Biogeography and community structure of abyssal scavenging Amphipoda (Crustacea) in the Pacific Ocean.

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Abstract

In 2015, we have collected more than 60,000 scavenging amphipod specimens during two expeditions to the Clarion-Clipperton fracture Zone (CCZ), in the Northeast (NE) Pacific and to the DISturbance and re-COLonisation (DisCOL) Experimental Area (DEA), a simulated mining impact disturbance proxy in the Peru basin, Southeast (SE) Pacific. Here, we compare biodiversity patterns of the larger specimens (> 15 mm) within and between these two oceanic basins. Eight scavenging amphipod species are shared between these two areas, thus indicating connectivity. We further provide evidence that disturbance proxies seem to negatively affect scavenging amphipod biodiversity, as illustrated by a reduced alpha biodiversity in the DEA (Simpson Index (D) = 0.62), when compared to the CCZ (D = 0.73) and particularly of the disturbance site in the DEA and the site geographically closest to it. Community compositions of the two basins differs, as evidenced by a Non-Metric Dimensional Scaling (NMDS) analysis of beta biodiversity. The NMDS also shows a further separation of the disturbance site (D1) from its neighbouring, undisturbed reference areas (D2, D3, D4 and D5) in the DEA. A single species, *Abyssorchomene gerulicorbis*, dominates the DEA with 60% of all individuals.

Keywords

JPIO Ecological Aspects of Deep-sea mining, Clarion Clipperton Fracture Zone, CCZ, DisCOL Experimental Area (DEA), Amphipoda.

Introduction

The abyssal deep sea (3000 - 6000 m) represents the largest ecosystem on the planet, with the abyssal seafloor covering approximately 54% of the Earth's solid surface (Rex et al. 1993; Gage & Tyler, 1991). Since it is one of the least investigated ecosystems, there are still extensive gaps in our knowledge of deep-sea fauna (German et al. 2011). Marine research has thus far focused on coastal areas, hydrothermal vents or chemosynthetic habitats, whereas open-ocean abyssal plains have been less extensively investigated (Ramirez-Llodra et al. 2010). This is unsurprising given the challenges of sampling this remote environment, which is impeded by several confounding factors. For example, deep-sea sampling is both financially expensive and labour intensive, and furthermore, constrained by the challenge of deploying equipment at low temperatures (0.01 - 4.0°C) and at high hydrostatic pressures (Sweetman et al. 2017). Therefore, to date very little of the deep sea has been sampled, and the oversampling in the North Atlantic basin has created a biased knowledge base (McClain & Hardy, 2010). Consequently, and owing to the low availability of data on deep-sea biodiversity, and with the inherent risk of under-sampling, it is difficult to estimate species richness in the deep sea.

In the traditional view of the deep sea, the abyss was considered to be homogeneous and many species were thought to have large biogeographical ranges, their dispersal aided by an apparent lack of barriers (Sanders, 1968). This hypothesis was challenged by the discovery of chemosynthetic habitats e.g. hydrothermal vents (Lonsdale, 1977), cold seeps (Paull et al. 1984), seasonal fluctuations in primary productivity (Billett et al. 1983) and erratic whale-falls (Smith et al. 1989). All of this research has demonstrated that the deep sea is an extremely heterogeneous environment and is controlled by many factors, including: Particulate Organic Carbon (POC) flux, water depth, flow regime, current circulation, seafloor topography (Laver et al. 1985) and also historical factors e.g. the opening of ocean basins (i.e. rifting), sea-level rise and fall, and periods of deep-sea anoxia (Smith et al. 2006). All of these can result in a mosaic of different communities (Levin et al. 2001), many of which do not follow a latitudinal gradient (Brandt et al. 2007).

It has also been established that dispersal ability of species on the one hand, and their actual geographic and bathymetric distribution range on the other, are not always linked, and are often dependent on habitat suitability, fragmentation, and ecological flexibility (Lester et al.

2007; Liow 2007). Therefore, although the deep-seafloor includes some of the largest contiguous features on the planet, the populations of many deep-sea species are spatially fragmented, and may become increasingly so with continued human disturbance (Hilario et al. 2015).

In the last decade, there has been an increased demand for exploitation of deep-sea resources e.g. rare earth element (REE) extraction (such as those concentrated in manganese nodule provinces) (Ramirez-Llodra et al. 2011). As a result, ecologists are increasingly asked to assess the ecological risks of these mining activities and to provide sustainable solutions for its mitigation, in order to prevent adverse changes to the deep-sea ecosystem (ISA, 2017).

Glover et al. (2001) showed that abyssal sediments can contain high biodiversity with more than 100 species of meiofaunal invertebrates (e.g. nematodes, copepods) and protists (e.g. foraminifers) found every square meter. In spite of this, our knowledge on the deep-sea ecosystem structure and functioning is still limited, and there is a paucity of data on the distribution, drivers and origins of deep-sea communities at global scales. This is especially true for the biogeography of deep-sea Amphipoda (Barnard 1961; Thurston 1990) and other invertebrates.

Although recent morphological and molecular studies have shed new light on the distribution and habitat niches of certain bentho-pelagic amphipods (e.g. *Eurythenes*) (Havermans, 2016; Narahara-Nakano et al. 2017), there is little published so far on how widespread other amphipod species may be. This lack of information on species richness and ecological uniqueness hampers the answering of crucial questions on recoverability of anthropogenic impacts. Ultimately this impedes ecologists from providing advice on sustainable deep-sea mining practices, thus, underpinning the need for this dedicated deep-sea ecosystem research.

Here, we present distribution patterns of scavenging deep-sea amphipod communities, with the first comparisons of their biogeography and community structures in two oceanic basins. These two basins are the research areas for simulating/studying the anthropogenic impacts of deep-sea nodule mining.

We are investigating whether there are differences and similarities in the species compositions of the two basins (e.g. richness, abundances), and further exploit a disturbance

125 experiment to compare the biodiversity of this mining impact proxy to the undisturbed

126 reference areas. We discuss the possible implications of our findings; aiming to use them to

127 formulate recommendations regarding the pending deep-sea mining of manganese nodule

128 activities in the NE Pacific ecosystem.

129 **Material and Methods**

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131 *Study area*

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133 We investigated the amphipod communities of two oceanic basins (Figure 1); (i) the Clarion-

134 Clipperton Fracture Zone (CCZ, six million km^2, 7000 km wide), an economically important

135 manganese nodule field in the NE Pacific, comprising several different contractor claim

136 areas, (who to date, have exploration licences only), and nine designated Areas of Particular

137 Ecological Interest (APEIs) as designated by the International Seabed Authority (ISA)

138 (Lodge et al, 2014) and (ii) the DISturbance and re-COLonisation (DisCOL) Experimental

139 Area (DEA, 11 km^2, 4 km wide), a mining disturbance proxy (also containing manganese

140 nodules) in the Peru Basin in the SE Pacific. In 1989, the DEA sediment bed was artificially

141 disturbed using a plough-harrow to create 78 track marks. These are supposed to simulate the

142 type of disruption which would be caused by a commercial mining operation (Appendix 1)

143 (Thiel, 1992). This baseline study was a new approach in deep-sea risk assessment and is still

144 ongoing today, providing us with crucial data from this long-term ecological experiment.

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Figure 1: Geographic locations of the two study areas, the Clarion-Clipperton fracture Zone (CCZ) (Northeast Pacific) and the DISturbance and re-COLonisation (DisCOL) Experimental Area (DEA) (Peru Basin, Southeast Pacific). There are nine Areas of Particular Ecological Interest (APEIs) in the CCZ region, which are illustrated by 400 x 400 km^2 white boxes. Grey boxes indicate the various contractor claim areas in the CCZ. We deployed eight amphipod traps across the CCZ, which is 16,000 km^2 and 7000 km wide, and five in the DEA, which encompasses 11 km^2 with a width of 4 km.

Sampling

In 2015 (26 years after the first impact in the DEA in 1989), two research expeditions with the "RV Sonne" visited the CCZ (cruise SO239) and revisited the DEA (cruise SO242-1 & SO242-2), to assess if and how the deep-sea faunal communities had recovered within the DEA, and to attempt to quantify their recolonization potential.

Amphipod samples were taken from the CCZ and DEA using a free-fall lander (120 x 120 x 120 cm), to which four plastic traps were attached (two 20 x 25 x 40 cm traps with four cm openings and two 25 x 40 x 60 cm traps with eight cm openings), ==baited for each station with an 800 g mixture of mackerel, squid and shrimp.== Using this specially designed deep-sea sampling equipment, more than 60,000 specimens of scavenging amphipods were collected from the CCZ and the DEA sites.

The baited trap was deployed eight times across the CCZ at a depth range of 4116 - 4932 m (samples C1 - C8), and five times in the DEA at a depth range of 4078 – 4307 m (samples D1

175   - D5; Figure 1, Table 1). In the CCZ, we sampled within three different contractor claim

176   areas (Table 1) to obtain a pre-disturbance baseline, and to then compare it with one of the

177   nine protected APEIs around the CCZ. In contrast, in the DEA, sampling was conducted once

178   within the disturbed area (D1), twice 10 km away (D2, D3) and twice 40 km away (D4, D5)

179   from D1 in four surrounding reference areas (see Figure 1).

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Table 1: Station overview.

Codes refer to the codes used in this paper in figures 1, 3, 4, 6 and Table 3. The original station code represents the cruise codes from (SO239 and SO232-1). Depth refers to water depth (m) on deployment. Nodule presence/absence information is known only for stations D3 and D4.

