

# Epidemiological study of *Cryptosporidium* at the wildlife-livestock and human interface in the western boundaries of the Kruger National Park

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## *Cryptosporidium* spp.

- Protozoan parasite
- Replicates in the small intestine of a large number of vertebrates (mammals, birds & reptiles)

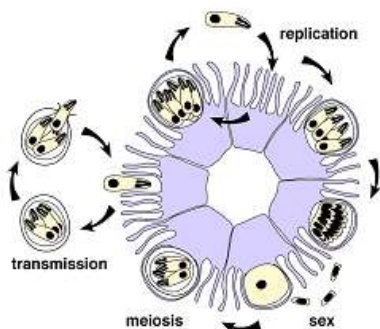
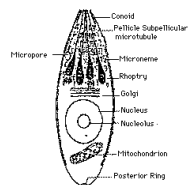


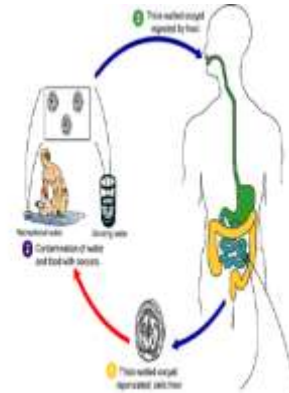
Figure 2.24: Apicomplex Structures



## *Cryptosporidium* spp.

Main source of infection: contaminated environment by faeces

- Oocysts: immediately infectious when excreted
- Very stable & survive up to 6 months in a moist & cool environment
- Unaffected by chlorine or other disinfectants added to water



## *Cryptosporidium* spp.

Phylum: *Apicomplexa*

16 species:

- C. andersoni
- C. baileyi
- C. bovis
- C. cervine\*
- C. canis\*
- C. felis\*
- C. galli
- C. hominis\*
- C. melagridis\*
- C. molnari
- C. muris\*
- C. parvum\*
- C. saurophilum
- C. serpentis
- C. suis\*
- C. wrairi

\* \* *Cryptosporidium* spp identified in human

\* Known to be of major zoonotic importance

## *Cryptosporidium* spp. in humans & animals

- Significant morbidity & mortality in young & immunocompromised individuals:

Human - among **young children** causes 45% of diarrheal deaths in Bangladesh, Brazil & in several African countries

Human - life-threatening diarrhea in **HIV positive patients** & therefore rural population in southern Africa with the highest prevalence of HIV/AIDS worldwide, are particularly at risk

Livestock - important cause of diarrhoea in calves (1 -3 week); adults shed the parasite without symptoms

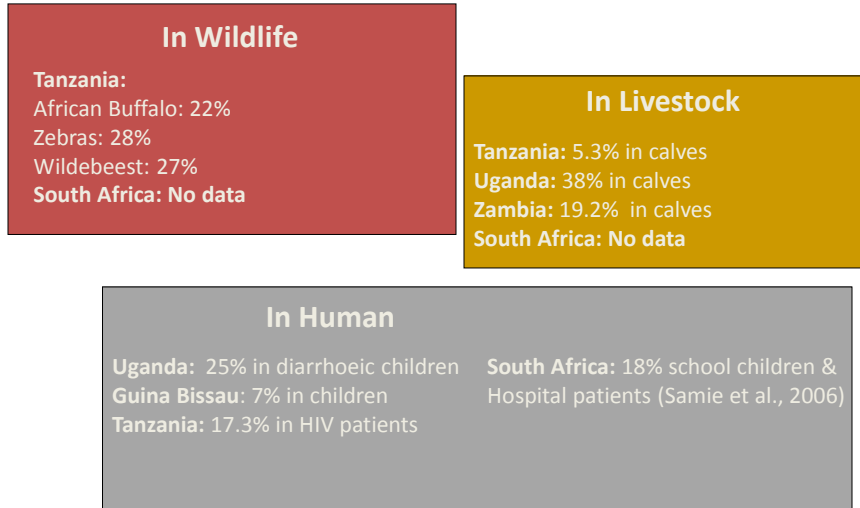
Wildlife – identified as reservoirs; infection appears to be asymptomatic

