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Diagnosis incidence and treatment of enterobius vericularis in North America

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Thesis

DIAGNOSIS, INCIDENCE AND TREATMENT of *ENTEROBIUS VERMICULARIS* in NORTH AMERICA

by

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TOPICAL OUTLINE

LIST OF ILLUSTRATIONS

I

INTRODUCTION

This thesis represents an effort to compile all the information and work available on Enterobius vermicularis. It was thought advisable, however, to restrict the incidence reporting to North America except for the noting of the world-wide distribution of the pinworm.

Approximately two-thirds of the sources used were those found in a thorough search of the Biological Abstracts - 1927 to 1948 inclusive. The remaining sources were gathered from the bibliographies of the original sources. Much valuable material was obtained from the more than twenty papers presented in various scientific periodicals by the personnel of the National Institute of Health, who from 1937 to 1944 undertook a deliberate, detailed study of enterobiasis in all of its phases and ramifications.

Information on the biology of the pinworm was included because it was felt that such information would be a helpful and a necessary supplement to the material on diagnosis, incidence and the treatment of enterobiasis. As a piece of original work to be incorporated into the thesis, the incidence of E. vermicularis was studied at an institution for boys in Boston.

II

BIOLOGY OF ENTEROBIUS VERMICULARIS

A. History and Importance

Sawitz, D'Antoni, Rhude and Lob (1940) stated,

> "Alexander of Tralles in the Sixth Century, ascribed to pinworms 'the priviledge over other helminths of being tormentors of any people of any age; and Leuckhart some seventy years ago, believed that' there may be only a few who do not harbor them at one time or another."

Cobb (1890) wrote that,

> "the common pinworm or threadworm parasitic in man, is the Nematode that has been longest known and is the one that comes most frequently under the notice of physicians and helminthologists."

Undoubtedly, millions of people in this country are infected with pinworms, and among the other nations of the world many millions of other persons are so infected.

B. Classification

The pinworm is also known in other parts of the world as the threadworm. Leiper (1926) disclosed that Linnaeus in 1758 called the species Ascaris vermicularis and that Leach in 1853 placed the helminth in the genus Enterobius. Stiles in 1905 placed this same worm in the genus Oxyuris. Earlier in the century Oxyuris vermicularis was used to designate the pinworm. Now, however, Enterobius vermicularis (L. 1758) Leach 1853 is the accepted and approved term.

According to Chandler (1944) the pinworm is classified as

follows:

Phylum Nemathelminthes - unsegmented roundworms.

Class Nematoda - triploblastic, no true segmentation, complete digestive tract, no circulatory or respiratory organs and dioecious.

Subclass Phasmida - phasmids present; excretory system present.

Order Ascaroidea - including most of the nematodes, mouth usually with 3 lips.

Genus and species - Enterobius vermicularis, (L. 1758) Leach, 1853.

C. Distribution

While this report is concerned with the North American continent, we must acknowledge the world wide distribution of Enterobius vermicularis. Below is given a list of countries outside of North America in which the pinworm has been identified as being present in the populace.

Table I. World-Wide Distribution of Enterobius.

AREA	INVESTIGATORS	SPECIFIC LOCATION IF ANY
Armenia	Plavtov (1926)	
Australia	Penfold and Penfold (1936)	State of Victoria
Belgian Congo	Fisher (1934)	
Bulgaria	Drenowsky (1943)	Southern Bulgaria
China	Schwartz (1926)	
Finland	Ehrstrom (1926)	

Formosa----------Ishii and Tei Tok Wa (1931)

Guam-------------Stoll, Chenoweth, and Peck (1947)

India------------Korke (1926)

Italy------------Pulle (1927)

Korea------------Mills (1927)

Madagascar-------Sice (1927)--------------Southern Madagascar

Philippines------Chanco (1939)

Roumania---------Nitzulesco (1939)

Russia-----------Dubrovinskii----------------Moscow

Dubrowinski (1928)----------Turkmenistan

Levin (1930)

Nevland-Dobrova (1930)--------Stalingrad

Shmeleva (1934)--------------Kalmuck

Volplenko (1943)-------------Baku

Zhukova-Florensova (1930)-----Kransnoyarsk

Sweden-----------Klercher (1927)--------------Southern Sweden

D. Morphology of Adult Pinworms and Ova

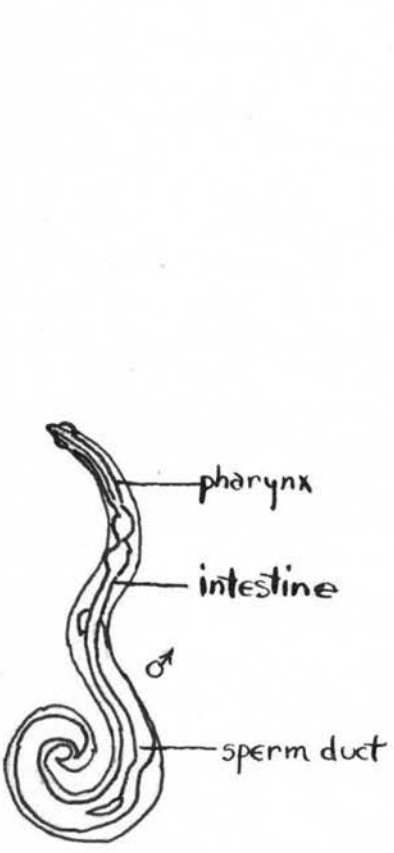


Fig.1
Adult male pinworm
(After Leuckart.)

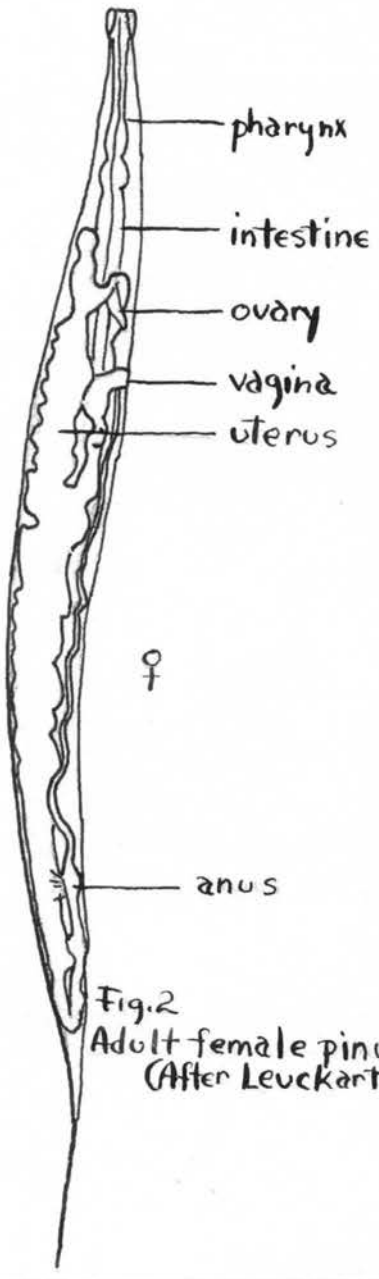


Fig.2
Adult female pinworm.
(After Leuckart.)

1. Male Pinworm

According to Craig and Faust (1937) the male pinworms are 2 to 5 millimeters long, 0.1 to 0.2 millimeters wide at their greatest diameter, and have their posterior ends curved ventrad. The male pinworm has a conspicuous spicule, but no gubernaculum. The bursa is greatly reduced and is supported by a pair of anterior pedunculated papillae and a pair of posterior papillae.

2. Female Pinworm

Craig and Faust (1937) put the size of the female pinworms at more than twice that of the males, measuring 8 to 13 millimeters by 0.3 to 0.5 millimeters. The tail portion is decidedly

attenuate and makes up one-third of the entire length. The vulva is located mid-ventraly one-third of the distance from the head region. The vagina is rather long and extends some distance posterior from the vulva before it joins the paired genital organs, made up of uteri, oviducts, and ovarian tubules.

E. Morphology of Pinworm Ova

Chandler (1944) described the ova as being clear, unstained, usually 55 by 30 μ, and not perfectly oval, being flattened on one side. Jacobs and Jones (1939) identified three layers in the cuticula of the egg. The outer layer composed of an unidentified protein substance, the middle of chitin compound, composed of two layers and the innermost layer being some steroid substance.

Both Jacobs and Jones (1939) and Cobb (1890) agree as to the existance of a definite hatching point on the shell proper but differing as to its nature. Jacobs and Jones state the hatching point is thin due to the absence of the chitin layer; while Cobb describes a porous area in the shell at which place he believes hatching to take place.

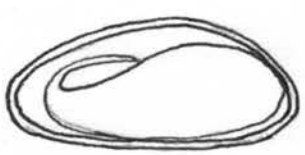


Fig. 3. Tadpole stage of pinworm ova. (After Chandler.)

F. Life Cycle

The life cycle of the pinworm is an exceedingly interesting one. Craig and Faust (1937) related that in the gravid female the uteri become distended and filled with eggs. The pressure of this egg mass on the esophagus causes the female to release her hold on the intestinal mucosa so that she migrates from place to place within the intestinal lumen, finally making her way through the anus to the perianol folds where mature and fully embryonated eggs are shed.

We are indebted to Bozicevich and Brady (1938) for a really detailed account of the migration and oviposition of the gravid female pinworms. They observed that the gravid females at 8:00 P.M. were located an inch away from the mucocutaneous junction and at 9:00 P.M. they had reached the junction. At this time, the boys being examined retired and the worms were found on the moist areas of the skin forward as far as the scrotum and in a 2.5 inch radius from the anus posteriorly and laterally. Locomotion was accomplished by alternate head and tail anchorages to the skin and mucosa. The worms which migrated to the body surface were far less active than were the ones present further up in the canal. After a few minutes exposure to the air, increased opacicity and considerably less activity was noticed. Eggs were discharged during violent uterine contractions by which large white egg masses were released. The eggs were expelled in cylindrical streams and frequently hung in festoons. Dessicated and

shriveled pinworms were sometimes found the next morning in the pajamas of the boys.

Most authorities, including Jones and Jacobs (1941), agree that the eggs deposited in the perianal region are in the tadpole stage and develop into the infective or ring-and-a-half stage after five to seven hours at a temperature of 36°C or at correspondingly longer times for temperatures lower than 36°C but above 20°C. It is not always necessary for the worms to undergo any development outside of the body although as Cobb states such is probably the normal case. Cobb proved this latter statement by inserting tadpole stage pinworm eggs into the stomach and observing that they hatched.

The means by which the infection may be acquired are many and diversified. Commonly it takes place by placing contaminated objects into the mouth. Cobb believed that anything which comes in contact with the eggs aids in their transportation. Cobb listed the three important vehicles as food, drink, and air. He likewise named many articles which are indiscriminately handled and lend themselves to easy infection; such as vegetables, fruits, bread, cakes, and pastry, butter, cigars, and cigarettes, drinks (non-alcoholic) and eating utensils. Ishii (1931) listed 17 kinds of vegetables as containing eggs of helminths among them Enterobius. Pipkin (1943) has cited the importance of insects as mechanical transmitters of Enterobius, and named several species of laboratory bred flies (Musca domestica, Lucilia pallesceno, Cochliomyia macellaria,

Phormia regina and Sarcophaga miserio) which were artificially soiled with contaminate material. External carriage of eggs of Enterobius vermicularis varied from 1.37 to 3.47 hours.

Cobb (1890), Chandler (1944), Cram, Reardon and Nolan (1937) and countless others believed that the infection is air borne and can be acquired by inhaling infectious dust. Schueffner and Swellengrebel (1947) performed experiments to demonstrate the existance of the mode of infection. Eggs of Enterobius vermicularis which had been collected from an untouched window sill (4 to 20 days untouched) were washed in a 2 per cent hydrochloric acid solution and centrifuged. Eight volunteers were fed this dust containing 40 to 80 eggs per person in bread. Six discharged pinworm ova thirty-six to fifty-three days after ingestion. The discharge lasted from twelve to twenty-six days. The experiments led the authors to maintain that "dust eggs" are infectious.

Man is the only animal which has the doubtful honor of playing host to Enterobius vermicularis. Jones and Nolan (1942) sought to infect vitamin A deficient rats with Enterobius. The worms failed to develop in any of the 36 albino rats on a vitamin A deficient diet or in any of the 40 control albino rats. In both groups some larvae were found in the gastro-intestinal tract after feeding the eggs. Pinworm larvae survived for a few days in the intestinal tract of guinea pigs and mice but it proved impossible to establish infections in either these animals or in dogs or in rhesus monkeys.

In the middle part of the nineteenth century a great

dispute arose among the parasitologists of the time. Did oxyuriasis take place by ingestion of infective eggs? Vix, (1860) Kuchenmeister, (1855) Goebel, (1922) believed the infection could be built up without the need of the gravid female worm ever migrating. They based this assumption on their simultaneous finding of eggs, larvae and adult worms within the large intestine. At first glance this observation would seem to point to the generation of pinworms without the females ever leaving the host by depositing and subsequent hatching of pinworm ova within the intestinal tract.

Zavadovskii and Shalimov (1929) disputed the theory of generation of pinworms within the intestine by taking eggs from the appendix and observing whether or not development took place in vitro. These eggs from the appendix did not develop, forcing the authors to the conclusion that the capacity to develop depends upon the stage of the embryo; the pre-tadpole stage (appendiceal eggs) being incapable of development. From the tadpole stage on, the eggs require oxygen, since oxygen is presumably not present in sufficient quantities in the lumen of the intestine multiplication within this organ is impossible. The voided eggs which were in the tadpole stage showed development.

Cobb (1890) sought to prove that infection was acquired by ingestion of eggs by demonstrating hatching of Oxyuris ova in the human stomach under normal conditions. A suction capsule was prepared to convey the ova of Oxyuris into the stomach. On reaching the stomach, gastric juice was admitted

and allowed to act on the eggs under normal conditions. The capsule was then recovered from the feces. Nearly all of the 12 or 13 eggs hatched. Greater development was seen in these eggs than in the controls which were left inside the dead and dry female worm. The largest larva was some 0.11 millimenters in length, the smallest barely developed beyond the tadpole stage at which they were inserted into the stomach.

Cobb in trying to rear pinworm eggs in artificial digestive juices (lacto peptone and other proprietary preparations) discovered fine pores in the egg shells. He discovered a distinct area, where the pores were very numerous which became the exit point for the larvae during hatching.

Fig. 4. longitudinal section through a pinworm ovum showing porous exit area for developing larva. x1300. (After Cobb.)

Cobb believes these pores to be present previous to any resultant action of digestive juices. In hatching, Cobb found that the embryo leaves the shell tail first. A fully developed embryo is doubled up on itself and the posterior bend located

in the anterior part of the shell leaves first, followed by the anterior part and ultimately the head.

Embryology of Pinworm

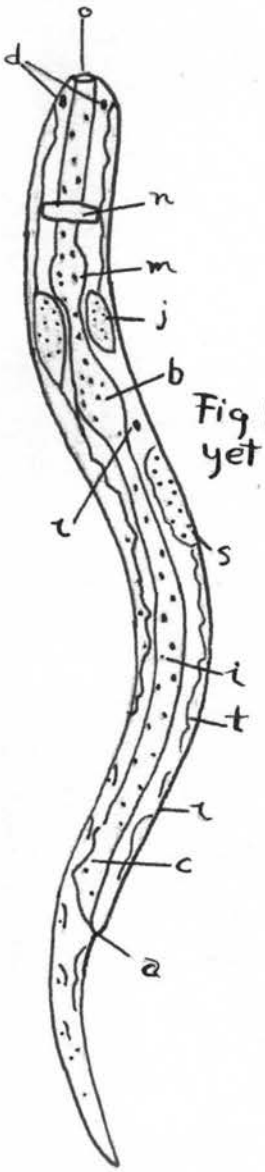


Fig 5. Tadpole stage

Fig 6. Ring and a half stage

Fig 7. Mature ovum

(Fig. 5-7 after Chandler)

Fig 8 Larva of Enterobius vermicularis not yet six hours old. x900. (After Cobb.)

Explanation of drawing

a- Anus
b- Posterior bulb of the esophagus
c- Constriction in intestine
d- two large cells at head
e- Ampulla
i- Intestine
j- Jugular cells
t- Muscle cells
m- median bulb
n- nerve ring
o- mouth
p- Porous area on the egg (Fig 4)
s- Sexual organs
t- Cuticula
v- Ventral gland

After hatching in the stomach, Craig and Faust (1937) related that the larvae undergo two moults while passing through the small bowel. The larvae may become ledged temporarily in the crypts and folds of the jejunum and upper parts of the ileum. Upon reaching the level of the intestine they mature and attach themselves to the intestinal mucosa.

The life cycle of Enterobius vermicularis is completed in two months.

III

DIAGNOSIS OF ENTEROBIUS VERMICULARIS

Sawitz, Odom and Lincicome (1939) report the difficulties encountered in the attempts at diagnosing Enterobius which are not found in the other intestinal helminthic parasites. The ova of Enterobius are deposited most commonly at night, by the gravid female worms, in the perianal folds.

Kuchenmeister (1855) advised that diagnostic activities for the presence of Enterobius be confined to fecal examinations. Davaine (1858) and Lambyl (1859) also advised fecal examinations to disclose the presence of Enterobius. However, it was Davaine in 1860 and Vix in the same year who first emphasized the anal region as the optimum site for diagnosing the presence of Enterobius. Vix (1860) and later Heller in 1876 obtained their best results by anal scrapings with various blunt instruments including scalpel handles, spatulas, and probes.

The methods available for diagnosing pinworm infections are several but can be divided into two major divisions, physical and serological techniques.

A. Physical Methods

1. Stool Examinations Via Flotation Techniques

Two types of flotation techniques which have been used in connection with pinworm diagnosis are (1) Willis - brine and

(2) the zinc sulphate centrifuge concentration techniques.

