Sustainable Marine Aquaculture in the Southern California Bight: A Case Study on Environmental and Regulatory Confidence: Final Report

California Sea Grant Project Number: NA14OAR4170075

Report Prepared by: J.E. (Jack) Rensel, Barry King and James Morris.

Contracting officer: Rose Madson, California Sea Grant College Program

Report Date: 20 September 2017

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Executive Summary

We conducted an environmental performance evaluation for a proposed open-ocean fish farm in the Southern California Bight (SCB) 7.2 km offshore of San Diego, California. The proposed farm would be surface cages over 95m depth where it would grow California yellowtail (*Seriola dorsalis*). The fish farm is intended to be an offshore aquaculture demonstration project that would include intensive and transparent monitoring of benthic and water column effects. The project would also serve as national case study for sustainable aquaculture development in the U.S.

This assessment focused on the use of an environmental model to forecast the probable water column and benthic effects of fish metabolic wastes from the farm. A properly sited and operated fish farm can be designed to produce quantities of wastes that can be readily assimilated into the marine food web with no adverse effects. Often in the past, this balance has sometimes not been achieved, or if so, only by trial and error that can involve expensive site reconfiguration and permit modification.

The overarching goal of this project was the analysis of probable effects of the proposed project and demonstration of the capabilities of aquaculture environmental modeling to California federal and state coastal managers. To ensure that the assessment focused on relevant questions by coastal managers, two workshops were held to inform the assessment and model development. The environmental model "*AquaModel*" was utilized in this project as the modeling framework. *AquaModel* was initially created to address open-ocean fish farming seabottom and water column effects. It has been widely used in different seas and locations and validated for both nearshore and offshore fish farms for several different species of fish. A standard approach is to calibrate and tune the model to a specific eco-region and habitat type.

To calibrate *AquaModel* for the SCB region, we utilized existing water and sediment quality databases. Site-specific data including an extensive nine-month current meter record and sediment data were obtained from the proposed site. A new physiology sub-model for California yellowtail providing baseline growth and feed conversion projections was developed using a temperature-specific growth model developed for kingfish (*Seriola lalandi*) aquaculture in Australia.

A ten-year fish production plan including stocking schedules, mortality, and harvest rates was created for *AquaModel* to predict farm effluent characteristics, environmental fate and effects. To assure that the model was adapted to local conditions, we evaluated the sensitivity of model response to key input parameters. *AquaModel* validation has been accomplished historically through iteration, by systematically varying input parameters until best fit between observed and predicted results was achieved. That was not possible for this project as the fish farm is not yet in operation.

The most sensitive model input parameters that created the most change in in the degree of seabottom sediment enrichment of total organic carbon (TOC) or sulfides was the water current-threshold current velocity for fish feces deposition and erosion on the seabottom. The same factors for waste fish feed produces significantly less change in model results for sediment TOC and sulfides.

Our sensitivity analysis in general illustrated that several fish fecal settings were more sensitive to change than those for waste fish feed, and that future analyses should have renewed emphasis on these factors for model calibration. We emphasize that the sensitivity analysis does not demonstrate what has been empirically proven elsewhere, that waste fish feed may have the most pronounced effects on sediments near fish farms if not kept to a low rate. In the U.S., federal regulations have for many years required the use of waste feed monitoring. This measure was supported by the industry. Pelleted fish feed is the single largest cost of fish farming in salmon and other large fish farming operations. It's loss can adversely affect the seabottom ecology and violation of benthic regulatory standards.

Overall, the *AquaModel* simulations reported here suggest that the proposed fish farm will not likely cause significant adverse impacts on water quality or seabottom invertebrate or fish communities. For example, at farm full operation (5,000 metric tons annual production) the maximum increase of sediment TOC concentration predicted directly beneath the cages was only 0.002%. Given the cool water temperatures and other conditions near the seabottom, this may result in a minor and beneficial increase in species diversity of the infauna invertebrate population or no measurable effect whatsoever. Increases of species diversity are considered by ecologists as beneficial because more diverse biotic communities are more stable and less likely to be perturbed by other factors.

Fish farms have a biological oxygen demand for their wastes as aerobic bacteria use oxygen to oxidize the organic carbon. As a result, siting of open-ocean farms in the SCB is best performed by selecting sites with the largest possible distribution of particulate wastes to allow food web assimilation that limits further oxygen drawdown. In the present study, there was a projected drawdown of dissolved oxygen underneath the cages, but not in adjacent areas. The level of sediment dissolved oxygen in the model simulations mostly remained above hypoxic (2 mg/L) levels and is a seasonal, not annual condition. The model was purposely constructed with the most conservative settings, including no use of waste particulate matter by zooplankton or wild fish species as the particles sink for long periods to the distant seabottom. In some seas, these are significant factors in reducing waste loading of the bottom and it probably will occur in the SCB, based on what is known about wild fish fecal wastes in the SCB region.

