Applicant: **Baum, Diane**
Organisation: **Ascension Island Government**
Funding Sought: **£199,300.00**

DPR10S2\1017

Barcoding an island – expanding genetic biomonitoring on Ascension

Identifying species is fundamental to biodiversity conservation. However, it is not easy. Modern molecular techniques offer a solution to this problem by providing species identification that is more accurate and efficient than standard taxonomic methods. Through this project Ascension will enter the DNA age. The development of reference barcodes, analysis tools and provision of training in DNA metabarcoding techniques will result in the Conservation Team on Ascension having the capacity to identify multiple species across a range of taxa.

PRIMARY APPLICANT DETAILS

Name	Diane
Surname	Baum
Website	[redacted]
Tel (Work)	[redacted]
Email (Work)	[redacted]
Address	[redacted]

CONTACT DETAILS

Title	Dr
Name	Tiffany
Surname	Simpson
Tel (Work)	[redacted]
Tel (Work)	[redacted]
Email (Work)	[redacted]
Address	[redacted]

Section 1 - Contact Details

PRIMARY APPLICANT DETAILS

Name	Diane
Surname	Baum
Website	[redacted]
Tel (Work)	[redacted]
Email (Work)	[redacted]
Address	[redacted]

CONTACT DETAILS

Title	Dr
Name	Tiffany
Surname	Simpson
Tel (Work)	[redacted]
Tel (Work)	[redacted]
Email (Work)	[redacted]
Address	[redacted]

GMS ORGANISATION

Type	**Organisation**
Name	Ascension Island Government
Phone	[redacted]
Email	[redacted]
Website	[redacted]
Address	[redacted]

Section 2 - Title, Dates & Budget Summary

Q3. Project title

Barcoding an island – expanding genetic biomonitoring on Ascension

What was your Stage 1 reference number? e.g. DPR10S1\1123

DPR10S1\1042

Q4. UKOT(s)

Which UK Overseas Territory(ies) will your project be working in?

☑ St Helena, Ascension and Tristan da Cunha*

*** if you have indicated a territory group with an asterisk, please give detail on which territories you are working on here:**

Ascension

Q4b. In addition to the UKOTs you have indicated, will your project directly benefit any other Territories or country(ies)?

⦿ No

Q5. Project dates

Start date:	End date:	Duration (e.g. 2 years, 3 months):
01 June 2022	31 January 2025	2 years 8 months

Q6. Budget summary

Year:	2022/23	2023/24	2024/25	Total request
Darwin funding request (Apr - Mar)	£67,100.00	£70,700.00	£61,500.00	**£** 199,300.00

Q6a. Do you have proposed matched funding arrangements?

⦿ Yes

What matched funding arrangements are proposed?

AIG staff time - [redacted]
AIG accommodation for project officer - [redacted]
The Natural History Museum (NHM) is providing [redacted] in-kind contributions by waiving the indirect costs associated with this work. Furthermore, the ongoing costs associated with archiving the specimens for future generations will be met by the NHM.

Q6b. Proposed matched funding as % of total project cost (total cost is the Darwin request plus other funding required to run the project). [redacted]

Q6c. If you have a significant amount of unconfirmed matched funding, please clarify how you fund the project if you don't manage to secure this?

The match funding listed above is all confirmed.

Section 3 - Project Summary and Conventions

Q7. Summary of Project

Please provide a brief summary of your project, its aims, and the key activities you plan to undertake. Please note that if you are successful, this wording may be used by Defra in communications.

Please write this summary for a non-technical audience.

Identifying species is fundamental to biodiversity conservation. However, it is not easy. Modern molecular techniques offer a solution to this problem by providing species identification that is more accurate and efficient than standard taxonomic methods. Through this project Ascension will enter the DNA age. The development of reference barcodes, analysis tools and provision of training in DNA metabarcoding techniques will result in the Conservation Team on Ascension having the capacity to identify multiple species across a range of taxa.

Q8. Environmental Conventions, Treaties and Agreements

Please detail how your project will contribute to the aims of the agreement(s) your project is targeting. What key OT Government priorities and themes will it address and how? You should refer to Articles or Programmes of Work here. You should also consider local, territory specific agreements and action plans here.

Letters of support from UKOT Government partners/stakeholders should also make clear reference to the agreements/action plans your project is contributing towards.

The Convention on Biological Diversity (CBD) is a global framework for conserving the three elements of biodiversity (genetic, species, and ecosystem diversity) while ensuring sustainable development and benefit sharing. The CBD has been extended to Ascension Island and requires that signatories:
"7(b) Monitor, through sampling and other techniques, the components of biological diversity."

DNA analysis is increasingly used to as a tool for biomonitoring, and can support the implementation of the CBD on Ascension Island in two main ways. First, through species identification, commonly referred to as DNA barcoding, whereby the island's biota can be both characterised and subsequently monitored with a level of speed, efficiency and taxonomic resolution not previously possible. Second, capacity for DNA analysis enables biodiversity within species to be assessed, and consequently the measurement and conservation of genetic diversity. While explicitly mentioned in the existing CBD targets (Aichi Target 13), The current zero-draft of the CBD CoP15 talks agreement are likely to result in much greater emphasis on the conservation of genetic diversity; a change that is being supported by the UK Government. Developing capacity for genetic monitoring will future-proof Ascension for forthcoming changes to CBD reporting requirements, and provide an example of how accessible, cutting-edge technologies can contribute to the aims of environmental conventions, treaties and agreements.

This international commitment to undertake monitoring is embedded in the Ascension's Species Action Plans and Protected Area Management Plans. The need to address the lack of data on Ascension's invertebrates and incorporate them into monitoring and conservation management programmes is identified as a priority in the new Ascension National Biodiversity Strategy and Action Plan (in prep.). This proposal will directly support the implementation of this UK OT priority by delivering a system capable of generating accurate, comparative long-term data that is sustainable both economically and in terms of its greater independence from personal taxonomic expertise.

