FIELD USE OF MOLLUSCICIDES IN SCHISTOSOMIASIS CONTROL PROGRAMMES

AN OPERATIONAL MANUAL FOR PROGRAMME MANAGERS

World Health Organization

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Abbrevations

GIS	geographical information system
GPS	global positioning system
FTA	fast technology for analysis (a paper filter used to enhance the preservation of nucleic acids)
DP	dry powder
EC	emulsifiable concentrate
GR	granule formulation
MDA	mass drug administration
SC	suspension concentrate
WP	wettable powder
WHOPES	WHO Pesticide Evaluation Scheme
WHO	World Health Organization

1. Introduction

Schistosomiasis, a parasitic disease caused by trematode worms, affects approximately 260 million people worldwide; more than 90% of cases occur in the WHO African Region (WHO, 2016). Schistosome parasites have complex life cycles entailing both mammalian hosts, including humans, and freshwater snails. The infection cannot be transmitted directly from person to person; it requires fresh water where the intermediate snail host species live and breed. Hence, snails are a crucial and necessary part of the life cycle that schistosomes depend upon for their development, multiplication and transmission. The 260 million individuals affected by schistosomiasis share in common that all have been exposed in freshwater to infective schistosome cercariae that have developed and emerged from snails. It follows that the distribution of the disease reflects in part the distribution of the genera and species of intermediate snail hosts that are compatible with the parasite. Without the presence of compatible snails, there can be no transmission; therefore much attention has focused on breaking the human–snail–human transmission cycle. In addition to snails, lack of adequate sanitation and clean water and, more importantly, human behaviour play an important role in the transmission of schistosomiasis through the release of eggs in the environment by infected people and contact with contaminated water.

Snail control, mainly by molluscicide but also by environmental modifications (Lardans and Dissous, 1998), was for many years the cornerstone of schistosomiasis control (WHO, 1992) and has contributed to many successful control outcomes (Rollinson et al., 2013; King et al., 2015). More recently, the focus of control has moved to anti-schistosomiasis chemotherapy, primarily to prevent morbidity in school-age children, who are often associated with the highest levels of schistosome infection. Regular treatment with praziquantel is now being successfully implemented in at-risk areas in the African Region, the Region of the Americas, the Eastern Mediterranean Region, the Western Pacific Region and the South-East Asia Region. Good overall progress is being made in the control of morbidity attributable to the disease in many parts of the world. However, chemotherapy alone rarely stops transmission of the causative parasite, and additional interventions must be integrated as programmes aim to reduce reinfection, lower prevalence and move towards elimination (Sokolow et al., 2016). While the attention and resources focused on anti- schistosomiasis chemotherapy over the past two decades have yielded many benefits, the focus on chemotherapy in Africa has perhaps hindered the development of new approaches for snail control and, consequently, led to a general decline in global malacological expertise.

The World Health Assembly has affirmed the feasibility of eliminating schistosomiasis from some Member States. The Sixty-fifth World Health Assembly in 2012 adopted resolution WHA65.21 on the elimination of schistosomiasis, urging Member States to take advantage of other ongoing activities to intensify schistosomiasis control programmes and to initiate elimination campaigns where appropriate, through strengthened health systems, preventive chemotherapy, provision of water and sanitation, as well as hygiene education and snail control.

Recent systematic reviews and meta-analysis of the impact of chemical-based molluscicide (King et al., 2015, Sokolow et al., 2016) have concluded that regular molluscicide is likely to contribute significantly towards elimination of schistosomiasis in high-risk areas. The WHO roadmap's new focus on "transmission control, wherever possible" (WHO, 2012a) reinforces the need to promote intermediate- host snail control to prevent schistosomiasis transmission. Hence, it is timely to reconsider snail control practices and to revisit procedures and guidelines.
This operational manual is intended to facilitate the reintroduction of practices and protocols for use of molluscicides in the field in schistosomiasis control programmes. It is complemented by guidelines on the laboratory and field testing of the efficacy of molluscicides for schistosomiasis control (WHO, 2017 [in preparation]).

The purpose of the manual is to assist programme managers with the introduction and implementation of a snail control intervention as part of an integrated schistosomiasis control and elimination program me, and as promoted by WHO's integrated vector management approach (WHO, 2012b).

Chemical snail control operations may be broken down into three phases:

- Phase 1: planning
- Phase 2: intervention
- Phase 3: monitoring and evaluation

2. Background on snail control

Snail control can be done using molluscicides and environmental or biological measures (McCullough et al., 1980; Lardans and Dissous, 1998; Sokolow et al., 2015, 2016). Molluscicides are chemicals or plant extracts (Mott, 1987) used to kill snails. Amphibious snail species especially can be controlled through environmental control because of the nature of their habitats. Such measures include stream channelization, seepage control, canal lining, canal relocation with deep burial of snails, proper drainage in irrigation schemes, vegetation removal, earth filling, ponding and improved agricultural practices. In irrigation schemes or aquaculture ponds, aquatic species especially may be controlled also by habitat modification, for example removal of aquatic vegetation, increased current speed in irrigation canals, prolonged drying of canals if possible, mud removal from ponds and others, to reduce snail density. Biological control may work under some circumstances.

Although many animal species belonging to a wide range of taxa may feed on snails, there are relatively few oligophagous snail predators; these are mainly some leech species, larvae of the family Sciomyzidae and some fish species. There has been much focus on the use of snail competitors, but generally these are introduced species and their use may therefore not be acceptable ecologically given the risk that some species may become invasive. Also, potential predators should be from within the major catchment area of the sites where used; recent studies on snail control using predators have focused on fish and some crustacean species (Sokolow et al., 2015).

3. Phase 1: Planning a snail control programme using molluscicides

3.1 Secure national regulatory approval for use of molluscicides

Molluscicides are chemicals or plant extracts that are used to control snails. They may be of synthetic or natural origin and indeed many plant molluscicides have been investigated (Mott, 1987). The control of freshwater snails requires molluscicides to be added to freshwater bodies (or on dried mud surfaces) that are frequently used by people and their livestock for everyday activities or for agriculture. As such, it is essential that molluscicides have undergone the necessary testing and evaluation for public health use. The WHO Pesticide Evaluation Scheme (WHOPES), set up in 1960, is the international programme that promotes and coordinates such evaluations. Testing of molluscicides is important because products are expected to meet their specifications, be effective and pose no unexpected risks in use.

Before using a molluscicide product, ensure that the product is registered with the national regulatory body. Regulatory authorities should be consulted before implementing snail control programmes. Currently, niclosamide is the only compound recommended by WHOPES for use as a molluscicide: it has also been approved, in a different formulation, as an anthelmintic drug for treatment of cestode infections, but this is beyond the scope of this manual. Many other compounds, both synthetic and of plant origin, have been used as molluscicides but do not have WHOPES approval; some products have regulatory approval for use in different countries.

3.2 Integrate snail control interventions into schistosomiasis control programmes

WHO promotes a strategy of focal or seasonal application of molluscicides rather than blanket application, since the latter is not only costly but also might cause more damage to the environment. The main objective of using molluscicides is to contribute – in combination with other interventions – to a significant reduction in transmission by reducing the snail population. A particular case is represented by transmission hot spots where several rounds of mass drug administration have not led to a significant decrease of the infection prevalence.

Other objectives include elimination of a newly introduced intermediate host snail in areas where it did not previously exist, or total elimination of snails from known transmission sites in low transmission areas to support interruption of transmission.
Snail control should be part of an integrated schistosomiasis control programme whose activities include preventive chemotherapy using praziquantel, behavioural and educational interventions, and improvements in the availability of safe water and sanitation. Data on

disease prevalence may already have been collected to locate areas with a known schistosomiasis problem; these data should be consulted for preparing risk maps of contact with water bodies by high-risk groups. Spatial distribution data may also be available to guide the intervention (Lai et al., 2015). If snail surveys have not been conducted recently, the data on disease mapping will be the primary lead as to snail location before field surveys are conducted to identify potential transmission sites.

The snail–parasite relationship is very specific. Only certain snail species primarily belonging to the genera *Bulinus*, *Biomphalaria*, *Oncomelania* and *Neotricula* play a role in transmission; certain species within each genus are responsible for transmission of a different schistosome species (Table 1 ; Figure 1). An initial step in the planning process is to gather information on the identity and distribution of potential intermediate snail hosts. This can be achieved by consulting the scientific literature and local reports on transmission of schistosomiasis in the endemic area. Local knowledge and expertise about the snails responsible for transmitting the disease can be useful and efforts should be made to consult those with previous experience.

