Supplementary Materials: Detection of a High-Density Brachiolaria-Stage Larval Population of Crown-of-Thorns Sea Star (*Acanthaster planci*) in Sekisei Lagoon (Okinawa, Japan)

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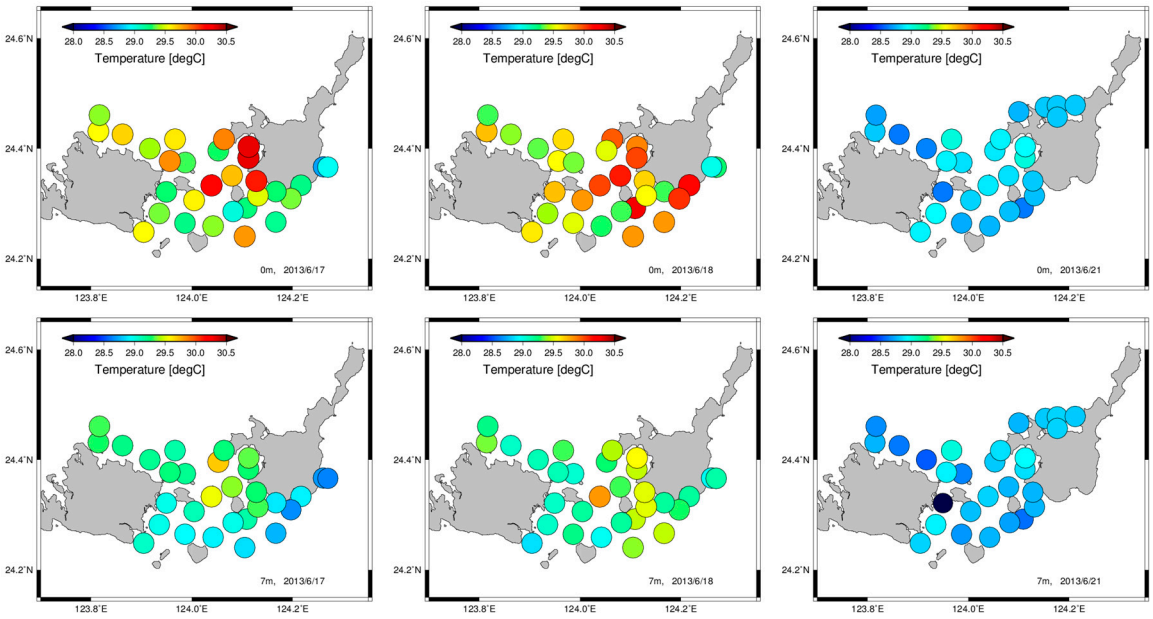
Table S1. Number of larvae isolated under a microscope (n = 225) and larvae identified using polymerase chain reaction (PCR) and sequencing analyses (n = 42 in total).

