RESEARCH NOTE

MICROFILARIA AND FILARIAL GRANULOMAS FROM FINE NEEDLE ASPIRATES: A STUDY OF 25 CASES

Sainath K Andola and A Akshatha Naik

Department of Pathology, Mahadevappa Rampure Medical College, Gulbarga, Karnataka, India

Abstract. The objective of this study was to assess the role of fine needle aspiration cytology (FNAC) for the diagnosis of filariasis. Cytological smears were studied during 2006-2009. Twenty-five cases with microfilaria were detected: 7 from lymph nodes, 5 from soft tissue, 4 from effusion fluids, 3 from breast tissue, 2 from liver tissue, 1 each from thyroid tissue, a cervicovaginal smear, a hernial sac and a spermatic cord cyst. Embryonated adult worms were seen in four cases and eosinophils were seen in 3 cases. In endemic areas the diagnosis of filariasis should be considered in the differential diagnosis of swelling. This study highlights the importance of screening smears for parasites even in the absence of clinical indications and shows eosinophils are not mandatory to diagnose microfilaria.

Keywords: FNAC, microfilaria, adult worm, embryoid bodies, eosinophils

INTRODUCTION

Filariasis is a major public health problem in many tropical and subtropical countries and is seen in India, China, Indonesia, Africa and the Far East (Park, 2007). It is estimated 600 million people are living in areas endemic for lymphatic filariasis in Southeast Asia (Park, 2007). Lymphatic filariasis is a major public health problem in India and is increasing yearly due to mismanagement of the environment (Park, 2007). The disease is endemic throughout India. An estimated 553.7 million people are at risk of lymphatic filariasis infection in 243 districts of India (Park, 2007).

The conventional diagnosis of filariasis relies on finding microfilariae on a peripheral blood smear; however, incidental detection of microfilariae in various cytological specimens has also led to the diagnosis in unsuspected case (Fernandes *et al*, 2007). The finding of microfilaria in fine needle aspirates is uncommon (Pandit *et al*, 1996). The literature contains occasional reports of finding microfilariae in various locations, including the thyroid, breast, skin and soft tissue swellings, epididymis, salivary glands, the liver, lymph nodes, ovarian cysts, urine, endoscopic brushings and effusion fluids (Yenkeshwar *et al*, 2006).

The present study was undertaken to analyze the microfilaria, adult filarial

Correspondence: Prof Sainath K Andola, Department of Pathology, MR Medical College, Gulbarga, Karnataka – 585105, India.
Tel: 08472-222948; Fax: 08472- 225085
E-mail: drskandola@gmail.com

worms and filarial granulomas in 25 cases from different locations diagnosed by fine needle aspiration cytology (FNAC) and exfoliative cytology.

MATERIALS AND METHODS

This study was conducted at The Department of Pathology, Mahadevappa Rampure Medical College, Gulbarga, India over a 4 year period (January 2006-December 2009). Microfilariae were identified in 25 cases, presenting at different sites. Aspirates were obtained by suction with a 22-23 gauge needle attached to a disposable syringe. In cases of cystic lesions, cyst content was aspirated and smears prepared after cytocentrifugation. Air dried smears were stained with Giemsa stain and smears fixed in 95% alcohol were stained with Papanicolaou stain.

RESULTS

Twenty-five cases of filariasis were diagnosed on routine FNAC material from various sites. Out of these 25 cases, the largest number of microfilaria (MF) positive cases were from lymph nodes (7) followed by soft tissue (5), effusion fluid (4), breast (3), liver (2), thyroid (1), cervicovaginal smear (1), hernial sac (1) and spermatic cord cyst (1) (Table 1).

The clinical presentations of these cases varied. The duration of symptoms varied from days to years. The most common age range at presentation was 20 to 30 years old, with a range of 17-65 years old. In the present study, most cases presented with swelling (Fig 1). In none of these cases was filariasis considered a diagnostic possibility. The aspirate was grossly clear in 16 of 25 cases, which included the effusions. Other aspirate specimens were hemorrhagic or purulent.