Table 1. Schistosome species and their intermediate snail hosts

Schistosome species infecting humans	Snail genus responsible for transmission
S. mansoni	*Biomphalaria*
S. haematobium, S. intercalatum, S. guineensis	*Bulinus*
S. japonicum	*Oncomelania*
S. mekongi	*Neotricula*

Freshwater snails may be distributed extensively across the country and be found in various habitats ranging from ponds, marshland, streams and irrigation canals to rivers and large lakes. It is important to be able to distil this information in order to focus snail control where it will be most effective. Consulting maps with geospatial data produced by geographical information systems (GIS) to locate water bodies associated with risk of infection will help to identify areas likely to be of interest.

The overall objective of the intervention must be to control snails in those places where people are in regular contact with water and where transmission is likely to occur. The "focal application" of molluscicide is the preferred approach as it minimizes the use of molluscicides in known transmission areas. Indeed, it is well recognized that schistosomiasis is a focal disease, and the prevalence and intensity of the infection can change dramatically from village to village.

In small water bodies, temporary elimination of the intermediate snail hosts may be feasible but this would be an impossible and unnecessary objective in larger water bodies. Indeed, molluscicide is for the most part not suitable for large water bodies. For example, it is not a feasible approach to control *Neotricula* spp., which transmit *S. mekongi* in certain locations in the large Mekong River. Previous experience suggests that large-scale or blan-

ket mollusciciding approaches as have been conducted in Egypt, Ghana and the Islamic Republic of Iran can be expensive, wasteful and ecologically unsound (Klumpp and Chu, 1987) whereas focal mollusciciding can be a cost–effective method in a variety of habitats. Snails have a high reproductive capacity and it is anticipated that populations will re-establish after focal mollusciciding; in most habitats, therefore, repeated molluscicide treatments will be required.

Snail control also increases awareness of the disease within the community and offers opportunities to reinforce health education interventions alongside preventive chemotherapy.

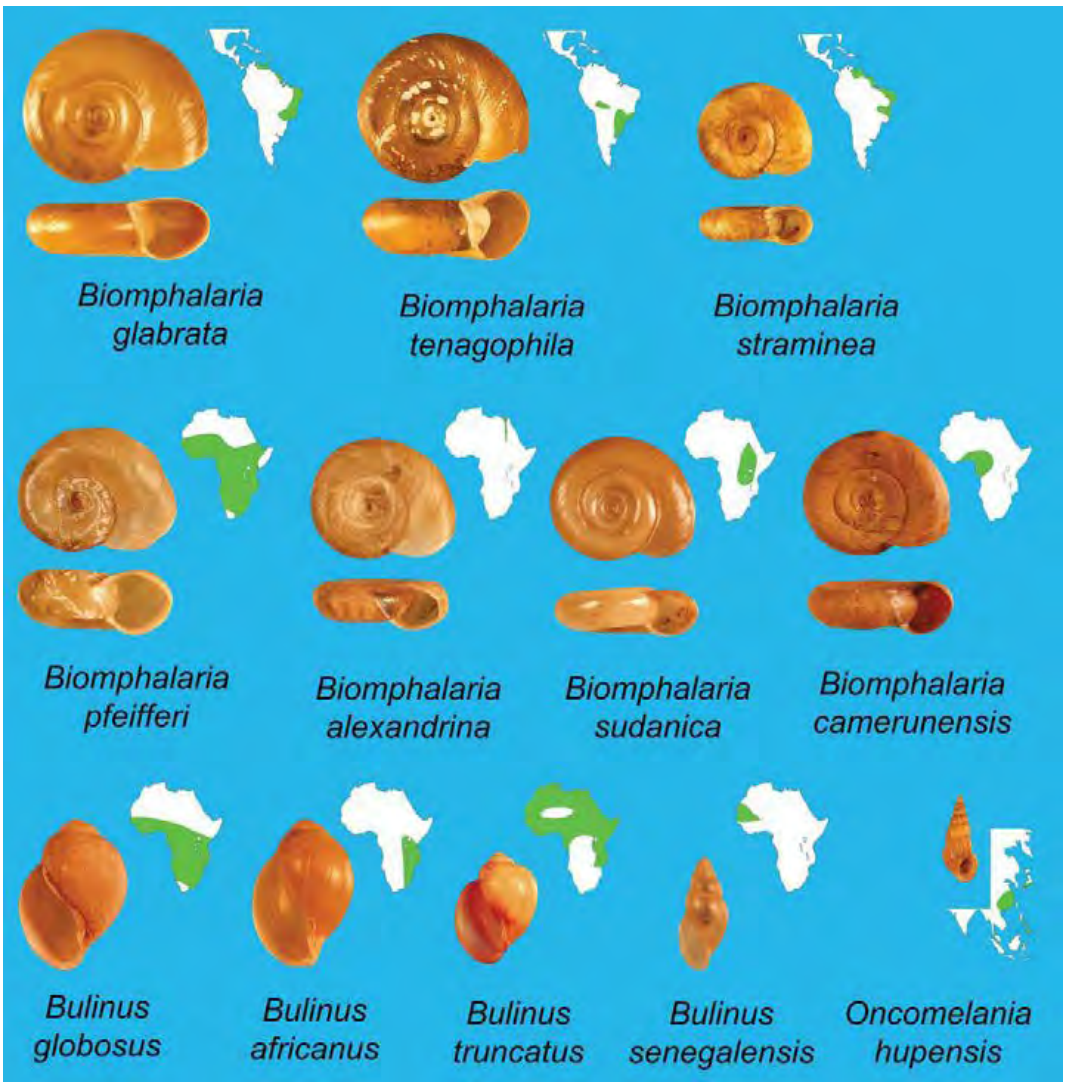


Figure 1. Intermediate snail host species of major global importance (adapted by Dr H Madsen from World Health Organization (1989) slide set: The intermediate snail hosts of African schistosomiasis. Taxonomy, ecology and control)

3.3 Establish a team responsible for snail control

There is a need to establish expertise and responsibilities within the team in snail sampling, snail identification, parasite screening in the laboratory, habitat monitoring, molluscicide application, and data collection and reporting.

Create a national vector control team or, if such a team already exists in the country, ensure that this team has expertise on snail control. Additional malacology training should be provided to the team in the framework of integrated vector management.

Many national disease control programmes will already have personnel familiar with the control of insect vectors of disease (mosquitoes, blackflies, sandflies) and other pests. It would benefit the schistosomiasis control programme if links could be made with other programmes and expertise shared, particularly in relation to spraying of chemical formulations. Application equipment is usually simple and cheap and can often be shared between vector control programmes.

A national coordinator for vector/snail control should be responsible for overseeing the activity. The person should be familiar with vector control and, ideally, have postgraduate and/or technical training. Their supervisor would be responsible for the overall running of the programme, including logistics and scheduling, staff finance, equipment and procurement and data handling.

Depending on the scale of the operation, snail teams should be established in subnational administrative regions. Snail teams will vary in size and number but it is recommended that each team include a supervisor, technical staff, a driver and a member of the community. Two or more support workers should be responsible for applications in the field and should have a technical background, ideally with experience of vector control. At least one trained technician will be required in the laboratory for handling of snails and checking for cercarial shedding.

The snail sampling and molluscicide application process can be demanding due to heavy and bulky equipment and the often "off-road" location of many transmission sites. Snail control will need to be deployed regularly and the operation, once set up, should be sustainable.

3.4 Build capacity of personnel involved in snail control

Teams should receive instruction relating to the biology and life-cycle of schistosomes, especially the essential role that snails play in transmitting schistosomiasis. Teams will need to be aware of how to interact with the local communities in order to explain their presence and the expected impact of the snail control intervention. Training will be required concerning the application of molluscicide, including the different methods available, how to achieve the correct concentration of molluscicide in the volume of water to be treated and on the monitoring of the effect of the molluscicide on snail populations.

Technical capacity of national and district level managers in risk assessment, programme planning, implementation, and monitoring and evaluation should be built. Training should take place centrally and ideally occur over 3–4 days with some field and hands-on laboratory practice.

A general training programme should include:

- Pre- and post- training examination
- Introduction to schistosomiasis and its importance
- Schistosomiasis control and integrating snail control interventions
- Understanding the schistosome life cycle
- The snails involved in transmission – biology and habitat preferences
- Snail identification
- Checking snails for infection
- What is a molluscicide?
- What are the risks and benefits of molluscicides?
- Available molluscicides for snail control
- Determining the best time to carry out snail control in the endemic area under study (coordinating with mass chemotherapy and any seasonality of transmission)
- Methods of application
- Calculating concentrations of molluscicide in different water bodies.
- Focal applications at known transmission spots rather than blanket approaches
- Field application – identifying all water bodies and human water contact points, planning and developing a schedule of activity
- Monitoring the site for snails before and after application – include the use of sentinel snails to determine efficiency of the molluscicide application
- Collecting cercariae on fast technology for analysis (FTA) cards for future analysis
- Keeping standardized and regular records of application
- Establishing good communication with the community
- Health and safety procedures associated with snail sampling and molluscicide application
- How to deal with potential problems reported by the local community – reports of dead fish, dead ducks, crop failure, etc.

