ensuring plants are high quality and by regularly harvesting adults, thereby reducing the two factors most likely to result in dispersal.

Two key aspects are fundamental to any successful rearing program.

Proximity to an adequate and constant water source—either a permanent water body or a reliable pumping system. Town supply water, tank water or water from ponds, rivers or wells may be used. If the supply is likely to be disrupted, storage facilities may be required.

Quality of host plants. Field studies have shown that water hyacinth plants which are healthy and have relatively high tissue nitrogen concentration yield more eggs and produce a higher proportion of adults with healthy ovaries than poorer quality (low nutrient) plants (Center 1994). Laboratory studies indicate that high quality plants can extend the duration of the reproductive period and increase the total reproductive output (Center 1994). If the quality of plants used during rearing deteriorates, insect production declines dramatically and insects are likely to disperse from the rearing area. Ultimately, the rearing process may collapse. Nutrient levels influence the growth rate and biomass of water hyacinth, which in turn influence the rate of population increase of the agent.

Where possible, starter colonies for mass-rearing programs should be obtained from neighbouring countries. This reduces both the cost of transporting insects, and the quarantine risks associated with moving insects, and possibly plant material, between regions or continents.

Mass rearing of *Neochetina* weevils has been carried out in large pools, in a technique developed by Commonwealth Scientific and Industrial Research Organisation (CSIRO) Entomology, Australia, and in smaller tubs, as developed by the International Institute of Tropical Agriculture (IITA), Benin.

7.2 Pool rearing

Large pools have been used successfully in a number of countries to rear *Neochetina* spp. for field release. Above-ground metal pools with plastic liners (2–3 m diameter, 60 cm deep, 3000–4000 L capacity) are easily handled and erected by two people, allow easy access to all areas of the water surface, and are large enough to moderate evaporation and overheating. Smaller corrugated iron (approx. 1000 L capacity) have been used successfully in Uganda (Ogwang and Molo 1997), but their smaller size makes them susceptible to overheating. Concrete pools are less easily transported and may need to be lined to prevent the leaching of lime which raises water pH.

Photo: M. Julien

Galvanised iron tanks have been used for rearing *Neochetina* spp. in Kenya

The stages in preparing and inoculating a rearing pool and harvesting *Neochetina* adults are described in Boxes 1a and 1b and in Table 4. Briefly, pools are filled with water and stocked with plants. Adult insects are added, allowed to oviposit, and the larvae develop through undisturbed. After 8–10 weeks the first generation of adults will start to emerge and feed on the leaf surface, and harvesting can commence.

Hand picking of adults from plants is laborious and if done regularly will cause damage to the plants. Insects can be harvested more easily by submerging plants beneath the water surface and collecting the adults as they swim to the surface. A sheet of wire mesh (covering about one third of the pool surface, see Box 1b) is ideal for this purpose and can be managed by one person. Larger mesh sheets covering the entire pool surface have also been used successfully (M. Hill, pers. comm.). Harvesting can continue weekly for approximately 10 months, at which time the pools are likely to require rejuvenation. If managed well the technique produces large numbers of adults with minimal handling. Some of the problems which may be encountered during pool rearing and some suggested solutions are described in Table 5.

Key points

The key to successful pool production is the regular (weekly) collection of adults.

Adult weevils should be harvested from the pool every week, even when not required for release. This will ensure the sustainability of the rearing process. If adults are not removed regularly, larval numbers will increase rapidly

Photo: M. Julien

Insects can be harvested from pools by submerging plants beneath the water surface and collecting adults as they swim to the surface—Angoram, Papua New Guinea

and plants will deteriorate beyond the point of recovery. Weevils which are collected should not be returned to the pools.

Regular and continued maintenance of pools is critical to ensure the continuity of the rearing process.

It is far better to check pools routinely than to try to restore a pool that has deteriorated. Maintenance should be carried out at least fortnightly, and can be done at the same time as insects are harvested. At this time, water levels are checked, plant contaminants removed and the density and condition of plants noted. Fertiliser should be added at intervals of approximately one month.

