INTERRUPTION OF TRANSMISSION OF O. VOLVULUS IN UGANDA

Transmission of Onchocerca volvulus by Simulium neavei in Mount Elgon Focus of Eastern Uganda Has Been Interrupted


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Abstract.

The study determined that Simulium neavei-transmitted onchocerciasis in Mount Elgon onchocerciasis focus had been interrupted. Annual mass treatment with ivermectin changed to two times per year along with vector elimination in 2007. Then, baseline microfilaria (mf) prevalence data of 1994 in five sentinel communities were compared with follow-up data in 2005 and 2011. Blood spots from 3,051 children obtained in 2009 were analyzed for exposure to Onchocerca volvulus immunoglobulin G4 antibodies. Fresh water crab host captures and blackflies collected indicated their infestation with larval stages of S. neavei and presence or absence of the vector, respectively. Mf rates dropped from 62.2% to 0.5%, and 1 (0.03%) of 3,051 children was positive for O. volvulus antibodies. Crab infestation dropped from 41.9% in 2007 to 0%, and S. neavei biting reduced to zero. Both remained zero for the next 3 years, confirming interruption of onchocerciasis transmission, and interventions were halted.

INTRODUCTION.

At the 2002 conference on the eradicability of onchocerciasis, global eradicability of onchocerciasis was deemed impossible using currently available tools (ivermectin and vector control) because of the challenges posed by the infection in Africa.¹ However, permanent interruption of transmission in isolated transmission foci, where feasible in Africa, was recommended, with the goal of
eventual elimination of the parasite and halting of interventions in those foci. The remaining meso- and hyperendemic foci in Africa under the assistance of African Program for Onchocerciasis Control (APOC) were advised to continue annual mass drug administration (MDA) of ivermectin (Mectizan; donated by Merck & Co.) with the goal of reaching a point of high-level control, where the disease resulting from the infection is no longer a public health problem. However, the donation of ivermectin and questions of how long the mass ivermectin treatment programs would be needed in endemic areas have long been a concern to the public health community in endemic African countries, including Uganda.

The immense health challenges of sustaining programs over time given frequent transfers of trained and committed personnel, shifting government policies, donor fatigue, and uncertain long-term political needed to justify long-term investment in the onchocerciasis control program prompted Uganda in 2007 to consider a move from control to an elimination approach. One determinant in this bold decision was the technical strength of Uganda’s Vector Control Division, consisting of dedicated personnel trained by the German GTZ, the Bernhard Nocht Institute for Tropical Medicine. Also, vector elimination efforts in the Itwara focus using focal ground larvicide application with Temephos (Abate) were successful. These findings, together with the Institutional Ministry of Health’s knowledge of the successful elimination of Simulium damnosum from the Victoria Nile focus that liberated 3 million people from the threat of onchocerciasis in 1970s, led to national confidence in declaring a countrywide elimination policy in 2007. In addition, there was, during this time period, evidence in the Americas that high-coverage semiannual MDA of ivermectin might actually kill the adult worms within 6.5 years. This time was a shorter time frame than necessary using annual treatments. The countrywide elimination policy for onchocerciasis used the strategy of a phased, integrated, and flexible program of aggressive mass treatment with ivermectin coupled with local vector elimination where ground larviciding was deemed technically feasible.

The Mount Elgon focus, the only onchocerciasis focus located in the eastern part of Uganda, was one of six Ugandan transmission foci initially targeted for the elimination effort. S. neavei, with larvae that occur in a phoretic association with the fresh water crabs Potomonautes niloticus and P. loveni in this area, are the vectors. Adult S. neavei require heavy forest canopy to survive and thrive.