3.5 Select molluscicide intervention sites

Molluscicides may be used during a control programme in a range of prevalence settings including:

- In “hot spots” (areas showing continued high prevalence and intensity of infection despite regular chemotherapy).
- When cases of schistosomiasis are identified in new areas not normally associated

with transmission or disease; a rapid snail control response would be justified to contain the infection and limit further spread.

- When elimination is the goal, molluscicide should be an integrated part of the control interventions and applications may need to be increased.

Three general strategies for snail control are in current use: focal, area, and radius control. The first approach may be valuable where transmission is limited to particular foci. It is adopted whenever stringent priorities have to be followed. This method restricts the application of the molluscicide to that "station" (a stretch of 500 m of the stream) in which infected snails are discovered (Ayad, 1976; Jordan and Webbe, 1982). Focal control thus depends upon knowledge of the transmission foci and periodic molluscicide must be continued. Transmission control based upon the essential focality of transmission is being successfully executed by molluscicide and surveillance. This highly cost–effective approach based upon accurate knowledge of human–water contact patterns may result in a considerable saving of expensive chemicals. It must be realized, however, that focal transmission control may be totally impracticable in a flowing water system with a high population density and diffuse domestic and occupational human water contact (Jordan and Webbe, 1982).

Area control or generalized control is likely to be the only practical approach if transmission is widespread in a watershed or irrigation system. In this approach, all snail habitats must be treated and initially it may prove more difficult than focal control. Snail control by periodic, area-wide molluscicide is now being successfully carried out in several major control programmes based upon irrigation and controlled water management. Area control has been restricted in Egypt, for budgetary reasons, to isolated sites, areas of land reclamation and areas converted from basin (flood) irrigation to perennial irrigation (Ayad, 1976). Arial application of molluscicides has been carried out in different endemic areas with varying degrees of success but apparently with high cost– effectiveness in Sudan (Barnish, 1970; Amin and Fenwick, 1977).

Radius control is followed in most other areas. This approach treats all snail-infested water bodies within a 500 m radius from the periphery of human settlements, because it is within that radius that the most frequent association between humans and snails occurs (Ayad, 1976).

Field visits should be made to familiarize the team with the range of water bodies and water contact sites in the endemic area. Discussions with village and community leaders to identify all existing water bodies and human–water contact points will be required. This is a good time to make contact with any environmental health or water committees and discuss with them the forthcoming plans for snail control.

Focal application of molluscicides will be the general rule and will be restricted to places that are commonly used for swimming, bathing, washing, etc., and to nearby habitats that may harbour snail populations, which may seed the transmission site. The local community will generally be able to inform the team about the whereabouts of common sites and also of

more private bathing areas that should be included. Take time to observe where water contact is taking place; this may differ with different sectors of the community (adult, children, male, female, agricultural workers, fishermen). Teams must be sensitive to local conditions and customs, including rights of way and access to private land, and must try to avoid disturbing food crops or interfering with other agricultural practices.

The area and size of water contact points should be mapped with a hand-held global positioning system (GPS) device and simple maps of the local transmission sites should be prepared. Photographs of the site should be taken, as these will provide a useful record of how the habitats change over the seasons. Most large rivers are not suitable habitats for intermediate snail hosts and are not often implicated in transmission, but smaller pools and associated swampy areas may be important. The construction of dams across river systems, both large and small, inevitably leads to still or slow flowing water that provides ecological changes that might favour transmission.

Meteorological data should be consulted to detect annual trends in rainfall and temperature. All freshwater bodies will vary according to season and some may completely dry out. Many snail species, especially *Bulinus* spp. and *Oncomelania* spp., can survive periods of drought. Water levels at potential transmission sites can change rapidly according to rainfall patterns and temperature, necessitating inspection of water contact sites at different times of the year.

3.6 Order equipment for molluscicide application and allocate budget

Before starting the intervention phase, it is important to order and purchase associated equipment. This is likely to include:

Protective equipment

- protective clothing for all field personnel (overalls, face masks and goggles);
- water boots (Wellington boots and/or waders); and
- gloves.

Equipment for snail sampling and identification

- hand-held GPS;
- camera;
- water meter (pH, salinity, conductivity, turbidity, velocity, oxygen content);
- tape measure;
- forceps;
- pots and trays for snail collections;
- notebook and pencils;
- laptop;

- snail scoops and sieves;
- cool box;
- binocular microscope for laboratory use;
- weighing balance; and
- quadrats, if sampling *Oncomelania* spp.

Equipment for application of molluscicides

- molluscicide (this may not be generally available in-country and should be ordered well in advance from the suppliers);
- mixing barrels;
- compression sprayers, knapsack sprayers;
- petrol-driven portable pumps;
- long hoses;
- solution dispensers of various types;
- a heavy-duty 4x4 vehicle will be required for transport of pumps and molluscicide plus snail team; and
- a trailer or wheelbarrow may be required to reach some water bodies.

Ensure that budget is available and sufficient to procure the molluscicide, the equipment needed, staff requirements and transport. Snail control should generally be considered a long-term programme, so activities need to be sustainable.

3.7 Ensure communication and information dissemination

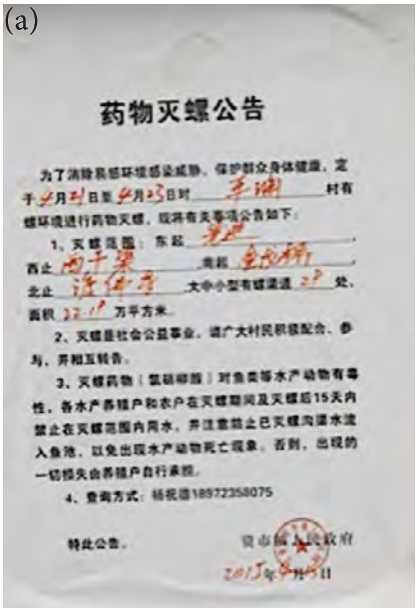
It is important to ensure that adequate notice is given of any molluscicide application and that local approval has been granted and in-country regulatory approval secured. When possible, local notices can be placed on community notice boards close to transmission sites or community oral announcements can be made (Figure 2). Information sheets could be provided to local farmers and others detailing the activity and the safety aspects. Programme managers should inform the community that:

- possible adverse effects to fish and amphibians may occur; and
- water use should be restricted for 24 h after molluscicide treatment. However, some programmes extend the restriction for more days.

Niclosamide is toxic to fish and amphibia (frogs and toads) but is safe to use at the recommended dosage in the presence of livestock and poultry. However, to avert complications arising from unrelated incidents, it is recommended that local farmers and others restrict access to areas being treated for a 24 -hour period. Niclosamide should be used carefully in closed water habitats where fish cannot escape.

Fish that are killed as a result of mollusciciding present no risk if eaten; the niclosamide levels are extremely low and are not toxic to humans (Andrews et al., 1983).

(a)



药物灭螺公告

特此公告。

(b) Notice is hereby given that molluscicide will be used in areas around Pingyuan village from 21 to 23 April in order to eliminate the threat of infected Oncomelania. Details are as follows:

1. Areas for snail killing: east of Xianjing, west to Xiganqu; south on Jinlong Bridge, north to Dufuo Temple. The area covered includes 29 big, medium and small ditches and channels, covering 221 900 m² in total.
2. Killing of these snails is a social and public welfare activity for public health; we hereby request villagers to cooperate, participate and notify each other.
3. Molluscicide (niclosamide) is toxic to fish and possibly to other organisms raised in aquaculture. Farmers and peasants are prohibited from using the water within the area treated from the beginning of the application and until 15 days after using niclosamide. Please pay attention to prevent the water treated with niclosamide from entering the ditches and channels and from flowing into the fish pond, to avoid killing fish. Otherwise, all losses shall be borne by farmers themselves.
4. Information request: Zhude Yang, 18972358075

Hereby notice
Zishi Town People's government
15 April 2015

Figure 2. (a) Notice of forthcoming snail control activities used in a *Schistosoma japonicum* control programme in China, and (b) translation

4. Phase 2: The intervention – snail sampling and application of molluscicides

4.1 Molluscicides

WHOPES recommends use of niclosamide emulsifiable concentrate (EC) and wettable powder (WP) for mollusciciding for which WHO specifications are available.[1] Niclosamide is a product specially developed for the control of freshwater snails, which act as intermediate hosts for schistosomiasis and other trematodes such as liver flukes (e.g. fascioliasis). Niclosamide acts against both snails and their eggs at low concentration, killing them in a few hours. The chemical also controls the free-living stages of the schistosome parasite found in water, the miracidia that hatch from the eggs, and the cercariae that develop in snails and infect humans directly through the skin.

