**Appendix S1 – Methodological Details**

***S1.1: Summary table of composition and site information for geographic samples.*** *Spp*. denotes species (HG = *H. gammarus*, PE = *P. elephas*); *N* denotes the individuals passing genotyping quality control; Lat. / Long. are approx. coordinates of origin; *H*e/ *H*o denote expected / observed levels of heterozygosity.

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| ***Spp.*** | **Region** | **Nation** | **Area** | **Code** | ***N*** | **Year** | **Tissue Clip** | **Lat.** | **Long.** | ***H*e** | ***H*o** |
| HG | East Med. | Greece | Thessaloniki, Macedonia | Mac | 7 | 2019 | Pereiopod | 40.17 | 23.54 | 0.14 | 0.12 |
| HG | East Med. | Greece | Chios & Skyros, Aegean | Aeg | 4 | 2001 | Pereiopod | 38.84 | 24.65 | 0.13 | 0.13 |
| HG | East Med. | Italy | Bari, Adriatic | Adr | 4 | 2018 | Pleopod | 41.34 | 16.56 | 0.14 | 0.13 |
| HG | West Med. | Italy | Tarquinia, Lazio | Laz | 7 | 2013 | Antennae | 42.00 | 11.78 | 0.15 | 0.15 |
| HG | West Med. | Italy | Olbia coast, Sardinia | Sar | 7 | 2017 | Pleopod | 40.77 | 9.73 | 0.15 | 0.14 |
| HG | West Med. | France | Calvi coast, Corsica | Csa | 4 | 2018 | Uropod | 42.75 | 8.91 | 0.15 | 0.14 |
| HG | Atlantic | Morocco | Tangiers coast | Tan | 7 | 2019 | Pleopod | 35.84 | -5.63 | 0.15 | 0.13 |
| HG | Atlantic | Portugal | Peniche, Oeste | Pen | 7 | 2019 | Pleopod | 39.41 | -9.51 | 0.15 | 0.13 |
| HG | Atlantic | Spain | Vigo, Galicia | Vig | 6 | 2017 | Pleopod | 42.49 | -8.99 | 0.15 | 0.12 |
| HG | Atlantic | France | Île de Ré, La Rochelle | Idr | 6 | 2017 | Uropod | 46.09 | -1.27 | 0.15 | 0.14 |
| HG | Atlantic | France | Brest, Finistère | Bre | 7 | 2019 | Pleopod | 48.31 | -4.84 | 0.15 | 0.13 |
| HG | Atlantic | Channel Is. | Jersey, English Channel | Jer | 9 | 2016 | Pleopod | 49.05 | -1.95 | 0.15 | 0.14 |
| HG | Atlantic | England | Isles of Scilly, Celtic Sea | Ios | 7 | 2016 | Pleopod | 49.90 | -6.36 | 0.15 | 0.14 |
| HG | Atlantic | England | Looe, Cornwall | Loo | 4 | 2016 | Pleopod | 50.29 | -4.30 | 0.15 | 0.14 |
| HG | Atlantic | England | Shoreham-by-Sea, Sussex | Sbs | 7 | 2016 | Pleopod | 50.81 | -0.26 | 0.15 | 0.14 |
| HG | Atlantic | England | Padstow, Cornwall | Pad | 8 | 2017 | Pleopod | 50.60 | -4.93 | 0.15 | 0.14 |
| HG | Atlantic | Wales | Dale, Pembrokeshire | Pem | 6 | 2016 | Pleopod | 51.81 | -5.29 | 0.15 | 0.14 |
| HG | Atlantic | Isle of Man | Peel, Isle of Man | Iom | 6 | 2016 | Pleopod | 54.21 | -4.75 | 0.15 | 0.14 |
| HG | Atlantic | Ireland | Cork, Munster | Cor | 9 | 2016 | Pleopod | 51.66 | -8.42 | 0.15 | 0.14 |
| HG | Atlantic | Ireland | Kilkieran, Galway | Kil | 9 | 2016 | Pleopod | 53.25 | -9.80 | 0.15 | 0.13 |
| HG | Atlantic | Ireland | Arranmore, Donegal | Don | 8 | 2016 | Uropod | 54.93 | -8.55 | 0.15 | 0.15 |
