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Article

Lymphatic filariasis elimination status: *Wuchereria bancrofti* infections in human populations after five effective rounds of mass drug administration in Zambia

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Abstract:

Lymphatic filariasis (LF), also commonly known as elephantiasis, is a neglected tropical disease (NTD) caused by filarial parasites. The disease is transmitted by a bite from infected mosquitoes. The bites of these infected mosquitoes deposit filarial parasites, *Wuchereria* or *Brugia* whose predilection site is the lymphatic system. The damage to the lymph system causes swelling in the legs, arms, and genitalia. A mapping survey conducted between 2003 and 2010 determined LF to be endemic in Zambia in 96 out of 116 districts. Elimination of LF is known to be possible by stopping the spread of the infection through large-scale preventive chemotherapy. Therefore, mass drug administration (MDA) with diethylcarbamazine citrate (DEC) (6 mg/kg) and Albendazole (400 mg) for Zambia has been conducted and implemented in all endemic districts with five effective rounds. Post-MDA pre-transmission assessment survey (pre-TAS) was conducted between 2021 and 2022 in 80 districts to determine the LF prevalence rate. We conducted a cross-sectional seroprevalence study involving 600 participants in each Evaluation Unit (EU) or each district. The study sites (sentinel and spot-check sites) were the districts which were the Implementation Unites (IUs) where MDA, a preventative chemotherapy against LF was conducted. These included 80 districts from the 9 provinces. A total of 47,235 people from sentinel and spot-check locations were tested. Of these, valid tests were 47,052 of which 27,762 (59%) were females and 19,290 (41%) were males. The survey revealed in the 79/80 endemic district a prevalence of *Wb* antigens of 0.14% and 0.0% prevalence of microfilariae. All the surveyed districts had an optimum prevalence of less than 2 percent, except for Chibombo district. The majority of participants that tested positive to *Wb* Ag were those that had 2, 3 and 4 rounds of MDA. Surprisingly, individuals that had 1 round of MDA were not found to have circulating antigens of *Wb*. The study showed that all the surveyed districts except for Chibombo, passed Pre-TAS. This further implies that there is need to conduct a TAS in these districts in order to decide whether to stop MDA or not.

Keywords: Lymphatic Filariasis**;** pre-TAS; Prevalence; Antigenaemia; Microfilariae; Zambia

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1. Introduction

Lymphatic Filariasis (LF), also known as "elephantiasis" is a deforming and disabling disease that is caused by roundworm parasites of the genera *Wuchereria* and *Brugia* that are transmitted by mosquitoes (Coulibaly et al. 2016; Dewi et al. 2015). It is an infection of human lymphatic system by filarial worms (Nutman 2013; Pfarr et al. 2009; Ramzy et al. 2019).

In Africa, the common filarial worm which causes this disease is the species *Wuchereria bancrofti* (Manguin et al. 2010; Small, Tisch, and Zimmerman 2014). In Western province of Zambia, the disease is locally known as "*Mbumba*" by the Lozi people. Whereas the Nsenga of Eastern province call it "*Msakasa*", while the Tumbuka call it "*Mchecha*" or "*Vimba*", which literally means "swelling". The disease is characterized by swelling (edema) mainly of the lower limbs with thickening of the skin and underlying tissues. Elephantiasis only results when the parasites (worms) lodge in the lymphatic system (Penzer 2003; Zulfiqar and Malik 2023). The swelling develops when the adult worms cause a partial or complete blockage of the flow of lymph, the fluid which drains the tissues and flows in the lymphatic vessels.

Lymphatic Filariasis is targeted for elimination as a public health problem by the year 2030 through the treatment of entire populations at-risk with repeated annual mass drug administration (MDA) (Santoso et al. 2020). Essential for programme success is defining and confirming the appropriate endpoint for MDA when transmission is presumed to have reached a level low enough that it cannot be sustained even in the absence of drug intervention (Chu et al. 2013). Guidelines advanced by World Health Organization (WHO) call for a pre-transmission assessment survey (pre-TAS) to determine if MDA can be stopped within an LF evaluation unit (EU) after at least five effective rounds of annual treatment.