The first studies of the Mount Elgon focus were conducted in the 1950s, when it was observed that there was a small extension of the focus into Kenya. On the Uganda side of the border, vector control activities against S. neavei were carried out in 1957, largely to protect a proposed coffee research station. The estimated size of Mount Elgon focus in Uganda at the time was 1,500 km², with onchocerciasis mean prevalence of 80%. Vector control activities consisted of 12 applications of DDT (18% e.c.) at 0.5 ppm/30 minutes at 14-day intervals from 140 dosing points, which resulted in a 98% reduction of S. neavei density. However, vector elimination was not achieved. Because of the political upheavals, the plan to launch coffee research in the area was dropped, and additional onchocerciasis control activities were abandoned. However, on the Kenya side, vector control activities in 1950s and early 1960s were successful, and onchocerciasis was eliminated there. The size of the Mount Elgon focus in Uganda in the mid-1960s had reduced to about 500 km² as a result of deforestation. As widespread deforestation continued, the size of Mount Elgon focus was determined to be only 250 km² (unpublished survey by the Uganda National Onchocerciasis Control Program, 1994).

Onchocerciasis control was relaunched in Mount Elgon in 1994 and continued for 18 years. This paper reports treatment and assessment activities conducted in support of the conclusion that onchocerca volvulus transmission was interrupted in Mount Elgon, and interventions were halted at the end of 2011.
METHODS.

History of recent assessments and treatment activities in the focus.

By 1994, the Mount Elgon focus was comprised of parts of Mbale and Sironko districts; subsequently, two more districts (Bududa and Manafua) were created from Mbale and Sironko districts (Figure 1). Rapid epidemiological assessment (REA) based on community nodule prevalence was done in 1994 in Mount Elgon to target ivermectin MDA. Samples of 30 resident adults ages 20 years and above who had lived in the community for at least 10 years were assessed for nodules. Annual mass treatment was launched in 1994 through community based treatment. In 1998, communities were empowered to make their own decisions under the community-directed treatment with Ivermectin (CDTI) approach.

Under the new 2007 elimination policy, the CDTI strategy continued to be used, but the frequency of ivermectin distribution was changed from one to two times per year, and there was an increase in the geographic scope of treatments to include hypoendemic areas (communities with nodule rates below 20%) that had not been included in mass treatment. Two times per year treatment was conducted for 5 years (2007–2011).

Treatment coverage was calculated by dividing treatments reported by the community-directed distributors (CDDs) by the total of all eligible persons (i.e., population minus children < 5 years who should not be treated with ivermectin) or the ultimate treatment goal (UTG) residing in all the at-risk communities within the onchocerciasis-endemic area. The entire expanded focus (including hypoendemic communities) had a combined population of about 328,862 people in 820 communities. The denominator (UTG) used to calculate coverage in this report includes the hypoendemic communities. Treatments in the Mount Elgon focus had been restricted to the priority meso- and hyperendemic areas from 1994 to 2006. As a result, the treatment coverage for the period for annual treatment could not reach the goal of 90% treatment coverage, because residents of hypoendemic areas were not included until 2007. With institution of two times per year treatment in 2007, the UTG under annual treatment doubled (hence, UTG2, which is number of treatments but not individual persons).

National criteria for interruption of transmission.

As part of the new elimination policy, the Ministry of Health (MOH) partnered with the Uganda Onchocerciasis Elimination Expert Advisory Committee (UOEEAC) in 2008. As a first task, the UOEEAC recommended national criteria for making decisions on where and when to halt interventions. The guidelines developed were based on a detailed review of the 2001 World Health Organization (WHO) criteria for certification of elimination of onchocerciasis, TDR studies, and guidelines used by the Onchocerciasis Elimination Program for the Americas. The Uganda criteria include three indicators.

Parasitological assessments.

Microfilaria (mf) prevalence in skin snips must be less than 5% in all sampled communities and less than 1% in 90% of sampled communities.

Serological assessments.

Infection rates in children must be < 0.1%. The prevalence of immunoglobulin G4 (IgG4) antibodies to Ov-16, a recombinant antigen of Onchocerca volvulus, was used as a measure of infection/exposure in children. A sample size of 3,000 children is needed to exclude 0.1% antibody prevalence with 95% confidence.
Entomological assessments.