| HG | Atlantic | Scotland | Harris, Hebrides | Heb | 6 | 2017 | Pleopod | 57.79 | -7.25 | 0.14 | 0.13 |
| HG | North Sea | England | Cromer, Norfolk | Cro | 7 | 2016 | Pleopod | 52.95 | 1.31 | 0.15 | 0.14 |
| HG | North Sea | England | Bridlington, Yorkshire | Brd | 7 | 2017 | Pleopod | 54.07 | -0.12 | 0.15 | 0.14 |
| HG | North Sea | Scotland | Eyemouth, Berwickshire | Eye | 7 | 2017 | Pleopod | 55.88 | -2.06 | 0.15 | 0.13 |
| HG | North Sea | Scotland | Shapinsay, Orkney | Ork | 6 | 2017 | Pleopod | 59.01 | -2.78 | 0.15 | 0.13 |
| HG | North Sea | Scotland | Scalloway, Shetland | She | 7 | 2017 | Pleopod | 60.12 | -1.43 | 0.15 | 0.14 |
| HG | North Sea | Germany | Helgoland, North Sea | Hel | 6 | 2017 | Pleopod | 54.18 | 7.90 | 0.14 | 0.13 |
| HG | North Sea | Sweden | Lysekil, Västra Götaland | Lys | 11 | 2017 | Pleopod | 58.26 | 11.38 | 0.15 | 0.13 |
| HG | North Sea | Norway | Flødevigen, Agder | Flo | 8 | 2019 | Pleopod | 58.42 | 8.76 | 0.15 | 0.14 |
| HG | North Sea | Norway | Bergen, Vestland | Ber | 6 | 2018 | Pleopod | 60.65 | 4.77 | 0.15 | 0.12 |
| HG | Norwegian Sea | Norway | Trondheim, Trøndelag | Tro | 4 | 2018 | Pleopod | 63.57 | 9.25 | 0.14 | 0.11 |
| PE | East Med. | Greece | Crete, Aegean Sea | Cre | 13 | 2020 | Pleopod | 35.45 | 24.90 | 0.10 | 0.09 |
| PE | East Med. | Croatia | Hvar, Šolta & Vis, Dalmatia | Dal | 30 | 2020 | Pleopod | 43.25 | 16.34 | 0.10 | 0.09 |
| PE | West Med. | Spain | Mallorca, Balearics | Mal | 8 | 2019 | EBP juvenile | 39.69 | 3.48 | 0.10 | 0.09 |
| PE | West Med. | Spain | Girona, Catalonia | Cat | 13 | 2019 | EBP juvenile | 42.12 | 3.29 | 0.10 | 0.10 |
| PE | West Med. | Spain | Columbretes Isles, Castellón | Col | 10 | 2019 | EBP juvenile | 39.89 | 0.68 | 0.11 | 0.10 |
| PE | Atlantic | Portugal | Peniche, Oeste | Pen | 32 | 2019 | Pleopod | 39.36 | -9.42 | 0.10 | 0.09 |
| PE | Atlantic | France | Perros-Guirec, Côtes-d'Armor | Amo | 21 | 2020 | Pleopod | 48.86 | -3.56 | 0.10 | 0.09 |
| PE | Atlantic | Channel Is. | Jersey, English Channel | Jer | 23 | 2020 | Pleopod | 49.10 | -2.27 | 0.10 | 0.10 |
| PE | Atlantic | England | Isles of Scilly, Celtic Sea | IoS | 34 | 2019 | Pleopod | 49.80 | -6.50 | 0.10 | 0.10 |
| PE | Atlantic | England | Falmouth, Cornwall | CwS | 29 | 2020 | Uropod | 50.09 | -5.01 | 0.10 | 0.10 |
| PE | Atlantic | England | Dartmouth, Devon | DvS | 32 | 2019 | Pleopod | 50.28 | -3.54 | 0.10 | 0.10 |
| PE | Atlantic | England | Newquay, Cornwall | CwN | 34 | 2020 | Pleopod | 50.62 | -5.29 | 0.10 | 0.09 |
| PE | Atlantic | Ireland | Shelf edge, Celtic Sea | Cel | 5 | 2019 | Pleopod | 51.00 | -8.42 | 0.10 | 0.09 |
| PE | Atlantic | Ireland | Kilkee, Clare | Cla | 35 | 2020 | Pleopod | 52.80 | -10.20 | 0.10 | 0.09 |
| PE | Atlantic | Scotland | Harris, Hebrides | Heb | 32 | 2020 | Pleopod | 58.12 | -6.22 | 0.10 | 0.09 |

