Dudgeon and Sheringham Shoal Offshore Wind Farm Extensions

Preliminary Environmental Information Report

Volume 3

Appendix 22.2 - Great Crested Newt Survey

April 2021

China Resources
Better Life Together

Masdar
A MUBADALA COMPANY

equinor

Title**:**	
Dudgeon and Sheringham Shoal Offshore Wind Farm Extensions Preliminary Environmental Information Report Appendix 22.2 2020 Great Crested Newt HSI and eDNA Survey Report 2020	
Document no.: PB8164-WFE-ZZ-ON-RP-Z-0002	
Date:	Classification
29th April 2021	**Final**
Prepared by:	
Royal HaskoningDHV	
Approved by:	Date:
Jo Rodriguez, Equinor	29th April 2021

WILD FRONTIER ECOLOGY

Sheringham Shoal and Dudgeon Extension Projects

Great Crested Newt HSI and eDNA Survey Report 2020

March 2021

Report produced by	Produced on behalf of
Written by: William Riddett BA ACIEEM Checked by: Robert Yaxley BSc CEcol CEnv MCIEEM Authorised by: Robert Yaxley BSc CEcol CEnv MCIEEM August 2020 Update authorised March 2021	Equinor New Energy Limited Forusbeen 50 4035 Stavanger Norway
Wild Frontier Ecology Ltd. Unit 2, Cold Blow Farm Great Snoring, Fakenham Norfolk, NR21 0HF Tel: 01328 864633 will@wildfrontier-ecology.co.uk	

The data which we have prepared and provided is accurate, and has been prepared and provided in accordance with the CIEEM's Code of Professional Conduct. We confirm that any opinions expressed are our best and professional bona fide opinions.

This report conforms to the British Standard 42020:2013 Biodiversity – Code of practice for planning and development.

CONTENTS

GLOSSARY OF TERMS

DCO	Development Consent Order
DEP	Dudgeon Extension Project
DLL	District Level Licence/Licensing
DNA	Deoxyribonucleic Acid
eDNA	Environmental DNA
EP1HS	Extended Phase 1 Habitat Survey
EPS	European Protected Species
ETG	Expert Topic Group
FCS	Favourable Conservation Status
GCN	Great Crested Newt
HSI	Habitat Suitability Index
NBIS	Norfolk Biodiversity Information Service
OS	Ordnance Survey
PEIR	Preliminary Environmental Information Report
SEP	Sheringham Shoal Extension Project
UCL	University College London
UCLPRRG	University College London Pond Restoration Research Group
WFE	Wild Frontier Ecology Ltd.

EXECUTIVE SUMMARY

Wild Frontier Ecology Ltd. was commissioned by Equinor New Energy Ltd. to complete great crested newt (GCN) surveys of ponds within and up to 250 metres of the Preliminary Environmental Information Report (PEIR) boundary associated with the proposed Dudgeon Offshore Wind Farm Extension Project (DEP) and Sheringham Shoal Offshore Wind Farm Extension Project (SEP) (see Glossary of Terms for abbreviation definitions). The surveys comprised Habitat Suitability Index (HSI) appraisals of all ponds for their suitability to support GCN, and environmental DNA (eDNA) surveys of all ponds to confirm the presence or absence of GCN. The HSI appraisals were undertaken between March and June 2020. All eDNA surveys were undertaken between 28th April and 30th June 2020, within the appropriate survey season, and by GCN licensed ecologists or accredited agents.

Ponds within the emerging PEIR boundary and up to 250 metres from it (hereafter referred to as the survey area) were identified using Ordnance Survey (OS) maps and other freely availably mapping programmes such as Google Earth. Any additional ponds that were noted during other ecological field surveys completed before July 2020 were also included in the 2020 GCN survey effort.

In early 2020, a total of 290 ponds were included within the survey scope. Access was arranged or attempted for all ponds, and a total of 161 ponds were HSI appraised and eDNA surveyed during the 2020 survey season. However, refinement of the PEIR boundary in December 2020 reduced the number of ponds within the relevant area (i.e. inside the refined PEIR boundary and its surrounding 250 metres) to 179 ponds, 98 of which have been surveyed. Refinement of the PEIR boundary in December 2020 brought an additional 52 new ponds (which had not been surveyed in 2020) into the survey area; these ponds will be surveyed in 2021, assuming landowner access is permitted.

The HSI appraisal of the 98 ponds which were accessible and surveyed in 2020 resulted in 19 being calculated within the Excellent suitability classification, 26 were classified as Good, 19 were Average, 23 were Below Average and 11 were Poor. A further two ponds (in addition to the aforementioned 98 ponds) could be remotely HSI appraised (e.g. from the banks of the ponds) but were not safely accessible for full eDNA survey. The HSI suitability of these ponds was Excellent (1) and Below Average (1).

Of the 98 ponds eDNA surveyed, 84 returned negative results indicating likely GCN absence. 14 ponds returned a positive result indicating GCN presence.

Of the remaining 81 ponds from the total of 179 ponds (i.e. those which were not eDNA surveyed), 38 ponds were visited and eDNA surveys were attempted but were not possible due to the ponds being dry, removed, unsafe to access or having other constraints which prevented water samples being taken. The remaining 43 ponds were not surveyed because landowner access was not granted at the time of the surveys.

The results of the HSI and eDNA surveys of the 98 ponds within the survey area are provided in full, below, as are the results of the HSI appraisals of the additional two ponds which could not be accessed for eDNA surveys. The individual reasons (e.g. pond dry, landowner access not granted, unsafe to access etc.) for not fully surveying each of the 81 ponds are also provided below. Maps are provided below (see Figures 1-12) showing the location and distribution of ponds surveyed and the positive/negative eDNA survey results. These maps also show the locations of the 52 newly identified ponds which have been brought inside the survey area following the December 2020 refinement of the PEIR boundary.

There are a number of clusters of ponds which returned positive results, suggesting the presence of GCN metapopulations in these areas, including around Swardeston, Ketteringham and Hethersett (see Figures 1 and 2), around Marlingford and Colton (see Figure 3) and around Bodham (see Figure 10).

A desk study comprising a data search with the Norfolk Biodiversity Information Service (NBIS) and a consultation with the University College London Pond Restoration Research Group (UCLPRRG) returned records of GCN within the survey area and further afield. These data largely corroborate the results of the 2020 eDNA surveys, with records of GCN distributed around the aforementioned metapopulation areas, particularly around Bodham. The one exception is around Saxthorpe and Itteringham where the 2020 eDNA surveys recorded one isolated positive result (pond PW166); the NBIS data search returned a GCN record from this same pond and another nearby pond (pond PN103, for which the 2020 eDNA survey received a negative result). This one positive eDNA result coupled with a nearby NBIS record may also indicate a metapopulation of GCN around this part of the PEIR boundary.

Further GCN surveys will be required as the project progresses and the PEIR boundary is refined further. Once a final Development Consent Order (DCO) boundary has been defined, a review of ponds within and up to 250m of those confirmed boundaries will be undertaken to ensure that all ponds have been identified and subject to a HSI appraisal and eDNA survey (assuming landowner access is granted). Ponds which fall outside of both the DCO boundary and its 250m surrounding buffer would be discounted and not subject to any further survey or assessment. Furthermore, any new ponds identified during the Extended Phase 1 Habitat Survey (or other field surveys) which are within the refined survey area will be incorporated into the scope of further HSI and eDNA surveys in 2021.

Further surveys would also be required to inform any European Protected Species (EPS) licensed mitigation, which could be necessary depending on the precise scope of construction works and their anticipated impacts on GCN and/or GCN habitat. Such surveys would need to be completed within the survey season immediately before the EPS mitigation licence is applied for, meaning they will likely be required shortly before construction works commence. EPS mitigation licences typically require presence/absence surveys (and corresponding population estimate surveys on ponds in which GCN are confirmed present) rather than eDNA surveys, to confirm GCN population sizes within ponds within relevant areas.

An alternative approach to any necessary licensed mitigation could involve the DEP and SEP achieving a District Level Licence (DLL). However, the PEIR boundary partly overlaps a DLL 'Red zone' around Upgate near Swannington; this may prohibit DLL as a viable option for DEP and SEP, but this will be confirmed through consultation with Natural England. DLL involves providing a Conservation Payment to fund a net increase in habitat for GCN across the landscape, rather than specifically within and around the PEIR boundary, as is involved in standard EPS mitigation licensing.

1. BACKGROUND

Equinor New Energy Limited (hereafter the Applicant) is proposing to extend the existing operational Dudgeon and Sheringham Offshore Wind Farms, named the Dudgeon Extension Project (hereafter DEP) and Sheringham Extension Project (hereafter SEP). DEP and SEP will consist of a number of offshore and onshore elements including the offshore wind turbines and subsea array cables, up to two offshore substations, offshore and onshore export cables, and a new area for up to two onshore substations to accommodate the connection of DEP and SEP to the transmission grid. A full description of DEP and SEP is provided within Chapter 5 Project Description. '

In August 2019, WFE was commissioned by the Applicant to undertake surveys to establish the presence and/or likely absence of GCN *Triturus cristatus* in ponds within and up to 250m of the PEIR boundary to inform an ecological impact assessment of the proposed onshore grid connection for the DEP and SEP. The current onshore proposals comprise a c.60km route with landfall location around Weybourne on the North Norfolk coast, with the route then running southwards and eventually eastwards around the west and south sides of Norwich, where it is to connect with a proposed onshore electricity substation, feeding into the National Grid near Norwich Main Substation.

There have been ongoing refinements to the PEIR boundary since August 2019; in general, the boundary is 200 metres wide, but with some wider sections such as around the landfall location and the substation search zone. The PEIR boundary will be refined further, and will become the finalised Development Consent Order (DCO) boundary.

Maps showing the survey area (i.e. the PEIR boundary plus the surrounding 250m area) are provided in Figures 1-12, below.

This report outlines the aims, methods and results of the surveys for GCN which have been completed in March to June 2020.

2. RELEVANT LEGISLATION AND POLICY

The GCN is fully protected in accordance with both national and international legislation. The species is listed under Annexes IV and II of European Directive 92/43/EEC, and Schedule 2 of The Conservation of Habitats and Species Regulations 2017 (as amended). EU laws supporting species protection are, from 31st January 2020, transposed into UK law and are referred to as The Conservation of Habitats and Species (Amendment) (EU Exit) Regulations Act 2019. The Act keeps in place all 'EU-derived domestic legislation' (clause 2), and incorporates 'direct EU legislation' such as EU environmental regulations into UK domestic law (clause 3). The GCN is also protected by Sections 9(4) and 9(5) of the Wildlife and Countryside Act 1981 as amended.

It is an offence to knowingly or recklessly kill, injure, disturb, handle or sell the animal, and this protection is afforded to all life stages. It is unlawful to deliberately or recklessly damage, destroy, or obstruct the access to any structure or place used for shelter or protection; this includes both the terrestrial and aquatic components of its habitat.

3. SURVEY METHODS

3.1. Desk Study

During the Terrestrial Ecology and Ornithology Expert Topic Group (ETG) meeting on 28th January 2020, attended by Natural England, the Environment Agency, Broadland District Council, Norfolk County Council, North Norfolk District Council and South Norfolk District Council, it was agreed that ponds within and up to 250m from the PEIR boundary should be surveyed for GCN.

Ponds within this survey area (i.e. within and up to 250m from the PEIR boundary) were identified from a desk-based review of Ordnance Survey (OS) maps and other freely available mapping software such as Google Earth. Ponds were mapped as points onto a Geographic Information System (GIS) programme (QGIS) and assigned a unique individual reference, typically a P (denoting Pond) followed by a three-digit number (e.g. P123) or a P and another letter followed by a three digit number (e.g. PW123).

At the time of the desk study (between January and March 2020) the PEIR boundary had three distinct sections: a southern section running from the proposed onshore substation zone northwards to the area around Swannington/Alderford, where the boundary diverged into a western section running north to a potential landfall location at Weybourne, and an eastern section running north-east to a potential landfall location at Bacton.

For those ponds in the southern section of the corridor (which would be used whichever east or west arm of the corridor was selected) a reference of Pxxx (e.g. P001, P002 etc.) was assigned. Ponds in the eastern section from the divergence point to the Bacton landfall were referenced PExxx and ponds in the western section from the divergence point to the Weybourne landfall were referenced PWxxx.

Whilst the HSI and eDNA surveys were underway, the western route was selected as the preferred option. This resulted in the removal from the survey scope of all 45 ponds in the eastern section. In addition, some sections of the PEIR boundary were realigned, which also resulted in 64 of the originally identified ponds now being located outside the GCN survey area and 130 new (previously unreferenced) ponds were now within the GCN survey area. These newly identified ponds were assigned references PNxxx (e.g. PN001, PN002 etc.).

Potential sites for the new onshore substation near the Norwich Main Substation were identified whilst the 2020 GCN HSI and eDNA survey effort was being undertaken. A 250m buffer around these potential onshore substation sites was applied and consequently all ponds within this onshore substation zone were added to the GCN survey area. The 10 newly identified ponds within these areas were referenced as PSxxx (e.g. PS001, PS002 etc.).

During the HSI and eDNA surveys, the field surveyors occasionally encountered new ponds which were not shown on OS maps and were not visible on aerial photographs, such as newly installed garden ponds or ponds within woodland. These 11 newly identified ponds were referenced as PXxxx (e.g. PX001, PX002 etc.).

In general, numbering started at 001 at the southern end of the PEIR boundary and increased moving northwards, so, for example, pond P001 is at the very southern end of the PEIR boundary (within the onshore substation zone) and pond PW204 is at the northernmost point of the boundary near the proposed landfall location near Weybourne.

At the time of the surveys between April and June 2020, a total of 290 ponds were identified within the PEIR boundary and the surrounding 250 metre buffer; this included all originally identified ponds (with references beginning P or PW), all newly identified ponds following the first refinement of the PEIR boundary (with references beginning PN), all ponds within the onshore substation zone and the surrounding 250 metre buffer (with references beginning PS) and all other ponds identified in the field (with references beginning PX). Surveys were completed of all accessible ponds, so excluding those to which landowner access was not granted or ponds which were dry, removed or unsafe to survey.

In December 2020, the PEIR boundary was further refined, resulting in the removal of 111 ponds of the 290 ponds which had been within the survey area based on the PEIR boundary as of April-June 2020. Ponds which were surveyed but now fall outside of the refined PEIR boundary and its surrounding 250 metres are not included in this report because they are no longer relevant to the GCN assessment. The boundary refinement brought 52 new ponds inside the survey area. These ponds had not previously been referenced and had not been surveyed in 2020. These have been identified, referenced and mapped using the same approach as outlined above. These 52 ponds are referenced PAxxx (e.g. PA001, PA002 etc.). As these 52 ponds were identified following a PEIR boundary refinement in December 2020, none of these ponds have yet been visited or surveyed. Surveys of these ponds are scheduled for April-June 2021 (assuming landowner access is permitted)

3.1.1. GCN Survey Data Provided by UCL Pond Restoration Research Group

One of the landowners of a parcel of land which the PEIR boundary passes through has connections to the University College London (UCL) Pond Restoration Research Group (UCLPRRG) which studies ponds and engages in the restoration and conservation of ponds in various parts of Norfolk, including part of the PEIR boundary. The studies include recording whether ponds support breeding GCN. Dr Carl Sayer of the UCLPRRG provided WFE with GCN survey data compiled between 2011 and 2020 for ponds between Baconsthorpe and Bodham[1]. A review of the data revealed seven of these ponds are within the survey area, including two which had not been surveyed by WFE in 2020, as landowner access had not been granted.