Microscopically (Figs 2-8), all mi-

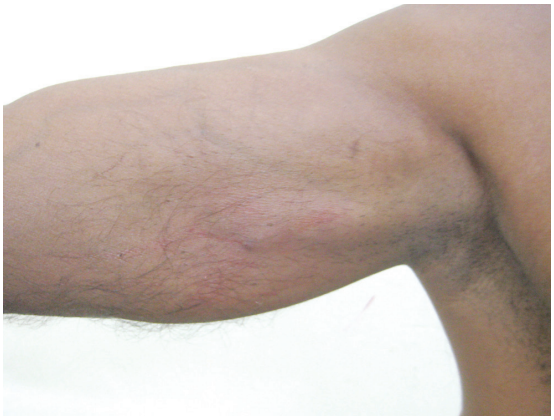

Fig 1–A filariasis patient with swelling of the right arm.

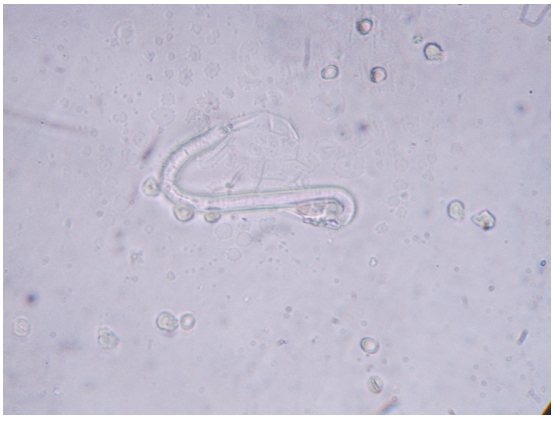

Fig 2–Wet mount of aspirate from the above patient (40x).

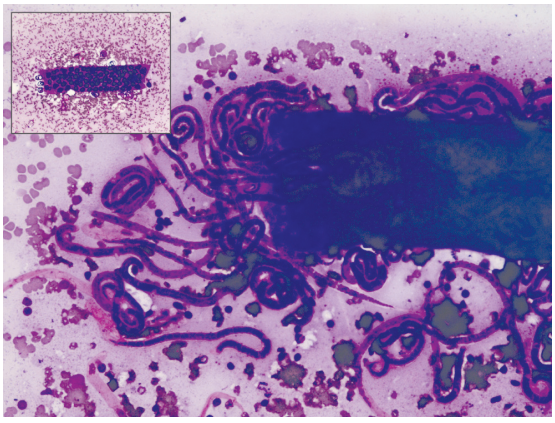

Fig 3–Aspirate from a soft tissue swelling showing an adult worm with splitting of the cuticle (40x, Giemsa). Inset- adult worm with embryoid bodies (40x, Giemsa).

Table 1
Anatomic location of microfilaria and adult worms.

Anatomic sites	Cases with microfilaria	Cases with adult worm	Cases with embryoid bodies	Total no. of cases	Percentage
Lymph node					
Cervical	1	-	-	7	28
Axillary	3	-	-		
Inguinal	3	-	-		
Soft tissues					
Neck	1	1	1	5	20
Midclavicular	1	-	-		
Arm	2	2	2		
Finger	1	-	-		
Breast	3	-	-	3	12
Liver	2	-	-	2	8
Thyroid	1	-	-	1	4
Spermatic cord cyst	1	-	-	1	4
Hernial sac	1	-	-	1	4
Cervicovaginal smear	1	-	-	1	4
Effusion fluid					
Pericardial	1	-	-	4	16
Pleural	1	-	-		
Ascitic	2	-	-		
Total	25 (100%)	4 (16%)	4 (16%)	25	100

crofilariae had sheaths and curved tails which were free of nuclei. The number of parasites varied from one to many per slide in various stages of development. A few specimens showed granulomas and inflammatory cells adherent to the microfilaria. Eosinophils were seen in only 4 cases. Adult worms were seen in 4 cases. Aspirate from soft tissues showed numerous adult worms with splitting of cuticles and embryoid bodies.

Six cases of malignancy were diagnosed along with the presence of microfilaria. They were ductal carcinoma of the breast, hepatocellular carcinoma, squamous cell carcinoma of the cervix from a cervicovaginal smear; adenocarcinoma cells were seen in pericardial fluid, ascitic fluid and pleural fluid (Table 2).