The Global Programme to Eliminate Lymphatic Filariasis is the largest public health intervention programme attempted to date through MDA (Ramaiah and Ottesen 2014). It is worth noting that MDA does not cure filarial infections, but it can reduce or interrupt transmission of new infections by clearing larval parasites from human blood so that they are not available for mosquitoes which are vectors.

Evaluation of the MDA is necessary to determine whether the programme has achieved its objective of reducing levels of LF microfilariae in endemic populations to an extent where transmission is likely no longer sustainable. Programmes must be able to assess whether MDA has succeeded in lowering the prevalence of infection to a level where recrudescence is unlikely to occur (Santoso et al. 2020). Thus, TASs are designed to help programme managers determine whether areas have reached this critical threshold of infection (Chu et al. 2013). While the TAS provides helpful evidence to national programmes regarding the decision to stop MDA, programme managers must thoughtfully consider the decision about whether to stop or continue MDA.

2. Materials and Methods

2.1. Study Design and Population

Using data from a cross sectional sero-prevalence community-based study conducted in eighty (80) evaluation units (EUs) we estimated the sero-prevalence of LF in Zambia. Each administrative/geographic district which was an implementation unit (IU) for MDA was designed to be an EU with two study sites each – a sentinel site and a spot-check site which was designated as a control site. The study enrolled persons who were aged 5 years and above at the time of the fifth MDA round. Each of the sentinel site or spot-check site was targeted to enroll 300 participants totaling 600 participants per district.

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The MDA was administered annually for five (5) years in every endemic district and an effective coverage was 65% and above.

2.2. Sampling Strategy

The study sites (sentinel and spot-check sites) were the districts which were the IUs where MDA, a preventative chemotherapy against LF was conducted. These include 80 districts from the 9 provinces that were surveyed namely: Central, Muchinga, Eastern, Copperbelt, Northwestern, Southern, Lusaka, Northern and Luapula provinces.

2.3. Timeline of the survey

The baseline data was already collected prior to the first round of the MDA (Mwase et al. 2014; Shawa et al. 2013). Data on microfilariae (Mf) prevalence or antigenemia (Ag) prevalence using blood-film microscopy and ICT (for *W. bancrofti* antigens) was also collected. The standard timeline used for the pre-TAS(s) is 6 months after the 5th round of LF MDA (Bah et al. 2020).

2.4 Determination of the prevalence of LF antigen

The Alere™ Filarial Testing Strips (FTS) were used to test for LF antigen. The middle or index finger of consenting individuals was cleaned using a cotton ball soaked in 70% alcohol. After drying, the tip of the finger was pricked using a sterile lancet and blood immediately collected using capillary tubes and loaded on sample pad of the FTS test. The results were then read by a trained timekeeper exactly after 10 minutes. The test requires only 75*ul* of blood to be collected from each participant and added to the card which gives a result in 10 minutes while still in the field. Thus, all participants had 75*ul* of blood withdrawn from them and used on an FTS to determine the prevalence of the LF antigen in the community.

2.5 Investigation of the intensity of microfilariae infection

Any participants who tested positive on the FTS had further blood samples (100*ul*) taken that evening of the day of sampling between 22:00 hours and 02:00 hours to detect the presence of *W. bancrofti* microfilariae. The blood samples were taken at night because of the Mf's behavior of nocturnal periodicity, whereby in the day, the microfilariae are in the deeper blood vessels and in the night, they are in the superficial capillaries. Therefore, for this purpose, samples were commonly taken between 22:00 hours and 02:00 hours.

Blood night sampling was arranged with the individual at the time of results feedback, informing them of their FTS result. In this traditional gold standard technique, blood smears for the detection of Mf, required the collection of 100ul of blood from each participant which were clearly labelled and stored in 1.5ml vials until further analysis in the laboratory. In the laboratory, the blood samples were evaluated for Mf intensity using the Sedgewick Rafter counting chamber. After examining the blood using the counting chambers, some of it was applied onto a slide, dried and stained (with Giemsa) before being examined (Hira 1976, 1977).

2.6. Questionnaire

A questionnaire capturing information (age, sex, received MDA, MDA round, province of residence, district of residence, nearest health facility, study site, village/settlement, blood test results for Antigenemia and blood test results for microfilariae) on the individuals tested and whether they had participated in the MDA or not was carried out and results recorded using an Open Data Kit (ODK) on android tablet and phone.

