



Sudan University of Science and Technology
College of Graduate Studies



Cytological Changes in Urinary Tract Cells mong
Patients Suffering from Schistosomiasis

التغيرات الخلوية لخلايا المجرى البولي عند المرضى المصابين
بالبلهارسيا البولية

A thesis submitted in partial fulfillment for the requirement of the
degree of M.Sc in histopathology and cytology department

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□ بسم الله الرحمن الرحيم

قال تعالى :

{قُلْ هَلْ يَسْتَوِي الَّذِينَ يَعْلَمُونَ وَالَّذِينَ لَا

يَعْلَمُونَ إِنَّمَا يَتَذَكَّرُ أُولُو الْأَلْبَابِ}

□ صدق الله العظيم

(سورة الزمر : 9)

Dedication

To my lovely mother for embracing me with an everlasting
emotion

To my great father who always behind me

To my lovely and my life (tasneem and sdan)_

To my brothers

To my sisters

To my friends

And to my colleagues

I dedicate this work with my best wishes to all

Acknowledgements

All my are in name of Allah , the most gracious and the most merciful

I would like to express ,my deep sincere gratitude and honest appreciation to my teacher and supervisor. Dr. Majdii Mansowr for his effort on supervision and encouragement throughout this work in completing this study.

Abstract

This study was a descriptive study (case and control) conducted in Khartoum state (Goze Elrimela) during the period from June (2010) —October (2014). This study was aimed to detect the cytological changes of urinary tract cells among Sudanese patients suffering from urinary schistosomiasis.. The secondary data was collected from books and internet. Random samples of 75 urine samples were taken from both male and female at a different ages of patients that effected by urinary schistosomiasis ranged from 12 to 56 years and 75 samples were taken from healthy population as control. Full voided urine in sterile container which processed and then stained by papanicolaue stain. Questionnaire containing essential patient identification data was used. Data were collected, from questionnaires and cytological results were analyzed using Statistical package for social sciences program and results were presented in frequencies and percentages in tables. The cytological changes were demonstrated in all patients sulfuring from urinary schistosomiasis 75(50%), inflammatory changes was noticed in72 patient (48%) and metaplastic changes in 3patients (2%).

الخلاصة

الدراسة عبارة عن بحث وصفي تجريبي أجري في منطقة الرميطة بولاية الخرطوم في الفترة من يونيو 2009 وحتى أكتوبر 2014. هدفت الدراسة الي بيان التغيرات الخلوية لخلايا الجهاز البولي بين المرضى السودانيين الذين يعانون من البلهارسيا البولية . تم الحصول على البيانات الثانوية من الكتب والمراجع ومن بعض مواقع الانترنت . تم أخذ عينات عشوائية من عينات البول من كلا الجنسين ومختلف الأعمار من المرضى الذين يعانون من البلهارسيا البولية وعينات أخذت من 75 من الأفراد الأصحاء . تم أخذ عينات في أنابيب معقمة وتمت معالجتها ومن ثم صبغها معملياً لإستخلاص النتائج . وقد استخدم استبيان يحتوى على البيانات الأساسية لتحديد هوية المرضى . حلت البيانات الأولية باستخدام برنامج الحزم الإحصائية للعلوم الإنسانية واستعرضت النتائج في شكل جداول للتكرارات والنسب المئوية . بينت النتائج أن هنالك تغيرات خلوية واضحة على خلايا الجهاز البولي للمرضى الذين يعانون من البلهارسيا البولية ، حيث سجلت 72 (48%) من الحالات الإلتهابية و3 (2%) حوول الخلية وتحول الخلية من سليمة إلى مريضة .

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Chapter One

Introduction

Chapter One

Introduction

1. Introduction

Cytology is the study of cells that are exfoliated from epithelial surfaces or shed from mucous membrane, or removed by physical means from various parts of the body, and cells that are found in Effusions. Diagnostic cytology is an art and science of the interpretation of the cells from human body (Bancroft JD. And Gunble M. (2002)) .Methods of preparation of cytology are rather simple than histology. The histological diagnosis is based on syntheses of entire evidence available rather than on changer in individual cell. Papanicolaou stain which has become the most popular stain for gynecological cytology, was originally develop to determine the changes that take place in squamouse epithelium of the female genital tract. In addition to Pap stain haematoxyline and Eosin, also Romanwesky stains can be used e.g. May Grun Giemsa (MGG) (Carleton's A. (1980)).Inflammation of the urinary tract due to *S.hematobium* may contribute to the propagation of the studies have shown the relationship between *S.haematobium* infection and the development of squamous cell carcinoma of the bladder (Badawi A F, et al 1992). Infection by *S.haematobium* - a member of this family - is particularly relevant due to its association to bladder cancer. In fact, while the disease can be

treated-in the sense that the parasites are killed - their calcified eggs can remain trapped in the bladder creating a chronic infection that is linked to the appearance of cancer. In fact, in regions where *S.haematobium* is endemic bladder cancer can be the most common cancer in men and the second in women, just behind breast cancer, accounting for as much as 30% of all cancer cases (Am J Pharmacol. (2006))

