Results of routine examinations for parasitic infections of humans from laboratory-submitted samples in Gauteng, North West and Mpumalanga provinces between 2009 and 2010

By

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SUMMARY

RESULTS OF ROUTINE EXAMINATIONS FOR PARASITIC INFECTIONS OF HUMANS FROM LABORATORY-SUBMITTED SAMPLES IN GAUTENG, NORTH WEST AND MPUMALANGA PROVINCES BETWEEN 2009 AND 2010

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Very few recent studies have been done in South Africa on the occurrence or prevalence of parasites in humans. Based on the results of routine examinations for parasitic infections conducted in the Microbiology Laboratory of Ampath in Pretoria, this study focuses on the spectrum of parasites diagnosed in samples from humans in Gauteng, North West and Mpumalanga provinces between 2009 and 2010. Database searches for results of samples in which parasites were positively identified were conducted using the laboratory's internal software system. Data of the positive results were exported and sorted according to date, specimen type, parasite identified, patient age, gender and geographic locality. Results showed that a total of 24 different species of parasites were identified in the laboratory over the two-year period. The overall numbers of parasites identified, according to the data searches, were 863 and 1061 in 2009 and 2010, respectively. The following parasites were identified: *Entamoeba coli*, *Entamoeba histolytica, Endolimax nana*, *Acanthamoeba* sp, *Giardia intestinalis*, *Trichomonas hominis, Trichomonas vaginalis*, *Chilomastix mesnili*, *Blastocystis hominis*, *Cryptosporidium* spp, *Cystoisospora belli*, *Sarcocystis* sp, *Enterobius vermicularis*, *Ascaris lumbricoides*, *Trichuris trichiura*, ancylostomatids (hookworm), *Taenia saginata*, *Taenia solium*, *Dipylidium caninum*, *Inermicapsifer madagascariensis*, *Bertiella studeri*, *Schistosoma haematobium*, *Echinococcus granulosus* and *Cordylobia anthropophaga*. In both years, the majority of cases originated from Gauteng Province followed by North West and Mpumalanga provinces. The data were summarized descriptively and compared with available published records.

Chapter 1
INTRODUCTION

Parasitism as a biological phenomenon is a form of relationship between two species that through evolution has proven to be enormously successful. Many parasites conveniently utilise their hosts without having to compete for survival in the broader ecosystem. This has required adaptation which has become so close that most parasites cannot exist without their hosts. A general prerequisite for the success of this coexistence is that the adverse effects do not kill the host, since death of the host would inevitably also impact negatively on the parasite. Parasitic infections are therefore rarely fatal under natural conditions in undisturbed ecosystems. In the medical context, parasitic infections and diseases respond well to treatment in most instances. Therapy however is not always readily available, especially in countries and areas with substandard healthcare systems.

Parasites are more prevalent in tropical and subtropical climates, favoured by uncontrolled population growth, urban expansion, inadequate governance and poor public health standards.[6] Key factors that negatively impact on the health of underprivileged communities globally include malnutrition, contaminated drinking water, general lack of hygiene and non-existing or inadequate sanitation.[22] In sub-Saharan Africa the mortality rate attributable to diarrhoeal diseases resulting from unsafe water, inadequate sanitation and hygiene was 629 000 in 2001, which compared to developed countries, is regarded as extremely high.[22] In the South African context, the beneficial climate combined with uncontrolled urbanization, settlements, population growth and inadequate municipal service delivery create the optimal environment for the occurrence of parasitic infections.

Diagnostics for parasitic infections of humans are done routinely at the Microbiology Laboratory of Ampath in Pretoria. Parasites have been identified in various samples such as faeces, urine, genital swabs, organ tissues, aspirates and contact lens fluids. Isolated and preserved helminths and arthropods are also sent to the laboratory for identification. There is a lack of recently published data on the current status of parasitic infections of humans in South Africa. In an attempt to remediate the lack of information, the aim of this project was to focus on the results of examinations for parasites obtained at the Microbiology Laboratory of Ampath from samples submitted from Gauteng, North West and Mpumalanga provinces between 2009 and 2010.

1.1 Literature review of parasites isolated at the Microbiology Laboratory of Ampath submitted from Gauteng, North West and Mpumalanga provinces between 2009 and 2010

1.1.1 Entamoeba coli

Entamoeba coli is a cosmopolitan, non-pathogenic protozoan parasite that inhabits the lumen of the large intestine of humans. Trophozoites are 15-50 µm in diameter and have a slow non-directional movement.[5] Mature cysts are 10-31 µm in diameter, contain eight nuclei and chromatoidals and are mostly identified in faecal samples.[5,24] Cysts are transmitted though faecal contamination, they are ingested and develop into trophozoites that become established in the lumen of the large intestine and multiply by binary fission.[5]

Worldwide prevalence has been estimated at 28% but it differs from one region to the next.[24] Regarding the prevalence in South Africa, only data of a single survey conducted in a primary school near Durban are available in which 56% of pupils were found positive for *E. coli* infection.[36]

1.1.2 Entamoeba histolytica

Entamoeba histolytica is a protozoan parasite of usually the colon and the cause of amoebic dysentery, also known as entamoebiasis.[33] It is unique amongst the human amoebae that the trophozoites of certain strains have the ability to invade and destroy almost any tissue.[5,13]
The most common tissues affected are the intestinal mucosa and the liver parenchyma and to a lesser extent brain and skin tissue.[13] It was stated by Stauffer & Ravdin (2003) that *E. histolytica* is possibly one of the foremost sources of diarrhoea in young children with considerable harmful health effects in this age group.

Trophozoites vary in size from 10-60 µm in diameter.[5,13] Trophozoites in dysenteric stools contain vacuoles with ingested red blood cells.[5,13] Mature cysts which are 10-18 µm in diameter contain 4 nuclei and 'cigar-shaped' chromatoidals.[5] Infection occurs via the faecal-oral route through ingestion of cysts in food or drinking water that has been contaminated with human faeces.[5] Cysts can be easily spread by food handlers.[13,24] After ingestion, cysts develop into trophozoites in the caecum and subsequently settle in the glandular crypts of the colon where they multiply by binary fission.[5,24] Depending on the pathogenicity of the strain involved as well as host-specific factors, trophozoites may invade the colonic mucosa as a primary site causing ulceration and inflammation from where they can spread to extraintestinal organs and tissues producing secondary lesions.[5,24]

Entamoeba histolytica has a worldwide distribution and is prevalent in countries with deprived socioeconomic conditions.[13] It has been estimated that over 600 million people harbour invasive strains of the amoeba but the infection only manifests clinically in about 20% of infected individuals.[7,24] The prevalence rate has been shown to be as high as 80% in some parts of the world.[24] It is regarded as the fourth leading cause of death and third leading cause of morbidity due to protozoal infections.[7]

In South Africa the prevalence of entamoebiasis is largely unknown. In a survey conducted on scholars of a primary school near Durban, a prevalence of 11.1% was determined.[36] In a community cross sectional survey in Durban conducted by Gathiram & Jackson (1987)[26], 1% of individuals were found to be infected without showing clinical signs. Eventually, 90% of the infected subjects tested negative on faecal examination within a year whereas the remaining 10% developed amoebic colitis.[26]

1.1.3 Endolimax nana

Endolimax nana is a common, non-pathogenic amoeba of the colon.[5] It has a cosmopolitan distribution.[5] Trophozoites are 6-15 µm in diameter, whereas mature cysts are 5-14 µm in diameter and contain four nuclei.[5,13,24] Infection is acquired by the ingestion of cysts present in contaminated food and water or other sources.[5] The prevalence of *E. nana* is similar to that of *E.coli*, prevalence is higher in warm, moist climates and in areas were there is inadequate personal hygiene.[5] In a survey conducted on children of a primary school located near Durban, Kvalsvig (1988)[36] found a prevalence of 32.5 % for *E. nana*.

1.1.4 Acanthamoeba sp

Acanthamoeba is present in all types of environments worldwide and are facultative parasites of man.[13] Although infections are rare, they are often severe or even fatal.[5,13] Life cycle stages comprise a trophic stage and a cyst stage which is highly resistant to desiccation and chlorine.[5,13]

The trophozoites of *Acanthamoeba* are extremely variable in size and shape and range from 10-60 µm in diameter.[5] Cysts are spherical and range from 7-25 µm in diameter.[5] Sources of infections appear to be dust and standing water.[5] Entry portals are probably inflamed, mechanically damaged or ulcerated skin, mucous membranes and cornea.[5]

Acanthamoeba species have been implicated as causative agents in granulomatous encephalitis, ocular infections such as keratitis and granulomatous lesions of the skin, kidneys, liver, spleen, uterus and prostate.[5,13] *Acanthamoeba* keratitis is being diagnosed more frequently since there is a correlation between infection and the wearing of contact lenses.[13]

1.1.5 Giardia intestinalis

Giardia intestinalis is the most widespread human protozoan flagellate enteropathogen with infection potentially causing acute or chronic diarrhoea and malabsorption.[13] The leaf-like trophozoite is dorsally rounded and has a ventral sucking disc that is used to anchor themselves to epithelial surfaces.[5,24] Trophozoites are 9-21 µm long and 5-15 wide.[5] Each trophozoite has four pairs of flagella and contains a pair of nuclei situated towards the anterior end lateral to the midline.[24] The ovoid cysts are 8-12 µm long and 7-10 µm wide and depending on their maturity they contain two or four nuclei with internal structures.[5]

The trophozoite inhabits the crypts of the proximal small intestine and multiplies by binary fission.[13,24] During transit in the colon, trophozoites encyst.[13,24] Environmentally resistant cysts are voided in the faeces and are the infective stage.[13,24] Cysts are transmitted through either intimate contact or via contaminated water or food.[13,24] Different strains of this parasite can also be found in other mammals, which include dogs, cats, cattle, pigs, sheep and horses.[58,59] However, the frequency of zoonotic transmission is uncertain and appears to be of little significance.[58,59]

Age is a major risk factor for susceptibility with infections more prevalent in infants and young children.[24,58] High prevalences of giardiasis are found in day-care centres, schools and residential establishments.[24,58] Outbreaks from exposure to contaminated swimming pools and other water sources are renowned.[24] Disease occurs sporadically in adults and outbreaks usually occur in groups of travellers.[24]

Giardiasis is found worldwide but prevalence is highest in developing countries, reaching up to 20-30%.[13] Two hundred million people suffer from clinical giardiasis in poor and average income nations annually and approximately 500 000 new cases are reported each year.[59] In South Africa, the prevalence of *G. intestinalis* was found to be 15.9 % in a primary school near Durban.[36] Adams *et al*. (2005)[1] found a prevalence of 17.3% in school children in Cape Town. *Giardia* cysts have been found in all types of water including surface water, sewage and treated effluents in South Africa.[34]

1.1.6 Trichomonas hominis

Trichomonas hominis is probably the most common intestinal flagellate of man inhabiting the caecal area of the colon.[5] It is regarded as largely non-pathogenic and has a trophozoite stage only, which is found exclusively in diarrhoeic stools.[5] Trophozoites vary in length from 5 to 14 µm and bear three to five anterior flagella.[13,24]

Another flagellum projects backwards and is joined to the body by an undulating membrane; an axostyle runs ventrally through the centre of the trophozoite and protrudes through the posterior end; there is a cytostome present opposite the undulating membrane and an ovoid nucleus near the anterior end of the trophozoite.[5,24] Infection occurs by ingestion of contaminated food. Once trophozoites reach the caecum they feed on enteric bacteria and multiply by binary fission.[5] The incidence of this parasite is generally low in different populations, ranging from 1% to 12%.[5] *Trichomonas hominis* was detected in only 0.5% of children in a survey in Nigeria; it was diagnosed exclusively in diarrhoeal stools.[46] Prevalence is not well documented in South Africa.