## *Cryptosporidium* spp. in humans & animals

No specific treatment available:

- Livestock - in calves quinazolina used in some European countries; its efficacy has not been demonstrated
- Humans – no drug effectively treats cryptosporidiosis; only reduces disease severity in some cases

## Prevalences of *Cryptosporidium* in Africa



## Objective

- Determine the zoonotic importance of *Cryptosporidium* spp. at the wildlife, livestock & human interface in KNP
- Understand the epidemiological patterns of *Cryptosporidium* spp. among the different compartments (wildlife, livestock and human)

## Research Questions

1. Are wildlife species in KNP a reservoir of *Cryptosporidium* ?
2. What is the prevalence of *Cryptosporidium* in livestock adjacent to KNP?
3. Is *Cryptosporidium* a neglected zoonosis in human communities living close to the KNP?
4. What are the epidemiological patterns of circulation and transmission of *Cryptosporidium* at the wildlife/livestock/human interface in KNP?

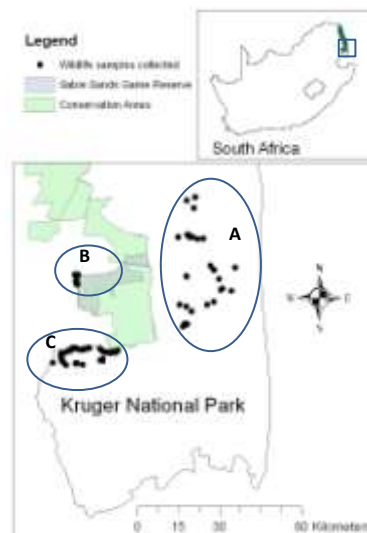
## Wildlife: Study Area

### Kruger National Park

Tshokwane (A), distant from KNP fence  
Skukuza (C), adjacent to fence

### Private game reserve

Sabi Sand (B), adjacent to fence



## Wildlife: Material & Methods

### Collection of faecal samples from wildlife :

- Three of the most commonly seen species
- Collected during dry & rainy seasons 2008/2009



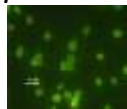
	Skukuza	Sabie Sand	Tshokwane	Total
Buffalo	103	71	92	<b>266</b>
Elephant	90	76	90	<b>256</b>
Impala	98	92	90	<b>280</b>
	<b>291</b>	<b>239</b>	<b>272</b>	<b>802</b>



## Wildlife: Material & Methods

### Laboratory analysis:

- immunofluorescent antibody (IFA) commercial kit (anti- *Cryptosporidium parvum* monoclonal antibody technique)



- Ziehl Neelsen (ZN) – detects all *Cryptosporidium spp.*



- Confirmation of Positives ZN & IFA with RT-PCR

## Wildlife: Results

Prevalence with Ziehl-Neelsen staining

	Skukuza	Sabi Sand	Tshokwane	Total/species
Elephant	<b>34.4%</b> (12/35)	<b>35.7%</b> (10/28)	<b>6.6%</b> (2/30)	<b>25.8%</b> (24/93)
Buffalo	<b>3.2%</b> (1/31)	<b>6.9%</b> (2/29)	<b>6.6%</b> (2/30)	<b>5.5%</b> (5/91)
Impala	<b>3.2%</b> (1/31)	<b>5.9%</b> (2/34)	<b>3.5%</b> (1/29)	<b>4.2%</b> (4/94)

Overall prevalence 11.8%

## Wildlife: Results

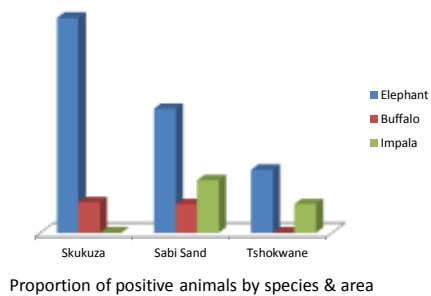
Prevalence with **direct immunofluorescent antibody test (IFA)**

