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Incidental Detection of Microfilaria in Clinically Unsuspected Patients: A Study of 21 Cases

KEYWORDS: Bone marrow, Endemic areas, Microfilaria, Peripheral smear, Cytology, Histopathology.

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ABSTRACT *Background: Filariasis is a vector borne disease and has been a major public health problem in India next only to malaria. Filariasis in India is caused by two closely related nematode worms–Wuchereria bancrofti and Brugia malayi. Aim: To assess status of filariasis in clinically unsuspected cases in endemic areas. Materials and Methods: A total of 21 cases, with age ranging from 4-72 years, were included in the present study. All cases did not have any clinical evidence or suspicion of Filariasis. Results: Of the total 21 cases, filariasis was detected in the peripheral smear and bone marrow samples (7 cases), cytology (11 cases) and histopathology samples (3 cases). Out of 7 cases of microfilariasis in hematological samples, 4 cases were in peripheral smears, 2 cases in bone marrow and in 1 case of microfilaria were detected both in the peripheral smear as well as the bone marrow. In cytology out of 11 cases, 3 cases presented with subcutaneous swelling, 2 cases with thyroid swelling, 4 cases with breast lump and 2 cases as lymphadenopathy. In histopathology out of 3 cases, 1 case was in the lymphnode and 2 cases in the epididymis. Eosinophilia was present in 16(76.2%) out of 21 cases in peripheral smear examined. Conclusions: Our study emphasized that keen search for parasites in every sample especially in endemic areas where patients present with fever, lump and with or without eosinophilia should always be done. This will help to diagnose patients in the acute phase of filariasis and to avoid the complication of the chronic phase besides starting the treatment as early as possible.*

INTRODUCTION

Filariasis is a vector borne disease and has been a major public health problem in India next only to malaria. Filariasis in India is caused by two closely related nematode worms - Wuchereria bancrofti and Brugia malayi. They are transmitted by the bite of the Culex mosquito. It is endemic in tropical countries such as India, China, Indonesia, parts of Asia and Africa affecting over 120 million people in 80 countries. [1]This problem is increasing every year due to gross environmental mismanagement. In India the areas affected include Uttar Pradesh, Bihar, Jharkhand, Andhra Pradesh, Orissa, Tamil Nadu, Kerala and Gujarat.[2]The disease mainly involves the lymphatic system of the body. Different literature reveal that microfilariae have been found in various locations including thyroid nodule, skin and soft tissue swelling, epididymis, breast, salivary gland, cervicovaginal smear, ovarian cyst, urine, lymph node, and effusion fluids (pleural, pericardial, peritoneal), laryngeal and pharyngeal brushings, lung, hydrocele fluid, bone marrow, bronchial washing, retroperitoneal tissue, brain aspirates and joint aspirates.[3-5]

MATERIALS AND METHODS:-

The study was conducted in the Department of Pathology, Rohilkhand Medical College and Hospital, Bareilly, Uttar Pradesh. A total of 21 cases, with the age ranging from 4-72 years with no clinical suspicion of filariasis were included in this study. All cases with a clinical suspicion of filariasis were excluded out from the study. Peripheral smear was examined in all the cases. In cystic lesions, the cyst content was aspirated and the smears prepared from the cyst fluid after cytocentrifugation were examined along with the aspiration performed from the cyst wall. Peripheral smears were fixed by air dry method and stained with Leishman stain. FNAC smears were wet fixed immediately in equal volume of Ether and 95% Alcohol and stained by Haematoxylin and Eosin stain and Papanicolaou stains, where as air dried smears were stained with Lieshman-Giemsa stain. For histopathological examination tissues were fixed in 10% buffered formalin. After routine processing Haematoxylin and Eosin stain was used to stain the slides.

