

Cruise Report – FK141109
Studying the Biology and Geology of the Mariana Trench: the deepest place
on earth (aka HADES-M)

Nov 9 to Dec 9, 2014

Executive summary

We had two overarching goals for this project: (1) to describe the patterns of hadal community structure and relate these patterns to environmental and physiological factors; and (2) to sample rocks, mud, and subducting-slab-derived fluids to understand crustal cycling, controls over seismicity and tsunamigenesis, and the evolution of continental crust. After the loss of HROV Nereus in May 2014 we modified our cruise plan to focus on sampling the biology and geology of the trench using landers. We used two baited cameras, two baited traps to sample the biota, an SOI lander fitted with Van Veen grabs to sample rocks and sediments and two 30l Niskin bottles to sample bottom water, and an SOI lander fitted with a coring respirometer to sample sediments and measure the activity of the sediment community. The Falkor's CTD was also used to sample water to a depth of 5000m on several occasions.

92 lander deployments and 4 CTD casts were completed on a transect beginning at roughly 5km depth south of Guam on the overriding plate, descending to the Sirena Deep at nearly 11km and back up the subducting Pacific plate to a depth of 5km. A few problems were encountered on the cruise. The hadal camera arrived but had been damaged during shipping but another camera was purchased and brought out to the ship by a small boat. The ship's radio directional finder, recently purchased, stop working but technician Paul Duncan fortunately had a hand held unit so we could listen for the landers surfacing beacons. Finally, on its sixth deployment the free vehicle coring respirometer (FVCR) was lost, presumed anchored in the mud.

Despite challenges the cruise was a resounding success. Prior study focused almost solely on the Challenger deep, so our cruise provided an unprecedented picture of the world's most famous and deepest trench, from top to bottom. Six measurements of sediment community oxygen consumption were made at 6800 and 7900m and the rates are perhaps the highest ever measured in the deep sea. This implies that there is a considerable amount of organic material in the sediments fueling this activity. Analysis of the sediment cores for microbial biomass, organic matter and meiofauna and macrofauna will proceed post cruise. Sediments were also collected by the rock grabber from 5025-10525m and these were partitioned for faunal, microbial, organic and genetic analyses.

Baited imaging landers and traps were very successful and provide some initial highlights. We documented the first snailfish and princaxellid amphipods in the Mariana trench. The common species of snailfish appears, upon inspection of captured specimens, to be a new species. In addition we made observations of the deepest living fish ever recorded at 8143m, a completely new species of snailfish. Video of this species was also made by the FVCRs task camera. Other depth records were made for rattail fishes, eelpouts, and the amphipod *Scopelocheirus* sp. The baited traps captured many of the animals seen in the imagery allowing for morphological verification of identifications. These samples also provide the basis for a variety of life history, genetic, reproductive, foodweb, and physiological studies.

Osmolality of fish blood was measured aboard to test the theory that osmoregulation in combination with the accumulation of piezolytes controls the lower depth limits of some animals including fishes. Measurements on the new Mariana snailfish confirmed the expectation that fishes become isosmotic at about 8200m and cannot live below that depth. In addition our imagery and trapping only found fishes to 8143m.

Depth zonation of the fauna in the Mariana trench is clearly evident. At shallower depths large fishes dominate and amphipods are sparser. At the middle trench, snailfish and giant amphipods are present. In the lower trench predatory prinxacellid amphipods and high densities of smaller amphipods such as *Hirondellia* and *Scopelocheirus* dominate. It is our working hypothesis that the lower depth limits of these animals are set by physiological capabilities but the upper limits are likely determined by predator-prey or competitive interactions much as intertidal community zonation is determined.

The deepest rock samples ever recovered from the Mariana forearc region were collected at 8720m by the rock grabber. These will be dated and analyzed in the laboratory. Samples of manganese nodules and altered pumice were recovered at 6957m on the subducting plate and rounded volcanic pebbles were recovered from 5025m. These samples may point to recent underwater volcanic activity from the nearby Caroline ridge, which points to underappreciated extent of this activity, but confirmation awaits laboratory analysis.

Finally, our unprecedented sample collections will now form the basis of considerable laboratory analysis which will likely result in many more important discoveries. Clearly, this cruise has drawn the first comprehensive picture of the Mariana Trench

Table of Contents

pg

Executive Summary	1
Science party details	4
Sampling map	5
Sediment sampling	
Community ecology and trophic function of sediment macrofauna and meiofauna residing in hadal environments. Jill Bourque (USGS)	6
Free Vehicle Coring Respirometer (FVCR) - Sediment Respiration Measurements. Clifton Nunnally (University of Hawaii)	11
Investigating the origin(s) and fate of sediment organic matter and bacterial biomass in the deepest marine environments on Earth. Eleanna Grammatopoulou (University of Aberdeen, Oceanlab)	15
Image sampling	
Scavenger assemblages investigated with the Hadal-Lander. Alan Jamieson, Thomas Linley (Oceanlab, University of Aberdeen)	21
Scavenging communities and environmental data from the Abyssal lander Thomas Linley (Oceanlab, University of Aberdeen)	27
Stand Alone Imaging Module – SAIM. Thomas Linley (Oceanlab, University of Aberdeen)	33
Animal Sampling	
Summary of fish captures. Mackenzie Gerringer (U. of Hawaii)	36
Photogrammetry. Thomas Linley (Oceanlab, University of Aberdeen)	38
Amphipods. Stuart Piertney (University of Aberdeen)	39
Reproduction in amphipods. Caitlin Plowman (Oregon Institute of Marine Biology (under Dr. Craig Young)	41
Investigations into pressure adaptations of hadal animals. Paul H. Yancey, Anna Downing, and Chloe Weinstock (Whitman College)	45
Microbial sampling and culturing. Logan Peoples (Scripps Institution of Oceanography)	48
Geological transect across the Mariana Trench at Sirena Deep. Patty Fryer (University of Hawaii)	51
Educational Outreach. Linda Tatreau	55
Media Summary from the Mariana Trench Press Release – Carlie Wiener	56

Table 1. Science party details for FK141109.

Name	Affiliation	General Research Area	Email Address
Jeff Drazen	Univ of Hawaii	physiology	jdrazen@hawaii.edu
Patty Fryer	Univ of Hawaii	Geology, multibeam	pfryer@hawaii.edu
Alan Jamieson	Univ of Aberdeen	megafauna landers/elevators	a.jamieson@abdn.ac.uk
Timothy Shank*	WHOI	comm structure and genetics	tshank@whoi.edu
Paul Yancey	Whitman College	osmolytes	yancey@whitman.edu
Stuart Piertney	Univ. of Aberdeen	Amphipod genetics	s.piertney@abdn.ac.uk
Clif Nunnally	Univ of Hawaii	FVCR, sediment processing	cnunn@hawaii.edu
Mackenzie Gerringer	Univ of Hawaii	physiology	mgerringer@hawaii.edu
Thomas Linley	Univ of Aberdeen	megafauna landers/elevators	t.linley@abdn.ac.uk
Eleanna Grammatopoulou	Univ of Aberdeen	sediment processing, CTD	r01eg13@abdn.ac.uk
Linda Tatreau	Guam – DOE	Education and outreach	lindian@teleguam.net
Chloe Weinstock	Whitman College	Osmolytes	weinstcl@whitman.edu
Caitlin Plowman	OIMB	Reproductive biology	cplowman@uoregon.edu
Anna Downing	Whitman College	Osmolytes	downinab@whitman.edu
Logan Peoples	SIO	Microbial culturing/comm. struc	peepsoccer2@gmail.com
Casey Machado	WHOI	Landers	cmachado@whoi.edu

* participated from shore.

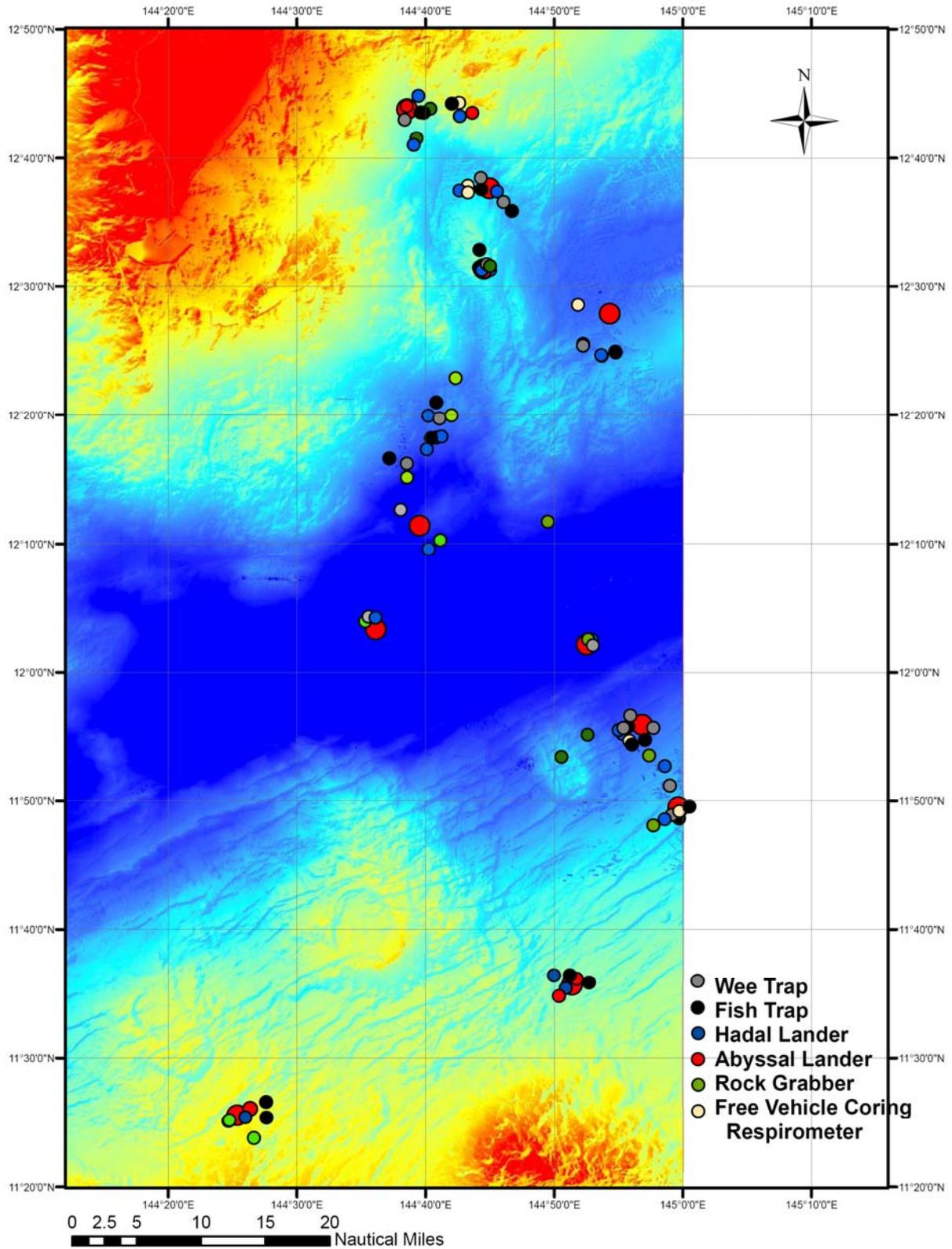


Figure 1. Sampling map for FK141109 showing 92 lander deployment locations.

Community ecology and trophic function of sediment macrofauna and meiofauna residing in hadal environments

Jill R. Bourque, U.S. Geological Survey

Objectives/Goals with Background

For vast portions of the deep sea, there is a strong relationship between decreased food supply (POC flux) and increased depth, which is associated with declining biomass of benthic organisms as food supply diminishes. This paradigm may be decoupled in hadal environments, often referred to as “depocenters”, where substantial quantities of POC can be concentrated on the sediment surface, relative to surrounding abyssal plain, possibly fueling high production of bacteria and higher order organisms. Trenches represent extremely heterogeneous environments, due to their topographic complexity, where organic matter is channeled and trapped on the seafloor. This high organic content can be an important driver for benthic communities, potentially promoting biomass for all groups from microbes to megafauna. They also represent extreme environments in terms of temperature and pressure. Thus, there are a variety of extrinsic and intrinsic factors that influence the community ecology within and among trenches. There are potential differences in sediment faunal communities residing in trenches below highly productive surface waters and adjacent to continental land masses, versus oligotrophic waters. Basic knowledge of the sediment community structure and functional attributes of benthos residing in deep-sea trenches is yet unknown. Key functional groups linking microbes to the megabenthos include sediment meiofauna (> 20 μm) and macrofauna (>300 μm). Our study is interested in examining how food quality and supply and pressure influence the sediment meiofaunal and macrofaunal community structure and function.

The project goal is to improve understanding of benthic macrofaunal and meiofaunal communities that reside in hadal environments. Specific objectives include:

1. To examine sediment meiofaunal and macrofaunal community structure (e.g., diversity, abundance, composition) and trophic ecology of infauna along a depth gradient using identification of functional groups through morphological analyses.
2. To couple the infaunal community analyses to other biological and environmental parameters, including estimates of bacterial and megafaunal biomass, community respiration, geological properties, and POC flux in order to develop a comprehensive understanding of the structure and function of hadal biological communities from microbes to megafauna.
3. Use phylogenetic and population genetic diversity of benthic infauna to assess levels of genetic divergence and evolutionarily independent lineages to assess the role of depth and topography in trenches in promoting the formation of species.

Sample Summary

Sediment Cores:

We successfully collected 6 cores over 3 deployments with the FVCR, ranging in depth from 6844-7942m. All cores were vertically sectioned at 1-cm intervals down to 10 cm, and 5-cm intervals thereafter. Two of the cores were split in half to maximize information output. The fate of each of the cores is shown in Table 2. Sediments in excess of 20 cm in 5 of the collected cores were preserved frozen for genetic analysis (TS/WHOI).

Table 2. Disposition of sediment cores collected in the Mariana Trench

Research Group	Analysis	Cores processed
Dan Mayor/University of Aberdeen	Sediment Organics/Bacterial Biomass	3.5
Amanda Demopoulos/USGS	Meiofaunal/Macrofaunal Community	1
Amanda Demopoulos/USGS	Pore Water Salinity/Redox	1*
Doug Bartlett/SIO	Microbial/Metagenomics	0.5
Patty Fryer/University of Hawaii	Sedimentology	0.5
Tim Shank/WHOI	Meiofaunal/Macrofaunal Genetics	0.5

*Subsamples of sedimentological/microbial cores

Grab and additional samples:

12 deployments of the grab samplers were successful in returning sediment from the trench environment (Table 3). Depths of grab samples ranged from 5025-10525m, encompassing the forearc area and the Pacific plate. 16 sediment samples were collected from the 12 deployments for community analysis and preserved in formalin, 14 sediment samples were collected for community genetic analysis and preserved in ethanol, and 13 sediment samples were collected for community genetic analysis and frozen at -80°C.

Additional sediment samples were collected from the FVCR in the form of sediment retained in one of the lander legs, and sediment that was retained on the core catchers. 2 samples were preserved each for genetic community analysis and faunal community analysis from sediment retained on the core catchers at 6844m and 7939m. An additional sample for both community and genetic analysis was preserved from the sediment in the lander leg at 6833m.

Preliminary Assessment:

A brown, globular organism (Figure 2) was collected from 8518m and preserved for genetic analysis. No other obvious macro or meiofauna were observed during processing of any of the sediment samples, so any community results will have to wait until samples are processed in the laboratory. However, there were noted differences in the sediment composition among our sampling locations (see sedimentological section). Sediment physical characteristics are known to affect faunal community composition, so we expect differences among these sediments regardless of differences in depth, organic supply, and location. The one core analyzed for redox potential suggests that sediments at 7942m were well oxygenated down to at least 10 cm.

Table 3. Collection information for grab and additional sediment samples.

Date	Gear	Site	Depth	Source	Formalin	Ethanol	Frozen
11/13/2014	RG02	1	6065	Grab	x	x	
11/14/2014	CR02	3	6844	Core Catcher	x	x	
11/17/2014	CR03	3	6833	Lander Leg	x	x	
11/19/2014	CR04	04B	7939	Core Catcher	x	x	
11/22/2014	RG05	6	7633	Grab	x	x	x
11/22/2014	RG06	6	6877	Grab	x	x	x
11/25/2014	RG07	7	6957	Grab	x	x	x
11/26/2014	RG08	7	7660	Grab	x	x	x
12/1/2014	RG10	09A	5025	Grab	x	x	x
12/2/2014	RG11	10	9067	Grab	x	x	x
12/3/2014	RG12	10A	8518	Grab	x	x	x
12/4/2014	RG13	11	8750	Grab	x	x	x
12/4/2014	WT13	11	8847	Grab	x		
12/5/2014	RG14	12	10525	Grab	x	x	x
12/5/2014	WT14	12	10250	Grab	x	x	x



Figure 2. Brown, globular organism collected from RG12 at 8518m. Photo: Paul Yancey

Sample IDs/list

All formalin-preserved samples collected by USGS are curated at the U.S. Geological Survey Southeast Ecological Science Center, 7920 NW 71st St, Gainesville, FL, USA. Each fraction of each core and individual subsamples of grab samples has been assigned a unique ID and registered in a sample database maintained by the Demopoulos Lab. Each core was assigned a

UH catalog number, and all USGS IDs have been cross-referenced to its associated UH number. A full list of samples curated by USGS is shown in Table 4.

All ethanol preserved and frozen sediment samples collected by WHOI are curated at the Woods Hole Oceanographic Institution, 2-34/2-40 Redfield Laboratory, MS#33, Woods Hole, MA, 02543. Each samples is registered with the UH catalog number and a full list of sediment samples curated by WHOI is shown in Table 5.

