

# **CRUISE REPORT**

## **Leg 8 Malaspina**

**BiO Sarmiento de Gamboa**

**Captain**

**David Domínguez Añino**

**27 January - 15 March 2011**

**Chief Scientist**

**Alonso Hernández Guerra**

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## Scientists

Alonso Hernández Guerra	Chief Scientist	ULPGC
Eugenio Fraile Nuez	IP, CTD+LADCP+XBT+Argo	IEO
Maria Dolores Pérez Hernández	CTD+Termo+Altim+XBT	ULPGC
Verónica Benítez Barrios	CTD+XBT+SMOS	ULPGC
David Sosa Trejo	LADCP+SADCP	ULPGC
José Francisco Domínguez Yanes	IP, Oxygen+nutrients	IEO
María D. García Lestón	Oxygen+nutrients	IEO
Iván Alonso González	Oxygen+nutrients	ULPGC
Amelia Serrano Martínez	Oxygen+nutrients	ULPGC
Alejandro Caballero Ramis	Autosal	UCA
Marcos Vázquez Rodríguez	Alcalinity+pH+pCO <sub>2</sub>	IIM-CSIC
Xosé Antonio Padín Álvarez	Alcalinity+pH+pCO <sub>2</sub>	IIM-CSIC
Elisa Fernández Guallart	Alcalinity+pH+pCO <sub>2</sub>	ICM-CSIC
Sofía Sal Bregua	Metabolism+DOC	IEO
Mar Benavides Gorostegui	N <sub>2</sub>	ULPGC
José Varela Romay	Plankton+POC+PON	IEO
Elena Mesa Cano	Isotopics 13C/12C	CSIC
Jesús M <sup>a</sup> Moreno Vidal	Spread of science	
María del Mar Díaz Sánchez	Spread of science	

## Watches

Chief Scientist: Alonso Hernández Guerra

ADCP: Antonio Martínez Marrero

Autosal and Thermosalinometer: Maria Dolores Pérez Hernández

Analysis oxygen+nutrients: José Escánez Escánez

<b>8-12 h</b>	<b>12-4 h</b>	<b>4-8 h</b>
<b>CTD+LADCP+XBT</b>		
Maria Dolores Pérez David Sosa	Verónica Benítez	Eugenio Fraile Nuez Alejandro Caballero
<b>Oxygen+nutrients+salinity</b>		
José Francisco Domínguez Amelia Serrano	Iván Alonso	María García
<b>Inorganic Carbon</b>		
Elisa Fernández	Marcos Vázquez	Xosé Antonio Padín

## **Acknowledgments**

First of all, I want to thank Captain David Domínguez for the good human and logistical collaboration to carry out this campaign. The synergy between us was so excellent that we solved all the little problems that always appear in such a long campaign. I also want to thank the good work and treatment of the members of the UTM. They promptly fixed any technical problems that appeared during the voyage. The BiO Sarmiento de Gamboa was an excellent ship to work with during the fifty days that the voyage lasted. The 16 scientists on board worked in perfect harmony and it has been one of the most enthusiastic groups that I have had the privilege of working with. Finally, to thank the presence of Jesus and Mary of outreach who taught us to look at science from another perspective.

## Hydrographic stations

Station	Lat Grad	Lat Min.	Lon Grad.	Lon Min.	Dist. miles	Depth m
Las Palmas	28	8. 50	-15	25. 50	112. 5	0
001	27	46. 51	-13	20. 57	4. 2	97
002	27	45. 06	-13	25. 08	3. 4	252
003	27	43. 90	-13	28. 67	3. 4	590
004	27	42. 74	-13	32. 27	6. 8	896
005	27	40. 42	-13	39. 46	9. 4	1156
006	27	37. 19	-13	49. 45	9. 4	1430
007	27	33. 96	-13	59. 45	11. 0	1864
008	27	30. 20	-14	11. 07	11. 0	2075
To get rid of PAR						
009	27	26. 45	-14	22. 70	11. 0	2252
010	27	22. 70	-14	34. 32	22. 0	2523
1 XBT between sta.						
011	27	15. 19	-14	57. 57	22. 0	2771
1 XBT between sta.						
012	27	7. 68	-15	20. 81	20. 1	3049
1 XBT between sta.						
013	27	0. 82	-15	42. 04	20. 1	3333
1 XBT between sta.						
014	26	53. 97	-16	3. 26	20. 1	3494
1 XBT between sta.						
015	26	47. 11	-16	24. 49	21. 2	3575
1 XBT between sta.						
016	26	39. 89	-16	46. 84	21. 2	3623
1 XBT between sta.						
017	26	32. 68	-17	9. 18	21. 3	3610
1 XBT between sta.						
018	26	25. 46	-17	31. 52	21. 3	3635
1 XBT between sta.						
019	26	18. 25	-17	53. 87	21. 3	3597
1 XBT between sta.						
020	26	11. 03	-18	16. 21	24. 9	3588
1 XBT between sta.						
021	26	2. 61	-18	42. 28	24. 9	2412
1 XBT between sta.						
022	25	54. 19	-19	8. 35	24. 9	3458
1 XBT between sta.						
023	25	45. 77	-19	34. 42	25. 0	3818
Argo 764						
1 XBT between sta.						
024	25	37. 35	-20	0. 48	25. 0	3487
1 XBT between sta.						
025	25	28. 93	-20	26. 55	29. 2	4307
1 XBT between sta.						
Done twice						
026	25	19. 11	-20	56. 96	29. 2	4436

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1 XBT between sta.						
027 1 XBT between sta.	25	9.29	-21	27.37	29.2	4606
028 1 XBT between sta.	24	59.47	-21	57.79	29.3	4713
029 Argo 765 1 XBT between sta.	24	49.64	-22	28.20	29.3	4817
030 1 XBT between sta.	24	39.82	-22	58.61	29.3	4941
031 2 XBT between sta.	24	30.00	-23	29.02	33.4	5023
032 2 XBT between sta.	24	30.00	-24	5.69	33.4	5108
033 Argo 769 2 XBT between sta.	24	30.00	-24	42.35	33.4	5212
034 Drifter 2 XBT between sta.	24	30.00	-25	19.02	33.4	5292
035 Drifter 2 XBT between sta.	24	30.00	-25	55.69	33.4	5336
036 2 XBT between sta.	24	30.00	-26	32.35	33.4	5437
037 2 XBT between sta. Done twice	24	30.00	-27	9.02	33.4	5458
038 Drifter 2 XBT between sta.	24	30.00	-27	45.68	33.4	5608
039 2 XBT between sta. Done twice	24	30.00	-28	22.34	33.4	5694
040 2 XBT between sta.	24	30.00	-28	59.01	33.4	5677
041 Argo 770 2 XBT between sta.	24	30.00	-29	35.68	33.4	5596
042 SMOS Drifter 2 XBT between sta.	24	30.00	-30	12.34	33.4	5566
043 2 XBT between sta.	24	30.00	-30	49.01	33.4	5884
044 2 XBT between sta.	24	30.00	-31	25.67	33.4	6022
045 2 XBT between sta.	24	30.00	-32	2.34	33.4	5628