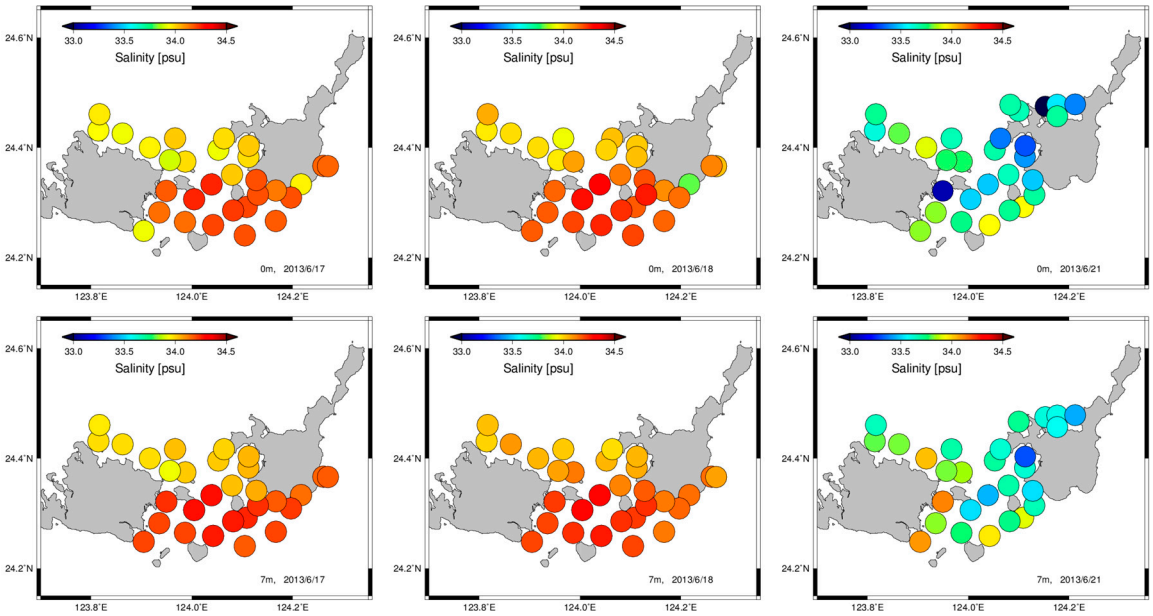
		17-Jun				18-Jun				21-Jun			
	Number of Larvae	First screening	Gastrula	Bipinnaria	Brachiolaria	First screening	Gastrula	Bipinnaria	Brachiolaria	First screening	Gastrula	Bipinnaria	Brachiolaria
SS02	Isolated								6				
	Identified as *A. planci*								0				
SS03	Isolated								9				
	Identified as *A. planci*								0				
SS05	Isolated				2								
	Identified as *A. planci*				0								
SS07	Isolated								5				
	Identified as *A. planci*	○							**1**				
SS10	Isolated												1
	Identified as *A. planci*	○											0
SS11	Isolated												
	Identified as *A. planci*					○							
SS13	Isolated						**3**						
	Identified as *A. planci*						3						
SS15	Isolated				5			1					
	Identified as *A. planci*				**1**			**1**					
SS17	Isolated		3		68		3	3	30			3	7
	Identified as *A. planci*		**1**		**16**		**2**	0	**8**			0	0
SS19	Isolated				2								13
	Identified as *A. planci*				**0**								**2**
SS20	Isolated		1	2	1		3	2	17			3	5
	Identified as *A. planci*		0	**2**	**1**		0	0	0			**1**	0
SS21	Isolated			1	2				2				
	Identified as *A. planci*			0	0	○			0				
SS26	Isolated												
	Identified as *A. planci*												
SS27	Isolated				5								
	Identified as *A. planci*				**0**								
SS28	Isolated										1	1	
	Identified as *A. planci*					○				○	0	**1**	
SS30	Isolated								2				1
	Identified as *A. planci*	○							0				0
SS31	Isolated												1
	Identified as *A. planci*									○			**1**
SS32	Isolated												1
	Identified as *A. planci*												**1**
SS33	Isolated												
	Identified as *A. planci*									○			
SS34	Isolated											1	5
	Identified as *A. planci*									○		0	0
SS35	Isolated											3	1
	Identified as *A. planci*											0	0

Table S2. The most closely related species obtained from larval DNA other than *Acanthaster planci* based on a BLAST search using the partial cytochrome oxidase subunit 1 (CO1) sequence. GS: gastrula, BP: bipinnaria, BR, brachiolaria. These stages were identified using echinoderm universal CO1 primers following the protocol of Arndt *et al*. (1996) [1].

