ORIGINAL ARTICLE

Mineral absorption in the black rhinoceros (*Diceros bicornis*) as compared with the domestic horse

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Summary

To test whether mineral recommendations for horses are likely to guarantee adequate mineral provision for black rhinoceroses (*Diceros bicornis*), we investigated the apparent absorption (aA) of macro- and microminerals in eight black rhinoceroses from three zoological institutions in a total of 32 feeding trials with total faecal collection, with additional data from three unpublished studies (18 feeding trials). Feeds and faeces were analysed for Ca, P, Mg, Na, K, Fe, Mn, Cu, Zn and Co. The resulting aA coefficients, and the linear relationships of apparently absorbable dietary mineral content to total dietary mineral content [per 100 g dry matter (DM)], were compared with data for domestic horses. Rhinoceroses had significantly higher aA coefficients for Ca and Mg (because of a higher calculated 'true' absorption), and lower ones for Na and K (because of calculated higher endogenous faecal losses). High absorption efficiency for divalent cations is hypothesized to be an adaptation to a natural diet of particularly high Ca:P ratio (approximately 14:1); an effective removal of Ca from the ingesta guarantees sufficient P availability at the fermentation site in the hindgut. Higher faecal losses of Na and K are hypothesized to be linked to a higher faecal bulk per DM intake in black rhinoceroses as compared with horses because of a generally lower digestive efficiency. There were no relevant differences in the absorption patterns of microminerals. In particular, there were no discernable differences in Fe absorption within the rhinoceroses for diets with and without tannin supplementation. Several of the zoo diets assessed in this study were deficient in Cu, Mn or Zn, and most contained excessive levels of Fe when compared with horse requirements. The findings of this study indicate that differences in mineral absorption between occur even between species of similar digestive anatomy; that in particular, Ca absorption might vary between hindgut fermenters with Ca:P ratio in their natural diet; that Na might be a particularly limiting factor in the ecology of free-ranging rhinoceroses; that moderate doses of tannins do not seem to markedly influence mineral absorption; and that diets for captive animals should contain adequate, but not excessive mineral levels.

Introduction

There has been recent interest in the mineral status of free-ranging and captive rhinoceroses (Dierenfeld et al., 2005). Zoo diets for rhinoceroses and elephants have been recorded that were deficient in different minerals when compared against horse maintenance requirements (Ange et al., 2001; Clauss et al., 2005a). It has been speculated that both a lack of mineral supplementation, e. g. for phosphorus, and an oversupplementation, e. g. with iron, could be involved in different disease syndromes observed in captive black rhinoceroses (*Diceros bicornis*) (Kock and Garnier, 1993; Paglia and Dennis, 1999).

Traditionally, the horse is considered the model animal for designing diets for other large hindgut fermenters, such as rhinoceroses or elephants (Oftedal et al., 1996). The validity of this concept has been confirmed for the grazing white (*Ceratotherium simum*) and Indian rhinoceros (*Rhinoceros unicornis*) (Kiefer, 2002; Clauss et al., 2005b), but not for elephants (Clauss et al., 2003) or the browsing black rhinoceros (Clauss et al., 2006a). Therefore, we wanted to generate data on the absorption of minerals in captive black rhinoceros, to facilitate a comparison with published data for domestic horses, to test whether differences in mineral absorption could be detected that would both have ecological relevance and necessitate a differentiated mineral supplementation regime for captive black rhinoceroses.

Black rhinoceroses are strict browsers (see Clauss et al., 2006a), and the mineral composition of their natural forage differs in several respects from both the natural diet of grazing equids and from horse recommendations. For example, the calcium (Ca) to phosphorus (P) ratio in forage of free-ranging black rhinoceros averages 14:1 (see *Discussion* for details), as compared with the 1.4:1 recommended for the maintenance of domestic horses (Meyer and Coenen, 2002). Therefore, it could be expected that different absorption efficiencies have evolved in black rhinoceroses as compared with grazers like horses.

The browse consumed by black rhinoceros additionally contains tannins (Loutit et al., 1987), which could in particular influence the absorption of iron (Paglia and Dennis, 1999). To test for an effect of moderate tannin supplementation on the apparent absorption (aA) of iron, two different tannin supplements were additionally used in the captive black rhinoceros of this study.

Methods

The principal setup of this study has been described in Clauss et al. (2005c, 2006a,b, 2007). Feeding trials were performed with eight black rhinoceroses from three zoological institutions. All animals were adult, and not reproducing (Table 1). Animals were kept individually, food intake was recorded by weighing offered feeds and leftovers for 7 days, and faecal

Table 1 Black rhinoceroses (*Diceros bicornis*) used for digestion trials. Body weights represent either actual weights or estimates (°). Diets used were conventional zoo diets with hay and concentrates (N), zoo diets supplemented with different proportions of browse (B), zoo diets with an addition of tannic acid to the pelleted compound feed (T) and zoo diets with an addition of quebracho to the pelleted compound feed (Q)

Animal no.	Studbook no.	Sex	Age (years)	BM (kg)	Diets fed	Study
1	451	f	10	1000/1065	N1, N2, T, Q	T. Woodfine (unpublished), this study
2	430	m	11	1093/1133	N1, N2, T, Q	T. Woodfine (unpublished), this study
3	533	m	6	1000°	N, T, Q	This study
4	532	f	7	1000°	N, T, Q	This study
5*	150	f	31	762	N1, N2, N3, T, Q	This study
6	217	f	≈30†	1000°	N1, N2, N3, T, Q	This study
7	318	m	19	1200°	N, N2, B1, B2, T	This study
8	662	m	5	900°	N1, N2, B1, B2, T	This study
9	428	f	10	1200°	N1, N2, T	T. Froeschle and M. Clauss (unpublished)
10	610	m	4	1200°	N1, N2, T	T. Froeschle and M. Clauss (unpublished)
11	240	f	27	1160	N1, N2, T	T. Froeschle and M. Clauss (unpublished)
12	438	m	9	1200°	N1, N2, T	T. Froeschle and M. Clauss (unpublished)
13	437	f	9	1200°	N1, N2, T	T. Froeschle and M. Clauss (unpublished)
14	376	m	12	1100	N	D. Paros and E. S. Dierenfeld (unpublished)
15	396	f	11	1200	N	D. Paros and E. S. Dierenfeld (unpublished)

*Animal had an oral abscess and received a particular diet (Hatt et al., 2004).
†Exact age unknown as animal was caught from the wild.

excretion by total collection for 5 days. Three to five different zoo rations were used per animal, consisting of varying proportions of roughage, concentrates and in some cases browse material (total trials, $n =$ 32). A total of eight animals (see Table 1) received basically identical diets twice, with the exception of the inclusion of 5% tannic acid (a source of hydrolysable tannins) in the pelleted diet compound; a total of six animals (see Table 1) received basically identical diets for a third time, with the exception of the inclusion of 5% quebracho (a source of condensed tannins). This resulted in an additional tannin source intake of 5–15 g/kg dry matter (DM). Adaptation periods for all diets lasted more than a month each. During the trial periods, the animals did not have access to mineral licks. A detailed description of all diets used in this study and in the unpublished studies, from which additional data were taken, is given in Clauss et al. (2006a, 2007).

