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EFFECTIVENESS OF UV-DISINFECTION AGAINST CERCARIA AT SELECTED WAVELEGNTH USING UV-LEDs (FOR SCHISTOSOMIASIS CONTROL)

BY

EDEN ERITREA

Thesis submitted to Addis Ababa University's African Centre of Excellence for Water Management (ACEWM) in partial fulfillment of the Master's Degree in Water Management with a Specialization in Water Quality Management

JUNE, 2021

ADDIS ABABA

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Thesis Approval Form

APPROVED BY BOARD OF EXAMINERS

This is to certify that we the undersigned, have examined this MSc thesis entitled **"EFFECTIVENESS OF UV-DISINFECTION AGAINST CERCARIA AT SELECTED WAVELEGNTH USING UV-LEDs FOR SCHISTOSOMIASIS CONTROL"** and that in our opinion; it is fully adequate, in scope and quality, as a MSc thesis for the degree of Master of Science in Water Management (Water Quality Management)

Advisor: Dr. Feleke Zewge

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Declaration

I, **Eden Eritrea**, declare that this research report is my unique effort and work, and that the findings have never been previously presented to Addis Ababa University or anywhere for the award of any academic distinction. Where help was requested, it was given. The findings, interpretations, and conclusions presented in this study do not reflect the opinions of the Addis Ababa, African Centre of Excellence for Water Management (ACEWM) or the MSc Examination Committee as a whole.

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Abstract

Due to a lack of water and sanitation, schistosomiasis is a neglected tropical water-based chronic illness that kills. It is spread to people through contact with cercariae-infected water on the skin while bathing, swimming, or in any other way. In earlier research, various studies, including UV irradiation, have been conducted to eradicate the diseases-causing pathogenic parasite known as cerceriae. However no studies were carried out using the newly emerging UV-technology such as UV-LEDs. Hence the objective of this study was to investigate the effectiveness of UVC-LEDs (with a wavelength of 255nm, 265nm and 285nm) to disinfect Schsitosoma cercariae. Snails which an intermediate host for Schsitosoma cercariae were collected from Hawassa (Tikur Wiha River) and also from Lake Ziway. The collected snails were prepared for shading cercariae and 100-120 cercariea were prepare in 200µL. The disinfection of cercarea was examined using UVC-LEDs at a wavelength of 255 nm, 265 nm and 285 nm respectively. Using fluence (UV-dose) of 1 mJ/ cm^2, 20 mJ/ cm^2 ,40 mJ/ cm^2, 60 mJ/ cm^2, 80 mJ/ cm^2, 100 mJ/ cm^2, 120 mJ/ cm^2,160 mJ/ cm^2, 200 mJ/ cm^2 and 300 mJ/ cm^2. Under controlled settings, the experiment was repeated three times in a solution containing 200µL of cercariae-infested water and 3.6 ml of bottled water.All fluence (UV-dose) showed no significance cercariae disinfection at a wavelength of 255 nm. But Wavelengths 265 nm and 285 nm showed significance cercariae disinfection. 1 log reduction was achieved at a fluence of 300 mJ/ cm^2 and wavelength of 265nm and 1.5 and 2 log reduction was achieved at fluence of 200 mJ/ cm^2 and 300 mJ/ cm^2 at wavelength of 285nm. It took 300 mJ/ cm^2 fluence (UV-dose) to achieve 2 log reduction which is high compared to fluence used in different studies to disinfect other pathogenic parasite. Therefore, in this study UVC-LEDs used is a promising technology to disinfect schsitosoma cercariae but requires further studies. Therefore, more research and different methodology is needed to enhance its use and effectiveness.

Key words**:** *Schistosomiasis, cercariae, fluence (UV-dose), disinfection, UV-LEDs*

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Acronyms

µl	Micro litter
DBPs	Disinfection by-products
DNA	Deoxyribonucleic acid
EPHI	Ethiopian Public Health Institute
LEDs	light-emitting diodes
mJ/cm^2 ($mW\ cm^{-2}$)	Milli joules per centimeter square
nm	Nanometer
PC	Preventative chemotherapy
PPE	Personal protective equipment
RNA	Ribonucleic acid
UV	Ultraviolet
THM	Trihalomethane

1. INTRODUCTION

1.1. Background

Water is a key natural resource that sustains life and the environment. It is essentially necessary for human growth and wellness (Kumar et al., 2019). Though its vital, the quality of water for a certain function can be influenced by disease-causing organisms and chemical compounds originating from both natural and manmade sources (Akafu et al., 2019). The existing population and movement patterns, as well as rural-to-urban migration, forced movement, worker migration, and the expansion of ecotourism, having all made a influence to the proliferation of water-related disease. In particular, in most developing nations, such as Ethiopia, there is an increasing public health risk that is directly or indirectly related to water contamination (Botelho, 2018).

Water-related illnesses are classified into four kinds depending on the disease's incidence and the microorganism responsible. The first is a waterborne disease, which spreads pathogens through fecal contamination of drinking water. Among them are typhoid, giardiasis, Cryptosporidium, and cholera. Water-washed disease, on the other hand, are infections that proliferate in areas where there is inadequate water for personal cleanliness, Scabies, Trachoma, and Shigella are among them. Water-related disease, which are vector-borne diseases that require the insect vector to have access to water, are the third type. Mosquitos transmit diseases such as malaria, onchocerciasis, and trypanosomiasis. Water-based diseases are the fourth type, in which the pathogenic organism spends part of its life cycle in water. Schistosomiasis infections are the greatest example of these types. It is a disease in which the causative organism cercariae must spend part of its life cycle in water and the rest of its life cycle outside of water. And it is prevalent in areas where contact with a source containing the pathogenic organism is intense and localized (Tigist, 2020).

Schistosomiasis, commonly known as bilharzias, is a parasitic infection triggered by flatworms (flukes) of the genus Schistosoma, which can cause acute and chronic disorders and is a major public health concern in many parts of the world. The term schistosomiasis is derived from two Greek words: schistos, which means split, and soma, which means body. The name was given

based on the morphology of the male worm and it was named by David Friedrich in 1858 (Di Bella et al., 2018)

The five schistosome species that cause disease are Schistosoma haematobium (S. haematobium), Schistosoma mansoni, Schistosoma japonicum, Schistosoma mekongi, and Schistosoma intercalatum. The most prominent trematodes from these species have been identified as S. haematobium, S. mansoni, and S. japonicum. *S. haematobium* and *S. mansoni* are common species found in Africa. Schistosomiasis is acquired though skin contact with cercariae-infested fresh water and when the cercariae burrows through the host skin to develop into schistosome worms. This disease is caused by a snail that has been infected with miracidia, which grew from eggs released by the urine or feces of an affected person. *Cercariae* are released from the snail after the miracidia went through two stages of development inside the snail.(Braun et al., 2020).

In many regions of the world, schistosomiasis infection has become a major public health concern. The infection begins in childhood and lasts until maturity, even when the infection has been terminated. More than 230 million people are believed to be infected with schistosomes, with an equal number of people suffering from chronic sickness as a result of their infection. As a result, the number of people suffering from infection-related schistosomiasis may be closer to 440 million (Colley et al., 2014).

Because schistosomiasis is becoming a global concern, numerous methods of control, including water storage, filtration, ultraviolet disinfection (UV), and chlorination, could be used. Chlorination (chlorine disinfection) has been shown to be an effective water treatment technology in a variety of settings, ranging from small-scale emergency situations employing chlorination tablets to large-scale municipal water treatment plants. It has led in a remarkable reduction in water-borne disease since its introduction into public water systems in the 1900s.(Braun et al., 2020).

Chlorine and its numerous compounds are potent oxidants that will kill or inactivate most pathogenic organisms harmful to human and animal life because the chemicals utilized are widely

available, inexpensive, effective, and simple to administer. In wastewater treatment, the chlorination procedure is the most often used method (Plant & Training, 2016).

Recently, UV radiation has gain more attention than conventional chemical disinfection such as chlorination. This is because UV disinfection doesn't need chemical addition, it doesn't have harmful disinfection by-product (DBPs) formation such as THM. Consequently, UV radiation is becoming attractive for inactivation of various microorganisms in water and it is being used for water disinfection (Song et al., 2016). Microorganisms are inactivated by UV-radiation in the UV wavelength range in a process of UV disinfection. The UV-C range (200-300 nm) is most effective for inactivating pathogens and is often known as the germicidal rang. The microorganisms are inactivated when UV is absorbed by their DNA and RNA and also it damages the cell membrane which leads to prevention of reproduction. UV fluence (mJ/cm^2), which is a product of the fluence rate and exposure time is used to determine the sensitivity of microorganisms to UV. There are different UV radiation source including sunlight the greatest source of UV radiation in the UV-A and UV-B range. Recently, UV light-emitting diodes (LEDs) is an emerging technology, which is considered to be environmentally friendly and efficient in disinfection (Braun et al., 2018).

In comparison to the efficiency of UV light in inactivating bacterial and viral pathogens, few study has been done on the effectiveness of UV light in inactivating helminth infectious illness, which are prevalent in many undeveloped countries. To prevent and eliminate helminth infectious illnesses like schistosomiasis, investigations employing UV light sources such as UV-C LED are required. Although UV light has been proven to be successful in inactivating Schistosoma spp., there is no standard procedure for measuring UV fluence from UV-C LEDs, necessitating additional research therefor researchers should make use of these devices and give a detailed methods section that specifies the device type and protocol used (Hazell et al., 2019b).

1.2. Problem Statement

Schistosomiasis is the most common parasite infection in Africa, the Middle East, Central and South America, the Caribbean, and the Far East. Due to the high number of patients and severity of the condition, schistosomiasis has a negative impact on a country's economic productivity (Kassa et al., 2005).

In terms of its public health and socioeconomic impact, Schistosomiasis is among the most neglected chronic tropical diseases next to malaria and is still the major helminth infection in many developing country at the beginning of the 21st century. The disease is endemic in more than 74 countries, of which 46 are found in Africa indicating that schistosomiasis is much more intense in Africa. The Sub-Saharan region of Africa is considered to be the major endemic area for the disease. Around 80-85% of schistosomiasis are found in these part of Africa where *S.haematobium, S.interalatum* and *S.mansoni* are endemic (Amisalu, 2010).

In Ethiopia, both rural and urban areas, schistosomiasis is considered endemic. The first schistosomiasis species, S.mansoni, was discovered in 1934, while S.haematobium was discovered much later in a few isolated areas. In the Omo, Awash, and Blue Nile River Valleys, S.mansoni, the fluke that causes the intestinal variant of the disease, is found (Isabel & Austin, 2016). According to Negussu et al., (2017), in Ethiopia there are an estimated 37.3 million people living in schistosomiasis endemic area and in 2013 there was 35,775,100 cases of schistosomiasis occurrence. Most transmission site of *S.mansoni* infection are in agricultural communities along streams between 1300 and 2000 m latitude. *S.mansoni* was also reported from these all administrative regions. Irrigation systems, lakes and small streams are the main site suitable for the intermediate host snail. Therefore the infection with *S.mansoni* is more chronic on people who live in a developing country that are greatly dependent on agricultural and irrigation activities alongside such streams and water bodies which are infested with the infections disease causing *S.mansoni* (Aemero, 2011).

Storage, UV light, heating, filtration, and chlorination can be used to treat cercariae infested water. (Braun et al., 2018). From these mechanisms UV disinfection is gaining increased interest since it is based on electromagnetic energy and in general it does not alter water quality. It has also been an attractive alternative to chemical methods since it avoids the by-product formed during chlorination. (Artichowicz et al., 2020). UV disinfection using UV LEDs is a promising technology for water and wastewater treatment and UV LEDs emitting UV-C irradiation have proven effective in inactivating different pathogenic microorganisms in water. UV-C LEDs has vast potential since they are smaller, lighter and less fragile than traditional mercury vapor lamps and also they consume less energy, provide easy access when it comes to turning on and off and are mercury free (S. E. Beck et al., 2017). Though these recent UV disinfection with UV-C LEDs technology could be realistic option for sustainable removal of cercariae in low-incoming regions, there is currently no detailed methods and standard fluence measurement protocol for determining UV fluence form UV-C LEDs to inactivate cercariae. Therefore, conducting this research is necessary to fill these gaps by providing information about UV fluence needed to remove cercariae using UV-C LEDs by using other developed standard fluence measurement protocol. And also optimization of UV disinfection is needed to set reliable design, implantation strategies and UV disinfection guidelines to inactivate cercariae and to control schistosomiasis.

