Moyvannan Electricity Substation

Environmental Impact Assessment Report

Annex 5.3: Aquatic Survey Report

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Aquatic ecological assessment of the Cross River, northwest of Athlone, Co. Roscommon

Prepared by Triturus Environmental Ltd.

for SLR Consulting Ireland Ltd.

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1. Introduction

1.1 Background

Triturus Environmental Ltd. were commissioned on behalf of SLR Consulting Ireland Ltd. to prepare a baseline fisheries and aquatic assessment of the Cross River, northwest of Athlone, Co. Roscommon (**Figure 2.1**). The survey was required to inform aquatic ecological constraints in light of a proposed grid cable route (GCR) crossing of the Cross River. The survey area was not situated in a European site. The closest downstream European sites with hydrological connectivity to the study area were the River Shannon Callows SAC (000216) and Middle Shannon Callows SPA (0004096). Both European sites are 10.8km downstream of the study area. The collated ecological data on aquatic species and habitats would inform the EIAR and NIS preparation for the proposed project.

A site visit to the Cross River was undertaken by Triturus Environmental Ltd. on the 7th February 2024. The surveys documented the physical habitat of the Cross River to determine its value as a nursery, spawning and holding area for fish of high conservation value. The survey would supplement pre-existing fisheries knowledge of the catchment collected by Inland Fisheries Ireland. The current survey also included environmental DNA (eDNA) collection to detect the presence of fish of high conservation including brook lamprey (*Lampetra planeri*), European eel (*Anguilla anguilla*) and Atlantic salmon (*Salmo salar*). Brown trout (*Salmo trutta*) were not tested for as they are known from the catchment based on historical fisheries surveys (Gordon et al. 2023; Kelly et al. 2017; Kelly et al. 2010). Furthermore the eDNA sampling also tested for white-clawed crayfish (*Austropotamobius pallipes*) given the survey was undertaken outside the recommended period for physical searching for this species. Additionally, the site survey documented the aquatic macrophyte and bryophyte assemblages of the Cross River inclusive of Annex I Habitat associations including floating river vegetation and hydrophillous tall herb. A macro-invertebrate sample was also collected to determine the presence of rare invertebrate species and also to determine biological water quality by Q sampling.

1.2 Fisheries asset of the survey area

The Cross River is a renowned recreational brown trout (*Salmo trutta*) fishery although historical drainage works (as recent as 2001) have impacted the fisheries habitat (O'Reilly, 2009). In addition to brown trout, the river is known to support perch (*Perca fluviatilis*), pike (*Esox lucius*), gudgeon (*Gobio gobio*), roach (*Rutilus rutilus*) and roach x bream hybrids (*R. rutilus* x *Abramis brama*) (Kelly et al., 2017; 2010). Lamprey (*Lampetra* sp.) and stone loach (*Barbatula barbatula*) are known in the lower reaches of the Cross River, whilst the heavily modified upper reaches support three-spined stickleback (*Gasterosteus aculeatus*) (Triturus 2021-2023 data). Lamprey (Lampetra sp.) are known from the Cross River and its tributary, Barr's Drain, in grid square M94 (Triturus, unpublished data).

1.3 Protected aquatic species

A comprehensive desktop review of available data from the National Parks and Wildlife Service (NPWS), National Biodiversity Data Centre (NBDC), Inland Fisheries Ireland (IFI), Botanical Society of Britain and Ireland (BSBI), Environmental Protection Agency (EPA) and Triturus databases for the 10km grid squares containing and adjoining the study area (i.e. M94) identified a low number of records for rare and or protected aquatic species within the vicinity of the proposed wind farm.

A sparse number of records for Annex II white-clawed crayfish (*Austropotamobius pallipes*) were available for 10km grid square M94, with the species known from the Cross River downstream of the study area but not overlapping the proposed GCR crossing.

A single record for the protected short-leaved water starwort (*Callitriche truncata*) was available for 2020 for Lough Ree but not in the Cross River (10km grid square M94) although the species is often found washed up in small patches along the shoreline with source populations not clearly identifiable (Paul Green, pers. comm.). However, whilst this macrophyte species is listed under the Flora (Protection) Order 2022 (S.I. No. 235/2022) and considered 'vulnerable' in Ireland (Wyse-Jackson et al., 2016), there was no hydrological connectivity with the proposed project and its known locations.