In the *S. neavei* foci, there should be a lack of positive crabs for larvae/pupae of *S. neavei* species in a series of surveys and absence of *S. neavei* collected in a defined focus over a period of 3 year.5


Baseline (pretreatment) mf rates were collected in four high-risk communities (determined based on high nodule rates from the wider 1994 nodule surveys described by Ndyomugeynyi28 in 1998). These results were compared with those results collected in the 2005 and 2011 follow-up surveys that used similar methods, with the exception that the 2011 survey added a fifth community. Another variant in the 2011 follow-up survey was that 87 children less than 10 years of age were examined from Buriri (8), Bubungi (27), Bunabutiti (22), Bunabatsu (15), and Bukikoso (15) communities. Two skin snips were taken from the iliac crest posteriorly for every selected adult.29 Skin snips were read under low (x40) magnification after a 12-hour incubation at ambient temperature in normal saline solution. The mf in individual skin snips were not quantified but recorded as positive or negative, and the prevalence was expressed as a percentage.

Assessments in 2010.

Serological assessments.

Children were randomly selected from 17 communities from four districts comprising the Mount Elgon focus. Each district was proportionally assigned sampling numbers based on population sizes from census recording in the community household register of 2008. Also, proportional sampling was applied at low levels of administrative structures, such as the subcounty and parish levels below the district level. The proportion of children selected for assessment by district was 23.2% (812) from Sironko District, 19.7% (689) from Mbale District, 10.9% (381) from Manafwa District, and 46.2% (1,617) from Bududa District. Age group samples sought were also proportionally allocated: under 5 years = 1,500 (42.8%), ≥ 5 to ≤ 10 years = 1,000 (28.6%), and > 10 to < 14 years = 1,000 (28.6%).

Sterile procedures were used to collect blood spots on Whatman No. 2 filter paper (Sigma). The blood samples were dried, separated by sheets of paper, systematically bundled, and stored in plastic bags in a cooler until they were returned to the laboratory and stored at 4°C before being processed for analysis. Sera were eluted from the dried spots and examined for the presence of Ov-16 IgG4 antibodies by enzyme-linked immunosorbent assay (ELISA) as previously described.25

Entomological activities.

Mapping of vector breeding sites and their upper elevation breeding limits began in March of 2007. Fresh water crabs (*P. niloticus* and *P. loveni*) in all the rivers in this focus were trapped and examined for infestation with immature stages of *S. neavei* in 2007 to determine the magnitude of crab infestation.5 There are over 20 rivers and tributary streams in the focus, but only the three major systems that had infested crabs with the immature stages of *S. neavei* were targeted for vector elimination: the Namatala, the Namufumbulo, and the Tsutsu and their tributaries (Figure 2, river system). Sironko river system was found unsuitable for crabs and subsequently, the breeding of *S. neavei*, because the surrounding forest had been destroyed (Figure 1).5, 30 Breeding of *S. neavei* was thought to be confined to the zone below 1,350 m, although adult *S. neavei* have been caught at altitudes as high as 2 100 m. Fresh water crabs were caught using locally designed funnel-shaped basket traps baited with fresh meat. They were left in rivers
for at least 1 hour and then immediately examined for crabs carrying larvae and pupae of *S. neavei*. The crabs carrying young stages of *S. neavei* were counted, and infestation rate was expressed as a percentage of the total catch. Crabs were returned to the stream immediately after examination. Crab monitoring continued one time per month until October of 2011.

**Adult S. neavei collection.**

Adult *S. neavei* collection following an MOH protocol based on WHO standards for full-day human landing catches of *S. neavei* (0700–1800 hours) were established at four catching sites: one on the Namufumbilo river system, two on the Namatala river system, and one on the Tsutsu river system (Figure 2). Female *S. neavei* seeking a blood meal settled on exposed legs of local collectors, who collected them in tubes before they bit. *S. neavei* collection was 2 days/week and 8 days/month until June of 2011, and the captures were preserved in ethanol. In a small study conducted in 2007, captured *S. neavei* were dissected to determine parous and infection rates before launching two times per year ivermectin treatments. Infection rate was defined as the proportion of *S. neavei* with all larval stages (L1, L2, and L3).

