

**A PROTOCOL FOR THE TRANSLOCATION OF THE FRESHWATER
PEARL MUSSEL
MARGARITIFERA MARGARITIFERA
IN THE RIVER SLANEY AT ENNISCORTHY, COUNTY WEXFORD**

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1.0 Introduction

Translocation is a last resort method of mitigation that is undertaken when mussels are in imminent danger of death or of habitat destruction. There is strong potential for failure to occur due to circumstances linked to the effects of chronic stress during translocation and establishment phase (Dickens *et al.* 2010, Teixeira *et al.* 2007). This protocol relies heavily on the Natural England translocation protocol produced in 2016 by Killeen & Moorkens.

Killeen & Moorkens (2016) outlined 15 likely combined contributors that can lead to the stress and ultimate death of translocated *Margaritifera* (Table 1). Results of previous translocation studies have noted very poor outcomes, with a number of studies reporting from 68% to 100% loss of individuals after 5 years (Figure 1). The process of translocation requires very careful consideration of donor sites in order to choose the habitat most suitable for the best possible outcome. Where a high percentage of mussels in a population or sub-population are likely to be negatively affected, or there is a very low chance of a positive translocation outcome, a precautionary approach is to captive breed a cohort of juvenile mussels from the adults prior to their translocation as this increases the numbers of individuals to be translocated, and very young juveniles have not become conditioned to any one habitat area. Short term captive breeding is described in Moorkens (2017).

Table 1 from Killeen & Moorkens (2016)

Factor Number	Factor	Potential cause of stress
1	Stress levels of donor mussels	Even when mussel habitat is in good condition, a prior negative event
2	Quality of donor habitat	Where donor habitat is excellent, translocated mussels may become stressed by responding to being moved to less optimum habitat. Where donor habitat is poor, mussels may already be stressed and not have the ability to adapt to new environment.
3	Collection and handling quality	Although they appear to be robust, mussels are easily stressed by over-handling, the period of emersion, and the quality of the temporary transport environment. Levels of cool box padding, cooling, and crowding can all contribute to stress.
4	Marking of mussels	In order to monitor translocation success, it is important to be able to clearly mark the mussels. This requires emersion of mussels to dry the shells to label them, which can be a source of stress.
5	Ease of transfer journey	The logistics of how the mussels have to be carried over land and road, the smoothness of the journey and the distance and time needed all contribute to stress levels.
6	Flow pattern differences in donor / receptor habitats	Mussels conditioned to living in fast flows will have strong muscular strength and may pull themselves out of slower flow areas in an attempt to move back to faster flows. Mussels conditioned to slower flows may not have the muscle tone quality to withstand faster flows and may be easily scoured out of the river bed and washed downstream.
7	Innate "righting response"	When mussels are "planted" in their normal two thirds buried position, they have an innate response to pull themselves out of the substrate and rebury themselves. This involves an additional stress and expense of energy reserves.
8	Flow conditions on the day or subsequent days	If translocations are made during high flow conditions or if flows increase significantly following translocation, the mussels are in higher danger of being washed downstream, especially if it follows a "righting" response.
9	Water temperature	Mussels have reduced metabolism and thus ability to move, burrow, right, and otherwise adjust to a more favourable position with decreasing water temperature.

		Very high temperatures are associated with oxygen reduction and mussel stress.
10	Time of year	Mussels have a complex life cycle and spend a high percentage of the year in gamete production. Females brood larval glochidia in their gills between June and September during which time they have reduced capacity for oxygen uptake and are very vulnerable to stress.
11	Similarity of receptor site	As mussels become adapted to their immediate environment, and most do not move during their lifetime, stress can occur from an inability to adapt to a change in flow, depth, turbidity and nutrient levels and of physical substrate type. Thus even a movement from poor habitat to good habitat may have an inevitable intrinsic level of stress.
12	Quality of receptor site	The correct choice of receptor site on a macro and micro scale presents the greatest challenge as all the aspects of appropriate macro and micro habitat need to be present, including appropriate flows at all times of year, suitable substrate conditions for adult and juvenile mussels, appropriate local hydrological function including provision of juvenile food sources, appropriate host fish densities and conditions appropriate to young host fish congregating close to mussels, juvenile mussel habitat in areas where host fish are likely to congregate in early summer, and the stability to maintain their ideal conditions without interruption for at least ten year intervals (time needed for juvenile mussels to be robust enough to withstand flowing open water).
13	Genetic suitability (mussels and fish)	The translocation of mussels should not compromise the genetic component of the receptor site, e.g. it should not bring a different genetic profile to an area that already has mussels of a different genetic adaptation. The translocated mussels should be demonstrated to be compatible with the host fish strain of the receptor locations.
14	Phenotypic suitability	Mussel shape is relatively plastic and adult mussels can form shapes that are well adapted to their river bed conditions, particularly their flow and substrate burial conditions. Preston <i>et al.</i> (2010) recommend that phenotypic characteristics and particularly shell shape variation is taken into consideration when considering the translocation of adult <i>Margaritifera</i> .
15	Future prospects	Any translocation receptor site should have long term resilience and not be likely to be especially vulnerable to the effects of climate change or in an area zoned for intended intensification of development.

