



WHITESTONE
solar farm

WHITESTONE SOLAR FARM

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ENVIRONMENTAL STATEMENT

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Prepared by:

ERM

Prepared for:

Whitestone Net Zero Ltd

Contents

6.13 Fisheries and White-clawed Crayfish eDNA Report.....	1
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Tables

Table 6.13.1 Aquatic eDNA Survey Progress as of March 2026	6
Table 6.13.2 Data Results from EA Ecology and Fish Data Explorer	7
Table 6.13.3 Species Detected in 16 Successful Samples via Metabarcoding Analysis.....	8
Table 6.13.4 Aquatic eDNA Analysis Results	10

Figures (See Appendix 6.13, Figures)

Figure Number	Figure Title
6.13.1	Aquatic eDNA Sample Locations

Annexes (See Volume A.1, Appendices)

Annex Number	Annex Title
6.13A	Legislation

Glossary

Term	Meaning
<i>Cable Corridor</i>	Corridors within which the high voltage cables would be constructed.
Development Consent Order (DCO)	A statutory order made by the relevant Secretary of State pursuant to The Planning Act 2008 to authorise a Nationally Significant Infrastructure Project which provides consent for the project and means that a range of other consents, such as planning permission and listed building consent, will not be required. A DCO can also include rights of compulsory acquisition
<i>Environment Statement (ES)</i>	Environmental Statement which presents the environmental information relating to the Proposed Development. The ES has been prepared in accordance with current EIA regulation.
<i>Long Lane 400kV Substation</i>	The new 400 kilovolt National Grid substation proposed on land immediately east of Long Lane, Brinsworth, S60 4JJ
<i>Multispecies Metabarcoding Analysis</i>	Metabarcoding is a modern molecular method used to detect and characterize multiple species within one eDNA sample simultaneously, by using DNA sequencing.

ENVIRONMENTAL STATEMENT

Term	Meaning
<i>Nationally Significant Infrastructure Project (NSIP)</i>	NSIPs are large scale major development projects in England or Wales which fall into the following categories: • Energy; • Transport; • Waste; • Waste Water; and • Water. The primary legislation which applies to NSIPs is called the Planning Act 2008. When these types of development meet the threshold described in the Planning Act 2008, they need a Development Consent Order before they can be built.
<i>Order Limits</i>	Total area comprising the Site and Cable Corridor.
<i>qPCR Analysis</i>	Quantitative Polymerase Chain Reaction (qPCR), also known as real-time PCR, is a molecular biology technique used to amplify and simultaneously quantify eDNA within a sample. The high specificity of the qPCR method makes it ideal for situations where a single target is required.
<i>Study Area</i>	This is an area which is defined for fish species and white-clawed crayfish surveys which includes the Order Limits as well as potential spatial and temporal considerations of the impacts on relevant receptors.
<i>The Applicant</i>	Whitestone Net Zero Ltd.
<i>The Application</i>	The Application to be submitted to the Secretary of State for Energy Security and Net Zero for a Development Consent Order.
<i>The Proposed Development</i>	The proposed Whitestone Solar Farm.
<i>The Site</i>	The land planned to be used for solar PV array and associated infrastructure, BESS, substation, and landscaping and habitat enhancement. The Site is split into W1, W2, and W3.

Acronyms

Acronym	Meaning
<i>AIL</i>	Abnormal Indivisible Load
<i>BESS</i>	Battery Energy Storage System
<i>DBRC</i>	Derbyshire Biological Records Centre
<i>DCO</i>	Development Consent Order
<i>EA</i>	Environment Agency
<i>eDNA</i>	Environmental Deoxyribonucleic Acid
<i>ERM</i>	Environmental Resources Management Ltd
<i>IUCN</i>	International Union for Conservation of Nature
<i>LRC</i>	Local Record Centres
<i>NERC</i>	Natural Environments and Rural Communities Act
<i>NSIP</i>	Nationally Significant Infrastructure Project
<i>PCR</i>	Polymerase Chain Reaction
<i>PV</i>	Photovoltaic
<i>QC</i>	Quality Control
<i>qPCR</i>	Quantitative Polymerase Chain Reaction
<i>RBRC</i>	Rotherham Biological Records Centre
<i>SAC</i>	Special Area of Conservation
<i>SAFFA</i>	Salmon and Freshwater Fisheries Act 1975
<i>SBRC</i>	Sheffield Biological Records Centre
<i>SL</i>	Sample Location
<i>WCC</i>	White-clawed Crayfish
<i>WFD</i>	Water Framework Directive
<i>W1</i>	Whitestone 1
<i>W2</i>	Whitestone 2
<i>W3</i>	Whitestone 3

Units

Units	Meaning
<i>ha</i>	Hectare
<i>km</i>	Kilometre
<i>kV</i>	Kilovolt
<i>L</i>	Litre
<i>m</i>	Metre
<i>MW</i>	Megawatt

