East Anglia ONE North Offshore Windfarm

Appendix 22.2

eDNA Survey Report

Preliminary Environmental Information
Volume 3
Document Reference – EA1N-DEVWF-ENV-REP-IBR-000291_002

Revision Summary					
Rev	**Date**	**Document Status**	**Prepared by**	**Checked by**	**Approved by**
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Description of Revisions			
Rev	**Page**	**Section**	**Description**
01	n/a	n/a	Final Draft

Table of Contents

Appendix 22.2 is additionally supported by:
Annex 1: eDNA result report
Annex 2: ***Figures 22.2.1*** and ***22.2.2***

Appendix 22.2 is supported by the figures listed below.

Figure Number	Title
Figure 22.2.1	Waterbody Locations
Figure 22.2.2	Waterbody Survey Results

Appendix 22.2 is supported by the tables listed below.

Table Number	Title
Table A22.1	Summary of Pond HSI Assessment
Table A22.2	Summary of Pond eDNA Survey

Glossary of Acronyms

EcIA	Ecological impact Assessment
eDNA	Environmental DNA
EPS	European Protected Species
EPSML	European Protected Species Mitigation License
ES	Environmental Statement
ETG	Expert Topic Group
HDD	Horizontal Directional Drilling
HRA	Habitats Regulations Assessment
HSI	Habitat Suitability Index
JNCC	Joint Nature Conservation Committee
km	Kilometres
m	Metres
NERC Act	Natural Environment and Rural Communities Act

Glossary of Terminology

Applicant	East Anglia ONE North Limited. ScottishPower Renewables is the parent company of East Anglia ONE North limited
Construction consolidation sites	Compounds which will contain laydown, storage and work areas for onshore construction works. The HDD construction compound will also be referred to as a construction consolidation site.
Development Area	Area containing all onshore and offshore infrastructure, transmission works, construction consolidation sites, and mitigation areas.
East Anglia ONE North project	The proposed project consisting of up to 67 wind turbines, up to four offshore electrical platforms, up to one offshore construction operation and maintenance platform, inter-array cables, platform link cables, up to one operational meteorological mast, up to two offshore export cables, fibre optic cables, landfall infrastructure, onshore cables and ducts, onshore substation, and National Grid infrastructure.
European site	Sites designated for nature conservation under the Habitats Directive and Birds Directive, as defined in regulation 8 of the Conservation of Habitats and Species Regulations 2017 and regulation 18 of the Conservation of Offshore Marine Habitats and Species Regulations 2017. These include candidate Special Areas of Conservation, Sites of Community Importance, Special Areas of Conservation and Special Protection Areas.
Evidence Plan Process	A voluntary consultation process with specialist stakeholders to agree the approach to the EIA and the information required to support HRA.
Horizontal directional drilling (HDD)	A method of cable installation where the cable is drilled beneath a feature without the need for trenching.
Jointing Bay	Underground structures constructed at regular intervals along the onshore cable route to join sections of cable and facilitate installation of the cables into the buried ducts.
Landfall	The area where the offshore export cables would make contact with land, and connect to the onshore cables.
Link boxes	Underground chambers or above ground cabinets next to the cable trench housing electrical earthing links.
Mitigation areas	Areas captured within the Development Area specifically for mitigating expected or anticipated impacts.
National Grid infrastructure	The proposed East Anglia ONE North project will require connection into an additional substation for ultimate connection to national electricity grid. The required National Grid infrastructure comprising a National Grid substation, connection to the existing electricity pylons and associated works will be consented as part of the proposed East Anglia ONE North project Development Consent Order but will be National Grid owned assets.
National Grid overhead line works	Works required to upgrade the existing electricity pylons and overhead lines to transport electricity from the National Grid substation to the national electricity grid

National Grid overhead line works area	The proposed area for National Grid overhead line realignment works.
National Grid substation	The substation (including all of the electrical equipment within it) necessary to connect the proposed East Anglia ONE North project to the national electricity grid which will be owned by National Grid but is being consented as part of the proposed East Anglia ONE North project Development Consent Order.
National Grid substation location	The proposed location of the National Grid substation required to connect the proposed East Anglia ONE North project to the national electricity grid.
Natura 2000 site	A site forming part of the network of sites made up of Special Areas of Conservation and Special Protection Areas designated respectively under the Habitats Directive and Birds Directive.
Onshore cable corridor	The corridor within which the onshore cable route will be located.
Onshore cable route	This is the construction swathe within the onshore cable corridor which would contain onshore cables as well as temporary ground required for construction which includes cable trenches, haul road and spoil storage areas.
Onshore cables	The cables which would bring electricity from landfall to the onshore substation. The onshore cable is comprised of up to six power cables and two fibre optic cables.
Proposed onshore development Area	Onshore transmission works, mitigation areas and temporary construction facilities such as access roads or construction consolidation sites and National Grid infrastructure.
Onshore infrastructure	The combined name for all infrastructure associated with the proposed East Anglia ONE North project from landfall to grid connection.
Onshore substation	The East Anglia ONE North substation and all of the electrical equipment, both within and connecting to the National Grid infrastructure
Onshore substation location	The proposed location of the onshore substation for the proposed East Anglia ONE North project.
Onshore study area	All onshore areas being considered for the placement of onshore infrastructure or temporary construction consolidation sites. This includes areas being considered for National Grid infrastructure, East Anglia ONE North onshore substation, onshore cable corridor and landfall.
Onshore transmission works	Landfall, onshore cable route and onshore substation location and National Grid substation location. This does not include temporary construction facilities such as access roads or construction consolidation sites.
Transition Bay	Underground structures at the landfall that house the joints between the offshore export cables and the onshore cables.

22.2 eDNA Survey Report

22.1 Introduction

22.1.1 Purpose of Survey

1. This report describes the approach and findings of great crested newt *Triturus cristatus* surveys undertaken in support of the proposed East Anglia ONE North project.

2. Royal HaskoningDHV was commissioned to undertake surveys of all ponds within the onshore study area for great crested newt in May and June 2018 (herein referred to as the onshore great crested newt survey area). This precluded the undertaking of great crested newt presence/absence field surveys using traditional techniques (as defined in English Nature 2001). Instead a survey was undertaken using the environmental DNA (eDNA) approach (Briggs et al. 2014). This is an approved valid method for great crested newt presence/absence survey and this approach was agreed with stakeholders at the Expert Topic Group (ETG) meeting in April 2018.

22.1.2 Scope of Works

3. The onshore great crested newt survey area incorporated all land within the onshore study area and should be noted that this includes a wider area than just the land within the East Anglia ONE North proposed onshore development area.

4. The scope of works for the 2018 onshore great crested newt survey was as follows:

   - Identify all ponds within the onshore study area (i.e. the onshore great crested newt survey area);
   - Complete Habitat Suitability Index (HSI) assessment of all potentially suitable ponds within the onshore great crested newt survey area to indicate their likely suitability for great crested newts; and
   - Undertake eDNA survey of all ponds with potential to support great crested newts in the onshore great crested newt survey area to determine likely presence or absence.

5. The methods and results of the great crested newt survey are reported in this appendix, along with an evaluation of the results to inform the subsequent Ecological Impact Assessment (EcIA). Due to the limitations associated with the eDNA method it was not possible to collect data on other species of amphibian

during the survey, so the remit of this report is restricted to great crested newt and other species of amphibian are not considered further.

