HIGH SCHOOL EDITION
Journal of Student Research
Volume 11 Issue 3 (2022)

Onchocerciasis Transmission in Africa and How It Can Be Resolved

Saketh Nallagatla[1], Rajagopal Appavu[#] and Jothsna Kethar[#]

[1]Gifted Gabber, USA
[#]Advisor

ABSTRACT

Onchocerciasis is a parasitic infection that affects the skin and eyes and is transmitted by black flies which predominantly affect rural areas. Onchocerciasis symptoms are caused by microfilariae that travel through the skin and eyes to cause lesions, dermatitis, and blindness. These microfilariae are key in diagnosing Ivermectin by their presence in the skin and eyes which can be determined with skin snips or exposure to certain chemicals. Onchocerciasis in Africa is spread along river areas and efforts to wipe it out have failed because transmission zones have not been fully mapped out, Onchocerciasis is coendemic with Loiasis in certain areas, Onchocerciasis-causing parasites are developing Ivermectin resistance, there is a lack of coordination among countries with shared transmission zones, violent conflict diverts focus from the elimination of Onchocerciasis transmission, and these countries lack the funding to perform large-scale, accurate diagnosis procedures and deliver treatments on a more individual basis.

Background Information

Onchocerciasis, or River Blindness, is a neglected tropical disease caused by parasitic nematodes that commonly affect those in rural African villages with skin lesions and eye inflammation. Neglected tropical diseases are diseases that primarily affect rural regions without consistent access to the advanced medical resources to fight these diseases. Although Onchocerciasis is very commonly transmitted in its endemic zones, it is not very virulent and there exist many instances of countries or groups of countries eliminating the spread of Onchocerciasis within their borders through the use of various carefully selected, tailor-made methods, such as: Mexico and certain African countries. Although there is a great wealth of knowledge available about why attempts to inhibit transmission of this disease in certain endemic zones have failed, this body of knowledge does not explain what must be done on an individual level to facilitate the cessation of Onchocerciasis transmission in African endemic zones. For an individual to adequately comprehend what actions one may take in order to provide assistance and facilitate the elimination Onchocerciasis, one must first understand the methods by which Onchocerciasis is contracted, the methods by which Onchocerciasis contraction is diagnosed, and the methods by which Onchocerciasis is treated after it is diagnosed, why previous attempts to stop Onchocerciasis transmission have failed, and why previous attempts stop onchocerciasis transmission have succeeded.

Onchocerciasis spreads through the transmission of parasitic worms known as *Onchocerca Volvulus* through the bites of infected black flies and is effectively treated through the use of Ivermectin and Doxycycline. It is vital to understand the symptoms, transmission methods, and treatment methods of Onchocerciasis to formulate an effective strategy to eliminate the disease. Onchocerciasis is the second leading infectious cause of blindness in the world, after trachoma. In total, 37 million people are thought to have active disease, with nearly all such cases in Africa where over 100 million people live at risk of new infection. *Onchocerca volvulus* lives only in humans and is not known to perform vital ecological services, making it a good candidate for elimination. Onchocerciasis causes a great deal of the cases of blindness in the world and eliminating it would alleviate

many of these cases. Most of the active cases of Onchocerciasis and populations at risk to Onchocerciasis exist in Africa, so efforts to eliminate Onchocerciasis can be focused on one region.

Onchocerciasis occurs when a black fly infected with Onchocerca Volvulus bites a human and the parasite is transferred to the human host. These flies typically live near fast-flowing rivers and only individuals living around these rivers are at risk of Onchocerciasis due to the need to be bitten multiple times to show symptoms due to the poor transmission efficiency of each bite. When deposited into the host's skin, the larvae mature and generate hundreds of thousands of microfilariae that travel through the skin and have the potential to reach the eye. As the microfilariae travel through the skin, they cause skin atrophy and discoloration which creates a great social stigma around this disease due to the changes sufferers experience. When the microfilariae reach the eye, they travel through the eye and attempt to migrate through the cornea. In this attempt to migrate, the parasites die off and release Wolbachia bacteria, a species of bacteria which allows them to reproduce, which causes eye damage and inflammation as an autoimmune response. This inflammation scars the cornea and renders it opaque which prevents vision out of the afflicted eye.