The model was also used to forecast how fast all traces of the fish farm would be gone from seabottom sediments if it was removed by harvesting all the fish at one time, while allowing the modeling processes to continue normally. Sediment TOC and sulfide concentrations returned to background levels within two weeks of fish removal and there were no effects whatsoever forecast by the three week point. These forecasts are consistent with several recovery rate field studies conducted in British Columbia that illustrated that faster recovery for fish farms located in areas with faster near-seabottom currents.

Analysis of projected water quality in and around the farm suggested that the near-surface average current flow of 19 cm/sec is sufficient to exchange the water within a single cage every 2.6 minutes and maintain concentrations of dissolved oxygen above 6.3 mg/L. Simulations of excreted ammonia from the cultured fish produced a maximum increase of 0.008 mg/L total ammonia at flow minimums but were rapidly diluted to immeasurable values near and inside the cage under all other flow regimes, resulting in no adverse effect on either the cultured fish or downstream biota. Ammonia is rapidly converted to nitrate in most oxygen rich aquatic environment by naturally occurring bacteria and other fauna.

Introduction

Offshore aquaculture in the U.S. is not yet developed but provides much promise for reducing the U.S. seafood trade deficit, revitalizing working waterfronts, and providing our nation with secure sources of seafood. This project provided an environmental performance evaluation of a proposed open-ocean fish farm 7.2 km offshore of San Diego in the Southern California Bight (SCB) with 95 meters water depth (Figure 1). The fish farm is being proposed by a consortium known as Rose Canyon Fisheries that includes the Hubbs SeaWorld Research Institute of San Diego, California.

The fish farm would be used to grow one or more species of native marine fish but focusing on California yellowtail (*Seriola dorsalis*) as a demonstration project where the operation and impacts would be closely monitored and reported. At build out, production would be approximately 5,000 metric tons per year. This is not a large fish farm compared to many that exist elsewhere in North America and elsewhere but would be closely monitored, modeled, and evaluated so that society and decision makers can judge the merits or drawbacks of open-ocean fish farming in southern California.