Ascension's Biosecurity Strategy was designed to function without the need for full species identification. However, detection and response capabilities would be greatly enhanced by the ability to identify new the introductions and proliferation of invasive non-native species (INNS), accurately and rapidly. This project will improve the ability of Ascension Island to enact its Biosecurity Strategy through rapid, on-site DNA testing.

Section 4 - Project Partners

Q9. Project Partners

Please list all the partners involved (including the Lead Partner) and explain their roles and responsibilities in the project. Describe the extent of their involvement at all stages, including project development.

This section should illustrate the capacity of partners to be involved in the project. Please provide Letters of Support for the lead partner and each partner or explain why this has not been included.

N.B: There is a file upload button at the bottom of this page for the upload of a cover letter and all letters of support.

Lead Partner name:	Ascension Island Government Conservation and Fisheries Directorate
Website address:	www.ascension.gov.ac
Details (including roles and responsibilities and capacity to engage with the project):	The Ascension Island Government Conservation and Fisheries Directorate (AIGCFD) is the only organisation undertaking biodiversity protection on the island and is responsible for ensuring Ascension's obligations under multinational environmental Agreements are met. It is the management authority for Ascension's protected areas and enforces legislation on species and habitat protection. The Directorate conducts long-term monitoring and research activities to aid in the management of biodiversity and biosecurity threats in both marine and terrestrial ecosystems. AIGCFD is the lead partner in this project and will be responsible for project and financial management and reporting. It will conduct a desktop study of available molecular reference sequences for species of interest and coordinate the creation of a sequence library by NHM from specimens already identified by classical taxonomic methods. These results will be used by AIGCFD to conduct metabarcoding of terrestrial and marine invertebrate bulk specimen and gut content DNA samples. AIGCFD will develop biomonitoring workflows, validate laboratory protocols and deliver training in laboratory techniques across the Directorate. AIGCFD will also be responsible for the management of data holdings and contribute to IBOL – BIOSCAN global platform by uploading reference sequences.
Have you included a Letter of Support from this organisation?	⦿ Yes
Have you provided a cover letter to address your Stage 1 feedback?	⦿ Yes

Do you have partners involved in the Project?

⦿ Yes

1. Partner Name:	University of Edinburgh (UoE), Royal (Dick) School of Veterinary Studies & the Roslin Institute
Website address:	www.ed.ac.uk/vet/conservation-science

Details (including roles and responsibilities and capacity to engage with the project):	The UoE Conservation Science group specialises in the development and application of DNA analysis methods to wildlife management and monitoring (https://www.ed.ac.uk/vet/conservation-science/conservation-genetics). During this project the UoE conservation genetics team will provide oversight and quality control of protocol development, assist with the design and validation of a suite of universal primers for Ascension invertebrate species and gut content analysis, and support training in genetic monitoring as required. Prof. Rob Ogden has over 20 years' experience of the application of DNA analysis to conservation management, biomonitoring and wildlife forensics, having worked on the development of overseas laboratory capacity in over a dozen countries. He contributes to policy advice in this field through membership of the IUCN conservation genetics specialist group and Scottish DNA Hub, aligned with the Defra DNA Centre of Excellence. Dr Melissa Marr is a post-doctoral research fellow in conservation genetics with significant laboratory and bioinformatic experience in the production and use of genetic data, including eDNA analysis and MinION sequencing. RO and MM will be ultimately responsible for the development of novel genetic monitoring tools within the project.
Have you included a Letter of Support from this organisation?	⊙ Yes

2. Partner Name:	The Natural History Museum (NHM)
Website address:	www.nhm.ac.uk
Details (including roles and responsibilities and capacity to engage with the project):	The Natural History Museum (NHM) in London has world leading expertise in specimen curation, taxonomic identification, and DNA analysis. In this project, NHM will undertake DNA extraction and -barcoding of up to 800 invertebrate species, depositing the data in the open access International Barcode of Life global repository (BOLD) to enable DNA-based surveillance. The NHM curatorial team will curate all voucher specimens and DNA extracts in our world class facilities, ensuring they are preserved and accessible to the global research community.
Have you included a Letter of Support from this organisation?	⊙ Yes

3. Partner Name:	*No Response*
Website address:	*No Response*
Details (including roles and responsibilities and capacity to engage with the project):	*No Response*
Have you included a Letter of Support from this organisation?	○ Yes ○ No

4. Partner Name:	*No Response*
Website address:	*No Response*
Details (including roles and responsibilities and capacity to engage with the project):	*No Response*
Have you included a Letter of Support from this organisation?	○ Yes ○ No

5. Partner Name:	*No Response*
Website address:	*No Response*

Details (including roles and responsibilities and capacity to engage with the project):	*No Response*
Have you included a Letter of Support from this organisation?	○ Yes ○ No

6. Partner Name:	*No Response*
Website address:	*No Response*
Details (including roles and responsibilities and capacity to engage with the project):	*No Response*
Have you included a Letter of Support from this organisation?	○ Yes ○ No

If you require more space to enter details regarding Partners involved in the Project, please use the text field below.

No Response

Please provide a cover letter responding to feedback received at Stage 1 if applicable and a combined PDF of all Letters of Support.

Darwin Plus 10 - Genetic bimonitoring-Letter responding to Stage 1 feedback final
10/01/2022
08:32:05
pdf 623.14 KB

DPLUS R10 Genetic biomonitoring Stage 2 Letters of support combined
08/01/2022
16:01:48
pdf 481.71 KB

Section 5 - Project Staff

Q10. Project Staff

Please identify the key staff on this project, their role and what % of their time they will be working on the project. Further information on who should be classified as key project staff can be found in the guidance.

Please provide 1 page CVs for these staff, or a 1 page job description or Terms of Reference for roles yet to be filled. These should match the names and roles in the budget spreadsheet. If your team is larger than 12 people please review if they are key project staff, or whether you can merge roles (e.g. 'admin and finance support') below, but provide a full table based on this template in the PDF of CVs you provide.

Name (First name, Surname)	Role	Organisation	% time on project	1 page CV or job description attached?
Diane Baum	**Project Leader**	AIGCFD	5	Checked
Tiffany Simpson	Project Co-lead	AIGCFD	15	Checked
To be recruited	Project Officer	AIGCFD	100	Checked
Robert Ogden	Senior Genetics Adviser	UoE	5	Checked

Do you require more fields?

