SURFACE ZOOPLANKTON ABUNDANCE AND DIVERSITY, AND THE SALINITY TOLERANCES OF THE SUBCLASS COPEPODA AND CRUSTACEAN NAUPLII IN MO'OREA, FRENCH POLYNESIA

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Abstract. Surface plankton tows were completed at select reef passes and lagoons over five weeks on the island of Mo'orea, French Polynesia. Differences in zooplankton species richness and abundance were analyzed based on location, salinity, and tide. Reef passes and lagoons varied in the average abundance of zooplankton as well as having different species prevalence. Lab experiments tested individual groups' tolerance to varying salinity levels that were found in Cook's Bay. There was a significant drop in copepod and crustacean nauplii populations when exposed to lower salinity levels.

Key words: Moorea, French Polynesia; Copepods; Veligers; Crustacean Nauplii; Crustacean; zooplankton abundance; zooplankton diversity; reef pass; lagoon; salinity tolerance

INTRODUCTION

Plankton is in the primary trophic level of the marine food web and constitutes a significant portion of the diet of many smaller marine organisms (Nybakken 1993) Zooplankton, in particular, plays a significant role in the transfer of energy to larger organisms (Lafontaine Y, 1994). Gathering data on zooplankton assemblages is important in understanding local marine productivity because of the role it plays in the marine food web. Certain conditions, such as low salinity levels due to increased rainfall, can affect zooplankton populations leading to a suit of effects on the entire marine ecosystem.

Plankton species and abundance differ with currents, depths, time of day, seasons, temperatures, and salinity (Otero and Carbery, 2005). Patterns of diel vertical migration show that different groups of plankton travel to different depths throughout the day (Ramos-Jiliberto and Gonzalez-Olivares, 2000). Accordingly, it is often difficult to get a representative sample of plankton, because surface daytime tows may miss the species that move between depths. Currents and seasonal weather shifts may also move plankton into habitats where they are not usually found. Extreme weather, such as hurricanes or large swells, can be very important in the transport of pelagic plankton into lagoons and vice-versa (Kaartvedt and Svendensen, 1990).

Identifying zooplankton species can be extremely difficult, especially because they may have different morphologies depending on the season and the geography. Significant morphological changes can also occur within their life cycle so that identifying plankton at different stages of development can be extremely challenging (Steedman 1974). For ecological comparisons on a large scale, such as in the comparison of passes and lagoons, species and genus within classes are generally combined and counted together because they often share similar characteristics and behaviors (Canepa, 1996).

The reef geography is crucial in the understanding of plankton distribution and abundance for islands surrounded by a barrier reef, such as Mo'orea. Passes have much more water movement with swell, wind and tide conditions while lagoons are generally less affected by pelagic waters. Sampling the sites of Opunohu and Pao Pao reef passes can provide a good indication of what kinds of zooplankton may be entering and exiting bay environments. One would predict that the differences in environmental and geographical conditions at reef passes and lagoons should cause a consistent and measurable change in zooplankton abundance and richness. I hypothesize that diversity should be higher in the lagoons because not only will there be species endemic to the shallower water but also currents, waves, and wind can bring in pelagic zooplankton. Population densities should be highest at the reef passes because tidal changes and currents bring surface water from many locations into a much smaller, narrow area before being dispersed into either the open ocean with an outgoing low tide, or the lagoons/bays with the incoming high tide.

I tested the copepod and crustacean nauplii for different salinity tolerances. I expect that there will be a lower survival rate the more the treatment is comprised of freshwater. The importance of this experiment lies in that there are many streams on Mo'orea and storm activity often results in increased brackish waters in river mouths and bays. I want to better understand the physiological ability of zooplankton to react to a rapid increase of freshwater. Further implications may help in the prediction of changes caused by climate change on zooplankton populations due to an expected increase in storm activity and rainfall.

MATERIALS AND METHODS

Study sites

Five sites on the island of Mo'orea, French Polynesia, Temae public beach, Pao Pao Pass, Opunohu Pass, the Sheraton Hotel lagoon, and the channel between Motu Fareone and Motu Tiahura (Fig. 1) were sampled six each for surface zooplankton. Plankton tows were completed on peak high and low tides during the same day for each location using the tides for Fare Ute Point on Tahiti around 25 km away.