Onchocerciasis has similar symptoms to a myriad of other diseases, and it is vital to properly diagnose Onchocerciasis infection before administering treatment. Onchocerciasis is characterized by the presence of visible microfilariae in the cornea, the presence of microfilariae in the skin, skin discoloration, and skin patch reactions with certain chemicals. By exposing patches of skin to diethylcarbamazine, the microfilariae in that patch is killed and provokes a feeling of hypersensitivity in the skin which is a clear sign of Onchocerciasis infection. Furthermore, there are tests that determine exposure to the parasite through monitoring antibodies, but these tests cannot differentiate between past and present infections and have given negative results when used to determine the status of the newly infected.

After diagnosis is performed successfully, there are a multitude of treatments that may be administered to treat Onchocerciasis. The most common treatment of Onchocerciasis is Ivermectin which only kills the microfilariae and not the actual parasites, but it is still effective because it prevents the parasites from reproducing and releasing more microfilariae and prevents further production of dermal and ocular lesions. A promising treatment is Doxycycline, an antibiotic that targets the Wolbachia bacteria that have a symbiotic relationship with Onchocerca Volvulus and enable the parasites to reproduce. This antibiotic permanently sterilizes the parasites and even decreases their lifespans and is commonly used alongside ivermectin. Although Doxycycline is a more effective treatment, it is impractical due to the length of time needed for repeated doses alongside the high possibility for reinfection when treating those living in areas where Onchocerciasis is endemic, such as rural villages near fast-flowing rivers. Other types of medication have shown promise, for example: Flubendazole is a benzimidazole that is traditionally used to treat gastrointestinal parasites, but tests conducted with injectable formulations of flubendazole show that it is highly effective in treating Onchocerca Volvulus, however, the injection sites frequently inflamed and the study was not continued.

Obstacles Preventing the Elimination of Onchocerciasis

Onchocerciasis has been targeted for elimination, but a number of socioeconomic and geographic features stop the disease from being truly wiped out despite the existence of a myriad of treatments. These factors are: transmission zones are not fully mapped out, Onchocerciasis is coendemic with Loiasis in certain areas, Onchocerca Volvulus is developing ivermectin resistance, lack of coordination in attempts to eliminate Onchocerciasis transmission, violent conflict in the region, and lack of funding.

Transmission zones that are considered high risk are well-mapped and have ivermectin treatment delivered forever. Transmission zones with lower infection rates have not been mapped and are thought to contribute to the prevalence of Onchocerciasis transmission. Multiple countries have developed their own mapping strategies and this lack of standardization hinders mapping attempts. Poor communication among multiple countries further contributes to the difficulty in sharing transmission zone data.

Onchocerciasis is coendemic with loiasis, a disease caused by the nematode Loa Loa. Ivermectin, the most effective treatment of Onchocerciasis, causes serious neurological harm to individuals with loiasis infections. Attempts to monitor areas of co endemicity to encourage ivermectin consumption have been unsuccessful due to widespread refusal to take ivermectin due to the fear of death from neurological harm. This issue makes the search for alternative methods of treatment and ways to separate loiasis from Onchocerciasis paramount to wiping out Onchocerciasis.

Another issue preventing the removal of Onchocerciasis is the development of Ivermectin resistance in several strains of the parasite. Ivermectin is the most common and effective treatment for Onchocerciasis due to its ability to kill Onchocerciasis-causing larvae and preventing them from doing more damage. Studies show that several genes responsible for the parasite's vulnerability to ivermectin have mutated which led to a great amount of variation of Onchocerciasis responses to ivermectin treatment. Ivermectin resistance is concerning because it is the best treatment for the original strain of Onchocerciasis and mutations may lead to the need for the development of new drugs.

One problem hindering efforts to eliminate Onchocerciasis is the poor coordination in large-scale attempts to eliminate Onchocerciasis. Multiple endemic countries have shared transmission zones because the rivers on their borders are breeding grounds for the parasites that cause Onchocerciasis. These areas increase Onchocerciasis transmission due to the fact that some countries on these borders may be successful but the countries on the other side of the borders may not fully eliminate Onchocerciasis in that area which can reintroduce Onchocerciasis into the previously cleaned areas. Black flies, the most common carrier of Onchocerciasis-causing parasites, have seasonal migration patterns that cause them to travel across borders and reintroduce Onchocerciasis to areas that previously eliminated the affliction.

