40 *Wuchereria bancrofti*

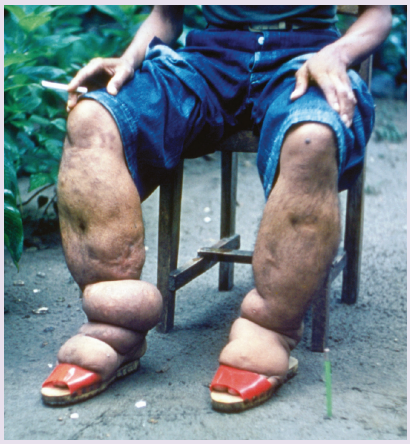

Figure 40.1 This man has more advanced swelling of his legs than in the case history. His bilateral lymphedema is more advanced and will not resolve with treatment. *From the Centers for Disease Control & Prevention, Atlanta, Georgia. Image is found in the Public Health Image Library #373.*

A 24-year-old man from Southern India presented with swelling of his left foot. The swelling diminished overnight. He was otherwise well with no fevers or systemic upset. Initially, there was concern that he had a deep vein **thrombosis**, but his D-dimers were not raised. There were a few enlarged lymph nodes in his groin and, on closer questioning, he reported previous episodes with the nodes becoming painful and swollen. In his hometown in India he recalled seeing individuals with grossly swollen legs. His full blood count revealed an **eosinophilia** of 1.4×10^9 L^{-1} (normal range $<0.45\times10^9$ L^{-1}). His filarial **serology** was positive. He was treated with a prolonged course of doxycycline. The eosinophil count fell to 0.4×10^9 L^{-1} and his swelling subsided (**Figure 40.1**).

1. WHAT IS THE CAUSATIVE AGENT, HOW DOES IT ENTER THE BODY AND HOW DOES IT SPREAD A) WITHIN THE BODY AND B) FROM PERSON TO PERSON?

CAUSATIVE AGENT

Wuchereria bancrofti is the causative agent of Bancroftian filariasis. It is a round worm, or nematode which, in the adult form, lives in lymphatic vessels. Females are about 4–10 cm long and males about 2–4 cm. However, they are very slender, being 0.25 and 0.3 mm in width, respectively. Other filarial nematodes that cause a similar illness are *Brugia malayi* and *B. timori*, which are confined to South-East Asia.

ENTRY AND SPREAD WITHIN THE BODY

Infection occurs with the bite of various species of mosquito vector (see below). Larvae enter the tissues and are thought to migrate along the lymphatics. They commonly reach the lymphatics draining to lymph nodes in the groin and sometimes the armpit. Adult worms mature, mate and, after about 8 months, females release ensheathed microfilariae from the ova in their uterine bodies. These circulate in the bloodstream and are available to be ingested when a mosquito takes a blood meal (**Figure 40.2**). The life cycle of *W. bancrofti*

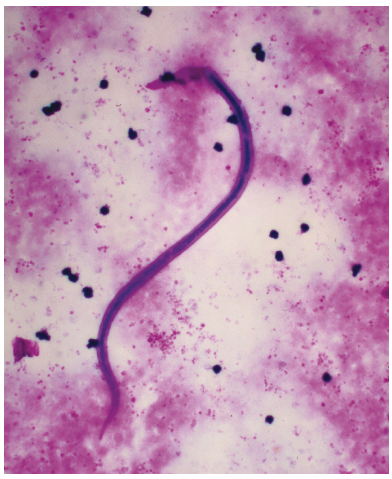

Figure 40.2 *Wuchereria bancrofti* microfilariae in a blood film. *From the Centers for Disease Control & Prevention, Atlanta, Georgia. Image is found in the Public Health Image Library #3008. Additional photographic credit is given to Dr Mae Melvine who took the photo in 1978.*

is shown in **Figure 40.3**. In areas where mosquitoes bite at night microfilariae appear in the bloodstream in maximum numbers around midnight. In other areas, mainly the Pacific Islands, mosquitos bite during the day and night, and microfilarial levels remain relatively constant in the bloodstream throughout the day. Microfilariae have a lifespan of about 1 year. Adult worms are capable of living up to 15 years, but usually for about 5 years.