1.4 EPA biological water quality data

There were two contemporary EPA biological monitoring stations on the Cross River downstream of the study area. Water quality was recorded as of good status (**Q4**) at a bridge site near Burnbrook (station RS26C100200) in 2023. Further downstream at a bridge upstream of the River Shannon confluence (station RS26C100400) the Cross River was of poor status (**Q3**).

The Cross River, comprises of the Cross_010, Cross_020, Cross_030 and Cross_040 river waterbodies, was of moderate status in the 2016-2021 period and was thus 'at risk' of not achieving good status. Agriculture, peat escapement and historical drainage are the main water quality pressures along the Cross River (EPA, 2019).

2. Methodology

2.1 Fisheries Assessment

A broad appraisal of the riverine habitat of the Cross River at the proposed GCR crossing (ITM 596174, 744372) was undertaken on the 18th December 2022 to evaluate the general fisheries habitat. The fisheries assessment was carried out by characterising the survey area (channel profile, flow profile, substrata & riparian habitat) relative to the known core determinants of good supporting fisheries habitat and or associated pressures (IFI, 2020; O' Grady, 2006; EA, 2003; Hendry & Cragg-Hine, 1997). This supported a broad overview of the general fisheries importance of the area by carrying out an assessment of the distribution and condition of the supporting habitats (i.e. nursery, spawning and holding) to inform the overall importance of the study area for fish. This was considered relative to the known habitat requirements of fish of high conservation value including salmonids, lamprey and European eel.

2.2 eDNA analysis

In order to support the physical fisheries habitat assessment, two no. composite water samples were collected from the Cross River on the 7th February 2024. The samples were analysed for Atlantic salmon, lamprey and European eel eDNA.

In accordance with best practice, a composite (500ml) water sample was collected from the sampling point, maximising the geographic spread at the site (20 x 25ml samples at each site), thus increasing the chance of detecting the target species' DNA. The composite sample was filtered on-site using a sterile proprietary eDNA sampling kit. The fixed sample was stored at room temperature and sent to the laboratory for analysis within 48 hours of collection. A total of n=12 qPCR replicates were analysed for each sample. Given the high sensitivity of eDNA analysis, a single positive qPCR replicate is considered as proof of the species' presence (termed qPCR No Threshold, or qPCR NT). Whilst an eDNA approach is not currently quantitative, the detection of the target species' DNA indicates the presence of the species at and or upstream of the sampling point. Please refer to Appendix A for full eDNA laboratory analysis methodology.

2.3 Q Sampling

A single macro-invertebrate sample was collected downstream of the bridge crossing and converted into a Q-rating as per Toner et al. (2005). The sample were taken with a standard kick sampling hand net (250mm width with, 500µm mesh size) from riffle/glide habitat utilising a three-minute sample effort. This included the washing of large cobble and or small boulder at each survey site, where present. All samples were elutriated and fixed in 70% ethanol for subsequent laboratory identification. Any rare invertebrate species were identified from the NPWS Red List publications for beetles (Foster et al., 2009), stoneflies (Feeley et al., 2020a), mayflies (Kelly-Quinn & Regan, 2012) and other relevant taxa (i.e. O'Connor, 2020; Byrne et al., 2009; Nelson et al., 2011).

An Ecological Quality Ratio (EQR) was also calculated for the riverine sample. This allows for the conversion of a Q rating class to a numerical value to correspond with targets as specified within the European Communities Environmental Objectives (Surface Water) Regulations (S.I. No. 272 of 2009)

as amended by the S.I. No. 77/2019. An EQR Ratio is expressed by a numerical value between 0 and 1 in the case of Q sampling by dividing the recorded Q rating by the maximum reference value (i.e. Q5 or 1.0 when converted to a numerical value). In the case of the Surface Water Regulations 2019, minimum targets for rivers are specified as 0.75 for Good Status (equivalent Q4) and 0.85 for High status (equivalent Q 4-5) (**Table 2.1**).