FIG. 1. Plankton drag sites around Mo'orea. "A" is the channel between Motu Fareone and Motu Tiahura. "B" is Opunohu Pass, "C" is the lagoon outside the Sheraton Hotel. "D" is Pao Pao Pass. and "E" is Temae

Sampling

In a two seat kayak, I would lower a plankton net 0.3m in diameter with 64 micron mesh, so it would be fully submerged and 15 feet behind the boat, then paddle for one minute in a predetermined direction based on where I could go in a straight line without having the net strike any underwater obstacles such as coral heads or buoys. Both kayaks had similar paddling speeds, an average of 60 meters per minute. This was tested in 10 trials for each kayak using a 100 meter transect tape. Each trial was within plus or minus one meter of 60 meters.

At the site, one 50ml water sample was collected for turbidity and salinity measurements while another 1.7 liters of ocean water was placed in a two liter ice cream container to be mixed with the plankton sample from the PVC bucket. 1.7 liters allowed the bucket to be fully emerged in water; and to extract the plankton four complete spin cycles were done underwater and then tipped into the container. The container was then homogenized using ten rapid shakes and a 50 ml subsample was taken incorporating all vertical layers of the sample by slowly filling the vile in a swooping motion. Half of this subsample was analyzed and all zooplankton were counted on the same day as collected to try and avoid any predation and the breakdown of organism tissue.

Species Identification

Identification tables and pictures found in *Coastal Marine Zooplankton* and *A Guide to Marine Coastal Plankton and Marine Invertebrate Larvae* were used to identify organisms. Pictures of samples were taken for future identification (Fig. 2). For this study, identification to the most accurate taxonomic level was used to reduce the error of misidentification.

Sampling regimen

The five sites were re-sampled two and four weeks later keeping tidal fluctuations similar. The tows also incorporated new and full moon's because of their affect on plankton activity (Canepa 1996**)**. The month difference in tows was also designed to show seasonal changes in plankton numbers and assemblages. Although the rainy season (December through March**)** had not fully started, heavy rain storms did provide different conditions to test in. To fully explore salinity changes on zooplankton, lab experiments were conducted to test specific group's tolerances.

Statistical tests

Two way ANOVA tests were used to compare the factors of tide, salinity, and location as well as the correlation between them, to overall zooplankton abundance, richness, and average numbers found of each of the four main groups; copepods, veligers, crustacean nauplii, and plankton "B" (Fig. 2).

FIG. 2. Zooplankton "B"

Salinity manipulations

The Subclass Copepoda and crustacean nauplii were tested by exposing them to different salinity levels. Samples were 25ml of water, enough to be homogenized and tested every 15 minutes over 1 hr but small enough so that analysis could be completed in the allotted 3 minutes per subsample. Every replicate would have one vial with 25 ml of salt water, one vial with 20 ml of saltwater and 5 ml of freshwater, one with 15 ml of saltwater and 10 ml of freshwater, and one with 5 ml of saltwater and 20 ml of freshwater. These solutions were chosen based on real salinity levels found in Cooks Bay. A plankton tow outside of the station or off the dock provided adequate live samples to test. Three vials with ocean water mixed with a plankton sample were used as an initial test to see if similar percentages of living target subjects would be found. All three had similar percentages so the plankton sample was homogenized and added equally to the four treatments. After adding between 75-125 target organisms into the solutions, 5ml samples would be taken from each every 15 minutes for analysis. The first sample at 0 minutes was taken before being mixed with the solution to have a reference as to how much the population declined. Vials were kept at a constant temperature of 28 degrees Celsius in the wet lab.

Species would be classified as alive and active, or as dead. Five replicates were completed for each species and a MANOVA test was used to look for significance over the entire hour. ANOVA and Tukey-Kramer HSD tests were used for each time period to test for significance between the means for each treatment.

RESULTS

Drag samples

There is a statistically significantly higher average number of zooplankton found in reef passes than in the lagoons as demonstrated by a p-value of 0.0098. The average mean at the passes and lagoons were 308 and 174 organisms, respectively. The tide was not found to have a significant influence on abundance with a p-value of 0.9808 and an average mean of 241 zooplankton found at high tide and 240 zooplankton found at low tide. There was no significant correlation between the factors of tide and lagoon/pass (Table 1). A good explanation of this is that there may be other variables that my study did not account for that affect abundance.

TABLE 1. ANOVA results for overall zooplankton abundance. Significant values are in bold and underlined.