The Willis - brine techniques as explained by Sawitz, Odom, and Lincicome (1939) consists of straining 2 grams of freshly passed feces into a glass jar together with 8 cc of distilled water. Two cc of this solution is pipetted into a Wasserman tube, water added, and centrifuged for forty-five seconds at 2,640 revolutions per minute. The supernate fluid is decanted, water added and the preparation recentrifuged. The procedure is repeated three times. Finally, after the last supernate fluid is poured off, brine (specific gravity of 1.2) is added, the solution is shaken, and a coverglass set on top of a full Wasserman tube. The solution is then centrifuged for forty-five seconds at 2,640 revolutions per minute, after which the coverglass is examined for the presence of pinworm ova.

In the zinc sulphate centrifuge flotation techniques, the same procedure is used as that employed in the brine techniques except that to the final centrifuge solution is added zinc sulphate (specific gravity of 1.18) instead of brine. The solution is then centrifuged for forty-five seconds at 2,640 revolutions per minute. A bacterial loop dipped into the levitated material, and the loopful placed on a microscopic slide and examined. The buoyancy of the pinworm ova was determined by Sawitz (1939) by tests with zinc sulphate solutions of various specific gravities. The values determined, however, are not necessarily the actual specific gravities, for in order to concentrate the eggs during centrifuging a solution of a higher specific gravity than its own is needed. The time

for centrifugation is a function, mathematically, of the difference between the specific gravity of the eggs and the solution. Pinworm eggs were found to be buoyed up in solutions of specific gravity of 1.115 or higher. At a solution specific gravity of 1.180, 97 per cent of the total eggs present in the solution rose to the surface layer and 70 per cent of the total eggs present on the first coverglass preparation. Thus it seems that in zinc sulphate solutions of pinworm eggs a solution specific gravity of 1.180 gives the best results.

2. Subungual and Rectal Endoscopic Examination

It has long been known that in cases of pinworm infections ova are frequently found under the fingernails of the infected persons. Levin (1930) made microscopic examination of subungual material taken from under the toe or finger nails which disclosed 87.6 per cent of the infected persons. Schuffner and Swellengrebel (1944) examined the dirt under the fingernails and in this way 35 per cent of the positives were diagnosed to be positive for pinworm ova. Still (1899) found a single ova underneath the fingernail of one of five children given subungual examination.

Sotolongo and Goldberg (1941) introduced a novel means of diagnosis of pinworm infections. By means of a delphic optic proctosigmoidoscope, they were able to see pinworms in the anal canal and internal sphincter areas.

3. Anal Swabs

The work done by Hall (1937) marks the turning point in the diagnostic history of Enterobius vermicularis. Up to this time Enterobius was regarded as of slight consequence in the overall picture of helminth infections of man. By use of the Hall NIH swab the realization of high incidence rates with pinworms was disclosed.

Hall (1937) in his classic work on types of anal swabs sets forth certain characteristics which the ideal swab for detecting the presence of pinworm ova should possess:

> "It should pick up pinworm eggs, when these are present, quite dependably; should deliver them to a microscope slide dependably, with the minimum amount of manipulation that involves dilution or possible loss of eggs and with safety to persons handling the swab; and should cause the patient little or no inconvenience in its use."

Hall tried many types of swabs before settling on the one type which he felt best met the above qualifications.

A. Cotton Swab

Absorbent cotton is twisted about one end of a wooden applicator, usually moistened before use. The swab was examined either by salt flotation or rubbing and rolling the cotton in a small amount of water on a slide; the released material was thereby examined on a slide. Examination showed that frequently the eggs were entangled in the cotton fibers and was not released on the slide. This type of swab is of small value because it fails to release the ova it picks up.

The flotation was more valuable on this score than the smear.

Dry cotton swabs were used and then placed in a $\frac{1}{10}$ normal solution of sodium hydroxide. Still, not all the eggs were released and cotton swab testing was abandoned.

B. Chamois Swab

Chamois swabs were used with the idea that a smoother surface would release its eggs much more readily than did the cotton. Chamois was rolled on an applicator and held to it by a rubber band. This type of swab proved to be impractical due to the fact that the wooden applicators were too fragile for the pressure needed to pick up the ova, and this rolled swab was not suited to the swabbing movements necessary for proper operation.

C. Rayon Swab

Rayon was used in swabs on the theory that a fabric mesh of smooth rayon would pick up eggs dependably and release them readily. A cotton swab was made, covered with rayon and fastened with a rubber band. The rayon swab was moistened and after use, rubbed and applied to some water on a slide. The swab was fairly satisfactory, but, it was noted that some of the eggs passed through the rayon mesh and collected in the cotton fibers. This particular type of swab failed to deliver dependably the eggs to the slide.

Variation of this type of swab was tried. A dry swab was

used, then placed in a vial along with a drop of $\frac{1}{10}$ normal sodium hydroxide to break up any fecal matter picked up and then the swab was examined in a small amount of the sodium hydroxide solution. This type of swab was an improvement but still did not dependably release its eggs to the slide.

Another variation involved the use of a dry swab. After use it was moistened in a water vial and then examined it in a small amount of the sodium hydroxide solution. This appeared to be the better of the three rayon swabs tried, but still did not prevent the passage of the eggs through the rayon mesh.

D. Celluloid Scrapers

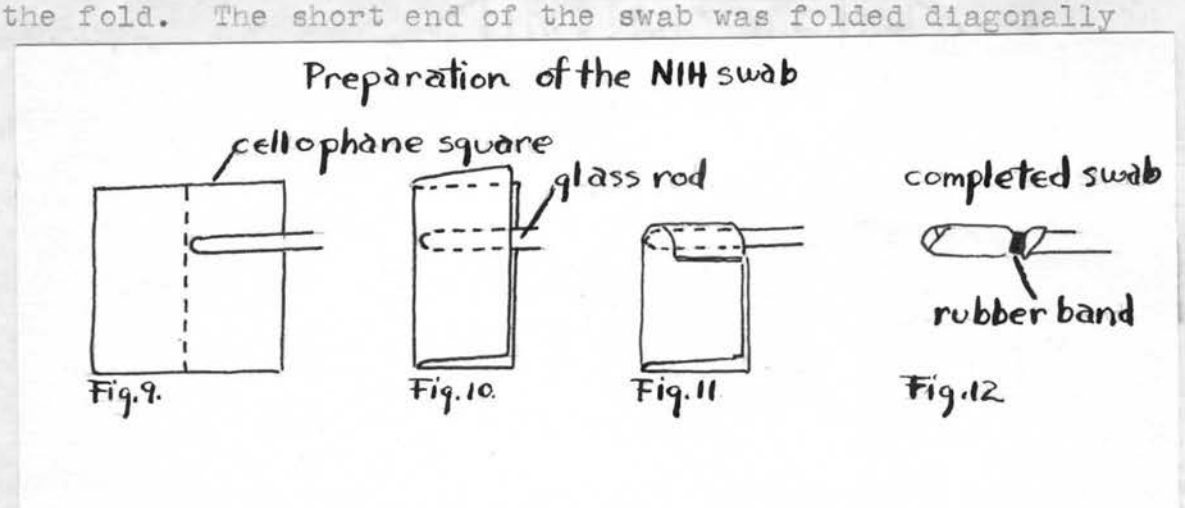
Use of rigid scraping devices has been described by a number of authors. Serbinow and Schulmann (1927) utilized a pointed spatula and a shaped match moistened with a 0.5 to 1 per cent solution of sodium hydroxide, which was lightly passed over the anal folds. After use the material was examined in sodium hydroxide or water. Bogajowlewski and Lewitzki (1929) used an ordinary match moistened with 1 per cent sodium hydroxide and the scrapings examined in the sodium hydroxide solution. When the scrapings were negative the edge of the slide as well as the match were used. Oleinikov (1929) stated that eggs did not adhere too well to a metal spatula but did adhere to a wooden one. Headlee (1935) made anal scrapings with a blunt instrument and examined the material in a drop of $\frac{1}{10}$ normal sodium hydroxide solution on a slide. For storage

after drying and in cases where longer storage of slides was necessary water was added.

Celluloid scrapers were tried by Hall (1937). Pieces of yellow celluloid were cut by scissors, rounded and the edge beveled on a whetstone. These scrapers were 4.5 centimeters long and 1 centimeter wide. When in use this celluloid scraper was held by a locking forceps. The scrapings were put in a vial and examined in a $\frac{1}{10}$ normal sodium hydroxide solution. The scraper was examined directly due to its transparency. However, this type of egg collector failed to transport eggs because of immersion in the vial.

E. Cellophane Swabs

A cellophane swab was made by taking a three centimeter square of the material and folding it in half to form two rectangles of equal size. A glass rod, 8 to 20 centimeters by 4 millimeters, was placed between the two rectangles perpendicularly to the fold about a third of the way from one end of the fold. The short end of the swab was folded diagonally



over the rod and the long end folded diagonally over the short end. A rubber band was used to fasten the swab to the rod, and the rod was pushed through a test tube stopper. The swab was used dry and worked as a swab and a scraper.

To make an examination for the presence of ova, the cellophane was pushed down from the glass rod and cut off with a pair of fine scissors. The cellophane was then flattened out with the rod and a pair of forceps, two drops of sodium hydroxide were added and the preparation covered with a coverglass. Eggs were readily visible through the transparent cellophane. No manipulation is needed to separate the eggs from this type of swab, a procedure necessitated by other types of swabs.

This type of swab was given extensive tests and found to be satisfactory. To eliminate possible laboratory contamination during cutting of the cellophane tip, a cellophane square was used which was smaller than the coverglass, thus eliminating the necessity of cutting the cellophane tip. This last type of cellophane swab meets all the theoretical requirements of a good anal swab. It picks up eggs dependably and faithfully delivers the entire egg mass to the slide with little manipulation. No effort is needed for separating the eggs from the swab. The swab is easily transported without loss of eggs, is safe for those handling it, and causes little discomfort to the patient. This cellophane swab was given the name the NIH (National Institute of Health) swab.

After the cellophane is removed the rod and stopper is

returned to the test tube and the forceps sterilized by heating. The test tube arrangement is subjected to dry heat to distort and destroy and eggs present and to prevent contamination upon subsequent reuse of the system.

Tenth normal sodium hydroxide is used to saponify fat in the adhered fecal particles and thereby to set the eggs free.

Several anal swabs are necessary to establish negativeness. Serbinow and Schulmann (1927) noted that Skrjabin reported in 1925 the rise in the number of positives as follows: first swab, 42 per cent positive; second swab, 75 per cent; third swab, 84.6 per cent; fourth swab, 94.4 per cent.

Hall (1937) stated that the number of ova present on a swab is not an indication of either the number of worms present or the seriousness of infection. Personal cleanliness may greatly cut down on the number of ova present perianaly.

The NIH swab was found by Sontolongo and Goldberg (1941) to be useful in demonstrating infections of Trichuris trichiura, (L.) Taenia saginata, (Goeze) Hymenolepis nana, (von Siebold) and Ascaris lumbricoides, (L.) as well as those of Enterobius vermicularis. Of 14 cases of Trichuris trichiura, 11 were diagnosed with the use of the NIH swab; 4 of 6 cases of Taenia saginata; 2 of 3 cases of Hymenolepis nana; and 2 of 4 cases of Ascaris were all diagnosed with the exclusive use of the NIH swab.

The use of multiple swabs is a marked feature of testing with the NIH technique for the presence of pinworm ova. Mazzotti and Quintanar (1943) stress the importance of repeated

examinations to disclose the cases with mild infection.

Sawitz and Karpinos (1942) presentedan entirely statistical approach to the problem of multiple swab testing with the NIH swab.

SYMBOLS USED:

N = total number of children examined

Ni = " " " " infected

p = probability of obtaining a positive test on a child using a single NIH swab.

q = (1 - p) = probability of failure to obtain a positive test on a child using a single NIH swab.

n = Number of NIH swabs made

E_T = true efficiency of NIH swab technique

E_O = observed " " " " "

R_T = true prevelancy rates

R_O = observed " "

This work was done in conjunction with examination of six children's homes in New Orleans. Seven NIH swabs were made to establish negativeness, in regard to infection with pinworms. With this data secured the following equations were developed and the proper relationship shown.

N_1P = number of positives disclosed by first NIH examination

Ni - Nip
or
Ni (1-p) = number of positives still undisclosed after the first swab

Ni (1-p)p
or
Ni ($p-p^2$) = number of positives the second NIH swab will disclose

$Ni(1-p) - Ni(p-p^2)$

or

$Ni(1-p)^2$ = number of positives still undisclosed after two NIH swabs therefore

$Ni(1-p)^n$ = number of positives remaining after "n" examinations

total number of individuals found positive after "n" examinations = $Ni - Ni(1-p)^n = Ni\ \ 1-1(1-p)^2$

Observed efficiency - $Eo = \frac{Ni\ p}{Ni\ 1-1(1-p)^n} = \frac{p}{1-(1-p)^n}$

True efficiency - $E_T = \frac{Ni\ p}{Ni}$ number of infectees disclosed after first swab

or

$E_T = p$

From this it can be seen that in the efficiency equations above Eo E_T because the number of infectees disclosed usually is less than the actual number infected.

True prevalance rate - $RT = \frac{100\ Ni}{N}$

Observed prevalence rate - $Ro = \frac{100\ Ni\ \ 1-1(1-p)^n}{N}$

$\frac{100\ Ni}{N} \times N$

$\frac{R_T}{Ro} = \frac{100\ Ni\ \ 1-1(1-p)^n}{N} = \frac{100\ Ni}{100\ Ni\ \ 1-(1-p)^n}$

$\frac{R_T}{Ro} = \frac{1}{1-1(1-p)^n}$

$R_T = \frac{Ro}{1(1-p)^n}$

The greater the efficiency of the testing generally the smaller are the number of tests required to carry out the

examination. When p= 0.2, 7 NIH swabs disclose 97 per cent of the infections; when p=0.5, 7 NIH swabs disclose 99 per cent of the infections. However, when p-0.04 even 15 NIH tests disclose less than 46 per cent of the actual number of infected cases.

Sawitz suggests that differences in efficiency may be due to differences in intensity of infection, use of the swab by different persons, and in the time of day at which the swab was made.

A permanent mount can be made for teaching use of microscope slides showing high counts of pinworm ova by a rather simple method proposed by Rhuhe (1940). The cellophane square, after being examined in saline, is lifted out by forceps and inverted in a drop of 0.5 per cent phenolic glycerine jelly upon a slide. A second drop of jelly is added to the upper surface of the cellophane square and a coverglass is then inverted upon it. Further sealing of the preparation is made with model airplane glue.

Folan (1939) gives specifications of the NIH anal swab and also a method of preparation and cleaning of the swab.

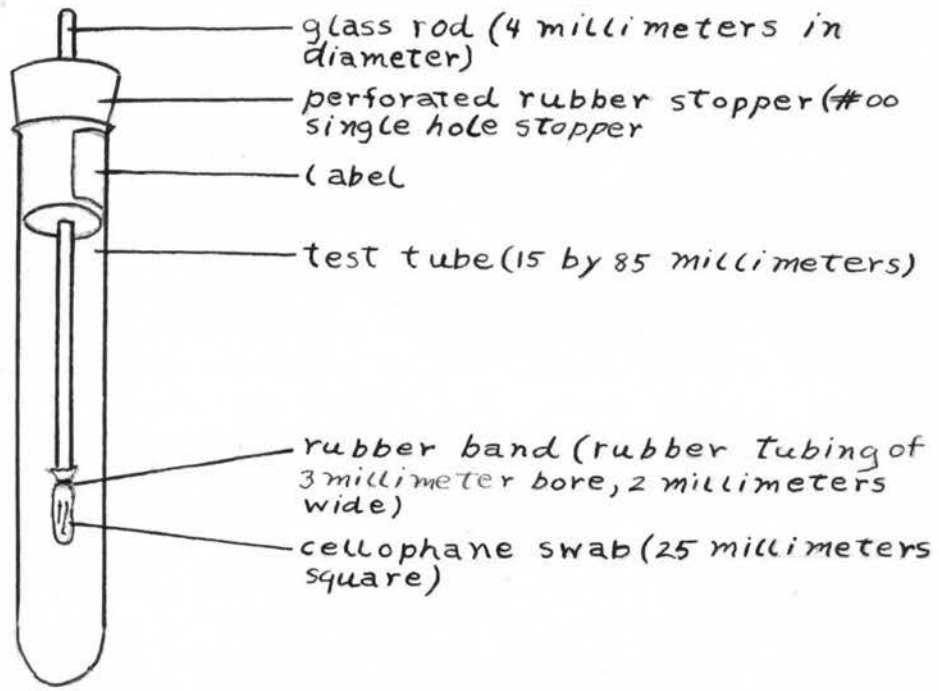


Fig. 13. NIH swab. (After Folan)

In using the NIH swab Folan suggested that the cellophane tip be stroked with an upward motion over the perianal region and across the anal opening. Morning examination is preferred in order to pick up any ova deposited during the night by gravid pinworms.

Careful cleansing of the anal swab and its housing is necessary to prevent any infectious material contaminating further anal swab examinations and thereby leading to faulty diagnosis. Ten per cent trisodium phosphate solution distorts the outer layer of the shell making it easily recognizable from untreated ova. In addition this cleaning solution makes possible the cleaning of that part of the glass.rod in the

rubber stopper by loosening the stopper.

The swabs and tubes immersed from 24 to 48 hours in two containers of three liter capacity.

After the sodium phosphate is poured off, the swabs are placed on a flat tray and autoclaved for 15 minutes under 15 pounds of pressure. After cooling, the rubber band is removed by forceps and the swabs are placed under running water for 20 minutes. The cleaning of the housing is the same as that of the swab proper. After autoclaving, the test tubes are washed in liquid soap and water, rinsed several times, and allowed to drain off.