PERSON-TO-PERSON SPREAD

Depending on geographic location, *W. bancrofti* is transmitted by *Anopheles*, *Aedes*, *Culex* or *Mansonia mosquitoes*. When mosquitoes ingest microfilariae-laden blood, the microfilariae pass into the midgut, shed their sheath, and penetrate the wall of the midgut to enter the body cavity. They migrate to muscles in the thorax and undergo two molts. The third stage larvae are the infective form and pass onto the mouth parts of the mosquito. Upon blood feeding, the larvae are deposited onto the skin surface and enter the host via the puncture made by the mosquito. Humans are the only host for *W. bancrofti* and the reservoir for infection is the local microfilaremic human population.

EPIDEMIOLOGY

Infections occur in Asia, Africa, Pacific Islands, South America, and the Caribbean. In 2000, the World Health Organization (WHO) estimated that there were 120 million cases of lymphatic filariasis in 80 countries, with 40 million individuals being severely incapacitated. Through elimination efforts, the estimated prevalence in 2018 was 51 million cases with about 38 million of these in South-East Asia. In endemic areas, infections establish from childhood and rates rise with increasing age.

2. WHAT IS THE HOST RESPONSE TO THE INFECTION AND WHAT IS THE DISEASE PATHOGENESIS?

Repeated mosquito bites expose individuals to the third stage larvae (L3). These go on to mature into adult worms, which mate and, in turn, release microfilariae. The immune response may be directed against infecting L3 larvae, adult worms or microfilariae and may involve antibody, lymphocytes, and other mononuclear cells. The immune response to each of