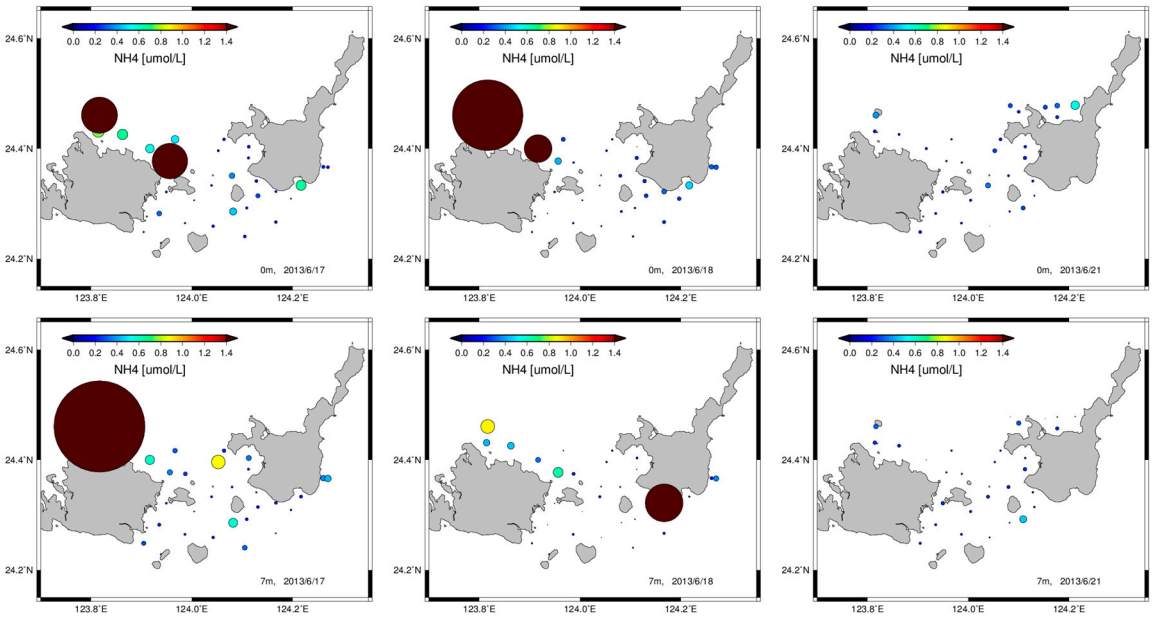
Individual number	Site	Date	Stage	Species	Most similar species	CO1 Query cover(%)	Ident(%)
105	SS17	18-Jun	BR	Culcita novaeguineae	Culcita novaeguineae	100	100
1	SS34	21-Jun	BP	Culcita novaeguineae	Culcita novaeguineae	96	96
141	SS20	18-Jun	BR	Linckia guildingi	Linckia guildingi	85	98
158	SS20	18-Jun	GS	Linckia laevigata	Linckia laevigata	85	100
108	SS17	18-Jun	BR	Mithrodia clavigera	Mithrodia clavigera	91	99
29	SS17	17-Jun	BR	Mithrodia clavigera	Mithrodia clavigera	81	99
39	SS17	17-Jun	BR	Mithrodia clavigera	Mithrodia clavigera	90	99
174	SS27	17-Jun	BR	Mithrodia clavigera	Mithrodia clavigera	84	98
34	SS17	17-Jun	BR	Mithrodia clavigera	Mithrodia clavigera	98	97
12	SS20	17-Jun	BP	Mithrodia sp.	Mithrodia bradleyi	56	92
42	SS17	17-Jun	BR	Mithrodia sp.	Mithrodia clavigera	85	91
182	SS34	21-Jun	BR	unknown	Arbacia lixula	13	91
89	SS17	21-Jun	BP	unknown	Asterinides sp.	84	82
97	SS15	17-Jun	BR	unknown	Choriaster sp.	97	83
99	SS15	17-Jun	BR	unknown	Choriaster sp.	93	80
16	SS17	17-Jun	BR	unknown	Choriaster sp.	93	78
20	SS17	17-Jun	BR	unknown	Choriaster sp.	95	83
23	SS17	17-Jun	BR	unknown	Choriaster sp.	95	82
30	SS17	17-Jun	BR	unknown	Choriaster sp.	94	80
40	SS17	17-Jun	BR	unknown	Choriaster sp.	98	82
46	SS17	17-Jun	BR	unknown	Choriaster sp.	93	80
47	SS17	17-Jun	BR	unknown	Choriaster sp.	94	82
56	SS17	17-Jun	BR	unknown	Choriaster sp.	90	81
57	SS17	17-Jun	BR	unknown	Choriaster sp.	99	82
62	SS17	17-Jun	BR	unknown	Choriaster sp.	49	81
66	SS17	17-Jun	BR	unknown	Choriaster sp.	50	81
70	SS17	17-Jun	BR	unknown	Choriaster sp.	95	79
72	SS17	17-Jun	BR	unknown	Choriaster sp.	89	82
104	SS17	18-Jun	BR	unknown	Choriaster sp.	80	81
