



Newall Green HS,
Greenbrow Rd,
Manchester,
M23 2SX

Great Crested Newt eDNA Survey

April 2023

Ref 23-10490

Rev 1



Quality Standards Control

The signatories below verify that this document has been prepared in accordance with our quality control requirements. These procedures do not affect the content and views expressed by the originator.

This document must only be treated as a draft unless it has been signed by the originators and approved by a director.

Revision	-	Rev 1
Date	20/04/2023	26/04/2023
Prepared by	G. Kelly	G. Kelly
Checked by	A. Gonzales	A. Gonzales
Authorised by	E. Snape	E. Snape

Limitations

Syntegra Consulting Ltd ("SC") has prepared this report for the sole use of the client in accordance with the agreement under which our services were performed. No other warranty, expressed or implied, is made as to the professional advice included in this report or any other services provided by SC.

The conclusions and recommendations contained in this report are based upon information provided by others and upon the assumption that all relevant information has been provided by those parties from whom it has been requested and that such information is accurate. Information obtained by SC has not been independently verified by SC, unless otherwise stated in the report.

The methodology adopted and the sources of information used by SC in providing its services are outlined in this report. The work described in this report was undertaken in **March 2023** and is based on the conditions encountered and the information available during the said period of time. The scope of this report and the services are accordingly factually limited by these circumstances.

Where assessments of works or costs identified in this report are made, such assessments are based upon the information available at the time and where appropriate are subject to further investigations or information which may become available.

SC disclaim any undertaking or obligation to advise any person of any change in any matter affecting the report, which may come or be brought to SC's attention after the date of the report.

Certain statements made in the report that are not historical facts may constitute estimates, projections or other forward-looking statements and even though they are based on reasonable assumptions as of the date of the report, such forward-looking statements by their nature involve risks and uncertainties that could cause actual results to differ materially from the results predicted. SC specifically does not guarantee or warrant any estimate or projections contained in this report.

Costs may vary outside the ranges quoted. Whilst cost estimates are provided for individual issues in this report these are based upon information at the time which can be incomplete. Cost estimates for such issues may therefore vary from those provided. Where costs are supplied, these estimates should be considered in aggregate only. No reliance should be made in relation to any division of aggregate costs, including in relation to any issue, site or other subdivision.

No allowance has been made for changes in prices or exchange rates or changes in any other conditions which may result in price fluctuations in the future. Where assessments of works or costs necessary to achieve compliance have been made, these are based upon measures which, in SC's experience, could normally be negotiated with the relevant authorities under present legislation and enforcement practice, assuming a proactive and reasonable approach by site management.

Forecast cost estimates do not include such costs associated with any negotiations, appeals or other non-technical actions associated with the agreement on measures to meet the requirements of the authorities, nor are potential business loss and interruption costs considered that may be incurred as part of any technical measures.

Copyright

© This report is the copyright of SC. Any unauthorised reproduction or usage by any person other than the addressee is strictly prohibited.

Table of Contents

Summary.....	5
Introduction	5
Methods.....	8
Results.....	9
Constraints and Recommendations	10
References.....	11
Appendix	12

1. Summary

We were commissioned on 30th March 2023 to undertake an eDNA survey to ascertain the presence/absence of great crested newt within the single pond at Dixon's Academy in Manchester.

The present eDNA survey returned a negative result for great crested newt DNA within this waterbody.

2. Introduction

2.1 Background

Following a 'Good' Habitat Suitability Indices score for great crested newt at the site pond within Dixon's Academy grounds (PEA Report, Syntegra, 2022, ref: 22-9204), we have carried out an eDNA survey for the species.

Specific to this report, redevelopment works of the existing Newall Green High School, Greenbrow Road, M23 2SX are planned for 2023 and 2024. The site comprises the disused school buildings and playing fields, plus all associated access areas.

Figure 1: Site Boundary



2.2 Personnel and Quality Assurance

All fieldwork is carried out in accordance with current best practice guidelines and under the supervision of senior staff and appropriately licensed ecologist, Georgina Kelly.

Georgina Kelly has over fifteen years professional experience within ecology and conservation. She has a wealth of experience undertaking ecological and protected species surveys. Working within both consultancy and research, Georgina has supported a variety of projects including large infrastructure projects and nationally important nature reserves. Georgina has held a great crested newt licence for nine years (2020-45934-CLS) and has thirteen years' experience with the species, providing survey training and guidance.

2.3 Status of the Great Crested Newt, *Triturus cristatus*

The great crested newt (GCN) is an internationally important species of conservation concern. Since the late 1940's great crested newt populations have experienced drastic declines throughout much of Europe, although populations are still found in many parts of the UK, which is considered to be a stronghold for the species.

