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Original Research Article

MULTISITE MICROFILARIA DETECTION USING FINE NEEDLE ASPIRATION CYTOLOGY (FNAC): A COMPREHENSIVE ANALYSIS

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Abstract

Introduction: Filariasis, a parasitic infection caused by several types of nematode worms, is a matter of great concern for public health in India, The disease is caused by microfilaria, which are released into the peripheral blood circulation with periodicity during the night. However, identifying microfilaria in routine peripheral blood smears, Fine Needle Aspiration (FNAC) smears, and body fluids is challenging due to their scarce nature. Our study aimed to underscore the significance of having a heightened suspicion of filarial infection in patients with swelling, particularly those from endemic regions. Furthermore, the study emphasised the importance of using FNAC as an economical and effective diagnostic tool for detecting microfilaria.

Materials and Methods: This retrospective study was conducted in the Pathology Department of ESIC Medical College and Hospital Gulbarga over a period of ten years (2013-2023). A 22-23 gauze needle made aspiration smears were stained by Papanicolaou and May-Grünwald Giemsa stain.

Results: In this study, 24 cases showed the presence of microfilaria, a type of parasitic worm, despite the lack of clinical suspicion. Of these cases, eight involved breast lumps, eight involved thyroid swelling, and eight involved lymph nodes. In fifteen of these cases, fragments of adult worms were also reported, indicating a potentially more severe infection. These findings suggest the need for more diligent screening and testing to detect and treat parasitic infections before they can cause further harm.

Conclusion: Filariasis is a rare disease that requires thorough screening of FNA smears, particularly in asymptomatic patients from endemic regions, to avoid missing incidental findings. Moreover, the study emphasises the cost-effectiveness and efficacy of FNAC in diagnosing microfilaria.

Keywords: Microfilaria, Thyroid, Breast, Lymph node, FNAC, Cytodiagnosis.

ISSN: 0975-3583, 0976-2833 VOL 15, ISSUE 07, 2024

INTRODUCTION

Filariasis, a parasitic infection caused by thread-like filarial nematode worms, is a significant public health challenge in India, with the country accounting for approximately 20% of the global burden. Microfilaria are released into the bloodstream with nocturnal periodicity, making it difficult to detect them in routine peripheral blood smears and body fluids. Fine Needle Aspiration (FNAC) Smears can be used to diagnose the disease. Endemic regions in India include all over India except a few areas in Northern India and Northeast India. In heavy parasitic load, they may appear in the blood, urine with chyle and sometimes in scrotal aspirates. Regions with the heaviest parasitic loads requiring chemotherapy are located in Southeast Asia and Africa 1.

Fine Needle Aspiration Cytology (FNAC) is a pivotal diagnostic tool for timely and accurate diagnosis of filariasis. The significance of FNAC in the diagnosis of filariasis cannot be overstated. Its use expedites the diagnosis process and enables prompt initiation of treatment, which is critical for the successful management of the disease.

The disease is transmitted through the Culex mosquito and is caused by two types of nematodes: W. bancrofti and B. malayi. Microfilariae are released into the peripheral blood circulation with nocturnal periodicity. Most men with filariasis have genital disease, commonly hydrocele, while women more frequently exhibit lymphoedema or elephantiasis of the leg. The lymphatic system is primarily affected by filariasis, with a preference for lower limbs, retroperitoneal tissues, spermatic cord, and epididymis. While most infected individuals are asymptomatic, a small number of cases with microfilaremia at various sites such as lymph nodes, breast lumps, bone marrow, bronchial aspirates, nipple secretions, pleural and pericardial fluids, ovarian cyst fluids, and cervicovaginal smears have been reported. In rare cases, microfilariae have been isolated from the thyroid.

Filariasis is a prevalent disease in tropical regions, particularly Southeast Asia, and poses a significant health hazard. The microfilaria that causes the disease can remain viable in the lymphatic system for a prolonged period. The disease can have asymptomatic and symptomatic manifestations, such as hydrocele, lymphatic channels, and acute adenolymphangitis 2. Filariasis is challenging to diagnose due to its rarity, and it is difficult to detect microfilaria in routine peripheral blood smears and body fluids. The diagnosis of filariasis typically involves demonstrating microfilaria in peripheral blood smears or performing Fine Needle Aspiration (FNAC) Smears.

Our study aimed to evaluate the effectiveness of fine-needle aspiration cytology (FNAC) in diagnosing filariasis in asymptomatic patients with palpable swelling but no clinical symptoms. Our objective was to stress the significance of being vigilant for filarial infection, particularly in patients from endemic areas, when encountering any swelling. The study also aimed to shed light on the cost-effectiveness of FNAC as a diagnostic tool for identifying microfilaria and to investigate the potential of FNAC in diagnosing filariasis in all conceivable sites.