The poor coordination in cross-border attempts to eliminate Onchocerciasis is further worsened by violent conflict in certain countries where Onchocerciasis is endemic. In countries with violent conflict, their healthcare system is not centralized and standardized to the degree required to perform large-scale elimination of Onchocerciasis. The required levels of epidemiological surveying and infrastructure are not in place in countries shattered by violent conflict.

Even countries with excellent plans to eliminate Onchocerciasis transmission can still fail to do so due to poor program implementation. Some examples of how these programs could be implemented poorly are: poor geographic drug distribution, frequency of ivermectin delivery, and supply mismanagement. Ivermectin deliveries have not been sufficiently distributed to reach levels required to minimize the prevalence of Onchocerciasis. The shortage of ivermectin and the lack of infrastructure needed to transport ivermectin to rural areas surrounded by natural obstacles that impede delivery of Ivermectin greatly hinder attempts to stop the transmission of Onchocerciasis in a permanent manner. Many individuals refuse to take ivermectin due to the aforementioned fear of neurological damage due to loiasis and this means the minimization of Onchocerciasis cannot be achieved in these endemic zones even if the required amount of ivermectin is delivered in a timely manner.

How Obstacles Preventing the Elimination of Onchocerciasis Can Be OverCome

All of these issues can be solved and have been solved in other areas where Onchocerciasis was previously endemic such as Mexico. Onchocerciasis in Mexico was endemic in three areas: Southern Chiapas, Northern Chiapas, and Oaxaca. Southern Chiapas was the most prevalent due to its great size and history of Onchocerciasis transmission. Mexico managed to stop Onchocerciasis through mass distributions of ivermectin. Mexico does not have the same challenges as Africa such as Loiasis coendemicity, violent conflict, lack of infrastructure in rural areas, and poor mapping of transmission zones; however, there are still issues that Mexico overcame

which are relevant to Onchocerciasis in Africa. Mexico overcame the resistance to ivermectin that Onchocerciasis developed by using doxycycline to kill the adult parasites whereas ivermectin only killed larvae and prevented further damage.

Doxycycline is an antibiotic that targets Wolbachia bacteria, a type of bacteria necessary for Onchocerciasis-causing parasites to survive. Studies show that Wolbachia is vital to the survival and fertility of Onchocerciasis-causing parasites due to their role in pathogenesis inducement. Doxycycline renders these parasites sterile for extended periods of time and allows them to die when they reach the end of their natural lifespan. This drug can overcome Onchocerciasis transmission even in areas where it is coendemic with Loiasis because it does not trigger the same neurological damage that Ivermectin does when exposed to Loiasis. Unfortunately, Loiasis parasites do not rely on Wolbachia to reproduce and Doxycycline is not an effective treatment for Loiasis.

The delivery of treatments like Ivermectin and Doxycycline are aimed at high-intensity transmission zones which have been thoroughly mapped, but less intense transmission zones are mapped with substandard accuracy and are not standardized across multiple organizations for ease of access. This issue is vital to the elimination of Onchocerciasis in Africa because even if high-intensity transmission zones are purged of Onchocerciasis, Onchocerciasis could still be reintroduced from one of the lower intensity transmission zones. This issue must be solved by standardizing mapping systems across a wide array of countries and healthcare organizations that deliver ivermectin and monitor Onchocerciasis transmission; furthermore, more resources must be allocated to the ivermectin delivery and the mapping of less intense transmission areas. This issue did not apply to Mexico because of their central infectious disease organization known as the Centro Nacional de Programas Preventivos y Control de Enfermedades; this organization used a standardized mapping system that allowed many members of the organization to rapidly understand the severity and distribution of Onchocerciasis transmission in newly mapped areas without the need to convert multiple mapping systems.

This lack of standardization is part of why there is poor coordination between countries with shared transmission zones. Governmental organizations responsible for eliminating Onchocerciasis transmission in shared transmission zones need to communicate with other organizations responsible for the areas of shared transmission zones outside of their jurisdiction in order to simultaneously eliminate the shared transmission zones at the same time to avoid reintroduction of Onchocerciasis through the part of the transmission zone that Onchocerciasis remained in. One reason African countries face these coordination issues is due to the prevalence of civil war and violent conflict in the region; this violent conflict ignores political pressure to end Onchocerciasis transmission and fragments healthcare systems. Mexico did not need to deal with violent conflict on the same scale as Africa as a whole and unfortunately cannot be used as a model to create a solution to this problem.

