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A review

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Effects of ocean acidification on the settlement and metamorphosis of marine invertebrate and fish larvae: a review

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Table S1 Field studies that explore aspects of the effects of elevated *p*CO_2 on settlement (or recruitment) of marine organisms at naturally occurring CO_2 vents. The table summarises the information available on the organism studied, the location of the CO_2 vent site and the different *p*CO_2 levels present, as well as the effects shown by each study. The table comprises information on different taxa of invertebrates, algae and bacterial communities. Although this review manuscript focuses on marine (invertebrate and fish) larvae, this table also includes other organisms that have previously been studied at the CO_2 vents.

Phylum, species	Vent site	*p*CO_2 levels	Effects found	References
Different taxa, benthic invertebrates and microfauna	Ischia, Italy	- Stations N1 & S1, normal pH conditions (8.06 - 8.15) - Stations N2 and S2, intermediate pH conditions with high pH fluctuations: range 7.27–7.99 and 7.49–7.89, - Stations N3 and S3, low pH conditions: range 7.26–7.60 and 7.08–7.79	Calcareous foraminiferans, serpulid polychaetes, gastropods and bivalves showed highly significant reductions in recruitment with increase in *p*CO_2	Cigliano et al. 2010
			Only one species of polychaete had higher abundances at the station presenting the lowest pH values, although a wide range of polychaetes and small crustaceans was able to settle and survive under these conditions.	
			A few taxa were particularly abundant at stations presenting intermediate amounts of *p*CO_2	

Phylum, species	Vent site	pCO_2 levels	Effects found	References
Algae	Ischia, Italy	- Normal pH = Sn (pH_T 8.06 ± SD 0.09) - Normal pH=Nn (pH_T 7.95 ± SD 0.06) - Medium pH = Sm (pH_T 7.75 ± SD 0.3) - Medium pH = Nm (pH_T 7.77 ± SD 0.19) - Low pH = Sl (pH_T 6.59 ± SD 0.51) - Low pH = Nl (pH_T 7.20 ± SD 0.36)	Decrease in number of species recorded in reduced seawater pH stations	Porzio et al. 2013
Benthic invertebrates (polychaetes, amphipod, isopod and tanaid crustaceans and molluscs)	Ischia, Italy	- Stations N1 & S1, normal pH conditions: 8.06 – 8.15 - Stations N2 & S2, intermediate pH conditions with high pH fluctuations: 7.27–7.99 and 7.49–7.89, - Stations N3 & S3, low pH conditions: 7.26–7.60 and 7.08–7.79	- No significant differences in # of organisms settled - Significant differences in diversity - Normal pH stations were significantly more diverse	Ricevuto et al. 2012
Microalgal assemblages (periphyton)	Vulcano Island, Italy	- Ambient pH (mean 8.18) - Intermediate pH (mean 8.05) - Low pH (mean 7.49)	- Significant changes in periphyton communities - Increase in chl a concentration and in diatom abundance with increasing pCO_2 - No change in cyanobacteria	Johnson et al. 2013
Benthic polychaete species: *Amphiglena mediterranea, Platynereis dumerilii, Syllis prolifera*	Ischia, Italy	- Control N1 = pH 8.0 ± 0.1 - Control S1 = pH 8.1 ± 0.1 - Intermediate N2 = pH 7.8 ± 0.2 - Intermediate S2 = pH 7.8 ± 0.3 - Acidified N3 = pH 7.2 ± 0.4 - Acidifed S3 = pH 6.6 ± 0.5	Significant increase in abundance of the three target species in reduced seawater pH conditions	Ricevuto et al. 2014
Bacterial and archaeal communities	Papua New Guinea	- low pCO_2: pH_T 7.97–8.14, 296–494ppm pCO_2 - high pCO_2: pH_T 7.73–8.00, 444–953ppm pCO_2	- Increased microbial richness with increase in pCO_2 - Shift in microbial composition along the natural gradient	Raulf et al. 2015

