCK et al., 1990a) and some effort has been made to ascertain effects of these procedures on biological parameters (KOCK et al., 1990b; KOCK and MORKEL, 1990).

Considerable effort has been put into understanding the red cell in this species as anaemia has been a concern in captive animals (MILLER, 1994; CHAPLIN et al., 1986; FAIRBANKS and MILLER, 1990; PAGLIA, 1993; PAGLIA and MILLER, 1992; PAGLIA and MILLER, 1992; PAGLIA et al., 1992; PAGLIA et al., 1986). Comparative data has shown some differences and similarities between wild and captive animals in relation to the red cell and reported pathology (KOCK et al., 1992).

This paper reports haematological findings from 1991-1994 when approximately 170 immobilisations of black rhino were undertaken in Kenya as part of translocation protocols and health management.

Materials and Methods

Free ranging black rhino of differing sex and age were immobilised using a dart rifle (Palmer Capchur) with metal darts containing etorphine (Pure etorphine - CVet Ltd), Hyaluronidase (Hylase) and xylazine (Rompun crystalline - Bayer) either from a ground tracking position or a helicopter (Bell Jet Long Ranger or a Hughes 500). Doses, induction times and periods of immobilisation are summarised in Table 1. Animals were clinically examined and blood and tissue samples taken . Anticoagulants used were ethylenediaminetetraacetic acid (EDTA) and lithium heparin for complete blood counts and plasma biochemistry, plain tubes were used for whole blood collection for serum. Samples were taken within 10 minutes of recumbency. Blood smears and blood separation were completed within 8 hours and haematological examination performed within 2 days in the majority of cases. Plasma and serum were frozen immediately after separation in liquid nitrogen for later analyses. Packed cell volumes were recorded immediately at the capture site using a field micro centrifuge (Compur) spinning for 7 minutes.

After capture the animals were recovered using narcotic antagonists (Nalorphine, diprenorphine - revivon Cvet Ltd) and loaded into a wooden transport crate which was winched up a ramp onto a lorry for translocation to various locations. Total transport times were recorded (Table 1). The animals were placed after transport into a boma at the release site where they remained for a period of 3 - 4 weeks. A second immobilisation was performed prior to final release for placement of radio-tracking transmitters in the horn and health checks. Doses, induction and immobilisation times are given in table 1. Blood samples were taken and processed as previously described.

For the comparative analyses between population sets in this paper data was selected from 39 PCV and 18 MCV paired samples out of a total of 138 samples for consistency and comparability to examine if transport time, altitude change or presence of trypanosomiasis had any impact on the red cell parameters.

Animals were translocated from the laikipia plateau or Nairobi National Park to other sites in the laikipia region, Nairobi or Tsavo National Park, East and West.

Table 1: Total doses(mg) of immobilising drugs used, anaesthetic induction, immobilisation and transport times (135 interventions on black rhinoceros)

DRUGS TOTAL DOSE RANGE	CAPTURE	BOMA
XYLAZINE mg	50 - 100	0
ETORPHINE mg	3 - 5	0.6 - 3
HYLASE i.u.	1500	0

ANAESTHETIC DATA n=135	ALL RHINO RANGE	AVERAGE AND STANDARD DEVIATION
INDUCTION TIMES	2.5 - 27 minutes	7.2 +/- 4 minutes
IMMOBILISATION PERIOD	14 - 120 minutes	41 +/- 19 minutes

TRANSPORT TIMES	HIGH ALTITUDE - LOW n=26	SAME ALTITUDE n=13
RANGE (minutes)	330 - 1080	60 - 300
MEDIAN	600	240
AVERAGE + S.E.	583 +/- 37	182.7 +/- 32

Results and discussion

A summary of the average haematological picture is shown in table 2 and 4 and compared with other data sets. Box whisker plots (figures 1-5) show data from different locations and comparisons of blood packed cell volume and mean cell volume changes between time of capture and release in each animal (paired samples). Two sets of PCV data were chosen for comparison as there appeared from inspection of the data to be a difference between; those transported for less than 6 hours with no significant altitude or daily mean temperature changes and no exposure to trypanosomes (H-H), the others animals transported for more than 6 hours with reduction in altitude, elevation in ambient temperature and exposure to trypanosomes (H-L). Statistical tests (SPSS 6.0 statistical software package) relating to the data sets are recorded on the figures.

In general the data from Kenya does not differ from other data sets (Table 2) (KOCK et al., 1990c; ZSL, 1991; NELSON and FOWLER, 1986) but it is notable that the mean haemoglobin levels are lower in the Kenyan animals and the percentage eosinophils is higher in the free living animals which might reflect a higher level of parasites.