22.1.3 Relevant Legislation

6. The great crested newt is listed under Schedule 5 of the Wildlife and Countryside Act 1981 (as amended) and Schedule 2 of the Conservation of Habitats and Species Regulations 2017 (as amended). This legislation, when taken together, results in a level of protection that prohibits the intentional, deliberate or reckless:

- Killing, injuring, taking or disturbance of great crested newts;
- Damaging, destroying or obstructing any place used by great crested newts for the purposes of breeding, sheltering or protection; and
- Selling and/or advertising for sale a great crested newt or any part thereof.

7. The great crested newt is listed as a species of principal importance for nature conservation in England in accordance with Section 41 of the Natural Environment and Rural Communities (NERC) Act 2006. Section 40 of the same Act requires that local and regional authorities have regard to the conservation of biodiversity in England, when carrying out their normal functions.

22.2 Survey Methods

22.2.1 Habitat Suitability (HSI Assessment)

8. A total of 38 potentially suitable water bodies (ponds and watercourses) were identified within the onshore great crested newt survey area (***Figure 22.2.1***). All 38 of these waterbodies were assessed for their potential to support great crested newt using the HSI in accordance with standard methodology (Oldham *et al*. 2000). These HSI assessments were undertaken during in May and June 2018.

9. The HSI assessment considers the following ten habitat attributes that are considered to influence the suitability of a pond for breeding great crested newts:

- Location – within a UK-wide context reflecting the differences in national distribution of this species;
- Area – water bodies between 100m^2 and 300m^2 in size are considered to represent the most suitable habitat for great crested newt;
- Drying – the number of years in which a pond dries over a ten-year period. Occasional drying kills fish which is beneficial for great crested newt, but the species predominantly favours ponds that do not dry out every year;

- Water quality – qualitative evidence-based assessment to infer good (diverse aquatic invertebrate assemblage), moderate (moderate invertebrate diversity), poor (low invertebrate diversity, few submerged plants) or bad (clearly polluted) water quality;
- Shade – percentage of pond perimeter shaded to at least 1m from the shore. Great crested newt favour lightly shaded water bodies;
- Waterfowl – qualitative evidence-based assessment of presence or absence and numbers is made. Large numbers of waterfowl can result in nutrient enrichment of the water and habitat damage, which is less favourable for great crested newt;
- Fish – qualitative evidence-based assessment of likely presence or absence is made. Great crested newt favour breeding ponds that do not support fish because their open-water swimming larvae are vulnerable to fish predation; and
- Number of waterbodies within 1 kilometre (km) – great crested newt populations are typically best developed where they have access to a network of ponds, and therefore the species is more likely to be found where there are several ponds within 1 km that are linked by suitable terrestrial habitat; and macrophyte cover – percentage of pond surface area occupied by macrophyte cover. Female great crested newts require aquatic vegetation for egg-laying.

22.2.2 eDNA Survey

10. Water samples were collected by Royal HaskoningDHV great crested newt licenced ecologists from all water bodies where landowner permission had been granted on the 30th May, 31st May and 29th June 2018. Each sample was then sent to Fera Science Solutions (FERA) for analysis for eDNA in accordance with approved field and laboratory protocols (Briggs et al. 2014). Water bodies were not entered by surveyors during sample collection, and new sterile equipment supplied by FERA was used to collect each water sample, to prevent contamination between samples.

11. The presence or absence of great crested newt from each of the surveyed water bodies was determined based on the results of the eDNA analysis. If eDNA is detected this provides confirmation of presence and the relevant water bodies are likely require further assessment. If eDNA is not detected then this provides high confidence that there is no reasonable likelihood of great crested newt being present in the relevant water bodies, and they require no further assessment with regards to this species.

22.2.3 Limitations

12. Surveys using the eDNA method have a benefit over traditional surveys in that they can be validly completed within a single visit to each relevant water body between mid-May and the end of June. Therefore, they can be programmed and completed later in the newt survey season when surveys using traditional methods are not possible. However, the eDNA sampling technique does not enable an estimate of population size class; rather it provides confirmation of presence or likely absence of great crested newts in the water body concerned. In addition, the method cannot currently be used to record the presence or absence of other species of amphibian present in the surveyed water bodies. In some circumstances, further survey is needed to allow estimation of the population size class, particularly where a European Protected Species Mitigation Licence (EPSML) may subsequently need to be obtained. However, for the purposes of this report consideration of the geographic location and relative distance of the only pond testing positive for great crested newt, population size class data is not relevant and no further survey work is necessary at this time. Therefore, reliance entirely on the result of eDNA survey is not a limitation to the current assessment presented in this report. The available data is sufficient to determine the potential consequences of the proposed East Anglia ONE North project for great crested newt.

13. Based on Natural England's standing advice on great crested newts, the window for collecting eDNA samples is 15th April to 30th June. All the eDNA samples were collected during this period and therefore are compliant with the eDNA methodology and analysis.

14. Only 32 of the 38 waterbodies had been granted landowner permission. Therefore, the remaining six waterbodies that were not accessed and therefore not surveyed in 2018 will be subject to future survey(s) once landowner permission has been obtained (dependent upon the results of the HSI assessment).

22.3 Results

22.3.1 2018 HSI Assessment

15. All 38 waterbodies identified to be within the onshore great crested newt survey area were subject to a HSI assessment. Of the 38 waterbodies assessed, 10 were noted as being dry and therefore scoped out from any further survey. The remaining 28 were therefore subject to an eDNA survey. However, five of these 28 waterbodies were not granted landowner access and consequently were not subject to an eDNA survey. The remaining 23 waterbodies were granted landowner access and where therefore subject to an eDNA survey.

16. The results of the HSI assessments for all 38 waterbodies is summarised in ***Table A22.1*** below, and their locations are shown on ***Figure 22.2.1***.

Table A22.1 Summary of Pond HSI Assessment

Water body (Pond) Reference	Pond Type	HSI Score	Scoped into eDNA survey (yes/no)
160	Small waterbody located within a corner of two arable fields and busy road. Oak, alder, hawthorn and dogs mercury present. No macrophytes present.	0.35	Yes but no access granted
140	Medium sized waterbody within a corner of an arable field. Connected to a dry ditch network. Hawthorn, common nettle, primrose, common hogweed and grass species present. Dogs mercury and common reed also present.	0.81	Yes
134	Medium sized waterbody within a residential garden. Hawthorn, goat willow, daffodils, primrose, common reed, broadleaved dock, water milfoil and soft rush present.	0.48	Yes but no access granted
133	Small waterbody within residential garden. Ash, goat willow and hawthorn present. No egg laying material noted and banks are steep sided. Daffodils, broadleaved dock and spear thistle also noted.	0.46	Yes but no access granted
135	Medium sized waterbody within residential garden next to arable fields. Waterbody is located within a grassland area surrounded by trees. Species include hawthorn and goat willow. Common reed and duckweed also present. Steep sided banks.	0.51	Yes
TN288	Waterbody (ditch) located between arable fields. Noted to be predominately dry at the time of the survey with steep sided banks.	0.82	No – Dry at time of survey
TN289	Waterbody (ditch) located between arable fields. Noted to be predominately dry at the time of the survey with steep sided banks.	0.82	No – Dry at time of survey
143	Waterbody (ditch) running around farmhouse and garden. Waterbody is fenced with steep sided banks. No egg laying material present.	0.48	Yes but no access granted
152	Small waterbody within a residential garden with steep sided concrete banks. Oak, hawthorn, lesser celandine, common dog violet, common hogweed, common nettle and primrose also present. Appears to be part of an old mill pond and ditch system.	0.72	Yes
139	Medium sized waterbody in arable field that is fenced off by trees, pheasant feeders and hides	0.81	Yes