Phylum, species	**Vent site**	***p*CO_2 levels**	**Effects found**	**References**
Microborers (cyanobacteria, chlorophytes, rhodophytes and fungi)	Maug, Commonwealth of the Northern Mariana Islands	- High *p*CO_2 site: mean pH 7.94±0.051 - Mid- *p*CO_2 site: mean pH 7.98 ± 0.027 - Control *p*CO_2 site: mean pH 8.04 ± 0.016	Increased colonization (settlement) of microbores at lower pH (higher *p*CO_2)	Enochs et al. 2016
Crustose Coralline Algae	Papua New Guinea	- Control sites: pH_T 8.02 - 7.98 / *p*CO_2 346 - 413 µatm - High CO_2 sites: pH_T 7.95 - 7.72/ 441 - 998 µatm	Total CCA cover decreased with increase in *p*CO_2	Fabricius et al. 2015
Vermetids (gastropoda)	Vulcano Island, Italy	- Low pH: ~7.31 - Mid pH: ~ 7.73 - High pH: ~ 8.03 - CTL_Vent: ~ 8.15	Recruitment success adversely affected at the Low and the Mid pH sites	Milazzo et al. 2014
Tropical coral recruits	Papua New Guinea	- Low pH: ~ 7.8 - High pH: ~ 8.0	- Reduced coral settlement and recruitment in presence of substrates (CCA) pre-conditioned at low pH. - Lower settlement associated with reduced CCA cover at reduced pH site.	Fabricius et al. 2017
Calcareous species of invertebrates Fleshy seaweeds	Ischia, Italy	- Control $pH_{north,south}$ = 8.0, 8.1 - Low $pH_{north,south}$ = 7.8, 7.8 - Extreme low pH < 7.4	- Early successional stages: Similar rates of recruitment of calcareous species at both ambient and low pH sites - Later succession: Fleshy seaweeds in low pH conditions	Kroeker et al. 2013a
Benthic rocky reef assemblages	Ischia, Italy	- Control $pH_{north,south}$ = 8.0, 8.1 - Low $pH_{north,south}$ = 7.8, 7.8 - Extreme low pH < 7.4	- Significantly different communities among pH zones after 32 months: - dominance of calcareous algae in ambient pH; - dominance of fleshy algae in low pH zones; - dominance of biofilm/filamentous algae and erect fleshy algae in extreme low pH zones;	Kroeker et al. 2013b

Phylum, species	**Vent site**	***p*CO_2 levels**	**Effects found**	**References**
			- greater variability in the communities in the ambient and extreme low pH zones.	
Crustose coralline algae	Ischia, Italy	- Control $pH_{north,south}$ = 8.0, 8.1 - Low $pH_{north,south}$ = 7.8, 7.8 - Extreme low pH < 7.4	- Lower CCA abundance in the low pH - CCA settled in extreme low pH were significantly smaller and presented altered mineralogy	Kamenos et al. 2016

Table S2: Single stressor laboratory studies that explore ocean acidification induced changes in larval settlement for different taxonomic groups. The table includes information on the pH treatments or pCO_2 levels tested, the effects found and the biogeographical region of origin of the species tested.

Phylum, species	**pH or pCO_2 range tested**	**Effects found**	**References**	**Region**
Cnidaria				
Acropora digitifera	- pH_T 8.0 - pH_T 7.6 - pH_T 7.3	Significant decline in metamorphosis rate under reduced seawater pH conditions after both short (2 h) and long (7 d) term exposure	Nakamura et al. 2011	Japan
Acropora gemmifera	- pH 8.1 - pH 7.8 - pH 7.5	Reduced settlement percentage in lower pH treatments	Yuan et al. 2018	China
Acropora millepora	- control (pH 8.04, 401 µatm CO_2) - Future CO_2 concentrations #1 (pH 7.79, 807 µatm) - Future CO_2 concentrations #2 (pH 7.60, 1299 µatm)	Negative effects on settlement due to increased pCO_2	Doropoulos et al. 2012	Australia
Acropora palmata	- average ambient pCO_2 conditions (~400 µatm) - Mid-CO_2 conditions (~560 µatm) - High-CO_2 conditions (~800 µatm)	Settlement was negatively impacted by increasing pCO_2	Albright et al. 2010	Florida, USA