Precise survey methodologies used by the UCLPRRG are not outlined in the report. However, the studies on which the GCN data is based date back to 2011 (before eDNA for GCN was known to be available), and relate to breeding GCN, which cannot be determined by eDNA surveys alone. Therefore, surveys are not expected to have used eDNA sampling; instead, more conventional pond survey techniques such as dip-netting or setting traps in the ponds overnight are thought to have been used.

3.1.2. NBIS GCN Records

A data search was completed with the Norfolk Biodiversity Information Service (NBIS) in January 2021 for all biological records (including of GCN) within the PEIR boundary and surrounding 2km area. A wider search area was used than the survey area (restricted to the PEIR boundary and the surrounding 250m) because some biological records are defined to a 1km grid square, so a wider search area is required to ensure all relevant records are obtained.

[1] Sayer C. (2020). *Threats to pond networks associated with the Equinor cable – Information provided by Carl Sayer and the Norfolk Ponds Project.* Unpublished report.

3.2. Habitat Suitability Index

All accessible ponds within the survey area were appraised for their suitability to support GCN using the Habitat Suitability Index (HSI) per Oldham (2000)[2] and the classification guide defined by the Amphibian and Reptile Groups of the United Kingdom (2010)[3]. All pond appraisals took place between March and June 2020.

The HSI is an indicative tool used to rate the suitability of ponds for GCN, based on ten characteristics and features such as size, water quality, vegetation cover and quality of surrounding terrestrial habitat. These features are assessed, classified according to prescribed criteria and assigned a numerical score. These scores allow the HSI to categorise ponds into one of five ratings which indicate their suitability for use by GCN. The five categories and the score parameters (between 1 and 0) are as follows:

- Excellent: >0.8
- Good: 0.7 – 0.79
- Average: 0.6 – 0.69
- Below average: 0.5 – 0.59
- Poor: <0.5

The HSI appraisals were completed by the following Wild Frontier Ecology staff (always working in pairs):

- Alex Lowe BSc MArborA
- Ptolemy McKinnon BSc MSc
- Justin Parry BSc
- Alice Petherick BA
- William Riddett BA ACIEEM (Natural England class licence reference 2015-19075-CLS-CLS).
- Graham Riley BSc ACIEEM (Natural England class licence reference 2019-43743-CLS-CLS)
- Katrina Salmon BSc
- Adam Stickler BSc MSc ACIEEM (Natural England class licence reference 2019-43544-CLS-CLS)
- Robert Yaxley BSc CEnv CEcol MCIEEM (Natural England class licence reference 2016-19382-CLS-CLS)

[2] Oldham R., Keeble J., Swan M. and Jeffcote, M. (2000). Evaluating the suitability of Habitat for Great Crested Newt (*Triturus cristatus*). Herpetological Journal 10: 143-155.

[3] ARG UK. (2010). ARG UK Advice Note 5, Great Crested Newt Habitat Suitability Index. Amphibian and Reptile Groups of the United Kingdom

All surveys were completed by Natural England licensed surveyors or accredited agents (i.e. surveyors permitted to do the surveys under the permission of the licence holder).

3.3. Presence/Absence Survey using eDNA Testing

Within the survey area, each accessible pond was surveyed to collect water samples for eDNA analysis using a SureScreen Diagnostics Ltd. eDNA sampling kit. The survey employed the methodology outlined by DEFRA[4], Natural England[5] and the Freshwater Habitats Trust[6].

Twenty water samples were taken from each pond using sterile equipment: samples were taken using gloves and a ladle from across all accessible parts of each pond, concentrating on areas which the surveyor considered had greatest potential to be used by GCN.

The surveyors did not enter the water in order to ensure there was no accidental contamination (e.g. from footwear), so all samples were collected by reaching into the pond from the shoreline. For each pond, the water samples were all poured into a mixing bag and combined. Water samples were then transferred with a pipette from the mixing bag into six sealed test tubes partly pre-filled with preservative. These tubes were resealed and then posted to SureScreen Diagnostics Ltd. for laboratory analysis. This process was completed for each surveyed pond. All surveys were completed between 28th April and 30th June 2020.

[4] http://randd.defra.gov.uk/Default.aspx?Menu=Menu&Module=More&Location=None&ProjectID=18650&FromSearch=Y&Publisher=1&SearchText=wc1067&SortString=ProjectCode&SortOrder=Asc&Paging=10#Description

[5] https://www.gov.uk/guidance/great-crested-newts-surveys-and-mitigation-for-development-projects

[6] Freshwater Habitats Trust (2015) *Pondnet: How to collect an eDNA sample*. Available online at https://freshwaterhabitats.org.uk/wp-content/uploads/2015/08/eDNA-method-protocol.pdf

4. RESULTS

4.1. Desk Study

In March 2020, a total of 249 ponds were initially identified within the survey area based on the PEIR boundary at that time. Refinement of the PEIR boundary resulted in the removal of 110 of the originally identified 249 ponds (as these were no longer within the survey area) and the addition of 130 new ponds. The addition of the onshore substation zone to the GCN survey area incorporated an additional 10 ponds. A further 11 ponds (which were not shown on maps) were found during field surveys. Therefore, a total of 290 ponds were identified within the survey area based on the PEIR boundary under consideration as of June 2020.

Of the 290 ponds, 89 are within the original southern section of the PEIR boundary (so have a P prefix), 50 are within the original western section of the PEIR boundary (so have a PW prefix), 130 are within the newly realigned sections of the PEIR boundary (so have a PN prefix) and 10 are within the onshore substation search area (so have a PS prefix). The 11 ponds identified within the survey area during field surveys have a PX prefix.

However, in December 2020 the PEIR boundary was further refined, resulting in the removal of 111 ponds which now fall outside of this boundary and its surrounding 250 metre buffer. The remaining 179 ponds remain relevant to the GCN assessment and therefore form the basis of this report. Another review of OS maps and aerial photographs identified an additional 52 ponds which are now within the survey area but had previously been outside it. These 52 ponds have been mapped and referenced but as they were brought inside the survey area in December 2020, they have not yet been surveyed.

4.1.1. GCN Survey Data Provided by UCL Pond Restoration Research Group

The data, which is appended in full, shows that the UCLPRRG studies have found breeding GCN within six of the seven ponds within the survey area. This includes two ponds which were not surveyed by WFE in 2020 (ponds PW179 and PW182) due to lack of landowner access. Records of breeding GCN were also provided for two ponds which were surveyed by WFE in 2020 but for which negative eDNA results were obtained (ponds PW181 and PW183). The results are included in the Notes column of Table 1, below.

4.1.2. GCN Data Provided by Norfolk Biodiversity Information Service

The NBIS data search returned 18 records of GCN within the PEIR boundary and the surrounding 250 metre buffer. These records were provided by NBIS with locations defined to a grid reference. These have been mapped and overlaid with the pond location maps to attempt to assign each NBIS GCN record to a known pond (none of the records are of terrestrial GCN, so are all assignable to a water-body). Some grid references are given to a low resolution so it is not certain which pond these records relate to based on their location alone. In such cases, the description of the record has been used to inform which pond the record is assigned to. Where there is a residual level of uncertainty as to which pond a record definitely relates to, this is listed in Table 1, below.

The records are clustered around Bodham, with 12 of the 18 NBIS GCN records attributed to five ponds (PW175, PW180 [7 records], PW181, PW182 [2 records] and PW183) in and around Pond Farm south of Bodham. Ponds PW175, PW181 and PW183 were surveyed by WFE in 2020 but negative eDNA results were returned. Pond PW180 was surveyed and returned a positive eDNA result. Pond PW182 was not accessible for survey by WFE in 2020.

Four of the records are clustered around Ketteringham, in ponds P016 (assumed to be this pond based on its written description), P024, P025 and another record which could also be in pond P024 or pond P025 but is not of sufficient accuracy to confidently assign to either. However, given the low accuracy grid reference of this record (defined to a 1km grid square only), it is possible it relates to a pond outside the survey area. In any case, the record would still relate to the same metapopulation. Ponds P016, P024 and P025 were all surveyed by WFE in 2020, but only P024 returned a positive eDNA result.

Two of the NBIS GCN records have been assigned to ponds PW166 and PN103 near Saxthorpe and Itteringham. Both these ponds were surveyed by WFE in 2020, with pond PW166 returning a positive eDNA result but pond PN103 returning a negative result.

Further records of GCN were provided by NBIS but mapping has revealed these are outside the survey area, so they are not included in results section of this report (see Table 1).

4.2. Habitat Suitability Index and eDNA Results

The results from the HSI appraisals are presented in Table 1, below, along with the eDNA results.

Full details of the HSI appraisals and pond photographs are appended as a separate document.

The eDNA analysis reports provided by SureScreen Diagnostics Ltd. are provided within this report (see below). The SureScreen Diagnostics Ltd. reports include results for many of the 111 ponds which were surveyed but which are now outside of the refined PEIR boundary and its surrounding 250 metre buffer.

Maps showing the locations of the ponds subject to the HSI and eDNA survey effort are provided in Figures 1-12.

Table 1: HSI Results (note: this table should be read in conjunction with Figures 1-12).

Pond Reference	HSI Score	HSI Classification	eDNA Result	SureScreen Sample Kit Reference	Notes
P001	0.56	Below average	Negative	5324	-
P002	0.60	Average	Negative	5322	-
P003	-	-	-	-	Pond dry
P004	-	-	-	-	Pond dry
P005	0.56	Below average	Negative	5319	-
P006	0.58	Below average	Negative	5326	-
P007	0.65	Average	Negative	5323	-
P008	-	-	-	-	Pond dry
P009	-	-	-	-	Pond dry
P010	0.58	Below average	Negative	2864	-
P011	-	-	-	-	Pond dry
P012	0.68	Average	Negative	5313	-
P014	0.76	Good	POSITIVE	683	-
P015	0.54	Below average	Negative	1301	-
P016	0.56	Below average	Negative	2862	NBIS record of GCN presence from 2008, likely relates to this pond (the grid reference of this record is of low accuracy but the pond description suggests it relates to this pond rather than PX001 which is also nearby).
P017	0.90	Excellent	Negative	700	-
P018	0.73	Good	POSITIVE	1335	-
P019	0.87	Excellent	Negative	704	-
P020	-	-	-	-	Access not granted
P021	0.71	Good	POSITIVE	1369	-
P022	0.80	Good	Negative	3548	-
P023	0.82	Excellent	Negative	3589	-
P024	0.81	Excellent	POSITIVE	3587	NBIS record of GCN presence from 2014. There is another NBIS record of GCN presence from 2006 inside the same 1km grid square as this pond. This may relate to this pond, to P025 or to another pond within the 1km grid square but outside the survey zone.

Pond Reference	HSI Score	HSI Classification	eDNA Result	SureScreen Sample Kit Reference	Notes
P025	0.93	Excellent	Negative	3549	NBIS record of GCN presence from 2014. There is another NBIS record of GCN presence from 2006 inside the same 1km grid square as this pond. This may relate to this pond, to P024 or to another pond within the 1km grid square but outside the survey zone.
P037	0.54	Below Average	Negative	2844	-
P038	0.57	Below Average	Negative	2845	-
P039	0.70	Average	Negative	3588	-
P040	0.79	Good	Negative	3546	-
P041	0.61	Average	Negative	2873	-
P042	0.62	Average	Negative	1350	-
P043	0.70	Average	Negative	2874	-
P049	-	-	-	-	Pond dry
P050	-	-	-	-	Access not granted
P051	-	-	-	-	Access not granted
P052	-	-	-	-	Access not granted
P053	-	-	-	-	Access not granted
P054	-	-	-	-	Access not granted
P055	-	-	-	-	Access not granted
P056	-	-	-	-	Access not granted
P057	-	-	-	-	Access not granted
P058	0.48	Poor	Negative	1303	-
P067	-	-	-	-	Pond dry
P068		-	-	-	Pond not accessible
P120	0.60	Below Average	POSITIVE	1316	Pond erroneously listed as PN120 in SureScreen report
P121	0.63	Average	Negative	1313	Pond erroneously listed as PN121 in SureScreen report
P122	0.60	Below Average	Negative	1305	-
P123	-	-	-	-	Pond dry
P130	0.53	Below Average	Negative	1329	-
P131	-	-	-	-	Pond dry
P132	0.71	Good	Negative	1331	-
P133	0.59	Below Average	Negative	1371	-
P134	0.71	Good	Negative	1336	-

Pond Reference	HSI Score	HSI Classification	eDNA Result	SureScreen Sample Kit Reference	Notes
P135	0.56	Below Average	Negative	1367	-
P138	0.51	Below Average	Negative	1351	-
P139	-	-	-	-	Pond dry
P140	-	-	-	-	Pond dry
P142	-	-	-	-	Pond dry
P143	0.75	Good	Negative	1341	-
P153	0.56	Below Average	Negative	3580	-
P154	0.72	Good	Negative	2881	-
PN001	0.40	Poor	Negative	1365	-
PN002	-	-	-	-	Pond dry
PN003	0.71	Good	Negative	3585	-
PN004	-	-	-	-	Pond dry
PN005	-	-	-	-	Access not granted
PN006	0.43	Poor	Negative	3542	-
PN007	0.86	Excellent	POSITIVE	3529	-
PN008	-	-	-	-	Pond dry
PN009	-	-	-	-	Pond dry
PN010	0.74	Good	Negative	3531	-
PN011	0.87	Excellent	POSITIVE	3530	-
PN012	0.66	Average	Negative	2850	-
PN013	0.54	Below Average	Negative	3540	-
PN014	-	-	-	-	Pond dry
PN015	0.74	Good	Negative	3537	-
PN016	0.83	Excellent	Negative	3544	-
PN017	0.82	Excellent	-	-	Pond not accessible for eDNA, but visible for HSI appraisal
PN018	0.59	Below Average	-	-	Pond not accessible for eDNA, but visible for HSI appraisal
PN019	0.44	Poor	Negative	3534	-
PN021	-	-	-	-	Pond dry
PN025	0.74	Good	Negative	2838	-
PN026	0.25	Poor	Negative	1306	-
PN027	-	-	-	-	Pond dry
PN029	0.42	Poor	Negative	1323	-
PN030	-	-	-	-	Pond dry

Pond Reference	HSI Score	HSI Classification	eDNA Result	SureScreen Sample Kit Reference	Notes
PN031	-	-	-	-	Pond dry
PN032	-	-	-	-	Pond dry
PN034	0.84	Excellent	Negative	1311	-
PN035	-	-	-	-	Access not granted
PN036	-	-	-	-	Access not granted
PN037	-	-	-	-	Access not granted
PN038	-	-	-	-	Pond dry
PN039		-	-	-	Access not granted
PN040	0.80	Excellent	POSITIVE	1338	-
PN041	0.61	Average	POSITIVE	1349	-
PN070	0.81	Excellent	Negative	3527	-
PN088	0.89	Excellent	Negative	3557	-
PN089	0.66	Average	Negative	1345	-
PN090	-	-	-	-	Access not granted
PN091	-	-	-	-	Access not granted
PN092	0.93	Excellent	Negative	3532	-
PN094	0.77	Good	Negative	2846	-
PN095	-	-	-	-	Access not granted
PN098	0.43	Poor	Negative	1327	-
PN099	-	-	-	-	Pond dry
PN100	-	-	-	-	Pond dry
PN101	0.78	Good	Negative	1322	-
PN102	-	-	-	-	Access not granted
PN103	0.67	Average	Negative	1339	NBIS record of GCN presence from 2009.
PN104	0.77	Good	Negative	3571	-
PN105	-	-	-	-	Access not granted
PN111	-	-	-	-	Access not granted
PN112	-	-	-	-	Access not granted
PN113	0.58	Below Average	POSITIVE	1375	-
PN121	-	-	-	-	Pond dry
PN122	-	-	-	-	Pond dry
PN126	0.80	Excellent	Negative	5136	-
PN128	-	-	-	-	Access not granted
PN129	-	-	-	-	Access not granted
PN130	-	-	-	-	Access not granted