	F Value	D.F.	P Value
Reef Pass/ Lagoon	7.7785	1	**<u>0.0098</u>**
High/Low Tide	0.0006	1	0.9808
Lagoon/pass *high/low	0.8457	1	0.3662

The four most prevalent groups had mixed results when comparing tide and location to their respected abundance. Copepods were found significantly more often at the reef passes (an average of 51 copepods) than the lagoons (an average of 26 copepods). The p-value was 0.0359. Tides and the relationship between tides and location were not significant, with a p-value of greater than 0.05 (Table 2) and an average abundance of 39 copepods at high tide and 38 copepods at low tide.

TABLE 2. ANOVA results for Copepod abundance. Significant values are in bold and underlined.

	F Value	D.F	P Value
Reef Pass/ Lagoon	4.8948	1	**<u>0.0359</u>**
High/Low Tide	0.0216	1	0.8844
Lagoon/pass *high/low	0.0307	1	0.8622

TABLE 3. ANOVA results for Veliger abundance

	F Value	D.F	P Value
Reef Pass/Lagoon	1.4760	1	0.2353
High/Low Tide	0.5907	1	0.4491
Lagoon/pass *high/low	2.82	1	0.1048

Location and tides and the correspondence between the two did not play a significant role in veliger abundance (Table 3) as there was an average of 124 and 86 veligers found at reef passes and lagoons respectively while there was an average of

92 and 118 veligers found at high and low tide, respectively.

Passes had significantly higher numbers of crustacean nauplii with a p-value of 0.0004 and a mean number of 91 per sample found at passes while only 12 on average were collected in the lagoons. Tide (65 and 37 crustacean nauplii found on average at high and low tide, respectively) and the correlation between tide and location did not have a significant role in its distribution (Table 4).

TABLE 4. ANOVA results for Crustacean Nauplii abundance. Significant values are in bold and underlined.

	F Value	D.F.	P Value
Reef Pass/Lagoon	16.8230	1	**<u>0.0004</u>**
High/Low Tide	2.0573	1	0.1634
Lagoon/pass *high/low	1.2754	1	0.2691

Only plankton "B" was found in significantly higher numbers in lagoons (a mean of 19 organisms as compared to only an average of 7 found in the pass samples) with a p-value of 0.0361, and once again tide (a mean of 8 and 19 plankton "B" found at high and low tide per sample, respectively) and tidal relations with location show no significant differences (Table 5).

There was a range of 7 to 13 species found in the tows, but their means had no significant differences when the factors of location, tide, and salinity (along with the interaction between each) were analyzed (Table 6).

TABLE 5. ANOVA results for plankton "B" abundance. Significant values are in bold and underlined.

	F Value	D.F.	P Value
Reef Pass/ Lagoon	4.8877	1	**<u>0.0361</u>**
High/Low Tide	3.8399	1	0.0608
Lagoon/pass *high/low	0.2627	1	0.6126

TABLE 6. ANOVA results for average species diversity

	F Value	D.F	P Value
Salinity T/C	0.0323	1	0.8588
Salinity*high/ low tide	0.2254	1	0.6395
Salinity*reef pass/lagoon	1.4462	1	0.2414
Lagoon/pass *high/low tide	3.2249	1	0.0857
Lagoon/pass	0.8066	1	0.3784
High/low tide	2.2371	1	0.1483

Higher salinity levels significantly increased total zooplankton numbers with a p-value of 0.0018. When salinity was tested

TABLE 7. ANOVA results for overall zooplankton abundance. Significant values are in bold and underlined.

	F Value	D.F	P Value
Salinity	12.1017	1	<u>**0.0018**</u>
Salinity*High/ Low Tide	0.0531	1	0.8196
Salinity*Lagoon/ pass	1.8226	1	0.1886

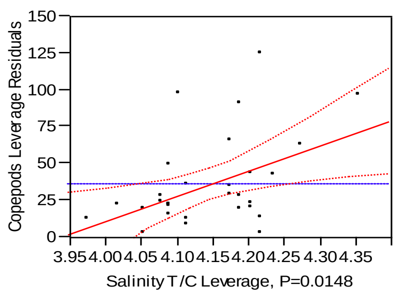

FIG. 3. ANOVA results showing a positive correlation between higher salinity levels and a greater abundance of copepods

with tide and with location (separately) there was no significant difference in abundance (Table 7). Of the four zooplankton groups, only copepod and crustacean nauplii were found in significantly larger quantities with an increase in salinity. The p-values were 0.0018 and 0.0045, respectively (Fig. 3 and Fig. 4). The relationship between the factors of salinity and tide and salinity and location had no significant change in the organisms' abundances (Appendix 1).