RESULT: - In the present study 21 cases of microfilaria were included, out of which 7 cases were haematological samples, 11 cases in cytology and 3 cases in histopathology. Out of 7 cases of haematological samples, 4 cases were in peripheral smears, 2 cases in bone marrow smears and in 1 case microfilaria was detected in both the peripheral smear as well as the bone marrow smears. In cytology out of 11 cases, 3 cases were presented as subcutaneous swelling, 2 case as thyroid swelling, 4 cases as breast lump and 2 cases as lymphnadenopathy. In histopathology out of 3 cases, 1 case presented with lymphadenopathy and 2 cases as epididymal cysts. In all the twenty one cases there were no clinical suspicions of filariasis. Peripheral smears were examined in all the cases, of which 16(76.2%) cases were associated with Eosinophilia. The cases showed age variation from 4-72 years. [Table 1]

Microscopic examination of Fine needle aspiration cytology smear of breast swelling showed sheathed microfilaria along with small cluster of ductal cells. Whereas aspirate from lymphnode tissue showed microfilariae in the background of mixed population of lymphoid cells in various stage of maturation. In histopathology, H & E stained sections showed microfilaria in lymphatic of infiltrating ductal carcinoma of breast and epididymal cysts. Whereas Leishman and Giemsa stained smears of bone marrow showed erthyroid hyperplasia with microfilaria. [Figure1a, b, c, d, e, f]

Fine needle aspiration cytology smear showed different types of inflammatory cell response around microfilaria in-

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cluding giant cells, polymorphs, plasma cells and eosinophils. Where as in histopathology H & E stained sections showed eosinophilic infiltration, giant cell, fibrosis, necrosis and granuloma around the microfilaria. [Figure 2a, b, c, d, e, f]

Table 1:-Showing distribution of cases according to clinical presentation and peripheral smear finding.

S.No	Age/ years	Sex	Specimen	Diagnosis	Blood (Eosinophilia)
1	30y	M	Peripheral smear	Microcytic hypochromic anaemia with microfilaria infection	Present
2	12y	M	Peripheral smear	Normocytic hypochromic anaemia with microfilaria infection	Present
3	5y	F	Peripheral smear	Normocytic hypochromic anaemia with thrombocytopenia with microfilaria infection	Absent
4	35y	M	Peripheral smear	Leucoerythroblastic reaction with thrombocytopenia,anaemia and presence of microfilaria	Absent
5	50y	M	Peripheral smear / Bone marrow	Erythroid hyperplasia with micronormoblastic reaction with microfilaria	Absent
6	35y	M	Bone marrow	Hypoplastic marrow with microfilaria	Absent
7	5y	F	Bone marrow	Hypocellular bone marrow with microfilarial infestation	Absent
8	28y	F	FNAC from right cubital fossa	Parasitic cyst(Microfilaria nodule)	Present
9	4y	M	FNAC from forearm swelling	Cystic lesion with microfilaria	Present
10	25y	M	FNAC from thyroid swelling	Colloid cyst with microfilaria infestation	Present
11	82y	F	FNAC from thyroid swelling	Colloid nodule with Microfilaria	Absent
12	18 y	M	FNAC from cervical Lymphnode	Microfilaria with hemorrhagic cyst	Present
13	7y	F	FNAC from cervical lymphnode	Reactive lymphadenitis with microfilaria infestation	Present
14	25y	M	FNAC from cervical swelling	Lymph cyst with microfilaria	Absent
15	40y	F	FNAC from breast lump	Granulomatous mastitis with microfilaria	Present
16	24y	F	FNAC right breast lump	Benign breast disease with microfilaria	Present
17	30y	F	FNAC from right breast swelling	Microfilaria cyst	Present
18	35y	F	FNAC from right breast swelling	Granulomatous inflammation with microfilaria	Present
19	40y	F	Histopathology from left axillary lymphnode	Metastasis ductal carcinoma of breast with microfilariasis	Present
20	16y	M	Histopathology from right epididymis	Filariasis infestation	Present
21	32 y	M	Histopathology from left epididymis	Filariasis infestation	Present

