Parasitic infections encountered by a cytopathologist: An institutional experience



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ABSTRACT

Background: Parasitic infections are one of the leading causes of morbidity and mortality worldwide. Cytological examination can be of great help in providing economical, accurate, and fast diagnosis. Aims and Objectives: The aim of the study was to analyze the cytomorphological features of various parasites on fine needle aspirates and vaginal smears. Materials and Methods: This study was a retrospective study of 5.5 years during the time period from July 2015 to December 2020. The study included cases which were already diagnosed as having or suspicious of various parasitic infections on cytological examination. Results: There were total of 37 specimens comprising of 28 fine needle aspiration cytology (FNAC) smears and nine vaginal smears. The parasites on FNAC included 17 cases of cysticercus, eight cases of filariasis, two cases of hydatid disease, and one case of leishmaniasis. All the nine vaginal smears were interpreted as trichomonas vaginitis. Conclusion: Cytomorphological examination helps in early definitive diagnosis and exclusion of various parasitic infections. Diagnosis is important as most of these parasites are treatable and have several systemic ramifications.

Key words: Fine needle aspiration cytology; Parasitic infections; Vaginal smears

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INTRODUCTION

Parasitic infections are serious public health problem worldwide. India is the second most populated country in the world with varied religious practices, food habits, literacy rates, and economic conditions. According to the WHO, India is home to the largest number of cases of major parasitic diseases. India accounts for nearly one-half of the world's prevalent cases of visceral leishmaniasis, one-third or more of the prevalent cases of cysticercosis and roughly one-quarter of the world's ascariasis and hookworm cases. Cytopathologist plays an important role in interpreting cytomorphological changes and early diagnosis.

In the literature, studies of parasite on cytology encompass mostly case studies and small case series.⁴ We present a detailed cytological study of 37 cases with their clinical correlation.

Aims and objectives

To analyze the cytomorphological features of various parasites on fine needle aspirates and vaginal smears.

MATERIALS AND METHODS

This is a retrospective study of 5.5 years duration from July 2015 to December 2020 conducted in Pathology Department of Integral Institute of Medical Sciences and Research, Lucknow, India.

Inclusion criteria

Cases which were already diagnosed as having or suspicious of various parasitic infections on cytological examination were included in the study.

The study included 37 specimens comprising of 28 fine needle aspiration cytology (FNAC) smears and nine vaginal

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smears. The clinical details were noted from the requisition forms. FNAC of superficial lesions was performed using 22 gauge needle attached to a 10 cc plastic syringe that was mounted on a handle for single hand grip. Deep lesions were aspirated by 23 G lumbar puncture needle under radiological guidance. The smears of the aspirated material were made. In case of cystic swelling, cytospin preparations of aspirated fluid were made. Few slides were spray-fixed with 95% alcohol and stained with hematoxylin and eosin stain (H and E), other slides were air dried, fixed in methanol, and stained with May Grunwald Giemsa stain. Special stains such as Ziehl-Neelsen stain were used whenever required. The vaginal smears were received from gynecology department with fixative in coplin jars. They were stained with Pap stain.

Cytopathological findings were correlated with histopathological diagnosis wherever available. Ethical clearance was obtained from Institutional Ethics Committee (letter no: IEC/IIMS and R/2020/03) in the month December 2020.

RESULTS

There were total of 37 specimens comprising of 28 FNAC smears and nine vaginal smears. The clinical and cytological features of all patients were tabulated (Table 1).

The parasites on FNAC included 17 cases of cysticercus, eight cases of filariasis, two cases of hydatid disease, and one case of leishmaniasis. All the nine vaginal smears were interpreted as trichomonas vaginitis.

Out of 17 cases of cysticercus 11 cases showed definitive evidence of cysticercosis in the form of bladder wall fragment with micronuclei (Figure 1a). All the cases showed presence of mixed inflammatory infiltrate in

varying proportion comprising of eosinophils, neutrophils, lymphocytes, histiocytes, epithelioid cell granuloma, and multinucleated giant cell. No hooklets or scolices were seen. In remaining 6 cases, no cytological evidence of parasite was seen. They were diagnosed on histology (Figure 1b). All the patients except one presented with single, firm, and non-tender subcutaneous nodules. One patient had multiple nodules. The majority of lesions were located on extremities. In majority of cases, aspirate was clear fluid while in few it was purulent.