Water body (Pond) Reference	Pond Type	HSI Score	Scoped into eDNA survey (yes/no)
	also present. Goat willow, ash, hawthorn, common hogweed, common nettle, bramble, broadleaved dock and primrose also present.		
153	Small heart shaped waterbody within corner of arable field with steep banks. Pipe inlet present with limited macrophytes. Scrub areas present.	0.50	No – Dry at time of survey
114	Long waterbody at edge of woodland. Leaf litter and large woody debris present. Absence of suitable egg laying material. Ground flora is limited to bramble and hogweed. Trees are predominately oak and sycamore interspersed with hawthorn.	0.62	Yes
113	Connected to Pond 114 and therefore similar waterbody description.	0.57	Yes
115	Connected to Pond 114 and therefore similar waterbody description.	0.62	Yes
112	Medium sized waterbody in residential garden. Common reed present. Concrete wall surrounding waterbody with fish also present.	0.32	Yes
123	Connected to Pond 114 and therefore similar waterbody description.	0.62	No – Dry at time of survey
125	Connected to Pond 114 and therefore similar waterbody description.	0.62	Yes
124	Connected to Pond 114 and therefore similar waterbody description.	0.62	No – Dry at time of survey
117	Waterbody with shallow banks and partially fenced. Goat willow, bramble, daffodils and common nettle also present.	0.39	Yes
120	Small waterbody within a woodland with a large amount of woody debris noted. Small area of water mint also present.	0.44	Yes
118	Elongated waterbody noted to be partially dry at the time of the survey.	0.38	Yes
119	Elongated waterbody noted to be partially dry at the time of the survey.	0.38	No – Dry at time of survey
137	Small waterbody at edge of arable field and surrounded by semi-improved grassland. Oak,	0.55	Yes

Water body (Pond) Reference	Pond Type	HSI Score	Scoped into eDNA survey (yes/no)
	hawthorn, bramble, common hogweed and common nettle also present.		
73	Waterbody within arable field and appeared to be subject to flooding. Surrounding vegetation consists of willow, bramble and dandelions.	0.60	Yes
NP3	Small pond within a deep depression within centre of arable field. Steep sided and access only possible from the northern end of the waterbody. Horse chestnut, hawthorn, oak, bramble, thistle, daffodils, common nettle, curled dock and cow parsley present around waterbody.	0.54	No - dry at time of survey
NP2	Small pond in corner of arable field and located within an area of marshy grassland. Grey willow, broadleaved dock and grass species present.	0.59	No – Dry at time of survey
86	Small waterbody on the edge of heathland surrounded by semi-improved grassland. Willow, broadleaved dock, oak, white clover, red dead nettle, lovage and watercress is present. Duckweed also present.	0.88	Yes but no access granted
NP1	Small waterbody within a wooded depression between two arable fields. Water quality observed to be very poor with surrounding vegetation predominately bracken, bramble, common nettle, oak and white willow.	0.39	Yes
2	Medium sized man-made waterbody that is concrete lined and located within residential property. Owner advised that goldfish are present. No macrophytes present and waterbody is surrounded by grassland that is regularly mown with limited herb species present.	0.49	Yes but no access granted
3	Small pond in residential garden. Waterbody is located within area of short grass.	0.48	Yes
6	Medium sized waterbody in agricultural field (in crop with beetroot). No shade around waterbody with common reed present. Water quality noted as being poor and subject to regular run-off from agricultural field.	0.51	Yes
158	Small waterbody within a depression within a woodland. Appears to be dry at times with a lack of surrounding terrestrial habitat.	0.21	Yes
4	Ephemeral waterbody within arable field. Shallow waterbody noted to be drying at the time of the survey. Poor water quality noted.	0.29	No – Dry at time of survey

Water body (Pond) Reference	Pond Type	HSI Score	Scoped into eDNA survey (yes/no)
9	Medium sized waterbody within a field depression. Water quality noted to be extremely poor with silty substrate. Hawthorn, bracken, bramble, grass species and reeds also present.	0.34	Yes
15	Ditch running through woodland with a sluggish flow. Steep banks present with no emergent vegetation or bankside vegetation.	0.58	No – Dry at time of survey
7	Small “puddle” in arable field with approximately 10cm of water at the time of the survey. No macrophytes present and appeared to experience dryness at times.	0.45	No – Dry at time of survey
18	Large pond within golf course. Bulrush and common reed present with areas of scrub around edges. Silver birch present. Surrounding habitat is regularly used by golfers but waterbody is connected to wider habitat.	0.52	Yes
19	Pond adjacent to golf course within woodland. Mainly oak, bramble, bracken, nettle and common reed present.	0.67	Yes

22.3.2 eDNA Survey

17. Of the 28 ponds scoped into requiring an eDNA survey, five were not granted access and therefore only 23 ponds were subject to the 2018 eDNA survey effort. Three of these ponds returned a positive result for great crested newt. The full eDNA results from the laboratory are provided in Annex 1 of this report and are summarised in ***Table A22.2*** below.

Table A22.2 Summary of Pond eDNA Survey

Water body (Pond) Reference	eDNA Positive for GCN?	FERA reference
140	Negative	S18-004115
135	Positive	S18-004091
152	Positive	S18-004122
139	Negative	S18-004120
114	Negative	S18-004118
113	Negative	S18-004121
115	Negative	S18-004123

Water body (Pond) Reference	eDNA Positive for GCN?	FERA reference
112	Negative	S18-004114
125	Negative	S18-004117
117	Positive	S18-004124
120	Negative	S18-004116
118	Negative	S18-004127
137	Negative	S18-004119
73	Negative	S18-004129
NP1	Negative	S18-004125
3	Negative	S18-004128
6	Negative	S18-004131
158	Negative	S18-004132
9	Negative	S18-004093
18	Negative	S18-004073
19	**Negative**	**S18-004076**

22.4 Conclusion

18. Three of the 21 ponds subject to the eDNA survey returned a positive result for great crested newts, namely:

- Pond 135;
- Pond 152; and
- Pond 117.

19. These are shown on ***Figure 22.2.2***.

20. Great crested newts are known to exist as meta-populations within small aggregations or clusters of ponds, i.e. individual great crested newts may travel between several ponds that are within proximity to one another (English Nature 2001). This allows genetic diversity within the breeding population and allows the population to move freely, depending on changing conditions. Based on this information, clusters of ponds within distinct areas can be classified as potentially supporting distinct meta-populations of great crested newts.

21. As these ponds are within or adjacent to areas where works associated with the proposed East Anglia ONE North project will be undertaken, further surveys for population class assessments will be undertaken in the appropriate survey window prior to the commencement of construction. The findings of which will inform and develop the EPSML and mitigation measures that will be required to ensure compliance with the legislation afforded to great crested newts. Furthermore, a Rapid Risk Assessment has been undertaken which concluded that a European Protected Species (EPS) licence also is likely to be required. Although none of these ponds would be directly affected or lost because of either proposed development, potential effects would be limited to effects on terrestrial habitat.

22. The remaining 18 ponds that were surveyed in 2018 returned a negative result for great crested newt eDNA, so there is no reasonable likelihood of this species being present in association with these ponds (***Figure 22.2.2***).

23. Of the 5 ponds which were not surveyed due to access constraints, if they fall within the proposed onshore development area to be taken forward for the final application, the strategy will be to progress access agreements with landowners with the aim of conducting eDNA surveys in 2019.