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046 2 XBT between sta.	24	30.00	-32	39.01	33.4	6349
047 2 XBT between sta.	24	30.00	-33	15.67	33.4	5931
048 Argo 771 2 XBT between sta.	24	30.00	-33	52.34	33.4	6062
049 1 XBT between sta.	24	30.00	-34	20.05	25.2	5654
050 1 XBT between sta.	24	30.00	-34	47.77	25.2	6190
051 1 XBT between sta. Drifter	24	30.00	-35	15.48	25.2	5414
052 1 XBT between sta.	24	30.00	-35	43.19	25.2	5631
053 1 XBT between sta.	24	30.00	-36	10.90	25.2	5826
054 1 XBT between sta.	24	30.00	-36	38.62	25.2	5050
055 1 XBT between sta. Drifter	24	30.00	-37	6.33	25.2	5746
056 1 XBT between sta.	24	30.00	-37	34.04	25.2	5830
057 1 XBT between sta.	24	30.00	-38	1.75	25.2	5519
058 1 XBT between sta.	24	30.00	-38	29.47	25.2	4568
059 1 XBT between sta.	24	30.00	-38	57.18	25.2	4010
060 1 XBT between sta.	24	30.00	-39	24.89	25.2	4800
061 1 XBT between sta. Drifter Done twice	24	30.00	-39	52.60	25.2	4740
062 1 XBT between sta.	24	30.00	-40	20.32	25.2	5091
063 1 XBT between sta. Argo 773	24	30.00	-40	48.03	25.2	4784
064 1 XBT between sta.	24	30.00	-41	15.74	25.2	4606
065 1 XBT between sta.	24	30.00	-41	43.45	25.2	4778
066 1 XBT between sta. Drifter	24	30.00	-42	11.17	25.2	3990
067	24	30.00	-42	38.88	25.2	4364

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1 XBT between sta. SMOS						
068 1 XBT between sta.	24	30.00	-43	6.59	25.2	3762
069 1 XBT between sta.	24	30.00	-43	34.30	25.2	4006
070 1 XBT between sta.	24	30.00	-44	2.02	25.2	4154
071 1 XBT between sta. Done twice	24	30.00	-44	29.73	25.2	3895
072 Drifter 1 XBT between sta.	24	30.00	-44	57.44	25.2	3622
073 1 XBT between sta.	24	30.00	-45	25.15	25.2	3078
074 1 XBT between sta.	24	30.00	-45	52.87	25.2	2419
075 1 XBT between sta.	24	30.00	-46	20.58	25.2	2779
076 Drifter 1 XBT between sta.	24	30.00	-46	48.29	25.2	3520
077 1 XBT between sta.	24	30.00	-47	16.00	25.2	4075
078 1 XBT between sta. Done twice	24	30.00	-47	43.72	25.2	3816
079 1 XBT between sta.	24	30.00	-48	11.43	25.2	4021
080 1 XBT between sta.	24	30.00	-48	39.14	25.2	4505
081 Drifter Argo 774 1 XBT between sta.	24	30.00	-49	6.85	25.2	4187
082 1 XBT between sta.	24	30.00	-49	34.57	25.2	4675
083 1 XBT between sta.	24	30.00	-50	2.28	25.2	4437
084 1 XBT between sta.	24	30.00	-50	29.99	25.2	5253
085 Drifter 1 XBT between sta.	24	30.00	-50	57.70	25.2	5424
086 1 XBT between sta.	24	30.00	-51	25.42	25.2	4925
087 1 XBT between sta.	24	30.00	-51	53.13	25.2	5074
088	24	30.00	-52	20.84	25.2	5406

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1 XBT between sta.						
089 Drifter 1 XBT between sta.	24	30.00	-52	48.55	33.5	5106
090 1 XBT between sta.	24	30.00	-53	25.33	33.5	5522
091 1 XBT between sta.	24	30.00	-54	2.11	33.5	6098
092 Drifter 1 XBT between sta.	24	30.00	-54	38.89	33.5	5752
093 Argo 775 1 XBT between sta.	24	30.00	-55	15.67	33.5	5954
094 1 XBT entre sta.	24	30.00	-55	52.44	33.5	5935
095 1 XBT between sta.	24	30.00	-56	29.22	33.5	6212
096 Drifter 1 XBT between sta.	24	30.00	-57	6.00	33.5	6162
097 1 XBT between sta.	24	30.00	-57	42.78	33.5	6176
098 Argo 776 1 XBT between sta.	24	30.00	-58	19.55	33.5	5901
099 Drifter 1 XBT between sta.	24	30.00	-58	56.33	33.5	5797
100 1 XBT between sta.	24	30.00	-59	33.11	33.5	5838
101 Drifter 1 XBT between sta.	24	30.00	-60	9.89	33.5	5941
102 Argo 777 1 XBT between sta.	24	30.00	-60	46.67	33.5	5818
103 Drifter 1 XBT between sta.	24	30.00	-61	23.45	33.5	5880
104 Drifter 1 XBT between sta.	24	30.00	-62	0.23	33.5	5885
105 1 XBT between sta.	24	30.00	-62	37.00	33.5	5917
106 Drifter 1 XBT between sta.	24	30.00	-63	13.78	33.5	5855
107 Argo 778	24	30.00	-63	50.56	33.5	5898

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1 XBT between sta.						
108	24	30.00	-64	27.34	33.5	5771
1 XBT between sta.						
109	24	30.00	-65	4.12	33.5	5816
1 XBT between sta.						
110	24	30.00	-65	40.90	33.5	5627
1 XBT between sta.						
111	24	30.00	-66	17.68	33.5	5275
1 XBT between sta.						
112	24	30.00	-66	54.46	33.5	5731
<b>Argo 779</b>						
1 XBT between sta.						
113	24	30.00	-67	31.23	33.5	5630
1 XBT between sta.						
114	24	30.00	-68	8.01	27.3	5738
1 XBT between sta.						
115	24	30.00	-68	38.00	27.3	5724
1 XBT between sta.						
116	24	30.00	-69	8.00	24.2	5635
1 XBT between sta.						
117	24	50.44	-69	22.23	32.2	5592
1 XBT between sta.						
118	25	17.69	-69	41.23	32.2	5548
1 XBT between sta.						
119	25	44.95	-70	0.23	32.2	5500
1 XBT between sta.						
120	26	12.20	-70	19.24	20.9	5505
1 XBT between sta.						
121	26	12.20	-70	42.58	20.9	5486
1 XBT between sta.						
122	26	12.20	-71	5.90	20.9	5500
1 XBT between sta.						
123	26	12.20	-71	29.24	20.9	5478
1 XBT between sta.						
124	26	12.20	-71	52.57	20.9	5373
1 XBT between sta.						
125	26	12.20	-72	15.91	20.9	5268
1 XBT between sta.						
126	26	12.20	-72	39.24	20.9	5228
1 XBT between sta.						
127	26	12.20	-73	2.57	20.9	5121
1 XBT between sta.						
128	26	12.20	-73	25.91	20.9	5151
1 XBT between sta.						
129	26	12.20	-73	49.24	15.7	5107
1 XBT between sta.						
130	26	12.20	-74	6.74	15.7	4890
1 XBT between sta.						
131	26	12.20	-74	24.24	15.7	4816
1 XBT between sta.						