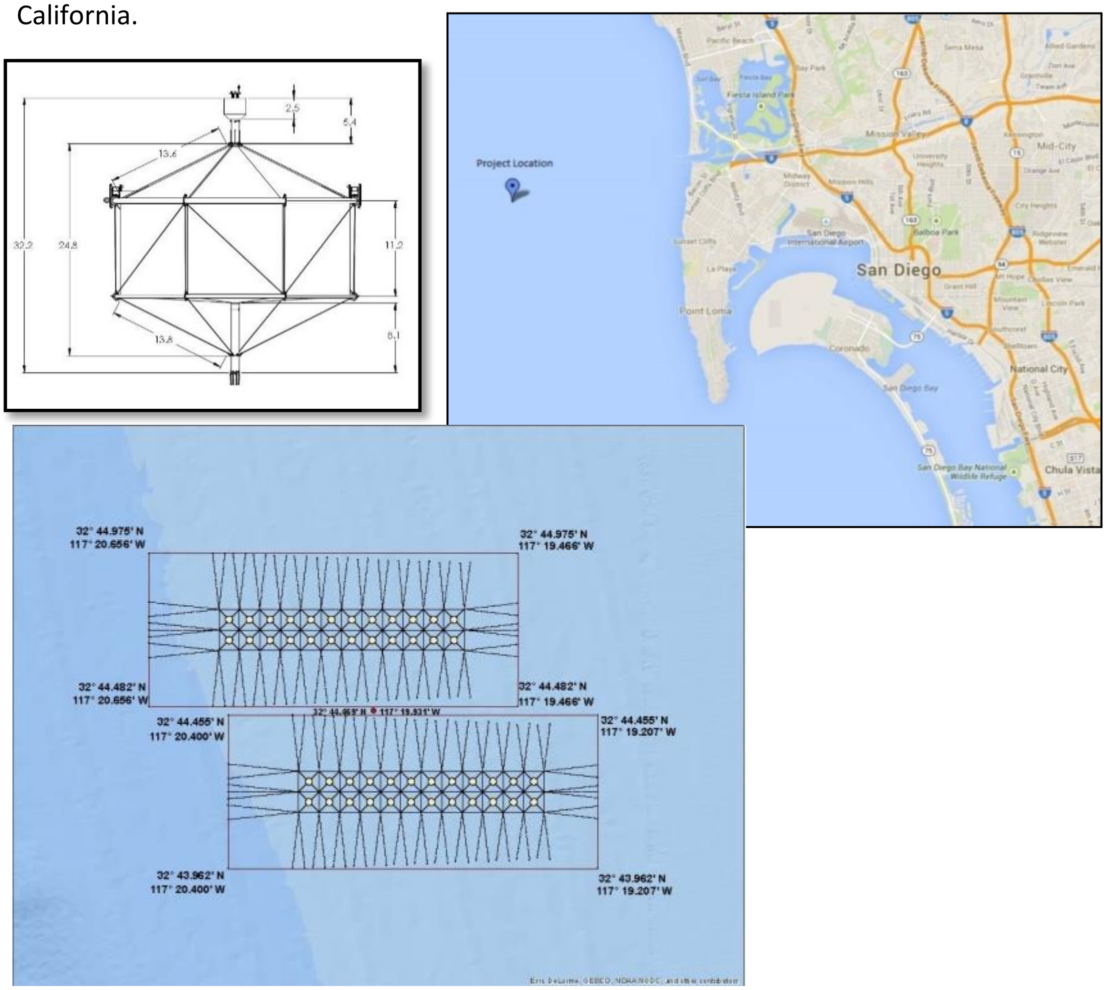


Figure 1. Project site vicinity (upper right), side profile of a single submersible cage (upper left), layout of cages and anchor lines (lower left) with current meter and central seabottom sampling locations (green star) and two other locations (red stars) used for baseline, seabottom sampling.

This project was also conducted to demonstrate the capabilities of aquaculture environmental modeling to California State and Federal agency regulatory and coastal managers through two workshops held in Long Beach California for state and federal agencies and selected academic or technical experts from the SCB region.

Our technical evaluation focused on the use of a software program to forecast the probable water column and benthic effects of fish metabolic wastes from the farm. A properly sited and operated fish farm should be designed to produce quantities of wastes that can be readily assimilated into the marine food web with no adverse effects. Often in the past, this balance has sometimes not been achieved or if so, only by trial and error that can involve expensive site reconfiguration and permit modification.

Fish aquaculture modeling has been in existence for several decades, but most models are never used operationally (routine use during permitting or operation) because of complexity and difficulty of use. However, several of the leading countries involved with salmon and marine fish farming now require the use of models for new site evaluation or existing site changes in configuration or expansion. Other countries are developing regional modeling approaches to evaluate coastal carrying capacity for fish farming for fish health biosecurity and eutrophication prevention or mitigation.

The model for this project needed to simulate many years of production and environmental effects, in this case nine years. The model also had to be able to forecast both water quality effects and changes if any to the seabottom benthic communities, simultaneously if possible. Ideally, the computer model would also be able to evaluate the possible cumulative effects of multiple net pens throughout broad coastal ecoregions. Coastal eutrophication is an increasing problem in many of our world's seas. Aquaculture can adversely affect nutrient loading, but its influence is usually dwarfed by terrestrial, riverine and atmospheric sources of nutrients that may cause algal blooms and subsurface hypoxia (lack of oxygen). Yet there are many locations where fish aquaculture can be practiced with no adverse effects, for example, areas where nutrient control of algal growth is not important as other factors such as light are the limiting factor.

The goal of modern aquaculture modeling is to allow for the sensible and scientifically-defensible siting and operation of fish farms to help feed the world with healthy seafood. The model used in this study has been under development for 17 years and is still being modified, corrected, improved and validated when possible and was built with all the above needs in mind. The subsequent sections of this report explain the modeling approach, methods of analyses, sensitivity of the model, and the modeling results with regard to farm impacts on water quality and seabottom sediments.

AquaModel Background

AquaModel is a computational tool for planning and evaluating aquaculture sites, both for single farms and multiple farms over broad geographic regions. It operates on Windows computers and provides a simple interface to enter environmental and operational information. Graphical outputs map the distribution over time of key parameters including oxygen, particulate organic and dissolved nutrient wastes, algal and plankton effects and dozens or other environmental and fish cultural/management parameters.

The single farm *AquaModel* version is simple to setup and operate and requires a current meter record or other single-location estimates of currents either from one depth or multiple depths. *AquaModel* has been used to simulate single (near field) and in some cases multiple (far field) fish farms over broad areas for Atlantic salmon (*Salmo salar,* 3 variations for west coast North America, East Coast N.A. and Chile), rainbow trout (*Oncorhynchus mykiss,* steelhead variety reared to large size in net cages), cobia (*Rachycentron canadum*), striped bass (*Morone saxatilis*), longfin yellowtail (*Seriola riviolani*), moi *Polydactylis sexfilis*), sea bream (*Sparus aurata*), hybrid grouper (two species), sablefish (*Anoplopoma fimbria*), and California yellowtail (*Seriola dorsalis*), similar to Kingfish, *S. lalandi*). The single farm version has been used by government agencies in the U.S. (NOAA during this project, in the Gulf of Mexico, and in Hawaii), Hong Kong and Chile. This modeling approach has also been used elsewhere in coastal waters of Atlantic Canada, Puget Sound, Washington, in the Atlantic Ocean near Portugal, the Sea of Oman in the Arabian Sea, the Belizean Lagoon and near the east coast of Puerto Rico (see www.AquaModel.org publications page). A multiple farm version of *AquaModel* used over large coastal areas is also available but it usually used by for or by governments or industry organizations that have the resources to provide needed 3D circulation model data and are interested in studying carrying capacity, zoning or farm site connectivity. *AquaModel* validation and region-specific calibration studies have been completed in some of these areas, most recently from open ocean waters near the Big Island of Hawaii (Rensel et al. 2015). The world's coastal seas can be categorized into "eco-regions" with shared traits and the model is then validated for the most optimum habitat(s) with each ecoregion to improve model output accuracy.