110	SS17	18-Jun	BP	unknown	Choriaster sp.	89	81
115	SS17	18-Jun	BR	unknown	Choriaster sp.	60	81
116	SS17	18-Jun	GS	unknown	Choriaster sp.	87	81
122	SS17	18-Jun	BR	unknown	Choriaster sp.	98	82
87	SS17	21-Jun	BR	unknown	Choriaster sp.	72	80
88	SS17	21-Jun	BR	unknown	Choriaster sp.	99	82
90	SS17	21-Jun	BP	unknown	Choriaster sp.	84	80
91	SS17	21-Jun	BR	unknown	Choriaster sp.	99	82
147	SS20	18-Jun	BP	unknown	Choriaster sp.	45	82
148	SS20	18-Jun	BR	unknown	Choriaster sp.	95	82
150	SS20	18-Jun	BR	unknown	Choriaster sp.	93	81
152	SS20	18-Jun	BP	unknown	Choriaster sp.	95	82
153	SS20	18-Jun	BR	unknown	Choriaster sp.	84	80
155	SS20	18-Jun	BR	unknown	Choriaster sp.	96	81
160	SS20	21-Jun	BR	unknown	Choriaster sp.	98	83
162	SS20	21-Jun	BR	unknown	Choriaster sp.	88	81
177	SS21	17-Jun	BP	unknown	Choriaster sp.	88	78
178	SS34	21-Jun	BR	unknown	Choriaster sp.	88	82
179	SS34	21-Jun	BR	unknown	Choriaster sp.	65	81
180	SS34	21-Jun	BP	unknown	Choriaster sp.	90	82
183	SS34	21-Jun	BR	unknown	Choriaster sp.	68	82
44	SS17	17-Jun	BR	unknown	Echinometra sp.	99	81
165	SS20	21-Jun	BR	unknown	Eleutherozoa sp.	99	97
143	SS20	18-Jun	BR	unknown	Heliocidaris erythrogramma	97	76
156	SS20	18-Jun	BR	unknown	Liza argentea	40	80
107	SS17	18-Jun	BR	unknown	Meridiastra calcar	57	82
86	SS17	17-Jun	BR	unknown	Neoferdina cumingi	73	83
96	SS17	21-Jun	BP	unknown	Ophionereis vittata	95	82
168	SS28	21-Jun	GS	unknown	Oreaster reticulatus	90	81
94	SS17	21-Jun	BR	unknown	Temnopleurus reevesii	96	79
35	SS17	17-Jun	BR	unknown	Zoroaster ophiactis	96	80
112	SS17	18-Jun	BP	unknown	Zoroaster ophiactis	80	78
118	SS17	18-Jun	BR	unknown	Zoroaster ophiactis	93	80



