Reconnect

epa Research
Climate - Water - Sustainability
Identifying pressures • Informing policy • Developing solutions

UCD DUBLIN

Issue 2: November 2017

Assessment of the extent and impact of obstacles on freshwater hydromorphology and connectivity in Ireland

Inside this issue:

Reconnect has just moved into its second year. As well as making great progress in many aspects of the research, we are delighted to say that we are engaging with the European project, AMBER (Adaptive Management of Barriers in European Rivers) and our records will contribute to their European atlas of river barriers. Please see page 6 for more information on some of the work being carried out by AMBER.

We have received a huge amount of help from many individuals over the last few months, so we would like to take this opportunity to thank all of the interns that have assisted us with lab and field work over the summer. In particular we would like to thank the staff of Inland Fisheries Ireland for helping us with our electrofishing surveys. The work would not have been done without their help.

Project Team:
Mary Kelly-Quinn[1], Jens Carlsson[2], Bernie Ball[2], Michael Bruen[3], Jonathan Turner[4], Craig Bullock[5], John O' Sullivan[3], Siobhán Atkinson[1,2], Colm Casserly[4].

1. *UCD School of Biology and Environmental Science, University College Dublin*
2. *Area52 Research Group, School of Biology and Environmental Science, Earth Institute, University College Dublin*
3. *UCD Dooge Centre for Water Resources Research, School of Civil Engineering, University College Dublin*
4. *UCD School of Geography, University College Dublin*
5. *UCD School of Architecture, Planning and Environmental Policy, University College Dublin*.

Further details of the project on our website or follow us on Twitter

Historical 25" map drawing of a weir on the Nore (photograph on the right) known as "The Basin". This weir was constructed in the late 1800s. Like many fish passes built at that time, the pass shown in this map is specifically designed for salmon and may not work for other native species (e.g eel, river lamprey, sea lamprey and cyprinids).

Reconnect Fish Surveys

River connectivity and river habitat are both important for sustaining healthy fish populations. Fish require access to spawning habitat in small streams but also need habitat for feeding. The Reconnect project team, together with Inland Fisheries Ireland, has surveyed fish populations upstream and downstream of river obstacles in order to establish (a) whether certain obstacles prevent the upstream movement of fish and (b) whether the habitat changes upstream of weirs (large, deep, slow flowing impoundments) impact on the age structure and density of salmonids.

Whether or not an obstacle in a river is causing a barrier to fish movement can be difficult to determine with electrofishing surveys alone. For example, in the case of brown trout it is difficult to establish impact because they are found throughout most rivers in Ireland, whether there are obstacles in the system or not. This is why we are using the presence of anadromous fish species (fish that move between marine and freshwater environments throughout their life cycle) such as salmon, sea trout and eels as indicators of the passability of an obstacle. Figure 1 shows an example of the results of an electrofishing survey on an obstacle in a tributary of the Slaney river. The fact that salmon were found below the obstacle and not above it suggests that it is an impassable barrier.

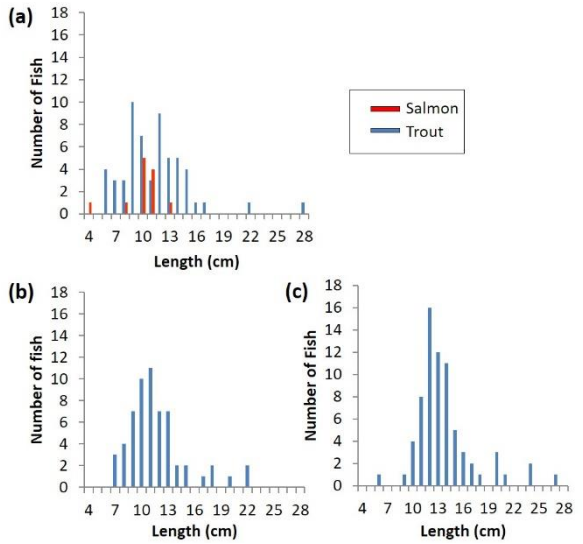


Fig. 1. Graph showing the length frequency distribution of salmon and trout (a) downstream and (b-c) upstream of a ford on a tributary of the Slaney river. Trout were present both upstream and downstream of the ford, while salmon were only found downstream.