1.3. Research Objective

1.3.1. Main objective

The main objective of the study is to examine the effectiveness of UV disinfection (UV-C LEDs) against cercariae for schistosomiasis control at selected wavelengths.

1.3.2. Specific objectives

The study is envisioned to pursue the following Specific Objectives:

1. To examine the effectiveness of UV-disinfection against schistosome *cercariae* at three selected wavelength using UV-C LEDs and determine the wavelength and exposure time required to inactivate schistosome cercariae using UV-C LEDs
2. To compute the UV-fluence, which is the product of the fluence rate and exposure time, required to inactivate schistsome cercariae.

1.4. Research Question

In pursuing the above objectives, the study will strive to answer the following questions:

1. Which wavelength form the UV-C LEDS is effective to inactivate schistosome cercariae?
2. What is the suitable wavelength and optimal exposure time require to inactivate schistosome cercariae?
3. At what UV-fluence value will the water be free from schistosome cercariae?

1.5. Significance of the Study

Treating the contaminated water plays a vital role in controlling schistosomiasis and UV disinfection using UV-C LEDs is found to be the best alternative to disinfect the cercariae infested water. And the finding of this study will help to set protocol, implantation strategies and UV disinfection guidelines at different wavelength regarding schistosomiasis control. It will also help to serve as a starting point for future research to further optimize the wavelength in lab condition and see how different wavelength contribute for the inactivation process. It also provides information of how to eradicate and prevent the disease using these new UV-C LEDs disinfection technology.

1.6. Scope and Limitation of the Study

The experiment was conducted in the chemistry laboratory of Addis Ababa University under the College of Natural and Computational Sciences. The samples were collected from Lake Ziway and Tikur Wuha Rivers in Hawassa, which are endemic to *S.mansoni* and also close to Addis Ababa city where the laboratory is located. This research demonstrated the efficiency of UV-C LEDs in inactivation against the causative agent for schistosomiasis that is cercariae, by comparing three different wavelength and identified the efficient wavelength and fluence needed to inactivate cercariae with UV-C LEDs. The effectiveness of UV-C LEDs to disinfect sistosome cercariae was not evaluated on water samples from endemic areas due to poor water quality and significant turbidity. Since this factor affect the UV light from reaching the target and therefore requiring high fluence (UV dose). Due to this pre-treatment is recommended when utilizing UV light to disinfect cercariae.

2. LITERATURE REVIEW

2.1. Historical Occurrence of Schistosomiasis in Human

Schistosomiasis has been affecting human health for at least 4000 years; characteristic symptoms are described in early Egyptian papyri and analysis revels immunological clue as to its presence in ancient mummies. The finding of schistosomiasis in human was first discovered in 1851 by the German parasitologists, Theodor Maximillian Bilharz and Carl Theodor Ernst while they were performing autopsy (examination of the body of dead person) at kasr-El-Ainy hospital in Cairo and it was first named *Distomum haematobium* but then a person known Cobbold named it 'Bilharzia' as a generic term for the parasite. Sixty-four years after the discovery of Bilharz, Robert Thomson Leiper understood the complete cycle of *Schistosoma spp*, with the recognition of aquatic snails as intermediate hosts of these trematodes and he also distinguished between *S.mansoni* and *S. haematobium* by their morphology, egg type, and snail host (Di Bella et al., 2018).

On the other hand SANDBACH, (1973) describes the evolution of schistosomiasis by mentioning the time when human population shifted from hunter-gathering economy to domestic agriculture.It is thought that the settlement of the societies changed to a place where there is slow moving water in which the snail host of the disease abound. It is thought that the *schistosome* parasite first evolved around the Great Lakes of East Africa. It is then distributed to almost all parts of the world due to rapid development of large-scale irrigation projects and migration of people from infected to uninfected areas.

2.2. Epidemiology of Schistosomiasis

Mostly the occurrence of schistosomiasis infection in human is due to 3 species of Schistosoma which are *Schistosoma haematobium (S. hae-matobium), S. mansoni, and S. japonicum*. S. mansoni is the most common genus responsible for human infection. Schistosomiasis is spread by

coming into touch with polluted fresh water carrying schistosome eggs, which hatch in the water and release free swimming schistosomes. Miracidia is a parasite that contaminate aquatic snails Biomphalaria pfeifferi, which are an intermediary host for S. mansoni to complete its life cycle and discharge cercariae into water, which can infect humans through contact with water for various domestic reasons (Tefera et al., 2020).

Schistosomiasis distribution became rapid throughout the world. According to Utzinger, (2014) 800 million people are at risk and more that 200 million people are infected with these blood-dwelling trematodes. The study of the global burden of disease in Schistosomiasis was expected to cause 3.3 million disability-adjusted life years in 2010, with the majority of cases occurring in Sub-Saharan Africa (Boussinesq et al., 2014). On the other hand Workineh et al., (2019) explained that among the three species of *schistosma*, *s.hematobium* is the cause for urinary schistosomiasis and *S.mansoni* and *S.japonium* cause intestinal schistosomiasis. Furthermore, it was mentioned that this infectious disease has been a severe public health problem in 77 poor tropical and subtropical countries, with over 240 million people sick and 700 million at risk of infection worldwide, with Africa bearing the brunt of the burden. Over 300,000 individuals die each year in Sub-Saharan Africa as a result of S.mansoni and S.hematobium.

The possible control of the disease and a strategy to control morbidity caused by schistosomiasis was authorised by WHO in 1984 through preventative chemotherapy with praziquantel, this is because it has an excellent acceptability and good ability to either cure or drastically reduce egg output but it does not guarantee prevention of reinfection therefore can't eliminate schistosomiasis alone from most regions. This is why additional control measurements should be integrated like snail control, disinfection of water containing the parasite that penetrates human skin (cercariae),habitat change, predators and also biological competitors.(Colley et al., 2014)

2.3. Distribution of Schistosomiasis in Different Countries

Japan managed to stop transmission of schistosomiasis and it is thought that transmission does not occur in Japan anymore yet there are many countries suffering from the infectious disease, with the introduction of *S.manosni* to Mauritania, Senegal and Somalia, intestinal schistosomiasis is found in 54 countries including the Arabian peninsular, Egypt, Libya, Sudan and *S.japoncium* is endemic in China, Indonesia and Philippines. *S.haematobium* is endemic in 53 countries in the Middle East and most of the Africa countries including Madagascar and Mauritius. (Chitsulo et al., 2000).

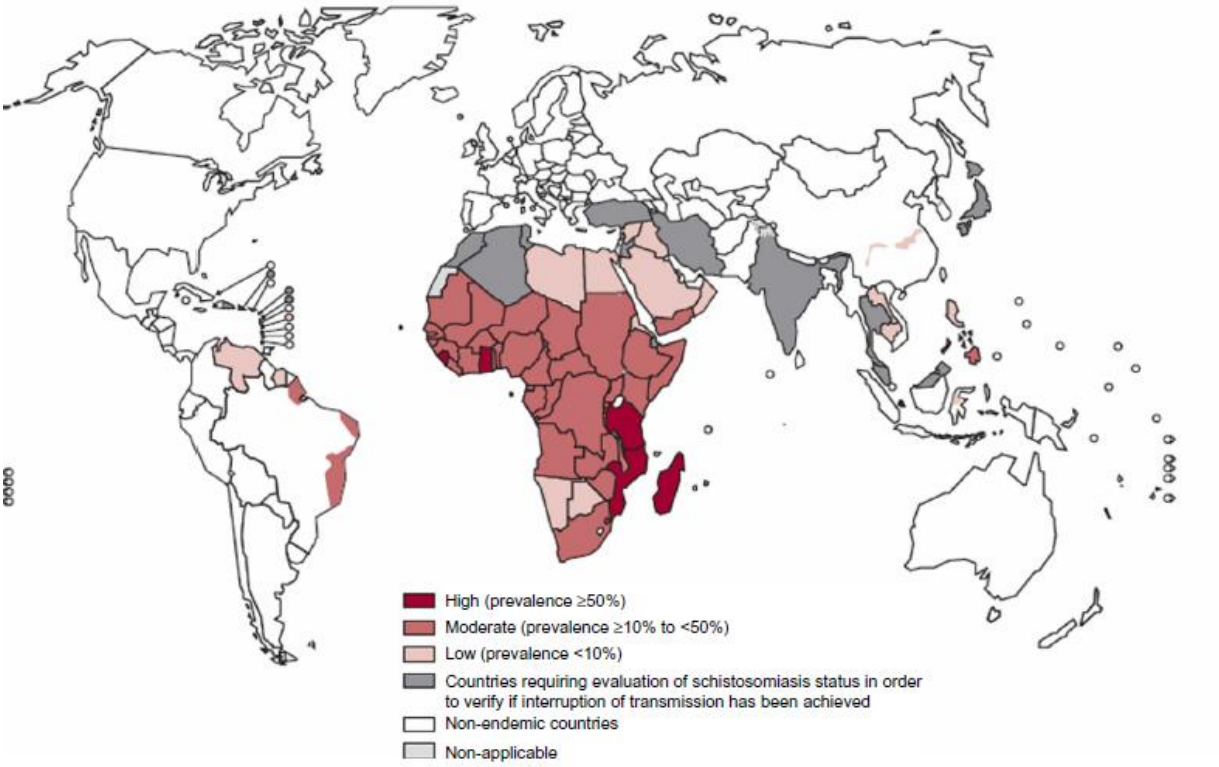


Figure 2-1 : Schistosomiasis prevalence in the world in 2010

2.4. Occurrence and Distribution of Schistosomiasis in Ethiopia

Schistosomiasis is also one of the most prevalent parasitic disease in Ethiopia. *S.mansoni* and *S.haematobium* are the species that are distributed throughout the country. *S.mansoni* is found mainly in altitudes between 1300-2000m above sea level and the responsible obligate intermediate host for the disease caused by *S.mansoni* is *Biomphalaria* (species of snail). The two species of the genus *Biomphalaria* responsible for transmitting *S.manosni* in Ethiopia are *B.sudanica* and *B.pfeifferi*, and *B.sudanica* has been documented from three locations in the rift valley: Ziway, Abaya lake, and the interference between Tikur wuha river and Awassa lake...(Alebie et al., 2014).

According to an Ethiopian Public Health Institute (EPHI) mapping survey in all regions of the country, there are an estimated 55.9 million people living in schistosomiasis endemic areas, including 5.0 million pre-school children, 17.7 million school-aged children, and 30.5 million adults. It was estimated that 35,775,100 cases of schistosomiasis occurred in Ethiopia in 2013 (EPHI et al., 2020).

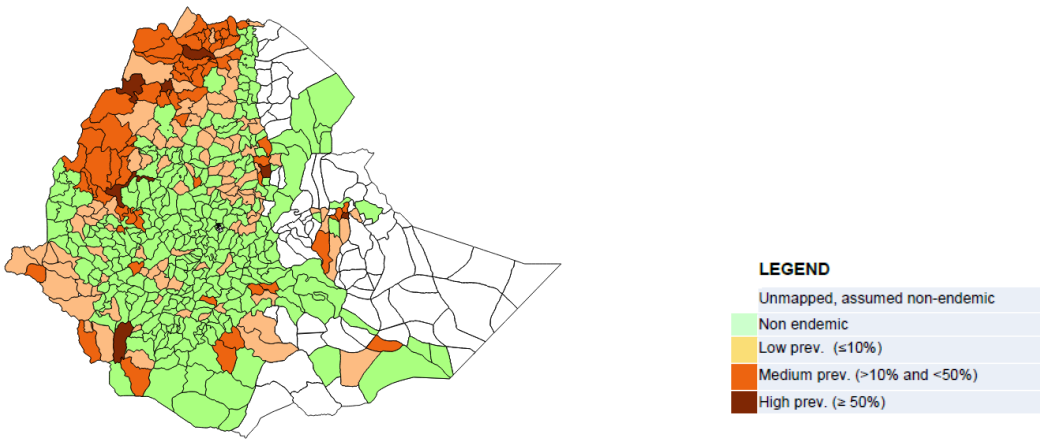


Figure 2-2 Combined data from EPHI and The Carter Center showing the distribution of schistosomiasis categorized according to WHO guidelines.(EPHI et al., 2020)

2.5. Control and Treatment Mechanism

Elimination of schistosomiasis has been successful in a number of countries including Japan but it remains a major public health challenge in many other countries. Therefore, different mechanisms have been used.(Chitsulo et al., 2000)

2.5.1. Chemotherapy

Preventive chemotherapy (PC) with praziquantel, which is effective, safe, and cost effective, is being used to control schistosomiasis. However, this treatment does not prevent subsequent reinfection, which means that if there is constant contact with water bodies infected with cercariae, this drug does not prevent or promise that reinfection will not occur (Braun et al., 2018).