⦿ Yes

Name (First name, Surname)	Role	Organisation	% time on project	1 page CV or job description attached?
Melissa Marr	DNA Technical Lead	UoE	15	Checked
Ben Price	DNA barcoding; Insect curation	NHM	5	Checked
Lauren Hughes	Non-insect invertebrate curation	NHM	5	Checked
No Response	*No Response*	*No Response*	0	Unchecked
No Response	*No Response*	*No Response*	0	Unchecked
No Response	*No Response*	*No Response*	0	Unchecked
No Response	*No Response*	*No Response*	0	Unchecked
No Response	*No Response*	*No Response*	0	Unchecked

Please provide 1 page CVs (or job description if yet to be recruited) for the Project staff listed above as a combined PDF.

Ensure the file is named clearly, consistent with the named individual and role above.

DPLUS R10 Genetic biomonitoring Stage 2 CVs combined
07/01/2022
09:25:30
pdf 966.81 KB

Have you attached all Project staff CVs?

Yes

Section 6 - Background & Methodology

Q11. Problems the project is trying to address

Please describe the problem your project is trying to address in terms of environment and climate issues in the UKOTs.

For example, what are the specific threats to the environment that the project will attempt to address? Why are they relevant, for whom? How did you identify these problems? How will your proposed project help?

Please cite the evidence you are using to support your assessment of the problem (references can be listed in your additional attached PDF document which can be uploaded at the bottom of the page).

Species are the basic unit of conservation and species identification underpins most monitoring and management efforts. Accurate identification is essential to determine the presence and status of species as well as their distribution, habitat associations and ecological interactions. Distinguishing native from non-native species and identifying the most high risk non-natives is also vital for an effective biosecurity response.

Ascension has relatively comprehensive catalogues of native and non-native vertebrates. This allows managers to quickly recognise an invasive lionfish, for example, and mount a rapid response. However, there is very limited information about invertebrate biodiversity. This means that if a new threat, such as a novel ant species arrives, it may have many years to establish and spread before we recognise it. An horizon scanning exercise identified three ant species as the highest risk potential invaders to Ascension (Roy et al. 2019), yet we would struggle to positively identify them. The very basic problem that invertebrate species are harder to identify than vertebrate ones is skewing our conservation efforts and hampering our biosecurity response.

Classical taxonomic techniques can also fail to provide the information required. Invertebrate species are often the foundation of food webs and the best early indicators of environmental pressures, such as climate change, so the ability to recognise and identify species is critical to conservation and management efforts. Gut content analysis is essential to understand ecosystem functioning, but morphological identification of samples biases the results in favour of species with hard parts that are less easily digestible. Partially or fully digested organisms are impossible to identify visually. Metabarcoding gut samples will increase the ability to identify the diets of indicator species from trace DNA from digested prey including fish invertebrates.

The turnover of staff on Ascension coupled with the multiple taxa and diverse origin of introduced species makes it impossible to maintain the level of classical taxonomic expertise required on island. Even where comprehensive surveys and reference collections have been established for certain taxa, using these can be challenging for generalist staff. Sending samples off for identification is logistically difficult, costly and it can take many months to receive the results reducing their utility for monitoring and biosecurity purposes.

These combined threats of climate change, invasive non-native species and our decreasing ability to monitor species on Ascension drive the need for on-island capacity to perform biodiversity monitoring using DNA analysis. To do this we need external support and the creation of a DNA reference library, something that has been identified as a key limitation to the application of genetic biomonitoring (Price at al. 2020). Achieving capacity in this area will significantly improve the island's ability to detect, monitor and thereby manage environmental and climate issues, while contributing to the continuous development of scientific skills on Ascension and demonstrating to the island community how applied research can help to understand and solve problems in our changing world.

Q12. Methodology

Describe the methods and approach you will use to achieve your intended Outcome and contribute towards your Impact. Provide information on:

- **How you have analysed historical and existing initiatives and are building on or taking work already done into account in project design. Please cite evidence where appropriate.**
- **The rationale for carrying out this work and a justification of your proposed methodology.**
- **How you will undertake the work (materials and methods).**
- **How you will manage the work (role and responsibilities, project management tools etc.)**

(This may be a repeat from Stage 1 but you may update or refine as necessary)

Historical initiatives
AIGCFD are establishing a DNA laboratory on the island. The equipment has already arrived on Ascension and the laboratory will be operational by April 2022. The facility will be led by AIGCFD's Marine Team Leader, who has a background in eDNA research. The current funding will allow the development of eDNA tools for monitoring the presence of native fish and marine biosecurity threats.

There is huge potential to expand the scope of the facility and apply it to other conservation priorities. DPLUS021 and DPLUS135 will provide tissue samples from verified specimens of marine and terrestrial invertebrates species on Ascension. These specimens have been or will be deposited with museums as part of these existing projects. The proposed project will build on that foundation to develop and apply novel genetic tools that will enable these species to be quickly and accurately identified through metabarcoding of samples. This will support the ongoing inshore monitoring that was initiated by DPLUS021and strengthen the Ascension biosecurity system developed through DPLUS096.

A recent report commissioned by the DEFRA DNA Centre of Excellence (Environment Agency 2021) highlighted the potential of DNA to support food web analysis. Barcoding of marine invertebrate species produced through this project would allow us to validate an Ascension marine ecosystem model that is being developed using isotope analysis.

Methodology rationale
This project will provide the tools and training required for Ascension independently to identify species using metabarcoding. External partners and funding will support the design and delivery of analytical protocols, prior to transfer and training on Ascension. Metabarcoding of samples by the Ascension laboratory will then be applied to both biodiversity monitoring and ecological research. Dissemination and outreach will complement the technical work to link the project outputs to local and international stakeholders.

