CHAPTER ONE

Introduction and Literature Review

1.1 Introduction:

The field of parasitology is often associated with tropical areas. However, many parasitic organisms that infect humans are world wide in distribution. Another factor to be considered is the increase in numbers of compromised patients. Those persons are very much at risk for certain parasitic problems (WHO , 1996).

Like in other developing countries, intestinal parasitic infections are one of the major health problems in Sudan. The prevalence rates of intestinal parasite infections are important in these country and produce basic data for the control of parasitic infections in the future (WHO , 1996).

Gastro-intestinal helminthes infections are transmitted mainly through drinking water or food intake. Organisms that are related to the two major parasitic groups, protozoa and helminthes, cause the most important and persistent health problems with a prevalence of particular significance among African children (WHO 1996)

Over 342 species of helminthes have been found in association with mammals, of which 197 would be considered as primary inhabitants of their gastro-intestinal tract (Crompton , 1999).

1-2 Hymenolepis nana:

1-2-1 Classification:

Kingdom : Animalia

Phylum : Platyhelminthes

Class : Cestoda

Order : Cyclophyllidea

Family : Hymenolepididae

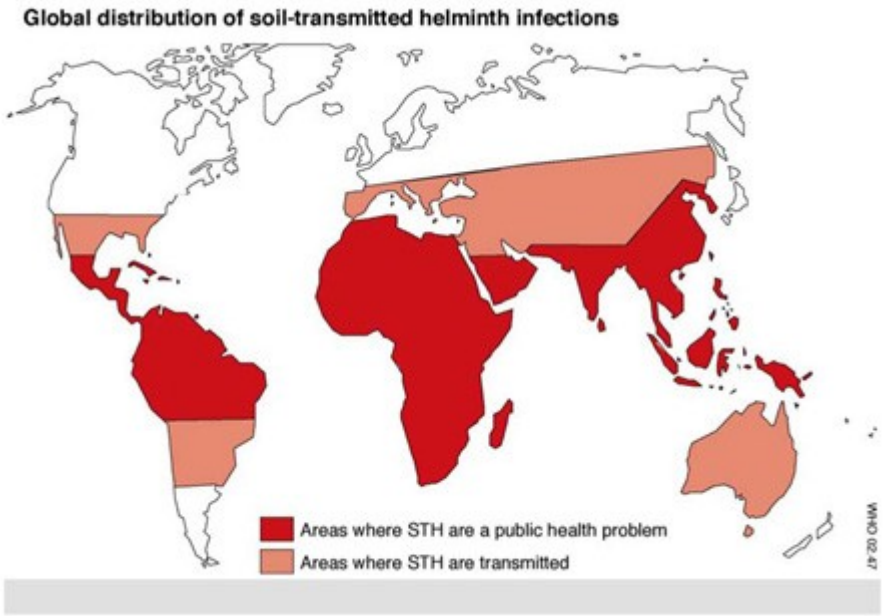
Genus : Hymenolepis

Species : Hymenolepis nana

Roberts and Janovy Jr (2000)

1-2-2 Distribution:

Hymenolepis nana can be found throughout the world, but is usually most common in temperate zones. It is the most common cestodes of human and is found wherever there are human inhabitants (Roberts and Janovy Jr 2000).



1-2-3 Morphology:

The adults of the dwarf tapeworm are 25 to 40 mm in length and 1 mm in width. This tapeworm is transparent, and has a long slender neck with segments wider than they are long. The genital pores are unilateral, or on the side of the segment. Each segment contains a single proglottid, which contains a single set of reproductive organs. On the scolex, a retractable rostellum with 20 to 30 hooks can be found. The scolex also has four suckers.

The cysticercoid has a tail, which is made of longitudinal fibers and is spade shape with the rest of worm still inside the cyst (Roberts and Janovy Jr 2000). The egg of *Hymenolepis nana* are round or slightly oval at about 40 – 60 micrometers X 30- 50 micrometers with 4 – 8 polar filaments spread out between the inner and the outer membranes (Ghaffar 2001). Unlike other taenia egg , the egg of *Hymenolepis nana* do not has a striated appearance (Roberts and Janovy Jr.2000) .

1-2-4 Life cycle:

A gravid proglottid contains fertilized eggs, which are expelled with the faeces (Cameron, 1956). However , most of the time, the eggs settles in the microvilli of the small intestine, hatch and the larva can develop to sexual maturity without ever leaving the host (Olsen,1974). An intermediate host is optional; *Hymenolepis nana* can go through its life-cycle with only one host . Then the eggs are ingested by the definitive host and hatch in the duodenum, releasing oncosphere which lie in the lymph channels of the of the villi, then the oncosphere develop into cysticercoid, which has a tail and well formed scolex , and it attaches to the small intestine and matures into an adult. The gravid proglottids then release and pass out through faeces along with eggs, or eggs can hatch and infect original host and start cycle over. Development of

Hymenolepis nana may also take place in the larval stage of the rat flea if it swallows the egg. A cysticercoid develop in the body of larva ,which persists through the metamorphosis into the adult flea , the rats become infected by swallowing the flea , also various insects , including fleas, beetles, cockroaches act as intermediate host . The egg hatches in midgut of the insect, and the onchosphere migrates into the haemocoele and there becomes acysticercoid , when the infected insect swallowed by suitable host , cysticercoid attach itself to the wall of the intestine and begins to bud off segments. (Davey .T. H, 1966).