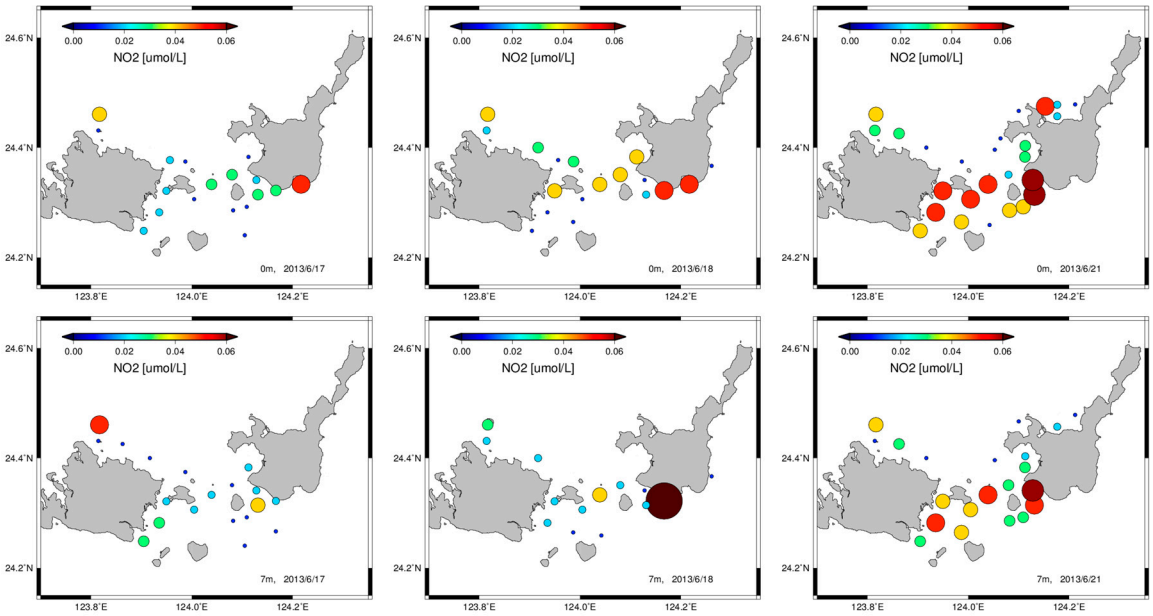
(**a**) Temperature



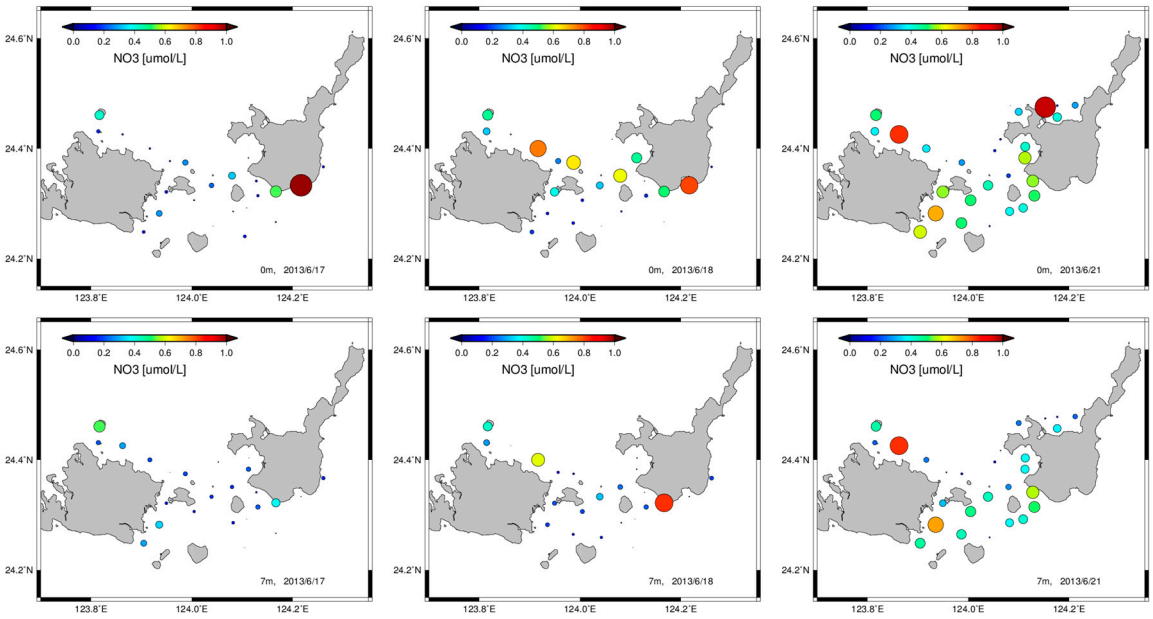
(**b**) Salinity


0.0 0.2 0.4 0.6 0.8 1.0 1.2 1.4
NH4 [umol/L]
0m, 2013/6/17
0m, 2013/6/18
0m, 2013/6/21
7m, 2013/6/17
7m, 2013/6/18
7m, 2013/6/21
24.6'N
24.4'N
24.2'N
123.8'E
124.0'E
124.2'E

