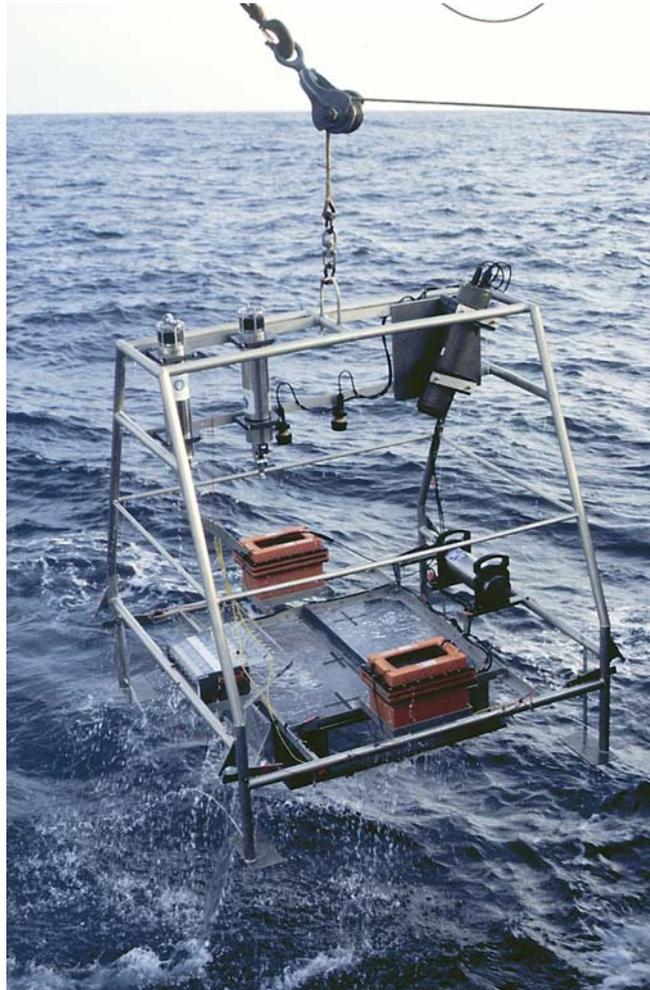




RRS *Discovery* Cruise 250

Metabolism, activity and distribution patterns in demersal deep-sea fish



15th September-10th October 2000

Principal Scientist: Dr Martin A Collins



Cover picture: The FRESP lander being brought on board RRS Discovery following a deployment to a depth of 4000 m.

Discovery 250 Scientific Party

Martin Collins	Aberdeen University
Monty Priede	Aberdeen University
Phil Bagley	Aberdeen University
David Bailey	Aberdeen University
Camila Henriques	Aberdeen University
John Pringle	Aberdeen University
Richard Paterson	Aberdeen University
Emma Battle	Aberdeen University
Oliver Yates	Aberdeen University
Andrew Stocks	Aberdeen University
Judith Brewster	Aberdeen University
Dave Billet	SOC
Ben Boorman	SOC
Ben Wigham	SOC
Kerry Howell	SOC
Francisco Solis-Marin	Southampton University
Catherine Pearson	Southampton University
Jean-Francois Rees	University of Louvain
Alexis de Kerchove	University of Louvain
Julian Partridge	University of Bristol
Amanda Brindley	Queen Mary College
Hans-Joachen Wagner	University of Tuebingen
Uli Mattheus	University of Tuebingen
Jason Scott	RVS (TLO)
Rob Lloyd	RVS
John Wynar	RVS
Alan Sherring	RVS
Bob Keogh	RVS

Ships Company

Robin Plumley	Master
Richard Warner	Chief Officer
Malcolm Graves	2 nd Officer
Annabel Evans	3 rd Officer
Ian McGill	Chief Engineer
Jim Royston	2 nd Engineer
Steve Bell	3 rd Engineer
Ray Perriam	3 rd Engineer
Dave Stewart	ETO (until 30 th September)
Greg Lewis	CPOD (Bosun)
Peter Bennet	POD
Dave Buffery	SG1A
John Dale	SG1A
Harry Hebson	SG1A
Steve Day	SG1A
Nigel Tuppenney	SG1A
Keith Pringle	Motorman
Clive Perry	Catering Manager
Peter Lynch	Chef
Wally Link	Steward
Mick Stephen	Steward
Andy Duncan	Steward

RRS *Discovery* Cruise 250 scientists and crew



Discover 250 Scientific Party, left to right: Ben Wigham, Dave Billet, Amanda Brindley, Emma Battle, Steve Day, Phil Bagley, Andy Stocks, John Dale, Oliver Yates, Catherine Pearson, Camila Henriques, David Bailey, Judith Brewster, Robin Plumley (Master), Steve Bell, Harry Hebson, Richard Paterson, John Pringle, Martin Collins, Jim Royston, Jean-Francois Rees, Hans-Joachen Wagner, Alexis de Kerchove, Julian Partridge, Uli Matheus. (Absent scientists: Francisco Solis-Marin, Kerry Howell, Ben Boorman, Jason Scott, Rob Lloyd, Bob Keogh, Alan Sheering & John Wynar).

Itinerary

Depart: Southampton, Empress Dock
Arrive: Southampton, Empress Dock

Friday September 15th 2000
Tuesday October 10th 2000

Background

The cruise was funded (20 days) by a NERC grant (GR3/12789: Metabolism, activity and distribution patterns of deep-sea demersal fishes: *In situ* oxygen consumption, activity and fast starts in relation to depth, season and temperature in the NE Atlantic and Eastern Mediterranean) awarded to Priede, Collins & Bagley. It was the first of a series of 5 cruises planned for the next three years. The main objectives of the project are:

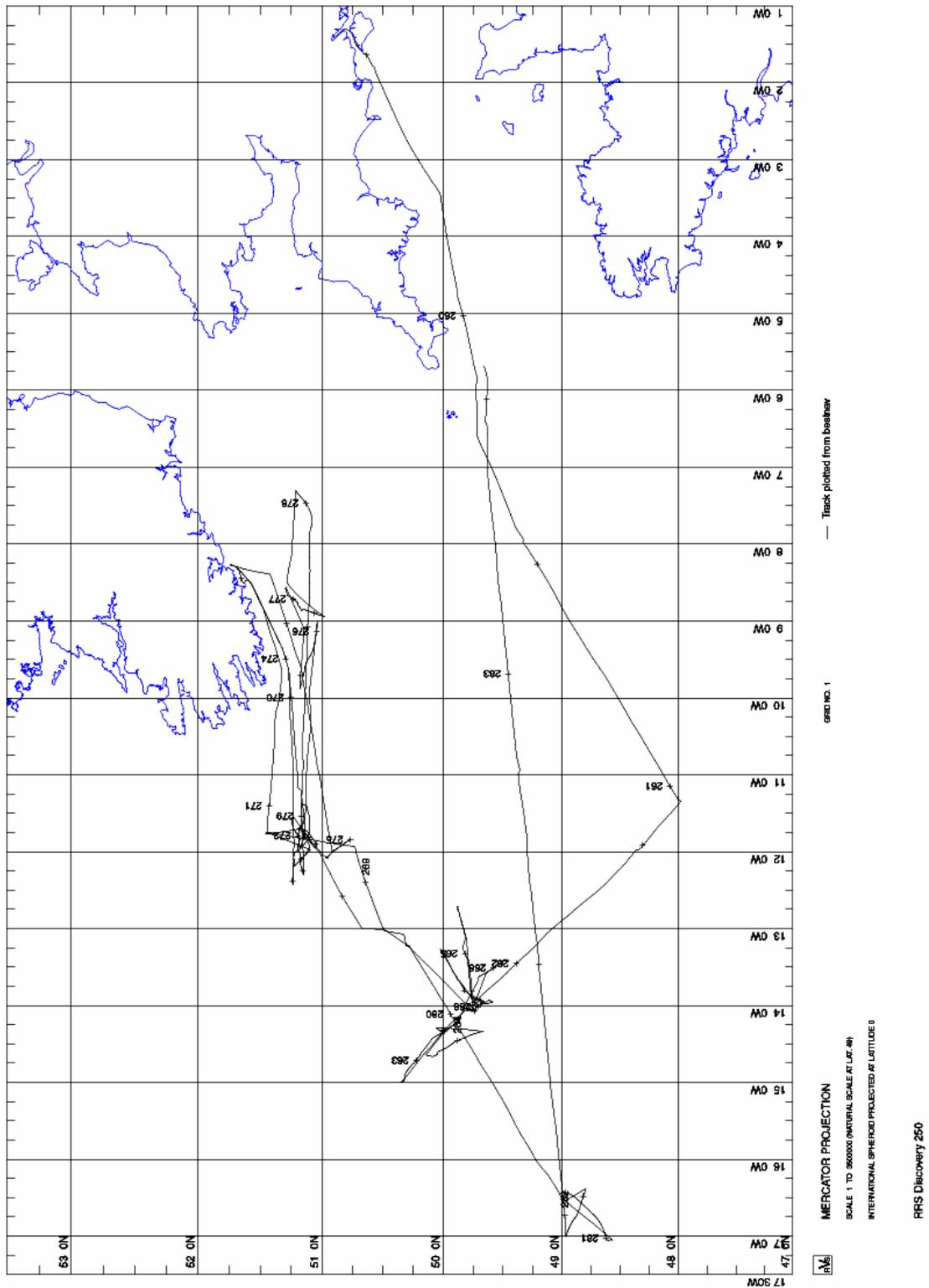
- 1. Determine routine metabolism and activity of demersal fishes in relation to seasonal and environmental parameters.** This objective to be achieved using autonomous lander vehicles to conduct experiments *in situ* on the ocean floor. The Aberdeen University Deep Ocean Submersible (AUDOS) will collect data on routine swimming speed and three new systems will collect data on resting metabolism (FRESP), fast starts (Video Lander) and long-term scavenger abundance and activity (DOBO). During the present cruise the AUDOS will be used routinely and the FRESP tested for the first time.
- 2. Temporal changes in demersal fish populations in the Porcupine Seabight.** This objective will be achieved using trawl sampling (OTSB) and baited camera data (AUDOS) to examine seasonal and inter-annual changes in the composition of the ichthyofauna.

An additional five days sea-time were funded by Dave Billet's group at SOC to allow time for sampling at PAP. Richard Lampitt (SOC) provided a further 1.5 days funding for mooring work.

Specific Objectives

1. Test the new FRESP lander with a series of deployments at various depths, to investigate routine oxygen consumption of scavenging fish.
2. Deploy the AUDOS at depths of 1000-4800 m to determine bathymetric trends in scavenging fauna composition and acoustically track scavenging fish.
3. Deploy the ISIT lander to investigate and quantify bioluminescent light at different depths in the PSB and at PAP.
4. Use the OTSB to determine the distribution and abundance of fish and invertebrates in the PSB (800-4200 m). In addition samples will be used for molecular analysis, enzyme assays and work on brain morphology and visual pigments.
5. Use the OTSB to determine the abundance of holothurians at PAP and, in particular, to determine if the dominance of *Amperima rosea* continues.
6. Obtain multi-core samples from PAP.
7. Service Bathysnap
8. Recover DRA mooring on the Goban Spur.
9. Service sediment trap at PAP.

The Cruise Track of Discovery 250



Cruise Narrative

Wednesday September 13th

0900: Mobilisation of Discovery began, with all of the Aberdeen University gear and much of the RVS and SOC loaded.

