A46 Coventry Junctions (Walsgrave) Scheme number: TR010066

6.3 Environmental Statement Appendices

Appendix 8.9 Great Crested Newt Report

APFP Regulations 5(2)(a)

Planning Act 2008

Infrastructure Planning (Applications: Prescribed Forms and Procedure) Regulations 2009

Volume 6

November 2024

Infrastructure Planning

Planning Act 2008

The Infrastructure Planning (Applications: Prescribed Forms and Procedure) Regulations 2009

A46 Coventry Junctions (Walsgrave)
Development Consent Order 202[x]

ENVIRONMENTAL STATEMENT APPENDICES
Appendix 8.9 Great Crested Newt Report

Regulation Number	Regulation 5(2)(a)
Planning Inspectorate Scheme Reference	TR010066
Application Document Reference	TR010066/APP/6.3
Author	A46 Coventry Junctions (Walsgrave), Project Team & National Highways

Version	Date	Status of Version
Rev 0	November 2024	Application Issue

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1. Introduction

1.1. Scheme overview

1.1.1. The A46 is part of the strategic road network forming a significant trade and export route between the East and West Midlands. As part of the Government's Road Investment Strategy (RIS2) 2020-2025, the A46 Walsgrave Junction (the 'Scheme') is being improved with the realignment of the carriageway and a new grade separated junction. This aims to increase the road's capacity to cater for future developments across the region and promote safety by separating local and long-distance traffic and reducing congestion.

1.1.2. At the time of writing, the Scheme has commenced the preliminary design stage. The great crested newt *Triturus cristatus* (GCN) survey undertaken during 2022 detailed within this report was undertaken in advance of the preliminary design stage and based upon the draft Order Limits as they were at the time. The update survey undertaken in 2024 and detailed within this report was undertaken during the preliminary design stage and based upon the Order Limits (the Scheme boundary).

1.1.3. Sweco were commissioned by Octavius Infrastructure on behalf of National Highways to undertake the GCN surveys for the Scheme.

1.2. Order Limits description

1.2.1. The Order Limits comprise an area of approximately 25ha of natural habitat located to the east of Coventry.

1.2.2. The habitats within the Order Limits include woodland, scrub, arable farmland and hedgerows.

1.3. Previous surveys

1.3.1. To inform the Scheme, GCN environmental DNA (eDNA) surveys were undertaken in 2016 (Highways England, 2018) on two ponds with GCN presence confirmed in pond 1 and absence confirmed in pond 2.

1.3.2. A scoping survey was undertaken on a further five ponds in 2018 with ponds 3, 4, 6 and 7 scoped out from further survey and pond 5 found to be dry. Ponds 1 and 2 were scoped in for further survey due to the GCN presence confirmed in pond 1 in 2016 and the close proximity of these ponds to each other (Highways England, 2018).

1.4. Purpose

1.4.1. This GCN report has been prepared by Sweco for National Highways and will be used to inform the biodiversity assessment of the Environmental Statement (ES). The report details GCN surveys undertaken in 2022 and April 2024.

2. Methodology

2.1. Desk study

2.1.1. The Multi-Agency Geographic Information for the Countryside (MAGIC, 2022) online database was used to obtain information on the presence of any ponds within 500m of the Order Limits to inform an assessment of habitat availability and connectivity for GCN.

2.1.2. In addition, the seven waterbodies identified by Highways England (2018) were reassessed to determine if further surveys were required.

2.2. Habitat suitability index

2.2.1. The habitat suitability index (HSI) assessment provides an objective method for assessing the suitability of a pond as habitat for GCN (Oldham et al, 2001; ARG UK, 2010). The system provides an index between 0 and 1, with 0 indicating unsuitable habitat and 1 optimal habitat. Ten suitability indices are used to calculate the index score, each representing a factor considered to affect GCN. Results from individual waterbodies are categorised as follows:

- <0.5 = poor
- 0.5 – 0.59 = below average
- 0.6 – 0.69 = average
- 0.7 – 0.79 = good
- >0.8 = excellent

2.2.2. During 2022 surveys a HSI assessment was undertaken on all ponds holding water. Dry ponds were not assessed as they are unsuitable for GCN. All ponds were re-visited during 2024 surveys and those which held water were subject to update HSI assessments.