1.1.7 Trichomonas vaginalis

Trichomonas vaginalis is a pathogenic flagellate of humans that normally inhabits the vagina and prostate gland.[5] It only exists in the trophozoite stage. It is 7-23 µm long, has four free flagella and a fifth one along the outer margin of an undulating membrane.[5] It is mainly transmitted during sexual intercourse (venereal transmission) or nonvenereal through communal bathing or sharing of bathing and toilet articles.[5,24] According to Beaver *et al.* (1984)[5], trichomoniasis is the most commonly acquired sexually transmitted infection. Infection can be spread to female infants as they pass through the birth canal.[5,24]

Most infections are generally asymptomatic or cause mild irritations; prevalence rates stated in the literature may thus not always reflect the true infection rates.[24] In the acute form, shortly after infection, there is desquamation of the vaginal epithelium, followed by leukocytic inflammation with the typical presenting clinical signs of vulvar pruritus, vaginal pruritus, dysuria and if heavy infections prevail, purulent vaginal discharge.[5,24] In the subsequent chronic stage the discharge looses its purulent appearance and infection persists for at least 2 years.[13,24] Trichomoniasis in men can be symptomless or manifest clinically as recurring urethritis with subsequent prostatitis.[5]

The parasite occurs worldwide in urban and rural areas.[13] It is frequently coexistent with other infections such as candidiasis, gonorrhoea, syphilis or human immunodeficiency virus infection (HIV).[13] Infected patients should be screened for other sexually transmitted diseases (STDs) which are often of greater clinical significance.[13]

In South Africa, several surveys have shown the coexistence of *T. vaginalis* with other STDs. According to Mhlongo *et al.* (2010)[39], trichomoniasis was the most frequently detected STD pathogen in a survey of patients with vaginal discharge syndromes. The prevalence was 19% in individuals from Cape Town and 34% in individuals of the Johannesburg study group.

A survey to assess the risk of being infected with a STD at the time of presentation for termination of pregnancy was conducted in a pregnancy clinic in Pretoria. A mixed infection with *T. vaginalis* and chlamydia was found in six out of nine women.[16] *Trichomonas vaginalis* was the most common pathogen found in women without visible vaginal discharge.[16]

1.1.8 Chilomastix mesnili

Chilomastix mesnili has a trophic and a cystic stage and is regarded as a widespread, non-pathogenic commensal that inhabits the caecum.[24] Cysts are easily recognized by their characteristic pear-shape appearance.[5,24] They are 7-10 µm long, 4-6 µm wide and have a slightly conical raised narrower end.[5,24] Trophozoites are pyriform, 6-20 µm long, 2-10 µm wide and contain 1 nucleus, 3 free anterior flagella and a well-defined cytostome.[5]

Transmission of cysts occurs via the faecal-oral route.[13,24] In the caecum, trophozoites multiply by binary fission.[5] The prevalence of *C. mesnelli* infection can range from 1% to 10% or more, depending on the age of the individuals and the population group.[5] Kvalsvig (1988)[36] reported a prevalence of 4.4% in pupils of a primary school near Durban.

1.1.9 Blastocystis hominis

Blastocystis hominis is a parasite that inhabits animal and human intestinal tracts.[57] The most common form seen in faeces are the hyaline cysts containing a central vacuole.[24] Cysts vary in size between 6-40 µm in diameter.[24] Transmission is thought to take place via the faecal-oral route possibly by contaminated food or water.[24] Infection with *B. hominis* has been successfully linked to the quality of drinking water.[56] It has also been considered to have zoonotic implications and it is thought that animals such as pigs, chickens and particularly dogs and cats, in which high prevalences have been reported act as major reservoirs for human infection.[57]

Different opinions exist about its pathogenicity; in diarrhoeic patients with large numbers of cysts shed, it is considered to be clinically important.[24] More than five cysts per high-power field of the microscope could indicate pathological involvement.[24] Infection has also been related to signs of diarrhoea and cutaneous rashes in immunocompromised individuals.[29] High prevalences of *Blastocystis* infection of up to 69% have been reported from economically poorer nations.[59]

1.1.10 Cryptosporidium spp

Cryptosporidiosis is one of the major public health concerns as it is increasingly implicated in outbreaks of water-borne diarrhoea among the poor of developing countries.[13] *Cryptosporidium* has been identified as a widespread cause of childhood diarrhoea.[13] The parasite colonises the brush border of epithelial cells of the small intestine and partially colon.[5]

Infection takes place by ingestion of sporulated oocysts. Sporozoites are released in the small intestine and are engulfed by the microvilli of the epithelial cells. In the brush border merogony, gamogony and subsequently sporogony takes place.[5] The latter will result in the formation of sporulated oocysts which are 4-5 µm in diameter and contain 4 sporozoites.[24] Oocysts are mostly excreted with faeces, however sporozoites of some oocysts are released inside the host which can result in a new infection of the epithelial cells (autoinfection).[24]

Cryptosporidium oocysts have been found in surface water, sewage and treated effluents in South Africa.[34] Studies in Durban showed that *Cryptosporidium* was the second most common enteric pathogen isolated from children admitted to hospital with gastroenteritis.[44] Infection rates varied between 1.2 and 20.9%.[44] There was a mortality of 10% in *Cryptosporidium*-infected children.[44] Prevalence of infection was shown to increase in the summer months.[44] A survey in the Venda region of the Limpopo province showed that age groups mostly affected by *Cryptosporidium* were 2- to 5-year-olds (28.6%) and 50- to 59-year-olds (50.0%).[51] HIV positive individuals included in the test group showed a 12.5% prevalence of infection.[51]

1.1.11 Cystoisospora belli

This coccidian is frequently associated with chronic diarrhoea in immunocompromised individuals, especially patients with AIDS.[13] Oocysts are elongate-ovoidal in shape and are 20-30 µm long and 10-19 µm wide.[5] Freshly passed oocysts are unsporulated and contain one or two sporoblasts. Following sporogony outside the host, oocysts become sporulated, with 2 sporocysts containing 4 sporozoites each.[5,13] The sporulated oocyst is the infective stage and infection is acquired by ingestion of food or drinking water contaminated with faeces. Development is intracellular and occurs in the intestinal epithelium.[5] As with other coccidian infections disease is usually self limiting, and infection can be prevented by maintaining good personal hygiene standards.[24] Infection is less common in developed countries; prevalence studies indicated that the prevalence in HIV patients with diarrhoea was 13% in Africa and the Caribbean, which was substantially higher than that of developed countries (0.5%).[13]

1.1.12 Sarcocystis spp

Sarcocystis spp follow an indirect life cycle. Two species are of relevance in humans, namely *Sarcocystis hominis* and *Sarcocystis suihominis.*[5] Humans act as definitive hosts for both.[5] Cattle (*S. hominis*) and pigs (*S. suihominis*) act as intermediate hosts with asexual reproduction first taking place in vascular endothelial cells (merogony) and subsequently in skeletal or cardiac muscle fibres (endodyogeny) which results in the formation of cysts.[5] The cysts forming in the musculature are the infective stage for humans.[5]

Infection is acquired by the consumption of uncooked beef or pork.[5] Released cystozoites enter cells of the lamina propria of the intestinal mucosa where they undergo gamogony and sporogony.[5] Sporulated, fully infective oocysts are shed in the faeces and are the infective stage for intermediate hosts.[5]

Infections in humans are accompanied by nausea, vomiting, abdominal pain and diarrhoea within 24 hours following the consumption of cyst-infested beef or pork.[24] Infections are more frequently seen in HIV-positive patients.[24] Oocysts are very delicate structures and often disintegrate in the gut, releasing the ovoid sporocysts and range in size from 13.6-16.4 µm by 8.3-10.6 µm.[5] Each sporocyst contains 4 elongate sporozoites and a granular residual body.[5,13] *Sarcocystis* infection in man is not well documented in South Africa, but its occurrence in domestic and sylvatic animals has however been reported in a number of studies.[47] This parasite has been characterised as one of the important food-borne parasites.[20]

1.1.13 Enterobius vermicularis

Commonly called the ‘pinworm’, this worm has a cosmopolitan distribution. It is found in areas with a temperate climate, and infections are more frequent in children.[24] Adult worms inhabit the caecum, appendix and adjacent portions of the ascending colon.[5] The male is 2-5 mm in length and has a strongly curved posterior end.[5,24] The female is 8-13 mm long and has a long, sharply pointed tail.[5,24] The size of eggs range from 50-60 µm by 20-30 µm, they are elongate-ovoidal, characteristically flattened on the one side and have a thick colourless shell.[5] Gravid females migrate down the colon, rectum and out of the anus, where they deposit the eggs on the skin of the perianal area.[5] A mean of 11 105 eggs can be deposited by a single female.[5] Eggs containing infective larvae are swallowed, they hatch upon reaching the intestine and larvae then develop into adult worms upon reaching the caecal region.[5]

Infection takes place by the ingestion of embryonated eggs through anus-to-mouth transmission, soiled clothes, contaminated linen or objects and through contaminated air.[5,24] In heavy infections there is marked anal pruritus caused by migrating female worms leaving the anus. Scratching of the anus can lead to secondary bacterial infection.[13,24]

Nervousness and insomnia have been attributed to enterobiasis in children.[5] Rectal discomfort can occur due to large numbers of worms in the rectum.[5] Urinary, genital and abdominal disturbances due to enterobiasis have been recorded in young girls and older women.[5] In a survey conducted on children in Cape Town, a prevalence of 0.6% was reported.[1]

