Rumen ciliates in the African (Cape) buffalo (*Syncerus caffer caffer*) living in the vicinity of the Orpen Gate entrance into Kruger National Park, South Africa

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Abstract

Samples of rumen contents were obtained from 10 African (Cape) buffalo living in the vicinity of the Orpen Gate entrance into Kruger National Park in South Africa. Total number of ciliate protozoa per animal ranged from 3.15 to 23.25 x 10^3. Forty three different species and forms were observed, of which 35 are a new host record. The total number of species and forms per animal varied from 10 to 17. *Eudiplodinium maggii* occurred in all 10 animals, followed by *Dasytricha ruminantium* in 9 animals. *Diplodinium posterovesiculatum, Ediplodinium magnodentatum* and *Ostracodinium mammosum* were present in 7 animals with all other species and forms occurring in 5 or less animals.

Key words: African buffalo, ciliate protozoa, rumen, South Africa, *Syncerus caffer*

Introduction

The African buffalo (*Syncerus caffer*) is not closely related to the Asian water buffalo or any of the other larger bovines. There are five subspecies, with *Syncerus caffer caffer* being the typical subspecies which is found primarily in South and East Africa. It is the largest of the subspecies, black in color and commonly called the Cape buffalo. The animals sampled in this study would belong to this subspecies. The other four subspecies are smaller in size and are found in Central and West Africa or the mountains of East Africa. Other than the report by Dogiel in 1932, on rumen protozoa from a single African buffalo in Uganda, no information was found on the fauna of this species. Based on location, it is presumed his animal was of the same subspecies. He reported a total of 21 species of ciliate protozoa.

Material and Methods

The samples were collected in a radius of 50 km from the Hans Hoheisen Wildlife Research Station (24^{o}28'55.60"S, 31^{o}23'09.40"E) next to the Orpen Gate of the Kruger National Park in South Africa during August till October of the year 2012.

For each animal, handfuls of rumen contents were taken randomly and the fluid squeezed out by making a fist with the thumb down so the fluid can run down into the container. This was repeated until a 200 ml sample was obtained. One hundred ml of Formalin was added to each sample for preservation.

In the laboratory, a 10ml subsample of the 300 ml protozoa sample was collected by using a pipette to stir the total sample and collecting 1ml at a time until 10 ml was acquired. Two drops of Brilliant Green Stain were added to this 10 ml sample and left overnight. Using a finnpipet, a 0.025 ml of the 10 ml stained sample was dropped on a microscope glass slide, then fitted with a cover slip and examined under a standard microscope for identification and counts of protozoa. However, protozoa concentrations at this dilution were too high for an accurate count. Thus, 4 ml of the 10 ml subsample was diluted to 10 ml with water and a 0.25 ml of this dilution counted as described above. Protozoa per ml of rumen fluid was calculated by multiplying the results of the counts by 150 (40 x 2.5 x 1.5).

Results

Concentration and generic distribution of protozoa in the 10 African (Cape) buffalo are shown in Table 1. Total concentration of protozoa ranged from 3.15 to 23.25 x 10^3, with a mean value of 8.79 ± 5.44 x 10^3. In general, the overall highest percentages of protozoa were in the genus *Eudiplodinium* (34.4 ± 9.3%), followed closely by *Dasytricha* (28.7± 16.1%). Animals 4 and 8 were the exception: *Eudiplodinium,* 30.9 and 12 % and *Dasytricha*, 54.8 and 54.0 %, respectively. *Diplodinium* were present in 9 animals (13.1 ± 8.2%) followed by *Ostracodinium*, (10.3 ± 7.0 %), present in all 10 animals. Of particular interest is the low overall percentage of *Entodinium,* 8.1 ± 10.3 %. Two animals, 5 and 8, were devoid of *Entodinium* and only one animal, 6, exceeded 13.7 %. *Metadinium, Enoploplastron and Epidinium* occurred sporadically in low percentages (1.4 ± 2.4 %, 0.4 ± 1.4 % and 3.4 ± 5.0 %, respectively).