There are several modified anal swabs, the evolution of which can be traced back to the prototype, the NIH. Graham (1941) used a loop

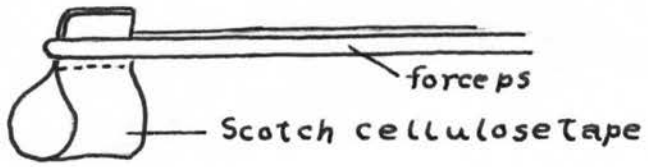


Fig. 14. Graham swab. (After Graham.)

of Scotch cellulose tape of half inch width and 8 centimeters long, folded down for about a centimeter at each end, adhesive surfaces together, to form two grips for handling. When using, the cellulose ribbon is held in forceps in a loop, adhesive surface out, and is patted down on the perianal skin. The adhesive surface picks up epithethial scales, fecal particles

and ova, if present. The tape is then placed lengthwise on a microscope slide, smoothed down with the forceps and examined with a 16 millimeter objective, as any other coverglass preparation. Three hundred and sixty inches of ½ inch scotch cellulose tape costs twenty-five cents (pre-war prices) and the cost of one swab is about ¼ of a cent. The swab can be made in ten seconds and the time required to examine it is about the same needed to examine ova on a fecal film. Graham believes his scotch tape swab meets Hall's (1937) requirements for a good swab, since it picks up a fair number of ova when present, deposits them on slide dependably and without much difficulty in manipulation, is easily transported and safe to handle, and causes little inconvenience to the patient.

Mazzotti (1945) in an examination of 2,989 cases, reports that the Graham swab, in addition to revealing the expected ova of pinworm, also discloses the presence of Ascaris, Trichuris, hookworm, and tapeworm ova.

The Frosst swab, developed by Charles E. Frosst & Co. of Montreal, is mentioned in a paper of Kuitunen - Ekbaum in discussing diagnosis of enterobiasis. This investigator cast considerable doubt on the value of the Frosst swab, in that the cellophane easily drops off the applicator before, during and after swabbing, and cannot be easily transported without the danger or losing material or contaminating the person handling the swab. The Frosst swab consists

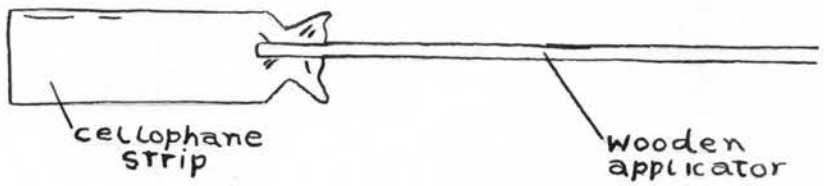


Fig. 15. Frosst swab. (After Kuitunen-Ehbaum (1942).)

of a cellophane strip an inch long and a ½ inch wide which is glued to the end of an applicator. The instructions for its use are as follows:

(1.) Patient is examined in the morning before bathing.

(2.) The anal and perianal region is stroked with the cellophane.

(3.) The cellophane is placed on a microscope slide having a drop of water.

(4.) Cover with a cover slip and examine.

The Sisk swab (1943) is a fairly simple affair, consisting of a rectangular piece of cellophane, an inch by an inch and a half, glued on to a wooden applicator. The cellophane swab is patted

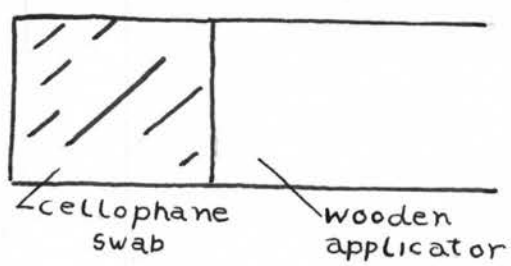


Fig. 16. Sisk swab.

against the perianal skin, the swab is cut loose and examined microscopically.

Jacobs (1942) tried a new type of Scotch tape technique. Using a wooden tongue depressor as the applicator, a piece of Scotch tape about the length of a microscope slide is attached adhesive end up over one end of the tongue blade, held in place, and applied along the perianal folds. The adhesive surface is then placed down on a slide and examined microscopically.

Von Hofe (1944) devised another type swab which utilized a test tube as an applicator. Scotch tape is cut into strips about 2.5 inches by 3/8 of an inch. The scotch tape is held over the closed end of a test tube with the index finger and the thumb with the adhesive surface of the tape exposed. This is applied to the anus and is rocked back and forth several times to obtain a smear. The cellophane Scotch tape is then placed on a glass slide, pressed down on the slide with the closed end of the test tube and examined microscopically. A

modification of the Von Hofe swab was the type used in connection with the original incidence work which was carried on in conjunction with this thesis. A later description will be given.

Differing from the approved cellophane swabs the glass club as devised by Schuffner and Swellengrebel (1943) does follow the general principle of perianal testing. The glass club consists of a glass tube 10 centimeters long with a ground spherical enlargement - 1.8 centimeters in diameter. It is wet and applied perianally, the egg suspension is transferred from the club to a slide by means of contact and examined immediately.

4. Comparison of the Efficiency of the Various Physical Diagnostic Techniques

Fecal Examinations

D'Antoni and Sawitz (1940) reported that direct fecal films detected less than 1 per cent of the true incidence of infection and concentration techniques, less than 25 per cent. Wright and Cram (1937) stated that fecal examinations detect 1 in 7 actual cases of pinworm infestation. The ineffectualness of fecal examinations in cases of enterobiasis is further noted by Craig and Faust (1937) who stated that pinworm ova are present in the feces in not over 5 per cent of infected persons. Serbinow and Schulmann (1927) pointed out the inadequacies of fecal examinations and recommended instead the examination of

anal scrapings.

The Cellophane Swabs

Faust, Sawitz, Tobie, Peres and Linicicome (1939) examined 189 fecal specimens via fecal concentration techniques and found 16 to be positive for Enterobius. They further stated that the NIH swab detects more than five times as many

anal scrapings.

The Cellophane Swabs

Faust, Sawitz, Tobie, Peres and Linicicome (1939) examined 189 fecal specimens via fecal concentration techniques and found 16 to be positive for Enterobius. They further stated that the NIH swab detects more than five times as many cases as can be detected by the best of the fecal flotation methods, the zinc sulphate centrifugal method.

Sotolongo and Goldberg (1941) compared the results of the NIH swab and fecal examinations. They presented 19 cases, aged 5 months to 39 years of age, all infected with pinworms. Fecal examinations disclosed 10 positives while the NIH swab showed the entire 19 cases to be positive.

Sawitz, Odom and Lincicome (1939) examined 136 institutionalized children using brine and zinc sulphate flotation methods and the NIH cellophane swab. The 131 children found to be positive using the three techniques served as the basis for testing the efficiency of these three techniques separately. The brine centrifugal flotation method showed the presence of 18 positives out of the 131 examined (13.7 per cent). Nineteen additional stools were examined giving 4 more positives. The zinc sulphate centrifugal flotation gave 23 positives, 14 were positive and 9 negative with the brine method. Sixteen additional stools were examined and 3 more positives were found. The anal swab results are summarized as follows:

The first swab examination gave 94 positives (71.8 per cent)
" second " " " 13 more positives (among the 37 negatives remaining)
The third swab examination gave 7 more positives (among the 24 negatives remaining)
The fourth swab examination gave 1 more positive (among the 17 negatives remaining)
The fifth swab examination gave 6 more positives (among the 16 negatives remaining)
The sixth swab examination gave 4 more positives (among the 10 negatives remaining)
The seventh swab examination gave 1 more positive (among the 5 negatives remaining)

Of the 131 children examined 127 or 96.9 per cent were found to be positive by the NIH swab method. The aforementioned figures along with further work on the diagnosis of Enterobius vermicularis by Schmidt in 1914, Skrajabin in 1925 and many other investigators shows that perianal testing is vastly superior to fecal flotation and concentration techniques. It has been shown that the NIH swab method yields its best results when repeated swab examinations up to seven times are performed. Beyond this number of examinations the number of negatives remains constant and no further positives are disclosed.

D'Antoni and Sawitz (1940) report that using 7 successive NIH swabs more than 99 per cent of the positive cases examined are detected.

A comparison between the two commonest types of anal

swabs, the Graham and the NIH swab, is available from two investigators. Kuitunen-Ekbaum (1942) reports that under clinical testing the NIH anal swab proved superior to the Graham swab for general use. The reasons he sets forth are (1), the NIH swab is firmer and (2), it picks up more eggs.

Mazzotti (1942) argues for the superiority of the Graham over Hall's NIH swab because of the simplicity of materials required and a greater accuracy in determination of actual infections.

With my limited experience in the use of the Graham and the NIH swab, I agree with Kuitunen-Ekbaum that the Graham swab is not firm enough for efficient and safe handling. However, I agree with Mazzotti that the Graham swab requires far simpler materials and procedures. It should be pointed out that both swabs are being used with a slight edge in popularity going to the Hall NIH swab.

Schuffner and Swellengrebel (1943) believe their glass club technique to be superior to the American NIH method, because of the larger number of ova picked up and seen and also the fact that an examination of a slide takes one-third as long as the examination of a cellophane swab slide.

5. Problems Encountered in the Diagnosis of Pinworm Infections

Artifacts which, according to Reardon (1938), greatly simulate the ova of Enterobius, are frequently seen in the observation of NIH or other cellophane swabs. These artifacts are not superficial, but are imbedded in the cellophane and

therefore cannot be dislodged by surface scratching. The artifacts are generally of a definite hyaline outline, usually ovoid, sometimes rounded, containing a dark amorphous mass. The resemblance, however, is only superficial. With slight experience they are easily differentiated and cause no further difficulty in identification. Among variable factors in diagnosis Cram (1939) mentioned the time of day at which pinworm migration takes place, the time the swab is made, the differences in exact location of ova by the pinworms, irregularity of worm migrations which may lead to the failure in picking up ova and classifying a large number of "infectees" as negative. To this might be added the human inaccuracies which may creep into the work, such as faulty handling of the swab, hasty examination of a slide (important in cases where only a few ova are picked up) and many other human failings which only add to the possibility of erroneous diagnosis.

B. Serological Methods of Diagnosis

Cienzynski (1928) experimented to determine whether cutaneous reactions are specific to Ascaris lumbricoides, Ascaris suum (Goeze), Trichuris trichura and Enterobius vermicularis. In order to discover specificity for enterobiasis, Cienzynski prepared two extracts (1) physiological salt solution of Enterobius and (2) alcoholic extract of Enterobius. Cienzynski injected these two extracts (.1cc doses) of Enterobius into six children. These six children were infected with Enterobius and Ascaris. Two of the children were likewise

infected with Trichocephalus. All but two of the children evidenced more or less intense cutaneous reactions, with reacted skin areas 1 to 4 centimeters in diameter. The reactions between alcoholic and physiological saline extracts did not differ appreciably. The extracts were found to contain ill-defined toxins. No specific reactions were observed therefore the testing was of no diagnostic value.

In a later work, Ciezynski (1928) prepared two additional antigenic extracts of Enterobius making a total of four which were used, (1) physiological salt solution of Enterobius, (2) alcoholic extract, (3) ether extract and (4) the use of the bodily fluids of Enterobius. His results were the same as those which occured in the earlier experiment. He found that the skin reactions to these antigens were not specific, parasitized and non-parasitized subjects reacting equally.

Schunfeld (1937) tested the hypersensitiveness of human skin to extracts of swine and human Ascaris. Immediate reactions and lesions occur as a late reaction, among them erythema migrans (redness). Repeated injections sensitized the skin so that 15 per cent of the patients went into anaphylactic shock. During the first hours the eosinophils of the peripheral blood increased 23 per cent. The reaction is a group reaction for Ascaris, Enterobius and Trichocephalus and persists even after the worms are gone.

Dermal and intradermal skin reactions in enterobiasis were observed by Wright and Bozicevich (1937). The antigen was prepared by washing the pinworms, drying them in vacuo

over sulphuric acid, grinding them into a dry powder, extracting the powder with ether, and further extracting using physiological saline or buffered saline solution. Dermal tests were made using a dilution of 1:100, and intra dermal tests with antigen dilutions of 1:500, 1:1000, 1:2000, 1:5000, 1:8000 and 1:10,000. Of 140 persons tested, 90 were found infected with pinworms, and 29 having helminth infections other than Enterobius were used as controls. A wheal formation 5 millimeters or more in diameter, with or without pseudopodia, with a negative control wheal, was considered to be a positive reaction for this pinworm antigen test. Of the 90 known cases of enterobiasis, 37 were shown to be positive for the dermal (1:100) test, 5 positive for the intradermal test using the 1:500 dilution, 2 positive for the 1:1000 antigen dilution, 1 to be positive with the 1:5000 antigen dilution. Eight negatives individuals of the dermal test were not intradermaly tested. Four pinworm cases were negative to both tests. The data presented shows the marked specificity of the dermal test and the unreliability of intradermal tests using antigen dilutions of 1:5000 or more. Frequently these 1:5000 antigen dilutions established positiveness in individuals infected with one or more species of worms, but not pinworms.

IV.

INCIDENCE OF ENTEROBIUS VERMICULARIS IN NORTH AMERICA

In previous discussions of the incidence and distribution of Enterobius vermicularis, it was noted that this helminth is found over the entire world among all races of men and knowing no national or geographic barriers. However, this study is restricted to the North American continent, which has been investigated as to incidence much more thoroughly than any other land mass. For purposes of convenience, the continent has been subdivided into five particular areas - Central America, West Indies, Canada, Alaska and the Aleutian Islands, and the United States.

A. Central American and Mexico

Sandground (1933) has published the only incidence report available on infections with pinworms in Yucatan, Central America. He noted that all persons who were given anthelmentic medications had pinworms present in the passed stools. For this reason Enterobius is believed to be universally present in the people of this area, probably because of the low hygienic levels encountered in the populace.

In Mexico we have more recent data concerning pinworm infestation. If we are to judge from the few scattered reports on hand the infection rates with pinworms run as high as those

encountered by Sandground in Yucatan.

Osorio, Teresa and Mazzotti (1940) examined children of three institutions in Mexico City with a single NIH swab and found that of the 684 children tested 51.6 per cent were infected with pinworms. Quite a contradictory report is noted from Galindez and Magdalena (1944) who reported 2 per cent of 600 patients studied to be positive for Enterobius. The diagnostic method employed was stool examination, which, as has been mentioned earlier, reveals only a small fraction of the actual number of infectees.

Mazzotti and Quintanar (1943) examined 1,551 children in Mexico City, 2-13 years of age. The first examination showed 41.8 per cent positive. They carried on repeated examinations of one group of 100 children which indicated that the infection rate might approach 95 per cent. The method employed was the Graham scotch tape technique, which they found to disclose a greater number of the infected cases than did the NIH swab.

Quintanar (1943) made examinations for Enterobius vermicularis ova on the external genitilia of 109 institutionalized girls in Mexico City. The Graham technique was used and 63.3 per cent of the girls were found to be infected.

B. West Indies

From the West Indies, the incidence reports are even sketchier than those reported from the Central American area. Two reports are available; one from Puerto Rico the other

Havana, Cuba.

Faust, Hoffman, Jones and Janer (1934) in a survey of intestinal parasites in endemic schistosomiasis areas of Puerto Rico, found in a single fecal examination of 1,003 persons in Trujillo Alto and environs 0.4 per cent to be infected with Enterobius vermicularis.

Sotolongo and Goldberger (1941) employed endoscopic examination methods carried out in 109 hospitalized cases in Havana. Enterobius was reported in 19 cases (17.4 per cent).

C. Canada

All of the published pinworm incidence reports from Canada were made in Toronto by Kuitunen-Ekbaum. In 1940, 843 children were examined by use of the NIH swab and 48 per cent were found to be infected with pinworms. An average of only 2.6 swabs was made per child, indicating a probably incidence of well over 50 per cent. Three years later (1943) 300 children and 56 adults were examined for enterobiasis. Sixty per cent of the children and 52 per cent of the adults were infected with pinworms.

D. Alaska and the Aleutian Area

Ashburn (1941) furnished us with the only information available as to the incidence of enterobiasis in the Alaska-Aleutian area, a series of 2,317 surgically removed appendices taken in United States government hospitals throughout the country and territories was made. The highest rate of

appendiceal enterobiasis was noted among the eskimos of the far North-23.91 per cent. This would seem to indicate that pinworm infection rates in the Arctic regions to be at least as high as those found elsewhere on the continent and also serves to indicate that enterobiasis is definitely not confined to tropical, semi-tropical or temperate regions.

E. United States

The United States has the most complete picture on incidence of enterobiasis of any other North American country. But even this is far from complete since no reports are available from the Pacific coast and South-Western areas of the country.

Mauss (1945) studied a series of 315 school and preschool children from Rapid City, South Dakota who were examined by use of the Jacob's swab. A single examination was performed and 124 children were found to be positive for pinworm ova.

Headlee has been responsible for the tabulation of incidence throughout the state of Indiana and Illinois. In 1935, 282 persons at the State Hospital at Kankakee, Illinois, were examined by studying anal scrapings microscopically, and 62 or 22 per cent were positive for Enterobius vermicularis. Headlee (1942) using smear and centrifuge techniques found the following:

TABLE II

Headlee's Survey on the Incidence of Enterobius in Indiana

Number of persons examined	Place samples were taken from	Percentage incidence of Enterobiasis
1,200	Logansport State Hospital	1.6
771	Evansville State Hospital	7.4
12	Terre Haute	0.0
87	Montgomery County	0.0
98	Warrick & Pike Counties	4.1
63	(Not given)	1.6
147	Purdue Students	0.0
319	Indiana University Medical Center Hospital	6.9
Total 2,697		percentage avg. of incidence 2.7

(Method utilizing perianal scrapings showed:)

80 (1948)	Riley Hospital	13.75

There is no doubt that if repeated anal swabs, either NIH or Graham, were made the incidence would have been much greater. If this had been done, the state-wide samplings would have greater meaning.

Faust (1936) examined 4,270 ambulatory, white clinic patients of New Orleans. Stool examination via salt concentrations was used and 2.4 per cent were found to be infected with Enterobius. Two-thirds of the diagnoses were based on the presence of worms in the passed stools. In autopsy

examinations of accident cases, Faust (1941) found Enterobius in one case out of the 202 examined.

Sawitz (1939), employing the far more efficient NIH swab technique, has given the following incidence report from a New Orleans' orphanage, in which 96.9 per cent of 131 institutionalized children examined were found to be positive for pinworms. Up to 7 swabs were given before a patient was admitted to be negative. A year later (1940) Sawitz, in a study of 491 institutionalized children examined by use of the NIH swab, observed an incidence of 74.3 per cent.