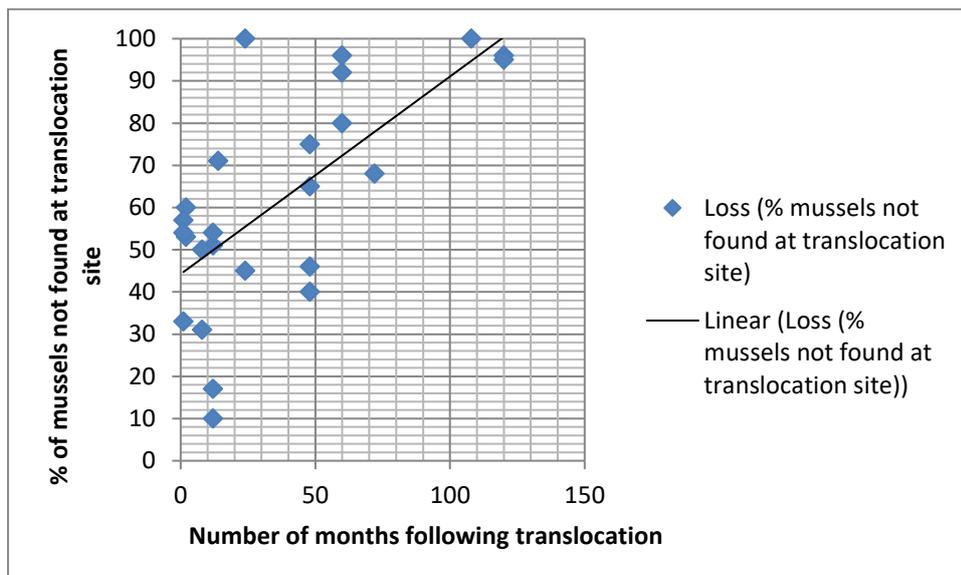


Figure 1 Loss of mussels from translocations over time (From Killeen & Moorkens, 2016)

The IUCN Species Survival Commission has published guidelines for reintroductions and conservation translocations (IUCN/SSC, 2013). The guidelines provide a basis for deciding when translocation is an acceptable option. They specify that risk analysis around a translocation should be proportional to the presumed risks, justification requires an especially high level of confidence regarding the organisms' performance after release, including over the long-term, with reassurance on its acceptability from the perspective of the release area's ecology, and the social and economic interests of its human communities. It notes that in any decision on whether to translocate or not, the absolute level of risk must be balanced against the scale of expected benefits. It concludes that where a high degree of uncertainty remains or it is not possible to assess reliably that a conservation introduction presents low risks, it should not proceed, and alternative conservation solutions should be sought.

Where a translocation is being considered because of a threat from a source other than those that could be considered to be "natural causes", as part of a plan or project, the potential impact of the translocation of the mussels should be considered as part of an NIS, which would then be used to inform the Appropriate Assessment undertaken by the regulating authority under Article 6 of the Habitat's Directive.

2.0 Choosing receptor sites

As the choice of receptor habitat plays a large role in any translocation outcome, it follows that a minimum level of information would be necessary to assist in the receptor site choice.

