

SPOROZOANS AS CAUSATIVE AGENTS OF PROTOZOAL DISEASE



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Abstract

Several species are classified as commensals, which implies that they do not normally do any harm to the host, whereas other species are considered pathogens and often cause sickness. Infections caused by protozoa may range from being absolutely innocuous to being lethal. Individuals whose immune systems can keep a parasite infection under control but not totally eliminate it are considered to be carriers of the illness and may transfer it on to other people. As eradication would reduce the individual's immunity to the parasite and result in a significant risk of reinfection, well-tolerated infections that occur in geographic regions with a high frequency are frequently not treated to eliminate the parasite. This is done because of the high frequency. As there are not enough trustworthy treatments, there are not enough immunizations that are successful, and there are other problems as well, such the difficulties of managing vectors, the World Health Organization has picked six ailments as the focus of intensified research and training. Malaria, trypanosomiasis, and leishmaniasis are examples of these sorts of disorders caused by protozoa.

Keywords: Sporozoans, Protozoal, Causative

INTRODUCTION

In the contemporary taxonomy, the Protozoa are classified as their own subkingdom inside the Protista kingdom; however, under the classical system, they were classed within the Animalia kingdom. The great majority of protozoa are free-living organisms, and there are around 50,000 different species of protozoa that have been described. Protozoa may be found in practically every setting conceivable. There is evidence in the form of fossilized shells discovered in sedimentary rocks showing protozoa lived in the time period before the Cambrian explosion. By creating microscopes out of simple lenses, Anton van Leeuwenhoek was the first person to observe protozoa. He performed this using his microscopes. Between the years 1674 and 1716, he not only characterized free-living protozoa, but also several forms of parasites that lived on animals, as well as *Giardia lamblia* that he had discovered in his own excrement. Many individuals are infected with one or more species of protozoa at some time in their lives, and the great majority of humans have protozoa dwelling in or on their bodies at some point in their lives. Several species are classified as commensals, which implies that they do not normally do any harm to the host, whereas other species are considered pathogens and often cause sickness. Infections caused by protozoa may range from being absolutely innocuous to being lethal. Individuals whose immune systems can keep a parasite infection under control but not totally eliminate it are considered to be carriers of the illness and may transfer it on to other people. As eradication would reduce the individual's immunity to the parasite and result in a significant risk of reinfection, well-tolerated infections that occur in geographic regions with a high frequency are frequently not treated to eliminate the parasite. This is done because of the high frequency.

In healthy persons, protozoan infections frequently go undetected or are small, but in immunocompromised patients, particularly those with acquired immune deficiency syndrome (AIDS), protozoan infections may represent a major danger to the patient's life (AIDS) (AIDS). There is evidence to demonstrate that the lungs of many otherwise healthy persons contain modest levels of the fungus *Pneumocystis carinii*. But, in immunocompromised patients, such as those with AIDS, this parasite may produce a sort of pneumonia that is virtually always lethal. *Toxoplasma gondii* is a common protozoan parasite that, in most instances, results in a fairly small acute illness, followed by a persistent latent infection that lasts for a long period. Toxoplasmic encephalitis is a potentially deadly illness that may afflict patients with AIDS. *Cryptosporidium* was initially described in the 19th century, but it wasn't until relatively