1.1.14 Ascaris lumbricoides

Ascaris lumbricoides is a cosmopolitan intestinal parasite of humans.[24] Man is the only definitive host, 1 in 4 of the world's population is regarded as infected and poor communities are more frequently affected.[7,24] The worm has a long tapering cylindrical body, is flesh-coloured with a whitish streak along its side.[5] Mature males are 15-31 cm long and 2-4 mm in diameter, the posterior end of the male is curved.[5] The female is larger than the male, 20-35 cm long by 3-6 mm in diameter.[5] Fertilized eggs are broadly ovoid with a thick transparent mammilated brownish shell.[5] Fertilized eggs are subspherical, 75 µm long and 50 µm wide, spherical forms are 60 µm in diameter.[5] Unfertilized eggs are elongate with a smooth shell and are 90 µm long.[5] Eggs are resistant to a wide range of chemicals and adverse environmental conditions.[5]

Eggs are shed with faeces into the environment and develop into infective, embryonated eggs.[5] Following ingestion of embryonated eggs, larvae hatch in the stomach and undergo a hepato-tracheal migration using the blood stream.[5] In the intestine, larvae develop into adult worms in the jejunum.[5] An adult worm has an average lifespan of 1 year.[5]

Children often get infected by ingesting eggs found in soil contaminated with human faeces.[24] Raw vegetables fertilized with human manure are also a common source of infection.[24] Disease is caused by migrating larvae or adult worms. Larvae migrating through the lungs can cause verminous pneumonia, also known as Löffler's syndrome.[13] Luminal infection by immature and adult worms is mostly asymptomatic.[13] However, heavy infections can cause impaction with resulting obstruction, volvulus or intussusception.[7] Aberrant migration into the appendix, bile duct, pancreatic duct or larynx may result in appendicitis, jaundice, pancreatitis or worms being vomited up.[7]

Worldwide prevalence is estimated to be 25%.[24] According to de Silva (2003)[17], the prevalence in sub-Saharan Africa is 25%, however, in some areas it is as high as 95%.[24] The prevalence of ascariasis in schoolchildren from rural KwaZulu-Natal was found to be 19.4%.[50] In Cape Town, the prevalence was found to be 24.8% among children of selected communities.[1] Appleton *et al*. (2009)[2] reported a prevalence of 81.7%–96.3% among slum-dwelling children in Durban.

1.1.15 Trichuris trichiura

Also known as the human whipworm, *T. trichiura* has a worldwide distribution.[5,24] Man is the only definitive host.[5] The adult female and male worms have a thickened posterior part and a thinner anterior part, giving members of the genus the appearance of a whip.[24] The male worm is 30-45 mm long, with a coiled tail.[5] Female worms are 35-50 mm long and are bluntly rounded at the posterior end.[5] The eggs are characteristically lemon-shaped and bear a mucoid plug at each pole; they measure 50-54 µm by 22-23 µm.[5,24]

The worm lives attached to the wall of the caecum, colon and rectum.[5] Eggs are passed with human faeces into the environment, where they become embryonated and infective within 10-14 days.[5] Humans become infected by ingestion of embryonated eggs.[5] The epidemiology is very much similar to that of *A. lumbricoides*. *Trichuris trichiura* is non-migratory.[7] Infection occurs directly from stale faeces by ingestion of the ova via contaminated soil.[13]

Symptomatic infection is usually only recognized in small children and only achieves public health importance in urban slums.[7] Main clinical effects are bloody diarrhoea without fever, rectal prolapse, anaemia, stunting growth, poor cognitive function and eosinophilia.[7] It has been estimated that 46 million people worldwide are infected with an annual mortality of 60 000.[13] Prevalence in sub-Saharan Africa was estimated to be 24% in 2003.[17] Following a survey on school children conducted in 2005 in Cape Town, South Africa, a prevalence of 50.6% was reported[1.] In another survey involving 2- to 10-year-old children in urban slums in Durban, a prevalence of infection of 54.5 to 86.2% was reported.[2]

1.1.16 Ancylostomatids (hookworm)

Human hookworm infection is mainly caused by *Ancylostoma duodenale* and *Necator americanus.*[5] Infections are widespread in tropical and subtropical climates.[5] The predilection site of the worms is the small intestine where they suck blood.[13] The worms are about 1 cm long and have a large buccal capsule armed with teeth (*A. duodenale*) or cutting plates (*N. americanus*) used to pierce the mucosa.[5,7] Eggs are shed in the faeces. A first larval stage hatches and develops into an infective third-stage larva which can survive for up to 3 months in the environment.[7] Infection is usually percutaneous.[5] Following infection, larvae undergo a blood-tracheal migration before settling in the small intestine where they develop into adults.[5,13] Clinical effects of the blood-sucking gut stages are the result of loss of iron and protein.[13]

The prevalence of hookworm infection for sub-Saharan Africa was reported to be 29% in 2003.[17] In South Africa, hookworm infection prevalence among school children in Cape Town was reported to be 0.08% only.[1] A mean prevalence of 4.7% is reported from children living in urban slums in Durban.[2] Hookworms have an affinity for moist hot conditions with coastal regions and areas with extremely high temperatures having higher hookworm prevalences.[32] Gold mines have also experienced major problems with infections, where the underground environment creates perfect conditions for larvae to develop.[13]

1.1.17 Taenia saginata

Taenia saginata, the beef tapeworm or unarmed tapeworm, utilizes humans as the only definitive host and cattle as the predominant intermediate host.[13] The worm typically consists of a scolex, a neck and a long strobila.[5,13] The scolex has 4 suckers but is devoid of a hooklet-bearing rostellum.[13] The gravid proglottids at the end of the strobila contain a uterus composed of 15-20 main lateral branches filled with eggs.[5] Adult worms of *T. saginata* are large, and can reach a length of up to 10 m.[5,13] Eggs are spherical, 31- 43 µm in diameter, with a thick striated embryophore (shell) and contain an oncosphere that has 3 pairs of hooklets.[5] They are morphologically identical to the eggs of *T. solium.*[5]

Human infection is acquired by eating undercooked beef containing cysticerci.[5,13,24] Following ingestion by the human host, the cysticercus develops into the strobilar stage in the small intestine and starts shedding proglottids 3 months after infection.[5,13] The gravid proglottids containing eggs detach from the strobila and are motile. They either leave the host spontaneously (*i.e.* without defaecation) or with the faeces.[13] Cattle get infected by ingesting eggs while grazing in areas contaminated with human effluent (sewage) or by handling from infected people which is important in feedlots.[5,13,24]

In humans, motile proglottids are noticeable when felt emerging from the anus or seen in faeces, which can cause a certain amount of discomfort to a patient.[24] Most infections are asymptomatic.[13,24] *Taenia saginata* has a cosmopolitan distribution and is prevalent where raw or undercooked beef is habitually consumed.[5,13] Beef containing cysticerci can have a significant economic impact since infected carcasses are chilled which impacts negatively on the meat quality and are subsequently downgraded. Economic losses in Iran, due to 0.02 % of infected carcasses being condemned, amounted to $400,000 annually.[59] In endemic areas in Africa economic losses are much higher, because of prevalences usually being greater than 20%.[59] The prevalence of *T. saginata* infection in humans has been shown to be extremely high in East Africa, particularly in Ethiopia and Sudan.[32]

1.1.18 Taenia solium

Similarly to the beef tapeworm, the only definitive host for *Taenia solium,* also known as the pork tapeworm or armed tapeworm of man, are humans.[5,13] The domestic pig acts as the predominant intermediate host.[5,13] Humans can also act as intermediate hosts.[5,13] The gross morphology of the adult worm is similar to that of *T. saginata*. In contrast to *T. saginata*, the scolex of the pork tapeworm shows typical taeniid features and bears an armed rostellum.[5]
The uterus in gravid proglottids has only of 7-13 main lateral branches.[5] Adult worms are 3-4 m in length.[5] Eggs are morphologically and morphometrically identical to those of *T. saginata.*[5,13]

Human infection with the adult worm is acquired by eating undercooked pork containing viable cysticerci.[13] Gravid proglottids containing eggs detach and are motile.[5] They either leave the host spontaneously or are shed with the faeces.[13] Intermediate hosts, domestic pigs or accidentally also humans, become infected by ingestion of eggs.[5]

Similar to *Taenia saginata*, infection with the strobilar stage of *T. solium* is trivial.[7] However, in the event of human cysticercosis, effects can be severe if the brain is affected.[7] Also known as cerebral cysticercosis or neurocysticercosis, infection clinically often presents as epilepsy.[5] In low or middle income countries where *T. solium* infection is endemic, approximately 30% of epilepsy cases may be caused by neurocysticercosis.[9]

A review of the status of human neurocysticercosis in eastern and southern Africa revealed the prevalence of infected pigs from slaughterhouses to be 0.5–25.7%, and faecal surveys in KwaZulu-Natal Province indicated a prevalence of <1 to 16.0%.[37] A study by Krecek *et al*. (2008)[35] revealed the prevalence of cysticercosis in pigs from 21 villages of the Eastern Cape Province to be 64.6%.

1.1.19 Dipylidium caninum

Dipylidium caninum is the most common tapeworm of dogs and cats in SA and worldwide.[52] Humans can act as accidental definitive hosts.[52] Intermediate hosts are fleas (*Ctenocephalides canis*, *C. felis* and *Pulex irritans*) and lice (*Trichodectes canis*).[5] The strobilar stage is 10-70 cm in length and has pumpkin-seed-shaped gravid proglottids.[5] Gravid proglottids contain egg capsules with each containing 2-38 eggs.[5] The proglottids are motile and can leave the definitive host spontaneously, *i.e.* without defecation.[5] They are often seen crawling on the surface of freshly-passed faeces.[24] Flea larvae ingest the eggs of the tapeworm while feeding on faecal debris and disintegrated proglottids.[24] The eggs hatch in the intestine of the flea and develop into cysticercoids.[5]

Human infection is very rare and mostly reported in children who become infected while playing with dogs and cats.[5,52] Infection takes place by accidentally swallowing adult fleas or by dogs and cats licking the face of humans and transferring crushed fleas.[24] Infection in humans is largely asymptomatic.[5,24] *Dipylidium caninum* has been reported in several surveys conducted on dogs and cats in communities in South Africa.[3,40,41,42]

1.1.20 Inermicapsifer madagascariensis

This anoplocephalid tapeworm parasitizes hyracoids, rodents, lagomorphs and occasionally humans in mainly Africa but also Cuba, Indian Ocean islands, SE Asia and elsewhere.[5,13,23,28,31] The life cycle is unknown.[5] The worm is 42 cm long and 2.6 mm wide.[5] The strobilar stage has rice grain-shaped gravid proglottids which contain eggs that are enclosed in capsules; each capsule contains 6 or more eggs.[5]

Human infections, reported from children and young adults only, are rare and largely asymptomatic.[5] In Africa, infections have been reported from Kenya, Democratic Republic of Congo, Zimbabwe, Zambia and South Africa.[5,12,23,27,28,31] This parasite is common in rodents south of the Sahara.[5]