2.3. eDNA survey

2.3.1. An eDNA survey was undertaken on 7 and 8 June 2022 by [redacted] and [redacted]. The weather during both days was dry and sunny. Water samples were taken from a total of five waterbodies (ponds 1, 2, 5, 9 and 10).

2.3.2. The 2024 surveys were undertaken by [redacted] and [redacted] on 15 April. The weather was dry and cloudy. Water samples were collected from a total of four waterbodies in 2024 including pond 9 which returned an inconclusive eDNA result in 2022, pond 13 which was dry in 2022 surveys and pond 14 which was scoped out in 2022 and 15.

2.3.3. All samples in 2022 and 2024 were taken following SureScreen Scientific's instructions for sampling GCN eDNA within Great Crested Newt eDNA Technical White Paper. The collection method is in line with guidance within Defra's *Analytical and methodological development for improved surveillance of the Great Crested Newt and technical advice note for field and laboratory sampling of great crested newt environmental DNA* (Biggs et al, 2014). The samples were then sent to SureScreen Scientifics for analysis.

2.4. Survey limitations

2.4.1. In 2024, the dense bankside vegetation surrounding pond 9 meant that only 30-40% of the pond was accessible for water sampling. This is not considered a limitation as the habitat within the pond is uniform throughout and different locations of the pond were still accessible for the collection of water samples.

2.4.2. Access permission was not obtained for a revisit of pond 4 during 2024 surveys. As this pond is located 560m from any works impacting suitable GCN habitat and was dry in 2022 this is not considered a constraint to the assessment of impacts of the Scheme upon GCN.

3. Results

3.1. Desk study

3.1.1. The desk study undertaken in 2022 identified 14 ponds within ~500m of the Order Limits which were subject to further scoping and assessment to determine the potential risk to GCN posed by the Scheme.

3.1.2. Of these 14 ponds, six (ponds 3, 4, 6, 7 and 12) were scoped out from further assessment due to the following:

- Pond 3 is a large lake called Coombe Pool which supports a fishery with limited emergent vegetation to provide cover for amphibians and is therefore unlikely to be suitable for GCN.
- Pond 4 is a small pond that was found to be dry during Highways England surveys in 2018. The pond is located approximately 430m from the Order Limits boundary in the centre of an arable field which is unsuitable habitat for GCN. The arable field was surveyed for badgers (*Meles meles*) in September 2022 and the pond was found to be dry with no aquatic vegetation present and therefore, the pond was scoped out.
- Ponds 6, 7 and 12 are present on the opposite side of the River Sowe which will act as a barrier to dispersal for GCN.

3.1.3. An additional pond (pond 15) was created in November 2022 and was therefore scoped in for survey in 2024.

3.2. Habitat suitability index

3.2.1. HSI assessments were undertaken on scoped in ponds in 2022 and 2024.

2022 HSI assessment

3.2.2. Nine ponds were visited on the 7 and 8 June 2022 and an assessment was made of the suitability of the waterbodies for GCN through a HSI assessment. Plates 1 – 6 below provide photographs of those ponds (ponds revisited in 2024 have been updated with most recent photographs) which were present and held water at the time of survey, and as such were subject to a HSI assessment (ponds 1, 2, 5, 9, 10 and 14). The full HSI assessment results are included in Appendix C.

3.2.3. The 2022 surveys found that pond 11 did not exist, and as such this feature was not re-visited in 2024.

3.2.4. During the 2022 survey pond 14 was scoped out of survey. The pond was created around January 2021 as a retention pond for the adjacent site compound associated with the construction works at the Binley Junction. The

pond had been present for one breeding season only at the time of the survey in 2022 and is located approximately 650m away from any other pond that is not separated by a significant dispersal barrier. Additionally, the pond was found to have a 0.56 HSI score (see Table 3-1) which represents a below average suitability and there was no aquatic vegetation for egg laying. As such GCN were not expected to have colonised the pond at the time of the 2022 surveys. Furthermore, pond 14 is located approximately 490m from the main works and works which will be undertaken within 490m from the pond include primarily in carriageway works with minor impacts to the verge habitats.