Thursday September 14th

0830: Continued loading the ship. Assigned the students and technicians responsibility for each of the landers. Remainder of scientific party arrived during the afternoon.

1500: Familiarisation and safety brief.

Friday September 15th

1015: Departed from SOC in heavy rain. Heading out towards the English Channel, aiming initially for the mouth of the Porcupine Seabight (4000 m).

1100: Scientific meeting to introduce scientific party to each other. Weather F4/5 with showers.

Provisional schedule for the cruise agreed with the captain. Aberdeen landers assembled in the hanger. Agreed to switch to GMT overnight.

1615: Boat drill, completed by 1700.

Saturday September 16th

0100: Clocks back to GMT. Weather moderated overnight and now calm and mostly clear.

1000: Briefing with captain, 1st officer, Jason Scott (RVS TLO) and Dave Billet to discuss plans for the cruise. Heading for 4000 m depth close to the location of the DRA mooring, in order to do CTD drops overnight and then recover the DRA mooring in the morning. Detailed schedule prepared for first two days.

1030: PES fish put in the water.

Sunday September 17th

0100: On CTD station.

0142: First CTD, with four water bottles and three acoustic releases. Completed and on board by 0428, with all three releases having fired.

0515: Second CTD station began, with three more Aberdeen releases, all released successfully. CTD back on deck at 0759.

0800: Proceeded to DRA mooring location.

1105: Hove to at DRA mooring location.

1118: Mooring released from 2076 m.

1139: First buoyancy on surface, grappled quickly but recovery was slightly delayed due to the mooring being tangled. Eventually on board at 1215 and transferred to the port container slot.

1250: Heading for the PSB, 4000 m location. Weather deteriorated through the afternoon and was F7 by the evening.

Monday September 18th

0730: On proposed FRESP station at, but weather poor and decided to deploy the ISIT instead. Landers moved out of the hanger to allow the ISIT out.

1012: ISIT deployed at 50 00 03 N, 14 20.24 W (13904#1). Wind had dropped but still heavy swell. Deployed with slightly more buoyancy than before, so descent monitored. Descended at 42 m/min.

1035: Proceeding to AUDOS location. Sunfish spotted on the surface, being pecked at by a variety of sea birds, tried to pick it up, but failed.

1300: Reached AUDOS station at 1400, but AUDOS not ready

1520: AUDOS deployment began, flag disappeared at 15:30 (13905#1).

1555: Weather too poor for FRESP, so heading to start OTSB.

1818: Began shooting OTSB at 4000 m (50 01.1N, 14 21.8 W; 13906#1)). Weather remains poor (F7), with no sign of immediate improvement.
2253: Net on bottom. Continued trawling overnight.

Tuesday September 19th

0235: Commenced hauling.
0638: Trawl on board in heavy weather (F7), but with no problems. Good catch of fish and invertebrates, but no *Amperima*. Invertebrate catch included *Psychropotes*, *Oneirophanta*, many asteroids and two *Grimpoteuthis* octopods. Fish catch dominated by *C. armatus*, with some *C. leptolepis*, a couple of *H. bathybius* and a single *Spectrunculus grandis*.
0652: Heading for the ISIT station, 3 hours away.
1000: Arrived at ISIT location, but weather too rough for recovery, so holding station. Remained close to the ISIT all day, but weather not suitable for recovery.
1700: Headed south towards a possible trawl location (4250 m) that will take us back towards the ISIT in the morning. Wind eased after dinner. Trawl prepared.
2045: Began shooting OTSB (13907#1). Trawling NW into swell.

Wednesday 20th September

0340: Commenced hauling, but OTSB did not come off the bottom easily and there was concern about snagging a sub-sea cable.
0805: Net hauled in and on deck, but cod end was lost, with a very clean tear. A few invertebrates in the net including a single *Amperima* and a piece of wood from a ship wreck. With just one net left we will aim to do some work at 2500 m next to ensure that we sample that depth. In the meantime the possibility of have spare nets sent to Cork is being investigated.
0815: Headed for the ISIT location.
0945: Arrived at ISIT.
0950: Released the ISIT, although the first release appeared not to fire. Swell now coming from the north, with 15 knot wind from the west. Navigation lights needed repair.
1120: ISIT on surface, and approached first into the swell, but this approach was abandoned and we approached instead into the wind. ISIT recovered smoothly.
1220: ISIT on deck.
1245: Headed for the AUDOS location. Wind remain light, but is forecast to increase later.
1415: Arrived at AUDOS.
1420: AUDOS released.
1550: AUDOS on surface.
1625: AUDOS on board. Moved the landers around deck.
1647: Proceeded towards the FRESP location.
1756: Deployed the FRESP in calm conditions and monitored it to the sea-floor. Found that the AUDOS camera had not operated properly during the deployment and that the new current meter had also failed! Fish tracking worked well however. Switched the Sensortec current meter to the AUDOS from the ISIT.
2046: Reached ISIT location.
2058: ISIT deployed (13909#1).

Thursday 21st September

0054: Commenced survey of OTSB station.
0241: Began shooting OTSB (13910#1, 2500 m). Problem with cable metering system slowed down the paying out of the main warp.
0539: Net on bottom.
0642: Net off bottom.
0853: Trawl brought on board, reasonable catch, with *C. guentheri*, *A. rostrata*, *H. bathybius*, *S. grandis* and *L. carapinus* and *C. armatus*. Invertebrates included large crab (*Neolithodes*) and lots of ophiuroids. Catch took a while to process due to ID problems and high diversity.
0920: Proceeded to ISIT station, but wind increasing again.
1300: Over ISIT location
1312: ISIT released.

1433: ISIT on surface, grappled at 14:45 and on board by 14:55. Secured the ISIT on board and proceeded to the location for the AUDOS deployment (49 40 N, 13 57 W).
1512: Deck secured and headed to AUDOS location.
1602: Hove to at AUDOS location.
1652: Deployed AUDOS (13911#1). There was a delay when half of the ballast fell off when it knocked the stern ramp, with one CAT lost. AUDOS brought back on board with the buoyancy trailing and new ballast fitted. Port crane leaked hydraulic fluid during deployment. A hose was split and needed replacement, so Discovery remained on station for 2 hours whilst repair was undertaken.
1854: Crane repairs complete, proceeded to CTD station.
1915: Arrived at CTD station and CTD in the water at 19:28 (13912#1). Collected three water bottles from 10 mab at 4023 m depth.
2201: CTD on board and headed for the ISIT deployment location. Wind blowing steady 30 knots.
2320: On ISIT station.
2334: ISIT deployed (13913#1). Headed towards trawl station.

Friday 22nd September

0143: Commenced survey of OTSB ground.
0338: Began shooting OTSB (13914#1, 3100 m) but there was a problem with door monitor, so brought back to the surface to be checked.
0730: Net on bottom.
0836: Commenced hauling
0855: Net of bottom. Arrangements being made to proceed to Cork to collect 3 OTSB nets, which are being sent from SOC.
1134: Trawl on deck, with large catch of the holothurian, *Benthothuria*. Reasonable fish catch with *C. armatus*, *H. macrochir*, *H. bathybius*, *C. leptolepis* and *C. carapinus*.
1200: Headed for the AUDOS location.
1331: Released AUDOS. AUDOS rose slowly as it had the Sentsortec current meter on, which is heavier than the Nortec.
1520: AUDOS on surface, on board at 15:45.
1600: Deck secure, headed for ISIT location to release it in time to surface before dusk.
1630: ISIT released.
1755: ISIT on surface, but weather had deteriorated with 35 knot winds. Managed to grapple mooring, but floats trailed to port with hauling line apparently trapped under rudder. Mooring released and ship turned for second attempt, however wind increased to gust at 45 knots and 2nd attempt abandoned. Ship held station with mooring in sight through dusk and into darkness. Wind eased slightly, but then increased to 50 knots at 22:30. Hoping it will have eased sufficiently by first light to recover it. Scientific watches operated through the night to monitor the ISIT on the surface.

Saturday 23rd September

0600: Hove to close to ISIT. Weather still poor. Wind moderated slightly but still blowing 30 knots.
1256: Wind eventually eased sufficiently to attempt recovery. The pellet was off, so the marker buoy had to be grappled.
1313: Grappled successfully and brought on board. The ropes at the surface end on the mooring had worn very badly. This was caused by rotation of the stainless steel eyes.
1320: ISIT finally on board.
1500: Deck secured, headed for AUDOS location.
1615: Deployed AUDOS (13915#1). One of the hydraulic hoses parted during deployment, so repair was required immediately afterwards. Remained hove to whilst crane repaired.
1848: Crane fixed. Crane repair caused trawl plans for the night to be altered. Selected new trawl location and headed for location to make sounding run.
2044: Commenced sounding run.
2143 Completed sounding run.
2215: Began shooting OTSB (13916#1, 2100 m).

Sunday 24th September

- 0035: OTSB on the bottom at 00:35.
0133: Hauling commenced.
0148: Net off the bottom (depth of 2023-2093 m).
0355: Net on deck. Small catch included over 100 fish, dominated by *Synphobranchus kaupi*, but also included *C. guentheri*, *C. mediterranea*, *H. macrochir*, a large *Hydrolagus affinis* and a small *Bathyraja*.
0415: All clear on deck, proceeded to FRESP location.
0616: Hove to at FRESP site.
0623: FRESP released. It rose very slowly, which caused some concern, but vehicle has large drag.
0905: FRESP on surface. Pellet buoy seen first, then very slowly the flag appeared.
0925: FRESP grappled and brought on board quickly (by 0935). No fish were trapped, and one of the doors was open, but otherwise it had operated as programmed. Video showed fish swimming around near the bait.
0940: Proceeded to AUDOS.
1005: AUDOS released.
1157: AUDOS on surface.
1216: AUDOS grappled. All on board by 12:45, and proceeded north to the next station, at 2500 m, in the Seabight.
1730: Arrived at AUDOS site and commenced deployment at 1745.
1755: AUDOS deployed (13917#1) and continued towards the FRESP station.
1927: On station for FRESP deployment.
2127: Commenced FRESP deployment (13918#1), flag disappeared at 21:30. Headed towards trawling location at 1900 m.

Monday 25th September

- 0141: Commenced sounding run at OTSB location.
0241: Completed sounding run, but when preparing to shoot the net was found to have a tear in the belly from the previous trawl station. Station cancelled.
0300: Proceeded to the Bathysnap (Deployed on Discovery 148, 8/8/00, 13880#1) location.
0635: Bathysnap released.
0655: Bathysnap on surface and on board at 07:05. Found a potential trawl location to the SW of the Bathysnap. The OTSB nets that were sent from Southampton to Cork have gone missing, but will proceed to Cork and hope that they turn up.
0940: Arrived at trawl location at 09:40 and headed NW to survey ground.
1050: Commenced shooting of OTSB (13919#1, depth 1550 m).
1242: Net on bottom.
1331: Commenced hauling.
1514: Net on board, with a large fish catch of *C. rupestris*, *S. kaupi*, some alepocephalids, *C. guentheri*, *C. mediterranea*, *T. murrayi*, and *Chimaera monstrosa*. Three octopus also caught (*S. syrtensis* and *G. verucosa*). Headed north-east in the direction of the ISIT site and Cork.
1652: Reached ISIT location.
1710: Deployed the ISIT at 1000 m at 17:10. Set off in the direction of Cork. Fish catch processing finally finished at 23:30.