Smith, Gill and McAlpine (1939) examined 637 white, institutionalized individuals in a survey of intestinal parasites in Alabama and found 65.15 per cent to be infected with pinworms.

In an extensive survey of fity-six countries in Florida, Leathers, Keller and McPhaul (1939) examined 29,064 fecal specimens. Of these 11 or 0.04 per cent indicated the presence of Enterobius vermicularis. This report once more demonstrates the inadequacies of fecal examination in testing for the presence of pinworms.

Keller, Leathers and Knox (1938) in a state-wide survey of 37,346 persons in the state of Georgia, diagnosis being based on stool examinations, found 0.7 per cent infected with pinworms. Byrd (1936) collected fecal samples from 257 college freshmen; the stools being preserved in formalin. Enterobius vermicularis was accounted for in only 1 case (0.38 per cent). Byrd (1936) in single fecal examinations on 537 individuals

on the relief rolls in the city of Athens, Georgia, found 0.186 per cent positive for Enterobius vermicularis. Reardon (1941) in an examination of 160 females of a Georgia state institution by use of the NIH swab found 53 per cent to be harboring pinworms.

Tennessee incidence reports of Enterobius vermicularis were all made on the basis of single fecal specimens and therefore are far from accurate. Rickard and Kerr (1926) reported 0.8 per cent of 1,491 individuals positive for pinworms. Canning (1933) in a sanitary survey of the Knox county Industrial School in Tennessee reported Enterobius vermicularis to be present among the inmates, but without giving any incidence figures.

Burrows (1943) reports that of 1,383 patients of the South Carolina State Hospital 4.73 per cent were infected with pinworms. Diagnosis was based on a single NIH swab. Undoubtedly multiple swabs would have given a much higher incidence.

Brown, Sheldon and Thurston (1940) employing a single NIH swab on several groups of persons found the following:

TABLE III

Brown, Sheldon and Thurston's Survey

on the Incidence of Enterobius in North Carolina

Number and character of group	Place where swabs were made	Percents of incidence
132 underpriviledged white boys	Charlotte, No. Carolina	32.8
67 white hospital patients	No. Carolina State Hospital	7.4
33 negro hospital patients	No. Carolina State Hospital	0.0
118 male students	University of North Carolina	0.0
350 Total	average incidence rate	13.7

The District of Columbia presents the most complete and thorough incidence work on Enterobius vermicularis of any other comparable size and population.

Bozicevich (1937) employing a single swab found 31 per cent of 230 boys in Washington, D.C. positive for pinworms. In 1938, Bozicevich and Brady in a study of 504 white boys in a boy's camp in the District of Columbia between the ages of 6 and 19 years of age found 57.3 per cent harboring pinworms. Jones (1941) in necropsy examination of 72 children at Washington, D.C. found 21 or 29.2 per cent infected with pinworms. Jacobs (1942) using a new scotch tape technique

examined 228 children and found 31.3 per cent positive for pinworm ova. Sisk (1943) employing a modified cellophane swab examined 62 children at the Mountain Orphanage and, after making two swabs, found 24 or 38 per cent infected with pinworms.

Cram and Folan (1939) examined 600 boys from 31 states, District of Columbia, and Puerto Rico recently arrived in Washington, D.C. It was shown that 4 NIH swabs doubled the efficiency of the diagnosis and that the Willis salt concentration method was 1/17 as effective as the NIH swab. The rate of infestation with pinworms was 12 per cent. Cram and Reardon (1939), examining 2,097 persons by up to 4 NIH swabs, found 861 or 41 per cent infected with pinworms.

Cram and Jones (1937), employing principally the NIH swab, studied 1,272 persons in Washington, D.C. One-half of the individuals (628) belonged to the general population. Of these 35.4 per cent were infected with pinworms. Of the remaining one-half belonging to an institutionalized group (644) 400 were examined at one time showing an incidence of 1.5 per cent; of the remaining 244, 217 showed an incidence of 7.8 per cent. The 27 girls examined showed no infection with pinworms. Cram and Nolan (1939) in examination of children in a private nursery school over an 18 month period found 58 (55 per cent) of the 106 children examined via NIH swab to be positive for pinworms.

Tansinsin (1930) in examination of 155 Pennsylvanian children found 3 (1.9 per cent) to be infected with Enterobius

vermicularis.

Weller and Sorenson (1941) examined 505 children between the ages of 2 to 12 by the use of the NIH swab method. The children were of two separate groups, the first being ward patients (118), and the remaining group medical outpatients (387). The swabs were taken between 9:00 A.M. and 5:00 P.M. Of 415 children examined by a single NIH swab, 74 or 18 per cent were found positive. In the selected group of 90 children two swabs were made and 23 or 21 per cent were found positive. Of the 118 ward patients, 25 or 21 per cent were positive; of the 387 outpatients, 72 or 18.6 per cent were positive. Among the 505 children examined 97 (19 per cent) were found positive for pinworm ova. Two reasons for the low incidence encountered in examination are, first, in most instances diagnosis was based on a single swab and, second, the times the examinations were made indicate that often they were made after the child had defecated thereby lessening the number of ova present in the perianal regions.

F. Summary on Incidence of Infection With Enterobius vermicularis in North America

I - North America

A - Central America

1. Yucatan - according to Sandground (1933) populace universally infected with Enterobius

2. Mexico - Osorio, Teresa and Mazzotti (1940)

684 examined--353 positive(51.6 per cent)

Mazzotti and Quintanar (1943)

1551 examined--------1480 positive (95 per cent)

Quintanar (1943)

109 examined-------- 69 positive (63 per cent)

Totals 2344 examined--------1902 positive (81.2 per cent)

B-West Indies

1. Puerto Rico - insufficient accurate data from which we can draw any sort of conclusion as to incidence

2. Cuba - Sontolongo and Goldberger (1941)

109 examined----19 positive (17.4 per cent)

Although evidence is lacking we can presume that the populace of the West Indies are at least moderately infected with Enterobius vermicularis.

C-Canada

1. Toronto - Kuitunen-Ekbaum (1940)

843 examined----405 positive (48 per cent)

Kuitunen-Ekbaum (1943)

356 examined----209 positive (58.8 per cent)

Totals 1199 examined----614 positive (51.2 per cent)

D-Alaska and Aluetians

According to Ashburn (1941) the highest rates of appendiceal enterobiasis occurs among the natives of this region so we may expect incidence rate to be high.

E, United States

1. Mid-West

a. South Dakota - Mauss (1945) - Single swab.

315 examined - 124 positive (39.4 per cent)

b. Illinois - Headlee (1935) - perianal scrapings

282 examined - 62 positive (22 per cent)

c. Indiana - Headlee (1942)

80 examined - 11 positive (13.75 per cent)

2. South-Eastern Area

a. Louisiana - Sawitz (1939)

131 examined - 127 positive (96.9 per cent)

Sawitz, D'Antoni, Rhude and Lob (1940)

491 examined - 365 positive (74.3 per cent)

b. Alabama - Smith, Gill and McAlpine (1939)

637 examined - 415 positive (65.15 per cent)

c. Georgia - Reardon (1941)

160 examined - 85 positive (53 per cent)

d. South Carolina - Burrows (1943)

1,383 examined - 66 positive (4.73 per cent)

e. North Carolina - Brown, Sheldon and Thurston (1940)

350 examined - 48 positive (13.7 per cent)

3. District of Columbia

Bozicevich (1937)

230 examined - 72 positive (31 per cent)

Bozicevich and Brady (1938)

504 examined - 290 positive (57.3 per cent)

Jones (1941)

72 examined - 21 positive (29.2 per cent)

Jacobs (1942)

228 examined - 72 positive (31.3 per cent)

Sisk (1943)

62 examined - 24 positive (38 per cent)

Cram and Foland (1939)

600 examined - 72 positive (12 per cent)

Cram and Reardon (1939)

2,097 examined - 861 positive (41 per cent)

Cram and Jones (1937)

1,272 examined - 245 positive (19.3 per cent)

Cram (1941)

1,525 examined - 859 positive (56.3 per cent)

Cram and Nolan (1939)

106 examined - 58 positive (55 per cent)

4. North-Eastern Area

a. Massachusetts - Weller and Sorenson (1941)

505 examined - 97 positive (19 per cent)

Totals 11,030 examined - 3,974 positive (36 per cent)

These figures are close to the probable incidence of Enterobius among the people of the Mid-Western and Eastern part of the United States. Since no data is present from the Western part of the nation, it might be a little preseumptuous to attempt a generalization of the incidence of Enterobius vermicularis in the United States. Yet it is probably true that it is the most common helminth infection in the United States, infecting people of all ages, races and socio-economic levels to a greater or lesser extent.

Epidemiology of Enterobius is an extremelly important

factor in a consideration of procedures to be followed in any course of treatment, and therefore should rightly be considered previous to a discussion of treatments of enterobiasis. Many incidence reports frequently contain data on age, sex and race, which are summarized below.

Our knowledge of Enterobius supports the view that very few infants under a year old are ever infected. Mazzotti and Quintanar (1943) found that of 500 children under the age of three years, 12 or 2.4 per cent were infected with pinworms, the youngest "infectee" being 7 months old. Kuitunen-Ekbaum (1943) found one child under 1 year of age positive for pinworms out of 300 children examined.

For the sake of convenience, the age groups after infancy are classified under three headings: preschool children, (1-5 years of age), school children (6-18 years of age), and adults. It can generally be said that there is a rapid rise in incidence as one goes from the pre-school children to the school children level.

TABLE IV

Incidence of Enterobius in Pre-School Children and School Children

Cram and Reardon (1939)	pre-school children	35 per cent
	school children	51 per cent
Cram and Nolan (1939)	2-3 years of age	43 per cent
	4-5 years of age	64 per cent
Weller and Sorenson (1941)	2-4 years of age	13 per cent
	5-9 years of age	29 per cent

According to Kuitunen-Ekbaum (1943) the 6 to 11 years old

period has the highest incidence.

Sawitz, D'Antoni, Rhude and Lob (1940) maintain that the age and incidence curve may be explained by means of direct contact. The upgrade in the 1 to 6 year group can be shown to be due to progressively greater contact with other children. From six years, contact is closer, the children do not wash as efficiently as in the older groups and the incidence runs the highest. The boys reach the personal cleanliness level at about the age of 15 and thereafter incidence rates are on the downgrade.

At adulthood, according to Cram and Reardon (1939), the incidence is about 22 per cent. Generally the older the age group the lower the incidence rates of pinworm infections. Mazzotti and Quintanar (1943) found that adults over the age of 60 to have an incidence rate of 5 per cent. The oldest person Mazzotti and Quintanar found to be infected with pinworms was 101 years of age.

A general age and incidence curve, according to the above information, would look something like this:

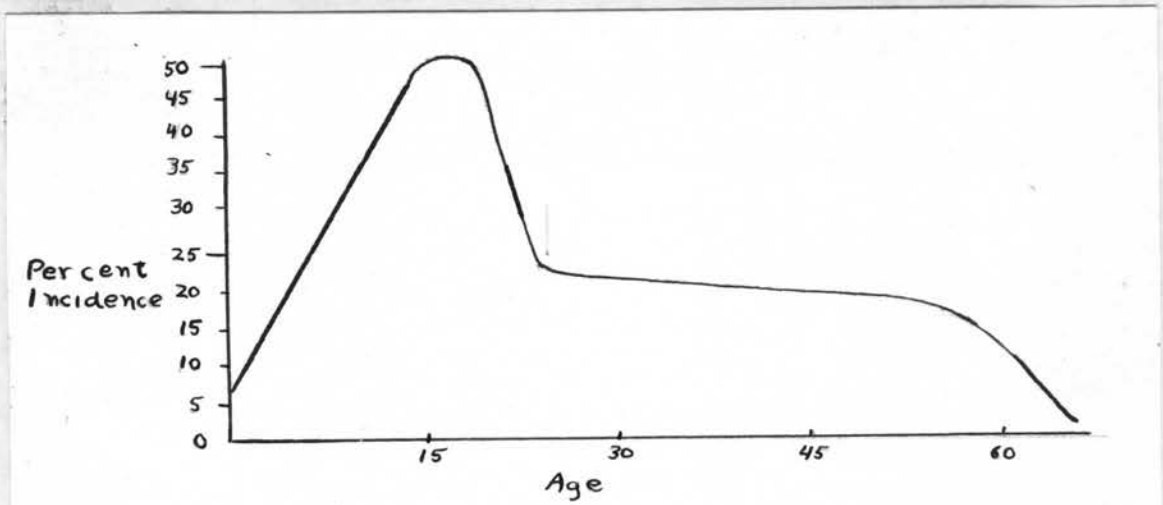


Fig. 17. Incidence and age graph.

Evidence as to which sex is the most heavily infested with pinworms is contradictory. Several workers in the field have indicated that females are more frequently infected by pinworms than are males. Generally it may be said that males have the higher incidence of enterobiasis than do females.

Burrows (1943), in a study of the intestinal parasites of mental patients, noted that with the one exception of Enterobius all helminths occured more frequently in females than in males. He also observed that of 343 negro females tested via the approved NIH swab not a single one was found to be positive for pinworms.

Sawitz, D'Antoni, Rhude and Lob (1940) maintained that generally girls had a lower incidence of infection than did boys expecially after the nine year level. Personal cleanliness with refraining from close contacts comes first to girls and results in a lowering of incidence around 9 years of age. As

has been mentioned before, boys reach this personal cleanliness level later at about 15 years of age.

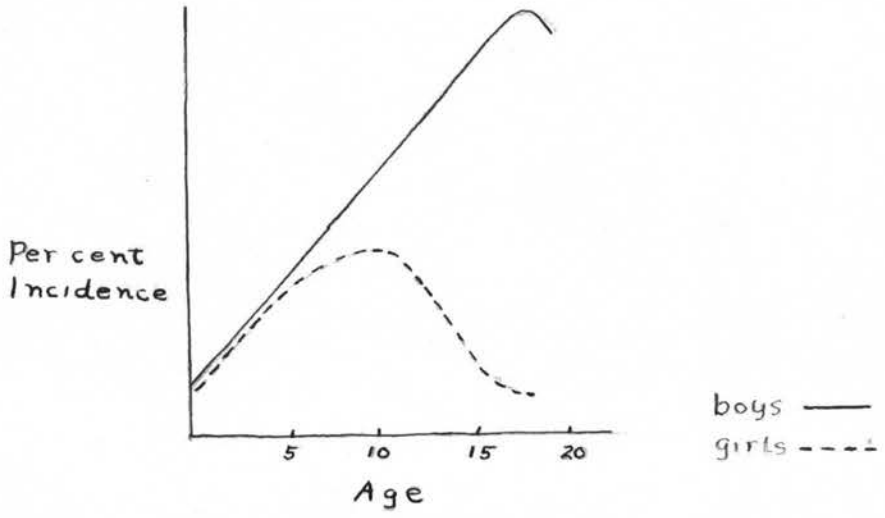


Fig. 18. Incidence and sex graph (based on above information.)

TABLE V

Incidence and Sex Chart

BOYS

	No.Exam.	No.+	% Incidence
Mauss (1945)	163	59	36.2
Brown (1941)	132	43	32.8
Sawitz, D'Antoni (1940)	341	303	88.8
Rhude, and Lob Bozicevich (1938)	504	290	57.3
Cram & Folan (1939)	600	72	12.0
Cram & Nolan (1939)	69	41	59.0
Total No. of boys	1,809	808	44.1

GIRLS

	No.Exam.	No. +	% Incidence
Mauss (1945)	153	65	42.8
Sawitz, D'Antoni (1940)	150	63	42.0
Rhude, and Lob Cram and Nolan (1939)	40	19	48.0
Total No. of girls	343	147	42.8

There occurs a very marked difference in incidence between races. The negro race has a much lower incidence than the white race. Due to some unknown and curious physiological factor, the negro, dispite his frequently low economic and hygienic level, is less frequently parasitized by Enterobius than his white bretheren.

As mentioned previously, Burrows (1943) failed to find a single case in 343 negro females examined. Brown (1940), while making a survey of intestinal nematodes in a hospitalized group, found 7.4 per cent of the 67 white patients positive for pinworms while not a single case could be found among 33 negro patients.

Sawitz, D'Antoni, Rhude and Lob (1940) found 302 (82.7 per cent) of 365 white children positive for pinworm ova, while of the 126 colored children examined 64 (50.6 per cent) were infected with pinworms.

Cram and Folan (1939) found that differences in incidence between negro and white were less pronounced in school children than in pre-school children and that an increase in the size of the sample showed the substantial difference in incidence rates between the white and negro race.

TABLE VI

WHITE		NEGRO	
Number Examined	Per Cent Infected	Number Examined	Per Cent Infected
476	41.2	90	21.1
2,582	41.9	789	15.5
2,895	41.5	1,099	12.9

Institutionalized individuals are more frequently parasitized by Enterobius vermicularis than are non-institutionalized persons. The chief reason for this seems to be the close proximity of a large number of confined persons which makes the presence of only a few persons harboring the parasite the starting point for an infection which will soon be acquired by the majority of the inmates.

Chandler (1944) states that general incidence seems to run between 33 to 50 per cent in children rising to 65 to 75 per cent in institutions.

Both Burrows (1943) and Headlee (1935) found that mentally deteriorated patients were generally more heavily infected than other types of institutionalized persons.

People of high socio-economic standings and superior hygienic levels are far from immune to enterobiasis. While low hygienic standards certainly are breeding places of pinworm infection, excellent hygienic facilities and high sanitary levels offer no insurance against its arrival in a household. Cram and Nolan (1939) in examination of a well equipped, white nursery over a period of eighteen months, found 55 per cent of the children, all from families of a high socio-economic level, positive for pinworm ova. The nature of pinworm infection is such that it is very easily acquired, once established quickly spread, and once spread difficult to eradicate.

The key to success in a particular course of treatment of infection with Enterobius vermicularis rests upon an important concept, namely, that infections with pinworms are not

isolated cases to be treated as such, but group and, more frequently, family situations. Kuitunen-Ekbaum (1943) found that of 34 families, examined by the use of the NIH swab, comprising 139 individuals, all or most of the members of 29 were infected, and in 26 of the families all the children were infected.