2.7. Statistical analysis

The Pearson's uncorrected Chi-square test was used to compare proportions at 5% significance level. Associations were established disregarding 'Don't know category.'

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2.8 Managing the positive cases

The *Wb Ag* positive cases were treated with a single dose of a combination of Albendazole (400 mg) plus diethylcarbamazine (6 mg/kg). This treatment regime is due to the fact that no investigations have been done to investigate the endemicity of Onchocerciasis caused by *Onchocerca volvulus* (River blindness) in Zambia, and thus Zambia is said to be non-endemic for Onchocerciasis. Therefore, the drugs used for MDA against LF are Diethylcarbamazine (DEC) and Albendazole (ALB)(Ottesen and Horton 2020).

3. Results

A total of 47,235 participants were tested and interviewed from 148 sites (70 sentinel and 78 spot-check sites) (Table 3.1). Of these, valid tests were 47,052 of which 27,762 (59%) were females and 19,290 (41%) were males. Most of the people sampled were older than 15 years.

Table *3.1.* **Number of endemic districts and tests conducted by province (n = 47,235)**

Province	Number of Endemic districts	Districts with +ve Wb Ag	Tests (A)	Valid tests (B)	% Valid tests (B/A*100) (C.)	Invalid tests (D)	% Invalid test (D/A*100)
Central	12	7	7,358	7,317	99.44	41	0.56
Copperbelt	9	2	5,044	5,042	99.96	2	0.04
Eastern	7	4	4,176	4,176	100.00	0	0.00
Luapula	6	5	3,587	3,550	98.97	37	1.03
Lusaka	6	1	3,674	3,670	99.89	4	0.11
Muchinga	6	2	2,869	2,828	98.57	41	1.43
N/Western	11	3	6,523	6,523	100.00	0	0.00
Northern	12	5	7,352	7,303	99.33	49	0.67
Southern	11	2	6,652	6,643	99.86	9	0.14
Zambia	**80**	**31**	**47,235**	**47,052**	**99.61**	**183**	**0.39**

The overall prevalence of *Wb Ag* was 0.14% (Table 3.2). The *Wb Ag* prevalence was higher (0.2%) in male than female participants. With regard to age, the older age group of above 15 years of age had higher (0.16%) prevalence of *Wb Ag* than the age group less than 15 years which had a prevalence of 0.11%. The highest provincial *Wb Ag* prevalence was in Central province at 0.29% while the lowest provincial *Wb Ag* prevalence was in Lusaka province at 0.03%.

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Table 3.2 Prevalence of *Wb* Ag by sex, age and province (n = 47,052)

Characteristics		Total	Positive (%)	(95% CI)	*p*-value
Total		47,052	65 (0.14)	0.14 [0.11, 0.18]	
Sex					
	Female	27,762	26 (0.09)	0.09 [0.06, 0.14]	0.002
	Male	19,290	39 (0.20)	0.20 [0.14, 0.28]	
Age (years)					
	≤15	17,750	19 (0.11)	0.11 [0.06, 0.17]	0.158
	>15	29,302	46 (0.16)	0.16 [0.11, 0.21]	
Province					
	Central	7,317	21 (0.29)	0.29 [0.18, 0.44]	<0.001
	Copperbelt	5,042	3 (0.06)	0.06 [0.02, 0.17]	
	Eastern	4,176	6 (0.14)	0.14 [0.05, 0.31]	
	Luapula	3,550	10 (0.28)	0.28 [0.15, 0.52]	
	Lusaka	3,670	1 (0.03)	0.03 [0.00, 0.01]	
	Muchinga	2,828	3 (0.11)	0.11 [0.02, 0.31]	
	N/Western	6,523	4 (0.06)	0.06 [0.02, 0.16]	
	Northern	7,303	15 (0.22)	0.20 [0.11, 0.34]	
	Southern	6,643	2 (0.03)	0.03 [0.00, 0.11]	

About 61.5% and 73.8% of the participants aged less than 15 years and above 15 years, respectively, received MDA (Table 3.3). Significantly (p<0.001), more older participants received MDA than younger participants. More female participants at 71.3% than male participants at 65.9% received MDA which finding was statistically significant (p <0.001). The *Wb Ag* positivity amongst the participants that received MDA was not related to the number of rounds or doses (p = 0.579)