Table 4. Sample of inventory for USGS curated samples. (temporary)

SampleID	Date	Gear	Site	Core	Depth	Type	Formalin	UH Number
FK-2014-200010	11/13/2014	RG02	1	RGY	6065	Sediment - grab	x	200010
FK-2014-200018	11/14/2014	CR02	3	MC3	6844	Core - macrofauna	x	200018
FK-2014-200030	11/17/2014	CR03	3	LL1	6833	Sediment - grab	x	200030
FK-2014-200057	11/19/2014	CR04	04B	MC3	7939	Core - macrofauna	x	200057
FK-2014-200067a	11/22/2014	RG05	6	RGY	7633	Sediment - grab	x	200067
FK-2014-200067b	11/22/2014	RG05	6	RGY	7633	Sediment - grab	x	200067
FK-2014-200068a	11/22/2014	RG06	6	RGY	6877	Sediment - grab	x	200068
FK-2014-200068b	11/22/2014	RG06	6	RGY	6877	Sediment - grab	x	200068
FK-2014-200075a	11/23/2014	CR05	6	MC1	7942	Core - macrofauna	x	200075
FK-2014-200075b	11/23/2014	CR05	6	MC1	7942	Core - macrofauna	x	200075
FK-2014-200075c	11/23/2014	CR05	6	MC1	7942	Core - macrofauna	x	200075
FK-2014-200075d	11/23/2014	CR05	6	MC1	7942	Core - macrofauna	x	200075
FK-2014-200075e	11/23/2014	CR05	6	MC1	7942	Core - macrofauna	x	200075
FK-2014-200075f	11/23/2014	CR05	6	MC1	7942	Core - macrofauna	x	200075
FK-2014-200075g	11/23/2014	CR05	6	MC1	7942	Core - macrofauna	x	200075
FK-2014-200075h	11/23/2014	CR05	6	MC1	7942	Core - macrofauna	x	200075
FK-2014-200075i	11/23/2014	CR05	6	MC1	7942	Core - macrofauna	x	200075
FK-2014-200075j	11/23/2014	CR05	6	MC1	7942	Core - macrofauna	x	200075
FK-2014-200089	11/25/2014	RG07	7	RGB	6957	Sediment - grab	x	200089
FK-2014-200090	11/26/2014	RG08	7	RGY	7660	Sediment - grab	x	200090
FK-2014-200091	11/26/2014	RG08	7	RGB	7660	Sediment - grab	x	200091
FK-2014-200109a	12/1/2014	RG10	09A	RGB	5025	Core - macrofauna	x	200109
FK-2014-200109b	12/1/2014	RG10	09A	RGB	5025	Core - macrofauna	x	200109
FK-2014-200109c	12/1/2014	RG10	09A	RGB	5025	Sediment - grab	x	200109
FK-2014-200110	12/1/2014	RG10	09A	RGY	5025	Sediment - grab	x	200110
FK-2014-200116a	12/2/2014	RG11	10	RGY	9067	Core - macrofauna	x	200116
FK-2014-200116b	12/2/2014	RG11	10	RGY	9067	Core - macrofauna	x	200116
FK-2014-200116c	12/2/2014	RG11	10	RGY	9067	Sediment - grab	x	200116
FK-2014-200116d	12/2/2014	RG11	10	RGY	9067	Sediment - grab	x	200116
FK-2014-200117	12/2/2014	RG11	10	RGB	9067	Sediment - grab	x	200117
FK-2014-200120a	12/3/2014	RG12	10A	RGY	8518	Core - macrofauna	x	200120
FK-2014-200120b	12/3/2014	RG12	10A	RGY	8518	Core - macrofauna	x	200120
FK-2014-200120c	12/3/2014	RG12	10A	RGY	8518	Sediment - grab	x	200120
FK-2014-200121	12/3/2014	RG12	10A	RGB	8518	Sediment - grab	x	200121
FK-2014-200125a	12/4/2014	RG13	11	RGY	8750	Core - macrofauna	x	200125
FK-2014-200125b	12/4/2014	RG13	11	RGY	8750	Core - macrofauna	x	200125
FK-2014-200125c	12/4/2014	RG13	11	RGY	8750	Sediment - grab	x	200125
FK-2014-200126	12/4/2014	WT13	11	RGY	8847	Sediment - grab	x	200126

Table 5. Sample of sediment-derived inventory for WHOI curated samples.

UH Number	Date	Gear	Site	Core	Depth	Type	Fraction	Ethanol	Frozen
200010	11/13/2014	RG02	1	-	6065	Animal	-	x	
200016	11/14/2014	CR02	3	MC4	6844	Core	20-26		x
200016	11/14/2014	CR02	3	MC4	6844	Core	26-31		x
200016	11/14/2014	CR02	3	MC4	6844	Core	31-35		x
200017	11/14/2014	CR02	3	MC2	6844	Core Catcher	-	x	
200030	11/17/2014	CR03	3	-	6833	Lander Leg	-	x	
200055	11/19/2014	CR04	04B	MC4	7939	Core	20-25		x
200055	11/19/2014	CR04	04B	MC4	7939	Core	25-29		x
200056	11/19/2014	CR04	04B	MC2	7939	Core Catcher	-	x	
200067	11/22/2014	RG05	6	-	7633	Grab	-	x	x
200068	11/22/2014	RG06	6	-	6877	Grab	-	x	x
200075	11/23/2014	CR05	6	MC1	7942	Core	10-15		x
200075	11/23/2014	CR05	6	MC1	7942	Core	15-20		x
200075	11/23/2014	CR05	6	MC1	7942	Core	20-25		x
200076	11/23/2014	CR05	6	MC2	7942	Core	0-1	x	
200076	11/23/2014	CR05	6	MC2	7942	Core	1-2	x	
200076	11/23/2014	CR05	6	MC2	7942	Core	2-3	x	
200076	11/23/2014	CR05	6	MC2	7942	Core	3-4	x	
200076	11/23/2014	CR05	6	MC2	7942	Core	4-5	x	
200076	11/23/2014	CR05	6	MC2	7942	Core	5-6	x	
200076	11/23/2014	CR05	6	MC2	7942	Core	6-7	x	
200076	11/23/2014	CR05	6	MC2	7942	Core	7-8	x	
200076	11/23/2014	CR05	6	MC2	7942	Core	8-9	x	
200076	11/23/2014	CR05	6	MC2	7942	Core	9-10	x	
200076	11/23/2014	CR05	6	MC2	7942	Core	10-15		x
200076	11/23/2014	CR05	6	MC2	7942	Core	15-20		x
200076	11/23/2014	CR05	6	MC2	7942	Core	20-27		x
200077	11/23/2014	CR05	6	MC3	7942	Core	20-24		x
200089	11/25/2014	RG07	7	-	6957	Grab	-	x	x
200090	11/26/2014	RG08	7	-	7660	Grab	-	x	x
200091	11/26/2014	RG08	7	-	7660	Grab	-	x	x
200109	12/1/2014	RG10	09A	-	5025	Grab	-	x	x
200116	12/2/2014	RG11	10	-	9067	Grab	-	x	x
200117	12/2/2014	RG11	10	-	9067	Grab	-	x	x
200120	12/3/2014	RG12	10A	-	8518	Grab	-	x	x
200121	12/3/2014	RG12	10A	-	8518	Grab	-	x	x
200122	12/3/2014	RG12	10A	-	8518	Animal	-	x	
200125	12/4/2014	RG13	11	-	8750	Grab	-	x	x
200128	12/5/2014	RG14	12	-	10525	Grab	-	x	x
200130	12/5/2014	WT14	12	-	10250	Grab	-	x	x

Free Vehicle Coring Respirometer (FVCR) - Sediment Respiration Measurements

Clifton Nunnally, University of Hawaii

Sediment community oxygen consumption (SCOC) was measured in situ using the FVCR lander during the HADES-Mariana cruise aboard the R/V Falkor. The FVCR was deployed six times (Table 6) but was only recovered after five. Details of the vehicle loss were recorded previously in a full report filed by lead Marine Technician Leighton Rolley.

Deployments CR01 and CR03 resulted in no cores collected or incubation experiments made. Deployments CR02 and CR04 recovered a single core only on which the lone successful SCOC measurement was made. CR05 accounted for the first fully successful deployment of the FVCR in which all 4 cores recovered mud and successfully conducted SCOC in situ measurements.

SCOC as measured in the collected cores was high on both sides of the trench. Single core measurements on the overriding plate were 2.00 and 2.10 $\text{mmol O}_2 \text{ m}^{-2} \text{ d}^{-1}$, at 6844 and 7939 meters respectively (Table 7). The average SCOC of four cores collected from 7942 meters on the downriding plate was 2.35 (SE \pm 2.79) $\text{mmol O}_2 \text{ m}^{-2} \text{ d}^{-1}$ (Table 6). These values are equivalent to 1.7, 1.8 and 1.9 $\text{mmol C m}^{-2} \text{ d}^{-1}$ remineralized by the sediments. The FVCR sites from the overriding plate returned dark, brown mud with little to no lamination of sediments (Figure 3). This contrasted to the heavily laminated sediments of the downriding plate which had alternating gray and brown layers of unequal bands (Figure 4).

The measured SCOC rates at 7 and 8 kilometer stations within the Mariana Trench account for 3 times the amount of oxygen consumed at abyssal depths in the central north Pacific (Appendix, Smith et al.). They are much closer to SCOC rates measured at 500 meters off of the organic rich Mississippi River delta (Appendix, Rowe et al.). In the failed cores respiration of muddy water accounted for very low oxygen consumption (26.36 and 150.78 $\mu\text{mol O}_2 \text{ m}^{-2} \text{ d}^{-1}$) which is much closer to the calculated oxygen consumption using microelectrode profiles of oxygen depletion in sediments of the Challenger Deep (Appendix, Glud et al.). Control rates were tiny compared to core respiration throughout all FVCR deployments, ranging from 1.80 to 17.43 $\mu\text{mol O}_2 \text{ d}^{-1}$ (Table 2), indicating very little microbial respiration in near bottom trench water.

Table 6. Details and the general results for the Free Vehicle Coring Respirometer deployments during theHADES-Mariana cruise.

STATION	DATE	LATITUDE	LONGITUDE	DEPTH	RESULT
CR01	10-Nov	12.73721	144.71625	4477	Landed on hard bottom; no cores.
CR02	12-Nov			6844	1 core collected with dark brown mud.
		12.63117	144.72149		
CR03	15-Nov	12.62229	144.72129	6833	Drive motor failure; no cores.
CR04	18-Nov			7939	1 core collected with dark brown mud.
		12.47649	144.86485		
CR05	21-Nov			7942	4 cores collected; gray and brown mud
		11.9118	144.9308		
CR06	24-Nov	11.8203	144.9956	6970	FVCR not recovered

Table 7. Results of SCOC measurements made by the FVCR.

CR 02	$\mu\text{mol-O}_2$ $\text{m}^{-2} \text{h}^{-1}$	$\mu\text{mol-O}_2$ $\text{m}^{-2} \text{d}^{-1}$	mmol-O_2 $\text{m}^{-2} \text{d}^{-1}$	$\text{mmol-C m}^{-2} \text{d}^{-1}$
Core 1	1.033	24.797	Respiration of suspended sediments	1.699
Core 2	No Data	No Data		
Core 3	1.163	27.918		
Core 4	83.264	1998.332	1.998	
Control	0.280	6.724		
NOTES: depth 6844m. 1 core recovered (#4). Mud was a dark, earthy brown with no evident laminations. 35 cm of sediment. Cores #1 and #3 did not collect sediment and measured oxygen consumption of suspended mud.				
CR 04	$\mu\text{mol-O}_2$ $\text{m}^{-2} \text{h}^{-1}$	$\mu\text{mol-O}_2$ $\text{m}^{-2} \text{d}^{-1}$	mmol-O_2 $\text{m}^{-2} \text{d}^{-1}$	$\text{mmol-C m}^{-2} \text{d}^{-1}$
Core 1	4.753	114.082	Respiration of suspended sediments	1.784
Core 2	7.060	169.442		
Core 3	7.034	168.810		
Core 4	87.431	2098.345	2.098	
Control	0.075	1.796		
NOTES: depth 7939m. 1 core recovered (#4). Mud was again a dark, earthy brown but had traces of small gray laminations that may be ash layers. 38 cm of sediment. Cores #1, #2 & #3 did not collect sediment and measured oxygen consumption of suspended mud.				
CR 05	$\mu\text{mol-O}_2$ $\text{m}^{-2} \text{h}^{-1}$	$\mu\text{mol-O}_2$ $\text{m}^{-2} \text{d}^{-1}$	mmol-O_2 $\text{m}^{-2} \text{d}^{-1}$	$\text{mmol-C m}^{-2} \text{d}^{-1}$
Core 1	100.895	2421.474	2.421	2.058
Core 2	95.654	2295.703	2.296	1.951
Core 3	103.961	2495.066	2.495	2.121
Core 4	91.364	2192.747	2.193	1.864
Control	0.726	17.434		
NOTES: depth 7942m. All 4 cores recovered. Mud was heavily laminated. Layers of alternating gray and brown mud present throughout the core. 24-29 cm of sediment.				

CR05 ($\mu\text{mol-O}_2 \text{m}^{-2} \text{d}^{-1}$)
 AVG 2.35
 St Error 0.067
 St Dev 0.134

Figure 3. Sediment cores recovered at a) CR02 and b) CR04 FVCR deployment sites at 6844 and 7939 meters, respectively.



Figure 4. Sediment core recovered by the FVCR at the CR05 site at 7942 meters.



Appendix:

Smith et al. $669.86 \mu\text{mol-O}_2 \text{ m}^{-2} \text{ d}^{-1}$ 4100 m AENP

Rowe et al. $2.68 \text{ mmol-O}_2 \text{ m}^{-2} \text{ d}^{-1}$ 500 m Miss. Canyon

Glud et al. $154 \mu\text{mol-O}_2 \text{ m}^{-2} \text{ d}^{-1}$ 11,000 m Challenger Deep

Investigating the origin(s) and fate of sediment organic matter and bacterial biomass in the deepest marine environments on Earth.

Eleanna Grammatopoulou, Dr. Daniel Mayor, Dr Barry Thornton- University of Aberdeen, Oceanlab

Objectives/Goals

The main objective of this research is to quantify the distribution of POC and bacteria biomass in the Mariana trench supporting one of the central hypotheses of the HADES programme, that seabed concentrations of organic carbon and bacterial biomass within the trench increase with depth, reaching maximums along the trench axis.

This objective will be met by the analysis of sediment cores, collected by the free vehicle coring respirometer (FVCR) and the rockgrabber. The sediment cores will be analyzed in terms of bacteria biomass and diversity as well as, origin of the food inputs i.e. accumulated organic matter (O.M). Benthic bacterial biomass will be calculated from sediment concentrations of the bacterial biomarker phospholipid fatty acids (PLFAs) and will be compared with direct bacteria counts under microscope examination. PLFAs will also be used for bacterial diversity, and the method will be compared by results obtained from advanced molecular techniques. In order to investigate the origin, age and fate of O.M. the biochemical composition of sediments will be analyzed. This analysis will include the quantity and quality of available O.M. i.e. the compounds that can be readily digested from the benthic fauna, total organic carbon and nitrogen and stable isotopic signatures.

It is believed that life in trenches is mainly dependent on organic matter (OM) supply from overlying productivity thus in addition to sediment O.M. the suspended particulate organic matter (sPOM) from the overlying water column will be also analyzed. The sPOM, a mixture of living organisms and detritus, will be investigated through the analysis of particle concentrated on filters from surface, chlorophyll maximum and abyssal seawater.

Number of samples collected

We successfully collected 6 cores over 3 deployments with the FVCR, ranging in depth from 6844-7942m. Two of the cores were split in half to maximize information output. From the total of 6 cores, 3.5 cores were allocated for the present work (Table 8).

Table 8. Sediment samples collected from FVCR.

Sample Nr	Dive #	Depth	Station #	Core #	Date	Horizon (cm)
1	CR02	6844	3	4	14/11/2014	0-1
2	CR02	6844	3	4	14/11/2014	1-2
3	CR02	6844	3	4	14/11/2014	2-3
4	CR02	6844	3	4	14/11/2014	3-4
5	CR02	6844	3	4	14/11/2014	4-5
6	CR02	6844	3	4	14/11/2014	5-6
7	CR02	6844	3	4	14/11/2014	6-7
8	CR02	6844	3	4	14/11/2014	7-8
9	CR02	6844	3	4	14/11/2014	8-9
10	CR02	6844	3	4	14/11/2014	9-10

11	CR02	6844	3	4	14/11/2014	10-15
12	CR02	6844	3	4	14/11/2014	15-19
13	CR04	7939	4b	4	19/11/2014	0-1
14	CR04	7939	4b	4	19/11/2014	1-2
15	CR04	7939	4b	4	19/11/2014	2-3
16	CR04	7939	4b	4	19/11/2014	3-4
17	CR04	7939	4b	4	19/11/2014	4-5
18	CR04	7939	4b	4	19/11/2014	5-6
19	CR04	7939	4b	4	19/11/2014	6-7
20	CR04	7939	4b	4	19/11/2014	7-8
21	CR04	7939	4b	4	19/11/2014	8-9
22	CR04	7939	4b	4	19/11/2014	9-10
23	CR04	7939	4b	4	19/11/2014	10-15
24	CR04	7939	4b	4	19/11/2014	15-20
25	CR05	7942	6	3	24/11/2014	0-1
26	CR05	7942	6	3	24/11/2014	1-2
27	CR05	7942	6	3	24/11/2014	2-3
28	CR05	7942	6	3	24/11/2014	3-4
29	CR05	7942	6	3	24/11/2014	4-5
30	CR05	7942	6	3	24/11/2014	5-10
31	CR05	7942	6	3	24/11/2014	10-15
32	CR05	7942	6	3	24/11/2014	15-20
33	CR06	7942	6	2	24/11/2014	0-1
34	CR07	7942	6	2	24/11/2014	1-2
35	CR08	7942	6	2	24/11/2014	2-3
36	CR09	7942	6	2	24/11/2014	3-4
37	CR10	7942	6	2	24/11/2014	4-5
38	CR11	7942	6	2	24/11/2014	5-6
39	CR12	7942	6	2	24/11/2014	6-7
40	CR13	7942	6	2	24/11/2014	7-8
41	CR14	7942	6	2	24/11/2014	8-9
42	CR15	7942	6	2	24/11/2014	9-10
43	CR16	7942	6	2	24/11/2014	10-15
44	CR17	7942	6	2	24/11/2014	15-20

Grab samples:

In total 12 deployments of the grab samplers were successful in returning sediment from the trench environment (Table 9). Depths of grab samples ranged from 5025-10525m, encompassing the forearc area and the Pacific plate. For the present work, sediment samples were collected from 7 deployments.

Table 9. Sediment samples collected from rockgrabber.