Deployment Code	Original Station Code	Depth (m)	Known geological features	Remarks	Residence Time (h:m:s)
C1	SO239-33	4122	Plains	German claim	100:20:00
C2	SO239-37	4116	Plains	German claim	125:49:00
C3	SO239-63	4354	Plains	German claim	66:07:00
C4	SO239-96	4388	Seamount	Inter Ocean Metals (IOM) claim	63:02:00
C5	SO239-123	4529	Plains	Belgian claim	26:12:00
C6	SO239-139	4516	Plains North/South + seamount to west	Belgian claim	56:33:00
C7	SO239-173	4934	Plains	French claim	79:40:00
C8	SO239-205	4855	Plains	Area of Particular Ecological Interest (APEI)	55:59:00
D1	SO242/1-8	4146	Plains	Disturbed	44:26:33
D2	SO242/1-30	4307	Plains	Undisturbed Reference	51:11:18
D3	SO242/1-55	4043	Seamount No nodules	Undisturbed Reference	25:09:09
D4	SO242/1-68	4078	Seamount No nodules	Undisturbed Reference	65:20:46
D5	SO242/1-106	4269	Plains	Undisturbed Reference	47:00:50

Processing

On recovery of the lander, all traps were disconnected and placed in pre-cooled (4°C) buckets of filtered seawater. All specimens were washed on board in a cool-climate laboratory (4°C), morphologically pre-sorted and fixed in molecular grade (95%) ethanol, before being stored at -20°C.

Detailed sorting and identification was performed using the morphological species concept (Futuyama, 1998) and the keys of Lowry & Killagen (2014) and Schulenberger & Barnard (1976), to separate the samples into taxonomic "morphotypes". The larger fraction (> 15 mm length) has been identified to the lowest possible taxonomic resolution. Species not assigned with certainty are denoted here by as affiliated species (e.g. *genus* aff. *species*) or conferred species (e.g. *genus* cf. *species*).

Specimens with a size of less than 15 mm length were excluded from the analysis, primarily because these were mostly juveniles, and their morphological differences were not sufficiently pronounced to allow an accurate identification to the species or even genus level. Also, all pelagic amphipod specimens were omitted which were accidentally caught when the sampling equipment was retrieved to the sea surface. Genera containing multiple and as yet unidentified species have been summarised as "spp."

Statistical analyses

Our null hypothesis (H_0) here is that there are no differences in the amphipod biodiversity of the two basins. To test this hypothesis, we firstly calculated the alpha biodiversity of the two basins was using the Simpson Index (D) (Simpson, 1949) (Appendix 4). Individual-based rarefaction curves were generated using the rarefy function of the vegan package in R 2.3.0 (R Core Team, 2013; Gotelli, 2001) to compare species richness across all thirteen sampling stations and to test for the completeness of sampling.

Secondly, to compare the beta biodiversity, we estimated the variability of the community compositions between sites. The Bray-Curtis dissimilarity metric (Bray & Curtis, 1957), was used to calculate differences between community compositions based on species densities, and the results were then visualised in 2D using a Non-Metric Dimensional Scaling (NMDS)

250 plot. The ANOSIM function in the vegan package of R (R Core Team, 2013; Taguchi & Ono,

251 2005) was used to test the statistical significance of the differences in species compositions

252 between the two study areas.

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Results

Basin biodiversity

In total, 6916 scavenging amphipods (> 15 mm) were collected from the thirteen trap deployments in the two study areas, representing nineteen morphotypes (Figure 2). In the CCZ, we collected 3932 individuals, which represent ten morphotypes. Five of these have been identified to the species level: *Abyssorchomene distinctus*, *A. gerulicorbis*, *Eurythenes sigmiferus*, *Paralicella caperesca* and *Valettietta tenuipes*. A further two are affiliated to a species: *Paracallisoma* aff. *alberti* and *Valettietta* cf. *gracilis*, and the remaining three are at least affiliated to a genus (Tables 2a and 2b). The 2984 individuals from the DEA represent eighteen morphotypes. Six of these have been identified to the species level: *Abyssorchomene distinctus*, *A. gerulicorbis*, *Eurythenes sigmiferus*, *Paralicella caperesca*, *Parandaniexis mirabilis* and *Tectovallopsis regelatus*. A further five which have been affiliated to a species: *Eurythenes* sp. 2. aff. *gryllus*, *Eurythenes* sp. 4. aff. *magellanicus*, *Paracallisoma* aff. *alberti*, *Stephonyx* sp. nov. aff. *arabiensis* and *Valettietta* cf. *gracilis* and the remaining seven identified to at least an affiliated genus (Tables 2a and 2b).

There are eight morphotypes shared between the basins: *Abyssorchomene distinctus, A. gerulicorbis*, *Abyssorchomene* spp., *Eurythenes sigmiferus*, *Eurythenes* spp., *Paracallisoma* aff. *alberti*, *Paralicella caperesca* and *Valettietta* cf. *gracilis* (Figure 2).

Two morphotypes were found only in the CCZ (*Hirondellea* sp. & *Valettietta tenuipes*), and eight morphotypes were found only in the DEA (*Eurythenes* sp. 1-4, gen. aff. *Cleonardo, Parandaniexis mirabilis, Stephonyx* sp. nov. aff. *arabiensis,* and *Tectovallopsis regelatus*) (Table 2).
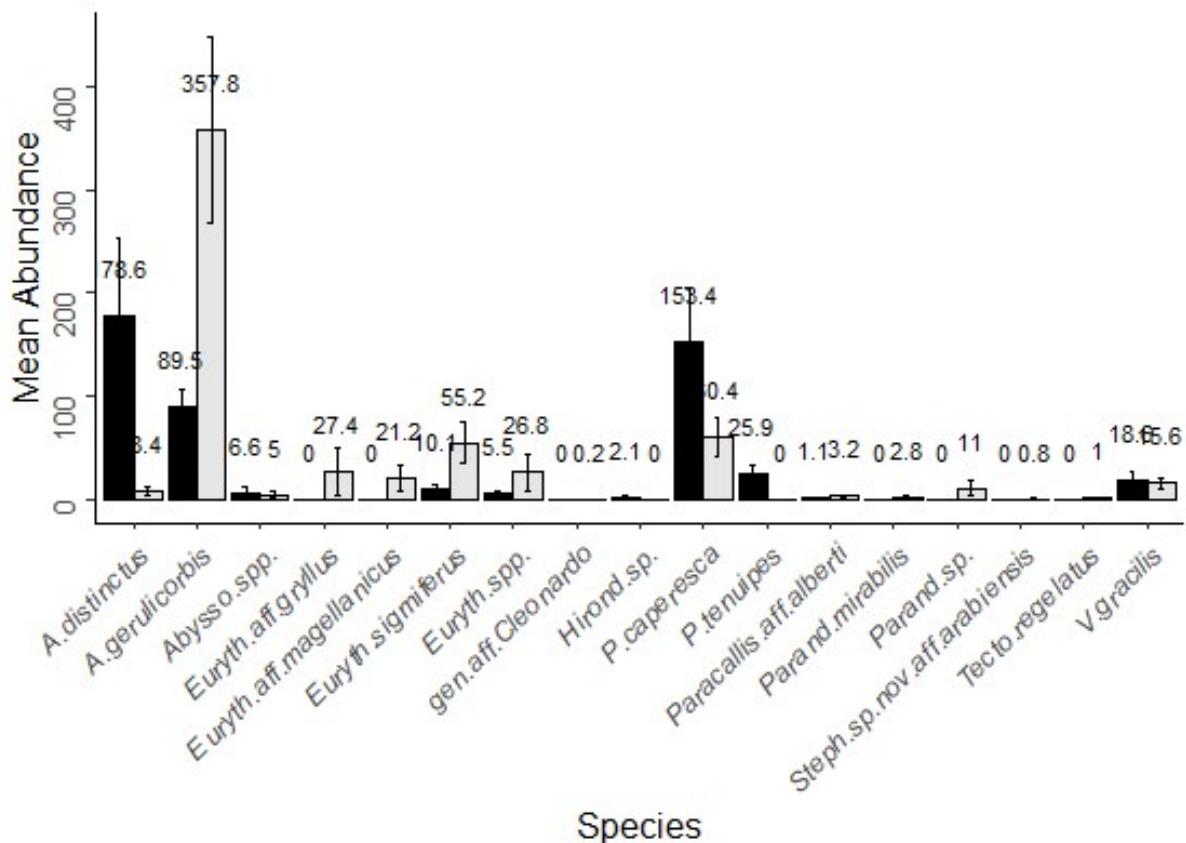
Figure 2: **Histogram showing the species assemblage for the scavenging community in the Clarion-Clipperton fracture Zone (CCZ) (black) and the DisCOL Experimental Area (DEA) (grey)**. The abundances of 17 morphotypes are shown.

Table 2a: Overview of morphotypes across the Clarion-Clipperton fracture Zone (CCZ) and DisCOL Experimental Area (DEA).

	Clarion-Clipperton fracture Zone	DisCOL Experimental Area
Total unique morphotypes collected	17 (10 found in the CCZ, 15 found in the DEA)	
Species possibly unique to this basin	2	7
Shared species between basins	8	

Table 2b: Distribution and abundances of morphotypes across the Clarion-Clipperton fracture Zone (CCZ) and DisCOL Experimental Area (DEA). Green = shared, Yellow = DEA only, Blue = CCZ only.

