

A303 Amesbury to Berwick Down

**Applicant's provision of technical reports supporting the
Environmental Information Review**

Great Crested Newt Survey Report (2021)

Document reference: Redetermination 2.7

Planning Act 2008

The Infrastructure Planning (Examination Procedure) Rules 2010

February 2022



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Executive Summary

Great crested newt surveys were undertaken in February-June 2021 to identify the presence/likely absence of great crested newt within 500 metres of the A303 Stonehenge (Amesbury to Berwick Down) Scheme. This report presents the methodology and results of the surveys.

A total of 18 waterbodies were assessed for their suitability for great crested newt. The majority of waterbodies were dry at the time of survey and therefore classified as not suitable for great crested newt. Four waterbodies were subject to a habitat suitability index assessment^{1,2}.

Habitat suitability index assessment concluded waterbodies 1, 3, 4 and 18 were suitable for great crested newt. Waterbody 4 was not subject to any further surveys for great crested newt due to the presence of high densities of stickleback fish likely to make the waterbody unsuitable for great crested newt. Water samples from waterbody 3 were sent for eDNA analysis, the result of which was negative, suggesting likely absence of great crested newt. Waterbody 18 was also not subject to any further surveys as the waterbody was dry for the remaining season after HSI assessment. It is therefore only likely to hold water during periods of heavy rainfall.

Waterbody 1 was subject to further surveys to identify the presence/likely absence of great crested newt and classify the population size. Great crested newt were found to be present during all surveys at waterbody 1 and the peak count identified 162 individuals. Population class estimates classify this as a 'Large' population. This waterbody had been recorded in the previous survey as having a 'Small' population.

This survey assessed waterbodies within 500 metres of the A303 Stonehenge (Amesbury to Berwick Down) Scheme and classified the population size of great crested newt in waterbody 1. It confirms that, except for waterbody 1, there are no other waterbodies for which mitigation will be needed for great crested newts during the construction of the scheme.

¹ Amphibian and Reptile Groups of the United Kingdom, ARG UK Advice Note 5, Great Crested Newt Habitat Suitability Index, May 2010.

² Oldham R.S., Keeble, J., Swan, M.J.S. & Jeffcote, M. (2000). Evaluating the suitability of habitat for the Great Crested Newt (*Triturus cristatus*) Herpetological Journal 10(4), 143-155.

1 Introduction

- 1.1.1 This report presents the results of the 2021 great crested newt surveys for the A303 Amesbury to Berwick Down Scheme (hereafter referred to as the 'Scheme'). Previous surveys were conducted in 2017-18 and identified 13 waterbodies within 575 metres of three potential route options with great crested newts present in one of these waterbodies³. The 2017-2018 surveys were used to inform the environmental assessment of the Scheme, which was included in the Environmental Statement which accompanied the DCO application in October 2018⁴. The 2021 survey is part of a programme of pre-construction surveys intended to keep the baseline data up to date, so it can be used to inform any application required for a European Protected Species licence for great crested newt during the construction period.
- 1.1.2 No ponds will be lost during the construction of the Scheme, but the Scheme will involve some temporary and permanent loss of terrestrial habitat within 500m of one pond, waterbody 1, which in 2017-2018 was recorded as having a breeding population of great crested newt (the 2021 surveys confirmed that this population is still present).
- 1.1.3 This report outlines the methodology and results for the great crested newt survey for the Scheme, carried out in 2021, which was undertaken to determine the presence or likely absence of great crested newt within 500 metres of the Scheme (see Figure 1) and the current status of the population in waterbody 1.
- 1.1.4 Photographs of the waterbodies are provided in Appendix A.

1.2 Legislation

- 1.2.1 Great crested newt are a European Protected Species (EPS), protected under the Conservation of Habitats and Species (amendment) (EU exit) Regulations 2019, known as the Habitats Regulations.
- 1.2.2 Under the Habitats Regulations, it is an offence to:
- deliberately capture, injure or kill any wild animal of an EPS;
 - deliberately disturb wild animals of any such species;
 - deliberately take or destroy the eggs of such an animal; or,
 - damage or destroy a breeding site or resting place of such an animal.
- 1.2.3 Great crested newt receives further protection through inclusion on Schedule 5 of the Wildlife and Countryside Act (WCA) 1981 (as amended) and the Countryside and Rights of Way (CRoW) Act 2000. Under these Acts it is an offence to:
- intentionally kill, injure or take any protected species;

³ Arup Atkins Joint Venture (2017) A303 Amesbury to Berwick Down Amphibian Survey Report 2016-2017

⁴ Highways England (2018) A303 Amesbury to Berwick Down TR010025, 6.3 Environmental Statement

- intentionally or recklessly damage, destroy or obstruct access to any structure or place which a protected species uses for shelter or protection; and,
- intentionally or recklessly disturb any protected species while it is occupying a structure or place which is uses for shelter or protection.

1.2.4 Actions which are prohibited by legislation can be made lawful on the approval and granting of a licence from Natural England (NE), subject to conditions.

1.2.5 Great crested newt as well as common toad (*Bufo bufo*) are listed as a Section 42 species of the Natural Environment and Rural Communities (NERC) Act 2006. The list of species on Section 42 is used to guide decision makers, including local and regional authorities, in implementing their duty under Section 40 of the Act, to have regard to the conservation of biodiversity in England.

2 Methodology

2.1 Survey Area

2.1.1 The survey area included all ponds within 500 metres of the Scheme, these were identified using aerial photography, OS maps, extended phase 1 habitat survey data⁵ and previous survey data⁶. The locations of each waterbody within the survey area are shown on Figure 1 and a description of each waterbody within the survey area is provided in Table 1.