And also Praziquantel has a number of drawbacks. For example, it is only effective against adult parasites, and a single treatment may not be sufficient to eliminate all worms, particularly in those with high-intensity infections. Similarly, even if the side effects of treatment are minimal, they might be quite painful for patients with significant worm burdens.(Secor & Montgomery, 2015)

2.5.2. Snail control

Snail control for Schistosomiasis transmission was an essential component of many regional schistosomiasis control efforts prior to the development of safe oral medication therapy (e.g., Praziquantel). During the 1950s, the most frequent strategy to snail control at transmission sites in Africa and the Americas was the use of chemical molluscicides, either as blanket or targeted treatments (King & Bertsch, 2015). According to Pointier, (2000) biological agents have been considered for integrated schistosomiasis-control in the last few decades as an alternative approach to chemical molluscicides and biological control using competitor snails has proven to be useful in the Caribbean area in controlling or even eliminating populations of the snail hosts of *schistosomes* in several types of habitat.

An Ethiopian pathologist Dr. Akiliu Lemma has also had a huge contribution in the area of snail control for eliminating schistosomiasis. He first noticed the mollusicidal property of ***Phytolacca dodecandra***, commonly known as endod, gopo berry, or African soapberry, during his study of the distribution and ecology of schistosomiasis-transmitting snails in the town of Adwa in the northern part of Ethiopia about 800 km from Addis Ababa. It was noticed that in places along rivers or streams where people washed clothes, there were comparatively more dead snails than there were in adjacent areas, whether upstream or downstream from these washing places. Subsequently, it was shown that a preparation of *P. doderandra*, widely used in Ethiopia instead of soap for laundering clothes, possesses molluscicidal properties(Lemma, 1970).

Though molluscicide application is the most cost-efficient and successful method of controlling snail populations for schistosomiasis control, There are limitation about the usage of molluscicides, including as their hazardous effect on macro- and microorganisms, as well as pollution of the environment.(Ross et al., 2014)

2.5.3. Storage

Cercariae cannot live without infecting a specific host for more than one or two days since they do not feed. The practice of storing water for 24–48 hours before use as a schistosome infection prevention technique has long been suggested, but it is time demanding and unpredictable (Grimes et al., 2015).

2.5.4. Chlorination

Most pathogenic organisms that are detrimental to human and animal life will be killed or inactivated by chlorine and its many forms. The most popular disinfection procedure for wastewater treatment is chlorination. A 'CT' value for chlorine inactivation is determined as the product of the residual chlorine dose (C) and contact time (T) required to inactivate a pathogen. For several waterborne diseases, CT values have been determined and can be utilized as a process monitoring parameter. Higher CT values indicate a higher chlorine tolerance (Braun et al., 2018). When cercariae-infected water is treated with chlorine solution, the cercariae move more slowly

and descend to the bottom of the container, and the movement of their bodies and tails gradually diminishes and eventually stops (Oliver et al., 1994). And also on the study done by (Braun et al., 2020), the effectiveness of chlorination against *S. mansoni* cercariae was tested and Chlorination tests show that S. mansoni cercariae are chlorine sensitive, with greater chlorine CT values required with higher pH and lower temperature.

Although chlorine is effective disinfectant against cercariae, it does have significant health and safety limitations, such as the chlorine residual, which is harmful to aquatic life even at low concentrations and may necessitate dechlorination, and some parasite species have shown resistance to low doses of chlorine (EPA, 1999).

2.5.5. UV disinfection

UV disinfection is a primary mechanism for inactivation of pathogenic organisms and the first solar UV disinfection was first discovered in the mid-to late 19^{th} century, at a time when contaminated water led to waterborne disease outbreaks and pandemics across Europe, Asia and the Americas. Then in 1877 two English scientists investigated the effect of light on microorganisms by exposing test tube of brown sugar solution to sunlight and monitoring for bacterial growth, bacterial growth was seen in the shaded sample but growth of the bacteria was inhabited after exposure of light for on moth. This experiment revealed that disinfection was dependent up on exposure time as well as intensity of the sunlight (S. Beck & Ph, 2016). Since then ultraviolet light has been very effective in disinfection and inactivation mechanisms of microorganisms. The mechanism involves absorption of ultraviolet light by DNA or RNA pyrimidine bases that is thymine or cytosine in DNA and uracil or cytosine in RNA, these will cause a photochemical reaction in which chemical dimer is formed between the two base and the dimer inhibits the formation of new DNA or RNA chains in the process of cell replication which will inhibit the microorganisms ability to replicate (Bolton et al., 2003).

Krakower was the first to investigate the direct effect of UV light on cercariae in 1940, when it came to inactivating cercariae with UV radiation. He discovered that 45 minutes of exposure to a mercury lamp was required to kill all cercariae in the infested sample, however he failed to specify the wavelength he utilized (Hazell et al., 2019b). Furthermore, according to Hazell et al., (2019b) review of Standen and Fuller's study, only four minutes were required to kill 100 percent of *S.mansoni* cercariae, but it is stated that the mercury lamp they used was very close to the sample and there is no evidence if they controlled the temperature..

There are different sources of UV light, from these sources low pressured mercury lamp is the most used UV light source in many studies. It emits monochromatic UV radiation at a wavelength of 254 nm and it has a wide application in drinking water and wastewater treatment plant both in developed and developing countries. The doses and efficacy needed using mercury-arc lamp are well-known for various microorganisms including bacteria, parasites and viruses. But this UV light source contains a very toxic chemical called mercury. There are also other different sources including sunlight and fluorescent lamps emitting different UV ranges including UV-C, the so-called germicidal range. Now a days there is a recent technology introduced as source of UV light in water sector which is known as UV-C LED. Various UV LEDs emit UV radiation at specific wavelength over the germicidal range (250-300 nm) and are considered as potential alternative method of UV delivery (Sholtes et al., 2016). This UV light source has more advantage than other UV light sources this because UV LEDs illumination offers a longer lifespan, is more reliable, and comes in smaller sizes, providing treatment plant designers the most design freedom(Grider, n.d.). According to Hazell et al., (2019b) In the near future, UV disinfection utilizing UV-C LED technology is expected to be a viable solution for long-term water treatment in low-income areas. It is also thought to provide safe water for activities involving water, such as bathing, washing, and improved hygiene. However, there is no standard method for calculating the UV fluence (UV dosage) required to kill schistosome cercariae with UV-C LEDs currently.

In UV disinfection process the fluence rate (UV dose) needed to inactivate microorganism is calculated as fluence multiplied by the exposure time. The fluence rate should be expressed in the

international system of unit Wm^{-2}. However, the unit mW cm^{-2} ($10Wm^{-2}$) is still quite common in many UV disinfection literatures (Bolton et al., 2003).

2.5.5.1. Corrections necessary when using UV lamps

Radiometer detectors are used to measure the irradiance that is released by the UV equipment. When UV exposure is done on a water media, the radiometer detector only provides a measure of the irradiance incident on the water at the center of the beam. In order to know the irradiance throughout the water media that is the average irradiance, several correction are required. Obtaining the average irradiance is very important since it provides an estimate of the average fluence rate at which a given microorganism is exposed and delivered fluence to a given sample can be also be calculated. The correction factors that are necessary while doing UV exposure experiment are Petri factor, Divergence factor, Water factor, Reflection and Sensor factor.(Bolton et al., 2016)

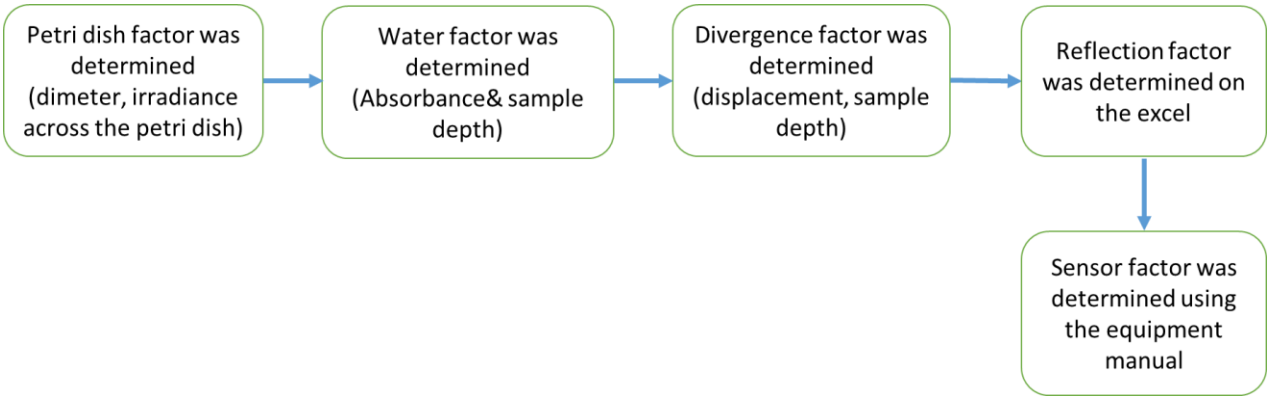


Figure 2-3 Schematic description of correction factor needed while using UV lamps

3. MATERIAL AND METHODS

3.1.Materials

3.1.1. Required Equipment

The following equipment's were employed to undertake the laboratory experiments:

- PearlLab Beam Triple Wavelength UV LED unit with Stage
- Radiometer and detector(international light ILT 2400)
- UV Spectrophotometer(1600 Series Single Beam UV-VIS spectrophotometer) and 10 mm quartz cuvette
- Small magnetic stirrer that fits beneath PearlLab Beam stage
- Small (e.g. 7 mm x 2 mm) Teflon coated magnetic stir bars
- Small (e.g. 30 mm diameter) Petri dishes
- Stereo microscope
- Small square of black card to cover sample (this acts as a shutter)
- Other basic lab equipment such as beakers, forceps, etc.

3.2.Methods

3.2.1. Sampling area

A live snail sample from the shores of Lake Zeway and the Tikur Wuha River near Hawassa, both of which are known to be endemic to schistsome cercariae, was collected. Both are about 166.6 and 279 kilometers from Addis Ababa, respectively, in the Rift valley basin (Tigist, 2020).

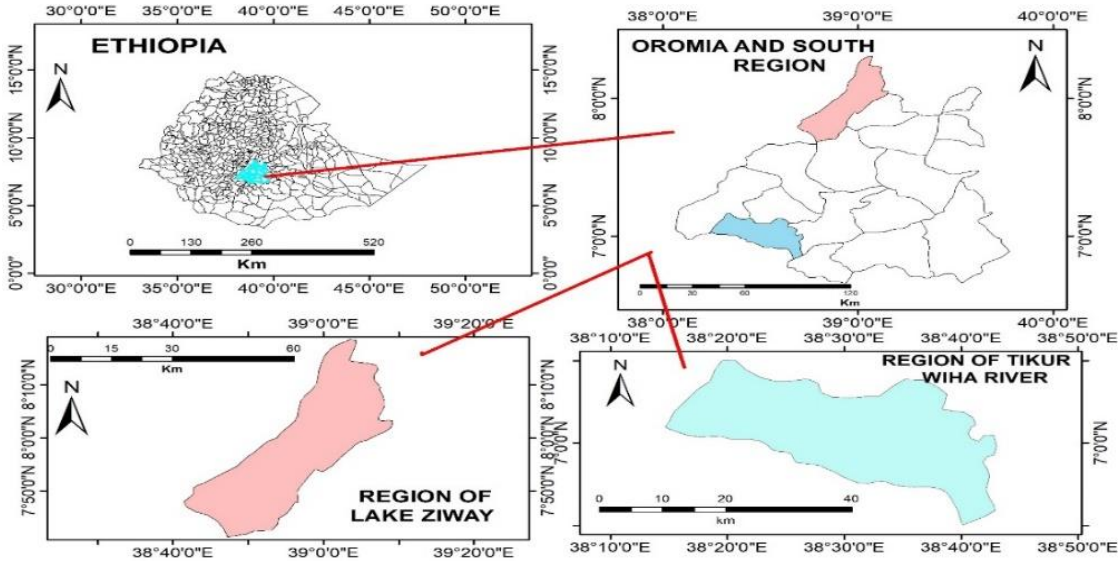


Figure 3-1: Map of sampling area

3.2.2. Sample collection

A bucket was filled with live snails collected at random from the banks of the Tikur Wiha River near Hawassa and Lake Ziway using a sieve with extended arms.

a) Tikur Wiha River site b) Lake Ziway site c) Collected alive snails

Figure 3-2 Sampling sites and collected alive snails

The snail genera *Biomphalaria* is the most common and widely distributed snail intermediated host for *S.mansoni*, while *Biomphalaria pfeifferi* is the most common and widely distributed snail intermediated host for *S.mansoni.* (Hailegebriel et al., 2020). These are snails that were collected for this study and are shown in the figure 4. While bringing the sample to the laboratory, water and aquatic macrophytes from the location were added to the bucket to simulate the snails' natural environment. Each day, the water and macrophytes were changed to keep the snail alive until cercariae were no longer shaded.