6.13 Fisheries and White-clawed Crayfish eDNA Report

Introduction

- 6.13.1 This Appendix has been prepared on behalf of Whitestone Net Zero Ltd ('the Applicant') to set out the methodology and results of the fisheries and white-clawed crayfish (*Austropotamobius pallipes*) Environmental Deoxyribonucleic Acid (eDNA) surveys in relation to the Development Consent Order (DCO) Application for the construction, operation, maintenance, and decommissioning of Whitestone Solar Farm (hereafter referred to as the 'Proposed Development').
- 6.13.2 This Appendix is supported by the following figures in **ES Volume 3, Figures [EN0110020/APP/6.19]**:
- **Figure 6.13.1: Aquatic eDNA Sample Locations**

The Order Limits

- 6.13.1 The extent of the Order Limits are described in **ES Volume 1, Chapter 3: The Site and Surrounding Area [EN0110020/APP/6.3]** and shown in **ES Volume 3, Figure 3.1: Order Limits [EN0110020/APP/6.19]**. The Proposed Development is described in **ES Volume 1, Chapter 5: The Proposed Development [EN0110020/APP/6.5]** and shown spatially on the **Works Plans [EN0110020/APP/2.3.1]**.

The Proposed Development

- 6.13.2 The Proposed Development involves the construction, operation and maintenance, and decommissioning of more than 100MW of solar photovoltaic (PV) array, Battery Energy Storage System (BESS), onsite substations and supporting infrastructure, and grid connection infrastructure. The grid connection infrastructure would connect the Proposed Development to the new National Grid substation at Brinsworth (Long Lane 400kV Substation).
- 6.13.3 As the Proposed Development would have a generating capacity in excess of 100MW, it is considered to be a Nationally Significant Infrastructure Project (NSIP) under the Planning Act 2008.
- 6.13.4 The Proposed Development would be located within the Order Limits. The Order Limits encompass the total area of the Proposed Development comprising the Site and Cable Corridors. The Site is specifically the land that is planned to be used for solar PV array and associated infrastructure, BESS, substations, landscaping and habitat enhancement. The Site is split into Whitestone 1 (W1), Whitestone 2 (W2) and Whitestone 3 (W3).
- 6.13.5 Highway Works are sections of the highway network that will contain localised improvements, such as improvements to road edge where it is deteriorated, or temporary highway and traffic works required to safely accommodate the Abnormal Indivisible Load (AIL) deliveries. These areas will support the movement of construction vehicles on narrower sections of the local highway network within parts of the construction vehicle routes to the Site, see **ES Volume 2, Chapter 13: Traffic and Transport [EN0110020/APP/6.13.1]**.

Legislation and Guidance

White-clawed Crayfish

- 6.13.6 White-clawed crayfish (WCC) are protected under the Wildlife and Countryside Act 1981 (as amended)¹ and are also included as a species of principal importance in England under section 41 of the Natural Environment and Rural Communities Act (2006)² meaning that they must be considered as part of planning decisions. WCC are listed as Endangered on the International Union for Conservation of Nature (IUCN) Red List due to the rapid decline in numbers found in recent decades³.
- 6.13.7 Developers therefore must comply with the legal protection of WCC and may need a license for any activity that has the potential to impact WCC. Developers may also need permission from the Environment Agency (EA) to trap WCC as part of their development proposal.
- 6.13.8 It is therefore important to know if WCC are present within the Order Limits, and to what extent they are likely to be impacted by the Proposed Development, so that suitable avoidance and mitigation measures can be implemented.

Fish species

- 6.13.9 Several fish species, including European eel (*Anguilla Anguilla*), Atlantic salmon (*Salmo salar*) and Brown trout (*Salmo trutta*) are afforded legal protection in the UK, summarised below.
- 6.13.10 It is therefore important to know what species of fish are present within the Order Limits and to what extent they are likely to be impacted by the Proposed Development, so that suitable avoidance and mitigation measures can be implemented.
- 6.13.11 Relevant legislation includes:
- Salmon and Freshwater Fisheries Act (SAFFA) 1975⁴
 - Wildlife and Countryside Act 1981 (as amended)¹
 - Conservation of Habitats and Species Regulations 2017(as amended) which includes Annex II of the Habitats Directive, transposed into UK Law⁵; and
 - The Eels Regulations 2009⁶.
- 6.13.12 All relevant legislation and species afforded legal protection are detailed in **Annex 6.13A**.

Survey Methods

- 6.13.13 A desk study and subsequent eDNA surveys were completed based on the Proposed Development boundary at the time, between July and December 2025. Both the desk study and surveys included the Site and Cable Corridor. The Order Limits have since changed and been refined for the Application; however, results are presented here in full based on the study area used, known from here as the 'eDNA Study Area'.

Desk Study

- 6.13.14 The EAs Ecology and Fish Data Explorer⁷ plus Local Record Centres (LRC) which the eDNA Study Area falls within were consulted in July 2025 for any records of protected or otherwise notable aquatic species within the last 10 years within 1km of the eDNA Study Area. The following LRCs were contacted:
- Derbyshire Biological Records Centre (DBRC)
 - Rotherham Biological Records Centre (RBRC); and
 - Sheffield Biological Records Centre (SBRC).
- 6.13.15 Any records returned older than 10 years old are considered historic and are not indicative of current populations within watercourses and have not been considered within this Appendix.