Sample Nr	Dive #	Depth	Station #	Date	Horizon
1	RG05	7633	6	22/11/2013	Whole core frozen
2	RG06	6877	6	22/11/2014	Whole core frozen
3	RG07	6957	7	25/11/2014	Whole core frozen
4	RG08	7660	7	26/11/2014	Whole core frozen
5	RG08	7660	7	26/11/2014	Whole core frozen
6	RG10	5025	9a	01/12/2014	0-1
7	RG10	5025	9a	01/12/2014	1-2
8	RG10	5025	9a	01/12/2014	2-3
9	RG10	5025	9a	01/12/2014	3-4
10	RG10	5025	9a	01/12/2014	4-5
11	RG10	5025	9a	01/12/2014	5-6
12	RG10	5025	9a	01/12/2014	6-7
13	RG10	5025	9a	01/12/2014	0-1
14	RG10	5025	9a	01/12/2014	1-2
15	RG10	5025	9a	01/12/2014	2-3
16	RG10	5025	9a	01/12/2014	3-4
17	RG10	5025	9a	01/12/2014	4-5
18	RG10	5025	9a	01/12/2014	5-6
19	RG10	5025	9a	01/12/2014	0-1
20	RG10	5025	9a	01/12/2014	1-2
21	RG10	5025	9a	01/12/2014	2-3
22	RG10	5025	9a	01/12/2014	3-4
23	RG10	5025	9a	01/12/2014	4-5
24	RG10	5025	9a	01/12/2014	5-6
25	RG10	5025	9a	01/12/2014	6-7
26	RG10	5025	9a	01/12/2014	7-8
27	RG10	5025	9a	01/12/2014	8-8.5
28	RG10	5025	9a	01/12/2014	0-1
29	RG10	5025	9a	01/12/2014	1-2
30	RG10	5025	9a	01/12/2014	2-3
31	RG10	5025	9a	01/12/2014	3-4
32	RG10	5025	9a	01/12/2014	4-5
33	RG10	5025	9a	01/12/2014	5-6
34	RG10	5025	9a	01/12/2014	6-6.5
41	RG11	9067	10	02/12/2014	0-1
42	RG11	9067	10	02/12/2014	0-1

43	RG11	9067	10	02/12/2014	1-2
44	RG11	9067	10	02/12/2014	2-3
45	RG11	9067	10	02/12/2014	3-4
46	RG11	9067	10	02/12/2014	4-5
47	RG11	9067	10	02/12/2014	5-6
48	RG11	9067	10	02/12/2014	6-7
49	RG11	9067	10	02/12/2014	7-8
50	RG11	9067	10	02/12/2014	0-1
51	RG11	9067	10	02/12/2014	1-2
52	RG11	9067	10	02/12/2014	2-3
53	RG11	9067	10	02/12/2014	3-4
54	RG11	9067	10	02/12/2014	4-5
55	RG11	9067	10	02/12/2014	5-6
56	RG11	9067	10	02/12/2014	6-7
57	RG11	9067	10	02/12/2014	7-8
58	RG11	9067	10	02/12/2014	8-8.3
59	RG11	9067	10	02/12/2014	0-1
60	RG11	9067	10	02/12/2014	1-2
61	RG11	9067	10	02/12/2014	2-3
62	RG11	9067	10	02/12/2014	3-4
63	RG11	9067	10	02/12/2014	4-5
64	RG11	9067	10	02/12/2014	5-6
65	RG11	9067	10	02/12/2014	6-7
66	RG11	9067	10	02/12/2014	7-8
67	RG11	9067	10	02/12/2014	8-8.5
68	RG11	9067	10	02/12/2014	0-1
69	RG11	9067	10	02/12/2014	1-2
70	RG11	9067	10	02/12/2014	2-3
71	RG11	9067	10	02/12/2014	3-4
72	RG11	9067	10	02/12/2014	4-5
73	RG11	9067	10	02/12/2014	5-6
74	RG11	9067	10	02/12/2014	6-7
75	RG11	9067	10	02/12/2014	7-8
76	RG11	9067	10	02/12/2014	0-1
77	RG11	9067	10	02/12/2014	1-2
78	RG11	9067	10	02/12/2014	2-3
79	RG11	9067	10	02/12/2014	3-4
80	RG11	9067	10	02/12/2014	4-5
81	RG11	9067	10	02/12/2014	5-6
82	RG11	9067	10	02/12/2014	6-7
83	RG11	9067	10	02/12/2014	7-7.5

84	RG12	8518	10A	03/12/2014	Whole core frozen
85	RG12	8518	10A	03/12/2014	Whole core frozen
86	RG12	8518	10A	03/12/2014	Whole core frozen
87	RG12	8518	10A	03/12/2014	Whole core frozen
88	RG13	8519	10A	04/12/2014	Whole core frozen
91	RG13	8750	11	04/12/2014	0-1
92	RG13	8750	11	04/12/2014	1-2
93	RG13	8750	11	04/12/2014	2-3
94	RG13	8750	11	04/12/2014	3-4
95	RG13	8750	11	04/12/2014	4-5
96	RG13	8750	11	04/12/2014	5-6
97	RG13	8750	11	04/12/2014	6-7
98	RG13	8750	11	04/12/2014	0-1
99	RG13	8750	11	04/12/2014	1-2
100	RG13	8750	11	04/12/2014	2-3
101	RG13	8750	11	04/12/2014	3-4
102	RG13	8750	11	04/12/2014	4-5
103	RG13	8750	11	04/12/2014	5-6
104	RG13	8750	11	04/12/2014	6-6.8
105	RG13	8750	11	04/12/2014	0-1
106	RG13	8750	11	04/12/2014	1-2
107	RG13	8750	11	04/12/2014	2-3
108	RG13	8750	11	04/12/2014	3-4
109	RG13	8750	11	04/12/2014	4-5
110	RG13	8750	11	04/12/2014	5-6
111	RG13	8750	11	04/12/2014	6-7
112	RG13	8750	11	04/12/2014	7-7.5
113	RG13	8750	11	04/12/2014	0-1
114	RG13	8750	11	04/12/2014	1-2
115	RG13	8750	11	04/12/2014	2-3
116	RG13	8750	11	04/12/2014	3-4
117	RG13	8750	11	04/12/2014	4-5
118	RG13	8750	11	04/12/2014	5-6
119	RG14	10525	12	05/12/2014	Whole core frozen
120	RG14	10525	12	05/12/2014	Whole core frozen
121	RG14	10525	12	05/12/2014	Whole core frozen
122	RG14	10525	12	05/12/2014	Whole core frozen
123	RG14	10525	12	05/12/2014	Whole core frozen

Sample summary

The sediment cores from FVCR and rock grabber were sliced at intervals of 1cm down to 10 cm and 5cm intervals thereafter. From each station, one sediment core was subsampled (4ml)

for microbiology analysis (stored at -80°C), when 3 cores were subsampled (1ml) for bacteria counts analysis (-20°C). The remained sediment was stored for PLFA and O.M. analysis (-20°C). The numbers of filters collected were in total 26 (Table 10). From these filters 9 were taken from the sea surface, 9 from chlorophyll maximum, 8 from 5000m. All filters were stored to -80°C.

Table 10. Water filters collected from sea surface (S.W.) and the CTD.

Sample #	Station #	Sample origin and deployment #	Depth(m)	Filter pore size (µm)	Volume (ml)
1	1	S.W. 1	10	0.7	3000
2	1	S.W.1	10	0.7	3000
3	1	S.W.1	10	0.7	3000
4	1b	CTD1	4886	0.7	13000
5	1b	CTD1	4886	0.7	13000
6	2	S.W.2	10	0.7	4000
7	2	S.W.2	10	0.7	4000
8	2	S.W.2	10	0.7	2500
9	3	S.W.2	10	0.7	4000
10	3	S.W.2	10	0.7	4000
11	4	CTD2	4886	0.7	14000
12	4	CTD2	4886	0.7	4000
13	4	CTD2	4886	0.7	11000
14	4	CTD2	115	0.7	3200
15	4	CTD2	115	0.7	3200
16	4	CTD2	115	0.7	3200
17	4	CTD2	4886	0.7	11500
18	5	CTD3	122	0.7	3500
19	5	CTD3	122	0.7	3500
20	5	CTD3	122	0.7	3500
21	5	CTD3	4917	0.7	9500
22	5	CTD3	4917	0.7	9000
23	5	CTD3	4917	0.7	5000
24	9a	CTD4	130	0.7	3500
25	9a	CTD4	130	0.7	3500
26	9a	CTD4	130	0.7	3500

Scavenger assemblages investigated with the Hadal-Lander

Alan Jamieson, Thomas Linley: Oceanlab, University of Aberdeen, U.K.

Summary

The Hadal-Lander was deployed 24 times during FK141109 and collected a total of 105 hours of video between 4998 and 10,545m. The camera system was successful in providing a wealth of imaging data of the large mobile communities spanning the entire depth range of the Mariana Trench, including many rare and new species, as well as many hitherto unseen behaviors. The upper trench (4500-6500m) was dominated by the ophidiid and macrourid fishes and the aristidean prawns, whereas the medium depths of the trench (6500-8000m) were dominated by the liparid fishes and supergiant amphipods. Beyond 8000, only small scavenging and predatory amphipods were observed albeit in high densities. The highlights were observing a putative new species of snailfish that dominated the upper trench plus an entirely new species that happened to be the deepest ever filmed (8145m). Also the quantity of behavioural information, particularly of the supergiants, decapods and *Princaxelia* amphipods were especially impressive.

Background

The contribution by the University of Aberdeen to the HADES-M expedition was the use of two deep sea baited camera systems (Abyssal-Lander and Hadal-Lander) and the baited trap dubbed 'Wee-Trap'. These vehicles were used to observe and record the zonation, abundance and behaviour of bait attending fauna across the entire depth range of the Kermadec Trench.

The Hadal-lander is an 11,000m rated baited video camera. Its basic delivery system comprises four pairs of 11,000m rated Vitrovex 17" spheres plus another two; one acting as a lazy buoy and the other as a dhan buoy in which recovery aids are connected (flag, VHF beacon and Xenon strobe). The lander descends by virtue of a negatively buoyant ballast weight, comprising 14 'Alvin plates' at 7.7kgs each. These weights are jettisoned by acoustic command from the surface via a tandem set of acoustic releases (IXSEA, Oceano 2500 ti-Deep). The scientific payload comprises a bespoke 3CCD Hitachi colour video camera (800 TV lines), controlled and logged autonomously by a custom built control system (NETmc Marine). Illumination is provided by two LED lamps in 6" vitrovex spheres. The camera is pre-programmed to take 1 minutes of video every 5 or 2.5 minutes throughout and is powered by a 12V lead acid battery (SeaBattery; DSP&L). The lander also has a 2.5 litre niskin water sampler that closes on ballast weight jettison. Three baited amphipod traps were secured to the base of the lander. Locally sourced mackerel was used for bait. An SBE-39 pressure and temperature sensor was logging at 30 seconds throughout.

Results

The Hadal-Lander was deployed 24 times during the cruise and collected a total of 105 hours of video between 4998 and 10,545m (Fig 5, Table 11). The camera system was successful in providing a wealth of imaging data of the large mobile communities spanning the trench, including many rare and new species, as well as many hitherto unseen behaviours.

The Hadal-Lander deployments in the early stages were not without problems. The new 3CCD camera was damaged upon transport to Guam and thus after being rebuilt provided usable data but out of focus (LH02-LH07). The first deployment failed due to software problems and the second because of exceptionally complex terrain causing the lander to tilt beyond view of the

seafloor. Deployments LH05 and LH06 were also greatly affected by the lander sinking into incredibly soft sediment and thus eventually burying the bait, although useful information was obtained. Upon receiving the new camera, there were still a few technical issues resulting in slightly out of focus images during LH08-LH11. The problem was resolved and the next 13 deployments were perfect. It is worth noting that the terrain on the fore arc made safe landing of the vehicle problematic, and despite reconfiguring the lander with a suspended ballast weight, there were many areas where deploying landers is risky. Having said that, the missing Hadal-Lander deployments from the first few stations was compensated by 4 successful abyssal lander deployments, and thus data integrity was maintained.

Once the camera issues were overcome, the Hadal-lander produced a vast quantity of high resolution video of many abyssal and hadal mobile animals. Generally, the stations between 4000 and 6500m deep were dominated by the cusk eel (ophidiid) *Bassozetus* sp. and the rat-tail (macrourid) *Coryphaenoides yaquinae*. Also ever present at these depths was the decapod *Benthesicymnus crenatus* and the lesser observed *Plesiopennaeus armatus*, being present at 5000m. From 6500 to 8000m, snailfish of the Liparidae family dominated alongside the 'supergiant amphipod' *Alicella gigantea*. It is highly likely this is a new species of snailfish. Furthermore, the video of the supergiant amphipods are extraordinarily rare. Another snailfish was observed at 8007 and 8145m making it the deepest fish seen alive albeit of unknown species. Beyond the nominal 8000m mark, no other organisms were observed other than scavenging amphipods of mixed species and the predatory amphipod of the *Princaxelia* genus (Family: Pardaliscidae). A generalised representation of zonation is shown in figure 6.

Table 11. Details of Hadal-Lander deployments, where 1 = time lander spent on seafloor, 2 = total number of 1-minute sequences on the seafloor, 3 = video timing intervals (1/5 = 1 minute every 5 minutes, 1/2.5 = 1 minute every 2.5 minutes), and 4 = total length of video on seafloor collected.

Station	Date	Latitude	Longitude	Depth (m)	Pressure (dbar)	Bottom time ¹	# of sequences ²	Interval ³	hhmm ⁴
LH01	10-Nov	12.74697'N	144.65566'E	4782	4863	12:33	0	1/5	0
LH02	11-Nov	12.72218'N	144.71029'E	4703	4782	12:55	0	1/5	0
LH03	12-Nov	12.52117'N	144.74701'E	6089	6211	11:11	134	1/5	02:24
LH04	13-Nov	12.52095'N	144.74368'E	6097	6220	11:00	130	1/5	02:10
LH05	14-Nov	12.62628'N	144.71080'E	6831	6980	14:01	166	1/5	02:46
LH06	15-Nov	12.62744'N	144.76111'E	6931	7084	12:52	152	1/5	02:32
LH07	16-Nov	12.41121'N	144.89291'E	7415	7587	33:29	397	1/5	06:17
LH08	18-Nov	12.40948'N	144.89458'E	7440	7615	10:40	126	1/5	02:06
LH09	20-Nov	12.30539'N	144.68709'E	7941	8136	15:57	374	1/2.5	06:14
LH10	21-Nov	11.92000'N	144.92250'E	8007	8206	32:04	471	1/2.5	07:51
LH11	23-Nov	11.92460'N	144.91530'E	8078	8279	09:41	227	1/2.5	03:47
LH12	24-Nov	11.80960'N	144.97660'E	7012	7168	11:09	266	1/2.5	04:26
LH13	25-Nov	11.87790'N	144.97710'E	7485	7661	11:00	257	1/2.5	04:17
LH14	26-Nov	11.59110'N	144.84730'E	6010	6130	10:11	239	1/2.5	03:59
LH15	27-Nov	11.60710'N	144.83310'E	6142	6267	35:50	831	1/2.5	13:51
LH16	29-Nov	11.41830'N	144.41220'E	5044	5133	13:56	327	1/2.5	05:27
LH17	30-Nov	11.42290'N	144.43340'E	4998	5085	13:06	301	1/2.5	05:01
LH18	01-Dec	11.91980'N	144.92300'E	8004	8202	11:13	262	1/2.5	04:22
LH19	02-Dec	12.04300'N	144.88230'E	9059	9306	10:31	124	1/5	02:04
LH20	03-Dec	12.15980'N	144.67080'E	8964	9207	13:02	154	1/5	02:34
LH21	04-Dec	12.07010'N	144.60220'E	10545	10872	10:07	238	1/2.5	03:58
LH22	05-Dec	12.28860'N	144.66870'E	8143	8347	13:32	317	1/2.5	05:17
LH23	06-Dec								
LH24	07-Dec								

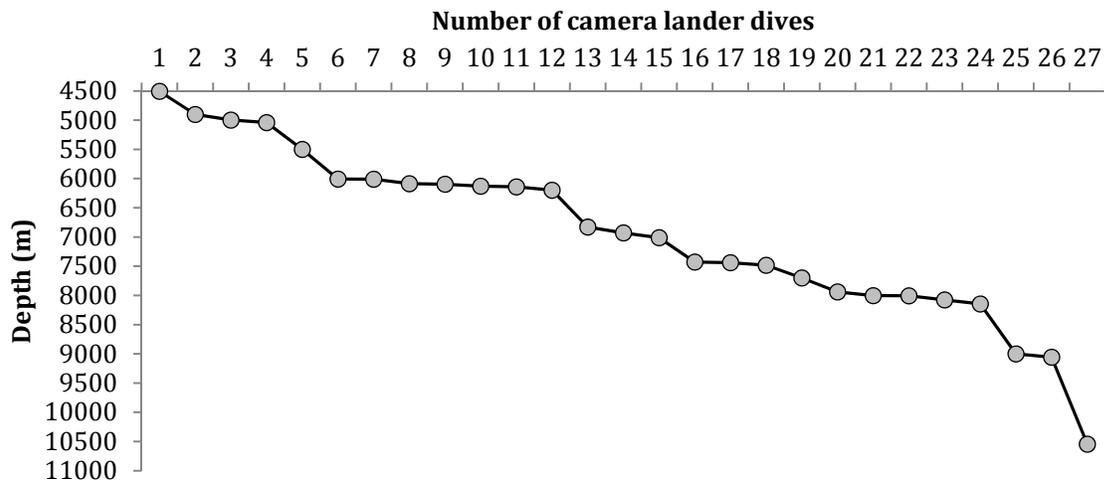


Figure 5. Combined Abyssal-Lander and Hadal-lander deployment depths showing full coverage of the bathymetric range.

St#	Depth	COY	BZ	BC	SF1	AG	PRA	PA	SF2
LA02	4506	x	x	x					
LA01	4902	x	x	x					
LH17	4998	x	x	x				x	
LH16	5041	x	x	x				x	
LH24	5500	x	x	x					
LA05	6008	x	x	x		x			
LH14	6010	x	x	x		x			
LH03	6087	x	x	x					
LH04	6096	x	x	x		x			
LA04	6130		x	x		x			
LH15	6141	x	x	x		x			
LA03	6198	x	x	x	x	x			
LH05	6829	x		x	x	x			
LH06	6929			x	x				
LH12	7011	x		x	x	x			
LH07	7428			x	x	x			
LH08	7439			x	x				
LH13	7483				x	x			
LH23	7700				x				
LH09	7939				x				
LH18	8003				x		x		
LH10	8006				x		x		x
LH11	8076				x		x		
LH22	8145						X		X
LH20	9000						x		
LH19	9057						x		
LH21	10545								

Figure 6. General zonation trends of the most frequently seen large mobile species. COY = *Coryphaenoides yaquinae*, BZ = *Bassozetus* sp., BC = *Benthesicymnus crenatus*, SF1 = snailfish species 1, AG = *Alicella gigantea*, PRA = *Princaxelia* sp. PA = *Plesiopennaeus armatus*, SF2 = Snailfish species 2.



Figure 7. (top left) swarms of amphipods at 10,545m (Sirena Deep), (top right) decapod, (middle left) Cusk eel and rat-tail, (middle right) snailfish, (bottom left) supergiant amphipods and (bottom right) the new species of snailfish at 8145m.

Scavenging communities and environmental data from the Abyssal lander

Thomas Linley – Oceanlab, University of Aberdeen

Materials and methods

The Abyssal-lander is a 6,000m rated baited stills camera. Its basic delivery system comprises three and a half pairs of 6,000m rated Vitrovex 17” spheres plus another two; one acting as a lazy buoy and the other as a dhan buoy in which recovery aids are connected (flag, VHF beacon and Xenon strobe). The lander descends by virtue of a negatively buoyant ballast weight, comprising 12 ‘Alvin plates’ at 7.7kgs each. These weights are jettisoned by acoustic command from the surface via a tandem set of acoustic releases (IXSEA, Oceano 2500 Universal). The scientific payload comprises a 5 megapixel stills camera and flash (Kongsberg Maritime, UK), The camera is pre-programmed to take 1 image every 60 seconds and is powered by a 24V lead acid battery (SeaBattery; DSP&L). Locally sourced Mackerel was used for bait. The lander also has an Anderaa Seaguard CTD probe that records every 30 seconds throughout.