1-2-5 Reproduction:

Hymenolepis nana, like all tapeworms, contain both male and female reproductive structures in each proglottid.This means that the dwarf tapeworm like other tapeworms is hermaphroditic. Each segment contains 3 testes and a single ovary. When a proglottid becomes old and unable to absorb any more nutrition, it is released and is passed through the host's digestive tract . This gravid proglottid contains the fertilized eggs, which are sometimes expelled with the feces . However, most of the time, the egg may also settle in the microvilli of the small intestine, hatch, and the larvae can develop to sexual maturity without ever leaving the host . (Cameron, 1956; Olsen, 1974; Roberts and Janovy Jr., 2000)

1-2-6 Biology:

Hymenolepis nana does not have a digestive system and each body segment has its own reproductive structures . Usually after it is ingested as an egg, it will lodge itself in the intestinal wall and then in five to six days, the cysticercoid emerges into the lumen of the small intestine, sheds it tail and becomes a mature tapeworm**.Cestodes** in general have sensory organs in the scolex, which are attached to

longitudinal nerves extending down the body. The nerves are attached to organs and the cestodes can detect tactile stimulation. **((Roberts and Janovy Jr., 2000.Brusca and Brusca, 2003)**

The dwarf tapeworm like all other tapeworms lacks a digestive system and feeds by absorption on nutrients in the intestinal lumen. They have non-specific carbohydrate requirements and it seems like they will absorb whatever is being passed through the intestine at that time . When it becomes an adult, it will attach to the intestinal walls with its suckers and toothed rostellum and have its segments reaching out into the intestinal space to absorb food . **(Cameron, 1956; Roberts and Janovy Jr., 2000)**

Hymenolepis nana is the most common cestode parasite of humans in the world . It lodges itself in the intestines and absorbs nutrients from the intestinal lumen . In human adults, the tapeworm is more of a nuisance than a health problem, but in small children, many *H. nana* can be dangerous. Usually it is the larva of this tapeworm that causes the most problem in children . The larva will burrow into the walls of the intestine, if there are enough tapeworms in the child, severe damage can be inflicted. This is done by absorbing all the nutrition from the food the child eats . Usually a single tapeworm will not cause any danger, but in small children, many tapeworms can become a problem . *Hymenolepis nana* usually will not cause deaths unless in extreme circumstances and usually in young children or in people who have weakened immune systems. In some parts of the world, individuals that are heavily infected are a result of internal autoinfection. (Cameron, 1956; Lapage, 1951; Olsen, 1974; Roberts and Janovy Jr., 2000)

1-2-7 Symptoms:

It is not clear that hymenolepiasis necessarily have any symptoms. The symptoms of hymenolepiasis are traditionally described as abdominal pain, loss of appetite (anorexia), itching around the anus, irritability and diarrhea. However, in one study of 25 patients conducted in Peru, successful treatment of the infection made no significant difference to symptoms. Some authorities report that heavily infected cases are more likely to be symptomatic. (Roberts and Janovy Jr 2000)

1-2-8 Signs and tests:

Examination of the stool for eggs and parasites confirms the diagnosis. The eggs and proglottids of *H. nana* are smaller than *H. diminuta*. Proglottids of both are relatively wide and have three testes. Identifying the parasites to the species level is often unnecessary from a medical perspective, as the treatment is the same for both. (Roberts and Janovy Jr 2000).

The laboratory diagnosis 0f hymenolepis nana infection is by finding the eggs in faeces . they can usually be found by direct examination of faeces. If required the eggs can be concentrated by formal ether concentration technique or saturated sodium chloride floatation technique.

Routinely faecal specimens are examined by direct wet technique. This involves, reporting the appearance of the specimen and identifying any parasitic worms or tapeworm segments, examining the specimen microscopically for motile parasites, helminth eggs, the direct examination of faeces is usually adequate to detect significant helminth infection. concentration techniques may be required to detect the eggs of *Hymenolepisnana,* to check whether treatment has been successful,also to

quantify intestinal parasites.(Monica , 1998) .

1-2-9 Treatment:

Praziquantel as a single dose (25 mg/kg) is the current treatment of choice for hymenolepiasis and has an efficacy of 96%. Single dose albendazole (400 mg) is also very efficacious (>95%). Niclosamide has also been used. (Roberts and Janovy Jr 2000)

A three-day course of nitazoxanide is 75–93% efficacious. The dose is 1g daily for adults and children over 12; 400mg daily for children aged 4 to 11 years; and 200mg daily for children aged 3 years or younger. (Roberts and Janovy Jr 2000)

1-2-10 Prevention:

Good hygiene, public health and sanitation programs, and elimination of rats help prevent the spread of hymenolepiasis.practising personal hygiene,espically the washing of hands before eating , providing laterines and encouraging their use espically by children , avoiding eating uncooked food which may be contaminated with human faeces. (Roberts and Janovy Jr 2000)

1.3International and national situation:

Mamo et al (1989) examined 2309 stool specimens from about 5% of the residents of Alaki in Ethiopia. The prevalence of various parasites was as follows: Taenia species (10.2%) and *Hymenolepis nana* (10.6%).

In Niger, Develoux et al (1986) examined 1748 children aged from 7 – 13 years from 6 districts. They found 1005 (57.5%) were infected. Among these, 37% were infected with protozoan parasites and 35.3% were infected with helminthes. The principal helminthes observed in their study were *Hymenolopis nana* 14.2%.

In Namibia, Evan and Joubert (1989) examined 4171 faecal sample of inpatient and out patient in the state hospital in Rundu Kavango ceritary Namibia over 29 month period and showed that common infection was by *H.nana* (1%) and *Taena spp* (0.9%). *H.nana* was most common in preschool children.

In Niger , Develoux. Et al (1986) examined 1748 children aged from 7-13 years from 6 districts they found 35.3% were infected with helminthes parasites . The principal helminthes observed in their study was *H.nana* (6.8 to 14.2).

In Egypt Elshazly et al (2006) , the study area included Mansoura city as an urban area and Gogar village as rural area , one thousand individual were randomly selected from each area , found Incidence of *H.nana in urban* 2.2 % and in rural 3.3 % . The infection rate of *H.nana* was relatively high in the rural than in the urban area .