Table 1: Description of waterbodies within the survey area

Waterbody number	Waterbody Description
1	Medium-sized waterbody located north of the A303 within fields and next to the River Till. The waterbody is surrounded by pasture habitat and arable fields to the south and east, with woodland and scrub habitat noted to the north.
2	This waterbody was surveyed in 2017 and scoped out, as it was river fed, as such, unlikely to be suitable for great crested newts. In addition, the waterbody is considered to be isolated from the main working area by the existing A303 and River Till.
3	Small waterbody located to the south of the A303, dries to a small area (1x1m hole) where a trench had been dug for water monitoring purposes was still wet.
4	Small waterbody/drain located north of the A303 and just west of the River Avon.
5	Ratfyn farm ditch adjacent to Avon with low flow, located directly adjacent to the A303 to the north and south (continues under road). The ditch is a drainage ditch and thus only contains water immediately after rainfall.
6	Small waterbody located north of the A303 and between the River Avon and the Sewage Works south west of Bulford.
7	Pond with ornamental waterfowl.
8	Large fishing pond located to the north of the A303 on the edge of the 500m buffer.
9	Large fishing pond located to the north of the A303.
10	Drainage ditch located along road and thus only contains water immediately after rainfall.
11	Drainage ditch located along road and thus only contains water immediately after rainfall.
12	Drainage ditch located along road and thus only contains water immediately after rainfall.
13	Drainage ditch located along road and thus only contains water immediately after rainfall.
14	Drainage ditch located along road and thus only contains water immediately after rainfall.
15	Drainage ditch located along road and thus only contains water immediately after rainfall.
16	Drainage ditch located along road and thus only contains water immediately after rainfall.
17	Drainage ditch located along road and thus only contains water immediately after rainfall.
18	Wet ditch running along the arable field. It is likely to only hold water during high rainfall.

⁵ Highways England (2018) A303 Amesbury to Berwick Down TR010025, 6.3 Environmental Statement Appendices, Appendix 8.3B Update surveys technical note, October 2018

⁶ Arup Atkins Joint Venture (2017) A303 Amesbury to Berwick Down Amphibian Survey Report 2016-2017

Waterbody number	Waterbody Description
Not Applicable	Previously described as a small waterbody located on the edge of Parsonage Down National Nature Reserve north of the A303, which almost certainly dries annually.

2.2 Habitat Suitability Index

- 2.2.1 Waterbodies 1, 3, 4 and 18 were assessed for their suitability to support great crested newt using the standard Habitat Suitability Index (HSI)^{7,8} methodology on 22 February 2021 and 19 March 2021.
- 2.2.2 As detailed in the limitations section all ponds that were dry at the time of survey were not assessed for their suitability to support great crested newt. The remaining ponds were not surveyed due to lack of access. As described in the limitations section, the lack of access to these waterbodies were not deemed a significant limitation to the assessment (see Appendix B for a summary of why each waterbody was scoped out from survey).
- 2.2.3 HSI has been designed to evaluate habitat quality to assess whether a waterbody is likely to provide suitable habitat for great crested newt as breeding ponds. The HSI is a numerical index, which ranges from 0 to 1. It is calculated using ten key habitat criteria and is based on the assumption that habitat quality determines great crested newt presence/absence. Using this standard approach, ponds with higher scores are considered more likely to support great crested newts compared to those with low scores (see Table 2 below). However, the system is not sufficiently precise to conclude that any particular pond with a high score will support great crested newt, or that any pond with a low score will not do so.

Table 2. Predicted presence of great crested newts based upon HSI results

HSI score	Pond suitability	Predicted occupancy
<0.5	Poor	0.03
0.5-0.59	Below average	0.20
0.6-0.69	Average	0.55
0.7-0.79	Good	0.79
>0.8	Excellent	0.93

2.3 eDNA Sampling

- 2.3.1 Waterbody 3 was subject to eDNA sampling and analysis on 17 May 2021, an eDNA survey on all other waterbodies within the survey area was scoped out due to their lack of suitability for great crested newt (see Appendix B for a summary of why each waterbody was scoped out from survey) or for their

⁷ Amphibian and Reptile Groups of the United Kingdom, ARG UK Advice Note 5, Great Crested Newt Habitat Suitability Index, May 2010.

⁸ Oldham, R.S., Keeble, J., Swan, M.J.S. & Jeffcote, M. (2000) Evaluating the suitability of habitat for the Great Crested Newt (*Triturus cristatus*). Herpetological Journal 10(4), 143-155.

known presence (waterbody 1). This testing followed standard methodology and best practice guidelines⁹, with the eDNA samples being tested at ADAS.

2.4 Population Size Class Assessment

Population size class assessments for great crested newt were carried out at waterbody 1 in accordance with the methodology described within the Great Crested Newt Mitigation Guidelines¹⁰. Six visits of the waterbody were undertaken with at least three visits undertaken mid-April to mid-May. All surveys were undertaken in late-April to late-June 2021 due to unseasonably low temperature in March and early April. The dates of survey visits and the weather conditions at the time can be found in Table 3.

Table 3. Weather conditions for each survey visit

Date	Visit number	Survey Methods	Time		Mean Air Temperature (°C)	Approximate area of waterbody surveyed	Limitations
			Start	End			
28/04/2021	1	Torch	21:04	22:01	9	40	Overnight temperature too cold to bottle trap. Heavily vegetated with <i>Ranunculus</i> sp. and watermint
11/05/2021 - 12/05/2021	2	Torch	21:00	22:00	8	40	Heavily vegetated with <i>Ranunculus</i> sp. and watermint
		Bottle trap	20:56	08:20	8	90	Could only trap the very edge. Most newts recorded in water >1m from edge.
17/05/2021	3	Torch	21:54	22:39	6	40	Heavily vegetated with <i>Ranunculus</i> sp. and watermint
07/06/2021	4	Torch	22:25	23:30	13	20	80% vegetation cover. Pond dried since previous survey, levels dropped by 10cm.
10/06/2021	5	Torch	22:00	22:45	17	5	<i>Ranunculus</i> sp. covered 95% of the waterbody. Cows had churned the waterbody. Turbidity 3/5.
30/06/2021	6	Torch	22:20	23:00	16	5	<i>Ranunculus</i> sp. covered ~95% of the waterbody. Cows had churned the waterbody.