The paired data did demonstrate a significant drop in PCV when animals were transported long distances to lower altitudes, higher ambient temperatures and with exposure to trypanosomes. Mean cell volumes did not change significantly. The animals transported shorter distances did show a downward trend but this was not significant statistically. A downward trend in PCV was also noted in Zimbabwe (KOCK et al., 1990b) after boma confinement (between 1-80 days) but the protocols for translocation were markedly different and the data is not directly comparable.

A reduction in PCV indicates a loss of red blood cells from circulation or relative anaemia. The lower value can be due to pathological anaemia fluid retention and adrenolytic drugs. The causes of anaemia can be due to haemorrhage, intravascular haemolysis, aplastic effects of toxins or irradiation and depression of bone marrow production (hypoproliferative).

In these rhino fluid retention is unlikely. The drugs used were consistent in both sampling periods so should not influence the difference which suggests that pathological anaemia was the cause. Depression anaemia is unlikely in the time frame of 3 weeks as is aplastic anaemia.

Haemorrhage or haemolysis are possible causes. In general the serum and plasma did not show evidence of recent haemolysis at the time of sampling but biochemistry remains pending on the samples. It may be possible to demonstrate changes in biochemical parameters to confirm a previous haemolytic crisis. There was no obvious frank haemorrhage associated with trauma or marked staining of the faeces suggesting lower bowel haemorrhage. There are many causes of haemorrhage but the most likely in this instance is a bleeding lesion internally. One possible cause of this is gastric ulceration.

Table 2 Comparisation of haematological values for wild and captive rhino average values unless range given

VARIABLE	KEN.BR	REF A	REF B	REF C	REF D BLACK	REF D WHITE
SAMPLE NUMBER	138		86		7	30
RBC (*10^12/l)	5.40	5.09	5.26	5.87-8.11	4.8	6.82
HAEMOGLOBIN (mg%)	13.41	15.2	16.1	13.1-19.5	14.7	17.03
MCV (fl)	81.83	84	82.5	55.2-67.6	89.5	68
MCH (pg)	24.17	29.84	30.9	19.5-27.1	31.6	25
MCHC (g/dl)	29.18	38.24	37.7	32.6-42.6	35.4	36.7
PACKED CELL VOLUME %	44	42	43	36.7-49.5	42	46.4
PLATELETS	422		210		314.2	419.4
WBC (*10^9/l)	9.02	9.34	11.5	6.1-15.6	8.5	8.6
NEUTROPHILS %	56.6	60.2	54	40-50	60	62
LYMPHOCYTES %	34	30	35	40	34	29
EOSINOPHILS %	5	1	5	0-2	3	5
BASOPHILS %	.4	2	0	0	0	0
MONOCYTES %	4	6	6	0-1	3	4

Ref A: C/BR captive black rhino MARUSKA E.J. et al., 1986;
Ref B: ZIM/BR wild Zimbabwe rhino KOCK M.D. et al., 1990c;
Ref C: C/WHR white rhino WALLACH J.D. and W.J. BOEVER 1983;
Ref D: C/BR C/WhR captive black and white rhino ZSL. 1991
KEN/BR Kenya wild black rhino

Table 3: Haematological values for SAM black rhino

Variable	**Nairobi**	**Tsavo**
Haemoglobin (mg%)	16	16
PCV (%)	48	48
WBC count (*10^9/l)	6.4	6.2
Neutrophils	3.7	3.7
Lymphocytes	1.8	1.9
Eosinophils	0.64	0.43
Basophils	0	0
Monocytes	0.26	0.43

Haemolytic anaemia is not uncommon in captive animals and the phenomena has led to considerable research in recent years, the results of which are suggestive of a tendency for the black rhino red cell to haemolyses under conditions of metabolic stress with an inherent acatalasaemia predisposing to lysis. Leptospirosis and hypophosphataemia have been implicated in the captive animal syndrome (MILLER, 1994) but are unlikely factors in these animals. Reports of haemosiderosis (KOCK et al., 1992) in captive stressed and translocated animals at relatively higher levels than in animals dying of natural causes or very recently translocated animals which died (<1 week) is suggestive that haemolysis may in part explain the drop in PCV observed from this data.

Causes for haemolysis can be blood parasites, bacterial or viral infection, drugs, toxic plants or venoms, intra-erythrocytic defects, immune mediated reactions, metabolic disease or disorder and water intoxication. Most of these can be excluded as very unlikely. The question is what was different about the two sets of translocated animals.