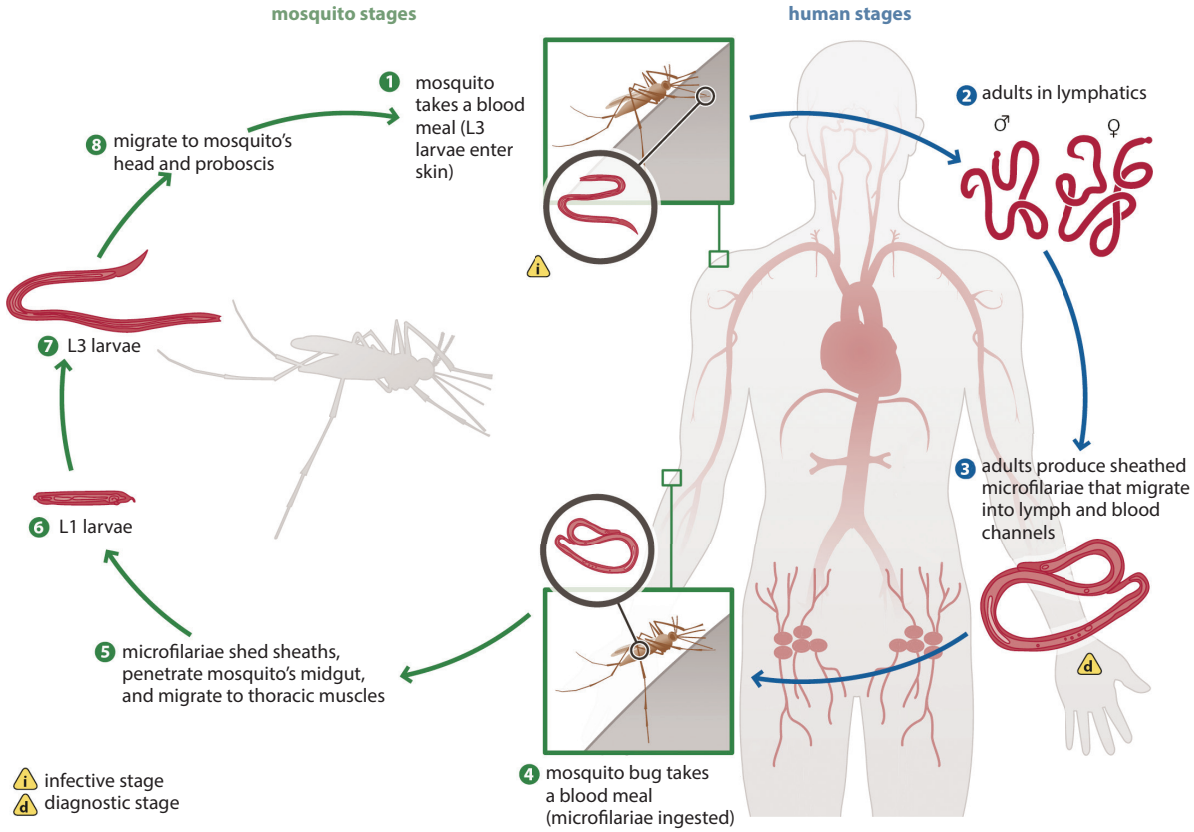

Figure 40.3 Life cycle of *Wuchereria bancrofti*. L3 larvae escape from the mosquito onto the skin surface at the time of taking a blood meal from humans (1). The larvae then enter the human host through the puncture made by the mosquito. The larvae mature into adults, which settle in lymphatics (2). Male and female adult worms mate and then females release ensheathed microfilariae larvae, which circulate in the blood (3). These may be ingested by mosquitoes feeding on humans (4). In the mosquito mid-gut, the sheaths are shed (5). Larvae then mature into the L3 form, which passes to the mosquito mouthparts (6-8). *From the Centers for Disease Control & Prevention, Atlanta, Georgia. Image is found in the Public Health Image Library #3425. Additional photographic credit is given to Alexander J da Silva, PhD, and Melanie Moser who created the image in 2003.*

these stages seems to be segregated. Some individuals will progressively stimulate a protective immune response to the L3 larvae. This may become so effective by adulthood that new infections can no longer occur. Adult worms present within lymphatics will eventually die and so, in an endemic community, there will be some who are immune and clear of infections.

In vitro, microfilariae can be killed by **IgE**-mediated degranulation of eosinophils and mast cells. This killing mechanism may be modified. In endemic areas, there will be mothers who have filariasis at the time of pregnancy. Microfilariae have been seen in cord blood and newborns have **IgM** and IgE antibodies to microfilarial antigen, as well as IgG. The IgM and IgE **antibody isotypes** do not cross the placenta and their production implies *in utero* microfilarial antigen exposure. It is postulated that this early antigen exposure modifies the subsequent immune response when children are exposed to microfilariae. In some studies in children, anti-microfilarial IgE-mediated degranulation of eosinophils and mast cells does not occur. It is thought that high titers of **IgG**4 block anti-microfilarial IgE in an antigen-specific way. This enables the survival of microfilariae. Peripheral blood lymphocytes taken from teenagers born to microfilaremic mothers do not proliferate *in vitro* and produce **interferon-γ** (**IFN-γ**) to the same extent as controls. There is a dominance of **Th-2** responses over **Th-1** responses, with potentially **interleukin** 10 (IL-10) and **transforming growth factor-β** (**TGF-β**) down-regulating Th-1 responses. At the molecular level, there is down-regulation of the expression of **Toll-like receptors** on T cells (TLR1, TLR2, TLR4, and TLR9), which means that they are less responsive to stimulation through these receptors. One contributor to immunomodulation is the release by adult worms of the phosphorylcholine containing molecule ES-62, which has a variety of immunosuppressive effects. Thus, by various possible means, the immune response to infection is down-regulated through childhood and adolescence. This results in persistent infection and microfilaremia.

Immune responses do not appear to be effective in killing adult worms. However, cumulative exposure to adult worms, alive or dying, stimulates inflammatory immune responses to adult worm antigens. Adult worms also carry an endosymbiont bacterial organism called *Wolbachia*. Antigens from *Wolbachia* also stimulate an immune response. These may reach a stage where intense inflammatory reactions occur around adult worms in lymphatics. Microfilarial sheath antigen also triggers an inflammatory reaction via TLR4, dendritic cell involvement, and a Th-1 response. Inflammatory reactions contribute to lymphatic pathology. This will not be the only contributory mechanism. Adult worms themselves can cause dilatation of the lymphatics, which is called **lymphangiectasia**, without an accompanying inflammatory response. Secondary bacterial infections have been shown to contribute to the maintenance of lymphatic pathology.

A small minority of individuals experience an immune response to microfilariae in the lung. The resulting inflammatory process results in the clinical condition of tropical pulmonary eosinophilia.

There is variation between individuals and populations in the pathogenesis of disease. There is a genetic component to immune responses, with familial aggregation of certain forms of the clinical disease. People who migrate into endemic areas do not have the possible benefit of a modified immune response from childhood. They experience stronger immune responses and more lymphatic pathology. The same applies to those in endemic areas not born to microfilaremic mothers.

3. WHAT IS THE TYPICAL CLINICAL PRESENTATION AND WHAT COMPLICATIONS CAN OCCUR?

There is a spectrum of clinical manifestations. The majority of individuals, about 70%, remain asymptomatic while others experience complications related to the adult worms or the microfilariae.