The overall project Methodology is divided into five sets of Activities (see Logframe for associated outputs):
1. Barcoding of existing island samples using single sample protocols.
To maximise the value of tissue samples and metadata (taxonomic ID, geographic origin etc) already collected, existing samples lacking reference sequence data and identified and vouchered taxonomically through projects DPLUS021, DPLUS135 will be collated, inventoried in the Ascension and Biodiversity database and subsampled for DNA analysis, prior to transport of samples to the NHM for accession and DNA barcoding. Resulting barcode data will be shared among partners and submitted to Open Access databases.

2. Development of DNA metabarcoding protocols.
We will develop and validate protocols for routine metabarcoding of mixed species samples, including eDNA samples, using Minion/Illumina sequencing (amplicon sequencing using sample specific indexes and universal primers for target groups). Project officer to deliver training in basic protocol elements (DNA extraction and PCR) across the AIGCFD team to create legacy of staff able to conduct this work.

3. Biodiversity monitoring via DNA metabarcoding.
DNA metabarcoding will be conducted on a variety of sample types using protocols established in Activity 2, including six-monthly sampling and analysis of terrestrial invertebrate traps (bulk insects), inshore marine light trapping (zooplankton) and settlement panels (sessile marine invertebrates). University of Edinburgh to analyse a proportion of the samples independently to QA the Ascension lab procedures.

4. Marine species ecological research.
AIGCFD will use current knowledge of Ascension food web dynamics to identify 10 indicator species for stomach content analysis to validate isotope ecosystem model. University of Edinburgh will assist in developing and validating universal DNA primers and blocking primers for the indicator species for subsequent application to gut content analysis using a metabarcoding approach.

5. Community engagement, schools outreach and stakeholder dissemination.
The Project Officer will organise school visits to the lab on Ascension to teach students about the applications of DNA techniques. We will use the combined technical outputs to create a blueprint for genetic biomonitoring in the UKOTs. Engagement with stakeholders including the Darwin Tree of Life and BioScan to assess collaborative opportunities and the involvement of Ascension Island in these initiatives.

Project management – roles and responsibilities
See Organogram in Supporting Materials for a graphical illustration of project roles.
The project will be managed by the two project co-leads at AIGCFD, who will responsible for line management of the project officer, interaction with project partners and external stakeholders, and reporting to Darwin Plus. Co-leadership combines policy and technical roles and reduces the risk of key-person dependency. Partner representatives for UoE and NHM will join monthly online management meetings and project progress will be monitoring against the Project Timetable (attached), with project risks and mitigation strategies discussed as part of the standing agenda.

If necessary, please provide supporting documentation e.g. maps, diagrams, and references etc., as a PDF using the File Upload below.

Environment Agency 2021 - Understanding_ecosystems_cropped
09/01/2022
15:49:20
pdf 686.84 KB

DPLUS R10 Genetic biomonitoring Stage 2 project management organogram
09/01/2022
15:46:39
pdf 45.43 KB

Price et al_2020_cropped
09/01/2022
15:46:17
pdf 281.92 KB

Final_CEH_report_Horizon_scanning (2)
09/01/2022
15:29:23
pdf 1.07 MB

Section 7 - Stakeholders and Beneficiaries

Q13. Project Stakeholders

Who are the stakeholders for this project and how have they been consulted (include local or host government support/engagement where relevant)? Briefly describe what support they will provide and how the project will engage with them.

The main stakeholders for this project are the AIGCFD Reserves, Marine and Biosecurity Teams who will gain skills in molecular identification techniques and benefit from the improved targeting of conservation effort this will allow. These teams have been involved in the development of the project plan to ensure the training and resources produced provide the support they need to incorporate these techniques into their routine work. This will secure sustainable benefits beyond the lifetime of the project. The project will not only improve their work on Ascension but also their future careers as they become adept at innovative methods that will soon be the cornerstone of conservation work.

The Ascension Island community are important stakeholders in this project as it improves the protection of the island's natural heritage and helps to reveal the true extent of its biodiversity. This proposal has been discussed with the elected Council and Administrator on Ascension to ensure there is strong support amongst policy makers. Social media and articles in the local press will be used to engage the community.

Other Overseas Territories and remote islands are grappling with the same challenge of species identification. By demonstrating that it is feasible to establish DNA extraction and metabarcoding capacity on Ascension and the applications of this for biodiversity protection, this project will provide a model for other territories. We will engage these

stakeholders through social media and peer networks to share learning and resources created during this project and advance their own initiatives.

Q14. Institutional Capacity

Describe the Lead Partner's capacity (and that of partner organisations where relevant) to deliver the project.

AIGCFD employs 21 members of staff with expertise in marine and terrestrial conservation who carry out almost all conservation activity undertaken on the island. Most have a background in biological sciences and have undertaken laboratory work as part of their careers or studies.

Dr Tiffany Simpson, the AIGCFD Marine Team Leader, is an expert in the use of molecular techniques for biodiversity monitoring and biosecurity surveillance. She has practical experience in developing and validating molecular diagnostic assays for high priority marine pests and using eDNA metabarcoding to generate biodiversity profiles from marine environmental samples

University of Edinburgh is a world-leading academic institution; the R(D)SVS and Roslin Institute is currently ranked as the 3rd best School of its type in the World OS Rankings. The state-of-the-art molecular genetics facilities at the Roslin Institute provide Prof Ogden's conservation genetics group with access to both the infrastructure and technical support needed for this project; the project staff themselves have the necessary expertise.

Natural History Museum, London is one of the world's three largest natural history museums. The institution has over 250 years of experience in the natural sciences and public engagement, with over 5 million visitors each year. The Department of Life Sciences comprises over 180 researchers exploring all major aspects of ecology, evolution, systematics and taxonomy. As a core member of the International Barcode of Life project (iBoL) and through our current work in collaboration with the Defra Centre of Excellence for DNA we have active high throughput pipelines for DNA barcoding.

Q15. Project beneficiaries

Who will your project benefit? You should consider the direct benefits as a result of your project as well as the broader indirect benefits which may come about as a result of your project achieving its Outputs and Outcome. The measurement of any benefits should be included in your project logframe.