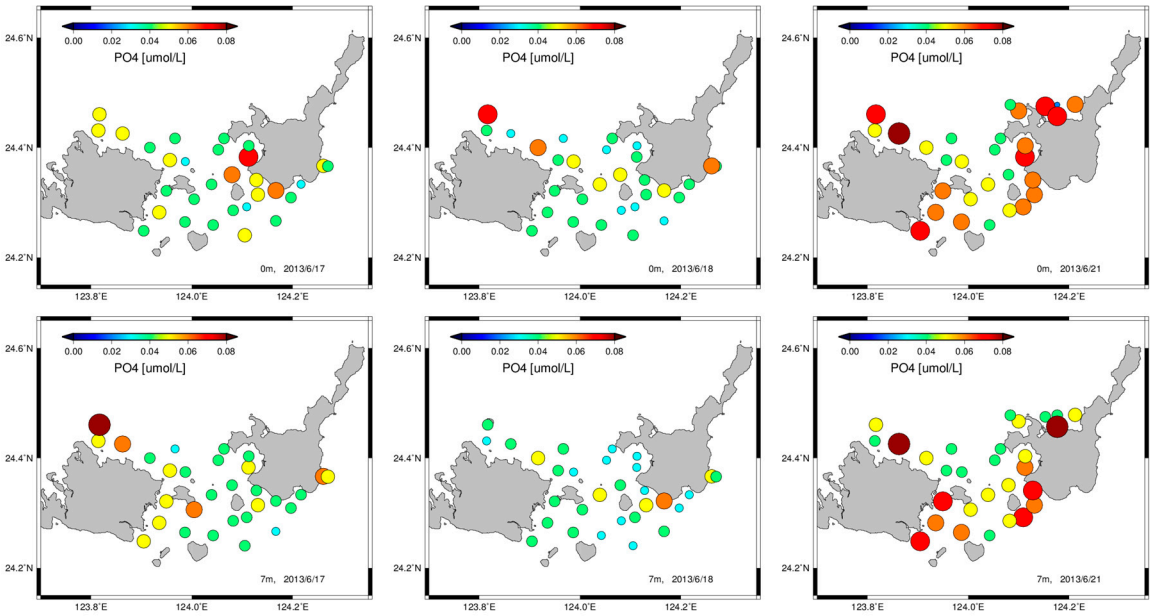
(**c**) NH_4^+


0.00 0.02 0.04 0.06
NO2 [umol/L]
0m, 2013/6/17
0m, 2013/6/18
0m, 2013/6/21
7m, 2013/6/17
7m, 2013/6/18
7m, 2013/6/21
24.6'N
24.4'N
24.2'N
123.8'E
124.0'E
124.2'E

