

A comparative study of gastrointestinal parasites between ranched and free ranging Burchell's zebra (*Equus burchelli antiquorum*) in Isiolo district, Kenya

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Parasites were collected from 20 Burchell's zebra, *Equus burchelli antiquorum*, from Isiolo district, Kenya. 10 were ranched animals from the Lewa Downs Ranch and 10 free ranging from the areas adjacent to the ranch to the north. The animals were culled from 4th to 18th June 1995. The gastrointestinal tract was removed from the carcass and separated as stomach, small and large intestines and searched for parasites. The abdominal cavity was scrutinized for *Setaria* species. The parasites were identified to genus and in some cases species level. All animals were infected with at least three genera of parasites of which at least one genus was a nematode. A total of 10 genera representing eight families were recovered from the two groups. These included six nematode families, *Strongylidae*, *Atractidae*, *Oxyuridae*, *Spiruridae*, *Setariidae*, and *Ascaridae*, one cestode family, *Anoplocephalidae* and one family of the larvae of *Gasterophilus* bot flies, *Gasterophilidae*. The most prevalent families were *Atractidae* (100%) and *Gasterophilidae* (100%). The principle nematode genera was *Crossocephalus* whose total burden, plus or minus the standard deviation, was $3,471,129 \pm 1,352,922$. *Setaria* species were recovered from the abdominal cavity of 45% of all the animals examined. The overall total worm burden was higher in the free ranging zebra $2,743,410 \pm 849,604$ than the ranched zebra, $787,669 \pm 246,006$. The range of individual genera varied from 0 to 269,225 in the free ranging group, which was higher than 0 to 77,890 in the ranched animals. From statistical analysis, no significant difference could be found between males and females. However, the burdens of genera *Strongylus*, *Triodontophorus*, *Crossocephalus* and *Parascaris* were significantly higher in free ranging animals.

Key words: Gastrointestinal, parasites, zebra, lewa downs ranch

Introduction

Some of the earlier documented parasitological work in zebra [1,18,21] led to the compilation of a checklist. This checklist was considered incomplete because many reports were incidental findings from sampling done at necropsy. Secondly, in earlier records, the host was not defined to species level [4], although these records were of substantial value as true observations. It was difficult to arrive at any assessment of the pathogenicity of many of these parasites. More work has since been carried out in Burchell's zebra giving attention to the total worm burdens, seasonal prevalence and life cycles [4,14,15]. Parasitological investigations such as these have been of value not only for their contribution to the baseline information in domestic equine helminthology [2], also suggests, a means of providing a system either for monitoring overpopulation, or disease in a population. However, for this you need a better understanding of the population dynamics of both the host and its nematode parasite.

The study area was selected following a study done by the Kenya Wildlife Service (KWS), Veterinary Units field veterinary staff [3], who between August and December 1991 reported morbidity and mortality among Burchell's (*Equus burchelli antiquorum*) and Grevyi zebra (*Equus Grevyi*). They found equine strongyloidosis to be a predisposing factor for nutritional stress and mortality on Lewa Downs Ranch in Isiolo District, Kenya. Mortality was first reported in 1989 and said to be occurring annually between August and December with the peak in September. The mortalities were associated with an overall increase of animals on the ranch, low precipitation and availability of only coarse lignified pasture low in nutrients. This raised particular concern about the long-term survival of the grevyi zebra, which are listed as endangered by the International Union for the Conservation of Nature. The initial study did not quantify the gastrointestinal parasite burden, only constituted a case response report. This study sought to extend the work already done. Burchell's zebra were used in

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this study since they are not endangered and were being cropped regularly on the ranch. The study area also presented an opportunity to make comparisons with free ranging Burchells zebra in areas adjacent to the ranch.

The objectives of the study were to determine, (a) The gastrointestinal parasite genera or species present in free ranging and ranches Burchell's zebra in Isiolo District of Kenya. (b) The total worm burden and the prevalence of the parasites present. (c) If 1 and 2 are affected by the system of husbandry. (d) The fecal egg count and how it correlated to the size of worm burden.