Taxa	C1	C2	C3	C4	C5	C6	C7	C8	D1	D2	D3	D4	D5
Abyssorchomene.distinctus	629	312	180	170	64	47	2	25	9	3	25	0	5
Abyssorchomene.gerulicorbis	73	47	48	107	71	65	184	121	351	143	522	178	595
Abyssorchomene .spp.	0	0	50	0	0	3	0	0	5	20	0	0	0
Eurythenes .aff. gryllus									119	0	9	0	9
Eurythenes .aff. magellanicus									0	0	59	0	47
Eurythenes.sigmiferus	9	3	35	11	12	5	0	6	30	61	127	36	22
Eurythenes .spp.	6	3	2	0	0	20	1	12	0	91	3	39	1
gen.aff.*Cleonardo*									1	0	0	0	0
Hirondellea .sp.	0	2	0	0	0	0	5	10					
Paracallisoma .aff.alberti	0	0	0	0	0	2	1	6	10	4	0	1	1
Paralicella.caperesca	104	4	114	152	255	75	63	460	86	108	80	21	7
Paralicella.tenuipes	22	0	14	42	43	9	19	58					
Parandania .sp.									5	2	42	5	1
Parandaniexis.mirabilis									11	0	3	0	0
Stephonyx .sp.nov.aff.arabiensis									0	4	0	0	0
Tectovallopsis.regelatus									5	0	0	0	0
Valettietta .cf.gracilis	75	11	29	3	2	5	1	23	2	29	17	1	29

Sampling completeness

Due to differences in allocated ship-times (CCZ cruise being 52 days and the DEA cruise being 29 days), the trap deployments were not identical, making it necessary to normalise deployment times. The resulting Catch Per Unit Effort (CPUE) plot (Figure 3,) shows that, with the exception of C5, all stations in the DEA yielded higher abundances/hr than the CCZ. The highest numbers of individuals/hr were collected at station D3. Overall, there is a moderate negative exponential correlation with increasing deployment times (R = 0.67, *p* = 0.01).
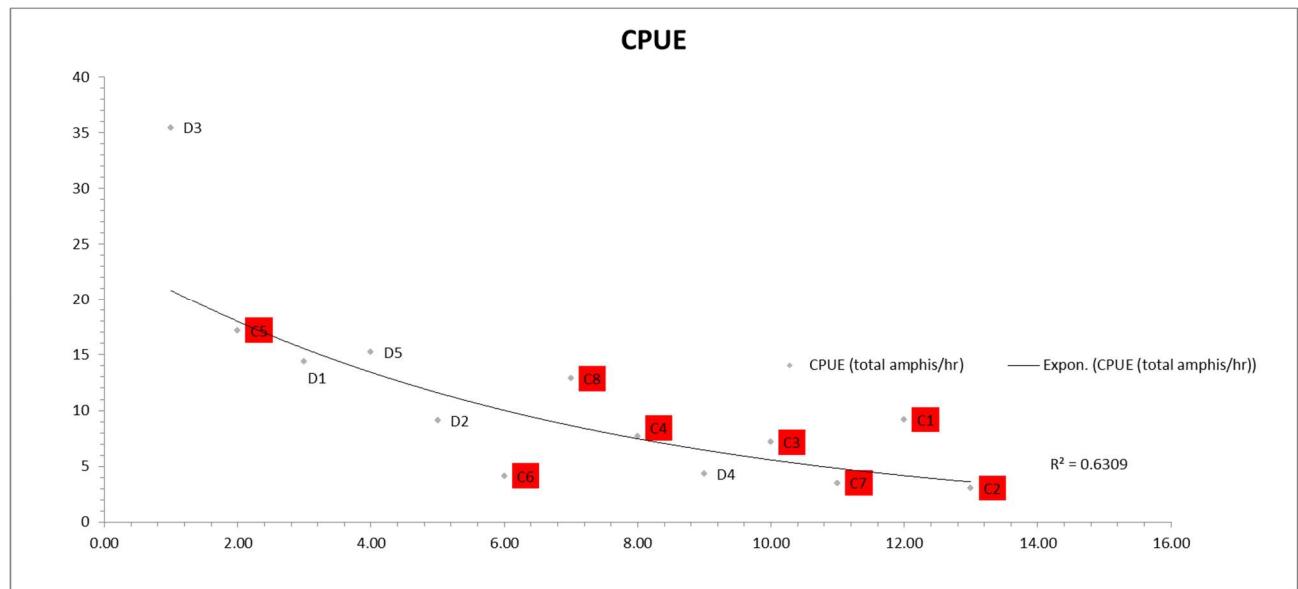
Figure 3: Catch Per Unit Effort (CPUE) illustrating the correlation between sampling time and number of individuals collected. Only the > 15mm fraction was included here to estimate number of collected individuals.

339   The rarefaction results (Figure 4) show that the curves for nine stations reach a plateau,

340   indicating that sampling effort was sufficient to assess diversity levels. These include all CCZ

341   stations except C7. In contrast, four of the five curves for the DEA (stations D1, D2, D4 and

342   D5) are unsaturated.

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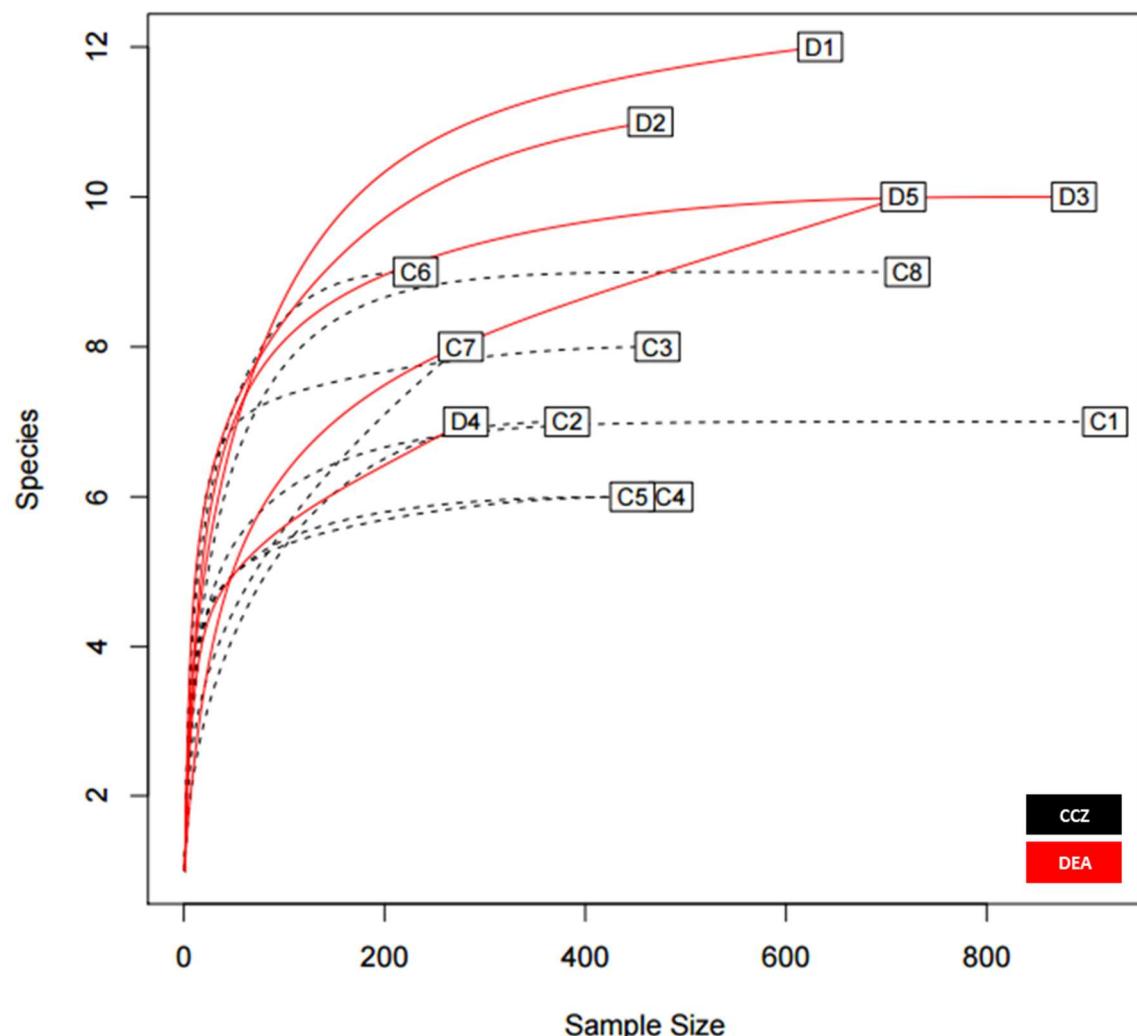
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345   **Figure 4: Species rarefaction curves for each of the 13 trap stations across both areas,**

346   **the Clarion-Clipperton fracture Zone and the DisCOL Experimental Area.** Only

347   individuals greater than 15 mm were considered here.

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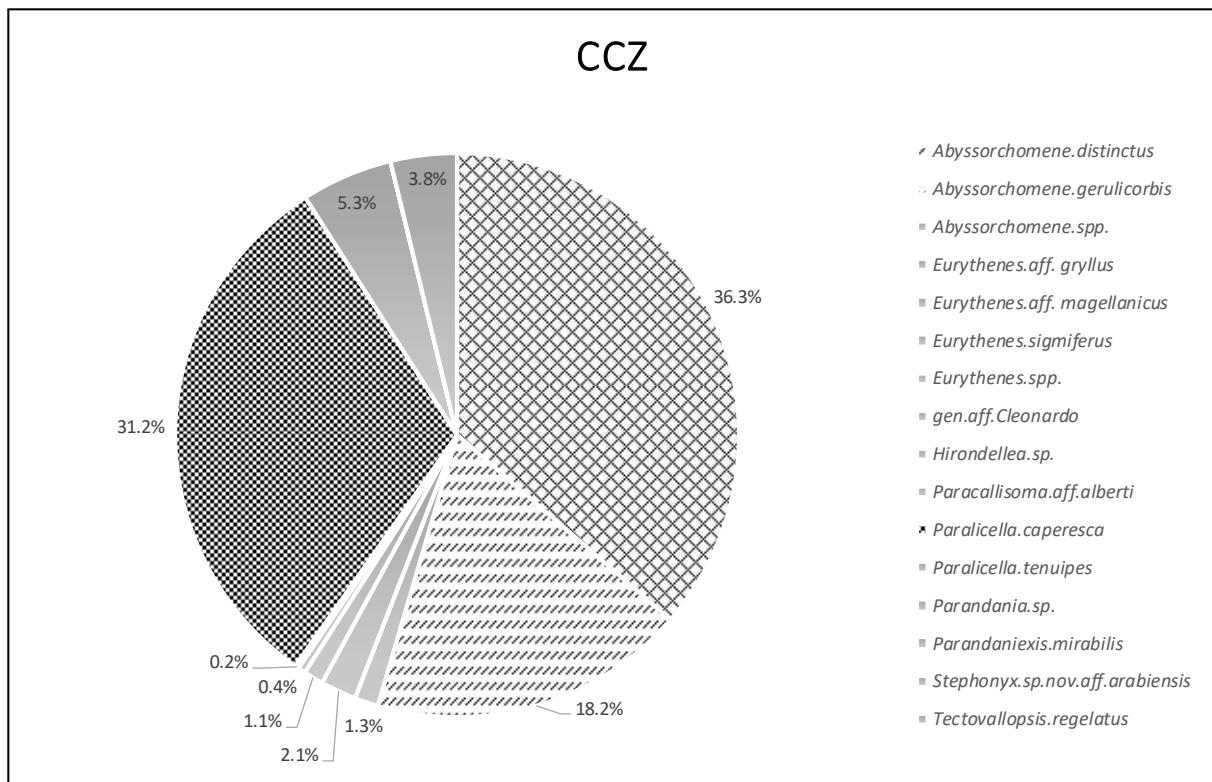
Biodiversity