As such, the severity of anthropogenic pollution can be determined based on deviation from target reference conditions (Feeley et al. 2020b). In this respect, 'High status' is defined as the biological, chemical and morphological conditions associated with no or very low human pressure, while at the other extreme 'Bad Status' would be representative of severe anthropogenic pressures on a river.

Table 2.1 Reference Categories for EPA Q-Ratings (Q1 to Q5)

Q Value	EQR	WFD Status	Pollution Status	Condition
Q5 or Q4-5	≥0.9	High Status	Unpolluted	Satisfactory
Q4	0.8	Good Status	Unpolluted	Satisfactory
Q3-4	0.7	Moderate Status	Slightly polluted	Unsatisfactory
Q2-3 or Q3	0.5-0.6	Poor	Moderately polluted	Unsatisfactory
Q1, Q1-2 or Q2	0.2-0.4	Bad	Seriously polluted	Unsatisfactory

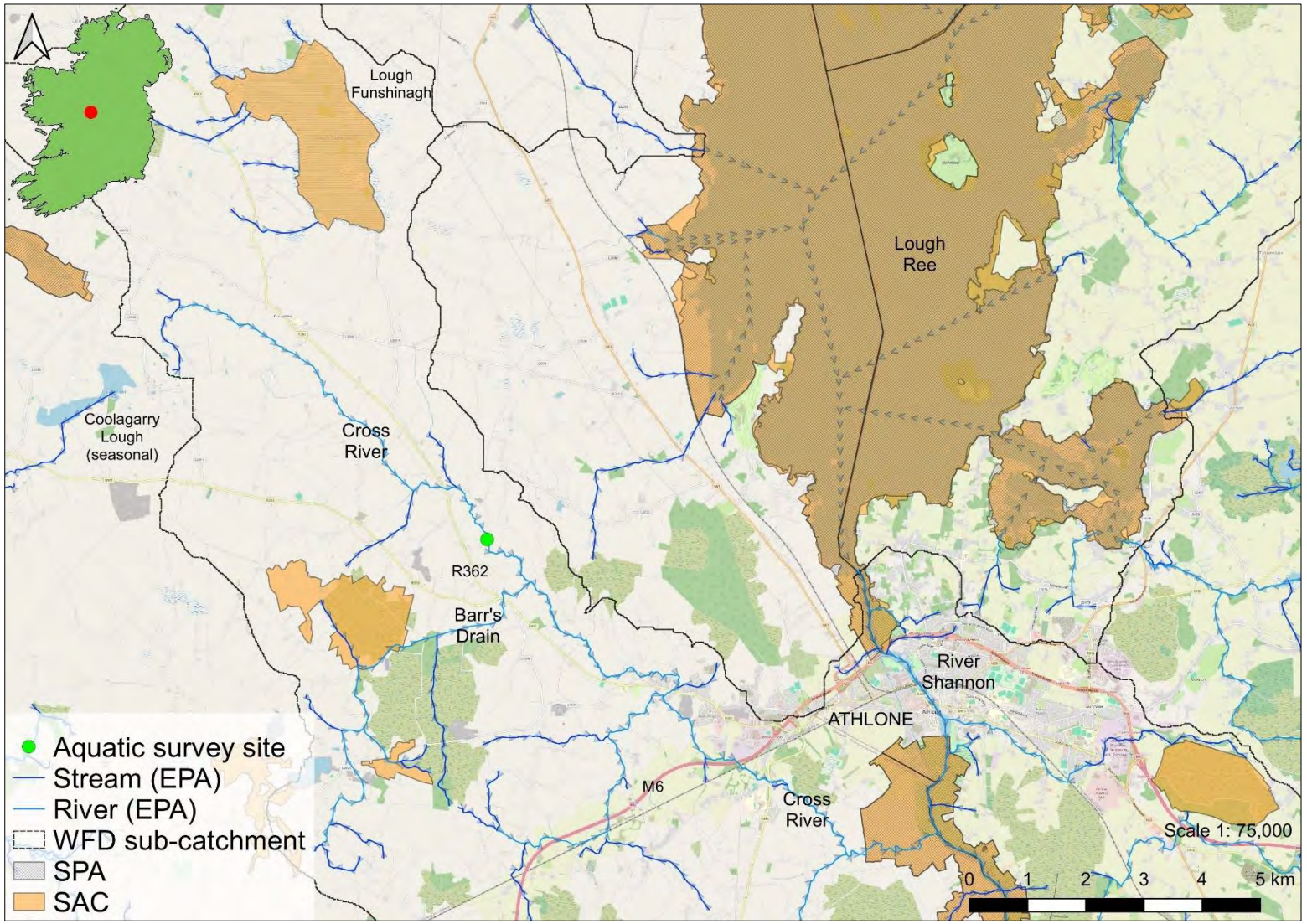

Figure 2.1 Location of the survey areas on the Cross River, northwest of Athlone, Co. Roscommon

3. Results

3.1 Aquatic Habitat Description

The Cross River was representative of very swift flowing lowland depositing watercourse (FW2). The watercourse was 5-6m wide and between 0.3-0.6m deep (**Plates 3.1-3.2**). The river had 1.5-2m high banks and was historically realigned and deepened with boulder bank reinforcements adjoining the bridge crossing. Despite historical alterations the Cross River exhibited good recovery and retained a semi-natural flow profile dominated by swift flowing riffle and glide with localised pool. The bed comprised of compacted small boulder and cobble with pockets of mixed coarse, medium and fine gravels with localised sand. Silt pockets were also present locally in pool slacks. The bed had moderate siltation despite high energy (silt plumes underfoot)with superficial silt deposition in the channel margins. Given the higher energy of the channel it only supported submerged water parsnip (*Berula erecta*) with localised water mint (*Mentha aquatica*) in the margins. The instream boulders supported the moss *Rhychostegium riparioides* and *Cinclodotus fontnaloides*. The macrophyte and bryophyte community was thus not representative of the Annex I habitat, Water courses of plain to montane levels with the Ranunculion fluitantis and Callitricho-Batrachion vegetation [3260].