To obtain representative faecal samples, the outer layer of dung balls was removed to avoid contamination of the sample. The rest of the material was thoroughly mixed, and a subsample representing 10% of the whole sample was taken and frozen at −20 °C. After thawing, all faecal samples were pooled per animal and feeding period. Representative samples of feeds and the pooled faecal samples were analysed for mineral content (Ca, P, Mg, Na, K, Fe, Mn, Cu, Zn and Co). All analyses were run in duplicate. To 0.5 g of sample, 5 ml of 65% HNO_3 was added for wet ashing (1200 mega High Performance Microwave, MLS, Milestone, Leutkirch, Germany). Mineral analysis was performed by inductively coupled plasma emission spectrometry (ICPES, JY66, Jobin Yvon, Longjumeau, France). As the results for Na were unexpected, new subsamples were dissolved by wet ashing and analysed by flame photometry (EFOX 5053, Eppendorf, Hamburg, Germany). There was no difference between results generated by the two methods. Here, therefore, only ICPES results are given. Additional data for adult, non-reproductive black rhinoceroses were available from three unpublished studies (D. Paros and E. S. Dierenfeld, $n = 2$; T. Woodfine and E. S. Dierenfeld, $n = 2$; T. Froeschle and M. Clauss, $n = 14$; not all minerals analysed in all cases), in which mineral levels had been measured by flame photometry and atomic absorption spectroscopy in different laboratories. These animals are also listed in Table 1; their diets were regular zoo diets based on hay and concentrates.

Apparent absorption of minerals was calculated using the formula, aA (%) = [mineral ingested (g) − mineral excreted (g)]/mineral ingested (g) × 100. Mineral content was plotted against absorbable mineral content in 100 g DM, and differences in the resulting regressions to those derived from literature data on domestic horses[1] as well as of calculated mean aA coefficients were tested by analysis of covariance (Huitema, 1980) and *U*-test, respectively, using the SPSS 12.0 statistical package (SPSS, Chicago, IL, USA). In these regressions, the regression slope (a) corresponds to the 'true' absorption coefficient, and the negative intercept (b) to the endogenous faecal losses (EFL) (Robbins, 1993). The significance level was set at p > 0.05.

Results

Dietary mineral contents, aA coefficients and the regression equations are summarized in Table 2; the respective data plots are depicted in Figs 1 and 2. Mean aA coefficients differed significantly for all macrominerals (Ca, P, Mg, Na and K), but not for microminerals (Fe, Mn, Cu and Zn).

Black rhinos had a significantly higher 'true' absorption efficiency for Ca and Mg than domestic horses. Regardless of a similar 'true' absorption efficiency for Na, black rhinos had significantly higher Na EFL (difference: 0.16 g/100 g DM). Potassium gave a similar pattern as Na, but in this case, the difference between the slopes was significant and therefore, intercepts could not be compared.

In general, comparisons were less conclusive for microminerals, with a larger data scatter, and less comparative data for domestic horses. For Co, no comparative horse data was available. The range of trace mineral content in captive black rhinoceros diets (Cu: 5.3–13.8 mg/kg DM; Mn: 30–117 mg/kg DM and Zn: 30–121 mg/kg DM) included diets that would be considered deficient in Cu, Mn or Zn for domestic horse maintenance. In contrast, Fe content (98–1009 mg/kg DM, mean: 374 mg/kg DM) was 2–25 times in excess of assumed horse maintenance requirements. The patterns in the graph plots did not differ in a systematic manner between black

[1]Data for domestic horses was taken from the following sources: Wolff (1884, 1887); Tangl (1902); Scheunert et al. (1923); Schryver et al. (1970, 1971); Wedemeyer (1970); Whitlock et al. (1970); Hintz and Schryver (1972, 1973, 1976); Ott et al. (1975); Weidenhaupt (1977); Mundt (1978); Pferdekamp (1978); Swartzman et al. (1978); Güldenhaupt (1979); Meyer and Ahlswede (1979); Schmidt (1980); Verthein (1981); Lindemann (1982); Meyer et al. (1982, 1993); Brems (1983); Günther (1984); Krull (1984); Teleb (1984); Smiley et al. (1985); Schulze (1987); Gomda (1988); Groenendyk et al. (1988); Kapusniak and Potter (1988); Bertone et al. (1989); Cymbaluk et al. (1989), Perez Noriega (1989), Cymbaluk (1990); Nehring (1991); Crozier et al. (1997); Stürmer (2005).

Table 2 Mineral absorption characteristics in domestic horses (*Equus caballus*, literature data; see *Methods* for sources) and black rhinoceroses (*Diceros bicornis*, data generated in the trials summarized in this study)

Mineral	Species	n*	Dietary mineral concentration, g/kg DM ± SD (minimum, maximum)	Apparent absorption†, % ± SD (minimum, maximum)	a‡	b‡	R†
Ca	*E. caballus*	85	9.2 ± 6.2 (0.7, 26.6)	26[a] ± 68 (−458, 70)	0.41[a]	−0.02	0.68***
	D. bicornis	50	8.2 ± 2.6 (3.6, 15.8)	80[b] ± 5 (65, 90)	0.84[b]	−0.03	0.97***
P	*E. caballus*	86	3.9 ± 2.2 (0.7, 13.9)	5[a] ± 28 (−123, 59)	0.45	−0.12[a]	0.65***
	D. bicornis	36	3.7 ± 0.9 (1.9, 5.3)	23[b] ± 10 (3, 50)	0.32	−0.03[b]	0.41***
Mg	*E. caballus*	162	1.8 ± 0.7 (0.2, 7.3)	35[a] ± 12 (−16, 67)	0.15[a]	0.03	0.17***
	D. bicornis	36	2.1 ± 0.6 (1.4, 4.0)	73[b] ± 6 (58, 84)	0.71[b]	0.00	0.92***
Na	*E. caballus*	163	2.4 ± 1.6 (0.1, 16.9)	56[a] ± 29 (−140, 95)	0.87	−0.06[a]	0.87***
	D. bicornis	36	3.9 ± 1.6 (1.5, 7.4)	15[b] ± 34 (−71, 72)	0.84	−0.22[b]	0.85***
K	*E. caballus*	166	15.2 ± 8.5 (0.5, 36.5)	78[a] ± 9 (45, 94)	0.88[a]	−0.11	0.98***
	D. bicornis	36	26.9 ± 7.8 (15.0, 43.7)	67[b] ± 13 (37, 88)	1.00[b]	−0.83	0.93***
Fe	*E. caballus*	18	258 ± 222 (77, 1083)	−42 ± 85 (−268, 54)	0.70[c]	−0.02	0.70***
	D. bicornis	50	374 ± 224 (98, 1009)	−40 ± 65 (−341, 32)	0.28[d]	−0.02	0.14***
Mn	*E. caballus*	18	107 ± 44 (45, 193)	−7 ± 28 (−69, 37)	0.23	−0.003[c]	0.09
	D. bicornis	36	65 ± 22 (30, 117)	5 ± 18 (−75, 28)	0.25	−0.001[d]	0.16***
Cu	*E. caballus*	21	18.9 ± 11.5 (4.0, 42.3)	23[c] ± 28 (−47, 69)	0.33	−0.03[a]	0.49***
	D. bicornis	50	9.5 ± 2.8 (5.3, 13.8)	14[d] ± 13 (−40, 35)	0.20	−0.05[b]	0.17***
Zn	*E. caballus*	21	64 ± 34 (17, 145)	−11[a] ± 35 (−122, 31)	0.00	−0.74[c]	0.00
	D. bicornis	36	54 ± 25 (30, 121)	19[b] ± 20 (−56, 48)	0.27	−0.38[d]	0.27***
Co	*D. bicornis*	29	0.49 ± 0.42 (0.21, 1.75)	9 ± 25 (−67, 41)	0.35	−0.01	0.83***

DM, dry matter; SD, standard deviation.