Rationale

Rationale:

Urinary schistosomiasis remains one of the increasing significant health issues. Symptoms and signs of the infection are quite affecting for both the health and the social status of individuals specially children. In the other hand the disease is basically associated with haematuria, which may lead to anaemia due to the chronic blood loss.

Study objective

General objective:

To detect cytological changes of urinary tract cells among patients suffering from schistosomiasis.

Specific objective:

- 1- To know the frequencies of each cytological change among schistomiasis patients.
- 2- To correlate these cytological changes with gender of patients and duration of the disease.

Chapter Two
Literature review

Chapter two

2. Literature review

2. 1 scientific background

2.1.1 Prevalence study of schistosomiasis in Sudan

Most epidemiologic studies regarding schistosomiasis in Sudan have been carried out in the Gezira-Managil area and in other central or northern areas of economic importance, while relatively few studies have been conducted in other parts of the country. The Upper Nile region is a swampy area and has been considered by the Sudanese authorities as being no endemic for bilharziasis(Amin MA, Omer AHS(1972)).

These data were based mainly on routine examination of urine in hospitals and dispensaries. Subsequently, suggested a 0.4%44% prevalence of *S.hematobium* infection in different areas of Southern Sudan .Since the 1970s, few observations on the prevalence of schistosomiasis in southern Sudan have been made (Montresor A,et al(2002)) .

2.1.2 Schistosomiasis

Schistosomiasis (informally known as Bilharzia) is a collective name of parasitic diseases caused by several species of trematode

belonging to the genus *Schistosoma*. Snails serve as the intermediary agent between mammalian hosts. Individuals within developing countries who cannot afford or obtain proper water and sanitation facilities are often exposed to water contaminated by the infected snails (Robbin and Cotran (2007)). Although it has a low mortality rate, schistosomiasis often is a chronic illness that can damage internal organs and, in children, impair growth and cognitive development. The urinary form of schistosomiasis is associated with increased risks for bladder cancer in adults. Schistosomiasis is the second most socioeconomically devastating parasitic disease after malaria (Robbin and Cotran (2007)). This disease is most commonly found in Asia, Africa, and South America, especially in areas where the water contains numerous freshwater snails, which may carry the parasite. Schistosomiasis affects almost 240 million people worldwide, and more than 700 million people live in endemic areas (Robbin and Cotran (2007)).

2.1.3 Pathophysiology

Penetration of the human skin occurs after the cercaria have attached to and explored the skin. The parasite secretes enzymes that break down the skin's protein to enable penetration of the cercarial head through the skin. As the cercaria penetrates the skin it transforms into a migrating schistosomulum stage. The newly

transformed schistosomulum may remain in the skin for two days before locating a post-capillary venule; from here the schistosomulum travels to the lungs where it undergoes further developmental changes necessary, for subsequent migration to the liver. Eight to ten days after penetration of the skin, the parasite migrates to the liver sinusoids: *S. japonicum* migrates more quickly than *S. mansoni*, and usually reaches the liver within eight days of penetration. Juvenile *S. inansoni* and *S. japonicum* worms develop an oral sucker after arriving at the liver, and it is during this period that the parasite begins to feed on red blood cells. The nearly-mature worms pair, with the longer female worm residing in the gynaecophoric channel of the shorter male. Adult worms are about 10 mm long. Worm pairs of *S. mansoni* and *S. japonicum* relocate to the mesenteric or rectal veins. *S. haematobium* schistosomula ultimately migrate from the liver to the perivesical venous plexus of the bladder, ureters, and kidneys through the hemorrhoidal plexus. Parasites reach maturity in six to eight weeks, at which time they begin to produce eggs. Adult *S. mansoni* pairs residing in the mesenteric vessels may produce up to 300 eggs per day during their reproductive lives. *S. japonicum* may produce up to 3,000 eggs per day. Many of the eggs pass through the walls of the blood vessels, and through the intestinal wall, to be passed out of the body in feces. *S. haematobium* eggs pass through

the ureteral or bladder wall and into the urine. Only mature eggs are capable of crossing into the digestive tract, possibly through the release of proteolytic enzymes, but also as a function of host immune response, which fosters local tissue ulceration. Up to half the eggs released by the worm pairs become trapped in the mesenteric veins, or will be washed back into the liver, where they will become lodged. Worm pairs can live in the body for an average of four and a half years, but may persist up to twenty years. Trapped eggs mature normally, secreting antigens that elicit a vigorous immune response. The eggs themselves do not damage the body. Rather it is the cellular infiltration resultant from the immune response that causes the pathology classically associated with schistosomiasis (Stothard, J. Russell; *et al.* (2005)).