***S1.2: Sample collection, sample verification and DNA quality control***

Lobsters (*Homarus gammarus*) were sampled during 2016-2020, except those from Lazio, Italy (collected 2013), Aegean Sea, Greece (collected 2001), and Macedonia, Greece (3/7 individuals collected 2001), where scarcity of sampling opportunities meant relying on tissues obtained from previous research. These temporal differences were considered inconsequential; two decades is within lobsters’ lifespan, and likely encompasses at most three generations. Given the recent discovery of Atlantic-origin lobsters in the area (Jenkins et al., 2020), all Corsican samples underwent a preliminary screening using diagnostic SNP assays (Jenkins et al., 2018) to ensure only individuals with Mediterranean descent were included. Crawfish (*Palinurus elehpas*)were sampled during 2019-2020. Tissues of *P. mauritanicus* and *P. charlestoni*, two congener spiny lobster species of similar morphology and overlapping spatial ranges, were also collected in 2019-20 to verify the taxonomic validity of putative *P. elephas* samples. All tissues were preserved in >95% ethanol and stored at 4°C prior to DNA extraction. Purity of all lobster and crawfish DNA extractions was assessed via a Nanodrop 1000 spectrophotometer, and integrity of high molecular weight DNA fragments was evaluated using gel electrophoresis. Lobster DNA yields were standardised using the Promega QuantiFluor-ONE and GloMax Discover Systems, while crawfish DNA yields were standardised using the Invitrogen Qubit 4 Fluorometer and dsDNA HS Assay Kit.

***S1.3: RAD library protocols***

RAD libraries for *Homarus gammarus* samples were prepared by Exeter Sequencing Service, UK, using Illumina Nextera XT barcodes. Genomic DNA (400 ng) from each sample was sheared to an average size of 1000 bp using a Covaris E220 sonicator, having previously optimised for a size range of 800-1000 bp. Fragmentation of eight samples was checked using a DNA 1000 screentape (Agilent). The NEBNext Ultra II DNA library preparation kit was used for end-repair, A-tailing and for ligating P2 adapters, and the reactions were purified using AMPure XP magnetic beads (Beckmann Coulter). P2-adapted DNA was then digested using the restriction enzyme SbfI (TGCA^GG) at 37oC for 4 hours and purified using AMPure XP beads to avoid heat-denaturing the enzyme, which can lead to a bias in the libraries. Phased P1 adapters were ligated to the digested fragments and unligated adapters were removed using AMPure XP beads. The P1 adapter was biotinylated at the 5’end of the top strand to enable capture with streptavidin beads. After washing away fragments not bound to the P1 adapter, the DNA was amplified by PCR to add Nextera XT multiplexing barcodes and flow cell attachment regions. Library quality and quantity was assessed using DNA screentapes. Equimolar pooling of the libraries was undertaken before size selection of libraries averaging 660 bp (inserts ~530 bp). The size-selected pool was quantified by qPCR and stored at -20˚C prior to sequencing of 150 base-pair (bp), paired-end reads on Illumina’s NovaSeq 6000 platform, using an SP Flowcell.

RAD libraries for *Palinurus elephas* were prepared by Floragenex Inc., USA. Genomic DNA from the population was digested with the restriction endonuclease SbfI (TGCA^GG) and processed into RAD libraries similar to the method of Baird *et al* (2008). Briefly, in 96-well format ~200 ng of genomic DNA per individual was digested for 60 min at 37°C in a 15 μL reaction with 20 units (U) of SbfI-HF (New England Biolabs [NEB]). After digestion, samples were heat-inactivated for 20 min at 80°C followed by addition of 2.0 μL of 100 nM P1 Adapter(s), a modified Solexa© adapter (Illumina, Inc.). P1 adapters each contained a unique multiplex sequence index (barcode), which is read during the first ten nucleotides of the Illumina sequence read. 1 uM P1 adapters were added to each sample along with 10x T4 DNA Ligase Buffer (Enzymatics, Inc), T4 DNA Ligase (high concentration [HC], Enzymatics, Inc), and 0.8 μL H2O which was then incubated at room temperature (RT) for 20 min. Samples were heat-inactivated for 20 min at 65°C, pooled, and randomly sheared with a Bioruptor (Diagenode) to an average size of 500 bp. Samples were then run out on a 1.5% agarose (Sigma), 0.5X TBE gel, and DNA 200 bp to 800 bp was isolated using a MinElute Gel Extraction Kit (Qiagen). One reaction of End Repair / dA-Tailing module (NEB) was used to polish the ends of the DNA. After subsequent purification, 1 μL of 1 μM P2 adapter, a divergent modified Solexa© adapter (Illumina, Inc.), was ligated to the obtained DNA fragments at RT. Samples were again purified and eluted in 15 μL. The eluate was quantified using a Qubit Fluorometer with the dsDNA HS assay kit (Invitrogen) and 200 ng of this product was used in 40 individual PCR amplifications (10 ng/reaction) with 25 μL 2x Phusion Master Mix (NEB), 2.5 μL of 10 μM modified Solexa© Amplification primer mix (Illumina, Inc.) and up to 22.5 μL H2O. The primers include a TruSeq i7 index that allows for increased multiplexing, and a unique barcode was assigned to each of the four plates processed. The amplified material for each barcode was pooled and run on a 1.5% agarose (Sigma), 0.5X TBE gel and DNA 300 bp to 900 bp was excised and purified as before. The library was quantified with a Qubit fluorometer and run on an Agilent Bioanalyzer with the High Sensitivity kit to determine size distribution, which was 639 bp for Plate 1, 648 bp Plate 2, 647 bp Plate 3, and 685 bp Plate 4. The libraries were diluted to 5 nM and pooled in equal portions. 2x100pb paired end sequencing was performed on the Illumina HighSeq 3000 platform at the Oregon State University Center facility for Quantitative Life Sciences.