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132 1 XBT between sta.	26	12. 20	-74	41. 74	15. 7	4583
133 1 XBT between sta.	26	12. 20	-74	59. 24	15. 7	4597
134 1 XBT between sta.	26	12. 20	-75	16. 74	15. 7	4639
135	26	12. 20	-75	34. 24	7. 8	4640
136	26	12. 20	-75	42. 97	7. 8	4714
137	26	12. 20	-75	51. 70	7. 8	4864
138	26	12. 20	-76	0. 42	7. 8	4756
139	26	12. 20	-76	9. 14	7. 8	4775
140	26	12. 20	-76	17. 87	7. 8	4716
141	26	12. 20	-76	26. 60	4. 8	4767
142	26	12. 20	-76	31. 95	4. 8	4703
143	26	12. 20	-76	37. 30	4. 8	4673
144	26	12. 20	-76	42. 65	4. 8	4637
145	26	12. 20	-76	48. 00	2. 1	4723
146	26	12. 20	-76	50. 29	2. 1	4723
147	26	12. 20	-76	52. 58	2. 1	3969
148	26	12. 20	-76	54. 86	2. 1	3969
149	26	12. 20	-76	57. 16	2. 1	3652
150	26	12. 20	-76	59. 44	2. 1	3386
151	26	12. 20	-77	1. 73	2. 1	3386
152	26	12. 20	-77	4. 02	2. 1	1634
153	26	12. 20	-77	6. 31	26. 9	1634
GBahama	27☐	0. 03	-77☐	12. 03	107. 7	-0
GBahama	27☐	27. 05	-79☐	9. 25	27. 1	-0
154	27	0. 03	-79	09. 87		348
155	27	0. 03	-79	11. 00	3. 4	452
156	27	0. 03	-79	14. 83	3. 4	584
157	27	0. 03	-79	18. 67	3. 4	634
158	27	0. 03	-79	22. 50	3. 4	707
159	27	0. 03	-79	26. 33	3. 4	743
160	27	0. 03	-79	30. 17	3. 4	743
161	27	0. 03	-79	34. 00	3. 4	667
162	27	0. 03	-79	37. 84	3. 4	596
163	27	0. 03	-79	41. 66	3. 4	467
164	27	0. 03	-79	45. 50	3. 4	427
165	27	0. 03	-79	49. 33	3. 4	368
166	27	0. 03	-79	53. 17	3. 4	236
167	27	0. 03	-79	57. 00	754. 7	43
S. Domingo	18	25. 50	-69	58. 50	0. 0	-0

## **Narrative**

According to the station locations shown in the previous table, only 8 hydrographic stations were carried out in USA national waters. It was stations 160 to 167. Thus, only these stations will be referred in this report.

**13th. March.** Hydrographic stations **160, 161, 162, 163 and 164** were successfully done. In the station **165**, Nyskin bottle 1 (323dbar) arrived open. Stations **166 and 167** were successfully done.

## **XBT, Drifters, SMOS and Argo**

No XBT, drifters, SMOS and Argo were launched in USA waters.

## Rosette+CTD+LADCP

### 1. Instrumentation

The CTD system used was a Sea-Bird 9/11 plus with dual T / S sensor, an altimeter, a 2 X 300 kHz LADCP system with RDI Master and Slave, 24 x 10L sampling bottles in the 24 positions of the rosette.

The processing applied to this data is in the Malaspina white paper and will not be repeated here.

### 2. Instrumentation handling procedure.

The operation of the CTD and the LADCP is performed from the same computer so there is no synchronization problem.

For each station, the launching of the CTD was the same. First of all, the bridge confirmed that we were on station. Shortly after receiving this confirmation, the CTD surface unit was turned on and the LADCP Master and Slave configuration files were entered. The rosette was held at a depth of approximately 10 meters for 3 minutes until the reading of the temperature and salinity data from the primary and secondary sensors was stable. Subsequently, the operator manually lowered the rosette to a depth of 50 m at 30 m/min. At that depth, the rosette automatically goes downward at 50 m/min to a maximum depth that was generally 100 m above the bottom of the ocean. Once at that depth, it was switched back to manual mode at a speed of 30 m/min ending at approximately 20 m from the bottom, indicated by the altimeter installed on the rosette. At this depth the first bottle was closed. Once closed, the rosette began to rise, stopping in the following depths to close the bottles:

Surface  
20-30 m above the DCM  
DCM  
75  
100 (Conditional 1)  
125  
150  
200 (Conditional 2)  
300  
400  
600  
800  
900 (Conditional 4)  
1000  
1100 (Conditional 3)  
1200

1300  
1500  
1750  
2000  
2500  
3000  
3500  
4000  
4500  
5000  
5500  
Bottom of the ocean (Never > 6000m)

The conditionals were closed according to the depth of the station. Data acquisition ceased when the instruments were on board. The processing of the CTD and LADCP data was done with different computers. The type of processing of both instruments is not described here because it is set out in the Malaspina white paper.

## **LADCP**

We have had problems with the RDI heads although all of them (4) were new or recently arrived after a review in RDI. The ones that broke came full of water. As shown in the LADCP report, the first head of the UTM-Slave (s / n: 15061) worked from station 1 to 24, the second head of the UTM-Master (s / n: 15330) worked between From stations 1 to 38, the first head of the ULPGC-Slave (s / n: 15231) worked between stations 25 to 41. From this station we only have the Master of the ULPGC.

## **LADCP REPORT**

### **Introduction:**

LADCP Model: WorkHorse Monitor 300 Khz

Ship: B/O Sarmiento de Gamboa

Cruise: Malaspina Leg 8

**CRUISE REPORT LEG 8 MALASPINA**

Date: 27/01/2011 to 07/02/2011

**Stations and Floodings**

CPU SERIAL NUMBER

CAST	MASTER (dnlooking)	SLAVE (uplooking)	DEPTH reached (metres)	BOTTOM	COMMENTS
1	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	78	87	
2	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	98	107	
3	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	500	520	
4	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	696	706	
5	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	1063	1073	
6	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	1422	1432	
7	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	1682	1693	
8	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	2022	2029	
9	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	2208	2218	
10	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	2450	2459	
11	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	2735	2744	
12	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	3002	3010	
13	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	3322	3330	
14	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	3460	3470	
15	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	3551	3560	
16	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	3606	3617	
17	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	3609	3618	
18	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	3611	3623	
19	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	3599	3605	
20	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	3487	3505	
21	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	3217	3229	
22	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	3440	3454	
23	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	3779	3792	
24	25 00 00 00 93 98 C9 09	CD 00 00 00 93 A8 D8 09	3970	3984	
25	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	4286	4295	Cast Repeated, ADCP SN#CD00000093A8D809 flooded
26	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	4430	4440	

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27	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	4572	4580	
28	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	5873	-	
29	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	4802	4812	
30	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	4925	4934	
31	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	5005	5015	
32	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	5108	5117	
33	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	5193	5202	
34	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	5275	5284	
35	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	5338	5346	
36	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	5418	5428	
37	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	5519	5528	
38	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	5598	5606	
39	25 00 00 00 93 98 C9 09	0C 00 00 06 42 BD 69 09	5644	5654	Cast Repeated, ADCP SN#250000009398C909 flooded
40	6D 00 00 02 FB 9A 85 09	0C 00 00 06 42 BD 69 09	5669	5822	
41	6D 00 00 02 FB 9A 85 09	0C 00 00 06 42 BD 69 09	5696	5929	
42	6D 00 00 02 FB 9A 85 09		5322	5403	ADCP SN#0C00000642BD6909 flooded
43	6D 00 00 02 FB 9A 85 09	no more ADCP to use	5764	5890	
44	6D 00 00 02 FB 9A 85 09		5684	5694	
45	still working				
	...				

## Instruments Report

### LADCP s/n 15061 (CD 00 00 00 93 A8 D8 09) – UTM

This instrument was initially orientated in the carousel as a Slave. After 24 hydrographic stations, we couldn't establish the communication with the system anymore. Once at the surface, we removed it from the carousel and we realized that a burn smelt come from the beam faces. We decided to open it and the system was completely full of water, as you can see on the figures enclosed. We haven't got any evidence that the water could be penetrated into the O-rings.