22.5 References

Briggs, J., Ewald, N., Valentini, A., Gaboriaund, C., Griffiths, R.A., Foster, J., Wilkinson, J., Arnett, A., Williams, P. & Dunn, F. (2014) Analytical and methodological development for improved surveillance of the Great Crested Newt. Appendix 5: Technical advice note for field and laboratory sampling of great crested newt (*Triturus cristatus*) E*nvironmental DNA*, Freshwater Habitats Trust, Oxford.

English Nature, (2001) *Great Crested Newt Mitigation Guidelines*. English Nature, Peterborough.

Natural England, (2016) Great Crested Newt Method Statement for EPS Licence Application, [Online], Available: https://www.gov.uk/government/publications/great-crested-newts-apply-for-a-mitigation-licence Accessed [July 31 2018].

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.

Annex 1: eDNA Result Report

DNA Analysis Report - Commercial in Confidence

Customer:	Royal Haskoning DHV
Address:	[redacted]
Contact:	[redacted]
Email:	[redacted]
Tel:	[redacted]
Report date:	12-Jul-2018
Order Number:	GCN18-0836
Samples:	Pond Water
Analysis requested:	Detection of Great Crested Newt eDNA from pond water.

Thank you for submitting your samples for analysis with the Fera eDNA testing service. The details of the analysis are as follows:

Method:

The method detects pond occupancy from great crested newts (GCN) using traces of DNA shed into the pond environment (eDNA). The detection of GCN eDNA is carried out using real time PCR to amplify part of the cytochrome 1 gene found in mitochondrial DNA. The method followed is detailed in Biggs J., et al, (2014). Analytical and methodological development for improved surveillance of the Great Crested Newt. Appendix 5. Technical advice note for field and laboratory sampling of great crested newt (Triturus cristatus) environmental DNA. Freshwater Habitats Trust, Oxford.

The limits of this method are as follows: 1) the results are based on analyses of the samples supplied by the client and as received by the laboratory, 2) any variation between the characteristics of this sample and a batch will depend on the sampling procedure used. 3) the method is qualitative and therefore the levels given in the score are for information only, they do not constitute the quantification of GCN DNA against a calibration curve, 4) a 'not detected' result does not exclude presence at levels below the limit of detection.

The results are defined as follows:

Positive:	DNA from the species was detected.
eDNA Score:	Number of positive replicates from a series of twelve.
Negative:	DNA from the species was not detected; in the case of negative samples the DNA extract is further tested for PCR inhibitors and degradation of the sample.
Inconclusive:	Controls indicate degradation or inhibition of the sample, therefore the lack of detection of GCN DNA is not conclusive evidence for determining the absence of the species in the sample provided.

DNA Analysis Report - Commercial in Confidence

CustomerReference	Fera Reference	GCN Detection	eDNA Score	Inhibition	Degradation
Pond 19	S18-004076	Negative	0	No	No
Pond 18	S18-004073	Negative	0	No	No
Pond 135	S18-004091	Positive	12	n/a	n/a

The results indicate that eDNA for great crested newts was detected in one of the samples and in the remaining samples eDNA was not detected (as detailed in the table above). Analysis was conducted in the presence of the following controls: 1) extraction blank, 2) appropriate positive and negative PCR controls for each of the TaqMan assays (GCN, Inhibition, and Degradation). All controls performed as expected.

This test procedure was developed using research funded by the Department of Environment, Food and Rural Affairs.

[redacted]

DNA Analysis Report - Commercial in Confidence

Customer:	Royal Haskoning DHV
Address:	[redacted]
Contact:	[redacted]
Email:	[redacted]
Tel:	[redacted]
Report date:	14-Jun-2018
Order Number:	GCN18-0836
Samples:	Pond Water
Analysis requested:	Detection of Great Crested Newt eDNA from pond water.

Thank you for submitting your samples for analysis with the Fera eDNA testing service. The details of the analysis are as follows:

Method:
The method detects pond occupancy from great crested newts (GCN) using traces of DNA shed into the pond environment (eDNA). The detection of GCN eDNA is carried out using real time PCR to amplify part of the cytochrome 1 gene found in mitochondrial DNA. The method followed is detailed in Biggs J., et al, (2014). Analytical and methodological development for improved surveillance of the Great Crested Newt. Appendix 5. Technical advice note for field and laboratory sampling of great crested newt (Triturus cristatus) environmental DNA. Freshwater Habitats Trust, Oxford.

The limits of this method are as follows: 1) the results are based on analyses of the samples supplied by the client and as received by the laboratory, 2) any variation between the characteristics of this sample and a batch will depend on the sampling procedure used. 3) the method is qualitative and therefore the levels given in the score are for information only, they do not constitute the quantification of GCN DNA against a calibration curve, 4) a 'not detected' result does not exclude presence at levels below the limit of detection.

The results are defined as follows:

Positive:	DNA from the species was detected.
eDNA Score:	Number of positive replicates from a series of twelve.
Negative:	DNA from the species was not detected; in the case of negative samples the DNA extract is further tested for PCR inhibitors and degradation of the sample.
Inconclusive:	Controls indicate degradation or inhibition of the sample, therefore the lack of detection of GCN DNA is not conclusive evidence for determining the absence of the species in the sample provided.

DNA Analysis Report - Commercial in Confidence

CustomerReference	Fera Reference	GCN Detection	eDNA Score	Inhibition	Degradation
P112	S18-004114	Negative	0	No	No
P140	S18-004115	Negative	0	No	No
P120	S18-004116	Negative	0	No	No
P125	S18-004117	Negative	0	No	No
P137	S18-004119	Negative	0	No	No
P114	S18-004118	Negative	0	No	No
P139	S18-004120	Negative	0	No	No
P113	S18-004121	Negative	0	No	No
P152	S18-004122	Positive	3	n/a	n/a
P115	S18-004123	Negative	0	No	No
P117	S18-004124	Positive	4	n/a	n/a
NP1	S18-004125	Negative	0	No	No
P9	S18-004126	Negative	0	No	No
P118	S18-004127	Negative	0	No	No
P3	S18-004128	Negative	0	No	No
P73	S18-004129	Negative	0	No	No
P6	S18-004131	Negative	0	No	No
P158	S18-004132	Negative	0	No	No