Figures 5a and b show that the scavenging community in the CCZ is dominated by three species, *A. distinctus* (36%)*, A. gerulicorbis* (18%) and *Paralicella caperesca* (31%)*,* whereas*,* in contrast, the DEA scavenging community is dominated by a single species, *A. gerulicorbis*, accounting for almost 60% of all specimens. The Simpson Index (D) for the entire CCZ area is (with 0.73), higher than the 0.616 that was calculated for the whole of the DEA area (Table 3). The biodiversity of each individual station was further explored (Table 3). In the CCZ, the lowest biodiversity was found at C3 and C6 (D = 0.23) and the highest at C2 (D = 0.67), respectively. In the DEA, the lowest biodiversity of D = 0.36 was found at station D1 (the site of the actual disturbance) and just south of the disturbance site at D2 (0.21), while the highest biodiversity was observed at D5 (D = 0.61) (Table 3).

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Figures 5a and b: Relative species abundances in the Clarion-Clipperton fracture Zone and the DisCOL Experimental Area. These abundances represent the greater than 15mm fraction of the scavenging amphipod community only.

Table 3: Comparison of biodiversity calculated using the Simpson Index (D), for the Clarion-Clipperton fracture Zone and DisCOL Experimental Area, and D for each station is shown for further comparisons within these areas.

Simpson Index (D) whole Clarion-Clipperton Fracture Zone	0.73
Simpson Index (D) whole DisCOL Experimental Area	0.62
C1 (D)	0.41
C2 (D)	0.68
C3 (D)	0.23
C4 (D)	0.27
C5 (D)	0.38
C6 (D)	0.23
C7 (D)	0.45
C8 (D)	0.44
D1 disturbed (D)	0.36
D2 ref 1 (D)	0.21
D3 ref 2 (D)	0.38
D4 ref 3 (D)	0.44
D5 ref 4 (D)	0.61

  Species composition

The NMDS shows that the communities of the two basins are clearly separated (ANOSIM: *p* = 0.002); Figure 6). The disturbed area in the DEA (D1) is showing a clear difference to the four reference areas (D2 - 5). When the communities between the two basins are compared, D2 appears to be most similar to the CCZ community, and more specifically to C6, C7 and C8. The reliability of the data ranking is supported by a low stress value of 0.01.
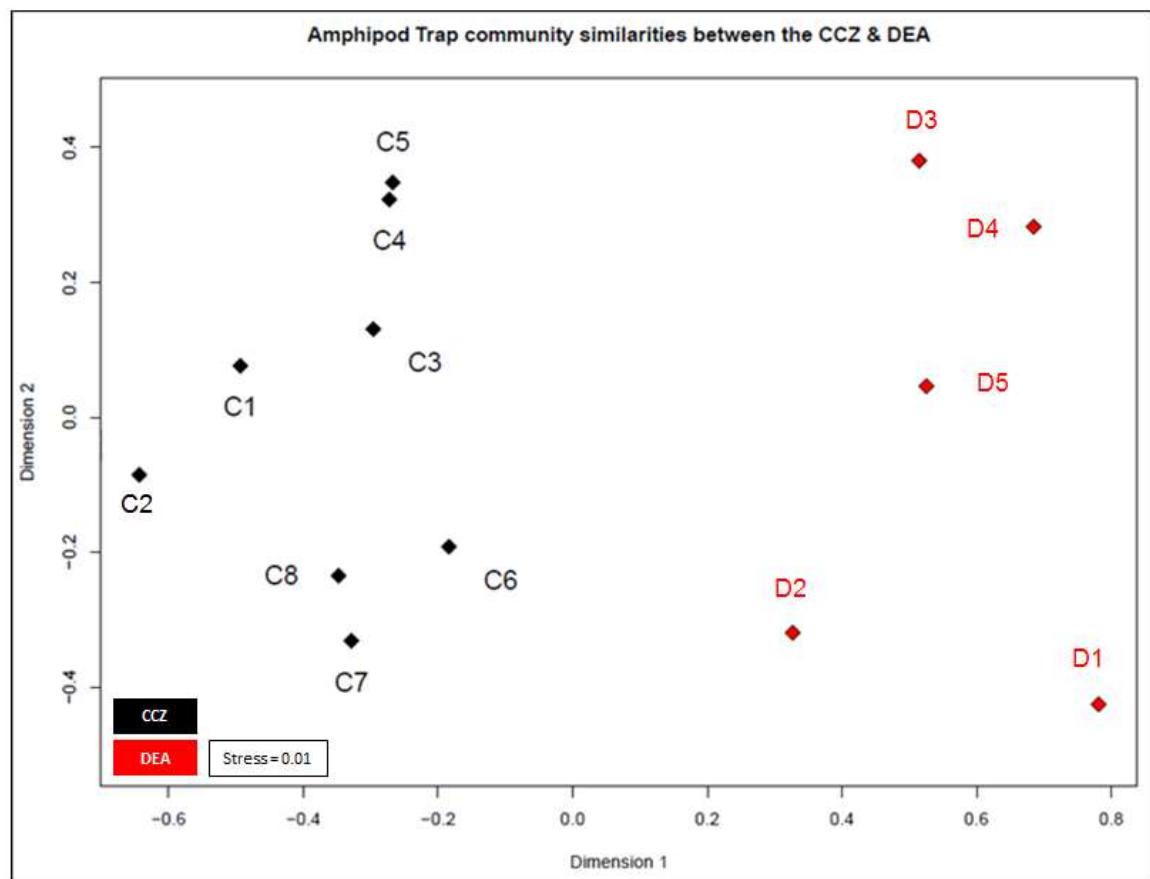
Figure 6 : NMDS plot showing the beta biodiversity (dis/similarities) for each of the thirteen amphipod trap sampling stations associated with the two basins, Clarion-Clipperton fracture Zone (CCZ) (black) and the DisCOL Experimental Area (DEA) (red). Data are supported by a low stress value of 0.01.

Discussion

An unexplored ecosystem

Although the most recent and comprehensive analysis of the Animal diversity of the World's oceans estimates a total of less than a million species over all depths (Appeltans et al. 2012), it is not currently known how many species actually inhabit the deep-sea. Regarding amphipods, only 328 benthic, demersal and benthopelagic species, belonging to 144 genera and 39 families. Over 7000 marine amphipod species have been found below 2000 m. These numbers are reduced to 173 known species, 87 genera and 37 families at depths below 3000 m, and 100 known species, 66 genera and 31 families are known to occur below 4000 m (Vader 2005; Brandt et al, 2012).

Lysianassoidea and their biogeography

The superfamily Lysianassoidea constitutes an important part of the abyssal amphipod fauna. Also, in our sampling, lysianassoid amphipods were collected in large numbers (99% of the samples taken in both basins). As a superfamily, they comprise 23% of all the species found below 2000 m, 35% of the species found below 3000 m and 31% of the species found below 4000 m (Brandt et al. 2012).

Many species in the Lysianassoidea occur in multiple abyssal basins, and some even have worldwide distributions (Thurston 1990). Despite the Ocean Biogeographic Information System (OBIS) database containing 615,650 records of Amphipoda, many of these are shelf or pelagic species, with very few records from the CCZ and DEA (OBIS, 2017). Here, we provide additional data for the known bathymetric range of the seven amphipods which we have identified to species level (*Abyssorchomene distinctus, Abyssorchomene gerulicorbis, Eurythenes sigmiferus, , Paralicella caperesca, Paralicella tenuipes Parandaniexis mirabilis & Tectovallopsis regelatus*) (Table 2b). In addition, we have found two possibly new species of *Eurythenes*, previously not known from these basins, which we plan to analyse further in the future.

While we only sampled *Hirondellea* sp. and *Paralicella tenuipes* in the CCZ, *Eurythenes* aff. *gryllus*, *Eurythenes* aff. *magellanicus*, *gen.* aff. *Cleonardo,* P*arandania* sp., *Parandaniexis*

453   *mirabilis, Stephonyx* sp. nov. aff. *arabiensis,* and *Tectovallopsis regelatus* only in the DEA,

454   we cannot conclude based on the current data only if these species are unique to their

455   respective basins without confirming these distribution patterns with additional sampling

456   campaigns.

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458   ***Catch Per Unit Effort***

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460   Despite the sampling campaign in the CCZ being twice as long as the DEA, the number of

461   individuals/species collected does not correlate positively with deployment effort. We assume

462   that this is rather an effect of abiotic and organic factors, such as the productivity-driven

463   gradients in the CCZ, which decrease from East-West and from North-South (Hannides &

464   Smith, 2003). This lack of correlation is supported by our findings for station C2 (with the

465   shortest deployment time), which shows the highest Simpson Index of all thirteen stations (D

466     $= 0.67$). Further evidence comes from the patterns visualised in Figure 3, which shows a

467   correlation of R = 0.67 for Catch Per Unit Effort (CPUE) and deployment times.