Sample Site Selection

- 6.13.16 A total of 29 sample sites were identified through the initial desk study with one further sample site being added in December 2025 following identification of a further hydrologically connected watercourse, bringing the total number of proposed sample locations within the eDNA Study Area to 30. Locations of all proposed sample sites can be seen in **ES Volume 3, Figure 6.13.1: Aquatic eDNA Sample Locations [EN0110020/APP/6.19]**.
- 6.13.17 Sites were chosen based on the following criteria
- Watercourses that the Cable Corridors would cross
 - Watercourses defined within the Water Framework Directive (WFD⁸)
 - Watercourse within 10m of the eDNA Study Area; and
 - Watercourses with connectivity to known species records (identified via desk study).
- 6.13.18 Experts at Nature Metrics were consulted and proposed sample sites were discussed prior to surveys being undertaken in order to determine the best method to survey for target species. NatureMetrics are a nature intelligence company specialising in eDNA analysis, in this instance through water sampling. It was decided with NatureMetrics that all sample locations would be assessed for fisheries species (including European Eel, sampled separately to the general suite for fisheries species). European eel are recommended by NatureMetrics to be sampled separately due to potential for interference from other species within the general fish suite.
- 6.13.19 Sample location (SL) 29, situated on the south edge of W3, was deemed to be at no risk from impacts during construction and operation of the Proposed Development and subsequently excluded from survey. The Applicant confirmed design changes in 2025 to allow the exclusion of SL29.
- 6.13.20 Five sample locations, at Broadbridge Dyke (SL25, SL28), Chesterfield Canal (SL26, SL30) and a final site Unnamed (SL27) were selected for WCC eDNA. Sample locations were chosen where there was potential connectivity to known species records, or suitable habitat was observed. All other sample locations were scoped out for WCC eDNA analysis as they were outside the known range⁹.

Environmental DNA (eDNA) Sampling Protocol

6.13.21 Aquatic eDNA sampling kits were purchased from Nature Metrics. A brief description of the sampling protocol is outlined below. Detailed sampling instructions can be found on the Nature Metrics website¹⁰.

6.13.22 Sampling protocol:

- Surveyors put on sterile nitrile gloves
- three litres of watercourse water was collected in sampling bag, avoiding sediment
- A Peristaltic pump was used to pump water through the eDNA filter until all three litres had passed through the filter or until the filter became clogged
- eDNA filter with any excess water was expelled and was then filled with preservation buffer solution and sealed
- Filter was placed into sample bag and sample manifest information sheet completed
- Sample was stored in a cool dry place, out of direct sunlight
- Collection by a courier was organised as soon as possible after sampling, ideally within 24 hours; and
- Sample was provided to Nature Metrics Lab within two weeks of sampling. If this was not possible then the sample was frozen as soon as possible after sampling to preserve eDNA and shipped when feasible.

Limitations

6.13.23 Two samples (SL8 and SL28) were lost in transit by couriers while being delivered to the Nature Metrics lab for analysis. However, another sample was taken approximately 1.4km downstream from SL28 at SL25 and so results for SL28 can be tentatively assumed from this. SL8 is associated with a Cable Corridor that is no longer under consideration, so there will be no impacts on the watercourse at this location. Due to this, the loss of both samples is considered only a minor limitation.

6.13.24 Four watercourses that were visited by surveyors were dry at the time of initial visits in 2025. However, it can be assumed that these watercourses are likely ephemeral dry ditches and only occasionally wet, making them unsuitable habitat and unlikely to contain the target species and migratory fish when wet. Therefore, being unable to collect an eDNA sample is considered only a minor limitation.

6.13.25 Land access constraints prevented surveyors from visiting seven of the sample locations. However, three of these sample locations are situated within proposed Cable Corridors that are no longer under consideration and do not need to be considered further. A lack of data from the remaining four watercourses means it should be assumed that target species could be present, and a precautionary approach to mitigation will be required. Therefore, this data constraint is considered only a minor limitation.

6.13.26 Based on the extensive survey work undertaken, it is considered that sufficient baseline has been recorded to base the assessment on and inform the design. Nonetheless, and in order to ensure readiness for implementation should development consent be granted, further survey work is ongoing to validate the

findings of this assessment. The Applicant will provide the Examining Authority with any outputs as part of that workstream (ensuring that sufficient time is provided for interested parties to comment on that material), but its approach in the ES has ensured that a reasonable worst-case scenario has been assessed through the use of public information, assumptions, and embedded mitigation (including requirements relating to avoidance measures, and requirements for pre-construction surveys which will inform post-consent activities). The Applicant is confident that the approach above meets the requirements of the Infrastructure Planning (Environmental Impact Assessment) Regulations 2017 in providing information reasonably required to reach a reasoned conclusion based on a reasonable worst case scenario, is consistent with established precedent and practice of accepted DCO applications, and relevant case law (in particular, *R (Jones) v Mansfield District Council* [2003] EWCA Civ 1408 at [39], and *National Trusts' Application* [2013] NIQB 60 at [70]).