Tuesday 26th September

- 0730: Arrived outside Cork Harbour and was met by the tug (Oyster Bank). The tug brought the hoses that were requested for the cranes, but the OTSBs had not arrived and the package of sampling bags sent from Cork were not brought out. Monty Priede disembarked to return to Aberdeen. Waited off Cork until 1100 in case the OTSBs could be located, but they could not be traced by TNT.
1100: Proceeded to Bullens Bay on the east side of the Old Head of Kinsale to launch the starboard life-boat.
1230: Off Bullens Bay, but the swell was too great to allow the lifeboat to be safely launched.
1300: PES fish deployed and Discovery headed back towards PSB. Weather deteriorated during passage to PSB.

1600: Brief scientific meeting to discuss the rest of cruise. Propose to collect the landers and head to PAP, to undertake work there, where the net is less likely to be damaged and return at the end of the trip to PSB.

Wednesday 27th September

0100: Arrived at Bathysnap location, weather poor.
0209: Bathysnap deployed (13921#1) in rapidly worsening conditions. Winds blowing up to 40 knots.
0245: Bathysnap on bottom.
0300: Proceeded to the ISIT location.
0500: Arrived ISIT location, wind 40 knots and no immediate prospect of recovering ISIT. Hove to all day.

Thursday 28th September

0600: Weather too poor to release landers or to trawl. Hove to all day in south-westerly and later westerly gales. Heavy rain squalls with 50 knot gusts. Pressure dropped to 975 as we hit centre of depression. In the evening we received notification that the trawls have arrived in Cork from RVS.

Friday 29th September

0600: Weather remains poor with winds still blowing at up to 40 knots, no prospect of work today. Wind now in the NW, but the forecast suggests that the weather will improve in 24 hrs. Dave Stewart (ships ETO) requested to go ashore as his father had passed away.
1300: Decided to head for Cork to drop Dave Stewart ashore and collect the nets at the same time.

Saturday 30th September

0750: Arrived outside Cork Harbour.
0800: The tug (Oyster Bank) arrived at 0800, dropped off the 3 OTSB nets and Dave Stewart disembarked.
0820: Departed Cork, heading back to PSB in relatively light winds. Swell remains from the north-west as we escape the lee of the Irish coast.
2255: Arrived at OTSB location and surveyed the ground in a SE direction.
2353: Survey complete.

Sunday 1st October

0011: Commenced shooting trawl (13922#1, 1933-1885 m). Towing NW into the wind.
0234: Net on bottom.
0322: Began hauling.
03:35: Net off bottom.
0536: Trawl on board.
0612: Clear on deck and headed off to the ISIT location.
0840: Approaching the ISIT location at 0800, weather was reasonable (F6) but on arrival wind rapidly increased to F9, making recovery impossible. Hove to as wind increased to F10.
1200: The wind and swell became too much for the ship to remain hove to, so vessel turned to run with the wind, heading back towards south coast of Ireland.
2200: SE of Fastnet, Discovery turned back into the wind. Wind still blowing F9-10, with extremely large seas. Remained hove to overnight, making slight westerly progress.

Monday 2nd October

0600: Hove to S of Fastnet, with the wind moderating rapidly.
1115: Turned to run downwind. Weather became calm, with light winds, but forecast for the next 24 hours poor, so captain advised against returning to PSB. Headed back east along the Irish coast, then SW away from the shore in view of predicted southerly gales.
1900: Hove to (51 17 N, 8 37 W) awaiting the forecast gales.

Tuesday 3rd October

- 0600: Hove to at 51 10 N, 8 51 W. Weather deteriorated rapidly in the early morning, to reach F10 at lunchtime from the south. Remained head to wind for the morning and early afternoon.
- 1500: Turned to run with the wind. Large waves taken over the after deck, resulting in damage and loss of one of the wooden crates used to store the trawls. Wind moderated in the evening. Past Kinsale Gas Field at 1900.
- 2130: Turned head to wind and hove-to.

Wednesday 4th October

- 0600: Weather moderated during the early hours of the morning, allowing slow progress back towards PSB, with the prospect of better weather. Large swell prevented full speed.
- 1615: Safety drill and quiz in the bar.

Thursday 5th October

- 0040: Passed over ISIT and confirmed acoustically that it was still on the sea-floor.
- 0100: Arrived at proposed OTSB station, but the wind had increased to 30 knots and the trawl was abandoned. Returned to ISIT location and hove-to.
- 0622: Released the ISIT, in a heavy swell.
- 0643: ISIT on the surface and grappled first time, however the line was lost after the pellet was removed and we turned and grappled it properly on the second attempt.
- 0730: ISIT on board.
- 0750: All clear and heading for FRESP. Port block fell from the A-frame shortly after recovery of ISIT, but fortunately no one near at the time. Heading SW to FRESP location, but forced to do a dog-leg course because of the swell.
- 1421: Released the FRESP from a range of 3 miles whilst approaching the location.
- 1559: FRESP on the surface and grappled quickly, but the pellet float came down the port side, delaying recovery.
- 1630: FRESP on board, but no fish trapped and two of the perspex doors cracked. Video and syringes worked fine. Secured the FRESP on the after deck and headed for the AUDOS location, although a dog-leg course was again required.
- 1745: Released the AUDOS before arrival at the location.
- 1852: AUDOS on the surface, just after dark. AUDOS recovered in calm conditions, although the ship had to manoeuvre on the bow thruster to get in close. Heading for PAP.

Friday 6th October

- 0930: Arrived at AUDOS deployment location at PAP at 09:30.
- 1019: Deployed the AUDOS at 10:19 at depth of 4814 m (13923#1). Headed for the sediment trap (55102#2).
- 1052: Sediment trap released.
- 1105: Sediment trap on surface, with the first set of buoyancy on board at 11:35.
- 1352: Recovery complete. Once deck was clear the OTSB was prepared.
- 1430: Commenced shooting OTSB
- 2000: Net on bottom.
- 2155: Commenced hauling.
- 2306: Net off bottom.

Saturday 7th October

- 0245: OTSB brought on board, but had not reached the sea-floor, so the only catch was a few midwater fish and invertebrates and a small *Cirrothauma murrayi*. Headed immediately for the AUDOS location.
- 0620: Arrived at the AUDOS site.
- 0631: Released AUDOS.
- 0844: AUDOS on the surface and quickly recovered. Headed south from the AUDOS station and prepared to shoot the trawl.
- 1052: Began shooting OTSB.

1545: Net on bottom.

1745: Commenced hauling.

1920: Net clear of bottom.

2233: OTSB on board, with good catch of holothurians, but with a lot of mud and few fish. Scientific programme completed and now heading for Southampton.

Sunday 8th October: On route for Southampton.

1000: Cruise debrief meeting.

Monday 9th October

On route for Southampton.

Tuesday 10th October

On route for Southampton.

1030: Arrived at SOC.

1200: Unloading commenced.

1600: All scientific gear offloaded.

Scientific Reports

1 FISH CATCHES FROM THE OTSB TRAWLS

David Bailey, Andy Stocks & Martin Collins, University of Aberdeen

Background

An important objective of the NERC grant GR3/12789 (Metabolism, activity and distribution patterns of deep-sea demersal fishes) is to investigate seasonal patterns in the distribution, abundance and metabolism of deep-sea fishes. The OTSB trawls provide one of the main methods of determining distribution and abundance of the deep-sea fish and provide material for studies of enzyme activities, condition indices, diet, growth and reproduction.

Methods

Seven OTSB trawls were completed at depths from 1000 to 4800m. All fish were identified, weighed, measured and individually labelled. Examples of each species were frozen and/or preserved in formalin. All other fish were available for the removal of tissue samples for analysis of enzyme activity, population genetics and brain and eye structure. These experiments will be discussed elsewhere.

Following the removal of tissue samples the animals were sexed and their reproductive maturity assessed. The gonads of mature animals were removed and preserved in formalin. Stomach fullness was determined and full stomachs frozen for later analysis. Otoliths were obtained from over 40% of fish caught. Complete livers were removed and weighed. Gills were also removed from a range of species.

Results

Fifty five species of fish were caught (Appendix III). The most abundant species were the eel *Synaphobranchus kaupi* and the grenadier *Coryphaenoides armatus*. The data will be utilised to determine seasonal patterns in distribution and abundance. New data was obtained on length-weight relationships in many of the species. Tissue samples were obtained from 33 species for future work on molecular phylogeny.

Summary

Despite the poor weather a large amount of basic data about the fish of PSB was collected and will be available for comparison with data from later cruises. This data and the samples collected will enable detailed analysis of seasonal cycles in growth and reproduction.

2 CEPHALOPODS FROM OTSB CATCHES

Martin Collins and Oliver Yates

Background

Cephalopods are common, but not abundant, members of the deep-sea benthic (incirrate octopods) and benthopelagic (cirrate octopods) fauna, however the taxonomy of this group is in disarray and virtually nothing is known about the ecology. A current NERC grant (GR8/04443: Taxonomic revision of the deep-sea octopods of the NE Atlantic) is investigating the taxonomy of the group and all cephalopod specimens caught were preserved to further this study.

Work undertaken

Twelve cephalopods, belonging to six species were obtained from the OTSB catches (Table 2.1). Specimens were measured and weighed while fresh and a tissue sample taken for molecular phylogenetic studies. Specimens were subsequently fixed in formalin and subsequently transferred to IMS (incirrates & squid) or Steadmans solution (cirrates).

Table 2.1 Cephalopods caught during Discovery 250.

Species	Depth caught	Notes
<i>Teuthowenia megalops</i>	1500 m	Damaged specimen
<i>Stauroteuthis syrtensis</i>	1500 m	1 male, 1 female in good condition
<i>Grimpoteuthis</i> sp 1	4000 m	2 specimens
<i>Grimpoteuthis</i> sp 2	4800 m	4 specimens in good condition
<i>Cirrothauma murrayi</i>	4800 m	Small specimen.
<i>Graneledone verrucosa</i>	1500 m	Large mature female

3. OTTER TRAWL - INVERTEBRATES

Dave Billet, SOC.

The Semi Balloon Otter Trawl (OTSB 14) with a headrope length of 14m and an effective fishing width of 8.6m was fished 8 times during the cruise. One trawl appeared to catch some wreckage on the seabed and the cod end was lost (Station 13907#1). The belly of the net was torn on another trawl (Station 13916#1). A third trawl (Station 13924#1) failed to reach the seabed despite having 11500m of wire out in 4850m water depth. Details of the dominant invertebrates for each net are presented below. Samples from the trawls were used for a wide variety of studies, including the molecular taxonomy of holothurians and asteroids, population genetics of holothurians, asteroids and decapod crustaceans, and the continuation of time-series sampling on the Porcupine Abyssal Plain for work on long-term change in abyssal ecosystems.

Station 13906#1. This trawl, at the base of the continental slope in the mouth of the Porcupine Seabight produced a good catch of invertebrates. Of particular note were the holothurian *Psychropotes longicauda*, the crustacean *Munidopsis* sp., a pagurid/zoanthid association, many scaphopods and the asteroids *Zoroaster longicauda*, *Freyella elegans*, *Hyphalaster inermis* and *Styracaster* spp. Loads of clinker (36 kg) had macerated many of the soft-bodied animals. The catch also contained sponges, small zoanthids, solitary corals, actiniarians, a small *Umbellula* pennatulid, large natant decapod crustaceans, gastropods, a small cephalopod, large echinothuriid echinoids, and several other holothurians, notably some small *Benthothuria funebris*, *Benthoodytes sordida*, *Oneirophanta mutabilis*, *Deima validum*, *Peniagone diaphana*, and, more abundantly than ever before, a small orange-red holothurian still to be identified.