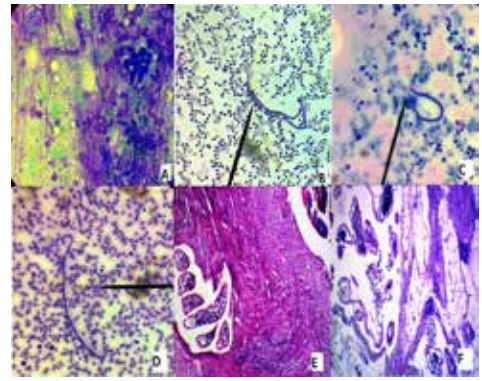

Figure: 1A- FNAC smear of breast swelling showing sheathed microfilaria (Leishman and Giemsa x400) 1B:- FNAC smear from cystic cervical swelling showing lymphocytes and microfilaria. (L and G x100) 1C:-Bone marrow smear showing microfilaria with marrow cells with erthyroid hyperplasia (L and G x100) 1D:- Leishman and Giemsa stain smear from thyroid swelling show microfilaria with pigment laden macrophages over a background of thin colloid (100x).1E:- H and E stained section showed microfilariae in epididymal cyst (400x). 1F:- Section show microfilaria in lymphatic of infiltrating ductal carcinoma of breast (H and E x 400).

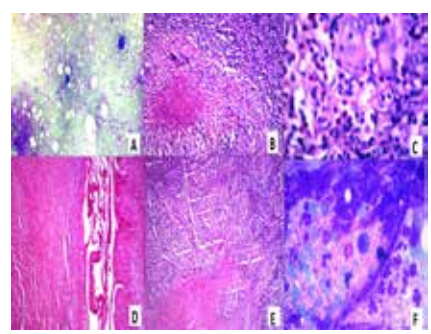

Figure 2A:- FNAC smear show giant cell and inflammatory cells around microfilaria.(L and G x100)1B:-Granuloma and giant cell around the microfilaria in histopathology.(H and E x400) 1C:-H and E stained section showing eosinophilic response around microfilaria.(400x) 1D:- Histopathology section showing fibrosis around microfilaria.(H and E x 400) 1E:- Section showing necrosis and fibrosis in epididymal cyst due to microfilaria (H and Ex 400).1F:- Leishman and Giemsa stain smear showing polymorphs and plasma cell around microfilaria.(400x).

DISCUSSION: - Wuchereria bancrofti is a human roundworm parasite, which is the major cause of lymphatic filariasis. Out of the three parasitic worms, Wuchereria bancrofti is more prevalent than Brugia malayi and Brugia timori to infect the lymphatic system. [6] Ancient Greek and Roman writers noted that there are similarities between the enlarged limbs and cracked skin of infected individuals to that of elephants. Since then, this condition has been commonly known as elephantiasis. [2, 6]

Lymphatic filariasis affects more than 90 million people worldwide and has been identified by WHO as the second leading cause of permanent and long term disability after leprosy.[7] Most people infected with Bancroftian filariasis are asymptomatic, since the development of symptoms relates to the increasing numbers of worms. The clinical course of lymphatic filariasis includes three distinct phases.

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Asymptomatic phase showing high microfilaremia infection but the individuals are asymptomatic. Second phase is the inflammatory phase in which the worms disrupt the flow of the lymph, causing lymphedema and individuals present with fever, chills, skin infections, painful lymph nodes, and tender skin of the lymphedematous extremity. Other symptoms that may be observed include orchitis and epididymitis. The third phase is chronic lymphedema disease or irreversible lymphedema in which the individuals present with lymph varices, lymph scrotum, hydrocele, chyluria and elephantiasis. Microfilariae are normally not present in this phase.

Filariasis has two stages- human stage and mosquito stage. Humans are the definitive host whereas mosquitoes are the intermediate host. In the human host, the infective third stage larvae enter during blood meals when mosquito punctures the skin, then they migrate to the peripheral lymphatic where they develop into adult male and female worms. The adult worm can survive for several years. Female worm produces a large number of microfilariae, which circulate in the blood and may be ingested by a feeding vector mosquito. After ingestion, the microfilariae lose their sheaths in the stomach and migrate to the thoracic muscle where they develop into the infective larvae over a period of 6-14 days. Finally larvae then migrate to the head and proboscis of the mosquito which infects the host during a bite. Adult worms of Wuchereria bancrofti are found in the lymphatic vessels and lymph nodes of human beings only, whereas larval forms (microfilaria) circulate in the peripheral blood. The major vectors are the Culex mosquitoes in most of the urban areas and Anopheles mosquitoes in the rural areas. Brugia malayi microfilariae are smaller, possess secondary kinks and contain nuclei while the Wuchereria bancrofti has a smooth curve and the tip is free of the nuclei. [8, 9]

Different laboratory tests are available to diagnose microfilaria including peripheral blood smear, immune chromatographic test, quantitative buffy coat, Ultrasonography, lymphoscintigraphy and histopathology. Fine needle aspiration cytology is not applicable for routine diagnosis of microfilaria but can be used for species identification on the basis of morphology.