- 6.13.27 Three samples failed quality control testing for European eel Quantitative Polymerase Chain Reaction (qPCR) analysis and subsequently gave inconclusive results. In the case of inconclusive results, it is assumed that target species are present and a precautionary approach to applying mitigation measures will be taken. Therefore, this is considered only a minor limitation.
- 6.13.28 Some detected eDNA was only identified down to genus level in the fisheries metabarcoding analysis. However, based on known ranges of species in England and Wales it is possible to make reasonable assumptions about the likely species the detected eDNA came from. Therefore, this considered only a minor limitation.
- 6.13.29 Seven of the 16 samples sent for fisheries metabarcoding analysis gave a 'no amplifiable DNA' result. This likely indicates no eDNA was collected in the sample, and that target species are not present in the sampled watercourses. Although less likely, this result could also be attributed to human errors in the sampling protocol, or the degradation of eDNA in the sample during transit and storage. It is not possible to know for certain the cause of this result, and so no conclusions can be drawn from it in isolation. Other aquatic assessment results can be considered in combination with these results to ascertain likelihood of these watercourses being of sufficient water quality to be able to support fisheries. For example, SL26 was positive for WCC, which is a species sensitive to pollution and requires good water quality and reliable water flow, so is a likely indicator of fisheries presence. A precautionary approach and professional judgement will be applied to reduce this limitation.

Survey Progress

6.13.30 At the time of writing, 18 of the 29 proposed sites have been sampled and results had been returned for 16 of these. Two samples were lost in transit by couriers; therefore, no results can be provided for those locations.

6.13.31 Four sample sites were identified as dry ditches during initial visits by surveyors and so water samples in those locations could not be taken. A further seven sample locations could not be accessed due to land access constraints. Watercourses which have not been surveyed at the time of writing will be considered in combination with other aquatic assessment results based on professional judgement and discussed further in **ES Volume 2, Chapter 6: Biodiversity and Nature Conservation [EN0110020/APP/6.6.1]**.

6.13.32 Survey progress is summarised in **Table 6.13.1** below.

Table 6.13.1 Aquatic eDNA Survey Progress as of March 2026

Solar Array / Cable Corridor	Area	Preferr ed Cable Corrido r?	Sampl e Locati on	Status
Solar Array	W1	N/A	1	Sampled 10/09/2025
Solar Array	W1	N/A	2	Sampled 10/09/2025
Cable Corridor	Non-preferred Cable Corridor	No	3	Not sampled – land access constraints
Cable Corridor	CRB	Yes	4	Sampled 16/12/2025
Cable Corridor	Non-preferred Cable Corridor	No	5	Not sampled – land access constraints
Cable Corridor	CRB	Yes	6	Not sampled – land access constraints
Cable Corridor	CRB	Yes	7	Not sampled – land access constraints
Cable Corridor	Non-preferred Cable Corridor	No	8	Sampled 18/09/2025 – sample lost by courier
Cable Corridor	Non-preferred Cable Corridor	No	9	Not sampled – land access constraints
Solar Array	W2	N/A	10	Not sampled -dry at time of visit 15/12/2025
Cable Corridor	CRC	Yes	11	Not sampled –dry at time of visit 15/12/2025
Cable Corridor	CRC	Yes	12	Sampled 15/12/2025
Cable Corridor	CRC	Yes	13	Sampled 13/11/2025
Cable Corridor	CRD-1	Yes	14	Not sampled – land access constraints
Cable Corridor	CRD-1	Yes	15	Not sampled – dry at time of visit 08/09/2025
Solar Array	W2	N/A	16	Sampled 09/09/2025
Solar Array	W2	N/A	17	Sampled 09/09/2025
Solar Array	W2	N/A	18	Sampled 09/09/2025

Solar Array / Cable Corridor	Area	Preferr ed Cable Corridor?	Sampl e Locati on	Status
Solar Array	W2	N/A	19	Not sampled – land access constraints
Solar Array	W2	N/A	20	Sampled 09/09/2025
Solar Array	W2	N/A	21	Sampled 09/09/2025
Solar Array	W2	N/A	22	Sampled 09/09/2025
Solar Array	W2	N/A	23	Sampled 09/09/2025
Solar Array	W2	N/A	24	Sampled 09/09/2025
Cable Corridor	Non-preferred cable route	No	25	Sampled 08/09/2025
Cable Corridor	Non-preferred cable route	No	26	Sampled 08/09/2025
Cable Corridor	CRL	Yes	27	Not sampled – dry at time of visit 08/09/2025
Solar Array	W3	N/A	28	Sampled 18/09/2025 – sample lost by courier
Solar Array	W3	N/A	29	Not sampled – location scoped out
Cable Corridor	CRL	Yes	30	Sampled 17/12/2025

Survey Findings

Desk Study

6.13.33 The EA Ecology and Fish Data explorer returned data from the search within 1km of the eDNA Study Area at one site, with notable species and count provided below in **Table 6.13.2**.

Table 6.13.2 Data Results from EA Ecology and Fish Data Explorer

Waterbody (Site Name)	Distance from eDNA Study Area Boundary	Species Recorded (Count)	Date
Ulley Brook (Downstream Ulley Reservoir (A618))	40m	Bullhead (4)	2017

6.13.34 Three LRC data requests were made in July 2024 with a 1km buffer around the eDNA Study Area. No notable species were recorded within the last ten years within a 1km buffer of the eDNA Study Area.

