

Case Report

Lymphatic filariasis: the importance of screening all peripheral blood smears in low power for detection of asymptomatic cases

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ABSTRACT

Lymphatic filariasis caused by the mosquito-borne, lymphatic-dwelling nematodes *Wuchereria bancrofti* and *Brugia malayi* is still a common tropical parasitic disease and 120 million people are affected in the world, of which two-third are in Asia. They cause high morbidity and mortality among humans. Irreversible “elephantiasis” is the major clinical manifestation for LF. Detection of microfilaria in peripheral blood is important. In addition to simple thick and thin blood smear tests, concentration techniques are used: mainly density gradient centrifugation, haemolysis and filtration of the blood through a polycarbonate membrane, which retains the parasite. Diagnosis has been revolutionized with the availability of circulating filarial antigen (CFA) tests which are easy to perform but are costly. Diethylcarbamazine (DEC) is the drug of choice for treating lymphatic filariasis. In the light of this information, hereby presenting a case series of 4 asymptomatic patients who were diagnosed with filariasis on peripheral blood smear examination. The article emphasizes the importance of low power scanning of every peripheral blood smear, especially when the laboratory is not facilitated with costly methods to detect microfilaria.

Keywords: Filariasis, Low cost, Low power scanning, Peripheral blood smear

INTRODUCTION

Filariidae have a remarkable specificity for their definitive mammalian host and obligate intermediate vector species. This specificity is frequently extended even to a favored or obligatory tissue within the host.

On the basis of the habitat of the adult worm stage, filarial parasites of medical importance may be classified into three groups: (i) the cutaneous group, including *Loa loa*, *O. volvulus*, *Mansonella perstans*, and *Dipetalonema streptocerca*; (ii) the lymphatic group, including *W. bancrofti*, *B. malayi*, and *Brugia timori*; and (iii) the body cavity group, including *Mansonella ozzardi*. *O. volvulus* is the cause of river blindness and several skin diseases. *L. loa* produces painful dermal reactions. *Wuchereria* and *Brugia* species cause elephantiasis and hydroceles. *M.*

ozzardi infection is associated with no or only minor disease manifestations.¹ Filariasis, a vector-borne disease, is common in tropical countries like India. *Wuchereria bancrofti* is the most widespread of the filarial organisms, infecting man. The parasite is endemic in both urban and rural areas of India.² Infection often begins during childhood but clinical filariasis is mainly a disease of adults.

LF is a major cause of disability, social stigmatization, and psychosocial and economic reductions in life opportunities. It is also a major burden on health and hospital resources. It may present as asymptomatic, acute, or chronic infections.³ Adult male and female worms of *Brugia* and *Wuchereria* species inhabit primarily the lumen of lymphatics. Adult worms of lymphatic filariae may live for 7 to 10 years, although cases of 40-year

longevity have been reported.⁴ Adult female filariae are ovoviviparous, producing thousands of microfilariae or first-stage larvae after fertilization by adult male parasites. In the cases of *Wuchereria* and *Brugia* species, microfilariae migrate from the lymphatics into the bloodstream.

These parasite stages are ingested by the intermediate arthropod host with the blood meal. Mosquitoes act as the intermediate hosts of *Wuchereria* and *Brugia* species. Ingested microfilariae penetrate the gut wall and eventually reach the thoracic musculature or fatty tissues of the invertebrate host, where they shorten by metamorphosis into sausage-shaped bodies 240 to 250µm in length in the case of lymphatic filariae. They enter the skin through the wound made by the insect.⁵

The majority of affected subjects show a clinically asymptomatic infection and harbour microfilariae or circulating filarial antigens (CFAs) in their peripheral blood.⁶ World Health Organization (WHO) considers LF to be a global health problem affecting over 120 million people in 73 countries in 2012.⁷ In 2000, the WHO established a global plan to eliminate LF: the 'Global Programme to Eliminate Lymphatic Filariasis in order to achieve the global goal of LF elimination as a public health problem by the year 2020.'⁸ The programme consists of annual mass drug administration (MDA) using a two drugs combination to treat the entire at-risk population.⁹ National Filaria Control Programme (NFPC) was launched in the country in 1955 with the objective of delimiting the problem and to undertake control measures in endemic areas.¹⁰

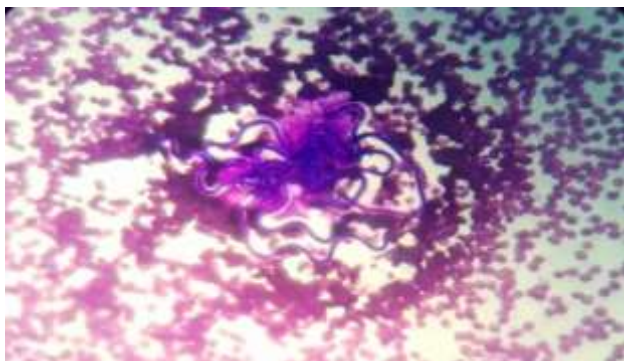


Figure 1: Clump of microfilaria in buffy coat preparation of an asymptomatic patient's blood sample.