Materials and Methods

Study area

During this study, 20 Burchell's zebra were culled. Ten were free ranging zebra from Isiolo district of Kenya. The district is in the eastern province and located on longitudes 36°50' and 39°50'E and latitudes 0°05' and 2°N. The district covers 25,605 square kilometer's. Ten were ranches zebras from Lewa Downs Game Ranch, which borders the semi-arid Isiolo district to the south. It lies between longitudes 32°E and 37°E and latitudes 0°10' and 0°17'N.

The area is dry and hot for most of the year. Rainfall is unreliable and scarce; however the two main rainfall seasons are the long rains between March and May and the short rains between November and December. The area receives an average rainfall of about 475 mm per annum with a range from 237 mm in some area to 698 mm per annum.

Animals

The 20 zebra were culled between the dates 4/7/95 and 18/7/95. Animals selected for shooting were judged to be of adult size and no discrimination by sex was made. The zebra were shot in the neck with a .308 calibre rifle. This resulted in instantaneous death in most cases. Upon examination, one free-ranging and three ranches zebra were found pregnant.

Helminth Recovery

The zebras were then skinned and eviscerated following techniques used in equids described [9,10]. Double ligatures were placed at the esophageal entrance to the stomach, the pylorus, and near the ileocecal junction. Cuts were made between the ligatures to divide the gastrointestinal tract into stomach, small intestines and large intestines. In this case the stomach and small intestines were placed into separate buckets that were graduated up to 10 liters. The combined large intestines were put into a large plastic tray measuring 170 × 135 × 33 centimeters. A small amount of feces were collected from the rectum into plastic fecal pots for fecal worm egg count. These were placed into a cool box and transported back to the field laboratory. They were placed in

a fridge and examined within 2 days. Fecal egg counts were carried out using MacMaster's Technique [20].

The abdominal cavity was scrutinized for *Setaria* species. Each organ was cut open and its ingesta emptied into the respective containers. The gastrointestinal walls were examined for worms adhering to the mucosa. The stomach and small intestines contents were diluted to 10 liters with water and the large intestines to 40 liters. Each container was thoroughly mixed and random samples were taken to form aliquots of 5% for stomach and small intestinal samples and 10% from the combined large intestines. Each aliquot was put into a sieve of 150 µm aperture and washed continuously until all fecal matter was washed off and the water was clear.

The gut washings were placed into polyurethane bags and marked with a felt pen then placed in a cool box and transported back to the laboratory. A few drops of 45% iodine were added to the washings before examination to kill and stain the helminths. The aliquots were examined in small portions at a time with the aid of a 30X magnification dissecting microscope. Each worm was picked out individually and placed into 10% buffered formalin. Most of the parasites could be seen macroscopically but *Crossocephalus* species required the use of microscopic examination. The total worm burdens were based on the aliquots taken of the complete specimen.

Helminth identification

Identification of large parasites was done under a dissecting microscope (30X) and smaller parasites under a compound microscope. Descriptions [7] were used for the identification of the genera *Strongylus*, *Triodontophorus*, *Cyathostomum*, *Oxyuris*, *Habronema*, *Seteria* and *Anoplocephala* [6] for *Crossocephalus*, and *Gasterophilus* larvae [22]. The helminths were examined to genus or species level and recorded separately.

The term prevalence, which is expressed as a percentage, is defined as the number of individuals of the host species infected with a particular parasite species divided by the number of hosts examined [11].

In the statistical analysis, ranked parasite burdens of each genera was compared using the Wilcoxon Rank Sum (Mann-Whitney) Test, to statistically determine if there was a significant difference between parasite burdens in males versus females. In addition, to see if being ranches or free ranging made any difference. This non-parametric test was used due to the small sample size and unevenly distributed data. Additionally, in order to discern the importance or contribution to the total variation of the number of parasites by the factors of sex or free ranging versus ranching, multiple regression was done. A computer statistical software application was used for the statistical analysis.