Deployments

The Abyssal Lander was deployed successfully five out of six times (Table 12). It captured over 3,000 seabed images. Gradual water ingress to the flash unit had been observed during the deeper deployments and attempts to better seal the housing only slowed the ingress. The humid environment within the housing caused corrosion and the eventual failure of the strobe element. Possible replacements were explored on board however the part is proprietary and an alternative was not found. Even if a replacement had been found it would not have addressed the root cause of water ingress.

Table 12. Abyssal lander deployments (Dep). Dates are given in the format DD/MM/YYYY, time is local (GMT + 10). Depth is provided from both the multibeam and the SeaGuard pressure sensor in meters. Stills is the number of images taken at the seabed and therefore the number of minutes recorded.

Station	Dep	Date Deployed	Time Deployed	Date Recovered	Time Recovered	Latitude (North)	Longitude (East)	Multibeam Depth	Sensor Depth	Stills
01	LA01	10/11/2014	16:43	11/11/2014	10:49	12.74366	144.64731	4897	4902	786
01	LA02	11/11/2014	17:49	12/11/2014	08:01	12.72555	144.72629	4575	4506	579
02	LA03	12/11/2014	17:03	13/11/2014	09:50	12.52415	144.73346	6081	6198	657
02	LA04	13/11/2014	17:13	14/11/2014	09:37	12.51938	144.74013	6058	6130	646
08	LA05	26/11/2014	19:27	27/11/2014	09:58	11.6025	144.8623	6023	6008	519
08	LA06	27/11/2014	15:11	29/11/2014	09:52	11.581	144.8395	5952	5925	NA

Environmental data

Seabed data

The SeaGuard CTD and RCM recorded environmental data at a 30 second resolution. The environmental data is outlined in Table 13. The force and direction of the seabed currents is further illustrated by Fig. 8. A tidal pattern is still detectable at the seabed (Fig.9)

Table 13. The environmental data recorded by the SeaGuard platform on the Abyssal Lander. Data are presented as a mean value for the duration on the bottom. Current speed is in cm.s^{-1} mean \pm standard deviation, temperature in $^{\circ}\text{C}$, salinity in PSU and depth in m.

Deployment	Current Speed	Temperature	Salinity	Depth
LA01	3.23±0.93	1.50	34.62	4902.26
LA02	10.67±0.94	1.47	34.64	4506.06
LA03	6.25±0.95	1.62	34.69	6197.96
LA04	7.64±0.95	1.61	34.65	6130.40
LA05	3.21±0.95	1.59	34.69	6007.91
LA06	5.33±1.02	1.58	34.67	5925.07

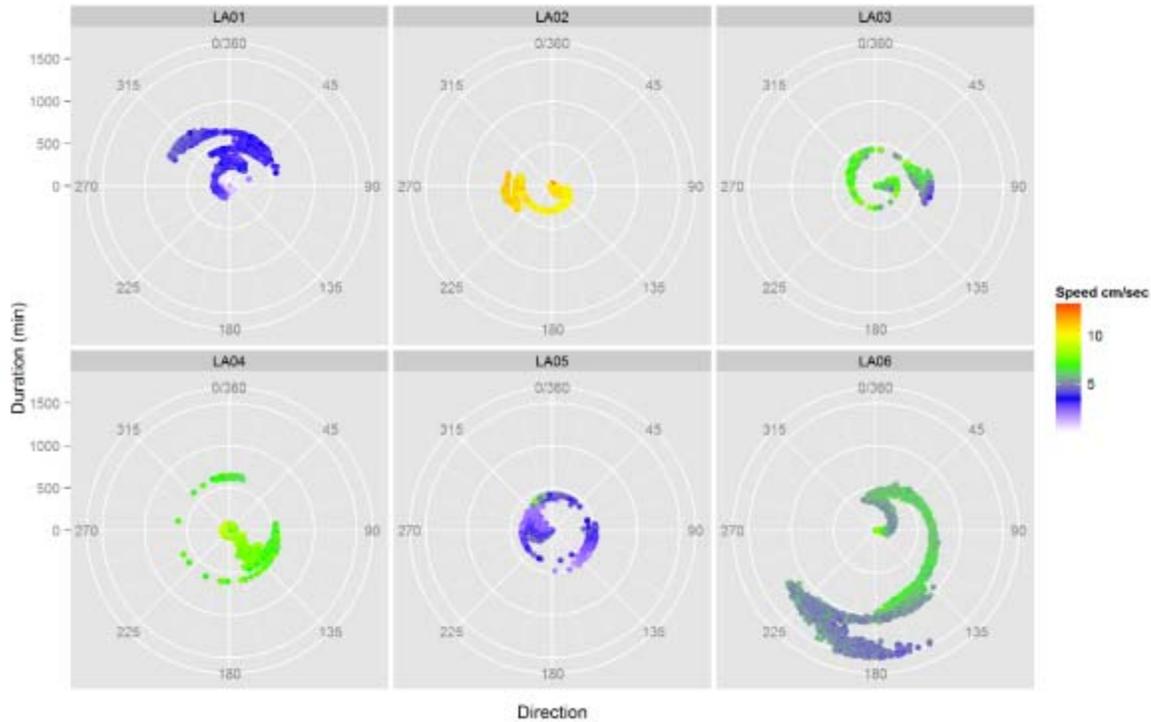


Fig. 8. The current speed and direction recorded by the Abyssal Lander. The direction of the current is expressed by its degrees on the circular plot (0/360 is equal to magnetic north). The magnitude of the current is expressed by the colour of the point and the distance from the centre of the plot is relative to the time that has passed during the deployment.

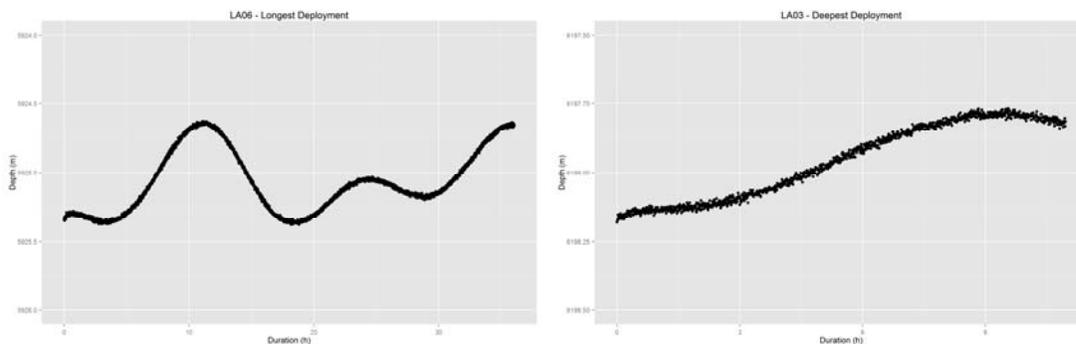


Fig. 9. Tidal cycle detectable at the seabed in the longest (left) and deepest (right) deployments.

Profile data

Water column profiles were very consistent for all deployments. The temperature range was very large (Fig. 10), from 32.3°C in the surface waters to the seabed temperatures of 1.47 to 1.62°C. The thermocline was fairly deep, a 20°C drop was observed over the first 250m.

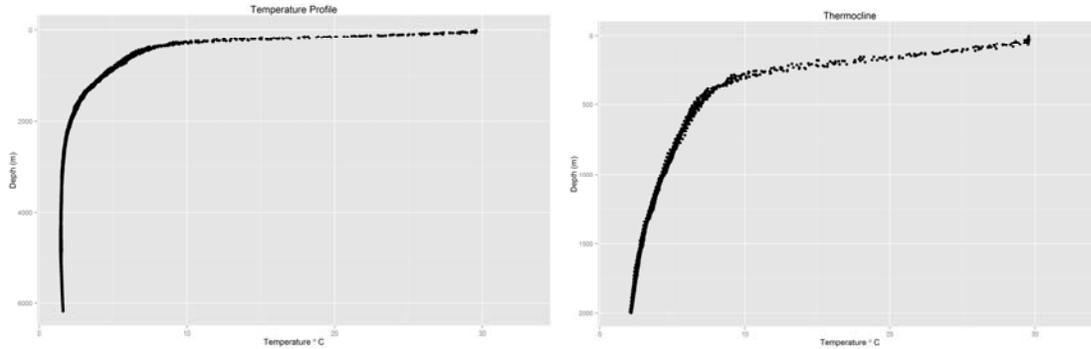


Fig. 10. The temperature profile recorded by the Abyssal Lander on ascent and decent (left) and a zoomed view of the thermocline (right).

Salinity showed the greatest variation in the surface waters (due to freshwater input and evaporation). Salinity generally increased with depth. Some variation was seen between deployments and even during the ascent and decent of the same deployment (Fig.11) indicating a temporal element.

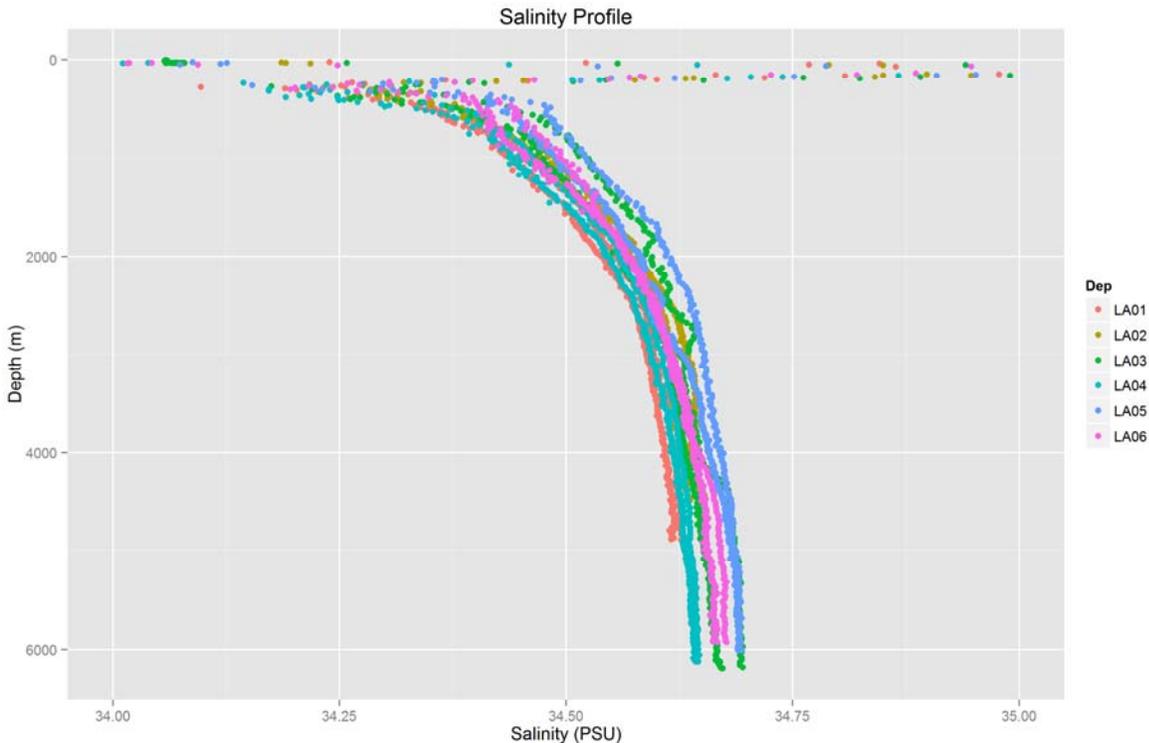


Fig. 11. The salinity profile recorded by the Abyssal Lander on ascent and decent for each of the deployments (Dep).

Biological Data

The Abyssal Lander takes a five megapixel image every minute of scavenging fauna attracted to bait. Deployments are presented by location and by increasing depth.

Overriding plate

4506m - LA02 – Station 1

The lander settled on a dark hummocky rock outcrop with a thin veneer of golden sediment (Fig.12). Observed megafauna species (Fig.13) in arrival order were: *Benthesicymnus crenatus*, slender cusk-eel, *Bassozetus sp.*, small rattail (likely juvenile *Coryphaenoides yaquinae*), and *C. yaquinae*.

4902m – LA01 – Station 1

The lander settled on open sandy seabed. The sediment appeared fine and homogenous. Dark rocks of varying size and shape were visible. Observed megafauna species in arrival order were: *B. crenatus*, small rattail (likely juvenile *C. yaquinae*), a very large *C. yaquinae*, *Bassozetus sp.* including some very small individuals and a slender rattail.

6130m – LA04 – Station 2

The lander settled on open sandy seabed. The sediment appeared fine and less homogenous than the previous station. Observed megafauna was dominated by *B. crenatus*, a single *Bassozetus sp.* was present for a large proportion of the deployment.

6196m – LA03 – Station 2

The seabed appeared as in the previous deployment. Observed megafauna species in arrival order were: *B. crenatus*, *Alicella gigantea*, blue-headed cusk-eel, hadal snailfish, *Bassozetus sp.*, *C. yaquinae* and small rattail (likely juvenile *C. yaquinae*).

Downriding plate

6008m – LA05 – Station 8

The lander settled on open sandy seabed. The sediment appeared fine and homogenous. Some animal burrows are visible. Observed megafauna species in arrival order were: *B. crenatus*, *Bassozetus sp.*, *C. yaquinae* (including a very large individual) and *A. gigantea*.

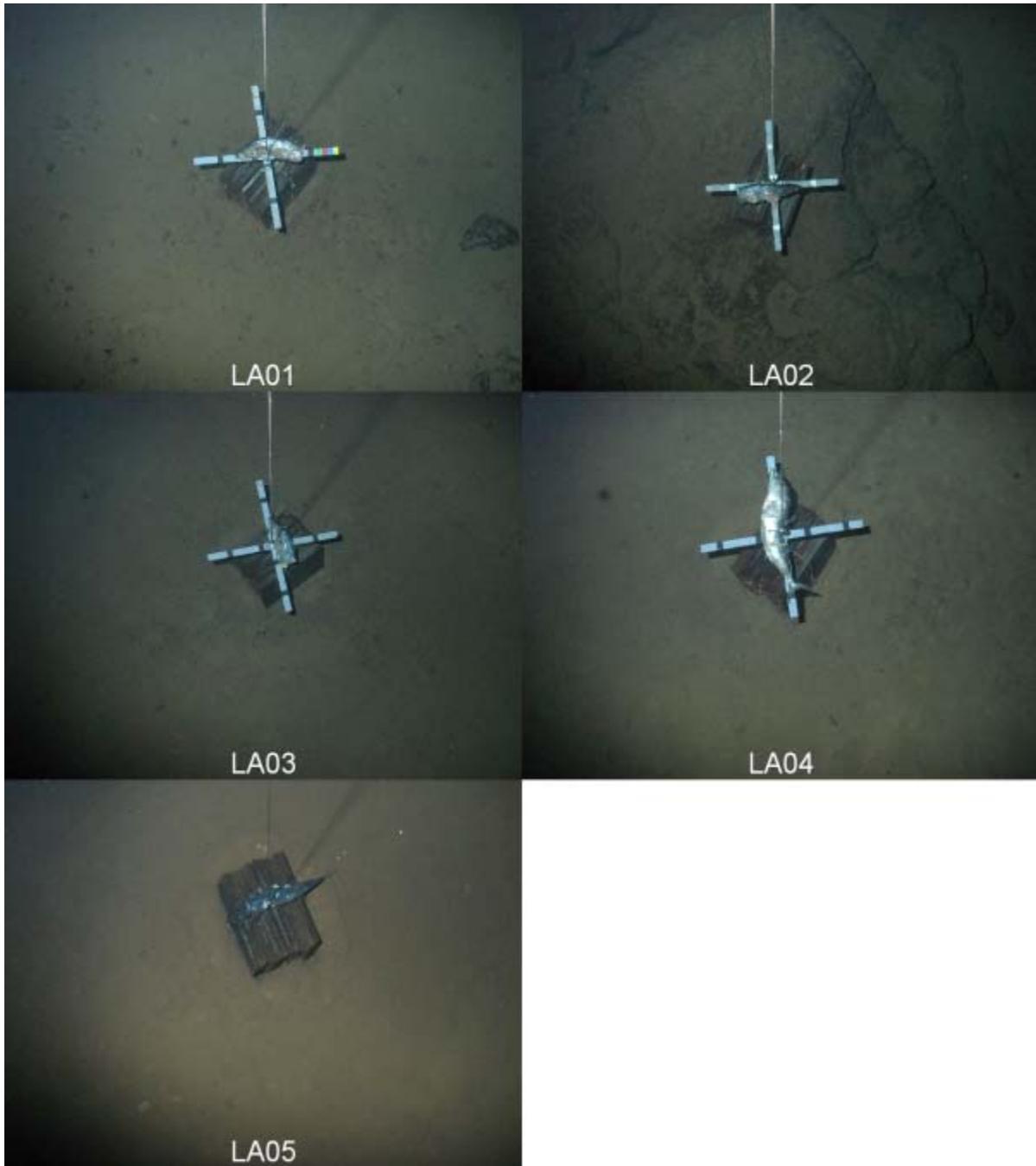


Fig. 12: Sediment types at the Abyssal Lander deployments. The scale bar is 10cm between the black tape.