Salinity Manipulations

Copepod populations decreased significantly when salinity approached fresh water levels. Table 8 shows that there is a

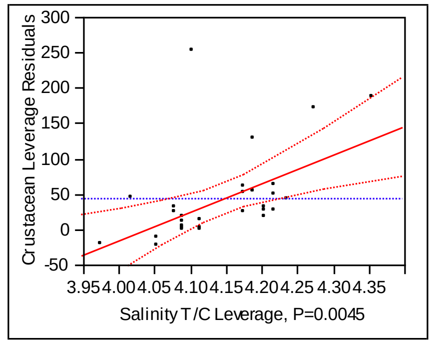

FIG. 4. ANOVA results showing significant positive correlation between higher salinity levels and a greater abundance of crustacean nauplii.

TABLE 8. Repeated measures ANOVA results for Copepod survivorship

	F Value	D.F	P Value
Between (F Test)	33.1417	3	<u>**<0.0001**</u>
Within (Wilks' Lambda)	6.6489	12	<u>**<0.0001**</u>
Time (F Test)	105.6726	4	<u>**<0.0001**</u>

statistical difference in the percentage of living copepods over the different treatment levels for the one hour. The Wilks'-Lambda test demonstrates a p-value of less than 0.0001. Fig. 5 shows that for the control of 0ml freshwater, the percentage of living copepods stays between 60% and 70%. It also demonstrates that the 5ml of freshwater treatment drops about 25% in the first 15 minutes, and then stays constant at around 40% living population for the next 45 minutes. The 10ml of freshwater treatment falls around 45% to around about 20%

TABLE 9. Repeated measures ANOVA results for Crustacean Nauplii survivorship

	F Value	D.F	P Value
Between (F Test)	62.0229	3	**<0.0001**
Within (Wilks' Lambda)	7.7921	12	**<0.0001**
Time (F Test)	48.0796	4	**<0.0001**

where it plateaus for the next 45 minutes. The 20ml treatment dramatically falls around 65% to about 0% living copepods where it levels off as well.

Crustacean nauplius populations died off significantly more when salinity approached fresh water levels. The Wilks'-Lambda statistical test demonstrates a statistically significant p-value of less than 0.0001 (Table 9). Although initial living populations were between 50% and 55% (Fig. 6) they follow similar paths as the copepod tests (Fig. 7), with the 5ml freshwater treatment falling to around 40%, the 10ml treatment dropping to around 20% and the 20ml freshwater treatment decreasing all the way to about 0% living population. More tests should be conducted to test for significance of the similarities between the two experiments' results.

For both experiments, individual ANOVA tests and Tukey-Kramer tests were run at each time point. Significant differences in means between the treatments were found except for a select few cases (primarily between the treatments of 5ml and 10ml of freshwater) when the Tukey-Kramer test showed no statistical significance. The ANOVA test still demonstrated a statistical significance overall and so the few non-significant relationships were not important to the experiments' significant results as a whole.

DISCUSSION

Reef passes and lagoons have significant differences in overall as well as individual group zooplankton abundance. Some groups (such as copepods and crustacean nauplii) are found more at the passes, while plankton "B" is found more often in the shallower lagoon waters. Yet other groups, such as veligers were found in similar quantities in both areas. My results support my hypothesis that passes have, on average, a greater number of zooplankton. It may be due to passes being a point of exchange in pelagic and lagoon waters. It may also be because the deeper water is a more suitable habitat for zooplankton, or one of many other variables.

Individual differences in numbers found are important because it may mean that some of the groups travel more than others. More research is needed to really understand the extent of these migrations as well as the fact that all groups are found in both locations, just in different numbers. The outliers of the data may be due to human error. One possibility is that if I did not homogenize the sample well enough, then the sample would have different species abundance and diversity. Along with the possibility of not having representative data, another variable in need of further study is the idea that zooplankton may be social animals, and may congregate together in swarms (Hamner and Carleton 1979). If this holds true in Mo'orea, then the large numbers of one group found may be due to towing through one of these "swarms". In reverse, some of the samples where I expected to find more organisms than I did may have been because I missed these swarms or even that they swam out of the path of the plankton net (Hamner and Carleton 1979). Towing in the same location and using the same speed during each tow