AquaModel simultaneously describes nutrient transformations by fish farms of both dissolved and particulate materials in the water column and sea bottom as shown in a conceptual approach drawing (Figure 2). A system of interlinked equations describe fish growth and physiology that integrates with flow field data to transport waste from farms, assimilate dissolved nutrients by plankton, and simulate the sinking, deposition, assimilation by benthic organisms as well as resuspension and mineralization of fish feces and uneaten feed. A mathematical description of fish growth and metabolism consists of a nutrient budget for carbon, oxygen, and nitrogen as determined by the size of the fish, water temperature, dissolved oxygen concentration, swimming speed, feed rate and composition. Optimal feeding rates for varying environmental conditions are provided as outputs in addition to all other simulation output data in tabular form.

AquaModel also provides a dynamic 4-dimensional display (3D plus time) of aquaculture and environmental processes and resides within the *EASy* Geographic Information System. This GIS was specifically designed for marine applications and provides interfaces to import diverse types of environmental data including satellite imagery, current meter data, modeled 3-D current data, bathymetry, and coastlines allowing site or regional-specific information to be incorporated into the simulations.

An example of dissolved oxygen concentration simulations at a 12-cage fish farm in British Columbia is provided in Figure 3. Plots surround the main image including oxygen transect (left center, along a user specified red line), a dissolved oxygen vertical profile through one of the pens (center, top), a time series of surface and bottom current speed and sediment organic carbon concentration transect (top right). Many other plots are available and all are updated at each user-specified time steps. Benthic outputs include sediment total organic carbon, oxygen and sulfide concentrations and virtual aerobic and anaerobic bacteria population abundance that respond to changes in temperature, organic carbon loading and effects of particle resuspension and transport.

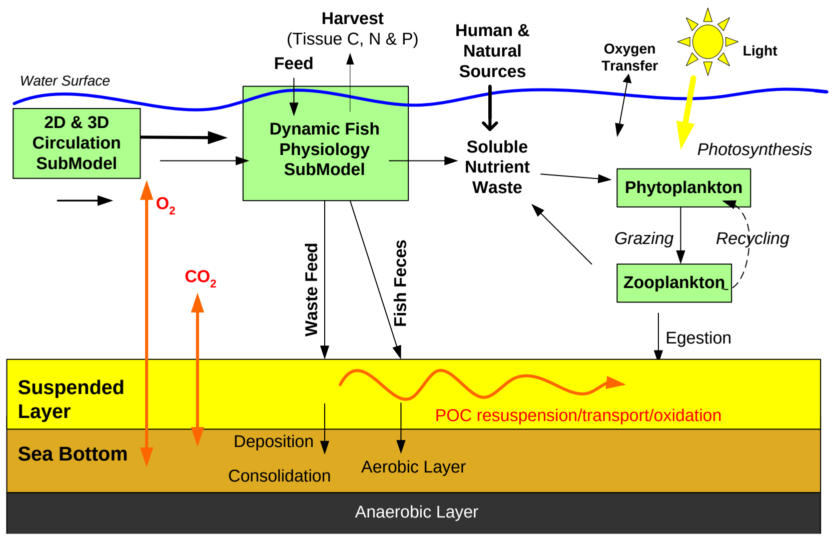


Figure 2. Simplified conceptual framework of *AquaModel* software.