Materials and Methods:

This retrospective study was conducted in the Pathology Department of ESIC Medical College and Hospital Gulbarga over a period of ten years (2013-2023). The patients included

ISSN: 0975-3583, 0976-2833 VOL 15, ISSUE 07, 2024

in the study underwent a comprehensive clinical evaluation and routine investigations. Cystic lesions were aspirated, the collected material was centrifuged, and smears were prepared. These smears were then stained using Leishman-Giemsa and PAP (Papanicolaou) stains. Air-dried smears were also stained with Leishman-Giemsa (LG) stain. The study included lesions from uncommon sites such as lymph nodes, thyroid, and breast. The cases were evaluated in conjunction with relevant history, physical examination findings, and other available investigations. The Fine Needle Aspiration Cytology (FNAC) process employs a 23-gauge needle and a 10 ml sterile syringe. The aspirated material and fluid were then smeared and stained with Leishman-Giemsa (LG) stain. Some FNAC smears were air-dried and stained with LG stain, while others were fixed in 95% ethanol, stained with PAP stain, and mounted in DPX with coverslips. The smears were examined under a light microscope. The parameters studied included clinical history, FNAC examination, laboratory investigations, and treatment records (if available).

Results:

The study presented 24 filariasis cases diagnosed using routine FNAC material from various sites. The majority of cases were reported in lymph nodes (8 cases), breast swellings (8 cases), and thyroid swellings (8 cases) [as shown in Table 1].

The smear test results revealed that microfilaria, which are larvae of the M. bancrofti species, have distinctive features that set them apart from other sheathed larvae. Their tails were free from nuclei and ensheathed, and some formed curves. The embryos had a blunt head and a pointed tail that projected slightly beyond the sheath. The body's central axis contained granular somatic cells or nuclei, which were absent at the tail tip. Additionally, male worms were smaller and thinner than female worms. Eosinophilia was present in six of the nine cases, and the prevalence of the disease was equal in males and females. Cytological diagnosis was used to institute microfilariasis treatment after detecting clear fluid, blood, or pus in the aspirated fluid. Lymph node smears showed microfilaria with reactive lymphoid cells in the background, consistent with previous studies.

Breast swelling due to microfilaria is an uncommon occurrence. Out of the total cases of breast aspirates, only eight were detected. In one such case, a 52-year-old woman presented with a lump in her right breast that had been there for three months and measured about 3 cm x 2 cm in the lower outer quadrant. Analysis of the breast swelling smears revealed the presence of multiple coiled and uncoiled microfilariae, benign ductal epithelial cells, and an inflammatory background. [Fig 1]

Out of the total cases of thyroid aspirates, only eight were detected. A case report describes the condition of a 35-year-old woman residing in a village near Hyderabad who had been experiencing a painless and gradual swelling of the thyroid gland for two years. Upon examination, a 4-cm by 3-cm thyroid nodule was detected over the right lobe. The nodule was soft to firm, non-tender, and moved with swallowing. The patient did not have any detectable cervical lymph nodes and was asymptomatic. The patient had a total leukocyte count of 13,200/mL with 76% neutrophils, 20% lymphocytes, and 4% eosinophils. The patient's erythrocyte sedimentation rate was 28 mm/hour, within the normal range. Additionally, the patient's thyroid function was determined to be normal.

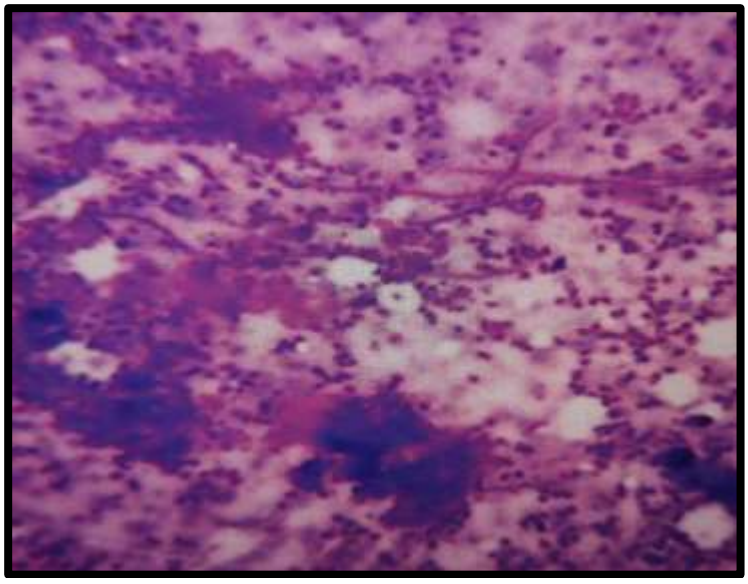
ISSN: 0975-3583, 0976-2833 VOL 15, ISSUE 07, 2024

A thyroid nodule was subjected to a fine needle aspiration, and the resulting smear showed the presence of blood-mixed colloid. Microscopic examination of the smear showed microfilaria larvae in the background of blood mixed-colloid, along with a few clusters of benign follicular cells and macrophages. The microfilariae had a sheath, which projected slightly beyond the larvae's body. The central axis of the larval body contained nuclei, which appeared as granules and were absent at the tip of the tail. The diagnosis was microfilaria of the thyroid, morphologically consistent with Wuchereria bancrofti. [Refer Fig 2 to Fig 4].