3.2.3. Equipment set up

Spectrophotometer (1600 Series Single Beam UV-VIS spectrophotometer) was set to required wavelength (to match the weighted average wavelength of the selected LEDs that is going to be used). Spectrophotometer was allowed to warm up for 30 minutes before use.

PearlLab Beam and Stage was set up as per the Aquisense instrument manual. The cap of the radiometer detector was removed and placed at the center of the stage. While the PearlLab Beam is in place (but with the UV LEDs off) the radiometer was adjusted to zero.

The radiometer detector was covered with black card and then the PearlLab Beam was switched to the required wavelength and the "shutter" button was used to switch on. Then it was left to warm up for 15 minutes (Bolton et al., 2016).

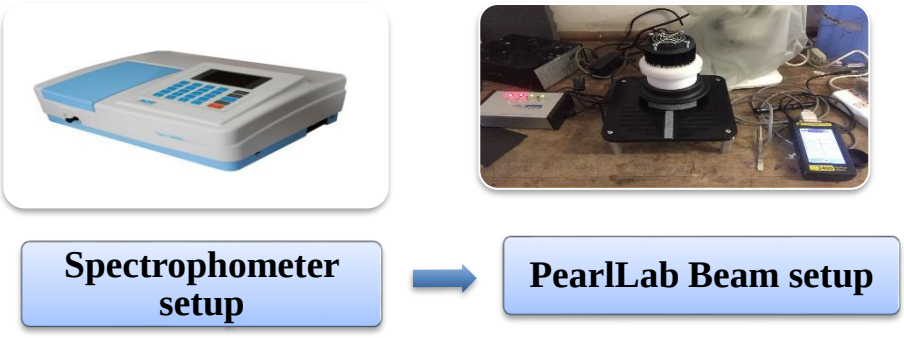


Figure 3-3 Equipment set up

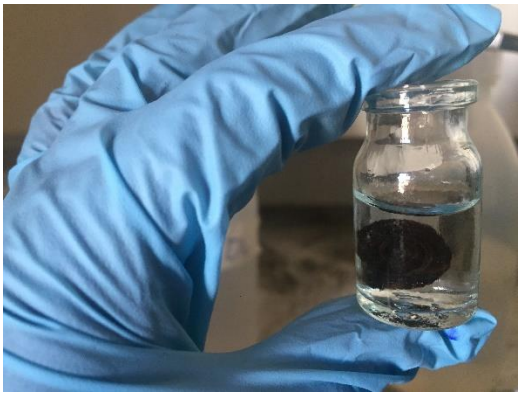
3.2.4. Monitoring UV system and adjustment of different parameters

Simple UV disinfection systems have little monitoring and control, often only a power on/off indictor and visual or audible alarms to indicate power or material failure within the machine. Daily checks has been made to ensure that the equipment is working.(DWI, 2016).

In addition, different correction factors were calculated such as Petri factor, Divergence factor, Water factor, Reflection factor and Sensor factor using a spreadsheet based on the protocol adapted by (Bolton et al., 2016). And UV disinfection system is relatively simple to operate and monitor yet UV disinfection will only be effective if certain constrains are met. The ideal water for use with UV has minimal dissolved substance, is free from turbidity and suspended solids and low in organic compound and color (Techincal Manual, 2019). As a result, bottled water was utilized, and the temperature in the room was maintained (220 and 240 ^{0}C) with a pH of around 7.

3.2.5. Cercariae preparation

A microscope with a magnification power of F10*/20 was used to select the infected snails from the obtained sample. Shedding snails were placed in a small bottle with a small amount of bottled water. The bottle was swirled gently to rinse snails, then forceps was used to transfer snails to a second clean small bottles containing 1-2 mL of bottled water. The snails were left to shed under direct light for about 50 minutes. When the amount of cercariae in the water was determined (100-120 cercariae/200µL), the snails were removed, and the cercariae-infested water was filtered using a 200 µm polyester filter to eliminate snail excrement.(Brady, 2016)



a) Preparation of snail for shedding

b) shedding snails under direct light

Figure 3-4 Cercariae Preparation

3.2.6. Irradiance Measurement and Calculation of Exposure Time

The internal diameter of the Petri dish was measured and entered into the spreadsheet. Based on the sample volume, stir bar volume, and Petri dish diameter, the depth of the sample was calculated and entered into the Excel 2016 spreadsheet.

In order to calculate for the average irradiance of the UV-LEDs, the irradiance thought the Petri dish that is going to be used was measured. In order to do this the diameter of the Petri dish was measured using a ruler and also the depth of the sample was calculated using the sample volume, stir bar volume and the diameter of the petri dish. It is calculated using the following equation.

$$\textbf{Depth of the sample} = \frac{\boldsymbol{sample\ volume + stir\ bar\ volume}}{\boldsymbol{\pi r^2}}$$

Using the grooves as a guide, the PearlLab Beam was moved across the stage on a 5 mm grid, taking irradiance readings as the points indicated on the spreadsheet. This data was also entered into Excel 2016 spreadsheet. The cap of radiometer sensor was replaced and moved to one side. Magnetic stirrer was placed under the stage of the PearLab Beam and the height was adjusted so that the sample surface will be at the same height as the measurement plane of the radiometer

detector. The irradiance of the LED light source was measured using a radiometer (international light ILT 2400) placing it under a small opening on the stage of PearlLab Beam. Since the diameter of the Petri dish is 50mm the irradiance was measured 25mm of each X and Y direction using the grooves as a guide, moving the PearlLab Beam across the stage on a 5 mm grid.

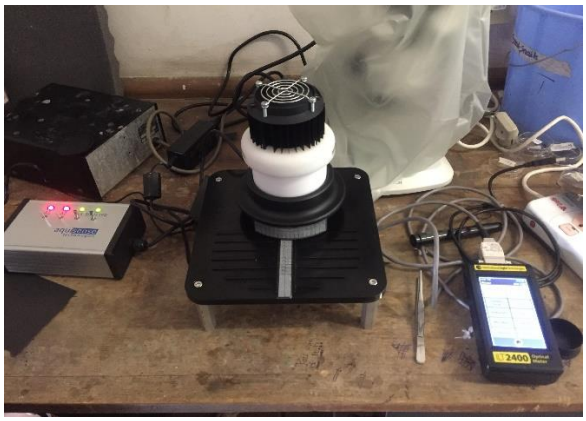


Figure 3-5 Irradiance measurement using UVC-LEDs

To measure the absorbance of the cercariae infested water sample, 3.8 mL of bottled water was added to a clean 10 mm quartz cuvette and was inserted into the spectrophotometer and the spectrophotometer was made zero. Then the cuvette was emptied and 3.6 mL of bottled water and 200 µL of cercariae infested water (total volume 3.8 mL) was added. The lid on the cuvette was put and inverted 3 times. Finally the cuvette was insert into spectrophotometer and the absorbance was measured. The above procedure was done in triplicate and the mean of the 3 absorbance readings was calculated and the acquired data was entered into the spreadsheet.

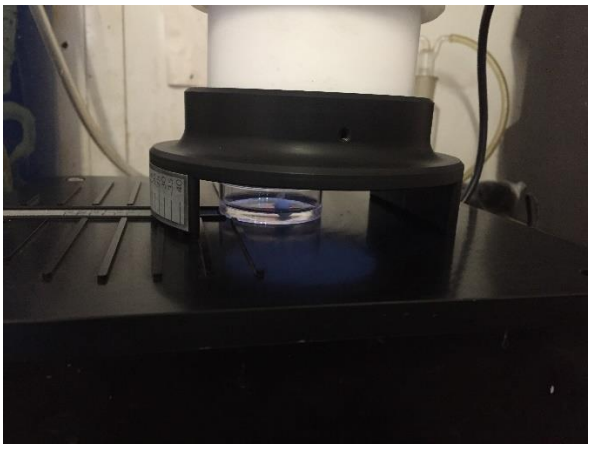
Finally the required exposure times was calculated based on the required fluence and the average fluence rate calculated by the spreadsheet and a randomised order of fluence(UV dose) for the exposures was established.(Bolton et al., 2003).

3.2.7. Exposure Procedure

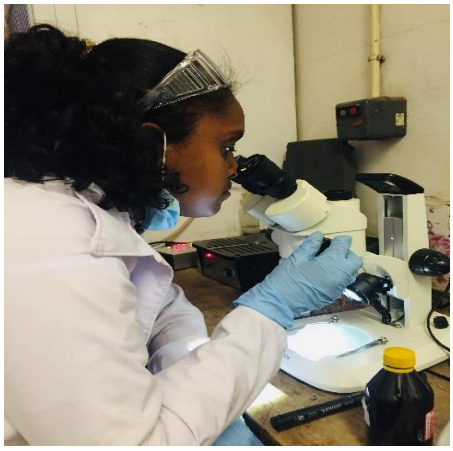
The PearlLab Beam was removed from the stage and the first Petri dish was placed on the center of the opening on the magnetic stirrer. A stir bar, 3.6 mL of bottled water, and 200 µL of *cercariae* infested water was added. The stirrer speed was set to the lowest speed so that the sample is gently mixed without causing a vortex. Then a piece of black card was placed over the stage opening so that it is completely covered. After covering the opening of the stage with the black card, the PearlLab Beam was replaced on to the stage in the correct centered position and the required exposure time on the timer was set, then as the timer is started the black card will be remove so that the sample is exposed to UV light. As soon as the time sounds, the black card was replaced and the Petri dish from the magnetic stirrer was removed and the stir bar was also removed carefully from the petri dish holding the sample by a larger magnet.

After exposing the sample to UV light, the dead cercariae was counted using microscope immediately and the number of the dead cercariae was noted down. Then the total number of cercariae was counted by adding 20 µL of Lugols iodine solution to the sample to fix and stain all cercariae. The above steps was repeated until all exposures have been carried out.

Once all exposures have been carried out, the irradiance was measured using the same method described above and the data was entered into Excel 2016 spreadsheet. Finally, the average fluence was calculated from the previous and later irradiance readings and exposure time that became exactly the same to the expected fluence, which proves that the output from UV-C LEDs is generally very stable (Bolton et al., 2003).



a) Exposing the sample under UV-LED light



b) Counting the dead cercariae

Figure 3-6 Exposure Procedure

3.2.8. Data analysis

The experiment was done in triplicate for each fluence and the average was taken for calculating the log reduction.

The log reduction was calculated for each sample using the following equation and the mean log reduction was calculated for each fluence after the calculation, fluence against log reduction was plotted

$$Log\ reduction = -\log_{10} \frac{\%\ Alive\ cercariae\ (sample)}{Mean\ \%\ alive\ cercariae\ (control)}$$

The mean log reduction was calculated for each fluence and it is plotted against fluence for all three wavelength using excel 2016. All the graph showed a mean value of log-reduction and some graphs also contains an error bar representing the standard deviation which is done using Excel 2016.

The correlation coefficient was also analyzed using Excel 2016 to describe the relationship between fluecne, log reduction and exposure time.