Fertilise monthly:

Plants require nutrients for healthy growth. To achieve this in rearing pools, between 100 and 200 g of a soluble complete fertiliser containing nitrogen, phosphorus, potassium and trace elements is added monthly to the pool water.

Table 4. Stages in setting up and maintaining a rearing pool

Setting up	
Requirements	▶ pool and liner ▶ level area slightly larger than the pool diameter for each pool ▶ adequate, regular water supply ▶ fine sand or soil for bedding under the pool liner
Prepare site	▶ smooth out bedding sand/soil to 5-10 cm depth ▶ apply broad-spectrum herbicide or layer of salt if weeds are present (e.g., *Cyperus rotundas*, nutgrass) ▶ mark a circle the diameter of the pool. Cut a piece of string to be half the diameter of the pool. Anchor one end of the string where the centre of the pool is to go and extend the string to mark a circle around the central point.
Construct pool	▶ unpack the pool parts, set out the pool wall around the marked circle and join the ends (follow manufacturers instructions) ▶ place the liner into the pool, unwrap around pool sides and press into joins between walls and base to remove creases ▶ fit clips to top of wall to hold liner in place
Prepare for plants	▶ fill pool with water to 20 cm below the top ▶ if the water is chlorinated allow pool to sit for several days to allow sunlight to reduce chlorine level. ▶ check for any leakage
Fertilise	▶ dilute 100–200 g soluble complete fertiliser in a bucket of water ▶ prepare iron supplement (if available) ▶ add fertiliser/s and stir through water column
Add plants	▶ collect field plants that are in good condition (healthy, undamaged) ▶ cover the pool surface with plants so that no water is visible between the plants ▶ remove other plants or insects ▶ allow plants to settle in for at least 3–4 days before adding insects
Inoculate with insects	▶ add approximately 200 young adults per pool ▶ distribute throughout pool

Table 4. (continued)

Harvesting–weekly (commencing 2–3 months after set up or when fresh feeding scars are visible)	
Submerge plants	▶ place mesh over plants and push below surface ▶ hold mesh below surface with weights (stones or bricks) ▶ skim off any weeds floating to surface with a sieve
Collect adults	▶ collect adults rising to the surface with a small sieve ▶ place adults in collecting container ▶ keep collecting container in shade ▶ when no more adults float to surface (after 20–40 minutes) raise mesh and repeat process over a new section of the pool ▶ record number of insects collected in each pool on each sampling date (Box 2) ▶ determine and record number of each weevil species present in collection
Maintenance–fortnightly from set up	
Pool maintenance	▶ check that no water is visible between plants, add new plants if necessary ▶ check water level is within 20 cm from top; top-up if necessary ▶ remove any contamination from other weeds (for floating weeds it is best done when plants are submerged for harvesting) and insects
Monitor plant quality	▶ check that plants are not sinking and look healthy without excessive feeding damage, chlorosis or necrosis
Review adult numbers	▶ if numbers are low for two consecutive weeks consider troubleshooting options
Fertilise–monthly from set up or as required	
Fertilise	▶ dilute 100–200 g soluble complete fertiliser in water ▶ prepare iron supplement (if available) ▶ add fertiliser/s at several points around pool perimeter
Rejuvenating pools - 9-10 monthly	
Clean and restock	▶ collect adults but retain for reinoculating pool ▶ remove water hyacinth plants ▶ drain water ▶ remove sludge build-up from base of pool ▶ check condition of liner, repair or replace as required ▶ set-up pool (see above)

Box 1a. The stages in preparing pools for rearing *Neochetina* weevils

Photo: M. Julien

1. Setting up Above-ground metal pools with plastic liners can be used for rearing. These should be erected on a level surface close to a water supply.

Photo: A. Wright

2. Filling the pools Pools can be filled with town supply water, tank water or water from ponds, rivers or wells. Pools should be filled to within approximately 20 cm of the top and this level should be maintained. If the water supply is chlorinated the filled pools should be allowed to sit for several days.

Photo: M. Julien

3. Mixing fertiliser A soluble complete fertiliser should be added when the pool is set up and monthly thereafter. The fertiliser should be diluted in water.