**CRUISE REPORT LEG 8 MALASPINA**

**LADCP s/n 15330 (25 00 00 00 93 98 C9 09) – UTM**

This instrument was initially orientated in the carousel as a Master. After 39 hydrographic stations, we couldn't establish the communication with the system anymore. Once in the surface, we removed it from the carousel and we realized that a burn smelt come from the beam faces. We decided to open it and the system was completely full of water, as you can see on the figures enclosed. We haven't got any evidence that the water could be penetrated into the O-rings.

**LADCP s/n 15231 (0C 00 00 06 42 BD 69 09) – ULPGC**

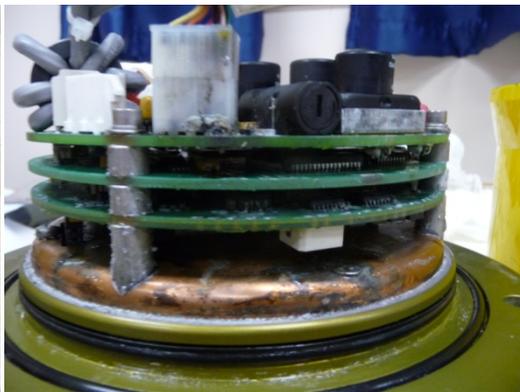
This instrument was initially orientated in the carousel as a Slave. After 16 hydrographic stations, we couldn't establish the communication with the system anymore. Ones in the surface, we removed it from the carousel and we realized that a burn smelt come from the beam faces. We decided to open it and the system was completely full of water, as you can see on the figures enclosed. We haven't got any evidence that the water could be penetrated into the O-rings.

**LADCP s/n 14794(6D 00 00 02 FB 9A 85 09) – ULPGC**

Still working

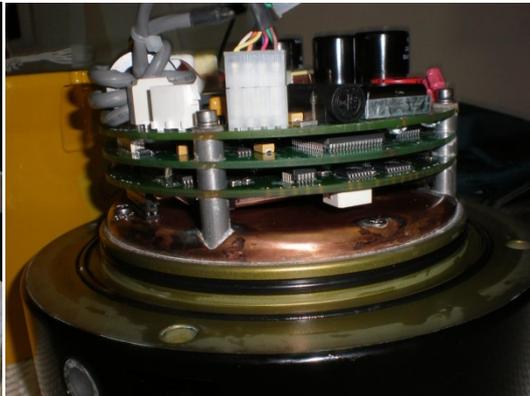
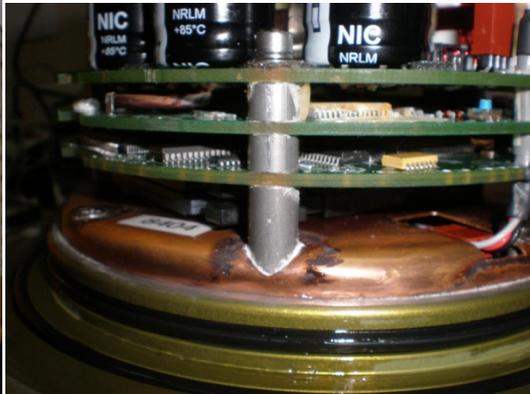
CRUISE REPORT LEG 8 MALASPINA

PHOTO LADCP s/n 15330 (UTM)



**CRUISE REPORT LEG 8 MALASPINA**

**PHOTOS LADCP s/n 15061 (UTM)**



PHOTOS LADCP s/n 15231 (ULPGC)



***CRUISE REPORT LEG 8 MALASPINA***

**Report Date 2011-02-10**

**B/O Sarmiento de Gamboa**

**David M. Domínguez Añine**

**(Captain Sarmiento de Gamboa)**

**Alonso Hernandez Guerra**

**(Chief Scientist)**

**Javier Vallo Rodríguez**

**(Cruise Technician Boss)**

## **AUTOSAL/PORTASAL**

It began using the IEO Autosal. In the first 31 stations, the slope was less than 1.0 and, therefore, the Autosal was not working properly. In stations 32-37, the same samples were calibrated by Autosal and Portasal that the UTM has installed on the ship. Slopes  $<1.0$  were obtained in the case of Autosal and Portasal. Due to the fact that we have used a dual sensors of CTD, stations 1-31 do not give significant differences in conductivity between the two sensors. From station 32 it will be calibrated with the data from the Portasal and after station 61 we will continue with the Portasal

## OXYGEN SENSOR CALIBRATION SBE43DO

The calibration of the SBE43DO oxygen sensor installed in the CTD has been performed following the procedure recommended by SeaBird and transcribed below. The procedure described in your Application Note is not correct.

- > It is possible that your range of DO concentration values is not large
- > enough to get a proper linear slope and offset correction. I would
- > recommend the following, having not seen any data.
- >
- > Follow APP Note 64-2, but \_use the 2nd method\_ that is shown. What you
- > would do there is the following:
- >
- > 1. Make sure you applied the hysteresis correction to your casts that
- > were deeper than 1000 dbar when you converted from SBE43 Oxygen Voltage
- > to Concentration. (see application Note 64-3 attached)
- >
- > 2. Ratio the Winkler values to the SBE43 oxygen concentration values at
- > each bottle stop.
- > Look at replicate Winkler STDEV first and make sure your bottle data
- > are suitable and that all is well with the chemistry.
- > Look at the stdev of the bottle sum oxygen data too, to be sure the
- > readings were stablized (carousel stop time was long enough to allow a
- > good sample)
- >
- > Compute:  $\text{Winkler value} / \text{SBE 43 Value}$  (same units i.e. ml/L, umol/kg)
- >
- > 3. Calculate the mean of this ratio and the STDEV for the cast
- > Value should be greater than 1
- > STDEV should be reasonably small (the ratio correction should be
- > very close regardless of the concentrations)
- > If STDEV is large, you might need to not use bottles and CTD data
- > from the steeper gradient regions...look for bottle stops where the
- > stdev in the Winkler and CTD measurements were low, and also where there
- > were not strong thermal or DO gradients that could dilute the bottle
- > value. Then remove those, and recalculate the ratio again.
- >
- > 4. Take the Mean ratio value (1.\*\*\*) and multiply this by your old SOC
- > value from your CON file (Use the SOC value used for calculating oxygen
- > concentration in DATCNV during this process).
- >
- > This is your newSOC value to use in an updated CON file (rename to keep
- > them separate).
- >
- > Note, you can also just multiply all the SBE 43 concentration data by
- > this ratio too, and compare the results with the bottles. But you will
- > want to reprocess the data...processing on SBE 43 oxygen voltage and

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- > then derive the concentration after all the T, C, and P, and DO
- > alignments are completed. Always process on SBE 43 Oxygen voltage...and
- > Derive oxygen concentration at the end. For Bottle comparisons, you
- > obviously need the SBE 43 concentrations, so that is why you convert it
- > in DATCNV...only for the bottle comparisons.
- >
- > Try this method of recomputing an SOC value. With this ratio method, you
- > only need a few good bottle CTD comparisons...as you are only correcting
- > the SLOPE (SOC) in the calibration. I would recommend using ratios made
- > in well mixed regions of the water column.
- >
- > Most people CANNOT get good VOFFSET corrections. In fact, we are
- > advising against that method, and will be updating the application note
- > soon to that effect. The VOFFSET is set by the electronics, and will
- > only change if we change the electronics or the sensor. In our
- > calibration fits, VOFFSET is fit with all the terms in the cal
- > equations, so sometimes, you might see small changes in VOFFSET between
- > factory calcs, even though nothing was done to the sensor. However, most
- > customers do not have enough data to correct VOFFSET and we are finding
- > many are trying to do so with a limited range and number of samples.
- >
- > Ratio method is better for most application, more accurate and much
- > easier...and you can see how good it is by looking at all the ratios
- > throughout your watercolumn...they should all agree fairly well.