1.1.21 Bertiella studeri

The genus *Bertiella* includes cestodes that parasitize primates, rodents and other mammals[18]. *Bertiella studeri* is a parasite of primates.[5] The tapeworm is accidentally transmitted to humans by the ingestion of oribatid mites, which are the intermediate hosts.[18] Oribatid mites are important components of the soil fauna and become infected by the ingestion of eggs shed in the faeces of definitive hosts.[18] *Bertiella studeri* is 275-300 mm long and 10 mm wide.[5] Gravid proglottids detach in groups of approximately 20 at one time from the mature strobilar stage.[5] The irregularly shaped ovoid eggs, 45-46 µm by 49-50 µm, contain an oncosphere covered by an inner shell with a typical bicornuate protrusion on one side.[5]

High rates of infections in children have been reported.[18] Infections in humans are mostly asymptomatic.[18] Occasionally, cases presenting with severe recurrent abdominal pain with intermittent vomiting have been reported.[18] Regarding Africa and its islands, human infections have been reported from Mauritius, East Africa, Democratic Republic of Congo, Gabon and South Africa.[18,23]

1.1.22 Schistosoma haematobium

Schistosoma haematobium causes genitourinary schistosomiasis, also known as urinary bilharziasis.[5] Natural infections, other than in man, have been found in baboons, monkeys, rats and pigs.[13] The predilection sites are the terminal venules in the wall of the bladder, the genitourinary system and the pelvic plexus.[13] The intermediate snail host in South Africa is *Bulinus africanus.*[13]

Adult flukes are white, roundworm-like and 1-2 cm long.[5] Eggs are ovoid, have a distinct terminal spine and measure 112-170 µm by 40-70 µm.[5] After oviposition, the already embryonated eggs break though the urinary bladder wall into the lumen and are then discharged in the urine.[5] Eggs hatch when the urine becomes diluted with water and free swimming miracidia will then infect the snail host upon contact.[5] Inside the snail host the miracidia develop into fork-tailed cercariae that are released from the snails and subsequently infect humans percutaneously when they enter infested freshwater bodies.[5,13]

Clinical signs seen in *S. haematobium* infection include haematuria, dysuria, and nutritional deficiencies caused by lesions of the bladder and kidney failure.[13] There is also an elevated risk of bladder cancer.[13] The female reproductive organs are frequently involved and genitourinary schistosomiasis is a risk factor for acquisition and transmission of HIV. Highest prevalences and intensity of *S. haematobium* infection are found in children with growth retardation being reported.[13] In an epidemiological study conducted in Limpopo Province, an infection rate of more than 70% was found in children under the age of 14 years.[62] Another study in Mpumalanga showed a prevalence of 35.1% in primary-school children.[43] In a survey conducted on schoolchildren in rural KwaZulu-Natal Province a prevalence of 68% was reported.[50]

1.1.23 Echinococcus granulosus

Echinococcus granulosus has a cosmopolitan distribution and belongs to the cestode family Taeniidae.[21,52] Dogs and sylvatic carnivores act as definitive hosts, carrying the strobilar stage in the small intestine.[5] The strobilar stage is only 3-6 mm long and consists of an armed scolex and 3 proglottids.[5] As for all the taeniids, gravid proglottids are motile and can leave the definitive host spontaneously.[49] Eggs are typically taeniid and cannot be differentiated.[5] Natural intermediate hosts are principally herbivorous domestic animals, especially cattle, sheep and goat which become infected by ingesting eggs while grazing.[5,21] The up to fist-size metacestode developing in intermediate hosts is known as a hydatid or hydatid cyst which is predominantly located in the liver and lungs.[5,7,21]

Infection in intermediate hosts is also referred to as hydatidosis or cystic echinococcosis.[21,24,60] In natural intermediate hosts, infection is asymptomatic and is only diagnosed during meat inspection.[21] Definitive hosts become infected by ingesting hydatids, with raw offal being the primary source of infection.[24] Most importantly, humans can also act as accidental intermediate hosts.[60] Humans become infected mostly through close contact with infected dogs whose coats are contaminated with eggs or through consumption of contaminated vegetables and drinking water.[52] Hydatid disease is considered to be the leading zoonotic disease of canine origin.[52] Similarly to natural intermediate hosts, human hydatidosis mostly affects the liver and lungs.[5,52] Cysts can grow to very large sizes in humans, with records of up to 50 cm in diameter.[52] Wide ranges of presenting symptoms and signs have been described in patients affected by cystic echinococcosis.[5,21,52]

This tapeworm is endemic in South Africa.[60] In South Africa it has been found that infection in animals was widely scattered and its distribution differed regionally, with human infections considered to be scarce.[38] A data search of the National Health Laboratory Service (NHLS) laboratory information system was done and results of echinococcosis serology, microscopy and histopathology tests were analysed.[60] Results showed that there was an overall prevalence rate of 17% in submitted diagnostic samples.[60] In South Africa, cases have been reported in children with hydatids found in the liver, central nervous system and maxillary antrum.[14] In 2006, a symptomatic abdominal mass was found between the uterine fundus and liver in a pregnant woman who later died postpartum due to complications caused by the hydatid cyst.[49]

1.1.24 Cordylobia anthropophaga

Cordylobia anthropophaga is a dipterous fly found throughout Africa and is commonly known as the ‘tumbu fly’ or ‘mango fly’.[5,24] It is stout, 6-12 mm long, dull yellowish to light brown and has 2 dark grey dorsal longitudinal stripes on the thorax.[7] The posterior abdominal segments are a darker brown-black colour than the anterior ones. The wings are slightly brownish.[7]

There are several larval instars.[24] The mature larva is 11-15 mm long and has a fat, oval shape. The body is covered with numerous black spines.[7] Females deposit eggs on dry soil or sand in shaded places, contaminated with urine or excreta of man, animals (rodents, dogs and monkeys) as well as on nappies of babies, and on clothes which are not properly cleaned.[48] As one of the myiasis-producing flies, the emerging 1st instar larva requires a living host to complete larval development. Typical hosts are dogs, humans and rodents.[48] First instar larvae enter the skin and cause the formation of boil-like, furuncular swellings with a central opening.[48] Final instar larvae leave the skin and pupate in the soil.[48] The condition developing in infected humans and other hosts is known as cutaneous myiasis.[5, 24,48]

This fly is found in sub-Saharan Africa and spreads into KwaZulu-Natal and the northern provinces of South Africa.[24] In Nigeria, infestation described as furuncular myiasis has been reported from neonates, children, and adults.[45]

Chapter 2
MATERIALS AND METHODS

2.1 Parasite identification

As part of the diagnostic services offered by the Ampath Microbiology Laboratory in Pretoria, parasitic infections were diagnosed in samples received from Gauteng, North West and Mpumalanga provinces. The diagnostic techniques employed were conducted according to the laboratory's standard operating procedures and included:

a. *Direct (microscopic) examination of faeces*
   Emulsions were prepared in 0.9% saline and Lugol's iodine solution for the examination of specimens for the presence of protozoa, helminth eggs and larvae according to the methods of Chapin[10] and Crede[15].

b. *Modified acid-fast stain technique*
   The modified acid-fast stain technique as specified by WHO (1991)[61] was used for the demonstration of *Cryptosporidium* oocysts in air-dried smears prepared from faecal samples.

c. *Direct saline smear method*
   This technique was used to demonstrate the presence of *T. vaginalis* trophozoites in genital swabs as well as *E. granulosus* protoscoleces and hooklets in brain, liver, pancreas tissues and aspirates.[10]

d. *Urine sedimentation technique*
   Urine concentration by means of centrifugal sedimentation and subsequent microscopical examination of the sediment was used for the detection of *S. haematobium* ova, following the techniques of Bradley[8] and Hanges[30].

e. *Acanthamoeba* sp *culture method*
   Culture of contact lenses and contact lens fluids for *Acanthamoeba* spp. was done according to the method of Chapin & Murray[11].

f. *Identification of helminths, protozoa and arthropods*
   Helminths, helminth ova, protozoa and arthropods were identified on published morphological criteria.

2.2 Data collection

Diagnostic results were entered into the internal data system 'Meditech 5.5' of Ampath. 'Meditech 5.5' is a healthcare information system software from MEDITECH South Africa (Pty) Ltd. The software is designed to capture, store and display administrative and clinical data used within a health care system.[53] At the Ampath microbiology laboratory in Pretoria, this software is used to order, manage, document and report on clinical pathology tests. Specific searches were conducted for the electronically stored results of samples in which parasites were positively identified.

2.3 Categorization of data

Data of the positive results were exported from the Meditech system into a Microsoft Office Excel worksheet. Further sorting was then conducted based on date, specimen type, parasite identified, patient age, gender and geographic locality using the Excel tools.

2.4 Data analysis

The data obtained from the information system were summarized descriptively and compared with available published records.

Chapter 3
RESULTS

3.1 Data collection and categorization results of parasites identified in the Ampath laboratory from the Gauteng, North West and Mpumalanga provinces

The data collected showed that a total of 24 different species of parasites was identified in the laboratory over the two-year period. The overall number of positive samples received according to the data searches, were 863 in 2009 and 1061 in 2010, (Tables 3.1 & 3.2). In both years, the majority of cases originated from Gauteng Province followed by North West and Mpumalanga provinces (Tables 3.1 & 3.2). The most frequently identified parasites were *B. hominis* followed by *G. intestinalis* and *Cryptosporidium* spp. (Tables 3.1 & 3.2).

3.1.1 Entamoeba coli

A total number of 25 samples positive for *E. coli* were received in 2009 and 31 samples in 2010, with most of them collected in Gauteng Province (Tables 3.1 & 3.2). Most samples were collected from male patients (Tables 3.3 & 3.4). Mixed infections with *Blastocystis*, *G. intestinalis*, *E. nana*, *C. mesnili* and *Cryptosporidium* spp were found (Table 3.5).

3.1.2 Entamoeba histolytica

Two *E. histolytica* positive samples were received in 2009 compared to one in 2010. All three samples were collected from patients in Gauteng Province of which two were females and one was male (Tables 3.1, 3.2, 3.3 and 3.4). The ages of the female patients were 45 and 59 years and the male patient was 5 years old.

3.1.3 Endolimax nana

In 2009, ten *E. nana* positive samples were received. All samples were collected from patients in Gauteng Province (Table 3.1). In 2010, 32 positive samples were received, with the majority collected in Gauteng Province followed by North West and Mpumalanga provinces (Table 3.2). Samples in Gauteng Province were mainly collected from male patients, whereas in the other two provinces samples originated equally from male and female patients (Tables 3.3 & 3.4). Mixed infections with *B. hominis*, *G. intestinalis*, *C. mesnili*, *E. coli* and *T. hominis* were found (Table 3.5).

3.1.4 Acanthamoeba sp

Only one case was diagnosed in 2009 in a 28-year-old female from Gauteng province (Table 3.7).