DISCUSSION

Filariasis is a major public health problem in many countries, including India (Chatterjee, 1980). Most filariasis is caused by nematodes *Wuchereria bancrofti*, *Brugia malayi*, *B. timori*, *Onchocerca volvulus*, *Mansonella perstans*, *M. streptocerca*, *M. ozzardi*, *Dirofilaria conjunctivae*, *D. magalhaesi*, *D. immitis* and *Loa-loa* (Chatterjee, 1980). *Wuchereria bancrofti* and *Brugia malayi* are the most common species seen in India. They are transmitted by the bite of the Culex mosquito (Chatterjee, 1980).

Wuchereria bancrofti and *Brugia malayi* parasites are largely confined to the tropics and sub-tropics, including northeastern India, the West Indies, Puerto Rico, southeastern China, Japan, the Pacific

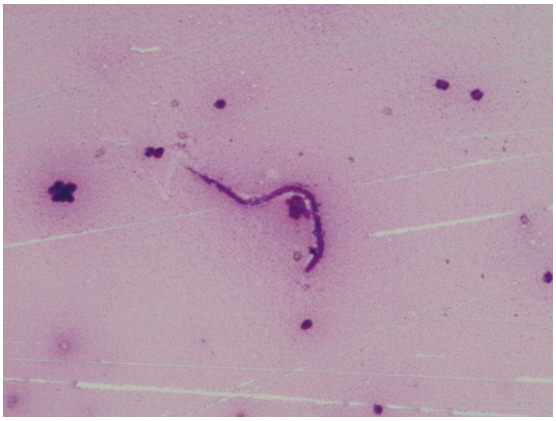

Fig 4–Aspirate from thyroid swelling showing microfilaria in the colloid background (40x, Giemsa).

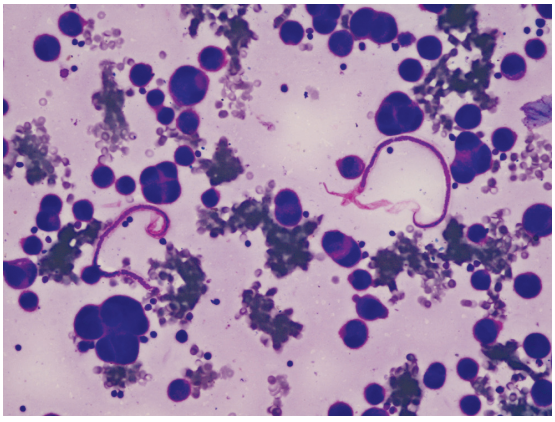

Fig 5–Pleural fluid showing microfilaria amidst adenocarcinoma cells (40x, Giemsa).

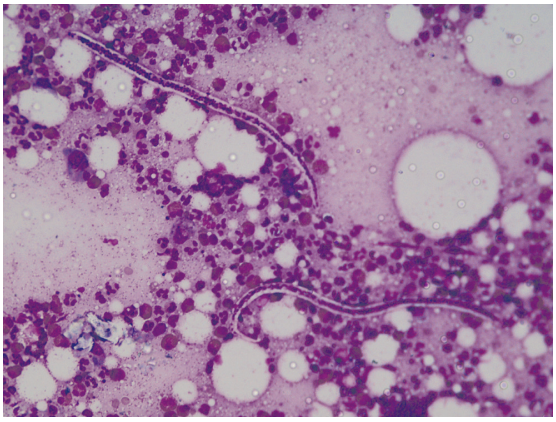

Fig 6–Microfilaria from a breast aspirate with eosinophils in the background (40x, Giemsa).

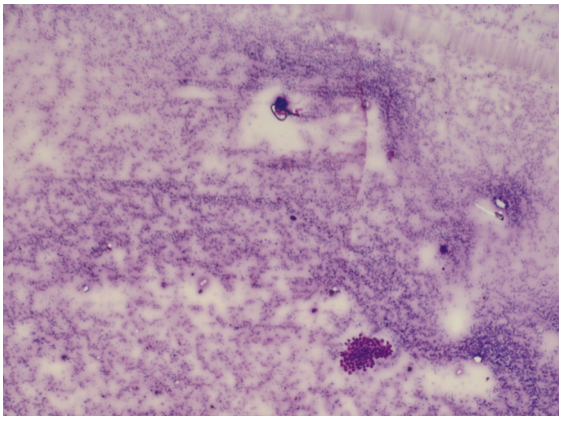

Fig 7–Microfilaria with duct carcinoma cells from breast aspirate (10x, Giemsa).