***Regression equations significant at p < 0.001 (regression analysis, *F*-test).

[a,b,c,d]Significant differences (a, b: p < 0.001; c, d: p < 0.05) in the respective parameter for this mineral (apparent absorption: *U*-test; a: ancova, test for interaction; b: ancova; test between groups).

*Number of observations.

†Defined as [mineral ingested (g) − mineral excreted (g)]/mineral ingested (g) × 100.

‡According to the regression equation: apparently absorbable mineral content = a × mineral content + b; unit: g/100 g DM for Ca, P, Mg, Na, K, Fe, Mn and mg/100 g DM for Cu, Zn, Co.

rhinos or domestic horses for Fe, Mn, Cu and Zn. There was no significant difference in the absorption pattern for Fe in black rhinoceroses between the three dietary tannin treatments (Fig. 3).

Discussion

Clearly, comparisons of measured aA for minerals, and in particular for trace minerals, must be regarded with caution, especially if results from different studies, or from different facilities, are pooled in one dataset. Apparent absorption coefficients are likely to reflect physiological regularities in the case of macrominerals; in the case of trace minerals, however, the potential influence of contaminations (of both diet and faeces), and differences in absorption in correspondence to the repletion state of the individual animal with the respective mineral, are factors that are likely to overrule any measurable correlation between dietary content and aA (Robbins, 1993). This difference between the macro- and the micro-

minerals is reflected in two characteristics of the dataset: the discrepancy in available data for domestic horses, and in the low aA coefficients for most trace minerals, which might reflect the fact that on average, the animals investigated were, at maintenance, in a metabolic balance of trace mineral absorption and endogenous faecal excretion. For this reason, trace minerals, in particularly Mn, have been suggested as alternative markers in digestion studies in primates, carnivores, edentates, rodents and reptiles, as their excretion should roughly equal intake at maintenance (reviewed in Hatt, 2003). With regards to the macrominerals, the uniformity of the data presented, in spite of the fact that data was pooled from different studies and facilities, supports the interpretation that the displayed patterns are indeed representative for black rhinoceroses. With respect to the statistical significance found in most comparisons, the physiological relevance of this difference should be assessed by the magnitude of the difference and a visual inspection of the scatter plots.
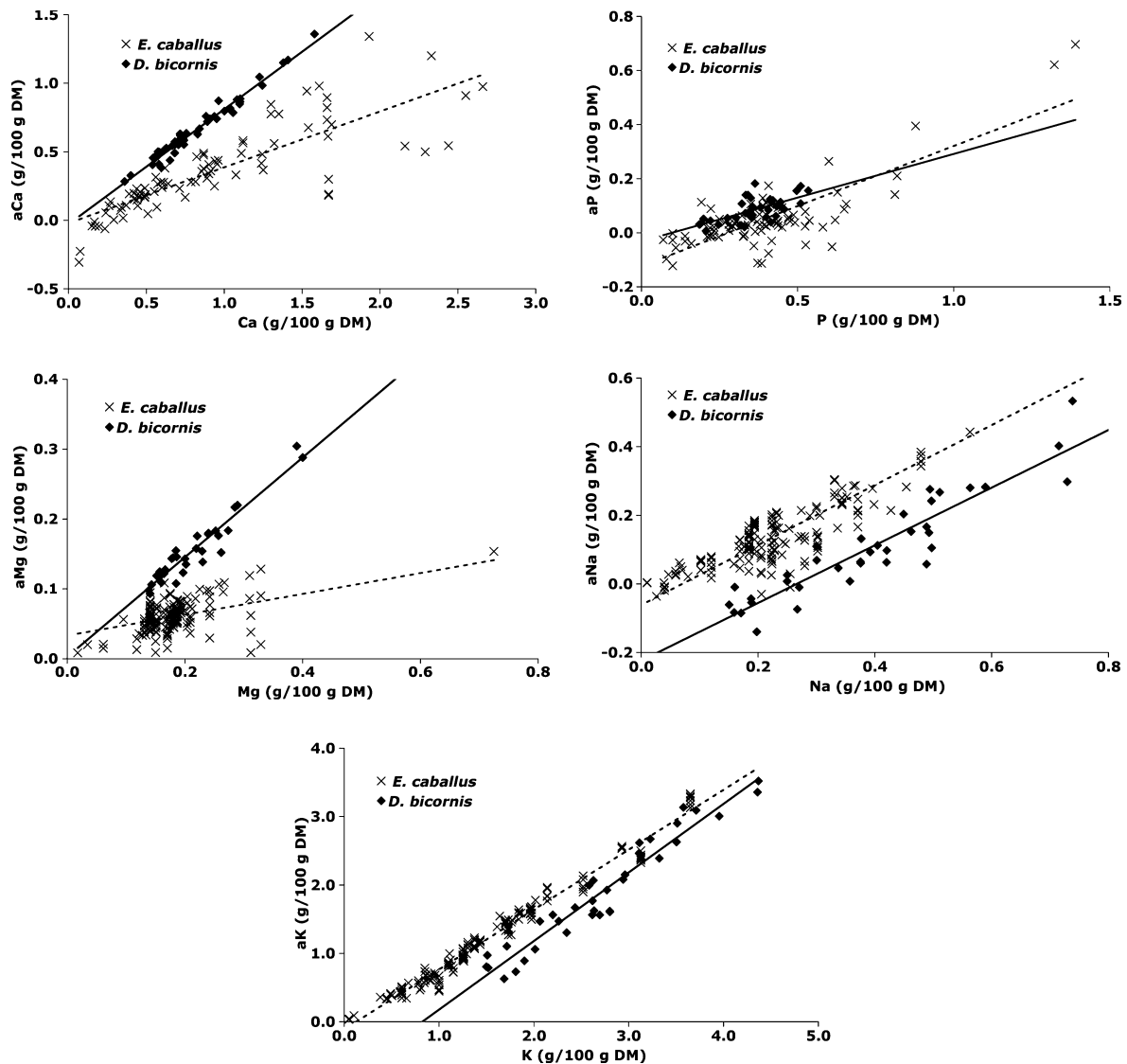
Fig. 1 Correlations between the mineral content and the absorbable mineral content (g/100 g dry matter) in horses (*Equus caballus*) and black rhinoceroses (*Diceros bicornis*) for Ca, P, Mg, Na and K. For significant differences between the species (see Table 1).