**Larviciding.**

Larviciding with temephos was conducted and supervised by expert MOH vector control teams at all sites where infested crabs were observed. First insecticide carry trials were carried out at 27 dosing points from October to November of 2007. During this period, river gauging to establish the discharge and determine dosing and booster points was done. Temephos was applied at a rate of 0.2–0.4 mg/L to reach a concentration of 0.1–0.3 ppm. The insecticide was pre-mixed in a 15-L knapsack sprayer and applied for a period of 30 minutes at established dosing points. Larviciding commenced in January of 2008 in all three river systems and continued for the next 12 months initially at 4-week intervals for 6 months and 8-week intervals thereafter until March of 2009. All 41 key dosing points plus an additional 6 booster points on these river systems were used in the first 6 months. Treatment at the booster points was discontinued when the program moved to 8-week treatments. The impact of the larviciding on *S. neavei* immature stages was assessed after 48 hours. When larviciding was stopped, surveillance through crab infestation and *S. neavei* collection continued through October of 2011 and June of 2011, respectively.

The MOH personnel also monitored crab populations in eight rivers where ground larviciding was not done in July of 2007, June of 2010, and June of 2011.

**Data analysis.**

Parasitological data from adults and children and serological data from children were entered and analyzed in Microsoft Excel. Where relevant, the data were entered and analyzed in Epi Info (CDC, Atlanta, GA) for a χ2 test of independence. The entomological data were also entered, analyzed, and graphically illustrated in Microsoft Excel.

**Ethical approval of the study protocols.**

Parasitological, serological, and entomological evaluations were approved by the MOH of Uganda. The Emory Institutional Review Board (11 438) classified the assessment activities as periodical program performance assessment (non-research). In parasitological and serological epidemiology studies, consent was obtained from all participants, and verbal assent was obtained from the parents of young children. All participating communities were educated about the importance of evaluations, and participants were assured that there would be no repercussions for refusing to participate. The *S. neavei* collectors were at least 20 years of age and provided written informed consent after they were informed of the nature of the work and told that they could opt out of the study if they so wished at any time without any repercussions.
RESULTS.

Mass treatment.

When mass treatments began in 1994, only 18,678 persons were treated. Treatments grew to 193,858 by 2006, the last year of annual treatment in hyper- and mesoendemic areas of Mount Elgon. When hypoendemic areas were added, treatments grew to over 270,000 treatments per round by 2011 (Figure 3). In terms of UTG coverage, the program reached no more than 73.5% in the period of 1994–2006. Beginning in 2007, when two times per year treatment was launched and hypoendemic areas were included, communities attained 90% of UTG in every treatment round every year from 2007 to 2011 (Figure 4).

Parasitological assessments.

Among adults, baseline skin mf prevalence fell by more than 95% from 62.2% (N = 320) in 1994 to 1.5% (N = 528) in the 2005 follow-up assessments (P < 0.0001) and 0.5% (N = 442) in the 2011 follow-up assessments. However, the two adults who tested mf-positive during 2011 were more than 70 years old (Table 1). The prevalence of palpable nodules also decreased by more than 95% from 59.2% (N = 120) at baseline assessments in 1994 to 3.2% in the 2005 follow-up assessments (P < 0.0001) and 1.4% (N = 442) in the 2011 follow-up assessments (Table 2). All 87 children (≤ 10 years of age) assessed in 2011 were negative for both mf and palpable nodules, indicating no recent transmission in the communities.

Serological assessments in children with Ov-16 antibody ELISA.