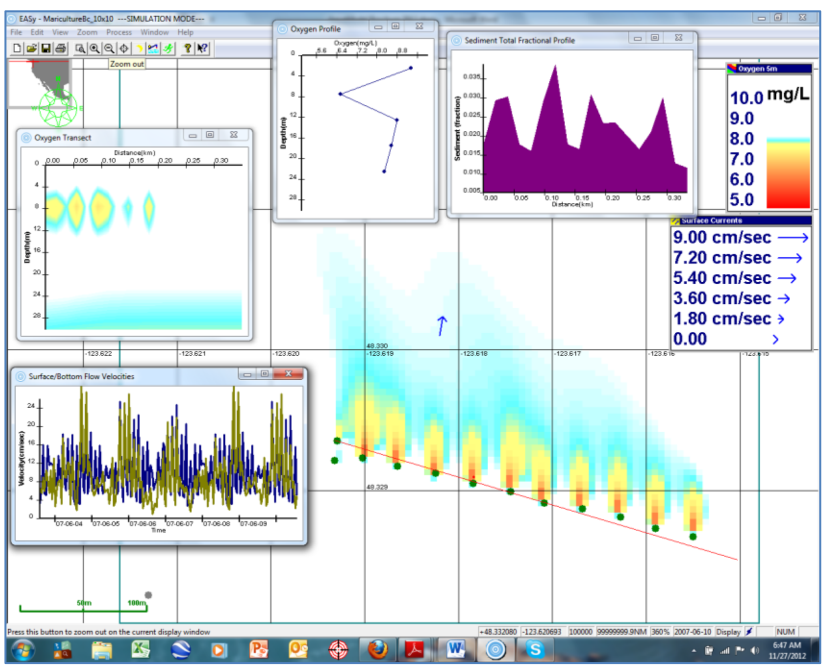


Figure 3. Screen print of model output from one time step showing water column effect of existing British Columbia fish farm described herein.

Fish Farm Production Planning

A fish farm stocking and harvest plan was developed and used to model the fish farm performance and effects. The plan included numbers and size of fish stocked, fish mortality, harvest rate and cage use required to meet the objective of about 5,000 metric tons of fish production per year. It was also necessary to provide for slowly ramping up fish production from a few cages to all cages over several years in order to utilize increasing production from an onshore juvenile production hatchery and adapt successfully to site-specific conditions. The plan included the following specifications and resulted in a schedule shown in Table 1.

- 70,000, 30 gram fish stocked per pen.
- 250 MT of 4 kg fish harvested per pen after about 2 years of ongrowing.
- 10% mortality, mostly as juveniles.
- Approximately two years to attain harvest size in California water temperatures.
- Four stockings per year in March, May , July, September
- 40 of the available 48 cage locations would be required.

Table 1. Annual fish stocking, harvest, and cage use specifications.

Year	Stocking Rate (MT)	Total Harvest Rate (MT)	New Cages Added	Cages in Operation
1	1000		4	4
2	1000		4	8
3	2500	1000	6	14
4	2500	1000	6	20
5	5000	2500	20	40
6	5000	2500		40
7	5000	5000		40
8	5000	5000		40
9	5000	5000		40
10	5000	5000		40

The above growth and stocking estimates were based on the use of a proprietary model of *Seriola lalandi* (kingfish) growth as cultured in New South Wales region of Australia. The growth model was run with near surface water temperatures common to the project site. Operational data were obtained necessary to ramp up production in a stepwise fashion to meet the fish production goals of the project (Figure 4 & 5). Based on these projections, data was obtained for fish stocking specific size and density, mortality rate and harvest amounts and timing of the model for a nine year plan of fish farm operation using overlapping cohorts of fish stocked and harvested at different times.

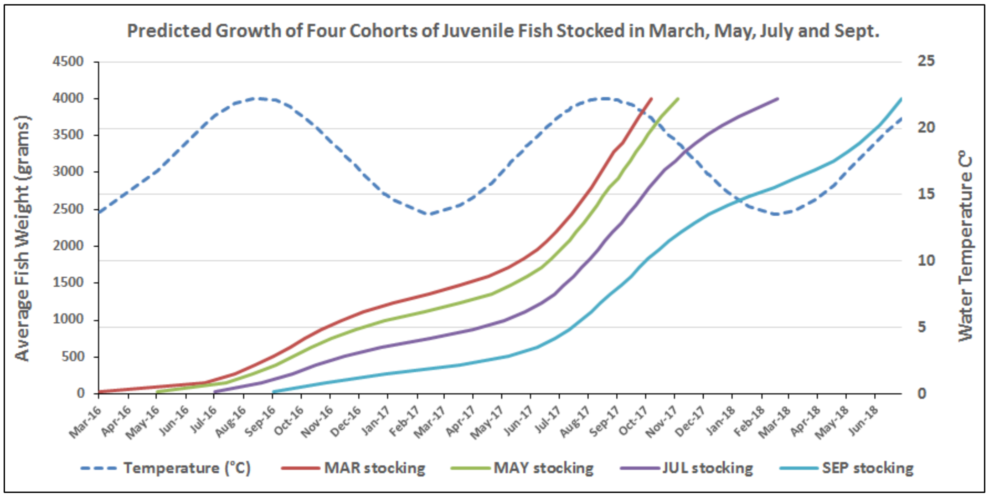


Figure 4. Projected growth of four cohorts of juvenile California Yellowtail stocked in the months of March, May, July and September estimated with SCB water temperatures from the New South Wales Australia growth prediction spreadsheet model.