In Sudan , Abu gusseisa (1995) examined 1500 stool samples from displaced people in Khartoum . He found that 374 (25%) egg , larvae , cyst of parasites .The genera of parasites identified in above survey included: *H.nana , Taena.spp*

Also in Sudan, Marnell et al (1992) conduct a parasitological survey of intestinal helminthes of refugees in Juba . They examined 241 fecal samples which revealed that 66% intestinal helminthes . The most common being *H.nana* (11%).

In a study survey for intestinal protozoa and helminthes infections carried out in a university community in Khartoum, Ibrahim (1999) reported an over all prevalence rate of 7.7% including *Hymenolepis nana* (13.3%), *Taenia saginata* (1.2%).

A parasitological survey among refugees residing in Juba area revealed that 66% of the population harbuored intestinal parasites (Marnell et al.1992).The parasites prevalence and intensity were analyzed in relation to sex, age , religion and occupation. Gender prevalence profile showed differences, females showed higher prevalence of *Hymenolepis nana* infection than males. In the some study, prevalence of parasites was also found to be higher among Christians than Muslims. None of the parasites showed any significant difference with regard to occupation although *Hymenolepis nana* was more prevalent among white collars and medical workers.

The health status of 239 refugees in the USA was evaluated. Of those, 36.7% were found positive for intestinal parasites. Anaemia was reported in 14% and esinophilia in 11%.

In a study of helminth parasite prevalence in Elgamair residential area (Omdurman town in Khartoum state) conducted by Ibrahim (1999), 1268 stool samples were collected from school children and patients reporting to health centers. 37.2% of the population was found to harbour intestinal helminthes. The most common parasites were *Hymenolepis nana* (11.6%), *Taenia saginata* (3.4%).

The study on gastrointestinal parasite infection conducted by Priscilla (2003) in school children of Juba revealed that nearly half of the

children were infected. The parasites involved were *Hymenolepis nana* (14%), Some of the children had infection with more than one parasite species. The study attributed the high worm manifestation to the lack of clean drinking water since most of those children (62.8%) drink surface water, (17.2%) drink water from the Nile and (45.6%) drink from untreated tap water.

Also the low educational level, and that lack of knowledge on personal hygiene as well as poor sanitation were the most important factors affecting parasite prevalence in that area. Regarding nutritional status as refuted to parasitic infection, the study revealed that (52%) of the children were mildly malnourished, (4.4%) moderately and (43.6%) reasonably fed. No severe malnourished or obese children were observed and that although high number of children was infected the parasite load was not heavy to affect the nutritional status. (Priscilla ,2003)

1-4 Haemoglobin:

Haemoglobin, is the iron – containing oxygen- transport metalloprotein in the red blood cells of the blood in vertebrates and other animals. In mammals the protein makes up about 97 % of the red cell's dry content , and around 35% of the total content (including water). Haemoglobin transport oxygen from the lungs to the rest of the body, such as muscles , where it release its load of oxygen . Haemoglobin also has a variety of other gas transport and effect- modulation duties, which vary from species to species (Hoffbrand and J.E Ppettit ,1988)

The name haemoglobin is the concentration of *haem* and *globin* , reflecting the fact that each subunit of haemoglobin is a globular protein with an embedded haem group , each haem group contains an iron atom, and this is responsible for the binding of oxygen . The most common type of haemoglobin in mammals contains four such subunits; each with one

haem group . In humans each haem group is able to bind one oxygen molecule , and thus , one haemoglobin molecule can bind four oxygen molecules (Hoffbrand and J.E Pettit ,1988)

It is increasingly recognized that intestinal helminthes may have a harmful impact on haemoglobin , growth and cognitive function of school age children . Previous studies have demonstrated this for heavy infection with *Hymenolepi.nana* , hookworms , *Schistosoma species* , *Trichuris trichiura* , and to a lesser degree *Ascaris lumbricoides*.(Steinberg, 2001) .

1-5 Leucocytes or White Blood cells:

White blood cells are the immune and defense cells of the blood. Generally most of their functions are performed in other tissues and they are merely transported in the blood. Any infection bacterial or parasitic increases number of white blood cells. High white blood cell counts may be due to inflammation. Leucocytes can be divided into two classes:

- Granulocytes.
- Agranulocytes.

(Hoffman , et al ,2005)

1-5-1Granulocytes:

Are named for their prominent and characteristic cytoplasmic granules as seen in standard blood smears. They are also known as polymorphonuclear leucoytes because their nuclei have several lobes. They are motile and phagocytic and can be divided into neutrophils, eosinophils and basophils. (Hoffman , et al ,2005)

1-5-1-1Neutrophils:

Neutrophils make up approximately 60% of blood leucocytes, they have segmented nuclei with 2 - 5 lobes, they from the first line of defense against most invading pathogenic organisms, particularly bacteria, they are highly phagocytic in their defensive role and in removal of damaged tissue after injury, they have mostly pale granules and some darker-

staining lysosomal granules. (Hoffman, et al, 2005)

The increase percentage of neutrophils due to the following, acute bacterial infection ,eclampsia -convulsion occurring with pregnancy associated high blood pressure, gout -acute arthritis with swelling and sever pain , resulting from disturbance of uric acid metabolism and characterized by an excess of uric acid in the blood and deposits of uric acid salts , usually in the joints of the feet and hands , especially in the big toe , rheumatoid arthritis -auto immune disease that caused chronic inflammation of the joints and tissues around joints , acute stress , thyroiditis -inflammation of thyroid gland which is located in front of neck , myelocytic leukemia -malignant disease in which too many white blood cells belonging to the myeloid line of cells are made in the bone marrow , the disease is due to the growth and evolution of an abnormal clone of cells contain a chromosome rearrangement known as the Philadelphia (Hoffman , et al ,2005).