Fig. 2. Large brown trout captured during an electrofishing survey.

Fig. 3 Sea trout (left) and flounder (middle) both found below a significant obstacle (right) in one of our study rivers. These are both anadromous fish species and so are good indicators of the migration barrier this particular obstacle is causing. The weir is approximately 2.5m high.

The ins and outs of environmental DNA (eDNA)

Environmental DNA (eDNA) is the collective term for DNA molecules that are released from living or dead organisms in the form of blood, skin, mucous, gametes or faeces and are freely present in the environment. eDNA can be extracted from water or soil samples, and used to detect the presence and relative abundance of target species. Research has shown that eDNA has been successful in detecting species in low numbers (e.g. rare or invasive species). It also has the potential to be a non-invasive alternative to traditional sampling methods.

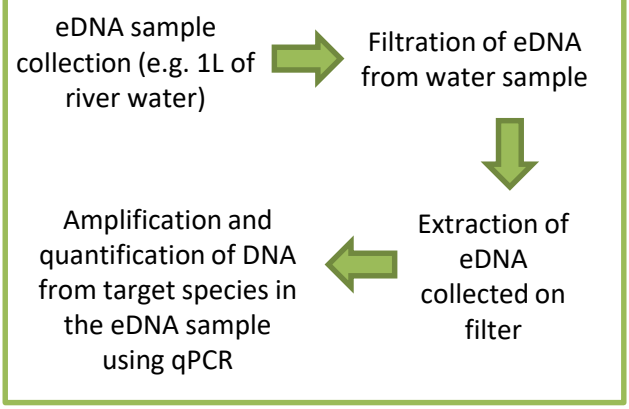
As part of Reconnect, water samples from rivers are collected for eDNA analysis in sterilised water bottles. The water is filtered on site using a pump (Fig. 4). The eDNA collected on the filter is preserved and extracted at a later stage in the laboratory.

To analyse eDNA samples, a technique known as quantitative Polymerase Chain Reaction (qPCR) is used (Fig. 5). Regular PCR, also known as "molecular photocopying", is a common technique used in molecular biology to amplify DNA fragments to obtain millions of copies of DNA. As part of a PCR reaction, *primers* are used to isolate a particular region of the DNA by binding to a complimentary DNA sequence. This in turn guides the DNA replication process. Primers can be designed so that they only bind to DNA sequences that are unique to a particular species. Quantitative PCR is similar to regular PCR, however, in addition to using species-specific primers, a fluorescent species-specific *probe* is added to the reaction. Similar to the primers, the probe will only bind to target sequences of DNA which are unique to a particular species. If DNA from this species is present in the eDNA sample, the probe will fluoresce as the target sequence is amplified. The DNA is quantified against a standard curve, which is generated using known concentrations of reference DNA from a sample of the target species (typically acquired from tissue, e.g. muscle, scales or fin clippings). If no DNA from the target species is present in the eDNA sample, the probe will not bind to DNA, and therefore will not fluoresce during the qPCR. The use of primers and probes in qPCR allows for increased specificity and sensitivity of the reaction.

Reconnect and eDNA

As part of Reconnect, eDNA water samples have been taken upstream and downstream of river obstacles. Primers and probes have been previously developed and deployed for Atlantic salmon (*Salmo salar)*, brown trout (*S. trutta*), freshwater pearl mussel (*Margaritifera margaritifera*), and sea lamprey (*Petromyzon marinus*). As part of Reconnect, primers for twaite and allis shad (*Alosa fallax*, and *A. alosa*), and for the white clawed crayfish (*Austropotamobius pallipes*) have been developed, and will be deployed in selected rivers.