Table 4: Haematological values for free ranging black rhino. Kenya 1992 - 1994

VARIABLE	NUMBER	MEAN	SD	VAR	MIN	MAX
RBC (*10^12/L)	76	5.4	.98	.95	3.3	7.8
HAEMAGLOBIN (mg%)	99	13.4	2.19	4.79	8	18.3
MCV (fl)	75	81.83	10.65	113.52	53.70	114.40
MCH (pg)	74	24.17	3.85	14.81	16.00	32.30
MCHC (g/dl)	79	29.18	4.56	20.84	12.20	40.70
PACKED CELL VOLUME %	120	43.74	5.76	33.23	28.50	55.00
PLATELETS	90	422.48	255.36		52	1097
WBC (*10^9/l)	117	9.02	3.06	9.36	3.60	17.60
NEUTROPHILS %	101	57	13	158	16	90
LYMPHOCYTES %	101	34	12	145	10	76
EOSINOPHILS %	101	5	3	12	0	18
BASOPHILS	100	0	1	1	0	4
MONOCYTES	101	4	4	13	0	20

There are four changes effecting the animals that might have been significant. Exposure to trypanosomes, change in altitude and ambient temperature and degree of transport stress. These factors theoretically could have contributed to a relative change in red cell to plasma ratios or lysis or sequestration of red cells from circulation.

Trypanosomes - previous studies (MIHOK et al., 1992a; MIHOK et al., 1992b) and work during these more recent translocations suggests that trypanosome infection becomes detectable at 3-4 weeks (direct microscopic inspection or xenodiagnosis) from an animal newly translocated from a trypanosome free area to where trypanosomes are endemic. It is possible that the parasite is cryptic in the early period and is causing damage to the red cells. This may in part explain the change in PCV observed without overt parasitaemia. The results of the 1992 MIHOK study showed a drop of 35% in PCV at 4 weeks in one rhino under a relatively heavy tsetse fly challenge (this animal recovered spontaneously without treatment) and T.brucei infection was implicated in the death of another animal. In the recent translocations at no time did animals appear ill or show symptoms of trypanosomiasis, they were not given any prophylactic treatment and after release thrived. The boma locations were different to the earlier reports and the fly challenge was probably lower although timings were oriented to periods when the vegetation was optimal and not selected to times of low fly activity. The change in boma location and protocols may in part explain the lack of clinical disease seen in recent translocations.

Altitude change - the animals went from areas at approximately 6000 to 1000 feet above sea level. In one reported study (JAIN, 1993), movement of cattle from 5000-9000 feet led to an increase in RBC of 6%. This was considered an underestimate as there was a seasonal effect occurring at the time of the relocation which usually led to a drop in numbers which may have reduced the actual potential rise. It can be speculated that a change to a lower altitude of a similar magnitude might lead to a drop in PCV. The drop in the data reported in this paper was of the order of 15% (Figure 3). It is possible that reduced production of erythropoeitin and reduced erythropoiesis would impact the PCV where the longevity of the red cell is of the order of 100 days. In 24 days without cell replacement we would expect a maximum drop of 24%.

Higher environmental temperatures - this might lead to increased water intake. The animals were stressed and high corticosteroid levels associated with this might have increased the thirst of the animals and water intake. Over hydration could lead to a relative drop in PCV. Examination of the plasma proteins will enable confirmation of this and is pending.

Stress - is difficult to quantify but clearly the procedure of darting a free living animal and placing it in a relatively small box, transporting it long distances on poor road surfaces for periods up to 14 hours and then putting it in a confined area with contact with conspecifics and humans at close quarters for 3-4 weeks is stressful. Cell metabolism is effected with a change in feed intake (volume and type), increased oxygen demand and oxidative stress (lactic acidosis) especially at the time of immobilisation with respiratory depressants such as narcotics being used and during transportation when there is considerable strain on the muscles. The only significant difference in the translocation protocols between the two groups of animals examined is in transport time as otherwise procedures and presumably stress factors were similar.
Sam - one translocated rhino was remarkable in that there was no change in PCV at all (Table 3). This animal was a tame hand reared individual that showed no stress and seemed totally relaxed in the crate, arrived in a similar state at the bomas and almost seemed to enjoy the journey and new surroundings. It was subjected to all the other factors mentioned and yet no alteration in PCV occurred.
This evidence is suggestive that transport stress might be the most important factor in the drop in PCV recorded in the wild translocated rhino.