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469   ***Biodiversity within basins***

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471   Figures 5a & b show clearly that the DEA scavenging community has reduced abundances of

472   all species including *A. distinctus* (1%) and *P. caperesca* (7%), and is now dominated by a

473   single species, *A. gerulicorbis*, accounting for 60% of the DEA community. This indicates an

474   interesting resilience and flexibility in the latter species. Similar patterns have been observed

475   in Potter Cove (Seefeldt et al. 2017), where following glacial retreat, a change in

476   sedimentation rates led to the dominance of a single amphipod scavenging species,

477   *Cheirimedon femoratus.*

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479   Although the assemblage of the two basins has some overlap in its amphipod diversity (as is

480   exemplified by the eight shared morphotypes), the sampling stations (and the two basins) are

481   heterogeneous concerning species compositions of the subdominant and rarer species

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483   Thus, we can observe some negative influence (possibly attributed to the disturbance in the

484   DEA) on the scavenging amphipod community. This reduced biodiversity is reflected in the

485   higher Simpson Index (D) for the CCZ (0.73) as compared to the DEA ($D = 0.62$; Table 3).

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487   To explore whether this reduced diversity in the DEA was truly a result of the simulated

488   disturbance, D was also calculated for each sampling station within each basin (Table 3).

489   In the CCZ, the APEI (C8) shows a moderate level of biodiversity ($D = 0.44$), indicating that

490   it is not optimally-placed for representing the biodiversity of the scavenging amphipod

491   community of the CCZ. Additionally, this pre-existing lower biodiversity (in comparison to

492   the contractor claim areas), indicates that the APEI may not serve well as a refugium for

493   amphipods post-disturbance. However, due to the fact that only one of the eight APEIs have

494   been investigated thus far, this APEI along with the remaining eight APEIs would need to be

495   (re-) sampled.

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497   Within the DEA, the lowest biodiversities are observed at the site of the disturbance (D1) and

498   south of it (D2; Table 3), indicating that the reduced biodiversity in the DEA could indeed be

499   caused by the simulated disturbance in 1989 (Thiel, 1992).

500

501   The highest abundances in the DEA were collected from station D5 ($n = 717$); this station

502   also has the highest Simpson Index within the DEA ($D = 0.61$) (Table 3). Side-scan sonar

503   imaging shows a seamount range to the North West (NW) of the disturbed area (D1)

504   (Appendix 3). Although the relief change is only 150m, the range extends laterally for several

505   kilometres (SO242-1 Cruise report, 2016) hampering dispersal across barriers such as sills,

506   canyons and ridges (Smith, et al. 2006; Blankenship et al. 2006; Etter et al. 2011). However,

507   recent studies have shown that due to their mobile nature, geographic isolation alone would

508   not pose a true barrier to bentho-pelagic amphipod species (Havermans, 2016; Ritchie et al.

509   2017), and thus, cannot explain why such a high number of large scavenging individuals was

510   collected at station D5.

511

512   ***Community similarities***

513

514   Scavenging amphipods are resilient and dispersive, but most importantly, they are highly

515   mobile (Ingram and Hessler 1983; Lörz et al. 2018). Often driven by their search for erratic

516   deposited feeding opportunities (Smith et al. 1989), they are probably less constrained by

517   local environmental abiotic conditions and seafloor topography.

518

519   Beta diversity can be regarded as the dissimilarities in species composition between spatially

520   different communities. As an indication for beta biodiversity, the NMDS (Figure 6) shows a

521   significant separation in the similarity index between the two basins (ANOSIM $p = 0.002$).

522   However, despite the dispersive and resilient nature of scavenging amphipods, their

523   biodiversity appears to have been affected by the disturbance experiment as evidenced by the

524   NMDS (Figure 6), where the disturbed area (D1) and the area closest to it (D2) are separated

525   from the remaining three reference sites (D3, D4 and D5).

526

527   In the CCZ, stations C1, C2, C3, C4 and C5 form one cluster in the NMDS (Figure 6), and

528   stations C6, C7 and C8 a second cluster. The CCZ is a geomorphologically very

529   heterogeneous region, with seamounts of 200 m altitude running from north-south. A barrier

530   of this height would be expected to affect sedimentation rates, nodule presence and currents.

531   Furthermore, the difference in depth from the eastern edge (3950m) and the western edge

532   (5150 m) is more than 1200 m. These combined factors very likely give rise to different

533   trends in species composition (Glover, et al. 2015). However, since it has been established

534   that bentho-pelagic amphipods are less sensitive to such barriers (Havermans, 2016), at this

535   stage, other biotic (e.g. the productivity gradient) and abiotic factors causing this separation

536   cannot be excluded as alternative explanations.

537

538   ***Dispersal and connectivity***

539

540   Whilst the NMDS (Figure 6) illustrates a visual separation of the two basins, there is also

541   some similarity in the amphipod fauna between the two areas, (as is obvious by the eight

542   shared species), indicating that the dispersal extent for these eight species might range up to

543   at least 3000 km. However, this hypothesis will need to be confirmed with subsequent

544   molecular analyses.

545

546

547   Abyssal amphipods have been shown to be able to travel actively at speeds of almost 4

548   cm/sec (Laver, 1985), even at temperatures as low as 3°C (Kankaanpää et al. 1995). It is

549   obvious that they are sufficiently strong to swim up weak currents since they can be found

550   several hundred meters above the seafloor searching pelagically for mates (e.g. *Eurythenes*

551   *gryllus* occurring up to 1800 m above the seafloor) (Thurston 1990; Havermans et al. 2013)

552   or following food-falls (Baldwin and Smith 1987).

553

However, it is apparent that the dispersal of abyssal amphipods is not always contingent on current direction, but also on passive dispersal. Amphipods can also be carried passively over long distances by stronger currents e.g. (the circumpolar current of the Southern Ocean) (Laver et al. 1985), but even weaker deep-sea currents have been suggested as a mechanism for deep-sea dispersal of amphipods (e.g. *Eurythenes gryllus* (Schüller and Ebbe 2007)). This coupled with their ability to follow odour-plumes (Ide et al. 2006; Premke, 2003), significantly increases the probability and extent of their dispersal (Conlan 1991; Highsmith, 1985). Specialist feeding adaptations for several species in our assemblages have been reported in Havermans & Smetacek, 2018). For example, the semi-tubular flap-like molars of the genera *Hirondellea* and *Eurythenes*, and the distendable foregut (*Abyssorchomene*) and midgut (*Paralicella*). It is not clear from our study in the absence of POC data for the areas of the trap deployments whether the biogeography of these specialised feeders is linked to the productivity gradients in these two basins.

The lack of a clear dispersal pattern is obvious from Figure 6, where station D2 is the station clustering closest with the CCZ basin in terms of species composition despite the fact that station D5 is geographically the shortest distance away from the CCZ.

Recent research on *Eurythenes gryllus* has demonstrated that it thrives in every ocean with a wide (pelagic – hadal) depth range. However, assumptions that individual morphotypes of this species belong to the same genetic lineage have been challenged (Havermans et al. 2013 & Havermans, 2016). Ritchie et al. (2016) demonstrated with microsatellite markers heterozygote deficiency in *Paralicella tenuipes,* which they attributed to cryptic diversity. It is likely that the connectivity of the eight species we observed as being shared between the CCZ and DEA (based on traditional morphological methods), can be explained by the presence of one or more cryptic amphipod species, which are being tested in our future molecular research.

Unfortunately, in the absence of data on deep-sea currents in the study area, especially between the CCZ and DEA, it is not yet possible to fully explain the drivers and mechanisms of amphipod dispersal between these particular basins.

The DisCOL Experimental Area as a proxy

Higher abundances of scavenging amphipods were collected from the CCZ (3932 individuals) as opposed to the DEA (2984 individuals). Yet, we have identified more morphotypes in the DEA (18) than in the CCZ (10), indicating that the DEA is more speciose, and thus, more biodiverse.

Although the DEA is more speciose, many of its morphotypes were collected in low abundances, with several of these being singletons or doubletons (collected from one or two sampling stations only). This is reflected in the rarefaction curves (Figure 4), which indicate thorough sampling in the CCZ with all but station C7 reaching asymptotes. In contrast, four stations in the DEA (D1, D2, D4 and D5) are unsaturated. This pattern suggests firstly that the less abundant species which are present at fewer stations only may not necessarily be rare species and secondly, that there could be as yet undetected biodiversity in the DEA. Therefore, the effects of mining impact could be even more pronounced than we observed in this study. However, as the seafloor environment is subject to seasonal fluctuations (Billett et al. 1983), it is hard to predict exactly what the effects will be at this stage.

Our preliminary (basin-scale) comparison of the scavenging communities of the two study areas shows that even if the DEA is a small-scale disturbance experiment, it is a very diverse area. Thus, the DEA is a well-chosen site for monitoring the impacts of disturbance and instrumental in its role as a proxy to assess impending mining activities in the CCZ.

Future research

At several stations in both basins, we collected amphipods in very high abundances (C1, C8, D3 & D5) (Table 2b). Since biotic production is contingent on the sinking flux of particles from the euphotic zone (Sweetman, 2017), the biodiversity differences at each of the thirteen stations could be driven by Particulate Organic Carbon (POC) or erratic whale-falls (Smith et al. 1989). However, not all feeding behaviour of scavenging amphipods is based on opportunistic or erratic availability of nutrients (Havermans & Smetacek, 2018). During future sampling campaigns, the POC of these areas should be monitored, along with experiments on different types of food-fall in addition to obtaining side-scan sonar and

abiotic data. This will provide a more comprehensive view of the food types required for these species to thrive in the deep sea.

It is not clear from our results whether substrate type (i.e. nodule/non-nodule) has any effect on the amphipod communities (Smith and Demopoulos, 2003) since this kind of data is only available for stations D3 and D4. To answer this question, resampling of the study areas in combination with an Ocean Floor Observation System (OFOBS) (video/camera) is required.

Although our study only addresses the scavenging amphipod species longer than 15 mm, we already find indications for a disturbance effect in the DEA. It is obvious that scavenging amphipods are only one of several bentho-pelagic indicator groups. Other truly benthic groups such as sponges or less dispersive amphipods (e.g. collected by epibenthic sledge (EBS)) may demonstrate an even more pronounced impact of mining activities and should be investigated in future studies.