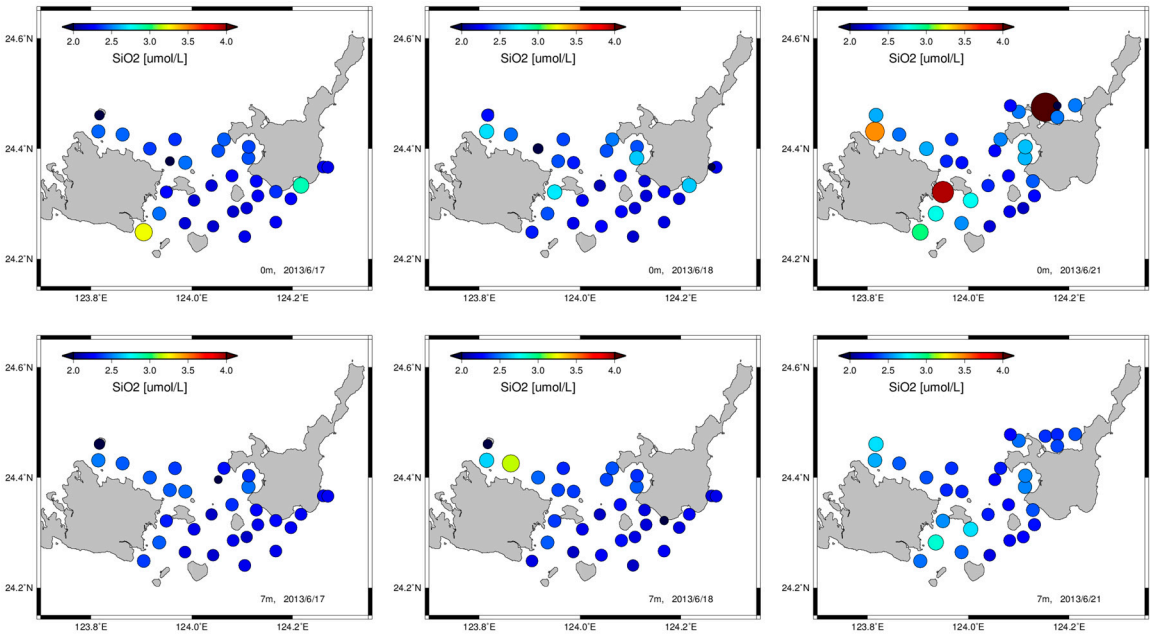
(**d**) NO_2^-


0.0 0.2 0.4 0.6 0.8 1.0
NO3 [umol/L]
24.6°N
24.4°N
24.2°N
123.8°E
124.0°E
124.2°E
0m, 2013/6/17
0m, 2013/6/18
0m, 2013/6/21
7m, 2013/6/17
7m, 2013/6/18
7m, 2013/6/21

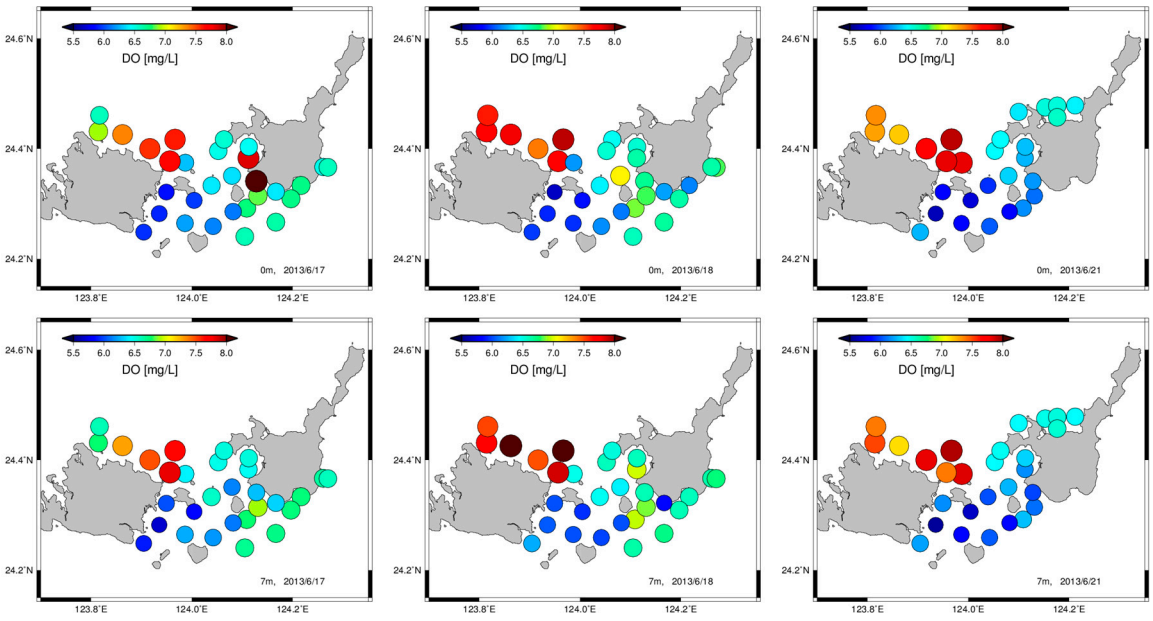
(**e**) NO_3^-


0.00 0.02 0.04 0.06 0.08
PO4 [umol/L]
24.6°N
24.4°N
24.2°N
123.8°E
124.0°E
124.2°E
0m, 2013/6/17
0m, 2013/6/18
0m, 2013/6/21
7m, 2013/6/17
7m, 2013/6/18
7m, 2013/6/21