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Summary

Effects of translocation on the black rhinoceros (Diceros bicornis) - red and white cell parameters

Haematology data taken from free-living black rhinoceros in Kenya during translocation procedures are consistent with average findings from animals in Zimbabwe and in general with captive animal data except that haemoglobin levels are generally lower and eosinophils higher than captive animals. There is evidence in the free-living black rhinoceros that a drop in PCV occurs between sampling animals at capture and 3-4 weeks later under certain conditions. The factors that lead to this include translocating them long distances to lower altitude. hotter and trypanosome endemic areas. The anaemia is most probably pathological involving the loss of red cells from circulation through sequestration or haemorrhage. The most likely central factor is transport stress leading to perhaps gastric ulceration with haemorrhage and metabolic problems consequential to general and metabolic (hypoxemia and lactic acidosis) stress affecting the red cell directly. Change in altitude, water intake. exposure to trypanosomes may all contribute to some degree in individual cases but the exception of Sam. a tame unstressed animal which showed no change in PCV suggests they have minimal effects in the time period involved.

Zusammenfassung

Umsetzung von Spitzmaulnashörnern (Diceros bicornis) - Auswirkungen auf das rote und weiße Blutbild

Die von wildlebenden Spitzmaulnashörnem in Kenia im Verlaufe von Umsetzungen in andere Lebensräume erhobenen hämatologischen Befunde stimmen mit den Durchschnittswerten bei Tieren in Zimbabwe sowie ganz allgemein bei Tieren in Gefangenschaft überein, allerdings mit der Einschränkung, daß die Hämoglobinspiegel allgemein niedriger und die Eosinophilen höher liegen als bei Tieren in Gefangenschaft.
Bei wildlebenden Spitzmaulnashömern konnte unter bestimmten Umständen von der Probenahme zum Zeitpunkt des Einfangens bis drei bzw. vier Wochen danach ein Abfall der Hämatokritwerte nachgewiesen werden. Dies war u.a. durch Umsetzungen über weitere Entfemungen in niedrigere Höhenlagen sowie wärmere Regionen und endemische Trypanosomgebiete bedingt. Die mit großer Wahrscheinlichkeit als pathologischer Faktor auftretende Anämie geht mit einem durch Isolierung und Blutungen verursachten Verlust an roten Blutzellen aus dem Kreislauf einher. Die Hauptrolle spielt hier wahrscheinlich der transportbedingte Stress, der unter Umständen zu Magengeschwüren mit Blutungen und Stoffwechselstörungen führt. Diese wiederum hängen mit dem allgemeinen Stress und mit Stoffwechselbelastungen zusammen (Hypoxämie und Laktoazidose), die sich unmittelbar auf die roten Blutzellen auswirken. Veränderungen in der Höhenlage, der

Wasseraufnahme und der Exposition gegenüber Trypanosomen können im Einzelfall von Bedeutung sein. Eine Ausnahme bildete Sam, ein gezähmtes, nicht gestreßtes Tier, daß keinerlei Veränderungen der Hämatokritwerte zeigte, so daß hier zumindest im Untersuchungszeitraum auf minimale Auswirkungen geschlossen werden kann.

Résumé

Le transfert de rhinocéros noirs (Diceros bicornis) - ses effets sur les données hématologiques rouges et blanches

Les hémogrammes obtenus à partir de rhinocéros noirs vivant en liberté au Kénya pendant leur transfert dans d'autres régions correspondent aux valeurs moyennes observées chez les animaux du Zimbabwé et en général aux données recueillies sur les animaux gardés à l'exception des taux d'hémoglobine généralement plus bas et des taux d'éosinophiles plus élevés que pour les animaux gardés en captivité.
En fonction des conditions données, il a été possible de déterminer, dans les prélèvements obtenus sur les rhinocéros noirs vivant en liberté une baisse des valeurs d'hématocrite à partir du moment de leur capture jusqu'à trois à quatre semaines plus tard. Les facteurs ayant causé cette baisse sont entre autres les transports sur de longues distances vers des régions à altitude plus basse, aux températures plus élevées et où la trypanosomiase est endémique. L'anémie probablement pathologique va de pair avec une perte de globules rouges causée par l'isolement et les hémorragies dans la circulation. Le facteur principal qui joue en l'occurrence est probablement le stress dû au transport aboutissant dans certaines conditions à des ulcérations avec hémorragies et troubles métaboliques consécutifs au stress général et métabolique (hypoxémie et acidose lactique) affectant les globles rouges directement. Le changement d'altitude, l'eau que les animaux boivent, l'exposition au risque de contamination par le trypanosome peuvent jouer un rôle dans des cas individuels mais l'exemple de Sam, d'un animal domestique non stressé qui ne présentait pas de changement dans ses taux d'hématocrite paraît montrer que ces facteurs n'ont qu'un effet minime sur la période considérée.

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