The main beneficiaries of this project will be AIGCFD and the protected species and habitats it safeguards. The ability to identify invertebrate species accurately and quickly will bring about transformative change for AIGCFD. Existing species identification work associated with our monitoring and biosecurity surveillance programmes will be made significantly more efficient and avoid the lengthy delays associated with ending samples to the UK. This will benefit Ascension's native and endemic biodiversity by allowing us to accurately record it and so create tools for better conservation and biosecurity planning. AIGCFD staff will gain training and experience in new skills that are set to become commonplace in conservation over the coming decade.

The school children and teachers at Ascension's school will benefit from access to a DNA laboratory that allows then to observe and participate in molecular studies that directly benefit the conservation of their island. This will add depth to their normal lessons and show the real world application of such techniques.

By publishing the resulting sequences and results generated from this project, the wider global science community will gain benefit from the increased data available from a normally inaccessible ecosystem.

Section 8 - Gender and Change Expected

Q16. Gender (optional)

How is your project working to reduce inequality between persons of different gender? At the very least, you should be able to provide reassurance that your proposed work is not increasing inequality. Have you analysed the context in which you are working to see how gender and other aspects of social inclusion might interact with the work you are proposing?

The AIGCFD staff upskilled through this project will be 64% female. The project leads are both women providing a positive example of female leadership. Appointment of the project officer will be in line with the Ascension Island Government code of management, which does not condone inequality or discrimination of any kind in the workplace.

We will make a dedicated effort to ensure events organised through this project are fully inclusive and take place at times and locations that do not present a barrier to particular groups attending. The range of communication methods, timings and activities will be designed to make them available and accessible to all members of the community ensuring no discrimination based on gender, religion, sexual orientation or disability.

Q17. Change expected

Detail the expected changed this work will deliver. You should identify what will change and who will benefit a) in short-term (i.e. during the life of the project) and b) in the long-term (after the project has ended). Please describe the changes for the environment and, where relevant, for people in the OTs, and how they are linked.

Monitoring and research that is currently impossible due to the resource required to identify samples would suddenly become feasible. This would enable us to conduct extensive and regular monitoring of our terrestrial and marine invertebrates to understand seasonal and spatial patterns in their distribution and to assess the impacts of threats and effectiveness of management measures. In particular, the ability to identify new non-native introductions rapidly will greatly enhance the opportunity to control and eradicate potentially damaging species.

In the long-term this project will ensure invertebrates are properly considered and protected in Ascension's conservation efforts. Invertebrates are likely to be much more sensitive indicators of change than vertebrates and so our adaptive management of entire ecosystems will be greatly improved as a result.

Embedding molecular techniques into AIGCFD's core work will open new possibilities as the technology develops. The AIGCFD staff trained will have the skills needed for the future of conservation. The project will demonstrate the feasibility of establishing genetic biomonitoring capacity on a remote island and the sharing of knowledge will encourage and support other UKOTs to follow this path.

Sharing the DNA barcodes of Ascension Island's unique biodiversity on an international platform will aid in the global understanding of biodiversity and our capacity to manage it.

Q18. Pathway to change

Detail the expected changes this work will deliver. You should identify what will change and who will benefit a) in the short-term (i.e. during the life of the project) and b) in the long-term (after the project has ended). Please describe the changes for the environment and, where relevant, for people in the OTs, and how they are linked.

DNA techniques will quickly become standard practice in conservation and this project will mean Ascension keeps pace with this change. Ascension is already demonstrating that it is feasible for a remote island to have its own sequencing capacity and the benefits that flow from that. However, the initial DNA barcoding and primer development is beyond the capacity of the Ascension equipment. This project will give us access to external capacity to develop an extensive suite of primers and create a strong partnership with the University of Edinburgh for future collaboration. Once the molecular reference collection is created and protocols established, then the skills required to conduct metabarcoding analyses will be within the capability of generalist AIGCFD staff. Importantly these skills can be passed from one member of staff to another ensuring a strong legacy of the project and the long-term retention of capacity on the island.

Q19. Exit strategy

State how the project will reach a stable and sustainable end point, and explain how the outcomes will be sustained, either through a continuation of activities, funding and support from other sources or because the activities will be mainstreamed in to "business as usual". Where individuals receive advanced training, for example, what will happen should that individual leave?

This project will provide the tools and skills for DNA extraction and metabarcoding to become mainstreamed into the work of AIGCFD. Well-tested protocols produced through this project and the training of all AIGCFD staff means there will be a reservoir of knowledge and experience that can be passed on and retained even with the relatively high turnover rates

seen within AIGCFD.

The new techniques will replace more time-consuming and costly standard taxonomic methods currently used making AIGCFD's activities more effective and more efficient. Costs associated with the future operation of the lab and purchase of consumables will come from the AIGCFD core budget and be offset by the savings against standard methods.

Q20. Ethics

Outline your approach to meeting Darwin's key principles for ethics as outlined in the guidance note. Additionally, are there any human rights and/or international humanitarian law risks in relation to your project? If there are, have you carried out an assessment of the impact of those risks, and of measures that may be taken in order to mitigate them?

This project will meet the relevant ethical requirements set out in the Darwin Plus guidance.

The project will be conducted in accordance with Ascension law. The project activities will be added as an extension to the existing research permit held by the AIGCFD and approved by the Administrator. This includes a data sharing requirement with AIG, though in this instance that is redundant given AIGCFD is part of the island government.

There is no specific Ascension legislation governing the use of genetic material, but in practice this is part of the assessment of research permit applications. This project is being led by AIG and so any benefit will accrue to the island.

AIG has an established health and safety policy that gives absolute primacy to the safety of people. This will be applied to all project activities and people working on the project.

AIG and UoE operate to the highest standards of academic integrity. The results and evidence collected through this project will be robust, accurately reported and freely shared.

Section 9 - Budget, Risk Management & Funding

Q21. Budget

Please complete the appropriate Excel spreadsheet, which provides the Budget for this application. Some of the questions earlier and below refer to the information in this spreadsheet. Note that there are different budget templates for grant requests under £100,000 and over £100,000.

- Budget form for projects under £100,000
- Budget form for projects over £100,000

Please refer to the Finance Guidance **for more information.**

Please ensure you include any co-financing figures in the Budget spreadsheet to clarify the full budget required to deliver this project.