Different studies show that microfilaria have been found in association with benign as well as malignant lesions. Malignant lesions include Ewing's sarcoma, Non Hodgkin's lymphoma, squamous cell carcinoma of maxillary antrum, craniopharyngioma of the third ventricle, transitional cell carcinoma of the urinary bladder, follicular carcinoma of the thyroid, Seminoma of undescended testis and Metastatic axillary lymphnode in ductal carcinoma of breast. [2, 10] Other sites associated with microfilariasis include thyroid, skin, soft tissues, epididymis, breast, lung, lymph nodes, and effusion fluids. [11]

In the present study microfilaria was detected in 7 cases of blood and bone marrow samples, out of which in 4 cases were peripheral smears, 2 cases was bone marrow smears whereas in 1 case microfilaria detected both in the peripheral smear as well as the bone marrow. Pradhan et al also reported microfilaria associated with anaemia in seven cases. [12] Similarly in our study all 7 cases showed microfilaria with associated anaemia.

Two cases of thyroid swelling on fine needle aspiration cytology showed microfilaria along with degenerated RBC and macrophages in a background of thin and thick colloid. Similarly Gupta et al and Varghese et al have also reported one case of microfilaria with coexistent colloid goitre. [11, 13] Whereas Mishra et al reported microfilaria in 3 cases of thyroid swelling diagnosed as colloid goitre. [1]

Fine needle aspiration cytology was done in three cases of subcutaneous swellings in the present study. One case showed microfilaria along with inflammatory cell including eosinophils, lymphocytes and macrophages. Another case showed only microfilaria and RBC. The third case shows microfilaria with lymphocytes in a background. Previous studies have also demonstrated microfilaria in subcutaneous swellings. [1, 14, 15]

In the present study 4 cases of Fine needle aspiration of breast swellings were carried out, of which 3 cases showed microfilaria along with few groups of benign ductal epithelial cells and inflammatory cells comprising of neutrophils, macrophages and giant cells. One case showed microfilaria with RBC in background. Varghese et al also reported that FNAC of cystic breast swelling showed clusters of microfilaria, lymphocytes and granular debris.[13] Yenkeshwar et al and Upadhyaya et al also reported microfilariae in breast lumps by FNAC smears. [14, 16]

Fine needle aspiration cytology from cervical lymph nodes demonstrates microfilariae in a background of reactive lymphoid cells in our study. Similar finding was reported by Varghese et al and Jindal et al. [13, 17] In histopathology a single case show metastatic deposits of ductal malignancy in lymphnode along with microfilaria in our study similarly Kolte et al reported metastatic deposits from papillary mucinous adenocarcinoma in supraclavicular lymphnode with microfilariae. [18]

In our study, out of 21 cases, 16(76.2%) cases showed associated eosinophilia in peripheral smear. These findings were consistent with observations made by previous authors. [19, 20] Mishra et al found eosinophilia in 8(33%) cases out of 24 cases reported. [1]The present study emphasis that there is no relationship between the percentage of eosinophils with microfilaria. Microfilaria may be found in any sample with or without Eosinophilia. Jindal et al and Agrawal et al reported 6% and 8% eosinophils in their studies. [17, 2]

CONCLUSION: -Filariasis is a disabling parasitic disease and can lead to different types of complications. Our study emphasized that careful search for parasites in every sample especially in endemic areas where patients present with fever, lump and with or without eosinophilia in peripheral smear examination, so that every patient should be diagnosed during the acute phase and complications during the chronic phase may be avoided. Furthermore, every patient may get treatment as early as possible.