⁹ Biggs et al. (2014) Technical Advice Note For Field and Laboratory Sampling of Great Crested Newt eDNA in Analytical and methodological development for improved surveillance of the Great Crested Newt. Defra Project WC1067. Appendix 5. Freshwater Habitats Trust, Oxford.

¹⁰ English Nature (2001). Great Crested Newt Mitigation Guidelines. English Nature.

2.4.1 During each visit, torch surveys were undertaken of the waterbody. During visit 2 bottle trapping were also undertaken. Detailed description of each methodology is as follows:

- Torching: The banks of the waterbody were walked after dark, using a high-powered torch (1 million candle power) to search for newts and other amphibians. Animals observed were identified to species, sex and life stage where possible.
- Bottle trapping: Bottle traps were placed around the perimeter of the waterbody at approximately 2 metre intervals (where suitable habitat and health and safety considerations allowed) shortly before dusk and checked the following morning to determine whether newts were present or absent. Each trap was made from a 2-litre plastic bottle with the top cut off and inverted, to make a funnel leading to the bottle. Bamboo canes were used to anchor the traps into the waterbody, taking care to ensure that each bottle included an air bubble. Any animals caught were immediately returned to the waterbody after identification.

2.4.2 On each occasion, surveys were led by an ecologist holding a NE survey licence for great crested newt.

2.4.3 Population size class was established using the maximum adult count per pond per survey visit, either through torching or bottle trapping, as follows:

- 'Small' for maximum counts up to 10;
- 'Medium' for maximum counts between 11 and 100; and,
- 'Large' for maximum counts over 100.

2.5 Survey Limitations

2.5.1 Waterbodies 5 and 10-17 were not subject to HSI assessment as they were dry at the time of survey and as such were considered not suitable for great crested newt. Access was not possible to waterbody 2 as no access consent was given, this waterbody was therefore not subject to HSI assessment. The requirement for surveys of waterbody 2 was ruled out as it is isolated from the main works by the A303 and River Till (which is perennial south of Winterbourne Stoke, whereas it is a seasonal, winterbourne to the north). HSI assessments were also not conducted on waterbodies 6-9 due to no access consent being given. These have been scoped out from further surveys for great crested newt due to being stocked fishing ponds, containing ornamental waterfowl and/or being dry at the time of survey.

2.5.2 Bottle trapping at waterbody 1 was only carried out on 11 May 2021 as livestock were present near the waterbody and using the waterbody for drinking. As a peak adult count was recorded during torching of this waterbody, this is not considered to limit the validity of the data.

2.5.3 As with many ecological surveys, the effectiveness of these surveys is subject to a range of seasonal, environmental and behavioural factors. The absence of evidence of a particular species in a survey should not be taken

as conclusive proof that the species is not present or that it will not be present in the future.

- 2.5.4 See Appendix B for a summary of why each waterbody was scoped out from further survey.

3 Results

3.1 HSI

3.1.1 A summary of the results of the HSI assessment are presented in Table 4 and detailed results are provided in Appendix C.

Table 4: Summary results of HSI assessment

Waterbody	HSI Score	HSI Category
1	0.49	Poor
3	0.50	Below average
4	0.25	Poor
18	0.31	Poor

3.2 eDNA

3.2.1 Samples from waterbody 3 were analysed for the presence of great crested newt eDNA. The result of this was negative, suggesting the absence of great crested newt (see Appendix D for the eDNA results letter). Further surveys of waterbody 3 were therefore not conducted.

3.2.2 No water samples from waterbody 1 were tested for the presence of great crested newt eDNA. This was considered not necessary given the known population present.

3.2.3 Waterbodies 4 and 18 were not surveyed for great crested newt eDNA. Waterbody 4 was considered unsuitable for great crested newt due to the high densities of stickleback fish recorded during the HSI assessment. Waterbody 18 was not surveyed for great crested newt eDNA as it was dry at the time of survey and therefore considered to only hold water during periods of heavy rainfall.

3.3 Population Size Class Assessment

3.3.1 Waterbody 1 was subject to great crested newt population size class assessment. Great crested newt were recorded as present during all surveys, as shown in Table 5. The maximum count of great crested newt was recorded during visit 2, at which point 103 and 59 male and female great crested newts were recorded, respectively, resulting in a peak count of 162 assessed as a population class estimate of 'Large'.

Table 5. Results of population size class assessment

Date	Visit number	Survey Method	GCN Male	GCN Female	Smooth Male	Smooth Female	Unknown newt	Other amphibians
28/04/2021	1	Torching	14	8	2	2	0	-
11/05/2021	2	Torching	103	59	8	5	0	Toad x2
12/05/2021	2	Bottle trapping	1	5	1	1	0	-
17/05/2021	3	Torching	86	55	13	13	3	Toad x1
07/06/2021	4	Torching	2	4	3	1	1	-
10/06/2021	5	Torching	2	1	1	2	0	Frog x2

Date	Visit number	Survey Method	GCN Male	GCN Female	Smooth Male	Smooth Female	Unknown newt	Other amphibians
30/06/2021	6	Torching	0	2				-