A common clinical manifestation of lymphatic filariasis is swelling in the scrotum, a **hydrocele**, whereby adult *W. bancrofti* localize in the scrotal lymphatics and lymphatics of the spermatic cord. Adult worms can also cause acute inflammation of lymph nodes and lymphatics, **adenolymphangitis**, accompanied by fever. Episodes of acute adenolymphangitis probably relate to the death of adult worms and settle over a week but can recur. The lymphatic channels become damaged and dilate (lymphangiectasia). Poor lymphatic drainage predisposes to secondary bacterial infection of the skin. When bacteria inflame afferent lymphatics, red streaks appear along their course (**lymphangitis**) and further lymphatic damage occurs. Eventually **lymphedema** can develop in the limbs or genitals, which can lead to **elephantiasis** (**Figures 40.1** and **40.4**). Lymphedema stretches the skin and causes it to thicken, with underlying **fibrosis**. Pushing a finger into the swollen leg does not create an indented pit. Cracks in the skin can lead to further

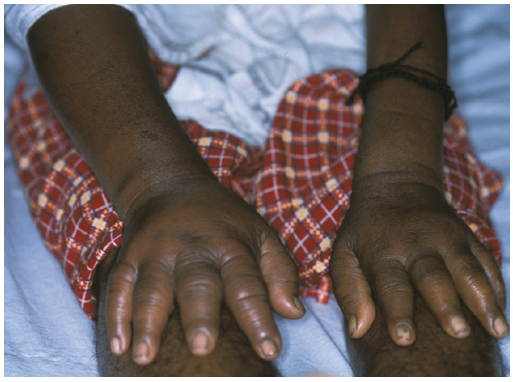

Figure 40.4 Lymphedematous arms. *From the World Health Organization, Special Programme for Research and Training in Tropical Diseases, http://www.who.int/tdr/index.html. Image #01021639. Additional photographic credit is given to Andy Crump who took the photograph in 2001 in India.*

secondary bacterial infection, aggravating the damage already done to the lymphatics. Sometimes blocked lymphatics within the abdomen can result in lymph discharging into the urinary tract. Urine takes on a milky appearance due to the fat content of lymph. This is called **chyluria**.

The presence of microfilariae in the bloodstream is in general asymptomatic but may rarely be associated with intermittent, nocturnal fevers. A strong immune response may be manifest in the lung. As microfilariae pass through the lung they cause an inflammatory reaction, with fever, cough, and wheeze and widespread inflammatory infiltrates on a chest X-ray. This is most pronounced at night. The eosinophil count is raised and there is a high titer of antifilarial antibodies. This respiratory manifestation is called tropical pulmonary eosinophilia (TPE) and is usually observed in Asia rather than Africa or South America. TPE is actually quite rare and affects about 1% of infected subjects. In TPE, microfilariae are actually scanty in the peripheral blood and are not found on blood films. Recurrent bouts of TPE damage the lungs with fibrosis. Bouts can be curtailed with a micro-filaricidal drug, diethylcarbamazine (see Section 5).

4. HOW IS THE DISEASE DIAGNOSED, AND WHAT IS THE DIFFERENTIAL DIAGNOSIS?

The diagnosis of lymphatic filariasis due to *W. bancrofti* may be based on visualizing the parasite, detecting antigen, serology or clinical features.

Adult worms are small and are rarely found in tissue. Careful ultrasound of the scrotum may sometimes reveal wriggling adults in lymphatic vessels. Microfilariae may be found in chyluria, but parasitologic diagnosis is usually based on finding microfilariae in a peripheral blood film. This requires nocturnal blood sampling in areas where lymphatic filariasis is nocturnally periodic. One can provoke microfilariae to appear in the bloodstream during the day by giving the drug diethylcarbamazine (DEC) and taking blood 30–60 minutes later. The DEC provocation test is as sensitive as a nocturnal blood film. As filariasis is a chronic infection, microfilariae may be present in the blood over prolonged periods. In endemic areas, microfilariae may be present in the blood when subjects succumb to another infection, such as malaria. Thus, a positive microfilarial blood film does not necessarily mean that the current illness is due to filariasis.