With the application of molecular techniques to identify cryptic species (Delić et al, 2017), more realistic estimates of biodiversity can be obtained (Schön et al. 2012), improving our current knowledge of the biodiversity of this area. If these improved estimates of biodiversity also include cryptic species, it is possible that the biological impact of manganese nodule mining on amphipod and other deep-sea faunal communities may turn out to be even higher.

Conclusions

In summary, this study on the scavenging amphipod community of two abyssal oceanic basins has demonstrated that amphipods are present in high abundances across the CCZ and DEA, with eight shared species and some species possibly being unique to their respective basin.

Our results have indicated that the simulated mining experiment probably had an impact on the biodiversity of these scavenging amphipods, as demonstrated by the low D of the DEA overall, at the disturbance site itself (D1), and the 60% dominance of *A. gerulicorbis* in this region.

Given the scarcity of sampling and industry experience of marine habitats at these depths, the formulation of effective regulations is challenging (International Seabed Authority, 2017).

652   Nonetheless, our study provides the first results on possible effects of disturbance activities

653   on the abyssal amphipod biodiversity of deep-sea basins.

654   **Sample and data availability**

655   Biological samples pertaining to this manuscript are stored at the Royal Belgian Institute of Natural
656   Sciences, and the data discussed in the manuscript are submitted to PANGEA.

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658

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References

Appeltans, W., Boxshall, G., Bouchet, P., Vanhoorneb., Decock, W., Warren, A., Collinsa., Kroh A., Schmidt−Rhaesa, A., Berta, A., Barber, A., Todaro, A., Gittenberger, A., Hoeksema, B., Swalla, B., Neuhaus, B., Hayward, B., Self−Sullivan, C., Fransen, C., Messing, C., Erséus, C., Emig, C., Boyko, C., Mah, C., Millsc., Nielsen, C., Jaume, D., Fautin, D., Domning, D. P., Gibson, D., Lazarus, D., Gordon, D., Opresko, D., Schwabe, E., Mac−Pherson E., Thuesen, E., Dahdouh−Guebas, F., Anderson, G., Poore, G., Williams, G., Walker−Smith, G., Read, G., Lambert, G., Paulay, G., Segers, H., Furuya, H., Bartsch, I.,Van Der Land, J., Reimer, J. D., Vanaverbeke, J., Saiz−aiz, Saike, J., Sair, J. D.,Pilger, J., Norenburg, J., Kolb, J., Schnabel, K. E., Meland, K., Fauchald, K., Cheng, L., Van Ofwe−Gen L., Błażewicz−Paszkowycz, M., Rius, M., Curini−Galletti, M., Schotte, M., Tasker, M. L., Angel, M. V., Osawa, M., Longshaw M., Guiry, M., Bailly, N., De Voogd, N., Bruce N., Shenkar, N., Garcia−Alvarez, O., Mclaughlin, P., Kirk, P., Davie, P., Ng, P. K. L., Schuchert P., Uetz P., Bock P., Pugh P., Lemaitrer., Kristensen R., Van Soestr., Bray, R., Bamber, R. N., Da Rocha, R. M., Hopcroft R., Stöhr, S., De Grave, S., Gerken, S., Gofas, S., Tyler, S., Ahyong, S., Wilson, S., Brandao S. N., Koenemann, S., Feist, S., Cairns, S., Timm, T., Cribb, T., Molodtsova, T., Chan, T.–Y., Iseto, T., Artois, T., Scarabino, V., Siegel, V., Eschmeyer, W., Hummon, W., Perrin W., Sterrer, W., Hernandez, F., Mees J., Costello, M. J.: The Magnitude of Global Marine Biodiversity, Curr. Biol., 22, 2189–2202, 2012.

Baldwin, R. J. & Smith, K. L. Jr.: Temporal variation in the catch rate, length, color, and sex of the necrophagus amphipod, *Eurythenes gryllus*, from the central and eastern North Pacific, Deep Sea Res. (part 1 Oceanogr. Res. Pap.), 34, 425–439, 1987.

Barnard, J. L.: Gammaridean Amphipoda from depth of 400 to 6000 m, Galathea. Rep., 5, 23–128, 1961.

Barnard, J. L. & Karaman, G.: The Families and Genera of Marine Gammaridean Amphipoda (Except Marine Gammaroids), Rec. Aust. Mus. Suppl. 13., 1–866, 1991.

708   Belyaev, G. M.: Deep-Sea Ocean Trenches and Their Fauna. Nauka Publishing House,
709   Moscow, 385 pp, 1989.

710

711   Billett, D. S. M., Lampitt, R. S., Rice, A. L. & Mantoura, R. F. C.: Seasonal sedimentation of
712   phytoplankton to the deep-sea benthos, Nature, 302, 520–522. 1983.

713   Blankenship, L. E. & Levin, L. A.: Extreme food webs: Foraging strategies and diets of
714   scavenging amphipods from the ocean's deepest 5 kilometers, Limnol. Oceanogr., 52,
715   1685–1697, 2007.

716

717   Blankenship, L. E., Yayanos, A. A., Cadien, D. B., and Levin, L. A.: Vertical zonation
718   patterns of scavenging amphipods from the Hadal zone of the Tonga and Kermadec
719   Trenches, Deep-Sea Res. Pt. I, 53, 48–61, doi:10.1016/j.dsr.2005.09.006, 2006.

720

721   Brandt, A., Gooday A. J., Brix S. B., Brökeland, W., Cedhagen, T., Choudhury, M.,
722   Cornelius, N., Danis, B., De Mesel, I., Diaz R. J., Gillan D. C., Ebbe B., Howe J.,
723   Janussen, D., Kaiser, S., Linse, K., Malyutina, M., Brandao, S., Pawlowski, J. &
724   Raupach, M.: The So Deep Sea: First Insights Into Biodiversity And Biogeography.
725   Nature, 447, 307–311, 2007.

726

727   Bray, J. R. & J. T. Curtis.: An ordination of upland forest communities of southern
728   Wisconsin, Ecol. Mono., 27, 325–349, 1957.

729

730   Brenke, N.: An epibenthic sledge for operations on marine soft bottom and bedrock, Mar.
731   Technol. Soc. J., 39, 10–21, 2005. hdl.handle.net/10.4031/002533205787444015.

732

733   Carmona, L., Malaquias, M. A. E., Gosliner, T. M., Pola, M. & Cervera, J. L. Amphi-Atlantic
734   distributions and cryptic species in Sacoglossan sea slugs, J. Molluscan Stud., 77,
735   401–412, 2011.

736

737   Conlan K. E.: Precopulatory mating behavior and sexual dimorphism in the amphipod
738   Crustacea, Hydrobiologia, 223, 255–282, 1991.

739

740 Delić, T., Trontelj, P., Rendoš, M. & Fišer., C.: The importance of naming cryptic species
741   and the conservation of endemic subterranean amphipods. Sci. Rep., 7, 3391, 2017.
742   https://doi.org/10.1038/s41598-017-02938-z
743
744
745 Etter, R. J., Boyle, E. E., Glazier, A., Jennings, R. M., Dutra, E. & Chase. M. R.
746   "Phylogeography of a Pan-Atlantic Abyssal Protobranch Bivalve: Implications for
747   Evolution in the Deep Atlantic." Molecular Ecology 20 (4): 829–843, 2011.
748   doi:http://dx.doi.org/10.1111/j.365-294X.2010.04978.x.
749
750 France, D. & Kocher, T.: Geographic and bathymetric patterns of mitochondrial 16S rRNA
751   sequence divergence among deep-sea amphipods, *Eurythenes gryllus*. Mar. Biol., 126,
752   633–643, 1996.
753
754 Futuyma, D. J.: Evolutionary Biology, ed. 3. Sinauer Associates, Sunderland MA, 751 pp.,
755   1998.
756
757 Gage, J. D. & Tyler, P. A.: Deep-Sea Biology: a natural history of organisms at the deep-sea
758   floor, Vol. Cambridge University press, Cambridge, 1991.
759
760 German C. R., Ramirez-Llodra, E., Baker, M. C., Tyler, P. A., and the Chess Scientific
761   Steering Committee.: Deep-Water Chemosynthetic Ecosystem Research during the
762   Census of Marine Life Decade and Beyond: A Proposed Deep Ocean Road Map,
763   PLoS ONE, 6, 2011. doi:10.1371/journal.pone.0023259.
764
765 Glover, A., Paterson, G., Bett, B., Gage, J., Sibuet, M., Sheader, M., Hawkins, L.: Patterns in
766   polychaete abundance and diversity from the Madeira Abyssal Plain, northeast
767   Atlantic. Deep Sea Res. (part 1 Oceanogr. Res. Pap.), 48, 217–236, 2001.
768
769 Glover, A., Smith, C. R., Paterson, G. J. L., Wilson, G. D. F., Hawkins, L. & Sheader, M.:
770   Polychaete species diversity in the central Pacific abyss: local and regional patterns,
771   and relationships with productivity, Mar. Ecol. Prog. Ser., 240, 157–170, 2002.
772

773 Gotelli, N. J. & Colwell, R., K.: Quantifying Biodiversity: Procedures and Pitfalls in the
774   Measurement and Comparison of Species Richness.", Ecol. Lett., 4, 379–91, 2001.
775   doi:10.1046/j.1461-0248.2001.00230.
776
777 Hannides, A., Smith, C. R.: The northeast abyssal Pacific plain. In: Biogeochemistry of
778   Marine Systems, K. B. Black and G. B Shimmield, eds., CRC Press, Boca Raton,
779   Florida, 208-237, 2003.
780
781 Havermans, C.: Have we so far only seen the tip of the iceberg? Exploring species diversity
782   and distribution of the giant amphipod Eurythenes, Biodiversity, 2016.
783 DOI: 10.1080/14888386.2016.1172257
784
785 Havermans, C & Smetacek, V. Bottom-up and top-down triggers of diversification: A new
786   look at the evolutionary ecology of scavenging amphipods in the deep sea. Progress in
787   Oceanogprahy, 164, 37-51, 2018.
788
789 Havermans, C., Sonet, G., d'Udekem d'Acoz, C., Nagy, Z. T., Martin, P., Brix, S., Riehl, T.,
790   Agrawal, S. & Held, C.: Genetic and morphological divergences in the cosmopolitan
791   deep-sea amphipod *Eurythenes gryllus* reveal a diverse abyss and a bipolar species.
792   PLoS ONE, 8, e74218, 2013.
793
794 Held, C. & Wägele, J. W.: Cryptic speciation in the giant Antarctic isopod *Glyptonotus*
795   *antarcticus* (Isopoda: Valvirfera: Chaetiliidae), Sci. Mar. 69 (Suppl. 2), 175–181,
796   2005.
797
798 Highsmith, R.: Floating and algal rafting as potential dispersal mechanisms in brooding
799   invertebrates, Mar. Ecol. Prog. Ser., 25, 169–179, 1985.
800
801 Hilário, A., Metaxas, A., Gaudron, S. M. ,Howell, K. L., Mercier, A., Mestre, N. C., Ross, R.
802   E. ,Thurnherr, A. M. & Young, C.: Estimating dispersal distance in the deep
803   sea:challenges and applications to marine reserves, Front. Mar. Sci., 2, 2015.
804   doi:10.3389/ fmars.2015.00006.