3.3.2 Other amphibians, including smooth newt, common toad and common frog, were also recorded during the surveys.

4 Conclusion

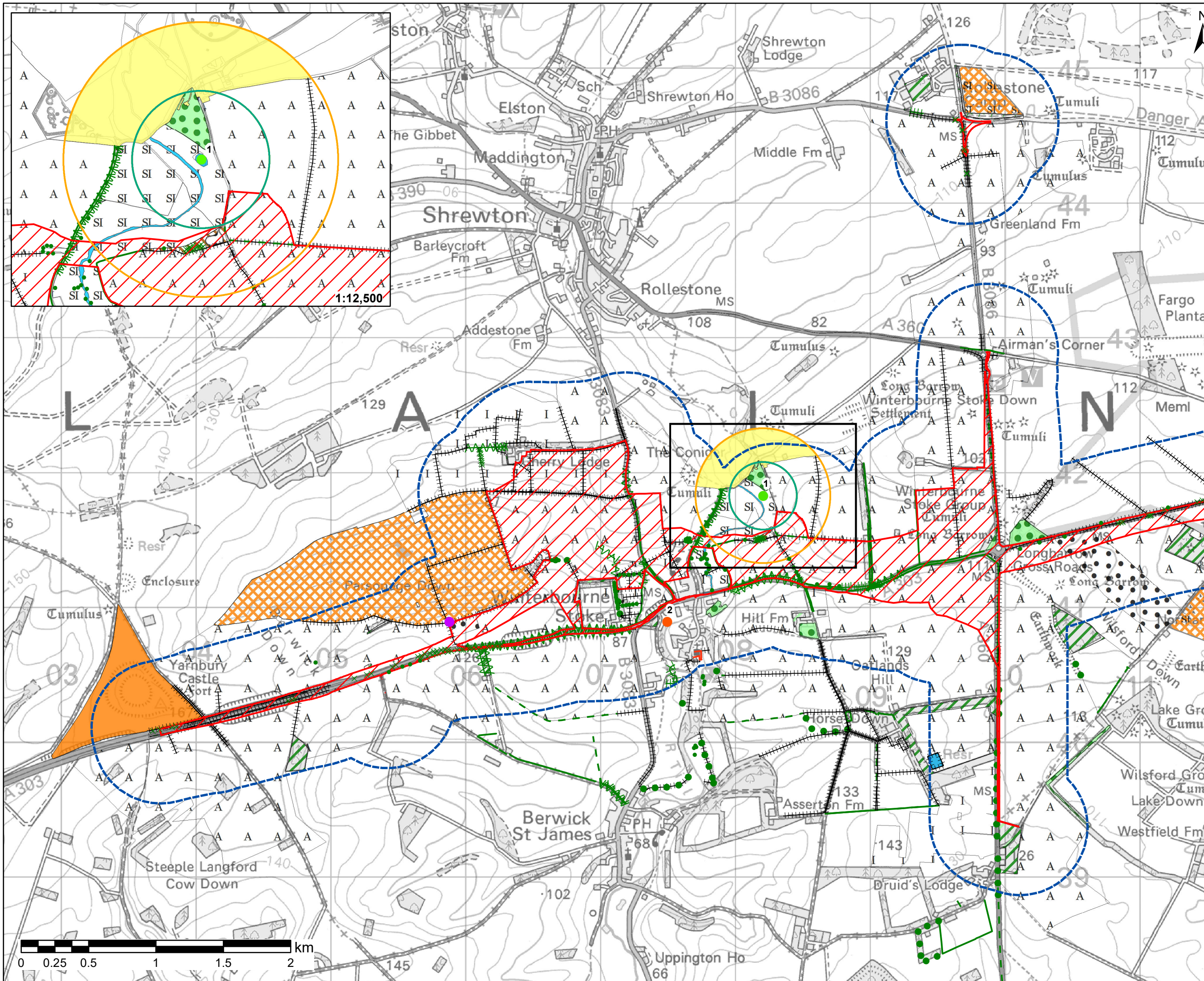
- 4.1.1 A summary of the survey results for the 2021 and 2016 surveys is provided in Appendix B. Waterbodies 2-18 were classified as not suitable for great crested newt a summary of why they were deemed not suitable is provided in Appendix B.
- 4.1.2 A population of great crested newt were confirmed to be present in waterbody 1. Population size class assessment classifies this waterbody to contain a 'Large' population of great crested newt. In the previous survey 2016¹¹ only a 'Small' population of great crested newts was recorded (peak count 10), compared to a peak count of 162 in 2021. The pond is in the valley of the River Till upstream of Winterbourne Stoke. It was dug to help provide a water supply for livestock when the river is dry¹². As well as being subject to varying climatic conditions from year to year, the pond is grazed and trampled by livestock. In addition, in wet years the River Till occasionally floods the pond and this may bring fish into the pond, although any fish will be lost if the pond dries up in late summer that year or a subsequent one. Taking these factors into account it is likely that the breeding population of great crested newts will also fluctuate in response to the conditions in both the current year and the breeding success in previous years. Any subsequent updating surveys may show population size class similar to this year, or down to Medium or Small size class. The 2021 survey confirms continuity of use of the pond by great crested newts.