If sufficient information is known on the flow / hydraulic regime, and if the desk top study indicates that there is sufficient potential for sustainable adult and juvenile habitat, the following protocol for field study can be recommended:

1. Use the desk top study to identify upstream and downstream limits for field studies.
2. A field study should be undertaken in two parts. Firstly, a **winter** high flow bank walkover should be undertaken to ensure the identified stretches do not have high flow constraints – highly drained and dirty inputs and / or chronic suspended solids issues can be clearly identified in these conditions, as can over deepened or bedrock restricted areas leading to excessively high flows. Caution should be taken as high flowing rivers are dangerous and a safe distance should be kept away from the water, which should not be entered during high flows.
3. The second field study should be undertaken during **summer** low flows, and an assessment should be made for river bed habitat suitability and quality, including:
 - a. River bed habitat suitability for adults and juveniles – clast range, compaction, scour levels
 - b. River bed habitat condition – algal and macrophyte levels (Refer to *Margaritifera* regulations 2009)
 - c. Adult mussel numbers present

- d. Near-bed velocity (refer to Moorkens & Killeen, 2014)
 - e. Redox potential (refer to Geist & Auerswald, 2007)
 - f. Suitable receptor sites should be mapped carefully and photographed.
4. A hydrological, hydrogeological and geomorphological risk assessment of the local mini-catchments supporting the proposed translocation sites should then be undertaken to assess the resilience of the local catchment area in its role to protect against sediment and nutrient pollution, and against the exacerbation of drought conditions (particularly through artificial drainage of the upper mini-catchments), and its ability to protect the mussel population through appropriate detritus food production and delivery (sufficient connectivity of undrained land delivering positive juvenile mussel nourishment), and, where appropriate, the replenishment of stone of favourable clast sizes. This study is not constrained by season.

More detailed field studies assessments are summarised in Killeen & Moorkens (2016) and Moorkens (2017). It must be understood that if all investigations at a site gave positive results, it is likely that a good population of *Margaritifera* would be likely to occur there already. However, the balance of positive and negative results provide the best indication not only of which sites are likely to result in success, but also what sort of ongoing conservation management might best improve the location for sustainable juvenile survival over time.

3.0 Protocol for the translocation and monitoring process

3.1 Timing

The translocations cannot proceed without the appropriate licenses (derogation and handling) from NPWS.

If short term breeding is included in the process, mussels are checked for brooding in July and brooding females are transferred to the captive breeding hatchery for approximately 9 weeks, and placed with host fish for the encystment process. Males and adults not destined for captive breeding are translocated directly to the receptor site; brooding females join them 9 weeks later. Mussels are moved in a period of relatively low flow and average air and water temperatures. The best time to carry out a mussel translocation would be from April through to late-June (without captive breeding), and July (with captive breeding). Before this time in winter/early spring the mussels are likely to have a lower metabolic rate and may not respond well to disturbance. From August the mussel glochidia will be too well developed to disturb. Glochidial release is normally in late August or into the first week of September. There is another window of opportunity until mid-October, but the mussels must have time to settle before temperatures decrease and flows increase.

To plan the right timing for the translocation, ensure:

- The translocation is carried out only when the river is relatively low and the turbidity at its lowest.
- If it is not possible to have full visibility at the receptor site, then the work should not be attempted.
- An accurate weather forecast is essential as the work should be carried out when there are clear skies and no heavy cloud cover.
- There should be no forecast for rain on the day of translocation or the subsequent 3 days.
- If the river flow increases before the mussels are settled, then they are very likely to be washed out.
- Where there is more than one translocation site, only complete multiple translocations if they are very close to one another and mussels will not undergo undue stress from delays, otherwise plan for multiple days.
- The translocation exercise should start as soon as there is sufficiently good daylight to allow for a full working day.

3.2 Preparation for the translocation day

Careful preparation is important to ensure that there are no delays that could cause unnecessary stress to the mussels, and that there is sufficient daylight to complete the translocation process.

- Ensure all licenses and permissions have been obtained.
- Ensure you have enough adequately trained and briefed personnel free to carry out the translocation. At least 2 people and preferably 3 should carry out the work and should all be available for the whole day(s). Females for breeding are translocated separately to a hatchery.
- Check that the weather forecast and river conditions are suitable the day before, and sufficient for the translocation day and the subsequent 3 days.
- Visit the translocation site to ensure flows and turbidity levels are low. Mark the selected translocation areas with bright white pebbles.
- Make sure all of the equipment has been gathered together and is ready to load into the vehicle.
- Make sure the vehicle has sufficient fuel for the day before collecting the mussels.

3.3 Collecting and marking mussels

Donor mussels will need to be collected from a wide area, which is time consuming. Two people should work together, one to locate the mussels with a bathyscope and the other to carry them once collected. The mussels should be removed from their substrate and gently placed into a net bag, and not thrown or dropped on top of each other. Emersion should be kept to a minimum and the bag of mussels should be kept within the water during the process to avoid temperature stress. No more than 4 mussels should be placed in each 25cm² net bag.