The data generated in this study suggest that differences in macromineral absorption exist between species, even in the face of a similar digestive anatomy (Stevens and Hume, 1995). Such differences could be explained by the fact that black rhinoceroses had to adapt, during evolution, to a diet that differed in mineral content from the diet usually ingested by grazing species (Table 3). When compared with horse maintenance recommendations, it is obvious that the natural diet of black rhinoceroses could be deficient in P, Na, Cu and Zn (Table 3). But, e. g. the Ca content of both native African browse and browse from the temperate zone is much higher than that usually found in grass (Table 3). The patterns observed in Fig. 1 for the two divalent cations Ca and Mg indicate a more efficient absorption of these minerals in black rhinoceroses as compared with horses. So far, the fact that most hindgut fermenting herbivores absorb more Ca from the gut than necessary to meet their requirements, and excrete the surplus in urine (Schryver et al., 1983; Clauss et al., 2005a), has not been explained satisfactorily (Leon and Belonje, 1979; Pitcher and Buffenstein, 1994). One potential explanation could be that Ca needs to be absorbed from the ingesta in the small intestine in large quantities to ensure P availability both in the small intestine and in the hindgut. Phosphorus is a major
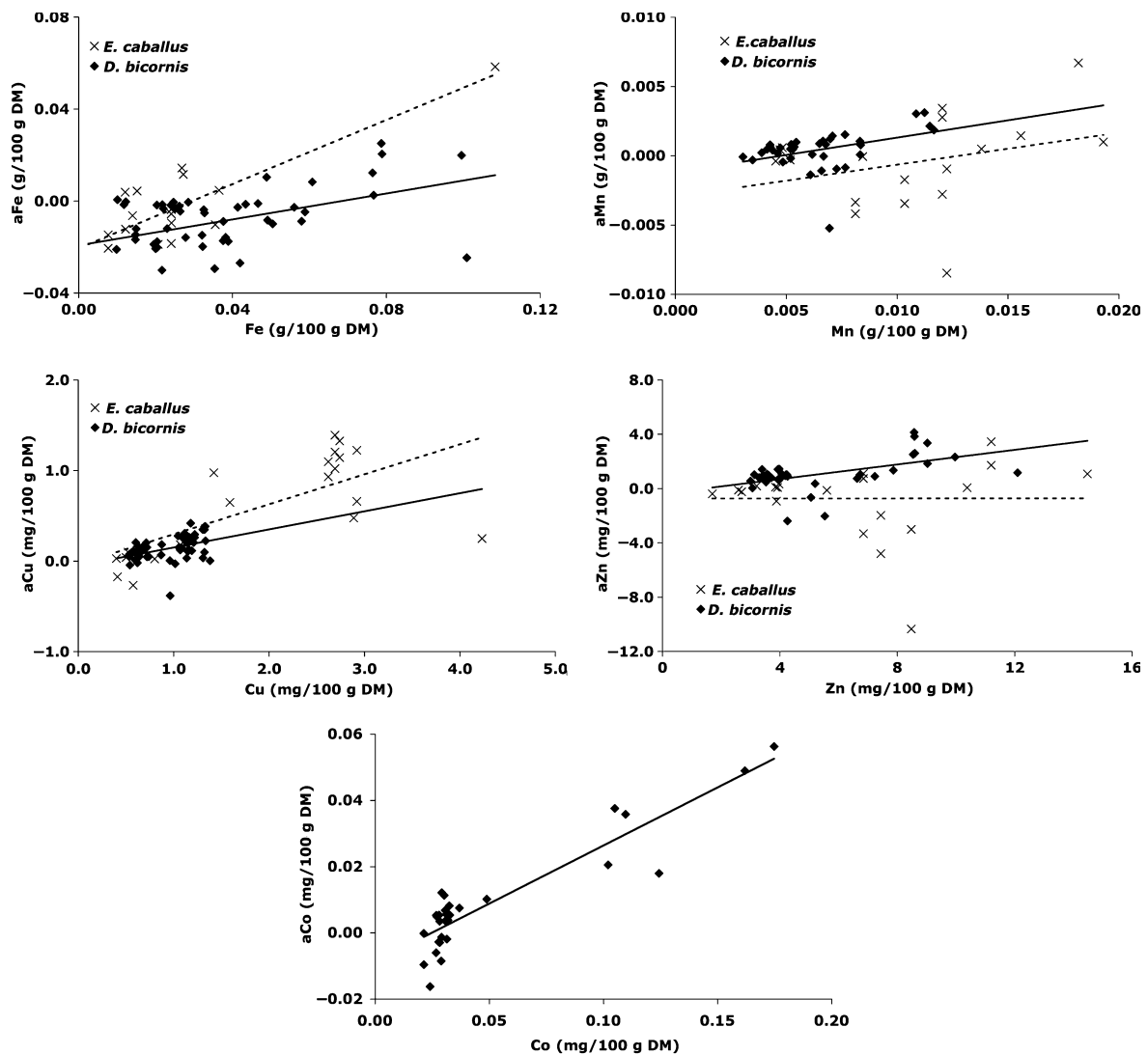
Fig. 2 Correlations between the mineral content and the absorbable mineral content (g/100 g dry matter) in horses (*Equus caballus*) and black rhinoceroses (*Diceros bicornis*) for Fe, Mn, Cu, Zn and Co. For significant differences between the species (see Table 1).

component of bacterial DM (Ca:P ratio in bacteria is approximately 1:100) (Cerkawski, 1976; Van Nevel and Demeyer, 1977; Merry and McAllan, 1983; Komisarczuk et al., 1987a) and therefore a limiting factor for bacterial growth and action. Low levels of available P can reduce microbial growth, organic matter and cellulose digestibility as well as the production of volatile fatty acids (Breves and Höller, 1987; Komisarczuk et al., 1987a,b) – a major energy source for herbivores. High intraluminal levels of Ca could, theoretically, render P unavailable through the formation of insoluble Ca–P complexes (Ritskes-Hoitinga et al., 2004). A species adapted to a diet with a particularly high Ca:P ratio, such as the black rhinoceros (c.f. Table 3), might therefore

be expected to display an even more effective Ca absorption efficacy (by active transport in the small intestine) than other hindgut fermenters adapted to diets with a lower Ca:P ratio. Comparative data from other herbivores is needed to corroborate this assumption. As the absorption mechanisms for Mg are known to be similar to those of Ca in both hindgut fermenters and ruminants (Hintz and Schryver, 1973; Reinhardt et al., 1988), it is understandable that they display a similar pattern in black rhinoceroses as well.

Regardless of the statistically significant differences in the calculated EFL for P (Table 2), the scatter plot does not suggest a general difference in P absorption between black rhinoceroses and horses (Fig. 1).
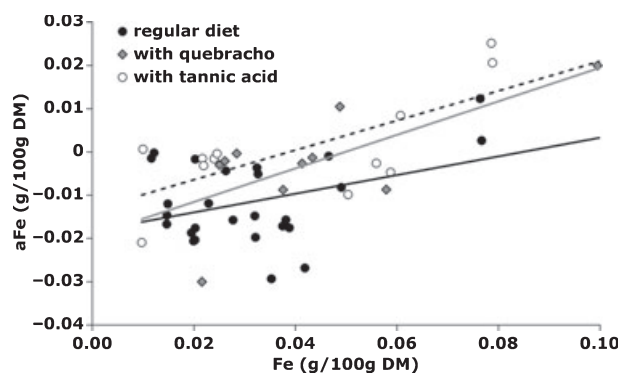
Fig. 3 Correlations between the iron (Fe) content and the absorbable Fe content [g/100 g dry matter (DM)] in black rhinoceroses (*Diceros bicornis*) for diets usually fed, and diets supplemented with either tannic acid or quebracho (at 5–15 g/kg DM).

Black rhinoceroses must have adapted to diets of low P content (Table 1), and diets in captivity do not seem to be particularly low in this mineral (Table 2). Therefore, the reported hypophosphatae-mia in diseased captive black rhinoceroses with hae-molytic anaemia or necrolytic dermatitis is unlikely to be primarily of dietary origin, unless one suspects that the P in captive diets is much less available than the P in native forages. Yet, both oral and par-enteral P supplementation has been successfully used to reverse these disease syndromes (Gillespie et al., 1990; Paglia, 1994; Paglia et al., 1996). Given the composition of the natural diet (low P), the absorption coefficients (not different from horses) and the current feeding practices (high P), it does not seem advisable to particularly supplement diets

of clinically healthy animals with P in excess of horse recommendations.