[1] http://www.who.int/whopes/quality/en/Niclosamide.pdf

Other molluscicide products not evaluated by WHOPES include niclosamide ethanolamine 4% dry powder (DP), 5% granule formulation (GR), and 50% wettable powder (WP), and metaldehyde 1% plus niclosamide-olamine 25% suspension concentrate (SC).

4.1.1 Niclosamide

Niclosamide toxicity has been assessed by WHO[2]. The 1983 monograph by Andrews et al. (1983) provides extensive information on the chemistry, biology and toxicology of niclosamide as well as its effects on non-targeted plant and animal species. Niclosamide's environmental impacts have been more recently reviewed in a study using niclosamide for

Figure 3. Preparation of molluscicide

[2] www.who.int/whopes/quality/en

controlling lamprey in the Great Lakes region of the United States of America by Dawson (2003), who concluded that there is minimal risk to humans and the environment provided its application is appropriately informed and supervised.

The manufacturers state that niclosamide must be stored in the closed original containers in a cool, dry place away from foods and feed and safely out of the reach of children. If niclosamide is used according to label recommendations it is unlikely to create undue human hazard. Niclosamide-treated water can be used for irrigating crops. This has been established in crops of bananas, cotton, sugar cane, wheat, maize, citrus fruits, beans, rice, lentils, barley and peanuts. Niclosamide does not accumulate in the soil. In water, the active particles quickly adsorb to plants and to the organic substances in mud followed by biological degradation by bacteria, yeasts and fungi, and breakdown due to light exposure.

Further research and development is encouraged to develop a broader portfolio of safe and effective products.

Molluscicides can be applied in a solid form (GR, WP formulations) in dried soil or marshlands. It is an efficient way of killing amphibious snails. The product is more durable than the WP form and it is mostly used for the control of *Oncomelania* snails.

Molluscicides can also be applied by placing bags or sacks filled with molluscicides in running water or along the margin of the water or rivers. The balls of molluscicide placed in the water will dissolve progressively.

4.1.2 Snail control and environmental impact

There is now general international recognition of the importance of conserving the world's biodiversity. Snail species, as many other organisms, are under threat in certain regions and while snail control can be an important intervention for schistosomiasis control, molluscicide programmes should take into account the impact on non-target snails and the wider aquatic ecosystem. It is important to gather as much knowledge as possible about the molluscan fauna in the study area, and every effort should be made to restrict molluscicide to known disease transmission sites. Places of high molluscan species endemicity such as the shores of Lake Malawi and Lake Tanganyika should be treated extremely cautiously (Brown and Kristensen, 1997).

Niclosamide is toxic to fish and amphibia (frogs and toads) but is safe to use at recommended dosage in the presence of livestock and poultry. In pristine areas, where the local fish and amphibian species may not have been described, an initial ecological survey is recommended to monitor any potential impact on biodiversity (Dawson, 2003). In areas where the human population may be using freshwater fish as a source of protein, an assessment of the likely economic impact should be made. General observation suggests that fish stocks quickly return to pre-treatment levels.

4.2 When to apply molluscicides

There are two major considerations for the timing of molluscicide applications so as to maximize the chance of reducing transmission. The first relates to the seasonal weather patterns, particularly rainfall and temperature, which influence the size of the snail populations and determine transmission patterns. The second relates to careful timing of molluscicide applications with ongoing population-based chemotherapy programmes.

4.2.1 Season

The main schistosomiasis transmission seasons are determined by rainfall and temperature patterns. Flooding, drought and low temperatures (below 18°C) tend to depress and interrupt transmission, although low water levels are likely to focus people at water contact sites and may intensify transmission.

- Local conditions will inevitably dictate the mollusciciding schedule: in some endemic areas, transmission may occur to a greater or lesser extent in permanent water bodies for 9 or more months of the year, while limited in other areas to seasonal pools for 3–4 months of the year.

- Timing of applications will be weather dependent and the snail control manager must decide on the correct time to treat according to the level of water at the potential transmission sites.

- The snail biology and the generation times of the snail hosts (depending on species, this can be 2–3 generations a year) should be considered. There may be optimal times for controlling adult breeding snails.

- Usually in Africa, most transmission takes place after the main rains in the early and mid to main dry season. When transmission is seasonal, it is recommended to carry out regular applications of molluscicide. Where focal application is conducted, several treatment cycles may be required per year according to transmission patterns. Sampling of snails is required to determine the interval between application cycles and should be conducted one month after application. If snails are present, reapplication should be done.

<u>Africa</u>

- More frequent applications may be needed in permanent water bodies.
- In seasonal pools with *Bulinus* spp., spray shortly after rains and snail re-emergence. Snails can carry the infection through aestivation during the dry period.
- Due to the varied ecological conditions by region the timing of molluscicide treatment will vary according to location.

Brazil

- Molluscicide is conducted shortly before chemotherapy and again a few weeks later.
- Due to the varied ecological settings the timing varies by region depending on surveys of snail populations.

China

- Molluscicide is normally conducted in April shortly before the peak transmission season and shortly before mass drug administration (MDA). In marshland, large vegetation should be cleared before spraying.
- Focal treatment is done shortly before MDA.
- Treatment is warranted when new transmission foci are detected.
- Treatment of water bodies is conducted once in April. Treatment of marshland is repeated three times with an interval of one week between treatments. Three treatments are conducted to increase the snail mortality to > 90%.

4.2.2 Mass chemotherapy

In countries where regular mass chemotherapy programmes are in place, ideally the timing of molluscicide application should also be linked to the delivery of population-based chemotherapy. If snail numbers can be reduced before the chemotherapy, patterns of reinfection should be reduced. The optimum time for preventive chemotherapy is when there is no risk of reinfection with schistosomes, i.e. when the snails have been reduced and transmission is halted. Hence, it is sensible to reduce snail populations before undertaking a population treatment campaign (Sturrock, 1995). It is important for chemotherapy and snail control teams to be fully integrated. Based on experience, snail control should be conducted 5–7 weeks before chemotherapy to reduce the transmission potential.

In China, the timing for mass chemotherapy is conducted 1 month after the end of the transmission season in November/December. Molluscicide is conducted before the start of the transmission season in April.

4.3 How to apply molluscicides

4.3.1 Preparation for application

It may be necessary to remove large vegetation from aquatic and marshland environments before applying molluscicides in order to improve the penetration of the molluscicide and enhance contact with snails.

- For the treatment of flowing streams, lakes and stagnant waters, knapsack sprayers or compression sprayers carried on the back of the person spraying can be used. Where compression sprayers are used they should be fitted with control flow valves and preferably ceramic nozzles.
- To achieve greater coverage, a motor powered spraying machine is preferred (Figure 3, Figure 4).

- For irrigation canals and rivers, it is preferable to use an automatic dispenser fitted with an agitator.
- Focal control in canals and flowing water can be done using sprayers, with the molluscicide dispersing downstream.
- For better application coverage in larger water bodies, boats may be used to transport sprayers and applicators.
- In China, drones have been used to apply molluscicides but there is a need for calibration.

Figure 4. Snail teams applying niclosamide in Zanzibar, United Republic of Tanzania against *Bulinus* spp. in a *Schistosoma haematobium* control programme (a), in Cameroon (b) and in Wuhan, China (c, d) against *Oncomelania* spp. in a *Schistosoma japonicum* control programme

4.3.2 Preparation and application of molluscicide

Molluscicide should be prepared according to the manufacturer's recommendations (Figure 3).

- Niclosamide WP 70 is sprayed at 1% (1 kg/100 litres of water); the solution can be prepared on site using previously weighed aliquots. In an 8-litre capacity sprayer this would equate to 80 g of niclosamide WP 70 per 8 litres. The spray mix should be constantly agitated (Annex 5).
- The desired concentration is calculated on the basis of achieving 1 ppm active ingredient for niclosamide WP 70 for 8 hours. Increased concentration will reduce the required time for maximum effectiveness.
- When doing focal applications in stagnant water, it will suffice to spray around the transmission site to a radius of at least 15 metres (at a concentration of 10 grams per litre) (Figure 4).
- It is necessary to evaluate the impact of the treatment as detailed below in phase 3.
- In each habitat it is essential to treat the banks and areas close to the shore; this will mean spraying from within the water body and often it will be necessary to walk along small streams and rivers spraying the banks on both sides.

4.3.3 Health and safety of applicators

- The applicators must wear personal protective clothing including overalls, gloves, boots and simple face masks, and goggles.
- Care must be taken to prevent operators from slipping over; each operator should have a back-up person to assist their movement in and out of the water body.
- All skin contact with water must be avoided to prevent schistosomiasis infection.
- Antiseptic must be available to the team in order to wash down any skin exposed to water from the site, as schistosome cercariae might be present.
- Handling, storage and disposal of waste containers should be done according to national regulations.
- The supervisor should be aware of non-swimmers in the team.
- Excess or out of date molluscicide should be disposed of according to the manufacturer's instructions.
- Team members should have regular health checks and be screened for schistosome infection.