Cram (1941) in her classic approach to the problem of the familial nature of pinworm infections, set forth a study of 320 family series. Of this number, 286 were white and the remaining 34 families negro. A total of 5,539 swabs were made, 3.7 swabs being the average for a white person and 3.5 swabs for a colored person. This, of course, is an insufficient number of NIH swabs to detect all cases of the infection, but sufficient to indicate familial nature of pinworm infections and to indicate again the higher incidence of pinworm infection among members of the white race than in members of the negro race.

In the 286 white families, 1,353 individuals were examined, and of 860 children, 618 (72 per cent) were positive. Of 493 adults examined, 179 (36 per cent) were infected with pinworms. The incidence in white children was twice that of the adult whites. In white families, multiple cases occurred three times more frequently than did the single case families.

In the 34 negro families, 172 persons were examined. Of 114 children 58 or (51 per cent) were infected while of the 58 adults examined only 4 or (7 per cent) were positive for pinworms. The number of single case negro families slightly

exceeded the number of multiple case familes.

The white multiple case families averaged 3.4 infected persons. Of the 67 single case families, 63 were represented by a single child, the other four being adults. In one-half of the white families examined, all of the children were infected.

It is evident from these data that if one member of a family is found positive for pinworms there is a strong likelihood that other members may be infected, that most or all of the children may be infected, and that one or both of the parents may be infected.

V

TREATMENT OF ENTEROBIUS VERMICULARIS

A. Pathology

1. Symptoms Frequently Encountered in Cases of Enterobiasis

The pathology connected with infections of Enterobius vermicularis is not of a serious nature. The symptoms very often seems to be the common complaints encountered in children whether they are infected or not.

Craig and Faust (1937) stated that young male and female worms may cause local irritations where they reside, and in sensative people their byproducts may be absorbed producing characteristic helminth toxemia.

The most common symptom of pinworm infections is the familiar pruritis ani or anal itch. MacArthur (1930) reported that the first and the most important cause of nocturnal pruritis ani is the presence of Enterobius.

Miller and Einhorn (1944) listed a few of the symptoms associated with enterobiasis such as pruritis, vaginal irritations, local secondary infections, wakefullness at night, pain on urination, noctiuria and enuresis. Many of the nervous reactions frequently coupled with pinworm infections were noted - anorexia, irritability, lethargy, nail biting, thumb sucking, gratting of teeth, headaches, tics and so forth. Gastro-intestinal symptoms including abdominal pain, diarrhea,

nausea probably arising as a result of irritations of the intestinal mucosa were noticed.

Wright and Bozicevich (1938) on page 615 detailed an additional list of symptoms encountered in cases of pinworm infections.

> "Gastro-intestinal symptoms, including nausea and vomiting, partial anorexia, depraved appetite, some cases of appendicitis, cases of a low grade colitis and proctitis, and abdominal pain of undetermined origin, particularly post prandial pain which frequently clears up following successful treatment for the removal of pinworms. The migration of the gravid female worms results frequently in anal irritation as evidenced by pruritis, pain and a tickling sensation, dermal irritation and skin lesions in the perianal region."

"Genito-urinary symptoms include nocturnal and diurnal enuresis, masturbation, and leukorrhea and vulvo-vaginitis due to the migration of female worms into the female genital organs."

Reaction to the migrations of the adult female pinworms vary. Wright and Bozicevich (1938) related that Goebel (1922) believed that the degree of reaction to the presence of pinworms generally, and particularly to the migration of gravid females, is more often a measurement of and is correlated to the nervous temperment of the individual. The more nervous individuals exhibit marked reaction while the more stolid individuals show less reaction and complain of no discomfort.

Using a rectal speculum, Sontolongo and Goldberg (1941) discovered that decided changes in the rectum took place in cases of enterobiasis. The rectal speculum showed a swollen hyperemic mucosa with slight bleeding points in the anal canal

and internal sphincter areas. The mucosa was seen to be swollen and its vessels engorged with glazing mucous patches in evidence. In cases of long periods of infection, chronic proctitis was seen from the internal sphincter all the way up to the Houston valve; the mucosa in this area being uneven having alternating dense and thin areas. Granulations of various sizes were also seen in this area. They state that generally infestations with Enterobius are those of a proctitis hemorrhagica or hyperemica.

Brady and Wright (1939) in a symptomatology study of two hundred patients infected with pinworms present a slightly different view of enterobiasis and associated symptoms. They agreed with the previous writers that pinworm infections may be the cause of gastro-intestinal symptoms in some people, but denied the existance of any relationship between appendicitis or abdominal pain and enterobiasis. They maintained that symptoms of worm migrations varied between mild tickling and extreme pain. Pruritis which occured frequently in the cases under observation was believed to be caused by allergic reactions. Enuresis was found as frequently among infected persons as in non-infected ones. Pinworms entering the vulva of the female frequently brought vaginal mucoid discharges in infected cases. Fourteen of 45 pinworm infected girls were shown by a single vulval swab at the introitus to have pinworm ova at that particular site. They likewise found that "biting of nails, sucking of a thumb, picking at a nose and grating of teeth were not found more frequently in infected persons than

in control groups." Inability and difficulty in sleeping was observed. Various behavior characteristics such as lack of cooperation, inattention, a feeling of inferiority and shame was noted by Brady and Wright (1939) as frequently accompaning pinworm infections.

Jacobs (1942) in an examination of 228 children discovered of the positive patients, 41.3 per cent had abdominal pains, 49.4 per cent were nervous, 41.4 per cent were underweight, 38.6 per cent were poor sleepers, 38.6 per cent had anorexia, 21.3 per cent pruritis ani, and 17.3 per cent enuresis.

In summary of the common complaints suffered by those infected by pinworms, it is safe to say that they are diverse and of such a wide scope as to demand the interest and attention of the mother, clinician, or pediatrician.

Frequently in considering helminth infections of man attention is given to disturbances in blood counts and chemistry. Plotnikov and Zerchaninov (1929) noted that infections with pinworms caused but slight changes in blood counts, many times being characterized by low monocyte counts, and inconstant eosinophilia, while no effect was seen in erythrocytic, bymphocytic, neutrophilic and on the total leucocytic count. Gruber (1930) substantiated this finding when reporting that Enterobius vermicularis caused the smallest leucocyte change in man of the helminth species observed. Brady and Wright (1939) report a slight eosinopilia but no anemias with Enterobius. Tuckwiller (1940) believes anemia may be a symptom of pinworm infestation, and Wright and Bozicivich (1938) associate

slight anemias and esinophilia with pinworm infections. Brady and Wright (1939) confirmed other findings by noting hemoglobin levels in 200 patients infected with Enterobius. Infections with pinworms are therefore held to have little if any effect upon either blood chemistry or blood cell counts.

2. Pinworms and Appendicitis

The role of pinworms in the etiology of appendicitis has had a long and interesting history. Fabricius (1634) first described the presence of Oxyuris vermicularis within the lumen of the appendix. During the 19th century, many investigators noted in autopsies and necropsies that in cases of pinworm infections many times the favorite site of these worms was the appendix. In a series of 200 necropsies, Still (1899) discovered 38 cases (19 per cent) of enterobiasis. Of these 38 cases no fewer than 25 (65.8 per cent) had pinworms located in the appendix. In one case, 111 worms were found in the appendix, many of them immature. Nearly a half century later Jones (1941) confirmed the frequency of appendiceal enterobiasis in children by finding 21 out of 72 post mortem examinations of children in two hospitals in Washington. In these 21 cases, 18 had the appendix parasitized; the caecum and colon were both infected in 12 and the small intestine 6 times, predominantly by male pinworms and only present when other parts of the gastro-intestinal tract were infected.

Much has been written since 1900 concerning the relationship of the pinworm to appendicitis. The incidence of

appendiceal enterobiasis, as reported by various investigators varies between 0 to 100 per cent. Crill (1906) did not find a single case in examination of 1,000 appendices, whereas Fischer (1923) found 46 positive cases in 110 appendices (41.8 per cent).

Of 144 appendices examined by Solov'ev (1927) Enterobius was found to be present in 67 cases. A total of three hundred and thirty-three female and 59 male worms were found to be present in the 67 infected cases. Among the females from the appendix, 180 were gravid and 41 ready to oviposit. For this reason Solov'ev considers Enterobius to be important in the etiology of appendicitis even more so than Trichocephalus because of the more frequent presence of the former helminth in appendicitis cases.

In a report of a study on 1,000 surgically removed appendices Schenken (1942) gives us some valuable information on the techniques of diagnosis of cases of appendiceal enterobiasis. The first group of 600 of the surgically removed appendices had their complete appendiceal contents emulsified in water, and then centrifuged at 500 revolutions per minute for one minute. In group II, 400 of the appendices were treated in the same way as the first group except that centrifugation was carried on until the supernate liquid was clear. It was found that 23.3 per cent of the 1,000 appendices examined were positive for pinworms. The second method with more extensive centrifugation was regarded as the more accurate of the two employed.

Gordon (1933) in an examination of 26,051 appendices found

311 or 1.19 per cent to show the presence of Enterobius vermicularis. Lopez-Fernandez (1946) found Enterobius in 32 (36.2 per cent) of 88 cases of appendicitis.

Rector (1943) reports that there are two factors responsible for variation of incidence on appendiceal enterobiasis. The first factor probably being due to differences in personal habits, environmental conditions and climate. The second and significant factor is that incidence of pinworms is usually higher among children than adults. Cecil and Bulkley (1912) found pinworms in 15 per cent of 129 unselected appendices removed from children between the ages of 2 to 15. Botaford, Hudson and Chamberlain (1939) reported an incidence of 5.3 per cent in an examination of 1,343 appendixes among children aged 1.5 to 12 years of age. Many other reports are noted - varying between .007 per cent (Wax and Cooper, 1941) to 18.1 per cent (Harris and Brown, 1925).

Diligent and systematic observations of removed appendices constitute a very important variable factor in the discrepencies in incidence reported. Harris and Brown (1925) used probably the greatest diligence and care in observation of any in the literature. After examining the fecal contents grossly with a hand lens, they were diluted in saline, centrifuged, and the sediment examined microscopically for the presence of parasites and ova. Goodale's (1934) method of examination consisted of opening up the organ longitudinaly, after formalin fixation, and spreading the contents out on a board for gross examination. Best results are obtained when the examination of the appendix

is made both grossly and microscopically, however.

Rector's (1943) survey was made on 210 appendixes in which the lumen was partially or completely patent. The age range was from 15 to 69 years of age with a median age of 29 years and a mean age of 31.08 years. Of the 210 appendices examined, 160 were from females and 50 from males. Many of the removed appendixed (188) were incidentally removed.

The appendices were opened in a fresh state in the laboratory and examined grossly on a glass slide by the use of teasing needles. The mucosa was examined grossly and a portion of the fecal contents was examined microscopically. The microscopic examination disclosed the presence of the parasites in three cases, which had not been detected grossly. The parasites were mostly males, only one female being found. No other parasites or ova were found.

Of the 210 appendices examined grossly and microscopically, 10 or 4.76 per cent were found to be infected with Enterobius vermicularis. The age range of the ten positives was 15 to 33 with an average age of 22.7 years. Eight of the 10 infected appendixes belonged to females.

Rector's work caused him to believe that the most important factor for the variation in incidence reports is due to the thoroughness with which the appendiceal contents are examined. Rector's experience, coupled with the results of Botsford, Hudson and Chamberlain (1939) lead him to believe that the expected incidence of appendiceal enterobiasis among adults should yield an incidence of 5 per cent and for the children

the incidence should range between 15 to 20 per cent.

Kuitunen-Ekbaum (1942) in a study of 100 surgically removed appendices of children, 6 months to 14 years old, Enterobius was found in 34 per cent. No correlation between infestation and occurance of appendicitis was found. Ova freed by atrophy of adult worms showed no advance stage of development. Twenty-seven cases of pinworms were found among the 57 normal appendices examined with from 1 to 290 pinworms per case.

Out of 35 acute appendices, 5 were positive numbering from 1 to 20 pinworms per case. In 5 chronic appendices, only 1 housed pinworms (5). Of two acute and chronic appendices one contained a single pinworm.

No other parasites or their ova were found in this series of 100 surgically removed appendices. The presence of Enterobius vermicularis according to Kuitunen-Ekbaum does not have any direct connection to cases of appendicitis and should properly be regarded as an accidental intruder, not an etiological agent.

3. Unusual Pathological Cases

The pinworm may be at times not the harmless intestinal worm so far depicted. Severe damage to an organ or the host himself may occur when the pinworm gets into an environment in which by nature he does not belong and flourishes to such an extent that great injury is done. Craig and Faust (1937) stated the wandering females may occasionally migrate against

the peristaltic current, enter the small intestine and may reach the stomach, esophagus and nares. If fully embryomated eggs are deposited in the duodenum and upper jejunum they can hatch and cause reinfection. Cases have been reported where gravid females have migrated from the vulva region up the vagina and uterus into the Fallopian tubes encysted there or escape the duct via the ostium and become encysted into the lining of the peritoneal cavity. In the tubules they may produce symptoms of salpingitis which may be present and felt for years.

Still (1899) reports a case of a child who died of pleurisy and acute pericarditis and complained before death of severe abdominal pain in the right iliac fossa. Post mortem examination disclosed the presence of 111 pinworms mostly immature in the lower part of the vermiform appendix.

Puccinelli (1929) tells of a case in which intestinal tissue contained pinworms which suggested to him that pneumatosis may be brought on by intestinal parasites. Afeeva (1927) in an autopsy examination of a case of acute peritonitis found Ascaris in the peritoneal cavity. In the wall of the ileum was found a leiomyoma (smooth muscle tumor) having many passages. In one passage Enterobius was found; other passages housed Ascaris.

An exceptional case of enterobiasis of the intestinal wall was reported by Biljmer (1946). The case involved a man 46 years old who died after four days of hospitalization of extreme diarrhoea and progressive uraemia. Post mortem

examination disclosed gangrenous ulceration of the rectum and colon and some ulcers in the ileum, caused by the presence of Enterobius. Microscopic examinations were made of various parts of the small and large intestine. Practically all of these enteric portions revealed the presence of Enterobius vermicularis. The infection was approximately 60 pinworms per square centimeter of intestinal wall. The proportion of males to females was approximately 5-6 to 1. In most of the worms examined, the males appeared to be one or two larval stages ahead of the females. The males averaged 1.677 millimeters in length and the females about 1.550 millimeters. The relatively small size of the females is explained by the fact that they were less mature than the males and frequently showing scanty development of the genitalia.

The worms were only occasionally found within the ulcers but frequently surrounding the ulcerous areas, enclosed in a common sheathlike structure.

Three possibilities as to the etiology of this condition are (1) Enterobius caused the gangrenous ulceration (2) Enterobius penetrated a pre-existing ulcer during life, (3) Enterobius invaded the ulcers after death. The second possibility is the most likely of the three. Although it is possible that the pinworms normally present in the intestinal lumen could carry bacteria with them, the finding of tunnels leading to the ulcers indicates that the ulcers were already present and that due to the crumbling of the host's bodily defenses, the worms were able to gain access into the

intestinal mucosa.

Tens of thousands of pinworms were found in the intestinal wall, especially that of the rectum. All the worms were in their larval stages but the males were one or two stages ahead of the females. The host tissue seemed to have reacted to this invasion by enclosing the worms in a connective tissue sheath and thereby localizing them.

An interesting sidelight to these cases of abnormal habitats of pinworms is evidenced by the work of Hoeppli and Hsiang (1931), who wished to view the reactions of host tissue to pinworms. They prepared a chamber made up of two celluloid plates fixed together and inserted it into the ear of a rabbit. The transparent chamber was introduced in such a way that the central blood vessels and nerves passed through the chamber. The worm during the first two days was observed to make slow movements. Peristaltic uterine contractions and subsequent expulsion of eggs was observed even though degeneration was going on in the other parts of the worm. Sectioning revealed the fact that the worm was embedded in a large mass of leucocytes. Small areas of hemorrhage were also seen. The pinworm previous to insertion in the chamber had been kept in a dilute zysol solution ($\frac{1}{500}$) for three minutes.

B. Prognosis

Dispite these serious cases where the pinworm demonstrates a latent ability to do great harm under the proper circumstances, an infection with pinworms is generally not of a

serious nature. Undoubtedly millions of Americans are now carrying them with apparently no large scale discomfort. The prognosis with Enterobius is good, unless, according to Craig and Faust (1937) the disease becomes chronic and sepsis in the perianal region becomes serious.

C. Control of Adult Pinworms

1. Physical

De Rivas (1938) advocated the rational treatment of parasitic infections of the large intestine by use of an intracolonic thermal method. Among the parasites eliminated by thermal treatment is the pinworm. Hot solution (50-53°C) is introduced into the lumen of the entire colon from 5 to 10 minutes. The solution consists of a 1-5000 copper sulphate solution with one ounce of glycerine per liter of solution. One to two liters of solution admitted at the rate of 100 to 150 cc per minute give the desired colonic temperature (45° to 47°C).

Non-medicated enemas (soapsuds and saline) are believed by Wright, Brady and Bozicevich (1939) to be of some value in treating pinworm infections in children and infants. Treatment in order to have worth must be continued for three to four week period or even longer every other night. Lutz (1888) recommended cold water enemas and found that they worked equally as well as some of the more elaborate medicated preparations.

Purgatives, according to Wright and Cram (1937), either in single or repeated doses in the treatment of pinworm infections, do not seem to be of any value. They further stated that cotton anal plugs to prevent the migration of gravid female pinworms into the perianal region, so that the ova will not be deposited, may be of some value, but in themselves will not erradicate the infection.

2. Chemical

A. Anal Ointments

Wright, Brady and Bozicevich (1939) believed that medicated anal ointments seem to be of little or no value in treatment of oxyuriasis, that is, in destruction of the gravid female worms or preventing migration or even rendering the ova non-viable. Anal ointments, however, do seem to be of value in the treatment of pruritis brought about by irritations of the gravid female movements in the anal and rectal areas.