recently that researchers identified widespread human infection. Another protozoan that has the potential to create serious complications in those living with AIDS is termed cryptosporidium. Before the discovery of AIDS, incidences of microsporidiosis in humans were exceedingly uncommon and infrequently described. It is presently considered a more frequent infection among persons suffering with AIDS. As additional in-depth studies of persons living with AIDS are done, it is likely that other atypical or uncommon protozoan infections may be detected in these individuals. Acanthamoeba species are amebas that are free-living and may be found in both water and soil. Cyst stages are able to travel through the air. Individuals who wear contact lenses have been proven to have an increased risk of having corneal ulcers caused by Acanthamoeba species. These ulcers may cause significant visual loss. It is believed that the parasites are disseminated via contaminated lens-cleaning solution. Nearly every case of primary amebic meningoencephalitis—a disorder that almost invariably ends in death—can be traced back to aquatic amoebas belonging to the genus Naegleria, which reside in freshwater habitats. It is thought that the amebas enter the body through the upper nasal tract when water from the environment is splashed over it during swimming or diving. Based on laboratory examinations of Acanthamoeba infections in cell cultures and in animals, it was able to predict human infections of this nature before they were found and reported. This was done by the use of statistical modeling. As there are not enough trustworthy treatments, there are not enough immunizations that are successful, and there are other problems as well, such the difficulties of managing vectors, the World Health Organization has picked six ailments as the focus of intensified research and training. Malaria, trypanosomiasis, and leishmaniasis are examples of these sorts of disorders caused by protozoa. Despite the fact that fresh understanding about these disorders has been gathered, the bulk of the obstacles connected with control remain to persist.

REVIEW LITERATURE

Raafat Mohamed Shaapan (2015) Toxoplasmosis, neosporosis, sarcosporidiosis (also known as sarcocystosis), and trypanosomiasis are some instances of prevalent zoonotic protozoal illnesses that may induce a miscarriage. Trypanosomiasis is also a form of sarcocystosis. Toxoplasma gondii, Neospora caninum, and Trypanosoma are all examples of single-celled protozoan parasites that are responsible for generating one of these diseases. Sarcocystis spp. is another example. It is generally accepted that toxoplasmosis is the disease that is responsible for the majority of miscarriages that occur in pregnant women as well as in a variety of female

animals all over the world. This is because toxoplasmosis can only be passed from one female animal to another through the female's urine. Antibodies directed specifically against *T. gondii* were detected in about one-third of the human population. It is possible for the infection to be passed on from mother to child via the placenta, which may result in congenitally infected newborns experiencing encephalitis, chorioretinitis, mental retardation, and loss of vision. In addition, the infection has the potential to induce stillbirths in animals as well as the mummification of fetuses that have been aborted. Neosporosis is a disease that may result in considerable financial losses for enterprises that grow dairy cattle and beef cattle. This is because the illness causes a drop in the amount of milk and meat that are produced. This illness is recognized as a key factor in the incidence of severe abortions in a broad variety of wild and domestic animals all over the globe, most notably cattle. These animals may be found in both wild and domestic environments.

Loeki Enggar Fitri (2016) Intestinal protozoan infections are a persistent danger to public health, and the populations of undeveloped nations that are located in tropical and subtropical parts of the world are more likely to be affected by these illnesses. It is still difficult to determine which protozoa are resident in the digestive system. This is particularly true in less developed nations, where there is a shortage of laboratory facilities, inadequate money for medical care, and the geographical isolation of populations. Conventional diagnostic procedures are based on microscopy and staining techniques, despite the fact that these approaches have a number of major drawbacks that seriously restrict their usefulness. These procedures are nonetheless commonly utilized. The lack of sensitivity and specificity of the traditional methods, which are both factors that contribute to the issue, is likely to be the primary cause of these limitations. When choosing diagnostic procedures, it is essential to take into consideration not only the purpose of the test but also the resources that are easily accessible as well as the species of parasite that is most likely to be found. Failing to do so could result in the selection of inappropriate diagnostic procedures. In this article, we will discuss the different immunodiagnosis and molecular diagnostic approaches for intestinal protozoa infection. Specifically, we will concentrate on *Entamoeba histolytica*, *Giardia duodenalis*, and *Cryptosporidium* spp. as our primary targets. In this article, we discuss not only the benefits and drawbacks of these techniques, but also the therapeutic applications that are most suited to make use of them.