4. RESULT AND DISCUSSION

4.1. Exposing cercariae infested water to UV light

The majority of the cercariae species seen in this study were human forms of schistosomiasis, however another type of cercariae was discovered that is likely to be a bird schistosome. This was discovered by comparing the morphology of cercariea examined in this study to that of cercariae species and morphology studied in prior studies. Such as a study conducted by Mohammed et al., (2016), illustrated the morphology of various cercariae species. The image below from the study showed two types of human schistosomiasis (*S.haematobium S.mansoni*) labeled a and b respectively. The majority of cercariae observed in this study have the same head and tail structure as b, which is identified in the study as *S.mansoni*.

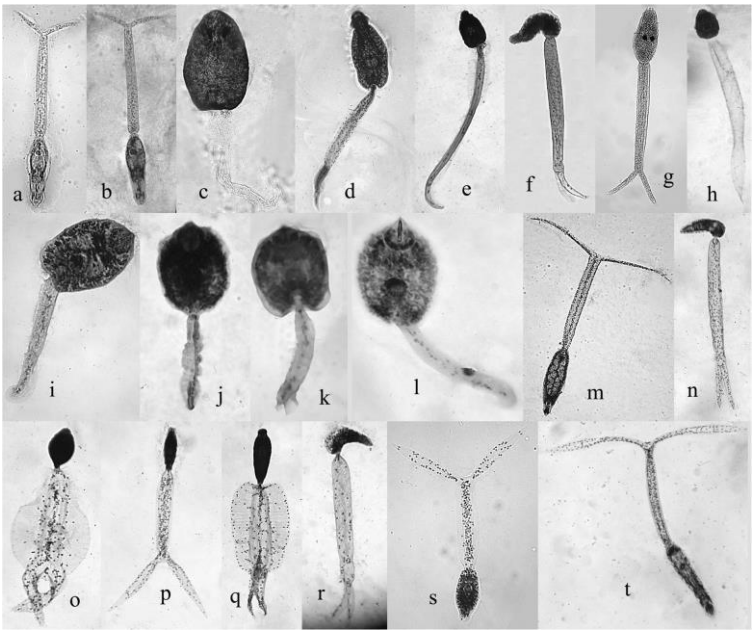


Figure 4-1: Morphotypes of cercariae recorded from five snail species in the East Nile locality, Khartoum, Sudan. The types are: - a) *Schistosoma haematobium* (or related species); b) *S. mansoni*.(Mohammed et al., 2016)

The sample containing cercariae were exposed to UV light emitted from a UVC-LEDs. But the cercariae with no exposure were moving fast by contracting and extracting their head and tail. Figure 8 showed the structure of cercariae with no exposure which is exactly the same as seen in this study, the first row is 40x magnification and second row is the same cercariae at 100x magnification.

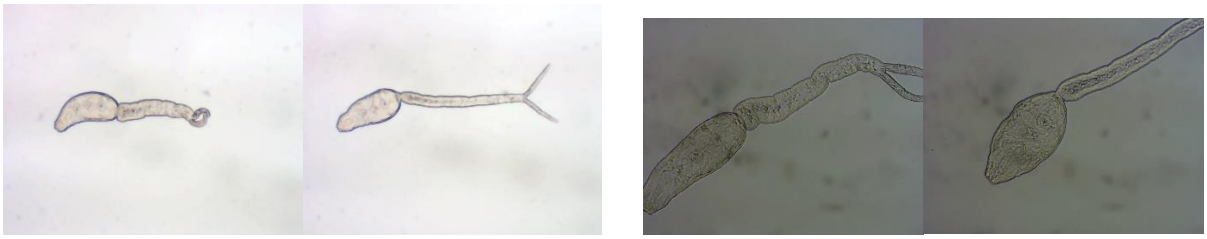


Figure 4-2: Structure of cercariae at 0mJ/cm^2 under a microscope (Lucinda Hazell)

After exposing the cercariae to a fluence of 100 mJ/cm^2 , contraction and extension of head/ oral sucker continued and the tail was rigid, straight with tightly curled fork which is an indicator that the cercariae was still alive but with slowed movement. This is shown in figure 4-3.

Figure 4-3: Structure of cercariae after exposure of UV- light at a fluence of 100 mJ/cm^2 and magnification of 100X

At the last fluence (300 mJ/cm^2) almost all cercariae died showing a ditched head and tail from the body. Both oral and ventral suckers were protruding and there was some liquid expelled from the base of the head at head/tail junction and the base of the head at the head/tail junction was possibly

a different shape to that of normal cercariae and the tail was fully relaxed. This is also illustrated in figure 4-4 below.

Figure 4-4: Structure of cercariae after exposure of UV- light at a fluence of 300mJ/cm^2

4.2. Computing the UV-Fluence for Disinfecting Cercariae of Schistosomiasis

The effect of UV light from UV-LEDS to inactivate cercariae was examined in the laboratory at three different wavelength 255 nm, 265 and 285 and in 10 doses (fluence) for each wavelength in the range between 1mJ/cm^2 and 300 mJ/cm^2.

In order to calculate the fluence needed to disinfect cercariae of schistosomiasis, the central irradiance was first determined by using radiometer (international light ILT 2400) and the result was 0.203 mW/cm^{2,} 0.237 mW/cm^2 and 0.740 mW/cm^2 for 255 nm, 265 nm and 285 nm, respectively.

The irradiance varies somewhat over the surface area of the liquid sample to be irradiated therefore the Petri dish factor was considered and the irradiance across the petri dish was measured by moving Pear Lab beam UV-LED progressively in 5 mm in +X,-X,+Y and –Y direction for 50 mm diameter Petri dish. The petri factor was found to be 0.889, 1.051 and 0.906 for 255 nm, 265nm and 285 nm wavelength, respectively

Water might also absorb UV light at a wavelength of interest therefore it was necessary to account for the decrease in irradiance. The water factor was also considered and put on excel sheet which was 0.999 for all wavelength.

The distance of the UV-LED and the sample was also considered, by measuring the distance from the top surface of the Petri dish to the center of the UV-LED. The value was added to the excel sheet. And also whenever a light passes from one medium to another a small fraction of beam is reflected off the interface of the media, therefore the water factor was also considered and according to Bolton et al., (2016) the reflection factor for water is 0.975.

After calculating the above correction factors necessary for calculating the average irradiance for PearLab Beam UV equipment using a prepared excel spreadsheet based on a paper published with a title of *standardization of methods for fluence determination* by (Bolton et al., 2003).The average irradiance of the each wavelength 255, 265 and 285 was determined to be 0.260 mW/cm^2, 0.297 mW/cm^2 and 0.694 mW/cm^2 respectively. Once the average irradiance in the water is calculated, this is the same as the average fluence rate (mW/cm^2), the inverse of this number is the exposure time (in seconds) to obtain a fluence (UV-does) of 1 mJ/cm^2.

Based the principle proposed by Bolton et al., (2003) the exposure time was calculated for each exposure that starts from 1mJ/cm^2 and goes to 300mJ/cm^2 and the experiment was done in triplicate for each exposure. Once the exposure time is known for one 1 mJ/cm^2 UV dose, then it can be calculated for any desired fluence by multiplying the exposure time of 1 mJ/cm^2 by the fluence desired. This is shown in the table below.

Table 4-1 Computed Fluence and Exposure time for 255 nm, 265 nm and 285n m wavelengths

Parameters	Exposure time(h:mm:ss)		
Fluence (mJ/cm^{-2})	255 nm	265 nm	285 nm
1	00:00:04	00:00:03	00:00:01
20	00:01:17	00:01:07	00:00:29
40	00:02:34	00:02:15	00:00:58
60	00:03:51	00:03:22	00:01:27
80	00:05:07	00:04:30	00:01:55
100	00:06:24	00:05:37	00:02:24
120	00:07:41	00:06:44	00:02:53
160	00:10:15	00:08:59	00:03:51
200	00:12:48	00:11:14	00:04:48
300	00:19:13	00:16:51	00:07:13

4.3. Determination of Wavelength to Inactivate Cercariae of Schistosomiasis

Using the above data of fluence (UV dose) form 1 mJ/cm^2 to 300 mJ/cm^2 and the exposure time for each fluence, the disinfection of cercariae of schistosomiasis was examined in this study. UVC-LED exposure was carried out at a water temperature of 23°C and a pH of around 7. The sample containing 100-120 cercariae in 3.8 ml of bottled water was tested under varied fluences (UV dose) and achieved an effective result of 99.9% cercariae inactivation

After the sample containing the cercariae infested water is exposed to the UVC light that comes from PearLab beam instrument, the result was examined under a microscope. It was visible under a microscope that when cercariae are exposure to UVC light their movement was slowing down and settling down to the bottom of a Petri dish showing that the cercariae's are dead. The number

of dead cercariae's were recorded after every exposure of each fluence for the given time shown in Table 1.

Figure 4-5 shows resistance of cercariae of schistosomiasis to UVC light at a fluence from 1-120 mJ/cm^2 in all three wavelengths throughout the experiment. The result shows more that 70% of alive cercariae in the sample are moving. Yet some were showing a weakened movement and sinking down to the bottom which indicates dying of cercariae.

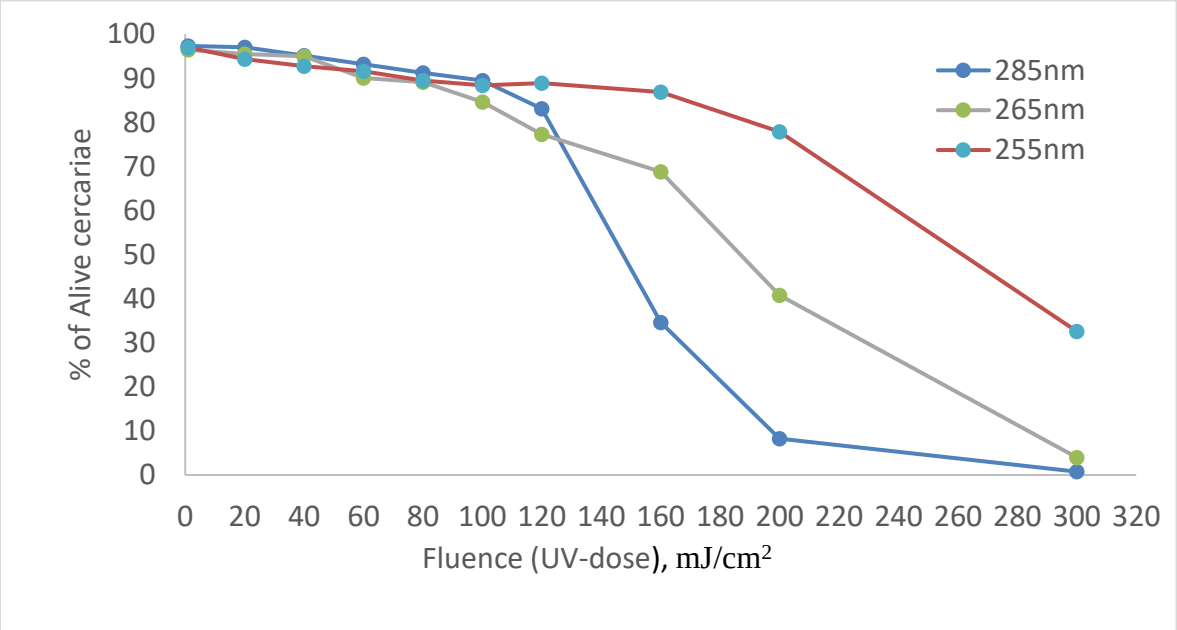


Figure 4-5: Percent of alive to total number of cercariae

At wavelengths of 265 nm and in a fluence of 160 mJ/cm^2 , 200 mJ/cm^2 and 300mJ/cm^2 , 68%, 40% and 4% of cercariae;s were able to survive respectively . And at same fluence at a wavelength of 285nm 34%, 8% and 0.7% of cercariae;s were able to survive. Which shows that the UV light exposed form UVC-LED is capable of disinfecting cercariae schistosomiasis.

The result also indicated that the capacity to disinfect cercariae of schistosomiasis using UVC-LEDs increased as the fluence (UV dose) increased. It was observed that as the fluence was

increasing with respect to the exposure time, the number of moving and alive cercariae decreased indicating that the more the fluence the less the number of alive cercariae within the sample solution. And this shows that the UV light emitted from UVC-LEDs are effective against disinfecting cercariae of schistosomiasis.