Results

Parasites recovered

Ten genera of gastrointestinal parasites were recovered and identified to the genera level and in some cases to the species level using various descriptions. The total and mean parasite burdens and their prevalence are presented in tabular form for the ranched zebra, and for the free ranging zebra. The 10 genera recovered were from eight families. Six nematode families found were *Strongylidae*, *Atractidae*, *Oxyuridae*, *Spiruridae*, *Setariidae*, and *Ascaridae*. One cestode family was recovered, *Anoplocephalidae*, and larvae of the fly *Gasterophilus* from the family *Gasterophilidae*. The genera *Habronema* and *Gasterophilus* were recovered from the stomach. *Parascaris*, *Crossocephalus*, *Anoplocephala* and *Gasterophilus* recovered from the small intestines. *Strongylus*, *Triodontophorus*, *Cyathostomum*, *Crossocephalus*, *Oxyuris* and *Anoplocephala* were recovered from the large intestines.

On the whole, the most prevalent families were the *Atractidae* (100%) and the *Gasterophilidae* (100%). The ranges of individual genera varied from an average of 0 to 77,890 from Lewa Downs Ranch zebra and 0 to 269,225 in the free ranging zebra. When comparing the two groups, the overall total parasite burden for all animals in the free ranging group of zebras was higher at $2,743,410 \pm 849,604$ SD with a range of 0 to 2,692,255. The ranched animals had a total worm burden of $787,669 \pm 246,006$ SD with a range of 0 to 778,904.

Strongylidae

Strongylinae

An overall 90% prevalence of strongylids was found from all the animals examined. Two species were recovered and identified to genus level. The species *Strongylus* was the second most abundant nematode and ranged from 280 to 4,740 for the free ranging zebra with 100% prevalence and 0 to 1,730 in the ranched zebra with 80% prevalence. *Triodontophorus* was recovered from only one animal on the ranch, which had a total worm burden of 710. In the free ranging group, there was a prevalence of 70% and burdens recovered ranged from 421 to 32,088.

Cyathostominae

Nematodes of the genus *Cyathostomum* were recovered from the large intestines of only one zebra from the ranched group which had a total worm burden of 1250. Filamentous prokaryotic organisms were noted attached to the anterior and posterior extremities of the nematodes of this family. This has been reported in her study on Burchell's zebra [4,5].

Atractidae

Only the genus *Crossocephalus* was consistently recovered

from the large intestines of all the animals from both groups. It was the most prevalent (100%), and abundant nematode recovered. The ranched animals had burdens ranging from 2506 to 170,872 and the free ranging zebra, 40,652 to 453,750.

Oxyuridae

Forty percent of all animals examined had *Oxyuris* nematodes. Two of the ranched animals were positive and had total worm burdens of 94 and 103. In the free ranging group, 6 animals were positive with burdens ranging from 520 to 2352.

Spiruridae

An overall of 30% of all animals were infected. Only members of the genus *Habronema* were recovered from the stomach. One zebra from the ranch was positive with a total count of 32 while 50% of the free ranging zebra were affected and had burdens ranging from 54 to 652. Some of these parasites were pulled out of nodules in the mucosa of the stomach.

Setariidae

There was a 45% overall infection rate. Two of the ranched animals had the nematode, *Setaria equina*. Each had 3 parasites. 70% of the free ranging zebra were infected with the same nematode and numbers ranged from 1 to 4.

Ascaridae

Parascaris equorum was recovered from 30% of all the animals. None of the ranched animals were positive for this nematode. The free ranging zebra had a range of 1 to 2.

Anoplocephalidae

Eighty percent of animals examined both ranched and free ranging were infected. Worms collected mainly from the caecum and colon. The only cestode recovered in the study was from the species *Anoplocephala Perfoliata*. In the ranched animals, they ranged from 12 to 71 and were found in all the animals. The free ranging group had burdens between 11 to 98 and 6 animals were affected. They left ulcerated and inflamed areas when detached from the mucosa.