European Eel eDNA

6.13.35 All 16 successful samples were tested for European Eel. Three samples – Anston Brook (SL21), Kearsley Brook (SL2) an Pinchmill Brook (SL12) returned inconclusive results, and the remaining 13 returned negative results. The inconclusive results were returned as they failed NatureMetrics internal inhibition tests. The eDNA was prevented from amplifying by substances already in the water – in these instances this is likely sourced from agricultural run-off – sediment or organic compounds – from surrounding fields. It is possible that the

environmental inhibitors i.e. run off was too high for the reaction to bypass. In the three inconclusive results, it should be assumed that in the absence of a negative result, European eel may be present in these watercourses.

White-clawed Crayfish eDNA

- 6.13.36 Of the five sample sites which were deemed suitable for WCC, three were tested with two being lost in transit. Results on Broadbridge Dyke (SL25) and Chesterfield Canal (SL26) both returned positive results, with a second site on Chesterfield Canal (SL30) returning a negative result. The remaining 11 sample sites were not tested for WCC presence.

Fisheries eDNA (excluding European Eel)

- 6.13.37 Across all 16 successful samples a total of at least 18 target species and at least a further 20 non-target species were detected, these species are listed below in **Table 6.13.3**. Results of the eDNA analysis at each sample location is detailed in **Table 6.13.4**.

Table 6.13.3 Species Detected in 16 Successful Samples via Metabarcoding Analysis

Common Name	Binomial Name
Target Species	
Common Bream	<i>Abramis brama</i>
White Bream	<i>Blicca bjoerkna</i>
Freshwater Sculpin spss., likely European Bullhead	<i>Cottus spss.</i>
Typical Carp spss., likely Common Carp	<i>Cyprinus spss.</i>
European Seabass	<i>Dicentrarchus labrax</i>
Northern Pike	<i>Esox lucius</i>
Three-Spined Stickleback	<i>Gasterosteus aculeatus</i>
Eurasian Ruffe	<i>Gymnocephalus cernua</i>
Eurasian Dace spss., likely Common Dace	<i>Leuciscus spss.</i>
Pacific Salmon/Trout spss., likely Rainbow Trout or Pink Salmon	<i>Oncorhynchus spss.</i>
European Perch	<i>Perca fluviatilis</i>
Eurasian Minnow	<i>Phoxinus phoxinus</i>
Ninespine Stickleback	<i>Pungitius pungitius</i>
Roach	<i>Rutilus rutilus</i>
Atlantic Salmon	<i>Salmo salar</i>
Gilt-head Seabream	<i>Sparus aurata</i>
Common Chub	<i>Squalius cephalus</i>
Tench	<i>Tinca tinca</i>
Non-Target Species	
Dabbling duck spss., likely Mallard	<i>Anas spss.</i>
Tit (bird) spss.	<i>Paridae spss.</i>
Common Chiffchaff	<i>Phylloscopus collybita</i>
Common Starling	<i>Sturnus vulgaris</i>
Wren (bird) spss.	<i>Troglodytes spss.</i>
Common Blackbird	<i>Turdus merula</i>

Common Name	Binomial Name
Song Thrush	<i>Turdus philomelos</i>
Wood Mouse	<i>Apodemus sylvaticus</i>
Roe Deer	<i>Capreolus capreolus</i>
European Hare	<i>Lepus europaeus</i>
European Badger	<i>Meles meles</i>
Vole spss.	<i>Microtus spss.</i>
Chinese Muntjac (INNS)	<i>Muntiacus reevesi</i>
American Mink (INNS)	<i>Mustela vison</i>
Eurasian Water Shrew	<i>Neomys fodiens</i>
European Rabbit	<i>Oryctolagus cuniculus</i>
Brown Rat (INNS)	<i>Rattus norvegicus</i>
Grey Squirrel (INNS)	<i>Sciurus carolinensis</i>
Red Fox	<i>Vulpes vulpes</i>
Smooth Newt	<i>Lissotriton vulgaris</i>

eDNA Species Interpretation

- 6.13.38 Four of the target species, including *Cottus spp.*, *Cyprinus spp.*, *Orcorhynchus spp.*, and *Leuciscus spp.*, were identified to Genus level only. In these cases, it is possible to make reasonable assumptions as to the species identified from eDNA analysis, based upon known ranges of fish species within these Genera¹¹.
- 6.13.39 The European bullhead (*Cottus gobio*) is the only member of *Cottus* native to England and Wales, making it the likely source of the detected eDNA for *Cottus sp.*
- 6.13.40 Additionally, common carp (*Cyprinus carpio*) is the only member of *Cyprinus* native to England and Wales, making it the likely source of the detected eDNA.
- 6.13.41 There are 17 species within the Genus *Orcorhynchus*, however none of these are native in England and Wales. Rainbow trout (*Oncorhynchus mykiss*) is a non-native species, introduced to farms in the UK in the early 20th century and has now become established in rivers and lakes throughout England and Wales¹² although it is most commonly found as a stocked fish in commercial fishing ponds. It is the likely source of the detected eDNA for this Genus. Pacific pink salmon (*Oncorhynchus gorbuscha*) is another non-native species known to be present in English rivers primarily in the Northeast¹³. Although much less common than the rainbow trout in English rivers, it is still a potential source of the detected eDNA.
- 6.13.42 The Common dace (*Leuciscus leuciscus*) is the only member of *Leuciscus* genus native to England and Wales, and is widespread in rivers throughout, making it the likely source of the detected eDNA.