2024 HSI assessment

3.2.5. HSI assessments were undertaken on ponds 1, 2, 9, 10, 13, 14 and 15 (Appendix C) on 15 April 2024. The ponds (excluding the following scoped out ponds; 3, 5, 6, 7, 12) were revisited in 2024 to ensure they remained in the same condition from 2022. Pond 8 was dry at the time of the survey therefore no HSI assessment was undertaken. Pond 4 was not subject to survey in 2024 (see Section 2.4) and pond 5 is no longer within the 500m study area for GCN following changes to the Order Limits during the preliminary design stage. The survey was undertaken by [redacted] (Senior Ecologist, Sweco, Natural England GCN class licence CL08 holder – registration number 2018-33651-CLS-CLS) and [redacted] (Ecologist, Sweco Natural England GCN class licence CL08 holder – registration number 2023-11072-CL08-GCN).

3.2.6. Updated results of the HSI surveys are shown in Table 3-1. Plates 7- 9 below provide photographs of those additional ponds visited in 2024.

3.2.7. In November 2022, pond 15 was created, approximately 40m south of pond 14. Due to the increase in potential breeding habitat and the increased length of time since the creation of pond 14 both ponds 14 and 15 were subject to survey in 2024.

Table 3-1 A46 Walsgrave Junction HSI results

Pond	OS grid reference	Description	HSI score/suitability
1	SP 39328 80259	The edge of the pond has aquatic vegetation dominated by bulrush *Typha latifolia* and rush *Juncus sp.* The pond is surrounded by dense scrub.	0.63 – Average
2	SP 39401 80285	The edge of the pond comprises a ring of emergent vegetation with a submerged species noted close to the centre. The pond is surrounded by dense scrub dominated by bramble *Rubus fruticosus*.	0.61 – Average

Pond	OS grid reference	Description	HSI score/suitability
5	SP 37684 78777	The pond is a small waterbody located within a woodland. There were no aquatic plants noted within the pond.	0.53 – Below average
8	SP 39376 80528	The pond was dry at the time of survey with no aquatic vegetation present.	N/A
9	SP 39310 81092	The pond was surrounded by dense bank vegetation and only 30-40% of the pond was accessible. Vegetation on the bank included bramble and hawthorn *Crataegus monogyna*. Sedge species were present within the pond.	0.62 – average
10	SP 38944 80177	The pond is located within a small section of woodland in an arable field. There were no aquatic plants noted within the pond.	0.50 – Below Average
11	SP 39573 80901	There was no pond present at this location. The habitat comprises tall ruderal vegetation dominated by nettle *Urtica dioica*.	N/A
13	SP 37568 79387	The pond is mostly dry, with standing water absent from the majority of the surface, with the exception of at the easternmost edge where there was approximately 20cm of water in a small area. The basin is dominated by extensive bulrush *Typha latifolia.* Other flora included willowherb *Epilobium*, meadowsweet *Filipendula ulmaria* and lesser celandine *Ficaria verna*. A small channel, dry at the time of survey and devoid of aquatic vegetation, connects the pond to the adjacent River Sowe suggesting that the basin may become wet during flood events.	0.49 – poor
14	SP 38775 78471	This pond was created around January 2021 as a retention pond for the adjacent works compound associated with the construction at the Binley Junction. The surrounding banks included grass and tall herbs. Limited submerged aquatic vegetation was present. The layer of algae covered the extent of the pond surface.	0.52 – below average
15	SP 38823 78394	This pond was created around November 2022 as a retention pond for the adjacent works compound. The moderately step banks were colonised by grasses and short herbaceous vegetation. Aquatic vegetation was entirely absent.	0.56 – below average

Plate 1 showing photo of pond 2 (2024)

Plate 2 showing updated photo of pond 1

Plate 3 showing pond 5

Plate 4 showing updated photo of pond 9

Plate 5 showing updated photo of pond 10

Plate 6 showing updated photo of pond 14

Plate 7 showing pond 8

Plate 8 showing pond 13

Plate 9 showing pond 15

3.3. eDNA survey

2022 eDNA results

3.3.1. In 2022, laboratory analysis of eDNA samples collected produced a positive result for GCN in pond 1 and a negative result for ponds 2, 5 and 10. An inconclusive result was returned for pond 9 as the sample failed an inhibition check.

