

## BIOASSAY DATA FOR MARINE POLLUTION USING SEA URCHIN EGGS, 1975

In 1975, six experiments for biological assay were made with sea urchin eggs to measure marine pollution around the Seto Marine Biological Laboratory.

I. Winter season, February 17. Eggs of *Hemicentrotus pulcherrimus* (A. Agassiz) were used, see Table 1.

II. Spring season, two experiments were made in March–May using *Hemicentrotus pulcherrimus* or *Anthocidaris crassispina* (A. Agassiz) eggs.

1. March 18, see Table 2.      2. May 23, see Table 3.

III. Summer season, two experiments were made in July–September, using *Anthocidaris crassispina* eggs.

1. July 28, see Table 4.      2. September 6, see Table 5.

IV. Autumn season, December 6. Eggs of *Pseudocentrotus depressus* (A. Agassiz) were used, see Table 6.

(Notes common to all tables: Fertilization membrane formation examined 3 min. after insemination; minutes and hours in parentheses respectively after First cleavage and Gastrulation indicate the time after insemination; the maturation state of gonads used was nearly ripe to full ripe; Degree of inhibitory effect 0 shows no inhibition, 1 a slight inhibition, 2 a weak and 3 a moderate inhibition by the sea water tested (see Publ. Seto Mar. Biol. Lab., XXI (5/6), p. 391, Table 8, 1974).

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Table 1. Results of the Feb. 17 experiment with eggs of *Hemicentrotus pulcherrimus*.  
Wind; NW1. Test water temperature; 20°C (warmed). 6 hrs. old eggs.

Location (depth)	Fertiliz.	First cleavage (90 min.)			Gastrulation (24 hrs.)			Other notes	Degree of inhibi- tory effect
	mem- brane for- mation	1 cell	2 cell (nor- mal)	multi- cell (poly- spermy)	perma- nent blastula	gastrula (nor- mal)	exo- gastrula	ab- normal develop.	
(m)	%	%	%	%	%	%	%		
Running sea water of Laboratory	99.5	1.0	99.0	0	0	100	0		0
	99.0	2.0	98.0	0	0.5	99.5	0		
Water from open sea side of Hatakejima Surface	99.5	0.5	99.5	0	0	100	0		0
	99.0	1.0	99.0	0	0	100	0		
Water from land side of Hatakejima Surface	98.5	2.5	97.5	0	2.5	97.5	0		0
	96.5	4.0	96.0	0	4.5	95.5	0		
Bottom (27)	95.0	5.0	95.0	0	4.0	96.0	0		1
	94.0	6.5	93.5	0	5.5	94.5	0		
Sea water from Tsuna- shirazu cove Surface	95.5	5.5	94.5	0	5.0	95.0	0		1
	94.0	7.5	92.5	0	6.5	93.5	0		
Bottom (5)	94.0	7.0	93.0	0	6.0	94.0	0		1
	92.5	8.0	92.0	0	7.5	92.5	0		

Table 2. Results of the Mar. 18 experiment with eggs of *Hemicentrotus pulcherrimus*.  
Wind; NW1. Test water temperature; 13°C. 8 hrs. old eggs.

Location (depth)	Fertiliz.	First cleavage (120 min.)			Gastrulation (24 hrs.)			Other notes	Degree of inhibi- tory effect
	mem- brane for- mation	1 cell	2 cell (nor- mal)	multi- cell (poly- spermy)	perma- nent blastula	gastrula (nor- mal)	exo- gastrula	ab- normal develop.	
(m)	%	%	%	%	%	%	%		
Running sea water of Laboratory	99.0	1.5	98.5	0					0
	98.5	3.0	97.0	0	1.0	99.0	0		
	99.5	1.5	98.5	0					
Water from open sea side of Hatakejima Surface	98.0	3.0	97.0	0					0
	96.5	4.5	95.5	0	1.5	98.5	0		
	96.0	7.0	93.0	0					
Water from land side of Hatakejima Surface	91.5	11.5	88.5	0					1
	88.0	15.5	84.5	0	7.5	92.5	0		
	86.5	16.5	83.5	0					
Bottom (27)	84.3	22.0	78.0	0					2
	87.5	18.5	81.5	0	13.0	87.0	0		
	84.0	21.0	79.0	0					
Sea water from Tsunashirazu cove Surface	91.0	11.5	88.5	0					1
	89.5	13.5	86.5	0	6.5	93.5	0		
	86.5	15.0	85.0	0					
Bottom (5)	83.5	20.5	79.5	0					2
	86.0	18.5	81.5	0	14.0	86.0	0		
	81.5	23.5	76.5	0					

Table 3. Results of the May 23 experiment with eggs of *Anthocidaris crassispinata*.  
Wind; O. Test water temperature; 21°C. 6 hrs. old eggs.

Location (depth)	Fertiliz.	First cleavage (65 min.)			Gastrulation (24 hrs.)			Other notes	Degree of inhibi- tory effect
	mem- brane for- mation	1 cell	2 cell (nor- mal)	multi- cell (poly- spermy)	perma- nent blastula	gastrula (nor- mal)	exo- gastrula	ab- normal develop.	
(m)	%	%	%	%	%	%	%		
Running sea water of Laboratory	99.5	1.0	99.0	0	0.5	99.5	0		0
	98.0	2.5	97.5	0	1.0	99.0	0		
Water from open sea side of Hatakejima Surface	99.0	1.0	99.0	0	0	100	0		0
	98.5	2.0	98.0	0	0.5	99.5	0		
Water from land side of Hatakejima Surface	98.0	3.0	97.0	0	4.0	96.0	0		1
	97.5	4.0	96.0	0	5.5	94.5	0		
Bottom (27)	97.5	3.0	97.0	0	9.0	91.0	0		1
	96.5	3.5	96.5	0	10.5	89.5	0		
Sea water from Tsuna- shirazu cove Surface	94.5	6.0	94.0	0	7.0	93.0	0		1
	94.0	7.0	93.0	0	8.5	91.5	0		
Bottom (5)	94.0	7.0	93.0	0	9.0	91.0	0		2
	91.5	9.0	91.0	0	11.5	88.5	0		

Table 4. Results of the July 28 experiment with eggs of *Anthocidaris crassispina*.  
Wind; SW1. Test water temperature; 28°C. 3 hrs. old eggs.