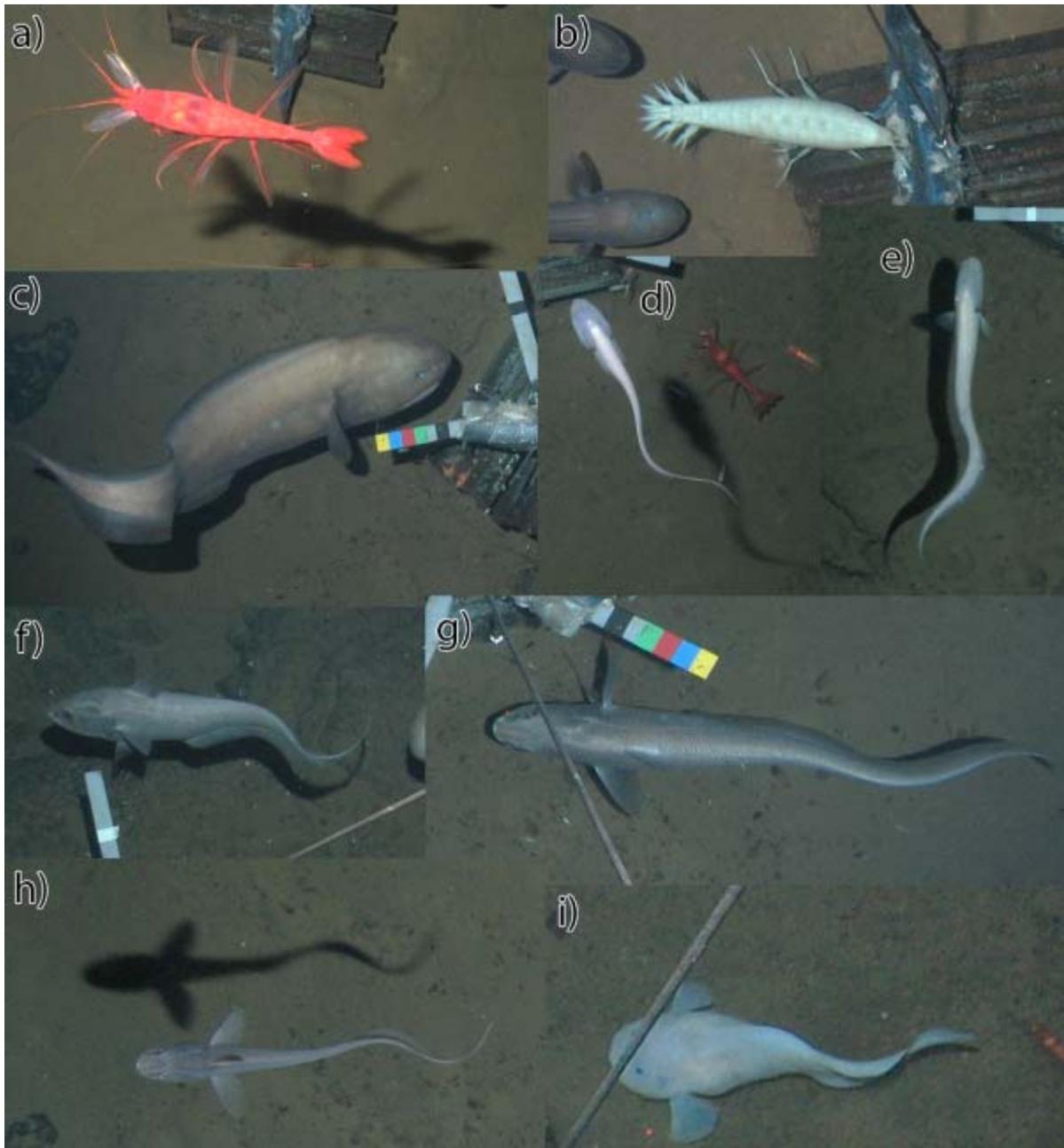


Fig. 13. Scavenging macrofauna attracted to the Abyssal lander; a) *Benthescymnus crenatus*, b) *Alicella gigantean*, c) *Bassozetus sp.*, d) blue-headed cusk-eel; possibly a juvenile of e) slender cusk-eel, f) *Coryphaenoides yaquinae*, g) slender rattail, h) small rattail, i) hadal snailfish.

Stand Alone Imaging Module – SAIM

Thomas Linley – Oceanlab, University of Aberdeen

Materials and methods

The Stand Alone Imaging Module (SAIM; Fig. 14) is a compact and low-cost near-field full ocean depth camera. It was used as a pilot technology on this cruise. It is based upon the Raspberry Pi (UK) microcomputer and camera (PiCam). Illumination is provided by three high-output LEDs mounted around the camera lens in a single port. The illumination is powered by a PP3 9V battery and the computer itself by two mobile phone travel chargers.



Fig. 14: The Stand Alone Imaging Module (SAIM)

The software was optimized during testing and clearer images were obtained in later deployments. Since there is a single port in the housing the outer face of the sapphire window caused reflection and some of the image is lost to the light from the LEDs bouncing back. Software changes mitigated this. The SAIM is a hybrid system able to take both video and stills. Several different sequences were tested. One minute of HD video and three five megapixel stills for each five minute cycle provided good data coverage.

Results

The SAIM is the first working prototype and was deployed opportunistically. The SAIM deployments are listed in Table 14. As this payload is added to another platform it is given the appropriate deployment numbers.

Table 14. SAIM deployments (Dep) attached to other platforms. Station is the location, Platform is the platform and deployment number that the SAIM was attached to. The date is in the format DD/MM/YY. Purpose is the intended reason for the recording. Notes are comments on the

quality of the deployment. Where species observed are marked “*” the species was seen but was not captured by the trap.

Dep	Station	Platform	Date	Purpose	Notes
SA01	02	RG02	12/11/14	Grab closure	Distance too great
SA02	03	WT03	15/11/14	Trap retention	<i>Alicella gigantean</i> , <i>Benthesicymnus crenatus</i> * and snailfish entering trap
SA03	04A	WT05	19/11/14	Trap retention	<i>Alicella gigantean</i> , <i>Benthesicymnus crenatus</i> * and snailfish entering trap
SA04	06	WT07	21/11/14	Trap retention	Recording error, some files missing
SA05	06	WT08	23/11/14	Trap retention	Early arrival only, insufficient battery charge. <i>Alicella gigantean</i> *, <i>Coryphaenoides yaquinae</i> * and snailfish.
SA06	07	WT09	24/11/14	Trap retention	<i>Alicella gigantean</i> , <i>Benthesicymnus crenatus</i> *, <i>Coryphaenoides yaquinae</i> * and snailfish.
SA07	07	WT10	25/11/14	Trap retention	Snailfish* entering trap
SA08	09A	TR17	30/11/14	Trap retention	No fish seen
SA09	06B	WT11	01/12/14	Trap retention	<i>Prinaxelia sp.</i> entering trap
SA10	11	WT13 – amphipod trap	03/12/14	Amphipod feeding	<i>Scopelocheiropsis Schellenberg</i> feeding
SA11	12	WT14 – amphipod trap	04/12/14	Amphipod feeding	Insufficient battery charge

Trap retention

The SAIM was placed across the entrance to both the Wee Trap and Trap to monitor animal retention and as additional presence/absence data (Fig.15).



Fig. 15. *Alicella gigantean* (left) and the hadal snailfish crossing the threshold of the Wee Trap.

Species were often observed that were not retained in the traps (Table X.X) increasing our presence/absence data. The video will be reviewed in an attempt to refine trap design.

Amphipod feeding

Due to the limited illumination the system was intended for macro imaging. This was best used when the SAIM was placed inside one of the amphipod traps within Wee Trap during deployment WT13 (Fig.16).



Fig. 16. SAIM within an amphipod trap. While bait is plentiful bait particles are suspended by the feeding amphipods (left). As the bait is removed there is evidence of cannibalism (right).

The SAIM continued to record on the ascent of WT13 and recorded (and time stamped) the death of the amphipods in the trap. The trap was released at 0933 local/ 23:33 GMT. Most movement had ceased by 1213 local/0213 GMT; after 2:40 (hh:mm). All visible movement had stopped at 1310 local/ 0310 GMT just 37min before the trap was recovered. The Wee Trap has an ascent rate of about $33\text{m}\cdot\text{min}^{-1}$. Amphipods from 8,847m have therefore been observed to reduce in activity at 3,567m depth and still show signs of life at the surface.

Products

The varied use of the SAIM will greatly aid in the development of the camera system. That an early stage prototype can gather useful scientific data is encouraging. The observations of animals around the trap will hopefully aid in the refinement in the trap design.

The recordings of feeding amphipods are most encouraging as this is the first time that they have been observed feeding so closely. This will provide information on feeding behavior and the nature of the reported cannibalism in these animals, particularly whether it is a natural phenomenon or caused by the animals being trapped together at high density. Estimations of when the amphipod samples were killed by being brought to the surface is extremely useful in analysis of the tissue samples taken and how much they are likely to have degraded.

Animal sampling summary

Baited traps were deployed 38 times on the cruise (Figure 17) to capture specimens of hadal animals to further the physiological, energetic, and life history goals of the project. In addition the samples allow verification of the identity of common species observed in the photo and video landers. There were two traps, the fish trap (or simply trap), the wee trap, and a cluster of three PVC tube shaped traps that were deployed on the foot of the hadal lander in most of its deployments. Between the these traps 37 specimens of a putative new species of snailfish, two rattails, *Coryphaenoides yaquinae*, three shrimp, likely *Benthiscymus crenatus*, 14 giant amphipods (*Alicella gigantea*), dozens of large Princaxellid amphipods, and thousands of smaller amphipods were captured (see Table 15 for large animal captures). Details of these captures follow.

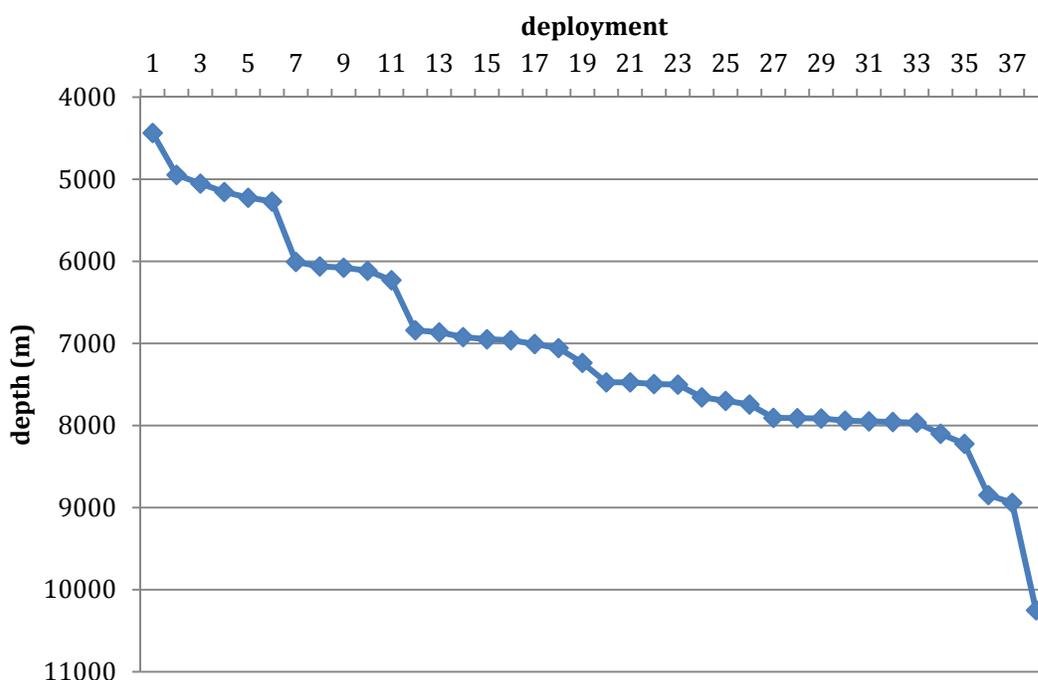


Figure 17. Distribution of trap sampling effort by depth in the Mariana Trench using the fish trap and wee trap.

Table 15. Summary of megafauna (excludes small amphipods) caught in the fish trap (TR) and wee trap (WT).

Deploy#	depth	latitude	longitude	fishes		crustaceans		
				C. yaquinae	snailfish	decapod	mysid	A.gigantea
TR02	4441	12.73644	144.70035	1				
TR04	6081	12.52613	144.73614	1				
TR05	7062	12.59786	144.77854		1			
TR06	6914	12.6339	144.7508		1	1		2
TR07	7497	12.42347	144.87058		9		1	4
TR08	7509	12.42556	144.91171		4			

TR09	7929	12.30274	144.67388		1			
TR10	7841	11.9128	144.9514		3			
TR12	6898	11.8107	144.9445		1		2	
TR13	6974	11.826	144.0088		3		3	
TR15	6062					2		
TR16	5053					1		
TR19	7626	12.2766	144.6202		2			
TR20	7652	12.3495	144.6813		4			
TR21	5225			2				
WT03	6961	12.61026	144.76839		2		2	
WT04	7495	12.41505	144.91187		1			
WT06	7949	12.3037	144.68038		1			
WT07	7907	11.9273	144.962		1			
WT08	7966	11.9297	144.9288		1			
WT09	6949	11.8147	144.9858		2		1	
Grand Total				4	37	4	1	14

Summary of fish captures (Mackenzie Gerringer, U. of Hawaii)

This cruise saw the first *in situ* records and captures of fishes in the Mariana Trench. In deployments of two traps, 33 individual fish were collected. Two of these were the macrourid, *Coryphaenoides yaquinae*, an important representative of the abyssal ecosystem. The other 31 fish, of the family Liparidae, are likely a new species of hadal snailfish, maybe endemic to this trench (Figure 18). Given the occurrence of snailfishes in other trenches and the marked depth zonation observed here and in other trenches, this is a particularly important finding. The collection of individuals of lengths 9.9 – 24.7 cm provide a distribution of life stage from what appears to be most of the depth range of the animal (6898 – 7974 meters). The fishes collected at the upper limits of the depth range are very likely the deepest fish ever caught.



Figure 18. Hadal snailfish species, likely undescribed. 7509 m. Mariana Trench.

Five specimens were preserved whole for taxonomic use. Muscle of fin clip samples were taken from all individuals for genetics. One whole individual was frozen for water content. Samples from these fish were collected for researchers at the University of Hawaii, WHOI, the

University of Aberdeen, and Scripps Institute of Oceanography. White muscle samples were taken to investigate metabolic enzyme activity as a function of depth, stable isotopes to illuminate trophic relationships in the hadal zone, physiological adaptations to pressure, and genomics. Hindgut samples and whole stomachs were taken for diet study and analysis of gut-associated microbes. Additional tissues preserved for research include liver, spleen, heart, gonad, otoliths, blood, and gelatinous tissue. These are likely the deepest-living vertebrate samples ever taken and will be valuable to the understanding of physiological and ecological trends with depth in and around the hadal zone.

Photogrammetry (Thomas Linley – Oceanlab, University of Aberdeen)

The photogrammetry methodology used on the HADES-K cruise has been further developed and 360 degree scans are now possible. Unfortunately the fragile nature of the snailfish and their translucent appearance made reconstruction very difficult. A partial reconstruction of the sucking disk (a key diagnostic feature) and a high quality 180 degree version were possible (Fig.19).



Fig. 19. Photogrammetric reconstructions of the hadal snailfish. Detail of the ventral side and sucking disk using the new methodology (left) and a lateral reconstruction using the old method (right).

Products

The models will be further worked on in the hope of resolving more of the 360 degree scan with the intention of including an interactive figure in the description of this new species.

Amphipod sampling (Stuart Piertney, University of Aberdeen)

Amphipod samples were collected from three baited funnel traps placed on the hadal lander (when in horizontal camera mode), two traps on the wee trap, and three traps (two internal and one external) on the fish trap. In all cases the bait comprised c. 100g of Pacific mackerel enclosed in a fine-mesh bag to allow an odour plume to emanate from the traps but prevent the larger amphipods from consuming the bait and contaminating downstream analyses with fish tissue. Amphipod traps were removed from the vehicles immediately after recovery onto deck and processed at 4°C. Individual amphipods were treated in one of five ways: 1) snap-frozen in liquid nitrogen for analyses on enzyme function and RNA extraction; 2) soaked in an excess volume of RNAlater for 24 hours with subsequent decanting and freezing at -80°C for future RNA extraction; 3) preserved in 95% ethanol for DNA extraction; 4) preserved in formalin for histology; 5) dissected to isolate gonads which were then preserved in gluteraldehyde.

In total, more than 3600 amphipods were collected from across 47 deployments ranging from 4421-10360 metres depth (Table 16). These hauls comprised a number of different species, which from initial identification based on gross morphology included *Hirondellea* sp., *Princaxelia* sp., *Alicella* sp., *Scopelocheirus* sp., *Eurythenes* sp. *Paralicella* sp. and *Abyssorchomene* sp.

Samples will be utilised to address a number of questions and issues:

Community structure and zonation with depth.

Samples from the hadal lander and wee trap will be identified to species level using diagnostic morphological features and DNA barcoding and counted. Assessment will then be made of how community structure alters with depth. New species of amphipod are expected, and these will be described for publication, named and associated DNA barcodes deposited into open access barcoding repositories (e.g. MarBOL).

Molecular phylogenetics

DNA sequences will be generated for all species from both nuclear (18S rRNA) and mitochondrial (16S rRNA and COI) amplicons that allow assessment of the evolutionary relationships with other hadal amphipod species and putative conspecifics from other hadal trenches.

Gene expression analysis

Isolation and subsequent sequencing of the mRNA population present in a tissue biopsy provides a snap-shot of the genes being upregulated in that individual at that time. Accumulated data across individuals can then be used to define the exome of a species, the component of the genome that is formed from the coding regions of genes. To this end, mRNA will be extracted from samples which was immediately stored in RNALater or snap frozen in liquid nitrogen. Species orthologues will be aligned and the ratio of synonymous to non-synonymous mutation calculated for each species as an indicator of selection.

Parasite identification and systematic classification

Individuals will be dissected to extract internal nematode parasites that will be identified using classical morphological features and placed into a broader taxonomic framework from DNA sequences obtained from both nuclear and mitochondrial amplicons.

Genomics

The nuclear genome of *Eurythenes* will be sequenced using shotgun and RAD-based approaches, then constructed and annotated through comparison with arthropod model scaffolds and *de novo* methodologies. Synteny with other arthropods can then be assessed, and regions of the genome showing signatures of selection to hadal environments identified.

Table 16. Numbers of amphipods preserved under different treatments from across deployments. LH = hadal lander; TR = fish trap; WT = wee trap.

Deployment	Depth (m)	EtOH	RNAlater	Snap frozen	Dissected	Formalin
LH01	4823	250	8	8		
LH02	4548	200	40	20		
LH14	5985	100+				
LH15	6112	200+		25		
LH16	5028	25				
LH17	5019	20				
LH18	8006	50		10		
LH19	9040	50		10		
LH20	8702	100+		20		
LH21	10360	26				
LH22	8126		10	10		
TR01	5231	100+	100+	90+		
TR02	4441	50	100	45		10
TR03	6230	50		50		1
TR04	6081	100	50	55	1	1
TR05	7092	100+	50	55	3	7
TR06	6914	4	4	40		
TR06	6914			5	1	
TR07	7497	100		55	2	
TR08	7509	60		30	1	10
TR09	7929	100+	10	65		10
TR09	7929					
TR10	7841	100+		35	2	10
TR11	8028	100+	3	24	5	7
TR12	6898	100+		40	1	
TR13	6974	100+		70	4	5
TR14	6034	100+	30	40		
TR15	6068	200+	50	80	2	
TR16	5072	80	25	30		
TR17	4994	50	30	40		
TR18	7912	100+	10	55		
TR19	7626	50	15	60		5
WT01	6115	~100	10	25		
WT02	6865	~100	25	10	3	

WT03	6961	200	10	10		
WT04	7495	40				
WT05	7502			52		
WT06	7949	100+	10	10		
WT07	7907	100+	15	15		
WT08	7966	30	5			
WT09	6949	100+	1	15	1	
WT10	7237	25				
WT11	8223	10				
WT12	8942	100+	25	50	5	5
WT13	8847	100	25	55	5	5
WT14	10250	30	25	60	3	9
WT15	7957	20				

Reproduction of hadal invertebrates

Caitlin Plowman – Oregon Institute of Marine Biology (under Dr. Craig Young)

Objectives and Methods:

My purpose on this cruise was to collect gonad samples from any hadal invertebrates that were found. Due to the sampling method (baited traps) this was limited to amphipods, with the exception of two shrimp. The gonad samples will be studied using Transmission Electron Microscopy (TEM) in order to learn about reproductive timing (seasonal vs. not), methods (larval type vs. direct development) and output (few vs. many). These samples were fixed in gluteraldehyde and will be post-fixed in osmium. Amphipods not big enough to dissect were preserved in formalin for paraffin histology. These will also be used to study the same type of questions. Learning more about reproduction will hopefully also lead to some insight to trench connectivity.