One method to solve this issue of political focus being diverted to the resolution of violent conflict rather than the elimination of Onchocerciasis transmission is the widespread education of people in rural areas about the dangers of Onchocerciasis which would generate enough public desire to eliminate Onchocerciasis in Africa. This can be accomplished in a multitude of ways, many of which have been proven to work, such as: Sending out teams of healthcare workers to teach isolated communities, distributing medical literature about health crises in the area, and hosting gatherings to teach people about the dangers of infectious diseases en masse. These methods would have the benefit of encouraging politicians to divert resources to the elimination of Onchocerciasis while also allowing individuals to protect themselves from the disease by taking precautions against it.

Alongside examples of successful elimination of Onchocerciasis transmission, there exist a variety of strategies to deliver treatment while reducing transmission that are tailor made for certain areas. One such strategy is an increase in Ivermectin delivery frequency which has the added benefits of greater reduction in microfilariae reproduction, greater reduction in microfilariae lifespan, and greater reduction in the prevalence of skin microfilariae. This strategy would reduce the likelihood of transmission through human vectors (skin-to-skin

contact) while also reducing the amount of time available for the parasites to create more skin and eye lesions. This strategy requires much more ivermectin than the current amount used for Onchocerciasis reduction programs currently. This lack of ivermectin would have to be remedied by investing more money into drug production while also creating initiatives for more developed countries with superior medical infrastructure to supply these African countries with ivermectin.

This strategy can be made more viable in these conditions through the use of more efficient distribution timing and the increased production of alternative Onchocerciasis treatments such as doxycycline and flubendazole. Ivermectin distribution most commonly occurs during the dry season, but this practice does not make the most efficient use of ivermectin because most instances of Onchocerciasis transmission occur during the wet seasons when the flies that carry Onchocerciasis-causing parasites breed. The production and testing of these drugs on a larger scale would make it much more feasible to eliminate Onchocerciasis transmission. Although doxycycline does not target the parasites directly, it will still eliminate them albeit without reducing the risk of transmission by killing skin microfilariae like ivermectin does. On the other hand, flubendazole is not well tested and previous tests with humans have caused swelling in the injection site and it cannot be used without further testing or modification despite its promising nature.

Another strategy that may be used to positively influence the success of Onchocerciasis elimination in Africa is a TNT strategy. TNT strategies are strategies that require diagnosis for infection before treatment regimens are chosen on an individual basis. This strategy would circumvent the adverse neurological damage caused by ivermectin when treating Onchocerciasis alongside Loiasis by first generating a superior understanding of each case of Onchocerciasis on an individual basis which would allow for administration of doxycycline or other alternative treatments that do not facilitate adverse neurological effects when exposed to Loiasis. This strategy has the indirect benefit of increasing the likelihood that locals take the treatments instead of wasting them out of distrust due to the mistaken belief that ivermectin causes adverse neurological effects when used in all circumstances rather than when it is used in the presence of Loiasis.

TNT strategies seem highly effective, but they require frequent, widespread, and thorough diagnosis procedures to be carried out. These diagnosis procedures can be performed in a variety of ways such as blood smears, examinations for the presence of ocular microfilariae, and examinations for the presence of dermal microfilariae. These methods of diagnosis are quite accurate but are not efficient when performed on the large scale required for a TNT strategy. The most efficient method of Onchocerciasis diagnosis on a large scale is the identification of the presence of Onchocerca Volvulus microfilariae in skin snips. Although the examination of skin snips is easily carried out on a wide scale, it requires trained personnel and is invasive.

After skin snip examinations are performed, the next step of a TNT strategy is to administer treatments on an individual basis. Ivermectin is delivered to those without Loiasis coinfection due to its immense efficacy in eliminating Onchocerciasis transmission by previously mentioned means. Doxycycline is administered to those with Loiasis coinfection because it is an antibiotic medication that is capable of killing the Wolbachia bacteria used by Onchocerca Volvulus to reproduce; this indirect method of curtailing transmission does not trigger the same adverse neurological effects caused by Loiasis exposure to Ivermectin. TNT strategies seem highly effective and would be excellent for the end of Onchocerciasis transmission in Africa due to their solution to Loiasis coendemicity, their ability to incorporate multiple types of drugs, their ability to benefit greatly from advances in diagnostic technology, and the greater increase in public trust garnered in comparison to the more traditional method of mass delivery of ivermectin administered by local communities rather than healthcare workers.