Pond Reference	HSI Score	HSI Classification	eDNA Result	SureScreen Sample Kit Reference	Notes
PN131	0.77	Good	Negative	1299	-
PS001	0.68	Average	Negative	1309	-
PS002	-	-	-	-	Pond dry
PS003	0.73	Good	Negative	2871	-
PS004	0.61	Average	Negative	2849	-
PS005	0.49	Poor	Negative	5312	Pond erroneously listed as P505 in SureScreen report
PS006	-	-	-	-	Pond dry
PS007	-	-	-	-	Pond dry
PS008	-	-	-	-	Pond dry
PS009	-	-	-	-	Pond dry
PS010	0.51	Below Average	Negative	1304	-
PW156	0.52	Below Average	Negative	1317	-
PW157	-	-	-	-	Pond dry
PW158	0.62	Average	Negative	1318	-
PW159	-	-	-	-	Access not granted
PW160	-	-	-	-	Access not granted
PW161	-	-	-	-	Access not granted
PW162	-	-	-	-	Access not granted
PW166	0.59	Below Average	POSITIVE	1298	NBIS record of GCN presence from 2009
PW167	0.85	Excellent	Negative	1302	-
PW168	0.75	Good	Negative	1283	-
PW169	0.71	Good	Negative	1282	-
PW170	0.76	Good	Negative	2882	-
PW171	-	-	-	-	Access not granted
PW172	-	-	-	-	Access not granted
PW173	-	-	-	-	Access not granted
PW174	-	-	-	-	Access not granted
PW175	0.86	Excellent	Negative	693	UCLPRRG has studied this pond but states the breeding GCN status as 'unknown'; their pond reference is POFA4. NBIS record of GCN presence from 2007
PW176	-	-	-	-	Access not granted

Pond Reference	HSI Score	HSI Classification	eDNA Result	SureScreen Sample Kit Reference	Notes
PW177	-	-	-	-	Access not granted
PW178	-	-	-	-	Access not granted
PW179	-	-	-	-	Access not granted. UCLPRRG confirmed GCN breeding in this pond; their pond reference is BAW02.
PW180	0.79	Good	POSITIVE	699	UCLPRRG confirmed GCN breeding in this pond; their pond reference is POFA2. 7x NBIS records of GCN presence from 2007
PW181	0.77	Good	Negative	676	UCLPRRG confirmed GCN breeding in this pond; their pond reference is POFA1. NBIS record of GCN presence from 2013 (record describes "hundreds of eggs" found during survey).
PW182	-	-	-	-	Access not granted. UCLPRRG confirmed GCN breeding in this pond; their pond reference is BAW01. 2x NBIS records of GCN presence from 2007
PW183	0.82	Excellent	Negative	679	UCLPRRG confirmed GCN breeding in this pond; their pond reference is POFA3. NBIS record of GCN presence from 2007
PW184	-	-	-	-	Access not granted
PW185	0.51	Below Average	POSITIVE	1370	-
PW186	0.69	Average	POSITIVE	3570	UCLPRRG confirmed GCN breeding in this pond; their pond reference is HART.
PW192	-	-	-	-	Access not granted
PW193	-	-	-	-	Access not granted

Pond Reference	HSI Score	HSI Classification	eDNA Result	SureScreen Sample Kit Reference	Notes
PW194	-	-	-	-	Access not granted
PW195	-	-	-	-	Access not granted
PW197	-	-	-	-	Access not granted
PW198	0.76	Good	Negative	1281	-
PW199	0.76	Good	Negative	1286	-
PW200	0.51	Below Average	Negative	1291	-
PW201	0.82	Excellent	Negative	1280	-
PW202	-	-	-	-	Pond dry
PW203	0.89	Excellent	Negative	1284	-
PW204	-	-	-	-	Pond not accessible because of Schedule 1 nesting birds using the pond
PX001	0.42	Poor	Negative	694	-
PX003	0.64	Average	Negative	1287	-
PX004	0.59	Below Average	Negative	1285	-
PX005	0.64	Average	Negative	705	-
PX007	0.36	Poor	Negative	1348	Pond erroneously listed as P138a in SureScreen report
PX009	0.47	Poor	Negative	3543	-
PX010	0.58	Below Average	Negative	3541	-
PX012	0.72	Good	Negative	5317	-

All ponds identified following the December 2020 refinement of the PEIR boundary (ponds with references from PA001 to PA052) have not yet been surveyed so are not listed in the above table.

Figure 1: Great Crested Newt Results Map (Onshore Substation Zone to East Carleton)

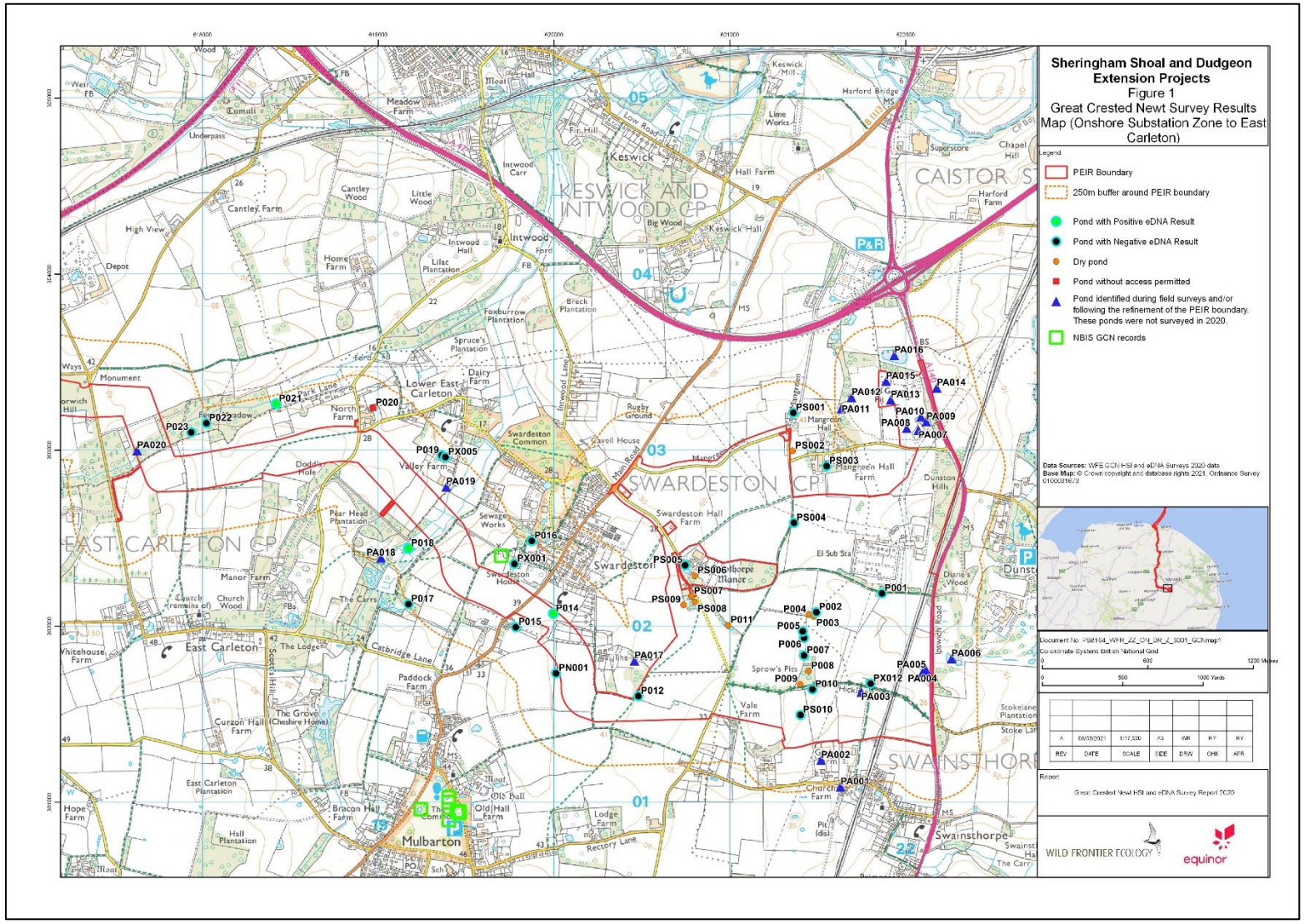


Figure 2: Great Crested Newt Results Map (East Carleton to Great Melton)

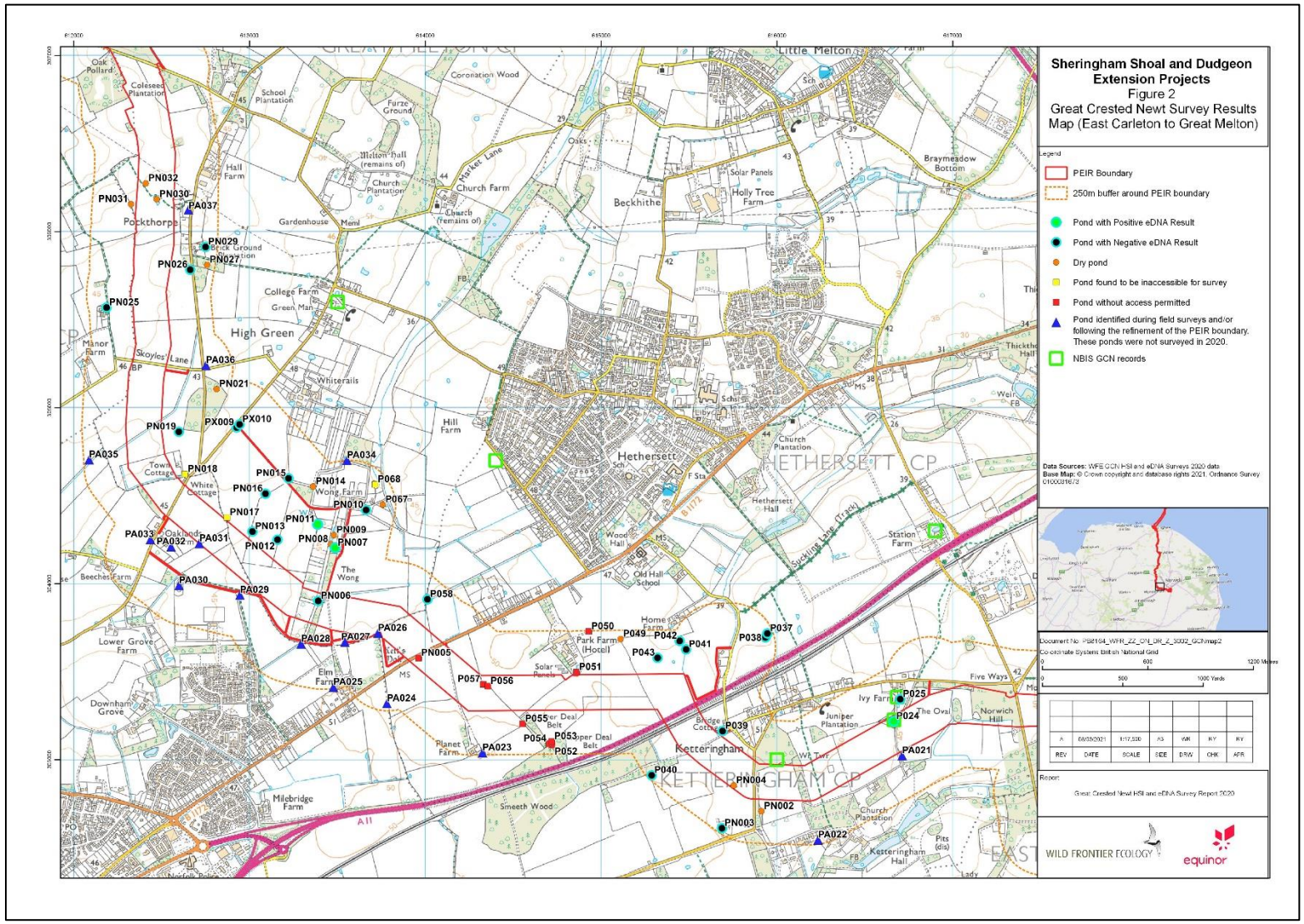


Figure 3: Great Crested Newt Survey Results Map (Great Melton to Easton)

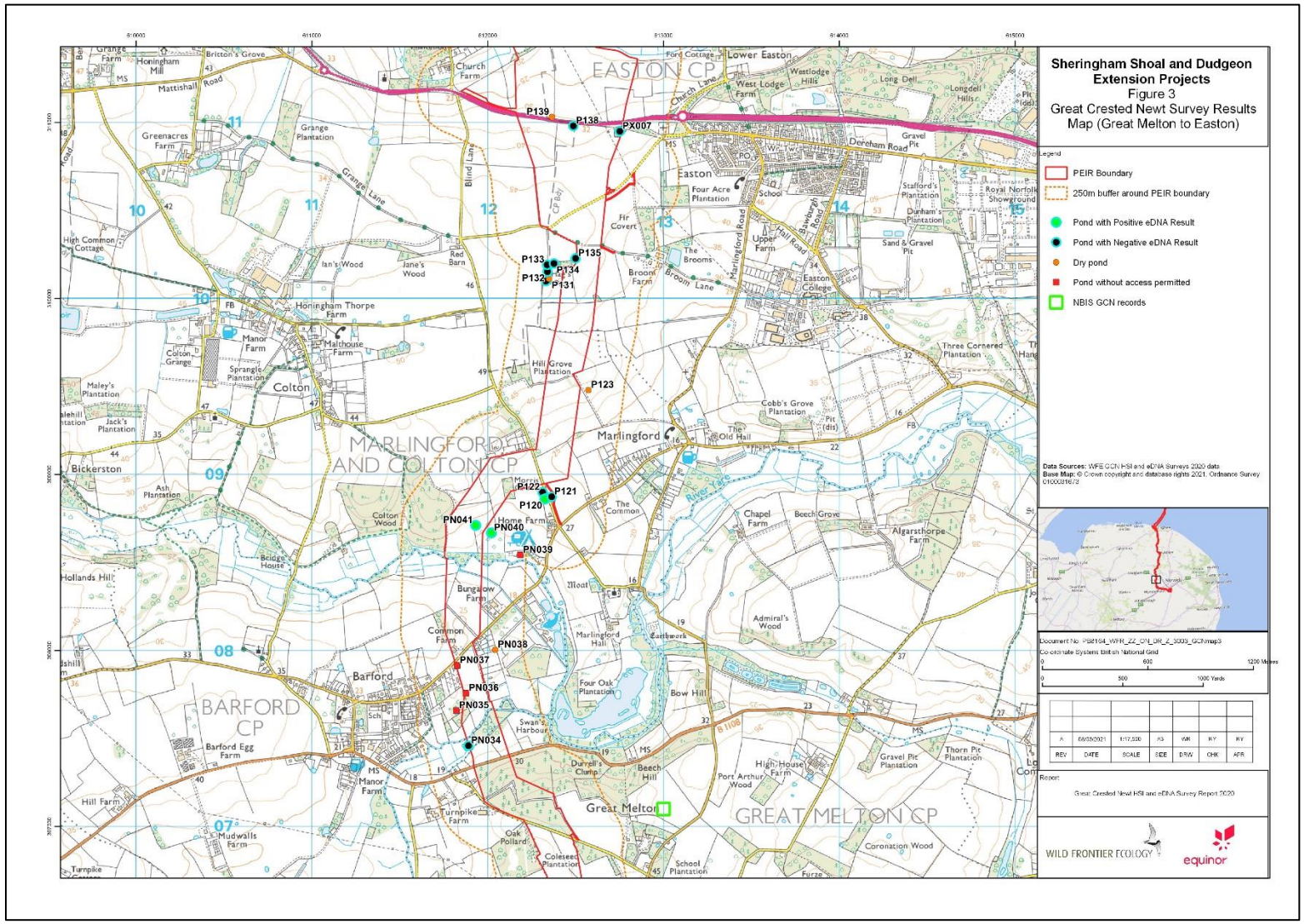


Figure 4: Great Crested Newt Survey Results Map (Easton to Weston Longville)