## Biogeochemistry section 24.5°N: study of the carbonic system in seawater

In leg number 8 of the Malaspina expedition, around parallel 24.5°N, sea water samples have been taken in order to carry out a detailed study of the CO<sub>2</sub> system throughout the entire section. The samples were collected in the entire water column by means of the use of CTD-rosette and following the established sampling plan.

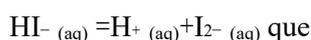
It is contemplated to derive the parameters of interest referring to CO<sub>2</sub> in seawater, including total inorganic carbon (TC), from pH and alkalinity (AT) measurements. The TC estimates obtained from the two variables measured during the cruise will be subjected to an internal consistency analysis, through their comparison with direct measurements of total inorganic carbon. These measurements will be obtained from samples that have also been collected and stored on board, for subsequent analysis on land, at the Marine Research Institute (IIM-CSIC) in Vigo.

The following table shows the number of stations in which each of the variables has been sampled, out of the total of the 167 stations carried out throughout the campaign:

	pH	A <sub>T</sub>	C <sub>T</sub>
Number of stat.	167	67	11

### PH ANALYSIS

The pH has been analyzed by the colorimetric spectrophotometric method (Clayton & Byrne, 1993) which consists of the addition of the m-cresol purple indicator to the seawater sample. The reaction of interest is:



Where I represents the indicator.

The total concentration of hydrogen ions in the sample is determined by the following equation:

$$\text{pH} = \text{p}K[\text{HI}^-] + \log_{10} [\text{I}^{2-}] / [\text{HI}^-]$$

The ratio  $[\text{I}^{2-}] / [\text{HI}^-]$  is calculated considering the Beer-Lambert law and the wavelengths of peak absorbance greater than the spectra of ions. For this, measurements are made at  $\lambda = 434, 578, 730 \text{ nm}$ .

The sample is taken directly from the rosette into special optical glass cuvettes with a volume of 28 mL and a 100 mm light path. Before the measurement, the samples are allowed to stabilize in the incubator at 25°C. Afterwards, 75  $\mu\text{L}$  of the indicator is added and the measurement is taken at the different wavelengths.

### ALCALINITY ( $A_T$ )

Samples have been collected in 600 ml glass bottles of water to determine alkalinity at 67 stations.

Total alkalinity ( $A_T$ ) was determined by automatic potentiometric titration with a hydrochloric acid solution ( $[\text{HCl}] = 0.1\text{M}$ ) at end point 4.4, using an automatic potentiometric titrator "Titrande Metrohm" with an aquatrode plus Pt-1000 (Metrohm 6.0257 .000).

The alkalinity analyzes were carried out in 17 sessions. The electrode was calibrated with a buffer made in seawater (Pérez et al, 2000). A volume of seawater stored in a 75-liter plastic bottle was used as substandard water. In order to verify the accuracy of the measurements, alkalinity determinations were made on  $\text{CO}_2$  reference samples (CRM) from batch 100, supplied by Dr Andrew Dickson from the University of California.

### TOTAL INORGANIC CARBON ( $C_T$ )

Along the 24.5°N parallel,  $C_T$  samples have been taken for subsequent coulumbimetric analysis (Johnson et al, 1993), with the IIM "SOMMA".

$C_T$  sampling is performed in much the same way as  $A_T$  sampling. With the help of a tube, the samples are taken into 600 mL flasks, avoiding the formation of bubbles, and 300  $\mu\text{L}$  of  $\text{HgCl}_2$  (sat) are added to prevent the formation of "fouling".

Regarding  $C_T$  analysis, a known volume of the seawater sample is acidified with  $\text{H}_3\text{PO}_4$  (8.5%) in an "extraction chamber" and the resulting  $\text{CO}_2$  gas is purged using  $\text{N}_2$  as inert gas. The

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purged CO<sub>2</sub> is extracted as much water vapor in a condenser and drying traps before its passage to the coulometric cell, where it reacts to form hydroxyethylcarbamic acid. The equipment determines the end point of the reaction by keeping the transmittance of the solution at a constant value.

As indicated above, the objective of carrying out this analysis is to check, with the CT concentrations obtained, the correlation between the measured data and those calculated from pH and AT.

## REFERENCES

Clayton & Byrne, Spectrophotometric seawater pH measurements: total hydrogen ion concentration scale calibration of m-cresol purple and at-sea results, *Deep-Sea Research* 1, Vol. 40, No. 10, pp. 2115-2129, 1993

Johnson, K. M., Wills K. D., Butler D. B., Johnson W. K., Wong. C. S.,. Coulometric total carbon dioxide analysis for marine studies: maximizing the performance of an automated gas extraction system and coulometric detector. *Marine Chemistry* 17: 1–21, 1993.

Mintrop, L., Pérez F. F., González-Davila M., Santana-Casiano M. J., Kortzinger, A., Alkalinity determination by potentiometry: Intercalibration using three different methods. *Ciencias Marinas* 26 (1): 23-37, 2000.

Pérez, F.F., A.F. Ríos, T. Rellán, M. Alvarez, Improvements in a fast potentiometric seawater alkalinity determination. *Ciencias Marinas*; 26, 463-478, 2000.

## Determination of dissolved oxygen and nutrient.

Methodologies used.

### 1) Dissolved oxygen.

Dissolved oxygen has been determined in all samples obtained by means of the classic Winkler method (1888) with the technical improvements provided by Carpenter (1965) and by Carritt and Carpenter (1966). The determination of the final point has been assessed visually, using a starch indicator.

For the calculation of results, the formula given by Carpenter (1965) modified by Culberson and adopted in this way by the WOCE program (1991) has been used.

### 2) Nutrients

For the determination of nutrients (nitrites, nitrates, silicates and phosphates), the techniques of automatic analysis by segmented continuous flow are used. For this purpose, a 4-channel Technicon-Bran-Luebbe autoanalyzer has been used with the following methodology:

Nitrates and nitrites: method described by Treguer and Le Corre.

Silicates: method described by A. R. Folkard.

Phosphates: United States Environmental Protection Agency (USEPA) method No. 365-5.

For the treatment of the results, a software was coupled to the channels of the autoanalyzer to obtain the data.

References.

Carpenter, J.H., (1965) The accuracy of the Winkler method for dissolved oxygen analysis. *Limnol. Oceanogr.* 10, 135-140.

Carrit, D.E. and Carpenter, J.H., (1966) Comparison and evaluation of currently employed modification of the Winkler method for determining dissolved oxygen in sea-water. A NASCO report. *J. Mar. Res.* 24, 286-318.

Trégner y Le Corze (1975) Manuel d'Utilisation de l'Autanalyseur Technicon AAll. Université de Bretagne Occidentale.

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Folkard, A.R. Automatic Analysis of Sea-water Nutrients Fisheries Research Technical Report N° 46. Ministry of Agriculture Fisheries and Food, Directorate of Fisheries Research. Lowestoft (1978).

Environmental Protection Agency. Method N° 365-5. USRPA 1997.