2.1.4Diagnosis:

Contemporary diagnosis involves detection of parasitic antigens by Enzyme linked immune sorbent assay; all that is required from the patient is a blood sample. This screening method is highly effective. Microscopic identification of eggs in the urine is another way of arriving at a positive diagnosis. For urine examination should be performed if *S. haematobium* is suspected. Investigation of *S. haematobium* should also include a pelvic x-ray as bladder wall calcification is highly characteristic of chronic

infection. Tissue biopsy of the bladder for *S. haematobium* may demonstrate eggs when urine examinations are negative. The eggs of *S. haematobium* are ellipsoidal with a terminal spine. Antibody detection can be useful in both clinical management and for epidemiologic surveys (Graaff, Van De (2002)).

2.2 Anatomy of urinary system:

The urinary organs include the kidneys, ureters, bladder and urethra. The urinary system is a group of organs in the body concerned with filtering out excess fluid and other substances from the blood stream. The substances filtered out from the body in the form of urine. Urine is liquid produced by the kidney, collected in the bladder and excreted through the urethra (Mader, Sylvia S. (2004)).

2.2.1 Kidney:

The kidneys are two small organs situated near the vertebral column at the small at the back. They are bean shaped about 10cm long and about (6.4cm) wide. Each kidney has dark brown outer cortex and light brown inner medulla. Each kidney contain 1 .2million filtering unites called nephron. A nephron is the basic structural and functional unite of the kidney. Nephrons eliminate wastes from the body, regulated blood volume and pressure,

control levels of electrolytes and metabolites, and regulate blood pH. Nephron is composed of glomeruli and along tubule that has three parts, which are the proximal convoluted tubule (McCance, et al. (1994)).

2.2.2 Ureters, urinary bladder and urethra:

The ureters are two tubes that drain urine from the kidneys to the bladder. The urinary bladder is hollow, muscular and distensible or elastic organ that sits on the pelvic floor (superior to the prostate in the males). On its anterior border lies the pubic symphysis and, on its posterior border, the vagina (in females) and rectum (in males). The urethra is a muscular tube that connects the bladder with the outside of the body (Betz SA, et al. (1993)).

2.3 Histology of urinary system:

The renal pelvis, ureters, and bladder are lined by the highly specialized urothelium or transitional epithelium. This epithelium is about seven layers thick and composed of cuboidal cells. The superficial cells, which are larger with multiple nuclei, are referred to as umbrella cells. The trigone of the bladder in up to 50% of women and small number of men has been shown to contain squamous cells. Areas of mucin-producing epithelium may also be

present, including Brunnes nests. These are generally thought to be normal variants (Coleman DV. (1975)).

2.4 Pathphysiology:

2.4.1 Urinary tract infection and obstruction:

The site of infection may be either in the kidneys themselves (pyelonephritis) or in the urinary bladder(cystitis). Bacteria (as evidenced by positive nitrite dipstick finding for some organism), haematouria and pyuria. Renal obstructions can cause disease in one of two ways. They may either gradually raise the intra tubular pressure until nephrones necrosis and chronic renal failure ensues, or they may predispose the urinary tract to repeated infections. Obstructions may be located in either the proximal or distal urinary tract. Causes of obstruction include neoplasia (such as prostate/bladder carcinoma or lymph node tumors constricting ureter) acquired disease! Such as renal calculi or urethral structures) and congenital deformities of lower urinary tract (Droese M, Voeth C(1975)).

2.4.2Glomerular diseases:

Disorders or diseases that directly damage the renal glomerulous, which include:

2.4.2.1 Acute glomerulonephritis:

Pathologic lesion in the acute glomerulonephritis primarily involve the glomerulus is cytological examination show large inflamed glomerulus with a decreased capillary lumen (Epstein JI, et al (1998)).

2.4.2.2 Chronic glomerulonephritis:

Length glomerular inflammation, whether form renal disease or form an idiopathic cause may lead to glomerular scaring eventual loss of operational nephrones (Highman W, Wilson E (1982)).