Gasterophilus larvae (Bots)

These were consistently present and attached to the glandular stomach of all zebra. They were sampled in fairly high numbers with ranges of 152 to 451 and 180 to 613 in the ranched and free ranging groups respectively.

The non-parametric Wilcoxon Rank Sum Test was applied at a 5% confidence level. When tested for sex, no significance could be demonstrated in the parasite burdens if the zebra was female or male. When tested for free ranging versus ranched animals, significance was shown for the

genera *Strongylus*, *Triodontophorus*, *Crossocephalus* and *Parascaris*. These were significantly higher in free-range zebra. There was not enough evidence to statistically demonstrate a difference in the total burdens of the genera *Cyathostomum*, *Oxyuris*, *Habronema*, *Setaria*, *Anoplocephala* and *Gasterophilus*. Results from multiple regression show that as far as free ranging versus ranching was concerned, significant difference was found for 3 genera of parasites. *Strongylus*, *Crossocephalus* and *Parascaris* were present in statistically significant higher burdens in the free ranging zebra. For *Triodontophorus*, *Oxyuris*, *Habronema*, and *Gasterophilus*, the p-values were close but not within the level of significance. If any difference existed between the two groups, there is too little data available to discern it. The test also showed that the burdens of genera *Setaria* and *Anoplocephala* were not significantly different whether the animal was ranching or free ranging.

Fecal egg counts

The fecal egg count in the ranching animals ranged from 650 to 1,800 with a mean egg count of $1,225 \pm 104.2$. The free-range animals had counts of 1,050 to 2,150 and a mean of $1,620 \pm 204$ using the Wilcoxon rank sum test. There was no significant difference in the total eggs per gram between the two groups. In comparing the mean total worm burdens to the total egg counts, there were generally higher egg counts in animals with higher worm burdens, however, no statistical difference could be found between the ranching and free ranging animals.

Discussion

Most genera of parasites were recovered from the large intestines including those that are of most parasitic importance such as the large strongyles. The large intestines therefore appear to be an important portion of the gastrointestinal tract as far as parasitism in zebras is concerned.

All zebra examined were infected with at least 3 genera of parasites of which at least one was a nematode. The genera of nematodes recovered were similar to those reported in other studies done on Burchell's zebra [4,5,14,15,16], but less diversity was recorded.

This may have been due to the fact that the study was conducted over a short period and therefore was not as intensive as other studies that have been done. In addition, having been carried out within one season only, it cannot present a complete picture of the nematodes that could be present in that group of zebras in Isiolo District as seasonal study would.

Of the large strongyles, *Craterostomum* and *Oesophagodontus* were not found in this group of zebra. In the sub-family *Cyathostominae*, only the genus *Cyathostomum* was recovered from one ranching animal. A previous study done at Lewa Downs on zebra [3] recovered other species of

Cyathostomes, namely *Cylindropharynx* and *Cylicocycylus* species which were not found in this study together with approximately 50 other species recorded in equine. Cyathostomes nearly always exist in equids however in this case, they must have gone undetected. For practical reasons, only small aliquots of the combined large intestinal contents could be examined making it possible to miss them. The burdens may also have been low due to other factors that affect strongylid numbers such as climatic and environmental factors. For instance, the arid climate may not support a great abundance of strongylids. Other host factors may have included age resistance.

Similar to the other studies done in Burchell's zebra that are mentioned in this paper, the family *Atractidae* was the most abundant recovered in this study. Only the species *Crossocephalus* was found. The species *Probstmayria* that has been recovered in other similar studies was not found. The high numbers of this species are attributable to its viviparous life cycle, which it completes entirely in one host. In addition, Scialdo-Krecek found that Atractids were more successful in an arid climate [15,16]. The numbers recovered could probably have been higher had a mucosal digestion been done.

The *Parascaris* species were found in low numbers. The Setariids were found in 45% of the animals examined. The prevalence of this parasite may have been higher however the parasite is difficult to locate within the abdominal cavity.