The most surprising finding of this study was the low aA coefficients measured for Na. The values generated in this study were confirmed by a second set of analyses with another laboratory method (see *Methods*), and the data from the unpublished studies matched the general pattern; these facts indicate that the results on Na are reliable. In general, Na is recognized as a limiting factor for many populations of free-ranging herbivores, and natural Na lick use has been observed in many wild herbivore species (Denton, 1984; Jones and Hanson, 1985). In our study, the difference between horses and black rhi-noceroses in Na absorption efficiency was not an effect of a less efficient 'true' absorption, but of higher EFL for Na. In terms of ecophysiology, the interesting question arises whether the high endog-enous faecal Na losses of black rhinoceroses are species-specific or represent an effect of feeding type, or increasing body size. Based on the theoretical grounds, Hellgren and Pitts (1997) suggested that dietary Na requirements in mammals should scale allometrically to the metabolic body weight. How-ever, comparative data is mostly lacking to test this or any other claim. Faecal Na excretion can be con-sidered an effect of both faecal bulk and faecal water content (Spiller et al., 1986; Robbins, 1993; McCabe et al., 1994, Kienzle et al., 2006). This means that the relation of faecal water and Na content is con-stant, and that therefore with increasing fresh faeces production, more Na is excreted, unless this increase is compensated by a reduction in faecal water

Table 3 Mineral content of the diet of free-ranging black rhinoceroses (*Diceros bicornis*) as compared with temperate browse, grass and recommendations for maintenance requirements in domestic horses

Mineral	Black rhinoceros forage* mean (range; n)	Temperate browse† mean (range; n)	Temperate grass† mean (range; n)	Maintenance recommendation for horses‡
g/kg DM				
Ca	17.6 (7.0–42.7; 25)	15.6 (9.3–23.8; 12)	4.8 (2.1–9.7; 14)	2.4
P	1.2 (0.4–2.0; 25)	2.7 (1.6–4.7; 12)	2.7 (2.0–3.1; 14)	1.7
Mg	2.7 (1.2–6.5; 18)	3.4 (2.0–6.9; 12)	1.5 (0.6–2.7; 13)	0.9
Na	0.14 (0.01–0.94; 18)	0.09 (0.01–0.31; 10)	0.05 (0.02–0.08; 5)	1.0
K	8.7 (2.8–17.7; 18)	14.9 (7.3–31.8; 11)	21.6 (16.0–27.0; 14)	3.0–6.0
mg/kg DM				
Fe	82 (12–215; 28)	120 (64–191; 12)	129 (46–391; 10)	40–70
Mn	60 (1–269; 28)	92 (14–248; 12)	74 (37–147; 9)	40
Cu	5 (1–12; 28)	11 (7–20; 12)	6 (4–9; 6)	10
Zn	14 (3–67; 28)	53 (13–121; 12)	19 (15–23; 5)	40
Co	6.3 (0.5–33.6; 10)	72 (21–159; 12)	26 (11–42; 5)	0.1

DM, dry matter.
*Joubert and Eloff (1971); Ghebremeskel et al. (1991); Dierenfeld et al. (1995).
†From DLG (1960).
‡From NRC (1989), Meyer and Coenen (2002).

content. Thus, species with generally lower apparent DM digestion coefficients and hence more faecal excretion should naturally have higher EFL for Na – such as elephants or black rhinoceroses as compared with white and Indian rhinos or horses (Clauss et al., 2003, 2005b, 2006a), or browsing as compared with grazing ruminants (Iason and Van Wieren, 1999, Pérez-Barberìa et al., 2004). In ruminants, another additional factor that influences Na EFL should be the faecal water content which can vary enormously between species (Clauss et al., 2004). In addition, free-ranging browsers might experience increased urinary Na losses as a consequence of secondary plant compounds in their natural diet (Robbins, 1993). To answer these questions, comparable data from other wildlife species would be warranted. To our knowledge, data on Na absorption in other large hindgut fermenters does not exist. Free-ranging elephants have been shown to be limited by available environmental Na, maybe more so than other (unspecified) species (Weir, 1972), and black rhinoceroses and elephants have been reported to exploit mineral licks (Weir, 1973; Mukinya, 1977), including the formation of caves by elephants (Bowell et al., 1996). In captivity, black rhinoceroses have low circulating Na levels as compared with values measured in free-ranging animals (Dierenfeld et al., 2005), and both black rhinoceroses and elephants (reviewed in Klemt, 2001) have circulating values that appear low compared with horse reference values. This might either reflect a particular physiological adaptation, or both high endogenous faecal Na losses and the observation that these animals are often not provided salt licks in captivity. As the absorption mechanisms for K are similar to those of Na, and endogenous faecal K losses are probably also a function of faecal bulk (Kienzle et al., 2006), the similarity in the patterns for these two minerals is understandable. Because of the difference in slopes, the relevant difference in EFL for K between horses and black rhinoceroses could not be evaluated statistically, but is numerically even larger than that for Na. However, in herbivores, K is not limiting (Robbins, 1993).

In contrast to the finding in macrominerals, and regardless of calculated statistical significances, a visual inspection of the micromineral scatter plots (Fig. 2) does not suggest fundamental differences in trace mineral absorption between horses and black rhinoceroses, with the potential exception of Mn. The potentially lower EFL of this element, as suggested in Table 2, would, however, be of no practical relevance. Iron absorption is a function of Fe status,

and captive black rhinoceroses are mostly overloaded with Fe (Paglia and Dennis, 1999; Dierenfeld et al., 2005). Therefore, the difference in Fe absorption between black rhinoceroses and horses, indicating a less efficient Fe absorption from the gut in black rhinoceroses (Table 2), might be understandable. It should be noted, however, that the steeper slope in horses results from one extreme measurement only (Fig. 2). The fact that black rhinoceroses are probably exclusively overloaded with Fe in captivity, linked with dietary Fe concentrations, explains why potential differences in Fe absorption are difficult to detect in conventional absorption trials. With respect to trace minerals, the composition of the diets used in captivity appears to be of primary interest, with potentially deficient contents in Mn, Cu or Zn in several, and extremely excessive Fe provision in most cases (except for grass-only diets). Low Mn and Cu status has been documented in individual captive animals (Dierenfeld et al., 2005). Although direct evidence for a correlation of excessive dietary Fe provision and iron overload in black rhinoceroses remains to be produced, it appears reasonable, in the face of the high incidence of the problem, to make any possible effort to reduce dietary Fe levels to horse maintenance requirements.

Tannin supplementation in this study did not have any detectable influence on Fe (or any other mineral) absorption patterns. On the one hand, this might reflect dietary tannin levels (0.5–1.5% DM) that were still low compared with values measured in African browse (e.g. up to 5.7%, Atkinson, 1995). Although it is known that dietary tannins will reduce iron absorption in other species (Brune et al., 1989), to our knowledge, no dose-related data exists from which an effect in black rhinoceros could be estimated. On the other hand, the tannin supplementation led to an increased production of tannin-binding salivary proteins in the same animals (Clauss et al., 2005c). This suggests that even such a moderate tannin supplementation as used in this study can trigger a physiological response. Additionally, the antioxidant capacity of the faeces of the animals of this study increased significantly on the quebracho tannin, but not on the tannic acid supplementation (Clauss et al., 2006b). It could not be demonstrated that this effect was due to an increased binding of Fe in the ingesta. Whether dietary tannins reduce the availability of Fe in black rhinoceros can be assumed but remains to be demonstrated.