Location (depth)	Fertiliz.	First cleavage (50 min.)			Gastrulation (15 hrs.)			Other notes	Degree of inhibi- tory effect
	mem- brane for- mation	1 cell	2 cell (nor- mal)	multi- cell (poly- spermy)	perma- nent blastula	gastrula (nor- mal)	exo- gastrula	ab- normal develop.	
(m)	%	%	%	%	%	%	%		
Running sea water of Laboratory	99.0	2.5	97.0	0.5					0
	97.5	4.0	95.0	1.0	0.5	99.5	0		
	98.0	2.5	97.0	0.5					
Water from open sea side of Hatakejima Surface	98.0	4.0	94.5	1.5					0
	91.5	8.0	90.5	1.5	1.0	99.0	0		
	96.5	6.0	92.0	2.0					
Bottom (25)	97.5	5.0	93.0	2.0					0
	91.5	8.0	90.5	1.5	1.5	98.5	0		
	92.0	8.0	90.0	2.0					
Water from land side of Hatakejima Surface	88.0	12.0	86.5	1.5					1
	86.0	16.5	81.5	2.0	5.0	95.0	0		
	87.5	14.0	82.5	3.5					
Bottom (27)	85.0	15.5	80.5	4.0					2
	83.5	17.5	76.0	6.5	13.0	87.0	0	slightly delay	
	83.0	17.0	78.5	4.5					
Sea water from Tsuna- shirazu cove Surface	81.0	27.0	70.5	2.5					2
	80.0	25.5	70.0	4.5	5.5	94.5	0		
	82.0	19.5	75.5	5.0					
Bottom (5)	78.5	30.5	64.5	5.0					3
	74.0	30.5	63.0	6.5	12.5	87.5	0	slightly delay	
	78.0	23.5	68.5	8.0					

Table 5. Results of the Sept. 6 experiment with eggs of *Anthocardia crassispina*.  
Wind; O. Test water temperature; 27°C. 3 hrs. old eggs.

Location (depth)	Fertiliz.	First cleavage (50 min.)			Gastrulation (15 hrs.)			Other notes	Degree of inhibi- tory effect
	mem- brane for- mation	1 cell	2 cell (nor- mal)	multi- cell (poly- spermy)	perma- nent blastula	gastrula (nor- mal)	exo- gastrula	ab- normal develop.	
(m)	%	%	%	%	%	%	%		
Running sea water of Laboratory	98.0	3.0	97.0	0					
	97.0	3.5	96.5	0	0.5	99.5	0	0	
	98.5	3.0	97.0	0					
Water from open sea side of Hatakejima Surface	99.0	2.0	98.0	0					
	97.5	3.0	97.0	0	0.5	99.5	0	0	
	98.0	2.5	97.5	0					
Water from land side of Hatakejima Surface	98.0	3.0	97.0	0					
	96.5	4.5	95.5	0	1.5	98.5	0	0	
	98.0	4.0	96.0	0					
Bottom (27)	97.0	6.0	94.0	0					
	95.0	6.5	93.5	0	5.5	94.5	0	1	
	96.5	7.0	93.0	0					
Sea water from Tsun- shirazu cove Surface	96.0	5.0	95.0	0					
	94.5	7.5	92.5	0	6.5	93.5	0	1	
	90.5	10.0	90.0	0					
Bottom (5)	86.0	15.0	85.0	0					
	87.0	14.0	86.0	0	11.5	88.5	0	2	
	85.0	17.0	83.0	0					

Table 6. Results of the Dec. 6 experiment with eggs of *Pseudocentrotus depressus*.  
Wind; O. Test water temperature; 14°C. 8 hrs. old eggs.

Location (depth)	Fertiliz.	First cleavage (120 min.)			Gastrulation (24 hrs.)			Other notes	Degree of inhibi- tory effect
	mem- brane for- mation	1 cell	2 cell (nor- mal)	multi- cell (poly- spermy)	perma- nent blastula	gastrula (nor- mal)	exo- gastrula	ab- normal develop.	
(m)	%	%	%	%	%	%	%		
Running sea water of Laboratory	98.5	2.5	97.5	0	0.5	99.5	0		0
	99.0	1.5	98.5	0					
Water from open sea side of Hatakejima Surface	99.0	1.5	98.5	0	1.0	99.0	0		0
	99.5	1.0	99.0	0					
Water from land side of Hatakejima Surface	98.0	3.0	97.0	0	1.5	98.5	0		0
	98.5	2.5	97.5	0					
Bottom (27)	96.0	5.0	95.0	0	1.5	98.5	0		0
	97.0	4.0	96.0	0					
Sea water from Tsuna- shirazu cove Surface	95.5	6.0	94.0	0	3.0	97.0	0		0
	96.5	6.5	93.5	0					
Bottom (5)	94.5	8.0	92.0	0	5.5	94.5	0		1
	95.0	7.0	93.0	0					