5. Phase 3: Monitoring and evaluation of molluscicide activities

5.1 Snail sampling: identification and parasite monitoring

The snail population should be sampled before and after molluscicide application.

- Investigate each site for the presence of freshwater snails in a standardized manner and make a collection. For *Bulinus* spp. and *Biomphalaria* spp. choose a sample area along the perimeter of the water body of around 15–20 metres, which can be measured with a tape, and instruct two trained staff to sample within that area for 15 minutes. Collect all snails and place them in a basin; count all species, alive and dead. Snails are most often found on vegetation, especially decaying leaves, sticks, water lilies and discarded plastics (Figure 5). Snails can be sampled using purpose built snail scoops and/or small hand-held sieves; snail shells tend to be fragile and so plastic or rounded forceps are best for handling them. Potential intermediate snail hosts should be transferred to the laboratory for checking for the presence of parasites. A different technique is employed for calculating the densities of amphibious *Oncomelania* spp., with numbers being assessed using quadrats.
- Temperature, pH, salinity and conductivity of the water should be measured and recorded at each of the sites, on all survey and sample days using standard protocols and forms (Annexes 1, 2 and 4). All collection sites will be located using a hand-held GPS device (Annexes 1 and 2).
- Identify the snails in the collection using appropriate snail identification keys (for the African fauna, the Danish Bilharziasis Laboratory (1980) snail keys are recommended).
- *Bulinus* spp. and *Biomphalaria* spp. should be examined in the laboratory for parasitic infection using the shedding method. For this purpose, the snails are placed individually in flat-bottomed glass vials or multi-welled plates containing de-chlorinated water, and exposed to indirect sunlight for a maximum duration of 4 hours. Cercarial shedding will be observed using a binocular microscope (Annex 2). Snails that do not shed cercariae on the first sunlight exposure will be re-exposed on the second day. Cercariae have a distinct circadian rhythm and the best time to isolate cercariae infecting humans is usually mid-morning. Based on their morphology, cercariae from *Bulinus* spp. will be categorized either as those of *S. haematobium* or those of other trematodes (non-*S. haematobium* cercariae) and from *Biomphalaria* spp. as *S. mansoni* or other trematodes. Freshwater snails act as hosts for a variety of trematodes and there are some helpful identification guides to differentiate cercariae; the most useful one for the African situation is that of Frandsen and Christensen (1984). Other closely-related animal schistosomes (e.g. *S. bovis, S. curassoni* from *Bulinus* spp. or *S. rodhaini* from *Biomphalaria* spp.) may also be observed, which are

not easily differentiated from the species infecting humans. Any domestic or wild animal contact at the transmission site should be noted.

- A different screening approach is taken for *Oncomelania* spp.; here, a subset of snails are squashed between glass sheets and then scanned under a binocular microscope for evidence of cercariae or sporocysts.
- If there is a need to keep cercariae for further identification or genotyping, then they can be collected using a pipette and placed on Whatman FTA cards for future molecular analysis (Emery et al., 2012). Similarly, collected snails can be preserved in small glass universal tubes containing 100% ethanol.

Different approaches are used to check the efficacy of the molluscicide application; while it is possible to determine the concentration of niclosamide in the water (Strufe, 1962), this approach is rarely needed for focal molluscicide operations where only small amounts of the chemical are periodically applied.

Figure 5. Sampling of aquatic snails

The following indicators are recommended for evaluation of the molluscicide in the long term:

- snail presence or absence;
- snail infection rates (cercaria shedding); and
- human infection where water contact takes place.

The selection of the sites to survey can be random, through sentinel sites previously identified by the programme among the sites that have been treated with molluscicides.

It is advisable to revisit the site within 1–4 weeks and re-survey the same area as before the application. If living intermediate snail hosts are recovered, then the area should be re-sprayed. The pre- and post- molluscicide population densities can be compared. Snail sampling should be conducted at as many sites as possible according to the capacity of the programme.

Future applications will depend on the recovery and reinvasion of the snail population. As a general rule, if intermediate snail hosts are present at the surveyed site, then all human contact points around the water body must be treated thoroughly. If no snails are found but snails were recorded at the time of the last survey, then all contact points should be sprayed. If snails have been absent during the past two surveys, then there is no need to treat. However, if snails appear to be absent and the area is a known transmission site, then the snail team can still decide to treat. The focality of schistosomiasis transmission is well recognized, as is the occurrence and persistence of "hot spot" areas. Increased molluscicide application may be required in these areas.

5.2 Use of sentinel sites for determining molluscicide effectiveness

According to the programme's capacity, sentinel sites should be established for longitudinal monitoring of snail densities and cercaria infection rates. The selected sites should be treated in the same way as the rest of the programme.

5.3 Parasitological information

Parasitological information from the human population in relation to prevalence and reinfection will be the final measure of the effectiveness of snail control in reducing transmission. If the prevalence and reinfection rates in villages or communities are not falling as anticipated, then the area should be resurveyed to determine whether there are water contact sites that may have been missed from the snail control programme and/or the snail control intervention should be intensified.

5.4 Resistance to molluscicides

There has been limited use of molluscicides in recent years and hence little concern about the development of resistance to niclosamide. Earlier work in Saint Lucia has demonstrated that Biomphalaria glabrata exposed to niclosamide at regular intervals for 9 years exhibited no evidence of resistance (Barnish and Prentice, 1981).

However, it is recommended that in areas where regular snail control is taking place, monitoring for resistance should be carried out every 2 years, or immediately if field data suggest a changing response to the chemical. This can be done by exposing snails to known concentrations of niclosamide in controlled laboratory settings; this will require establishing diagnostic concentrations for resistance monitoring (see WHO, 2017 [in preparation]). Resistance monitoring should be done by the national research institution. This can be done by monitoring the susceptibility of wild populations of snails over the period of the intervention or by using the diagnostic concentration when established by WHO. Comparison can be made with a molluscicide-susceptible reference laboratory colony of snails.

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Annex 1. Form for field collection of snails

Collector's name: Date of collection (dd/mm/yy):

GENERAL LOCALITY INFORMATION	
Water body name:	Site code:
Water body type: pond ☐ lake ☐ dam ☐ rice paddy ☐ marsh ☐ stream ☐ river ☐ canal ☐ permanent ☐ temporary ☐	District name: Nearby village/treatment strategy:
GPS coordinates: Latitude: (N)/(S) Longitude: (E)/(W) Altitude: m	Notes:

WATER PROPERTIES DATA	
Temperature:	TDS (mg/L):
pH:	Conductivity (ms):
Salinity (g/L):	Dissolved oxygen (mg/L, ppm):
Flow rate: > 1m/s ☐ 0.5–1m/s ☐ < 0.5m/s ☐ still ☐	Depth: < 0.5 m ☐ 0.5–1m ☐ > 1 m ☐
Water level: Flooded ☐ Normal ☐ Low ☐ Dry ☐	Season: Dry ☐ Rainy ☐

COLLECTION INFORMATION	
Number of collectors:	Number of dredges (if applicable):
Time in:	Time out:

ECOLOGICAL/EPIDEMIOLOGICAL DATA	
Substrate: mud ☐ sand ☐ gravel ☐ rock ☐ concrete ☐ peat ☐ roots ☐ Notes:	Domestic animals: cow ☐ sheep ☐ goat ☐ pig ☐ horse ☐ donkey ☐ camel ☐ dogs ☐ Notes:
Snails present: Notes:	Wild animals: monkey/ape ☐ ungulate ☐ carnivore ☐ water bird ☐ other ☐ Notes:
Main vegetation present (e.g. reeds/ grasses): Notes/species identification:	Activities: washing/bathing ☐ washing clothes ☐ washing dishes ☐ washing car /bike ☐ collecting water ☐ fording ☐ swimming/playing ☐ fishing ☐ rice cultivation ☐ other farming ☐ sanitation ☐ other ☐ Notes:

SNAIL DATA	
Snail species collected:	Number of snails collected:
Number of infected snails:	Expected schistosome species:
Date of first shedding attempt (dd/mm/yy):	Notes:

Annex 2. Cercaria collection form

Site name/code:

Snail collection date (dd/mm/yy):

Infected snail ID	Date of shedding (dd/mm/yy)	Snail species/ collection method	Time of shedding (morning/ evening)	Name of cercariae collector	No. of cercariae /FTA (and no. of water controls)	FTA* card ID	Fate of snail specimen (preserved?)	Notes:

* FTA: fast technology for analysis

Annex 3. Niclosamide application form

Sprayers's name: Control date (dd/mm/yy):