3.1.5 Giardia intestinalis

Giardia intestinalis was the second most identified parasite in the two-year period, with 277 and 282 identified cases in 2009 and 2010 respectively (Tables 3.1 & 3.2). The number of positive samples received from Gauteng Province was far greater than from the other two provinces (Tables 3.1 & 3.2). Positive samples originated almost equally from male and female patients.

The *Giardia intestinalis* data were grouped according to the ages of patients and the months of the year when samples were received and constructed into a graph. The majority of positive samples received originated from patients between the ages of 0-5 years (Figs. 3.1 & 3.2). The number of cases diagnosed in 2009 was highest between the months of April and July, whereas in 2010 the number of cases peaked towards the end of the year between August and December (Figs. 3.1 & 3.2). Several cases of mixed infections with mainly *B. hominis* and *Cryptosporidium* spp. were identified (Table 3.5).

3.1.6 Trichomonas hominis

The few cases of *T. hominis* infections mostly originated from Gauteng Province (Tables 3.1 & 3.2). In 2009, positive samples originated almost equally from male and female patients (Table 3.3). In 2010, six out of the nine samples originated from female patients (Table 3.4). *Trichomonas hominis* was found in mixed infections with *B. hominis*, *C. mesnili* and *E. nana* (Table 3.5).

3.1.7 Trichomonas vaginalis

Only samples from female patients were received. Over the two-year period a total of 16 infections were diagnosed. Nine of the cases were patients in Gauteng province, three in North West and four in Mpumalanga provinces. The ages of patients varied between 23 to 59 years. (Table 3.6)

3.1.8 Chilomastix mesnili

Chilomastix mesnili was found in patients from Gauteng and North West provinces. Infections were more frequent in Gauteng Province in 2009 and 2010 (Tables 3.1 & 3.2). In 2009 a larger amount of positive samples originating from male patients was received (Table 3.3). In 2010, positive samples originated equally from male and female patients. Mixed infections occurred with *B. hominis*, *G. intestinalis*, *T. hominis* and *E. coli* (Table 3.5).

3.1.9 Blastocystis hominis

Blastocystis hominis was the main parasite reported in both of the study years. A total of 417 samples were found positive in 2009 compared to 559 samples in 2010. Most samples were received from Gauteng Province (Tables 3.1 & 3.2). Samples originated almost equally from male and female patients.

In 2009, *B. hominis* was found mostly in samples from elderly patients over 50 years. Positive samples were mostly received in April, May, July and October (Fig. 3.3). Similarly in 2010, most samples were received from older patients but mostly during the end of the year (Fig 3.4). Positive samples were also received from patients in the age groups of 0-5 years and 36-50 years (Figs. 3.3 & 3.4). In August 2010, there was a peak in the number of cases reported from babies and adult age groups (Fig. 3.4). Only a few positive samples were received from patients between the ages of 14 to 20 years (Figs. 3.3 & 3.4). Mixed infections, particularly with *G. intestinalis* and *Cryptosporidium* spp. were found (Table 3.5).

3.1.10 Cryptosporidium spp

Cryptosporidium spp were the third most common parasites diagnosed over the two-year period. In both years a total of 84 infections were diagnosed, with the majority of samples originating from Gauteng Province (Tables 3.1 & 3.2). Slightly more samples came from male patients (Tables 3.3 & 3.4). Positive samples from North West Province in 2009 originated equally from male and female patients. In Mpumalanga Province positive samples originated more from male patients (Table 3.3). In 2010, more females than males from the North West Province were diagnosed with infection, whereas in Mpumalanga, 2 males and 1 female patient were diagnosed with infection.

The *Cryptosporidium* spp data were grouped according to the ages of patients and the months of the year when samples were received and constructed into a graph. The majority of infections occurred in patients between the ages of 0-5 years (Figs. 3.5 & 3.6). In both years there was a greater frequency in the number of cases in the months of May to July and in almost all age groups there was an increase in the amount of infections in May (Figs 3.5 & 3.6).

3.1.11 Cystoisospora belli

Three isolated cases of infection with *C. belli* were reported over the two year period, two cases from North West Province in 2009 and one from Gauteng Province in 2010 (Tables 3.1 & 3.2). The patients from North West Province were a 44-year-old male and 39-year-old female. The patient from Gauteng Province was a 65-year-old female. (Table 3.7)

3.1.12 Sarcocystis spp

Sarcocystis infection was diagnosed only in a 35-year-old female patient from North West Province (Tables 3.1 & 3.7).

3.1.13 Enterobius vermicularis

Only five positive samples were received during the two-year period (Tables 3.1 & 3.2). In 2009 a positive sample from a 4-year-old female from Gauteng Province was received (Table 3.7). The other four cases were diagnosed in 2010, namely three patients from Gauteng Province and one from North West Province (Table 3.7). The patients from Gauteng Province were a 1-year-old male, an 8-year-old male and an 11-year-old female (Table 3.7). The patient from the North West Province was a 37-year-old male (Table 3.7).

3.1.14 Ascaris lumbricoides

Infection with *A. lumbricoides* was only diagnosed on three occasions (Table 3.1 & 3.2). In 2009 a 57-year-old patient from Mpumalanga was found positive (Table 3.7). The cases in 2010 were two female patients from Gauteng Province of 1 and 19 years (Table 3.7).

3.1.15 Trichuris trichiura

Only one case of infection with *T. trichiura* was found. The patient was a 55-year-old male from Mpumalanga Province (Table 3.7).

3.1.16 Ancylostomatids (hookworm)

Hookworm infections were diagnosed on two occasions, in a 50-year-old male patient from Mpumalanga Province in 2009 and in a 27-year-old male from Gauteng Province in 2010.

3.1.17 Taenia saginata

In 2009, five cases of infection with the beef tapeworm were diagnosed. Four of the infections occurred in patients from Gauteng Province and one in a patient from Mpumalanga Province (Table 3.1). In 2010, twelve infections were diagnosed, 11 in patients from Gauteng Province and 1 patient from North West Province (Table 3.2). The majority of *T. saginata* infections were found in adult males (Fig. 3.7).

3.1.18 Taenia spp

Diagnosis of infection with *Taenia* spp was reported when tapeworm segments could not be properly identified to species level or when only taeniid eggs were observed on wet mounts. The possibility of infection with *T. solium* could thus not be ruled out. All three of the reported cases falling in this category were from patients in Gauteng Province.

3.1.19 Dipylidium caninum

Dipylidium caninum segments were identified in the faeces of a 5-month-old male originating from Gauteng Province in 2010 (Table 3.7).

3.1.20 Inermicapsifer madagascariensis

Infection with this tapeworm was reported in a 2-year-old male from Mpumalanga Province in 2010 (Table 3.7).

3.1.21 Bertiella studeri

Infection was diagnosed twice in 2010 in a 6-year-old female and a 27-year-old male from Gauteng Province (Table 3.7).

3.1.22 Schistosoma haematobium

Schistosoma haematobium was diagnosed in 8 samples in 2009 which originated from four patients in Gauteng Province, three patients in Mpumalanga Province and one patient in North West Province (Table 3.1). In 2010, the total number of infections diagnosed was 13, the amount of infections in each province was almost evenly distributed (Table3.2). Infections were relatively scattered throughout both years (Fig. 3.8).

3.1.23 Echinococcus granulosus

One case of cystic echinococcosis was diagnosed during the two-year period. *Echinococcus granulosus* hooklets were observed in pus drained from a liver abscess of a 61-year-old male from North West Province (Table 3.7).

3.1.24 Cordylobia anthropophaga

Three infections with *C. anthropophaga* larvae were diagnosed over the two-year period (Table3.1 & 3.2). In 2009 larvae were identified from a 37-year-old male residing in Gauteng Province (Table 3.7). In 2010 another two cases were identified in two female patients from Gauteng Province, a 37-year-old and a 3-year-old.

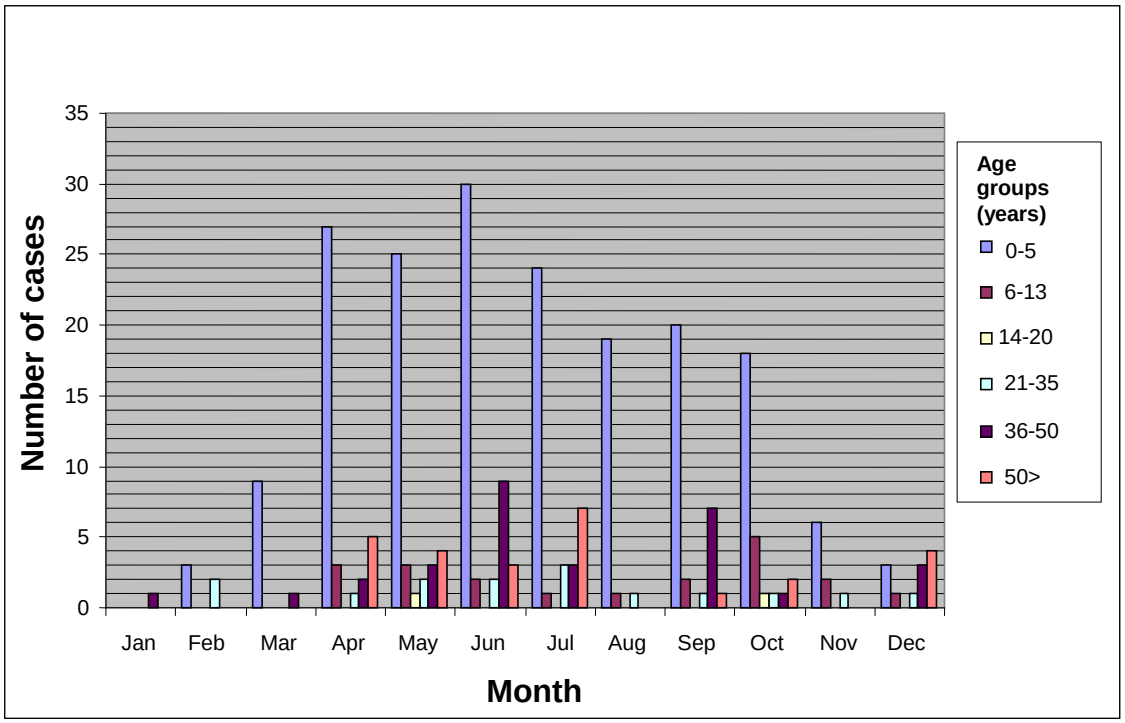

Figure 3.1: Number of *Giardia intestinalis* infections reported monthly in 2009 grouped according to patient age groups