The basic process of eDNA collection and detection from a water sample



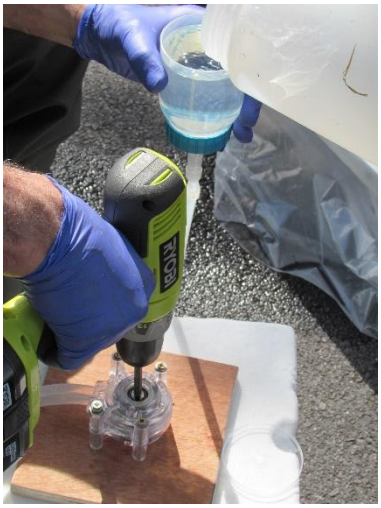
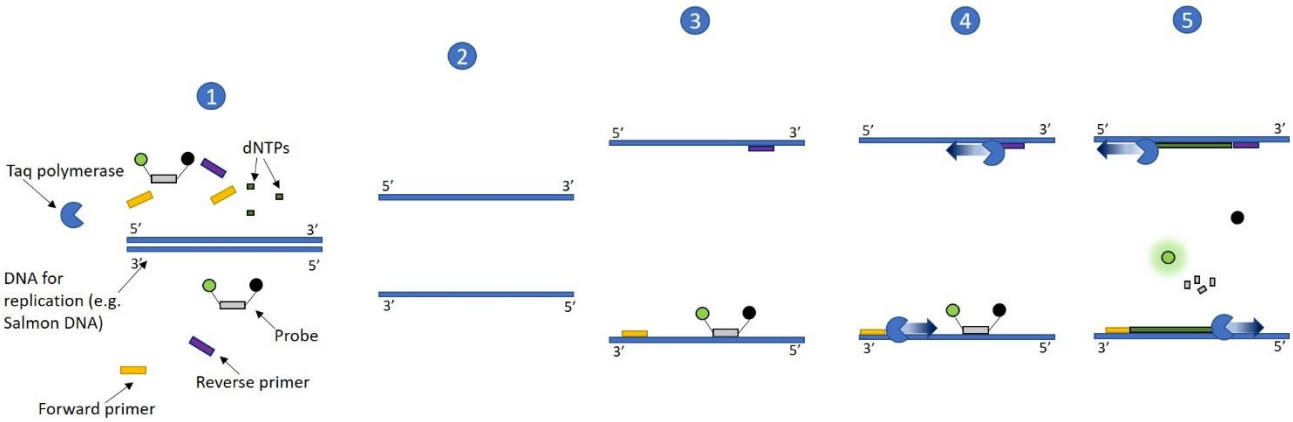


Fig. 4. Bernie Ball from the Reconnect project team filtering an eDNA water sample on site using a peristaltic pump powered by a hand drill.



1. Double-stranded DNA for amplification is mixed with qPCR reagents including species specific primers and probe. The probe has a fluorescent reporter dye (pictured in green) and a quencher (black) attached to it. The quencher prevents the reporter dye from fluorescing.
2. Double-stranded DNA is denatured into single-stranded DNA using high temperatures
3. The qPCR temperature is lowered and the probes and primers bind to specific regions of the single-stranded DNA
4. The enzyme Taq polymerase synthesises complimentary DNA strands using dNTPs (the building blocks of DNA) and the forward and reverse primers as starting points.
5. As it encounters the probe, the Taq polymerase enzyme digests the probe, releasing the reporter dye and quencher. The reporter dye will fluoresce once it is separated from the quencher.

Fig. 5. Diagram showing the steps that occur during a qPCR run. Steps 2-5 are repeated throughout the qPCR run, resulting in exponential amplification of the target DNA sequence. The fluorescence can be measured, giving an estimate of the relative biomass of a particular species in a water sample. As mentioned above, probes and primers will not bind to non-target DNA. In such a case, no amplification would occur, and therefore no fluorescence would be observed. This is why it is necessary to have a quencher attached to the probe as well as a fluorescent reporter dye.