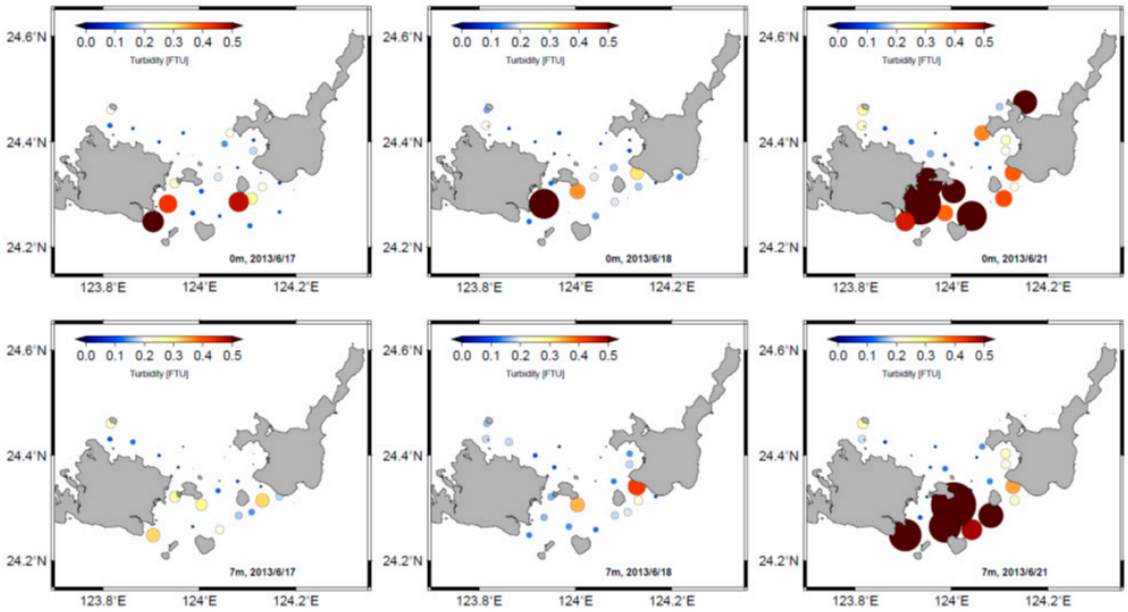
(**f**) PO_4^{3-}


2.0 2.5 3.0 3.5 4.0
SiO2 [umol/L]
24.6°N
24.4°N
24.2°N
123.8°E
124.0°E
124.2°E
0m, 2013/6/17
0m, 2013/6/18
0m, 2013/6/21
7m, 2013/6/17
7m, 2013/6/18
7m, 2013/6/21

(**g**) SiO_2


5.5 6.0 6.5 7.0 7.5 8.0
DO [mg/L]
24.6°N
24.4°N
24.2°N
123.8°E
124.0°E
124.2°E
0m, 2013/6/17
0m, 2013/6/18
0m, 2013/6/21
7m, 2013/6/17
7m, 2013/6/18
7m, 2013/6/21

(**h**) DO



(**i**) Turbidity

Figure S1. Distributions of temperature (**a**); salinity (**b**); nutrient concentrations (**c**–**g**); NH_4^+, NO_2^-, NO_3^-, PO_4^{3-}, and SiO_2); (**h**) dissolved oxygen (DO); and (**i**) turbidity at the surface and at a depth of 7 m in Sekisei Lagoon during 3 days of sampling.

References

1. Arndt, A.; Marquez, C.; Lambert, P.; Smith, M.J. Molecular phylogeny of Eastern Pacific sea cucumbers (Echinodermata: Holothuroidea) based on mitochondrial DNA sequence. *Mol. Phylogenet. Evol*. **1996**, *6*, 425–437.