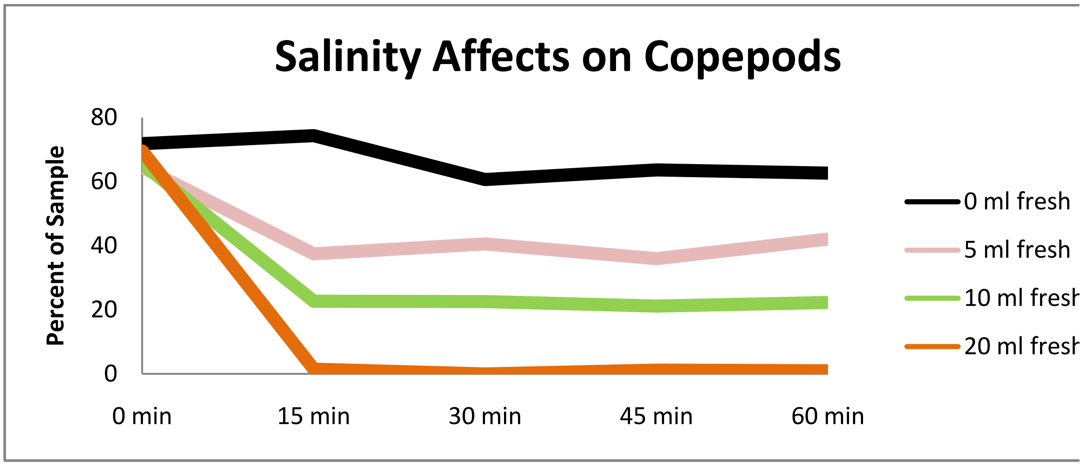

FIG. 5. Copepod living percentage with different salinity treatments

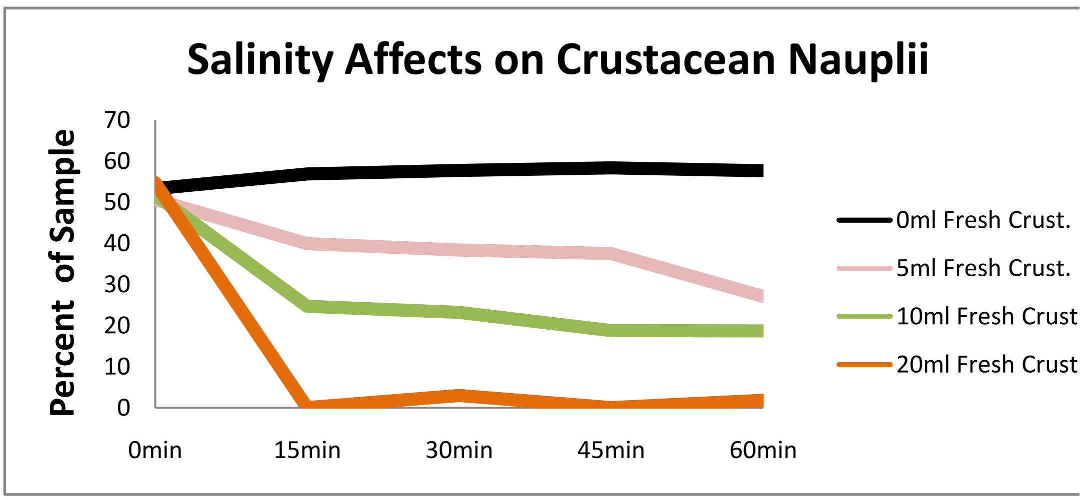

FIG. 6. Crustacean nauplii living percentage with different salinity treatments

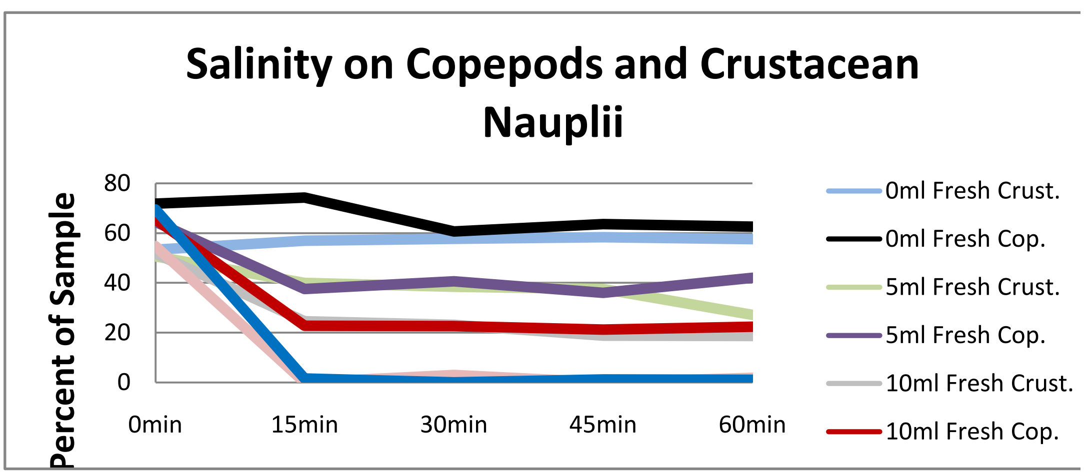

FIG. 7. Comparison of Copepod and Crustacean nauplii living percentage with different salinity treatments

was designed to cut down on sample error, but this did not control for zooplankton's ability to swim away.

Although the data is not conclusive about species diversity when compared to different factors, it still is of interest for different factors, it is of interest for further research. It is apparent that some species prefer either the passes or the lagoons, and there was a slight trend towards having more species found at lower tide especially in the passes. My data does not support my hypothesis that lagoons have greater species richness, but more data, especially from different depth ranges as well as time changes, is needed to check for statistical significance.

An increase of freshwater lowers the percentage of copepods and crustacean nauplii that are able to survive (Fig. 7), supporting my hypothesis that there is a positive correlation between lower salinity levels and an increase in death rates. Copepods and crustacean nauplii are two of the three most abundant species I found around Mo'orea, meaning that any large influx of freshwater may have devastating impacts on the marine environment.

The lack of significance between some treatments at the same testing time may be corrected with more replicated. Some of the samples may have been exposed to heavier shaking for homogenization, and since they are not being replaced or used in other treatments, each organism has its own tolerance to salinity and to disturbance so small amounts of variation are expected.

Copepods are one; if not the most, important group in the transformation of energy to higher marine levels (Hamner and Carleton 1979), meaning that any large drop in population will have significant affects on the rest of the ecosystem with a major drop in food availability for small marine organisms. The significant results in the two groups' stress test are a good