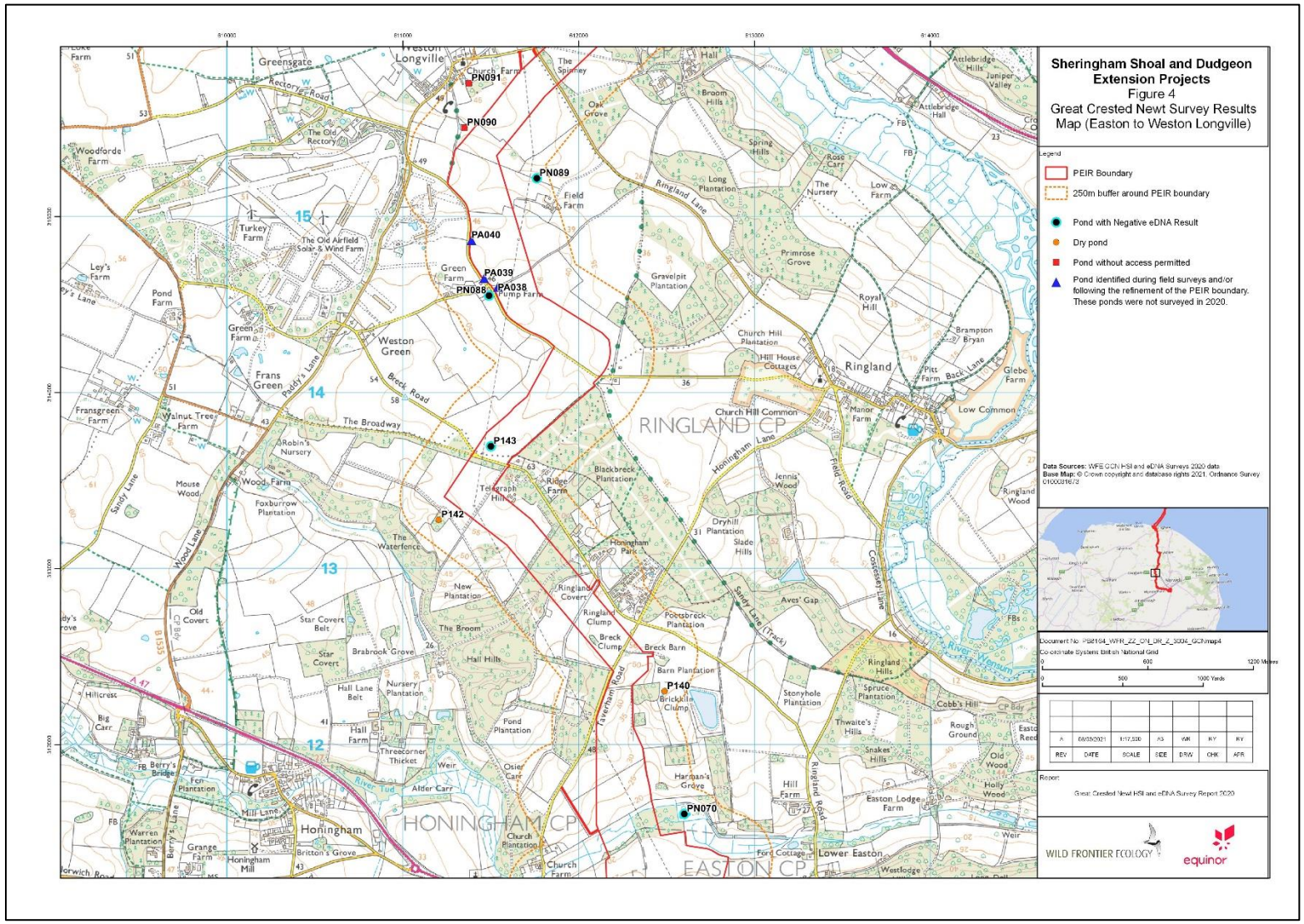


Figure 5: Great Crested Newt Survey Results Map (Weston Longville to Brandiston)

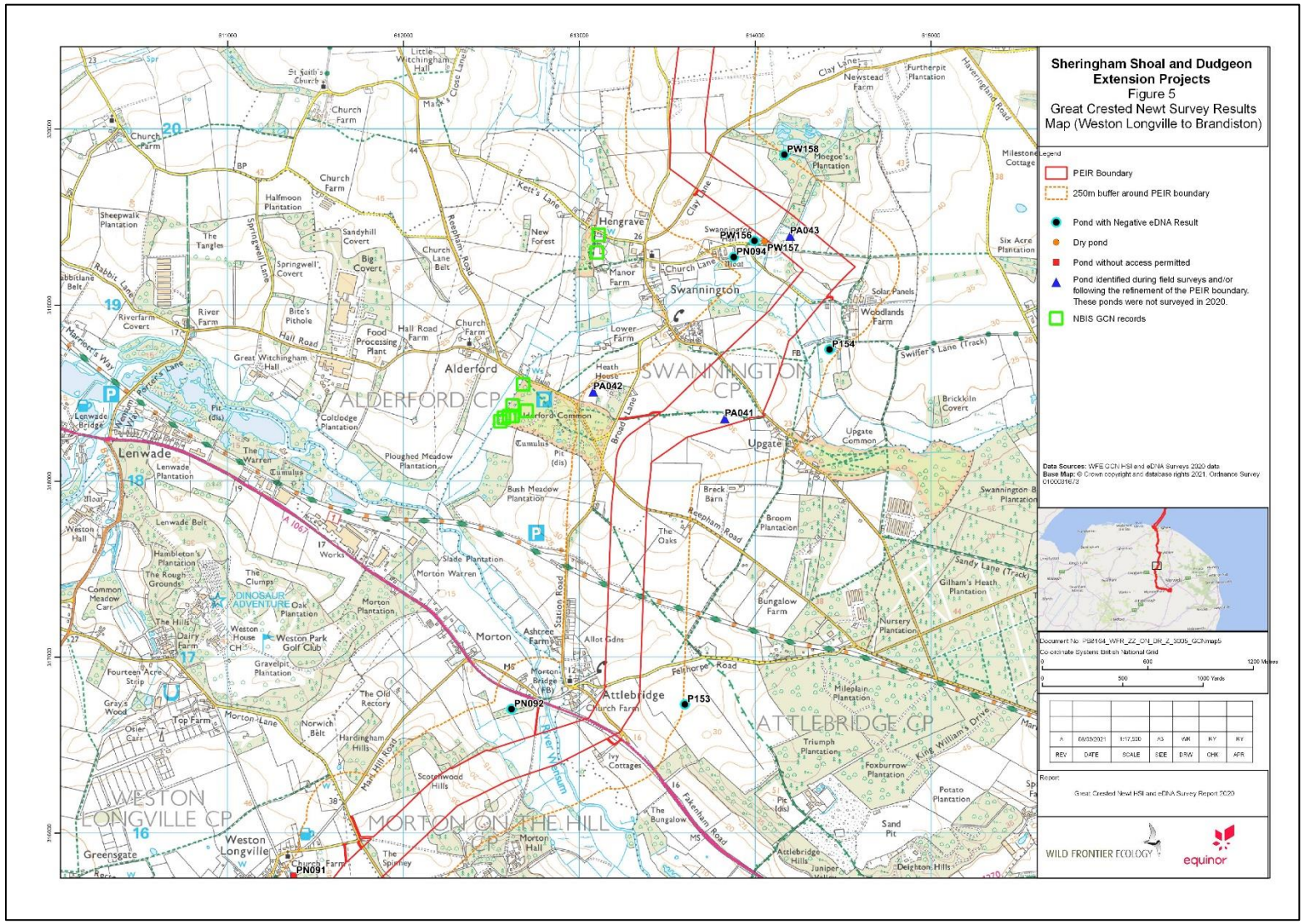


Figure 6: Great Crested Newt Survey Results Map (Brandiston to Cawston)

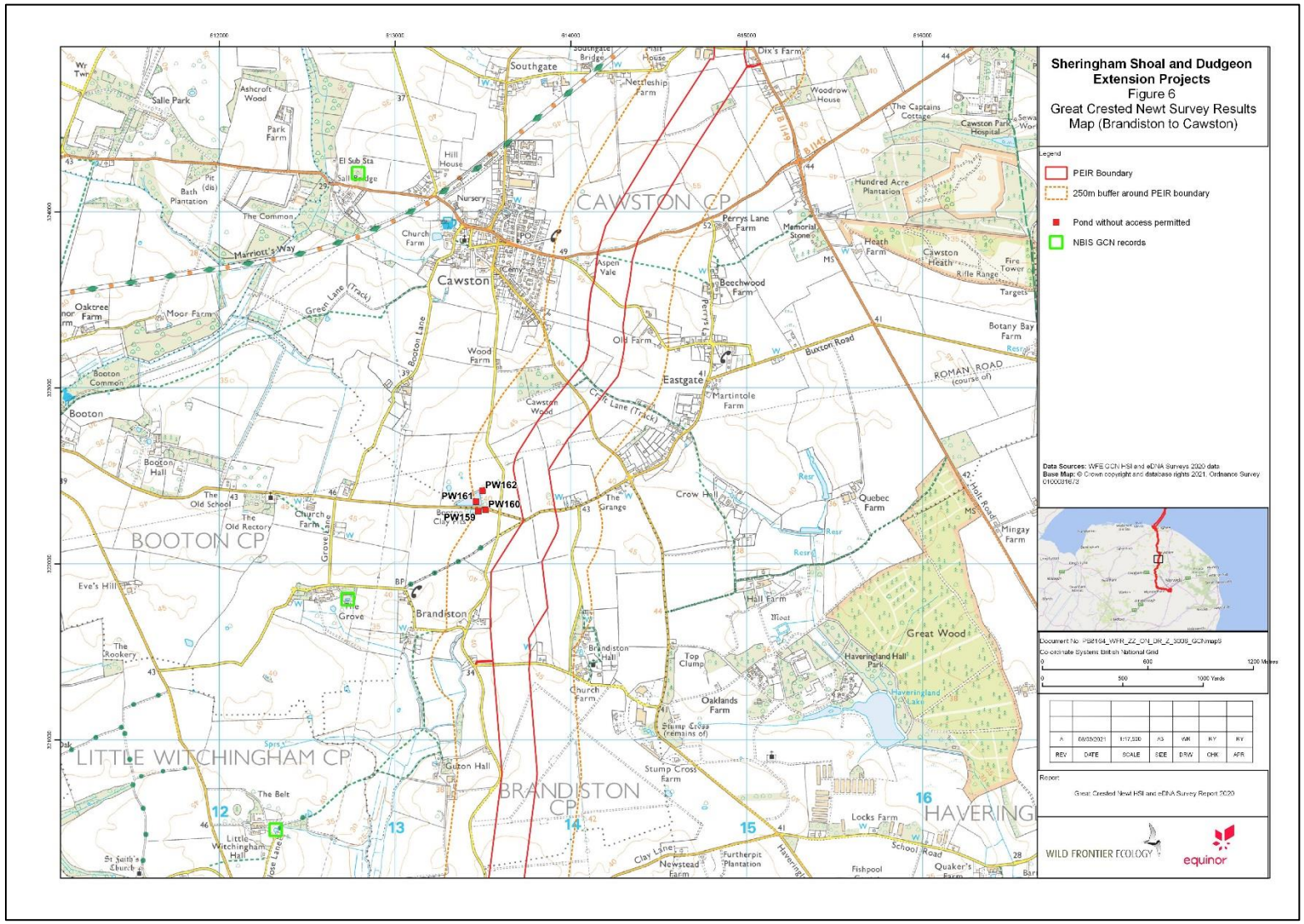


Figure 7: Great Crested Newt Survey Results Map (Cawston to Oulton)

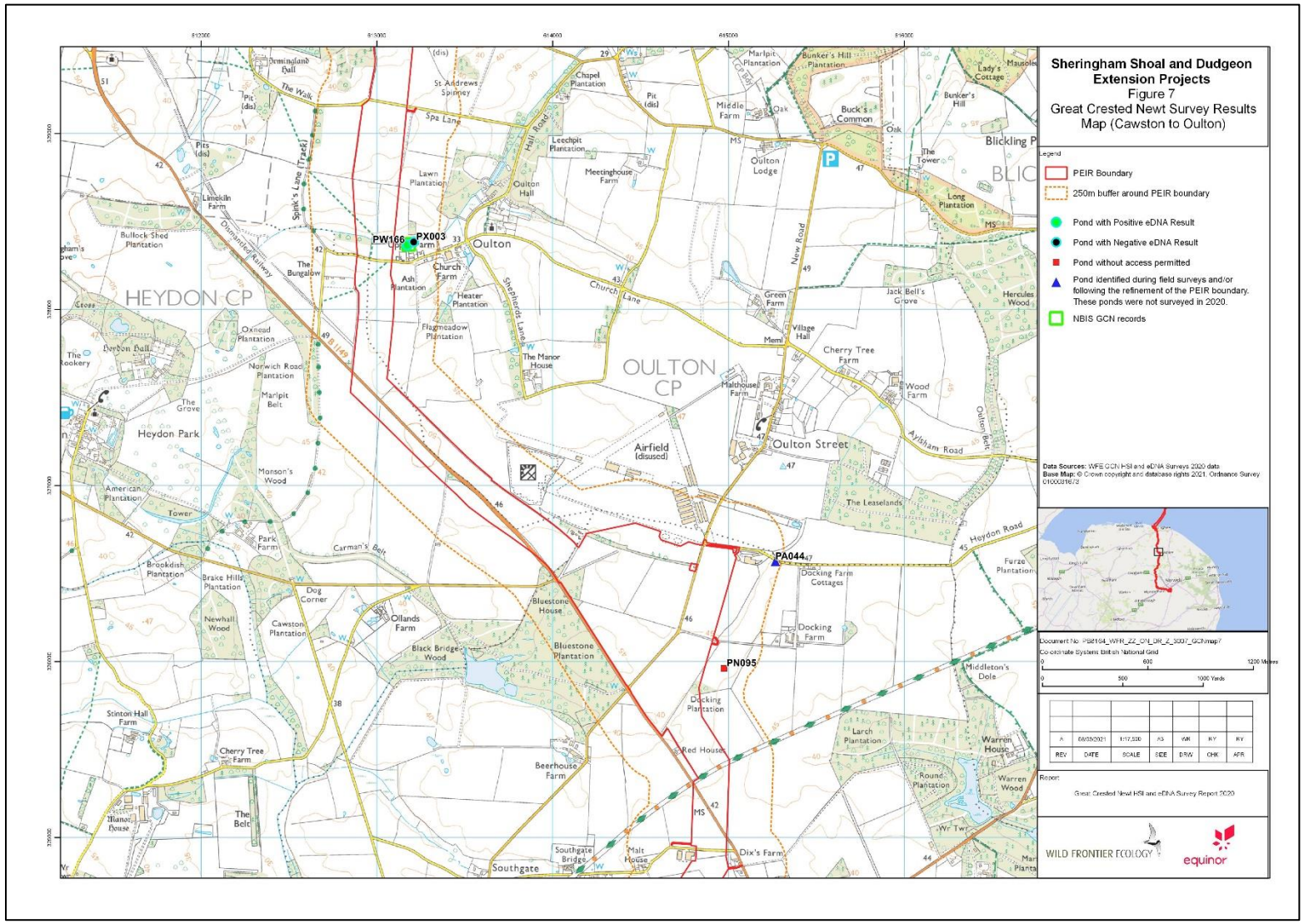


Figure 8: Great Crested Newt Survey Results Map (Oulton to Little Barningham)