The three acknowledged species within the *Habronema/Draschia* complex are ubiquitous presumably due to the universal distribution of their major intermediate hosts *Muscae domestica* and *Stomoxys calcitrans* [17]. *Draschia* species were not found in this study although it has been reported in high prevalence in equids in tropical Africa. The intensities reported are related to the abundance of their vectors, highest numbers occurring in the summer months [14,15,16]. Thirty percent of the animals were infected with *Habronema* species. The limited numbers of *Habronema* found in this study may have been because the weather may not have favored their vectors. Also since few *Habronema* species are found free in the stomach after death [8]. Most are still associated with the mucosa and during this study; a mucosal digestion was not done.

The oxyurids were not found in very high numbers and this was to be expected, as large numbers are usually found in foals.

Other work on Burchell's zebra [14] also recovered cestodes of the genus *Anoplocephala*. These were found consistently in this study in numbers ranging from 11 to 98. The clinical significance of cestode infections is usually difficult to ascertain. Most pathology attributed to these parasites is from natural infections and other contributing factors cannot be ruled out [8].

The statistical results seem to suggest that there is no difference in the parasite burdens if the animal is a male or

female. Also that there are higher burdens of particular genera of parasites (*Strongylus*, *Triodontophorus*, *Crossocephalus* and *Parascaris*) in the free-range zebra compared to the ranch. However, it is difficult to draw any conclusions from this analysis due to the fact that our sample size was too small to demonstrate conclusively a clear difference in our comparisons. Additionally, there was an unequal number between the females and males. A larger sample size would have to be used to obtain results that one can make inferences from.

Some genera recorded in other studies were not recovered in this study. *Strongyloides westeri* in the family *Strongyloididae* has been reported in Burchell's zebra [4], [5] recovered from small intestines. It is a nematode common in young foals although occasionally reported in adults [7].

No helminths from the family *Trichostrongylidae* were recovered. *Trichostrongylus axei* Cobbold, 1879 is a widespread parasite of equids occurring in most parts of the world [7]. It has been reported in Burchell's zebra however the location of its host is not given [13]. Although widespread, few reports of its occurrence in equids are available. One of the reasons for its reported low prevalence or absence in some studies may be due to its relatively small size compared to other nematodes of equids, making it easily overlooked [12]. *Trichostrongylus thomasi*, Monnig 1932 has also been reported in Burchell's zebra in Kruger National Park, South Africa [4,5].

The trematodes of *Gastrodiscus* species, which have been reported in the small intestines of equine, were not recovered in this study. It has been reported recovered from a Grevy's zebra [19]. This parasite normally occurs in Africa and requires an intermediary host, a fresh water mollusc. The most likely reason for its absence was the lack of its intermediate host in an arid area such as Isiolo District.

Free ranging wild animals have evolved over millions of years and under normal conditions live in a state of equilibrium with their parasites. Although young and stressed animals with large burdens may suffer from ill health, this resistance to helminth infestations in adult wildlife species probably results from immunity stimulated by the continuous challenge in the first year of life. In this study, all the animals examined were apparently healthy. It was expected that the study would find a higher gastrointestinal burden in ranch animals due to the stress they undergo of confinement with poor grazing and high populations.

However, it was found that the free ranging animals had higher parasite burdens. This difference was partly attributed to the change in the management practice on the ranch since 1989 when equine strongyloidosis was diagnosed on the ranch. The predominant genera of parasites identified at that time were *Strongylus*, *Trichostrongylus*, *Cylindropharynx* and *Cylicocyclus*. Management practices that were advised

and adopted on the ranch at that time included reducing herbivore density by continual removal of Burchell's zebra through a cropping program. In addition, pasture management by encouraging rotational grazing through controlled burning. This helped to reduce the number of infective larvae on the ground possibly leading to a reduction of parasites that have free living stages such as the strongylids and oxyurids. Since the pasture burning started the zebra mortality has reduced with improved overall quality of pasture and reduced the nutritional stress.

This study should serve as a preliminary study to other epidemiological studies, which look into parasite burdens in the zebra through the different seasons.

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