Hall (1923) stated that anal washes or ointments have been recommended to destroy worms and eggs, to prevent their contamination of bedding and clothing, to relieve itching and to cut down on the transfere of infectious materials from the hands to the mouth. Among the ointments and washes used are the official unguentum phenolis, 3 per cent phenol ointment, unguentum hydrargyri; 1:10,000 bichloride of mercury solution; silver nitrate ointment; camphor, thymol and quinine ointment; or a paradichlorobenzene ointment.

Fernan-Nunez (1927) reported that the most efficacious oral remedy against Enterobius proved to be the fresh sap of the tree Ficus laurifolia (L.). Oil of chenopodium was most effective against worms but did not expell them from the caecum. Intervenous injections of oil of chenopodium were most useful in difficult cases. With the use of this latter treatment a boy 19 years of age expelled 639 pinworms in a period of seven days.

B. Methylene Blue

Pakenham-Walsh, (1944) treated three cases of enterobiasis with methylene blue. One case resistant to phenothiazine was treated successfully with 3 grams T.D.S. for a week without ill effects to herself or the nursing baby.

C. Santonin

Wright, Brady and Bozicevich (1939) carried on experimentation with the use of santonin in treating twenty cases of enterobiasis. Each patient received a single dose of the drug daily for a ten day period. Dosage was based not only on the chronological age but also physical condition of the individual, the latter being frequently more important, since many of the children were undernourished and poorly developed and were therefore given relatively small doses. Thirteen of the cases were children between the ages of 5 to 14 and the remainder adults with the exception of one 17 years old. Dosages varied from $\frac{1}{2}$ grain per day for most children to 1 grain per day for

adults. After an interval of from seven to twenty-seven days, post-treatment swabs were used, 55 per cent of the cases were negative. Actually the per cent of negatives was probably less than 50 per cent due to the insufficient number of post treatment swabs. However, after treatment nine were still positive indicating that santonin as a treatment of enterobiasis is far from specific, and would prove to be far from satisfactory for familial situations.

D. Tetrachlorethylene

Hall and Agustine (1929) first recommended using tetrachlorethylene in pinworm infections. They succeeded in expelling 5,544 pinworms from one patient and 1,191 pinworms from a soldier following treatment with a single dose of 4cc of this drug.

Wright, Bozicevich and Rose (1937) treated eleven children between the ages of 4 and 15. They were given a light supper of non-fatty foods the night before treatment, after which at 9:00 P.M. they were given soapsuds enemas. No breakfast was given the following morning and treatment started at 7:00 A.M. The dosages for children under 12 years of age was 1cc for each year of apparent age. For children over 12, dosages were given commensurate with the age, weight and physical condition of the patient. The drug was administered in a magnesium sulphate solution to those over 10 years of age. An average of 352 worms per patient was passed. Of the 11 treated 6 were found to be negative after the period

of treatment.

Wright, Bozicevich and Gordon (1937) treated 47 boys infested with pinworms with single doses of orally administered tetrachlorethylene. The dosages were the same as those reported in the previous paper. Magnesium citrate instead of magnesium sulphate was used as the base solution in order to decrease the amount of reactions incurred in the use of the latter. Two post treatment swabs were made fourteen to twenty-one days after treatment and 30 or 68.2 per cent were negative. The number of worms expelled varied from 1 to 4,958; the mean being 255.8 worms. Tetrachlorethylene was believed by the authors to be probably of more value in treating light pinworm infestations rather than heavy infestations.

Wright and Bozicevich (1938) follow up the previous paper by a study of clinical improvements following single dose administrations of tetrachlorethylene. Clinical improvement was measured by gains in weight after treatment, amelioration of certain pretreatment symptoms exhibited by the individual, and any gain in scholastic standing. The degree of pinworm infestation was evidenced by the number of worms passed. Part of lightly infected individuals showed little or no clinical improvement after treatment although they were shown to be negative, after post-treatment anal swabs. On the other hand heavily infected individuals who passed relatively large numbers of worms (up to 5,000) showed clinical improvement after treatment, even though they still remained positive upon post-treatment checks.

Thirty-seven of the 44 boys gained an average of 2.74 pounds each in the two weeks following treatment, 4 boys lost an average of 3 pounds each during this same period and the remaining 3 boys showed no change in weight.

Information supplied by the parents indicated clinical improvement in a varying percentage of the boys treated, and amelioration of certain symptoms associated with pinworm infections.

A slight improvement in scholastic standing was noted, in information gathered from teachers of 24 boys treated. Ten of the 24 exhibited marked improvement in their social attitude and scholastic standing within the two months' period following treatment, no change was noted in 9 of the boys either in social attitude of scholastic standing, 2 of the boys exhibited worse behavior and the remaining 3 boys indicated mixed changes in attitude for both better and worse. The teachers were ignorant of the fact that these boys had received treatment, and therefore their answers to the questions put to them were regarded as spontaneous and unbiased.

Wright and Cram (1937) report that tetrachlorethylene is the best single dose drug for treatment of pinworm infections. It is much safer than oil of chenopodium or carbon tetrachloride, and can be used safely in treating children. It is soluble enough in water to act on the larval forms in the small intestine and yet insoluble enough so that it can be carried into the large intestine to act on both the pre-adult and adult worms present there. Tetrachlorethylene is administered in a

magnesium citrate base solution which is readily taken by children who do not object to its taste. Furthermore, use of a magnesium citrate solution eliminates the reactions such as nausea, vomiting, headache, dizziness, and frequent abdominal discomforts, encountered when sodium and magnesium sulphate are used as bases.

E. Phenothiazine

Phenothiazine is a thiazine dye related to Lauth's violet, and methylene blue. It is fine, smooth, pale lemon-yellow powder, insoluble in water and almost tasteless.

Manson-Bhar (1940) reported that children under 8 years of age were given a dosage of 2 grams daily for seven days. Half this dosage or 1 gram per day for seven days was given to children under 4 (about 17 grams per pound of body weight). Adults were given 4 to 8 grams of phenothiazine daily for from seven to ten days. Granular preparation sweetened in limejuice was the method of administration of the drug for children, while adults were given tablets.

No dietary restrictions were made. Cotton gloves, to prevent contaminated fingers from coming in contact with the mouth, and mercurial anal ointments were not used and enemas were eliminated during the course of treatment. No toxic cases developed.

Phenothiazine was administered to four children infected with Enterobius. A boy aged 4 received 5 grams daily for seven days and three girls aged 7 to 8 received 1 gram daily

for seven days. During the first two days of treatment active pinworms were observed in freshly passed stools. On the third day the worms passed were far more sluggish and the remainder dead; some were stained red. In the three girl cases, cures were effected. In the fourth case, the boy aged 4 suffered a relapse but an increased dosage, 1 gram daily for seven days produced a permanent cure.

Phenothiazine was then administered by Manson-Bahr to a family in which both the mother and the father had been infected for at least six years. Quassia chips, santonin, beta-napthol and hexylresorcinol had been tried without avail. The father and mother both received 2 grams of phenothiazine daily for the five day period. Sodium sulphate was given after each dose of phenothiazine. Both mother and daughter were cured. The father suffered a relapse but when given a course of treatment involving the use of 4 grams daily a cure was effected.

An adult aged 47 complaining of loss of weight, mental irritation, and anal itching, was discovered to be heavily infected with Enterobius. He was given 4 grams of phenothiazine for four days. On the third and the following days, dead worms were passed and the treatment was begun again with apparently permanent results.

Because of the above fact, Manson-Bahr believed that greater dosage than 4 grams is necessary for adults if permanent cures are to effected; and therefore recommended an 8 gram daily course of treatment for five days as being the best treatment for adults infected with pinworms.

Kuitunen-Ekbaum (1941) reported a treatment study of 89 children and 9 adults who were given oral administrations of recrystallized phenothiazine. He presented the following total dosage plan:

4 - 5 grams for 24 children (2 - 5 years of age)

6 grams for 31 children (6 - 8 years of age)

8 grams for 34 children (9 - 14 years of age)

5 - 10 grams for 9 adults

Eighty-six per cent of the children and 89 per cent of the adults were cured by the first course of treatment. After the second course of treatment practically 100 per cent cures were effected. From 7 to 10 post treatment NIH swabs were used to demonstrate efficacy of treatment. Kuitunen-Ekbaum used smaller total dosages than those employed by Manson-Bahr. The course of treatment was five days.

Miller and Allen (1942) treated 50 children infected with pinworms with phenothiazine. The dosage was 1 gram per day for from six to eight days. In this study, 64 per cent of the children were cured. In another experiment, 23 children were treated with phenothiazine on three consecutive days - 2.5, 2.5 and 2 grams being given in that order.

Miller discovered that phenothiazine produced a definite but in most cases a slight anemia in at least half of the children treated. The hemoglobin level reached its lowest level four to seven days after treatment but returned to normal in three to four weeks.

According to Miller, phenothiazine is more effective when

given in relatively larger doses over a shorter period of time.

Sisk (1943) treated a group of 62 children and four adults, 24 of whom were positive for pinworms, with phenothiazine. Adults and children over 6 years of age received 1 gram a day for six days, then an eight day rest, followed by a six day treatment of 1 gram of phenothiazine per day. Children from 1 to 6 years of age received the same treatment except they were given smaller doses, 0.5 grams per day. Children under 1 year of age were given 0.25 grams per day. Post treatment swabs were taken and 4 children were shown to be positive. An 83 per cent cure was achieved.

Bercowitz, Page and de Beer (1943) examined 24 outpatients for blood and urine disturbances after administration of a 40 gram dose of phenothiazine three times daily over a ten day period. Three cases showed a decrease of 1,000,000 erythrocytes per cubic centimeter, 9 patients showed a 10 per cent decrease in hemoglobin, 6 patients showed a trace of albumin in the urine, 2 patients cylindroids, and 1 patient hyaline and cellular casts. Two patients developed nausea and vomiting.

Most (1943) reported that phenothiazine is an effective drug in the treatment of pinworm infections. The drug has been given to more than 200 patients and caused no noteworthy toxic reactions. The recommended dosage is 300 milligrams per kilogram of body weight. This is to be administered over a three day period. Most recommended that phenothiazine be given to patients normally refractive to gentian violet.

F. Gentian Violet

Faust and Yao in 1927 introduced gentian violet theropy in treatment of helminthic intestinal infections. Wright and his associates (1938) used gentian violet in the treatment of enterobiasis and obtained a 91.8 per cent cure. They recommended treatment with gentian violet (Seal-Ins), a trade name, over a period of eight days, followed by a seven day rest period, after which another eight day period of treatment was repeated. The adult's dose was two - 32 milligram tablets, three times daily; the children's dose was 10 milligrams for each year of apparent age, divided and adminstered in three doses.

Wright and Brady (1938) tried enteric-coated gentian violet capsules on 31 persons known to be infected with pinworms. Of the 31 treated, 25 (80.8 per cent) remained negative on the 7 consecutive post treatment swabs which began forty-two days after completion of the treatment period. These enteric-coated capsules are superior to the previously used tablets and are better tolerated by the patient. The dosage and length of treatment was the same as that used in the earlier study.

D'Antoni and Sawitz (1940) undertook to determine the value of gentian violet therapy for treatment of enterobiasis under institutional conditions. Medicinal treatments were carried on in two of the institutions examined, the other institution being treated by the use of enforced hygienic

measures (see section on hygienic control). The drug used for the chemotheraputically treated institutions was gentian violet (Seal-Ins) medicinal - ½ grain tablets. Tablets of the four hour type, which are said to dissolve in the caecal region, were used.

Institution N.H. was treated with medicinal gentian violet (Seal-Ins) without any specific hygienic measures being taken. The 122 boys in the institution were of the following classificational groups: 32 large boys (14 to 18 years of age), 31 of whom had enterobiasis; 50 medium sized boys (11 to 15 years of age), 49 of whom were positive for pinworms; and 37 small boys (12 to 17 years of age), 35 of whom were infected with pinworms.

The large boys received 1 grain, three times daily before meals for eight days followed by a seven day rest period and a subsequent eight day treatment period. The medium sized boys received 1 grain, three times daily before meals for five days followed by a three day rest period; the procedure was repeated until twenty treatment days had been passed covering four entire courses of treatment. The small boys received ½ grain three times daily before meals continuously for a thirty-five day period. To prevent the possibility of reinfection, the medication procedure of the three groups was terminated at the same time. Post treatment NIH swabs showed that 94 per cent of the large boys, 90 per cent of the medium-sized boys and 89 per cent of the small boys previously infected with pinworms were now pinworm free.

In institution P.E., 21 of the 26 white girls aged 5 to 21 had enterobiasis. Five girls weighing a hundred pounds or less received ½ grain, three times daily for fourteen days. Hygienic measures were employed similar to those enforced in the institution treated by hygienic measures solely. Immediately following treatment 7 NIH swabs were made indicating a 100 per cent cure.

Symptom reactions during medication with gentian violet Seal-Ins, were never of a serious nature and generally the drug was well tolerated. More than half of those, however, who were given 1 gram doses three times daily, experienced abdominal cramps, nausea and a loss of appetite. Manufacturers of Seal-Ins suggest administration of the drug an hour before a meal because an empty stomach will dissolve the drug in the time specified while food present in the stomach probably prolongs the time necessary for absorption.

D'Antoni and Sawitz (1940) believe this work demonstrates the value of mass treatments and the continuance of treatment until all the individuals are rid of pinworms, since a single positive individual can result in reinfection of the others.

Miller and his associates (1940) treated 29 school children aged 6 to 13 with gentian violet pills. The children 6 to 9 were given two 3/20 grain tablets three times daily, and those 10 to 13 years of age received one ½ grain tablet, three times daily. Three post treatment swabs indicated a 90 per cent cure. Vomiting occurred in two children, one of whom was in error given more than the prescribed dose, the other child

of five was given three, 3/20 grain tablets daily for nine days.

Wright and Brady (1940) examined a series of cases comprising 224 individuals who were oral administrations of gentian violet enteric coated capsules. Adult and children's dosage and course of treatment was the same as employed by these same workers in 1938. Of the 224 individuals treated, 189 or 84.5 per cent were found to be negative for pinworms after 7 NIH swabs. Some patients suffered from nausea, vomiting, diarrhea, and abdominal pain. The reactions were only temporary and were quickly overcome by a reduction in dosage or a rest of a couple of days before resumption of treatment.

When Ascaris infections are concomitant and various other disorders, such as moderate to severe cardiac, hepatic and renal disease, alcoholism and gastro-intestinal infections are present, gentian violet therapy is not recommended.

Minimum lethal dose for rabbits is 22 milligrams per kilogram of body weight. One dog tolerated a dose of 35.4 milligrams per kilogram of body weight for eighteen days.

Gentian violet therapy seems to be of considerable value in treatment of pinworm infections. It is cheap, efficacious, and easily administered.

G. Hexylresorcinol

Lampson, Brown and Ward (1935) in an extensive investigation of the parasiticidal actions of the alkylhydroxybenzenes found that crystaline hexylresorcinol showed the best results in treatment of pinworm infestation.

Brown (1932) was the first to use hexylresorcinol in the treatment of Enterobius vermicularis. Six adults infected with pinworms were given 1 gram of hexylresorcinol orally. Administration with the drug was made early in the morning, breakfast ommitted, and no food taken for six hours following treatment. Three weeks after treatment adult worms or ova was found in the stools of all six patients. Brown has set forth three conclusions which he has arrived at from his investigation.

1. Hexylresorcinol is very active against Enterobius, a 1-1000 suspension killing them in two minutes.
2. Five out of 6 persons treated with hexylresorcinol pills and enemas were Enterobius free. Two out of 3 persons treated by enemas only were freed of pinworms. This suggests that oral administrations should be coupled with enemas to achieve the best results.
3. Suggested procedure for treatment

   Treat twice in the morning as follows:

   a. Omit breakfast and partake of no food until noon.
   b. Hexylresorcinol pills (1 grain per year of age up to 10, then 1 grain doses for all above this age). Drink plenty of water.
   c. Soapsuds enema followed by an enema of 1 part crystalline hexylresorcinol to 1000 cc of water; this enema to be given and restrained for 5 minutes.
   d. Bed sheets to be boiled twice weekly to destroy any eggs passed on them.

Wright and Cram (1937) relate that Robbins (1934) found

that after giving a 1 gram dose of hexylresorcinol 18 per cent was excreted in the urine and 64 per cent in the feces, showing that the drug does reach the large intestine in fairly large concentrations. Hexylresorcinol administered orally and in enema form or in enema form alone seems to be quite effective in some cases of pinworm infections. With young children under 10 treated with enemas of hexylresorcinol alone, 5 grams to one liter of water, excellent results were obtained in some cases. Hexylresorcinol has two definite disadvantages: first, it is expensive, and, second, in cases where enemas alone are given treatment must be continued over long periods of time.

Spaak (1936) recommended hospitalization of the patient for three days and then administration of hexylresorcinol for three days. On the first day the patient received a purgative, on the second day hexylresorcinol was given orally and by enema, and on the third day by enema alone. This treatment is repeated in fourteen days.

Wright, Brady and Bozicevich (1939) treated 27 individuals positive for Enterobius with a varying number of hexylresorcinol enemas in a dilution of 1:2000. Eighteen or 67 per cent were negative on post-treatment swabs. Hexylresorcinol enemas proved of considerable value in treatment of Enterobius infections. A minimum of ten such enemas over a three week period is necessary to expell the helminths and ova of Enterobius vermicularis.

Wright and his associates tried single oral administrations of hexylresorcinol in the form of Caprokol pills, this proved

ineffective in treatment of 4 cases of pinworm infection. Dosage varied between 0.4 to 0.8 of a gram.

In treatment of three cases of pinworm infestation, Wright and his associates used hexylresorcinol orally and with enemas. Only one of the three proved to be negative by post-treatment and swabs. Apparently, oral treatment by hexylresorcinol in the form of Caprokol pills, even when combined with hexylresorcinol enemas, does not constitute an effective treatment of pinworm infections.