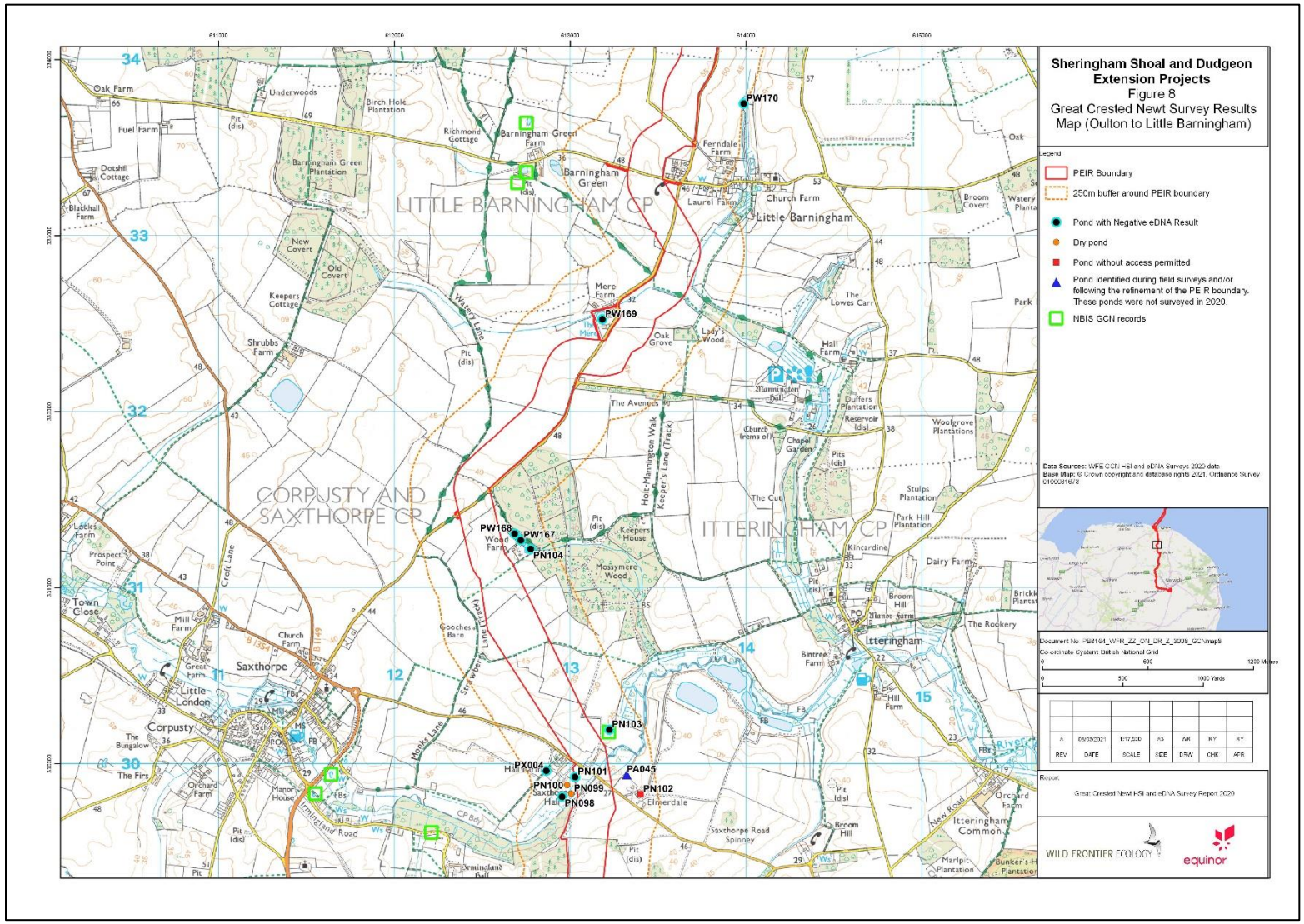


Figure 9: Great Crested Newt Survey Results Map (Little Barningham to Baconsthorpe)

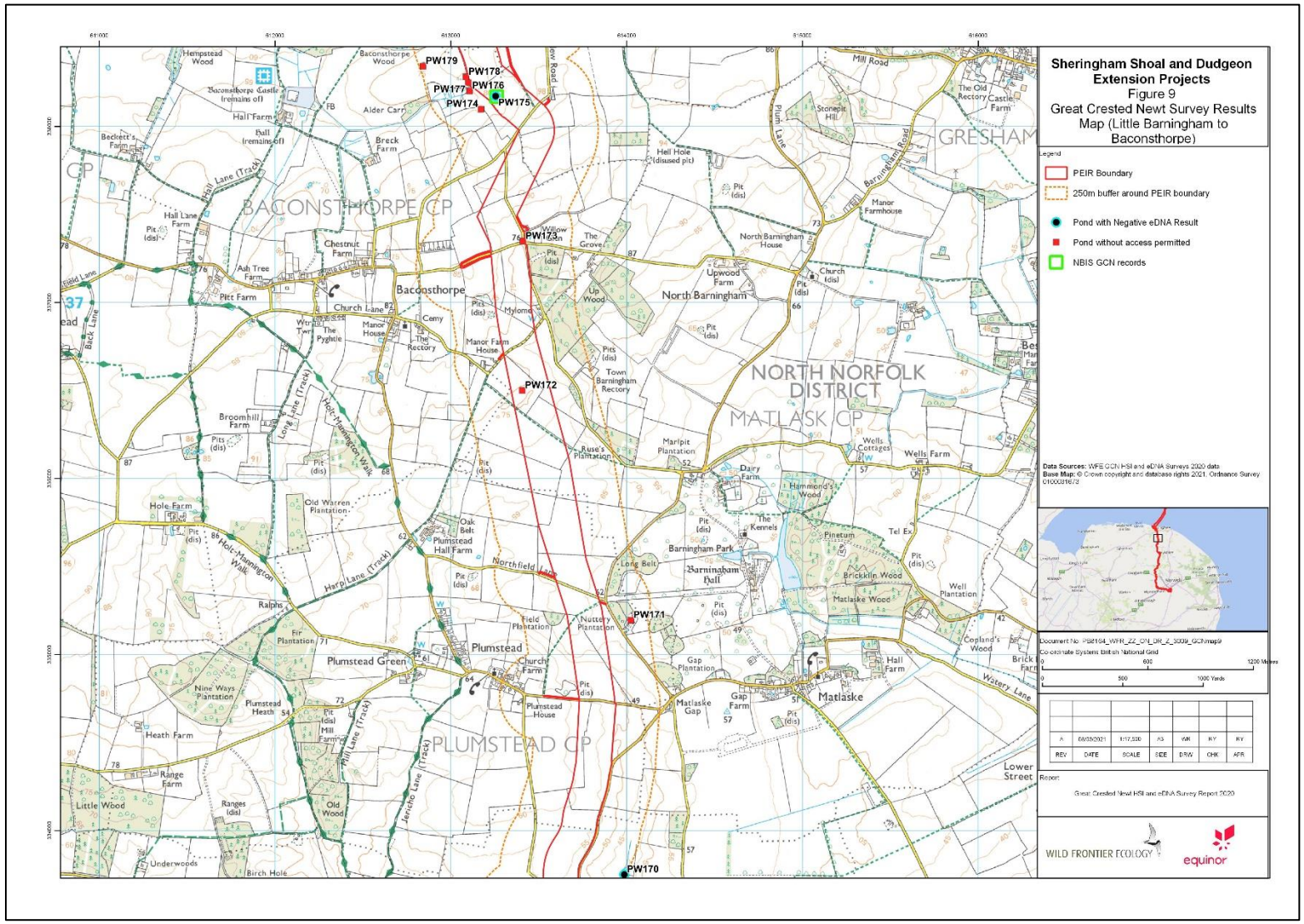


Figure 10: Great Crested Newt Survey Results Map (Baconsthorpe to Bodham)

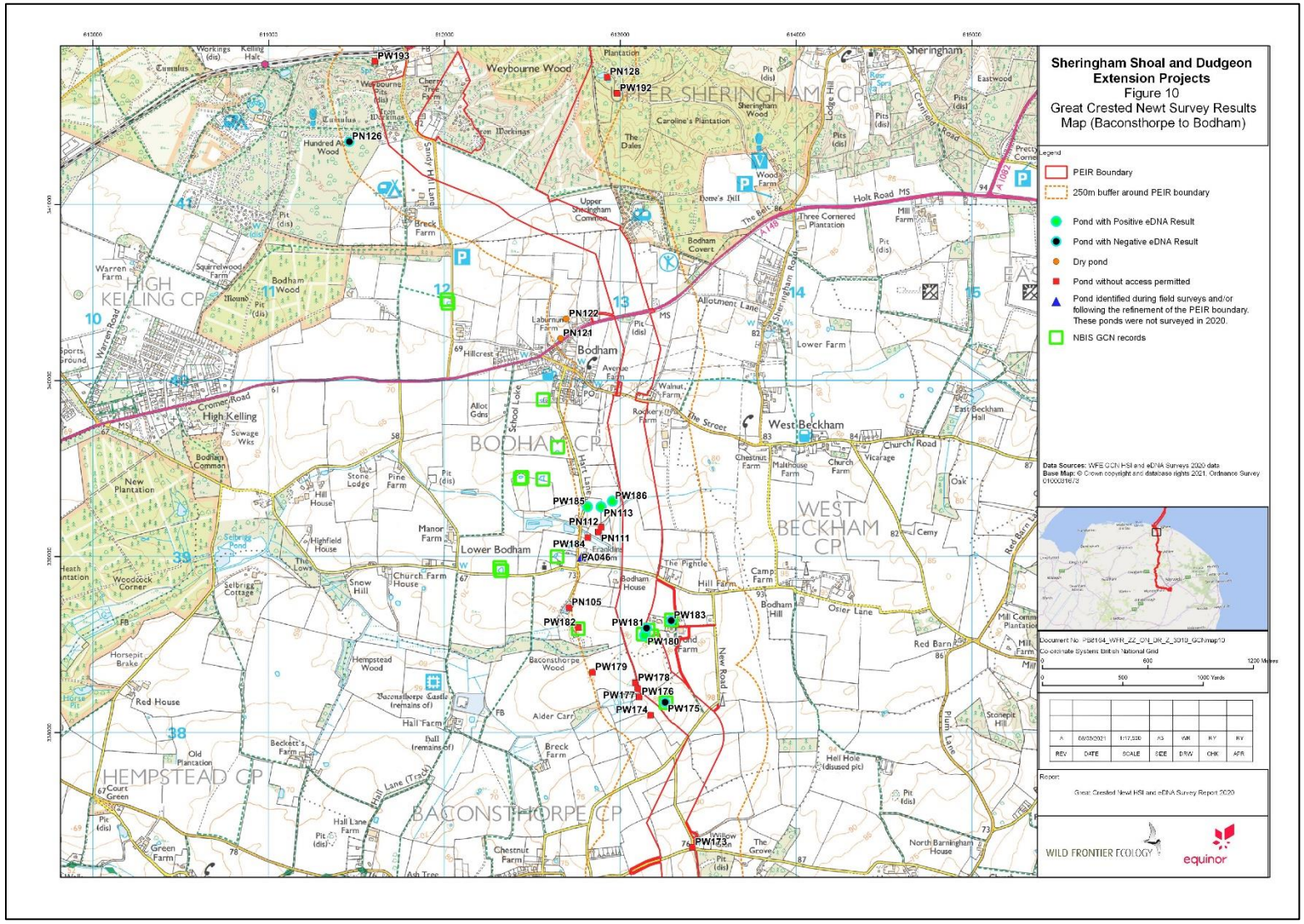


Figure 11: Great Crested Newt Survey Results Map (Bodham to Landfall Location)