Photo: M. Julien

4. Adding fertiliser Fertiliser should be added to the pool water and stirred well, not applied to the plant foliage.

Photo: M. Julien

5. Adding plants and insects Water hyacinth plants should be chosen which are healthy with minimal insect damage, and should be crowded into the pool so that water is not visible from above. To commence breeding, a starter colony of 200 adults (100 females) per pool is recommended.

Photo: M. Hill

6. A rearing facility A series of rearing pools set up near Lusaka, Zambia.

Box 1b. The stages in harvesting *Neochetina* weevils from rearing pools

Photo: M. Julien

1. Preparing to harvest Numerous fresh feeding scars seen on young leaves 8–10 weeks after initial stocking indicate that a new generation of adults is emerging and that harvesting should commence.

Photo: M. Julien

2. Mesh used to submerge the water hyacinth A sheet of wire mesh can be used to force plants below the water surface. The edges of the mesh should be covered to prevent damage to the plastic pool liner. Hosepipe around the edges, held in place with tape or string, provides good protection.

Photo: M. Griffiths

3. Placing the mesh The mesh should be placed over the plants and pushed down to submerge the water hyacinth.

Photo: M. Griffiths

4. Submerging the plants The mesh can be held beneath the water surface by weights (heavy bricks or rocks). Unwanted weeds will float to the surface and can be skimmed off.

Photo: A. White

5. Collecting weevils Within a few minutes weevils will float to the water surface where they can be collected using a small strainer. Collection should commence within, at most, 10 minutes of submersion and can continue for 20 to 40 minutes. Insects should be harvested weekly even if not required for release.

Photo: M. Julien

6. Containing collected weevils during harvest Collected adults can be held in plastic containers with several water hyacinth leaves. Care must be taken not to leave the container in direct sunlight during or after collecting as overheating will kill the insects.

Table 5. Troubleshooting during pool rearing

Problem/Symptom	Possible cause	Possible solution
Pool factors		
▶ pool losing water	▷ hole in pool	■ patch hole or replace plastic liner
	▷ excessive evaporation	■ provide shade
▶ water surface visible	▷ too few plants	■ add new plants
▶ excessive algal growth	▷ too much fertiliser	■ reduce amount of fertiliser added
	▷ too few plants allowing sunlight on water surface	■ add new plants
Plant factors		
▶ yellow or light green plants	▷ nutrient deficiency	■ fertilise
▶ blue colour to roots	▷ nutrient deficiency	■ fertilise
▶ spots or streaks of chlorosis or necrosis on leaves	▷ nutrient deficiency	■ fertilise
▶ thinning plant density–water visible between plants	▷ insect density too high	■ add new plants/harvest insects
▶ plants dying and/or sinking	▷ insect density too high	■ add new plants/harvest insects
▶ excessive stunting of plants	▷ insect density too high	■ add new plants/harvest insects
▶ excessive spider mite	▷ plants stressed	■ fertilise, add new plants/harvest insects
Insect factors		
▶ insect production poor	▷ plant quality deteriorated	■ see plant factors above
	▷ too hot	■ provide shade
	▷ too cold	■ insulate pool exterior
▶ high proportion of *N. eichhorniae* in *N. bruchi* pools	▷ natural invasion by *N. eichhorniae*	■ rejuvenate pools and restock with *N. bruchi*, assess whether plant quality has deteriorated or pools require more regular harvesting to limit dispersal, consider moving pools further apart or covering pools
▶ collected adults die before release	▷ overheating	■ keep collecting container in shade during collection and until release

The addition of an iron supplement such as iron sulphate or iron chelate, if available, will further enhance the growth of water hyacinth, which requires higher levels of iron than that provided by most complete fertilisers. The amount of fertiliser required will vary according to the condition of the plants, with more required during periods of active growth. Over-fertilising, especially when plant density is low, promotes algal growth.

Optimise water temperature:

Production from pools is optimal when water temperature beneath the weed mat is approximately 25°C. At cooler temperatures weed growth, insect development and, ultimately, production figures will decline. Electric immersion heaters can be used to heat pools but if not available, insulating material such as grass or straw packed around the sides of the pool may reduce heat loss. Overheating will lead to death of plants and insects. If air temperatures above 35°C are common, partial shading of pools is recommended.