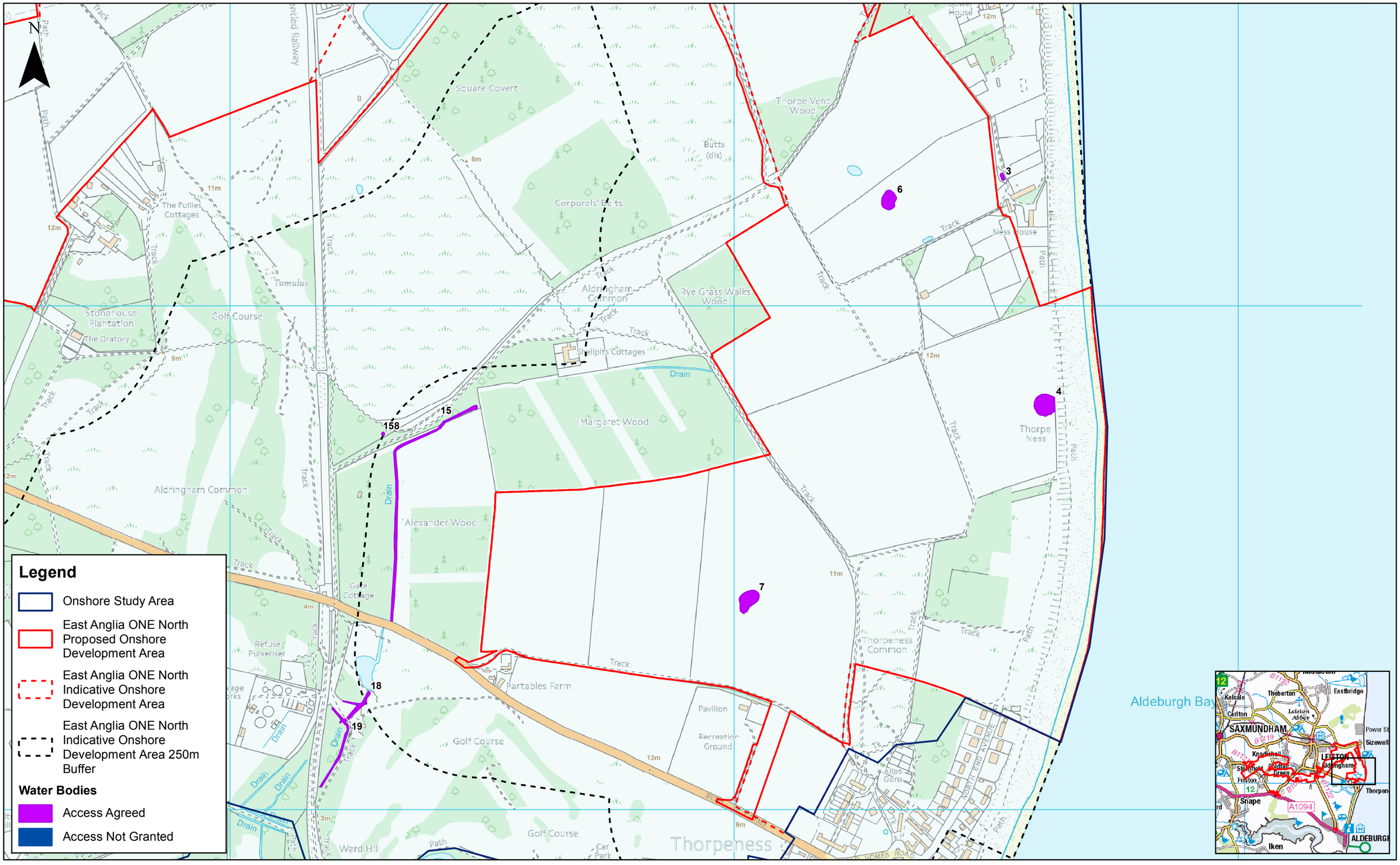
The results indicate that eDNA for great crested newts was detected in two of the samples and in the remaining samples eDNA was not detected (as detailed in the table above).

Analysis was conducted in the presence of the following controls: 1) extraction blank, 2) appropriate positive and negative PCR controls for each of the TaqMan assays (GCN, Inhibition, and Degradation). All controls performed as expected.

This test procedure was developed using research funded by the Department of Environment, Food and Rural Affairs.

[redacted]

Annex 2: Supporting Figures



Rev	Date	By	Comment		
				Prepared:	FC
1	21/11/2018	FC	First Issue.	Checked:	CS
				Approved:	AH

1:7,000 Scale @ A3

0 200 400 Metres

East Anglia ONE North

Access Status for Water Bodies

Drg No	EA1N-DEV-DRG-IBR-000373
Rev	1
Date	21/11/18
Figure	22.2.1a

Coordinate System: BNG Datum: OSGB36

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N

Legend

- Onshore Study Area
- East Anglia ONE North Proposed Onshore Development Area
- East Anglia ONE North Indicative Onshore Development Area
- East Anglia ONE North Indicative Onshore Development Area 250m Buffer

Water Bodies

- Access Agreed
- Access Not Granted

Rev	Date	By	Comment
1	21/11/2018	FC	First Issue.

Prepared: FC
Checked: CS
Approved: AH

1:7,000
Scale @ A3

0 200 400 Metres

East Anglia ONE North

Access Status for Water Bodies

Drg No	EA1N-DEV-DRG-IBR-000373
Rev	1
Date	21/11/18
Figure	22.2.1b

Coordinate System: BNG
Datum: OSGB36

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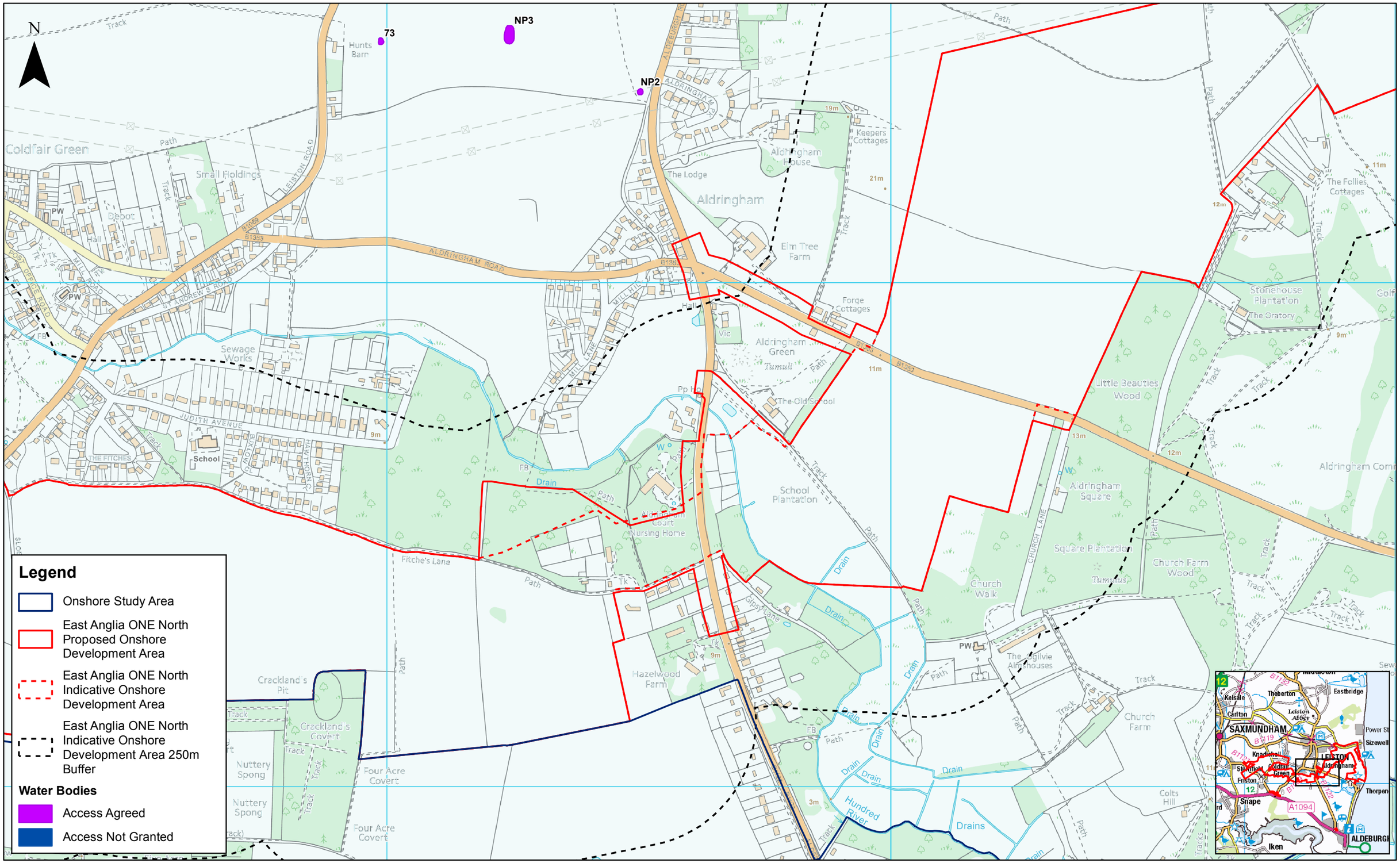