A total of 43 different species and forms of protozoa were observed in the 10 buffalo, Table 2. The number of species and forms observed in individual animals varied between 10 and 17 with a mean of 12 ± 2.4. *Eudiplodinium maggii* was present in all 10 buffalo, and comprised

Table 1. Concentration and generic distribution of protozoa in rumen contents of 10 African (Cape) buffalo

	Buffalo number										Overall
	1	2	3	4	5	6	7	8	9	10	mean
Total protozoa per ml (x10$^{3)}$	4.20	9.45	10.95	23.25	4.65	3.15	7.50	7.50	10.65	6.60	8.79±5.44
Generic distribution (%)											
Entodinium	10.8	7.9	13.7	0.6	0	33.4	4	0	4.2	6.8	8.1±10.3
Diplodinium	25	17.5	15	8.9	25.6	4.8	14	10	9.8	0	13.1±8.2
Eudiplodinium	39.1	35	34.2	30.9	38.6	42.9	28	12	42.3	40.9	34.4±9.3
Ostracodinium	3.6	6.4	6.8	3.1	6.4	14.3	16	22	19.6	4.6	10.3±7.0
Metadinium	0	3.2	0	0	6.4	4.8	0	0	0	0	1.4±2.4
Enoploplastron	0	0	0	0	0	0	0	0	0	4.5	0.4±1.4
Epidinium	0	0	2.7	1.2	0	0	14	2	2.8	11.4	3.4±5.0
Dasytricha	21.4	30.2	27.4	54.8	22.6	0	24	54	21.1	31.8	28.7±16.1

Table 2. Percentage species distribution of protozoa in the rumen contents of ten African (Cape) buffalos.

Family Subfamily Genus Species Form	Dogiel (1932)	Animal number										Occurrence
		1	2	3	4	5	6	7	8	9	10	
		% of total protozoa										
OPHRYOSCOLECIDAE												
Entodiniinae												
Entodinium												
bicarinatum (da Cunha, 1914)	+	-	-	-	-	-	-	-	-	-		-
biconcavum (K & M[a], 1930)	-	-	-	-	-	-	-	-	-	1.4	2.3	2/10
bimastus (Dogiel, 192)	+	-	-	-	-	-	-	-	-	-	-	-
birostratum (Wertheim, 1935)	+	-	-	-	-	-	-	-	-	-	-	-
bovis (Wertheim, 1935)	-	-	-	-	-	-	4.8	-	-	-	-	1/10
bursa (Stein, 1858)	-	-	-	-	-	-	4.8	-	-	-	-	1/10
ciculum (Dehority, 1979)	-	-	-	-	-	-	-	-	-	-	4.5	1/10
dubardi (Buisson, 1923)	-	3.6	7.9	-	-	-	9.5	-	-	-	-	3/10
furca (da Cunha, 1914)	+	-	-	12.3	-	-	4.8	-	-	1.4	-	3/10
indicum (K & M[a], 1930)	-	-	-	-	-	-	9.5	4.0	-	-	-	2/10
longinucleatum (Dogiel, 1925)	-	-	-	-	0.6	-	-	-	-	1.4	-	2/10
nanellum (Dogiel, 1923)	-	-	-	1.4	-	-	-	-	-	-	-	1/10
parvum Buisson, 1923)	+	-	-	-	-	-	-	-	-	-	-	-
rectangulatum (K & M, 1930)	-	3.6	-	-	-	-	-	-	-	-	-	1/10
rostratum (Fiorentini, 1889)												
rostratum (Dogiel, 1927)	+	-	-	-	-	-	-	-	-	-	-	-
bifidum (Dogiel, 1927)	+	-	-	-	-	-	-	-	-	-	-	-
triacum (Buisson,1923)												
dextrum (Dogiel, 1927)	+	-	-	-	-	-	-	-	-	-	-	-
unidentified species		3.6[b]	-	-	-	-	-	-	-	-	-	-
Diplodiniinae												
Diplodinium												
anisacanthum (da Cunha, 1914))	+	-	-	-	-	6.4	-	4.0	4.0	-	-	3/10
anacanthum (Dogiel, 1927)	-	-	1.6	-	0.6	-	-	-	-	-	-	2/10
monocanthum (Dogiel, 1927)	-	-	-	2.7	-	-	-	-	-	-	-	1/10
tetracanthum (Dogiel, 1927)	-	3.6	1.6	-	-	-	-	-	-	-	-	2/10
pentacanthum (Dogiel, 1927)	-	3.6	-	-	1.3	-	-	-	-	-	-	2/10
bilobosum (Dogiel, 1927)	+	7.1	1.6	4.1	-	6.4	-	-	-	-	-	4/10