In conclusion, the results of this study indicate significant differences in macromineral absorption between black rhinoceroses and horses that are of

ecophysiological relevance. For the management of captive black rhinoceroses, the results imply that diets designed according to horse requirements should be adequate, with the exception of Na. Therefore, additional salt licks should be provided. Given the particularly effective Ca absorption in black rhinoceroses, which is interpreted as an adaptation to the high Ca:P ratios in their native forage, the use of lucerne hay in captivity appears adequate. It should be assured that trace mineral levels, especially those of Cu and Zn, are not lower, and those of Fe not excessively higher than the recommended values for horse maintenance.

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References

Ange, K.; Crissey, S. D.; Doyle, C.; Lance, K.; Hintz, H., 2001: A survey of African and Asian elephant diets and measured body dimensions compared to their estimated nutrient requirements. *Proceedings of the Nutrition Advisory Group (NAG)* **4**, Lake Buena Vista, FL, 5–14.

Atkinson, S. J., 1995: *Maintenance of captive black rhinoceros (Diceros bicornis) on indigenous browse in Zimbabwe: energetics, nutrition and implications for conservation.* MSc, University of Zimbabwe, Harare.

Bertone, A. L.; Van Soest, P. J.; Stashak, T. S., 1989: Digestion, fecal, and blood variables associated with extensive colon resection in the horse. *American Journal of Veterinary Research* **50**, 253–258.

Bowell, R. J.; Warren, A.; Redmond, I., 1996: Formation of cave salts and utilization by elephants in the Mount Elgon region, Kenya. In: J. D. Appleton, R. Fuge, G. J. H. McCall (eds), *Environmental Geochemistry and Health.* Geological Society Special Publication No. 113, London, pp. 63–79.

Brems, R., 1983: *Untersuchungen zur Verdaulichkeit verschiedener Rationen bei aktiv eingesetzten Sportpferden mit Hilfe einer modifizierten Indikatormethode und Überprüfung des Einflusses auf Gesundheit und Leistung.* Dissertation, University of Giessen, Germany.

Breves, G.; Höller, H., 1987: Effects of dietary phosphorus depletion in sheep on dry matter and organic matter digestibility. *Journal of Animal Physiology and Animal Nutrition* **58**, 281–286.

Brune, M.; Rossander, L.; Hallberg, L., 1989: Iron absorption and phenolic compounds: importance of different phenolic structures. *European Journal of Clinical Nutrition* **43**, 547–558.

Cerkawski, J., 1976: Chemical composition of microbial matter in the rumen. *Journal of the Science of Food and Agriculture* **27**, 621–632.

Clauss, M.; Loehlein, W.; Kienzle, E.; Wiesner, H., 2003: Studies on feed digestibilities in captive Asian elephants. *Journal of Animal Physiology and Animal Nutrition* **87**, 160–173.

Clauss, M.; Lechner-Doll, M.; Streich, W. J., 2004: Differences in the range of faecal dry matter content between feeding types of captive wild ruminants. *Acta Theriologica* **49**, 259–267.

Clauss, M.; Polster, C.; Kienzle, E.; Wiesner, H.; Baumgartner, K.; von Houwald, F.; Streich, W. J.; Dierenfeld, E. S., 2005a: Energy and mineral nutrition and water intake in the captive Indian rhinoceros. *Zoo Biology* **24**, 1–14.

Clauss, M.; Polster, C.; Kienzle, E.; Wiesner, H.; Baumgartner, K.; von Houwald, F.; Streich, W. J.; Dierenfeld, E. S., 2005b: Studies on digestive physiology and feed digestibilities in captive Indian rhinoceros. *Journal of Animal Physiology and Animal Nutrition* **89**, 229–237.

Clauss, M.; Gehrke, J.; Hatt, J. M.; Dierenfeld, E. S.; Flach, E. J.; Hermes, R.; Castell, J.; Streich, W. J.; Fickel, J., 2005c: Tannin-binding salivary proteins in three captive rhinoceros species. *Comparative Biochemistry and Physiology. A, Comparative Physiology* **140**, 67–72.

Clauss, M.; Castell, J. C.; Kienzle, E.; Dierenfeld, E. S.; Flach, E. J.; Behlert, O.; Ortmann, S.; Streich, W. J.; Hummel, J.; Hatt, J. M., 2006a: Digestion coefficients achieved by the black rhinoceros (*Diceros bicornis*), a large browsing hindgut fermenter. *Journal of Animal Physiology and Animal Nutrition* **90**, 325–334.

Clauss, M.; Pellegrini, N.; Castell, J. C.; Kienzle, E.; Dierenfeld, E. S.; Hummel, J.; Flach, E. J.; Streich, W. J.; Hatt, J. M., 2006b: Antioxidant status of feces of captive black rhinoceros (*Diceros bicornis*) in relation to dietary tannin supplementation. *The Journal of Veterinary Medical Science Series A* **53**, 319–322.

Clauss, M.; Castell, J.C.; Kienzle, E.; Dierenfeld, E.S.; Flach, E. J.; Behlert, O.; Ortmann, S.; Streich, W. J.; Hummel, J.; Hatt, J.-M., 2007: The influence of tannin supplementation on digestive performance in captive

black rhinoceros (*Diceros bicornis*). *Journal of Animal Physiology and Animal Nutrition* (in press).

Crozier, J. A.; Allen, V. G.; Jack, N. E.; Fontenot, J. P.; Cochran, M.A., 1997: Digestibility, apparent mineral absorption, and voluntary intake by horses fed alfalfa, tall fescue, and Caucasian bluestem. *Journal of Animal Science* 75, 1651–1658.

Cymbaluk, N. F., 1990: Comparison of forage digestion by cattle and horses. *Canadian Journal of Animal Science* 70, 601–610.

Cymbaluk, N. F.; Christison, G. I.; Leach, D. H., 1989: Nutrient utilization by limit- and ad libitum-fed growing horses. *Journal of Animal Science* 67, 414–425.

Denton, D., 1984: *The Hunger for Salt*. Springer, Berlin.

Dierenfeld, E. S.; du Toit, R.; Braselton, W.E., 1995: Nutrient composition of selected browses consumed by black rhinoceros in the Zambezi Valley, Zimbabwe. *Journal of Zoo and Wildlife Medicine* 26, 220–230.

Dierenfeld, E. S.; Atkinson, S.; Craig, A. M.; Walker, K. C.; Streich, W. J.; Clauss, M., 2005: Mineral concentrations in serum/plasma and liver tissue of captive and free-ranging rhinoceros species. *Zoo Biology* 24, 51–72.

DLG, 1960: *Futterwerttabellen der DLG Mineralstoffe*. DLG-Verlags-GmbH, Frankfurt.

Ghebremeskel, K.; Williams, G.; Brett, R. A.; Burek, R.; Harbige, L. S., 1991: Nutrient composition of plants most favoured by black rhinoceros in the wild. *Comparative Biochemistry and Physiology. A, Comparative Physiology* 98, 529–534.

Gillespie, D.; Burton, M.; Gosselin, S.; Godfrey, B.; Munson, L., 1990: An unusual case of ulcerative stomatitis and prolonged pregnancy in a black rhinoceros. *Proceedings of AAZV*, South Padre Island, TX, 361–363.

Gomda, Y. M., 1988: *Untersuchungen über die renale, fäkale und kutane Wasser- und Elektrolytausscheidung bei Pferden in Abhängigkeit von Fütterungszeit, Futtermenge sowie Bewegungsleistung*. Dissertation, TiHo Hannover, Germany.