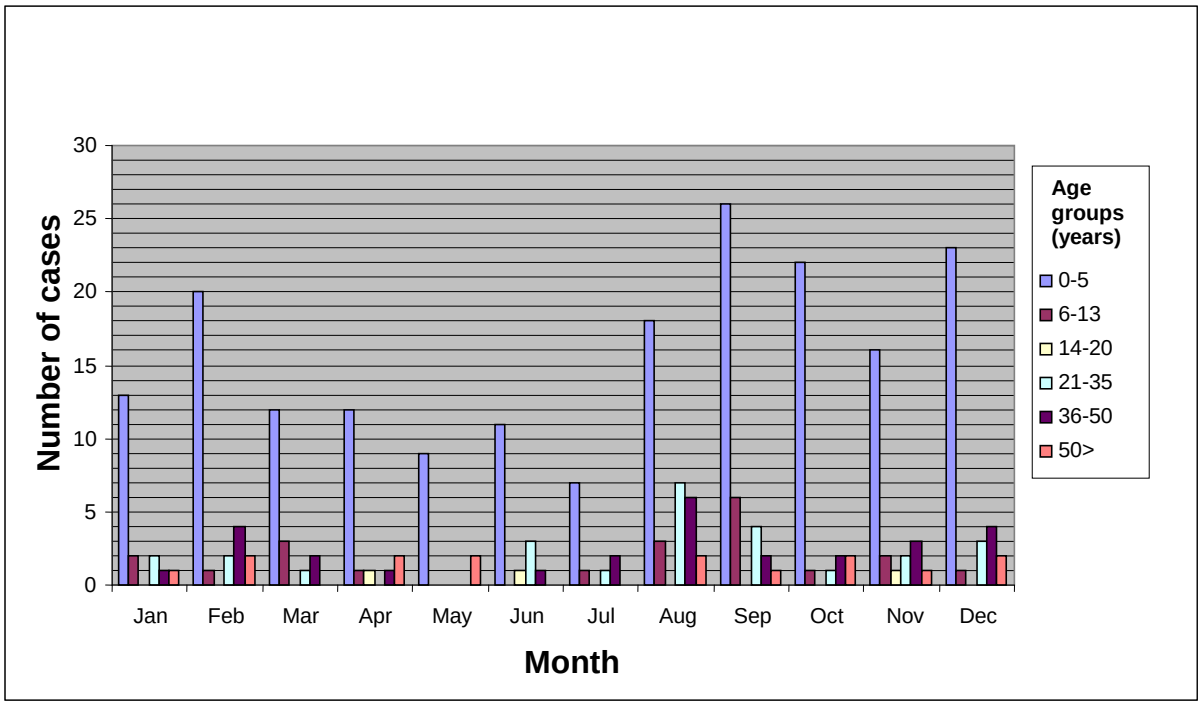

Figure 3.2: Number of *Giardia intestinalis* infections reported monthly in 2010 grouped according to patient age groups

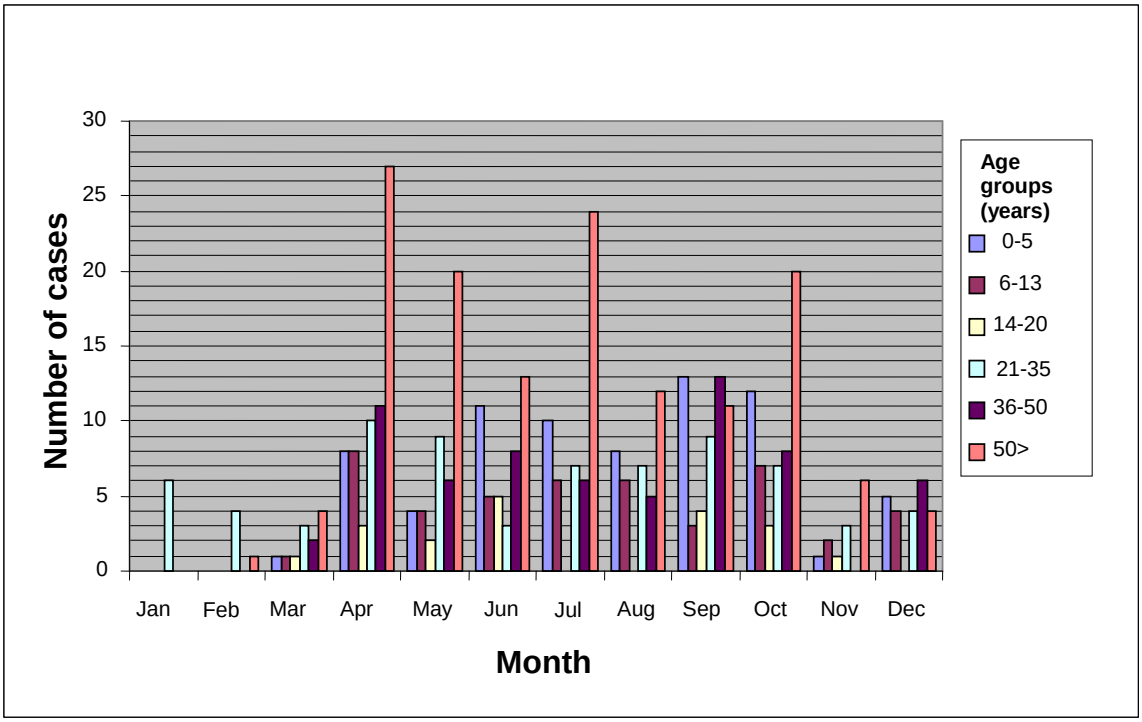

Figure 3.3: Number of *Blastocystis hominis* infections reported monthly in 2009 grouped according to patient age groups

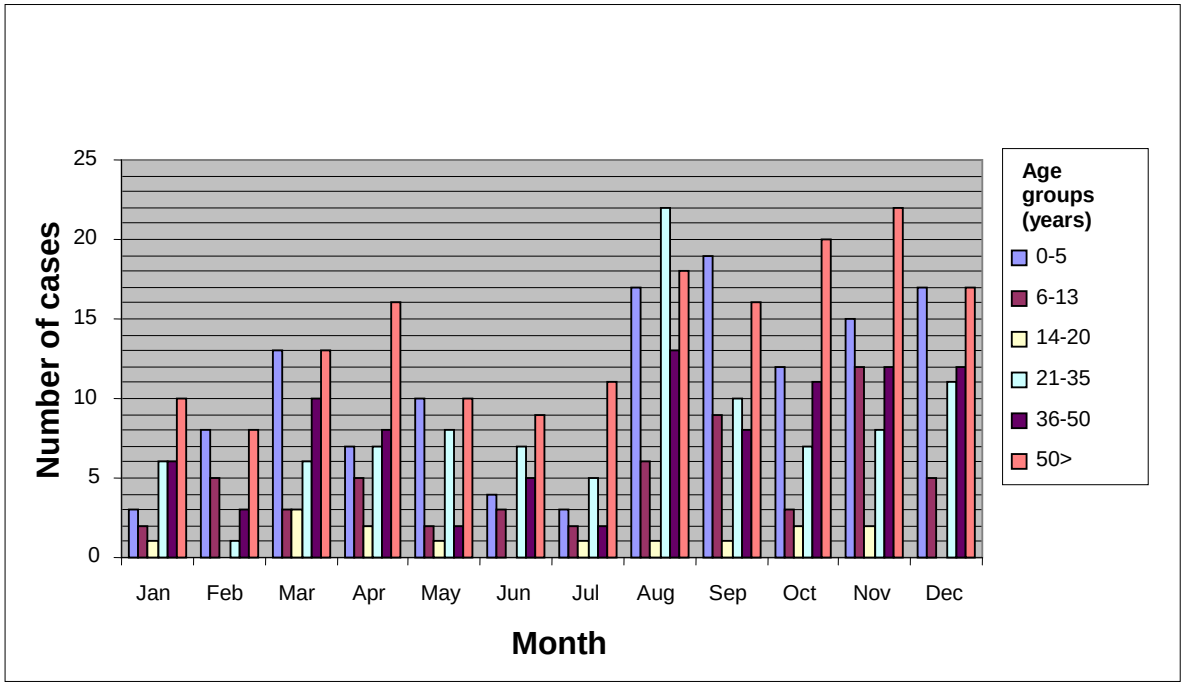

Figure 3.4: Number of *Blastocystis hominis* infections reported monthly in 2010 grouped according to patient age groups

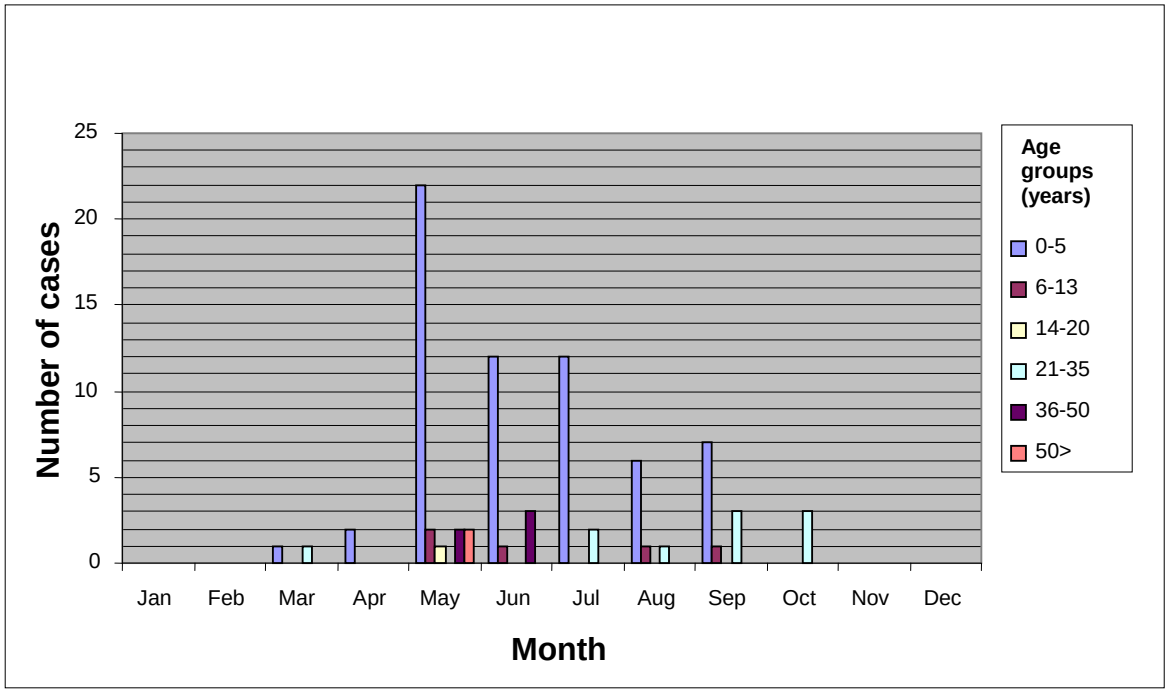

Figure 3.5: Number of *Cryptosporidium* spp infections reported monthly in 2009 grouped according to patient age groups