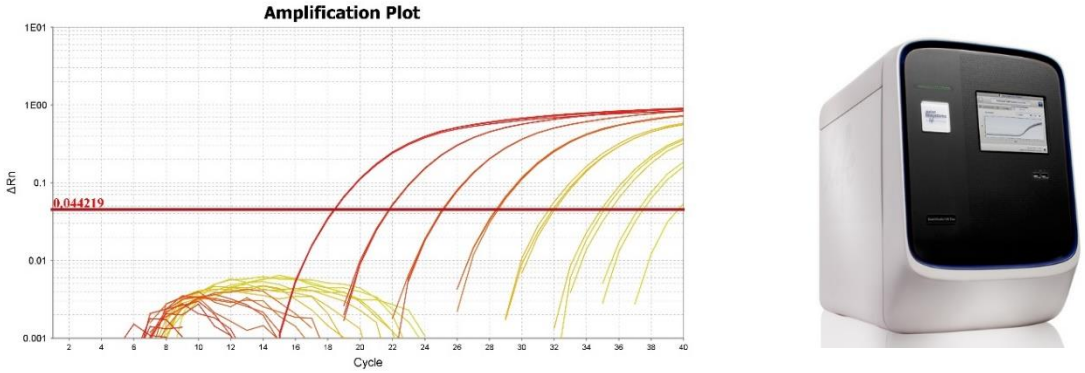


Fig. 6. A sample output from a qPCR (machine pictured on right) run. The graph shows a serial dilution (1 in 10) of salmon DNA (with a known starting concentration). The standard curve generated from this can be used to calculate salmon DNA concentrations from eDNA samples.

River Obstacles and Citizen Science via a Mobile App – an update

The River Obstacles app was introduced to Ireland just over a year ago. Since then, 98 river obstacle record uploads have been recorded in 16 counties (Figs. 7 & 8). The data will be used to help in the development of a national georeferenced map of river obstacles in Ireland, which will ultimately be used during the systematic prioritization of obstacles for removal or modification.

The app is free to download and easy to use. One simply takes a photograph of the obstacle and notes some details. The app uses the GPS facility built into a smartphone to record the location. It is also possible to upload details on obstacles via the online platform on the River Obstacles website here. The river obstacles app can be downloaded from Google Play and the iTunes App Store. Links to the app can also be found on the Reconnect Facebook page. A huge thank you to everyone who has contributed to this research so far!

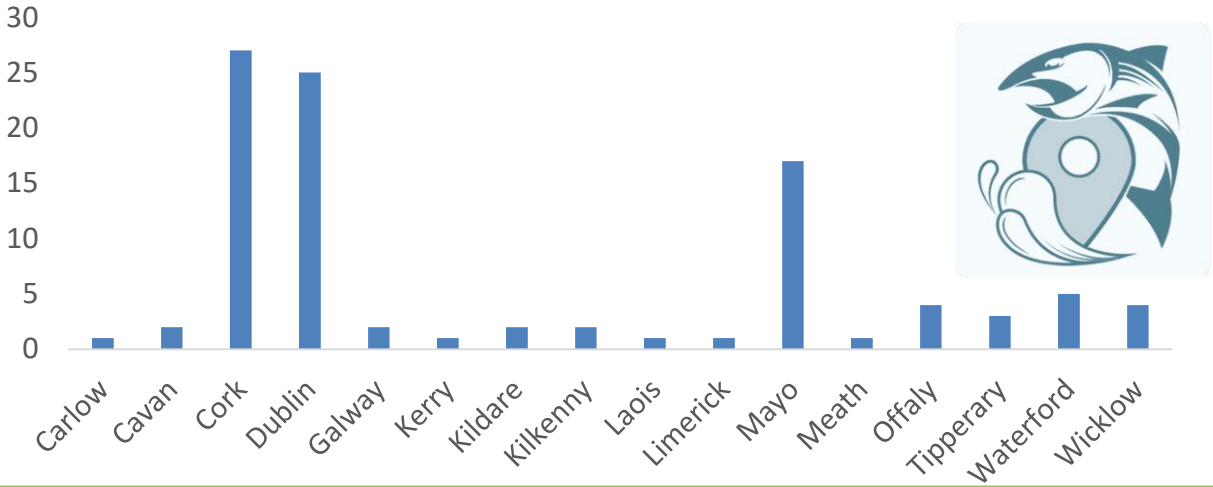


Fig. 7. A graph showing the number of river obstacle records per county (July 2016 – October 2017)