Before transportation, mussels should be measured (length, using callipers) and labelled using one of the following methods:

- Dymo™ tape with unique numbers attached with/embedded in superglue or epoxy resin. This has been successfully used in several mussel translocations although some tags do become detached or wear and become indecipherable within a short number of years. The procedure does take time, only a few should be dried at any one time and the adhesive also requires time to dry, all of which places stress on the mussels.

- Engraving tool – this does not require the mussels to be dried so emersion is kept to a minimum. However, there have been reports that engraving through the shell periostracum may accelerate erosion of the shell.
- Permanent gel or “gold paint” pen. This method has been used in Germany but again requires thorough drying of the shell both before and after application of the number. Additional dots of pen marks should be made on both valves close to each mussel’s siphon area, so that marked mussels can be seen without lifting them out of the substrate. We have no information on how long the paint remains before wearing off.
- PIT tags – passive integrated transponder PIT tags are small, inert microchips with an electromagnetic coil encapsulated in glass and with a unique code. They are cheap and easy-to-deploy devices used widely as a method of increasing recapture rates and for long-term monitoring, and are increasingly being used to monitor translocated freshwater mussels (e.g. Kurth *et al.* 2007, Wilson *et al.* 2011).

3.4 Transportation of mussels

The key to successful transportation is to provide the mussels with conditions in which stress will be kept to a minimum.

Mussels cannot be moved to or from multiple translocation sites on the same occasion unless the translocation sites are located very close together. If they are separated by excessive distance, and / or accessibility is difficult, or there are delays at the first site, more than one translocation trip is needed to give the mussels the best chance to have a stress-free journey.

The methods used to hold the mussels during transportation depend entirely on the distance being travelled and the ambient temperature on the day.

If the distance to be travelled is less than 20 km and less than 30 minutes driving time then the mussels may be placed in a cool box (or large buckets, or large tanks) on a cushion of towels wetted with river water on top of 2 or 3 ice packs. Box or bucket lids cannot be closed or sealed in any way.

If travel times or distances are greater, or ambient temperature is >20°C then the mussels should be transported in cold boxes (or large buckets, or large tanks) filled with river water. Again the box should be cushioned with towels and the mussels placed in net bags (containing 4 mussels each) to prevent too much movement during transport. If the oxygen in the water is likely to become depleted then battery powered aerators should also be fixed in the boxes. Do not seal down the lid.

It is important to drive straight to the translocation site.

3.5 Placement of mussels in the receptor site

Mussels should be placed in their net bags into the river in cool, shaded flowing water whilst the next phase is underway.

The exact locations of the receptor habitats will have been clearly marked. Care must be taken to ensure that mussels are placed in stable, un-compacted substrate, buried appropriately with siphons facing the flow. Do not force the mussels into the substrate, a trowel can be used to open up a space in the gravels.

The mussels are buried to at least half of their shell length. The presence of a 'tide-mark' formed by algae or a diatom coating may indicate the depth to which they were buried at the donor site. However, if the donor mussels were stressed they have risen to an unnaturally high level in the substrate, and may need deeper burial in a faster receptor site. Even if mussels are correctly buried they may perform a "righting" response, and attempt to lift out of the substrate and rebury again.

In less stable habitats, the placement of some larger clasts around the newly buried mussels may enhance the stability of the substrate.

The mussels should be observed to check that they settle into natural siphon function (should be within one hour).

Take GPS, fixed point references and photographs of site and underwater to assist in relocation of the exact site for monitoring purposes.

Return to the site within the following 2 days to ensure mussels have not dug themselves out and have been washed into totally unsuitable habitat. There may have been some movement and repositioning so a further set of monitoring photographs should be taken.

3.6 Follow-up monitoring

Given the acknowledged poor success rate of translocations, it is very important that there is adequate monitoring to inform ongoing improvements in the translocation process.

Translocated mussels should be monitored as a minimum after one month, six months, one year and then ideally at least annually for five more years (until 6th year post translocation).

The mussels and habitat should be photographed, counted, checked for labels, and their habitat assessed for quality and condition, and redox potential measurements taken.

On the 5th year monitoring round, the habitat area should be checked carefully for emerging juveniles and in a subset of the habitats a demographic excavation of approximately 20 x 20cm should be undertaken.

Juvenile searches should be repeated during the 6th monitoring round.

Annual monitoring should be undertaken in good survey conditions during low flow summer / early autumn conditions.

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