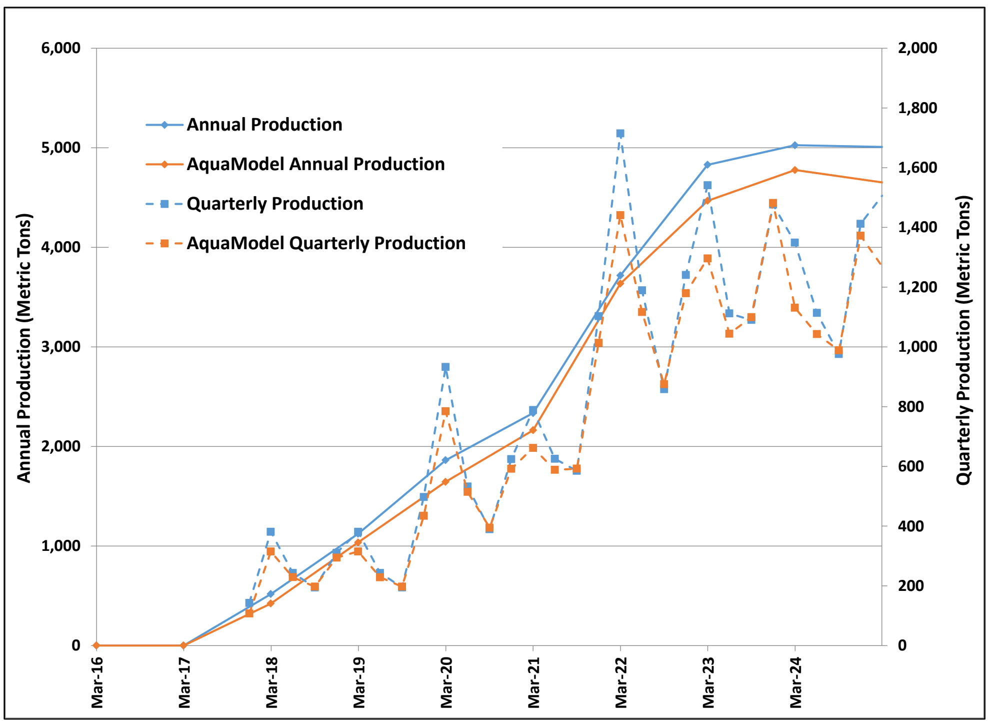


Figure 5. Rose Canyon Fisheries fish farm total and single example cage biomass over the 9 year simulation.

RCF Model Regional Calibration

After completing an operational plan and input file for *AquaModel*, it was necessary to adjust the model setup and calibration for the Southern California Bite region and in particular for the type of habitat found near the proposed fish farm site. Although *AquaModel* had been used ten years previously in a nearby location (Kiefer et.al 2007), the model structure and function had extensive alterations in the following decade. Setup and calibration is done separately for water column and benthic considerations, with some overlap and for physiological characteristic of the fish species to be cultivated.

The calibration included several categories of input data, model settings and plan preparation that are routinely developed for *AquaModel* application at single sites intended to be representative of similar habitat throughout a specific region:

- GIS data that included bathymetry and spatial analysis or use data
- LOCATION was 4.5 miles (7.2 km) offshore of San Diego in 95-meter water
- FARM, PENS, FISH SPECIES were 40 ocean pens ~11,000 m^3 each, California Yellowtail
- AMBIENT PHYSICAL CONDITIONS included winds, temperature, currents, sampled water quality, sampled sediment chemistry and physical factors
- STOCKING PLAN included 30g juvenile fish at stocking, 4 kg harvest, ramped up to 5000 MT/yr. (~11 million lbs.)
- MODEL PARAMETERIZATION included over 35 parameters selected from peer review literature and validated sites around the world. When a range of optimum parameter values were available, the most conservative value was chosen providing highest confidence in model output.
- SIMULATION model external time step of 30 min time steps for 9 years, covering 27 km^2 ocean
- MODEL EXECUTION involved 3 days to runs on highest clock rate, 64 bit, 32 core, Windows 10 computer, 0.5 Gbyte output file size per run.

The next several sections address the above topics in more detail.

Model Parameter Description

The model configuration settings used in this study are considered to be appropriate for the types of sediment present at this site (about half sand and half silt/clay) and other factors such as average current speed. Some of these configuration settings are known with relatively high certainty (e.g., waste feed or fish fecal settling rates through the water column). Other factors are less well known (e.g., resuspension rates of particulate wastes that are temporarily deposited on the seabottom. Selection of settings from the less-well-known category is driven in part on prior trial and error model validation and tuning studies of existing fish farms, both inshore and offshore locations (Cromey et al. 2002a, Cromey et al. 2002b, Chamberlain and Stucchi 2007, Kiefer et al. 2011, O'Brien et al. 2011, Rensel et al. 2013, Rensel et al. 2015).