Figures and Supporting Evidence

Authors	Year published	Country	Sample size	Setting	Outcomes
Turner *et al.* [46]	2010	Cameroon	104 subjects	Doxycycline 200mg/day for 6 weeks alone, *vs.* Doxycycline 200mg/day for 6 weeks + Ivermectin 150 µg/kg (4 months after Doxycyclin); *vs.* Ivermectin alone	1 year of follow up: Doxycycline + ivermectin had lower skin microfiliaria prevalence (23.9%) than doxycycline (61.9%) and ivermectin only (78.4%), *P*<0.005. 21 months of follow-up: skin microfiliaria prevalence was smaller in the Doxycycline + ivermectin (10.9%) and doxycycline alone (33.3%) than the ivermectin group (78.4%), *P*<0.05.
Hoerauf *et al.* [47]	2009	Ghana	30 subjects	Doxycycline 100mg/day for 5 or 6 weeks *vs.* control (untreated)	(A) Worms: Doxycycline group had more dead female (49% *vs.* 16%, *P*<0.0001) and male (27% *vs.* 5%, *P*<0.034) worms than untreated group. (B) *Wolbachia*: Doxycycline group had 24% female and 38% male worms containing *Wolbachia*, while the untreated group had 98% and 86%, respectively (*P*<0.0001). * The authors considered that worms containing several *Wolbachia* acquired the bacteria after Doxycycline treatment.
Specht *et al.* [48]	2009	Ghana	72 subjects	Doxycycline 100–200mg/day for 4, 5 or 6 weeks *vs.* control (untreated), followed by ivermectin	Only worms acquired after Doxycycline treatment showed microfiliaria production; only newly acquired *O. volvulus* were classified as having several live *Wolbachia*
Hoerauf *et al.* [50]	2008	Ghana	67 subjects	Doxycycline 200mg/day for 4 or 6 weeks (*vs.* placebo only), followed by Ivermectin (150 µg/kg) 6 months later	Skin nodules: (A) Quantitative PCR: Reduction on *Wolbachia* load in worm tissue in both Doxycycline groups compared to placebo (*P*<0.05). (B) Immunology: At both treatment groups, higher proportion of dead female worms compared to the placebo group (*P*<0.05)
Specht *et al.* [49]	2008	Ghana	26 subjects	Three groups: Rifampicin for 2 weeks, Rifampicin for 4 weeks, and control (untreated)	Reduction of *Wolbachia*-colonized female *O. volvulus* after 2 weeks (66%) and 4 weeks of Rifampicin (21%) compared to control (92%, *P*<0.0002)
Richards *et al.* [30,51]	2007	Guatemala	73 subjects	Four groups: rifampin, azithromycin, rifampin + azithromycin, and control (vitamins). Drugs taken for 5 days, followed by Ivermectin (single dose). Patients evaluated 9 months later	No significant differences in the percentage of live females with *Wolbachia* and live females reproductively active (*P*>0.05)
Hoerauf *et al.* [52]	2008	Ghana	40 patients	Azithromycin for 6 weeks (250mg daily, or 1200mg once a week), nodules examined after 6 and 12 months	After 1 year, the group that received 250mg daily presented a reduction of the number of female worms containing elevated number of *Wolbachia* compared to untreated patients (65% *vs.* 92%, *P*<0.0001)
Debrah *et al.* [53]	2006	Ghana	60 subjects	Doxycycline 200mg/day for 2, 4 or 6 weeks; control (untreated). Some subjects received Ivermectin 150 µg/kg 8 months after first Doxycycline dose	Compared to controls, reduction in skin microfiliaria load seen in the 4-week group (*P* = 0.0039) after 1.5 years of first dose.
Hoerauf *et al.* [54]	2003	Ghana	99 subjects	Doxycycline 100mg/day for 6 weeks or control (no treated), followed by ivermectin 2 or 6 months later	Reduction of live female worms with numerous *Wolbachia* compared to the control group (88% *vs.* 0–2%, *P*<0.0008); reduction of number of female worms with microfilariae production compared to controls (56% *vs.* 1–4%, *P*<0.0001); reduction of number of male worms containing sperm after 11 months of first dose of doxycycline, compared to control (89% vs. 63%, *P*<0.002) and number of female worms containing sperm in their uterus (56% *vs.* 15%, *P*<0.002) after 11 months of first dose of doxycycline.
Hoerauf *et al.* [55]	2001	Ghana	88 subjects	Doxycycline 100mg/day for 6 weeks; Ivermectin 150 µg/kg	Group that received ivermectin + doxycycline had a smaller microfilaridermia than ivermectin only, after 1 year of initial treatment (0.22 *vs.* 1.89 microfiliaria/mg skin, *P*<0.0001)
Hoerauf *et al.* [56]	2000	Ghana	35 subjects	Doxycycline 100mg/day for 6 weeks *vs.* control (untreated)	Skin nodules: Treatment group had fewer female worms (*P*<0.03), fewer live females containing *Wolbachia* (*P*<0.0001), fewer live females with intact embryogenesis (*P*<0.0001) than control group and fewer nodules with live microfilariae (*P*<0.0001)