Out of the total cases of lymph node aspirates, only eight were detected. The case of a 33-year-old female patient is described, who presented with a gradual onset of swelling in the right side of the neck accompanied by pain, weakness, and fever during the night for the last six months. On examination, a palpable lump of size 3 cm × 2 cm was observed over the right supraclavicular region, which was firm, non-tender, and mobile, with a globular shape. The patient's menstrual cycle was regular, with an average flow. Upon examination of the neck using ultrasonography (USG), a lymph node in the right supraclavicular region was found to be enlarged. The lymph node showed a loss of fatty hilum. The complete blood count was normal, and the peripheral smear examination revealed a normal count and morphology of cells with no abnormal findings. The ESR was 20 mm/hr. The patient was given a provisional diagnosis of a case of cervical lymphadenopathy under evaluation.
The patient underwent FNAC, which revealed the presence of an ensheathed, coiled, and slightly curved microfilaria (Figure 5). The background contained red blood cells (RBCs).
The incidental finding of microfilaria in the lymph node highlights the importance of thorough screening in patients with absent clinical manifestations of filariasis and microfilaremia in the blood.

Table 1: Sites of Microfilaria

Site	No of cases	Percentage
Breast	08	33.33%
Thyroid	08	33.33%
Lymph node	08	33.33%
Total	24	100%



ISSN: 0975-3583, 0976-2833 VOL 15, ISSUE 07, 2024

Figure 1: The sample displays numerous coiled and uncoiled microfilariae, benign ductal epithelial cells, and an inflammatory background

Figure 2: Photograph of a 4-cm × 3-cm thyroid nodule: soft to firm and non-tender, moving with swallowing.

Figure 3: Photomicrograph of microfilaria in a colloid background (May Grunwald Giemsa (MGG), 400×).

ISSN: 0975-3583, 0976-2833 VOL 15, ISSUE 07, 2024

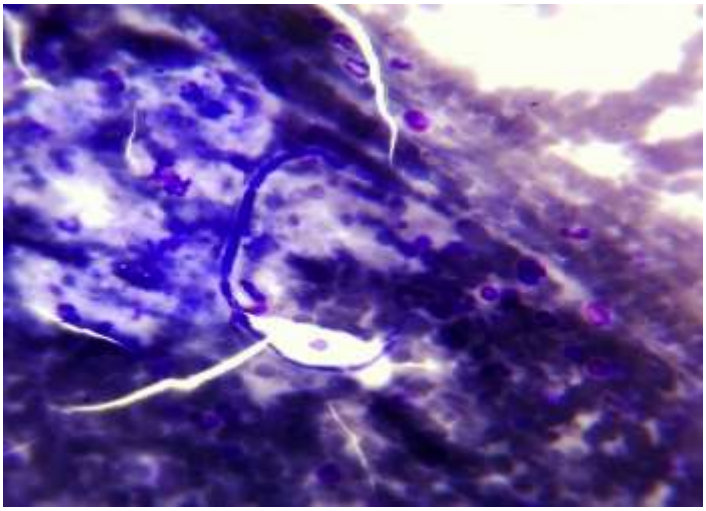


Figure 4: Photomicrograph of microfilaria with background showing scattered thyroid follicular cells (May Grunwald Giemsa (MGG), 400×).