2024 eDNA results

3.3.2. In 2024, laboratory analysis of eDNA samples collected produced a positive result for GCN in pond 15 and a negative result for ponds 9, 13 and 14.

4. Conclusions and recommendations

4.1. Discussion

4.1.1. Surveys have confirmed GCN presence within pond 1 and pond 15 in the study area. Given that GCN were found absent from ponds immediately adjacent to ponds 1 and 15 (ponds 2 and 14 respectively) it is assumed that the GCN populations in ponds 1 and 15 are present in low numbers.

4.1.2. Pond 1 is located on the edge of an arable field approximately 430m east of the Order Limits. Unsuitable arable land is present between pond 1 and the Order Limits. The pond is approximately 420m from the Order Limits along the nearby hedgerow which would be the single suitable commuting corridor for GCN from the pond to the Order Limits. Suitable GCN habitat in the Order Limits within 500m of pond 1 is limited to the highway boundary woodland. As such it is considered unlikely that GCN from pond 1 are present in terrestrial habitat within the Order Limits, however GCN absence cannot be confirmed.

4.1.3. Pond 15 is approximately 95m from the Order Limits however is approximately 430m from the Order Limits where works other than in-carriageway works (within hard standing which is unsuitable habitat) are proposed. It is considered unlikely GCN will use the terrestrial habitat to be impacted in the Order Limits within 500m of pond 15 as this is a significant distance from the pond (430m), however GCN absence cannot be confirmed. Coventry Road (which is kerbed) may also act as a dispersal barrier for GCN which may commute north from pond 15.

4.2. Recommendations

4.2.1. Works within 500m of ponds 1 and 15 include works impacting highway boundary habitat suitable for GCN to facilitate the Scheme. However, due to the distance of these habitat impacts from the ponds (>250m) and the small area of GCN habitat lost within 500m of the ponds (<5ha) no population size class surveys are required.

4.2.2. In order to mitigate against injury/mortality to GCN during construction it is recommended that appropriate mitigation measures, which will be detailed within the First Iteration Environmental Management Plan (EMP), are adhered to during construction. These measures would be applicable to works within suitable habitat within 500m of the GCN ponds and include timing of works to avoid impacts where feasible and supervision of vegetation clearance during the active season by an ECoW.

Construction is programmed to commence in September 2026. Update surveys will be undertaken in the survey season of 2025. A review of the validity of 2024 survey data will be undertaken in advance of these update surveys.

