

MCI DEVELOPMENTS LIMITED

15 Beecham Court
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Date: 18th December 2024
Reference: 81-734-L1

WHITE-CLAWED CRAYFISH SURVEY TRUBS FARM, ROCHDALE

BACKGROUND

E3P were instructed by MCI Developments Limited to produce a Letter Statement detailing the results of a white-clawed crayfish (*Austropotamobius pallipes*) eDNA survey of the Rochdale Canal adjacent to Trubs Farm, Rochdale, hereafter referred to as 'the site'.

A Preliminary Ecological Appraisal was undertaken by E3P in March 2024 (report reference: 81-734-R1). During the desk study, it was identified that the Rochdale Canal Site of Special Scientific Interest (SSSI), Special Area of Conservation (SAC) and Site of Biological Interest (SBI), which is designated for floating water-plantain (*Luronium natans*), borders the eastern boundary of the site.

It was recommended that a 5 m buffer be maintained from the Rochdale Canal due to an historic record of white-clawed crayfish and the known presence of the species within the canal. As a 5 m buffer cannot be maintained, therefore it was recommended that white-clawed crayfish surveys be carried out following best practice guidance. The survey was to include hand searching and torch light searches, as well as baited traps if permission was granted by the Environment Agency. Following correspondence with the Canal and River Trust, neither baited trapping nor netting could be undertaken, this was to protect any floating water-plantain present within the Rochdale Canal.

WHITE-CLAWED CRAYFISH SURVEY

The white-clawed crayfish survey was undertaken on 21st October 2024 by Mike Bryant, Principal Ecologist at Ecology Elements Ltd, and Lowri Thomas BSc (Hons), Senior Ecologist at E3P. Mike holds a Natural England White-clawed Crayfish Class Licence (2015-14637-CLS-CLS) and has experience undertaking multiple white-clawed crayfish surveys at a wide variety of sites across the UK.

The survey comprised torch light searches of the Rochdale Canal, as well as water sample extraction and lab analysis for white-clawed crayfish eDNA. A total of 20 water samples were taken along the Rochdale Canal, as per the instructions set out in the 'Filtration eDNA Sample Form' by SureScreen Scientifics. Please see Appendix I for the extent of the white-clawed crayfish survey.

LIMITATIONS

The main channel of the canal was too deep to successfully torch. As such, torch searches were limited to a spur of the canal (see Appendix I). The torch search of the spur was restricted to areas where gaps in the marginal and emergent vegetation were present.



RESULTS

No white-clawed crayfish were identified during the torch search of the canal. Furthermore, the results of the eDNA survey received on 04th November 2024, returned as **NEGATIVE** for the presence of white-clawed crayfish.

Please see Appendix II for the eDNA Survey Results.

CONCLUSIONS AND RECOMMENDATIONS

White-clawed crayfish have been confirmed to be absent from the stretch of the Rochdale Canal adjacent to the site. The survey results will be valid for two years to inform a planning application. As such, no further consideration regarding white-clawed crayfish is required. However, the pollution prevention measures as detailed in the Preliminary Ecological Appraisal (report reference: 81-734-R1), will need to be followed throughout development.

Yours sincerely,
For and on behalf of E3P Ltd

Conor Evans
Consultant Ecologist

APPENDIX I

WHITE-CLAWED CRAYFISH

SURVEY EXTENT

Key:

 Site boundary

 Area of canal torch searched

 eDNA Sample Section

Spur

Notes

Issue: 1	Revision: 1	Date: 04/12/2024	Drawn: CE	Authorised: CK
Client: MCI Developments Ltd			Job No. 81-734	Date: 04/12/2024
			Drawing No. 81-734-010	Scale: 1:4000 @ A4
Job title: Trubs Farm, Rochdale			Drawing title: White-clawed Crayfish Survey Extent	



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APPENDIX II

WHITE-CLAWED CRAYFISH

EDNA SURVEY RESULTS

Folio No: 3615-2024
Purchase Order: 009800
Contact: E3P
Issue Date: 04.11.2024
Received Date: 23.10.2024

eDNA Report

Technical Report



SureScreen Scientifics

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
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FK2787	Trubsfarm Rochdale - Rochdale Canal		White-clawed crayfish	Pass	Negative	0
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Matters affecting result: none

Reported by:Chelsea Warner

Approved by: Consuela Sopronyi



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.