	Skukuza	Sabi Sand	Tshokwane	Total/species
Elephant	<b>8%</b> (4/50)	<b>0%</b> (0/44)	<b>4%</b> (2/50)	<b>4.2%</b> (6/144)
Buffalo	<b>2%</b> (1/50)	<b>2.5%</b> (1/40)	<b>0%</b> (0/50)	<b>1.4%</b> (2/140)
Impala	<b>0%</b> (0/50)	<b>5%</b> (3/59)	<b>0%</b> (0/50)	<b>1.8%</b> (3/161)

Overall prevalence 2.5%

## Results wildlife

Variable	Level	OR	95% C.I. (OR)	P-Value
Area	Tshokwane	1*	-	-
	Sabi Sand	3.6	1.2;11.1	0.023
	Skukuza	2.9	1.0; 8.8	0.057
Species	Impla	1*	-	-
	Buffalo	1.3	0.3; 5.2	0.665
	Elephant	8.4	2.8;25.8	< 0.001



- Prevalence was significantly higher in elephant than other species.
- Prevalence was higher in areas close to the fence

## Wildlife: Main Conclusions

- *Cryptosporidium* spp. are present in KNP wildlife
  - Potential source of infection to livestock & human at the interface?
  - Is wildlife close to the fence more exposed?
- Prevalence might be higher than the one observed
  - Low sensitivity & specificity of ZN
  - Samples may contain genotypes that could not be detected
  - Analyzed samples: collected during dry season





## Wildlife: Way forward

- Detection of all *Cryptosporidium* spp circulating in the wildlife population: PCR, genotyping & subtyping
- Analysis of seasonal variations (only dry season samples have been analyzed)

## Livestock: Study Area



Bushbuckridge: 10 diptanks (●) located in close proximity to the fences of KNP & Sabi Sand



## Livestock



- 1000 samples randomly selected
- Collected in rainy and dry season
- All ages
- immunofluorescent antibody (IFA) commercial kit (anti- *Cryptosporidium parvum* monoclonal antibody technique)

### Results from dry season

- Overall prevalence: **1% (3/300)**

## Livestock: Main Conclusion



### Low prevalence in cattle (1%):

- IFA test specific for the detection of *Cryptosporidium parvum* oocysts (species of major zoonotic importance)
- Other *Cryptosporidium* spp. might circulate in cattle

## Livestock: Way forward



- PCR, genotyping & subtyping to detect and characterize *Cryptosporidium* strains in the cattle population.
- Analysis of seasonal variations (only dry season samples have been analyzed)

## Human communities: Study Area

7 clinics located in communities in close proximity to KNP:

Belfast Clinic

Justicia Clinic

Lillydale Clinic

Lillydale Private Hospital

Agincourt Clinic

Calcutta Clinic

Oakley Clinic

## Human communities: Way forward

- Collection of human faecal samples (approx. 200)
- PCR, genotyping & subtyping of human samples to detect the presence of *Cryptosporidium* and the different *Cryptosporidium* spp.
- Determine the zoonotic importance of *Cryptosporidium* spp from livestock and wildlife origin.

## Expected Results

- Detect different *Cryptosporidium* spp. circulating in the wildlife, livestock & human population
- Establish links between *Cryptosporidium* spp. in humans, livestock & wildlife.
- Comparison of different diagnostic methods

## Activities 2011

- **March 2011:** Collection of human faecal samples in the study area (7 clinics in Bushbuckridge).
- **April – August 2011:** PCR, genotyping & subtyping of *Cryptosporidium* of wildlife, cattle & human samples at National Center for Emerging and zoonotic Infectious Diseases, Centers for Disease Control and Prevention (CDC), Atlanta, USA



## Acknowledgement

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**Thank You**