The riparian areas supported occasional reed canary grass (*Phalaris arundinacea*), grey willow (*Salix cinerea* sp. *oleifolia*) and gorse (*Ulex europaeus*). The channel was bordered by improved pasture (GA1) for sheep grazing that was the dominant bordering land use. While the channel had some value for white-clawed crayfish (*Austropotambius pallipes*) none were recorded present and no crayfish remains were observed in otter spraint recorded in the vicinity of the bridge crossing. The eDNA sample also returned a negative result for crayfish (**section 3.3**) supporting the absence of the species in the survey area despite known historical records downstream (refer to desktop review).

The Cross River in the vicinity of the GCR crossing was a good quality salmonid nursery with mixed cohorts brown trout observed during the survey supporting the known value of the river as a brown trout fishery. The mixed coarse substrata and swift flows provided ample refugia for salmonids. However, the moderate sedimentation and compaction of the bed reduced spawning quality to moderate despite locally good patches of spawning habitat being present both upstream and downstream of the bridge. Holding habitat was good locally in deeper glide and in pool. The channel had some moderate quality eel habitat that was reduced because of the compaction of coarse bed substrata. The eDNA sampling did not detect eel supporting the species absence from the river. The channel had localised lamprey ammocoetes burial habitat despite high energy restricted to superficial silts. Improved burial habitat may exist further upstream and or downstream of the study area given that lamprey distribution is often patchy in rivers that have had historical drainage alterations. The channel however supported good spawning for brook lamprey and the eDNA showed a strong positive trace of brook lamprey (**section 3.4**).

Plate 3.1 Cross River upstream of the bridge crossing

Plate 3.2 Cross River downstream of the bridge crossing

3.2 Otter Survey

Otter spraint was recorded c. 10m downstream of the bridge on marginal boulders (ITM 596174, 744360) and on the concrete ledge under the bridge structure (ITM 596171, 744367). The otter spraint had both roach and salmonid remains but not white-clawed crayfish. No other otter signs were recorded within 150m of the proposed GCR crossing inclusive of breeding and or resting areas. While there was some suitability for an otter couch area under the dry western arch (given secluded void space) no otter signs were recorded.