2.4.3 Kidney stones:

It is solid concentration (crystal aggregation) of dissolved minerals in urine found inside the kidney or ureters. Calcium can aggravate the development of the kidney stones, since the most common type of stone is calcium oxalate. Other examples of Kidney stones include struvite (Magnesium, ammonium and phosphate), uric acid, calcium phosphate and cysteine (Kahan AV, et al (1980)).

2.5 Normal urine cytology:

Transitional epithelial cells are present in all urine specimens. In voided urine they occur singly or in the form of loosely cohesive

cluster or sheets. They vary considerably in size and the cytoplasm is opaque, granular or vacuolated. Renal tubular epithelium cells are rarely found in voided or catheterized urine, except in cases of renal transplant reaction or renal parenchymal disease (Koss LG, (1996)).

2.6 Cytopathological appearance of urine:

2.6.1 Benign changes:

In the normal situation, urine contains very few cells, the large umbrella cells 30-50 mm in diameter, polygonal in shape with two or more round to oval nuclei with fine granular chromatin. The cytoplasm abundant, usually blue in staining, deeper layers cells show a more round shape with single nucleus occupying about one third to one half of the cell, the chromatin is finely granules, often with small nucleus, the cytoplasm stain blue\green. In the presence of infection or inflammation, the cell show degenerative changes include, pyknosis and karyorrhexis and associated polymorphs, lymphocyte and cell debris may obscure cell details. Squamous metaplasia may occur in the bladder and the cell appearance is similar with that of the cervix. Columnar cells from the glandular elements of the urinary tract including renal tubule are also seen. Cytomegalovirus and polyoma virus are infection that may be identified by cell changes (Koss LG, (1992)).

2.6.2 Mild and severe dysplasia:

Mild dysplasia is characterized by normal or near normal thickness of the epithelium, usually less than seven layers. Slight increases in the number, slight crowding of the cell have nuclei with coarsely granular chromatin. In severe dysplasia the urothelium is strikingly abnormal (Murphy WM, et al (1981)).

2.6.3 Malignant change:

Transitional cell carcinoma is the most common type of tumors. Malignant urothelial cell will show enlarged, hyperchromatic nuclei with coarsely clumped chromatin almost opaque with little chromatin pattern discernible. The nuclei may show marked-pleomorphism with irregular shape. The cytoplasm is usually basophilic but can be eosinophilic (Koss LG. (1996)).

2.6.3.1 Adenocarcinoma :

is usually composed of typical cuboidal cells in a papillary formation with hyperchromatic nuclei and vacuolated cytoplasm. Squamous carcinoma in the bladder is rare and cytological difficult to distinguish from the urothelial carcinoma, however if keratinized cell particularly a nucleate cell, are seen this should be as a differential diagnosis (Koss LG. et al (1984)).

2.6.3.2 Papillary transitional cell carcinoma:

Papillary transitional cell carcinoma include non-invasive tumors that resemble papillomas, but are characterized by thicker urothelium .Slight abnormality in the architecting with occasionally broader papillae and greater cytological atypical than is seen in papillomas (Koss LG. et al (1984).

2.6.3.3 Squamous cell carcinoma of the bladder:

Squamous cell carcinoma present component of poorly differentiated transitional cell carcinoma. But it 2%to 3% of patient it is the principle primary tumor. The percentage is much high in the region of the world in which schistooma haematobium commonly infected the bladder and where over one half of bladder cancer is squamous in type. Many squamous cell carcinoma may occurring in the other of the regions are associated with squamous metaplasia, and occasionally with squamous metaplasia found in the bladder Diverticular. Most tumors are keratinizing; cytological the urine specimen contains keratinizing malignant epithelial cell. If the tumors are highly grade and if the keratinization is not readily apparent in malignant cell, they are often associated with atypical metaplatic squamous cells.

2.7 Lab diagnosis:

2.7.1 Urine samples:

Voided urine is usually submitted for cytological examination for the diagnosis of tumors or for monitoring patient with past history of tumors. Collection morning urine specimen has the advantage of highest cellularity, but with the disadvantage of marked cell degeneration. Specimen from the morning second voiding is usually the best. Three samples obtained on three consecutive days are diagnostically advisable (Raab SS, et al (1994)). The catheterization urine is often rich in desquamated cell which may be appearing in single or clusters either polyhedral or smaller elongated (Raab SS, et al (1982)). The bladder wash has been introduced on large scale for purpose of DNA analysis by flow cytometry in bladder cancer (Wiener HG et al)

2.7.2 Urine fixation:

Most cytological preparations are stained by papanicolaou method, and in order to optimally preserve cell detail, without, distortion, the smear must be wet-fixed rapidly before any air drying occurs. Papanicolaou is original fixative of equal parts of 95 per cent ethyl alcohol and ether has been largely abandoned, because of fire hazards associated with ether. Most that require fixation in which

smears made from the centrifuge sediment were put into 95% ethanol before drying, spray fixation, in which a fixative was sprayed onto the wet smear and modified two step fixations, in which centrifuge sediment was treated twice with alcoholic carbowax (Wojcik EM, et al (1999)). Fixatives before smears were prepared Epostis fixative for urine is best and use equal volume of fixative added to the urine.