In the endemic areas the majority of affected subjects show a clinically asymptomatic infection. It is important to know that even at this stage of the disease abnormalities of the lymphatic vessels such as dilatation appear to be irreversible even after treatment.¹¹ Microscopy remains the cornerstone of the laboratory diagnosis of infections due to blood and tissue parasites. Microscopy is performed on thick and thin blood smears or buffy coat films stained with Giemsa.¹²

The morphology of *W. bancrofti* is the most significant differentiation from other species. The microfilariae, or larval stage of *W. bancrofti*, are sheathed, and range from approximately 245 to 300 µm. As adults, the males range from 2.5 to 4 cm, and the females range from 5 to 10 cm. One end of the round body is blunt, while the other is pointed.

Nuclei do not appear at the end of the tail, which is a major difference from other microfilariae. Generally, microfilariae range from 200 to 275 µm. Adult size is not well-known, as very few have actually been found. Microfilariae of *B. malayi* are sheathed like *W. bancrofti*, and have a very similar shape. However, the nuclei extends nearly to the tip of the tail, a characteristic not shared with *W. bancrofti*.¹³

The only unequivocal means of ascertaining active filarial infection is by demonstrating parasites in host tissue. In the cases of lymphatic filariasis this is most commonly achieved by detection of microfilariae in the bloodstream. Blood samples should be obtained at a time of day consistent with the known periodicity of microfilariae in the specific geographic region, which is between 2200 and 0200 h for nocturnally periodic forms of *brugian* and *bancroftian* filariasis.



Figure 2: Microfilaria in Giemsa stained blood smear: the central nuclear column does not extend to the tip of its tail (*Wuchereria bancrofti*).

Various techniques for identification of microfilariae in blood have been utilized. These range from a Giemsa smear of a 20-µl blood sample obtained by finger prick to concentration methods utilizing a larger volume of blood. The Knott concentration method involves fixing a 1-ml sample of anticoagulated blood with 4 volumes of Formalin. Sediment is then prepared and examined microscopically for microfilariae.¹⁴

The Nuclepore (Nuclepore Corp., Pleasanton, Calif.) method uses a polycarbonate filter through which 1 to 2 ml of anticoagulated blood is passed. The filter is then removed from the supporting chamber and placed on a microscope slide for counting of parasites.¹⁵ Recently ultrasonography has helped to locate and visualise the

movements of living adult filarial worms of *W. bancrofti* in the scrotal lymphatics of asymptomatic males with microfilaraemia. The constant thrashing movement of the adult worms in their 'nests' in the scrotal lymphatics is described as the 'filaria dance sign'.¹⁶

After injecting radiolabelled albumin or dextran in the web space of the toes, the structural changes are imaged using a gamma camera. Lymphatic dilatation, dermal back flow and obstruction can be directly demonstrated in the oedematous limbs by this method. Lymphoscintigraphy has shown that even in the early, clinically asymptomatic stage of the disease, lymphatic abnormalities in the affected limbs of people harboring microfilaria may occur.¹⁷

Highly sensitive and specific filarial antigen detection assays, both as card test and in ELISA based format are now available for the diagnosis of *W. bancrofti* infection. This test is positive in early stages of the disease when the adult worms are alive and becomes negative once they are dead.¹⁶ DNA probes using Polymerase Chain Reaction (PCR): These tests are of high specificity and sensitivity, and are able to detect parasite DNA in humans as well as vectors in both bancroftian and brugian filariasis.¹⁸ Hereby, presenting a case report of 4 asymptomatic cases that were diagnosed on routine low power scanning of peripheral blood smear.

CASE REPORT

Case 1

Blood sample of a 28 year old pregnant lady admitted to labour ward was sent for routine haematological analysis. A single microfilaria was noticed at the tail of the peripheral blood smear on routine low power (4x) scanning. Further investigation on high power (40x) revealed the central row of nuclei which were conspicuously absent from the tip of the tail, typical of *W. bancrofti*. On examining the wet film, live microfilariae kept on moving creating a blank space around them. The other hematological parameters were as follows: Hb 9.21g/dl; PCV 29.04%; MCV 54.68fl; MCH 32.22pg; MCHC 17.57; TLC 6400/cmm; DLC N55 L40 M1 E4 B 0; There was no eosinophilia.