Plate 3.3 Otter spraint with salmonid and roach remains

3.3 Biological Water Quality (Q Sampling)

A Q-samples was collected at a single location downstream of the road bridge crossing on the Cross River on the 7th February 2024. The species composition was converted into Environmental Protection Agency (EPA) Q rating by grouping the species assemblage into water quality classes based on their known pollution sensitivities. Following the methodology of Toner et al. (2005), the Environmental Protection Agency (EPA) group invertebrates into classes whereby pollution intolerant species are denoted class A, and species with greater pollution tolerance fall into successive classes (B through E, respectively). As such, the presence or absence of these groups and their relative abundance facilitates an assessment of biological river health. Good status (Q4) unpolluted water quality is achieved according to the EPA if at least one Group A taxon is present in, at least, fair numbers (5-10% total sample composition). Group B taxa may be common or absent and *Baetis rhodani* (large dark olive mayfly) is often dominant (Toner et al. 2005). Other Group C taxa are never excessive and group D / E taxa are present in small numbers or absent (Toner et al., 2005). Our results are discussed in this context to interpret potential changes in the macroinvertebrate community composition. Furthermore, the Q sample results were converted into an Ecological Quality Ratio to reference with

the standards specified in the European Communities Environmental Objectives (Surface Water) (Amendment) Regulations S.I. No. 77/2019.

The invertebrate sample from the Cross River had fair numbers of EPA group A mayfly species including both *Ephemera danica* and *Heptagenia sulphurea* (i.e. very clean water indicator species). The Cross River also supported the clean water indicator cased caddis species *Silo pallipes* and *Agapetus fuscipes*, both being clean water EPA group B species. The sample also had numerous EPA group C (moderate water quality indicator species) including the caseless caddis *Hydropsyche instabilis* and the riffle beetle species *Elmis aenea* and *Limnius volkmari.* The invertebrate sample at the Cross River had a representative Q-rating of **Q4 (good status**) based on the sample composition recorded (i.e. good numbers of clean water Group A and B species). Rivers with Q4 ratings have an equivalent EQR of **0.8** and therefore meets the target EQR of ≥0.75 good status target of the Water Framework Directive (2000/60/EEC) as prescribed by the European Communities Environmental Objectives (Surface Water) (Amendment) Regulations S.I. No. 77/2019. No rare invertebrate species were recorded according to the NPWS Red List publications for beetles (Foster et al., 2009), stoneflies (Feeley et al., 2020a), mayflies (Kelly-Quinn & Regan, 2012) and other relevant taxa (i.e. O'Connor, 2020; Byrne et al., 2009; Nelson et al., 2011) (**Table 3.1**).

Table 3.1 Macro-invertebrate species composition for the Cross River northwest of Athlone

Taxon	Family	Binomial name	Abundance	EPA Groups
Ephemeroptera	Ephemeridae	*Ephemera danica*	2	**A**
Ephemeroptera	Heptageniidae	*Heptagenia sulphurea*	5	**A**
Trichoptera	Goeridae	*Silo pallipes*	1	**B**
Trichoptera	Glossosomatidae	*Agapetus fuscipes*	2	**B**
Ephemeroptera	Baetidae	*Baetis rhodani*	9	**C**
Trichoptera	Hydropsychidae	*Hydropsyche instabilis*	3	**C**
Crustacea	Gammaridae	*Gammarus duebeni*	22	**C**
Coleoptera	Elmidae	*Elmis aenea*	6	**C**
Coleoptera	Elmidae	*Limnius volckmari*	1	**C**
Coleoptera	Gyrinidae	Gyrinidae larva	1	**C**
Diptera	Simuliidae	sp. indet.	5	**C**
Abundance			**57**	
Taxon Richness			**11**	
Q-rating			**Q4**	
Ecological Quality Ration (EQR)			**0.7**	
WFD status			**Good**	

3.4 eDNA Analysis

Very strong eDNA signatures were present for brook lamprey in the Cross River (12 out of 12 qPCR replicates, for both) (**Table 3.2; Appendix A**). This is considered as evidence of the presence of the species in the vicinity of the study area. No salmon, eel or white-clawed crayfish eDNA was detected for brook lamprey at either the upstream or downstream sites (i.e. 0 out of 12 qPCR replicates for each sample). This is considered as evidence of these species absence from the study area (**Table 3.2**).