Figure 1. The effectiveness of Doxycycline as a treatment of Onchocerciasis due to its ability to kill Wolbachia bacteria.

All the cases of Onchocerciasis treated with Doxycycline showed a visible decline in the prevalence of dermal and ocular microfilariae. This level of efficacy combined with Doxycycline's ability to stop Onchocerciasis transmission even in the presence of Loiasis makes it an extremely useful tool in overcoming Onchocerciasis in areas where it is coendemic with Loiasis. African governments can overcome the obstacle of coendemicity by allocating more resources to the distribution and creation of this drug. Unfortunately, overuse can allow Wolbachia bacteria to develop resistance to the antibiotic which means Doxycycline should only be used in cases of coinfections.

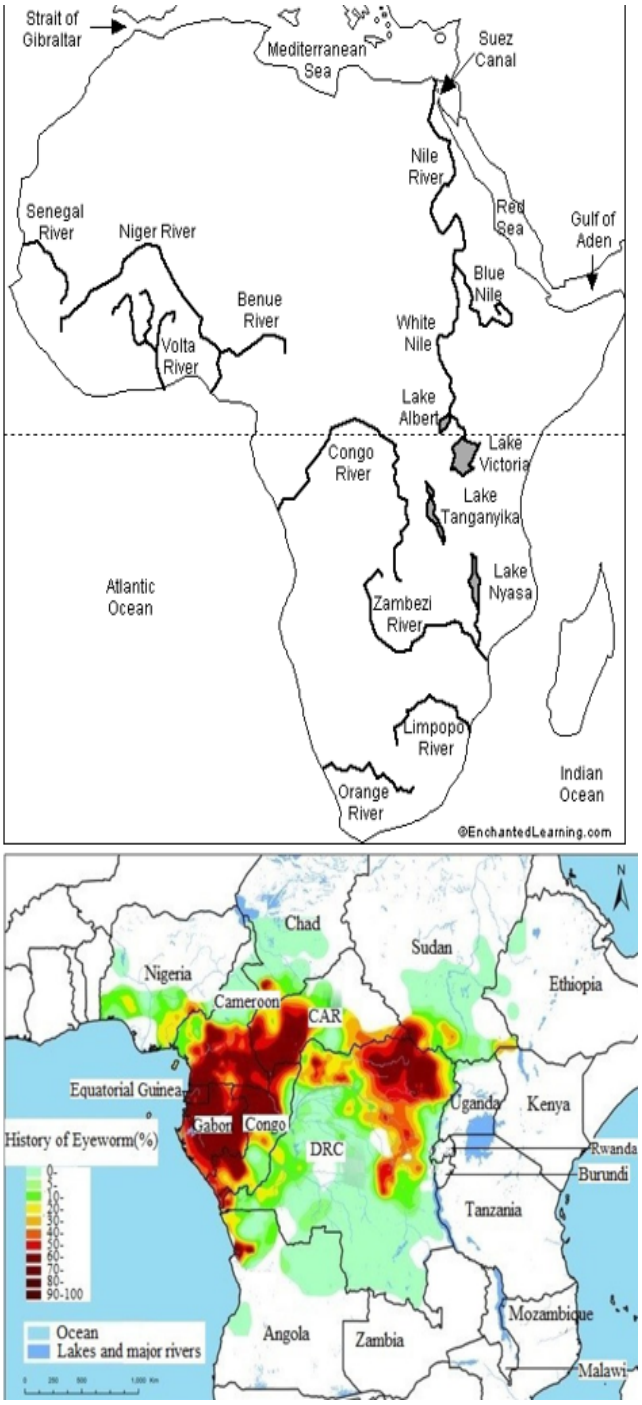

Figure 2. Prevalence of Onchocerciasis in Africa

The maps above depict the distribution of rivers alongside the intensity of Onchocerciasis in certain regions of Africa. It is plainly visible that Onchocerciasis is most prevalent alongside rivers and many of these rivers lie on the borders of countries which further exacerbates the difficulty of eliminating Onchocerciasis. These maps illustrate the necessity of the cooperation of countries with shared transmission zones. If one transmission zone is not fully cleansed of Onchocerciasis, it will quickly revert to its previous state unless all countries in that transmission zone put in the work to eliminate Onchocerciasis.

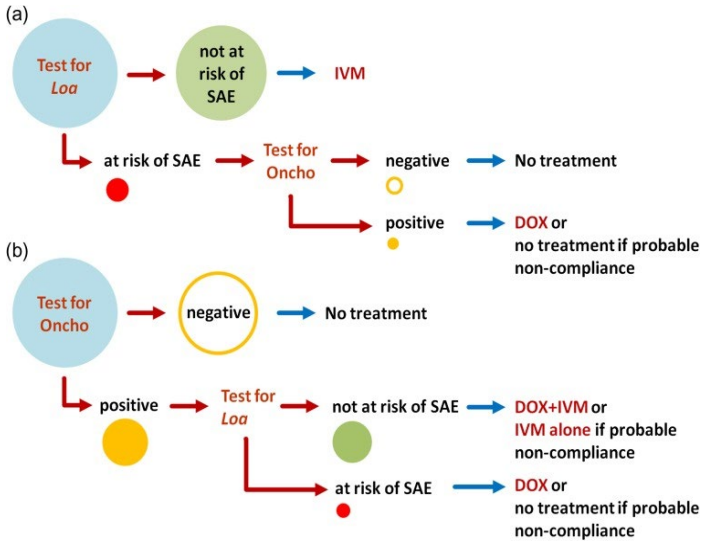

Figure 3. Treatment flowchart for Onchocerciasis coinfections with Loiasis

Loiasis is a great obstacle in the elimination of Onchoerciasis because Ivermectin, the most common treatment for Onchocerciasis, causes great neurological damage when used on someone with a Loiasis infection. It is necessary to spend resources and manpower on testing each patient for both Loiasis and Onchocerciasis to deliver the most efficient treatment possible. This highlights the need for less demanding methods of diagnosis that can be performed on a wide scale for both accurate and quick treatment decisions.