indicator that other marine organisms may be affected by salinity levels in similar ways. This is important when considering current climate changes, because if there is increased storm activity on a large scale, then the increased flow of freshwater flow into the ocean may have an impact on plankton composition. More research is needed to see whether increased rainfall and storm activity will affect the zooplankton populations.

Zooplankton is an underexplored key group of small animals; therefore there are almost infinite possibilities for further research. Looking into energy transformation between phytoplankton to zooplankton, then on to higher trophic levels is one topic to look into. Also, different observational studies are necessary to better understand the social structure of different groups. Studying the few organisms that seem to be healthy and fine in the 20ml of freshwater treatment could lead to interesting discoveries on individual tolerances. Lifespan studies, physiological studies, and exploring how great a distance they can travel could further the understanding of the importance of zooplankton. More stress tests could give scholars a better idea of what to be careful of in the future if natural conditions continue to change. More studies could be focused on the interactions between different species at different levels of development, and different zooplankton hierarchies could be established based off of predator/prey interaction.

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APPENDIX 1. ANOVA results for Copepod, Veliger, Crustacean Nauplii, and plankton "B" abundance. Values in bold and underlined represent a statistically significant level of below 0.05.

	Source	F Value	D.F.	P Value
Copepods	Salinity	6.8183	1	**<u>0.0148</u>**
	Salinity*lagoon/reef pass	3.1384	1	0.0882
	Salinity*High/Low tide	3.6038	1	0.688
Veligers	Salinity	1.71	1	0.2024
	Salinity*lagoon/reef pass	0.1283	1	0.7231
	Salinity*High/Low tide	0.1007	1	0.7535
Crustacean Nauplii	Salinity	9.6654	1	**<u>0.0045</u>**
	Salinity*lagoon/reef pass	0.9416	1	0.3408
	Salinity*High/Low tide	3.5619	1	0.0703
Crustacean "B"	Salinity	2.6112	1	0.1182
	Salinity*lagoon/reef pass	2.5047	1	0.1256
	Salinity*High/Low tide	0.0246	1	0.8765