Table 6.13.4 Aquatic eDNA Analysis Results

Area	Sample Location	Sample Date	Kit ID	European Eel	White-clawed Crayfish	Fisheries Summary	Non-Target Species	Comments on Results
W1	1	10 September 2025	PSI-01-06269	Negative	Not Tested	No amplifiable DNA	No amplifiable DNA	None
W1	2	10 September 2025	PSI-01-06254	Inconclusive	Not Tested	No amplifiable DNA	No amplifiable DNA	As the result is inconclusive for eels, it is assumed they are present. Therefore SAFFA 1975 and Eels Regs 2009 would apply.
CRB	4	16 December 2025	PSI-01-06280	Negative	Not Tested	Target species present: • European Seabass • Gilt-head Seabream • Atlantic Salmon	Non-target species present: • Smooth Newt; • Common Starling; • Common Blackbird; • Song Thrush; • Roe Deer; • Chinese Muntjac; • Red Fox; • European Badger; • American Mink; • European Hare; • European Rabbit; • Wood Mouse; • Brown Rat; • Grey Squirrel; • Duck spss; • Wren spss; and • Vole spss.	The fisheries assessment at this site was likely as the result of introduced DNA from bird sources or campers leaving behind food sources. SAFFA 1975 and EELS 2009 Regs would not apply here for European Seabass or Gilt-headed Seabream. Although Atlantic salmon could be similarly introduced via the methods mentioned above, as it is a protected species in the UK, SAFFA1975 and EELS regs 2009 should apply. Terrestrial protected and notable species including European badger and hare species were recorded. Invasive species including American mink have been recorded, a negative indicator for water vole presence. Brown rat also a negative indicator.
Non-preferred route	8	18 September 2025	PSI-01-06255	Sample Lost	Sample Lost	Sample Lost	Sample Lost	As the sample was lost, it is assumed protected and/or priority species may be present and that SAFFA 1975 and Eels Regs 2009 would apply.
CRC	12	15 December 2025	PSI-01-06268	Inconclusive	Not Tested	No amplifiable DNA	No amplifiable DNA	None
CRC	13	13 November 2025	PSI-01-06258	Negative	Not Tested	No amplifiable DNA	No amplifiable DNA	None

ENVIRONMENTAL STATEMENT

Area	Sample Location	Sample Date	Kit ID	European Eel	White-clawed Crayfish	Fisheries Summary	Non-Target Species	Comments on Results
W2	16	09 September 2025	PSI-01-06261	Negative	Not Tested	Target species present: • White Bream; • Eurasian Minnow; • Roach; • Common Chub; • Three-spined; • Stickleback; • Nine spined; • Stickleback; • European Perch; and • Freshwater sculpin (Bullhead).	None	Water quality is of sufficient quality to support fisheries. White bream may have been introduced at this location, as it is not in its usual range. Freshwater sculpins (i.e. bullhead species) present. These are a priority species. Provision of general fisheries habitat protection under SAFFA 1975 apply.
W2	17	09 September 2025	PSI-01-06273	Negative	Not Tested	Target species present: • Freshwater Sculpin (Bullhead).	Non-target species present: • Common Blackbird	Yes, freshwater sculpins (i.e. bullhead species) present. These are priority species. Provision of general fisheries habitat protection under SAFFA 1975
W2	18	09 September 2025	PSI-01-06264	Negative	Not Tested	No amplifiable DNA	No amplifiable DNA	None
W2	20	09 September 2025	PSI-01-06260	Negative	Not Tested	Sample passed all Quality Control (QC), no target species reported	Non-target species present: • Tit spp; • Common Chiffchaff; • Common Blackbird; • Roe Deer; • European Hare; • Vole spp; • Brown Rat; and • Grey Squirrel.	Not identified as supporting fisheries. European hare, a terrestrial priority species was identified in this location.
W2	21	09 September 2025	PSI-01-06272	Inconclusive	Not Tested	No amplifiable DNA	No amplifiable DNA	As the result was inconclusive for eels, as a precautionary principle, it is assumed that protected or priority species may be present and that SAFFA 1975 and Eels Regs 2009 apply.
W2	22	09 September 2025	PSI-01-06274	Negative	Not Tested	Target species present: • Three-spined; • Stickleback; and • Freshwater Sculpin (bullhead).	Non-target species present: • Duck spp; • Brown Rat; and • Eurasian Water Shrew	Bullhead is present, which is a priority species. Presence of brown rat is a negative indicator for water vole presence. Water shrew is an indicator of clean waters and is protected from trapping under Schedule 6 of the WCA 1981. Provision of general fisheries habitat protection under SAFFA 1975.