To delay decomposition of urine use the following methods of preservation refrigeration, preservative, hydrochloric acid, Glacial acetic acid, Boric acid and other preservatives used include formaldehyde, toluiden, and thymol (Wojcik EM, et al (1997)).

2.7.3 Cytological staining:

Consistency and reliability in staining are the cornerstones of cytological interpretation. Subtle cell appearances are constantly being compared. And assessed during screening, the universal stain or cytological preparation is the papanicolaou stain. Harris haematoxlen is the optimum nuclear stain; EAO gives the subtle range green, blue and pink hues to the cell cytoplasm. Papanicolaou stain which has become the most popular stain for gynecological cytology was originally develop to determine the

changes that take place in the squamous epithelium of the female genital tract .Good definition of nuclear details. Cytoplasm transparency and indication of cellular differentiation. But no specific staining of nuclear and cytoplasm proteins. Staining solution is stable and the haematinmetal relationship is hard to control (Bancroft JD. And Gunble M. (2002)).

Examination of smears for the inflammatory cells found that inflammatory cells were detected in all cases while meta plastic cells were found in 17 (14.2%) of the study group (Hassan Sidigg , May (2007)

Chapter Three

Material and method

Chapter Three

Material and method

3.1 Study design:

This was descriptive study aimed to detect the cytological changes in urinary tract cells of patient suffering from schistosomiasis.

3.2 Study area:

This study was conducted in Khartoum state (Goz Elrimela) during the period from June2010 -Octobers 2014.

3.3 Study population:

About 75 samples urine were taken from (all ages and both sex) patients suffering from schistosomiasis and remaining 75 samples were taken from health population as control. Questioner containing essential patient identification data was used. .

3.4 Sample size:

About 150 urine samples were included in this study.

3.5 Sample collection and preparations:

Full voided urine samples were collected from patients complaining from schistosomiasis and healthy people. Each urine

sample was centrifuged at 1500 RPM for 5 minutes; the supernatant was discharged from the tube, and the sediment was placed on the one end of the slides, and then smears were made using a spreader and immediately fixed in 95% alcohol for 15 minutes then stained by Papanicolaostain.

3.6 Staining procedure:

Smears were treated with 95% alcohol for minute then treated with 70% alcohol for two minutes, rinsed in distilled water for three minutes and then treated with filtered harris haematoxyline for four minutes, rinsed in tap water for one minute and treated with 1% acid alcohol for three Seconds, then in tap water for two minutes treated with ammoniated water for 1minute , then rinsed in tap water for three minutes , treated in 70% alcohol for two minutes, then treated with 95% alcohol for two minutes, after that they were stained with OG6 solution for two minutes, -then treated with 95% for two minutes treated with 95% alcohol for two minutes and then stained with EA 50 solution for three minutes then treated with 95% alcohol for two minutes and then treated with absolute alcohol for three minutes, and after that they were treated with xylene for five minutes, finally they were mounted in DPX.

3.7 Result interpretation:

All quality control measures were adopted in this study. Smears were initially examined by light microscope using x10 then x40. For the assessment of changes in cells appearance which encounter in the smears, the characteristics of these cells were compared with the illustrated photographs in atlas comprehensive cytopathology (1991).

3.8 Statistical analysis:

All information about the study population was entered a computer, as well as the obtained results, the data was analyzed using SPSS program. Frequencies, chi-square, cross tabulation values were calculated.

3.9 Ethical consideration:

Each individual was told about importance the research during the interview and all them were agree to participate in this study.