A decreased percentage of neutrophils may be due to the following: Aplastic anemia-due to failure of the bone marrow to produce blood cells , including red and white blood cells as well as platelets , chemotherapy -treatment of cancer with drugs that can destroy cancer cells , Influenza -an illness caused by viruses that infect the respiratory tract , widespread bacterial infection e.g. (Mycobacterium) , radiation therapy -high energy rays are used to damage cancer cells and stop them from growing and dividing .(Hoffman , et al , 2005)

1-5-1-2 Eosinophils:

Eosinophils make up 1-2% of circulating leucocytes,Have bilobed nuclei. Eosinophils have bright red lysosomal granules which contain cytotoxic compounds such as eosinoplip cation protein, major basic

protein peroxidase as well as common lysosomol enzymes. They are primarily involved in defense against parasitic infection, particularly helminthes and protozoa. They are attracted by substances released by mast cells and activated lymphocytes. They may regulate aspects of the hyper sensitivity (allergic reactions) and immune reactions through inhibiting mast cell - secreted histamine (via histaminases) and vasoactive substances, inhibiting mast cell degranulation and through phagocytosis of antigen – antibody complexes. They accumulate in tissues and increase in blood (eosinophilia) in a number of circumstances e.g. hay fever, asthma, eczema and parasitic infections.

An increased percentage of eosinophils may be due to the following: Allergic reaction -hypersensitive response of the immune system of an allergic individual to substance, parasitic infection (helminthes – *Schistosoma mansoni* , *H.nana*), Hodgkin's disease -type of lymphoma , a cancer that develops in lymph system . (Hoffman , et al , 2005)

1-5-1-2-1Eosinophils and parasitic infection:

Eosinophils have been shown to be potent effecter cells for the killing of helminthes parasites invitro cultures. However an in vivo role for eosinophils has been more difficult to establish. Early data showed close association between eosinophils and damaged or dead parasites in histological sections , and significant correlation between resistance to parasite , and the capacity to induce eosinophilia after infection(Klion,and Nutman ,2004) .

Increases in the number of mast cells and eosinophils have long been

recogonized as characteristic features of infection with helminth parasites . Initially, it was thought that eosinophils could function as regulatory cells to dampen down the inflammatory reactions caused by mast cells degranulation . The demonstration that purified eosinophils could kill the early schistosomula stage of Schistosoma mansoni in in vitro cultures redirected eosinophil research towards an examination of their role as major effector cells in resistance to parasitic infections. Subsequently in vitro killing by eosinophils was also demonstrated for a variety of nematode parasites of human and veterinary importance . In most cases, eosinophil mediated killing was most effective against the larval stages and required cooperation with antibody and complement for maximum killing capacity (Klion ,And Nutman , 2004)

1-5-1-3 Basophil:

Basophils make up fewer than 1% of circulating leucocytes. they have bilobed nuclei often obscured by granules. They have deep blue granules that contain vasoactive substances (serotonin, histamine) and heparin (an anticoagulant). They are activated in allergic reactions through surface IgE receptors to release the contents of their granules and cause local tissue reactions and symptoms associated with acute hypersensitivity reactions(Mitre ,and Nutman , 2004)

1-5-1-3-1 Basophils and human parasitic infection

While basophilia is often found in animal models of parasitic infection , it has not yet been established whether it occurs in parasite-infected humans . The relationship between basophilia and parasitic infection in human has been investigated by reviewing charts from 668 patients with confirmed parasitic infection , 472 with only

helminthes, 146 with only protozoa, and 50 with both helminthes and protozoa infections , and from 50 patients without parasitic infection (Mitre and, Nutman , 2004).

Basophilia occurred in only four of the 668 patients (0.6%), and there were no statistically significance in the percentage of patients with basophilia or in the absolute basophil counts among either the helminth-infected , protozoa- infected , or uninfected population . Analysis with regard to relative basophil levels revealed that basophils constituted more than 3% of the peripheral white blood cell population in only four patients. Thus basophilia occurs only rarely in human parasitic infection and is consequently not a useful clinical marker in the evaluation of suspected parasitic disease. (Mitre and, Nutman, 2004)

1-5-2 Agranulocytes

1-5-2-1 Lymphocytes:

Lymphocytes make up 20-40% of circulating leucocytes. They play central roles in the generation of specific immune response. They are comprised of two main types, B cells and T cells, which perform different but linked roles in the generation of immune response. They are mostly have round, condensed nuclei typical of cells with little biosynthetic activity, cytoplasm forms only a narrow rim around the nucleus. About 3% of circulating lymphocytes are larger and represent activated B cells en route to tissue where they will become plasma cells or a small population of lymphocytes called natural killer cells. They have variable life – span with some cells living many years (e.g. memory cells) . (Hoffman R ,Benz et al 2005)

An increased percentage of lymphocytes may be due to the

following :Chronic bacterial infection e.g. (tuber closes) , Infectious hepatitis -Inflammation of liver , due to infection of hepatitis viruses (ABCD,E), Infectious mononucleosis -illness caused by the Epstein – Barr virus EBV , Lymphocytic leukemia (cancer of lymph) ,Viral infection (such as infectious mononucleosis, mumps, measles) , Recovery from a bacterial infection (Hoffman , et al ,2005).