Determining the wavelength and exposure time required to inactivate cercariae of schistosomiasis using UV-C LEDS was one of the objective of this research. Based on the results acquired form the laboratory experiment, there were active cercariae of schistosomiasis at a wavelength of 255 nm until the last does 300 mJ/cm^2 exposed for 19 min and 13 seconds which is a high dose when comparing to other studies done. For example, Tian et al. (2010) conducted an immunization trial to examine the use of UV-attenuated cercariae to generate a vaccine in contradiction of human schistosomiasis. At a wavelength of 254 nm, a 1 log reduction was obtained in this investigation.

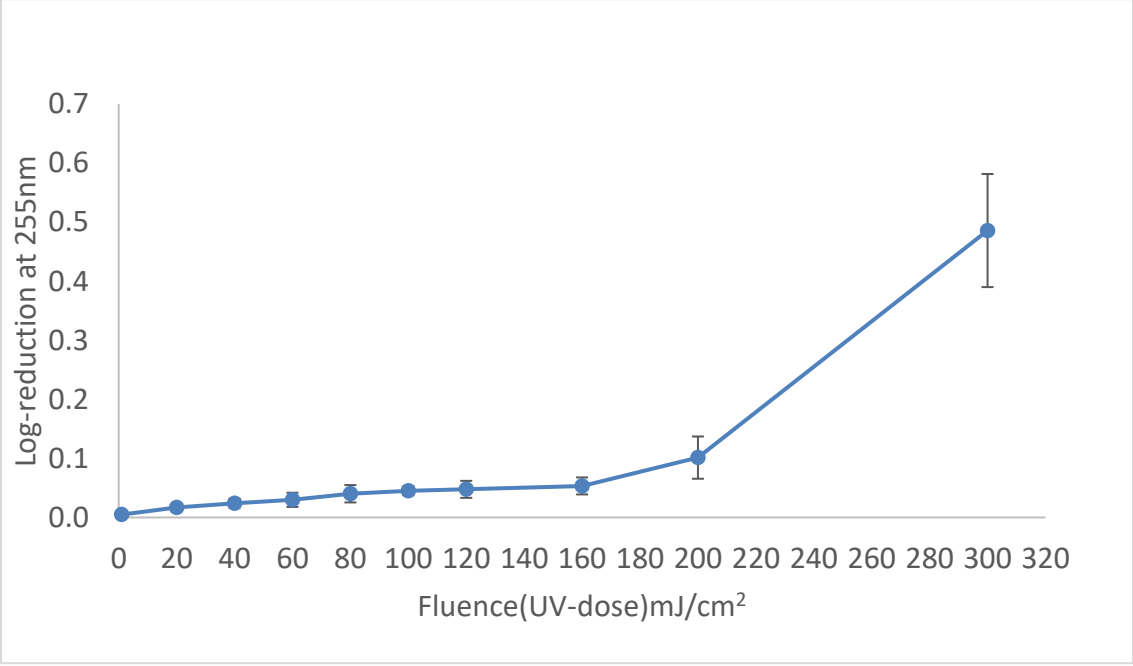


Figure 4-6: Log reduction at a wavelength of 255 nm.

Figure 4-6 shows a plot of fluence against log-reduction for a wavelength of 255 nm. When doing the exposure of the cercaria*e* infested sample to a 255 nm wavelength of UV-light using PearLab beam, there were alive cercariae's until the last fluence 300 mJ/cm^2 which is the highest UV-dose. More than 30% of the cercariae was alive and the log-reduction was below 0.5 which indicates that the disinfection is not effective at a wavelength of 255 nm. This value of log reduction is lower when compared to Tian et al., (2010) and According to the review done by (Hazell et al., 2019b) in these experiments the cercariae were exposed to a fluence of 5–14 mJ/cm^2 , which is very low compared to 300mJ/cm^2 but high enough to cause damage and stop development into adult worms. The UV dose is low in the previous study because of how the inactivation was measured, the inactivation in Tian et al., (2010) study was measured by worm burden while in this study it was measured by number of dead cercariae.

But this does not mean the UVC-LED at a wavelength of 255nm can't disinfect but it needs higher dose and exposure time. This relationship is presented in Tables 4-2 and 4-3 showing the correlation between fluence and time to log-reduction, respectively.

Table 4-2 : Correlation coefficient between Fluence and Log reduction

Parameter	*Fluence*	*Log-reduction at 255 nm*
Fluence	1	
Log-reduction at 255 nm	0.84	1

Table 4-3 Correlation coefficient of Exposure time and Log reduction

Parameters	*Exposure time*	*Log-reduction*
Exposure time	1	
Log-reduction at 255nm	0.84	1

The result in Table 4-2 shows that there is strong positive relationship between fluence and Log-reduction having 0.842 correlation coefficient. Table 4-3 shows again strong positive relationship between exposure time and Log-reduction of 0.841 correlation coefficient. Both Tables indicate

that when fluence and exposure time increased the Log-reduction will also increase since there is strong positive correlation. Significance level

Exposure done with the second wavelength 265 nm, was more effective than 255 nm. A 1.5 Log reduction was achieved at exposure time of 16 min: 51 sec in 300 mJ/cm^{-2} fluence. At this wavelength alive cercariae were observed until 200 mJ/cm^{-2} showing 40% of cercariae alive in the sample solution. Yet their movement was weakened and most of them have settled down to the bottom of the Petri dish.

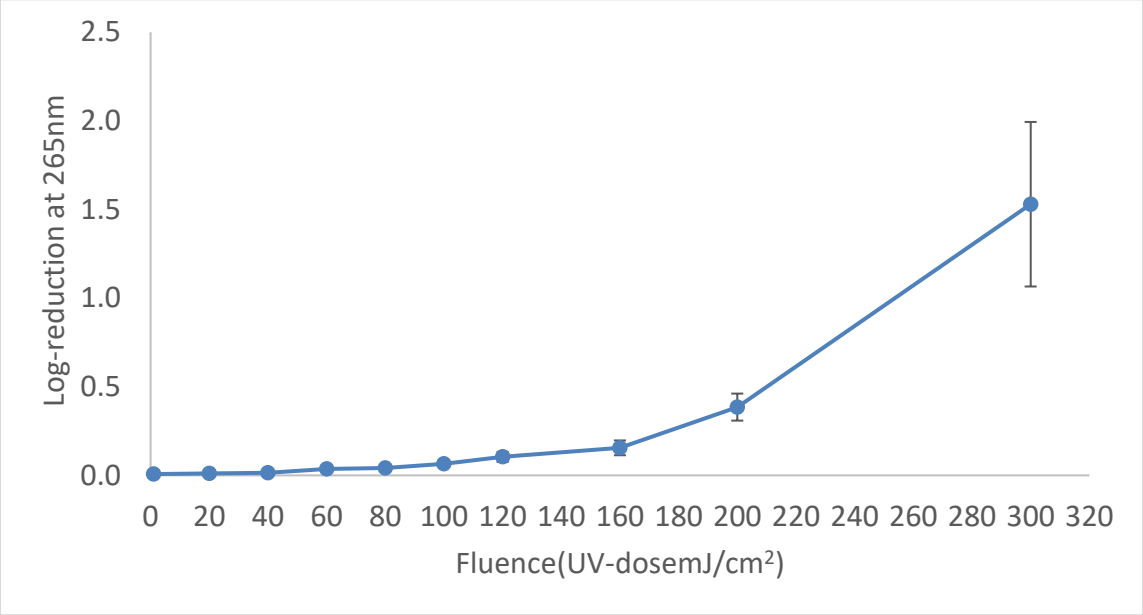


Figure 4-7: Log reduction for a wavelength of 265 nm

Figure 4-7 shows a reduction of 1.5 log was achieved at 300 mJ/cm^{-2}. At the first wavelength 255 nm almost 0.5 log reduction was achieved by the exposure done for 19 min and 13 sec and when it comes to the second wavelength 265 nm a 1.5 log reduction was achieved by the exposure done for 16 min and 51sec. This shows that as the wavelength increased form 255 nm to 265 nm the ability to disinfect cercariae of schistosomiasis has also increased.

In the third and last wavelength in the PearLab beam UVC-LEDs which is 285 nm, much more disinfection was achieved as compared to the other two wavelengths (255 nm, 265 nm) as shown in the next Figure 16. On this wavelength log-reduction of 2 that is 99% of inactivation was achieved at 300 mJ/cm^{-2} at exposure time of 7 min and 13 sec.

And also 1-log reduction that is 90% of inactivation was achieved at 200 mJ/cm^{-2} at exposure time of 4 min and 48 sec which is short exposure time than the other wavelength to achieve 1 log-reductions.

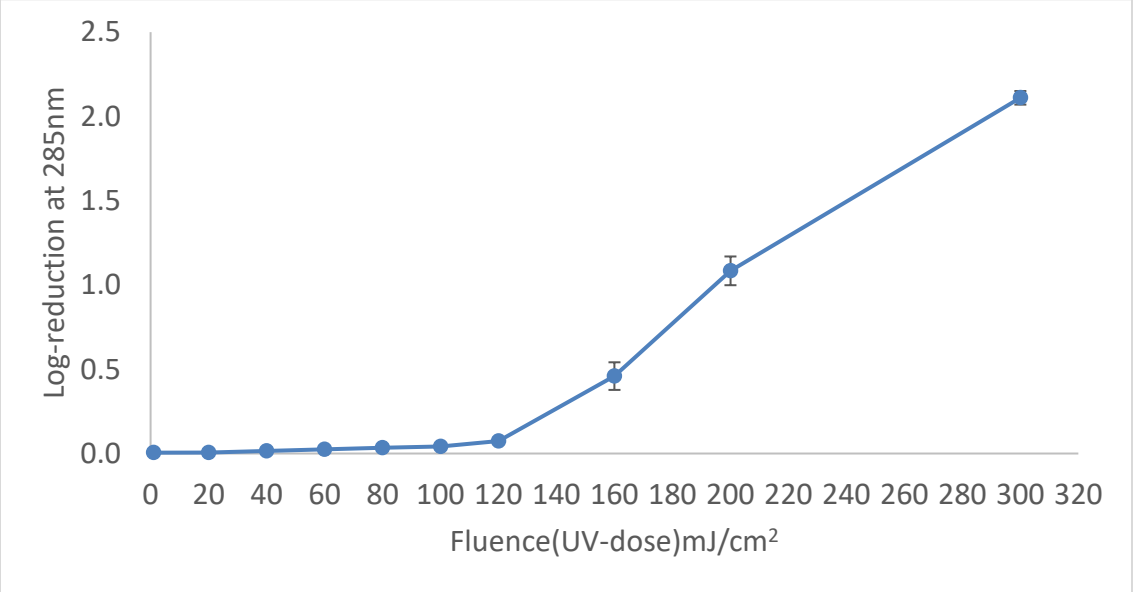


Figure 4-8: Log reduction for a wavelength of 285 nm

As shown in Figure 4-8 more than 38% of viable cercariae was noticed until fluence 160 mJ/cm^2 and a log-reduction below 0.5, but the number of dead cercariae start to increase drastically starting form 200 mJ/cm^2 to 300 mJ/cm^2. At 200 mJ/cm^2 6-9% of cercariae were alive and 1.1 log-reduction was achieved and at 300 mJ/cm^2 0.7-0.8% 0f alive cercariae were noticed and 2.1 log-reduction was achieved showing almost all cercariae are dead at 300 mJ/cm^2.

When researching the efficacy of UV-LEDs and comparing all three wavelengths on their potential to disinfect schistosomiasis cercariae, it was found that UV-LEDs can be used to disinfect schistosomiasis cercariae. Among the three wavelengths examined in this study, 285 nm was found to be the most effective in achieving 99 percent cercariae inactivation with a 300 mJ/cm^2 UV-dose and 7 minutes and 13 seconds of exposure time. The log-reduction verses UV-dose for all wavelength 255, 265 and 285 nm is shown in Figure 4-9.