ENVIRONMENTAL STATEMENT

Area	Sample Location	Sample Date	Kit ID	European Eel	White-clawed Crayfish	Fisheries Summary	Non-Target Species	Comments on Results
W2	23	09 September 2025	PSI-01-06265	Negative	Not Tested	Target species present: • Roach; • Three-spined; • Stickleback; and • Freshwater Sculpin (bullhead).	Non-target species present: • Wren spss; • Common Blackbird; • Roe Deer; and • Brown Rat	Bullhead is present, which is a priority species. Provision of general fisheries habitat protection under SAFFA 1975. Presence of brown rat is a negative indicator for water vole.
W2	24	09 September 2025	PSI-01-06275	Negative	Not Tested	Target species present: • Roach; • Three-spined; • Stickleback; and • Freshwater Sculpin (bullhead).	None	Bullhead is present, which is a priority species.. Provision of general fisheries habitat protection under SAFFA 1975
Non-preferred route	25	08 September 2025	PSI-01-06276	Negative	Positive	Target species present: • Three-spined; • Stickleback; and • European Perch	None	White-clawed crayfish present, which is a protected species under Schedule 5, WCA 1981. Provision of general fisheries habitat protection and navigation under SAFFA 1975. Mitigation or licensing may apply where there are works in the locality.
Non-preferred route	26	08 September 2025	PSI-01-06270	Negative	Positive	No amplifiable DNA	No amplifiable DNA	White-clawed crayfish present, which is a protected species under Schedule 5, WCA 1981. Unclear whether other fisheries species are present, however WCC need clean water to be present, so it should be assumed in the absence of other data that other freshwater fish are also present and that the provision of general fisheries habitat protection and navigation under SAFFA 1975 applies. Mitigation or licensing may apply where there are works in the locality.
W3	28	18 September 2025	PSI-01-06271	Sample Lost	Sample Lost	Sample Lost	Sample Lost	As sample lost, assume protected or priority species may be present and that SAFFA 1975 and Eels Regs 2009 would apply.
CRL	30	17 December 2025	PSI-01-06256	Negative	Negative	Target species present: • Common Bream; • <i>Cyprinus</i> spp. (likely common carp); • <i>Orcorhynchus</i> spp. (likely Rainbow Trout or Pink Salmon);	Non-target species present: • Common Blackbird • Brown Rat	Presence of brown rat is a negative indicator for water vole. Fish associated with clean waters present. Provision of general fisheries habitat protection and navigation under SAFFA 1975 applies.

ENVIRONMENTAL STATEMENT

Area	Sample Location	Sample Date	Kit ID	European Eel	White-clawed Crayfish	Fisheries Summary	Non-Target Species	Comments on Results
						• <i>Leuciscus</i> spp. (likely Eurasian Dace); • Tench; • Northern Pike; • Eurasian Ruffe; • Roach; • Three-spined Stickleback; and • European Perch		

Format and Interpretation of Reporting Results

White-clawed Crayfish & European Eel

6.13.43 WCC and European Eel eDNA was tested for via single species qPCR analysis. This methodology can yield the following results:

- Positive: target eDNA was detected;
- Negative: sample passed QC tests and target eDNA was **not** detected; and
- Inconclusive: Target eDNA was **not** detected, however sample failed QC tests meaning this could be a false negative.

Fish Species

6.13.44 Fish species eDNA in water samples was tested for via multi-species metabarcoding analysis. This methodology allows for eDNA from multiple species to be detected simultaneously via a single testing protocol and can yield the following results¹⁴:

- No amplifiable DNA - DNA sequence amplification was not successful. This is due to a low concentration of DNA, or Polymerase Chain Reaction (PCR) inhibition. Low concentration of DNA usually occurs because there are no target species in the area. However, no amplifiable DNA can also result from a small volume of water filtered or DNA degradation within the sample. PCR inhibition occurs because substances were present that limit PCR. Target groups that are rare, or at very low abundance, will often not be detected
- Data QC step not passed - DNA sequence amplification was successful, but the number of DNA sequences for target DNA was below Nature Metrics' minimum reporting thresholds. Samples with low numbers of DNA sequences are removed as they are not considered representative of the sampling location. The metabarcoding technology does not guarantee that data is generated from 100% of samples, and a low percentage of samples do not generate data. In cases where a sample does not yield robust data, it is not always possible to pinpoint exact causes
- Target species detected: sample passed all QC tests and target eDNA was detected; and
- Target species not detected: sample passed all QC tests and target eDNA was not detected

Summary and Conclusions

- 6.13.45 The desk study results provided records of European Bullhead in 2023 in Ulley Brook within 100m of the Order Limits and in 2017. No other species were reported within 1km of the Order Limits from the consulted LRCs and the EA Ecology and Fish data explorer within the last ten years.
- 6.13.46 WCC survey results are indicative of a population in and around W3, with positive eDNA results for the species within both Broadbridge Dike and the Chesterfield Canal.
- 6.13.47 Thirteen out of 16 negative results suggest that European eel is likely absent from most of the watercourses within the Order Limits. As the remaining three samples were inconclusive, it is necessary to assume the presence of European eel in these localities – Kearsley Brook, Pinchmill Brook and Anston Brook – and to ensure compliance with the requirements of the Eels (England and Wales) Regulations 2009.
- 6.13.48 Multispecies barcoding analysis confirms the presence of freshwater fish species across the Order Limits. In instances where results are inconclusive—such as when no amplifiable eDNA is detected—it is necessary to assume target species are present.
- 6.13.49 Within the Proposed Development, two species protected under Annex II of the Habitats Directive have been identified. Atlantic Salmon, an IUCN Endangered species in Great Britain was detected at SL4 – Firsby Brook along the Cable Corridor. Although this DNA could have been introduced or erroneous as marine species were recorded at this site, it is necessary to assume the presence and to ensure compliance with the SAFFA 1975 regulations. *Cottus* species (likely European Bullhead) were identified at Ulley Brook (SL16, SL17), Cramfit Brook (SL22) and Anston Brook (SL23, SL24). Targeted mitigation strategies will be implemented for construction activities that could affect watercourses where Annex II species are confirmed or considered likely to be present. As none of these watercourses are designated Special Area of Conservation (SACs), both species are considered within **ES Volume 2, Chapter 6: Biodiversity and Nature Conservation [EN0110020/APP/6.6.1]** as a priority species at policy level.
- 6.13.50 Where samples were lost or not undertaken at the time of writing this report, watercourses will be considered in combination with other aquatic assessment results based on professional judgement, discussed further in **ES Volume 2, Chapter 6: Biodiversity and Nature Conservation [EN0110020/APP/6.6.1]**.
- 6.13.51 Appropriate mitigation measures will be implemented for any construction activities that could potentially affect watercourses where White-clawed crayfish, European eel or notable fisheries species may be present in line with appropriate legislation. Further details are provided in **ES Volume 2, Chapter 6: Biodiversity and Nature Conservation [EN0110020/APP/6.6.1]**.