5. References

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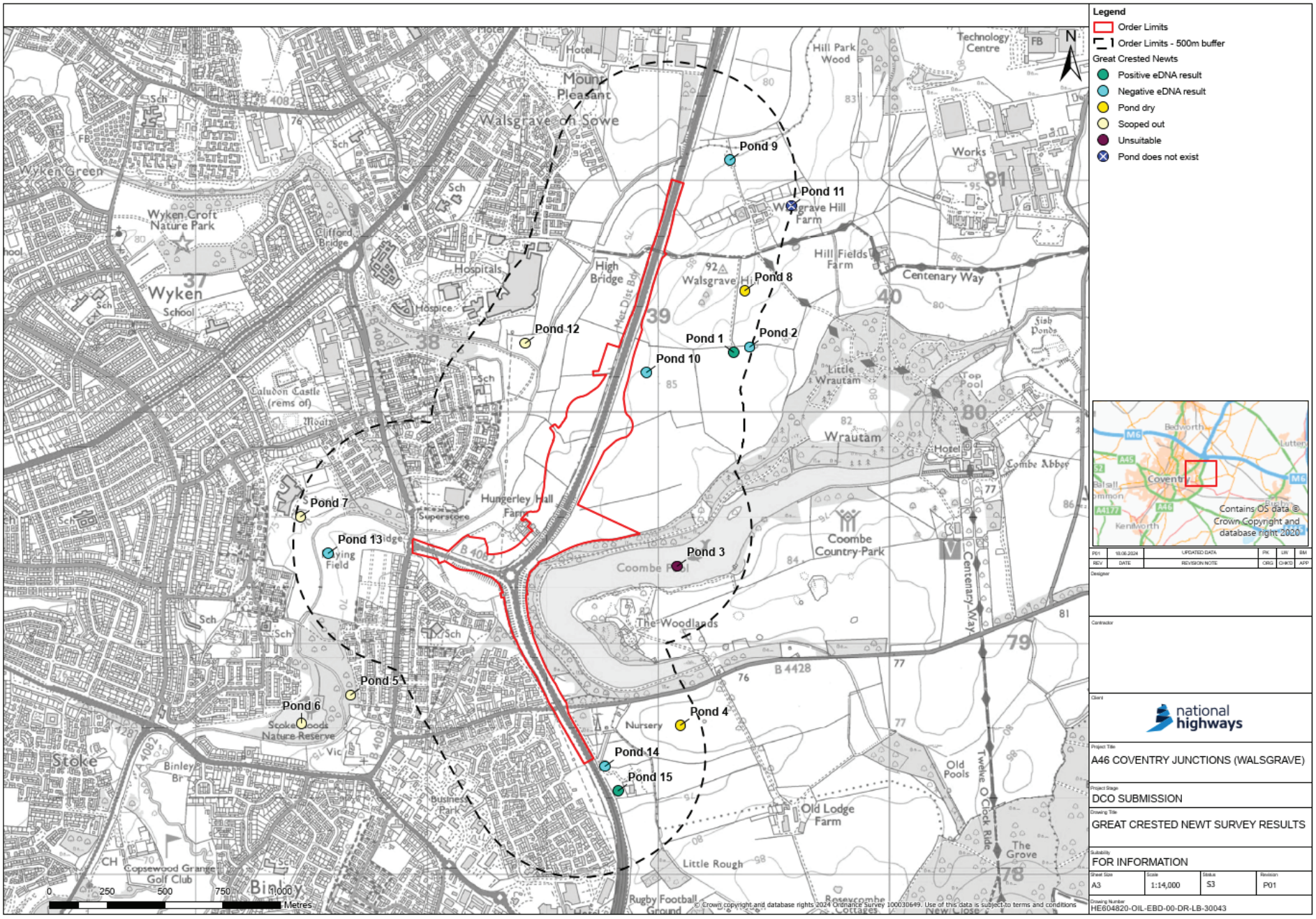
Oldham, R.S.; Keeble, J.; Swan, M.J.S. & Jeffcote M. (2001) Evaluating the suitability of habitat for the great crested newt (*Triturus cristatus*). The Herpetological Journal 10 (4), 143-155.

Appendix A. Great Crested Newt eDNA Survey Results


Legend
Order Limits
Order Limits - 500m buffer
Great Crested Newts
Positive eDNA result
Negative eDNA result
Pond dry
Scoped out
Unsuitable
Pond does not exist
Pond 1
Pond 2
Pond 3
Pond 4
Pond 5
Pond 6
Pond 7
Pond 8
Pond 9
Pond 10
Pond 11
Pond 12
Pond 13
Pond 14
Pond 15
Walsgrave on Sowe
Wyken
Stoke
Coombe Country Park
Wrautam
Hungerley Hall Farm
Contains OS data © Crown Copyright and database right 2020
national highways
A46 COVENTRY JUNCTIONS (WALSGRAVE)
DCO SUBMISSION
GREAT CRESTED NEWT SURVEY RESULTS
FOR INFORMATION
A3
1:14,000
S3
P01
HE604820-OIL-EBD-00-DR-LB-30043
0 250 500 750 1000 Metres
© Crown copyright and database rights 2024 Ordnance Survey 100030649. Use of this data is subject to terms and conditions