A decreased percentage of lymphocytes may be due to the following :

Chemotherapy -treatment of cancer with drugs , HIV infection -Human immunodeficiency virus , the cause of AIDS (acquired immunodeficiency syndrome) , Leukemia -cancer of the blood cells , the growth and development of the blood cells are abnormal , Radiation therapy high energy rays are used to damage the cancer cells and stop them from growing , Sepsis -blood stream infection , the presence of bacteria and infectious organism in blood. (Hoffman ,et al ,2005)

1-5-2-2 Monocytes:

Monocytes make up 2 – 4% of leucocytes. They are precursors of tissue macrophages. They are large cells with indented nuclei . They are capable of phagocytising, engulfing and destroying bacteria and various materials recognized as foreign. (Hoffman ,et al, 2005)

An increased percentage of monocytes may be due to the following :

Chronic inflammatory disease (TB) , Parasitic infection (helminthes , *A . lumbercoids , H.nana , S . mansoni*) , Tuberculosis-infectious disease caused by bacteria whose scientific name is Mycobacterium tuberculosis ,

commonly infect the lung , Viral infection (for example, infectious mononucleosis, mumps, measles) (Hoffman ,et al ,2005)

1-6 Objective :

The objectives of our study are the followings:

- To detect the occurrence of *Hymenolepis nama* in Khartoum state.
- To detect the effect of *Hymenopepis nana* infection on Haemogeobin, Total white blood cells count and differential white blood cells.

Chapter Two

Chapter two

Materials and Method -2

:2-1study area

.The study was carried out in hospitals of Khartoum state

:Study population 2-2

The study 168 samples (stool – blood) according to questionnaire and stool examination they divided into three groups, infected group, infected before group and control group, all samples collected from adult.

:Collection of the specimens 2.3

Faecal specimens were collected in a wide mouth stool containers, and they were labelled with the patient name and laboratory number. Blood specimens were collected by taking venous blood from the patient using a syringe ,then the blood was mixed with suitable anticoagulant .(EDTA) before any investigations

:Direct wet preparation technique 2.4

The collected stool from the patients were examined by mixing a small portion of stool taken with a wood stick with a drop of normal saline on a slide and covered with cover glass slip. The prepared slide were examined systematically under the microscope using (x 10), and the .high power magnification (x 40) for observation of more details

2 .5 formal ether concentration technique :

Aproximately, one gram of faeces was collected from different parts of the specimen. The faeces was emulsified in about 4 ml. of formol water (10 % Fomalehyde) in a conical tube. Further 4 ml. was added and mixed. The resulting suspension was strained through the sieve. The filtrated sample was poured back into a conical tube. Debris trapped in the sieve was discarded and a 4ml. of diethyl ether was added and the tube was stoppered and shaken. Then the tube was centrifuged at 2000 rpm for two minutes. Thus, the preparation was separated into four layers, ether at the top followed by faecal debris and formaldehyde and the sediment in the bottom. All layers were removed except the sediment.Then the seiment was transferred onto a slide and covered with .(a cover glass and was examined microscopically using (x10) and (x40

Haemoglobin concentration :2 6.

ml of blood was taken by 0.02 ml pipette from a blood 0.02 specimen well mixed in a container with suitable anticoagulant (EDTA).Then the specimen was added to 4 ml of Drabkin solution in a test tube, this was allowed to stand for at least 3 minutes, then the absorbnce is measured on a colorimeter. The absorbance was read against .either distilled water or reagent blank at a wave length of 540

:Total white blood cells count 2.7

ml of blood was added to 0.38 ml of Glacial acetic acid in a 0.02 test tube, this was allowed to stand for 5 minutes, then a drop of the solution was transferred to the counting chamber ,covered with a cover glass, and placed under the microscope using (x 10).The four corner squares of the counting area were counted for white blood cells, the total number is multiplied by 50 to obtain the total number of cells per cubic

.millimeter

Differential leucocytes count :2 8.

A thin blood film was prepared by placing o drop of blood onto one end of a clean grease-free slide , the drop was allowed to spread along it , then the slide and the spreader slide was hold at a suitable angle 45◦ , then the spreader was pushed along the slide drawing the blood .behind it until the whole drop was smeared

Then the thin blood film was stained with Leishman stain, which .is a Romanowsky – type stain designed to differentiate leucocytes

Then the cells were counted in a series of complete longitudinal strip of the film until 100 cells were counted. All types of leucocytes were differentiated and classified. The count

.of each cell type was recorded and then expressed as percentage

:Statistical Analysis 2-9

The data was analyzed by using SPSS computer program., one way ANOVA.

Chapter Three

Chapter Three

Result

specimens (stool , blood) were examined ,68 stool 168 specimens were found to be positive for *Hymenolepis.nana* ,the rest were negatives , among negatives and according to questionnaire 50

infected before with *Hymenolepis.nana* ,and 50 were non infected with *hymenolepis nana* and this group used as control . the prevalence of (*hymenolepis nana* was found to be 40.5% (table 1

:*Table (1):* Number of examined samples and prevalence of *H. nana*

Part	Infected with *Hymenolepisnana*	Infected before	Control	Prevalence of *Hymenplepisna na*
No of samples	68	50	50	40.5

The percentage between males and females was found to (be63.7% male and 36.3% female (table 2

***Table (2)* The percentage between male and female:**

Sex	Male	Female	Total
Frequency	107	61	168
Percent	63.7%	36.3%	100%

Table (3) shows that the mean of hemoglobin was found to be 9.8 g/dl among those who were infected with H.nana, 11.2g/dl among those who were infected before , and 12.9 among the control . The significance (difference between groups (P<0.000)(appendix 5

:*Table (3)* The mean value of hemoglobin in three different groups

Part	Infected with *hymenolepisnana*	Infected before	Control	I
Mean of Hemoglobin	9.8	11.9	12.9	M Fe

The mean of total white blood cells count was found to be9.944X10 ^{3} among those who were infected with *H.nana* ,6.362X10 ^{3} among those who were infected before , and 3.646X10 ^{3} among the control (table 4) . (The significance difference between groups (P<0.000) (appendix 5

Table (4)**The mean value of T.W.B.C count in three different groups :**

Part	Infected with *H.nana*	Infected before	Control	Norma range
Mean of Total white blood cells count	X10^{3}9.944	X10^{3}6.362	X10^{3}3.646	3x10^{3} ------- 9x10^{3}