Rev	Date	By	Comment
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Prepared: FC
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1:7,000 Scale @ A3

0 200 400 Metres

East Anglia ONE North

Access Status for Water Bodies

Drg No	EA1N-DEV-DRG-IBR-000373
Rev	1
Date	21/11/18
Figure	22.2.1c

Coordinate System: BNG Datum: OSGB36

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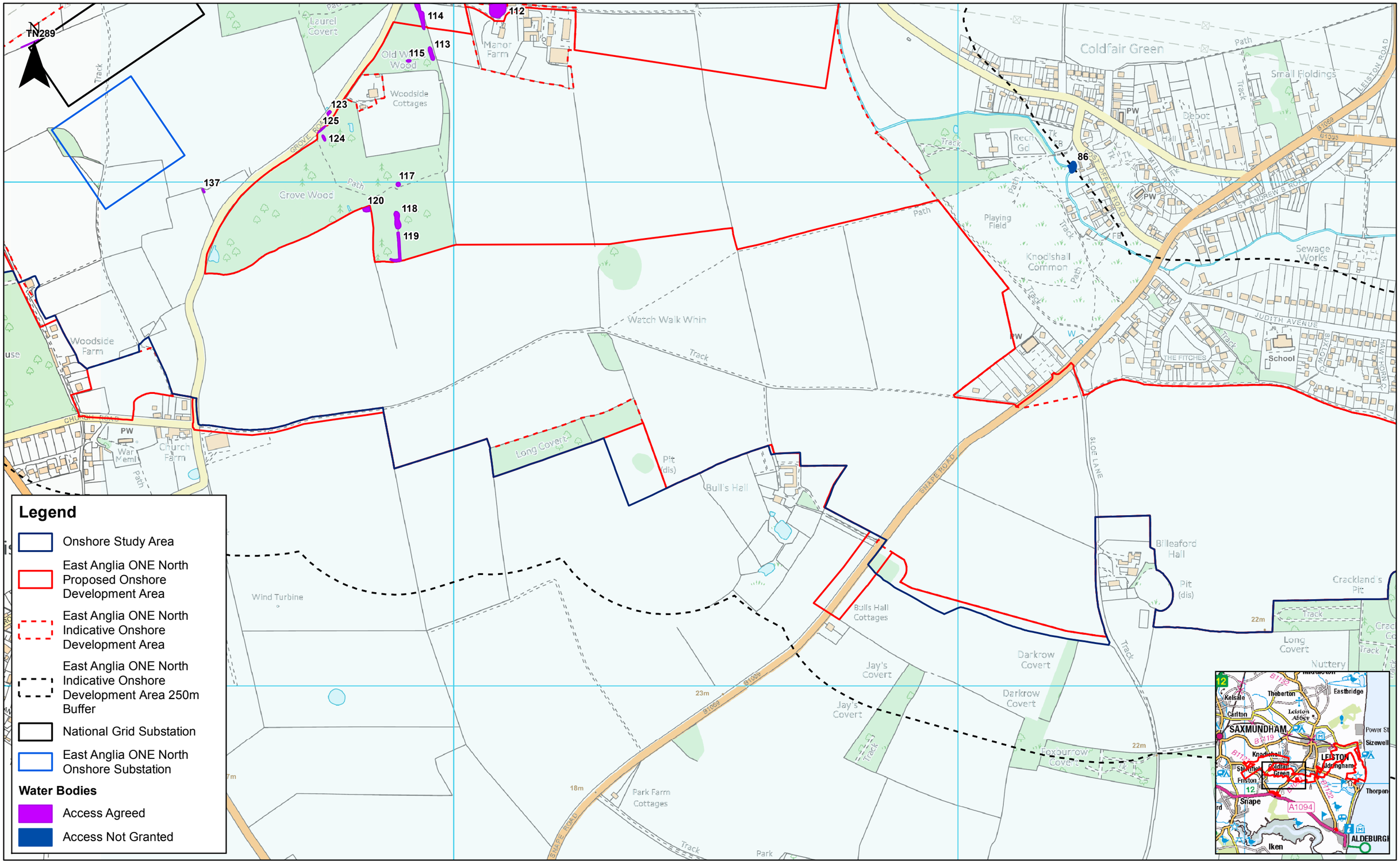

Legend

- Onshore Study Area
- East Anglia ONE North Proposed Onshore Development Area
- East Anglia ONE North Indicative Onshore Development Area
- East Anglia ONE North Indicative Onshore Development Area 250m Buffer
- National Grid Substation
- East Anglia ONE North Onshore Substation

Water Bodies

- Access Agreed
- Access Not Granted

SCOTTISHPOWER RENEWABLES

Rev	Date	By	Comment
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1:7,000
Scale @ A3

0 200 400 Metres

East Anglia ONE North

Access Status for Water Bodies

Drg No	EA1N-DEV-DRG-IBR-000373
Rev	1
Date	21/11/18
Figure	22.2.1d

Coordinate System: BNG Datum: OSGB36

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N

Legend

- Onshore Study Area
- East Anglia ONE North Proposed Onshore Development Area
- East Anglia ONE North Indicative Onshore Development Area
- East Anglia ONE North Indicative Onshore Development Area 250m Buffer
- National Grid Substation
- East Anglia ONE North Onshore Substation

Water Bodies

- Access Agreed
- Access Not Granted

SCOTTISHPOWER RENEWABLES

Rev	Date	By	Comment
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1:7,000 Scale @ A3