Katherine T. Andrews, (2014) Parasitic illnesses have a significant negative influence not only on human health but also on society and the economy, and they are especially prevalent in tropical areas of the globe. The majority of parasite-related morbidity and mortality, which is estimated to amount to 1.1 million deaths yearly, is caused by diseases that are caused by protozoa and helminths. Some examples of these diseases are malaria and schistosomiasis. Because of the absence of approved vaccinations, the worldwide burden of these illnesses is made even worse, which is why it is essential to develop safe and effective medications for their prevention and treatment. Regrettably, in areas where medications are accessible, their efficacy is rapidly being called into question due to the development of drug resistance in parasites. The demand for novel medications is the primary driver of research into antiparasitic drug development all around the world. In order to assure a sustained pipeline of lead compounds, a variety of creative tactics are required. In this review, we will focus on major human parasitic protozoan diseases such as malaria, trypanosomiasis, toxoplasmosis, cryptosporidiosis, and leishmaniasis as we discuss one of these approaches, which is drug repurposing or repositioning. In other words, we will repurpose or reposition existing drugs.

RESEARCH METHODOLOGY

MATERIALS AND METHODS

Source of protozoa and surrogates

The heat-inactivated *C. parvum* oocysts were provided by the Wisconsin State Laboratory of Hygiene in Madison, Wisconsin, while the *G. lamblia* cysts were given by Waterborne Inc. in New Orleans, Louisiana. Oocysts and cysts were collected no more than one week before their separate usage in both the settling column study and the mesocosm tank research. Dragon Green (DG) microspheres (Product No. FC07F/5493, Bangs Laboratories, Inc., Fishers, IN) and Glacial Blue (GB) microspheres (Product No. PC06N/8319, Bangs Laboratories, Inc., Fishers, IN) have both been investigated in the past as possible surrogate particles for *Toxoplasma gondii* oocysts due to the surface properties of each of these microspheres.

SETTLING COLUMN EXPERIMENTS

Tests were performed in vertical settling columns in order to investigate the impact of hydrologic factors on the settling velocities. Each settling column had the ability to hold one liter (L) of water and was outfitted with sample apertures that were positioned one hundred and

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three hundred millimeters (mm) below the surface of the water. Over the course of the research, counts of protozoa and microspheres were carried out at each of the ports in order to determine whether or not there was a discernible difference in the rates at which particles settled depending on the various kinds of water conditions. The effects of water type, salt level, and water temperature on the settling velocity of parasites and microspheres were measured by evaluating a total of eight different treatment conditions using a completely blocked design and carrying out each evaluation in triplicate. This allowed the researchers to determine the significance of these three factors. Throughout the course of the research, both reverse osmosis filtered and distilled water from a Milli-Q® water system and environmental water gathered from Tembladero Slough in Castroville, California were used. It was found that "pure" water and "environmental" water were both found to have the same properties. In order to parameterize the variability in salinity levels in coastal wetlands caused by the interaction of freshwater and marine influences, "low salinity" at baseline saline levels (0 ppt in Milli-Q® water and 0.1 ppt in environmental water) and "high salinity" at 30 ppt, which was achieved through the addition of salt (Coralife Scientific Grade Marine Salt®, Franklin, WI), were evaluated. "Low salinity" was defined as sa The findings of these assessments were compared and contrasted with one another. In addition, the effects of water temperatures of 4 and 27 degrees Celsius were investigated for each distinct kind of water and salt level in order to determine how temperature affects the settling qualities of protozoa and microspheres. The goal of this investigation was to determine how temperature affects the settling qualities of protozoa and microspheres. Immediately before to each experiment, 1×10^6 of *C. parvum* oocysts, *G. lamblia* cysts, and *T. gondii* oocyst surrogates (DG and GB) were mixed together in a 1 L container of water and homogenized by shaking. This was done to ensure that the mixture was consistent. This was done in order to guarantee that each of the constituent parts was dispersed uniformly over the whole.