GENERAL LOCALITY INFORMATION	
Water body name:	Site code:
Water body type: pond ☐ lake ☐ dam ☐ rice paddy ☐ marsh ☐ stream ☐ river ☐ canal ☐ permanent ☐ temporary ☐	District name: Region / island:
GPS coordinates: Latitude: (N)/(S) Longitude: (E)/(W) Altitude: m	Intervention arm: Notes:
Nearby village:	

APPLICATION INFORMATION	
Method of application 8 litre Hudson sprayer ☐ 10 litre Hudson sprayer ☐ Power sprayer ☐ Barrel drip ☐	Dosage: Number of tanks used: Volume of water sprayed: Barrels used:

ECOLOGICAL INFORMATION	
Number in team:	Time of application:
Duration of application:	
Snails species present: Notes:	Domestic animals: cow ☐ sheep ☐ goat ☐ pig ☐ horse ☐ donkey ☐ camel ☐ dogs ☐ Notes:
Other animals present: fish ☐ beetles ☐ Frogs/toads ☐ Other invertebrates ☐ Notes:	Wild animals: monkey/ape ☐ ungulate ☐ carnivore ☐ water bird ☐ other ☐ Notes:
Notes:	

SNAIL AND INFECTION INFORMATION	
Snail genus collected:	Number of snails collected:
Number of infected snails:	Expected schistosome species:
Date of first shedding attempt (dd/mm/yy):	Notes:

Annex 4. Protocol for the collection of snails, shedding of cercariae and storage onto fast technology for analysis (FTA) cards for future DNA analysis

1. Snail collection

1. Assess designated collection sites, using local knowledge and transmission information.
2. Put on protective clothing and gloves.
3. Collect snails using metal mesh paddle scoops; handle length dependent on the depth of the site.
4. At sites where snails are found directly on submerged rocks/leaves, pick off by hand using forceps. In deeper waters, where possible, use a dredge to collect snails. Sampling should be semi-quantitative, with 2–3 collectors scooping for approximately 15 minutes' duration and/or up to three dredges per site. It is important to note the distance from shore for each of the dredge attempts and try to keep this relatively close to the shore (i.e. no further than 10 m), if too far offshore there is potential for missing important transmission areas.
5. Inspect snails and identify species/genus level based on the shell shape and size. Place in plastic containers filled with a little water for later processing. Label these plastic containers with collection site and date, additionally with collection method (i.e. dredge or scoop).
6. Fill in the snail collection form with the location information: date, time, season (wet/dry) and global positioning system (GPS) coordinates. Record water chemistry variables such as water temperature (°C), conductivity (measured in micro-Siemens, or mS), total dissolved solids (TDS, measured in mg per litre [mg/L]), salinity (g/L) and pH using a hand held water meter. Note down units for the chemistry as may vary from one meter to another. Also, note the habitat type, substrate, water depth and flow rate and distance from shore for dredging activities.
7. Note down the presence or absence of all genera of snails found; there will be several genera present dependent on the country. It is important to include notes on vegetation types, human activities, and domestic and wild animal present.
8. When travelling long distances, wrap snail pots with damp cloths and place in a cool box (or out of sunlight) for transport. This will keep the snails cooler, decreasing death and stress of the snails.

2. Snail shedding and storage in the laboratory

1. Remove snails from the plastic jars/containers, one site at a time, into sorting trays. Place all potential host snails into 12 or 24 welled trays containing 3 ml of water wells (if low numbers, put in individual wells; if many snails, start off by placing 2–5 in each well, then, if shedding, place individually into separate wells). Leave the trays in indirect sunlight/under a lamp for 2–3 hours. If the laboratory is air conditioned it would be advisable to turn off the air conditioning or at least to set the temperature warmer (> 24 °C) when attempting shedding; this should induce shedding. Ideally, shedding should be attempted once during the morning to middle of the day, as this has been reported to be the hours of peak cercarial production for *Schistosoma mansoni* (Kazibwe et al., 2010) and once at dusk to look for presence of any other (animal) schistosome species (e.g. *S. rodhaini*).

2. Following exposure to light for shedding, examine the wells under a dissecting microscope for presence of *Schistosoma* cercariae. If shedding is occurring, make sure the infected snail is in a separate well of its own. Use a 20 µl pipette set at 3 µl to collect individual cercariae and place directly onto a fast technology for analysis (FTA) card. Continue until you have approximately 20–40 cercariae on one FTA card. Make sure the cercariae spots do not touch each other. Allow the FTA card to dry for one hour. Then place in a sealable plastic bag. The DNA should be stable at ambient temperature without damage. **Change pipette tips between each snail** to avoid cross-sample contamination.

   Note: Freshwater snails are used as the host for many different parasites and therefore all/any cercariae emerging from snails must be noted down (i.e. *Paramphistomum* spp., *Strigea* spp.). If there is time, collect a few on an FTA card, making sure to write on the card and the log sheet that they are non-schistosome cercariae.

3. Label FTA cards

   This is vital for the samples to be analysed and stored within the schistosome repository correctly. Please make sure that the cards are labelled with:
   - snail ID number;
   - site name;
   - site code;
   - date of collection of cercariae (shedding date not snail collection date); and
   - number of cercariae.

   Additionally, if more than one sample is put on one card, clearly indicate which dots belong to which sample.

4. Water control samples for cercariae FTA cards

   Leave a small part of the card clear; in this area pipette 6 drops of water onto the card (using a P20, set at 3 µl). This should be bottled water from the well the cercariae have been collected from. With a pencil indicate which the water control samples are. This control is used to test that there is no genetic material contaminating results in the water used. You only need to do this for a few cards per day.

Fill in the snail form

- Record the species of snail collected.
- Record the total number of snails collected per species.
- Record the total number of snails that were shedding human schistosome cercariae.

3. Storage of snail samples

Cercarial shedding should be attempted on at least 2 days; a total of 50 snails should be stored in ethanol from each collection site. Make sure that any snails that are not being fixed and kept are killed and disposed of very carefully, so they cannot escape into local water bodies (preferably dispose of into incineration bins).

- From each site, a maximum of 50 snails should be fixed in ethanol including ALL shedding snails.
- Shedding snails should be stored separately from non-shedding snails. Additionally, separate out snails shedding animal schistosomes from human schistosomes.
- Storage should proceed as follows:
- Remove snails from water and place pots of 15–20 snails in a freezer for a few minutes; this will relax the snails so they can quickly be fixed in ethanol without retraction into their shells. Alternatively relax snails with a few menthol crystals.
- Remove from freezer and add 70–100% ethanol to each glass universal.
- Shedding snails can be stored together in one pot. Ensure correct labelling glass universal.
- Label the outside of the tube with masking tape and marker pen. Place parchment paper inside the tube with information written in pencil.
- Change ethanol after a few days to preserve snail tissue for future use.

How to label snails

- Date
- Village code
- Snail collection sub-site (sequential number 1,2,3,4)
- If shedding or non-shedding (Use a "+" for positive and "–" for negative).
- If shedding human schistosome or animal schistosome (draw shape of animal schistosome
- It is important to store snails with a label on the outside of the tube but also parchment paper on the inside written on in pencil.

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Annex 5. Application of niclosamide in static and flowing habitats

GUIDELINES FOR MOLLUSCICIDING OPERATIONS USING NICLOSAMIDE (BAYLUSCIDE®)

1. Niclosamide (Bayluscide®) and its properties

The active molluscicidal ingredient (a.i.) in Bayluscide® is niclosamide. Bayluscide® is a product specially developed for the destruction of freshwater snails which transmit schistosomiasis and some other trematode infections. This molluscicide can kill both the snails and their egg masses at very low concentrations (ppm or $mg.l^{-1}$) within a few hours; at such levels Bayluscide® can also kill miracidia and cercariae, the larval stages of schistosomes which may occur in the transmission foci.

Niclosamide is available in two formulations:

(i) Bayluscide® wettable powder (WP) with 70% efficiency (a.i. 70%) and

(ii) Bayluscide® emulsifiable concentrate (EC) with 25% efficiency (a.i. 25%). This product is sometimes called Clonitralide®.

As a general rule, Bayluscide® WP should be mixed at not less than 1:20 (WT/Vol.) in water and the EC formulation should be diluted to at least 1:15 (Vol./Vol.) in spraying equipment in order to facilitate improved dispersion.

Further practical information on the application of Bayluscide® is given in the following two documents which may be sent to the reader by request to the address given for each:

(i) "Bayluscide® molluscicide for the control of freshwater snails, vectors of bilharziasis" (7 pages, in English and French). Bayer AG Technical Document, Bayluscide, Sparte Pflanzenschutz, Anwendungstecknik, Beratung, 5090 Leverkusen, Bayerwerk, Germany.

(ii) *Bilharzia. A Manual for Health Workers in Malawi* 64 pages. 3rd Edition, 1986. Published with support from CIBA GEIGY Ltd. on behalf of the Ministry of Health, Lilongwe, Malawi, for the National Bilharzia Control Programme.