¹¹ Arup Atkins Joint Venture (2017) A303 Amesbury to Berwick Down Amphibian Survey Report 2016-2017

¹² Landowner (date unknown) personal communication

5 Figures

Figure 1: Survey Map



NOTES / LEGEND

- DCO Area
- GCN Survey Area
- 250m buffer
- 500m buffer

Waterbodies survey status

- GCN present- Large population
- Not suitable for GCN – scoped out
- No longer present

Phase 1 Habitats

- Broadleaved parkland/scattered tree
- Scrub - scattered
- Broadleaved parkland/scattered trees
- Defunct hedge - native species-rich
- Defunct hedge - species-poor
- Fence
- Hedge with trees - native species-rich
- Hedge with trees - species-poor
- Intact hedge - native species-rich
- Intact hedge - species-poor
- Wall
- Bare ground
- Broadleaved woodland - plantation
- Buildings
- Calcareous grassland - semi-improved
- Calcareous grassland - unimproved
- Cultivated/disturbed land - arable
- Hardstanding
- Improved grassland
- Mixed woodland - plantation
- Mixed woodland - semi-natural
- Neutral grassland - unimproved
- Not surveyed
- Other tall herb and fern - ruderal
- Poor semi-improved grassland
- Scrub - dense/continuous
- Scrub - scattered
- Standing water
- Running water

Purpose of issue: FOR INFORMATION

Client: Highways England
Working on behalf of:

Project Title: A303 AMESBURY TO BERWICK DOWN

Drawing Title: FIGURE 1 SURVEY MAP

Designed AM	Drawn AM	Checked CC	Approved HM	Date 28/10/21
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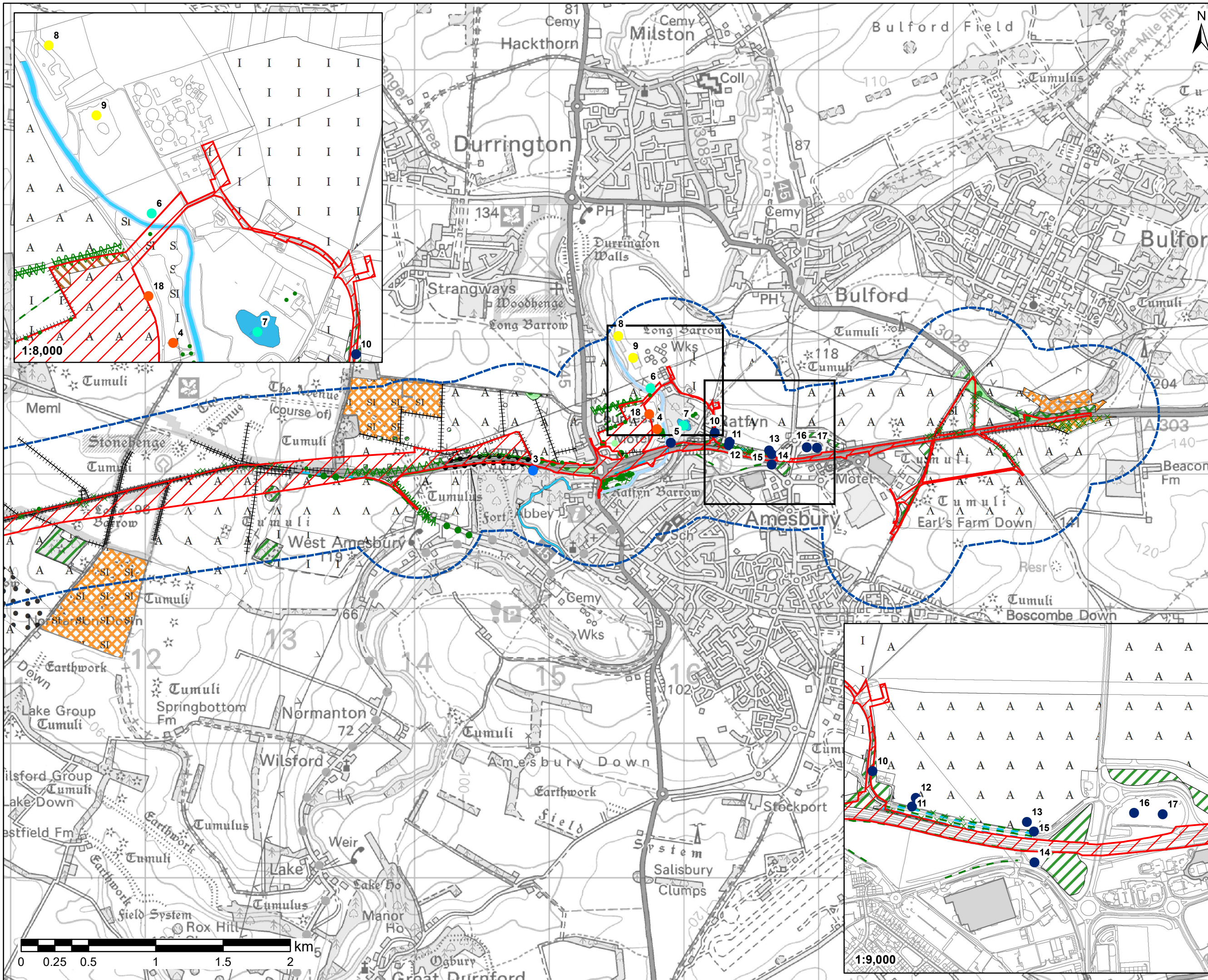
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- NOTES / LEGEND**
- DCO Area
 - GCN Survey Area
- Waterbodies survey status**
- Not suitable for GCN – scoped out
 - eDNA negative result
 - Dry - scoped out
 - No access – scoped out during 2017 survey
 - No access – scoped out fishing pond and isolated
- Phase 1 Habitats**
- Broadleaved parkland/scattered tree
 - x Scrub - scattered
 - Broadleaved parkland/scattered trees
 - - - Defunct hedge - native species-rich
 - - - Defunct hedge - species-poor
 - - - Dry ditch
 - Earth bank
 - Fence
 - - - Hedge with trees - native species-rich
 - - - Hedge with trees - species-poor
 - - - Intact hedge - native species-rich
 - - - Intact hedge - species-poor
 - - - Marginal and inundation - marginal vegetation
 - Mixed parkland/scattered trees
 - x Scrub - scattered
 - Bare ground
 - - - Broadleaved woodland - plantation
 - x Calcareous grassland - semi-improved
 - x Calcareous grassland - unimproved
 - Cultivated/disturbed land - arable
 - Hardstanding
 - - - Improved grassland
 - - - Mixed woodland - plantation
 - - - Mixed woodland - semi-natural
 - - - Other tall herb and fern - ruderal
 - - - Poor semi-improved grassland
 - x Scrub - dense/continuous
 - x Scrub - scattered
 - Standing water
 - - - Running water