DATA ANALYSIS

In this particular region, *Giardia* and *Cryptosporidium* are both rather common. The presence of *Giardia* was confirmed by epifluorescent microscopy in all three of the study's subjects: mountain gorillas, domestic cattle, and forest buffalo (Table 4.1). In all, 9% of mountain gorilla feces (11 out of 130) were confirmed to be positive for the parasite *Giardia*. Moreover, the infection was discovered in fecal samples from 33% of the gorilla groups (5 out of 15 groups that were studied). Two of these five infected groups were habituated for the sake of tourism,

while the other three were habituated for the purpose of research. It was determined that all of the positive groups that were detected were more often located in the Kinigi portion of the park. In a way very dissimilar to this, there was evidence of *Giardia* in six percent (eight out of thirteen⁵) of the fecal samples obtained from calves. At least one year had passed since the infected cows were born in every case. Of the total number of cattle for whom demographic information was available, 7 out of 114 female cattle and 1 out of 19 male cattle tested positive for *Giardia*. In male cattle, the percentage of positive tests was 1 out of 19. Out of the whole number of pregnant women who were tested, not a single one of them had a positive result for *Giardia*.

One of the animals that tested positive for *Giardia* was found to have been living in the Shingiro sector, while four of the animals that tested positive for *Giardia* were found in the Nyange sector. *Giardia* was found in one out of every fifty-five samples of forest buffalo, which corresponds to a positivity rate of 2%; the *Giardia*-positive forest buffalo was included in the sample that was taken from the Kinigi area. There were particles that seemed to be cryptosporidium in one percent of mountain gorilla samples (one out of 130), three percent of cow samples (four out of 135), and thirty-six percent of buffalo samples (20 out of 55). Due to the fact that the sample only included a trace amount (0.0001) of these Cryptosporidium-like particles. There was no noticeable grouping detected among the *Giardia*-positive cattle or buffalo samples that were analyzed. When the Bernoulli multinomial approach was used to analyze each species, the significance of the *Giardia*-positive cluster was not altered. It was located in the Kinigi area as the lone cluster of gorillas, and it included the sick buffalo sample (relative risk = 5.25, P 0.0001). characterization on a molecular level. Using the use of molecular techniques, we were able to ascertain the *Giardia* genotype of each positive sample. This was done because taking into consideration the genetic diversity of *Giardia* across host species is an essential component to contemplate when one is thinking about shared parasitism. Due to insufficient DNA being able to be recovered from low numbers of cysts, particularly from slide scrapings, only six out of eleven gorilla samples, one out of eight cow samples, and none of the buffalo sample were able to have PCR products acquired from them. This was the case for gorilla samples, cow samples, and buffalo samples respectively. In spite of this, the sequencing analysis of the GDH locus of the *Giardia* that was found in the samples of the mountain gorillas indicated that five of the samples were of the B-IV subtype, representing three separate family groupings, and that one sample from a third family group was of the B-III subtype. The B-III subtype was found in one of the samples. Sequence analysis of the giardin

locus was used to verify the B-IV subtype in five of the samples, and it was used to confirm the assemblage B in the sixth sample. Sequence analysis of the cow sample utilizing both GDH and -giardin loci was able to identify Giardia assemblage E, which is known to be host specific to cattle. This particular strain of Giardia is only found in cattle.

DISCUSSION

Mountain gorillas who are still living in the Virunga Massif must struggle with a variety of biological restrictions, one of which is the fact that they must share their limited environment with humans. This is one of the challenges they face (Woodford et al., 2002, Cranfield, 2008). Even though gastrointestinal disease is not a primary cause of gorilla mortality (Cranfield, 2008), and in many cases it may be asymptomatic (Gillespie et al., 2004), it is important to consider the long term effects that gastrointestinal parasites may have for general health, survival, and reproduction. This is because gastrointestinal disease is not a primary cause of mortality in gorillas. This is due to the fact that gastrointestinal illness is not among the leading causes of death in gorillas (Gillespie and Chapman, 2006). Mountain gorillas may experience changes in the transmission rates and virulence of gastrointestinal parasites as a consequence of their continual interactions and contact with humans and other animals (Gillespie et al., 2004).