Table 3.3. Participation in MDA according to age, sex and MDA rounds (n = 47,052)

Age (years)	Total	Don't know (%)	No (%)	Yes (%)	p-value
≤15	17,750	305 (1.7)	6,534 (36.8)	10,911 (61.5)	<0.001
>15	29,302	128 (0.4)	7,563 (25.8)	21,611 (73.8)	
Total	**47,052**	**433 (0.9)**	**14,097 (30.0)**	**32,522 (69.1)**	

Sex	Total	Don't know (%)	No (%)	Yes (%)	p-value
Female	27,762	264 (1.0)	7,697 (27.7)	19,801 (71.3)	<0.001
Male	19,290	169 (0.9)	6,400 (33.2)	12,721 (65.9)	
Total	**47,052**	**433 (0.9)**	**14,097 (30.0)**	**32,522 (69.1)**	

MDA Round (A)	Total received MDA. (B)	Proportion received MDA. (C)	Number of Wb Ag + ve cases (D)	Prevalence, % [D/B*100] (E)	p-value
1	13,482	41.46	0	0.00	0.579

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2	12,304	37.83	17	0.13
3	5,036	15.48	17	0.34
4	1,027	3.16	10	0.97
5	673	2.07	0	0.00
	47,522	**100.00**	**44**	**0.14**

The positivity amongst participants that received MDA were 0.14% and amongst participants that did not receive MDA was also 0.14% (Table 3.4). Therefore, there existed no statistically significant difference (p= 0.803) in the proportion of *Wb* Ag positivity between the two groups.

Table 3.4. *Wb* Ag positivity by MDA (n = 47,052)

	valid tests	***Wb* Ag.+ ve**	***Wb* Ag. + ve %**	**p-value**
With MDA	32,522	44	0.14	0.803
Without MDA	14,530	21	0.14	
	47,052	**65**	**0.14**	

4. Discussion

It has been shown and therefore accepted that LF can be eliminated as a public health problem after a minimum of five effective rounds of MDA and demonstrating low prevalence in succeeding assessments (Niles et al. 2021). The first assessments recommended by WHO are sentinel and spot-check sites, referred to as pre-TAS, which are conducted in each implementation unit after MDA (Burgert-Brucker et al. 2020). All IUs should have had at least 5 effective (≥65%) rounds of MDA and in all sentinel and sport-check sites the prevalence of Mf should be <1% or the *Wb Ag* be <2% at all sites after the last effective round (Deribe et al. 2020). Failure to pass pre-TAS means that further rounds of MDA are required(Burgert-Brucker et al. 2020).

Following the successful conducting of five rounds of LF MDA, Zambia went ahead and conducted a Pre-TAS. The survey revealed in the 79/80 endemic districts a prevalence of *Wb Ag* of 0.14% which on the overall this *Wb Ag* prevalence is less than the target 2%. and 0.0% prevalence of Mf which is within the target of less than 1%. Of the districts surveyed, it was only Chibombo which had the highest prevalence of 2% *Wb Ag*. This is the only district whose Wb Ag prevalence failed to meet the threshold of less than 2% antigenemia for an EU/IU to pass a Pre-TAS. This could then suggest that all the surveyed districts except for Chibombo, passed Pre-TAS. This further implies that there is need to conduct a TAS in these districts to decide whether to stop MDA or not. The results also indicate that in Zambia, Chibombo is the only district where MDA should be continued. When a country or an IU fails to meet the established thresholds in a pre-TAS, at least two more rounds of MDA must be implemented, this should be the case with Chibombo district (Deribe et al. 2020).

The finding that no individuals that were positive on FTS had Mf is not surprising. This is because individuals infected by filarial worms may be described as either microfilaraemic or amicrofilaraemic, depending on whether microfilariae can be found in their peripheral blood. Filariasis is diagnosed in microfilaraemic cases primarily through direct observation of microfilariae in the peripheral blood. The other reaon is that it could be due to the impact of the MDA and other cross-cutting interventions like that of malaria vector control interventions (Berg, Kelly-Hope, and Lindsay 2013).