## PROTOCOL FOR OBTAINING AND PRESERVING SAMPLES OF PARTICULATE ORGANIC MATTER (POC AND PON)

### Material

- 1.- Filtration system consisting of: (i) Vacuum pump (we use an Eyela, which does not need quitasato), (ii) Filtration train, with filter funnels for 25 mm diameter filters (we use a chain manufactured by ourselves, with 25 mm polycarbonate Gelman funnels, to which we have added airtight lids with an upper hole to connect the hose. The hose connects the funnel with the bottle where the sample is. Then, the water passes directly from the bottle to the funnel (once the hose is primed - due to the vacuum generated).
- 2.- Whatman GF / F 25 mm filters. 0.7  $\mu$ m. The filters must be pre-calcined in a Muffle oven at 450° for 12 hours, and stored in a piece of precalcined aluminum foil, taking care not to get contaminated. Aluminum foil squares must also be pre-calcined, which will be used to preserve the filters individually after collecting the samples. Another less economical but better option is to use small Petri dishes to store each filter separately in a box.
- 3.- 15 and 2-liter polyethylene bottles (see Methodology point 5 for the reason for having these two sizes), cleaned with 10% HCl acid and rinsed with distilled and sea water previously (own sample).
- 4.- Silicone hoses for sampling from Niskin bottles to bottles.
- 5.- Large plastic funnel and 0.5 or 1L plastic test tubes (The test tube is used to measure the volume of water in the bottle that has not been filtered and subtract it (in case the filter is clogged by excess MO during the filtration).
- 6.- 3 Filter clips
- 7.- Nitrile / vinyl gloves.

### Sampling, Filtration and Conservation

NOTE: From this step, clean gloves and tweezers should be used during the below process to handle the samples so that they are not contaminated.

- 1.- Before starting with the sampling, check that the filtration funnels and the 4 and 20 liter jugs are well clean.
- 2.- Check that the filtration system works perfectly (pump, trains, etc.).

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- 3.- 10 minutes before uploading the rosette, write down all the data on the chart (longitude, latitude, start time, depths, etc.).
  - 4.- Once the rosette is on the desk and well anchored, place the clean hoses at the outlet of the Niskin bottles, ensuring that the hoses do not touch the cover or any other place to not contaminate the sample.
  - 5.- The Niskin bottles are emptied into the 15-liter jugs, through the hoses, always writing down the number of the jugs and the depth to which they correspond on the chart. It is very important to completely empty the Niskin into the bottle, including the water that remains below the "spigot" (the tap), since the "denser" POC settles quickly and remains concentrated there. This is the reason for emptying all the water that remains (after sampling for other analyzes) into a 15 L jug, even though we will only need 4 L.
  - 6.- When the Niskin bottles are finished emptying and the hoses are removed, stored the bottles in a clean place and closes them to avoid possible contamination.
  - 7.- Shake the 15-liter jugs (gently) to resuspend any precipitate that may have formed and, therefore, homogenize the sample.
  - 8.- Transfer 4 liters from the 15-liter jug to the 4-liter jug with the help of a plastic funnel to avoid wasting the sample volume. Also ensuring that the number of the jugs coincide.
  - 9.- Remove the 25 mm Whatman filters from the stage, which have previously been pre-calcined in the flask, avoiding that they do not touch anything avoiding to contaminate them.
  - 10.- Place the filter in the funnel, making sure to take only one per funnel (\* See comment in point 17) and that the roughest part of the filter is placed face up. Screw the funnel tightly.
  - 11.- Pour the contents of the 4-liter bottle into its corresponding filtering funnel (or, as in our case, connect the bottle with the funnel through the hoses). Connect the pump, open the tap and start filtering.
- NOTE: The funnel should not run out of water at any time during filtration to prevent the filter containing the sample from drying out.
- 12.- Once we have finished filtering the entire volume of water, the filter is left to dry for a few seconds in the funnel (to suck up the water that remains in the filter)
  - 13.- Place the filter on a clean filter paper (absorbent paper sheet). Remove possible contaminating elements from the filter (zooplankton, fluff, etc.) - Fold the filter in half with the tweezers, pressing them on the lower face of the bent filter, so that the paper absorbs the residual water that has the filter.
  - 14.- A piece of pre-calcined platen is removed from the bag, ensuring that the bag containing this material is open for as little time as possible to avoid contamination. The filter is packed in the precalcined platen.
  - 15.- Label the station, cast and depth on the stage, avoiding breaking the stage when marking it (this could contaminate the sample). If you have Petri dishes, label on the lid.

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16.- Introduce all the samples in a -20°C freezer (or in a -80°C freezer) for their conservation.

17.- DOC adsorption: The GF / F filters adsorb DOC and DON during the filtration process (in quantities > 1 µmol / DOC filter), so this quantity must be quantified to subtract it to the amount of organic matter that suits us to give the CHN analyzer when passing the sample in order to obtain the real value of POC and PON (see attached work by Turnewitsh et al. 2007).

## **PROTOCOL FOR COLLECTION AND CONSERVATION OF TOTAL ORGANIC CARBON (TOC) SAMPLES.**

### Necessary material

- Sample collection chart indicating: date of collection, CTD launch time, station number, CTD number, Niskin bottle number, depth of each bottle in db, observations for each bottle and observations for the station
- Polyethylene or nitrile gloves
- 10 ml glass vials sealed with aluminum foil, calcined at 450°C for 24 hours and labels for the identification of the samples.
- Micro-dispenser of phosphoric acid (H<sub>3</sub>PO<sub>4</sub>) 25%
- Plastic racks for transporting the ampoules and collecting samples on the cover
- Butane burner and bottle for heat sealing of the ampoules
- Safety glasses
- 25 cm stainless steel tweezers for heat sealing of the ampoules
- Metal rack to deposit the ampoules after heat sealing
- Zip bags for storing ampoules

### Collection of samples

Before collecting the samples, the chart is filled and each ampoule is labeled with the name of the cruise, the date of collection, the station, the depth and the Niskin bottle number.

Then, using gloves and a laminar flow hood, the samplings are placed in a plastic rack, as many ampoules as depths have been chosen. The aluminum cover is removed and 50 µL of 25% phosphoric acid is added to each one with the micro-dispenser. Once the acid has been added, cover them with a sheet of aluminum foil.

Samples are taken directly from the Niskin bottle corresponding to each ampoule, always wearing gloves, and never rinsing or using tubes. Before filling the ampoule, the Niskin bottle is opened and a little water is allowed to run. The ampoule is then filled to about 5 mm below the cut line. At the end they are covered again with a sheet of aluminum foil.

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After collecting the samples, the ampoules are immediately heat sealed. Sealing must be carried out in a laboratory with a clean atmosphere and free of organic solvents, especially acetone. With the help of stainless steel tweezers, the blisters are sealed to the flame of the lighter. After 15 minutes, the blisters are checked for water leaks.

In the event that any of the ampoules are cracked and lose water, it is broken by the cut line and the sample is transferred to another ampoule, sealing it again.

When all ampoules are properly sealed they are packaged in zip bags layered inside cardboard boxes. Finally, they are stored in a refrigerator at 4°C.