It's possible that this touch might potentially serve as a conduit for the transfer of other illnesses. According to the findings of our study, the current incidence of Giardia in mountain gorillas is 9% (11/130), which is an increase from the 3% prevalence (2/70) that was discovered in 1997. (Sleeman et al., 2000). On the other hand, a separate research that was carried out in 2003 discovered that the actual prevalence was 10% (16/169). (Gaffikin et al., 2003). On the other hand, we came to the conclusion that the prevalence of Giardia in domestic cattle in the region has decreased from 16% (25 / 153) in 2003 to 6% (8 / 135) in recent years. This was the result that we reached after doing our research (Gaffikin et al., 2003). Although pre-weaned calves have a greater risk of becoming infected with Giardia and producing a greater number of cysts, none of the domestic cows that tested positive for Giardia in this investigation were younger than one year of age. Thompson (2000) found that pre-weaned calves have a greater risk of becoming infected with Giardia and producing a greater number of cysts. The investigation that was conducted in 2003 did not provide any information on the age distribution (Gaffikin et al., 2003). Despite the fact that Cryptosporidium has been found in cattle and mountain gorillas in the past (Nizeyi et al., 1999, Nizeyi et al., 2002, Gaffikin et al., 2003), our

investigation did not uncover any verified cases of *Cryptosporidium* infections in either species. It is an interesting fact to note that *Cryptosporidium* has been discovered in cattle and mountain gorillas in the past. In addition, we discovered a little quantity of *Giardia* in the woodland buffalo, with a frequency of around 2%. (1 out of 55). This is the very first occasion that *Giardia* has been identified in a host, and it is extremely likely that it will not be the last. Due to the cross-sectional methodology of this research as well as the little amount of feces that was analyzed, it is likely that the prevalence of both *Giardia* and *Cryptosporidium* in our study species was understated. This is a possibility. This is particularly true when taking into consideration the fact that infected animals may only shed a little amount of parasites sometimes or only do so very seldom. In addition, it is possible that we underestimated the prevalence of parasites in this species since we oversampled buffalo due to the fact that individuals were not visible before to collection. This is because we did not see any buffalo individuals before we collected them. Because of this, it's possible that we underestimated the number of buffalo who were infected with the disease. This study demonstrated a reduced prevalence of *Giardia* in cattle compared to previous published data, which may imply that adjustments in livestock management that were implemented after earlier studies were carried out are decreasing the parasite prevalence in cattle.

The practice of zerograzing is a technique of livestock management that involves the imposition of mandatory constraints on cattle range and the use of mowed grass in lieu of pastured grazing for livestock. This way of managing livestock is also known as rotational grazing. The government of Rwanda started a program in 2006 called "one cow per family" with the goals of reducing the prevalence of chronic childhood malnutrition, increasing the level of food security in households, and producing new sources of income. The program's name comes from the Rwandan proverb "one cow equals one family." Farmers that took part in this program and were awarded cows were required to devise a method of herd management that prohibits the use of grazing at any time (Kim et al., 2012). Not only has it been proved that this management strategy reduces the demands placed on land utilization in areas with a low carrying capacity, but it has also been established that it reduces the prevalence of a number of diseases that are specific to cattle as well as zoonotic diseases. For example, a tick survey that was carried out in order to develop a control strategy for East Coast fever (*Theileria parva*; ECF) revealed that the widespread use of zero-grazing in the northwest of Rwanda resulted in a delayed age-at-first-contact with ticks. This was discovered as a direct result of the delay in the age at which animals first came into contact with ticks. As a direct result of this, there were

a significantly lower number of confirmed cases of East Coast fever in Rwanda's northwest region in compared to other areas of the nation (Bazarusanga et al., 2007). In addition to this, researchers discovered that countries in sub-Saharan Africa that practiced zero-grazing had a reduced overall prevalence of brucellosis (McDermott and Arimi, 2002).