Purpose of Issue: FOR INFORMATION

Client: Highways England
 Working on behalf of:

Project Title: A303 AMESBURY TO BERWICK DOWN

Drawing Title: FIGURE 1 SURVEY MAP

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Scheme Wide	SCHEME WIDE	DR	GI	0822			
Location		Type	Role	Number			

Appendices

Appendix A: Photographs



Photograph 1: Waterbody 1



Photograph 2: Waterbody 1



Photograph 3: Waterbody 3



Photograph 4: Waterbody 18



Photograph 5: Waterbody 18

Appendix B: Waterbody GCN Survey Summary

Waterbody number	HSI	eDNA	Population Estimate Surveys	Summary Notes	Result
1	Last undertaken in 2021 with a score of 0.49, poor.	Last undertaken in 2016 with a result of positive	Surveys in 2021 identified a Large population, surveys undertaken in 2017 identified a Small population.	Medium-sized waterbody located north of the A303 within fields and next to the River Till. The waterbody is surrounded by pasture habitat and arable fields to the south and east, with woodland and scrub habitat noted to the north.	GCN present- Large population
2	Last undertaken in 2016 with a score of 0.68, average	Last undertaken in 2016 with a result of negative	Not Applicable	This waterbody was surveyed in 2017 and scoped out, as it was river fed, as such, unlikely to be suitable for great crested newts. In addition, the waterbody is considered to be isolated from the main working area by the existing A303 and River Till. Any works on the A303 itself would be minimal, associated with resurfacing / painting and not trigger the requirement of a licence.- Scoped out for further survey in 2021	Not suitable for GCN – scoped out
3	Last undertaken in 2021 with a score of 0.50, below average	Last undertaken in 2021 with a result of negative	Not Applicable	Small waterbody located to the south of the A303. At the time of the 2021 HSI the waterbody was present. - Scoped in for further surveys. When the waterbody was surveyed in 2021, it was mainly dry, with only a small area (1x1m hole) where a trench had been dug for water monitoring purposes was still wet. For completeness an eDNA survey was completed and the result was negative, as such, no further surveys undertaken.	eDNA negative result
4	Last undertaken in 2021 with a score of 0.25	Last undertaken in 2016 with a result of negative	Not Applicable	Small waterbody/drain located north of the A303 and just west of the River Avon. The waterbody was scoped in for further survey. However, when undertaking the first survey, the waterbody was scoped out from further surveys due to the presence of 1000's of stickleback fish making it not suitable for GCN. At high water levels the waterbody is hydrologically connected to the River Avon.	Not suitable for GCN – scoped out
5	Last undertaken in 2021 but waterbody was dry so assessment not undertaken	Not Applicable	Surveyed undertaken in 2017 confirmed GCN likely absent. Not Applicable in 2021 as dry.	Ratfyn farm ditch adjacent to Avon with low flow, located directly adjacent to the A303 to the north and south (continues under road). The ditch is a drainage ditch and thus only contains water immediately after rainfall. The ditch was observed to be dry during the 2021 HSI surveys.	Dry- scoped out

6	Last undertaken in 2016 with a score of 0.39, poor	Not Applicable	Not Applicable	Small waterbody located north of the A303 and between the River Avon and the Sewage Works south west of Bulford. Recorded in previous reports but not on OS maps. Obstructed by vegetation and dry at the time of the HSI in 2017 /2018 and previous surveys. No access permitted at time of 2021 survey, however, scoped out as not considered suitable for GCN. This is not considered to constitute a constraint to the validity of the results, as works are not planned to be undertaken within 500m of the waterbody that would trigger the requirement of a licence (being mainly limited to maintenance works, as the electricity connection will be connected via the River Avon bridge rather than crossing the River Avon.	No access – scoped out during 2017 survey
7	Last undertaken in 2017 with a score of 0.47, poor	Not Applicable	Not Applicable	Pond with ornamental waterfowl. The pond was scoped out from further survey in 2017, as it had a HSI of 0.47. Access was not permitted in 2021. Observations from adjacent land indicates that the status would not have changed.	No access – scoped out during 2017 survey
8	Not Applicable	Not Applicable	Not Applicable	Large fishing pond located to the north of the A303 on the edge of the 500m buffer. The waterbody is isolated from the main works by the River Avon and no works will occur within 500m of the pond as the River Avon crossing is no longer required as part of the Scheme.	No access – scoped out fishing pond and isolated
9	Not Applicable	Not Applicable	Not Applicable	Large fishing pond located to the north of the A303. The waterbody is isolated from the main works by the River Avon and no works will occur within 500m of the pond as the River Avon crossing is no longer required as part of the Scheme.	No access – scoped out fishing pond and isolated
10	Dry at time of survey therefore HSI not undertaken	Not Applicable	Not Applicable	Drainage ditch located along road and thus only contains water immediately after rainfall.	Dry- scoped out
11	Dry at time of survey therefore HSI not undertaken	Not Applicable	Not Applicable	Drainage ditch located along road and thus only contains water immediately after rainfall.	Dry- scoped out
12	Dry at time of survey therefore HSI not undertaken	Not Applicable	Not Applicable	Drainage ditch located along road and thus only contains water immediately after rainfall.	Dry- scoped out