consors (Dogiel, 1925)	-	-	-	-	2.6	3.2	-	-	-	-	-	2/10
dentatum (Stein, 1858)	-	-	1.6	-	1.3	-	-	-	4.0	4.2	-	4/10
flabellum (K & M, 1932)												
aspinosum (D & P[d], 1974)	-	-	-	-	0.6	3.2	-	-	2.0	-	-	3/10
lobatum (K & M, 1932)	-	-	-	-	-	-	-	-	-	1.4	-	1/10
posterovesiculatum (Dogiel, 1927)	-	7.1	7.9	4.1	0.6	3.2	-	10.0	-	4.2	-	7/10
rectangulatum (K & M, 1932)	-	3.6	3.2	-	1.9	3.2	4.8	-	-	-	-	5/10
unidentified species		-	-	4.1[b]	-	-	-	-	-	-	-	-
Eudiplodinium												
dilobum (Dogiel, 1927)	-	-	-	-	1.3	-	-	-	-	-	-	1/10
impalae (Dogiel, 1925)	-	7.1	-	-	-	-	-	-	-	-	-	1/10
maggi (Fiorentini, 1889)	+	32.0	31.8	34.2	29.0	32.2	28.6	22.0	10.0	25.4	25.0	10/10
magnodentatum (K & M, 1932)	-	-	3.2	-	-	3.2	14.3	2.0	2.0	16.9	11.4	7/10
neglectum (Dogiel, 1925)	+	-	-	-	-	-	-	-	-	-	-	-
rostratum (Fiorentini, 1889)	+	-	-	-	0.6	3.2	-	4.0	-	-	4.5	4/10
Ostracodinium												
clipeolum (K & M, 1932)	_	-	-	-	0.6	-	-	-	-	4.2	2.3	3/10
dentatum (Fiorentini, 1889)	+	-	-	-	-	-	-	-	-	-	-	-
gauri (Kofoid & Christenson, 1933)	-	3.6	-	-	-	-	-	-	-	-	-	1/10
gracile (Dogiel, 1925)	-	-	-	4.1	-	-	-	-	-	-	-	1/10
monolobum (Dogiel, 1927)	+	-	-	-	-	-	-	-	-	-	-	-
gladiator (Dogiel, 1925)	-	-	-	-	-	-	-	-	4.0	1.4	-	2/10
mammosum (Railliet, 1890)	-	-	3.2	2.7	1.9	-	-	12.0	16.0	4.2	2.3	7/10
nanum (Dogiel, 1925)	-	-	1.6	-	-	6.4	-	-	-	-	-	2/10
stokyi (Buisson, 1923)	-	-	-	-	0.6	-	-	-	-	-	-	1/10
ventricosum (Buisson, 1923)	-	-	1.6	-	-	-	-	-	2.0	9.8	-	3/10
venustem (K & M, 1932)	-	-	-	-	-	-	14.3	4.0	-	-	-	2/10
Metadinium												
medium (A & M[c],1914)	+	-	3.2	-	-	6.4	4.8	-	-	-	-	3/10
fissilaminatum (Imai et al., 1992)	+	-	-	-	-	-	-	-	-	-	-	-
Enoploplastron species	-	-	-	-	-	-	-	-	-	-	- 4.5[e]	- 1/10
Epidinium												
cattanei (Fiorentini, 1889)	-	-	-	-	-	-	-	-	-	1.4	-	1/10
caudatum (Fiorentini, 1889)	+	-	-	-	0.6	-	-	14.0	-	1.4	9.1	4/10
tricaudatum (Sharp, 1914)	+	-	-	-	-	-	-	-	-	-	-	-
quadricaudatum (Sharp, 1914)	-	-	-	-	0.6	-	-	-	-	-	2.3	2/10

parvicaudatum (A & M, 1914)	-	-	-	2.7	-	-	-	-	2.0	-	-	2/10
ISOTRICHIDAE												
Dasytricha												
ruminantium (Schuberg, 1888)	+	21.4	30.2	27.4	54.8	22.6	-	24.0	54.0	21.1	31.8	9/10
BLEPHAROCORYTHIDAE												
Blepharocorys												
bovis (Dogiel, 1926)	+	-	-	-	-	-	-	-	-	-	-	-
Total number of species and forms observed	21	11	14	10	17	12	10	10	10	15	11	