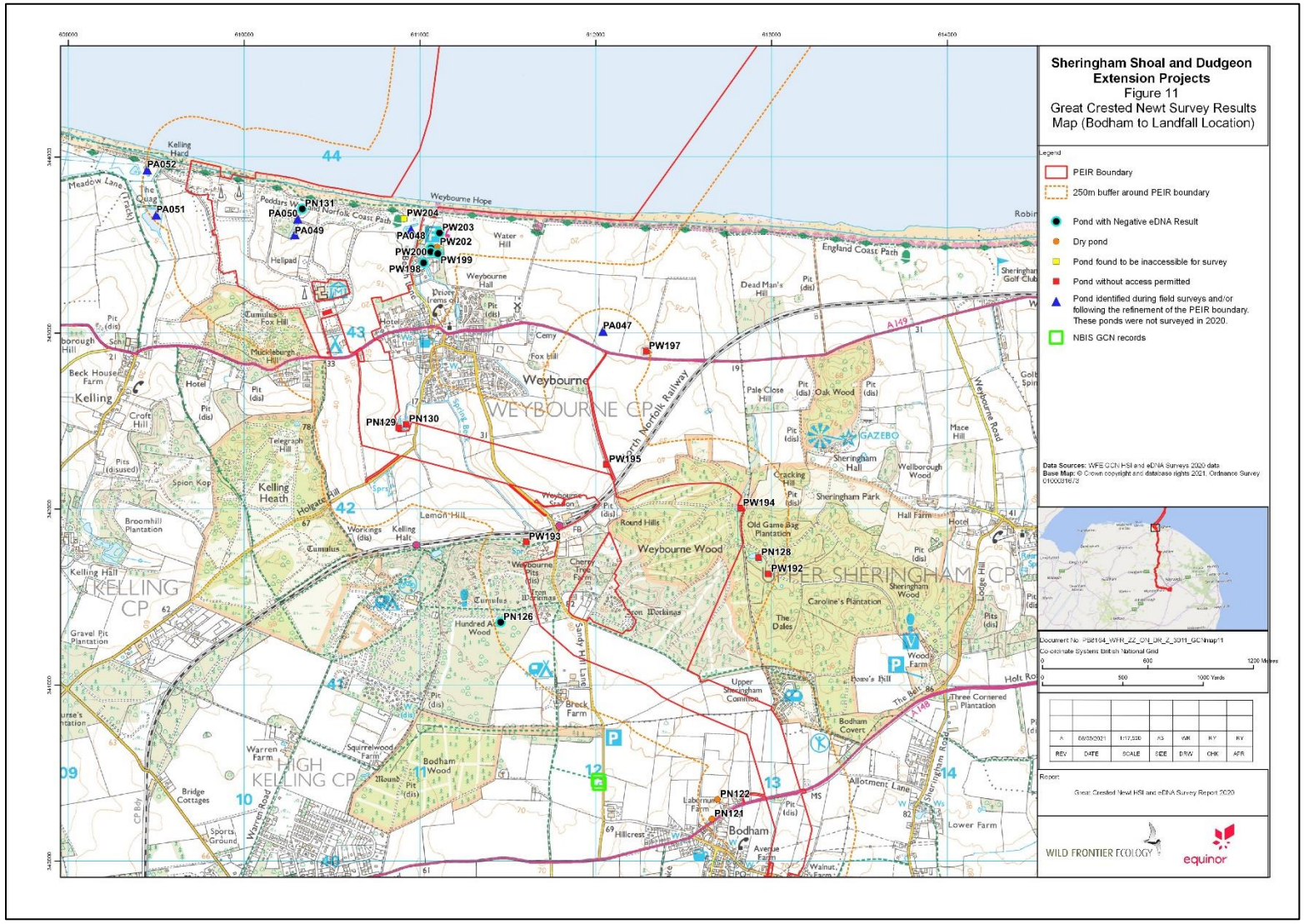
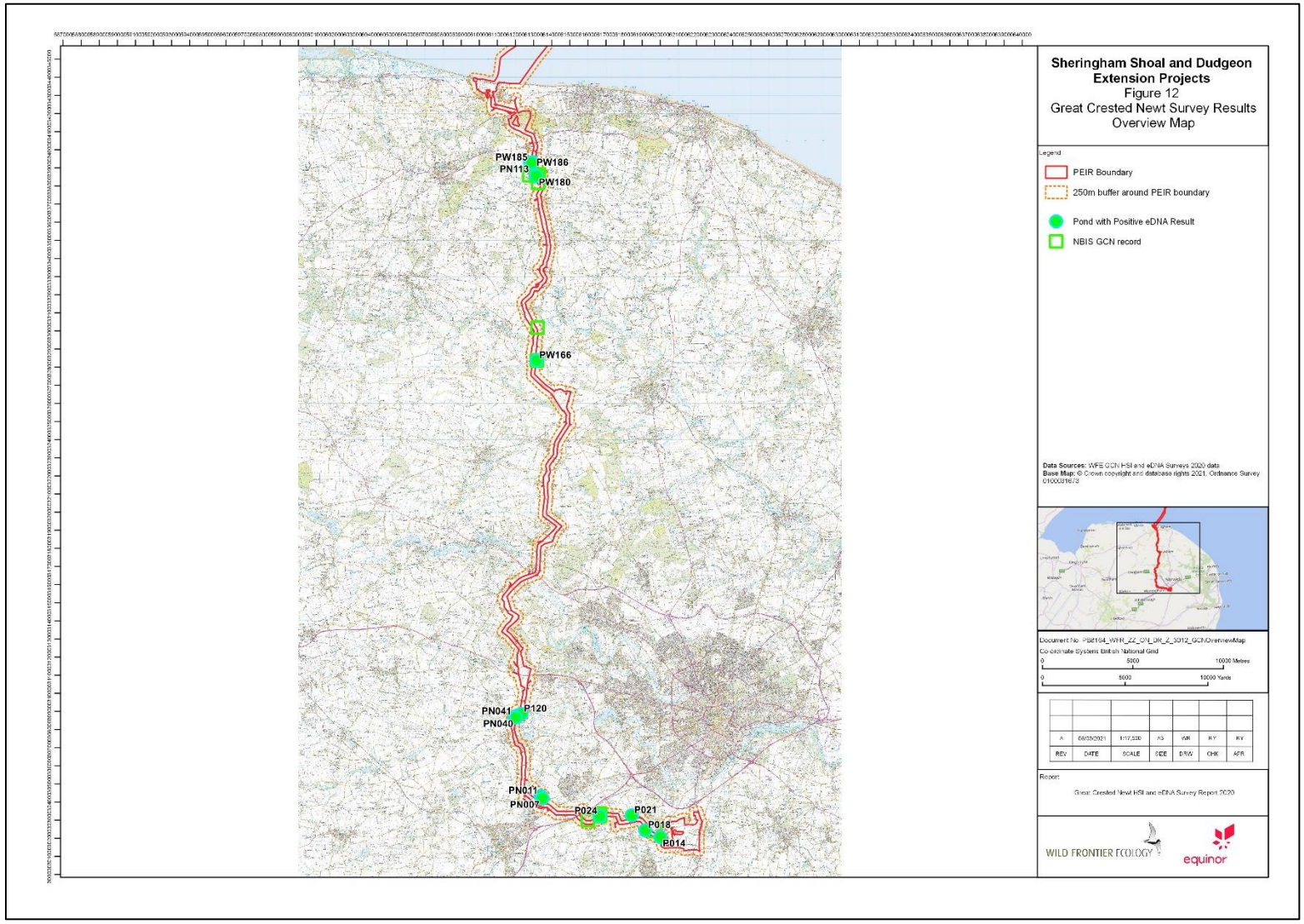


Figure 12: Great Crested Newt Survey Results Overview Map (only showing positive GCN records)



4.3. Constraints and Limitations of Survey

The main constraint to the 2020 GCN HSI and eDNA survey effort related to the limited landowner access at the time of the survey. Three ponds were not physically accessible due to issues such as dangerously steep/unstable banks, impenetrable vegetation around the pond or protected nesting birds using the pond (which meant the pond could not be accessed due to the risk of disturbing the birds). Two of these ponds could be HSI appraised from distance but none of the three was accessible for eDNA surveys.

Thirty-four ponds were found to be dry so could not be surveyed. However, this is not considered a significant constraint because any such ponds are likely unsuitable for use as a breeding pond by GCN if dry during the breeding season (March to June).

There are inherent constraints to the eDNA surveys, such as potential natural contamination, such as from birds or other wildlife transferring eDNA between ponds, which could lead to false Positives. Conversely, there is also the potential for false Negatives for various reasons. For example, access for eDNA sampling at some ponds was severely restricted such as by steep banks, unstable ground, dense vegetation, fences etc., meaning the sampling may have not collected water from parts of the pond used by GCN. This has been acknowledged as a potential constraint partly because WFE has previously surveyed a number of relevant ponds within the survey area (but for other development proposals in the past) which have confirmed GCN presence, yet eDNA sampling in 2020 has returned Negative results. It is possible that GCN are no longer present in some such ponds, but equally it should be noted that eDNA surveys could have returned false Negatives.

SureScreen data returned four incorrect pond references; in these instances, they are acknowledged in Table 1. As each sample kit has a unique 4-digit reference, inconsistencies in pond referencing could be readily corrected because surveyors recorded which kit was used at each pond.

4.4. Further Survey Requirements and Expiry Dates

Government guidelines[7] state that "*Survey data provided by the developer should be less than 2-4 survey seasons old, depending on the extent of the effects.*" For the DEP and SEP, it is considered, as a precaution, that two survey seasons would be an acceptable period of validity of the collected data. The HSI and eDNA survey results should therefore be regarded as valid for up to two years from the date the surveys were undertaken, meaning the data will begin to 'expire' from mid-April 2022, and by the end of June 2022 it will likely be considered invalid in terms of suitability to support a DCO application. However, as the DCO application is due to be submitted in late 2021, the survey data from 2020 will be valid at that time meaning no updates to the survey are expected to be necessary to support this application.

Nevertheless, further surveys of any newly identified and therefore un-surveyed ponds (following refinement of the PEIR boundary and completion of the EP1HS) are due to be completed from mid-April to June 2021. Surveys will also be attempted on ponds which were found to be dry during 2020, as they may hold water in 2021, particularly as many of the ponds were surveyed in late June 2020 when there is an increased likelihood of ponds having dried out.

[7] https://www.gov.uk/guidance/great-crested-newts-surveys-and-mitigation-for-development-projects

The 2021 surveys will involve the same approach as the 2020 surveys, comprising HSI appraisals and eDNA surveys. The survey area (in which surveys of relevant ponds would be completed/attempted) would be the refined PEIR/DCO boundary and the surrounding 250 metre buffer.

EPS mitigation licences may need to be obtained to legally permit works which would impact GCN or their habitat and ensure maintenance of the conservation statuses of local GCN populations. If required, EPS mitigation licence applications will need to be supported with survey data from the most recent survey season. Clearly, construction works which may require EPS licensing will not be completed before April/May/June 2021, meaning the 2020 data will not be able to fully inform any necessary EPS mitigation licence applications. Assuming EPS mitigation licences are required, further surveys will be necessary and must be completed in the survey season before licensable works are due to commence; for example, if licensable construction works are scheduled to commence in October 2025, surveys of relevant/nearby ponds would need to be completed in March-June 2025 to inform the licence application.

Although the precise scope of any such surveys is not currently defined, it is anticipated that any surveys to inform EPS mitigation licence application/s will comprise presence/absence surveys (and corresponding population estimate surveys on ponds in which GCN are confirmed present) of relevant ponds, rather than eDNA surveys. The decision as to which ponds will be incorporated within the scope of EPS-licence related surveys will be made on a case-by-case basis once the precise scope of construction works (and the corresponding predicted impact on GCN) is determined.

An alternative approach to any necessary licensed mitigation could involve the DEP and SEP achieving a DLL. However, the PEIR boundary partly overlaps a DLL 'Red zone' around Upgate near Swannington; this may prohibit DLL as a viable option for DEP and SEP, but this will be confirmed through consultation with Natural England. DLL involves providing a Conservation Payment to fund a net increase in habitat for GCN across the landscape, rather than specifically within and around the PEIR boundary, as is involved in standard EPS mitigation licensing. DLL does not necessarily require completion of GCN surveys to inform the licence application, so the costs and time requirements for presence/absence and population estimate surveys would not necessarily be incurred if taking the DLL approach.

5. CONCLUSIONS

The GCN surveys have confirmed that the species is present in localised parts of the survey area. Although the 2020 HSI and eDNA survey coverage is incomplete due to restricted landowner access and post-survey revisions to the survey area, the survey results obtained to date indicate a number of apparent clusters of ponds supporting GCN, which likely indicate the presence of metapopulations in these areas. From a review of the spatial distribution of ponds with Positive eDNA results and other records from NBIS and the UCLPRRG, these clusters are located in the following general areas:

- Between Swardeston, Ketteringham and Hethersett: ponds P014, P018, P021, P024, PN007 and PN011. Pond P016 has a likely NBIS biological record from 2008 and pond P025 has a NBIS biological record from 2014; these ponds may also support this metapopulation.
- Between Marlingford and Colton: ponds PN040, PN041 and P120.
- South of Bodham: ponds PW180, PW185, PW186 and PN113. Ponds PW175, PW179, PW181, PW182 and PW183 also have various records of GCN presence according to NBIS and UCLPRRG data; these ponds are also likely to support the same GCN metapopulation.

There may also be a metapopulation around Oulton and Saxthorpe. A positive eDNA result was returned for pond PW166 (and there is a NBIS biological record of GCN presence for this pond), and NBIS returned a record of GCN presence at pond PN103, located approximately 1.8km north of PW166.

Further refinement of the PEIR boundary is ongoing and will become the finalised DCO application boundary in due course. Once this boundary is refined, a review of ponds within and up to 250m from the DCO boundary will be undertaken, and further surveys of ponds within the refined survey area will be completed. The findings of these future surveys will be used to inform the GCN impact assessment. In addition, the data from the future surveys will inform the mitigation and enhancement approach to ensure maintenance of the conservation status of the local GCN populations.

Annex 1: SureScreen Diagnostics Ltd. Reports

SureScreen Scientifics

Folio No: E7133
Report No: 1
Purchase Order: 2020/08
Client: WILD FRONTIER ECOLOGY
Contact: Will Riddett

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: 04/05/2020
Date Reported: 13/05/2020
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
0676	Equinor PW181, Pond Farm, Bodham		Pass	Pass	Pass	Negative	0
0683	Equinor Pond 14, Swardestone Pond 14		Pass	Pass	Pass	Positive	12
0687	Equinor PO111, Waiton Equinor		Pass	Pass	Pass	Negative	0
0693	Equinor PW175, Pond Farm, Bodham		Pass	Pass	Pass	Negative	0

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 3

SureScreen Scientifics

0694	Equinor PX1, Waring Swardestone		Pass	Pass	Pass	Negative	0
0699	Equinor PW180, Bodham Pond Farm		Pass	Pass	Pass	Positive	1
0700	Equinor Pond 17, Swardestone Pond 17		Pass	Pass	Pass	Negative	0
0704	Equinor PO19, Mr Cooke, Old Nursery, Swardestone		Pass	Pass	Pass	Negative	0
0705	Equinor PX5, Old Nursery, Swardestone		Pass	Pass	Pass	Negative	0

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

Reported by: Chris Troth **Approved by:** Sarah Evans

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

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

SureScreen Scientifics

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

SureScreen Scientifics

Folio No: E7140
Report No: 1
Purchase Order: 2020/08
Client: WILD FRONTIER ECOLOGY
Contact: Will Riddett

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: 04/05/2020
Date Reported: 12/05/2020
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
0677	Equinor Pond 48, Home Farm Ketteringham		Pass	Pass	Pass	Negative	0
0679	Equinor PW183, Bodham Pond Farm		Pass	Pass	Pass	Negative	0

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

Reported by: Sarah Evans **Approved by:** Chris Troth

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

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.

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

SureScreen Scientifics

Folio No: E7302
Report No: 1
Purchase Order: 2020/08
Client: WILD FRONTIER ECOLOGY
Contact: Will Riddett

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: 12/05/2020
Date Reported: 19/05/2020
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
1282	Equinor PW169, Harris, Matlaske Road		Pass	Pass	Pass	Negative	0
1283	Equinor Pond PW168, Brooks Pond		Pass	Pass	Pass	Negative	0
1300	Equinor Pond 100, Markham		Pass	Pass	Pass	Negative	0
1301	Equinor PO15, Land at Swardestone		Pass	Pass	Pass	Negative	0
1302	Equinor PW167, Brooks,		Pass	Pass	Pass	Negative	0

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

SureScreen Scientifics

Mossymere Wood

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

Reported by: Chris Troth **Approved by:** Sarah Evans

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.

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

The standard text in SureScreen reports on Methodology and Interpretation of Results is included in every report they issue, but to reduce duplication, this text is not included in full for the following eDNA reports. As shown by the first two reports, this text is the same, standard text in each report.