13	Dry at time of survey therefore HSI not undertaken	Not Applicable	Not Applicable	Drainage ditch located along road and thus only contains water immediately after rainfall.	Dry- scoped out
14	Dry at time of survey therefore HSI not undertaken	Not Applicable	Not Applicable	Drainage ditch located along road and thus only contains water immediately after rainfall.	Dry- scoped out
15	Dry at time of survey therefore HSI not undertaken	Not Applicable	Not Applicable	Drainage ditch located along road and thus only contains water immediately after rainfall.	Dry- scoped out
16	Dry at time of survey therefore HSI not undertaken	Not Applicable	Not Applicable	Drainage ditch located along road and thus only contains water immediately after rainfall.	Dry- scoped out
17	Dry at time of survey therefore HSI not undertaken	Not Applicable	Not Applicable	Drainage ditch located along road and thus only contains water immediately after rainfall.	Dry- scoped out
18	Last undertaken in 2021 with a score of 0.31.	Not Applicable	Not Applicable	Wet ditch running along the arable field. The ditch was wet during the HSI surveys, yet was dry when presence / absence surveys commenced and throughout the survey period, further surveys were scoped out at this point. It is likely to only hold water during high rainfall.	Not suitable for GCN – scoped out
Not Applicable	Last undertaken in 2017 with a score of 0.40, poor.	Not Applicable	Not Applicable	Previously described as a small waterbody located on the edge of Parsonage Down National Nature Reserve north of the A303, which almost certainly dries annually. Very isolated from other ponds (only one within 1km not including those on the distal side of barriers). Limited and poor terrestrial habitat nearby.	No longer present

Appendix C: Detailed results of HSI assessment

HSI criteria	HSI Criteria description	Waterbody 1		Waterbody 3		Waterbody 4		Waterbody 18	
		Resulting criteria	Suitability index score	Resulting criteria	Suitability index score	Resulting criteria	Suitability index score	Resulting criteria	Suitability index score
Location	Ponds are scored according to their geographic location	A	1.00	A	1.00	A	1.00	A	1.00
Pond Area	m ² to nearest 50 m ²	450	0.91	150	0.30	50	0.10	150	0.30
Pond Drying	Never dries; rarely dries; sometimes dries; or, dries annually.	Sometimes dries	0.50	Rarely dries	1.00	Rarely dries	1.00	Dries annually	0.10
Water Quality	Based on abundance and diversity of invertebrate community: Good; Moderate; Poor; or, Bad.	Good	1.00	Moderate	0.67	Poor	0.33	Poor	0.33
Shade	Estimate percentage perimeter shaded (May – September).	10%	1.00	<60%	0.80	<60%	1.00	95%	0.30
Waterfowl	Absent; Minor; or, Major.	Minor	0.67	Minor	0.67	Absent	1.00	Absent	1.00
Fish	Absent; Possible; Minor; or, Major.	Absent	1.00	Absent	1.00	Major	0.01	Absent	1.00
Ponds	Number of ponds within 1 km of survey pond not separated by major barriers divided by pi.	<0.1	0.01	<0.1	0.01	<0.1	0.01	<0.1	0.01
Terrestrial Habitat	Good; Moderate; Poor; or, Bad.	Moderate	0.67	Good	1.00	Moderate	0.67	Moderate	0.67
Macrophytes	Estimated percentage value of macrophyte cover.	30%	0.60	70-80%	1.00	10%	0.40	10%	0.40
Resulting HSI Score		0.49		0.50		0.25		0.31	
Resulting HSI Category		Poor		Below average		Poor		Poor	

Appendix D: eDNA Results Letter

Client: Roz Owen,
Aecom



ADAS
Spring Lodge
172 Chester Road
Helsby
WA6 0AR

Tel: 01159 516747
Email: Helen.Rees@adas.co.uk

www.adas.uk

Sample ID: ADAS-0359 Condition on Receipt: Good Volume: Passed
Client Identifier: Amesbury A303 Description: pond water samples in preservative
Date of Receipt: 24/05/2021 Material Tested: eDNA from pond water samples

Determinant	Result	Method	Date of Analysis
Inhibition Control [†]	2 of 2	Real Time PCR	25/05/2021
Degradation Control [§]	Within Limits	Real Time PCR	25/05/2021
Great Crested Newt*	0 of 12 (GCN negative)	Real Time PCR	25/05/2021
Negative PCR Control (Nuclease Free Water)	0 of 4	Real Time PCR	As above for GCN
Positive PCR Control (GCN DNA 10 ⁻⁴ ng/μL) [#]	4 of 4	Real Time PCR	As above for GCN

Report Prepared by: Dr Helen Rees Report Issued by: Dr Ben Maddison

Signed:  Signed: 

Position: Director: Biotechnology Position: MD: Biotechnology

Date of preparation: 26/05/2021 Date of issue: 26/05/2021

eDNA analysis was carried out in accordance with the stipulated methodology found in the Technical Advice Note (WC1067 Appendix 5 Technical Advice Note) published by DEFRA and adopted by Natural England.

** If all PCR controls and extraction blanks give the expected results a sample is considered: negative for great crested newt if all of the replicates are negative; positive for great crested newt if one or more of the replicates are positive.*

[†] Recorded as the number of positive replicate reactions at expected C_t value. If the expected C_t value is not achieved, the sample is considered inhibited and is diluted as per the technical advice note prior to amplification with great crested newt primer and probes.

[§] No degradation is expected within time frame of kit preparation, sample collection and analysis.

[#] Additional positive controls (10⁻¹, 10⁻², 10⁻³ ng/μL) are also routinely run, results not shown here.

Appendix 1: Interpretation of results

Sample Condition

Upon sample receipt we score your samples according to quality: good, low sediment, medium sediment, high sediment, white precipitate, and presence of algae.