Chapter Four

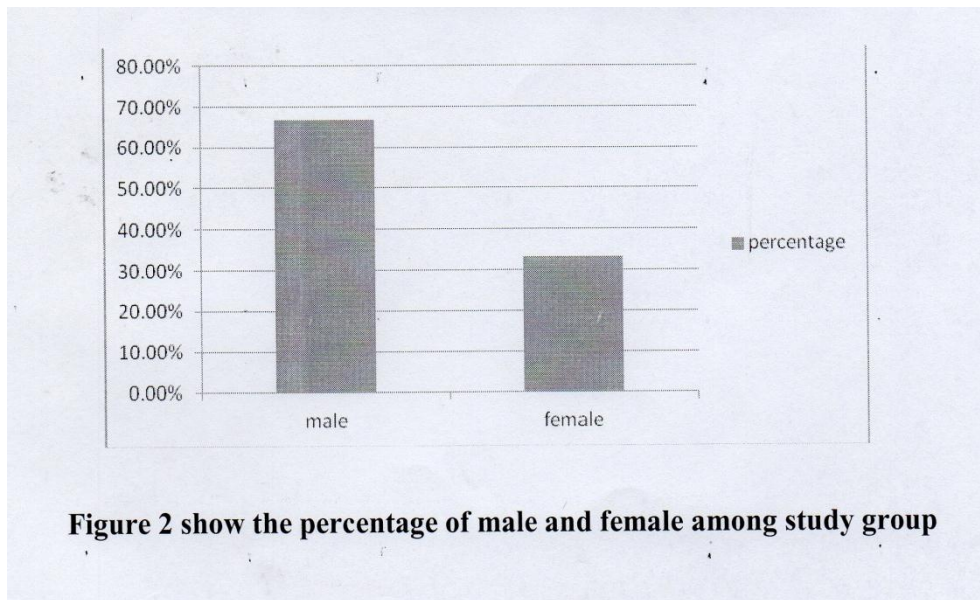
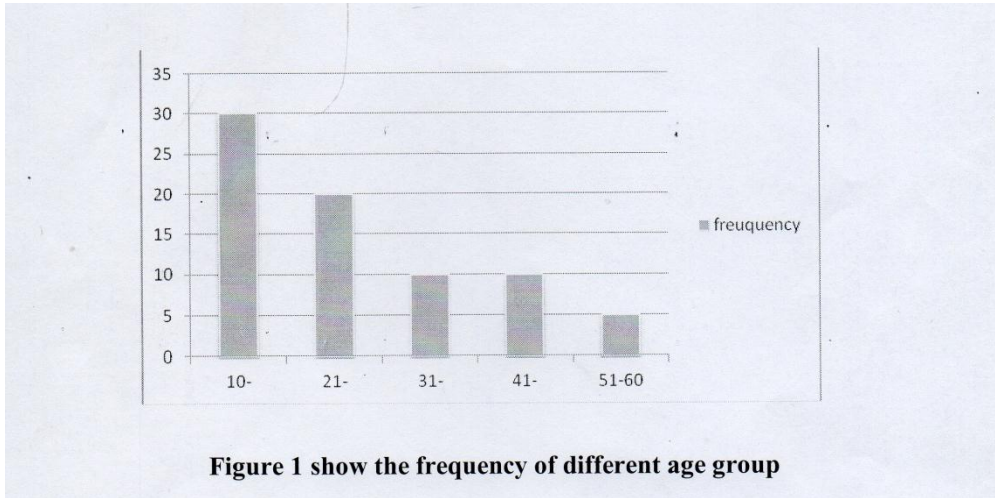
Result

Chapter four

Results

4. Result:

The age of the study group range between 12 to 56 years, the mean range was 24.2 years and standard deviation is 9.6. There was different age groups; most of patients were within the age group (10-20), (21-30), (31-40), (41-50), (51-60) and the frequency of this groups is 30(20%), 20(13.3%), 10 (6.7%) 10(6.7), 5(3.3%) respectively (see in figure1). The frequency of male is 100(66.7%) and female is 50(33.3%) (See figure2). With regard to cytological finding 75(50%) of patients were negative, the cytological changes with notice that the other study group 72(48%) reported with inflammatory changes and 3(2%) show metaplastic changes (show in the figure3). The inflammatory change of age group and duration of infection by month show (5-20), (21-36) are 46% (n69), 2 % (n3) respectively. And the metaplastic change of age group (5-20), (21-36) are 0 % (n0) 2% (n3) respectively (show table 1). The negative results among male and female are 31.3%(n47) , 18.7%(n28) respectively , the inflammatory changes are 34.7(n52) , 13.3(n20) and the metaplastic changes are 1.3(n2) , .7(n1) respectively (show table 2).