The mean of differential white blood cells count was found in granulocyte , 61% neutrophils , 2% eosinophils, 0% basophils among those who were infected with H.nana , 60% neutrophils , .46% eosinophils , 0%basophils among those who were infected before , and 64% neutrophils, .22%eosinophils , 0% basophils among control(table 5) . The significance difference between groups neutrophils(P< 0.502) ,

(eosinophils(P< 0.000) , basophils no significant (appendix 5

Table (5):**The mean value of granulocytes white blood cells in three different groups:**

Infected with H.nana	Infected before	Control	Normal range
61	60	61	60%
2	0.46	0.22	1-2%
0.00	0.00	0.00	Fewer than 1%

The mean of differential white blood cells count , Agranulocytes , 4% monocytes ,32% lymphocytes among those who were infected with H.nana , 2% monocytes , 34% lymphocytes among those who were infected before with Hymenolepis.nana , and 2%monocytes , 29% lymphocytes among control (table 6) . The significance difference between groups , monocytes(P< 0.000) , lymphocytes(P< 0.000) ((appendix 5

Table (6):**The mean value of a granulocytes white blood cells in three different groups:**

Infected with H.nana	Infected before	Control	Norma lrange
5.7	3.6	3	1-4%
30	34	35	20-40%

Chapter four

Chapter four

4-1 Discussion

The study showed that there is a significant effect of helminthic infection on the hemoglobin and total white blood cells count and differential white blood cells.

Mean value of hemoglobin was found to be (9.8 g /dl) among those who were infected with *H.nana* , (11.9 g/dl) in those who were infected before and (12.9 g/dl) among the control group (table 3) , and this values were observed statistically with highly significant difference with P value (0.000) (appendix 5) , this result agree with (Steinberg,MA 2001) , stated that Intestinal helminthes may have harmful impact on hemoglobin . The health status of 239 refugees in the USA was evaluated. Of those, 36.7% were found positive for intestinal parasites. Anaemia was reported in 14%. Normal range 12-17g/dl for male and 11-14g/dl for female

The mean of total white blood cells count was found to be ($9.944X10^3$) in the blood samples of those who were infected with *H.nana* , ($6.362X10^3$) among those who had previous infection , and ($3.646X10^3$) among the control (table 4).and this values were observed statistically with highly significant difference , with P value (0.000) (appendix 5) , and this agree with(Hoffman , et al ,2005) .who stated that any infection bacterial or parasitic increases number of white blood cells. High white blood cell counts may be due to inflammation . Normal range ($3X10^3$-$9X10^3$).

There were normal in neutrophils as compared with normal value 60% (table 5).and this agree with(Hoffman , et al ,2005) who stated that the increase percentage of neutrophils due to acute bacterial infection .No significant (0.502) (appendix 5) .

Eosinophils were increased among the infected patients and were normal among the control and who infected before (table 5) . and this values were observed statistically with highly significant difference with P value (0.000) (appendix 5), and this result agree with (Klion, and , Nutman , 2003) , Stated that early data showed close association between eosinophils and damage dead parasites in histological section , and significant correlation between resistance to parasites , and the capacity to induce eosinophilia after infection . The health status of 239 refugees in the USA was evaluated. Of those, 36.7% were found positive for intestinal parasites. was reported esinophilia in 11%) . Normal range (1-2%).

Monocytes were increase among those who were infected with *H.nana* , and was noticed to be normal in the samples of patients with previous attack and control (table 6) . and this values were observed statistically with highly significant difference with P value(0.000) (appendix 5) , and this result agree with (Hoffman.R .Benz et al 2005), an increase percentage of monocytes may be due to parasitic infection , helminthes and protozoa. Normal range (2-4%).

Lymphocytes were noticed to be normal with variation between groups of samples (table 5), normal range (20-40%).

Result of study showed that infection with *H.nan*a is common in males, who were more affected than female with percentage 63.7% for males, and 36.3% for females (table 2).

The prevalence of *hymenolepis nana* among 168 adults were found to be 40.5% which was very high and this may be due to the poor hygiene and sanitation among the people.

4-2 *Conclusions*

1. *Hymenolepis nana* are highly prevalent among the population of Khartoum (from the result table 1 the prevalence 40.5%), high prevalence may put the health service under attention.
2. Direct wet preparation technique was found to be sufficient for detection of *Hymenolepis nana* when there is infection.
3. Formal ether concentration technique is still the best method for detection of *Hymenolepis nana* in infected individuals for its high sensitivity but time consuming.
4. *Hymenolepis nana* infection reduce hemoglobin concentration may cause anemia, increase total white blood cell count, increase eosinophils and monocytes.
5. *Hymenolepis nana* infection has no effect on neutroplils, lymphoytes and basophips.

4-3 Recommendation

1. Improving the hygienic standards among the population in Khartoum to help in controlling *Hymenolepis nana.*
2. Good hygiene, public health and sanitation programs are very useful in elimination of *Hymenolepis nana* infection.
3. The use of formal ether concentration technique for the detection of *Hymenolepis nana* as it is highly sensitive, easy to apply and cheap but time consuming.
4. Control Rats which are common host of *Hymenolepis nana* and in addition to that control the insect vector.
5. Mass treatment in endemic areas of Khartoum.