The results showed that more women were interviewed than men. Similarly, more females participated in MDA than men. The reason for this could partly be that the surveys were done at the beginning of the rainy season (December to January) and so most men could have been busy preparing their crop (maize) fields. The other reason is that generally, when you examine census population figures in Zambia, there are more women

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than there are men (Zambia Census Report 2022 n.d.). Thus, any given sample is bound to have more females than men as a result of this skewed population structure.

It is interesting to note that of the participants tested, more were older than 15 years. This may mean that the older age group paid more attention to taking the MDA drugs implying that sensitization was adequate. The majority of tested participants were those with 1 and 2 rounds of MDA. This may mean that not all people have had all five rounds of MDA. Even if the MDA coverages in most of these districts were above 65% as per the WHO threshold for achieving elimination, not everyone participated in every MDA campaign. However, it was interesting to note that those that had 5 rounds of MDA were not found to have circulating antigens of *Wb.*

These results are encouraging and suggest that the LF MDA programme has contributed to reducing the prevalence of circulating microfilaria in the endemic districts. However, it may be difficult to attribute the reduced prevalence solely to the success of the LF MDA programme. This could be so because the other interventions that target mosquitos such as the distribution of insecticide-treated mosquito nets (ITNs) as was the case with the Rollback Malaria Programme could have greatly impacted our findings. This is for the simple reason that ITNs play a significant role (if used correctly and consistently) in reducing mosquito bites thereby reducing the transmission of the parasite as it is equally transmitted by mosquitoes (Berg, Kelly-Hope, and Lindsay 2013).

That said, it can be concluded that the LF MDA in Zambia has had a great impact as it has managed to reduce the prevalence of LF to almost zero and that LF MDA may no longer be necessary in 78 of the 80 LF endemic districts of Zambia. That Chibombo is the only district that requires LF MDA. It also suffices to state that this success has probably resulted from consented effort by all the stakeholders involved. It is therefore, tempting with the results of this pre-TAS from these 9 provinces to suggest that Zambia should move into conducting the TAS to ascertain if MDA should be stopped or not.

5. Conclusion

There is a significant decline in LF transmission in Zambia after five effective rounds of MDA. The country qualifies to conduct TAS 1 in the 78 districts of the 80 endemics districts.

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Informed Consent Statement: Written consent and accent were made available to all the participants with electronic signature on ODK enabled questionnaires.

Data Availability Statement: Data is available on request.