NB: Please state all costs by financial year (1 April to 31 March) and in GBP. Darwin Plus cannot agree any increase in grants once awarded.

Copy of Genetic biomonitorig stage 2 budget Final
10/01/2022
13:48:58
xlsx 80.84 KB

Q22. Financial Risk Management

This question considers the financial risks to the project. Explain how you have considered the risks and threats that may be relevant to the successful financial delivery of this project. This includes risks such as fraud, bribery or corruption, but may also include the risk of fluctuating foreign exchange, delays in procurement or recruitment and internal financial processes such as storage of financial data.

Ascension Island Government financial statements are audited annually which involves obtaining evidence about the amounts and disclosures in the financial statements sufficient to give reasonable assurance that the financial statements are free from material misstatement, whether caused by fraud or error. This includes an assessment of: whether the accounting policies are appropriate to Ascension Island Government's circumstances and have been consistently applied and adequately disclosed; the reasonableness of significant accounting estimates made; and the overall presentation of the financial statements. In addition, the auditors read all the financial and non-financial information in the Introduction to identify material inconsistencies with the audited financial statements and to identify any information that is apparently materially incorrect based on, or materially inconsistent with, the knowledge acquired by them in the course of performing the audit.

All externally funded projects are also managed under Ascension Island Government's financial regulations including the contract regulations for procuring, or tendering for works or goods, and the accounting officer is responsible for ensuring these regulations are followed.

Q23. Funding

Q23a. Is this a new initiative or a development of existing work (funded through any source)?

⦿ Development of existing work

Please provide details:

This work builds on grants from the Pew Charitable Trusts, Blue Belt Programme and Blue Marine Foundation that have enabled the establishment of a DNA laboratory on Ascension. The funding has been used to purchase and convert a shipping container into a dedicated laboratory space and to equip it for DNA extraction and sequencing. This existing project has also supported the development of primers and the purchase of consumables to allow marine eDNA metabarcoding to be carried out for the identification and monitoring of Ascension's inshore and offshore fish assemblages.

There are many further applications of this technology for AIGCFD's work and we are keen to realise the full potential of the laboratory and build the skills of the team.

Q23b. Are you aware of any other individuals/organisations/projects carrying out or applying for funding for similar work?

⦿ No

Section 10 - Finance

Q24. Financial Controls

Please demonstrate your capacity to manage the level of funds you are requesting. Who is responsible for managing the funds? What experience do they have? What arrangements are in place for auditing expenditure?

Grant payments will be administered through Ascension Island Government's bank account, with project expenditures tracked by the AIG Finance Department. AIG has a fully dedicated financial accounting and management team. The Government currently manages capital and reserves of [redacted]. The Finance and Conservation Departments have jointly managed many biodiversity conservation projects, large and small, over the last 10 years, including those funded by RSPB, OTEP, Blue Marine Foundation and Darwin Plus. AIG's main accounts are subject to an annual, independent financial audit and a specific audit would be undertaken for this project.

Q25. Balance of budget spend

Defra are keen to see as much Darwin Plus funding as possible directly benefiting OT communities and economies. While it is appreciated that this is not always possible every effort should be made for funds to remain in territory.

Explain the thinking behind your budget in terms of where Darwin Plus funds will be spent. What benefits will the Territory/ies see from your budget? What level of the award do you expect will be spent locally? Please explain the decisions behind any Darwin Plus funding that will not be spent locally and how those costs are important for the project.

A large proportion of the budget ([redacted]) will be spent on employing a dedicated project officer to provide the short-term additional capacity required to carry out the initial laboratory work required on-island to prepare samples for barcoding, develop protocols and deliver training. These activities are central to the success of the project and AIGCFD will need support on Ascension to deliver them.

The other major expenditure ([redacted]) is the expert assistance from UoE and NHM to conduct the barcoding and oversee the development of protocols and training material. It will not be possible to conduct the long-chain sequencing required for species barcoding within Ascension's laboratory and so an external partner is required for this. Support from UoE will be vital for the project officer and provide confidence that the methods developed are scientifically robust.

Consumables account for [redacted] of the total budget and are required to conduct the DNA extraction, barcoding and metabarcoding. In future the cost of consumables for routine metabarcoding work will be absorbed into the AIGCFD core budget, but there is a large upfront need for consumables to develop the methods that can only be resourced from external funding.

Q26. Capital Items

If you plan to purchase capital items with Darwin Plus funding, please indicate what you anticipate will happen to the items following project end. If you are requesting more than 10% capital costs, please provide your justification here.

A total of [redacted] of the total budget will be spent on capital items. This will be used to purchase a laptop computer for the project officer, a small portable sequencing device that will expand our metabarcoding capacity and basic laboratory equipment such as glassware and specimen tubes. At the end of the project the computer and equipment will remain in the Ascension DNA laboratory to support AIGCFD's ongoing use of DNA metabarcoding for species identification.

Q27. Value for Money

Please describe why you consider your application to be good value for money including justification of why the measures you will adopt will secure value for money.

AIGCFD has used its previous experience of managing projects on the island to ensure that costs are realistic. Staff costs are in line with AIG salaries and having a full-time dedicated project officer based in Ascension offers the most efficient use of budget. Travel costs are unavoidably high given the difficult access to Ascension, but could reduce if the island's runway is repaired during the project.

This project will make full use of laboratory facilities established through other grants. It will link to past (DPLUS021) and current (DPLUS135) specimen collection projects to extract full value from them and mean this project can be focused on DNA sequencing costs.

Specialist expertise from UoE and NHM will be obtained through remote communication tools to allow experts to contribute without the need for expensive international travel.

Q28. Outputs of the project and Open Access

All outputs from Darwin Plus projects should be made available on-line and free to users whenever possible. Please outline how you will achieve this and detail any specific costs you are seeking from Darwin Plus to fund this.

AIGCFD already operates an open access data management policy and will ensure that all outputs are made accessible online through the AIGCFD website and publicised through AIGCFD social media. Sequences produced through this project will be shared globally through the International Barcode of Life BIOSCAN platform.