805 Ide, K.,Takahashi, K., Nakano, T., & Minoru, S. & Omori, M. Chemoreceptive foraging in a
806   shallow-water scavenging lysianassid amphipod: Role of amino acids in the location
807   of carrion in Scopelocheirus onagawae. Marine Ecology-progress Series, 2006.
808 MAR ECOL-PROGR SER. 317. 193-202. 10.3354/meps317193.
809
810 International Seabed Authority. A Discussion Paper on Developing a Regulatory Framework
811   for Mineral Exploitation in the Area (Env. Matt.), 1–102, 2017.
812
813 Kankaanpää, H., Laurén, M., Mattson, M. & Lindström, M.: Effects of bleached kraft mill
814   effluents on the swimming activity of Monoporeia affinis (Crustacea, Pmphipoda)
815   lindström, Chemosphere, 31, 4455–4473, 1995.
816
817 Laver, M. B., Olsson, M. S., Edelman, J. L. & Smith K. L. Jr.: Swimming rates of scavenging
818   deep−sea amphipods recorded with a free−vehicle video camera. Deep−Sea Res. II,
819   32, 1135–1142,1985.
820
821 Lester, S. E., Ruttenberg, B. I., Gaines, S. D. & Kinlan B. P.: The relationship between
822   dispersal ability and geographic range size. Ecol. Lett, 10, 745–758, 2007.
823
824 Levin, L. A., Etter, R.J., Rex, M. A., Gooday, A. J., Smith, C. R., Pineda, J., Stuart, C. T.,
825   Hessler, R. R. & Pawson, D.: Environmental influences on regional deep-sea species
826   diversity, Annu. Rev. Ecol. Evol. Syst., 32, 51–93, 2001.
827
828 Liow, L. H.: Does versatility as measured by geographic range, bathymetric range and
829   morphological variability contribute to taxon longevity?, Glob. Ecol. Bio., 16, 117–
830   128, 2007.
831
832 Lodge, M., Johnson, D., Le Gurun, G., Wengler, M., Weaver, P. & Gunn, V.: Seabed mining:
833   International Seabed Authority environmental management plan for the Clarion–
834   Clipperton Zone. A partnership approach, Mar. Pol., 49, 66–72, 2014.
835
836 Lonsdale, P.: Clustering of suspension-feeding macrobenthos near abyssal hydrothermal
837   vents at oceanic spreading centers, Deep-Sea Res., 24, 857–863, 1977.

838   Lörz A-N, Jażdżewska AM, Brandt A. A new predator connecting the abyssal with the hadal

839   in the Kuril-Kamchatka Trench, NW Pacific. Costello M, ed. PeerJ. 2018;6:e4887.

840   doi:10.7717/peerj.4887.

841

842

843   Managing Impacts of Deep Sea Resource Exploitation (MIDAS) Summary Report:

844   Biodiversity in the Clarion-Clipperton Zone, 1–2, 2016.

845

846   McClain, C. R. & Hardy, S. M.: The dynamics of biogeographic ranges in the deep sea, Proc.

847   Roy. Soc. B: Bio. Sci., 277, 3533–3546, 2010.

848

849   Narahara-Nakano, Y., Nakano, T. & Tomikawa, K. Deep-sea amphipod genus Eurythenes

850   from Japan, with a description of a new Eurythenes species from off Hokkaido

851   (Crustacea: Amphipoda: Lysianassoidea), Marine Biodiversity, 1867-1616, 2017. Mar

852   Biodiv DOI 10.1007/s12526-017-0758-4

853

854   Paull, C. K., Hecker, B., Commeau, R., Freeman-Lynde, R. P., Neumann, C., Corso, W. P.,

855   Golubic, S., Hook, J. E., Sikes, E. & Curray, J.: Biological communities at the Florida

856   escarpment resemble hydrothermal vent taxa, Science, 226, 965–967, 1984.

857   Premke K, Muyakshin S, Klages M & Wegner J. Evidence for long-range chemoreceptive

858   tracking of food odour in deep-sea scavengers by scanning sonar data. J Exp Mar Biol

859   Ecol, 285:283–294, 2003.

860

861   R Core Team. R: A language and environment for statistical computing. R Foundation for

862   Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, 2013.

863   URL http://www.R-project.org/.

864

865   Ramirez-Llodra, E., Brandt, A., Danovaro, R., De Mol, B., Escobar, E., German, C. R.,

866   Levin, L. A., Martinez Arbizu, P., Menot, L., Buhl-Mortensen, P., Narayanaswamy,

867   B. E., Smith, C. R., Tittensor, D. P., Tyler, P. A., Vanreusel, A. & Vecchione, M.:

868   Deep, diverse and definitely different: unique attributes of the world's largest

869   ecosystem, Biogeosciences, 7, 2851–2899, 2010.

870

871 Ramirez-Llodra, E., Tyler, P. A., Baker, M. C., Bergstad, O. A., Clark, M. R. & Escobar, E.:

872 Man and the last great wilderness: human impact on the Deep Sea, PLoSONE, 6,

873 2011. doi:10.1371/journal.pone.0022588.

874

875 Rex, M. A., Stuart, C. T., Hessler, R., R., Allen, J. A., Sanders, H. L. & Wilson, G. D. F.:

876 Global-scale latitudinal patterns of species diversity in the deep-sea benthos, Nature,

877 365, 636–639, 1993.

878 Ritchie, H., Jamieson, A. J. & Piertney, S. B. Isolation and Characterization of Microsatellite

879 DNA Markers in the Deep-Sea Amphipod Paralicella tenuipes by Illumina MiSeq

880 Sequencing, Journal of Heredity, 367 – 371, 2016.

881

882 Sanders H. L.: Marine benthic diversity: a comparative study, Am. Nat., 102, 243–282, 1968.

883

884 Schön I, Pinto R. L., Halse, S., Smith, A. J. & Martens, K.: Cryptic Species in Putative

885 Ancient Asexual Darwinulids (Crustacea, Ostracoda), PLoS ONE, 7, e39844.

886 doi:10.1371/journal.pone.0039844. 2012.

887

888 Schulenberger, E. & Barnard, J. L.: Clarification of the Abyssal Amphipod, *Paralicella*

889 *tenuipes* Chevreux, Crustaceana, 31, 267–274, 1976.

890

891 Schüller, M. & Ebbe, B.: Global distributional patterns of selected deep−sea Polychaeta

892 (Annelida) from the Southern Ocean, Deep−Sea Res. II, 54, 1737–1751, 2007.

893

894 Simpson, E. H.: Measurement of diversity, Nature, 163, 688, 1949.

895

896 Smith, C. R., Kukert, H., Wheatcroft, R. A., Jumars, P. A. & Deming, J. W.: Vent fauna on

897 whale remains, Nature, 341, 27–28, 1989.

898

899 Smith, C. R. & A.W. J. Demopoulos.: Ecology of the deep Pacific Ocean floor. In:

900 Ecosystems of the World Volume 28: Ecosystems of the Deep Ocean, P. A. Tyler,

901 ed., Elsevier, Amsterdam, pp. 179–218, 2003.

902

903   Smith, C. R., Drazen J. & Mincks, S. L.: Deep-sea Biodiversity and Biogeography:
904   Perspectives from the Abyss. International Seabed Authority Seamount Biodiversity
905   Symposium, 1–13, 2006.
906

907   Somero, G. N.: Adaptations to high hydrostatic pressure, Ann. rev. physiol., 54, 57–577.
908   1992.
909

910   Stuart, C., Rex, M. & Etter, R.: Large scale spatial and temporal patterns of deep-sea
911   biodiversity. Ecosystems of the World Volume 28: Ecosys. Deep Oc., P. A. Tyler, ed.,
912   Elsevier, Amsterdam, 295–311, 2003.
913

914   Taguchi Y. H. & Oono, Y.: Relational patterns of gene expression via non-metric
915   multidimensional scaling analysis, Bioinformatics, 21, 730–740, 2005.
916

917   Thiel, H.: Deep-sea Environmental Disturbance and Recovery Potential, Int. Revue ges.
918   Hydrobiol. Hydrogr., 77, 331–339, 1992. doi:10.1002/iroh.19920770213.
919

920   Thurston, M. H.: Abyssal necrophagous amphipods (Crustacea: Amphipoda) in the northeast
921   and tropical Atlantic Ocean, Prog. Oceanogr., 24, 257–274, 1990. doi:10.1016/0079-
922   6611(90)90036-2.
923

924   Vader, W.: How many amphipods species? 6th International Crustacean Congress, Glasgow,
925   Scotland, 18–22 July 2005, 143, 2005.
926

927   Wilson, G. D. F., Hessler R.: Speciation in the deep sea, Ann. Rev. Ecol. Syst., 18, 185–207,
928   1987.
929

930   Zardus, J. D., Etter, R. J., Chase, M. R., Rex, M. A. & Boyle, E. E. Bathymetric and
931   geographic population structure in the pan-Atlantic deep-sea bivalve *Deminucula*
932   *atacellana* (Schenck, 1939), Mol. Ecol., 15, 639–651, 2006.
933

934

Figure captions

Figure 1: Geographic locations of the two study areas, the Clarion-Clipperton fracture Zone (CCZ) (Northeast Pacific) and the DISturbance and re-COLonisation (DisCOL) Experimental Area (DEA) (Peru Basin, Southeast Pacific). There are nine Areas of Particular Ecological Interest (APEIs) in the CCZ region, which are illustrated by 400 x 400 km^2 white boxes. Grey boxes indicate the various contractor claim areas in the CCZ. We deployed eight amphipod traps across the CCZ, which is 16,000 km^2 and 7000 km wide, and five in the DEA, which encompasses 11 km^2 with a width of 4 km.