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References

1. Bah, Yakuba M. et al. 2020. "Achievements and Challenges of Lymphatic Filariasis Elimination in Sierra Leone." *PLoS neglected tropical diseases* 14(12): e0008877.
2. Berg, Henk van den, Louise A. Kelly-Hope, and Steve W. Lindsay. 2013. "Malaria and Lymphatic Filariasis: The Case for Integrated Vector Management." *The Lancet Infectious Diseases* 13(1): 89–94. https://www.thelancet.com/journals/laninf/article/PIIS1473-3099(12)70148-2/fulltext (April 7, 2023).
3. Burgert-Brucker, Clara R. et al. 2020. "Risk Factors Associated with Failing Pre-Transmission Assessment Surveys (Pre-TAS) in Lymphatic Filariasis Elimination Programs: Results of a Multi-Country Analysis." *PLoS neglected tropical diseases* 14(6): e0008301.
4. Chu, Brian K. et al. 2013. "Transmission Assessment Surveys (TAS) to Define Endpoints for Lymphatic Filariasis Mass Drug Administration: A Multicenter Evaluation." *PLoS neglected tropical diseases* 7(12): e2584.
5. Coulibaly, Yaya I. et al. 2016. "Dynamics of Antigenemia and Transmission Intensity of Wuchereria Bancrofti Following Cessation of Mass Drug Administration in a Formerly Highly Endemic Region of Mali." *Parasites & Vectors* 9(1): 628.
6. Deribe, Kebede et al. 2020. "African Regional Progress and Status of the Programme to Eliminate Lymphatic Filariasis: 2000-2020." *International Health* 13(Suppl 1): S22–27.
7. Dewi, Rita M. et al. 2015. "Brugia Rapid™ Antibody Responses in Communities of Indonesia in Relation to the Results of 'transmission Assessment Surveys' (TAS) for the Lymphatic Filariasis Elimination Program." *Parasites & Vectors* 8: 499.
8. Hira, P. R. 1976. "Bancroftian Filariasis. An Autochthonous Case in Zambia." *Medical Journal of Zambia* 10(6): 160–64.
9. ———. 1977. "Wuchereria Bancrofti: The Staining of the Microfilarial Sheath in Giemsa and Haematoxylin for Diagnosis." *Medical Journal of Zambia* 11(4): 93–96.
10. Manguin, S., M. J. Bangs, J. Pothikasikorn, and T. Chareonviriyaphap. 2010. "Review on Global Co-Transmission of Human Plasmodium Species and Wuchereria Bancrofti by Anopheles Mosquitoes." *Infection, Genetics and Evolution: Journal of Molecular Epidemiology and Evolutionary Genetics in Infectious Diseases* 10(2): 159–77.
11. Mwase, Enala T. et al. 2014. "Mapping the Geographical Distribution of Lymphatic Filariasis in Zambia." *PLoS neglected tropical diseases* 8(2): e2714.
12. Niles, Reza A. et al. 2021. "Assessing Factors Influencing Communities' Acceptability of Mass Drug Administration for the Elimination of Lymphatic Filariasis in Guyana." *PLoS neglected tropical diseases* 15(9): e0009596.
13. Nutman, Thomas B. 2013. "Insights into the Pathogenesis of Disease in Human Lymphatic Filariasis." *Lymphatic Research and Biology* 11(3): 144–48.
14. Ottesen, Eric A, and John Horton. 2020. "Setting the Stage for a Global Programme to Eliminate Lymphatic Filariasis: The First 125 Years (1875–2000)." *International Health* 13(Supplement_1): S3–9. https://academic.oup.com/inthealth/article/13/Supplement_1/S3/6043667 (April 11, 2023).
15. Penzer, Rebecca. 2003. "Lymphoedema." *Nursing Standard (Royal College of Nursing (Great Britain): 1987)* 17(35): 45–51; quiz 52, 55.
16. Pfarr, K. M., A. Y. Debrah, S. Specht, and A. Hoerauf. 2009. "Filariasis and Lymphoedema." *Parasite Immunology* 31(11): 664–72.
17. Ramaiah, K. D., and Eric A. Ottesen. 2014. "Progress and Impact of 13 Years of the Global Programme to Eliminate Lymphatic Filariasis on Reducing the Burden of Filarial Disease." *PLoS neglected tropical diseases* 8(11): e3319.
18. Ramzy, Reda M. R., Hussein A. Kamal, Mohamed A. Hassan, and Ayat A. Haggag. 2019. "Elimination of Lymphatic Filariasis as a Public Health Problem from the Arab Republic of Egypt." *Acta Tropica* 199: 105121.
19. Santoso, null et al. 2020. "Risk of Recrudescence of Lymphatic Filariasis after Post-MDA Surveillance in Brugia Malayi Endemic Belitung District, Indonesia." *The Korean Journal of Parasitology* 58(6): 627–34.
20. Shawa, Sheila Tamara, Enala T. Mwase, Erling M. Pedersen, and Paul E. Simonsen. 2013. "Lymphatic Filariasis in Luangwa District, South-East Zambia." *Parasites & Vectors* 6(1): 299.
21. Small, Scott T., Daniel J. Tisch, and Peter A. Zimmerman. 2014. "Molecular Epidemiology, Phylogeny and Evolution of the Filarial Nematode Wuchereria Bancrofti." *Infection, Genetics and Evolution: Journal of Molecular Epidemiology and Evolutionary Genetics in Infectious Diseases* 28: 33–43.
22. "Zambia Census Report 2022." *Bing*. https://www.bing.com/search?pglt=41&q=zambia+census+report+2022&cvid=bd801f301e1c4792a9d9cc2361aa5946&aqs=edge..69i57j0l7j69i60j69i11004.11120j0j1&FORM=ANNAB1&PC=EDGEDBB (April 10, 2023).
23. Zulfiqar, Hassam, and Ahmad Malik. 2023. "Bancroftian Filariasis." In *StatPearls*, Treasure Island (FL): StatPearls Publishing. http://www.ncbi.nlm.nih.gov/books/NBK547682/ (April 8, 2023).