2. Application of niclosamide

Cost–effective mollusciciding operations require that as far as possible the correct dosage is applied; either too much or too little molluscicide is wasteful and inefficient. The correct dose (D) is derived from the product of the concentration (C) of the chemical in the waterbody and the time (T) during which the snail hosts are exposed: thus

$$D = C \times T \quad \text{and hence } C = \frac{D}{T}$$

In most field situations the dose recommended for niclosamide can be accepted as satisfactory, but in certain transmission sites it may be necessary to increase it slightly or, for example, to use the 25% E.C. formulation mixed with diesel oil, instead of the 70% W.P., in order to achieve better dispersion of the molluscicide in waterbodies where the aquatic vegetation is exceptionally dense. Niclosamide is highly toxic to the snail hosts at a concentration of 0.5 parts per million ($0.5\ mg.l^{-1}$) if the water volume to be treated is accurately measured and the exposure time is 24 hours.

In transmission sites with stationary water (ponds, borrow-pits, small dams and reservoirs, etc.) the first procedure is to obtain a reasonably accurate estimation of the volume of water in cubic metres. While a satisfactory estimation of the surface area can usually be quickly obtained, measurements to determine the average depth are more time-consuming and they may need, particularly in larger waterbodies, to take account of considerable variations in depth. Inaccurate measurements of the volume of water in snail habitats larger, for example, than 10.000 m^3 can be both costly and wasteful. In fact, at the present time, molluscicide operations in very large waterbodies are limited to specific transmission sites.

The following description of the application of Bayluscide, given in the document entitled *Bilharzia: A Manual for Health Workers in Malawi* (Anon., 1986), is exemplary.

Static water habitats

The makers of Bayluscide recommend that it should be mixed in the proportion of 1 gramme of active ingredient to two million parts (cubic centimeters) of water. This is the same as half a gramme to one million parts of water, or 0.5 parts per million (ppm). In practice, because only 70% of the powder contains the active chemical this must be allowed for when calculating the amount of chemical needed. Knowing these facts, (which never change), the only thing we have to measure when treating static water is the amount of water to treat, i.e., the volume. This is calculated by measuring the average length by the average width by the average depth (all in metres), thus:

$$Volume\,(m^3) = length \times width \times depth$$

When the volume is known the amount of chemical necessary if treating with Bayluscide 70% wettable powder is:

$$Volume\ of\ waterbody \times 0.5 \times \frac{100}{70} g$$

Or if using Bayluscide 25% emulsifiable concentrate, the amount of chemical needed is:

$$Volume\ of\ waterbody \times 0.5 \times \frac{100}{25} cm^3$$

Example

A swamp with bilharzia has an average length of 100 metres, an average width of 40 metres, and an average depth of 3/4 of a metre. How much chemical is needed to treat it? From the equation above, and using Bayluscide 70% WP, amount of chemical needed =

$$(100 \times 40 \times 0.75) \times 0.5 \times \frac{100}{70} = 2142.86 g\ \ or\ \ 2.14\,kg$$

Using Bayluscide 25% EC, amount of chemical needed =

$$(100 \times 40 \times 0.75) \times .05 \times \frac{100}{25} = 6000\,cm^3$$

In locations where water is static, such as swamps, dams and ponds, treatment is best carried out using sprayers. These are also useful in flowing-water habitats where vegetation obstructs the flow or in drains where the flow is so sluggish that the

molluscicide will travel only a short distance before becoming ineffective. Because drains are generally of varying cross-section, and the depth of water in them can range from almost nothing to shallow ponds, it is difficult to estimate accurately the amount of chemical required to treat them effectively. For this reason it is better to overdose than underdose. For places where the flow is very slow, 30 mg Bayluscide in 10 litres of water sprayed generously over the whole surface of the water should be sufficient. The powder is placed in the sprayer with about 1 litre of water and thoroughly shaken before the remainder of the water is added. Spraying should be carried out systematically, and the entire surface of the water should be treated.

Where drains are difficult to treat because of large volumes of water, or vegetation makes access to them impossible, they can be treated by the "dam and flush" technique. This involves damming the drain at suitable points (e.g. a road bridge), and then treating the trapped water heavily (10 ppm) by blanket spraying the surface. The water is left for two hours while the water upstream is treated in the normal way (moving from mouth to source), and the dam is then breached and the entire drain flushed. This method saves chemical and has a longer-lasting effect.

For large dams and lakes, where the water is deep and there is a submerged vegetation, only the edges need be sprayed for a distance of about 5 metres from the shore. This is often best carried out from a boat.

However, as already mentioned, it is very important to remember the reservations concerning the cost-effectiveness of carrying out molluscicide in large waterbodies. If chemical treatment does seem to be desirable it is probably best restricted to actual transmission sites where infected snails, cercariae and miracidia are concentrated, often only seasonally.

At foci, where molluscicide:
is beyond the range of knapsack sprayers, stirrup pumps can be used. When these are unobtainable, old maize cobs soaked in a concentrated solution of Bayluscide for a week and thrown into the middle of the pond are effective.

Knapsack sprayers, as their name implies, are carried on the back of the worker, and there are models where the chemical can be pumped out by hand, while others dispense the chemical under pressure. Stirrup pumps are designed to enable the chemical to be dispensed under high pressure so that it can be projected long distances.

Flowing water habitats

In general the cost of molluscicide in flowing waterbodies is more expensive than in comparable stationary transmission sites.

Chemical control of flowing water is commonly carried out by drip-feed technique. This involves the steady introduction of the chemical for a number of hours into flowing water at the source of the system. The aim is to use the flow of the water to carry the chemical throughout the system. It requires that sufficient chemical is introduced at the source to ensure that by the time the chemical front reaches the end of the scheme it is still of high enough concentration to kill the snails and their eggs. In practice the water seldom flows uninterrupted from one end of the scheme to another. There may be ponds or storage dams midway to break the flow, or in irrigation schemes there may be "dead ends" of canals, where no flow occurs unless water is being drawn from them. These factors should

be taken into account and allowed for when applying molluscicide by drip-feed. Such situations may require booster dispensers at intermediary dams, and knapsack spraying of the tail ends of static water canals, or isolated stagnant pools in natural water bodies. These places are usually primary transmission points. Dispensers should be set up at narrow or turbulent points in a stream canal to ensure complete mixing of the chemical with the water.

Chemical introduced into flowing water will be immediately carried away from the point of application, and therefore must be applied for a period of time to compensate for this. If not, it will not be in contact with any snails present long enough to kill them. The recommended time that flowing water should be treated with Bayluscide at a concentration of 1 ppm is 8 hours. The only measurement that concerns us in this situation is how fast the stream or river is flowing. This can be measured using a flow metre. However, such instruments may not be easy to obtain, and for the purposes of snail control the following procedure to measure the rate of flow or discharge is quite adequate:

(i) Measure and mark off a 20 metre stretch of the river or canal below the point where you will introduce the chemical.

(ii) Drop in a float (green twig or float) above the first mark and measure the time in seconds that it takes for the float to travel the distance between the two marks.

(iii) Repeat the operation six times and note the fastest speed.

(iv) Measure the cross-sectional area of the canal/stream. This is obtained by measuring the average width and depth, and multiplying by 0.85. The latter figure is a constant as the average flow velocity amounts to about 85% of the maximum velocity measured.
Now the Discharge (D) = speed x cross section x 0.85 m^3/sec.
Knowing the discharge, the amount of chemical (Bayluscide 70% WP) needed to treat a flowing water habitat is

$$D \times 1 \times \frac{100}{70} \textit{ grams per second}$$

We have decided to treat for 6 hours (rather than 8 hours) and therefore the amount of chemical needed is

$$D \times 1 \times \frac{100}{70} \times 6 \times 60 \times 60 \ g$$

Example
A stream is found to have a discharge of 6 m^3/sec. How much Bayluscide 70% WP is needed to treat it?
From the equation above, the amount of chemical needed =

$$6 \times 1 \times \frac{100}{70} \times 6 \times 60 \times 60 = 185.14 \ kg$$

A convenient dispenser is a 200 litre drum that can be set to empty its contents every 30 minutes. Thus, to discharge the full amount of chemical, the drum will need to be emptied 12 times (12 x 30 mins = 6 hours). Therefore the amount of chemical that should be mixed at each filing of Bayluscide 70% WP should be:
This amount of chemical is measured out, preferably in the laboratory beforehand and stored in 12 bags of 15.42 kg each, and added to the drum when 1/4 full of water.

$$\frac{185}{12} = 15.42\ kg$$

When the powder has been thoroughly mixed, the remainder of the water is added, and the drum is ready for discharging. It is obvious that no break in the discharge should occur once the drip-feed has started or some water will pass by untreated. To avoid this two dispensers should be used side by side, the one being recharged while the other is discharging. Frequent stirring of the contents of the drum during discharge is also recommended to avoid the chemical settling out.