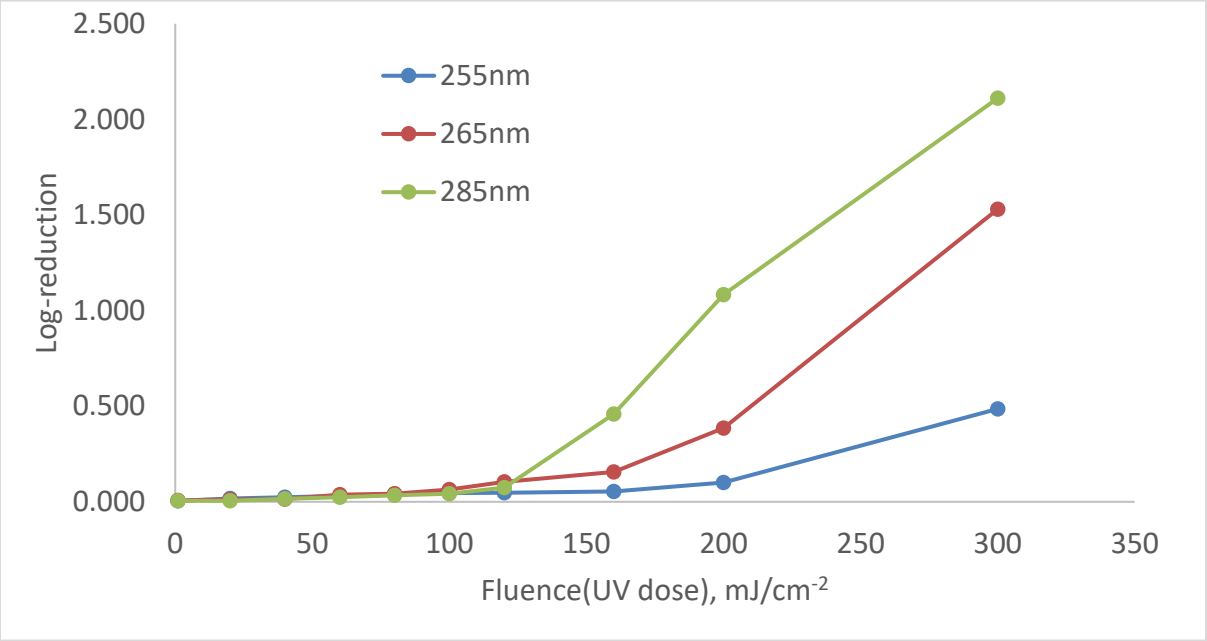


Figure 4-9: Log reduction for all three wavelength 255nm, 265nm and 285nm

And also the correlation coefficient in Table 4-4 below shows that there is a strong relationship between all wavelength and fluence (UV dose), which indicates the increment of inactivation of cercariae as the wavelength and the corresponding fluence increased.

Table 4-4 Correlation coefficient of Fluence and Log reduction for all wavelength

Parameter	*Fluence*	*Log-reduction at 255 nm*	*Log-reduction at 265 nm*	*Log-reduction at 285 nm*
Fluence	1			
Log-reduction at 255 nm	0.84	1		
Log-reduction at 265 nm	0.86	0.99	1	
Log-reduction at 285 nm	0.916	0.93	0.96	1

It is also be seen in Table 4-4 that the Log-reduction at a wavelength of 285 nm has a very strong positive correlation of 0.91 with the fluence, indicating that more inactivation could be achieved at 285 nm.

The influence of exposure time on inactivation of cercariae was also examined in this study. The result showed that as the wavelength increased the exposure time became less. When the wavelength increased from 255 nm to 285 nm, the exposure time needed to kill the cercariae became less. This is because protein has a relative peak in UV absorbance at 280nm due to the absorption of aromatic amino acids. As a result, it is reasonable to conclude that LEDs emitting at 280nm may cause protein damage, resulting in improved efficacy.(Wickham et al., 2019)

In this study a wavelength of 255 nm took 19 minutes and 13 seconds to achieve 0.5 log reduction and less time that 16 minutes and 51 seconds was needed to achieve 1.53 log-reduction in a wavelength of 265 nm. The last wavelength 285nm took 7mintes and 13 seconds to achieve 2.1 log-reduction and 4mintes and 48seconds to achieve 1 log reduction. When we compare these three wavelength based on the exposure time, wavelength 285nm took less time than the other two for achieving 2 log-reduction that is 99% inactivation of cercariae of schistosomiasis.

When the exposure time increased the log- reduction was increasing for each wavelength for instance for a wavelength of 255nm the log reduction increased from 0.005 to 0.486 as the exposure time increased from 4 seconds to 19mintes and 13 seconds. For a wavelength of 265 the log reduction increased from 0.01 to1.5 as the exposure time increased from 3 seconds to 16minutes and 51 seconds and for a wavelength of 285nm the log reduction increased from 0.005 to 2 as the exposure time increased form 1 second to 7 minutes and 13 seconds. Showing that the exposure time and the log-reduction in this study have a positive relationship and this is proved by analysing the correlation coefficient. The correlation of time and log-reduction for a wavelength of 255 nm, 265 nm and 285 nm is show on Table 4-5, 4-6 and 4-7 respectively.

Table 4-5 Correlation of exposure time and log reduction for a wavelength 255nm

Parameters	*Exposure time*	*Log-reduction*
Exposure time	1	
Log-reduction	0.84	1

Table 4-6 Correlation of exposure time and log reduction for a wavelength 265nm

Parameter (265 nm)	*Exposure time*	*Log-reduction*
Exposure time	1	
Log-reduction	0.86	1

Table 4-7 Correlation of exposure time and log reduction for wavelength 285n

Parameter (285 nm)	*Exposure time*	*Log-reduction*
Exposure time	1	
Log-reduction	0.91	1

Table 4-5 shows that there is a significant positive correlation between the exposure time taken when applying UV light on the sample of cercariae infested water and the log-reduction with *r* (8)

= 0.84 and $p < 0.002$. Table 4-6 shows that there is also a significant positive correlation between the exposure time taken when applying UV light on the sample of cercariae infested water and the log-reduction with $r(8) = 0.86$ and $p < 0.001$. Table 4-7 also has the same relationship with $r(8) = 0.91$ and $p < 0.001$. The significance was determined using a regression statistics table generated from Excel 2016.

4.4. Effectiveness of UV-disinfection against cercariae of schistosomiasis at three selected wavelength using UV-C LEDs

This study presented the reaction of cercariae of schistosomiasis to UV irradiation from UVC-LEDs. It has been found that the dose needed to disinfect cercariae of schistosomiasis effectively was 300 mJ/cm^{-2} at a wavelength of 285 nm. This UV dose (Fluence) is high when comparing to the doses needed to disinfect other disease causing infectious parasites.

Escherichia coli is one of the most common infection causing pathogenic parasite which causes disease in human through fecal-oral route of transmission and on the study done by (Nyangaresi et al., 2018). The fluence required to attain above 3.0-log and 4.0-log inactivation was 8.78 and 10.09 mJ/cm^{-2} at a wavelength of 267 nm and 15.35 and 23.04 mJ/cm^{-2} at a wavelength of 275 nm.

The effect of UV light on the most chemically resistant protozoan parasites, such as Cryptosporidium and Giardia, was also investigated. Cryptosporidium and Giardia are protozoan parasites that are known to cause gastrointestinal disorders in both healthy people and those who are immunocompromised. In the study by Adeyemo et al., 2019), it required 83.2 mJ/cm^2 to disinfect *Cryptosporidium* to achieve 2-log reduction and 20.8 mJ/cm^2 for disinfection of *Giardia* to achieve again 2 log-reduction both using a wavelength of 254 nm. The study also mentioned that *Cryptosporidium* was resistant to UV irradiation as it needs high UV-dose 83.2 mJ/ cm^2.

Again on the systematic review by (Hazell et al., 2019a), the effect of UV irradiation was investigated for many WASH (water, sanitation and hygiene) related helminth including *Ascaris* species which is responsible for Ascariasis, a diseases that causes an infection on the small intestine. According to the study, fluences of more than 80 mJ/cm^2 were required to achieve a 1-log inactivation, which was more than twice the fluence allowed by certain European countries for the treatment of publicly supplied drinking water. And (Hazell et al., 2019a) come to the

conclusion that UV-disinfection may not be the most effective way of water treatment for these helminths due to the high fluence (UV dose) requirements.

In a chapter review done by (Stanfield et al., 2003) it is mentioned UV disinfection has been shown to be effective in inactivating bacteria and viruses. And usually a dose of 40mJ/cm^2 is accepted as being sufficient for efficient treatment. In the review it was stated that a UV dose of 40 mJ/cm^2 will reduce vegetative bacteria by 4 to 8 logs and Virus inactivation is by 3 to 6 logs. Protozoa are more resistance to UV disinfection, yet for cryptosporidium oocysts at a UV dose of 410J/m^2 (41 mJ/cm^2) log reduction of 4 can be achieved.

In the present study, three wavelengths were tested to examine their effectiveness to inactivate cercariae of schistosomiasis. These wavelengths are 255 nm, 265 and 285 and the two wavelengths showed an ability to disinfect by achieving 1 log reduction at a fluence of 300 mJ/cm^2 for a wavelength of 265n m and 1.2 log-reduction at a fluence of 200 mJ/cm^2 and 300 mJ/cm^2 respectively for a wavelength 285 nm.

When the present results are compared with all the previous studies done on UV irradiation using different pathogenic organisms, the UV-dose (fluence) needed to disinfect cercariae of schistosomiasis is high. Based on the results from the mentioned studies the fluence needed to kill cercariae (300 mJ/cm^2) is high, this might be due to the characteristics of cercariae such as size and number of cells. The length of a *S. mansoni* cercariae is approximately 500 μm, but it contracts and elongates almost continuously and the cercariae of the most studied species (S. mansoni) possesses about 1,000 cells, with numerous cell types, ranging from sensory cells, muscle cells, nerve cells, support cells and others. Many of the organ systems found in the adult worm are already formed, in miniature, in the body of the cercariae (Toledo & Fried, 2014).

5. CONCLUSION AND RECOMMENDATION

5.3. CONCLUSION

From the result acquired above, all the three wavelength showed a change on the number of alive cercariae in the sample when UV- light was exposed. But only the two wavelength 265nm and 285nm showed a 1 and 2 log reduction which is 90% and 99% disinfection of cercariae of schistosomiasis. The last wavelength 285n m showed a high disinfection which is 99% disinfection in another word 2 log-reduction at a very high fluecne that is 300 mJ/cm^2.

The lower the fluence the lower the disinfection of cercariae in all wavelength, therefore the higher disinfection was at 300 mJ/cm^2 at a wavelength of 285 nm and the lowest was at 1 mJ/cm^2 at a wavelength of 255nm. At 285 nm a 1 and 2 log-reduction was achieved at a fluence of 200 mJ/cm^2 and 300 mJ/cm^2 respectively. And at 265nm a 1 log-reduction was achieved at the same 300 mJ/cm^2 fluence. From these the optimum wavelength and fluence can be 285nm and 300 mJ/cm^2 respectively. Which is a very high fluence when compared to fluence needed by other pathogenic organism.

The impact of exposure time was also studied for each wavelength 255nm, 265nm and 285nm and explained in the graph. It was found out that as the wavelength increased the time needed to disinfect cercariae decreased and as fluence and exposure time increased for a specific wavelength the log-reduction also increased.

Therefore, based on this study 99% of cercariae of schistosomiasis can be disinfected at a wavelength of 285 nm using 300 mJ/cm^2 at water temperature of 24^0C and pH 7.2. Yet the fluence needed to disinfect cercariae of schistosomiasis from the sample in this study was 300 mJ/cm^2 which is considered to be high. Therefore UV disinfection using UV-C LEDs for inactivating cercariae of schistosomiasis needs further research considering the high fluence required.

5.4. RECOMMENDATION

The present experiment has been carried out under controlled conditions in a laboratory. Irradiating cercariae-infected water in a Petri dish with bottled water was used in the experiments. As a result, UV fluence observed at the water's surface was considered to be UV fluence reaching the cercariea. When it comes to the field, however, this is not the case since the suspended particles and water absorb the UV light, limiting the depth of UV penetration. Also, when the turbidity and depth of the water rise, the UV fluence required to kill schistosomiasis cercariae rises.