Eight cases of pinworm infestation were treated by 1 ounce of hexylresorcinol jelly 1:1000 anally administered at bedtime over varying lengths of time. Six of the 8 were still positive after treatment and one of the 2 negatives was a doubtful negative. Hexylresorcinol jelly was shown to be non-effective in killing gravid females located in the rectum. It did not always prevent worm migration to the rectum or render the ova deposited there inactivated, since they readily hatched when placed in a moist chamber.

Wright, Brady and Bozicevich sum up by stating that hexylresorcinol enemas are of value if treatment is continued over suitable length of time. They repeat Wright and Cram's (1937) view that there are two disadvantages in use of the drug: first, it is expensive and, second, in order to be effective treatment must proceed for a considerable length of time.

Regardless of the disadvantages in the use of hexylresorcinol, its efficiency in treating infections of Enterobius

vermicularis has resulted in its being retained by the medical profession as the recommended therapy. The adult dosage is given as a 0.2 gram pill. A single treatment consists of 5 pills taken successively.

D. Control of Pinworm Ova

1. Physical

Shalimov (1935) studied the effects of ultraviolet light on the development of the eggs of parasitic worms with the use of a mercury-quartz lamp. The irradiation distance was 50 centimeters. All the eggs of Enterobius vermicularis were killed in five minutes. The ultraviolet rays were found to disturb the delicate physicochemical processes within the egg. The effectiveness of the lamp was found to depend upon its power.

Hollaender, Jones and Jacobs (1940) employed monochromatic ultraviolet in order to observe its effect on pinworm ova. Eggs of Enterobius vermicularis were evenly irradiated in a beam of monochromatic ultraviolet light by rotating the exposed dish and by blowing a stream of air against the surface to insure uniformity of treatment of all the free floating eggs. Technical difficulties and not individual resistances are responsible for the inability to kill the remaining few of the eggs.

Variability in response to irradiation does occur, however, in eggs taken from different patients. Hollaender and his

associates state:

> "If we realize the probable variation of the sources from which pinworms come, the nutritional level of the patient, the different type of food the patient has eaten, the PH of the intestine, the variation of the intestinal flora, and also perhaps of equal importance, the variation in the methods and time of obtaining the eggs and bringing them to the laboratory, this variation in response of eggs from different sources is certainly not surprising."

The variation of different egg batches applies only to the absolute energy, in eggs, of killing at specific wave-lengths. The inactivation of the eggs is not dependent on the wave-length. The eggs showed their greatest sensitivity to the radiation of 2280 Å, and a slight maximum at 2805 Å.

The egg of Enterobius, as has been pointed out in an earlier part of the paper, is a fairly complicated structure composed of three shell layers: the outermost being protein, the second, chitin, and the innermost layer lipoid. The egg is not completely surrounded by these three egg layers. The hatching area of the egg proper seems to be devoid of a chitin layer. Most of the radiation seems to be absorbed by the proteinaceous top layer.

Hollaender, Jones and Jacobs (1940) theorize that the use of ultraviolet radiations could produce an inhibition of hatching by a hardening of the protein layer; second, by a change in composition of the lipoid membrane; third, by irradiations injurying the embryo and lastly by possible toxic substances formed within the embryo as a result of this radiation. Death and the failure to hatch is probably due to a combination of these factors and not to any single effect

produced by irradiation.

Ultraviolet radiations tend to be efficient in destroying tadpole stage embryo but of only limited value in killing infective stage eggs. Pinworm eggs are most sensitive at the shortest wave-length tested, i.e. 2280 Å, less so at 2650 Å and a slightly increased sensitivity at 2805 Å. Bacteria and fungus spores have their greatest sensitivity at 2650 Å. It takes one egg at 2650 Å to produce 50 per cent killings in pinworm eggs.

In a follow up article, Jones, Jacobs and Hollaender (1940) investigated the effects of monochromatic ultraviolet radiations on (1) hatching rate, (2) eggs stored after irradiation, (3) the survival of larvae hatched from irradiated eggs, and (4) the further development of immature eggs.

Eggs of Enterobius vermicularis in the infective ring and a half stage, and a few in the tadpole stages were rotated in small Syracuse watch glasses so that the exposed eggs were subjected to a uniform and measurable quantity of monochromatic ultraviolet radiations. Most of the tests were made with ultraviolet radiations with wavelengths of 2650 Å.

(1) Effect on hatching rate:

Controls and irradiated eggs were examined in a four hour hatching period. Most of the controls hatched in the first fifty minute interval. Control and only slightly irradiated eggs hatched at about the same rates. Most of the irradiated eggs had a slow start in hatching with a rapid increase in hatching rate after the first hour and with a drop in the

hatching rate in the last hour.

Ninety per cent of the controls hatched by the time the first observation had been made. The irradiated eggs hatched at a much slower rate. Apparently, irradiation of a sufficient intensity is successful in retarding hatching.

(2) Effect of storage on ability to hatch

Eggs in control dishes stored at 5°C retained their hatching ability while those at 24° to 26°C showed a drop in some but not all cases. Irradiated eggs exhibit a great drop in ability to hatch after eighteen and forty hours of storage. This indicates the presence of sublethal effects in irradiated ova.

(3) Effect on survival of larvae

Fifty-four larvae hatched from an irradiated egg dish, after a half to one and a quarter hours, a survival rate of but 35 per cent was noted.

One hundred and nine larvae hatched as active larvae from a control egg dish, after a half to one and a half hours, a survival rate of 95 per cent was in evidence. Radiation of pinworm eggs seem to increase the dying rate of the hatched larvae.

(4) Effect on immature eggs

Immature eggs of the tadpole stage to be more succeptable to irradiation than do more advanced and developed eggs. Three dishes of immature egg dishes were irradiated at 2650 Å and showed after irradiation a decreased ability to develop into the infective one and a half ring stage larvae. Besides

the three egg shell layers, the infective stage ring and a half larvae have several protein layers in the cuticula and hence are better protected than are the immature larvae forms to ultra-violet irradiation.

It is evident by these two papers on radiation with ultraviolet light that while the number of eggs killed outright is small in comparison with the number that hatch these larvae exhibit sublethal effects in a longer hatching time, a decreased ability to hatch after storage for varying lengths of time and an increased mortality of those which do manage to hatch.

2. Chemical

The amount of work on chemical treatment of pinworm ova is small, and indicates that a considerable amount of work remains to be done.

Jacobs and Jones (1939) stated that because of the makeup of the egg membranes of Enterobius an effective ovicide should possess lipoid and protein solvent properties.

Most authorities agree that hygienic measures in treatment of prevention of pinworm infections should involve frequent washing of hands with soap and water. Schuffner and Swellengrebel (1943) relate that during the recent war heavier infestations with pinworms in Europe were directly attributable to the acute shortage of soap. Whether the action of soap and water on eggs of pinworms is chemical or physical or a combination of the two is difficult to say. The soap and water would probably have an effect on the surface tension of the

eggs, leaving them more vunerable to the caustic action of any free sodium hydroxide present in the soap.

Nolan and Jones (1942) presented a study of the survival of eggs of Enterobius vermicularis exposed to household fumigants. They exposed pinworm eggs on a dry base or shallow water to "Cyanegg" (minimum of 96 per cent of sodium cyanide) in a closed chamber at the rate of 1 pound per thousand cubic feet of space for twenty-one hours. The eggs twenty feet from the "Cyanegg" survived at the same rate as the controls. Paradichlorobenzene and napthalene were likewise used and found to be ineffective in changing the survival rates of the pinworm eggs.

The use of anal ointments in connection with control of adult pinworms has been mentioned earlier. The various anal ointments, Mercuric phenols and so forth, seem to be ineffective in rendering the ova non-viable (Wright, Brady, and Bozicevich, 1939).

3. Hygienic

Prophylaxis or prevention alone is not very effective in pinworm infections. Cram (1939) noted that theraputic coupled with prophylactic measures resulted in a lowering of the incidence substantially during the last 6 months of the 18 month period of examination of children in a private nursery. The prophylactic measures taken included disinfection of toilets and bowl with Lysol as well as sprinkling it on the floor before sweeping. Cots and blankets were aired in the

sunshine. The children were required to use individual towels and to wash their hands after the use of the toilet and before eating.

Sawitz, D'Antoni, Rhude and Lob (1940), in their study of the epidemiology of Enterobius in 471 children of whom 74.3 per cent were infected, employed strict hygienic measures, such as frequent house cleaning, changing of underwear and bed-sheets, and frequent bathing, but found that it did not disrupt the transmission routes of Enterobius. A cleanliness period of six weeks was tried out in one home, but no decrease in incidence was apparent after that time.

D'Antoni and Sawitz (1940) employed non-medicinal therapy in an institution made up of 23 girls and 35 boys of which 38 per cent were found to be infected by pinworms. The non-medicinal treatment involved rigid hygienic measures. The rooms were washed daily with hot soap and water, the children were required during sleep, to wear short cotton pants, which were changed and sterilized daily, and nail brushes were used by the children. Finally, two showers were required daily. This non-medicinal, hygienic treatment was put into effect for six weeks. Since the ova's viability outside the body was determined by Lentze (1932) to be one week, it was therefore thought that a six week control period should be more than effective in reducing the number of ova outside the body.

The results of this rigid, hygienic, six week period was an increase from 38 to 51 per cent in the incidence of Enterobius.

In another institution D'Antoni and Sawitz (1940) combined medicinal and non-medicinal procedures. The hygienic measures employed were similar to those enforced in the first institution. Immediately after treatment with gentian violet the 26 girls of the institution, 21 of whom were positive before treatment had been initiated, were given 7 NIH anal swabs indicating a 100 per cent cure.

Hall (1923) reported hygienic measures, such as washing hands after defecation and upon eating, sterilization of bed clothing, underwear and bedding, to be of value.

From the above data, it is readily apparent that hygienic measures alone will not clear up pinworm infections and that when used with medication best results are achieved.

Wright and Cram (1937) stated that because of the wide distribution of pinworm ova in such places as underwear, night clothing, other clothing, bed linen, towels, wash cloths, soap, floors, upholstery, and furniture it appears to be impossible in a practical sense to impose sufficiently rigourous hygienic measures to prevent reinfection especially in lower economic dwellings where many people are crowded into a relatively short space of a few rooms.

F. Reasons for Difficulty in Treating Enterobiasis

There are several reasons why it is difficult to treat enterobiasis: first, the great number of eggs which are liberated by the female during oviposition; second, the distribution of eggs throughout the household; third, the

resistance of the eggs; fourth, the familial group concept; and lastly the need of a better chemotherapatic agent.

Sisk (1943) examined two mature females as to egg content and counted 9,860 and 10,240 ova respectively.

Reardon (1938) completed a study on the egg counts in 20 gravid female specimens of Enterobius. Most of the worms were fixed in 10 per cent formalin; two in Bouin's and one in hot alcohol; measurements of the worms were made with a stage micrometer and dissecting scope. All worms were previously soaked in a tenth normal solution of sodium hydroxide for twenty-four hours, and then were stained in a blue rayon dye for another twenty-four hours. Counts were made on a special Rhodium-plated 2 by 3 inch slide squared off in one millimeter sections. On each of four such slides, three drops of glycerine were placed. A previously stained worm was then placed on the slide and dissected microscopically with an iris knife in quarters one of which was allowed to remain on the slide and the other three portions placed in the other three glycerine slides. Each worm portion was teased with the iris knife and a dissecting needle until a level dispersion of ova was achieved. The dissecting instruments were passed into glycerine to release eggs adhering to their surfaces, and 24 by 50 millimeter cover slips were applied to the preparation. The ova were counted under a compound microscope with 5 x ocular and 16 millimeter objective lens. The egg count was accomplished by means of a Veeder hand counter.

Worms having the highest egg count were secured from

specimens recently expelled in stools after treatment with gentian violet. Variation in egg count may be correlated with the size of the gravid female specimens. Counts made in the 20 gravid females show from 4,672 to 16,888 eggs per worm, the arithmetical mean being 11,105.

From these findings it is evident why an infected household may harbor many thousands of ova and that there is real danger from infectious material. Cobb (1890) wrote on the matter of "contagion" in enterobiasis:

> "The abundance of the eggs about dwellings is difficult to overstate, and is easily illustrated by a calculation. Reckoning 50 female worms to the individual (with 20,000 eggs in each female) an average which I am certain is exceeded in many localities, we have a population of 250,000, the enormous number of two hundred and fifty thousand million eggs, which if distributed evenly over 20 square miles, would furnish four to five hundred eggs to the square foot."

Reardon taking 11,000 eggs per female worm as the mean assumes that a slight case of oxyuriasis with 5 worms involved, the startling figure of 385,000 eggs would be present in a household at the end of a week's time. Such figures explain why Enterobius vermicularis has been long considered by many competent parasitologists as the foremost pathogenic animal parasite of man.

Oleinikof (1929) had made the first study of Enterobius ova in dust. He collected samples of dust, from the floors, benches, and window sills of a schoolroom with a camel hair brush dipped in glycerine. The brush was washed into a test tube and the washings centrifuges. Ova from Enterobius was recovered from many of the dust samples.

Nolan and Reardon (1939) collected dust samples from seven houses in which heavily infected person or persons lived. Of 241 dust samples taken 221 (91.7 per cent) were found to contain pinworm ova. Samples were taken from all the rooms at different levels and locations. The 20 negative samples are not regarded as significant since other samples from the same room were positive. The recovered ova were in all stages of development from tadpoles to fully developed embryos. About one-fourth of the ova examined were found so deteriorated as to make their developmental stage impossible to ascertain; of the remaining three-fourths, 75 per cent contained fully developed embryo and 17 per cent undeveloped tadpole stages. About one-half of the ova examined were considered viable or only recently dead. Twenty-four of the microscopically determined "viables" were incubated in gastric juice at 37°C with these results: 9 failed to hatch, 1 did not hatch but was active, and 14 either completely or partially hatched.

The location of ova on high mouldings of doorways and windows indicates, according to Nolan and Reardon, transportation of viable ova by air currents and the possibility of infection through inhalation. This supposition was later proved by an experiment performed by Schuffner and Swellengrebel (1946).

In 1944, Schuffner and Swellengrebel demonstrated the presence of eggs in the dust of rooms by gathering dust from 10 square decimeters, suspending it in water, filtering through wire gauze and centrifuging for one minute at moderate speeds.

The sediment was suspended in a solution of zinc chloride in a narrow vial. In 45 minutes, nearly all the eggs had risen to the surface, adhered to the coverglass on the surface and readily counted. Large quantities were found in schoolrooms and even dining rooms, where the children stayed only a relatively short time.

It has been noticed earlier that the eggs of Enterobius are practically refractory to the common household fumigant, "Cyanegg". Are they affected by such natural factors as changes in temperature and humidity? To answer this question Jones and Jacobs (1941) performed a series of experiments in order to see precisely what effect temperature and humidity changes had on the viability of the infective ring and a half stage of the ova. A summary graph of their results is given below. The chart is of necessity an approximation.

Fig. 19. Representation of survival rates of pinworm ova under dry conditions (30-54 per cent humidity), and exposed to temperature changes.