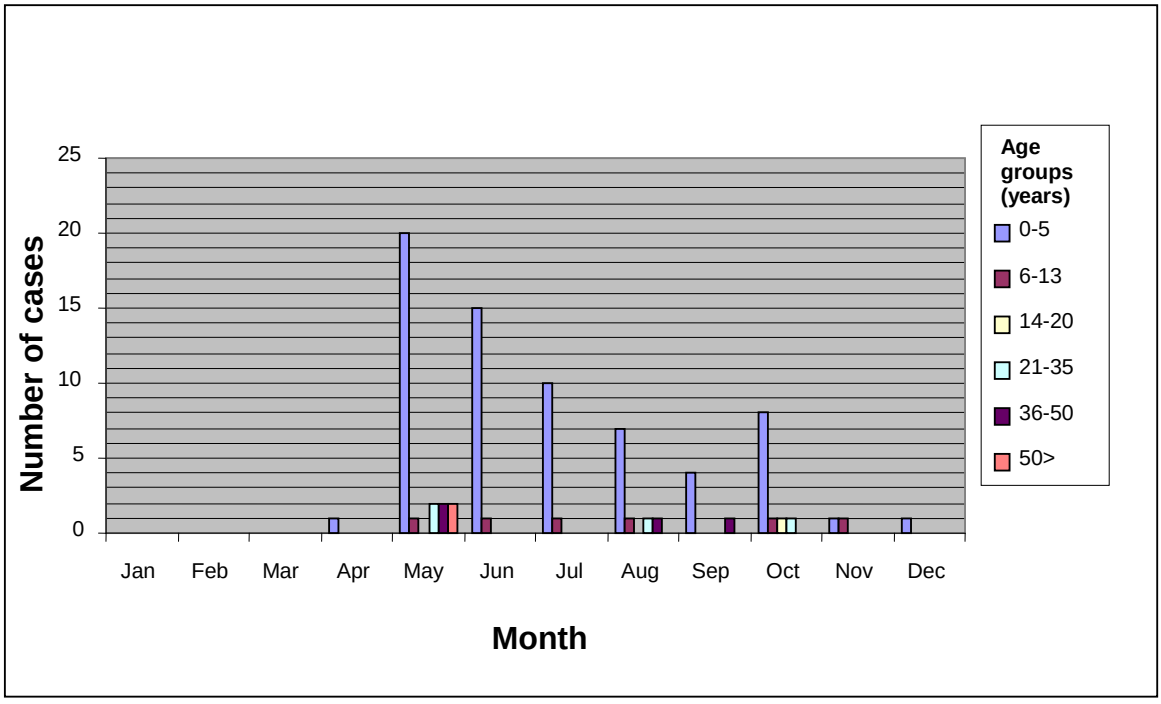

Figure 3.6: Number of *Cryptosporidium* spp infections reported monthly in 2010 grouped according to patient age groups

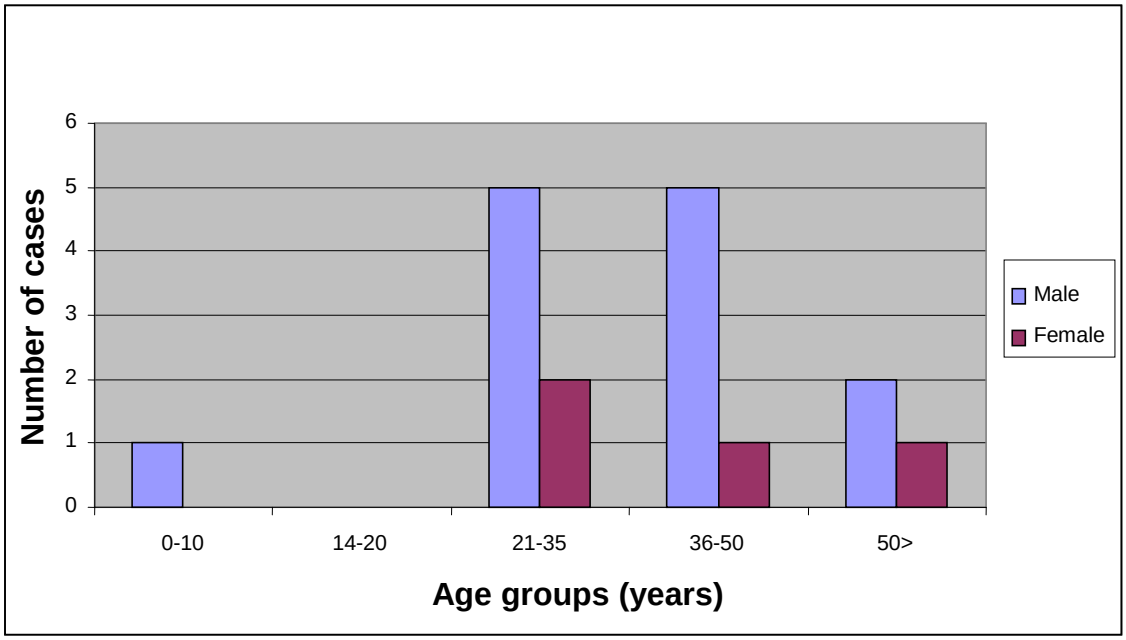

Figure 3.7: Number of *Taenia saginata* infections reported in 2009 and 2010 grouped according to patient age groups and gender

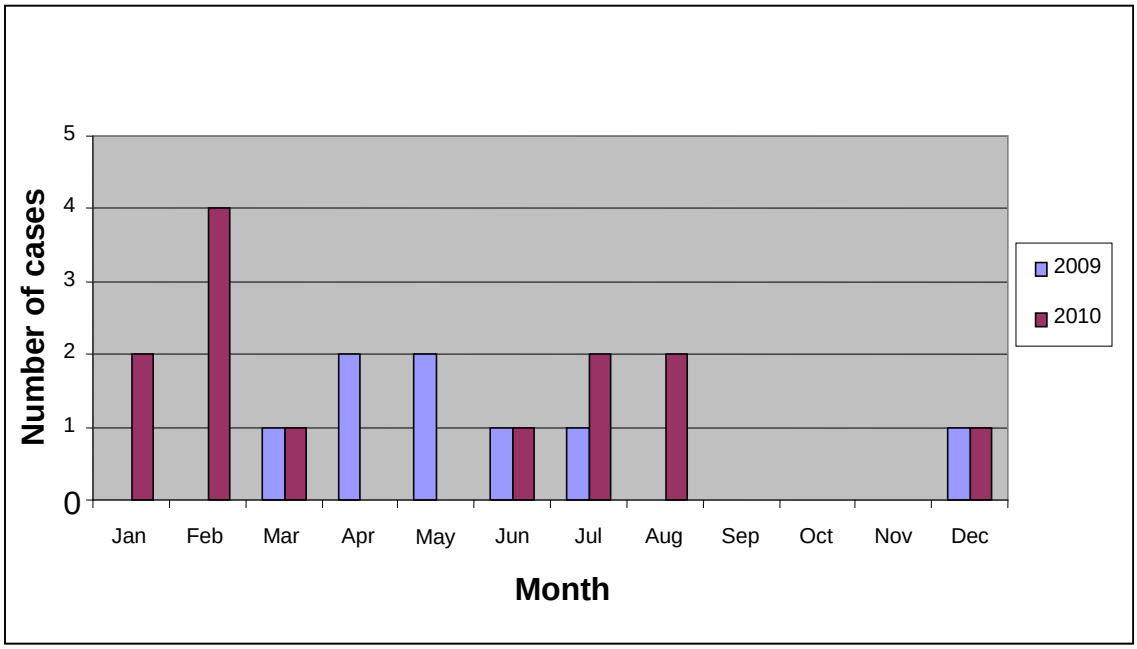

Figure 3.8: Number of *Schistosoma haematobium* infections reported in 2009 and 2010 grouped according to year and month

Table 3.1: Overall number of parasites identified in 2009 from patients in Gauteng, North West and Mpumalanga provinces

Parasite	Province			TOTAL
	Gauteng	North West	Mpumalanga	
Entamoeba coli	20	4	1	**25**
Entamoeba histolytica	2	0	0	**2**
Endolimax nana	10	0	0	**10**
Giardia intestinalis	240	21	16	**277**
Trichomonas hominis	7	1	1	**9**
Chilomastix mesnili	5	3	0	**8**
Blastocystis hominis	367	25	25	**417**
***Cryptosporidium* spp**	71	4	9	**84**
Cystoisospora belli	0	2	0	**2**
***Sarcocystis* spp**	0	1	0	**1**
Trichomonas vaginalis	5	2	2	**9**
Enterobius vermicularis	1	0	0	**1**
Ascaris lumbricoides	0	0	1	**1**
Trichuris trichiura	0	0	0	**0**
Ancylostomatids (hookworm)	0	0	1	**1**
***Taenia* spp**	2	0	0	**2**
Taenia saginata	4	0	1	**5**
Dipylidium caninum	0	0	0	**0**
Inermicapsifer madagascariensis	0	0	0	**0**
Bertiella studeri	0	0	0	**0**
Echinococcus granulosus	0	0	0	**0**
Schistosoma haematobium	4	1	3	**8**
***Acanthamoeba* sp**	0	0	0	**0**
Cordylobia anthropophaga	1	0	0	**1**
TOTAL	**739**	**64**	**60**	**863**

Table 3.2 Overall number of parasites identified in 2010 from patients in Gauteng, North West and Mpumalanga provinces

Parasite	Province			TOTAL
	Gauteng	North West	Mpumalanga	
Entamoeba coli	24	4	3	**31**
Entamoeba histolytica	1	0	0	**1**
Endolimax nana	25	5	2	**32**
Giardia intestinalis	248	18	16	**282**
Trichomonas hominis	9	0	0	**9**
Chilomastix mesnili	12	1	0	**13**
Blastocystis hominis	479	41	39	**559**
***Cryptosporidium* spp**	76	5	3	**84**
Cystoisospora belli	1	0	0	**1**
***Sarcocystis* spp**	0	0	0	**0**
Trichomonas vaginalis	4	1	2	**7**
Enterobius vermicularis	3	1	0	**4**
Ascaris lumbricoides	2	0	0	**2**
Trichuris trichiura	0	0	1	**1**
Ancylostomatids (hookworm)	1	0	0	**1**
***Taenia* spp**	1	0	0	**1**
Taenia saginata	11	1	0	**12**
Dipylidium caninum	1	0	0	**1**
Inermicapsifer madagascariensis	0	0	1	**1**
Bertiella studeri	2	0	0	**2**
Echinococcus granulosus	0	1	0	**1**
Schistosoma haematobium	4	4	5	**13**
***Acanthamoeba* sp**	1	0	0	**1**
Cordylobia anthropophaga	2	0	0	**2**
TOTAL	**907**	**82**	**72**	**1061**