## NITROGEN FIXATION REPORT

*Mar Benavides Gorostegui (ULPGC)*

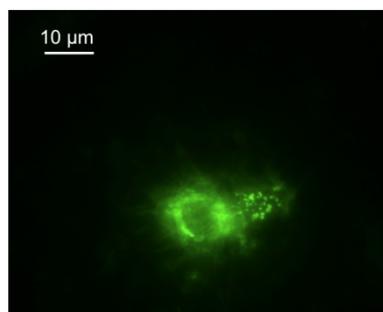
### Introduction

Nitrogen fixation is the process by which atmospheric nitrogen (N<sub>2</sub>) is reduced to ammonia, an essential substrate for primary ocean producers. This pathway for nitrogenous nutrients to the ocean surface is estimated to support up to 50% of primary production in oligotrophic oceans. Therefore, diazotrophic nitrogen fixation plays a key role in the ocean nitrogen cycle, which ultimately directly affects the carbon cycle.

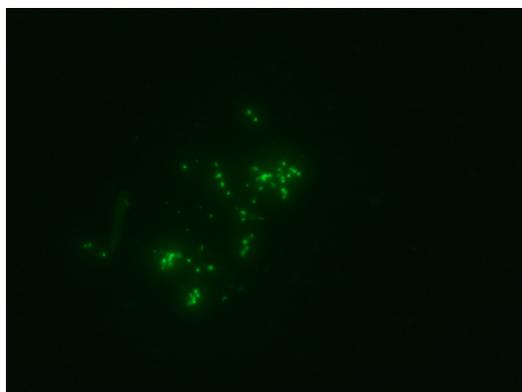
Nitrogen-fixing organisms (or diazotrophs) are primarily cyanobacteria (filamentous, symbiont, or unicellular), but various species of bacteria and archaea are also capable of carrying out this process. At present, the distribution and diazotrophic activity of the colonial cyanobacterium *Trichodesmium*, which is widely distributed throughout the tropical and subtropical areas of the oceans, is known in considerable detail. However, in the last decade, several genetic studies have demonstrated the presence of nitrogen-fixing unicellular cyanobacteria in the Atlantic and Pacific oceans.



Cianobacteria colonial *Trichodesmium thiebautii*



Cianobacterias unicelulares simbiotes  
(TSA-FISH Nitro821)



Cianobacterias unicelulares  
(TSA-FISH Nitro821)

Nitrogen fixation studies have focused mainly on the Northwest Atlantic, where *Trichodesmium* prevails compared to smaller fixatives. In the Northeast Atlantic, the proximity of the Sahara desert and the contributions of atmospheric dust (rich in iron) suggests that the diazotrophic activity and the abundance of *Trichodesmium* should be high. However, the population density of this colonial cyanobacterium is low and the few studies on nitrogen fixation find rates often 1 or 2 orders of magnitude lower than those of the Northwest Atlantic. However, recently, fixation rates associated with diazotrophs <10 µm have been found in waters of the Iberian and African outcrops of a magnitude comparable to those on the eastern flank of the North Atlantic subtropical gyre.

The apparent imbalance in the oceanic nitrogen cycle may be due in large part to a methodological error. Only in the last decade have global biogeochemical models begun to consider nitrogen fixation as a relevant variable. However, the methodologies currently used only consider total fixation (assimilation of nitrogen as biomass) and therefore largely underestimate fixation, since much of the fixed nitrogen is lost as dissolved organic nitrogen (DON) within a few hours after having been fixed. DON release rates in *Trichodesmium* often reach 40-80% of the total set. There are currently no studies that determine the rate of DON release in diazotrophs <10 µm. It is important to consider that this DON feeds not only bacterial production, but also primary production.

Therefore, the main objective of the experiments carried out in this campaign is to determine the relative importance of fixers > 10 µm compared to <10 µm both in their total fixation and in the loss of nitrogen fixed in the form of DON.

**Specific objectives**

- Determine the variability of diazotrophic nitrogen fixation in the subtropical North Atlantic
- Determine the relative importance of the binding activity of colonial cyanobacteria or symbionts (> 10 µm) versus unicellular cyanobacteria and heterotrophic diazotrophs (<10 µm).
- Study the diversity of the diazotrophic community of the subtropical North Atlantic

**Sampled stations**

<b>Station</b>	<b>Date</b>	<b>Latitude</b>	<b>Longitude</b>	<b>Time Niskin 30L</b>
1	28/01/2011	27.78	-13.34	08:25
13	29/01/2011	27.02	-15.70	19:25
17	30/01/2011	26.55	-17.15	16:15
21	31/01/2011	26.02	-18.76	18:18
24	01/02/2011	25.63	-20.00	12:30
28	02/02/2011	24.99	-21.97	22:30

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30	03/02/2011	24.66	-22.98	12:47
34	04/02/2011	24.50	-25.32	18:00
36	05/02/2011	24.50	-26.54	09:15
39	06/02/2011	24.50	-28.37	22:12
41	07/02/2011	24.50	-29.59	18:53
39	06/02/2011	24.50	-28.37	22:12
41	07/02/2011	24.50	-29.59	18:53
44	08/02/2011	24.50	-31.43	21:00
45	09/02/2011	24.50	-32.04	08:50
49	10/02/2011	24.50	-34.33	19:15
52	11/02/2011	24.50	-35.72	16:05
56	12/02/2011	24.50	-37.57	20:00
58	13/02/2011	24.50	-38.49	08:00
62	14/02/2011	24.50	-40.00	21:08
65	15/02/2011	24.50	-41.72	16:15
70	16/02/2011	24.50	-44.04	22:15
71	17/02/2011	24.50	-44.50	13:30
77	18/02/2011	24.50	-0.79	20:10
78	19/02/2011	24.50	-47.73	14:14
83	20/02/2011	24.50	-50.04	21:30
85	21/02/2011	24.50	-50.96	12:15
89	22/02/2011	24.50	-52.81	19:00
91	23/02/2011	24.50	-54.03	11:56
95	24/02/2011	24.50	-56.49	19:56
97	25/02/2011	24.50	-57.72	11:25
102	26/02/2011	24.50	-60.68	22:30
104	27/02/2011	24.50	-62.01	18:00
108	28/02/2011	24.50	-64.46	21:22
110	01/03/2011	24.50	-65.68	17:00
114	02/03/2011	24.50	-68.14	23:30
116	03/03/2011	24.50	-69.13	14:00
121	05/03/2011	26.20	-70.71	11:56
126	06/03/2011	26.20	-72.65	22:30
129	07/03/2011	26.20	-73.82	13:15
135	08/03/2011	26.20	-75.57	21:00
139	09/03/2011	26.20	-76.15	15:10

## **Implemented techniques**

### Fractional nitrogen fixation

Original methodology: Mohr et al., 2010.

- Sample surface water (~ 5 m) with Niskin 30 L
- Transfer 5 L to a carafe using silicone tubes, avoiding the formation of bubbles
- Fraction <10 µm
  - Add 200 mL of filtered seawater enriched with 15N<sub>2</sub> to a 2 L polycarbonate (PC) bottle
  - Pass 2 L of sample through a 10 µm sieve connected to a funnel, in turn placed on the PC bottle
- Fraction > 10 µm fraction
  - Add 200 mL of filtered sea water enriched with 15N<sub>2</sub> to a 2 L PC bottle
  - Carefully recover biological material in the sieve with water sprayer and transfer to the 2 L PC bottle
  - Fill the bottle up to 2 L with filtered sea water
- Incubate for 3-4 h (“on-deck” incubators)
- Filter the contents of each bottle through previously muffled 25 mm GF / F filters, save the filtrate
- Store the GF / Fs in sterile Eppendorf vials at -20°C
- Filter the filtrate through 0.2 µm PC filters
- Store the filtrate in 3 50 mL polypropylene tubes, wrap the cap with Parafilm and freeze (-20°C)

Preparation of seawater enriched in 15N<sub>2</sub>

- Take 1 L of water from the ship's continuous system in a 1 L PC bottle
- Add a magnet (for stirring) and connect to a vacuum system to degas the water
- Close the bottle with a screw cap with built-in septum
- Inject 10 mL of 15N<sub>2</sub> through a sealed syringe, injecting one syringe tip at a time to allow for pressure equilibrium across the septum
- Shake the bottle until the bubble dissolves and place in the on-deck incubator until use.
- Fill 2 capsule vials of 10 mL with the enriched water, fixing it with 100 µl HgCl<sub>2</sub>, encapsulate and store in the dark at room temperature.