Case 2

An 18 year old male patient admitted with acute gastroenteritis was found to be another case of asymptomatic filariasis. Similar wet mount was prepared from the blood drop to see live microflaria. Also a buffy coat was prepared and stained with a Geimsa stain and observed under microscope. A small clump of microflaria was noticed. His haemoglobin level was 10.23g/dl while total and differential leucocyte count was within normal range. There was no eosinophilia. On retrospective analysis he was found to be a resident of Bihar which has highest endemicity (over 17%) of microflaria.

Case 3

This case was a pre-operative sample of a 38 year old male patient in surgical ward, who was posted for inguinal hernioplasty. After confirming the diagnosis on high power (40x), a wet film and a buffy coat examination followed. This was another case with absolutely no symptoms. His haemoglobin was 9.89g/dl with a normal range total and differential count. Again no eosinophilia.

Case 4

The last case of this series was suffering from fever, clinically suspected to be of viral origin. On routine blood sample analysis microfilaria was detected as with the previous cases and the diagnosis was reaffirmed with a wet mount and a buffy coat study. All of his haematological parameters were normal, thereby raising no suspicion of filariasis.

DISCUSSION

These cases of microfilariae were incidental findings while reporting routine haematological blood smears. These 4 cases were positive out of 500 blood smears examined over a period of 3 months. Out of the various methods to identify microfilariae, microscopy is the cornerstone of laboratory diagnosis. Examination of peripheral blood smears and/or buffy coat preparations is very easy and cost-effective. The other methods based on detecting filarial antigen assays (for e.g. ELISA) are highly sensitive but costly. Polymerase Chain Reaction (PCR) a highly specific and sensitive test is not only costly but also time-consuming. In tropical countries like India where filariasis is endemic, it may not be possible for all laboratories to perform expensive tests.

Microscopy helps with the accurate and cost-effective diagnosis of lymphatic filariasis. Peripheral blood eosinophilia is considered to be a useful diagnostic clue to screen the blood smear for filariasis, but was conspicuously absent in my findings. Peripheral blood eosinophilia is a common haematological finding in filariasis. But in a majority of the reported cases eosinophilia was absent.²⁰⁻²² The absence of peripheral blood eosinophilia in these cases may be attributed to the oxidative stress which was associated with the chronic and occult filariasis, which had caused altered immune responses.²³ Anaemia and pancytopenia were the other frequent peripheral blood findings.²⁰⁻²⁴

In this case series too anemia was a consistent feature. It has been estimated in December 2006, that the total population at risk of LF was estimated to be 1,254 million in 83 endemic countries of which 64% was contributed by South-East Asia Region (SEAR) alone. In India it is estimated 554.2 million populations are at risk of LF.²⁵

Lymphatic filariasis is so called because the adult worm lives in the lymphatic system of the definitive host and microfilaria (MF) is released and circulated in the peripheral blood. Lymphangitis, lymphadenitis, and lymphedema resulting in elephantiasis are various manifestations of lymphatic filariasis. Filariasis is treated by diethylcarbamazine therapy.²⁶ Most people with microfilaraemia do not show signs or symptoms of the disease but are important source of infection in the community. Thus, disease and infection do not necessarily accompany each other.²⁷

CONCLUSION

This case series emphasizes the importance of screening all peripheral blood smears in low power for the detection of asymptomatic cases of lymphatic filariasis. Thus morbidity associated with filariasis can be significantly reduced. Also it is a cost effective method and can be done in any laboratory set-up. Though eosinophilia is considered to be associated with filariasis none of our cases exhibited such a finding. Low power screening is an important step in haematological slide analysis which should be performed on all peripheral blood smears.