Table 3.2 eDNA results from samples collected from the Cross River, northwest Athlone, Co. Roscommon (positive qPCR replicates out of 12 in parentheses)

Laboratory Sample ID	Target Species	Sample Integrity Check	Number Positive qPCR Replicates
11689	White-clawed crayfish (*Austropotamobius pallipes*)	Pass	Negative (0/12)
	European eel (*Anguilla anguilla*)	Pass	Negative (0/12)
	Atlantic salmon (*Salmo salar*)	Pass	Negative (0/12)
	Brook Lamprey (*Lampetra planeri*)	Pass	**Positive (12/12)**

4. Discussion

The Cross River at the study area was a semi-natural lowland depositing watercourse (FW2) that had evident historical drainage modifications. Nonetheless, the Cross River remained of high value for fish of high conservation value. Brook lamprey (*Lampetra planeri*) were detected via eDNA sampling and with good spawning and moderate quality nursery habitat for the species present in the vicinity of the proposed GCR crossing. While nursery habitat was 'patchy' (limited to more localised superficial sand and silt) spawning habitat was more extensive given mixed medium and fine gravels between coarser bed substrata. However, in pools and depositional areas >100m downstream of the crossing more significant lamprey habitat exists (pers. obs.)

Good quality spawning and nursery habitat for brown trout was also present both upstream and downstream of the proposed GCR crossing, with valuable holding areas for migratory adults present downstream (in deeper glide and pool habitat). No Atlantic salmon were recorded in eDNA and while salmon can and enter parts of the middle Shannon including the River Suck the species densities are either very low or the species does not occur in the Cross River. The species has not been recorded by Inland Fisheries Ireland from the river in historical surveys (Gordon et al. 2023; Kelly et al. 2017, Kelly et al. 2010).

The survey area (especially deep glide) downstream of the proposed GCR was of moderate suitability for Red-listed (King et al., 2011) and critically endangered (Pike et al., 2020) European eel, but the species was not recorded in eDNA sampling. Downstream barriers including Meelick Weir and Ardnacrusha Dam restrict the passage of eel into the middle River Shannon catchment and likely explains the very low density or absence of eels in the study area.

Although some good habitat suitability was present in terms of instream refugia for white-clawed crayfish (i.e. boulders and cobble), none were recorded during the survey in eDNA sampling. Furthermore, no crayfish remains were detected in otter spraint. However, historical crayfish records exist for the Cross River downstream of the study area and thus populations may exist further downstream (NBDC & NWPS data). Additionally, no other rare or protected macro-invertebrate species (according to national red lists) were recorded in the samples taken from the Cross River at the study area. In terms of biological water quality, the Cross River achieved **Q4** (good status) due to the presence of fair numbers of pollution sensitive (EPA group A) mayflies and also cased caddis (EPA group B). Thus, the Cross River in the study area was meeting the target good status (≥Q4) requirements of the European Union Environmental Objectives (Surface Waters) (Amendment) Regulations 2019 and the Water Framework Directive (2000/60/EC).

Two otter spraint sites were recorded under the bridge crossing and on marginal boulders downstream of the bridge crossing on the Cross River. The survey area was considered to provide good foraging and commuting habitat although no breeding and or resting areas were recorded within 150m of the proposed GCR crossing, likely due to more limited riparian cover (much of the banks downstream of the crossing were sparse and open. A search of the riparian boulder revetments and dry arch of the bridge adjoining the GCR crossing did not identify any potential holt sites. Furthermore, the very hard ground of the modified banks, compacted during historical drainage works also offered limited the potential for holt excavation on the riverbanks.

The macrophyte and bryophyte community was thus not representative of the Annex I habitat, Water courses of plain to montane levels with the Ranunculion fluitantis and Callitricho-Batrachion vegetation [3260].