### Total nitrogen fixation

Original methodology: Montoya et al., 1996.

- Transfer 2 L of sample (in duplicate) to a 2 L PC bottle with silicone tube avoiding the formation of bubbles
- Inject 2 mL of 15N<sub>2</sub> with a sealed syringe (as in the previous methodology)
- Incubate 24 h
- Filter through muffled GF / F filters
- Store the GF / Fs in sterile Eppendorf vials at -20°C

### DNA

- Transfer 10 L of sample to a carafe
- Connect 3 x 47 mm filter holders with 10, 3 and 0.2 µm PC filters
- Store filters in sterile cryovials
- Store at -80°C

Cytometry to determine the abundance of UCYN

Original methodology: Zehr et al., 2008.

- Sample 1 L of surface water
- Filter it on a 0.2 µm PC filter
- Insert the filter into a 15 mL centrifuge tube with 10 mL of sample
- Vortex for 30 seconds
- Transfer 4.5 mL to 5 mL cryovials (in duplicate)
- Fix with 500 µl of 20% paraformaldehyde
- Freeze with liquid nitrogen
- Store at -80°C

**References**

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Montoya JP, Voss M, Kähler P, Capone D (1996) A simple, high-precision, high-sensitivity tracer assay for N<sub>2</sub> fixation. Appl Environ Microbiol 62:986-993

Zehr JP, Bench SR, Carter BJ, Hewson I, Niazi F, Shi T, Tripp HJ, Affourtit JP (2008) Globally Distributed Uncultivated Oceanic N<sub>2</sub>-Fixing Cyanobacteria Lack Oxygenic Photosystem II. Science 322:1110-1112

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**Malaspina project, Leg 8: Sarmiento de Gamboa**

**Group: Biogeochemistry of stable isotopes, Andalusian Institute of Earth Sciences (CSIC), Granada**

**Main researcher: Antonio Delgado Huertas**

During this leg, it has been done:

- 1- Sampling for DIC
- 2- Primary production experiments

### 1- SAMPLING FOR DIC

All stations (1-167) have been sampled except the following: 2, 9, 23, 29, 61, 74, 148

The samples are taken in 12 ml vials, which already have some grains of mercury chloride in advance, so that the sample is fixed at the moment it is taken. The vials are stored, and once on the ground, they will be analyzed, obtaining the concentration of DIC (dissolved inorganic carbon) and its isotopic ratio.

### 2- PRIMARY PRODUCTION EXPERIMENTS (GPP-<sup>18</sup>O en protocol)

Primary production experiments have been carried out in the deck incubators, in coordination with the experiments of Sofía Sal.

The objective is to calculate the gross primary production by tracing the water with the <sup>18</sup>O isotope.

The process can be summarized as follows:

- The first rosette in the morning is sampled per day taking samples from 3 Niskin bottles: DCM, intermediate depth and surface.
- 6 vials (12 ml each) for each Niskin (i.e. 18 vials per rosette). Of these 6 vials, 3 are fixed with 12 ml of HgCl<sub>2</sub> (for each vial). The other 3 vials are "labeled" with 12 ml of labeled water (H<sub>2</sub><sup>18</sup>O) in each vial.
- The 9 marked vials (3 for each depth) are placed, together with the Sofía winkler bottles, in the 3 deck incubators. They will be removed after 24 hours of incubation, being then fixed with 12 ml of HgCl<sub>2</sub> (for each vial), thus stopping any metabolic process.

Once on land, the gross primary production (from <sup>18</sup>O produced during photosynthesis) will be determined, on the one hand, and its isotopic ratio on the other.

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The stations sampled for this purpose have been:

11-16-20-24-26-36-38-40-43-45-48-51-55-58-64-68-71.1-78.1-82-88-91-94-97-100-103-109-  
112-115-120-124-128-132-137-143-149-154

**METABOLISM, CYTOMETRY AND IMAGE ANALYSIS**

Sofía Sal

The methodology for determining the gross, net and respiration primary production rates of the planktonic community was carried out using the Winkler method.

This achieves an estimate of the variation and biogeography of the metabolism of the pelagic community.

The procedure was as follows: Three samples were chosen in the first rosette in the morning: surface (at 5 m), the depth of the DCM (maximum of chlorophyll) and an intermediate depth that corresponded to the 20% light. Assuming that 1% light reaches the depth of the DCM, and by means of the light attenuation constant  $K_d$ , the intermediate depth was calculated.

For sampling, 21 replicates in 115 ml winkler bottles were taken from each Niskin bottle, 7 to measure the initial oxygen concentration, 7 to measure the final one in light and 7 to measure the final oxygen concentration in the dark. In this way, we can make a balance between production and respiration. After sampling, the initial 7 replicates of each depth were fixed with 1 ml of Manganese Sulfate solution, followed by 1 ml of Iodine Sodium. In this way, the oxygen concentration remains unchanged. These samples were allowed to settle submerged in water and darkness for 6-8 hours or until the precipitate. Meanwhile, the rest of the bottles were incubated in a tank connected to the ship's continuum: the samples corresponding to the shallowest depth were incubated, the 7 clear bottles were incubated uncovered and the 7 dark ones were wrapped in black plastic bags so that did not receive any light. The other two depths, DCM and intermediate, were incubated in methacrylate and PVC tubes, connected to refrigerators that maintained the temperature corresponding to each depth. To simulate the light, filters were placed around the clear tubes, while the samples that were to be kept in the dark were placed in opaque PVC tubes. A Li-cor sensor connected daily during daylight hours recorded the PAR daily. After 24 hours, the samples were taken out of the incubators and fixed in the same way as the initial ones. After sedimentation of the precipitate, the oxygen concentration was measured by means of a Titrand, using 0.01 N Thiosulfate as titrator and analyzing 50 ml aliquots of each sample.

Parallel to the metabolism experiments, a 110 ml sample was fixed with lugol acetic from each depth to subsequently perform phytoplankton image analysis. Likewise, a sample by depth was analyzed by the FacsAria cytometer to perform analysis of the size structure of the phytoplankton, measuring the spectrum within the range of nano- and picoplankton. The same

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replica of each of the samples was fixed with P + G and frozen at -80°C, so that later, and on land, it was possible to carry out cytometric analysis again in case of any error.

### **TOC**

The collection and conservation of water samples was also carried out for the simultaneous analysis of total organic carbon (TOC) and total nitrogen (TN) by the catalytic oxidation method at high temperature, which will be carried out in the base laboratory.

Daily, and in the same rosette that the metabolism samples were taken, 12 depths were chosen, from which 2 replicates of each were taken in glass ampoules to which 50 µL of 25% H<sub>3</sub>PO<sub>4</sub> was previously added (with micro-dispenser). The blisters were covered with a sheet of aluminum foil.

Subsequently, the ampoules were heat sealed and stored in the refrigerator at -4°C.