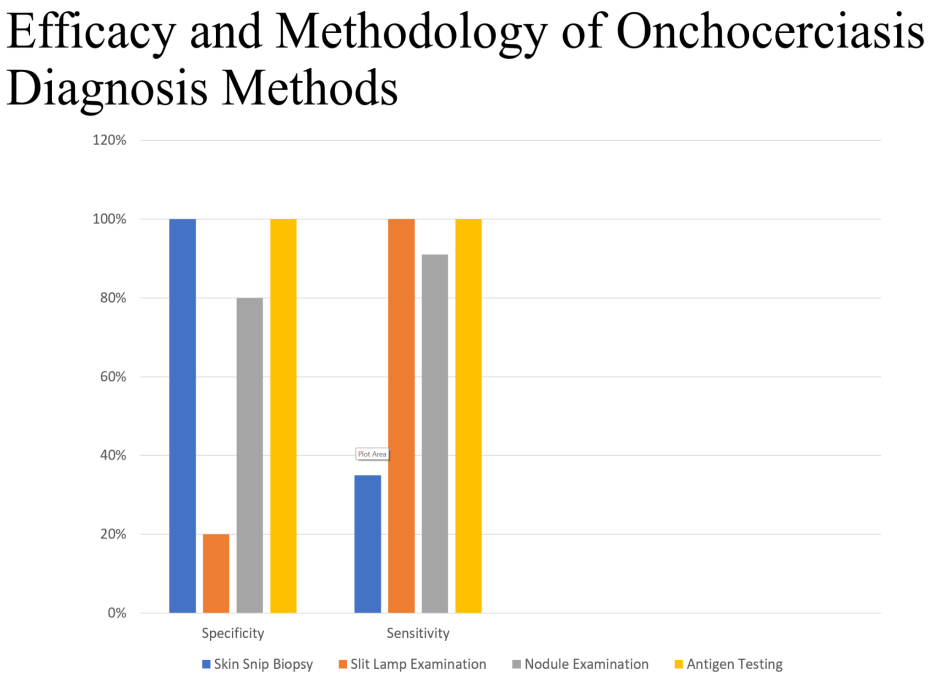

Figure 4. Individual Contribution

Conclusion

African Onchocerciasis transmission is a large problem with many obstacles blocking the path to its elimination but it is not impossible to solve. As demonstrated by countries like Mexico, it is possible to eliminate Onchocerciasis and it is only a matter of proper resource allocation, planning, and coordination. Many obstacles hindering the progress of Onchocerciasis elimination have already been overcome in other countries and African countries need to follow their examples while further improving their healthcare systems by standardizing mapping techniques and creating more infrastructure that can be used to expedite the delivery of treatments to the rural areas where Onchocerciasis is most common.

Acknowledgments

I would like to thank my advisor for the valuable insight provided to me on this topic.

References

Boussinesq, M., Fobi, G., & Kuesel, A. C. (2018, March 1). *Alternative treatment strategies to accelerate the elimination of Onchocerciasis*. International health. Retrieved January 16, 2022, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5881258/

Centers for Disease Control and Prevention (CDC). (2013, May 24). *Progress toward elimination of onchocerciasis in the Americas - 1993-2012*. MMWR. Morbidity and mortality weekly report. Retrieved January 16, 2022, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4604938/

Gebrezgabiher, G., Mekonnen, Z., Yewhalaw, D., & Hailu, A. (2019, July 4). *Reaching the last mile: Main challenges relating to and recommendations to accelerate onchocerciasis elimination in Africa*. Infectious diseases of poverty. Retrieved January 16, 2022, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6609392/

Mackenzie, C., Kollmann, M., Specht, S., & Sodhalon, Y. (2018). *River blindness: Reducing the prevalence of clinical disease*. Community eye health. Retrieved January 16, 2022, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5998396/