SureScreen Scientifics

Folio No: E7322
Report No: 1
Purchase Order: 2020/08
Client: WILD FRONTIER ECOLOGY
Contact: Katrina Salmon

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: 13/05/2020
Date Reported: 20/05/2020
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
1280	201, Equinor		Pass	Pass	Pass	Negative	0
1281	Equinor PW198, Preston		Pass	Pass	Pass	Negative	0
1284	PW203 Equinor, Preston		Pass	Pass	Pass	Negative	0
1286	199, Equinor - Preston		Pass	Pass	Pass	Negative	0
1291	PW200, Equinor, Preston		Pass	Pass	Pass	Negative	0

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

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

SureScreen Scientifics

Folio No: E7446
Report No: 1
Purchase Order: 2020/08
Client: WILD FRONTIER ECOLOGY
Contact: Will Riddett

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: 20/05/2020
Date Reported: 26/05/2020
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
1285	Equinor Pond PX4, Equinor Pond House, Saxthorpe		Pass	Pass	Pass	Negative	0
1287	Equinor PX3, Rowe, Oulton		Pass	Pass	Pass	Negative	0
1298	Equinor PW166, Equinor Pond NR Oulton		Pass	Pass	Pass	Positive	1
1303	Equinor P058, Equinor Land at Heathersett		Pass	Pass	Pass	Negative	0

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

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

SureScreen Scientifics

Folio No: E7536
Report No: 1
Purchase Order: 2020/08
Client: WILD FRONTIER ECOLOGY
Contact: Will Riddett, Katrina Salmon

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: 28/05/2020
Date Reported: 08/06/2020
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
1299	Equinor PN131, Equinor Muckleburgh Collection		Pass	Pass	Pass	Negative	0
1333	PN108, Equinor		Pass	Pass	Pass	Positive	11
1335	PN117, Equinor		Pass	Pass	Pass	Positive	12
1337	PN115, Equinor		Pass	Pass	Pass	Negative	0
1339	Equinor PN103, Agnew Saxthorpe		Pass	Pass	Pass	Negative	0
1370	Equinor PW185,		Pass	Pass	Pass	Positive	3

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

SureScreen Scientifics

	Knowles Bodham						
1372	PN116, Equinor		Pass	Pass	Pass	Positive	2
1375	Equinor PN113, Equinor near Bodham		Pass	Pass	Pass	Positive	5

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

Reported by: Chris Troth **Approved by:** Sarah Evans

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.

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

SureScreen Scientifics

Folio No: E7613
Report No: 1
Purchase Order: 2020/08
Client: WILD FRONTIER ECOLOGY
Contact: Will Riddett

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: 04/06/2020
Date Reported: 11/06/2020
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
1289	Equinor PN050, Alston Honingham		Pass	Pass	Pass	Negative	0
1328	PN060, Equinor		Pass	Pass	Pass	Negative	0
1330	P136, Equinor		Pass	Pass	Pass	Negative	0
1332	PN062, Equinor		Pass	Pass	Pass	Negative	0
1340	PN065, Equinor		Pass	Pass	Pass	Negative	0
1341	Equinor P143, Ebony Weston Green		Pass	Pass	Pass	Negative	0
1342	P124,		Pass	Pass	Pass	Negative	0

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

SureScreen Scientifics

	Equinor					
1343	P125, Equinor	Pass	Pass	Pass	Negative	0
1344	Equinor P129, Equinor Nr Honingham	Pass	Pass	Pass	Negative	0
1345	Equinor PN089, Wales Weston Longville	Pass	Pass	Pass	Negative	0
1346	Equinor PW164, Friend, Cawston	Pass	Pass	Pass	Negative	0
1347	Equinor Pond PW165, Equinor Nr Cawston	Pass	Pass	Pass	Negative	0
1348	P138a, Equinor	Pass	Pass	Pass	Negative	0
1351	P138, Equinor	Pass	Pass	Pass	Negative	0
1364	Equinor P127, Alston Honingham	Pass	Pass	Pass	Positive	2
1366	Equinor, Alston Honingham	Pass	Pass	Pass	Negative	0
1368	Equinor, Alston, Honingham	Pass	Pass	Pass	Negative	0
2867	PN052, Equinor	Pass	Pass	Pass	Negative	0
2868	PN061, Equinor	Pass	Pass	Pass	Negative	0

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

Reported by: Sarah Evans **Approved by:** Chris Troth

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

SureScreen Scientifics

Folio No: E7687
Report No: 1
Purchase Order: 2020/08
Client: WILD FRONTIER ECOLOGY
Contact: Will Riddett

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: 10/06/2020
Date Reported: 17/06/2020
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
1365	Equinor PN001, Swardeston Peddars Pies		Pass	Pass	Pass	Negative	0
2861	Equinor P013, Srokowski Swardeston		Pass	Pass	Pass	Negative	0
2866	Equinor PN067, Honingham		Pass	Pass	Pass	Negative	0
2870	Equinor PN068, Honingham		Pass	Pass	Pass	Negative	0
2878	Equinor PN063, Honingham		Pass	Pass	Pass	Negative	0

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

SureScreen Scientifics

2879	Equinor PN064, Honingham Thorpe		Pass	Pass	Pass	Negative	0
2880	Equinor PN057, Honingham Fishing Lake		Pass	Pass	Pass	Negative	0
2881	Equinor P154, Mutimer Swannington		Pass	Pass	Pass	Negative	0
2882	Equinor PW170, Little Barningham		Pass	Pass	Pass	Negative	0

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

Reported by: Chris Troth **Approved by:** Sarah Evans

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

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

SureScreen Scientifics

Folio No: E7782
Report No: 1
Purchase Order: 2020/08
Client: WILD FRONTIER ECOLOGY
Contact: Will Riddett

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: 12/06/2020
Date Reported: 19/06/2020
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
1350	Equinor P042, Day Ketteringham		Pass	Pass	Pass	Negative	0
2839	Equinor P033, Day Ketteringham		Pass	Pass	Pass	Negative	0
2840	Equinor P032, Day Ketteringham		Pass	Pass	Pass	Positive	2
2841	Equinor P035, Day, Heathersett		Pass	Pass	Pass	Negative	0
2844	Equinor P037, NR Heathersett		Pass	Pass	Pass	Negative	0
2845	Equinor P038,		Pass	Pass	Pass	Negative	0

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

SureScreen Scientifics

	Day Ketteringham					
2856	Equinor P047, NR Hethersett	Pass	Pass	Pass	Negative	0
2872	Equinor P045, Day Ketteringham	Pass	Pass	Pass	Negative	0
2873	Equinor P041, NR Hethersett	Pass	Pass	Pass	Negative	0
2874	Equinor P043, Day Ketteringham	Pass	Pass	Pass	Negative	0

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

Reported by: Sarah Evans **Approved by:** Chris Troth

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

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

SureScreen Scientifics

Folio No: E7823
Report No: 1
Purchase Order: 2020/08
Client: WILD FRONTIER ECOLOGY
Contact: Will Riddett, Katrina Salmon

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: 16/06/2020
Date Reported: 25/06/2020
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
2850	PN012, Wong Farm		Pass	Pass	Pass	Negative	0
2860	PN054, Equinor		Pass	Pass	Pass	Negative	0
2862	Equinor P016, Near Swarestone, Old Rectory		Pass	Pass	Pass	Negative	0
2875	Equinor PX, Poachers Rest, Colston		Pass	Pass	Pass	Negative	0
3529	Equinor PN07, Wong Farm		Pass	Pass	Pass	Positive	3
3530	Equinor PN011, Wong Farm		Pass	Pass	Pass	Positive	4

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

SureScreen Scientifics

3531	PN010, Wong Farm		Pass	Pass	Pass	Negative	0
3534	Equinor PN019, Wong Farm		Pass	Pass	Pass	Negative	0
3537	Equinor PN015, Wong Farm		Pass	Pass	Pass	Negative	0
3538	Equinor PN022, Whiterail Farm		Pass	Pass	Pass	Positive	2
3539	Equinor PN020, Whiterail Farm		Pass	Pass	Pass	Negative	0
3540	Equinor PN013, Wong Farm		Pass	Pass	Pass	Negative	0
3541	Equinor PX10, Whiterail Farm		Pass	Pass	Pass	Negative	0
3542	PN06, Wong Farm		Pass	Pass	Pass	Negative	0
3543	Equinor PX9, Wong Farm		Pass	Pass	Pass	Negative	0
3544	PN016, Wong Farm		Pass	Pass	Pass	Negative	0
3562	PN084, Equinor - Easton Estate		Pass	Pass	Pass	Negative	0
3563	Equinor PN132, Equinor - Easton Estate		Pass	Pass	Pass	Negative	0
3565	PN080, Equinor - Easton Estate		Pass	Pass	Pass	Negative	0
3567	PN079, Equinor - Easton Estate		Pass	Pass	Pass	Negative	0

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

SureScreen Scientifics

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

Reported by: Chris Troth **Approved by:** Sarah Evans

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

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

SureScreen Scientifics

Folio No: E7915
Report No: 1
Purchase Order: 2020/08
Client: WILD FRONTIER ECOLOGY
Contact: Will Riddett, Katrina Salmon

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: 23/06/2020
Date Reported: 29/06/2020
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
1309	Equinor PS01, Mangreen PS01 Substation		Pass	Pass	Pass	Negative	0
2838	Equinor PN025, Betts High Green		Pass	Pass	Pass	Negative	0
2846	Equinor PN094, Swannington Weston		Pass	Pass	Pass	Negative	0
2849	Equinor PS04, Mangreen PS04 Susbstation		Pass	Pass	Pass	Negative	0
2863	Equinor P028, Day		Pass	Pass	Pass	Negative	0

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

SureScreen Scientifics

	Ketteringham						
2864	Equinor P010, Mangreen, Hickling Lane		Pass	Pass	Pass	Negative	0
2869	Equinor P060, Hethersett Richardson		Pass	Pass	Pass	Negative	0
2871	Equinor PS03, Mangreen PS03 Substation		Pass	Pass	Pass	Negative	0
2876	P029, Equinor		Pass	Pass	Pass	Negative	0
3527	PN070, Equinor		Pass	Pass	Pass	Negative	0
3528	PN048, Equinor		Pass	Pass	Pass	Negative	0
3532	Equinor PN092, The Lodge, Morton on the Hill		Pass	Pass	Pass	Negative	0
3536	Equinor PN096, Oulton Hall		Pass	Pass	Pass	Negative	0
3545	PW155, Equinor		Pass	Pass	Pass	Negative	0
3564	PN097, Equinor		Pass	Pass	Pass	Negative	0
3568	PN081, Equinor - Easton Estate		Pass	Pass	Pass	Negative	0

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

Reported by: Chris Troth **Approved by:** Sarah Evans

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

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

SureScreen Scientifics

Folio No: E8119
Report No: 1
Purchase Order: 2020/08
Client: WILD FRONTIER ECOLOGY
Contact: Will Riddett

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: 29/06/2020
Date Reported: 07/07/2020
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
1304	EQUINOR PS10		Pass	Pass	Pass	Negative	0
1305	P122		Pass	Pass	Pass	Negative	0
1306	PNO26		Pass	Pass	Pass	Negative	0
1307	PNO71		Pass	Pass	Pass	Negative	0
1311	EQUINOR PNO34		Pass	Pass	Pass	Negative	0
1313	PN121		Pass	Pass	Pass	Negative	0
1315	PNO24		Pass	Pass	Pass	Positive	1
1316	PN120		Pass	Pass	Pass	Positive	1
1317	EQUINOR PW156		Pass	Pass	Pass	Negative	0
1318	EQUINOR		Pass	Pass	Pass	Negative	0

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

SureScreen Scientifics

	PW158						
1319	PN072		Pass	Pass	Pass	Negative	0
1320	PN023		Pass	Pass	Pass	Positive	2
1322	EQUINOR PN101		Pass	Pass	Pass	Negative	0
1323	PN029		Pass	Pass	Pass	Negative	0
1325	P119		Pass	Pass	Pass	Negative	0
1327	PN098		Pass	Pass	Pass	Negative	0
2847	PN119		Pass	Pass	Pass	Negative	0
2848	EQUINOR PN119		Pass	Pass	Pass	Negative	0

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

Reported by: Sarah Evans **Approved by:** Chris Troth

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

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

SureScreen Scientifics

Folio No: E8453
Report No: 1
Purchase Order: 2020/08
Client: WILD FRONTIER ECOLOGY
Contact: Will Riddett, Alex Lowe

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: 08/07/2020
Date Reported: 22/07/2020
Matters Affecting Results: None

Lab Sample No.	Site Name	O/S Reference	SIC	DC	IC	Result	Positive Replicates
3546	Equinor P040, Horner Ketteringham	-	Pass	Pass	Pass	Negative	0
3548	Equinor P022, Moores Ketteringham	-	Pass	Pass	Pass	Negative	0
3549	Equinor P025, Moores, Ketteringham	-	Pass	Pass	Pass	Negative	0
3550	Equinor P026, Moores, Ketteringham	-	Pass	Pass	Pass	Positive	5
3552	Equinor P027, Moores Ketteringham	-	Pass	Pass	Pass	Negative	0
3556	PN125,	-	Pass	Pass	Pass	Negative	0

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

SureScreen Scientifics

	Kelling Health						
3557	Equinor PN088, Weston White	-	Pass	Pass	Pass	Negative	0
3570	Equinor PW186, Thurtle Bodham	-	Pass	Pass	Pass	Positive	12
3571	Equinor PN104, Brooks Saxthorpe	-	Pass	Pass	Pass	Negative	0
3579	Equinor PN053, Colton Curtis	-	Pass	Pass	Pass	Negative	0
3580	Equinor P153, Wensum Dacre	-	Pass	Pass	Pass	Negative	0
3582	Equinor PX11, Scales Colton	-	Pass	Pass	Pass	Negative	0
3583	Equinor PN043, Scales Colton	-	Pass	Pass	Pass	Negative	0
3585	PN003, Horner, Ketteringham	-	Pass	Pass	Pass	Negative	0
3586	PN047 Equinor, Scales, Colton	-	Pass	Pass	Pass	Negative	0
3587	Equinor P024, Moores, Ketteringham	-	Pass	Pass	Pass	Positive	1
3588	Equinor P039, Ketteringham, Horner	-	Pass	Pass	Pass	Negative	0
3589	Equinor P023, Moores Ketteringham	-	Pass	Pass	Pass	Negative	0
3590	Equinor P030, Moores Ketteringham	-	Pass	Pass	Pass	Negative	0

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

SureScreen Scientifics

3593	Equinor PN046, Scales Colton	-	Pass	Pass	Pass	Positive	1
5312	P505, Equinor Substation	-	Pass	Pass	Pass	Negative	0
5313	P012, Equinor Substation	-	Pass	Pass	Pass	Negative	0
5314	P510, Equinor Substation	-	Pass	Pass	Pass	Negative	0
5315	P010, Equinor	-	Pass	Pass	Pass	Negative	0
5316	PN126, Kelling Health	-	Pass	Pass	Pass	Negative	0
5317	P509, Equinor Substation	-	Pass	Pass	Pass	Negative	0
5319	P005, Equinor Substation	-	Pass	Pass	Pass	Negative	0
5322	P002, Equinor Substation	-	Pass	Pass	Pass	Negative	0
5323	P007, Equinor Substation	-	Pass	Pass	Pass	Negative	0
5324	P001, Equinor Substation	-	Pass	Pass	Pass	Negative	0
5326	P006, Equinor Substation	-	Pass	Pass	Pass	Negative	0
5329	PN127, Kelling Health	-	Pass	Pass	Pass	Negative	0

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

Reported by: Chris Troth **Approved by:** Sarah Evans

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-

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

Annex 2: UCL Pond Restoration Research Group Information

Threats to pond networks associated with the Equnior cable – Information provided by Carl Sayer and the Norfolk Ponds Project

This letter provides some information to inform on the proposed Equinor cable corridor for North Norfolk in terms of its intersection with farmland ponds. The particular focus of this is letter is Great Crested Newt (GCN) and initially data is provided for the Bodham-Baconsthorpe (**Fig. 1, Fig. 2**) and Heydon (**Fig. 3**) areas, both of which are in or close to the proposed cable corridor and which have also been the subject of major landscape-scale pond restoration projects of the Norfolk Ponds Project (NPP). Note that further information is held on many other species groups: plants, invertebrates, other amphibians, birds, bats, fish (including eels) for many of the ponds that can be made available.