There are three reasons as to why sediment should be avoided:

1. It is possible for DNA to persist within the sediment for longer than it would if it was floating in the water which could lead to a false positive result i.e. in this case GCN not recently present but present a long time ago
2. In some cases sediment can cause inhibition of the PCR analysis used to detect GCN eDNA within samples which could lead to an indeterminate result.
3. In some cases sediment can interfere with the DNA extraction procedure resulting in poor recovery of the eDNA which in turn can lead to an indeterminate result.

Algae can make the DNA extraction more difficult to perform so if it can be avoided then this is helpful.

Sometimes samples contain a white precipitate which we have found makes the recovery of eDNA very difficult. This precipitate can be present in such high amounts that it interferes with the eDNA extraction process meaning that we cannot recover the degradation control (nor most likely the eDNA itself) at sufficient levels for the control to be within the acceptable limits for the assay, therefore we have to classify these type of samples as indeterminate.

What do my results mean?

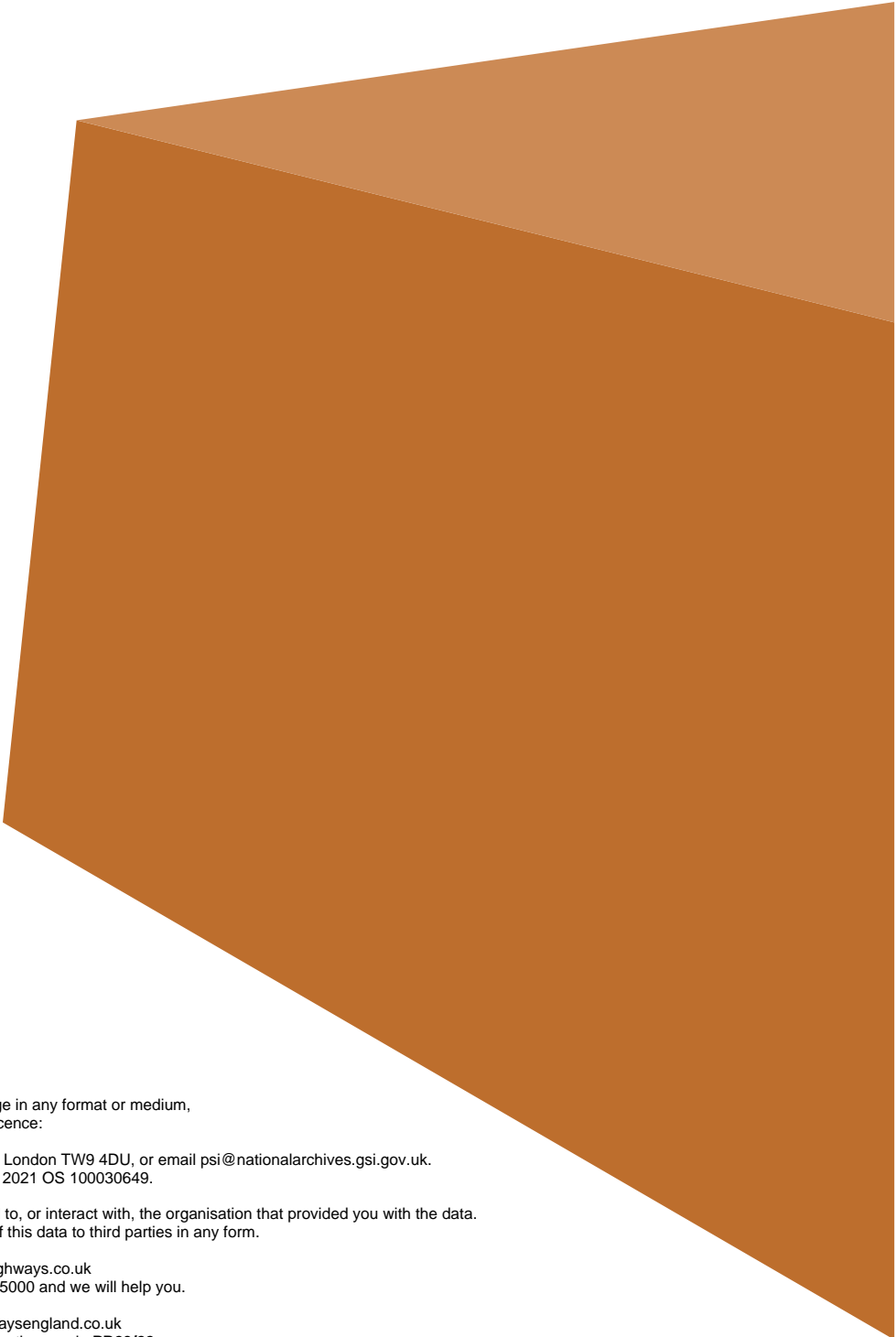
A positive result means that great crested newts are present in the water or have been present in the water in the recent past (eDNA degrades over around 7-21 days).

A negative result means that DNA from the great crested newt has not been detected in your sample.

On occasion an inconclusive result will be issued. This occurs where the DNA from the great crested newt has not been detected but the controls have indicated that either: the sample has been degraded and/or the eDNA was not fully extracted (poor recovery); or the PCR inhibited in some way. This may be due to the water chemistry or may be due to the presence of high levels of sediment in samples which can interfere with the DNA extraction process. A re-test could be performed but a fresh sample would need to be obtained. We have successfully performed re-tests on samples which have had high sediment content on the first collection and low sediment content (through improved sample collection) on the re-test. If water chemistry was the cause of the indeterminate then a re-test would most likely also return an inconclusive result.

The results will be recorded as indeterminate if the GCN result is negative and the degradation result is recorded as:

1. evidence of decay - meaning that the degradation control was outside of accepted limits
2. evidence of degradation or residual inhibition - meaning that the degradation control was outside of accepted limits but that this could have been due to inhibitors not being removed sufficiently by the dilution of inhibited samples (according to the technical advice note)



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