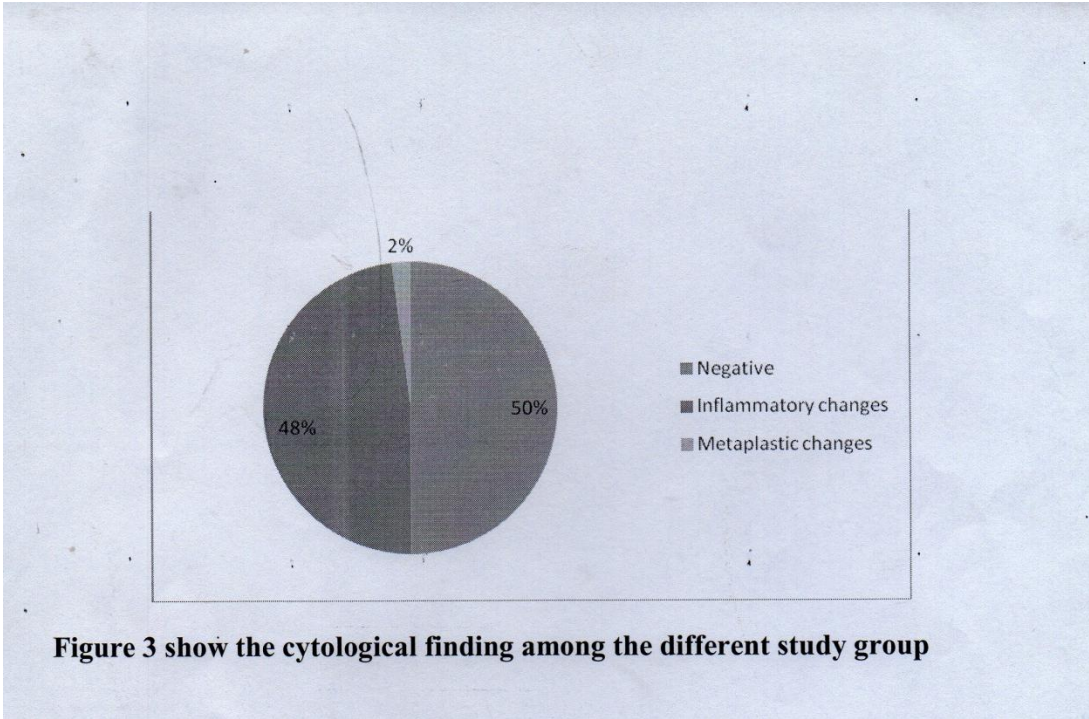


Figure 3 show the cytological finding among the different study group

Table (1) show cytological changes among age group of infection of duration by months by using crosstabulation.

Duration of infection by months	Inflammatory changes	Metaplastic changes
5-20	46%(n69)	0%(n0)
21-36	2%(n3)	2%(n3)

P. value (0.004) (significant).

Table (2) show cytological changes among male and female

Sex	Negative result	Inflammatory changes	Metaplastic changes
Male	31.3(n47)	34.7%(n52)	1.3(n2)
Female	18.7(n28)	13.3%(n20)	.7(n1)

P. value (0.005) (significant).

Chapter Five

Discussion

Chapter Five

Discussion

Referring to the results it can be clearly observed that schistosomiasis in Sudan seems to be a normal practice. The cytological results showed the clear change in cells of the different urinary tract system comparing to the normal cells which ensured - the negative effects of schistosomiasis on the urinary tract system. Almost the majority of respondents were suffering from inflammation and metaplastic change. Also these cytological changes in urinary tract cells appears more clearly when we used Papnicolau stain, this indicated that Papanicolau stain is more convenient.

Our findings from the cytological changes of urinary tract cells affected by schistosomiasis among different age groups shown the inflammatory changes in 72(48%) were detected among cases. Furthermore, the metaplastic changes in 3(2%) were detected among cases, and these finding are similar to those of (Hassan elsidigg May (2007).

Chapter Six

Conclusions and Recommendation

Chapter Six

Conclusions and Recommendation

6.1 Conclusions: This study confirms previous findings that urine cytology provides accurate information regarding the inflammatory and metaplastic changes. There are significant differences in cytological changes of schistosomiasis among different age group. Our data suggest that cytological findings may predict bladder cancer.

8.2 Recommendation:

Further researches are needed to illustrate the effect of urinary schistosomiasis motherly tract cells. Further study is needed with large sample size, using more advance techniques and types of stains to attend more accurate results.

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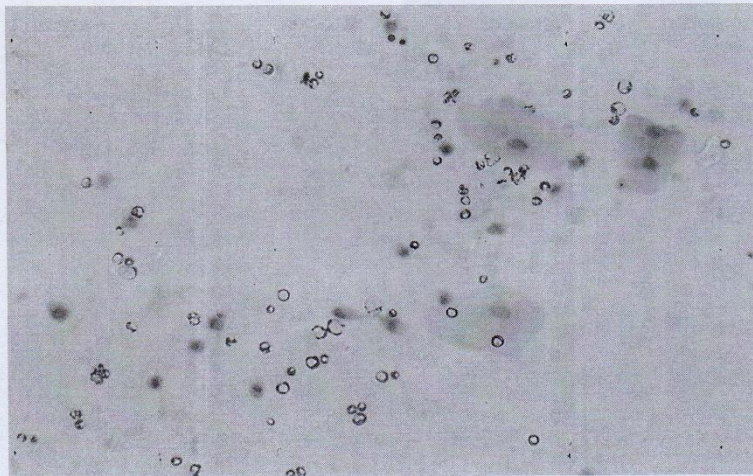
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Appendices