Phylum, species	pH or *p*CO_2 range tested	Effects found	References	Region
Acropora selago	- Ambient (pH 7.98, 447 µatm CO_2) - Mid elevated treatment (pH 7.81, 705 µatm CO_2) - High elevated treatment (pH 7.60, 1214 µatm CO_2)	Negative effects on settlement. Settlement decreased with increase in *p*$CO_2$for all CCA taxa.	Doropoulos & Diaz-Pulido 2013	Australia
Acropora spicifera	- Control *p*CO_2: ± 250 uatm - High *p*CO_2: + 900 uatm	No significant effects on settlement	Foster et al. 2015	Australia
Acropora tenuis	- Control pH - Reduced pH 7.6 (1000 µatm CO_2)	No effects on settlement	Kurihara 2008	Japan
Acropora millepora	*Experiment 1:* Larval settlement on pre conditioned CCA (pH 8.1, pH 7.9, pH 7.7 and pH 7.5) pH treatments: 8.1, 7.9, 7.7 and 7.5	Reduced settlement on CCA pre-exposed to different pH treatments	Webster et al. 2013b	Australia
	Experiment 2: Larval settlement on conditioned CCA at ambient pH (8.1) pH treatments: 8.1, 7.9, 7.7 and 7.5	No significant effects on settlement		
Acropora millepora & Acropora tenuis	*Experiment 3:* Larval settlement on CCA extract. Larvae preconditioned for 24h in different treatments (pH 8.1, pH 7.9, pH 7.7 and pH 7.5) Experimental pH treatments: 8.1, 7.9, 7.7 and 7.5	No significant effects on settlement		
Pocillopora damicornis	- Control pH = 8.1 - Treatment 1: pH = 7.9 - Treatment 2: pH = 7.6	Decrease in pH caused a strong decline in larval settlement rates, with the lowest rate at pH 7.6. At pH 7.9 and 7.6 all larvae were unable to complete	Viyakarn et al. 2015	Thailand

Phylum, species	pH or pCO_2 range tested	Effects found	References	Region
		metamorphosis		
Porites asteroides	- pH 8.1 - pH 7.6	No significant effects of reduced pH on coral settlement	Olsen et al. 2015	Florida, USA
Porites asteroides	- present pCO_2 conditions (380 ppm) - projected pCO_2 for the year 2065 (560 ppm) - projected pCO_2 for the year 2100 (720 ppm)	No direct correlation between increase of pCO_2 and decrease on settlement	Albright et al. 2008	Florida, USA
Porites astreoides	- ambient pCO_2 conditions (380 µatm) - middle century predicted pCO_2 (560 µatm) - end of century predicted pCO_2 (800 µatm)	- Significant effects on settlement, but only in tiles preconditioned in high pCO_2 seawater conditions - indirect effects by altering settlement substrate	Albright & Langdon 2011	Florida, USA
Annelida				
Hydroides elegans	- Control: pH_{NBS} 8.17 - Reduced: pH_{NBS} 7.56	No effects on settlement and metamorphosis	Lane et al. 2013	Hong Kong
Mollusca				

Phylum, species	**pH or pCO_2 range tested**	**Effects found**	**References**	**Region**
Gastropoda				
Haliotis diversicolor and *H. discus hannai*	- Control pCO_2: 447 µatm ~ pH_{NBS} 8.15 - High pCO_2: 1500 µatm ~ pH_{NBS} 7.7 2000 µatm ~ pH_{NBS} 7.6 3000 µatm ~ pH_{NBS} 7.4	Reduced survival and metamorphosis	Guo et al. 2015	China
Crepidula fornicata	- pH_T 7.51 - pH_T 7.71 - pH_T 7.96	Significantly higher settlement and metamorphosis at lower pH	Dooley & Pires 2015	Washington, USA
Bivalvia				
Macoma balthica	- Ambient pH_T1 = 7.94 - Ambient pH_T2 = 7.94 - pH_T3 = 7.80 - pH_T4 = 7.51 - pH_T5 = 7.43	Negative effects on settlement due to reduced pH	Jansson et al. 2016	Baltic Sea
Bryozoa				
Bugula neritina	Range pH treatments (8.0 to 6.5)	- Time to settle increased with reduction in pH. - No lethal effects showed at lower pH.	Pecquet et al. 2017	Hong Kong
Arthropoda				
Maxillopoda				
Amphibalanus amphitrite	- Ambient pH 8.2 - Low pH 7.4	No influence on settlement due to reduced pH	McDonald et al. 2009	North Carolina, USA