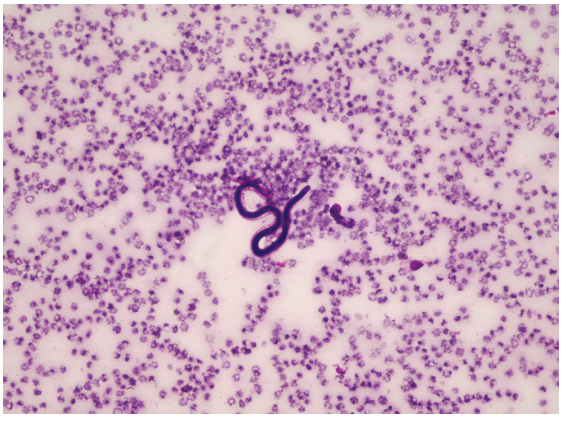

Fig 8–Microfilaria from finger cyst aspirate (40x, Giemsa).

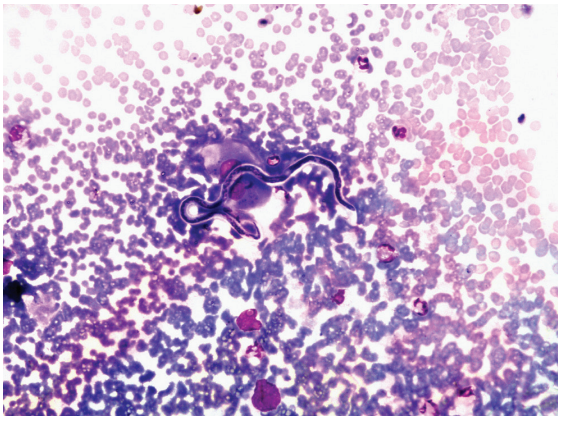

Fig 9–Microfilaria from liver aspirate with hepatocellular carcinoma (40x, Giemsa).

Table 2
Clinical and cytological findings.

Features	Lymph node (*n*=7)	Soft tissue (*n*=5)	Breast (*n*=3)	Liver (*n*=2)	Thyroid (*n*=1)	Vaginal (*n*=1)	Effusion (*n*=4)	Spermatic cord cyst (*n*=1)	Hernia sac (*n*=1)
Clinical findings									
Swelling	7	5	3	-	1	-	-	1	1
Pain	1	2	1	-	-	-	-	-	-
Fever	2	-	-	-	-	-	-	-	-
Cytological findings									
Gross									
Hemorrhagic									
Aspirate	1	3	1	2	-	1	-	-	-
Pus aspirate	-	-	-	-	-	-	-	-	1
Fluid aspirate	6	2	2	-	1	-	4	1	-
Microscopy									
Lymphocytes	3	1	-	-	-	-	-	-	-
Neutrophils	3	2	-	-	-	-	-	-	-
Eosinophils	1	2	1	-	-	-	-	-	-
Granuloma	2	1	-	-	-	-	-	-	-
Microfilaria	7	5	3	2	1	4	1	1	1
Adult worms and embryonal bodies	1	3	-	-	-	-	-	-	-
Associated malignancy	-	-	Duct Ca (1)	H.C. Ca (1)	-	Sq Cell Ca (1)	Adenoca (3)	-	-
Associated conditions	-	-	Fibrocystic disease (2)	-	Colloid goiter (1)	-	-	-	-

Table 3
Comparison of filarial infections among several studies.

Features	Yenkeshwar *et al* (1999-2000)	Mitra *et al* (2006-2007)	Present study (2006-2009)
	n=22 (%)	*n*=24 (%)	*n*=25 (%)
Anatomic site			
Lymph node	2 (9.1)	6 (25.0)	7 (28)
Soft tissue	7 (31.8)	2 (8.3)	5 (20)
Breast	3 (13.6)	8 (33.3)	3 (12)
Liver	-	-	2 (8)
Thyroid	3 (13.6)	3 (12.5)	1 (4)
Cervicovaginal smear	1 (04.6)	-	1 (4)
Effusion fluid	3 (13.6)	1 (4.2)	4 (16)
Others	3 (13.6)	4 (16.7)	2 (8)
Clinical findings			
Swelling	16 (72.7)	23 (95.8)	18 (72)
Pain	7 (31.8)	14 (58.3)	4 (16)
Fever	5 (22.7)	2 (8.3)	2 (8)
Cytological findings			
Gross			
Hemorrhagic fluid	4 (18.2)	5 (20.8)	8 (32)
Purulent fluid	3 (13.6)	-	1 (4)
Clear fluid	9 (40.9)	6 (25.0)	16 (64)
Microscopy			
Eosinophils	7 (31.8)	15 (62.5)	4 (16)
Granuloma	4 (18.2)	3 (12.5)	3 (12)
Adult worm and embryoid bodies	2 (9.1)	-	4 (16)
Associated malignancy	6 (27.3)	-	3 (12)

Islands, North Africa and South America. In India, it is distributed chiefly along the banks of large rivers (except the Indus) (Chatterjee, 1980).