Working towards the goals of the NPP a major activity of the UCL Pond Restoration Research Group (PRRG) has been the restoration of farmland ponds with the aim of increasing aquatic and terrestrial biodiversity and aquatic connectivity. A key issue tackled is pond terrestrialisation as, since the 1960s/70s period, with the cessation of traditional pond management practices, Norfolk farmland ponds have become highly overgrown with negative impacts for species due to habitat uniformity (almost all ponds are overgrown presenting a lack of aquatic plant habitat) and because overgrown ponds afford very harsh (anoxic) conditions to aquatic species (Sayer *et al.,* 2012; 2013; Sayer, 2014). Thus the project has re-instated management and undertaken major scrub and sediment removal for networks of ponds. The project started its work on the Bodham area, with Sayer's Black Pit (**Fig. 2**) the first restored pond. The results of the project have been startling which substantial measured gains for aquatic plants, invertebrates, amphibians (including the protected Great Crested Newt), farmland birds and pollinator communities across the ponds (Sayer *et al.,* 2012; 2013; Sayer, 2014; Lewis-Phillips *et al.* 2019, 2020 and see **Fig. 1**).

At Bodham-Baconsthorpe the UCL PRRG has been working on several ponds (with 11 ponds restored since 2011), some 19 of which have recorded Great Crested Newt presence (**Table 1**) and highly diverse communities of plants (e.g. **Fig. 1**), invertebrates, dragonflies, pollinators and farmland birds. Note we have data for all of these groups for several of the ponds. Important pond plants include *Nitella opaca, Nitella flexilis and Tolypella glomerata* and the rare (Norfolk BAP species) crucian carp (*Carassius carassius*) is present in four ponds in this area.

At the Heydon pond landscape (**Fig. 3**) Great Crested Newt has been recorded at some 9 ponds (**Table 2**). Some rare plants have been recorded from these ponds, including *Najas marina* (PYES1), *Hottonia palustris* (HEY97), *Oenanthe aquatica* (several ponds) and *Tolypella glomerata* (several ponds), with one restored pond (COLG3) supporting a remarkable 6 charophytes species. In addition the crucian carp has been recorded at 3 of the Heydon ponds. Thus there are very valuable restored pond landscapes adjacent to and within the cable route that will be compromised.

Given our major interests in Norfolk farmland ponds and in the network of hedges, woodland and meadows that form important corridors and connections between ponds that will be damaged and compromised by the cable works and in particular given the proximity of the Equinor cable route to our two major pond restoration areas at Bodham and Heydon we are obviously very concerned regarding potential impacts.

Yours sincerely

The UCL Pond Restoration Research Group
c/o Dr. Carl Sayer, Department of Geography, University College London, Gower Street London, WC1E 6BT, Tel: 07766717245, e-mail: c.sayer@ucl.ac.uk

Pond name	Pond code	NGR	Date restored by NPP	Great Crested Newt known to be (breeding)
Hart Lane Pond	HART	TG 12952 39304	Not restored	Yes
Beckett's Farm Pond	BECK	TG 1110 3765	Restored Sept. 2014	Yes
Shooting Close Pond	SHOOT	TG 1135 3780	Restored Sept. 2014	Yes
Sayer's Black Pit	SABA	TG 1265 3960	Restored Sept. 2011	Yes
Sayer's New Pond	SAYNE	TG 12561 39892	Restored Sept. 2011	Yes
Bodham Mystery Pit	MYST	TG 1260 3945	Restored Sept. 2011	Yes
Mystery Pit Friend	MYSTF	TG 12430 39444	Not restored	Yes
Church Farm Pond 1	CHFA1	TG 11704 38768	Not restored	No
Church Farm Pond 2	CHFA2	TG 11886 38818	Restored Sept. 2017	Yes
Church Farm Pond 3	CHFA3	TG 11735 38720	Restored Sept. 2017	Yes
Church Farm Pond 4	CHFA4	TG 11874 38908	Not restored	Yes
Baconsthorpe Wood S. Pond	BAWO2	TG 12846 38343	Restored Nov. 2017	Yes
Breck Farm Pond	BRECK	TG 12591 37622	Not restored	Yes
Baconsthorpe Wood N. Pond	BAWO1	TG 12759 38591	Not restored	Yes
New Road Pond	NROAD	TG 12882 37684	Restored Nov. 2017	No
Skylark Pond	SKYLA	TG 11060 38332	Restored Sept. 2017	Yes
Wrong Close Pond	WRONG	TG 1160 3750	Not restored	Yes
Rail Pit	RAIL	TG 1235 3890	Not restored	Yes
Pond Farm Pond 1	POFA1	TG 1315 3860	Not restored	Yes
Pond Farm Pond 2	POFA2	TG 1315 3855	Not restored	Yes
Pond Farm Pond 3	POFA3	TG 1330 3865	Not restored	Yes
Pond Farm Pond 4	POFA4	TG 1325 3815	Restored Sept. 2010	Unknown

Table 1. Key Norfolk Pond Project study ponds within or close to the proposed Equinor cable corridor in the Bodham-Baconsthorpe area detailing known sites for Great Crested Newt (GCN) breeding as recorded by Carl Sayer & Ewan Shilland of the Pond Restoration Research Group at UCL. All sites with a Yes for breeding had GCN eggs.

Pond name	Pond code	NGR	Date restored by NPP	Great Crested Newt known to be (breeding)
Colgreen Field Pond 1	COLG1	TG 10448 26942	Not restored	No
Colgreen Field Pond 2	COLG2	TG 10426 26632	Restored Sept. 2015	Yes
Colgreen Field Pond 3	COLG3	TG 10553 26790	Restored Sept. 2015	No
Colgreen Field Pond 4	COLG4	TG 10462 26806	Restored Sept. 2015	Yes
Heydon Pond 102	HEY102	TG 10706 27106	Restored Sept. 2018	No
Heydon Pond 103	HEY103	TG 10650 27056	Not restored	No
Heydon Pond 97	HEY97	TG 10720 26945	Restored Sept. 2016	No
Heydon Pond 96	HEY96	TG 10786 26862	Restored Sept. 2016	No
Bonfire Field Pond	BONF	TG 1095 126778	Restored Sept. 2015	Yes
Heydon Pond 94	HEY94	TG 11057 26722	Not restored	Yes
Heydon Pond 93	HEY93	TG11363 28269	Restored Sept. 2016	Yes
Bullock Shed Pond 1	BULLS1	TG 11267 28319	Not restored	No
Bullock Shed Pond 2	BULLS2	TG 11108 28326	Not restored	No
Heydon Pond 90	HEY90	TG 11133 28459	Not restored	Yes
Heydon Pond 89	HEY89	TG 11363 28268	Not restored	No
Holly Grove Pond	HOLLY	TG 10707 27940	Not restored	No
Dairy Farm Pond 1	DAIRY1	TG 10501 27652	Not restored	Yes
Dairy Farm Pond 2	DAIRY2	TG 10534 27732	Restored Sept. 2018	Yes
Dairy Farm Pond 3	DAIRY3	TG 10598 27758	Not restored	No
Dairy Farm Pond 4	DAIRY4	TG 10627 27783	Not restored	Yes
Cinders Hill Pond	CIND	TG 10908 28756	Restored Sept. 2012 + managed Feb. 2015	No
Pyes Pit 1	PYES1	TG 1330 2555	Restored Feb. 2015	No
Pyes Pit 2	PYES2	TG 1340 2535	Restored Feb. 2015	Yes

Table 2. Key Norfolk Pond Project study ponds close to the proposed Equinor cable corridor in the Bodham-Baconsthorpe area detailing known sites for Great Crested Newt (GCN) breeding as recorded by Carl Sayer & Ewan Shilland of the Pond Restoration Research Group at UCL. All sites with a Yes for breeding had GCN eggs.

Figure 1. Shooting Close Pond (TG 1135 3780), a small Great Crested Newt supporting farmland pond close to the Orsted cable corridor in the Bodham-Baconsthorpe area before (a), during (b) and two years after restoration (c) by scrub and sediment removal in September 2014.

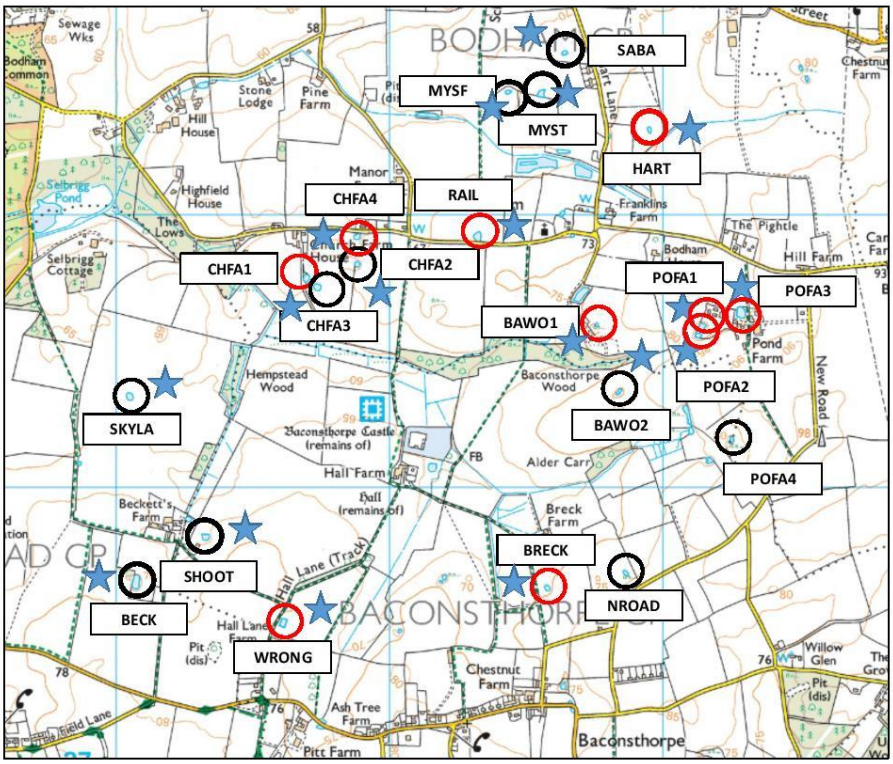


Figure 2. Ponds in the Bodham-Baconsthorpe Norfolk Ponds Project pond landscape as discussed in the text and detailed in Table 1. Ponds circled black have been restored by the NPP and ponds with a blue asterisk next to them are known to support Great Crested Newt as recorded by Carl Sayer & Ewan Shilland of the Pond Restoration Research Group at UCL.

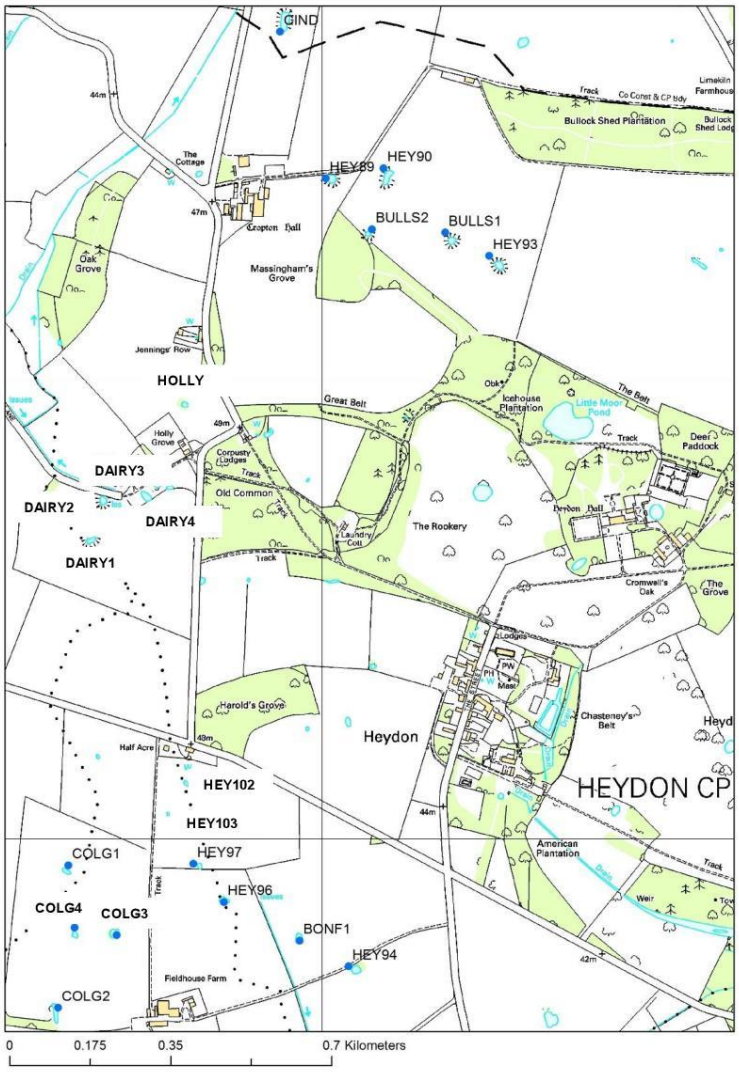


Figure 3. Ponds in the Heydon area Norfolk Ponds Project pond landscape. Note that all of CIND, PYES1, PYES2, COLG1, COLG3, COLG4, HEY97, HEY96, BONF1 and HEY93 have all been restored since 2012. Great Crested Newt breeding is known for ponds BONF, HEY94, COLG3, COLG2, HEY93, DAIRY1, DAIRY4 and PYES2 and we expect the species to spread further throughout the pond network in the coming years. Note the NPP is monitoring the ponds closely in this respect each year.

References

Lewis-Phillips, J., Brooks, S., Sayer, C., McCrea, R., Siriwardena, G., Axmacher, J.C. (2019). Pond management enhances the local abundance and species richness of farmland bird communities. *Agriculture, Ecosystems and Environment*, 273, 130-140.

Lewis-Phillips, J., Brooks, S.J., Sayer, C.D., Patmore, I.R., Hilton, G., Harrison, A., ...Axmacher, J.C. (2020). Ponds as insect chimneys: restoring overgrown farmland ponds benefits birds through elevated productivity of emerging aquatic insects. *Biological Conservation*, 108253,

Sayer, C.D., Copp, G.H., Emson, D., Godard, M.J., Zieba, G., Wesley, K.J. (2011). Towards the conservation of crucian carp *Carassius carassius*: understanding the extent and causes of decline within part of its native English range. *Journal of Fish Biology*, 79 (6), 1608-1624. doi:10.1111/j.1095-8649.2011.03059.x

Sayer, C.D., Andrews, K., Shilland, E., Edmonds, N., Edmonds-Brown, R., Patmore, I., Emson, D. & Axmacher, J.A. (2012). The role of pond management for biodiversity conservation in an agricultural landscape. *Aquatic Conservation: Marine and Freshwater Ecosystems*, 22 (5), 626-638. doi:10.1002/aqc.2254

Sayer, C.D., Shilland, E.M., Greaves, H., Dawson, B., Patmore, I., Emson, D., Alderton, E., Robinson, P., Andrews, K., Axmacher, J.A., Wiik, E. (2013). Managing Britain's ponds - conservation lessons from a Norfolk farm. *British Wildlife* 25(1), 21-28.

Sayer, C.D. (2014). Conservation of aquatic landscapes: ponds, lakes, and rivers as integrated systems. *WIRE's: Water*, 1 (6), 573-585. doi:10.1002/wat2.1045