Station 13907#1. Although the cod end was lost on this net there were a number of animals caught on the mesh including some sponges, tunicates (*Culeolus*), asteroids and holothurians (*Amperima rosea* and *Pseudostichopus villosus*).

Station 13910#1. This trawl at mid-slope depths between the Gollum Channel System and the Goban Spur produced another good and varied catch. Notable invertebrates included a giant spider crab *Neolithodes*, countless ophiuroids (*Ophiomusium lymani*), a good number of *Glyphocrangon ?sculpta* and several *Munidopsis* sp. In addition there were a few *Bathybiaster vexillifer* and a solitary *Psychropotes depressa*.

Station 13914#1. This trawl close to the steep slopes in the mouth of the Porcupine Seabight produced a giant catch of the holothurian *Benthothuria funebris*. As the cod end came in it looked as though there was a large fish catch. While there were some 140 fish, the bulk of the catch was made up the rugby-ball shaped holothurian which soon started to slide all over the deck. Specimens were frozen and preserved directly in IMS. Previous experience of preservation in formalin had shown that the holothurian became very fragile. The catch was also notable for several specimens of *Peniagone azorica*, a species common in the Rockall Trough at this depth, but which had only been recorded a couple of times in the Porcupine Seabight. There were also a few specimens of the benthopelagic

holothurian *Peniagone diaphana*. Several *Neolithodes* spider crabs were present, as well as the asteroid *Hymenaster pellucidus (membranaceus)*, echinothuriid echinoids and some pagurid crabs.

Station 13916#1. This trawl, on the southern slopes of the Goban Spur just above steep slopes extending down to the abyssal plain, produced an interesting, if small, mixed catch of invertebrates and an excellent fish catch, even though the belly of the net had been torn across the width of the net. There were some notable absentees, such as the holothurian *Paelopatides grisea*, that might be expected at this depth in the centre of the Seabight. Centre stage for the invertebrates was an extremely large sponge. There were numerous crustacean decapods belonging to the genera *Glyphocrangon* and *Polycheles*. There were a few small *Geryon tridens*, presumably at the bottom of its ontogenetic upslope migration. Fragments of a large stalked crinoid were recovered as well as several pycnogonids. Surprisingly for this depth, holothurians were rare, with just a few *Benthogone rosea* and a handful of a *Pseudostichopus*, species that appeared to be different from the two species encountered on the abyssal plain. These are the shallowest records of *Pseudostichopus* made in the PSB area.

Station 13919#1. A good fish and invertebrate catch was obtained at mid-slope depths in the northeast of the Porcupine Seabight. The invertebrates were dominated by the slimy holothurian *Benthogone rosea*. The catch also contained several other holothurians, notably *Mesothuria lactea*, *Mesothuria* sp. and *Paroriza pallens*. The catch also contained a large stalked sponge, a few gravid specimens of *Neolithodes*, several *Polycheles*, and the asteroids *Persephonaster patagiatus*, *Plinthaster dentatus* and *Pectinaster filholi*. There was also an interesting long-armed ophiuroid associated with a gorgonian.

Station 13922#1. A good catch of invertebrates, if fewer fish this time, was obtained from the centre of the Porcupine Seabight. Holothurians dominated the catch with a high numbers of the large synallactid *Paelopatides grisea* (418) and the elasipodid *Benthogone rosea* (334). Other notable fauna included a number of actinarians, the decapod crustaceans *Polycheles* and *Glyphocrangon*, the ophiuroid *Ophiomusium lymani*, and the echinoids *Echinus affinis* and *Phormosoma placenta*.

Station 13924#1. This was the first trawl on the Porcupine Abyssal Plain. Despite having 11500m of wire out the net failed to ground and the cod end had only a few benthopelagic animals, notably the benthopelagic holothurian *Peniagone diaphana* and the octopod *Cirrothauma murrayi*.

Station 13925#1. This trawl produced a good, if muddy, catch with all the usual fauna from the Porcupine Abyssal Plain. Holothurians dominated, mainly *Psychropotes longicauda*, *Oneirophanta mutabilis* and *Pseudostichopus villosus*. *Amperima rosea* was also present, but not in the large numbers encountered in recent years. Other holothurians included *Peniagone diaphana*, *Molpadia blakei*, *Deima validum*, *Ellipinion* sp., *Paroriza prouhoi*, *Mesothuria candelabri*, *Benthodytes* sp., *Protankyra brychia* and a second species of *Pseudostichopus*. There were also 4 small *Grimptoteuthis* spp. octopods, a large and a small *Umbellula* pennatulids, several actinarians (many attached to pieces of clinker) and the asteroids *Hyphalaster inermis*, *Styracaster* sp. and *Freyastera* sp.

4 FUNCTIONAL MORPHOLOGY OF DEEP-SEA FISH GILLS

Judith Brewster, Aberdeen University

Deep-sea fish are generally considered to be inactive and slow swimmers. This would therefore suggest low oxygen requirements and hence reduced gill area. The aim of this study is to investigate bathymetric trends in gill surface area and structure. During the course of the cruise, gills were collected from a variety of different species at different depths and the table below outlines the species and number of fish that gills were collected from.

Species	Gills collected
<i>Bathysaurus ferox</i>	2
<i>Bathytroctes microlepis</i>	2
<i>Bellocia foefedi</i>	2
<i>Concara murrayi</i>	1
<i>Coryphaenoides (Nematonurus) armatus</i>	4
<i>Coryphaenoides (Lionurus) carapinus</i>	2
<i>Coryphaenoides guentheri</i>	1
<i>Coryphaenoides leptolepis</i>	1
<i>Coryphaenoides (Profundicola) mediterraneus</i>	3
<i>Coryphaenoides rupestris</i>	13
<i>Halosauropsis macrochir</i>	1
<i>Histiobranchus bathybius</i>	2
<i>Poromitra capito</i>	1
<i>Trachyrincus murrayi</i>	2
<i>Ceolorynchus occa</i>	1

Table 4.1 Species and number of fish from which gills were collected.

Back in Aberdeen, the surface area of the gill filament will be estimated and the number of secondary lamellae on each filament counted. It will then be possible to look at the relationships between fish of the same species but of differing lengths and weights. It will also be possible to compare fish of same species but at different depths as well as comparing fish of similar sizes but of different species. The size and shape of the gill rakers will also be investigated and comparisons will be made between different species.

5 HOLOTHURIAN GUT CONTENT ANALYSIS AND POPULATION GENETICS

Ben Wigham, SOC.

Samples were taken from the one trawl at the Porcupine Abyssal Plain site; station 13925#1.

(a) Gut contents.

The analysis of gut sediments from holothurians, using HPLC and flurometry techniques, can identify, qualitatively and quantitatively, chlorophyll a and its breakdown pigments. Six species of holothurian were selected for this analysis, all with varying tentacle structure and proposed modes of feeding. The six species selected were:

1. *Amperima rosea*
2. *Oneirophanta mutabilis*
3. *Psychropotes longicauda*
4. *Pseudostichopus villosus*
5. *Pseudostichopus* SP.
6. *Molpadia blakei*

10 specimens of each animal were selected for dissection, with the exception of *Amperima rosea* where 18 samples were taken. The guts of the three larger species (2,3,4) were divided into anterior and posterior halves and the sediment was removed from the gut and frozen (-70°C). In the case of the remaining animals, total gut sediment was removed and frozen as one sample.

This work will accompany that already in progress on the observed *Amperima* 'bloom' of 1996-98. It is believed that changes in the deposition of phytodetritus to the abyssal plain may have provided a trigger for this observed population explosion. Therefore we are planning to investigate feeding selectivity in abyssal holothurians, as has previously been observed in their bathyal counterparts (Billett et al., 1988).

(b) Population Genetics.

All specimens of *Amperima rosea* were removed to the CT lab (4°C) as soon as possible after sorting. Hopefully this action will prevent the degradation of enzymes and the digestion of DNA brought about by the increase in temperature and exposure of the specimens to UV light.

50 Specimens were dissected and sections of tissue were placed in 2ml vials and immersed in 95% ethanol and refrigerated. The material taken from the *Amperima* specimens will complement that taken in April 1999 during Challenger cruise 142.

The application of various molecular techniques will enable us to study the spatial genetic structure of the *Amperima* population(s) on the Porcupine Abyssal Plain. By comparing the results against the Hardy-Weinberg equilibrium we will be able to establish whether *Amperima* is sexually reproducing and outbreeding.

6 ECOLOGY AND MOLECULAR TAXONOMY OF ECHINODERMS IN THE NORTH ATLANTIC

Francisco Alonso Solis-Marin, SOC.

The central aim of my research programme involved in cruise Discovery 250 is to clarify the taxonomy of selected groups of deep-sea echinoderms (especially Holothurians) from the North Atlantic using molecular techniques and clarify the ecology and life history of these and any cryptic species identified.

There are a number of species that remain taxonomically problematical, and until there is clarification of the taxonomy the ecology and life history work cannot be completed. A number of taxa also show overlapping depth distribution of species comprising the genus and this has led to speculation about the evolution of these taxa and their ability to invade the deep sea.

During Discovery 250 tissue samples (mainly muscles) were taken for DNA analysis from 19 holothurian species.

List of holothurian species sampled during cruise Discovery 250

Mesothuria cathedralis

Mesothuria lactea

Mesothuria sp.

Paroriza pallens

Paroriza prouhoi

Benthogone rosea

Pseudostichopus villosus

Pseudostichopus sp.

Bentbodytes sanguinolenta

Benthothuria sp.

Benthothuria funebris

Peniagone azorica

Peniagone diaphana

Psychropotes longicauda

Scotoplanes depressa

Protankyra brychia

Oneirophanta mutabilis mutabilis

Deima validum

Molpadia blakei

7 STARFISH TAXONOMY AND DIETARY ANALYSIS

Kerry Howell, SOC.

Background

Starfish (Echinodermata – Asteroidea) are abundant members of the deep-sea benthic megafauna. The work undertaken on this cruise was concerned with utilising molecular methods to solve problems in the taxonomy of this group. Specimens were also sampled for fatty acid or carbon nitrogen isotopic analysis of diets.

Work undertaken

From 6 trawls 9 species were dissected and/or frozen and tissue samples taken from five species. Those that have been dissected and frozen had their guts removed. These samples will be later used in an analysis of diet using either fatty acid analysis or carbon nitrogen isotopic analysis. The tissue samples were taken from species whose taxonomy is in question. They will be used for 16S genetic testing in order to determine whether they are legitimate separate species or not. Gonad and a small section of arm have been preserved in 100% ethanol for this analysis. Those species which have been frozen whole are being kept as back up samples for all types of analysis to be performed.