In total, 3,051 blood spots were obtained, exceeding the sampling scheme goals for the younger age group (1,113, under 5 years; 1,032, ≥ 5 to ≤ 10 years) but not reaching the goal for the group of > 10 to < 15 years (906). Only one older child in the > 10 to < 14 years age group tested positive (0.03% with upper 95% confidence interval [95% CI] = 0.128%) (Table 3). By age groups, the results in children under 5 years were 0% (upper 95% CI = 0.172%), > 5 and ≤ 10 years were 0% (upper 95% CI = 0.185%), and > 10 and ≤ 14 years were 0.11% (upper 95% CI = 0.430%). In the 10 years and under cohort (N = 2,145), the rate was 0% (upper 95% CI = 0.089%).

Entomological assessments.

Monitoring of fresh water crabs.

The infestation level of about 41.9% (range = 47–59%) from April to December of 2007 declined to 27.7% (range = 15.6–34.6%) from January to April of 2008 with the commencement of larviciding. After April of 2008, the decline was dramatic, and the last infested crab was captured in August of 2008. Thereafter, no infested crab with larval stages of *S. neavei* was captured for the next 3 years (Figure 5). However, a progressive decline in the number of crabs captured to almost extinction during monitoring was noted, even after larviciding was halted in March of 2009. Monitoring of crab populations in eight river systems where no larviciding was done revealed a similar trend, with 698 crabs being capture in 2007; then, in June of 2010, only 43 crabs were capture, whereas in July of 2011, no crab was captured (Table 4).
S. neavei *adult fly collection.*

*S. neavei* collections from April of 2007 show a year-round biting pattern, with a peak biting period between June and October. With the commencement of larviciding in January of 2008, a progressive decline in *S. neavei* collection was observed, and the last *S. neavei* were collected in June of 2008. The mean biting rate in 2007 of five *S. neavei* per man-hour had quickly been reduced to zero after commencement of ground larviciding. The vector population did not recover for a period of 3 years since July of 2008 (Figure 6). The very limited dissections in 2006 of 57 parous *S. neavei* showed an infection rate of 7.0%.

**DISCUSSION.**

In the Mount Elgon onchocerciasis focus in eastern Uganda, the human adult infection rates in sentinel communities in 2011 averaged below 1%, with only one community being above 1% (1.5% in Buriri). Children under 10 years of age from 17 communities selected from throughout the focus in 2010 show no serological evidence of recent infection or exposure to onchocerciasis. The last crab with larval stages of *S. neavei* was captured in August of 2008, and no infested crab has been captured since that time. Also, no *S. neavei* was collected for 3 years between July of 2008 and July of 2011. Larviciding halted in March of 2009, and therefore, it seems that the *S. neavei* population is incapable of recovering.

Absence of the vector (as far as we were able to measure) from the Mount Elgon focus means that onchocerciasis transmission has been interrupted. The Ugandan entomological and epidemiological criteria for interruption of transmission have been met.

In the Mount Elgon focus, communities using the CDTI approach showed that they are capable of distributing ivermectin two times per year while maintaining treatment coverage exceeding 90%. The capacity of CDTI to incorporate (without collapsing) a 6-month treatment strategy in Africa has been debated (Boatin and Richards, unpublished data). This study, however, is the second report from Uganda that documents the ability of a CDTI program to rapidly expand from annual to 6-month treatments (the first being from Wadalai21). The debate about two times per year treatment should move away from asking the research question about whether CDTI can tolerate two times per year to an operational/epidemiological question of when and where two times per year treatment needs to be implemented to advance the elimination agenda.