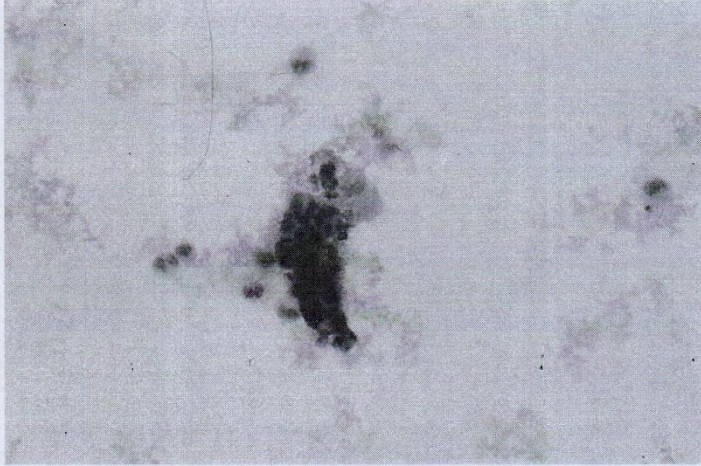
Appendices



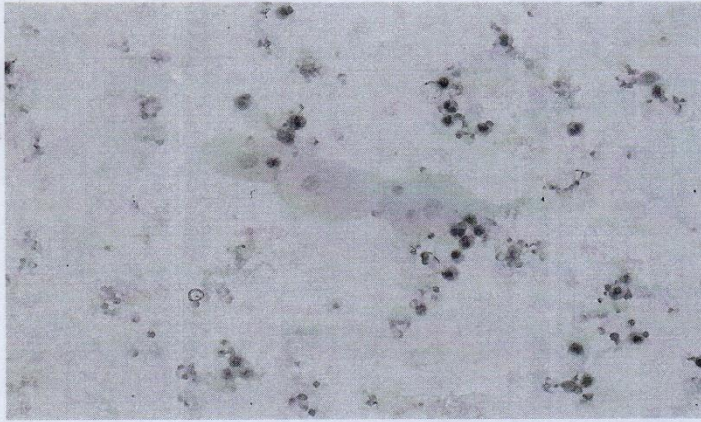
Appendix (1): showed ova of schistosoma hemtabium in urine cytology smear stain by papanicoleuo^{ed} stain (X40).



Appendix (2): showed inflammatory cells in urine cytology smear stain by papanicoleuo stain (X40).



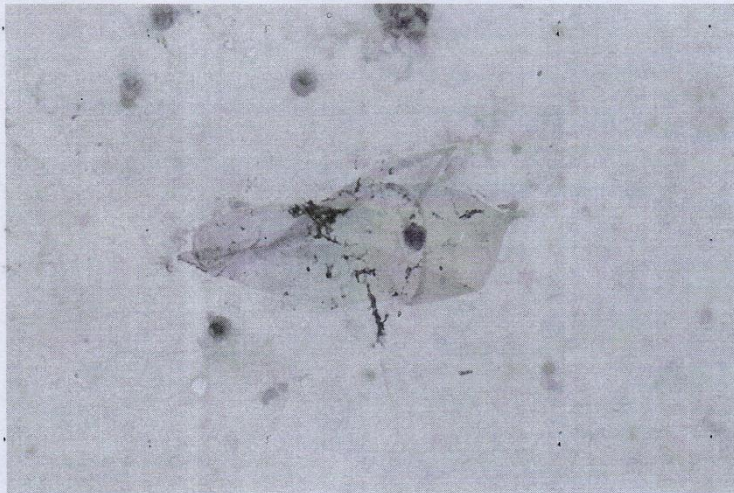
Appendix (3): showed meracidium in urine cytology smear stain by papanicolaou stain (X40).



Appendix (4): showed inflammatory cells with normal transitional epithelial cells in urine cytology smear stain by papanicolaou stain (X40).



Appendix (5): showed inflammatory cells with normal transitional epithelial cells in urine cytology smear stain by papanicoleuo stain (X40).



Appendix (6): showed ova of schistosoma hemtabium in urine cytology smear stain by papanicoleuo stain (X40).

Appendix (7) Perinuclear halo in urine cytology smear by pap stain