Samples taken

Trawl number	Species	Number	Treatment	Purpose
13906#1	<i>Zoroaster longicauda</i>	20	tissue sample	genetics
13906#1	<i>Zoroaster longicauda</i>	20	frozen	back up
13906#1	<i>Dytaster grandis grandis</i>	9	guts removed and frozen	diet
13910#1	<i>Hymenaster membranaceus</i>	10	tissue sample	genetics
13910#1	<i>Hymenaster membranaceus</i>	10	frozen	back up
13914#1	<i>Hymenaster</i> sp.	3	tissue sample	genetics
13914#1	<i>Hymenaster</i> sp.	3	frozen	back up
13919#1	<i>Zoroaster fulgens</i>	7	tissue sample	genetics
13919#1	<i>Zoroaster fulgens</i>	7	frozen	back up
13919#1	<i>Plutonaster bifrons</i>	8	guts removed and frozen	diet
13922#1	<i>Plutonaster bifrons</i>	12	guts removed and frozen	diet
13922#1	<i>Plutonaster bifrons</i>	9	tissue sample	genetics
13922#1	<i>Plutonaster bifrons</i>	9	frozen	back up
13922#1	<i>Benthopecten simplex</i>	23	frozen	possible diet
13925#1	<i>Hyphalaster inermis</i>	20	guts removed and frozen	diet
13925#1	<i>Styracaster chuni</i>	16	guts removed and frozen	diet
13925#1	<i>Dytaster grandis grandis</i>	1	guts removed and frozen	diet

8 POPULATION GENETICS OF DEEP-SEA FISH

Catherine Pearson (on behalf of Alex Rogers), SOC.

Background:

Many deep-sea fish have extensive bathymetric and geographic ranges, however it is not known to what extent populations are mixed both spatially and seasonally. Population genetics may be able to resolve geographic, bathymetric and seasonal patterns of distribution and determine the degree of isolation of populations.

Objectives:

To collect DNA samples from all specimens of deep-sea fish, particularly grenadiers captured during the cruise. 1cm³ tissue samples were taken and stored in ethanol in a 2ml eppendorf tube. These samples will be used for DNA analysis of the genetic population structure of these deep sea fishes.

Results

From seven OTSB hauls a total of 28 species of fish were sampled:

<i>Coryphaenoides armatus</i>	<i>Alepocephalus rostratus</i>
<i>C.guntheri</i>	<i>Hydrolagus mirabilis</i>
<i>C.rupestris</i>	<i>Alepocephalus productus</i>
<i>C.leptolepis</i>	<i>Borostomias antarcticus</i>
<i>C.mediterraneus</i>	<i>Trachyrinus murrayi</i>
<i>C.profundicola</i>	<i>Bathyroctes microlepis</i>
<i>Histiobranchus bathybius</i>	<i>Bathyraja richardsoni</i>
<i>Halosauropsis macrochir</i>	<i>Coelorhynchus occa</i>
<i>Lionurus carapinus</i>	<i>Cataetyx laticeps</i>
<i>C.brevibarbis</i>	<i>Bathysaurus mollis</i>
<i>Spectrunculus grandis</i>	<i>Antimora rostrata</i>
<i>Belloccia koefedi</i>	<i>Synaphobranchus kaupi</i>
<i>Conacara murrayi</i>	<i>Bathysaurus ferox</i>

In addition, 83 samples of *Glyphocrangon* sp. and 37 samples of *Munidopsis* sp. were collected.

Tissue was stored as for the fish. It is hoped that some genetic work may also be conducted on these specimens.

9 ENZYME ACTIVITIES AND ANTIOXIDATIVE DEFENCE MECHANISMS IN DEEP-SEA FISHES

Jean-François Rees and Alexis de Kerchove, University of Louvain

The objectives of our participation to the cruise were:

1. The study of enzymatic activities related to the energy supply in tissues of deep-sea fishes and their depth- and season-dependant variations.
2. The adaptations of antioxidative defence mechanisms in the blood and tissues of these fishes.

1 Enzymatic activities related to the energy supply in tissues of deep-sea fishes and their depth- and season-dependant variations.

Deep-sea fishes live in a food-limited environment. Several studies have shown that metabolic enzymes activities in muscles of active pelagic swimmers decrease with increasing depth. In order to determine whether this also occurs in deep-sea demersal fishes, the levels of metabolic enzymes will be measured in muscle and other fish tissues. Tissues (blood, brain, eyes, heart, liver, white and red muscles) were collected from a total 350 individuals belonging to 25 species (see table). The tissues will be homogenised, centrifuged and activities of lactate dehydrogenase, citrate synthase, pyruvate kinase will be assayed in the supernatant at 4 °C. The protein content of each tissue will be determined. The scaling of these activities with the size of the fish will be studied within each species.

Table 9.1 Deep-sea fishes sampled for enzyme activity studies.

species	13906 4000m	13910 2500m	13914 3050m	13916 2050m	13919 1540m	13922 1900m	13922 4845m	Total
ALB					3			3
ALP				1				1
ALR					3	1		4
ANR		7		3		4		14
BEK							1	1
BRR				1				1
BTM							1	1
BSF		3		1		1		5
BSM			1					1
CAL						1		1
CNR						1		1
COA	58	6	41				6	111
COB		8						8
COC		4						4
COG		7		8	8	15		38
COM				1		5	4	10
COR					26	5		31
HAM		19	14	3		2		38
HIB		7	3	19			1	30
NOB		1			3			4
POC		1						1
POR			2			5		7
SPG		4				2		6
SYK		1				19		20
TRM					9			9
							Sum:	350

2. The adaptations of antioxidative defence mechanisms in the blood and tissues of these fishes.

All extant organisms are endowed with an arsenal of mechanisms aimed at protecting tissues against the toxicity of oxygen reactive species (ROS), such as superoxide anion and peroxide. If these defence mechanisms cannot cope with the ROS continuously generated by oxidative metabolic processes, cell constituents (lipids, nucleic acids, proteins) will be damaged and cells undergo necrosis or apoptosis. Our previous work in pelagic fishes indicates that levels of antioxidative defence paralleled that of the oxidative metabolic activities. Whereas activities of defence mechanisms in demersal species could similarly be adjusted to the threat posed by the metabolically produced ROS, the high levels of organochlorine pollutants and mono-oxygenase activities previously found in some species could favour high levels of antioxidative protections.

The levels of antioxidative enzymes (superoxide dismutase, catalase and glutathione peroxidase) and free radical scavengers (glutathione, coelenterazine, coelenteramine, vitamin E) will be determined in the tissue homogenates. The total antioxidant potential of the plasma will be measured. The depth-dependence of these antioxidative indexes will be investigated. The levels of cytochrome P-450 mono-oxygenases and organochlorine contaminants will be analysed.

10 MOLECULAR ADAPTATIONS TO HIGH PRESSURE AND LOW TEMPERATURE IN DEEP-SEA FISH

Amanda Brindley, Queen Mary College, London

This is a joint project with Professor Martin J Warren, School of Biological Sciences, Queen Mary, University of London, Professor David M Hunt, Department of Molecular Genetics, Institute of Ophthalmology, University College London and Dr Julian C Partridge, School of Biological Sciences, University of Bristol. This project is funded by the BBSRC for three years from September 2000.

Background

The physical properties of the deep-sea create an environment which is characterised by high pressures and low temperatures. Very little is known about the molecular changes that allow the proteins of deep-sea fish to function in such an environment, although it is clear that molecular adaptations have taken place, leading to alterations in the thermal and kinetic properties of proteins.

Pressure induced denaturation of single chain proteins occurs at lower pressures than those observed in the deepest areas of the ocean. Furthermore for oligomeric proteins dissociation occurs at much lower pressures. Previous studies have shown that proteins of deep-sea fish have increased resistance to thermal denaturation relative to shallow-water homologs. Thermal stability is thought to be due to the evolution of especially rigid proteins that are able to resist disruption of tertiary and quaternary structure under high pressure. The effect of low temperature is thought to result in cold adapted proteins that have weaker intramolecular interactions that produce more flexible molecular edifices capable of performing catalysis at a lower energy cost. It would appear that there is a dichotomy between adaptations for high pressure and those for low temperature.

The objective of this project is to contribute to the understanding of the structural adaptations of certain deep-sea fish enzymes that permit them to operate at both high pressures and low temperatures. Three enzymes have been selected for this work; lactate dehydrogenase (LDH), 5-aminolaevulinic acid dehydrogenase (ALAD) and porphobilinogen deaminase (PBGD).

Work on board ship

Whole liver and large body muscle samples have been collected from the following species; *Coryphaenoides armatus*, *Coryphaenoides guentheri*, *Coryphaenoides rupestris*, *Coryphaenoides leptolepis*, *Coryphaenoides carapinus*, *Antimora rostrata*, which represent the Order Gadiformes over a depth range of 1250m to 4800m, and the species *Histiobranchus bathybius* and *Synaphobranchus kaupi* which represent the Order Anguiliformes over a depth range of 1250m to 4800m. These have been stored at -75°C in order to carry out enzyme assays at a later date. Small liver and muscle samples were also taken from these species and stored in RNA later™ at -20°C to isolate mRNA for cDNA synthesis for recombinant protein production on return to London. Small liver and muscle samples were also isolated from other species, *Alepocephalus bairdii*, *Alepocephalus rostratus*, *Halosauropsis*

macrochir, *Spectrunculus grandis* and stored in RNAlater at -20°C for sequence analysis on return to London.

Liver and muscle samples from *Coryphaenoides armatus* and *Histiobranchus bathybius* were homogenised and assayed for the presence of LDH, ALAD and PBGD activity using spectrophotometric assays. All three enzyme activities were detected in both muscle and liver from both species.

Future Work

***In vivo* assay**

Each liver and muscle sample will be assayed for each individual enzyme activity in order to determine standard kinetic constants, the relationship of activity with temperature and with pH. As far as possible these will be carried out at atmospheric and at high pressure. There will also be comparisons with shallow water equivalents from fish in the same Orders.

Amino acid sequence determination and comparison

mRNA will be isolated from liver and muscle and cDNA generated for libraries. These will be screened with orthologous sequences from other species for the three enzymes, all positive clones will be sequenced. The deduced amino acid sequences from deep sea fish will be compared with those from shallow water fish and other vertebrates and any candidate substitutions noted. Any patterns in the substitutions will be pursued to determine whether a particular residue is important or a particular region of the protein is substantially altered. Sequences will be studied within Families and Orders comparing deep-sea species and shallow-water species.

***In vitro* analysis of recombinant proteins**

Complete cDNA sequences will be used for the *in vitro* synthesis of each of the enzymes from several species. The enzymes will be over-expressed as recombinant proteins in *E.coli* and purified using standard techniques. Comparisons of the recombinant and tissue homogenate enzymes will be made with respect to K_m , temperature rate profiles, pH dependency and thermal stability, in order to determine if the recombinant proteins fold properly. Purified protein will also allow accurate rate constants to be determined at both atmospheric and high pressure. The large quantities of pure protein available will provide sufficient material to pursue crystallisation trials, ultimately leading to the determination of the three-dimensional crystal structure of the enzymes. Comparisons of the structures of the pressure / cold adapted enzymes with their normal counterparts will enable any functional differences in enzyme properties to be identified and evaluated in structural terms.

11 SPECTRAL SENSITIVITY OF DEEP SEA FISH VISUAL PIGMENTS AND THE EFFECT OF PRESSURE

Julian Partridge, Bristol University.

Background. Visual pigments are the photosensitive pigments found in the retinas of both vertebrates and invertebrates. Visual pigments consist of a protein moiety, opsin, and a chromophore, which is an aldehyde of vitamin A. Opsins are highly conserved proteins and, in common with all members of the class of G-protein linked cell membrane receptors, have seven trans-membrane helices. In visual pigments these helices form a binding pocket in which the chromophore is held.