Annex 6.13A Legislation

Salmon and Freshwater Fisheries Act (SAFFA) 1975

- 6.13.52 SAFFA protects freshwater fish, with a strong focus on salmon and trout.
- 6.13.53 Primary concern with regards to causing an offence under SAFFA will be causing fish mortalities, damage to or disturbance of spawning habitats, or obstruction to fish passage because of the proposed works, for example polluting the water course through elevated sediment levels, fuel spills, noise etc. The following are considered the relevant sections of SAFFA that are directly applicable to the Proposed Development;
- 6.13.54 Part 1 (2) Roe, spawning and unclean fish, etc.
- 6.13.55 Subject to subsection (5), any person who, except in the exercise of a legal right to take materials from any waters, wilfully disturbs any spawn or spawning fish, or any bed, bank or shallow on which any spawn or spawning fish may be, shall be guilty of an offence.
- 6.13.56 Part 1 (4) Poisonous matter and polluting effluent.
- 6.13.57 4.2.15 Subject to subsection (2), any person who causes or knowingly permits to flow, or puts or knowingly permits to be put, into any waters containing fish or into any tributaries of waters containing fish, any liquid or solid matter to such an extent as to cause the waters to be poisonous or injurious to fish or the spawning grounds, spawn or food of fish, shall be guilty of an offence.

The Eels (England and Wales) Regulations 2009

- 6.13.58 These Regulations implement the EU Eel Recovery Regulation 1100/2007/EC in England and Wales, to help return the stock of the European eel to safe biological limits. These Regulations came into force on 15 January 2010 for England and Wales.
- 6.13.59 The requirements of the regulations are:
- The requirement to notify the Environment Agency of the construction, alteration or maintenance of any structure likely to affect the passage of eels
 - Where any such structure exists, the requirement to construct and operate an eel pass to allow the free passage of eels
 - The removal of any obstruction, if deemed necessary
 - The use of eel screens to exclude eels from water abstraction and discharge points; and
 - If necessary, the use of a by-wash to return excluded eels to the waters they came from.

Wildlife and Countryside Act 1981 (as amended)

- 6.13.60 The following fish are protected under Schedule 5 of the Wildlife and Countryside Act 1981 (*as amended*)
- Allis shad (*Alosa alosa*)
 - Twaite shad (*Alosa fallax*)

- Vendace (*Coregonus albula*)
- Whitefish (*Coregonus laveratus*)
- European sturgeon (*Acipenser sturio*); and,
- Burbot (*Lota lota*).

6.13.61 White clawed crayfish are also afforded protection under Schedule 5 of the Wildlife and Countryside Act 1981 (*as amended*), as such it is an offence to:

- Intentionally kill, injure, or take white-clawed crayfish
- Possess or control any live or dead specimen or anything derived from a white-clawed crayfish
- Intentionally or recklessly damage, destroy, or obstruct any structure or place used for shelter or protection by white-clawed crayfish; or,
- Intentionally or recklessly disturb white clawed crayfish whilst occupying a structure or place which is uses for protection.

Habitats Directive Annex II (part of the Conservation of Habitats and Species Regulations 2017)

6.13.62 The following fish species are listed on Annex II of the Habitats Directive, these species are of conservation importance within Europe, and to ensure conservation of these species core areas of their habitat requires designation of SAC:

- Sea lamprey (*Petromyzon marinus*)
- Brook lamprey (*Lamprey planeri*)
- River lamprey (*Lampetra fluviatilis*)
- Allis shad
- Twaite shad
- Atlantic salmon
- Spined loach (*Corbitis taenia*); and
- Bullhead

Natural Environment and Rural Communities (NERC) Act 2006²

6.13.63 All Annex II listed species above are also listed as a Species of Principal Importance under Section 41 of the Natural Environment and Rural Communities (NERC) Act 2006. As such they are a conservation priority in England.

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- ¹³ Environment Agency (2025). Pacific Pink Salmon (*Oncorhynchus gorbuscha*) Advisory note - 2025 update. Available at:  (Accessed March 2026).
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WHITESTONE
solar farm

Contact

Whitestone Net Zero Ltd

info@whitestonesolarfarm.co.uk

0800 688 9936