AIGCFD is also part of a SAERI-hosted information network where data from the South Atlantic OTs can be easily discovered and accessed online by external users (http://www.south-atlantic-research.org/ims-gis). All data from this project will be documented within this online metadata system. Project partners will provide additional support to widen accessibility and will publish information on their websites when appropriate.

Section 11 - Safeguarding

Q29. Safeguarding

Projects funded through Darwin Plus must fully protect vulnerable people all of the time, wherever they work. In order to provide assurance of this, projects are required to have appropriate safeguarding polices in place. Please confirm the lead organisation has the following policies in place and that these are available on request:

Please upload the lead partner's Safeguarding Policy as a PDF on the certification page.

We have a safeguarding policy, which includes a statement of our commitment to safeguarding and a zero tolerance statement on bullying, harassment and sexual exploitation and abuse	Checked
We have attached a copy of our safeguarding policy to this application	Checked
We keep a detailed register of safeguarding issues raised and how they were dealt with	Checked
We have clear investigation and disciplinary procedures to use when allegations and complaints are made, and have clear processes in place for when a disclosure is made	Checked
We share our safeguarding policy with downstream partners	Checked
We have a whistle-blowing policy which protects whistle-blowers from reprisals and includes clear processes for dealing with concerns raised	Checked
We have a Code of Conduct in place for staff and volunteers that sets out clear expectations of behaviors - inside and outside of the work place - and make clear what will happen in the event of non-compliance or breach of these standards	Checked

Please outline how you will implement your policies in practice and ensure that downstream partners apply the same standards as the lead organisation.

All members of AIGCFD and partner organisation staff working on the project will be required to read AIG's Safeguarding Policy and state they are aware of procedures for raising issues or making a complaint. All AIGCFD staff members complete Safeguarding training. A clause will be included in all partner agreements requiring them to have their own safeguarding policy, Code of Conduct and register.

Section 12 - Logical Framework

Q30. Logical Framework

Darwin Plus projects will be required to monitor (and report against) their progress towards their expected Outputs and Outcome. This section sets out the

expected Outputs and Outcome of your project, how you expect to measure progress against these and how we can verify this.

- Stage 2 Logframe Template

Please complete your full logframe in the separate Word template and upload as a PDF using the file upload below – **please do not edit the template structure other than adding additional Outputs if needed as a logframe submitted in a different format may make your application ineligible**. Copy your Impact, Outcome and Output statements and your activities below - these should be the same as in your uploaded logframe.

Please upload your logframe as a PDF document.

R10-DPlus-St2-Logical-Framework-Genetic biomonitoring
07/01/2022
10:39:55
pdf 127.55 KB

Impact:

Ascension acquires the long-term capacity to identify species in difficult taxa enabling a strategic reprioritisation of conservation efforts and increasing the efficacy of biosecurity surveillance and ecosystem analysis.

Outcome:

Ascension has the on-island capability to identify species using DNA metabarcoding and the training materials and corporate memory to ensure this capacity is maintained in the long-term.

Project Outputs

Output 1:

DNA primers developed for detection and identification of principal Ascension Island terrestrial and marine invertebrate species.

Output 2:

AIGCFD staff able to carry out DNA extraction and metabarcoding.

Output 3:

Metabarcoding used to identify terrestrial and marine invertebrate species as part of AIGCFD monitoring and biosecurity surveillance activities.

Output 4:

Gut content analysis using metabarcoding techniques undertaken to validate isotope-based ecosystem model.

Output 5:

Secondary school students on Ascension understand how DNA biomonitoring techniques are carried out and their application for conservation.

Do you require more Output fields?

It is advised to have less than 6 Outputs since this level of detail can be provided at the Activity level.

◉ No

Activities

Each activity is numbered according to the Output that it will contribute towards, for example 1.1, 1.2 and 1.3 are contributing to Output 1.

1.1 Locate all verified invertebrate specimens from past and current studies and obtain tissue sample from each.

1.2 Genetic sample identification number added to Ascension Biodiversity database.
1.3 Barcoding of all samples collated in activity 1.1 by NHM.
1.4 University of Edinburgh to develop a suite of primers that allow discrimination of species collated in output 1.1.
2.1 Write and test protocols for DNA extraction and metabarcoding.
2.2 Create training reference documents and deliver practical training course in DNA extraction and metabarcoding for ten members of AIGCFD.
2.3 Metabarcoding results from AIGCFD staff quality assured by comparing with those from Project Officer and University of Edinburgh.
3.1 AIGCFD staff collect monthly samples from 3 pitfall traps, 2 malaise traps, 3 inshore settlement panels and 2 light traps over six month period and preserve samples in ethanol.
3.2 Metabarcoding of samples collected in output 3.1 by trained AIGCFD staff
3.3 Pass any detections of high priority invasive species to AIG Biosecurity Team for response action.
3.4 Produce summary report listing species detected in samples by metabarcoding.
4.1 Select 10 indicator marine species for gut contents analysis.
4.2 University of Edinburgh to develop blocking primers for the indicator species
4.3 Conduct metabarcoding analysis on gut contents of ten individuals from each of the ten indicator species.
4.4 Conduct traditional gut content analysis on same samples and compare the results of the different methods in a report.
5.1 Organise school visits to the AIG DNA lab and lead practical lessons on DNA extraction.

Section 13 - Implementation Timetable

Q31. Provide a project implementation timetable that shows the key milestones in project activities

Provide a project implementation timetable that shows the key milestones in project activities. Complete the Word template as appropriate to describe the intended workplan for your project, and upload as a PDF.

Implementation Timetable Template

Please add/remove columns to reflect the length of your project. For each activity (add/remove rows as appropriate) indicate the number of months it will last, and fill/shade only the quarters in which an activity will be carried out.

DPLUS R10 Genetic biomonitoring Stage 2 Timetable
05/01/2022
12:03:13
pdf 481.06 KB

Section 14 - Monitoring and Evaluation

Q32. Monitoring and evaluation (M&E)

Describe, referring to the Indicators, how the progress of the project will be monitored and evaluated, making reference to who is responsible for the project's M&E.