The visual pigments of some 180 species of deep sea fish have been measured and approximately 90% of these have in their retinas only rod photoreceptors, containing a shortwave sensitive visual pigment. Typically, the wavelength of maximum absorbance (λ_{\max}) of these pigments is between 470 nm and 490 nm. Such pigments confer high sensitivity to both downwelling daylight (relevant only to mesopelagic depths in most oceans), and bioluminescence. The exact λ_{\max} of any visual pigment is determined by the electrostatic interactions between the chromophore and key amino acids in the opsin. Recently, the genes encoding the opsins of some 60 species of deep-sea fishes have been sequenced. The way in which these opsin amino acid sequences affect spectral tuning in these visual pigments is thus very well characterised.

Nevertheless, all spectrographic work on these pigments has hitherto been undertaken at normal atmospheric pressure. Given the facts that: (i) pressure is known to perturb the function and/or tertiary structure of many proteins, and (ii) that the λ_{\max} of a visual pigment is dependent on electrostatic interactions between the opsin and the chromophore, which will be altered by changes in interaction distance, it is reasonable to expect pressure to affect λ_{\max} values. In other words, are the previously measured λ_{\max} values of deep sea fish the same as those that occur *in vivo*, at depth and under pressure?

Work on board ship

1. **Collection of retinae.** Fishes, captured at depths ranging from 1250 m to 4000 m, were removed from the trawl as soon as possible, placed in a light-tight bag, and transferred to a dark room where eyes were removed under dim red light. The eyes of 54 individuals of 12 species were collected in this way, and frozen at -75°C .
2. **Fixation of eyes.** The eyes of 15 individuals of 8 species were removed and fixed in 4% formalin for histological purposes.
3. **Lens measurements.** The diameter of the ocular lenses of 40 individuals of 8 species were measured using an electronic calliper gauge to determining the effective pupil diameters of their eyes.

Future work

1. **Visual pigments.** The absorbance spectra of the collected visual pigments will be measured as detergent extracts or suspensions of purified rod outer segment membranes. Measurements will be made at both 1 bar and in a purpose-built pressure chamber which can be placed within the sample

chamber of a Shimadzu spectrophotometer. This chamber is capable of producing pressures up to 460 bar (equivalent to approximately 4600 m depth of sea water). Such measurements will determine if pressure affects the λ_{\max} values of deep sea fish visual pigments. These data will be compared with equivalent measurements obtained from shallow water species. In order to reduce the potentially confounding influence of phylogeny, the chosen deep and shallow water species will be closely related. If pressure effects are found, these will be related to the amino acid sequences of the opsins.

2. **Fixed tissue.** Eyes fixed in formalin will be cryosectioned and measurements made of the dimensions of rod outer segments, and the number of retinal banks.

These data, together with pupil measurements, will be used to test the predictions of a computer model that I have previously written which suggests that the eyes of deep-sea fishes, including their visual pigments, pupil diameters and rod outer segments dimensions, are optimised in the course of evolution for the detection of bioluminescent sources at maximum visualisation range. The understanding of eye design, and hence the visual capabilities of deep-sea fishes, is fundamental to our understanding of both intra- an inter-specific communication and interactions in these animals.

Acknowledgements

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I would like to thank Professor I.G. Monty Priede and Dr Martin Collins for the invitation to take part in *Discovery* cruise 250, and Dr Martin Collins for being an excellent PSO in the trying circumstances of exceptionally bad weather. I also thank the officers and crew of the *RRS Discovery* for making this cruise both safe and effective, despite the difficult conditions.

12 SEASONAL CHANGES IN THE SENSORY MODES OF THE BRAINS OF DEEP-SEA FISH

H.-J. Wagner & U. Mattheus, University of Tuebingen, Germany.

The first aim of our experiments is to investigate the sensory biology of deep demersal fish and the impact seasonal changes may have on the relative importance of the various sensory modes. In mesopelagic species, especially the olfactory system has been shown to vary in accordance with a sexual cycle, which in turn would be linked to seasonal variations. The second main objective concerns the role of the pineal gland in the control of seasonality of demersal fish. In all vertebrates this part of the forebrain plays a decisive part in synchronising reproductive activity to the yearly seasons by secreting the effector hormone melatonin.

1. Sensory brain areas in demersal fish

1.1. Relative size of sensory lobes. The primary concern during the present cruise was to establish baseline data for the relative volumes of the four major sensory lobes, namely the olfactory bulb, the optic tectum, the facial and the vagal lobes. For this purpose, we aimed at collecting brains of as many different species as possible. This would allow us to determine the average relative volume of each structure and the deviation from these values in the individual species. We were able to collect the brains of 28 species during the present cruise, which brings the total number of demersal species available for analysis (including those from DISCO 243) to 35 and corresponds roughly to one half of the demersal species. After identification and basic measurements of the catch by the Aberdeen group, we removed either entire heads or brains for fixation in buffered formaldehyde. The heads were further dissected to expose the size and topography of the cranial nerves as well as the morphology of the brain in situ. The preparations as well as the isolated brains will be photographed in the lateral and dorsal aspect and processed for digital morphometry.

1.2. Seasonal changes in individual sensory systems. These data will form the basis for comparison with the same species caught in future cruises during spring time in order to find out about potential seasonal changes in individual sensory systems. Further variables to be considered for analysis are the size and state of sexual maturity of a given species, and possibly related to this, the depth distribution. In this respect, it may turn out to be necessary to focus on few representative species, like *C. (N.) armatus*, *H. bathybius*, and *S. kaupi* in order to establish a detailed profile of brain development and to interpret seasonal changes on this more comprehensive background.

1.3. Comparison with mesopelagics. The findings on the sensory brain areas in deep demersal fish will also be compared with similar data on the brains of mesopelagic fish. Preliminary observations of the fish collected during last year's cruises suggest that vision is by far the dominant sense in mesopelagic fish, whereas, in the demersal fish, there is a markedly higher degree of variability, with a major role of the olfactory system, and the vagal lobes (chemical senses).

2. Pineals in demersal fish

2.1. Identification and localisation. The first and elementary task was the identification and localisation of pineal glands in demersal fish. This turned out to be of no major difficulty in all of the eel-like species where there was not much space between the brain and the skull, and here consequently the pineal stalk was short. In addition, in these species the pineal was comparatively large. By contrast, in the grenadier fish, in general, there is a huge room filled with gelatinous

material between the skull and the brain. Since the pineal proper is localised in the arachnoidea attached to the skull, the thin and ephemeral pineal stalk can have a length of up to several cm; consequently the pineal is often strongly displaced with respect to the brain proper.

2.2. Morphological analysis of pineals. We have identified pineal glands in all of the species collected and in each one, fixed several specimens for histological and ultrastructural analysis. Previous preliminary observations have shown in (presumed) pineals of *C. (N.) armatus* the presence of parenchymatous tissue with a small lumen, conspicuous blood vessels, and several myelinated nerve fibres. At the same time, photoreceptor-like outer segments which are common in mesopelagic pineals were conspicuously absent in the two specimens analysed to date. We have to confirm these observations in additional specimens and conduct a comparative morphological analysis of all the species collected. Of major interest is the presence of outer segments and the amount of sympathetic afferents.

2.3. Melatonin seasonality. Melatonin is the neurochemical messenger to mediate the cyclical effects of light and dark (in surface or terrestrial) animals; furthermore, its role is to synchronise reproduction and seasonality. It is secreted by photoreceptor cells in the retina and the pineal gland, resp. pinealocytes which have lost their photoreceptive capacity (mammals, demersal fish?). We have collected several dozens of eel pineals and 20 pineals of *C. (N.) armatus* and intend to assay them for melatonin content. Possible variations dependent on the light and dark phase will be of major interest. In view of the importance of melatonin in controlling the effects of seasonal cycles on sexual activity, it is necessary to perform the same type of analysis in material obtained during spring cruises.

2.4. Molecular biology of melatonin synthesizing enzymes. In collaboration with Dr. L. Williams (The Rowett Institute, Aberdeen) we plan to study the molecular biology of melatonin synthesising enzymes (N-acetyltransferase, Hydroxy-O-methyltransferase) and the potential presence of clock genes (Per). Therefore, about half of the material cryofixed for biochemistry will be directed to her laboratory.

2.5. Pineal rhodopsins. Several pineals of *S. kaupi* have also been sent to Prof. D. Hunt (Inst. of Ophthalmology, London) for characterisation and molecular biology of potential pineal rhodopsins. This will be of particular interest in the context of the ongoing analysis of mesopelagic pineals collected during DISCO 243 and may shed light on the evolution of these molecules.

3. Further projects:

3.1. Microscopic analysis of sensory-motor co-ordination. The experiments conducted with the FRESP lander include the analysis of the quick start response in fish trapped in the instrument. This involves sensory-motor co-ordination processes, the morphological basis of which may be accessible by studying our brain collection. We intend to perform a microscopic analysis of the brains of the main species involved and study in particular the morphology of the cerebellum as a general site of such a co-ordination, and the Mauthner cells of the rhombencephalon which are responsible for mediating the startle response.

3.2. Retinal ganglion cells in multibank, all-rod retinae. Retinal ganglion cells in deep sea fish are interesting for two reasons: Their topography yields clues on potential areas of retinal specialisation

(areae, foveae), and the study of their morphological differentiation may indicate functional differences. We have recently shown that in the mesopelagic *Anoplogaster*, there are (at least) two types of retinal ganglion cell, one of which shows marked differences in regional density, and the other not. We have collected several retinae in as fresh a state as possible and applied fluorescent tracer molecules to the optic nerves in the hope that they reach the ganglion cell perikarya by retrograde axonal transport (hence fresh, living material) and label the ganglion cells selectively.

Species list (available on request)

13 BIOLUMINESCENCE IN THE BENTHIC BOUNDARY LAYER

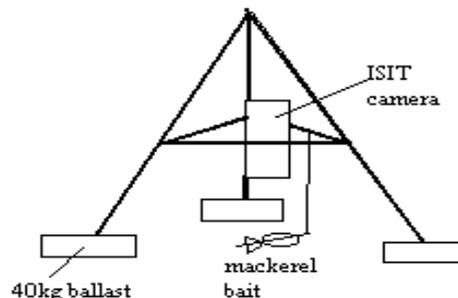
Emma Battle, University of Aberdeen.

Background

The ISIT lander is designed specifically to record deep-sea bioluminescence, using an ISIT (intensified silicon intensifying tube) camera. There are three ways to set up the ISIT camera:

- 1: **baited** – using 1 mackerel, slashed and tied in position 80cm below the vertically mounted camera
- 2: **splat screen** – using a wire mesh screen placed 50cm below the vertically mounted camera that records luminescence as organisms impact on the screen
- 3: **rotator** – the camera is mounted horizontally on a turntable which maintains the camera's position into the current.

Work on Discovery 250



During D250 the ISIT was deployed four times at stations ranging in depth from 1001 to 4099m in the Porcupine Seabight. Only the baited version was used. Bad weather prevented further deployments taking place in the Seabight or Abyssal Plain.

The videos from each deployment were reviewed on board. The first three deployments at 4000 m at the foot of the PSB, showed low levels of bioluminescence compared with similar experiments undertaken on a previous cruise at Cape Verde (D243). The final deployment in the north of the Seabight at 1000 m produced large amounts of bioluminescence, with many bright and persistent flashes observed. A variety of types of bioluminescence were seen, some of which may have been excreted. The videos will be examined in detail at Aberdeen University.