Preliminary Findings:

Many of the amphipods dissected (Table 17) did not contain visible gonads. This could be the result of a size threshold, but that seems unlikely since from many sites, the amphipods dissected were in the same size range, yet some had gonads and some did not. There could also be an age threshold, but at this time we have no way to verify or deny that. Another hypothesis is that as an adaptation to ephemeral food sources, amphipods only invest energy into producing and maintaining gonads when food is plentiful. Again, there is not currently enough data to test this hypothesis.

Gonads from amphipods in deeper depths, 9000 meters and down, were bright purple in color, while all other amphipod gonads collected were yellow (figure 20). These amphipods appeared to be a *Hirondellea* species that is not found in shallower depths, so this could be a species or genus level characteristic, but not enough information exists at this time to confirm that hypothesis.

The first ever gonad samples from *Alicella gigantea* were obtained on this cruise. Some *A. gigantea* specimens had red vesicles on their ventral surface where brooded eggs would be kept. Initial thoughts were that these vesicles were brooded eggs, but that has since shifted. Current conclusion is that they are unknown structures. They contain liquid and are firmly attached to the joint where the leg meets the ventral surface (scissors were needed to detach

them; Figure 20). The most found on one *A. gigantea* was 6 and the least was 2. After repeatedly finding them on *A. gigantea*, they were then noticed on smaller (2-6 cm) amphipods, possibly *Hirondellea sp.* and *Scopelocheirus sp.* Dr. Alan Jamison hypothesized that they could be glandular swellings that are a result of the decrease in pressure. Dr. Paul Yancey found they had a higher osmotic pressure than seawater, which could induce a rush of seawater into the vesicles causing them to swell. More information is needed to conclude exactly what they are, but samples were taken for TEM and for paraffin histology.

Table 17. Information about the reproductive samples taken on the HADES – M cruise, FK141109, from 9 November to 6 December 2014, in the Mariana Trench.

Site	Depth (m)	Gear (Deployment)	Animal	ID #	# dissected	# w/ gonads	# in formalin	Notes
1	5231	Fish Trap (TR01)	Amphipod	200004	0	0	13	
1	4441	Fish Trap (TR02)	Amphipod	200007	0	0	10	
2	6230	Fish Trap (TR03)	Amphipod	200009	1	0	0	
2	6081	Fish Trap (TR04)	Amphipod	200014	4	1	9	
3	6865	Wee Trap (WT02)	Amphipod	200019	4	3	0	
3	7062	Fish Trap (TR05)	Amphipod	200020	3	3	7	
3	6917	Fish Trap (TR06)	Supergiant Amph	200026	1	1	0	
4a	7497	Fish Trap (TR07)	Supergiant Amph	200034b	2	2	0	
4a	7509	Fish Trap (TR08)	Amphipod	200046	2	1	10	
5a	7929	Fish Trap (TR09)	Amphipod	200061	5	0	10	
6	7841	Fish Trap (TR10)	Amphipod	200069	3	2	10	<i>Princaxellia</i>
7	8028	Fish Trap (TR11)	Amphipod	200082	6	5	3	<i>Princaxellia</i>
7	6949	Wee Trap (WT09)	Supergiant Amph	200083a	1	1	0	
7	6898	Fish Trap (TR12)	Supergiant Amph	200086a	1	1	0	
7	6974	Fish Trap (TR13)	Amphipod	200093	6	4	5	
6b	8223	Wee Trap (WT11)	Amphipod	200115 200101/	0	0	5	
8	6068	Fish Trap (TR15)	Shrimp	2	2	2	0	
10a	8942	Wee Trap (WT12)	Amphipod	200119	12	4	5	Purple
11	8847	Wee Trap (WT13)	Amphipod	200124	8	3	5	Purple
12	10250	Wee Trap (WT14)	Amphipod	200129	5	3	5	Purple
13	7975	Wee Trap (WT15)	Amphipod	200135	0	0	5	Purple

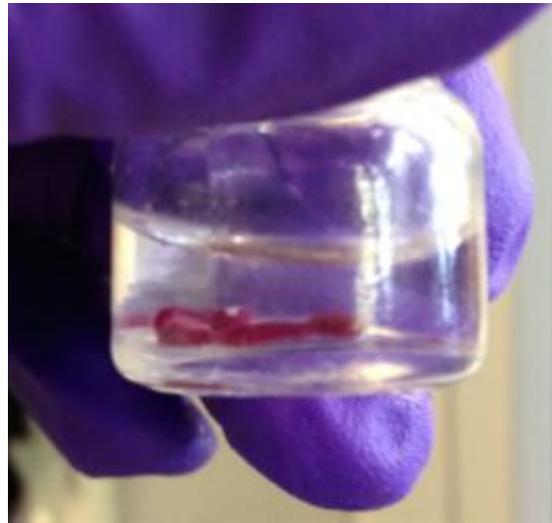


Figure 20. (top left) *Alicella gigantea* with ventral vesicles. (top right) Purple amphipod gonads. (bottom) Yellow amphipod gonads

Investigations into pressure adaptations of hadal animals

Paul H. Yancey (co-P.I.); Anna Downing, Chloe Weinstock (undergraduate assistants), Whitman College

Goals

In this study, we continue to investigate the role hydrostatic pressure, which distorts biomolecules including proteins, in species distributions in the hadal zone. Pressure adaptations fall into two types of mechanisms: i) intrinsic, in which proteins have evolved structural changes to make them more pressure-resistant; and ii) extrinsic, in which proteins are protected from pressure perturbations by "piezolytes," small organic molecules originally discovered as osmolytes (molecules used to regulate cellular water-solute balance). The latter hypothesis was developed in the P.I.'s laboratory with bony fish, whose cells were found to have the osmolyte trimethylamine N-oxide (TMAO) increasing linearly with depth. Work in pressure chambers revealed that TMAO does indeed stabilize deep-sea proteins against pressure perturbations, more than other common invertebrate osmolytes such as taurine and glycine. Note that protein stabilizers are of interest for medical and biotechnology applications.

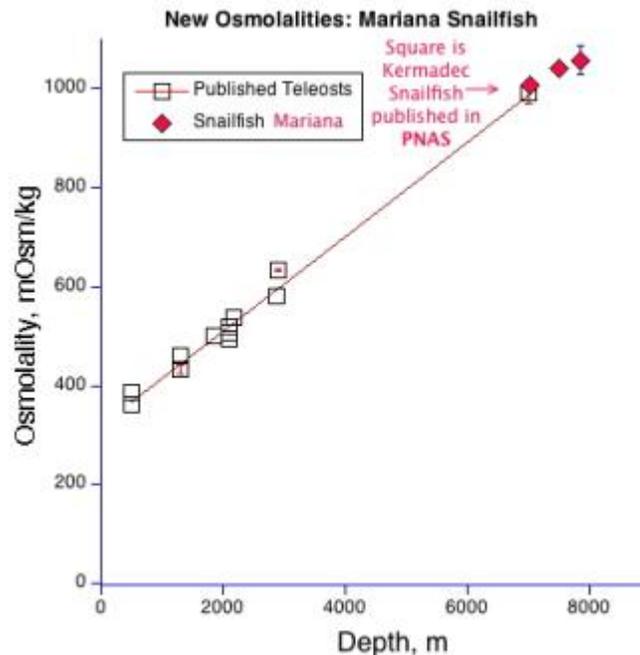
The specific model animals for further testing of these ideas, with results of this cruise, are:

1) Bony Fish: As osmoregulators, shallow teleosts (the major group of bony fish) are hypo-osmotic to seawater (osmolality ~350 milliosmolar or mOsm vs 1000 – 1100 mOsm for full seawater). This osmotic gradient is dehydrating (seawater's high NaCl osmotically draws water out); so these fish must drink seawater and use gill transporters to remove NaCl. However, as TMAO increases with depth, fish osmolality rises. Curiously, no fish of any kind have been found in any study (trawls, baited landers and traps, submersibles) in the deepest regions of the hadal zone. The best documented depth record for a fish is the Japan Trench snailfish *Pseudoliparis amblymopsis* filmed at 7700 m (2008, HADEEP project, Jamieson et al.). We have proposed, based on extrapolations of fish osmolalities, that for fish to enter the deepest waters, TMAO would need to be so concentrated that it would reverse the fishes' osmotic gradient, drawing water into their bodies. Since no fully marine fish are known that can cope with water influx (due in part to inability of their kidneys to remove excess water), this may not be possible.

This hypothesis was supported by analysis of 5 snailfish *Notoliparis kermadecensis* caught at 7000 m in the Kermadec Trench by collaborators Alan Jamieson (Aberdeen), Ashley Rowden (NIWA New Zealand) and Mackenzie Gerringer (then my undergraduate researcher at Whitman College). The fishes' osmolalities (990 mOsm) and TMAO levels almost precisely fit our hypothesis, extrapolating to an estimated isosmotic limit at ~8200 m. Published: Yancey, P.H., M. Gerringer, A. Rowden, J. Drazen, A. Jamieson (2014). Proc. Natl. Acad. Sci. USA (PNAS) 111: 4461-4465.

On this cruise: We filmed and captured a number of Mariana snailfish, probably a new species, down to 8075 m, a new depth record for fish. Mackenzie Gerringer and Thom Linley obtained blood from 7 fish near 7000 m, 2 near 7500 m, and 2 at 7841m. Anna and I measured their osmolalities on the ship (Wescor 5500 osmometer) and results are plotted in the graph here. These data fit the hypothesis quite well and suggest it might be widely applicable rather than trench-specific

Muscle and gel tissues were dissected and frozen at -80°C (by Mackenzie, Thom and Chloe Weinstock). See TABLE of Fish Specimens (submitted separately).



2) Crustacea: Certain taxa are able to go to full ocean depth (~11,000 m), including amphipods (Crustacea) and sea cucumbers (Echinoderms: holothurians). How they can go where fish apparently cannot? Invertebrates are very different osmotically: they are osmoconformers, that is, isosmotic at 1000 – 1100 mOsm, due to high NaCl in body fluids and organic osmolytes such as taurine and glycine inside cells in shallow species. Neither the osmolytes in deep species, which might serve as piezolytes, nor possible intrinsic adaptations, have been studied well. Preliminary results with amphipods caught in the Kermadec Trench from 1500 to almost 10,000 m (HADES-K, Apr.-May 2014) by Gemma Wallace and I revealed depth-related increases in numerous osmolytes, including TMAO, glycerophosphocholine (GPC), scyllo-inositol, and at least 2 unidentified major solutes.

On this cruise: Only crustaceans were obtainable with traps. Amphipods of several species including "supergiants" were obtained from numerous depths from 5000 down to ~10,600 m, sorted by Stuart Piertney, Anna Downing and Caitlin Plowman, and frozen whole for future laboratory analysis of potential piezolytes and for intrinsic protein adaptations to pressure.

In addition, 2 *Benthescymus* decapods were obtained at 6068 m and dissected for tissues. Decapods, like fish, do not appear to reach the deepest hadal zone. Tail muscle from a mysid from 7500 m was also obtained; mysids appear to go deeper than decapods. Osmolytes as potential piezolytes will be analyzed in these as well. As decapods, mysids and amphipods are all Crustacea, they will be an interesting test set. See TABLES of Crustacean Specimens (submitted separately).

Photographs: The Mariana Snailfish; Benthescymus shrimp;
Amphipod from 10,500 m (Sirena Deep)



Future work: for fish and amphipods, we will conduct laboratory analyses of osmolytes and intrinsic protein adaptations to pressure. Having comparable specimens from two trenches may reveal universal adaptations. Since only TMAO has been shown to be a piezolyte in pressure chambers, other osmolytes in amphipods will be identified and tested for piezolyte properties, if any. GPC is a known protein stabilizer in mammalian kidneys, where it protects proteins from the unfolding effects of the waste product urea. Scyllo-inositol is currently being tested to treat the mis-folded amyloid proteins of Alzheimer's Disease. Thus both these solutes enhance proper protein folding and may also do so at high pressure.

Microbial sampling and culturing

Logan Peoples, Scripps Institution of Oceanography

One goal of this expedition was to determine how pressure affects the microbial communities present in the Mariana Trench. These goals were accomplished by evaluating community composition, culture-independent estimates of microbial activity, and culture-dependent viable counts. Samples processed include water from ~1m above the sediment-water interface using the Rock Grabber, pelagic water using a CTD rosette, sediment from the FVCR, Rock Grabber, and Wee Trap, and amphipod-associated microbial communities from the Wee Trap. Major outcomes of this work include: 1) identification of the microbial communities present in an ocean trench 2) an understanding of how these communities change within a trench across depth gradients 3) identification of both community and single-cell physiological adaptations to life in a trench 4) the deepest estimates of pelagic microbial activity 5) novel pressure-adapted microbial isolates.

Water samples were collected for community composition and metagenomics through filtration of ~60L per site onto 3.0, 0.2, and 0.1 μM filters (Table 18) This will allow for comparisons between particle-associated microbes and free-living microbes on different filter sizes. Metagenomics will provide insight into how these communities are physiologically adapted to life in a trench environment. This is the second such experiment detailing how the benthic boundary layer microbial community changes down a trench (with the first being on the HADES Kermadec Trench cruise earlier this year). This will complement sediment community composition and organics analyses done on this cruise and may allow us to disentangle autochthonous vs allochthonous community members and their roles in bringing organic matter into the trench. Water, sediment, and amphipod-associated microbes were collected for single cell genomic analysis, which allows for genome amplification of individual cells. While metagenomics shows whole community metabolic potential it tends to exclude individuals present in low abundance. Previous use of this technique in the Mariana Trench revealed highly diverse and novel microbes.

While the above techniques shows us who is there and what they might be doing, they are incapable of telling us whether these microbes are actually important in these communities. Therefore water samples were incubated using homopropargylglycine (HPG), a methionine analog that is incorporated into newly produced protein, to estimate microbial activity in the deep ocean. Samples were incubated under atmospheric pressure, their *in situ* pressure, and 110 MPa to determine if communities are adapted to their *in situ* pressure. The deepest measurements, prior to these experiments, evaluating the affects of pressure on microbial activity were at 5km depth. Water, sediment, and amphipod-associated microbes were inoculated on Zobell 2216 marine medium to determine viable counts. Samples were incubated under atmospheric pressure, *in situ* pressure, and 110 MPa to again assess the importance of hydrostatic pressure. 2216 is the most common marine medium used for culturing but it's use tends to allow for the growth of a few select individuals from the deep ocean. Therefore these viable counts will be compared to measurements of microbial activity to identify whether these communities are full of pressure-adapted microbes that we simply can't culture. Some sediment samples were amended with sodium acetate and incubated under their *in situ* pressure as previous single cell genomic analysis in the Mariana Trench revealed the presence of a novel, obligately deep sea

microbe capable of acetate oxidation that is yet to be cultured. Water samples were collected for bacterial and viral counts to provide abundance estimates. Water samples were preserved for general nutrient analysis (nitrate, phosphate, silica, nitrite, ammonium).

Table 18. Water samples collected for microbial work

Sample depth (m)	Sample type	Viability?	Activity?	Large volume filtration?	Single cell?	Bacterial counts?	Viral Counts?	General Nutrients?	Enrichment?
0	Underway	X	X	--	--	--	--	--	--
130	CTD	--	--	--	--	--	--	--	--
130	CTD	--	--	X	--	--	--	--	--
500	CTD	--	--	X	--	--	--	--	--
4616	Lander water	X	X	X	X	X	X	X	--
4886	CTD	--	--	X	--	--	--	X	--
4886	CTD	X	X	X	X	X	X	X	--
5025	Lander water	X	X	X	X	X	X	X	--
5025	Lander water, sediment	X	--	X	X	X	X	X	--
6065	Lander water	X	X	X	X	X	X	X	--
6833	Sediment	--	--	--	X	--	--	--	X
6844	Sediment	X	--	--	X	--	--	--	--
6877	Lander water	--	--	X	X	X	X	X	--
6957	Lander water	X	X	X	X	X	X	X	X
7502	Sediment	--	--	--	X	--	--	--	--
7502	Surface water	X	X	--	--	--	--	X	--
7591	Lander water	X	X	X	X	X	X	X	--
7618	CTD water	--	--	X	--	--	--	--	--
7633	Lander water, sediment	X	X	X	X	X	X	X	--
7660	Lander water	--	--	X	X	X	X	X	--
7929	Amphipods	X	X	--	X	--	--	--	X
7939	Sediment	X	--	--	X	--	--	X	X
7942	Sediment	X	X	--	X	--	--	X	X
8028	Amphipods	X	--	--	X	--	--	--	X
8518	Lander water, sediment	X	--	X	X	X	X	X	--
8720	Lander water	--	--	X	X	X	X	X	--
8750	Lander water, sediment	X	X	X	X	X	X	X	--
8847	Amphipods	X	X	--	X	--	--	--	X
9067	Lander water, sediment	X	X	X	X	X	X	X	X
10250	Sediment, amphipods	X	--	--	X	--	--	--	X
10520	Lander water, sediment	X	X	X	X	X	X	X	X

Geological transect across the Mariana Trench at Sirena Deep

Patricia Fryer, SOEST/HIGP, University of Hawaii at Manoa

Background and Objectives

Trench geology is fundamental to our understanding of plate dynamics and formation yet we lack samples from their deepest locations. Geologic investigations were advanced over the last two decades with dives using the Shinkai 6500 submersible (Kaiko was used principally for biological investigations before its loss). Subduction zones like the Mariana system expose mantle peridotite on the deep inner trench wall. These mantle peridotites are serpentized to varying degrees and seeps associated with them support communities of benthic organisms. Recently an active seep area was found at ~5800 m and we suspected that they may exist at greater depths. Severe technical challenges with extremes of hydrostatic pressure have prevented major advances in geological studies, and relegated hadal regions to among the most poorly investigated places on Earth.