0 200 400 Metres

East Anglia ONE North

Access Status for Water Bodies

Drg No	EA1N-DEV-DRG-IBR-000373
Rev	1
Date	21/11/18
Figure	22.2.1e

Coordinate System: BNG
Datum: OSGB36

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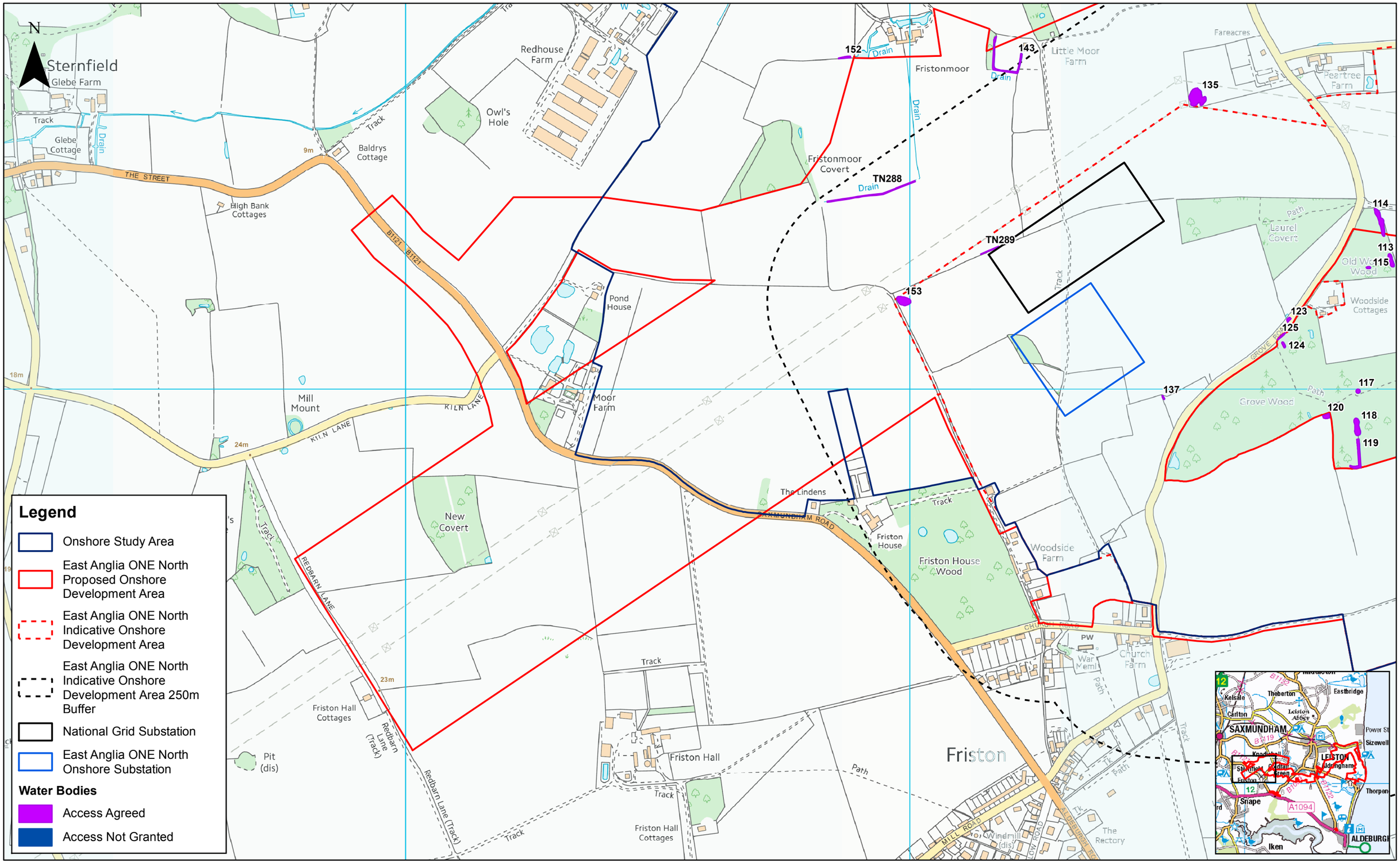

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Rev	**Date**	**By**	**Comment**		Approved:	AH			

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Rev	1
Date	21/11/18
Figure	22.2.1f

Coordinate System: BNG Datum: OSGB36

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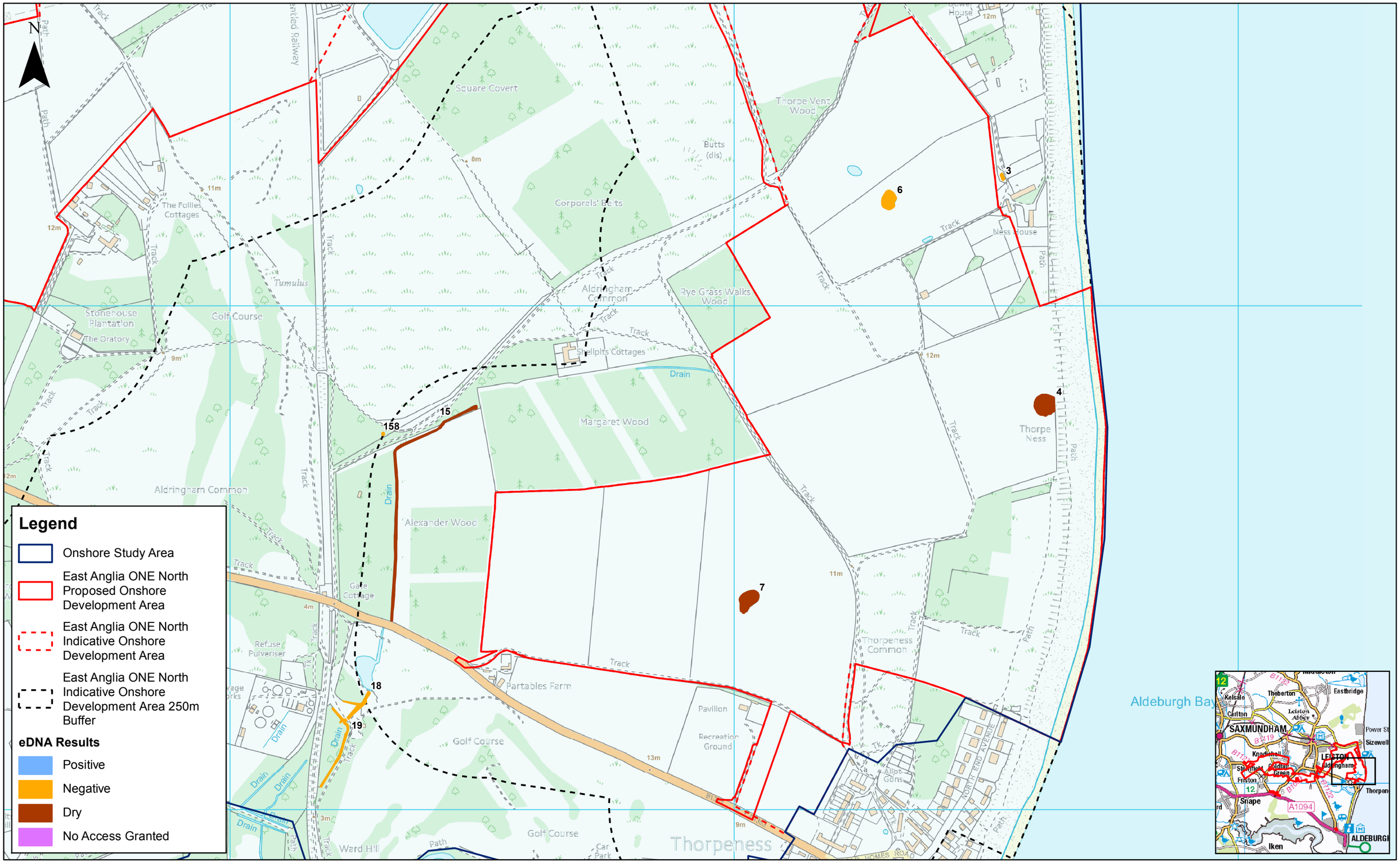

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Scale @ A3

0 200 400 Metres

East Anglia ONE North

eDNA Survey Results

Drg No	EA1N-DEV-DRG-IBR-000374
Rev	1
Date	21/11/18
Figure	22.2.2a

Coordinate System: BNG
Datum: OSGB36

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N

Legend

- Onshore Study Area
- East Anglia ONE North Proposed Onshore Development Area
- East Anglia ONE North Indicative Onshore Development Area
- East Anglia ONE North Indicative Onshore Development Area 250m Buffer

eDNA Results

- Positive
- Negative
- Dry
- No Access Granted

NP1

9

2

SCOTTISHPOWER RENEWABLES

Rev	Date	By	Comment		
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Metres 0 200 400