Figure 2: **Histogram showing the species assemblage for the scavenging community in the Clarion-Clipperton fracture Zone (CCZ) (black) and the DisCOL Experimental Area (DEA) (grey)**. The abundances of 19 morphotypes are shown.

Figure 3: Catch Per Unit Effort (CPUE) illustrating the correlation between sampling time and number of individuals collected. Only the > 15 mm fraction was included here to estimate number of collected individuals**.**

Figure 4: Species rarefaction curves for each of the 13 trap stations across both areas, the Clarion-Clipperton fracture Zone and the DisCOL Experimental Area. Only individuals longer than 15 mm were considered here.

Figures 5a and b: Relative species abundances in the Clarion-Clipperton fracture Zone and the DisCOL Experimental Area. These abundances represent the greater than 15mm fraction of the scavenging amphipod community only.

Figure 6: NMDS plot showing the beta biodiversity (dis/similarities) for each of the thirteen amphipod trap sampling stations associated with the two basins, Clarion-Clipperton fracture Zone (CCZ) (black) and the DisCOL Experimental Area (DEA) (red). Data are supported by a low stress value of 0.01.

Table captions

Table 1: Station overview. Codes refer to the codes used in this paper in figures 1, 3, 4, 6 and Table 3. The original station code represents the cruise codes from (SO239 and SO232-1). Depth refers to water depth (m) on deployment. Nodule presence/absence information is known only for stations D3 and D4.

Table 2a: Overview of morphotypes across the Clarion-Clipperton fracture Zone (CCZ) and DisCOL Experimental Area (DEA).

Table 2b: Overview of morphotypes across the Clarion-Clipperton fracture Zone (CCZ) and DisCOL Experimental Area (DEA).

Table 3: Comparison of biodiversity calculated using the Simpson Index (D), for the Clarion-Clipperton fracture Zone and DisCOL Experimental Area, and D for each station is shown for further comparisons within these areas.

Appendix/Electronic Supplementary Information (ESM) captions

Appendix 1: Multibeam scan - Showing the location of the 78 track marks created by the plough harrow in the DisCOL Experimental Area to simulate manganese nodule extraction activity (D1)

Appendix 2 – Photograph showing the baited free-fall lander trap designed and deployed by RBINS.

Appendix 3 - Side-scan sonar image of site D5 showing possible seamount barriers. View from NW (top) to SE (bottom). Contours are every 25 m. (Source: GEOMAR, 2015).

Appendix 4 – Calculation of alpha biodiversity used in this manuscript.

1016 **Appendix 1: Multibeam scan** - Showing the location of the 78 track marks created by the
1017 plough harrow in the DisCOL Experimental Area to simulate manganese nodule extraction
1018 activity.

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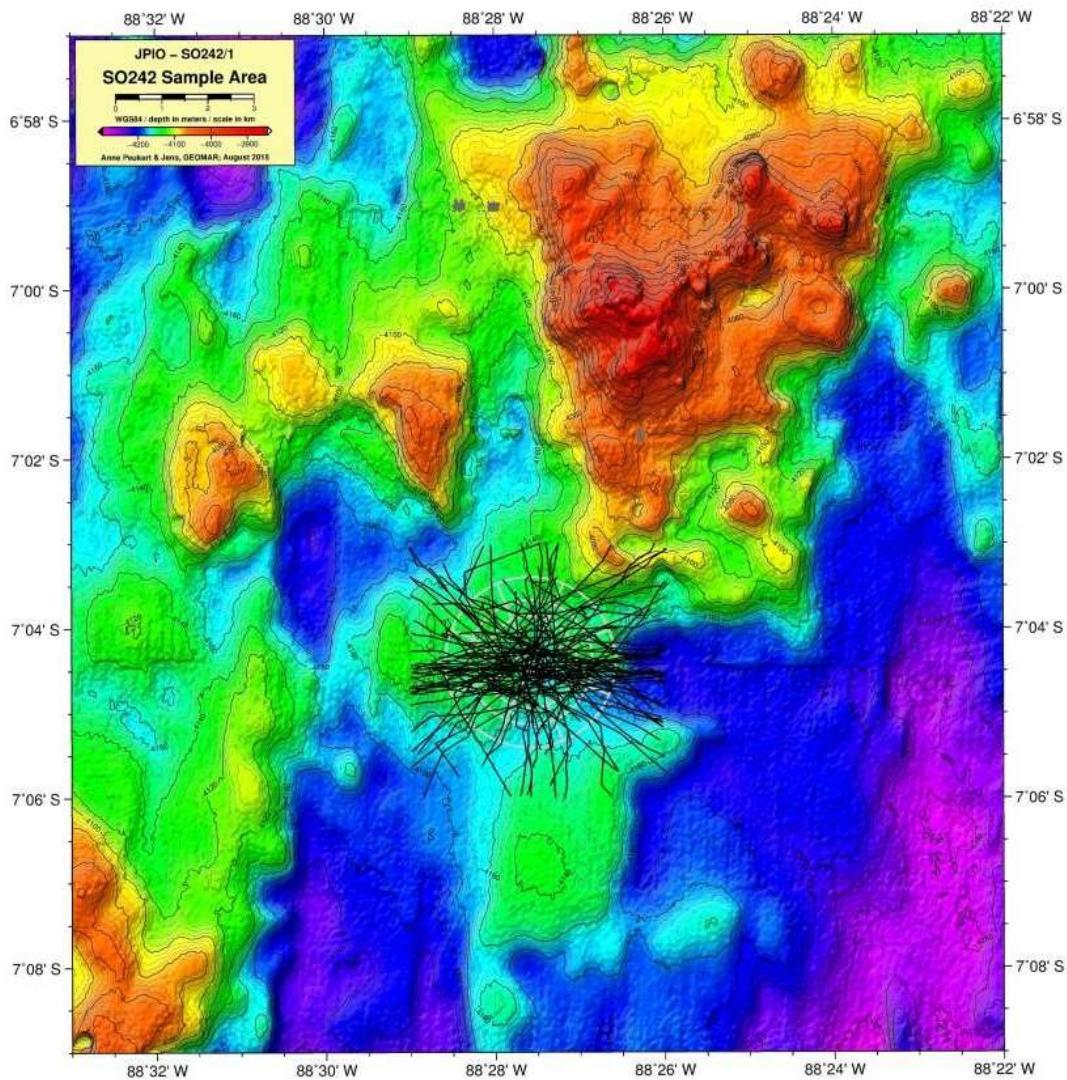
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1028   **Appendix 2** – Photograph showing the baited free-fall lander trap designed and deployed by
1029   RBINS.

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1043   **Appendix 3** - Side-scan sonar image of site D5 showing possible seamount barriers. View
1044   from NW (top) to SE (bottom). Contours are every 25 m. (Source: GEOMAR, 2015).

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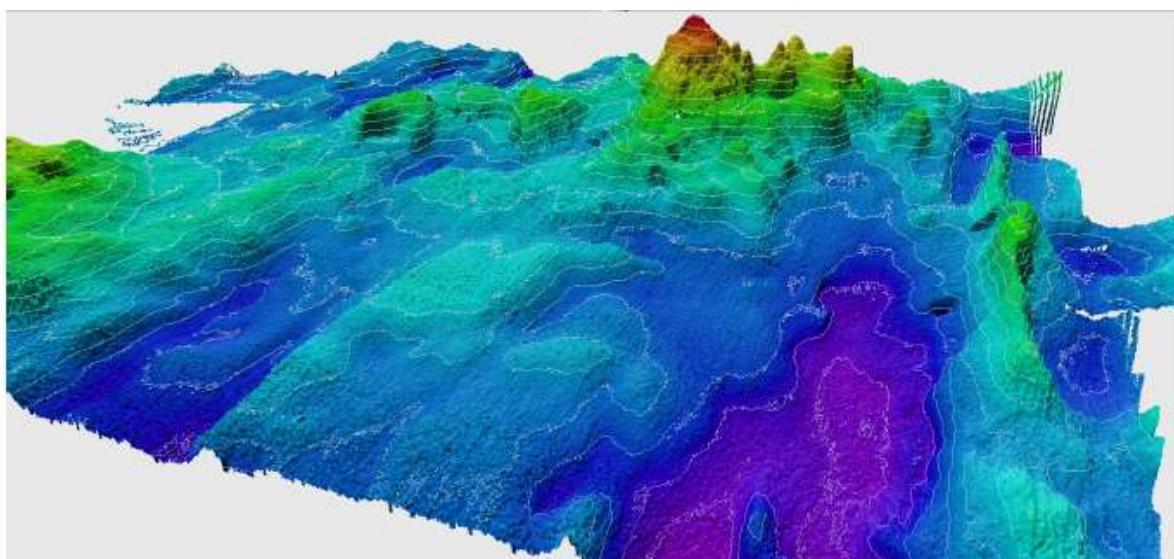
1057   **Appendix 4** - Calculation of alpha biodiversity used in this manuscript.

1058   Simpson Index

1059   Simpson Diversity Index (D) = $1 - \dfrac{\sum n(n-1)}{N(N-1)} =$

1060   D = Diversity Index

1061   n = number of individuals in each particular species

1062   N = Total number in community

1063   A high value of D indicates a high species diversity.

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