According to Lukuyu et al. (2009), a zero-grazing management system may have the best chance of being effectively implemented with small landholders who possess a limited number of cattle among their families. The farms that may be seen in the region around Volcanoes National Park are shown here in an appropriate manner thanks to this description. There is some evidence to suggest that a reduction in parasitism is an additional benefit of this strategy, despite the fact that the objective of this study was not to investigate the positive effects that zero-grazing has on the health of cattle in this region. The fact that the prevalence of *Giardia* has decreased ever since zero-grazing was introduced is one piece of evidence that supports this hypothesis. *Giardia* was discovered to infect our study species in two different genotypic assemblages: the multi-host species genotype assemblage B was found in mountain gorillas, and the cow-specific assemblage E was identified in cattle. Both of these assemblages were confirmed to be infected with *Giardia*. Within the B assemblage, the BIII and B-IV subtypes were uncovered as separate but related discoveries. Our findings suggest that there may not have been any transmission between species, which is in contrast to the findings of a previous study (Graczyk et al.) that found assemblage A in mountain gorillas, humans, and cattle in Bwindi Impenetrable National Park and suggested that there may have been cross-transmission between the species. Our findings suggest that there may not have been any transmission between species. In contrast to the SSU-rDNA locus that was used in the previous work, which had poor phylogenetic resolution and was prone to misclassification bias, we chose two loci that were sensitive enough to differentiate between different assemblages. This is in contrast to the SSU-rDNA locus that was used in the previous work. Our discovery of assemblage B is a fascinating one since it has been seen in a wide range of mammalian species, including humans, making it one of the most interesting of our discoveries. It has been demonstrated that ensemble B infects non-human primates (NHP): assemblage B was identified in 48% of the samples from a review of isolates from 31 captive NHP species, including western and eastern gorillas. In spite of the fact that very little is known about the *Giardia* species variety seen in mountain gorillas, it has been determined that assemblage B is responsible for infecting non-human primates (NHP).

Moreover, it has been shown that assemblage subtype B-IV has a role in the transmission of NHP (colobus and guenon monkeys) from humans to wild monkeys in Uganda, particularly when combined with a considerable level of ecological overlap (Johnston et al., 2010). The results of the genotypic subtype demonstrate that there is a possibility that humans play a part in the cycle of transmission of *Giardia* into the population of mountain gorillas. It is probable that the transmission cycle of *Giardia* into the mountain gorilla population was not specifically identified by this investigation. Comparing the geographic distribution of B-III and B-IV in humans, a survey of 1,658 human *Giardia* isolates taken from the ZOOPNET-database found that B-III was found in 81% of the samples taken from Africa, while B-IV was found in 19% of the samples taken from that continent. *Giardia* sequences were analyzed, and out of all of them, 83% were classified as belonging to the assemblage subtype B-IV, and 17% were classified as belonging to the assemblage subtype BIII. The opposite trend was seen in samples that originated in North America, with just 14% of samples classed as B-III and 86% of samples classified as B-IV respectively. The distributions in other geographic regions fall somewhere in the center of these two extremes (Sprong et al., 2009). Previous investigations have led researchers to theorize that members of the local community and employees of the park could be responsible for spreading gastrointestinal parasites among the gorillas. This is due in part to the poor sanitation that exists in the towns as well as the regular excursions that park staff make into the gorilla habitat (Graczyk et al., 2002). Given the relative prevalence of the subtypes of *Giardia* that we observed in this study, it may also be appropriate to examine tourists and researchers for their potential role in the transmission cycle of *Giardia* in this area. This hypothesis is based on the fact that the relative prevalence of the subtypes of *Giardia* that we observed in this study. This is the case as a result of the findings of this research, which examined the relative prevalence of the subtypes of *Giardia*. Although a number of studies (Graczyk et al., 2002, Chapman et al., 2005, Goldberg et al., 2007) highlight the fact that human interaction with mountain gorillas is a significant risk factor for the transmission of zoonotic diseases, it is essential to keep in mind that the conservation status of the mountain gorilla is highly dependent upon contact with humans. This is because mountain gorillas live in close proximity to humans in their natural habitats. This contact includes the implementation of behavioral research programs that involve daily human observation, anti-poaching patrols to protect the integrity of the park and the wildlife community within it, and gorilla ecotourism, which generates significant revenue for both the park and surrounding communities while simultaneously educating global citizens about these endangered animals.