2.4 Legislation and Policy

Great crested newts are protected under Schedule 2 of the EC Council Directive "*Conservation of Natural Habitats and Wild Fauna and Flora*" (the Habitats Directive) as a European Protected Species (EPS). This legislation has been transposed into UK legislation through the Conservation of Habitats and Species Regulations 2017.

Regulation 41 of the Conservation of Habitats and Species Regulations 2010 makes it an offence to:

- *Deliberately capture, injure, or kill great crested newts or destroy their eggs.*
- *Deliberately disturb great crested newts.*
- *Damage or destroy a breeding site or resting place of a great crested newt; this applies whether newts are present or not.*

In addition to the above protection, great crested newts are protected under Schedule 5 of the Wildlife & Countryside Act 1981 (as amended). In addition to the above, it lists the following as additional offences:

- *Disturbance of an animal whilst it is occupying a place which it uses for shelter or protection.*
- *Obstruct access to any structure or place which an animal uses for shelter or protection.*
- *Possess or control any live or dead specimen or anything derived from a great crested newt.*

The above legislation applies to all life-stages of the species, from egg through larvae to sub-adults and adult newts.

The great crested newt is also listed as a priority species within the UK Biodiversity Action Plan and under the NERC Act 2006.

2.4.1 Purpose of Legislation

The purpose of the legislation is to maintain and restore great crested newt populations within their natural range. This implies that the habitats on which they rely, and the ecology of their life cycles should not be compromised by human activities. Where activities have the potential to compromise great crested newt populations, measures are required to be put in place to avoid impacts or compensate and mitigate for those impacts.

3. Methods

3.1 Presence/absence eDNA Survey

The single pond on site was assessed for great crested newt presence or absence using eDNA sampling techniques. This method requires the collection of water samples which are then sent to a laboratory for analysis.

This technique aims to detect DNA that is present in the environment in which an animal lives. Great crested newt DNA can enter ponds via their faeces, urine and skin cells. In aquatic environments, this DNA may persist for weeks and can be collected through water samples and analysed to determine the presence or absence of the species. Natural England accepts eDNA test results as evidence of presence or absence of great crested newt.

The survey was undertaken on 13th April 2023 by Georgina Kelly, a licensed great crested newt worker. This is during the optimal period for GCN eDNA survey.

Survey protocol followed the recommendations by Natural England. In summary, this involves the following steps:

20 samples were taken from the waterbody. The location of sub-samples were spaced as evenly as possible around the waterbody margin. Subsamples generally targeted areas with potential egg laying substrate (e.g. submerged vegetation) and open water areas which newts may be using for displaying. Prior to sampling, the water column was mixed by gently using a ladle to stir through the entire water column, whilst avoiding disturbing the sediment on the bed of the waterbody. Sampling of very shallow water was avoided where possible (less than 5-10 cm deep).

Once all samples had been collected, the sample bag was secured and shaken vigorously for 10 seconds. This mixed any DNA across the whole water sample. Using a pipette, c15mL of water was taken from the bag and transferred to a sterile tube containing 35mL of ethanol to preserve the eDNA sample.

Preserved samples were then stored at a cool ambient temperature before being returned to Surescreen Scientific on the 14th April 2023 for analysis.

4. Results

The survey pond is located to the northern side of the existing school building, partially lying beneath the most north-easterly unit (NGR: SJ 80924 87527). The pond lies within an enclosed garden area that separates the existing buildings from one of the carpark areas. There are no signs of recent management to the marginal vegetation or the surrounding garden. This is a medium-sized pond with relatively dense vegetation, including flag iris (*Iris pseudocorus*), bulrush (*Typha latifolia*), and marginal willows (*Salix* spp.).

Figure 2: The Survey Pond



The laboratory at Surescreen Scientifics returned a **negative** result for great crested newt eDNA. A copy of the laboratory results is provided in the Appendix.

Consequently, it is considered very unlikely that great crested newt is present within the pond at this time.

5. Constraints and Recommendations

The results of the survey are pertinent to the single survey pond only.

While presence of GCN has not been confirmed within the survey pond at this time, the site has been found to provide potential terrestrial habitat for great crested newt and other amphibians, particularly within the areas of grassland, scrub and hedgerows. Considering the significant records in nearby ponds (including waterbodies at Davenport Green to the southwest of the site that support the largest known population in the Trafford area), the potential for the planned development to have an impact on any great crested newts present in and around the site remains. Please refer to the PEA survey for both historic records and assessment of terrestrial habitat for GCN (Syntegra, 2022).

If changes to work plans occur, it is recommended that further impact assessments are made to identify the likelihood of impact to great crested newts or other amphibians.