Groenendyk, S.; English, P. B.; Abetz, I., 1988: External balance of water and electrolytes in the horse. *Equine Veterinary Journal* 20, 189–193.

Güldenhaupt, V., 1979: *Veträglichkeit und Verdaulichkeit eines Alleinfutters für Pferde in Kombination mit Stroh*. Dissertation, TiHo Hannover, Germany.

Günther, C., 1984: *Untersuchungen über die Verdaulichkeit und Verträglichkeit von Hafer, Quetschhafer, Gerste und Mais beim Pferd*. Dissertation, TiHo Hannover, Germany.

Hatt, J. M., 2003: Markers in zoo animal nutrition with special emphasis on n-alkanes. In: A. Fidgett, M. Clauss, U. Ganslosser, J. M. Hatt, J., Nijboer (eds), *Zoo Animal Nutrition*, Vol. II. Filander, Fürth, Germany, pp. 167–180.

Hatt, J. M.; Wenker, C.; Castell, J.; Clauss, M., 2004: Dietary and veterinary management of a lingual abscess in a geriatric captive black rhino (*Diceros bicornis*) with iron storage disease. *Proceedings of the European Association of Zoo Wildlife Veterinaria* 5, 339–340.

Hellgren, E. C.; Pitts, W. J., 1997: Sodium economy in white-tailed deer (*Odocoileus virginianus*). *Physiological Zoology* 70, 547–555.

Hintz, H. F.; Schryver, H. F., 1972: Availability to ponies of calcium and phosphorus from various supplements. *Journal of Animal Science* 34, 979–980.

Hintz, H. F.; Schryver, H. F., 1973: Magnesium, calcium and phosphorus metabolism in ponies fed varying levels of magnesium. *Journal of Animal Science* 37, 927–930.

Hintz, H. F.; Schryver, H. F., 1976: Potassium metabolism in ponies. *Journal of Animal Science* 42, 637–643.

Huitema, B. E., 1980: *The Analysis of Covariance and Alternatives*. John Wiley & Sons, New York.

Iason, G.; Van Wieren, S., 1999: Digestive and ingestive adaptations of mammalian herbivores to low-quality forage. In: H. Olff, V. Brown, R. Drent (eds), *Herbivores: Between Plants and Predators*. Blackwell Scientific, Oxford, pp. 337–369.

Jones, R. L.; Hanson, H. C., 1985: *Mineral Licks, Geophagy, and Biogeochemistry of North American Ungulates*. Iowa State University Press, Ames.

Joubert, E.; Eloff, F. C., 1971: Notes on the ecology and behaviour of the black rhinoceros in South West Africa. *Madoqua* 1, 5–53.

Kapusniak, L. J.; Potter, G. D., 1988: Calcium, magnesium and phosphorus absorption from the small and large intestine of ponies fed elevated amounts of aluminium. *Journal of Equine Veterinary Science* 8, 305–309.

Kiefer, B., 2002: *Qualität und Verdaulichkeit der vom Breitmaulnashorn aufgenommenen Nahrung*. Dissertation, LMU Munich, Germany.

Kienzle, E.; Dobenecker, B.; Wichert, B.; Schuster, S., 2006: Effect of fecal water and dry matter excretion on fecal mineral excretion in dogs studied in a fibre model. *Journal of Nutrition* 136, 2001S–2003S.

Klemt, A., 2001: *Klinisch-chemische Blutwerte Asiatischer Elefanten aus Zoologischen Gärten Deutschlands*. Dissertation, University of Leipzig, Germany.

Kock, R. A.; Garnier, J., 1993: Veterinary management of three species of rhinoceros in zoological collections. In: O. A. Ryder (ed.), *Rhinoceros Biology and Conservation*. San Diego Zoological Society, San Diego, CA, USA, pp. 325–338.

Komisarczuk, S.; Durand, M.; Beaumatin, P.; Hannequart, G., 1987a: Effects of phosphorus deficiency on rumen microbial activity associated with the solid and liquid phase of a fermenter (RUSITECH). *Reproduction, Nutrition, Development* 27, 907–919.

Komisarczuk, S.; Merry, R.; McAllan, A., 1987b: Effect of different levels of phosphorus on rumen microbial fermentation and synthesis determined using a continuous culture technique. *British Journal of Nutrition* **57**, 279–290.

Krull, H. D., 1984: *Untersuchungen über Aufnahme und Verdaulichkeit von Grünfutter beim Pferd*. Dissertation, TiHo Hannover, Germany.

Leon, B.; Belonje, P. C., 1979: Calcium, phosphorus and magnesium excretion in the rock hyrax. *Comparative Biochemistry and Physiology. A, Comparative Physiology* **64**, 67–72.

Lindemann, G., 1982: *Untersuchungen über den Einfluss von Lactose- und Stärkezulagen auf die Verdaulichkeit von NH_3-aufgeschlossenem Stroh beim Pferd*. Dissertation, TiHo Hannover, Germany.

Loutit, B. D.; Louw, G. N.; Seely, M. K., 1987: First approximation of food preferences and the chemical composition of the diet of the desert-dwelling black rhinoceros (*Diceros bicornis*). *Madoqua* **15**, 35–54.

McCabe, R.; Smith, M. J.; Dwyer, T. M., 1994: Faecal dry weight and potassium are related to faecal sodium and plasma aldosterone in rats chronically fed on varying amounts of sodium or potassium chlorides. *British Journal of Nutrition* **72**, 325–337.

Merry, R.; McAllan, A., 1983: A comparison of the chemical composition of mixed bacteria harvested from the liquid and solid fraction of rumen bacteria. *British Journal of Nutrition* **50**, 701–709.

Meyer, H.; Ahlswede, L., 1979: Untersuchungen über den Na-Stoffwechsel des Pferdes. *Zbl Veterinary Medicine A* **26**, 212–220.

Meyer, H.; Coenen, M., 2002: *Pferdefütterung*. Parey, Berlin.

Meyer, H.; Schmidt, M.; Lindemann, G.; Muus, H., 1982: Praecaecale und postileale Verdaulichkeit von Mengen- (Ca, P, Mg) und Spurenelementen (Cu, Zn, Mn) beim Pferd. *Advances in Animal Phsyiology and Animal Nutrition* **13**, 61–69.

Meyer, H.; Stadermann, B.; Radicke, S.; Kienzle, E.; Nyari, A., 1993: Untersuchungen zum Einfluss der Futterart auf Füllung und Zusammensetzung des Verdauungskanals sowie postprandiale Blut- und Harnparameter. *Pferdeheilkd* **9**, 15–25.

Mukinya, J. G., 1977: Feeding and drinking habits of the black rhinoceros in Masai Mara Game Reserve. *East African Wildlife Journal* **15**, 125–128.

Mundt, H. C., 1978: *Untersuchungen über die Verdaulichkeit von aufgeschlossenem Stroh für Pferde*. Dissertation, TiHo Hannover, Germany.

National Research Council, 1989: *Nutrient Requirements of Horses*, 5th edn. National Academy Press, Washington.

Nehring, T., 1991: *Einfluss der Futterart auf die Nettoabsorption von Calcium sowie Magnesium und Phosphor beim Pferd*. Dissertation, TiHo Hannover, Germany.

Oftedal, O. T.; Baer, D. J.; Allen, M. E., 1996: The feeding and nutrition of herbivores. In: D. G. Kleimann, M. E. Allen, K. V., Thompson, S. Lumpkin (eds), *Wild Mammals in Captivity: Principles and Techniques*. University of Chicago Press, Chicago, pp. 129–138.