Phylum, species	pH or pCO_2 range tested	Effects found	References	Region
Balanus amphitrite	- Ambient pH: 8.2 - Low pH: 7.6	No influence on settlement due to reduced pH	Campanati 2016	Hong Kong
Malacostraca				
Stenopus hispidus	- 400 ppm CO_2 (current-day control) - 700 ppm CO_2 - 850 ppm CO_2	Significant negative effects in ability to recognize conspecific cues important for settlement	Lecchini et al. 2017	French Polynesia Japan
Echinodermata				
Echinoidea				
Paracentrotus lividus	- Present average pH 8.1 - Average predicted for 2100, pH 7.7 - Extreme predicted for 2100, pH 7.4	- Delay in larval settlement at pH 7.7 - No successful settlement at pH 7.4	García et al. 2015	Mediterranean and NE Atlantic Ocean
Strongylocentrotus droebachiensis	- pH_T = 8.07 (~ 361 μatm CO_2) - pH_T = 7.69 (~ 942 μatm CO_2)	No significant effect of pCO_2 in larval settlement	Dupont et al. 2013	Boreal coastal ecosystems
Asteroidea				

Phylum, species	pH or pCO_2 range tested	Effects found	References	Region
Acanthaster cf. solaris	- Pre-industrial pH_{NBS} 8.25 target - Present conditions pH_{NBS} 8.1 target - Future medium conditions pH_{NBS} 8.0 target - Future low conditions pH_{NBS} 7.9 target	- Significant negative effects on settlement when settlement substrates were previously conditionned in the different pH treatments. - No significant effects on settlement only due to water chemistry (settlement experiment 2)	Uthicke et al. 2013	Australia
Fish				
Amphiprion percula	- Control pH 8.15 - Predictions for the year 2100: pH 7.8 - Low pH 7.6	Significant reduction in homing behaviour due to loss in olfactory capacity	Munday et al. 2009	Australia
Chromis viridis	- 400 ppm CO_2 (current-day control) - 700 ppm CO_2 - 850 ppm CO_2	Significant negative effects in ability to recognize conspecific cues important for settlement	Lecchini et al. 2017	French Polynesia Japan

Phylum, species	**pH or pCO_2 range tested**	**Effects found**	**References**	**Region**
Lates calcarifer	pH 8.19 ~ 400 μatm CO_2 pH 7.70 ~1675 μatm CO_2	Negative effects of elevated pCO_2 in the auditory preference of the fish at time of settlement	(Rossi et al. 2015)	Tropical areas between eastern Indian Ocean to the western Central Pacific
Lates calcarifer	- pH 8.13 ~ 465 μatm CO_2 - pH 7.70 ~1477 μatm CO_2	Negative effects of reduced pH on attraction by larval fish towards physico-chemical cues for settlement	Pistevos et al. 2017	Tropical areas between eastern Indian Ocean to the western Central Pacific
Pomacentrus amboinensis	- 440 ppm CO_2 (current-day control) - 700 ppm CO_2 - 850 ppm CO_2	Impairement in olfactory discrimination of settlement cues	Devine et al. 2012	Australia

Phylum, species	**pH or pCO_2 range tested**	**Effects found**	**References**	**Region**
Pomatoschistus pictus	- pH 8.06 ~ 530 μatm CO_2 - pH 7.66 ~ 1500 μatm CO_2	Negative effects of high CO_2 in auditory responses important for settlement	Castro et al. 2017	Eastern Atlantic Ocean and Mediterranean Sea

Table S3: Multiple stressor laboratory studies that explore changes in settlement caused by ocean acidification (OA) in conjunction with a second stressor. This table includes information on taxa, OA treatment levels and other stressors tested, as well as biogeographical region of origin of the species tested.