***S1.4: Taxonomic verification of crawfish***

The crawfish data (including interspecific controls) underwent preliminary treatment to ensure taxonomic viability of all putative crawfish as *Palinurus elephas*, rather than the congener species *P. mauritanicus* or *P. charlestoni*. SNP filtering followed the criteria of the main datasets, except for a SNP retention criterion of 75% overall presence, following which DAPC assessment of the resultant 11,122 SNP genotypes confirmed all putative *P. elephas* samples as being genetically distinct from congener species (Supplementary Material S2a). Additionally, one *P. elephas* attributed to a Mediterranean stock actually clustered with Atlantic individuals (S2a). This sample was putatively of Tunisian origin, but was sourced from a holding facility in France and appears not to be of Mediterranean descent, so was removed from the dataset along with all interspecific controls ahead of downstream analysis.

***S1.5: Filtering of reads and RAD loci***

Raw reads were trimmed of adapters and poly-G tails using fastp v0.20 (Chen et al., 2018), before being dephased. Reads were then cleaned, quality filtered and truncated to 97bp using the Stacks v2.54 (Catchen et al., 2011; 2013; Rochette et al., 2019) program *process\_radtags*, following which samples with <300,000 retained reads were removed. The Stacks pipeline *denovo\_map* was used to build RAD loci with optimised parameters for allowable stack mismatches within and between individuals (M/n = 2 for both species) to maximise SNP retention (Paris et al., 2017). The Stacksprogram *populations* was run to filter, isolate and process SNP genotypes.

***S1.6: Protocols and parameters of population genetic analyses***

Some of the site coordinates supplied for the capture of lobsters and crawfish for tissue collection were close to coastlines, at depths of 2-9m according to interrogation via the *getNOAA.bathy* and *get.depth* functions of the R package marmap v1.0.4 (Pante & Simon‐Bouhet, 2013). When run using these original coordinates and a minimum depth of 2m, the *lc.dist* function produced pairwise paths that occasionally traversed landmasses or other impenetrable barriers, and environmental data was unavailable for some sites due to the coarser resolution of the depth-averaged BioOracle dataset (5 arcminutes; ~9.2km2 at the equator). To ensure least coast distances between sites maintained oceanic paths, and to ensure mean depth within BioOracle pixels was negative to facilitate extraction of fully marine environmental data, some site co-ordinates were adjusted (generally seaward) within 5km to ensure depth was negative and normalised to 10-120m.

Overall p-values associated to Mantel tests of IBD, carried out between matrices of pairwise genetic (*F*ST) and geographic (km) distance, were estimated across 10,000 MCMC permutations via the *mantel.rtest* function of ade4 v1.7-15 (Thioulouse et al., 2018) in R. To minimise the possibility of false positives in the detection of SNP outliers potentially under selection by Bayescan v2.1 (Foll & Gaggiotti, 2008), the expectation that site-wise variations in allele frequencies arose from neutral demographic effects was considered 10,000x more likely than effects of selection, and the False Discovery Rate was controlled equivalent to *p* <0.05 (Benjamini & Hochberg, 1995).

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