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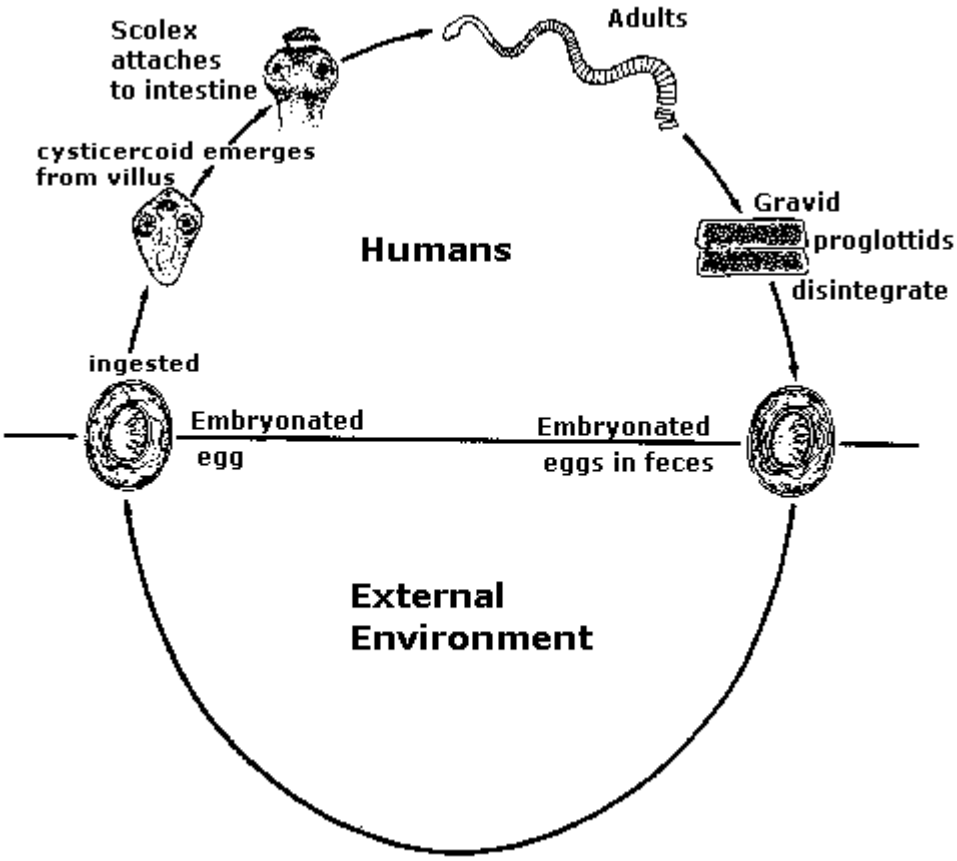
6- Appendix