Parameter	Units	Parameter Components	Relative Uncertainty (1 low – 3 high)
Sediment carbon factors	grams C m^2	1. Sediment aerobic carbon factor 2. Sediment anaerobic carbon factor*	1
Sediment carbon assimilation maximum rate coefficient	per day (d^{-1})	3. Sediment carbon maximum aerobic assimilation rate coefficient 4. Sediment carbon maximum anaerobic assimilation coefficient*	2
Waste deposition & resuspension thresholds	centimeters per second (cm s^{-1})	5. Fish fecal deposition velocity threshold 6. Fish fecal resuspension velocity threshold 7. Waste fish feed deposition velocity threshold 8. Waste fish feed resuspension velocity threshold	2
Erosion rate constants**	g carbon m^2 d^{-1}	9. Fish fecal erosion rate coefficient 10. Waste feed erosion rate coefficient	3
Sediment consolidation rate	fraction d^{-1}	11. Fish fecal consolidation rate 12. Waste fish feed consolidation rate	2
Fish fecal settling rate	centimeters per second (cm s^{-1})	13. Mean velocity fish feces settling rate (uncertainty varies by fish species)	1 - 2

*Asterisk indicates not required in this situation due to a lack of anaerobic conditions at pens or reference areas in measured and all model outcomes. ** These are not fixed rates but rather part of a computational system that includes varying near bottom flow rates and other factors.

Current Flow Data

Physical water current flow is a critically important model input required for accurate fish farm modeling. Inshore waters often have repeating tides that are adequately represented by a fortnightly (14 day) or lunar tidal cycle (28 day) current record. At open ocean locations, tides may not be the dominant factor so that longer current flow records or accurate circulation model data is needed to forecast flows and effects of a fish farm on sediment and water column conditions. In the case of the RCF project, an exceptionally long and high quality current flow record was available.

Staff of Hubbs SeaWorld Research Institution provided extensive current meter data from the center of the proposed site (M. Shane, HSWRI technical report). A bottom mounted TRDI Acoustic Doppler Current Profiler (ADCP) was deployed on the seabottom at 82 m (271fsw) near the center of the proposed net pen site to collect data in 28 each, mostly 3 meter high bins. The first deployments began on 15 October 2014 and extended to 11 March 2015 when the unit was recovered. After ADCP recovery and maintenance, a second deployment commenced on 1 May 2015 and was completed on 29 September 2015. The sampling interval throughout was four times per hour. To compensate for the missing 20 days of data, ten days prior and ten days after the hiatus were copied to fill the void.

The available current meter data in ASCII form was converted to spreadsheet format using the TRDI (manufacturer's) software and arrayed into columns of matching depths for velocity and direction of each depth bin. Basic statistical summaries were constructed for each depth bin and plots made for inspection of outliers and missing cells. Near surface bin data was discarded as is normally done due to surface backscatter interference but data from 6 meters deep were evaluated and found to pass quality control and assurance tests Table 1. A small number of missing cells occurred but they were very infrequent and usually only from single or two adjacent ensembles at one depth.

Table 1. Basic statistics for current meter data before *AquaModel* replacement of missing cells

Depth (meters)	78	75	72	69	66	63	60	57	54	51	48	45	42
Mean Velocity cm/s	**9.3**	**10.0**	**10.1**	**10.1**	**10.2**	**10.3**	**10.5**	**10.7**	**11.1**	**11.5**	**11.9**	**12.4**	**13.0**
Standard Deviation	4.8	5.4	5.7	6.0	6.4	6.7	7.0	7.1	7.3	7.4	7.6	7.8	8.1
Coefficient of Variation	1.9	1.8	1.8	1.7	1.6	1.5	1.5	1.5	1.5	1.6	1.6	1.6	1.6
10th Percentile cm/s	3.6	3.7	3.6	3.6	3.3	3.2	3.3	3.4	3.6	3.7	3.8	3.9	4.1
90th Percentile cm/s	15.8	17.3	17.8	18.3	18.8	19.3	19.8	20.1	20.6	21.4	22.4	23.5	24.7
Total N	35041	35041	35041	35041	35041	35041	35041	35041	35041	35041	35041	35041	35041
Missing Cells*	0	0	0	0	0	0	0	0	0	0	0	0	0
Count > 75 cm/s	0	0	0	0	0	0	0	0	0	0	0	0	0
Count > 100 cm/s	0	0	0	0	0	0	0	0	0	0	0	0	0

Depth (meters)	39	36	33	30	27	24	21	18	15	12	9	6
Mean Velocity cm/s	**13.6**	**14.3**	**14.9**	**15.6**	**16.3**	**17.0**	**17.5**	**18.1**	**19.2**	**19.4**	**19.7**	**17.9**
Standard Deviation	8.5	8.9	9.4	9.8	10.3	10.9	11.5	12.0	13.4	13.1	13.8	11.1
Coefficient of Variation	1.6	1.6	1.6	1.6	1.6	1.6	1.5	1.5	1.4	1.5	1.4	1.6
10th Percentile cm/s	4.3	4.4	4.7	4.9	5.2	5.2	5.2	5.4	5.2	5.4	5.1	5.7
90th Percentile cm/s	26.1	27.5	29.1	30.7	32.3	33.7	35.1	36.2	39.1	38.6	40.2	32.5
Total N	35041	35041	35041	35039	35034	35007	34989	34953	34826	34857	34843	35034
Missing Cells*	0	0	0	2	7	34	52	88	215	184	198	7
Count > 75 cm/s	0	0	0	0	0	0	0	0	69	36	62	77
Count > 100 cm/s	0	0	0	0	0	0	0	0	0	0	0	28