East Anglia ONE North

eDNA Survey Results

Drg No	EA1N-DEV-DRG-IBR-000374
Rev	1
Date	21/11/18
Figure	22.2.2b

Coordinate System: BNG Datum: OSGB36

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N

NP3

73

NP2

Legend

- Onshore Study Area
- East Anglia ONE North Proposed Onshore Development Area
- East Anglia ONE North Indicative Onshore Development Area
- East Anglia ONE North Indicative Onshore Development Area 250m Buffer

eDNA Results

- Positive
- Negative
- Dry
- No Access Granted

				Prepared:	FC	1:7,000 Scale @ A3
1	21/11/2018	FC	First Issue.	Checked:	CS	Metres 0 200 400
Rev	Date	By	Comment	Approved:	AH	

East Anglia ONE North

eDNA Survey Results

Drg No	EA1N-DEV-DRG-IBR-000374
Rev	1
Date	21/11/18
Figure	22.2.2c

Coordinate System: BNG Datum: OSGB36

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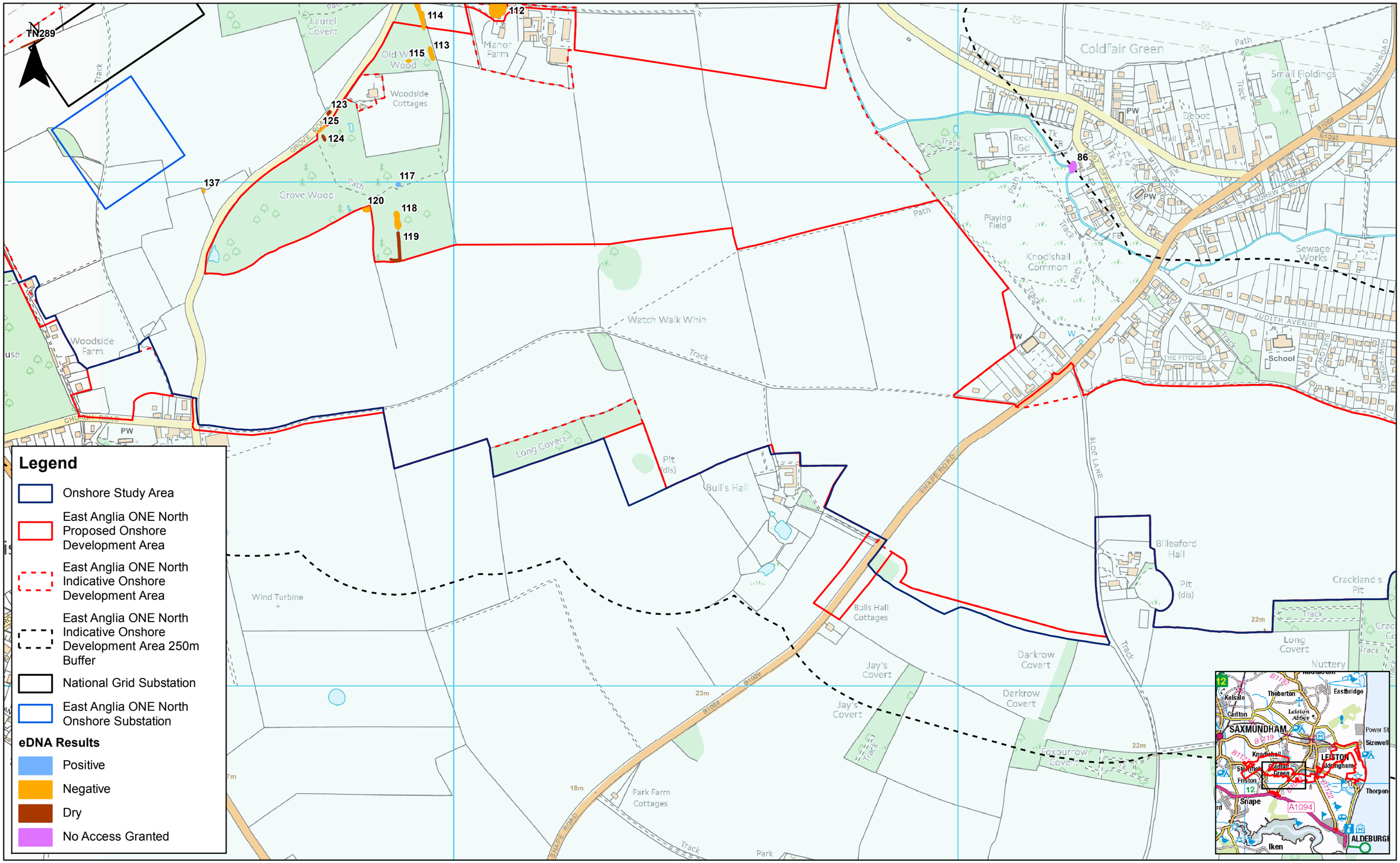

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1:7,000 Scale @ A3

0 200 400 Metres

East Anglia ONE North

eDNA Survey Results

Drg No	EA1N-DEV-DRG-IBR-000374
Rev	1
Date	21/11/18
Figure	22.2.2d

Coordinate System: BNG Datum: OSGB36

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N

Legend

- Onshore Study Area
- East Anglia ONE North Proposed Onshore Development Area
- East Anglia ONE North Indicative Onshore Development Area
- East Anglia ONE North Indicative Onshore Development Area 250m Buffer
- National Grid Substation
- East Anglia ONE North Onshore Substation

eDNA Results

- Positive
- Negative
- Dry
- No Access Granted

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0 200 400 Metres

East Anglia ONE North

eDNA Survey Results

Drg No	EA1N-DEV-DRG-IBR-000374
Rev	1
Date	21/11/18
Figure	22.2.2e

Coordinate System: BNG Datum: OSGB36

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N

Sternfield

Glebe Farm

Track

Glebe Cottage

Drain

THE STREET

High Bank Cottages

Baldrys Cottage

9m

Track

Redhouse Farm

Owl's Hole

B1121

B1121

Pond House

Moor Farm

Mill Mount

18m

KILN LANE

KILN LANE

24m

New Covert

Track

REDBARN LANE

23m

Friston Hall Cottages

Redbarn Lane (Track)

Pit (dis)

Redbarn Lane (Track)

Track

Friston Hall

Track

Friston Hall Cottages

Path

Track

The Lindens

SAXMUNDHAM ROAD

Friston House

Friston House Wood

Track

Woodside Farm

CHURCH ROAD

PW War Mem

Church Farm

Path

Track

Friston

Path

MILL ROAD

LOW ROAD

Windmill (dis)

ALDEBURGH RD

The Rectory

Fristonmoor

Drain

Drain

Drain

Fristonmoor Covert

Drain

Track

Little Moor Farm

Fareacres

Pearetree Farm

Path

Laurel Covert

Old Wood

Woodside Cottages

GROVE RD

Path

Grove Wood

Path

152

143

135

TN288

TN289

153

114

113

115

123

125

124

137

117

120

118

119

Legend

- Onshore Study Area
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eDNA Results

- Positive
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- Dry
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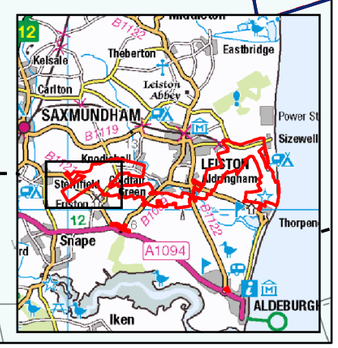

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1:7,000 Scale @ A3

0 200 400 Metres

East Anglia ONE North

eDNA Survey Results

Drg No	EA1N-DEV-DRG-IBR-000374
Rev	1
Date	21/11/18
Figure	22.2.2f

Coordinate System: BNG Datum: OSGB36

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