At its fourth meeting in August of 2011, the UOEEAC reviewed and discussed the Mount Elgon focus survey data with respect to national parasitological, serological, and entomological elimination criteria.22 The UOEEAC concluded that the Mount Elgon focus had met the Ugandan national criteria for transmission interruption and recommended to the MOH that interventions be halted. Subsequently, the MOH’s National Certification Committee accepted the UOEEAC recommendation, and in 2012, ivermectin mass treatments in Mount Elgon were halted. If, during a 3-year post-treatment surveillance (PTS) phase, no evidence for renewed transmission is noted, the focus could be declared free of onchocerciasis.21–25

The 2009 serological results from the Ov-16 antibody survey among children were discussed by the UOEEAC at length. The national criteria call for < 0.1% seroprevalence, and although the serosurvey found a prevalence of 0.03%, the upper bound of the 95% CI was just above 0.1% (at 0.128%). The UOEEAC concluded, however, that the one positive child was in an age group that likely represented an older transmission event, given the absence of infection in younger children. The UOEEAC unanimously recommended to stop interventions in Mount Elgon, despite the one positive child in the survey, while also urging any subsequent serosurveys to sample only children less than 10 years of age so as to avoid this problem in future.24,33 The national guidelines now require the seroprevalence of children under 10 years of age to be less than 0.1%.21
It was evident that deforestation rendered a large area unsuitable for *S. neavei* breeding and survival, resulting in progressive shrinking of the Mount Elgon focus from 1,500 km² in the 1950s to the current 250 km². In fact, tree felling and bush clearing was used in the 1940s and early 1950s successfully to control *S. neavei* in the Nyanza region of Kenya. Deforestation is an important factor in reducing onchocerciasis in areas where the sole vector is *S. neavei*. Deforestation was exacerbated by the population pressure on land (currently at about 1,315 persons/km²) in and around Mount Elgon. There was also evidence of declining crab host populations of *P. niloticus* and *P. loven*. Previous studies in western Uganda had implicated deforestation as a major factor driving the decline of the fresh water crab population. Our surveys conducted from 2007 to 2011 documented a dramatic decline in crab populations in both treated and untreated rivers, indicating that the decline that we observed was not related to the larviciding with temephos to control the vector population. On reflection, deforestation in the Mount Elgon focus may have also contributed to the interruption of transmission of onchocerciasis.

**CONCLUSION.**

Transmission interruption in Mount Elgon validates the Ugandan MOH decision in 2007 to adopt an aggressive tactical approach to onchocerciasis elimination, including semiannual mass treatment of the afflicted communities with ivermectin and vector elimination/control efforts where feasible. Onchocerciasis elimination as a strategy should mean the reconfiguration of static onchocerciasis control programs to more flexible ones in an attempt to speed up the elimination process, and it should convey to all partners that elimination means that it can no longer be business as usual.

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FIGURE 1. Map of Mount Elgon onchocerciasis focus of eastern Uganda.

FIGURE 2. Gauging and dosing points as well as fly-catching sites on river systems where S. neavei breeds in Mount Elgon focus.


FIGURE 4. History of percent treatment coverage with ivermectin annually from 1994 to 2005 (UTG) and two times per year (UTG2) from 2006 to 2011 in Mount Elgon onchocerciasis focus in Uganda.

FIGURE 5. Trend of crab infestation from April of 2007 to September of 2011.


Tables:

TABLE 1. Trend of mf (microscopy) prevalence among adults (20 years and above) from 1994 baseline (N = 320) to follow-up surveys in the same communities in 2005 (N = 528) and 2011 (N = 442)

TABLE 2. Trend of nodule prevalence among adults (20 years and above) from 1994 baseline (N = 120) to follow-up surveys in the same communities in 2005 (N = 528) and 2011 (N = 442)

TABLE 3. Prevalence of O. volvulus Ov-16 IgG4 antibodies in children 1 to 14 years of age (N = 3,051) in 2010 by district and age groups in Mount Elgon onchocerciasis focus

TABLE 4. A record of fresh water crab captures in 2007, 2010, and 2011 in Mount Elgon focus in rivers where larviciding with temephos (Abate) was not done

*All Figures and Tables can be found in the online PDF of the article. The latest version is available here: http://www.ajtmh.org/content/early/2014/03/27/ajtmh.13-0501.full.pdf+html