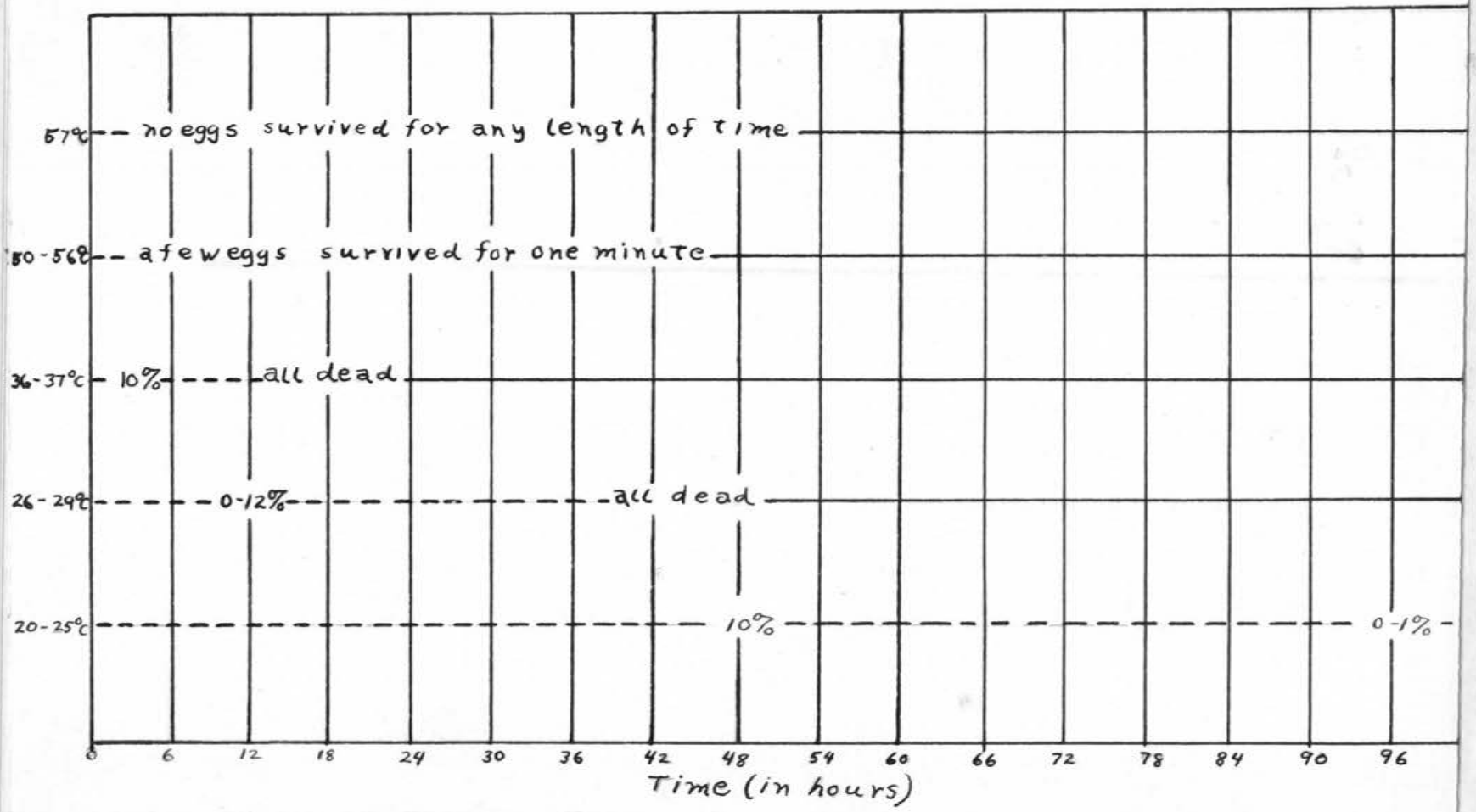
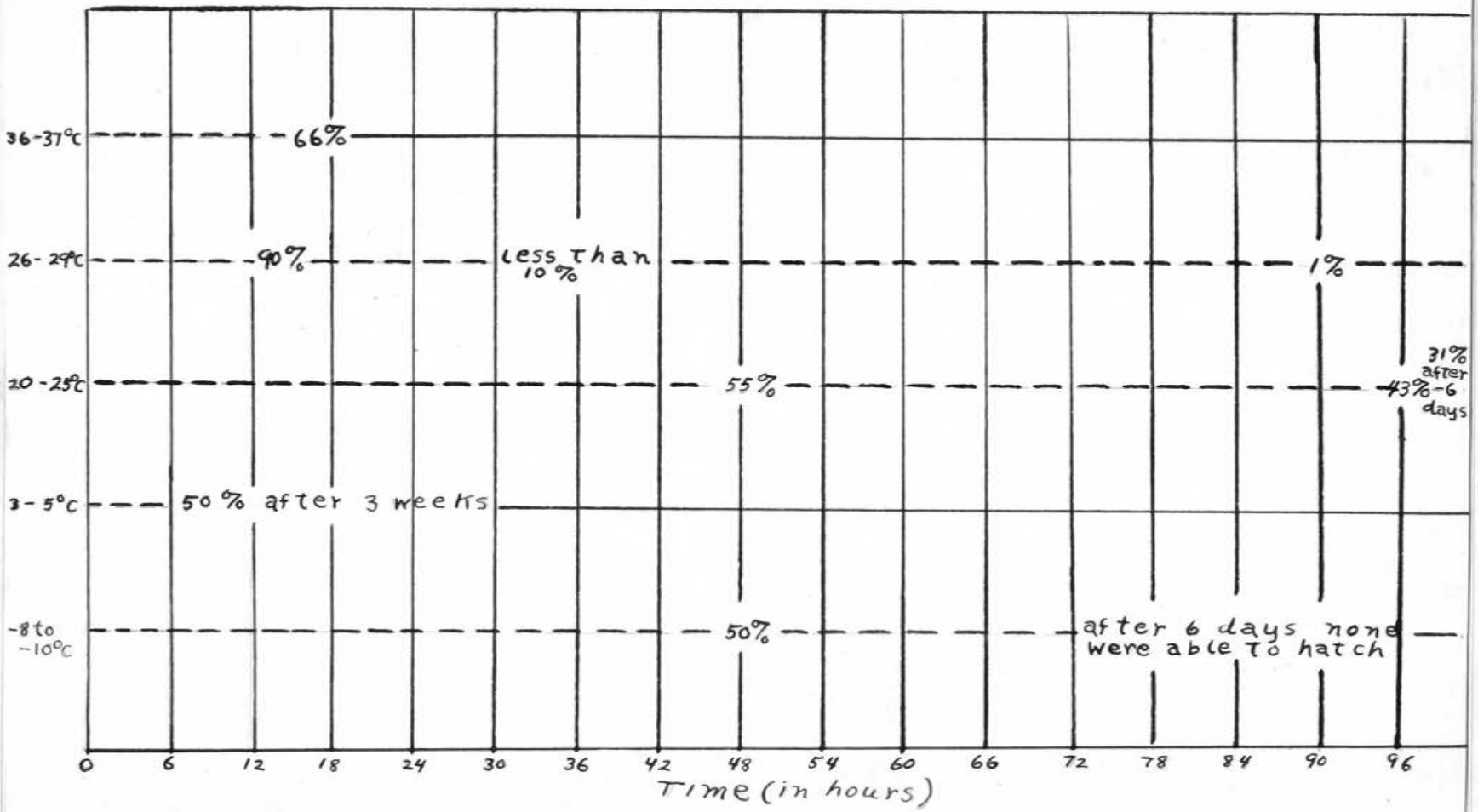


Fig. 20. Representation of survival rates of pinworm ova under moist conditions (53-91 percent humidity), and exposed to temperature changes.



It is evident that drying conditions and high temperatures are the most valuable for control measures. The effect of drying is thought by Jones and Jacobs to be brought about by a loss of water from the egg membranes or the embryo itself or both. Loss of water from the shell membranes, especially the innermost, might result in a change in permeability and thereby interfere with hatching and certain vital life processes going on within the egg proper.

The eggs of Enterobius vermicularis survive, fairly well, changes in temperature and humidity. When it is realized that no such extremes as were conducted in the experiment are likely to be met in vivo, and that the average home's temperature and humidity would not present any great obstacle to the survival of pinworm eggs, the great problem in control is self-evident.

Earlier in the paper notice was made of the familial nature of enterobiasis (Cram, 1941). This is a most important concept. Group situations must be appreciated if control measures are ever to be effective.

To illustrate the difficulty encountered in the group situation, Wright, Jones and Cram (1937) presented the problems posed by a single family of seven individuals, all of who were infected with pinworms. The family was observed over a ten months' period. The family belonged to a low income group. Various treatments were given and approximately 800 examinations made. Anal swabs were used in diagnosis rather than fecal or fingernail examination.

At some time during the period of observation five of this

family group were freed of the infestation for varying lengths of time. The remaining two were never negative for more than ten to fifteen days at a time, and at no time in the examination was all of the family negative. Apparently reinfection was constantly taking place due to the residual cases of infection present.

The study indicates once more the familial situation in regard to pinworm infections and the necessity of treating simultaneously all the members of a family. The study also indicates the difficulties encountered under conditions of inadequate income, restricted living space, and above all, poor sanitary conditions.

Lastly there seems to still be a necessity for a better drug to treat enterobiasis. According to Wright and Cram (1937), in enterobiasis, expelling 90 per cent of the worms is of only transitory relief, for, unless the patient is of the none too common type which will carry out the tedious and complex procedure necessary to prevent reinfection, the residual infection will build up or even surpass its former levels. At present there is not avilable any anthelmintic drug which in a single dose can safely and surely expell all the worms, larval and adult, from the host. Since a 100 per cent effective anthelmitic drug has not yet been found, multiple dose therapy with relatively non-toxic drugs used daily over a period of time seems to be the best way to treat the infection. Repeated treatments have the advantage of preventing reinfection during treatment by getting rid of the larval worms ingested

with infected material and of inabling the drug to reach the rather inaccessible areas of the large intestine and calcum.

Because of the great egg production (probably about 10,000 per worm), the wide distribution of these eggs throughout an infected household, the resistance of the eggs to either chemical or physical control agents, the familial nature of the infection plus the subtle factor of residual infections, and lastly the absence of a really efficient, non-toxic and easily administered drug, Enterobius is well established as the foremost helminth pathogen of man and will undoubtedly be with us for many years to come.

VI

CLINICAL SURVEY OF 36 INSTITUTIONALIZED BOYS WITH THE USE OF A MODIFIED ANAL SWAB

A. Introduction

In conjunction with a survey of the available literature on Enterobius vermicularis, it was thought that some original research work might be an important adjunct to the thesis. It was decided that the work would consist of incidence work done on a group of individuals in the Boston area. The only previous work done in Boston and in Massachusetts was performed by Weller and Sorenson (1941) who examined 505 children at Children's Hospital with the use of, in most cases, a single NIH swab and found 19 per cent positive.

B. Materials and Techniques

It was felt that the NIH swab would prove too complicated for easy use and the Graham swab was eliminated because it was not firm and difficulty in handling it may lead to inaccuracies. In further readings of the literature the Von Hofe swab was described. The idea of using a test tube as an applicator seemed to be of value and the swab devised was built along that general line.

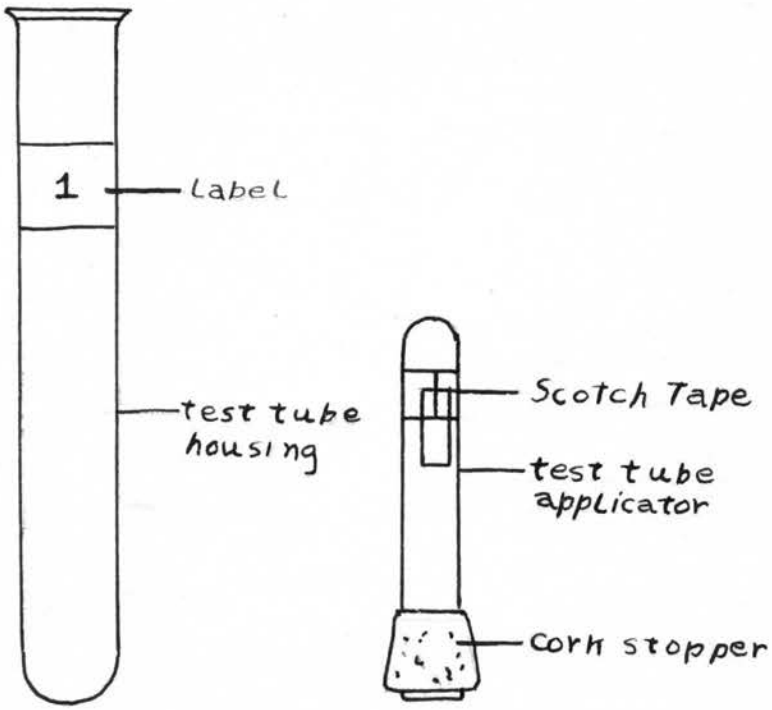


Fig. 21. Modified anal swab used in research problem.

The swab consists of a small test tube held in a larger test tube before and after using to cut down on egg loss or contamination of other swabs. The Scotch tape is fixed on the small test tube by cutting a length of Scotch tape slightly longer than the circumference of the small test tube and joining the ends snugly over the test tube so that the sticky surface faces out. Then a smaller strip of tape is attached to the joined seam and the test tube in the manner shown above. When the time comes to examine the used swab, the swab is withdrawn from the holder and the cellophane ring is placed seam up on a microscope slide. A scalpel or a razor blade is used to cut along the line of the joined ends, and the freed

Scotch tape is smoothed out on the slide. The swab is rotated until the rest of the tape is attached and the rest of the joined ends cut loose and discarded.

A powerful cleaning solution was prepared by adding 500 cc of commercial sulphuric acid slowly and with a great deal of stirring, so as to insure complete reaction, to 5 grams of sodium dichromate. The chromic acid evolved is an effective oxidizer of all organic material within contact and therefore an ideal cleaning solution.

A wooden box five and a half inches square and seven inches deep was built to carry swabs to and from the laboratory. Sixteen swabs could be carried in this way conveniently.

The study was carried on at the Catholic Boys Guidance Center located at 102 Fenway Road, Boston. Here in excellent surroundings a group of 36 boys, 13 to 17 years of age, the average being 14.8, were housed. These boys came from many parts of greater Boston. The institution is a stone structure comprising four floors in a residential area of the city. Of the 36 boys in the institution 14 lived in double rooms the other 22 boys in single rooms.

C. Procedure

For convenience it was decided that the boys be divided into three groups,, these to be tested on successive weeks. The testing began on January 18, 1949 and was completed February 3, 1949. The testing was performed early in the morning, (6:30 to 6:45 A.M.) before the boys had risen or

performed their toilet. The procedure for testing was fairly simple. The boys upon being wakened got out of bed, took down their pajama bottoms and spread their buttocks. The test tube applicator was withdrawn from the holder and the Scotch tape tip was placed against the perianal area and rocked back and forth several times in order to obtain a smear. The applicator was then replaced in the holder, transported to the laboratory and smoothed down on the surface of a microscope slide, as was previously described. Examination was made under low power, 100x.

D. Results

Exactly one-half or 18 of the 36 boys examined were found to be positive for pinworm ova. A total of 98 swabs were prepared, 2.7 per boy. Up to four swabs were given in order to establish negativeness. Three of the 18 negatives received only 3 swabs, one in the first group and two in the second group. No increase in incidence was noted among the boys living in double rooms as compared with those living in single rooms. Of 22 boys living in single rooms 13 or 59 per cent were positive for pinworms while of the 14 boys in double rooms only 5 or 35.7 per cent were infected.

In group I comprising 13 boys, 8 or 61.5 per cent were found to be positive. In group II 4 or 30.8 per cent of the 13 boys examined were positive for pinworms. In group III 6 or 60 per cent out of the 10 boys.were positive.

E. Summary of Findings and Conclusions

Group I had not only highest incidence rate of the three groups tested but also the positive microscopic slides had generally a great many more ova than those of either of the other two groups. No quantitative data on this observation was made.

TABLE VII

Summary Chart on Results of Research Problem

NUMBER OF POSITIVES

	1st swab	2nd swab	3rd swab	4th swab	Total
Group I (13 boys)	6	0	2	0	8
Group II (13 boys)	0	2	2	0	4
Group III (10 boys)	2	4	0	0	6
	8	6	4	0	18

This chart is convenient and useful in that by reading across and down the results of the tests are readily apparent. Reading across discloses how many boys in the individual groups were found to be positive at each swab. Reading down we find the total number of boys found positive in the three groups at any particular swab. This later fact gives us the clue as to the efficiency of the swab.

Of the 18 positives found, 8 or 44 per cent were disclosed

after the first swab. On the second swab 6 more positives were disclosed running the total to 14 or 78 per cent of the total number of positives. The third swab disclosed 4 more positives making up the full 18 or 100 per cent of all the positives found. The fourth swab made on 17 boys disclosed not a single additional positive.

It should be realized, however, that the negative fourth

after the first swab. On the second swab 6 more positives were disclosed running the total to 14 or 78 per cent of the total number of positives. The third swab disclosed 4 more positives making up the full 18 or 100 per cent of all the positives found. The fourth swab made on 17 boys disclosed not a single additional positive.

It should be realized, however, that the negative fourth swabs do not mean that with the aid of this swab cases of enterobiasis can be diagnosed on the basis of three consecutive swabs. From the above data it is impossible to predict what a fifth, sixth or seventh swab would have disclosed. Perhaps many more positives would have discovered. Undoubtedly much more work is needed to demonstrate the efficiency of this modified anal swab.

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THESIS ABSTRACT

The adult male pinworm measures from 2 to 5 millimeters in length, and has its terminal end curved ventrad. The female pinworm is more than twice as large as the male, 8 - 13 millimeters in length, and has its tail portion decidedly attenuate. Pinworm ova is characteristically flattened on one side, and measures 55 by 30 μ.

Examination of the feces show only a small percentage of the actual number of cases infected with pinworms. D'Antoni and Sawitz (1940) reported diagnosing less than 1 per cent of the true incidence using direct fecal films, and less than 25 per cent using fecal concentration techniques.

The use of swabs is the present accepted means for diagnosing pinworm infections. Many swabs have been devised, but only two are in general use (1) the NIH swab, and (2) the Graham swab. These two swabs require a total of 7 consecutive swabs before an individual is considered to be negative.

Serological testing seems to be of little value in diagnosing the presence of Enterobius. The reactions encountered upon injection of various sorts of extracts of pinworms into individuals are not specific and therefore of little diagnostic value.

The incidence of Enterobius in North America can be regarded as high, probably infecting half the people of North America.

Central America and Mexico seems to be the most heavily

infected area of the continent. According to Sandground (1933) and Mazzotti and his associates (1940) and (1943), the majority of central americans and mexicans are probably harboring pinworms.

In the Canadian-Alaska areas, the incidence of pinworms is high, according to Kuitunen-Ekbaum (1940) and (1943) and Ashburn (1941).

The numerous papers presented on the incidence of pinworms in various parts of the United States supports the view that possibly 40 per cent or more of the people in the United States are infected with pinworms.

The injury that results from infections with Enterobius is slight. Gravid female migrations may set up local irritations in the anal area known as pruritis ani. This latter phenomenon is often used as a basis for diagnosing pinworms. MacArthur (1930) reported that the first and most important cause of nocturnal pruritis is the presence of Enterobius. Numberous other complaints are associated with pinworm infections anorexia, irritability, headaches, gastro-intestinal symptoms and so forth.

Anal washes and ointments were once highly favored in treating pinworm infections, but they are, for the most part, no longer used. Wright, Brady and Bozicevich (1939) reported anal ointments to be of doubtful value in treating cases of enterobiasis since they failed either to destroy ovipositing females or render the ova non-viable.

Santonin was shown by Wright, Brady and Bozicevich (1939)

to be far from effective in treating pinworm infections.

Tetrachlorethylene is probably the best single dose drug for the treatment of Enterobius, but in large doses it is not well tolerated by the patients especially children.

Phenothiazine therapy is the standard treatment for enterobiasis in Europe and appears to be quite effective in ridding individuals of pinworms. With the use of phenothiazine nearly 90 per cent cures are effected.

Hexylresorcinol is probably the best all around drug available in pinworm therapy. The drug is better tolerated in large doses than most anthelmintics. It has, however, two major disadvantages, (1) it is expensive, and (2) if treatment is to be effective it must proceed for a considerable length of time.

Hygenic measures alone will not clear up infections with pinworms. Best results are obtained when medication is coupled with hygenic controls.

Thirty-six institutionalized boys, between the ages of 13 and 17, were examined with the use of a modified anal swab. The swab consisted of a small test tube having a ring of Scotch tape affixed to its terminal end. The applicator tube when not in use was inserted into a larger, housing test tube.

The boys were examined in the morning, 6:30-6:45 A.M. For the sake of convenience the boys were divided into three groups, group I - 13 boys, group II - 13 boys, and group III - 10 boys. Of the 36 boys examined on the basis of four swabs, 18 or 50.0 per cent were found to be infected with pinworms. The negative

fourth swabs were not regarded as evidence for the diagnosis of pinworm infections using three consecutive swabs.