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Disease prevention measures have been implemented to reduce disease transmission from humans to gorillas either directly or via environmental transmission (Cranfield, 2008), but these have been primarily enforced for those who come into daily contact with the animals, such as the researchers or the park rangers. This is because these measures are intended to reduce disease transmission from humans to gorillas either directly or via environmental transmission. Because of tourism, an area that is home to an endangered species draws visitors from all over the world. This puts the species in a precarious position since it is more likely to get diseases from visitors. There have only been a few of studies conducted on the part that tourism plays in the spread of zoonotic diseases among primates, which is an important aspect of primate-based ecotourism (Muehlenbein and Ancrenaz, 2009, Muehlenbein et al., 2010).

Giardia was found in all of the species that were evaluated for this study in and around Rwanda's Volcanoes National Park, including mountain gorillas, cattle, and forest buffalo. This discovery was made possible by the fact that this research was conducted in Rwanda. Since the first investigations on the incidence of Giardia in mountain gorillas were carried out between seven and thirteen years ago, there has been no change in this parameter. On the other hand, the prevalence of Giardia in cattle was shown to be decreased in this research. This may be because modifications in livestock management have been applied since the prior investigations were conducted. In addition to this, the genetic subtype analysis that has been reported here implies that tourism or research may have a more significant role in the disease transmission of these gastrointestinal parasites. As a consequence of this, both of these approaches to study on gastrointestinal parasites should be included into any future studies that are conducted on the topic. These future studies should more deeply examine the role of human-primate contact on disease transmission to endangered populations.

Table 1 : Giardia Prevalence In Mountain Gorillas (*Gorilla Beringei Beringei*), Forest Buffalo (*Syncerus Caffer*) And Domestic Cattle In Three Sectors Of Volcanoes National Park, Rwanda, As Determined By Direct Fluorescent Antibody Testing.

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Sampled Group	Mountain Gorilla			Domestic Cattle			Forest Buffalo		
	n	Number Positive	Prevalence (%)	n	Number Positive	Prevalence (%)	n	Number Positive	Prevalence (%)
<i>Overall</i>	130	11	9	135	8	6	55	1	2
<i>Sector</i>									
Kinigi	50	11	9	50	3	6	28	1	4
Shingiro	50	0	0	37	1	3	16	0	0
Nyange	30	0	0	48	4	8	11	0	0
<i>Age</i>									
Pre-weaned (<1 year)				24	0	0			
Post-weaned (>1 year)				111	8	7			
<i>Body Condition Score</i>									
1				8	1	13			
2				46	2	4			
3				63	4	6			
4				15	1	7			
<i>Sex</i>									
Male				19	1	5			
Female (pregnant)				14	0	0			
Female (not pregnant)				100	7	7			

Table 2 Giardia Genotypes In Mountain Gorilla (*Gorilla Beringei Beringei*), Forest Buffalo (*Syncerus Caffer*), And Domestic Cattle In Volcanoes National Park, Rwanda Using Glutamate Dehydrogenase (Gdh) And B-Giardin Loci To Determine Assemblage And Subtype Genetic Information