Rodríguez-Pérez, M. A., Fernández-Santos, N. A., Orozco-Algarra, M. E., Rodríguez-Atanacio, J. A., Domínguez-Vázquez, A., Rodríguez-Morales, K. B., Real-Najarro, O., Prado-Velasco, F. G., Cupp, E. W., Richards, F. O., Hassan, H. K., González-Roldán, J. F., Kuri-Morales, P. A., & Unnasch, T. R. (2015, July 10). *Elimination of onchocerciasis from Mexico*. PLoS neglected tropical diseases. Retrieved January 16, 2022, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4498594/

Tekle, A. H., Zouré, H. G. M., Noma, M., Boussinesq, M., Coffeng, L. E., Stolk, W. A., & Remme, J. H. F. (2016, June 27). *Progress towards onchocerciasis elimination in the participating countries of the African programme for Onchocerciasis Control: Epidemiological Evaluation Results*. Infectious diseases of poverty. Retrieved January 16, 2022, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4924267/

Winthrop, K. L., Furtado, J. M., Silva, J. C., Resnikoff, S., & Lansingh, V. C. (2011, April). *River blindness: An old disease on the brink of elimination and control*. Journal of global infectious diseases. Retrieved January 16, 2022, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3125028/

Darvin Scott Smith, M. D. (2021, July 19). *Onchocerciasis (river blindness) workup: Laboratory Studies, Imaging Studies, other tests*. Onchocerciasis (River Blindness) Workup: Laboratory Studies, Imaging Studies, Other Tests. Retrieved January 22, 2022, from https://emedicine.medscape.com/article/224309-workup#:~:text=Traditionally%2C%20a%20diagnosis%20of%20onchocerciasis,in%20early%20stages%20of%20infection.

Taylor, H. R., Munoz, B., Keyvan-Larijani, E., & Greene, B. M. (1989, October 1). *Reliability of detection of microfilariae in skin snips in the diagnosis of onchocerciasis*. AJTMH. Retrieved January 22, 2022, from https://www.ajtmh.org/view/journals/tpmd/41/4/article-p467.xml
Udall, D. N. (2007, January 1). *Recent updates on Onchocerciasis: Diagnosis and treatment*. OUP Academic. Retrieved January 22, 2022, from https://academic.oup.com/cid/article/44/1/53/433399?login=true