The connection between geologic processes and ecosystems in the Mariana Trench region is a function of several factors. Depth and topographic differences among trench and abyssal regions have promoted the formation of genetically distinct species of organisms, acquiring detailed bathymetric and seafloor sonar backscatter data provide constraints on the dispersal of organisms that are constrained by depth (pressure adaptation) to exist in limited bathymetric zones. Augmentation of existing bathymetric data in the region of the transect explored on this cruise will help to guide the biologists in their interpretations of overall ecosystem

Deep exposures of peridotite that have been observed thus far by the *Nereus ROV* and deep landers show that it has been permeated by fluids and serpentized. The fluid could be either sea water or derived from the subducted slab and samples of the rocks and any seep fluids can be used to distinguish these. It was hoped that fluids collected from any mud flows and sediments near seeps at depths greater than 6500 m and closer to the trench than ~30 km would provide near end-member compositions of slab-derived fluids for identification of the dewatering reactions at shallow depths in a subduction zone. After failure of the FVCR lander to release from the sea floor, we were unable to obtain cores that might have helped address this objective.

The geological architecture (sequences of rock types and structures) of Island arcs is accepted as a model for the earliest (Archean) development of continental crust and increasingly is suggested as a site of development of the earliest life on Earth. The architecture of the “arc” lithosphere is currently based on seismic velocity data alone. Composition and textural analysis of rock and sediment samples from the deep inner forearc will advance realistic modeling of the history of middle to lower crust and upper mantle of the arc. Hydrogen and methane from serpentization reactions fuel subsurface microbial populations. We anticipated that it is likely there may be seeps on the inner trench slope in the vicinity of the Sirena Deep because of the faulting associated with the canyon east of Santa Rosa Bank. If so, seep communities, as observed elsewhere on the inner trench slope, might be associated with them. Attempts to recover evidence for such seeps was hampered again by the loss of the FVCR lander, however sediments recovered in several grab sampler deployments did recover serpentized peridotite sands and pebbles.

The project goal was to advance the concept of growth of the arc lithosphere, refine structural provinces of the region and determine how geological processes might influence environments for life in the hadal zone. This was to be accomplished by examining sediment and rocks collected along the transect and by collecting bathymetric data that will refine existing data sets and enhance the ability to interpret geologic processes on the overriding Mariana forearc and the subducting oceanic lithosphere.

Sample Summary

Sediment Cores:

Six core samples were recovered in 3 deployments with the FVCR (Table 6), for depths (ranging from 6844-7942m). As described in J. Bourque's cruise report, all cores were vertically sectioned at 1-cm intervals down to 10 cm, and 5-cm intervals thereafter. For details of disposition of sediment in cores please see table 2 of Bourque's report. Those taken by Fryer will be used for stratigraphic and lithologic analysis post cruise.

Grab and additional samples:

We used Van Veen grab samplers initially on the WHOI lander. These were first deployed with galvanic releases to prevent possible anchoring of the grabs to the seafloor (and potential loss of the lander). The galvanic releases were abandoned after Nov. 17 when both grab samplers were lost. A Van Veen grab sampler was obtained from the EPA office in Guam and brought to the ship on Nov. 18. The ship's engineers fabricated 2 more grab samplers based on the design of the one brought from Guam. We deployed grab samplers on the WHOI lander 16 times and added a grab to the Wee Trap for an additional 5 deployments. We recovered sediments in a total of 12 of these deployments (see table 3 of Bourque's cruise report). Sediment for stratigraphic and lithologic analysis was obtained from all of these recoveries.

Rocks were recovered from 3 of the grab samples. Samples of manganese nodules and altered pumice (Fig. 21) were recovered in grab #7 from 6957 on the subducting plate. Samples of rounded volcanic pebbles were recovered in grab #10 from 5025 m on the subducting plate (Fig 22). Samples of angular volcanic rocks (Fig. 23) were recovered in grab #15 from 8720 m on the Mariana forearc region (area between the trench axis and the volcanic arc). These are the deepest samples ever recovered from the Mariana forearc region.

Details of sample locations are listed in Bourque's cruise report.

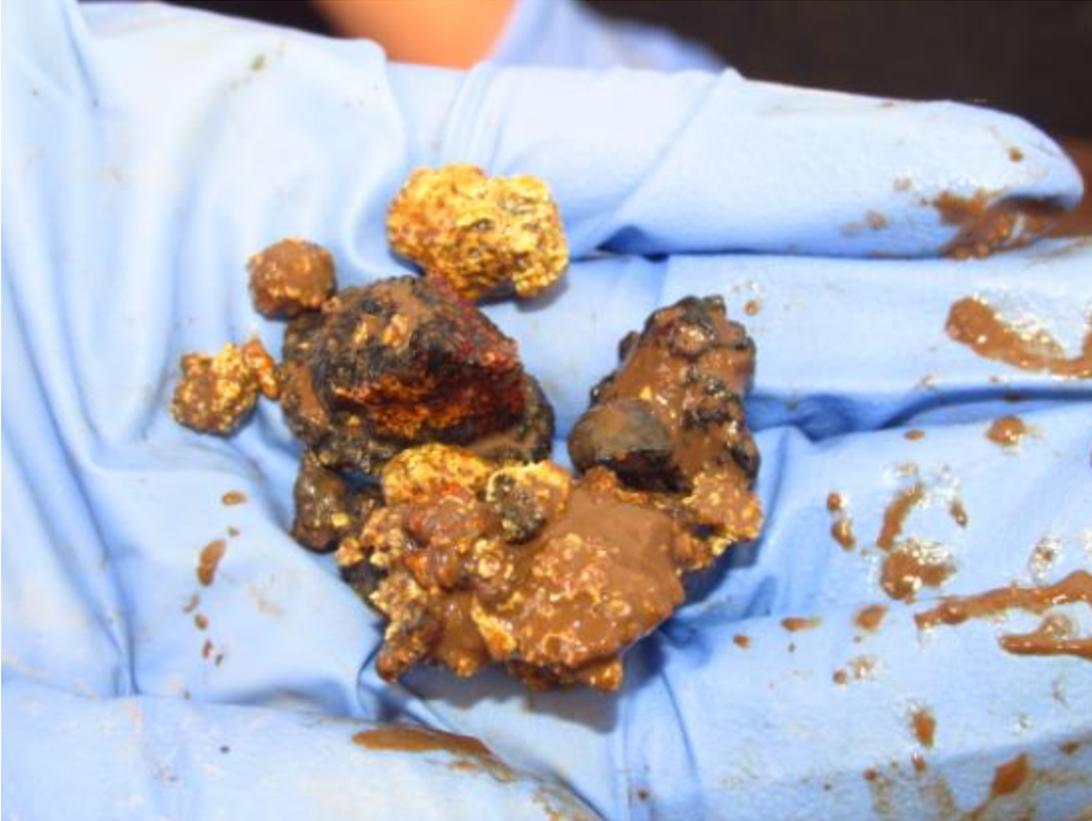


Figure 21. pieces of manganese nodules (dark) and altered pumice (yellow) from RG #07.



Figure 22. Rounded pebbles of volcanic rock coated with a chilled margin of volcanic glass. Collected from the subducting plate in RG #10.



Figure 23. Angular rock fragments from the Mariana forearc in RG #15.

Educational Outreach

Linda Tatreau

Prior to the expedition, SOI Carlie Weiner gave presentations about the cruise to 2 high school classes, a class at the community college, a teacher seminar, and the University of Guam Marine Lab. She was the featured speaker for Science Sunday at the T. Stell Newman Visitor Center and spoke at a meeting of the Fisherman's Co-op. She also appeared on the radio program Where We Live and the TV program The Buzz.

All members of the science team (17) wrote one or more articles for SOI web page Exploring the Marina Trench cruise logs, for a total of 28 articles. Each was accompanied by relevant photos. Also, 4 to 6 daily photos were posted to the website during the entire expedition.

“Sirena Deep” on Facebook, created specifically for this adventure, has 206 “friends.” Posts of current events are made 2 to 3 times daily, always with photos. Sirena has received hundreds of “likes,” dozens of comments and many questions via personal message.

Questions about the cruise have come via Facebook and personal e-mail. More than forty relevant and thought-provoking questions have been received and answered. Other questions, variations on the 40, have also been answered directly to the person asking.

Two successful Skype sessions were conducted with Guam marine biology classes: one at the community college and another with a high school class. Thanks to the ETO who made this possible.

Approximately 225 students from two high schools (9 classes) decorated styrofoam cups after learning about this cruise. The cups went to the bottom of the Sirena Deep. Linda Tatreau will give presentations to each of those classes when she gives the students their pressurized cups.

FK141109 – Media Summary from the Mariana Trench Press Release (as of 12/29/2014)

Carlie Wiener (SOI)

Summary

- Over 151 Articles from 23 countries (United Kingdom, United States, Germany, Australia, Indonesia, China, Canada, Netherlands, Spain, South America, Armania, Iran, Germany, India, Italy, Slovakia, France, Poland, Israel, Russia, Austria, Guam, and International sites)
- Over 123,085 Facebook likes related to release
- Over 3684 Twitter likes
- Over 470 Google Plus likes
- The press release video made #1 watched videos on Facebook Dec 20;
- The story was 'most trending' on Facebook Dec 19
- Over 1, 438, 076 million You Tube views of the SOI videos from the Press Release and 2, 317, 691 views of the same video on the OceanLab You Tube site (As of Dec. 29, 2014).

	Date	Site	Link	Analytics			Country
				FB	Tw	G P	
1.	Dec. 18	BBC	http://www.bbc.com/news/science-environment-30541065	0	0	0	U.K.
2	Dec. 18	KHON	http://www.takepart.com/video/2014/12/27/ghost-try-snailfish-video-shows-worlds-deepest-dwelling-fish-swimming-mariana	1k	23	0	HI, U.S.
3.	Dec. 18	Hawaii Public Radio	http://hpr2.org/post/new-discoveries-deep-mariana-trench	0	0	0	HI, U.S.
4.	Dec. 19	Speigel Wissenschaft	http://www.spiegel.de/wissenschaft/natur/marianengraben-forscher-filmen-fisch-in-8143-metern-tiefe-a-1009623.html	172	59	0	Germany
5.	Dec. 19	NBC	http://www.nbcchicago.com/news/national-international/286417331.html	3	0	22	Chicago, U.S.
6.	Dec. 19	Nature World News	http://www.natureworldnews.com/articles/11332/20141219/ghostly-new-fish-species-discovered-in-darkest-ocean-depths-yet.htm	32	6	30	Int.
7.	Dec. 19	ABC News	http://abcnews.go.com/International/scientists-found-deepest-fish-miles/story?id=27717832	7.3k	641	17	U.S.
8.	Dec. 19	Independent	http://www.independent.co.uk/news/science/video-shows-snailfish-swimming-deeper-than-any-other-creature-at-8000m-beneath-pacific-9937291.html	0	0	0	Int.
9.	Dec. 19	Sci News.com	http://www.sci-news.com/biology/science-footage-worlds-deepest-dwelling-fish-mariana-trench-02362.html	0	0	0	Int.
10.	Dec.19	The Conversation	http://theconversation.com/how-we-found-worlds-deepest-fish-in-the-mariana-trench-and-why-we-must-keep-exploring-35743	21	57	0	U.S.
11.	Dec. 19	ABC News – San Francisco	http://abc7news.com/science/palo-alto-ocean-institute-discovers-new-fish-at-record-depth/444432/	0	0	0	San Fran. U.S.
12.	Dec. 19	Washington Post	http://www.washingtonpost.com/news/science/wp/2014/12/19/ghostly-new-fish-discovered-at-record-breaking-depths/?hpid=z4	0	0	0	WA, US
13.	Dec. 19	National Geographic	http://news.nationalgeographic.com/news/2014/12/141219-deepest-fish-mariana-trench-animal-ocean-science/	14K	37	24	Int.
14.	Dec. 19	New Scientist	http://www.newscientist.com/article/dn26719-weird-sea-ghost-breaks-record-for-deepest-living-fish.html?full=true&print=true#.VJg6wfVEPg	287	121	66	U.S., U.K., Australia
15.	Dec. 19	NBC News	http://www.nbcnews.com/science/weird-science/sea-ghost-scientists-spot-deepest-living-fish-mariana-trench-n271861	0	0	0	U.S.
16.	Dec. 19	LA Times	http://www.latimes.com/science/sciencenow/la-sci-sn-mariana-trench-	0	0	0	L.A., U.S.

			deep-sea-fish-20141219-story.html				
17.	Dec.19	USA Today	http://www.usatoday.com/story/tech/sciencefair/2014/12/19/deepest-fish-discovered-pacific/20659917/	3707	51	0	U.S
18.	Dec. 19	The Province	http://www.theprovince.com/news/world/record+deepest+fish+ever+found+after+Snailfish+discovered/10667497/story.html	0	0	2	B.C., Canada
19.	Dec. 19	US News	http://www.usnews.com/news/articles/2014/12/19/mariana-trench-is-home-to-deep-water-fish	0	0	0	U.S.
20.	Dec. 19	Discovery News	http://news.discovery.com/animals/worlds-deepest-fish-filmed-in-the-mariana-trench-141219.htm	2.6K	126	44	U.S.
21.	Dec.19	Mirror	http://www.mirror.co.uk/news/world-news/worlds-deepest-living-fish-discovered-swimming-4841183	0	0	0	UK
22.	Dec.19	Daily Mail. Com	http://www.dailymail.co.uk/sciencetech/article-2880612/World-s-deepest-fish-Ghostly-snailfish-27-000ft-deep-bottom-Pacific-s-Mariana-Trench.html	971	0	0	U.K.
23.	Dec. 19	IFLS	http://www.iflscience.com/plants-and-animals/bizarre-ghost-animal-new-record-deepest-living-fish	40.4k	156	0	U.S.
24.	Dec. 19	I09	http://io9.com/a-bizarre-new-species-of-fish-has-been-discovered-at-a-1673109432	1.5K	0	0	Int.
25.	Dec. 19	United Press Inter.	http://www.upi.com/Science_News/2014/12/19/Researchers-film-fish-at-record-setting-depth/8561418998164/	0	0	0	Int.
26.	Dec. 19	Voice Chronicle	http://www.voicechronicle.com/201412-new-species-of-fish-discovered-in-mariana-trench	0	0	0	Int.
27.	Dec. 19	The Examiner	http://www.examiner.com/article/snailfish-nicknamed-sea-ghost-breaks-record-for-deepest-living-fish	0	0	0	U.S.
28.	Dec. 19	WECT - NBC	http://www.wect.com/story/27670194/bizarre-looking-creature-breaks-record-for-worlds-deepest-fish	79	0	0	NC, U.S.
29.	Dec. 19	National Monitor	http://natmonitor.com/2014/12/19/new-fish-species-found-at-a-depth-of-5-miles-in-the-mariana-trench/	1048	0	0	D.C., U.S.
30.	Dec. 19	MTV News	http://www.mtv.com/news/2032691/deepest-dwellingfish-mariana-trench/	0	0	0	U.S.
31.	Dec. 19	Techaeris	http://techaeris.com/2014/12/19/newly-discovered-snailfish-found-near-deepest-possible-location/	2	1	0	Int.
32.	Dec. 19	Design and Trend	http://www.designntrend.com/articles/31808/20141219/new-transparent-fish-discovered-smashes-record-for-deepest-fish.htm	24	0	0	Int.
33.	Dec. 19	KaLeo UH Magazine	http://www.kaleo.org/news/uh-scientists-discover-new-species-in-mariana-trench-fish-in/article_f94bc6fc-87ca-11e4-a7cc-f7b53f74621b.html	5	3	0	HI, U.S.
34.	Dec.19	KVEW TV	http://www.kvewtv.com/article/2014/dec/19/whitman-college-professor-and-students-discover-ne/	0	0	0	WA, U.S.

35.	Dec. 19	Utah Peoples Post	http://www.utahpeoplespost.com/2014/12/researchers-found-deepest-fish-ever-recorded-video/	0	0	0	UT, U.S.
36.	Dec. 19	KULR - NBC	http://www.kulr8.com/story/27672501/deepest-dwelling-creatures-on-earth-discovered-by-whitman-college-studentsstaff	0	0	0	WA, U.S.
37.	Dec. 19	Jobs N Hire	http://www.jobshire.com/articles/15880/20141219/worlds-deepest-living-fish-scientists-discover-elusive-snailfish-8000-meters-below-sea-video.htm	0	0	0	U.S.
38.	Dec. 19	Headlines & Global News	http://www.hngn.com/articles/53432/20141219/worlds-deepest-fish-discovered-5-miles-below-oceans-surface-video.htm	0	0	0	U.S.
39.	Dec. 19	All Voice	http://www.allvoices.com/article/100002895	0	0	0	U.S.
40.	Dec. 19	Epoch Times	http://www.theepochtimes.com/n3/1152545-worlds-deepest-dwelling-fish-discovered/	29	20	0	U.S.
41.	Dec. 19	Capital OCT	http://www.capitalotc.com/scientists-went-on-a-journey-to-discover-life-forms-in-the-deepest-parts-of-the-pacific-ocean/26680/	0	0	0	U.S.
42.	Dec. 19	Yahoo News	http://finance.yahoo.com/news/first-footage-worlds-deepest-dwelling-102800361.html	0	0	0	U.S.
43.	Dec. 19	Popular Science	http://www.popsoci.com/found-worlds-newest-deepest-dwelling-fish	65	0	0	U.S.
44.	Dec. 19	Morning Vertical	http://www.morningvertical.com/researchers-have-discovered-the-worlds-deepest-living-fish-in-mariana-trench-watch-video/245069/	0	0	0	TX, U.S.
45.	Dec. 19	Canada Journal	http://canadajournal.net/science/worlds-deepest-fish-video-aberdeen-university-sets-record-deepest-fish-20868-2014/	0	0	0	Canada
46.	Dec. 19	Evening Express	http://www.eveningexpress.co.uk/news/local/video-aberdeen-scientists-film-creatures-from-deepest-place-on-earth-1.746386	0	0	0	U.K.
47.	Dec. 19	STV	http://news.stv.tv/north/304127-aberdeen-university-scientists-discover-deepest-fish-in-mariana-trench/	380	14	0	Scotland
48.	Dec. 19	Scotsman	http://www.scotsman.com/news/transport/aberdeen-university-sets-record-for-deepest-fish-1-3639226	16	22	0	Scotland
49.	Dec. 19	The Guardian	http://www.theguardian.com/environment/2014/dec/19/snailfish-depth-record-pacific-mariana-trench	0	0	0	U.S.
50.	Dec. 19	Maine News	http://newsmaine.net/21762-bizarre-looking-creature-sets-new-record-world-s-deepest-fish	0	0	0	U.S.
51.	Dec. 19	Tech Times	http://www.techtimes.com/articles/22581/20141219/new-fish-discovered-lurking-in-depths-of-mariana-trench.htm	51	7	0	NY, U.S.
52.	Dec. 19	China Topix	http://www.chinatopix.com/articles/28778/20141219/new-species-of-worlds-deepest-fish-discovered-in-the-mariana-trench.htm	25	0	1	China
53.	Dec. 19	Hawaii News Now	http://www.hawaiinewsnow.com/story/27670310/university-of-hawaii-scientists-observe-deep-fish	4	0	0	HI, U.S.
54.	Dec. 19	Repubblica	http://www.repubblica.it/scienze/2014/12/19/news/pesce_record_profo	58	7	2	South