There are now tests for filarial antigen. If positive, they indicate current infection. Antigen is present in the blood in the day and the night and there is no need for nocturnal blood sampling. Current tests are highly sensitive and specific for Bancroftian filariasis. There are two commercial antigen tests. One is an **enzyme-linked immunosorbent assay** (**ELISA**) and the other an immunochromato-graphic card test. Serologic tests for antifilarial antibodies have suffered from poor sensitivity and specificity. In endemic areas, it is difficult to distinguish between current and past infection. Tests have suffered from cross-reactions with other parasites.

Eosinophilia is common and reflects the responses to all life stages. Elephantiasis, once established, is unmistakable, but earlier stages of infection have differential diagnoses and can warrant specific investigation.

Across the clinical spectrum of filariasis, there are differentials for different manifestations, including intermittent fevers, **lymphadenopathy**, **edema**, cough, wheeze, and eosinophilia. **Febrile** illness may be due to a long list of infections. Those that do not settle spontaneously include malaria, tuberculosis, and HIV. The differential diagnosis of lymphadenopathy is discussed with the *Toxoplasma* case. A swollen limb may be due to local **cellulitis**, a deep vein thrombosis, heart failure or low protein states from **nephrotic syndrome** or **cirrhosis**. Edema once it has reached the stage of elephantiasis in most endemic areas is unmistakable. In certain parts of Africa, walking barefoot introduces silicates and minerals into the skin. When these pass along the lymphatics and to lymph nodes there is a fibrotic reaction that can cause lymphatic obstruction and gross swelling. This is called **podoconiosis** and can be as severe as elephantiasis. Intermittent cough, wheeze, and eosinophilia have to be differentiated from asthma, some connective tissue diseases, **allergic alveolitis**, and lung migratory phases of intestinal nematode infections. Eosinophilia is discussed with the *Schistosoma* case.

5. HOW IS THE DISEASE MANAGED AND PREVENTED?

MANAGEMENT

There are four key drugs that have been used to treat filariasis, and each has differing effects on microfilariae and on adult worms. Drug efficacy can be monitored by blood films, ultrasound examination for adult worms, and antigen tests.

DEC has been used for several decades. It kills about 70% of microfilariae and about 50% of adult worms. It has been the backbone of attempts to eradicate filariasis. It has to be used in repeated annual cycles to reduce transmission and adult worm numbers. Ultimately, eradication of infection requires death of all adult worms, either naturally or through treatment. DEC can cause headaches and gastrointestinal (GI) upset. Dying adult worms excite episodes of adenolymphangitis. DEC is used to abort episodes of TPE due to microfilariae in the lung. However, DEC should not be used in co-infections with onchocerciasis and used with caution with loiasis due to adverse inflammatory reactions.

Albendazole can be used with DEC and ivermectin. Ivermectin is very good at killing microfilariae in the short term but has no effect on adult worms. Microfilariae therefore reappear in the blood rapidly.

Living within adult *W. bancrofti* are bacterial endosymbionts called *Wolbachia*. *Wolbachia* can be killed

with the antibiotic doxycycline and if this antibiotic is used in patients with filariasis adult *W. bancrofti* die. In a trial of an 8-week course of doxycycline, there was an 80% reduction in adult worms and a loss of microfilariae in the blood 14 months after treatment. Doxycycline is avoided in pregnancy, breast-feeding mothers, and children under 12 years because of its effects on bones and teeth. These features preclude the use of doxycycline in mass treatment programs. The use of doxycycline in the case history is not typical of treatment worldwide.

Beyond specific antifilarial drugs, subjects with lymphedema need a lot of help and support in the care of their swollen limb(s) (**Figure 40.5**). Secondary bacterial infections need to be avoided. This requires attention to skin care, elevation of the leg to reduce swelling if possible, and prompt use of antibiotics if infection occurs. Sometimes surgery has been employed, but with limited success for swollen legs. The most effective treatment for patients suffering from hydrocele is surgery.

PREVENTION

Since 2000, the Global Progam to Eliminate Lymphatic Filariasis (GPELF) has continued elimination efforts. The mainstay of elimination efforts is periodic mass drug administration (MDA). Entire, defined populations are given chemotherapy irrespective of whether they are microfilaremic or amicrofilaremic. This is simpler than testing every individual by blood films. Repeated MDA at annual intervals is intended to reduce levels of microfilaremia so that it is less likely for mosquitoes to transmit infection. A high level of population coverage is required for this to work. MDA must also be repeated for a number of years, usually 4–6 years to cover the average lifespan of adults and their release of microfiliariae. However, in recent years, trials have shown that a single dose, three-drug combination of DEC, albendazole, and ivermectin cleared microfilariae for three years in the majority of recipients.