[a]Kofoid & MaLennan.
[b]Cells distorted, could only be identified to the genus level. Not included in total number of species observed.
[c]Awerinzew & Mutafowa.
[d]Dehority & Potter.
[e]Could not be identified to species level, but was included in total number of species since all protozoa observed in this genus were similar.

between 10 and 34.2% of the total population. Next most prevalent was *Dasytricha ruminantium*, which occurred in 9 of the animals and ranged from 21.1 to 54.8% of the total population. Three species were found in 7 of the 10 buffalo: *Diplodinium posterovesiculatum* (0.6 to 10% of total); *Eudiplodinium magnodentatum* (2.0 to 16.9%); and *Ostracodinium mammosum* (1.9 to 16%). All of the remaining 38 species occurred in 5 or less animals.

Discussion

The indigenous ruminants in East Africa can be classified as browsers or concentrate selectors, as grazers or roughage eaters or as mixed feeders (Hoffman 1988). When ruminants consume a diet containing 60% or more of concentrates (i.e., grains or highly digestible browse), there is generally a decrease in the concentration of total protozoa and an increase in the percentage of *Entodinium* (Dehority and Orpen 1997). This can result in an "*Entodinium* - only" fauna or in some cases, with a 100% concentrate diet, complete elimination of protozoa (Dehority 1990, Dennis et al. 1983, Vance et al. 1972). In contrast, the grazers and mixed feeders tend to have a lower percentage of *Entodinium* with a more diverse fauna. Dehority (2003) studied eight species of indigenous wild ruminants from Kenya and found their fauna to match fairly well the expected distribution based on feeding type. Although it has been suggested that these differences between fauna and feeding type result from the lowering of rumen pH by the rapid bacterial fermentation of concentrate-type substrates, studies by Franzolin and Dehority (1996) and Dehority (2005) do not support this concept. Perhaps a lowering of rumen pH causes environmental changes in the rumen which favor growth of *Entodinium* species. Rumen contents from 10 wildebeest (four Blue and 6 Black), living in South Africa and consuming natural forages were studied by Booyse and Dehority (2012), and the percentage *Entodinium* was found to range between 0 and22.4. In the present study with African buffalo, also described as a grazer

or grass eater, the percentage of *Entodinium* ranged from 0 in two animals up to 33.4%. The major difference between the fauna in these two animal species appeared to be the percentage of *Dasytricha.* Except for one buffalo without *Dasytricha,* percentages in the other nine ranged from 21.1 up to 54.8% with an overall mean of 28.7 ± 16.1%. *Dasytricha* in four blue wildebeest ranged from 12.2 to 29.8% and 4.9% were present in a single black wildebeest.

Dogiel (1932) identified 21 species of protozoa in the single African buffalo from Uganda. The number of species in a single animal in the present study ranged from 10 to 17 and only eight of the 21 species listed by Dogiel were present in the 10 South African buffalo. For individual animals the highest number of species also found in the Uganda buffalo was 6, five in four animals and three in the remaining four buffalo. Most probably this difference in the fauna between the two sites is related to differences in the vegetation available in the two different areas. Also, no information is given on the subspecies of the animal from Uganda.

The overall species distribution in the South African buffalo is of interest, in that in all but one animal (number 6), *Dasytricha ruminantium* and *Eudiplodinium maggii* comprised 46% or more of the total population (mean of nine animals = 58.8 ± 11.4%). Buffalo number six was free of *Dasytricha,* but contained the highest percentage of *Entodinium* and *Eudiplodinium* species.

Buffalo numbers five and eight were *Entodinium*-free. In animal five, *Diplodinium, Eudiplodinium* and *Dasytricha* accounted for 86.8% of the total fauna. For animal 8, 76 % of the fauna were *Ostracodinium and Dasytricha*. In comparison (Booyse and Dehority 2012), one wildebeest was *Entodinium*-free, with *Diplodinium bubalidus, Ostracodium damaliscus and Ostracodinium gracile* comprising 98% of the total. Although diets were different, Towne et al. (1990) sampled 364 steers in feedlot fed high concentrate, of which 47 were *Entodinium*-free.

Isotricha was the predominant species replacing *Entodinium,* followed by either *Polyplastron* or *Epidinium.* It is of interest that the two genera, *Isotricha* and *Polyplastron,* have not been found in this study or in the wildebeest from South Africa (Booyse and Dehority 2012). Neither were they present in the fauna of the hartebeest or wildebeest sampled in Kenya (Dehority 2003).

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