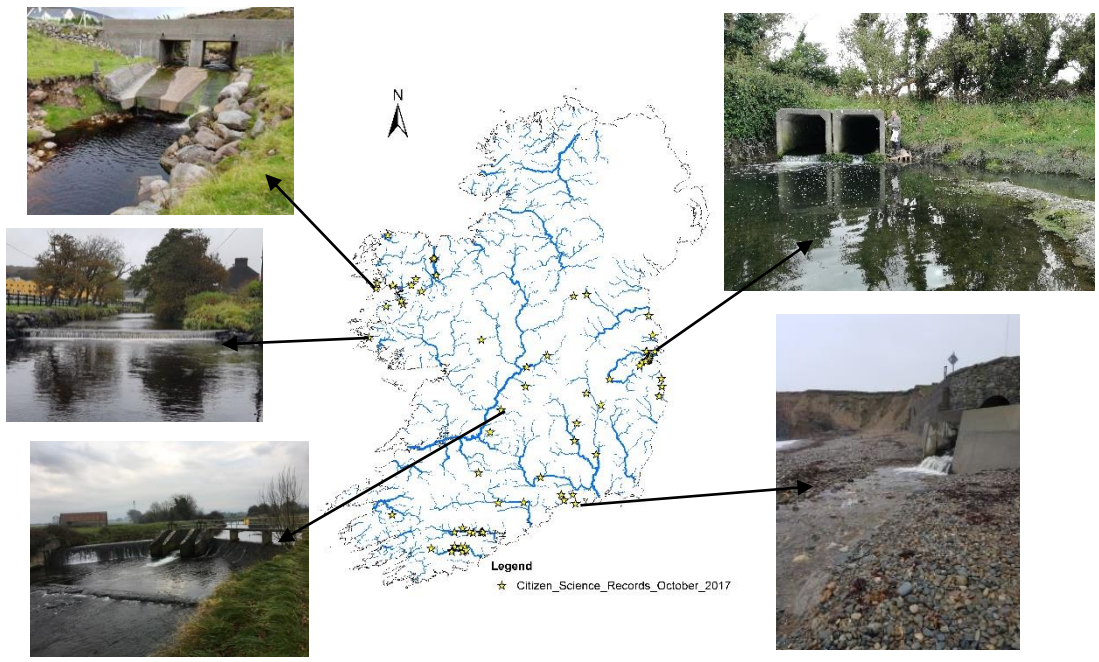


Fig. 8. A map of Ireland showing the locations of the River Obstacle app uploads, with some examples of the types of obstacles recorded.

The AMBER Project – a European wide project addressing river obstacle issues

Prepared by Joost van Deelen

This project has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No. 689682.

The AMBER (Adaptive Management of Barriers in European Rivers) project seeks to apply adaptive management to the operation of dams and barriers in European rivers to achieve a more efficient restoration of stream connectivity, and address impacts caused by river fragmentation. High level of river fragmentation is a key driver for not reaching good ecological status in many water bodies in the European Union. The AMBER project will deliver valuable knowledge and outputs on river fragmentation in Europe that could contribute to the review discussion on the Water Framework Directive (WFD).

The specific objectives of the AMBER project are to:

1. build a **Europe-wide atlas of barriers** to the migration and movement of fish and other aquatic organisms.
2. develop **state of the art methods to monitor barrier passability** and **methods to manage barriers** to reduce ecological effects (this includes aspects like planning, mitigation, removal of barriers).
3. develop adaptive and comprehensive barrier management and decision-making **guidance for NGOs, regulators and industry** incorporating: ecological impacts, cost-benefit analysis, sociological and economic factors, and ecological/economic modelling.

AMBER is a research project with a budget of €6.2 million euro, funded through the European Union's Horizon 2020 Research and Innovation Programme. The project consortium consists of 20 institutions. They include large hydropower businesses, rivers authorities, non-governmental organisations, universities and the European Joint Research Centre. These institutions are spread throughout Europe including Poland, Italy, Germany, UK, Ireland, Netherlands, Spain, France, Switzerland, Denmark and Sweden.

A large number of stakeholders and end-users of the various outputs of the AMBER project have been identified representing interests not only in every single European Member State, but also in several European Free Trade Association countries.

The project has started on the 1st of June 2016 and will end on the 31st of May 2020. To stay up to date on progress, visit http://amber.international/ or follow AMBER on Twitter and Facebook or sign up for the newsletter.

For more information and updates on the Reconnect project, click on the below icons:

Project contact: Assoc. Prof. Mary Kelly-Quinn
School of Biology & Environmental Science, Science Centre West, UCD, Belfield, Dublin 4, Ireland.
Tel: +353 (0)1 716 2337 Email: mary.kelly-quinn@ucd.ie

Newsletter Editor: Siobhán Atkinson