REFERENCES:-

1. Mishra RK, Mitra SK, Verma P. Cytological diagnosis of microfilariae in filariasis endemic areas of eastern Uttar Pradesh. Journal of Cytology.2009; 26(1): 11-14.
2. Agrawal R, Kumar P. Incidental detection of filarial worm in a metastatic axillary Lymphnode from ductal carcinoma breast. Med J DY Patil Univ 2015; 8(4):499-501.
3. Mitra SK, Mishra RK, Verma P: Cytological diagnosis of microfilariae in filariasis endemic areas of eastern Uttar Pradesh. J Cytol 2009; 26(1): 11-14.
4. Sujathan K, Abraham EK: Breast lump suspected for Carcinoma Diagnosed as Filarial Granuloma by FNAC: A Report of Two Cases. Austral-Asian Journal of Cancer 2005; 4(1): 57-59.

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5. Jha A, Aryal G, Pant AD, Adhikari RC, Sayami G: Cytological diagnosis of bancroftian filariasis in lesions clinically anticipated as neoplastic. Nepal Med Coll J 2008; 10(2): 108-114.
6. Melrose WD. "Lymphatic Filariasis: New insight into an old disease." Int J Parasitol 2002; 32 (8): 947-60.
7. Dayal A, Selvaraju K: Filariasis of the Breast. Web Med Central Surgery; 2010; 1(11): WMC00942.
8. Park K. Epidemiology of communicable disease. In: Textbook of Preventive and Social Medicine. 18th ed, Banarsidas Bhanot Publishers: Jabalpur; 2005:211–16.
9. Chatterjee KD. Phyllum Nematohelminthes, class Nematoda. In: Parasitology (Protozoology and Helminthology) in Relation to Clinical Medicine. 12th ed. Calcutta: Chatterjee Medical Publishers; 1980:190–199.
10. Katti TV, Athanikar VS, Ananthrao AS, Rathod CV. Cytodiagnosis of microfilarial lymphadenitis coexistent with metastatic squamous cell carcinoma in a left cervical lymphnode: An unusual presentation. Ann Nigerian Med 2012; 6(1):47-9.
11. Gupta S, Gupta R, Bansal B, Singh S, Gupta K, Kudesia M. Significance of incidental detection of filariasis on aspiration smears: a case series. Diagn Cytopathol. 2010; 38(7):517-20.
12. Pradhan S, Lahiri VL, Eihence BR, Singh KN. Microfilaria of Wuchereria bancrofti in bone marrow smears. Am J Trop Med Hyg 1976; 25(1):199-200.
13. Varghese R, Raghuveer CV, Pai MR, Bansal R. Microfilariae in cytologic smears: a report of six cases. Acta Cytol 1996; 40 (2):299-301.
14. Yenkeshwar PN, Kumbhalkar DT, Bobhate SK. Microfilariae in fine needle aspirates: A report of 22 cases. Indian J Pathol Microbiol 2006; 49(3):365-9.
15. Oza H, Bhalodia J, Shah A, Modi P. Mid arm swelling- A rare presentation of filariasis. NJMR 2014; 4(3):256-58.
16. Upadhyaya V, Upadhyaya DN, Sarkar S. An interesting case of breast filariasis. Indian J Radiol Imaging 2006; 16(4):915-7.
17. Jindal A, Sukheeja D, Midya M. Cervical lymphadenopathy in a Child –An unusual presentation of filariasis. IJSRP 2014; 4(8).
18. Kolte SS, Satarkar RN, Mane PM. Microfilaria concomitant with metastatic deposits of adenocarcinoma in lymph node fine needle aspiration cytology: A chance finding. J Cytol 2010; 27(2):78-80.
19. Walter A, Krishnaswami H, Cariappa A. Microfilariae of Wuchereria bancrofti in cytologic smears. Acta Cytol 1983; 27(4):432–6.
20. Hira PR, Lindberg LG, Ryd W, Behbehani K. Cytologic diagnosis of Bancroftian filariasis in a non-endemic area. Acta Cytol 1988; 32(2):267–9.