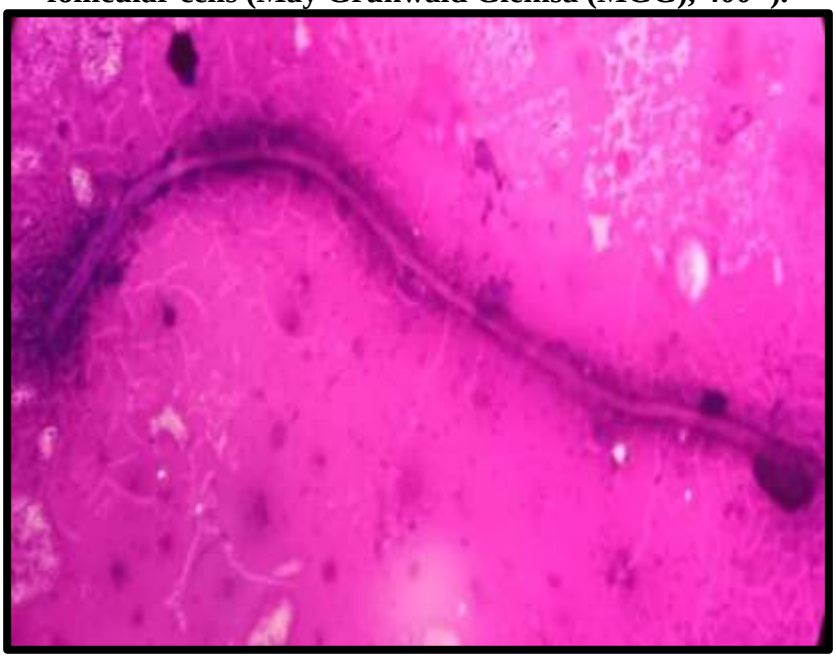


Figure 5: Microfilaria in lymph node. An ensheathed, coiled, and slightly curved microfilaria was observed. The background contained red blood cells

Discussion:

Filariasis is a significant public health concern in India, caused by nematodes such as W. bancrofti, B.malayi, B. timori, Loa-loa, O. volvulus, M. perstans, and M.ozzardi 3. While most infected individuals are asymptomatic, W. bancrofti and B. malayi account for 95% and 5% of cases, respectively. Adult worms of these species reside mainly in lymphatic channels of the spermatic cord, retroperitoneum, lower extremities, and breast 4. The female worm measures 8-10 cm in length by 0.2-0.3 cm in thickness, while the male measures 2.5-4 cm in length by 0.1 mm. The embryo size is 290 micrometres long and 6-7 micrometres thick.

ISSN: 0975-3583, 0976-2833 VOL 15, ISSUE 07, 2024

Out of all the cases of fine needle aspiration cytology (FNAC), only 24 showed the presence of microfilaria. The discovery of microfilaria in the mentioned sites was not significant and was found incidentally. Other researchers have also conducted similar studies, showing a very low incidence of microfilaria from these superficial sites 5. Our findings indicate that the aetiology of microfilaria is attributed to Wuchereria bancrofti, which aligns with the results of the nationwide study conducted in India, where this pathogen was identified as the causative agent in 95% of the cases 6.

Out of 24 cases, 16 showed the presence of eosinophilia. Based on our research, we have found that filariasis can occur even in the absence of microfilaremia, which is consistent with the findings of previous studies. Microfilaria, accompanied by reactive lymphoid cells, was detected in lymph node aspirates. These cytological features were similar to those that Varghese et al 7 and Joshi et al 8 reported.

Identifying microfilaria in breast swelling is an infrequent occurrence that has only been reported as an accidental discovery by a handful of researchers 9,10. Cases of breast lumps with tenderness have been observed in patients where microfilariae were detected in FNAC smears of breast swelling.

Colloid aspiration was performed on eight cases of thyroid swellings. The cytological examination of the aspirated material revealed that five cases had features indicative of colloid goitre, while the other showed lymphocytic thyroiditis with rare microfilaria. The findings are consistent with the earlier research conducted by Varghese 7, Sodhani 4, Chowdry 11, and Yenkeshwar 3. These results further reinforce the validity of the prior study, lending support to its conclusions and contributing to the broader body of knowledge in this area.

In 12 out of 24 cases, adherent inflammatory cells were observed in microfilariae. This phenomenon was previously reported by Pandit et al 12. and Walter et al 13. Additionally, inflammatory cells were observed surrounding degenerated microfilaria and coiled larvae.

Although new techniques such as Filaria antigen detection tests, PCR, and FISH are highly specific, they are expensive and only available in specialised centres, making them inaccessible to many people. These techniques are mainly used when there is a pre-existing suspicion of tissue filariasis.

Conclusion:

Filariasis is a rare condition requiring high suspicion and careful screening of FNA smears, especially in asymptomatic patients from endemic areas. It is important to notice this incidental finding, as early diagnosis can help reduce morbidity before the onset of lymphatic filariasis. FNAC is a cost-effective tool that can be used to diagnose microfilaria. As such, it is an important aspect of managing these cases, as it can help avoid unnecessary surgical interventions. Although filariasis incidence is quite high, microfilaria is uncommon in FNAC smears. Therefore, it is important to thoroughly examine FNAC smears for the presence of microfilaria, even in situations where the clinician has not suspected the disease. FNA is a simple procedure that can detect the hidden burden of microfilaria and guide proper management, sparing unnecessary investigations. Filariasis should be considered a differential diagnosis of swelling at any site, and early diagnosis by demonstration of parasites can reduce significant morbidity in due course of the disease.

ISSN: 0975-3583, 0976-2833 VOL 15, ISSUE 07, 2024

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