Species	n	Assemblage result by GDH	Accession # and Reference	Assemblage result by β -giardin	Accession # and Reference
Mountain Gorilla	5	BIV*	JF773755 (Lebbad et al., 2010)	BIV*	AB618785 (Abe and Teramoto, 2012)
Mountain Gorilla	1	BIII*	DQ923581 (Robertson et al., 2007)	B**	EU014389 (Teodorovic et al., 2007)
Domestic Cattle	1	E*	DQ18265 (Langkjaer et al., 2007)	E*	DQ116621 (Di Giovanni et al., 2006)

*100% match to reference sequence

**90% match to reference sequence

CONCLUSION

The epidemiology of fecal protozoa, which is at the crossroads of human, animal, and environmental health, is made up of complicated ecosystems that are impacted by a range of different situations. This field of study is at the intersection of human, animal, and environmental health. It is possible for the presence of protozoa in humans and animals, in addition to environmental factors such as hydrologic (salinity, flow rate, temperature), and landscape effects such as vegetation, to have an effect on the baseline prevalence and dynamics of *Cryptosporidium* and *Giardia* in the environment. These effects can be either positive or negative. The existence of protozoa in human and animal bodies have the potential to moderate these effects. In addition, the condition of these components is in a constant state of transition owing to the fact that the environment around them is always changing.

REFERENCES

1. Atwill, E.R., Tate, K.W., Pereira, M.D., Bartolome, J. and Nader, G. (2006) Efficacy of natural grassland buffers for removal of *Cryptosporidium parvum* in rangeland runoff. *Journal of Food Protection* 69(1), 177-184.
2. Dai, X. and Boll, J. (2006) Settling velocity of *Cryptosporidium parvum* and *Giardia lamblia*. *Water Research* 40(6), 1321-1325.
3. Davies, C.M., Ferguson, C.M., Kaucner, C., Krogh, M., Altavilla, N., Deere, D.A. and Ashbolt, N.J. (2004) Dispersion and transport of *Cryptosporidium* oocysts from fecal pats under simulated rainfall events. *Applied and Environmental Microbiology* 70(2), 1151-1159.
4. Dubey, J.P. and Beattie, C.P. (1988) *Toxoplasmosis of Animals and Man*, CRC Press, Boca Raton, FL.
5. Dumètre, A., Aubert, D., Puech, P.H., Hohweyer, J., Azas, N. and Villena, I. (2012) Interaction forces drive the environmental transmission of pathogenic protozoa. *Applied and Environmental Microbiology* 78(4), 905-912.
6. Fayer, R., Dubey, J.P. and Lindsay, D.S. (2004) Zoonotic protozoa: from land to sea. *Trends in Parasitology* 20(11), 531-536.
7. Furness, B., Beach, M. and Roberts, J. (2000) Giardiasis surveillance—United States, 1992–1997. *MMWR. CDC surveillance summaries : Morbidity and mortality weekly report. CDC surveillance summaries / Centers for Disease Control* 49(7), 1-13.
8. Hogan, J.N., Daniels, M.E., Watson, F.G., Conrad, P.A., Oates, S.C., Miller, M.A., Hardin, D., Byrne, B.A., Dominik, C., Melli, A., Jessup, D.A. and Miller, W.A. (2012) Longitudinal Poisson regression to evaluate *Cryptosporidium*, *Giardia*, and fecal indicator bacteria epidemiology in coastal California wetlands. *Applied and Environmental Microbiology* 78(10), 3606-3613.
9. Juranek, D. (1995) *Cryptosporidiosis: sources of infection and guidelines for prevention. Clinical Infectious Diseases*, 57-61.

12. Kao, C.M., Wang, J.Y., Lee, H.Y. and Wen, C.K. (2001) Application of a constructed wetland for non-point source pollution control. *Water Science and Technology* 44(11-12), 585-590.