Table 3.3: Protozoa identified in faecal samples of patients from Gauteng, North West and Mpumalanga provinces in 2009

Parasite	Province and Gender [a]					
	Gauteng		North West		Mpumalanga	
	M	F	M	F	M	F
Entamoeba coli	11	9	3	1	0	1
Entamoeba histolytica	0	2	0	0	0	0
Endolimax nana	7	3	0	0	0	0
Giardia intestinalis	121	119	10	11	7	9
Trichomonas hominis	3	4	1	0	0	1
Chilomastix mesnili	4	1	2	1	0	0
Blastocystis hominis	189	178	13	12	10	15
Cryptosporidium spp	40	31	2	2	6	3

[a] **Male (M) and Female (F)**

Table 3.4: Protozoa identified in faecal samples of patients from Gauteng, North West and Mpumalanga provinces in 2010

Parasite	Province and Gender [a]					
	Gauteng		North West		Mpumalanga	
	M	F	M	F	M	F
Entamoeba coli	13	11	3	1	3	0
Entamoeba histolytica	1	0	0	0	0	0
Endolimax nana	15	10	2	3	1	1
Giardia intestinalis	118	130	12	6	9	7
Trichomonas hominis	3	6	0	0	0	0
Chilomastix mesnili	6	6	0	1	0	0
Blastocystis hominis	244	235	19	22	19	20
Cryptosporidium spp	39	37	1	4	2	1

[a] **Male (M) and Female (F)**

Table 3.5: Mixed protozoal infections diagnosed in faecal samples of patients from Gauteng, North West and Mpumalanga provinces in 2009 and 2010

Parasite combination	Number of cases
Blastocystis hominis & Chilomastix mesnili	3
Blastocystis hominis & Entamoeba coli	7
Blastocystis hominis & Cryptosporidium spp	11
Blastocystis hominis & Endolimax nana	4
Blastocystis hominis & Giardia intestinalis	22
Blastocystis hominis & Trichomonas hominis	1
Chilomastix mesnili & Giardia intestinalis	2
Chilomastix mesnili & Trichomonas hominis	1
Cryptosporidium spp *& Entamoeba coli*	1
Cryptosporidium spp *& Giardia intestinalis*	8
Endolimax nana & Entamoeba coli	3
Endolimax nana & Giardia intestinalis	2
Endolimax nana & Trichomonas hominis	1
Entamoeba coli & Giardia intestinalis	2
Blastocystis hominis, Chilomastix mesnili & Endolimax nana	1
Blastocystis hominis, Cryptosporidium spp *& Giardia intestinalis*	1
Blastocystis hominis, Chilomastix mesnili, Entamoeba coli & Giardia intestinalis	1
TOTAL	**71**

Table 3.6: *Trichomonas vaginalis* infections diagnosed in patients from Gauteng, North West and Mpumalanga provinces in 2009 and 2010

Year	Age [a]	Province [b]
2009	23 y	GP
	28 y	GP
	29 y	GP
	40 y	GP
	45 y	GP
	39 y	NW
	47 y	NW
	33 y	MP
	43 y	MP
2010	47 y	GP
	49 y	GP
	50 y	GP
	59 y	GP
	30 y	NW
	33 y	MP
	37 y	MP

[a] Year (y)
[b] Gauteng Province (GP), North West Province (NW), Mpumalanga Province (MP)

Table 3.7: Parasites isolated only occasionally in 2009 and 2010, grouped according to patient age and gender

Year	Parasite	Age [a]	Gender [b]	Province [c]
2009	***Ascaris lumbricoides***	57 y	F	MP
	Enterobius vermicularis	4 y	F	GP
	Hookworm	50 y	M	MP
	Cystoisospora belli	44 y	M	NW
		39 y	F	NW
	***Sarcocystis* spp**	35 y	F	NW
	Cordylobia anthropophaga	37 y	M	GP
2010	***Acanthamoeba* sp**	28 y	F	GP
	Ascaris lumbricoides	1 y	F	GP
		19y	F	GP
	Bertiella studeri	6 y	F	GP
		27 y	M	GP
	Dipylidium caninum	5 m	M	GP
	Echinococcus granulosus	61 y	M	NW
	Enterobius vermicularis	1 y	M	GP
		11 y	F	GP
		8 y	M	GP
		37 y	M	NW
	Hookworm	27 y	M	GP
	Inermicapsifer madagascariensis	2 y	M	MP
	Cystoisospora belli	65 y	F	GP
	Trichuris trichiura	55 y	M	MP
	Cordylobia anthropophaga	39 y	F	GP
		3 y	F	GP

[a] **Year (y) and Months (m)**
[b] **Male (M) and Female (F)**
[c] **Gauteng Province (GP), North West Province (NW), Mpumalanga Province (MP)**

Chapter 4
DISCUSSION

4.1 Origin of patients

Very few recent studies have been carried out in South Africa on the occurrence or prevalence of parasites in humans. The most recent surveys only focus on specific parasites in set target regions. Most of the literature found, were surveys conducted in provinces other than those considered in this study. The results indicate that most of the parasitic infections diagnosed were from patients residing in Gauteng Province. A reason for this could be that the Ampath Microbiology laboratory in Pretoria mostly receives samples from patients in this province. Another factor could be that the human population of Gauteng Province exceeds those of North West and Mpumalanga provinces by far. Ampath is a private pathology laboratory, thus all samples are submitted by private physicians or hospitals. Also to consider is that people in Gauteng have better access to doctors and hospitals. However, the fact that most parasites were found in samples from people residing in Gauteng Province does not necessarily mean that infections were acquired there. Infections could have been acquired while travelling or working in other provinces of the country, unfortunately, information regarding patient history is very seldom available.

4.2 Protozoal infections

Blastocystis hominis was found in abundance. The majority of faecal specimens received in the laboratory originated from patients with gastrointestinal upsets. Those cases in which *B. hominis* was isolated only, strongly suggest the pathogenic potential of the parasite. Since the majority of samples received were collected from over-50-year-old patients, age appears to be a risk factor for infections.

Giardia intestinalis and *Cryptosporidium spp* infections were mostly identified in samples received from infants and children between the ages of 0-5 years which correlates with data in the literature.[51,58] Regarding *Cryptosporidium*, surveys are required in South Africa to determine the species and genotype involved in human infections, which would give an indication on the epidemiology and significance of cryptosporidiosis in humans.

Several cases of infections with protozoa generally regarded as non-pathogenic were diagnosed. Parasites identified included *Entamoeba coli*, *Endolimax nana*, *Chilomastix mesnili* and *Trichomonas hominis*. Because of the cosmopolitan distributions of these organisms, this finding was to be expected. *Entamoeba coli* and *E. nana* were more frequently diagnosed than *C. mesnili* and *T. hominis*. A survey by Kvalsvig (1988)[36] showed similar results, with *E. coli* and *E. nana* found to be more prevalent than *C. mesnili*.

Only 16 cases of *T. vaginalis* infection were diagnosed in the 2-year period, conclusions cannot be made from this study whether the low amount of cases might reflect the effectiveness of STD education programmes in South Africa.

A single case of infection with *Acanthamoeba* sp was diagnosed in the 2-year period. The literature indicates that infection with *Acanthamoeba* sp is infrequent.[5,13]

Infections with *Cystoisospora belli* (3 cases) and *Sarcocystis* spp (1 case) were rarely diagnosed, which is in accordance to the literature confirming their uncommonness in developed communites.[13]

4.3 Mixed protozoal infections

The majority of mixed infections were with *B. hominis*, *G. intestinalis*, *Cryptosporidium* spp and *E. coli.* According to the literature, contaminated water seems to be the major source of infection for these parasites.[5,7,56] Surveys should be conducted in the respective provinces to evaluate the status of municipal water quality.

4.4 Helminth infections

Ascaris lumbricoides, hookworm species and *T. trichiura* were identified only occasionally. In other surveys, prevalences of these parasites in South Africa were high.[1,2] However, in those surveys, emphasis was predominantly placed on economically deprived communities. It is clear that infections with the above-mentioned parasites can be directly linked to the socioeconomic status of a community.

Surveys conducted on the status of schistosomosis in South Africa in the past have revealed high prevalences of *S. haematobium* infections in the rural areas of KwaZulu-Natal, Limpopo and Mpumalanga provinces.[43,50,62] At the Ampath laboratory a total of 21 cases of *S. haematobium* infection was diagnosed. It was not possible to determine where infections were most likely contracted. The results motivate to conduct surveys to determine the current prevalence status of schistosomosis in particularly Gauteng and North West provinces as well as to determine the distribution of the intermediate hosts *Bulinus africanus* and *Bulinus globosus*.

Enterobius vermicularis was almost exclusively identified in samples received from children, which is in accordance with the literature[24].

Regarding the *Taenia* spp, there were no confirmed infections with the strobilar stage of *T. solium*. However, on three occasions taeniid segments could not be properly identified or only taeniid eggs were found in faecal samples. As the public has been sensitized extensively

over the years regarding the microbial hazards of consuming undercooked pork, the result was to be expected. Infection with *Taenia saginata* was diagnosed in 17 cases with most samples originating from Gauteng Province. Human strobilar infection is contracted by consuming raw or undercooked beef infested with cysticerci. Since bovine cysticercosis originates from ingesting eggs shed in human faeces, infection is a strong indicator of inadequate sanitation and sewage management.[5,13]

Only a single case of cystic echinococcosis (hydatid disease) was diagnosed in the 2-year period. The finding and identification of *E. granulosus* larval hooklets in a specimen collected from the liver of a patient is in accordance with the literature, since 65-70% of hydatids develop in the liver of humans and other intermediate hosts.[52]

Although commonly encountered in dogs and cats, human infections with *D. caninum* appear to be very rare and are mostly reported in children.[5,24,52] A single infection diagnosed in a 5-month old baby supports these findings.

Similarly, human infections with *Bertiella studeri* and *Inermicapsifer madagascariensis* were diagnosed only on 3 occasions. Both are anoplocephalid tapeworms, which commonly utilize oribatid mites as intermediate hosts. The oribatids *Scheloribates laevigatus* and *Galumna* spp have been identified as intermediate hosts of *B. studeri*. However, the life cycle of the *I. madagascariensis* is unknown. Humans acting as accidental definitive hosts, can only become infected by ingesting oribatid mites or, in the case of *I. madagascariensis*, a so-far unknown arthropod intermediate host carrying cysticercoids.[5,18] A source of infection could be mite-infested vegetables grown in areas populated by primates (*B. studeri*) or rodents, lagomorphs and hyracoids (*I. madagascariensis*).

4.5 Arthropod infestations

Cutaneous larva migrans caused by larvae of *Cordylobia anthropophaga* is commonly encountered in KwaZulu-Natal and the northern provinces of South Africa[24]. In the two-year period only three cases were diagnosed from patients in Gauteng Province.

Chapter 5
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