Ott, E. A.; Feater, J. P.; Panco, P. J., 1975: Effect of calcium and phosphorus levels on availability of trace minerals. *Proceedings of Equine Nutrition and Physiology Symposium* **4**, 61–67.

Paglia, D. E., 1994: Rationale for phosphate supplementation in prevention and therapy of haemolytic anemia in the African black rhinoceros. *Proceedings of the Zoological Society of South Africa, Symposium on Contemporary South African Zoology*, Pietermariztburg, pp. 107.

Paglia, D. E.; Dennis, P., 1999: Role of chronic iron overload in multiple disorders of captive black rhinoceroses. *Proceedings of AAZV*, Columbus, Ohio, 163–171.

Paglia, D. E.; Miller, R. E.; Renner, S. W., 1996: Is impairment of oxidant neutralization the common denominator among diverse diseases of black rhinoceroses? *Proceedings of AAZV*, Puerto Vallart, Mexico, 37–41.

Perez Noriega, H. R., 1989: *Untersuchungen über den postprandialen Wasser- und Elektrolythaushalt des Pferdes unter Variation des Wasser- und Futterangebotes*. Dissertation, TiHo Hannover, Germany.

Pérez-Barberìa, F. J.; Elston, D. A.; Gordon, I. J.; Illius, A. W., 2004: The evolution of phylogenetic differences in the efficiency of digestion in ruminants. *Proceedings of the Royal Society London B* **271**, 1081–1090.

Pferdekamp, M., 1978: *Einfluss steigender Proteinmengen auf den Stoffwechsel des Pferdes*. Dissertation, TiHo Hannover, Germany.

Pitcher, T.; Buffenstein, R., 1994: Passive uptake in the small intestine and active uptake in the hindgut contribute to highly efficient mineral metabolism of the common mole rat. *British Journal of Nutrition* **71**, 573–582.

Reinhardt, T. A.; Horst, R. L.; Goff, J. P., 1988: Calcium, phosphorus, and magnesium homeostasis in ruminants. *The Veterinary Clinics of North America. Food Animal Practice* **4**, 331–350.

Ritskes-Hoitinga, J.; Grooten, H.; Wienk, K.; Peters, M.; Lemmens, A.; Beynen, A., 2004: Lowering dietary phosphorus concentrations reduces kidney calcification, but does not adversely affect growth, mineral metabolism, and bone development in growing rabbits. *British Journal of Nutrition* **91**, 367–376.

Robbins, C. T., 1993: *Wildlife Feeding and Nutrition*. Academic Press, San Diego.

Scheunert, A.; Schattke, A.; Weise, M., 1923: Über den Kalk- und Phosphorsäurestoffwechsel des Pferdes bei normaler Fütterung. *Biochemische Zeitschrift* **139**, 1–10.

Schmidt, M., 1980: *Untersuchungen über die Verträglichkeit und Verdaulichkeit eines pelletierten Mischfutters für Pferde*

in Kombination mit Heu und NH$_3$-aufgeschlossenem Stroh. Dissertation, TiHo Hannover, Germany.

Schryver, H. F.; Craig, P. H.; Hintz, H. F., 1970: Calcium metabolism in ponies fed varying levels of calcium. *Journal of Nutrition* **100**, 955–964.

Schryver, H. F.; Hintz, H. F.; Craig, P. H., 1971: Calcium metabolism in ponies fed a high phosphorus diet. *Journal of Nutrition* **101**, 259–264.

Schryver, H. F.; Foose, T. J.; Williams, J.; Hintz, H. F., 1983: Calcium excretion in the feces of ungulates. *Comparative Biochemistry and Physiology. A, Comparative Physiology* **74**, 375–379.

Schulze, K., 1987: *Untersuchungen zur Verdaulichkeit und Energiebewertung von Mischfuttermitteln für Pferde*. Dissertation, TiHo Hannover, Germany.

Smiley, L. K.; Greene, L. W.; Potter, G. D.; Arnold, F. F., 1985: Digestibility of calcium in alfalfa hay in the equine small intestine. *Proceedings of Equine Nutrition and Physiology Symposium* **9**, 124–128.

Spiller, G. A.; Story, J. A.; Wong, L. G.; Nunes, J. D.; Alton, M.; Petro, M. S.; Furomoto, J. D.; Whittam, J. H.; Scala, J., 1986: Effect of increasing levels of hard wheat fiber on fecal weight, minerals, steroids and gastrointestinal transit time in healthy young women. *Journal of Nutrition* **116**, 778–785.

Stevens, C. E.; Hume, I. D., 1995: *Comparative Physiology of the Vertebrate Digestive System*. Cambridge University Press, Cambridge.

Stürmer, K., 2005: *Untersuchungen zum Einfluss der Fütterung auf den Säure-Basen-Haushalt bei Ponies*. Dissertation, University of Munich, Germany.

Swartzman, J. A.; Hintz, H. F.; Schryver, H. F., 1978: Inhibition of calcium absorption in ponies fed diets containing oxalic acid. *American Journal of Veterinary Research* **39**, 1621–1623.

Tangl, F., 1902: Mitteilung aus der Königlich Ungarischen tierphysiologischen Versuchsstation in Budapest. III. Beitrag zur Kenntnis des anorganischen Stoffwechsels beim Pferde. *Landwirtsch Versuchsstation* **57**, 367–404.

Teleb, H., 1984: *Untersuchungen über den intestinalen Ca-Stoffwechsel bei Pferd nach variierender Ca-Zufuhr und einer Oxalatzulage*. Dissertation, TiHo Hannover, Germany.

Van Nevel, C.; Demeyer, D., 1977: Determination of rumen microbial growth in vitro from 32P-labelled phosphate incorporation. *British Journal of Nutrition* **38**, 101–114.

Verthein, B., 1981: *Auswirkungen einer Enzymgabe auf die Futterverdaulichkeit beim Pferd*. Dissertation, TiHo Hannover, Germany.

Wedemeyer, H. C. v., 1970: *Untersuchungen zum Calcium-, Phosphor- und Natrium-Umsatz des erwachsenen Pferdes*. Dissertation, University of Göttingen, Germany.

Weidenhaupt, K., 1977: *Untersuchungen zum Kaliumstoffwechsel des Pferden*. Dissertation, TiHo Hannover, Germany.

Weir, J. S., 1972: Spatial distribution of elephants in an African National Park in relation to environmental sodium. *Oikos* **23**, 1–13.

Weir, J. S., 1973: Exploitation of water soluble soil sodium by elephants in Murchison Falls National Park, Uganda. *East African Wildlife Journal* **11**, 1–7.

Whitlock, R. H.; Schryver, H. F.; Krook, L.; Hintz, H. F.; Craig, P. H., 1970: The effects of dietary calcium in horses. *Proceedings of the American Association of Equine Practitioners*, Lexington, Kentucky, **16**, 127–134.

Wolff, E., 1884: Pferde-Fütterungsversuche, 13. Bericht: Vergleichende Versuche mit Pferd und Hammel über die Verdaulichkeit von Wiesenheu und Kleeheu, nebst Beobachtungen über die Ausscheidung der Mineralstoffe bei Pferden. *Landw. Jb.* **13**, 271–290.

Wolff, E., 1887: Pferde-Fütterungsversuche, 15. Bericht: Versuche über die Leistungsfähigkeit des Pferdes bei stickstoffreicherem und stickstoffärmeren Futter, sowie über den Kreislauf der Mineralstoffe im Körper dieses Tieres. *Landw. Jb.* **16** (Suppl. 3), 1,49,95.