Darwin Initiative projects are expected to be adaptive and you should detail how the monitoring and evaluation will feed into the delivery of the project including its management. M&E is expected to be built into the project and not an 'add' on. It is as important to measure for negative impacts as it is for positive impact. Additionally, please indicate an approximate budget and level of effort (person days) to be spent on M&E (see Finance Guidance**).**

The creation of an M&E framework will be a high priority at the beginning of the project. The framework will be tied to the indicators and verification methods set out in the logframe and the timetable included in this application. The AIGCFD project lead will be responsible for drawing up this framework and overall management of the M&E process with input from project partners on specific work packages.

M&E of project progress will be carried out quarterly through a meeting with all project partners to assess progress in delivering the activities shown in the project timetable and achieving outputs according to the logframe indicators. Where important milestones are missed, all relevant project partners will agree actions to regain the original timetable and prevent other outputs being delayed as a consequence.

An adaptive approach will be taken whereby actions that are failing to produce the required outputs and outcomes will be reviewed and revised during virtual meetings of the project partners.

Total project budget for M&E in GBP (this may include Staff, Travel and Subsistence costs)	[redacted]
Number of days planned for M&E	[redacted]
Percentage of total project budget set aside for M&E (%)	[redacted]

Section 15 - Lead Partner Track Record

Q33. Lead Partner track record

Has your organisation been awarded a Darwin Initiative award before (for the purposes of this question, being a partner does not count)?

⦿ Yes

If yes, please provide details of the most recent awards (up to 6 examples).

Reference No	Project Leader	Title
DPLUS135	Diane Baum	From pseudoscorpions to crickets: securing Ascensions Island's unique invertebrates
DPLUS134	Diane Baum	Repelling the invader: turning the tide on Ascension's Mexican thorn
DPLUS113	Diane Baum	Climate Resilience and Conservation of Ascension Biodiversity
DPLUS096	Diane Baum	Building Ascension's Biosecurity Capacity
DPLUS063	Dr Sam Weber	The Ascension Island Ocean Sanctuary
DPLUS047	Mike Howarth	Reduce, reuse, recycle – developing a waste management strategy for

Have you provided the requested signed audited/independently examined accounts?

If yes, please upload these on the certification page. Note that this is not required from Government Agencies.

⦿ No

If no, please provide details.

AIGCFD is a Government Agency.

Section 16 - Certification

Certification

On behalf of the

trustees

of

Ascension Island Government

I apply for a grant of

[redacted]

I certify that, to the best of our knowledge and belief, the statements made by us in this application are true and the information provided is correct. I am aware that this application form will form the basis of the project schedule should this application be successful.

(This form should be signed by an individual authorised by the applicant institution to submit applications and sign contracts on their behalf.)

- **I have enclosed CVs for project key project personnel, letters of support, budget and project implementation timetable (uploaded at appropriate points in application).**
- **Our last two sets of signed audited/independently verified accounts and annual report are also enclosed.**

Checked

Name	Diane Elizabeth Baum
Position in the organisation	Director of Conservation and Fisheries
Signature (please upload e-signature)	Dee signature 07/01/2022 10:50:48 jpg 7.12 KB
Date	10 January 2022

Please upload the Lead Partner's Safeguarding Policy as a PDF.

AI Child Protection procedures 2015 final (June 2015)
08/01/2022
16:04:46
pdf 220 KB

Please attach the requested signed audited/independently examined accounts.

Financial Statements 310320-compressed
08/01/2022
16:03:46
pdf 4.6 MB

Section 17 - Submission Checklist

Checklist for submission

	Check
I have read the Guidance documents, including the "Guidance Notes for Applicants" and "Finance Guidance**".**	Checked
I have read, and can meet, the current Terms and Conditions **for this fund.**	Checked
I have provided actual start and end dates for this proposed project.	Checked
I have provided a budget based on UK government financial years i.e. 1 April – 31 March and in GBP.	Checked
I have checked that the budget is complete, correctly adds up and I have included the correct final total at the start of the application.	Checked
The application has been signed by a suitably authorised individual (clear electronic or scanned signatures are acceptable).	Checked
I have attached my completed logframe and timeline as a PDF using the templates provided.	Checked
I have included a 1 page CV or job description for all the Project staff identified at Question 11, including the Project Leader, or provided an explanation of why not.	Checked
I have included a letter of support from the Lead Partner and main partner organisation(s) identified at Question 10, or an explanation of why not.	Checked
I have included a cover letter from the Lead Partner, outlining how any feedback at Stage 1 has been addressed where relevant.	Checked
I have included a signed copy of the last 2 years annual report and accounts for the Lead Partner, or provided an explanation if not.	Checked
I have checked the Darwin Plus website **immediately prior to submission to ensure there are no late updates.**	Checked
I have read and understood the Privacy Notice on the Darwin Plus website**.**	Checked

We would like to keep in touch!

Please check this box if you would be happy for the lead applicant (Flexi-Grant Account Holder) and project leader (if different) to be added to our mailing list. Through our mailing list we share updates on upcoming and current application rounds under the Darwin Initiative, Darwin Plus and our sister grant scheme, the IWT Challenge Fund. We also provide occasional updates on other UK Government activities related to biodiversity conservation and share our quarterly project newsletter. You are free to unsubscribe at any time.

Checked

Data protection and use of personal data

Information supplied in this application form, including personal data, will be used by Defra as set out in the latest copy of the Privacy Notice for Darwin, Darwin Plus and the Illegal Wildlife Trade Challenge Fund available here. This Privacy Notice must be provided to all individuals whose personal data is supplied in the application form. Some information, but not personal data, may be used when publicising the Darwin Initiative including project details (usually title, lead partner, location, and total grant value) on the GOV.UK and other websites.

Information relating to the project or its results may also be released on request, including under the 2004 Environmental Information Regulations and the Freedom of Information Act 2000. However, Defra will not permit any unwarranted breach of confidentiality nor will we act in contravention of our obligations under the General Data Protection Regulation (Regulation (EU) 2016/679).