Appendix B. SureScreen Scientifics technical report

SureScreen Scientifics

Folio No: E14017
Report No: 1
Purchase Order: PO-22-652-00823
Client: SWECO
Contact: [redacted]

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: 08/06/2022
Date Reported: 17/06/2022
Matters Affecting Results: 4358 - Degradation or inhibition

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
4358	Pond 9 A46	SP 393 810	Pass	Pass	Fail	Inconclusive	0
4361	Pond 10 A46	SP 389 801	Pass	Pass	Pass	Negative	0
4362	Pond 2 A46	SP 393 802	Pass	Pass	Pass	Negative	0
4363	Pond 1 A46	SP 393 802	Pass	Pass	Pass	Positive	9

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

Reported by: [redacted] **Approved by:** [redacted]

Forensic Scientists and Consultant Engineers
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SureScreen Scientifics

Folio No: E14018
Report No: 1
Purchase Order: PO-22-652-00831
Client: SWECO
Contact: [redacted]

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: 08/06/2022
Date Reported: 15/06/2022
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
2600	Pond 5 A46	SP 376 787	Pass	Pass	Pass	Negative	0

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

Reported by: [redacted] **Approved by:** [redacted]

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Company Registration No. 08950940

SureScreen Scientifics

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.

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Company Registration No. 08950940

Follo No: 182-2024
Purchase Order: PO-24-652-00553
Contact: Sweco Services UK Ltd
Issue Date: 18.04.2024

GCN Report

Technical Report

SureScreen Scientifics

Folio No: 182-2024
Purchase Order: PO-24-652-00553
Contact: Sweco Services UK Ltd
Issue Date: 18.04.2024

SureScreen Scientifics

GCN eDNA Analysis

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 analyzing water samples, we can detect these small traces of environmental DNA (eDNA) to confirm GCN habitation or establish GCN absence.

Results

Lab ID	Site Name	OS Reference	Degradation Check	Inhibition Check	Result	Positive Replicates
GCN0927	A46 Walsgrave/ Pond 15		Pass	Pass	Positive	4/12
GCN0928	A46 Walsgrave/ Pond 9		Pass	Pass	Negative	0/12
GCN0929	A46 Walsgrave/ Pond 14		Pass	Pass	Negative	0/12
GCN0930	A46 Walsgrave/ Pond 13		Pass	Pass	Negative	0/12

Matters affecting result: none

Reported by: [redacted] Approved by: [redacted]

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Folio No:	182-2024
Purchase Order:	PO-24-652-00553
Contact:	Sweco Services UK Ltd
Issue Date:	18.04.2024

SureScreen Scientifics

Methodology

The samples detailed above have been analyzed 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 tube which then undergoes DNA extraction. The extracted sample is then analyzed 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 attention to detail to prevent the 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 analytical 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: 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 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.

Inhibition Check: **Pass/Fail.** The presence of inhibitors within a sample is assessed using a DNA marker. 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 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 the WC1067 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.
Inconclusive: Controls indicate inhibition or degradation of the sample, resulting in the inability to provide conclusive evidence for GCN presence or absence.

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Appendix C. GCN HSI Results

Pond ref	Pond 1	Pond 2	Pond 5	Pond 9	Pond 10	Pond 13	Pond 14	Pond 15
SI1	1	1	1	1	1	1	1	1
SI2	0.8	0.95	0.3	0.2	0.3	0.1	0.9	0.9
SI1	0.9	0.9	0.5	0.9	0.9	0.1	0.9	0.9
SI4	1	0.67	0.33	0.67	0.33	0.67	0.33	0.67
SI5	1	0.9	0.3	1	0.2	1	1	1
SI6	0.67	0.67	1	0.67	1	1	0.67	0.67
SI7	0.33	0.33	1	0.67	1	1	0.67	0.67
SI8	0.6	0.6	0.4	1	0.6	0.4	0.1	0.1
SI9	0.33	0.33	1	0.33	0.33	0.33	0.33	0.33
SI10	0.3	0.3	0.3	0.5	0.3	0.9	0.35	0.4
HSI	0.63	0.61	0.53	0.62	0.50	0.49	0.52	0.56