Future cruises shall attempt to identify the bioluminescent organism(s) and to trap them with the further possibility of examining luminescence in the laboratory.

14 IDENTIFICATION AND ACOUSTIC TRACKING OF SCAVENGING FISH IN THE PORCUPINE SEABIGHT

Camila Henriques, John Pringle and Phil Bagley, University of Aberdeen

Background

The Aberdeen University Deep Ocean Submersible (AUDOS) is designed to photograph and track scavenging fishes. One of the principal aims of the NERC grant (GR3/12789, Metabolism, activity and distribution patterns of deep-sea demersal fishes) is to use the AUDOS to determine routine swimming speeds of scavenging fish, particularly *C. (N.) armatus* at 4000 m and *A. rostrata* at 2500 m. In addition the photographic data provides information on the scavenging fauna for bathymetric, geographic and seasonal comparisons.

AUDOS Configuration

The AUDOS consisted of:

1. Aluminium Lander Frame
2. 15v Flash Battery + Casing
3. Twin Mors AR +RT Acoustic Releases
4. Ocean Instrumentation M7 Stills Camera + Flash Unit
5. Three Hydrophones
6. Control and Data Logging Computer
7. Nortek Acoustic Doppler Current Meter (replaced by Sontec current meter after deployment 1)

Buoyancy

Buoyancy was provided by a single large block of syntactic foam, with a single trimsyn float, marker buoy and syntactic pellet. A radio beacon, strobe and flag were mounted on the marker buoy to aid recovery

Deployment and recovery procedures

The lander was deployed over the stern. The pellet buoy was lowered first by hand followed by the flag mast and trimsyn float. Both the syntactic foam and the lander were lowered on the cranes and released into the water using 10 tonne quick releases.

For recovery a grappling line was thrown over the starboard side to catch the floating rope linking the pellet buoy to the flag mast. The pellet was disconnected and the mooring connected to a line taken from the auxiliary deck winch through a block on the starboard crane and around the stern. The mooring was then brought in over the stern, with care taken to ensure that the main propeller was stopped. The entire mooring was hauled on to the winch drum, with the marker buoy and trimsim float disconnected as they came on board. When the large float came on board it was stoppered off on deck

and disconnected to allow the vehicle to be brought on board. Problems were encountered disconnecting the main float, and an extra shackle and swivel were added to the outboard side on the last deployment.

Work Undertaken

Due to the weather conditions only five AUDOS deployments were achieved during this cruise, with three deployments at 4000 m, one at 2500 m and one at 4800 m.

Deployment 1 (Station 13905): Deployed at 4033 m at the mouth of the Porcupine Seabight with the new Nortek current meter. The new Nortek current meter failed during the deployment, and when retrieved it was found to contain a considerable quantity of salt water and one of the battery leads was completely corroded. The meter was allowed to dry and a new battery inserted and an attempt to retrieve the data prior to failure was made, but was unsuccessful due to PC's inability to communicate with current meter, which would indicate catastrophic failure and poor likelihood of being able to carry out repairs. M7 Camera failed, and on retrieval it would appear that the failure was due to salt water contamination of the connector for cable from battery to flash unit, the faulty connector was on the flash unit side. On retrieval the connector was smeared with silicone grease and all conducting components were well cleaned. After downloading the CAT (Code Aided Transponder) Data some concern was expressed over maximum operating distance and the need for tuning to optimum frequency (77 kHz).

Deployment 2 (Station 13911): Deployed at the mouth of the Seabight at 4124 m, with the failed Nortek current meter replaced with the Sensortec meter. Again M7 Camera failed due to salt water contamination, this time the cable between the flash unit and the battery was replaced.

Deployment 3 (Station 13915): Deployed at 4090 m at the mouth of the Seabight. All equipment operated successfully and full set of data was obtained.

Deployment 4 (Station 13917): Deployed at 2612 m in the Porcupine Seabight. All equipment operated successfully and full set of data was obtained. Remained on the seafloor for 10 days due to poor weather conditions preventing recovery. Strip of film from the end of the sequence developed and showed the grenadier *Coryphaenoides armatus* and blue-hake *Antimora rostrata*.

Deployment 5 (Station 13923): Deployed at 4848 m on the Porcupine Abyssal Plain. The M7 Camera failed after eight shots and it would appear that only 75% of the CAT data was logged. Further investigation is planned on return to Aberdeen

15 FISH RESPIROMETER, FRESP

David Bailey and Monty Priede, University of Aberdeen

Background

In situ respirometry forms part of the NERC grant (GR3/12789) investigating seasonal changes in deep-sea fish behaviour, physiology and distribution. A new lander has been developed and was tested during cruise D250. Aims of the trials included testing the mechanical and electronic systems under field conditions, evaluation of oxygen analysis systems and recording of fish behaviour in order to optimise the set-up of the respirometer for future cruises.

Due to adverse weather conditions, and as FRESP was a new vehicle, it was only deployed twice during D250. Despite this most of the above aims were achieved.

Methods

The respirometer consists of a 1.4x1.4x0.4m perspex chamber with a transparent upper surface. On deployment the sides of the chamber are held open to allow fish to enter, at a preset time the doors close sealing the chamber and trapping the fish within. The mechanical sampler is capable of taking up to eight 48ml water samples at pre-set times while a housed digital video camera records animal behaviour in and around the chamber. Timing of chamber closure was selected on the basis of likely fish arrival times with water samples taken at intervals for the following 50 hours.

Colorimetric and electrode oxygen analysis systems were tested for accuracy and reliability with seawater of known oxygen contents and used to measure the oxygen content of respirometer samples.

Results

1. Lander systems. All electronic and mechanical systems worked effectively, the only failure being damage to the chamber doors during deployment 13918. Stronger doors will be fitted.

2. Fish Behaviour. Low numbers of fish were recorded within the chamber prior to door closure with none being trapped by the respirometer during the two trial deployments. Future deployments will not illuminate the lander with white light prior to door closure and a fish trap will be incorporated into the chamber so that all fish attracted will be retained. The trap will also size-select the animals to be caught, greatly simplifying the analysis of the results. Much useful data for the design of future respirometers was obtained. Fast-start escape responses observed will be used in the planning and design of a new *in situ* fast-start experiment as part of the same NERC grant.

3. Oxygen analysis systems. The Acuvac colorimetric oxygen analysis system was simple and easy to use and gave consistent and accurate results with samples of known oxygen content. In the absence of a successful incubation its suitability for the current project is uncertain. Further tests will be

undertaken on return to the UK, a variety of alternative methods are under consideration. The Oxygen electrode proved less satisfactory with the readings being very unstable under the test conditions.

Summary

Much has been achieved through just two deployments with a range of modifications planned for the next cruise. Better weather would have enabled modification in the field and would probably have allowed some initial data collection.

16 BATHYSNAP

Dave Billet, SOC.

The time-lapse camera system Bathysnap was recovered from a location in the north-east of the Porcupine Seabight during the cruise. The camera had been deployed approximately 40 days earlier during a previous Discovery cruise (D248) on the edge of a deep, giant, carbonate mound. The Bathysnap was recovered without incident, but on deck the connector to the camera was found to have suffered recent damage. All the film had passed through the camera indicating a successful deployment. The frame interval had been set at 40 photographs per day and hopefully photographed the activity of coral communities living on the carbonate mound.

The Bathysnap was re-deployed on almost exactly the same position. The frame interval was set at 5 frames per day. The Bathysnap will be recovered during a cruise in 2001.

17 SEDIMENT TRAPS

Dave Billet, SOC.

A sediment trap mooring was recovered from a location on the Porcupine Abyssal Plain during the cruise. The sediment trap mooring was part of a long-term study spanning 11 years of organic matter flux through the water column to the seabed. The sediment traps had been placed at 1000m, 3000m and 4700m (100 m above bottom).

Recovery of the mooring was fairly uneventful, but the two lower moorings both came in upside down because the current meter and sediment traps had become entangled with lines attached to subsurface buoyancy. All the traps appeared to have worked successfully. Interestingly there was evidence for a late pulse of organic matter in October 1999. In addition, whereas during 1997 to 1998 the major flux period had been in July-August, the major flux period occurred much earlier in 2000. The trap at 3000m showed a peak in May, while the deeper trap at 4700m showed a peak in June.

The sediment traps were prepared for another deployment, but because of the shortage of time caused by the bad weather experienced by the cruise, and the failure of one of the trawls in this area, it was not possible to put the mooring back in the water.

18 RVS Technical Report

Jason Scott, RVS Technical Liaison Officer

The cruise consisted of 4 disciplines;

1. Lander deployment / recovery
2. OTSB trawling
3. CTDs'
4. Mooring recovery

1. Three different types of landers were used during the cruise (AUDOS, ISIT and FRESP).

These were all deployed through the stern gantry using the ACTA 30TM cranes and general purpose winch. The port crane blew 2 hoses on deployment of lander, which was probably due to large swell pulling the float prior to release. No other problems occurred.

2. OTSB trawls used the 20T winch system which worked fine during the cruise. The encoder belt drive parted on one drop which caused the wire out / speed to stop counting. A new belt was fitted and no other problems occurred.

3. There were 3 CTD drops for bottom water retrieval and acoustic release testing to a maximum of 4800m. No problems.

4. The sediment trap mooring was recovered at the end of the cruise using the stern cranes and the double barrel winch. Due to lack of time available this was not re-deployed.

19 STUDENT REPORTS

Judith Brewster, Aberdeen University

I felt that the cruise was a fantastic opportunity and gave me an insight into what a marine biologist might expect if they chose to follow a career in deep-sea biology. I certainly learnt a great deal on the cruise, not only about deep-sea fish, more so than I could have in a book. The cruise also gave me the chance to collect data for my fourth year project in what could be considered extreme conditions.

Apart from the initial bout of seasickness, I had a wonderful time on the cruise and I would like to thank Martin and Monty for giving me this opportunity, the other scientists and crew for making it an enjoyable cruise and also a big thanks to the galley crew who gave us something to look forward to when we were in the middle of a hurricane!!!

Oliver Yates, Aberdeen University

My time spent on the RRS Discovery has been excellent. I have enjoyed all aspects of the work and operations on deck plus been thoroughly satisfied with the general happenings on the ship. My time has been spent as the cephalopod man which would have been rather relaxed if it wasn't for helping out in the melee of fish processing as well. There has been an amazing increase in my understanding of

marine biology just by being part of this cruise not to mention the introduction to so many other aspects of deep-sea biology. However, if there is one thing that this experience has taught me it is that the weather in Aberdeen is not all that bad after all!

I feel it is important to give thanks to all aboard the Discovery without exception. The guys in the galley were fantastic, comedy moments in the saloon during horrific storms were calmly dealt with as the chefs cooked on! All the crew that kept the ship moving safely did a sterling effort and there wasn't a single moment where I thought we were in trouble, well..... Special thanks to Martin Collins and Monty Priede for providing the chance of sailing in the first place. This has been a superb voyage, I can only hope to be part of another in the near future especially if the same scientists and crew are on board again. I hope to see you all again soon

Andy Stocks, Aberdeen University

Three and a half weeks ago I set sail, onboard RRS Discovery, from Southampton with nothing but University lectures and laboratories as experience in the field of marine biology, that has now changed. My time onboard has taught me how to apply my knowledge gained at University in the real world of data collection and scientific research. The majority of data collected will be used in my chosen honours thesis. I would like to thank everyone who sailed on cruise D250 both crew and scientific compliment alike. Special thanks go out to the galley staff who provided us all with 'Mother' standard meals every day even when the storm outside was reaching force 11 status. Chiefly I would like to thank Both Monty and Martin initially for choosing me and for sparing so much of their valuable time helping me with my thesis project. I'm sure all the fantastic experiences I've gained throughout this cruise will be invaluable in the future. Once again I thank you all very much and will look back on my time aboard with very fond memories of you all.