Phylum, species	**OA Treatments**	**Other stressor tested**	**Effects on settlement**	**References**	**Region**
Porifera					
Carteriospongia foliascens	- pH_T = 8.1 - pH_T = 7.8 - pH_T = 7.6	T = 28.5°C T = 30°C T = 31.5°C	No significant effects	Bennett et al. 2016	Australia
Cnidaria					
Acropora spicifera	- Control pCO_2 = ± 250 μatm - High pCO_2= ± 900 μatm	Control T = 24°C High T = 27°C	No significant effects	Foster et al. 2015	Subtropical - Western Australia
Porites astreoides	- pH = 8.0 - pH = 7.85	- Contact with the alga *Stypopodium zonale* - Two settlement substrates: - CCA - Tiles woith biofilm	No significant effects	Campbell et al. 2017	Caribbean

Phylum, species	OA Treatments	Other stressor tested	Effects on settlement	References	Region
Porites astreoides	- pH = 8.1 - pH = 7.6	Two Temperatures: - Ambient (28°C), - Elevated (31°C) Two settlement substrates: - Plastic algal mimic - Live Dictyota spp.	No significant effects	Olsen et al. 2015	Caribbean
Porites panamensis	- Control pH = 8.08 - Low pH = 7.85	Control T = 28.4°C High T = 29.6 °C	No significant effects	Anlauf et al. 2011	Tropical Eastern Pacific
Mollusca					
Gastropoda					
Crepidula onyx	- Control pH = 8.00 - Medium pH = 7.7 - Low pH = 7.3	Algae grown at - Control pH = 8.00 - Medium pH = 7.7 - Low pH = 7.3	- No effects of pH or diet alone on the settlement - Enhanced settlement at low pH + diet low quality	(Maboloc & Chan 2017)	Hong Kong
Bivalvia					

Phylum, species	**OA Treatments**	**Other stressor tested**	**Effects on settlement**	**References**	**Region**
Magallana gigas (formerly *Crassostrea gigas*)	- pH_{NBS} 8.04 - pH_{NBS} 7.47	- Temperature: 24 and 30 °C - Salinity: 15 and 25 psu	- Reduced metamorphosis due to carryover effects in larvae - Significant delays in pre- and post-settlement growth.	Ko et al. 2014	China
Arthropoda					
Maxillopoda					
Amphibalanus improvisus	$p CO_2$ = 400 µatm $p CO_2$ = 1250 µatm $p CO_2$ = 3250 µatm	- T = 12°C - T = 20°C - T = 27°C	No significant effects	(Pansch et al. 2012)	Baltic Sea
Balanus amphitrite	$p CO_2$ = 400µatm $p CO_2$ = 750 µatm $p CO_2$ = 1500 µatm	- Four temperatures (28°C, 30°C, 32°C and 34°C) - Two nutrient conditions (unenriched and enriched)	- No direct effect on settlement due to acidification alone - Reduced settlement due to warming individually or in combination with acidification	(Baragi & Anil 2017)	India
Echinodermata					
Echinoidea					
Arbacia lixula	pH_T = 8.09 pH_T = 7.69	T = 16.0 °C T = 17.5 °C T = 19.0 °C	Reduced settlement	(Wangensteen et al. 2013)	Mediterranean and tropical Eastern Atlantic

Phylum, species	OA Treatments	Other stressor tested	Effects on settlement	References	Region
Heliocidaris erythrogramma	pH = 7.8 pH = 7.6	T = +2 °C T = +4 °C	No effects on settlement in control pH treatments at both temperatures (Effects on settlement not reported for low pH treatments)	(Byrne, Ho, et al. 2011)	Subtropical Australia
Heliocidaris erythrogramma	pH treatments at which the CCA were conditioned prior to larval settlement assays : pH = 8.1 pH = 7.6	Temperature: - 20.5°C - 24 °C	Reduction in larval settlement when placed in presence of CCA conditioned under any of the elevated temperatures	(Huggett et al. 2018)	Subtropical Australia

Phylum, species	**OA Treatments**	**Other stressor tested**	**Effects on settlement**	**References**	**Region**
Paracentrotus lividus	pH = 8.1 pH = 7.7 pH = 7.4	T = 19°C T = 20.5°C T = 22.5°C	- No settlement observed at the lowest T and pH. - Unsuccessful settlement at the highest T and lowest pH (7.4). - The rest of the results show no significant effect on settlement due to reduced pH. - Enhanced settlement at 20.5°C	(García et al. 2015)	Mediterranean and NE Atlantic Ocean, from Ireland to Canary Islands

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