Adult worms live in the lymphatics of definitive hosts and the microfilaria are released into the peripheral blood. The most frequently involved lymphatics are the lower limbs, retroperitoneal tissue, spermatic cord, epidydimis and mammary glands (Yenkeshwar *et al*, 2006). Filariasis causes a variety of diseases, including asymptomatic microfilaremia, acute lymphangitis, edema of limbs and genitalia and tropical eosinophilia (Chowdry *et al*, 2008). The larval forms of *W. bancrofti* and *B. malayi* circulate in the body until they are removed by an intermediate host. Microfilaria appear in tissue fluid and exfoliated surface material due to lymphatic and vascular obstruction and subsequent extravasation (Chowdry *et al*, 2008).

In endemic areas people become infected early in life and develop microfilaremia, reaching a peak between 15-20 years old (Mitra *et al*, 2009). Most infected people in endemic areas are asymptomatic

but are an important source of infection in the community (Jayaram, 1996; Varghese *et al*, 1996) Filariasis can exist without microfilaremia. Thus, infection and disease do not necessarily accompany each other (Varghese *et al*, 1996). Although the incidence of filariasis is high on the Indian subcontinent, the finding of microfilaria on fine needle aspirate samples is unusual (Varghese *et al*, 1996; Chowdry *et al*, 2008).

In the present study, the majority of cases presented with swelling; filariasis was not considered in the differential diagnosis. Swelling was also a major clinical symptom seen in studies by Yenkeshwar *et al* (2006) and Mitra *et al* (2009). As in our study, only a minority of cases had eosinophils in the aspirate; hence, eosinophils are not necessary to diagnose microfilaria.

The phenomenon of cell adherence may reflect the immune status of the patient in regard to his or her filarial disease. Cell adherence to the microfilaria with *W. bancrofti* infection was first described by Pandit *et al* (1996) who noted cell adherence to the microfilaria of *W. bancrofti* in the serum of a microfilaremic patient with elephantiasis. This could be due to filarial antibodies in the sera of these patients, since the sera of apparently healthy individuals in endemic areas shows the same effect (Varghese *et al*, 1996; Chowdry *et al*, 2008).

Various authors have expressed the opinion that because these parasites circulate in the vascular and lymphatic systems, their appearance in tissue fluid and exfoliated surface material can occur only under conditions of lymphatic and vascular obstruction, causing extravasation of blood and release of microfilariae into the blood circulation (Varghese *et al*, 1996; Yenkeshwar *et al*, 2006; Chowdry *et al*, 2008). Such aberrant migration to these "dead end" sites is probably determined by local factors, such as lymphatic blockage by scars or tumors and damage to the vessel wall by inflammation, trauma or stasis. In the present study, a common site for microfilaria was the lymph nodes. In two similar studies conducted by Yenkeshawar *et al* (2006) and Mitra *et al* (2009) the common sites were soft tissue and breast, respectively. This could be attributed to no site preponderance for microfilaria. Thus, it can be seen anywhere in the body (Table 3).

The rich blood supply of tumors could increase the concentration of the parasite at that site (Gupta *et al*, 2001). The larvae may be present in the vasculature and aspiration may lead to rupture of vessels resulting in hemorrhage and release of microfilariae (Ahluwalia *et al*, 2003). The presence of microfilaria within a neoplasm is a chance association. The patient probably had subclinical filariasis when the tumor developed.

In conclusion, endemic areas the diagnosis of filariasis should be considered in the differential diagnosis of swelling, not only of the skin, but in other sites as well. Careful screening with FNAC is helpful to detect microfilaria even in asymptomatic patients. This study highlights the importance of screening smears for parasites even in the absence of clinical indications and shows that eosinophils are not necessary to diagnose microfilaria.

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