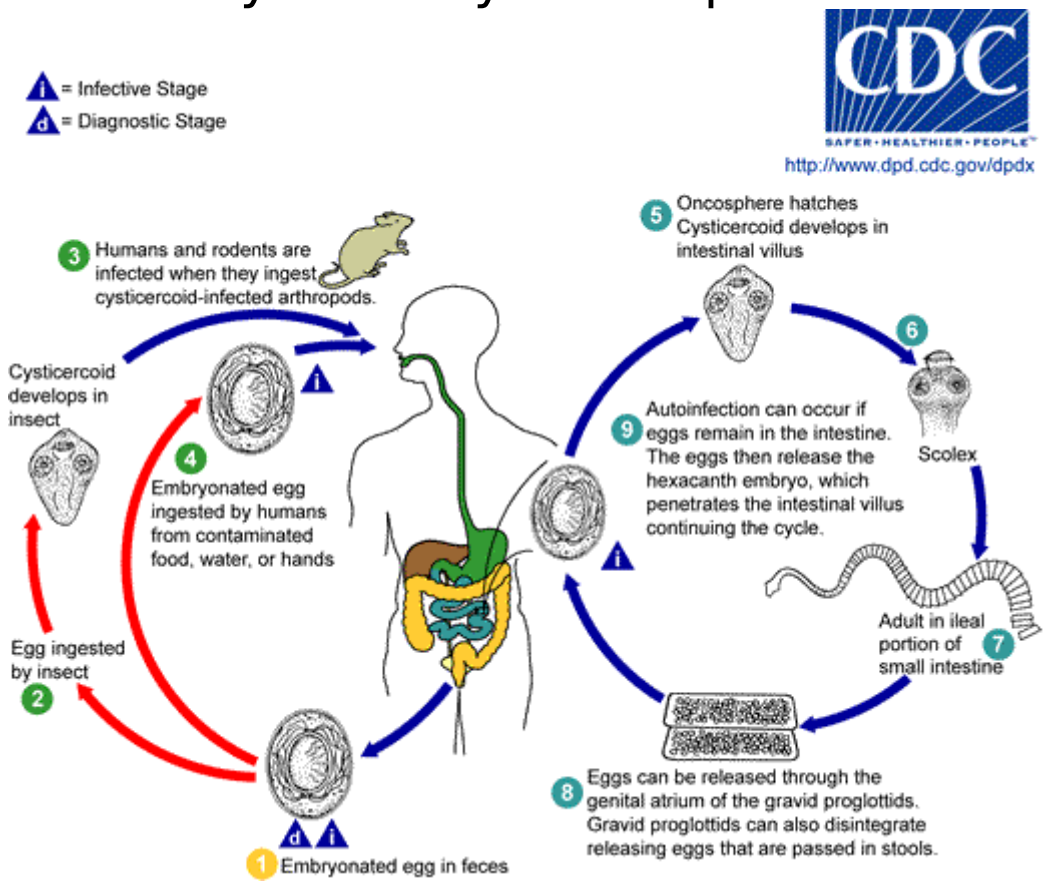
Appendix-1

Life cycle of Hymenolepisnana

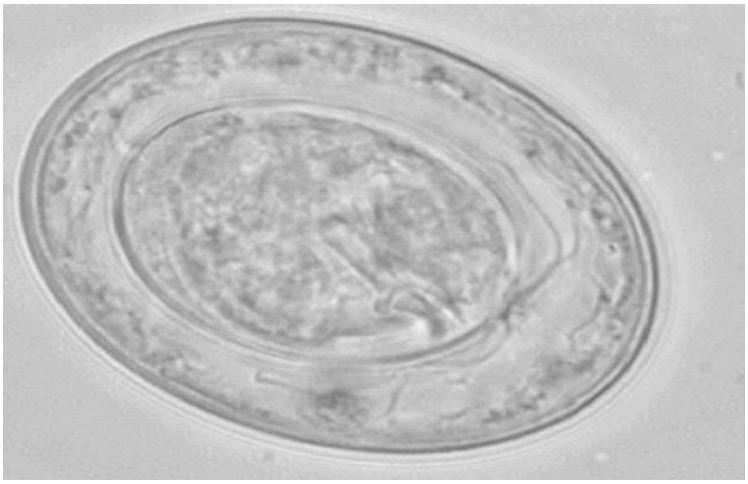


Appendix-2

Color Life cycle of Hymenolepis nana



Appendix -3
Eggs of Hymenolepis nana



3-1Egg of *H .nana* in wet preparation

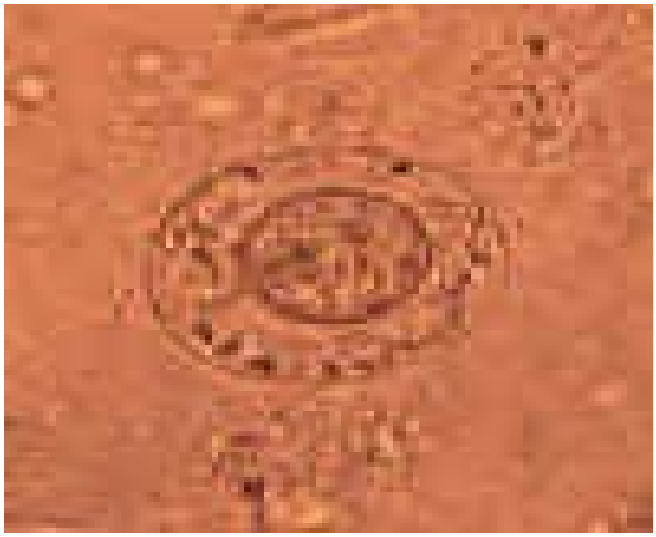
3-2 Color egg of *H.nana* stained by Trichrom stain

Appendix: 4

3-3 Egg *of H.diminuta*

Egg *of H. nana* stained by permanent stain 3-4

Appendix: 4

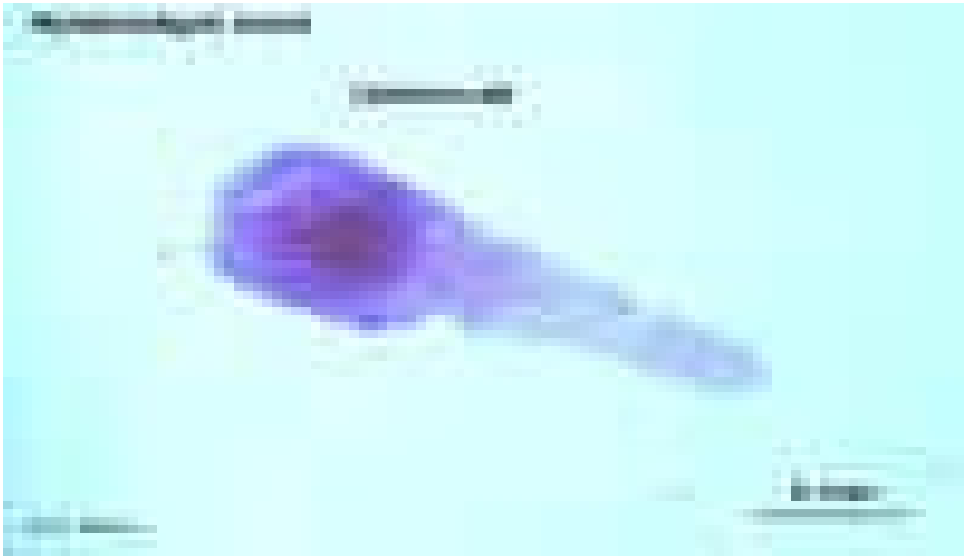


Egg of *H.nana* wet preparation stain with iodine 4-1

Three Adult worm *H.nana* 4-2

Scolex of adult worm 5-3

Adult wor*m H.nana* **4-3**



Scolex of adult worm **4-4**

Appendix: 5

:Statistical analysis

		Significance
Haemoglobin	Between Groups	.000 +
	Within Groups	
	Total	
TWBCs *	Between Groups	.000 +
	Within Groups	
	Total	
Neutrophils	Between Groups	.502 -
	Within Groups	
	Total	
Lymphocytes	Between Groups	.000 +
	Within Groups	
	Total	
Monocytes	Between Groups	.000 +
	Within Groups	
	Total	
Eosinophils	Between Groups	.000 +
	Within Groups	
	Total	
BASophils	Between Groups	.
	Within Groups	
	Total	Significance

* Total white blood cells

+ Significant differences

- No significant differences

Apendex6

:Equipments -1

.Glass slides .1

Cover glass .2

.Wood sticks .3

.(Glass tubes.(12x75mm .4

.Conical glass tubes .5

.Plastic sieve .6

7. Blood containers (EDTA)

.Stool containers .8

9. 10ml. pipette

1ml. pipette .10

.0.02ml. pipette .11

.Capillary tubes .12

.Light microscope .13

.Centrifuge .14

.Colorimeter .15

.Neubar Counting Chamber .16

Syringes .17

.Blood lancets .18

:Reagents -2

.(Normal saline (0.9 Nacl .1

.Formaldehyde .2

.Diethyl ether .3

.Drabkin solution .4

.(Glacial Acetic acid (2% .5

.Leishman stain .6

.(Disodium hydrogen phosphate (buffer .7

.Haemoglobin standard .8