Where this is deemed likely to impact GCN, it is recommended that construction works are carried out under a Natural England Protect Species Licence. The licence application process may require additional surveys to be carried out of any ponds within 500m of the proposed works.

6. References

Baker, et al. (2011) *Amphibian Habitat Management Handbook*

Froglife (2001) *Great Crested Newt Conservation Handbook*

Gent, A. and Gibson, S. (1998). *Herpetofauna Workers' Manual*. Peterborough, UK. Joint Nature Conservation Committee

Oldham R.S., Keeble J., Swan M.J.S. & Jeffcote M. (2000). *Evaluating the suitability of habitat for the Great Crested Newt (Triturus cristatus)*. Herpetological Journal 10 (4), p143-155.

UK Biodiversity Action Plan: Available: www.ukbap-reporting.org.uk/plans/national.asp

Wildlife & Countryside Act (1981)

7. Appendix

eDNA result



Folio No: E16481
Report No: 1
Purchase Order: DIXON
Client: Terra Ecology
Contact: Georgina Kelly

TECHNICAL REPORT

ANALYSIS OF ENVIRONMENTAL DNA IN POND WATER FOR THE DETECTION OF GREAT CRESTED NEWTS (TRITURUS CRISTATUS)

SUMMARY

When great crested newts (GCN), *Triturus cristatus*, inhabit a pond, 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 GCN habitation or establish GCN absence.

RESULTS

Date sample received at Laboratory: 18/04/2023
Date Reported: 19/04/2023
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
0848	DIXON ACADEMY		Pass	Pass	Pass	Negative	0

If you have any questions regarding results, please contact us: ForensicEcology@surescreen.com

Reported by: Chris Troth

Approved by: Jennifer Higginbottom



Forensic Scientists and Consultant Engineers
SureScreen Scientifics Ltd, Morley Retreat, Church Lane, Morley, Derbyshire, DE7 6DE
UK Tel: +44 (0)1332 292003 Email: scientifics@surescreen.com
Company Registration No. 08950940

Page 1 of 2



METHODOLOGY

The samples detailed above have been analysed for the presence of GCN eDNA following the protocol stated in DEFRA WC1067 'Analytical and methodological development for improved surveillance of the Great Crested Newt, Appendix 5.' (Biggs et al. 2014). Each of the 6 sub-sample tubes are first centrifuged and pooled together into a single sample which then undergoes DNA extraction. The extracted sample is then analysed using real time PCR (qPCR), which uses species-specific molecular markers to amplify GCN DNA within a sample. These markers are unique to GCN DNA, meaning that there should be no detection of closely related species.

If GCN DNA is present, the DNA is amplified up to a detectable level, resulting in positive species detection. If GCN 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 risk of contamination. 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 and reported. Stages of the DNA analysis are also conducted in different buildings at our premises for added security.

SureScreen Scientifics Ltd is ISO9001 accredited and participate in Natural England's proficiency testing scheme for GCN eDNA testing. We also carry out regular inter-laboratory checks on accuracy of results as part of our quality control procedures.

INTERPRETATION OF RESULTS

- SIC:** **Sample Integrity Check** [Pass/Fail]
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.
- DC:** **Degradation Check** [Pass/Fail]
Analysis of the spiked DNA marker to see if there has been degradation of the kit or sample between the date it was made to the date of analysis. Degradation of the spiked DNA marker may lead indicate a risk of false negative results.
- IC:** **Inhibition Check** [Pass/Fail]
The presence of inhibitors within a sample are assessed using a DNA marker. If inhibition is detected, samples are purified and re-analysed. Inhibitors cannot always be removed, if the inhibition check fails, the sample should be re-collected.
- Result:** **Presence of GCN eDNA** [Positive/Negative/Inconclusive]
Positive: GCN DNA was identified within the sample, indicative of GCN presence within the sampling location at the time the sample was taken or within the recent past at the sampling location.
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 GCN presence. It may be assumed that small fractions of positive analyses suggest low level presence, but this cannot currently be used for population studies. In accordance with Natural England protocol, even a score of 1/12 is declared positive. 0/12 indicates negative GCN presence.
Negative: GCN eDNA was not detected or is below the threshold detection level and the test result should be considered as evidence of GCN absence, however, does not exclude the potential for GCN presence below the limit of detection.



Forensic Scientists and Consultant Engineers
SureScreen Scientifics Ltd, Morley Retreat, Church Lane, Morley, Derbyshire, DE7 6DE
UK Tel: +44 (0)1332 292003 Email: scientifics@surescreen.com
Company Registration No. 08950940

Page 2 of 2