All eight cases of filariasis showed presence of microfilaria and inflammatory cells against a fluid background on cytology (Figure 2). Peripheral blood smear does not show microfilaria; however, eosinophilia was present. The sites were axillary and inguinal lymph node, breast, thyroid, and scrotum. Both the cases of hydatid were aspirated from liver. The aspirates were blood mixed and necrotic. The smears showed protoscolices, hooklets, and fragment of laminated membrane on a background of inflammatory cells and necrotic debris (Figure 3a). Histological examination in one of the cases showed acellular laminated membrane with protoscolices (Figure 3b).

One case of leishmaniasis was found in splenic aspirate and showed extracellular amastigote forms. The vaginal smears showed scattered pear-shaped organism along with mature squamous cells and neutrophils (Figure 4).

DISCUSSION

Parasites causing diseases in human can be broadly classified in two groups: Helminths and protozoa.5 Protozoa are single-celled microorganism consisting of trichomonas, entamoeba, and leishmania etc., while helminths are multicellular worms such as echinococcus, taenia, and ascaris. In India poverty, unhygienic living

S. No.	Diagnosis	No of cases	Age (years)	Sex ratio (M: F)	Clinical Features	Procedure	Site	Material	Cytology Feature
1.	Cysticercosis	17	16–47	1:1.12	Subcutaneous swelling	FNAC	Arm, chest, abdominal wall, neck	Clear/ purulent	Bladder wall, granuloma, Eo, N, L, Epithelioid cells
2.	Filaria	8	22–59	3:1	Fever, swelling	FNAC	L.N., breast, thyroid, scrotum	Fluid/ blood mixed	Microfilaria, Eo
3.	Hydatid cyst	2	20–36	1:0	Abdominal pain	Guided FNAC	Liver	Blood mixed/ pus like	Acellular laminated membrane
4.	Leishmania	1	39	1:0	Fever Splenomegaly	Guided FNAC	Spleen	Blood mixed	LD bodies, L
5.	Trichomonas vaginitis	9	28–45	0:1	Greenish-yellow Discharge	Pap smear (Manual Scrapping)	Vagina	Recieved fixed slides	Pear shaped flagellated parasite

conditions, contaminated water and food, animal rearing practices, and poor personal hygiene lead to spread of parasitic infections.² *Trichomonas vaginalis* is a sexually transmitted infection and the most common parasite encountered in routine cytology.⁶ Cytopathologist can play an important role in diagnosis of superficial and deep inflammatory swellings caused by parasites such as filaria, echinococcus, cysticercus, and leishmania by FNAC and exfoliative cytology.⁷

In this study, cysticercosis was the most common parasitic infestation. Cysticercosis in humans is caused by larval stage of Taenia solium. Humans are definitive host while pigs are intermediate host. However, humans can become intermediate host by consumption of water and raw vegetables contaminated by eggs or by ingestion of larvae in undercooked pork.8 Larvae after evagination in small intestine can pass into human bloodstream and lymphatic system and then establish into skeletal muscle, subcutaneous tissue, central nervous system (neurocysticercosis), eye, lung, liver and occasionally heart, pancreas, and thyroid.9 Neurocysticercosis is one of the main causes of epilepsy in India. 10 Demonstration of parasitic bladder wall fragments, calcospherules, scolex, and hooklets helps in cytomorphological diagnosis of cysticercosis.¹¹ In our study, parasitic bladder wall fragments

a

Figure 1: (a) Fine needle aspiration cytology smear demonstrating bladder fragment (Giemsa $100\times$). (b) Cysticercus cellulose (Haematoxylin and eosin $400\times$)

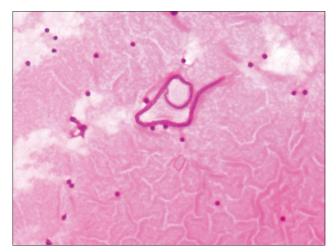


Figure 2: Microfilaria in a fluid background (Haematoxylin and eosin 100×)

were demonstrated on 11 cases. Rest six cases showed presence of mixed inflammatory cells and were reported as suspicious of parasitic infection and were confirmed by follow-up histopathological examination.