Temperature and pH have no major direct impact on UV performance but UV transmittance is affected by dissolved and particulate matter therefore it is best to apply pre-treatment before applying UV light (Templeton, 1986). As a result, different water quality matrices must be addressed when using UV-LEDs on a small or community scale. If the water quality is poor, such as high turbidity or organic matter content, attention should be given to pre-treatment methods that can be applied at home or on a small scale, such as filtering and sedimentation (Hazell et al., 2019a)

According to the findings of the study, UVC-LEDs can be used as a disinfectant. However, the approach utilized in this study was to count the number of dead cercariae in order to determine the effectiveness of UV-light emitted by UV-LEDs. UV-irradiation typically damages an organism's DNA and RNA, disrupting proper functioning. This means that the organisms might still be alive, but they are damaged and unable to cause disease. As a result, more studies and methodologies are needed to investigate the mechanisms of DNA and RNA damages.

Finally, it is strongly recommended to increase public awareness of this diseases, its transmission route, to provide public bathrooms in endemic regions, and to prevent open defecation and discharge of human and animal waste to surface water bodies.

6. REFERENCE

Adeyemo, F. E., Singh, G., Reddy, P., Bux, F., & Stenström, T. A. (2019). Efficiency of chlorine and UV in the inactivation of Cryptosporidium and Giardia in wastewater. *PLoS ONE*, *14*(5), 1–22. https://doi.org/10.1371/journal.pone.0216040

Aemero, M. (2011). *Genetic Diversity , Multiplicity of Infection and Population Structure of Schistosoma mansoni Isolates of Ethiopia within Human Hosts*. ADDIS ABABA UNIVERSITY.

Akafu, T., Chimdi, A., & Gomoro, K. (2019). Removal of Fluoride from Drinking Water by Sorption Using Diatomite Modified with Aluminum Hydroxide. *Journal of Analytical Methods in Chemistry*, *2019*. https://doi.org/10.1155/2019/4831926

Alebie, G., Erko, B., Aemero, M., & Petros, B. (2014). Epidemiological study on Schistosoma mansoni infection in Sanja area , Amhara region , Ethiopia. *Parasites & Vectors*, 1–10.

Amisalu, G. (2010). *Biomedical science)* (Issue June). ABABA UNIVERSITY.

Artichowicz, W., Luczkiewicz, A., & Sawicki, J. M. (2020). Analysis of the radiation dose in UV-disinfection flow reactors. *Water (Switzerland)*, *12*(1). https://doi.org/10.3390/w12010231

Beck, S. E., Ryu, H., Boczek, L. A., Cashdollar, J. L., Jeanis, K. M., Rosenblum, J. S., Lawal, O. R., & Linden, K. G. (2017). Evaluating UV-C LED disinfection performance and investigating potential dual-wavelength synergy. *Water Research*, *109*, 207–216. https://doi.org/10.1016/j.watres.2016.11.024

Beck, S., & Ph, D. (2016). *UV LED Disinfection 101*. *20*(1), 4–9.

Bolton, J. R., Beck, S., & Linden, K. G. (2016). *Protocol for the determination of fluence (UV dose) using a low-pressure or low-pressure high-output UV lamp in bench- scale collimated beam ultraviolet experiments*. *2003*, 1–9.

Bolton, J. R., Linden, K. G., & Asce, M. (2003). *Standardization of Methods for Fluence „ UV Dose … Determination in Bench-Scale UV Experiments*. *129*(March), 209–215.

Boussinesq, M., Brooker, S. J., Brown, A. S., Buckle, G., Budke, C. M., King, C. H., Lozano, R., Jasrasaria, R., Johns, N. E., Keiser, J., Pion, D. S., Pullan, R. L., & Ramaiah, K. D. (2014). *The Global Burden of Disease Study 2010 : Interpretation and Implications for the Neglected Tropical Diseases*. *8*(7). https://doi.org/10.1371/journal.pntd.0002865

Brady, M. F. J. and F. J. (2016). *The Removal of the Cercariae of Schistosoma mansoni from Water by Filtration through Diatomaceous Silica in a Small Model Filter*. *61*(43), 1538–1543.

Braun, L., Daniel, Y., Sylivester, Dessalegne, M., Zerefa, Maru, M., Allan, F., Zewge, F., Aidan, M. E., Kinung, S., & Templeton, M. R. (2020). *Chlorination of Schistosoma mansoni cercariae*. 1–16. https://doi.org/10.1371/journal.pntd.0008665

Braun, L., Grimes, J. E. T., & Templeton, M. R. (2018). *The effectiveness of water treatment processes against schistosome cercariae : A systematic review*. 1–22. https://doi.org/10.1371/journal.pntd.0006364

Chitsulo, L., Engels, D., Montresor, A., & Savioli, L. (2000). The global status of schistosomiasis and its control. *Acta Tropica*, *77*, 41–51.

Colley, D. G., Bustinduy, A. L., Secor, W. E., & King, C. H. (2014). Human schistosomiasis. *The Lancet*, *6736*(13), 1–12. https://doi.org/10.1016/S0140-6736(13)61949-2

Di Bella, S., Riccardi, N., Giacobbe, D. R., & Luzzati, R. (2018). History of schistosomiasis (bilharziasis) in humans: from Egyptian medical papyri to molecular biology on mummies. *Pathogens and Global Health*, *112*(5), 268–273. https://doi.org/10.1080/20477724.2018.1495357

DWI. (2016). *Guide to the assessment of UV disinfection systems for local authorities*. 1–6. http://dwi.defra.gov.uk/private-water-supply/regs-guidance/Guidance/UV-la.pdf

EPA, U. S. E. P. A. (1999). *wastewater technology fact sheet chlorine disinfcetion*.

EPHI, E. P. H. I., (FMOH), F. M. O. H., Initiative, S. C., & (PCD), P. F. C. D. (2020). *Mapping of schistosomiasis and soil-transmitted helminthiasis in Ethiopia*. 1–11.

Grider, B. (n.d.). *Advantages of Ultraviolet LED Lighting*.

Grimes, J. E. T., Croll, D., Harrison, W. E., Utzinger, J., Freeman, M. C., & Templeton, M. R. (2015). *The roles of water , sanitation and hygiene in reducing schistosomiasis : a review*. 1–16. https://doi.org/10.1186/s13071-015-0766-9

Hailegebriel, T., Nibret, E., & Munshea, A. (2020). Prevalence of Schistosoma mansoni and S. haematobium in Snail Intermediate Hosts in Africa: A Systematic Review and Meta-analysis. *Journal of Tropical Medicine, 2020*. https://doi.org/10.1155/2020/8850840

Hazell, L., Braun, L., & Templeton, M. R. (2019a). *Ultraviolet sensitivity of WASH (water , sanitation , and hygiene) -related helminths : A systematic review*. 1–19.

https://doi.org/10.1371/journal.pntd.0007777

Hazell, L., Braun, L., & Templeton, M. R. (2019b). Ultraviolet sensitivity of WASH (water, sanitation, and hygiene)-related helminths: A systematic review. *PLoS Neglected Tropical Diseases*, *13*(9), 1–19. https://doi.org/10.1371/journal.pntd.0007777

Isabel, J., & Austin, R. (2016). *Ethiopia Prevalence of Schistosomiasis*. 2015–2016.

Kassa, L., Omer, A., Tafesse, W., Taye, T., Kebebew, F., & Beker, A. (2005). *Schistosomiasis*.

King, C. H., & Bertsch, D. (2015). Historical Perspective: Snail Control to Prevent Schistosomiasis. *PLoS Neglected Tropical Diseases*, *9*(4), 2–7. https://doi.org/10.1371/journal.pntd.0003657

Lemma, A. (1970). Laboratory and field evaluation of the molluscicidal properties of Phytolacca dodecandra. *Bulletin of the World Health Organization*, *42*(4), 597–612.

Mohammed, N. A. I., Madsen, H., & Ahmed, A. A. A. R. M. (2016). Types of trematodes infecting freshwater snails found in irrigation canals in the East Nile locality, Khartoum, Sudan. *Infectious Diseases of Poverty*, *5*(1), 1–10. https://doi.org/10.1186/s40249-016-0108-y

Negussu, N., Mengistu, B., Kebede, B., Deribe, K., Ejigu, E., Tadesse, G., Mekete, K., & Sileshi, M. (2017). *Ethiopia Schistosomiasis and Soil-Transmitted Helminthes Control Programme : Progress and Prospects Europe PMC Funders Group Ethiopia Schistosomiasis and Soil-Transmitted Helminthes Control Programme : Progress and Prospects*. *May 2018*.

Nyangaresi, P. O., Qin, Y., Chen, G., Zhang, B., Lu, Y., & Shen, L. (2018). Effects of single and combined UV-LEDs on inactivation and subsequent reactivation of E. coli in water disinfection. *Water Research*, *147*, 331–341. https://doi.org/10.1016/j.watres.2018.10.014

Oliver, J., Biaggi, N., & River, J. (1994). *The effect of Chlorine on the Motility and Infectivity of the Cercariae of Schistosoma Mansoni.pdf*.

Pointier, J. P. (2000). Biological control of the intermediate snail hosts of schistosomiasis in the Caribbean area. *Acta Tropica*, *77*, 53–60.

Ross, A., Inobaya, M., Olveda, R., Chau, T., & Olveda, D. (2014). Prevention and control of schistosomiasis: a current perspective. *Research and Reports in Tropical Medicine*, 65. https://doi.org/10.2147/rrtm.s44274

SANDBACH, F. R. (1973). *THE HISTORY OF SCHISTOSOMIASIS RESEARCH*. 259–275.

Secor, W. E., & Montgomery, S. P. (2015). Something old, something new: Is praziquantel enough for schistosomiasis control? *Future Medicinal Chemistry*, *7*(6), 681–684. https://doi.org/10.4155/fmc.15.9

Sholtes, K. A., Lowe, K., Walters, G. W., Sobsey, M. D., Linden, K. G., & Casanova, L. M. (2016). Comparison of ultraviolet light-emitting diodes and low-pressure mercury-arc lamps for disinfection of water. *Environmental Technology (United Kingdom)*, *37*(17), 2183–2188. https://doi.org/10.1080/09593330.2016.1144798

Song, K., Mohseni, M., & Taghipour, F. (2016). Application of ultraviolet light-emitting diodes (UV-LEDs) for water disinfection: A review. *Water Research*, *94*, 341–349. https://doi.org/10.1016/j.watres.2016.03.003

Stanfield, G., Lechevallier, M., & Snozzi, M. (2003). Treatment efficiency. *Assessing Microbial Safety of Drinking Water: Improving Approaches and Methods*, *9789264099*, 159–178. https://doi.org/10.1787/9789264099470-en

Techincal Manual, P. water supplies. (2019). *Disinfection by UV Radiation Technical Guidance*.

Tefera, A., Belay, T., & Id, M. B. (2020). *Epidemiology of Schistosoma mansoni infection and associated risk factors among school children attending primary schools nearby rivers in Jimma town , an urban setting , Southwest Ethiopia*. 1–15. https://doi.org/10.1371/journal.pone.0228007

Templeton, D. M. (1986). Basic Principles of Uv-Disinfection of Water. *Ozone: Science and Engineering*, *9*(4).

Tian, F., Lin, D., Wu, J., Gao, Y., Zhang, D., Ji, M., & Wu, G. (2010). Immune events associated with high level protection against Schistosoma japonicum infection in pigs immunized with UV-attenuated cercariae. *PLoS ONE*, *5*(10), 1–12. https://doi.org/10.1371/journal.pone.0013408

Tigist, A. (2020). *Efficiency of chlorine disinfection against cercaria removal for schistosomasis control: Optimization of chlorine dose and contact time* (pp. 1–79).

Toledo, R., & Fried, B. (2014). *Digenetic Trematodes*.

Utzinger, J. (2014). Control and Elimination of Neglected Tropical Diseases in Africa. *International Journal of Infectious Diseases 21S*, 1–460. https://doi.org/10.1016/j.ijid.2014.03.502

Wickham, J., Stehman, S., Gass, L., Dewitz, J., Sorenson, D., Granneman, B., Ross, R., & Baer,

L. (2019). Evaluating UV-C LED disinfection performance and investigating potential dual-wavelength synergy. *Advances in Ecological Research*, *60*, 1–24. https://doi.org/10.1016/j.watres.2016.11.024.Evaluating

Workineh, L., Yimer, M., Gelaye, W., & Muleta, D. (2019). The magnitude of Schistosoma mansoni and its associated risk factors among Sebatamit primary school children , rural Bahir Dar , Northwest Ethiopia : a cross - sectional study. *BMC Research Notes*, 1–6. https://doi.org/10.1186/s13104-019-4498-3