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REFERENCES

- Denham DA, McGreevy PB. Brugian filariasis. *Adv Parasitol.* 1977;17:243-308.
- Arora VK, Gowrinath K. Pleural effusion due to lymphatic filariasis. *Indian J Chest Dis Allied Sci.* 1994;36:159-61.
- Palumbo E. Filariasis: diagnosis, treatment and prevention. *Acta Biomed.* 2008;79:106-9.
- Leeuwijn RS. Microfilaremia in Surinamese living in Amsterdam. *Trop. Geogr. Med.* 1962;14:355-60.
- Nanduri J, Kazura J. Clinical and laboratory aspects of filariasis. *Clinical Microbiology Reviews.* 1989;2(1):39-50.
- Fernando SD, Rodrigo C, Rajapakse S. Current evidence on the use of antifilarial agents in the management of bancroftian filariasis. *J Trop Med.* 2011;2011:175941.
- World Health Organization. *Weekly Epidemiol Rep.* 2012;37:345-56.
- World Health Organization. *Preparing and implementing a national plan to eliminate lymphatic filariasis.* Geneva: WHO. 2000.
- Addiss D. The global alliance to eliminate lymphatic filariasis. The 6th Meeting of the global alliance to eliminate lymphatic filariasis: a half-time review of lymphatic filariasis elimination and its integration with the control of other neglected tropical diseases. *Parasit Vectors.* 2010;3:100.
- Sabesan S, Palaniyandi M, Das PK, Michael E. Mapping of lymphatic filariasis in India. *Ann Trop Med Parasitol.* 2000;94:591-606.
- Freedman DO, de Almeida Filho PJ, Besh S, Maia e Silva MC, Braga C, Maciel A. Lymphoscintigraphic analysis of lymphatic abnormalities in symptomatic and asymptomatic human filariasis. *J Inf Dis.* 1994;170:927-33.
- Rosenblatt JE. Laboratory diagnosis of infections due to blood and tissue parasites. *Clin Infect Dis.* 2009;49:1103-8.
- John DT, Petri WA. "*Wuchereria bancrofti*." *Medical Parasitology.* 9th ed. St. Louis, Missouri: Saunders Elsevier. 2006:274-85.
- Ottesen EA. "Filariases and tropical eosinophilia" in *Tropical and Geographic Medicine* (K.S. Warren and A.A.F. Mahmoud, eds). McGraw-Hill Book Co., New York. 1984;390-412.
- Desowitz RS, Hitchcock JC. Hyperendemic bancroftian filariasis in the Kingdom of Tonga: the application of the membrane-filter technique to an age-stratified blood survey. *Am. J. Trop. Med. Hyg.* 1974;23:877-79.
- Amaral F, Dreyer G, Figueredo-Silva J, Noroes J, Cavalcanti A, Samico SC, et al. Live adult worms detected by ultrasonography in human bancroftian filariasis. *Am J Trop Med Hyg.* 1994;50:735-57.
- McCarthy J. Diagnosis of lymphatic filarial infection. In: *Lymphatic Filariasis*, ed. Nutman TB, Imperial College Press, London. 2000;127-41.
- Weil G, Lammie PJ, Weiss N. The ICT filariasis test: A rapid format antigen test for diagnosis of bancroftian filariasis. *Parasitol Today.* 1997;13:401-4.
- McCarthy J. Diagnosis of lymphatic filarial infection. In: *Lymphatic Filariasis*, ed. Nutman TB, Imperial College Press, London. 2000;127-41.
- Sharma S, Rawat A, Chowhan A. Microfilariae in bone marrow aspiration smears; their correlation with marrow hypoplasia: a report of six cases. *Indian J Pathol Microbiol.* 2006;49:566-68.
- Pradhan S, Lahiri VL, Elhence BR, Singh KN. The microfilariae of *Wuchereria bancrofti* in bone marrow smears. *Am J Trop Med Hyg.* 1976;25:199-200.
- Shenoi U, Pai RR, Pai U, Nandi GK, Adhikari P. Microfilariae in bone marrow aspiration smears. *Acta Cytol.* 1998;42:815-16.
- Pal BK, Kulkarni S, Bhandari Y, Ganesh BB, Goswami K, Reddy MV. Lymphatic filariasis: a possible pathophysiological nexus with oxidative stress. *Trans R Soc Trop Med Hyg.* 2006;100:650-55.
- Hemachandran M, Varma N, Varma S. Aplastic anaemia following a varicella infection with a coexistent microfilaraemia of *Wuchereria bancrofti*- a case report. *Indian J Pathol Microbiol.* 2003;46:662-63.
- Weekly epidemiological record. Geneva: World Health Organization. 2007;82(42):361-80.
- Park K. *Textbook of Preventive and Social Medicine.* 21st ed. Jabalpur: Banarsidas Bhanot Publishers; 2011. Epidemiology of communicable disease. 244-50.
- Azad K, Arora R, Gupta K, Sharma U. Lymphatic filariasis: Aspiration of adult gravid female worm from a soft tissue swelling. *J Cytol.* 2010;27(4):156-7.

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