With regard to mixing the spray, the manufacturer (Bayer) recommends that Bayluscide 70% is sprayed at 1% (1 kg/100 litres water). The spray mixture should be constantly agitated. While mixing the spray it is advisable to wear a mask. For the application of Bayluscide 25% EC a dilution with water in a ratio of 1:10 to 1:60 is recommended, the latter being optimal. Bayluscide 25% EC displays an excellent spreading effect when used in combination with diesel oil at a ratio of 8.5 parts of Bayluscide 25% EC to 1.5 parts of diesel oil (see the document entitled "Bayluscide®. Molluscicide for the control of freshwater snails, vectors of bilharziasis", published by Bayer.

In some irrigation schemes it may be possible to hold the treated water in the canals/drains/dams for a period of time to allow the chemical longer contact with the snails. In order to do this we need to know the discharge of the pump or weir and the time it takes to fill the system with Bayluscide-treated water. The method for measuring the discharge is described above. The time it takes to fill the system will depend on the size of the pump/weir feeding it, and it can be calculated thus:

$$\textit{Time required to fill system} = \frac{\textit{Size of dam } (m^3)}{\textit{Capacity of pump } (m^3 \textit{ per second})}$$

Example
Size of dam and canals = 70,000 m^3. Capacity of pump = 6 m^3/sec. Therefore, time to fill the system =

$$\frac{70\,000}{6 \times 3600} = 3.24\ hours = 3\tfrac{1}{4}\ hours$$

If the volume of water in the system is unknown the time taken to fill it can be calculated by first emptying the canals/dams and then measuring how long it takes for them to be filled up again. With this information it is possible to calculate the amount of chemical required as described above.

The following procedure should be adopted when carrying out a drip-feed by the above method:

(i) Inform irrigation and agricultural personnel well in advance to ensure that the day chosen for the drip-feed will suit their programme.

(ii) The day before drip-feeding switch off the pumps, (close the weir), and open the gates to the fields to reduce the level of the water in the canals as much as possible. This will reduce the amount of untreated water already present, and ensure more thorough penetration of the chemical subsequently.

(iii) On the day of the drip-feed close all the outlet gates, and switch on the pumps and the drip-feed together.

(iv) When the system is full switch off both pumps and dispensers and hold the water in the canals/dams for a further 24 hours before releasing it. During this time any parts where the treated water is unlikely to reach should be treated using knapsack sprayers.

Annex 5 - Table 1

Quantity of Bayluscide WP 70% needed to give a concentration of 1 PPM in stationary water

Volume of water in cubic metres	Quantity of Bayluscide WP 70% needed				
1	0.00143	kilograms	OR	1.43	grams[1]
2	0.00286	"	"	2.86	"
3	0.00429	"	"	4.29	"
4	0.00572	"	"	5.72	"
5	0.00715	"	"	7.15	"
6	0.00856	"	"	8.56	"
7	0.01001	"	"	10.01	"
8	0.01144	"	"	11.44	"
9	0.01287	"	"	12.87	"
10	0.01430	"	"	14.30	"
20	0.0286	"	"	28.6	"
30	0.0429	"	"	42.9	"
40	0.0572	"	"	57.2	"
50	0.0715	"	"	71.5	"
60	0.0856	"	"	85.6	"
70	0.1001	"	"	100.1	"
80	0.1144	"	"	114.4	"
90	0.1287	"	"	128.7	"
100	0.143	"	"	143.0	"
200	0.286	"	"	286.0	"
300	0.429	"	"	429.0	"
400	0.572	"	"	572.0	"
500	0.715	"	"	715.0	"
600	0.856	"	"	856.0	"
700	1.001	"	"		
800	1.144	"	"		
900	1.287	"	"		
1000	1.430	"	"		

NOTE: The efficiency of Bayluscide wettable powder is 70%. This means that to provide 1 gram of active Bayluscide, 1.43 g of the Bayluscide powder is needed.

[1] In field situations, the molluscicide will be measured to the nearest gram.

Annex 5 - Table 2

Quantity of Bayluscide WP 70% needed to give a concentration of 1 PPM in flowing water

1 cubic meter = 1000 litres

Flow of water in cubic metres per second		Period of continuous application	Quantity of Bayluscide WP needed		
0.01	(or 10 litres)	8 hours	0.4116	kg	(or 411.6 g)
0.02	" 20 "	"	0.8232	kg	(or 823.2 g)
0.03	" 30 "	"	1.235	kg	
0.04	" 40 "	"	1.641	kg	
0.05	" 50 "	"	2.058	kg	
0.06	" 60 "	"	2.470	kg	
0.07	" 70 "	"	2.882	kg	
0.08	" 80 "	"	3.293	kg	
0.09	" 90 "	"	3.705	kg	
0.10	" 100 "	"	4.116	kg	
0.20	" 200 "	"	8.232	kg	
0.30	" 300 "	"	12.348	kg	
0.40	" 400 "	"	16.464	kg	
0.50	" 500 "	"	20.580	kg	
0.60	" 600 "	"	24.696	kg	
0.70	" 700 "	"	28.812	kg	
0.80	" 800 "	"	32.928	kg	
0.90	" 900 "	"	37.044	kg	
1.00	" 1000 "	"	41.160	kg	
2	Cubic metres	"	82.320	kg	
3	" "	"	123.480	kg	
4	" "	"	164.640	kg	
5	" "	"	205.800	kg	
6	" "	"	246.960	kg	
7	" "	"	288.120	kg	
8	" "	"	329.280	kg	
9	" "	"	370.440	kg	
10	" "	"	411.600	kg	

NOTE: The efficiency of Bayluscide wettable powder is 70%. This means that one part of the powder contains only 0.7 part of active Bayluscide. In other words, to get a concentration of 1 ppm of active Bayluscide in the water, 1.43 grams of Bayluscide WP must be applied to every cubic metre of water.

Annex 5 - Table 3

Quantity of Bayluscide EC 25% needed to give a concentration of 1 PPM in stationary water (ponds and pools, etc.)

Volume of water in cubic metres	Quantity of Bayluscide WP 70% needed				
1	0.004	kilograms	OR	4	grams[1]
2	0.008	"	"	8	"
3	0.012	"	"	12	"
4	0.016	"	"	16	"
5	0.020	"	"	20	"
6	0.024	"	"	24	"
7	0.028	"	"	28	"
8	0.032	"	"	32	"
9	0.036	"	"	36	"
10	0.040	"	"	40	"
20	0.080	"	"	80	"
30	0.120	"	"	120	"
40	0.160	"	"	160	"
50	0.200	"	"	200	"
60	0.240	"	"	240	"
70	0.280	"	"	280	"
80	0.320	"	"	320	"
90	0.360	"	"	360	"
100	0.400	"	"	400	"
200	0.800	"	"	800	"
300	1.200	"	"		
400	1.600	"	"		
500	2.000	"	"		
600	2.400	"	"		
700	2.800	"	"		
800	3.200	"	"		
900	3.600	"	"		
1000	4.000	"	"		

NOTE: To get a concentration of 1 ppm, 4 g of the emulsifiable concentrate would be needed to treat each cubic metre of water.

Annex 5 - Table 4

Quantity of Bayluscide EC 25% needed to give a concentration of 1 PPM in flowing water

1 cubic meter = 1000 litres

Discharge or flow per second					Period of continuous application	Quantity of Bayluscide EC 25% required for entire period of application
0.001	cubic meters	(or	1	litre)	8 hours	115.2 g
0.002		"	2	"	"	230.4 g
0.003		"	3	"	"	345.6 g
0.004		"	4	"	"	460.8 g
0.005		"	5	"	"	576.0 g
0.006		"	6	"	"	691.2 g
0.007		"	7	"	"	806.4 g
0.008		"	8	"	"	921.6 g
0.009		"	9	"	"	1.0368 kg
0.010		"	10	"	"	1.152 kg
0.020		"	20	"	"	2.304 kg
0.030		"	30	"	"	3.456 kg
0.040		"	40	"	"	4.608 kg
0.050		"	50	"	"	5.760 kg
0.060		"	60	"	"	6.912 kg
0.070		"	70	"	"	8.064 kg
0.080		"	80	"	"	9.216 kg
0.090		"	90	"	"	10.368 kg
0.100		"	100	"	"	11.520 kg
0.200		"	200	"	"	23.040 kg
0.300		"	300	"	"	34.560 kg
0.400		"	400	"	"	46.080 kg
0.500		"	500	"	"	57.600 kg
0.600		"	600	"	"	69.120 kg
0.700		"	700	"	"	80.640 kg
0.800		"	800	"	"	92.160 kg
0.900		"	900	"	"	103.680 kg
1.000		"	1000	"	"	115.200 kg

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