Filariasis is a mosquito-borne disease caused by *Wuchereria bancrofti* and *Brugia malayi*. According to the WHO, filariasis is the leading cause of long-term physical disability.¹² The chronic manifestations of disease are lymphedema, elephantiasis, and hydrocele. Human is the definitive host and mosquito is the intermediate host. The sites commonly involved are lungs, effusion fluids, epididymis, lymph node, thyroid, skin, and soft tissue.¹³ On cytology identification of microfilariae, fragments of adult worms or ova facilitate the diagnosis of filariasis in mass lesions.¹⁴ In the present study, the smears revealed microfilaria, lymphocytes, and histiocytes on a fluid background.

Hydatid disease or echinococcosis is predominantly caused by larva of *Echinococcus granulosus*. Dog is definitive host and sheep is intermediate host. Humans become incidental host by consumption of eggs in fecal-matter of infected canine. The most common site for hydatid disease is liver (70%) followed by lung (20%) and other organs (10%) such as

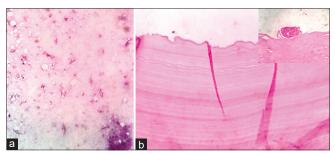


Figure 3: (a) Fine needle aspiration cytology smears showing hooklets (Haematoxylin and eosin (H and E) 400×), (b) Acellular laminated membrane in hydatid cyst. Inset showing scolex (H and E 100×)

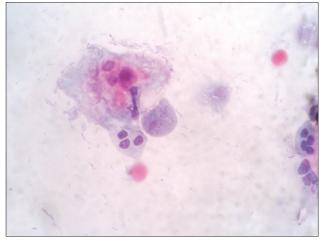


Figure 4: Trichomonas in vaginal smear (PAP 400x)

spleen, muscles, kidney, brain, and bone marrow.¹⁵ Hydatid disease remains asymptomatic for long durations and later may cause hepatomegaly, jaundice, cholangitis, and rupture. Cytodiagnosis of hydatid disease requires demonstration of laminated membrane, hooklets, scolices, and calcified spherules.¹⁶ In our study, the smears showed multiple scolices and scattered hooklets. FNAC as a diagnostic modality was considered unsafe as spillage of cyst contents could lead to acute anaphylactic reaction; however, none of our cases had any adverse reaction. Kaylap et al., also reported no complications in a study of 22 cases of primary subcutaneous cyst.¹⁷

Leishmaniasis is caused by a protozoan parasite Leishmania and transmitted to human by the bite of sandfly. The three main forms of disease are visceral (kala-azar), cutaneous, and mucocutaneous leishmaniasis. Clinical features of visceral leishmaniasis are fever, splenomegaly, hepatomegaly, weight loss, and anemia and are fatal if left untreated. Demonstration of amastigote form in tissue remains the gold standard of diagnosis. Bone marrow smears have sensitivity of 60–85% while splenic aspirates have sensitivity more than 90%. 19

Trichomoniasis is one of the most common curable nonviral sexually transmitted infections in the world caused by *T. vaginalis*, a flagellated parasitic protozoa infecting the urogenital tract of both women and men.²⁰ The majority of infected patients are asymptomatic.²¹ Symptomatic women have foul smelling yellow-green discharge, dysuria, and abdominal pain while symptoms in men include urethral discharge and dysuria.^{21,22} Identification of trichomonads on Pap smear is diagnostic in high prevalence settings such as STD clinics and has specificity of 95% and sensitivity of 60%.²³

Limitations of the study

This is a single center study, so cytological features of only few parasites were analyzed, subject to prevalence of parasites in the area. Multicenter studies may be planned to study the spectrum of cytomorphological features of various parasitic infections.

CONCLUSION

In a developing country like India where sophisticated diagnostic methods are not available to entire population, cytological samples obtained by fine needle aspiration and exfoliative sources are imperative in diagnosis of parasitic infections and preventing fatal complications.

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Authors Contribution:

PJ- Concept and design of the study, literature search, data acquisition, interpreted the result, reviewed the literature, manuscript preparation, and prepared first draft of study; JI, NSK, PT, MS, and MA- Concept, literature search, data analysis, manuscript editing, and manuscript review

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