Appendix I. Discovery 250-Station List

Station	Gear	Start				End				Min	Max	Comments
		Date	Time	Latitude	Longitude	Date	Time	Latitude	Longitude			
13903#1	CTD+WB	17/09/00	01:42	48 00.32 N	11 19.04 W	17/09/00	04:29	47 59.9 N	11 16.74 W	0	4216	Four water bottles and three acoustic releases
13903#2	CTD	17/09/00	05:15	47 59.71 N	11 20.07 W	17/09/00	07:59	47 59.15 N	11 21.12 W	0	4214	Three releases tested., all OK.
55101#1	DERA Mooring	09/09/99	10:43	48 18.59 N	11 55.08 N	17/09/00	12:15	48 18.59 N	11 55.08 W	2076	2076	Mooring recovered, slightly tangled
13904#1	ISIT	18/09/00	10:12	50 00.03 N	14 20.24 W	20/09/00	09:55	50 00.03 N	14 20.24 W	4014	4014	Deployed OK in F6. On surface at 11:20, on deck at 12:20.
13905#1	AUDOS	18/09/00	15:29	49 53.06 N	14 13.02 W	20/09/00	14:20	49 53.06 N	14 13.03 W	4033	4033	Deployed OK in F6/F7. Surface at 15:50, on board 16:25.
13906#1	OTSB	18/09/00	22:53	50 11.92 N	14 39.56 W	19/09/00	02:35	50 17.52 N	14 49.87 W	3986	4016	Good catch of fish and invertebrates Distance run 16.27 km.
13907#1	OTSB	20/09/00	01:27	49 55.85 N	14 30.34 W	20/09/00	04:50	50 02.64 N	14 36.04 W	4128	4220	Cod end lost, caught on wreck. Single piece of wreck recovered
13908#1	FRESP	20/09/00	17:56	49 49.99 N	14 03.32 W	24/09/00	06:23	49 49.99 N	14 03.29 W	3991	3991	Sank at 36 m/min. On bottom at 19:46; surface at 09:05. No fish trapped.
13909#1	ISIT	20/09/00	20:58	49 44.97 N	14 02.93 W	21/09/00	13:08	49 44.97 N	14 02.93 W	4072	4072	Deployed with bait. Little bioluminescence seen.
13910#1	OTSB	21/09/00	05:36	49 50.55 N	12 56.84 W	21/09/00	06:37	49 49.88 N	13 00.42 W	2456	2467	Good catch of fish and invertebrates. Distance run 4.43 km
13911#1	AUDOS	21/09/00	16:51	49 39.54 N	13 57.07 W	22/09/00	13:31	49 39.54 N	13 57.07 W	4124	4124	Part of ballast dropped during deployment: replaced. Camera failed.
13912#1	CTD + WB	21/09/00	19:28	49 36.80 N	13 55.95 W	21/09/00	22:02	49 35.41 N	13 56.97 W	0	4034	CTD with 3 water bottles fired 10 mab
13913#1	ISIT	21/09/00	23:34	49 42.49 N	14 00.19 W	22/09/00	16:30	49 42.49 N	14 00.19 W	4115	4115	Recovery attempted on 22/9/00 aborted, on surface until 13:20 on 23/9/00
13914#1	OTSB	22/09/00	07:29	49 54.84 N	13 34.34 W	22/09/00	08:55	49 52.88 N	13 38.99 W	2981	3115	Good catch of <i>C.armatus</i> and <i>Benthothuria</i> . Distance run 6.785 km
13915#1	AUDOS	23/09/00	16:14	49 44.17 N	14 04.70 W	24/09/00	10:05	49 44.17 N	14 04.70 W	4090	4090	Port crane hose failed during deployment. Surface at 11:57.
13916#1	OTSB	24/09/00	00:35	49 36.29 N	13 32.22 W	24/09/00	01:48	49 38.96 N	13 34.73 W	2023	2093	Reasonable catch of fish and inverts, net belly damaged. Distance run 5.79 km.
13917#1	AUDOS	24/09/00	17:55	50 17.90 N	13 13.14 W	05/10/00	17:40	50 17.90 N	13 13.14 W	2612	2612	Surface at 18:52. Good photos and tracking data.

13918#1	FRESP	24/09/00	21:34	50 29.93 N	15 00.12 W	05/10/00	14:21	20 29.93 N	15 00.12 W	2452	2452	Surface at 15:59. Door broken, but good video of fish.
13880#1	BATHYSNAP	08/08/00	08:09	51 26.77N	11 45.22 W	25/09/00	06:40	51 26.77N	11 45.22 W	884	884	Recovered in good conditions, but camera lead damaged.
13919#1	OTSB	25/09/00	12:39	51 08.89 N	12 03.92 W	25/10/00	13:46	51 07.34 N	12 00.56 W	1537	1545	Good catch of fish and invertebrates. Distance run 5.04 km, Course 126.
13920#1	ISIT	25/09/00	17:11	51 10.40 N	11 40.93 W	05/10/00	06:22	51 10.40 N	11 40.93 W	1002	1002	Mackerel bait. Excellent bioluminescence on bait.
13921#1	BATHYSNAP	27/09/00	02:31	51 26.77 N	11 45.13 W							5 frames per day. Deployed in 35 knot winds.
13922#1	OTSB	01/10/00	02:34	50 53.88 N	11 58.44 W	01/10/00	03:35	50 55.63 N	12 01.52 W	1885	1933	Good catch of fish and invertebrates. Distance run 4.99 km.
13923#1	AUDOS	06/10/00	10:19	48 59.77 N	16 30.01 W	07/10/00	06:31	48 59.77 N	16 30.01 W	4848	4848	Camera failed after 13 shots.
55102#2	SEDIMENT TRAP	08/09/99	12:00	48 58.43 N	16 25.82 W	06/10/00	10:51	48 58.43 N	16 25.82 W	4837	4837	Surface 11:05; all on board 13:52
13924#1	OTSB	06/10/00	19:55	48 42.18 N	16 46.04 W	06/10/00	22:05	48 38.83 N	16 55.00 W	4790	4784	Did not reach bottom, some benthopelagic animals caught, ~50mab.
13925#1	OTSB	07/10/00	15:45	48 53.46 N	16 45.86 W	07/10/00	19:20	48 56.79 N	16 54.73 W	4835	4845	Distance run 16.30 km, course 300. Good invertebrates catch.

Note: Lander start times are the time the flag disappears beneath the surface, except for Bathysnap, where start times are on seafloor. End times are release times.

APPENDIX II NAMES, ADDRESSES AND E-MAILS OF ALL SCIENTIFIC PARTICIPANTS.

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Appendix III. Fish Species Caught During Discovery 250.

Species	CODE	Family	Tissue
<i>Alepocephalus agassizii</i>	ALA	Alepocephalidae	
<i>Alepocephalus bairdii</i>	ALB	Alepocephalidae	
<i>Alepocephalus productus</i>	ALP	Alepocephalidae	+
<i>Alepocephalus rostratus</i>	ALR	Alepocephalidae	+
<i>Bellocia koefedi</i>	BEK	Alepocephalidae	+
<i>Bathytroctes microlepis</i>	BTM	Alepocephalidae	+
<i>Conacara</i> sp.	CN?	Alepocephalidae	+
<i>Conacara macroptera</i>	CNM	Alepocephalidae	
<i>Conacara murrayi</i>	CNR	Alepocephalidae	
<i>Conacara salmonea</i>	CNS	Alepocephalidae	
<i>Narctes stomias</i>	NAS	Alepocephalidae	
<i>Xenodermichtys copei</i>	XEC	Alepocephalidae	+
<i>Borostomias antarcticus</i>	BOA	Astronesthidae	
<i>Bathylagus euryops</i>	BLE	Bathylagidae	+
<i>Cataetx laticeps</i>	CAL	Bythidae	+
<i>Platyberyx opalescens</i>	PLO	Caristiidae	
<i>Chauliodus sloanii</i>	CSS	Chauliodontidae	
<i>Chiasmodon niger</i>	CIN	Chiasmodontidae	
<i>Kali macrodon</i>	KAM	Chiasmodontidae	
<i>Chimaera monstrosa</i>	CHM	Chimeridae	
<i>Hydrolagus mirabilis</i>	HYM	Chimeridae	+
<i>Bathypterois dubius</i>	BPD	Chlorophthalmidae	+
<i>Gonostoma bathyphylum</i>	GOB	Gonostomatidae	+
<i>Halosaurusopsis macrochir</i>	HAM	Halosauridae	+
<i>Leptoichthys agassizii</i>	LPA	Leptoichthyidae	
<i>Coelorhynchus occa</i>	CLL	Macrouridae	
<i>Coryphaenoides armatus</i>	COA	Macrouridae	+
<i>Coryphaenoides brevibarbis</i>	COB	Macrouridae	
<i>Lionurus carapinus</i>	COC	Macrouridae	+
<i>Coryphaenoides guentheri</i>	COG	Macrouridae	+
<i>Coryphaenoides leptolepis</i>	COL	Macrouridae	+
<i>Coryphaenoides mediterraneus</i>	COM	Macrouridae	+
<i>Coryphaenoides profundicola</i>	COP	Macrouridae	+
<i>Coryphaenoides rupestris</i>	COR	Macrouridae	+
<i>Nezumia aequalis</i>	NEA	Macrouridae	+
<i>Trachyrincus murrayi</i>	TRM	Macrouridae	+
<i>Malacosteus niger</i>	MAN	Malacosteidae	
<i>Poromitra capito</i>	PMC	Melamphidae	+
<i>Antimora rostrata</i>	ANR	Moridae	+
<i>Lampanyctus</i> sp.	LA?	Myctophidae	
<i>Lampanyctus macdonaldi</i>	LAM	Myctophidae	
<i>Myctophidae</i>	MC?	Myctophidae	
<i>Nemichthys scolopaceus</i>	NES	Nemichthidae	+
<i>Notocanthus bonapartei</i>	NOB	Notocanthidae	
<i>Notocanthus chemnitzii</i>	NOC	Notocanthidae	
<i>Polyacanthonotus challengerii</i>	POC	Notocanthidae	
<i>Polyacanthonotus rissoanus</i>	POR	Notocanthidae	+
<i>Spectrunculus grandis</i>	SPG	Ophidae	+
<i>Cottunculus thompsonii</i>	CTT	Psychrolutidae	
<i>Bathyraja richardsoni</i>	BRR	Rajidae	
<i>Etmopterus</i>	ET?	Squalidae	+
<i>Histiobranchus bathybius</i>	HIB	Synphobranchidae	+
<i>Synphobranchus kaupii</i>	SYK	Synphobranchidae	
<i>Bathysaurus ferax</i>	BSF	Synodontidae	+
<i>Bathysaurus mollis</i>	BSM	Synodontidae	
<i>Lycodes</i> sp.	LC?	Zoarcidae	

Appendix IV. Weather conditions during Discovery 250

Cruise D250

Wind Summary

R.R.S. DISCOVERY