Overall, the high value fisheries habitat (including brown trout and lamprey) in the context of the Shannon catchment and the presence of important water dependant species such as otter inclusive of historical crayfish records indicates the Cross River is of County Importance.

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6. Appendix A – Copy of eDNA laboratory report

eDNA Analysis

Summary

When aquatic organisms inhabit a waterbody such as a pond, lake or river they continuously release small amounts of their DNA into the environment. By collecting and analysing water samples, we can detect these small traces of environmental DNA (eDNA) to confirm the presence or absence of the target species within the waterbody.

Results

Lab ID	Site Name	OS Reference	Target Species	Sample Integrity Check	Result	Positive Replicates
11689	Cross River		Atlantic salmon	Pass	Negative	0
			Brook lamprey	Pass	Positive	12
			European eel	Pass	Negative	0
			White-clawed crayfish	Pass	Negative	0

Matters affecting result: none

Reported by:pos

Approved by: Chelsea Warner

Triturus

Folio No:	75-2024
Purchase Order:	FEB24_CROSS
Contact:	Triturus Environmental Ltd
Issue Date:	27.02.2024

SureScreen Scientifics

Methodology

Samples have been analyzed for the presence of target species eDNA following readily available and scientifically published eDNA assays and protocols.

The analysis is conducted in two phases. The sample first goes through an extraction process where the filter is incubated in order to obtain any DNA within the sample. The extracted sample is then tested via real-time PCR (also called q-PCR) for each of the selected target species. This process uses species-specific molecular markers (known as primers) to amplify a select part of the DNA, allowing it to be detected and measured in 'real time' as the analytical process develops. qPCR combines amplification and detection of target DNA into a single step. With qPCR, fluorescent dyes specific to the target sequence are used to label targeted PCR products during thermal cycling. The accumulation of fluorescent signals during this reaction is measured for fast and objective data analysis. The primers used in this process are specific to a part of mitochondrial DNA only found in each individual species. Separate primers are used for each of the species, ensuring no DNA from any other species present in the water is amplified. If target species DNA is present, the DNA is amplified up to a detectable level, resulting in positive species detection. If target DNA is not present then amplification does not occur, and a negative result is recorded.

Analysis of eDNA requires scrupulous attention to detail to prevent the risk of false positive and false negative results. True positive controls, negative controls, and spiked synthetic DNA are included in every analysis and these have to be correct before any result is declared. Stages of the analysis are also conducted in different buildings at our premises for added security. SureScreen Scientifics Ltd is ISO9001 accredited and participates in Natural England's proficiency testing scheme for GCN eDNA testing.

Interpretation of Results

Sample Integrity Check:

Laboratory Arrival:

When samples are received in the laboratory, they are inspected for any tube leakage, suitability of sample (not too much mud or weed etc.) and absence of any factors that could potentially lead to inconclusive results. Any samples which fail this test are rejected and eliminated before analysis.

Degradation and Inhibition check:

Analysis of the spiked DNA marker to see if there has been degradation or inhibition of the kit or sample, between the date it was made to the date of analysis. Degradation of the spiked DNA marker may indicate a risk of false negative results. If inhibition is detected, samples are purified and re-analyzed. Inhibitors cannot always be removed, if the inhibition check fails, the sample should be re-collected.

Result:

Presence of eDNA (Positive/Negative/Inconclusive)

Positive: DNA was identified within the sample, indicative of species presence within the sampling location at the time the sample was taken or within the recent past.

Positive Replicates: Number of positive qPCR replicates out of a series of 12. If one or more of these are found to be positive the pond is declared positive for species presence. It may be assumed that small fractions of positive analyses suggest low level presence, but this cannot currently be used for population studies. Even a score as low as 1/12 is declared positive. 0/12 indicates negative species presence.

Negative: eDNA was not detected or is below the threshold detection level and the test result should be considered as evidence of species absence, however, does not exclude the potential for species presence below the limit of detection.

Inconclusive: Controls indicate inhibition or degradation of the sample, resulting in the inability to provide conclusive evidence for species presence or absence.

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