Remove plant contaminants:

Plant species other than water hyacinth should be removed from pools. Submersion of the plants to harvest adults provides an ideal opportunity to remove populations of *Salvinia molesta* (salvinia or water fern), *Azolla* spp., *Lemna* and *Spirodela* spp. (duck weeds) and algae which will float to the surface and can be skimmed off with a sieve. Larger weedy contaminants including *Utricularia* spp. (bladderwort), *Pistia stratiotes* (water lettuce) and larger *Salvinia molesta* plants require hand-weeding.

Remove animal contaminants:

Generalist feeders such as larvae of *Spodoptera* spp. can be excluded with the use of tight-fitting gauze covers placed over the pool at night and held tightly around the pool lip. Spider mite can be controlled using non-insecticidal miticide. Predatory mites are not an effective control option as they do not tolerate submersion during harvesting of *Neochetina* spp. Mosquitoes will breed in pools, causing potential health problems. Insectivorous fish have been tried in pools in Australia but were unsuccessful because the fish, at high densities, also fed upon *Neochetina* pupae.

Maintain high plant density:

At all times plants should cover the pool area so that the water surface is not visible. If the plants are too thin new plants should be added to achieve the correct density. Regular declines in plant density may indicate that adults need to be harvested more frequently or that additional fertiliser is required.

The numbers of adults collected and the relative proportions of the two *Neochetina* species in a pool should be monitored continuously.

In addition to knowing the number of insects being released into the field it is important to monitor the productivity of the pools as an indicator of pool quality. Although insect numbers are likely to fluctuate from week to week, a continual decline over a number of weeks may indicate problems in the rearing process and that the maintenance and harvesting programs should be reviewed.

In addition to total insect numbers it is important to monitor the relative abundance of the two *Neochetina* spp. Experience has shown that if *N. eichhorniae* adults invade a *N. bruchi* pool they often come to dominate. Invasion can occur from nearby natural infestations or from neighbouring pools and is more likely to occur if plant quality in the source pool is poor, encouraging adults to disperse in search of better quality plants. It is important to monitor the numbers of each species collected and, when the levels of *N. eichhorniae* increase to as much as 50% of the population, to review rearing. At high levels it becomes necessary to rejuvenate the pools and restock with *N. bruchi*. At this time it may also be beneficial to move the rearing pools further apart to limit dispersal between pools.

A suggested recording sheet to monitor insect numbers and relative abundance is shown in Box 2.

If managed well, rearing pools should be sustainable for up to ten months.

If pools are kept filled with water and are fertilised, and adults are harvested regularly, the system should be sustainable for 9–10 months. After this time plant quality may start to deteriorate and production figures will decline. Pools should be rejuvenated by draining, cleaning, refilling with water and restocking with new plants and insects (see Table 4). Plants can be collected from field infestations or from neighbouring pools which have been established for shorter periods. The plastic liners can be patched if leaks develop but will probably require replacing after 3–4 years. The longevity of the liner can be extended by maintaining the water level to within 20 cm of the top, reducing the risk of exposure and deterioration of the liner.

Multiple pools

A set-up including 4–6 pools can be maintained and harvested by two or three people working 2–3 days/week and, if well managed, will produce large quantities of insects (425–1100 adults/pool/month) for field release. The average for 5 pools in PNG was about 700 adults/month/pool (see Table 6). When operating a number of pools, cleaning and rejuvenating should be staggered so that not all pools are restarted at the same time. This prevents a decline in the numbers of insects harvested and spreads the workload over a longer period.

7.3 Tub rearing

The tub-rearing technique was initially developed by IITA in Benin and has subsequently been modified for use in other countries (O. Ajuonu, pers. comm.; G. Ochiel, pers. comm.; Ogwang and Molo 1997; van Thielen et al. 1994). The following account is based on these sources.

Photo M. Julien

An operation including four to six pools can be maintained and harvested by two people

Box 2. Rearing productivity sheet — biological control of water hyacinth

Date	No. and percent of each weevil spp.								Total adults	Fertiliser added	Comments
	Pool 1				Pool 2						
	Ne	%	Nb	%	Ne	%	Nb	%			