			ndit-103295643/				America
55.	Dec. 19	The Post	http://nieuws.thepostonline.nl/2014/12/19/diepst-levende-vissen-ontdekt-en-gefilmd/	6	3	0	Amsterdam
56.	Dec. 19	La Vanguardia	http://www.lavanguardia.com/natural/20141219/54421982299/descubr-en-nuevo-pez-record-mundial-de-vida-en-el-aguas-profundas.html	563	50	13	Spain
57.	Dec. 19	FOX News	http://www.fox8live.com/story/27670194/bizarre-looking-creature-breaks-record-for-worlds-deepest-fish	0	0	0	New OrL. U.S.
58.	Dec 19	NPR - KPCC	http://www.scpr.org/news/2014/12/19/48778/7-miles-beneath-the-sea-s-surface-who-goes-there/	6	2	0	U.S.
59.	Dec. 19	Whitman College News	http://www.whitman.edu/newsroom/whitman-college-professor-and-students-discover-new-species-in-the-deepest-trench-on-earth	0	0	0	WA, U.S.
60.	Dec. 19	University of Oregon	http://around.uoregon.edu/content/students-probe-worlds-deepest-oceans-research-trip	0	0	0	OR, U.S.
61.	Dec. 19	NPR	http://www.npr.org/2014/12/19/371670931/7-miles-beneath-the-sea-s-surface-who-goes-there	0	0	0	U.S.
62.	Dec. 20	International Biz. Times (IBT)	http://www.ibtimes.com/ghost-sea-creature-breaks-record-deepest-fish-ocean-1764100	106	5	0	Int.
63.	Dec. 20	Houston Chronicle	http://www.chron.com/houston/article/Reports-Strange-new-fish-found-at-5970555.php	99	28	8	Houston, US
64.	Dec.20	Metro	http://metro.co.uk/2014/12/20/scientists-discover-worlds-deepest-fish-4994834/	0	0	0	UK
65.	Dec.20	Newsday	http://www.newsday.com/news/nation/mariana-trench-expedition-spots-fish-5-miles-below-surface-of-pacific-ocean-scientists-say-1.9734336?view=print	0	0	0	U.S.
66.	Dec. 20	9 News.com.au	http://www.9news.com.au/world/2014/12/20/09/48/bizaree-ghost-fish-discovered-at-record-low-depths	6K	33	0	Australia
67.	Dec. 20	Vox	http://www.vox.com/2014/12/19/7423645/deep-fish-record	56	52	0	Int.
68.	Dec. 20	National Post	http://news.nationalpost.com/2014/12/19/new-record-for-deepest-fish-ever-found-after-new-snailfish-discovered-8145m-down-the-mariana-trench/	224	0	0	Canada
69.	Dec. 20	Telegraph UK	http://www.telegraph.co.uk/news/worldnews/australiaandthepacific/11303017/Worlds-deepest-fish-found-in-Pacific-Oceans-Mariana-Trench.html	11k	425	0	U.K.
70.	Dec. 20	Football Examiner	http://thefootballexaminer.com/technology-22/record-researchers-filmed-fish-in-eight-kilometers-deep-6589.html	0	43	0	Int.
71.	Dec. 20	Macro Insider	http://www.macroinsider.com/technology/new-fish-discovered-lurking-in-depths-of-mariana-trench-h700.html	13	5	3	TX, U.S.
72.	Dec. 20	Big Think	http://bigthink.com/ideafeed/scientists-discover-fish-living-deeper-	371	62	23	NY, U.S.

			than-any-previously-recorded				
73.	Dec. 20	Mother Nature Net.	http://www.mnn.com/earth-matters/animals/stories/worlds-deepest-fish-discovered-5-miles-under-the-ocean-surface	2.3K	106	0	Int.
74.	Dec. 20	Tweak Town	http://www.tweaktown.com/news/42143/new-ghostly-snailfish-swimming-great-depths-mariana-trench/index.html	60	4	2	CA, U.S.
75.	Dec. 20	California Diver	http://californiadiver.com/new-fish-species-discovered-at-the-bottom-of-the-mariana-trench-26000-feet-deep/	0	0	0	CA, U.S.
76.	Dec.20	Pioneer News	http://www.piercepioneer.com/new-fish-species-discovered-bottom-mariana-trench-2/36089	0	0	0	U.S.
77.	Dec.20	PBS News Hour	http://www.pbs.org/newshour/runtdown/new-fish-species-dwells-deep-sea/	938	48	0	U.S.
78.	Dec. 20	Sports Act	http://sportact.net/worlds-deepest-fish-found-pacific-oceans-mariana-trench-watch-5707-2014/	0	0	0	Int.
79.	Dec. 20	Northern Voices	http://nvonews.com/ethereal-deep-sea-fish-lives-5-miles-underwater-report/	11,027	0	0	Int.
80.	Dec. 20	Raw Story	http://www.rawstory.com/rs/2014/12/how-we-found-worlds-deepest-fish-in-the-mariana-trench-and-why-we-must-keep-exploring/	0	0	0	U.S.
81.	Dec. 20	Inquisitr	http://www.inquisitr.com/1692978/new-species-of-angel-like-deep-sea-fish-caught-on-tape-discovered-by-accident-video/	9	6	2	U.S.
82.	Dec. 20	Digital Journal	http://www.digitaljournal.com/science/video-world-s-deepest-living-fish-26-000-ft-in-mariana-trench/article/421538	28	5	2	Int.
83.	Dec. 20	Armen Press	http://armenpress.am/eng/news/788656/new-record-ethereal-deep-sea-fish-lives-5-miles-underwater.html	0	0	0	Armania
84.	Dec. 20	AOL	http://travel.aol.co.uk/2014/12/20/world-deepest-fish-found-snailfish-weird-animals/	11	0	0	U.K.
85.	Dec. 20	Dunya News	http://dunyaNews.tv/index.php/en/WeirdNews/251014-Scientists-discover-the-worlds-deepest-fish-	0	0	0	Iran
86.	Dec. 20	Apex Tribune	http://www.apextribune.com/expedition-finds-new-fish-species-at-a-depth-of-5-miles-in-mariana-trench/22511/	0	0	0	VA, U.S.
87.	Dec. 20	Wall Street	http://www.wallstreetotc.com/scientists-discovered-weird-snailfish-in-the-mariana-trench/213534/	0	0	0	LA, U.S.
88.	Dec. 20	Perez Hilton	http://perez Hilton.com/2014-12-20-new-fish-species-discovered-ocean-deep#.VJ7OUvVFA	54	0	0	U.S.
89.	Dec. 20	Science Daily	http://www.sciencedaily.com/releases/2014/12/141220040544.htm	0	0	0	U.S.
90.	Dec. 20	Frankfurter Allgemeine	http://www.faz.net/aktuell/wissen/rekordfund-bei-8143-metern-die-gespenster-der-tiefe-13332330.html	381	22	0	Germany
91.	Dec. 21	ABC News – Social Climber	http://abcnews.go.com/US/worlds-deepest-fish-found-mariana-trench/story?id=27747990 Top Search Trends	0	0	0	U.S.

92.	Dec. 21	Full-time Whistle	http://full-timewhistle.com/science-27/ghostly-fish-breaks-ocean-depth-record-556.html	0	0	0	
93.	Dec.21	Guardian Liberty Voice	http://guardianlv.com/2014/12/mariana-trench-the-deepest-point-on-earth/	4	58	2	Vegas, U.S.
94.	Dec. 21	Inferse	http://www.inferse.com/21330/mermaid-like-sailfish-found-8143-meters-sea-level-mariana-trench-video/	8	0	3	
95.	Dec. 21	Flight Centric	http://www.flightcentric.com/fantastic-ghost-fish-found-breaking-the-record-for-deepest-living-fish-news-1	0	0	0	
96.	Dec. 21	Weather Channel	http://www.weather.com/science/nature/news/mariana-trench-fish-record	0	0	0	U.S.
97.	Dec. 21	CB Sport	http://www.cbsport.org/2014/12/fish-discovered-mariana/	0	0	0	U.K.
98.	Dec. 21	Geek Infinite	http://geekinfinite.com/news/2014/12/researchers-unveiled-new-fish-species-living-26715-feet-underneath-sea-surface/	0	0	0	
99.	Dec. 21	West Texas News	http://wtexas.com/content/14121393-scientists-find-deep-living-fish-mariana-trench	0	0	0	TX, U.S.
100	Dec. 21	The Capital Wide	http://www.thecapitalwide.com/1797/live-fish-species-found-under-the-ocean-surface/	35	29	5	Int.
101	Dec.21	Geek	http://www.geek.com/science/researchers-find-tissue-paper-fish-at-record-depth-in-mariana-trench-1612145/	0	0	0	Int.
102	Dec.21	Dehli Daily News	http://www.delhidailynews.com/news/Scientists-discover-worlds-deepest-dwelling-fish-species-1419175990/	4.9k	740	17 0	India
103	Dec.21	Science Recorder	http://www.sciencerecorder.com/news/new-species-of-fish-found-at-record-breaking-depths-in-mariana-trench/	0	0	0	VA, U.S.
104	Dec.21	Tasnim News	http://www.tasnimnews.com/English/Home/Single/595307	0	0	0	Iran
105	Dec.21	America Herald	http://www.americaherald.com/worlds-deepest-ever-snailfish-found-5-miles-down-in-mariana-trench/21589/	0	0	0	U.S.
106	Dec. 22	News.com.au - science	http://www.news.com.au/technology/science/scientists-film-worlds-deepest-fish-during-mariana-trench-expedition/story-fnjwkt0b-1227163623655	0	0	0	Australia
107	Dec. 22	Perfect Science	http://perfscience.com/content/214910-scientists-hawaii-find-sailfish-deep-underground-waters-mariana-trench	0	0	0	Ohio, U.S.
108	Dec. 22	Morning Ticker	http://www.morningticker.com/researchers-found-worlds-deepest-unidentified-fish-five-miles-below-the-surface/15525/	0	0	0	U.S.
109	Dec. 22	Science Work News	http://www.scienceworldreport.com/articles/20340/20141222/new-species-discovered-deepest-trench-earths-oceans.htm	8	10	0	N.Y., U.S.
110	Dec. 22	Empire State Tribune	http://www.esbtrib.com/2014/12/22/3016/record-breaking-discovery-life-forms-5-miles-deep-marianas-trench/	0	0	0	
111	Dec. 22	Frontline Desk	http://www.frontlinedesk.com/2014122446-tissue-like-fish-found-extreme-depth-mariana-trench/	0	0	0	

112	Dec. 22	Mod Vive	http://www.modvive.com/2014/12/21/worlds-deepest-fish-found-dwelling-mariana-trench/	824	3	2	
113	Dec. 22	Marianna Trench Variety	http://www.mvariety.com/cnmi/cnmi-news/local/72212-world-s-deepest-fish-found-in-marianas-trench	0	0	0	Guam
114	Dec. 22	Science World Report	http://www.scienceworldreport.com/articles/20340/20141222/new-species-discovered-deepest-trench-earths-oceans.htm	8	10	0	Int.
115	Dec.22	Press TV	http://www.presstv.ir/detail/2014/12/22/391387/new-species-found-in-western-pacific/	10	32	15	Iran
116	Dec. 22	Scientific Computing	http://www.scientificcomputing.com/news/2014/12/new-species-found-greatest-depths-ever-recorded	2	1	0	U.S.
117	Dec. 22	Cienciay Biologia	http://cienciaybiologia.com/nuevas-y-sorprendentes-especies-de-peces-y-anfipodos-en-la-fosa-de-las-marianas/	19	10	3	Spain
118	Dec. 22	Focus It	http://www.focus.it/ambiente/animali/il-pesce-fantasma-nella-fossa-delle-mariane	0	0	0	Italy
119	Dec. 22	Green Report It	http://www.greenreport.it/news/aree-protette-e-biodiversita/fossa-delle-mariane-scoperto-pesce-fantasma-vive-fondo-tutti-video/#prettyPhoto[videospot]/0/	40	7	0	Italy
120	Dec. 22	Ch. 11 NBC	http://www.11alive.com/story/news/weird/2014/12/22/deepest-fish-discovered-pacific/20788401/	997	8	0	Atlanta, U.S.
121	Dec. 23	NY Daily News	http://www.nydailynews.com/news/world/fish-found-historic-depth-27-000-feet-article-1.2051915	18	6	0	New York, U.S.
122	Dec. 23	Belfast Telegraph	http://www.belfasttelegraph.co.uk/news/world-news/deepest-fish-found-by-scientists-at-27000ft-30850900.html	145	50	4	Ireland
123	Dec. 23	Toronto Sun	http://www.torontosun.com/2014/12/19/the-sea-ghost-scientists-discover-new-fish	3	7	0	Toronto Canada
124	Dec. 23	UH News	http://www.uhm.hawaii.edu/news/article.php?aId=6926	0	0	0	Hawaii
125	Dec. 23	Huffington Post	http://www.huffingtonpost.com/2014/12/19/deepest-fish-discovered-mariana-trench-snailfish_n_6357680.html	6.8k	87	0	U.S.
126	Dec. 23	CNN Indonesia	http://www.cnnindonesia.com/teknologi/20141223111235-199-19966/udang-super-besar-terekam-di-laut-terdalam-di-bumi/	0	0	0	Indonesia
127	Dec. 23	Webnoviny Slovak News	http://www.webnoviny.sk/zaujímavosti/clanok/903736-video-vedci-nafilmovali-neznamy-druh-ryby-v-rekordnej-hlbke/	0	0	0	Slovakia
128	Dec.24	Veo Verde	http://www.veoverde.com/2014/12/descubren-nuevos-peces-y-crustaceos-que-viven-en-el-lugar-mas-profundo-de-la-tierra/	263	312	0	Spain
129	Dec. 24	Saipan Tribune	http://www.saipantribune.com/index.php/marianas-trench-may-biological-hot-spot/	4	3	0	Saipan
130	Dec. 24	American	http://americanlivewire.com/2014-12-22-new-species-found-greatest-	1	2	1	U.S.

		Livewire	depths-ever-recorded-mariana-trench-video/				
131	Dec. 24	Maxi Sciences	http://www.maxisciences.com/poisson/un-poisson-inconnu-decouvert-a-plus-de-8000-metres-de-profondeur_art34040.html	764	0	0	France
132	Dec.25	NPR	http://www.npr.org/blogs/thetwo-way/2014/12/25/372894314/unexpected-life-found-in-the-oceans-deepest-trench	0	0	0	U.S.
133	Dec.25	Tri-City Herald	http://www.tri-cityherald.com/2014/12/25/3329596_whitman-professor-among-researchers.html?rh=1	0	0	0	WA, U.S.
134	Dec.25	KUAM	http://www.kuam.com/story/27708583/2014/12/26/scientists-make-groundbreaking-discovery-in-marianas-trench	16	0	3	Guam
135	Dec. 25	Wyborcza	http://wyborcza.pl/1,75248,17182160,Ryba_niczym_duch_odkryta_na_rekordowej_glebokosci.html	5	15	0	Poland
136	Dec. 25	Jerusalem Dispatch	http://www.jerusalemdispatch.com/scientists-discover-worlds-deepest-fish-565.html	0	0	0	Israel
137	Dec. 26	Customs Today	http://customstoday.com.pk/tissue-like-fish-discovered-at-depth-of-8145-meters-in-mariana-trench-3/	0	0	0	U.S.
138	Dec. 26	Care 2	http://www.care2.com/causes/mariana-trench-discovery-proves-we-need-to-explore-the-ocean.html	102	35	0	CA, U.S.
139	Dec. 26	Catholic Online	http://www.catholic.org/news/green/story.php?id=58173	0	0	0	U.S.
140	Dec. 26	New Hampshire Voice	http://nhv.us/content/14121247-mariana-trench-turns-out-be-biological-hot-spot	0	0	0	NH, U.S.
141	Dec. 27	Walla Walla Union Bulletin	http://union-bulletin.com/news/2014/dec/27/whitman-researchers-discover-new-deep-sea-species/	0	0	0	WA, U.S.
142	Dec. 27	Capital Berg	http://www.capitalberg.com/researchers-found-angel-like-fish-deep-ocean-trench/22053/	0	0	0	TX, U.S.
143	Dec. 27	National Monitor	http://natmonitor.com/2014/12/27/angel-like-fish-discovered-in-deepest-ocean-trenchvideo/	1062	0	0	DC, U.S.
144	Dec. 27	Tech Gen Magazine	http://www.techgenmag.com/2014/12/27/mystery-fish-sets-new-record-deepest-swimming-marine-life/	9	8	0	U.S.
145	Dec. 27	Market Biz. News	http://marketbusinessnews.com/first-time-supergiant-amphipod-filmed-alive-5-miles-underwater/42466	0	1	1	U.S.
146	Dec. 27	SMN Weekly	http://www.smnweekly.com/new-species-of-fish-found-below-8000-meters/6613/	0	0	0	U.S.
147	Dec. 27	Boxing Dispatch	http://boxingdispatch.com/sci-tech-3/scientists-discovered-new-species-of-fish-8000-meters-underwater-19.html	7	0	0	U.S.
148	Dec. 27	Take Part	http://www.takepart.com/video/2014/12/27/ghost-try-snailfish-video-	0	0	0	U.S.

			shows-worlds-deepest-dwelling-fish-swimming-mariana				
149	Dec. 27	Kronen Zeitung		0	0	0	Austria
150	Dec. 29	Tekno	http://www.tempo.co/read/news/2014/12/22/061630250/Spesies-Baru-Ditemukan-di-Parit-Terdalam-Bumi	0	0	0	Indonesia
151	Dec. 29	KM RU	http://www.km.ru/science-tech/2014/12/29/issledovaniya-rossiiskikh-i-zarubezhnykh-uchenykh/753013-na-dne-marianskoi-v	0	0	0	Russia
152	Jan.3	Earth Sky	http://earthsky.org/earth/new-video-shows-life-in-the-deepest-ocean	122	116	62	U.S.
153	Jan.5	UH News	http://www.hawaii.edu/news/2015/01/05/new-species-and-surprising-findings-in-the-mariana-trench/	0	0	0	HI, U.S.
154	Jan. 5	ElMundo	http://www.elmundo.es/ciencia/2015/01/05/54a98cd722601d473b8b4582.html	76	15	5	Spain
155	Jan. 7	AAAS	http://news.sciencemag.org/biology/2015/01/video-gelatinous-sac-gives-deep-sea-fish-swimmer-s-body	33	18	4	U.S.
156	Jan. 8	Scientific American	http://blogs.scientificamerican.com/artful-amoeba/2015/01/08/deepest-fish-features-angel-wings-tentacles-and-amazing-ability-to-perform-under-pressure/?print=true	21	9	6	U.S.