Figure 40.5 Woman carefully washing her lymphedematous leg to reduce the chances of secondary bacterial infection. *From the World Health Organization, Special Programme for Research and Training in Tropical Diseases, http://www.who.int/tdr/index.html. Image #01021471. Additional photographic credit is given to Andy Crump who took the photograph in 2001 in Haiti.*

In areas where **onchocerciasis** is present, DEC causes severe reactions in individuals co-infected with *Onchocerca volvulus*. Therefore, in these areas, ivermectin is used in combination with albendazole as part of MDA.

MDA has to work in conjunction with mosquito control in the elimination of lymphatic filariasis. This has a broader context in the prevention of malaria and other mosquito-borne diseases. Efforts in mosquito control and MDA work better combined than in isolation.

SUMMARY

1. WHAT IS THE CAUSATIVE AGENT, HOW DOES IT ENTER THE BODY, AND HOW DOES IT SPREAD A) WITHIN THE BODY AND B) FROM PERSON TO PERSON?

- Bancroftian, lymphatic filariasis is caused by *Wuchereria bancrofti*, a tissue-dwelling round worm or nematode.
- Male and female adult worms live in the lymphatics.
- Female worms release larvae called microfilariae which enter the bloodstream.
- Various species of mosquitoes ingest microfilariae in the blood and pass infection onto others.
- *Wuchereria* is only found in humans, and there is no animal reservoir.

2. WHAT IS THE HOST RESPONSE TO THE INFECTION AND WHAT IS THE DISEASE PATHOGENESIS?

- In endemic areas, pregnant mothers may have chronic filarial infection and the fetus is exposed to filarial antigen. This early exposure may modify the subsequent immune response in children.
- IgG4 antibodies may block the effects of IgE in an antigen-specific manner.
- Th-1 and Th-2 responses can be down-regulated in children and adolescents.
- Repeated exposure to third-stage larvae stimulates immunity, which may prevent further infection in older subjects.

Continued...

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- Some adult individuals also develop immune responses to worms that cause lymphatic pathology.
- In Asia, there are occasional individuals who mount a strong anti-microfilaremic response, which causes intermittent pulmonary pathology with a marked peripheral eosinophilia.

3. WHAT IS THE TYPICAL CLINICAL PRESENTATION AND WHAT COMPLICATIONS CAN OCCUR?

- Infection is often asymptomatic.
- Adult worm death can cause intermittent inflammatory episodes of lymph nodes and lymphatics.
- Lymphatic damage leads to lymphedema and potentially gross swelling of dependent parts.
- Microfilariae in the blood can cause intermittent fevers.
- Tropical pulmonary eosinophilia is the clinical manifestation of a strong immune response to microfilariae with fever, cough, wheeze, and infiltrates on a chest X-ray.

4. HOW IS THE DISEASE DIAGNOSED, AND WHAT IS THE DIFFERENTIAL DIAGNOSIS?

- The presence of microfilariae in a blood film indicates current infection.
- Microfilariae may be present in the blood over long periods and therefore care must be taken in attributing the current illness to filariasis.
- Filarial antigen tests, if positive, also indicate current infection.
- In endemic areas, gross lymphedema and hydrocele is usually diagnostic of lymphatic filariasis.

5. HOW IS THE DISEASE MANAGED AND PREVENTED?

- Diethylcarbamazine (DEC) and ivermectin kill microfilariae. These are often used in conjunction with albendazole to maximize killing of adult worms.
- Doxycycline can kill adult worms through its effect on the endosymbiont bacteria, *Wolbachia*. When these bacteria are killed, the adult worms die.
- Patients complicated with gross lymphedema require careful attention to their swollen limbs and prevention of secondary infection.
- Control programs have involved repeated, annual community-wide use of combinations of drugs to reduce microfilarial numbers for transmission by mosquitoes.

FURTHER READING

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GAELF, Global Programme to Eliminate Lymphatic Filariasis: www.filariasis.org/

World Health Organization, Lymphatic filariasis (elephantiasis): https://www.who.int/health-topics/lymphatic-filariasis

Students can test their knowledge of this case study by visiting the Instructor and Student Resources: [www.routledge.com/cw/lydyard] where several multiple choice questions can be found.