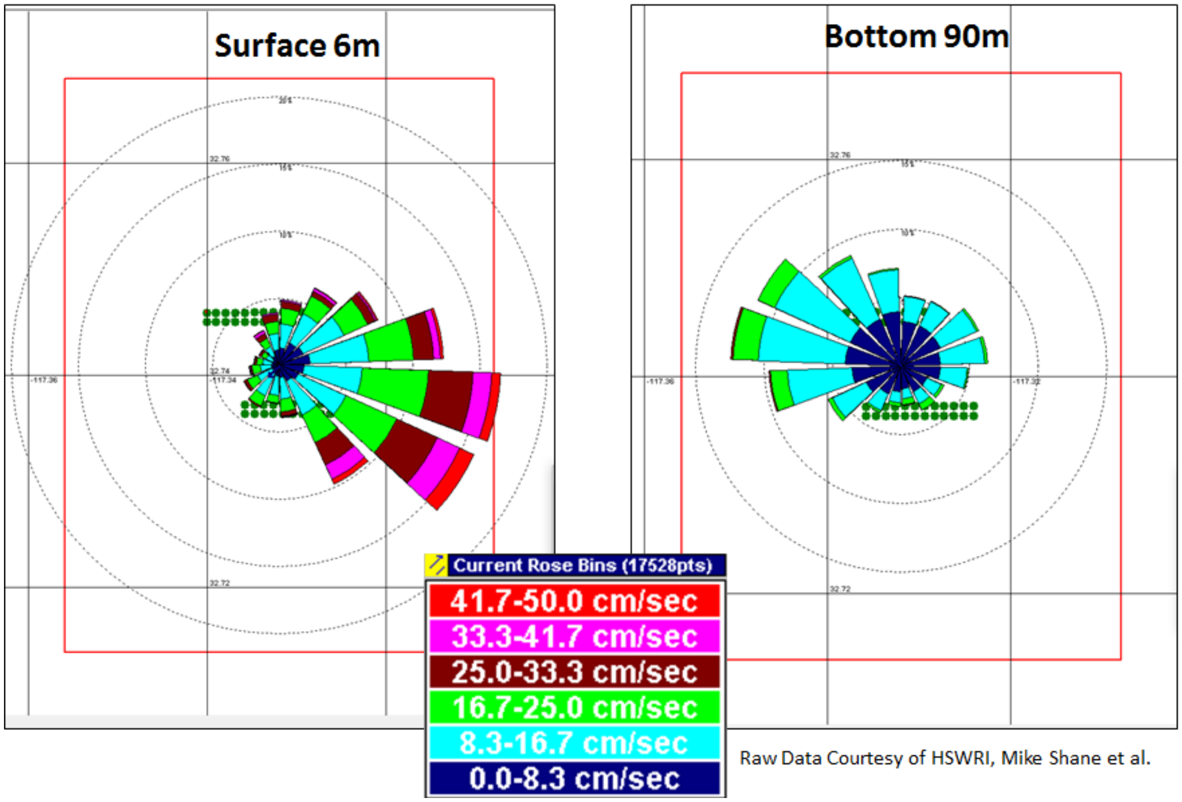
After inspection of the data and additional plotting, the data was loaded into *AquaModel* for further analysis. Missing cell data were provided by an *AquaModel* utility that interpolates speed and direction between available data surrounding the few blank cells. As Table 1 indicates, the missing cells were all in the upper water column, as expected, and were relatively infrequent and occurring as single blanks bracketed by useful data so that the interpolation utility use was reasonable. The maximum number of missing cells was for the 9 meter depth bin, with 0.6% of the data missing before interpolation in that single depth bin. Overall, about 0.1% of the data were missing for all depth bins.

In order to graphically summarize current flows at the project site, we used an *AquaModel* utility that produces water current “vector rose” diagrams. Vectors are a combination of direction and magnitude velocity; “rose” diagram refers to having a 360 degree polar view of the graphic. These are constructed by the model for specific depth bins, including near surface and near bottom as shown and for subsurface at 15 m depth in Figure 6.

The vector diagrams summarize the direction and magnitude of flow, in this case for an entire year period. The near surface vectors show a strong east-southeasterly flow most of the time. Current velocity at times exceeded 42 cm/s or about ¾ nautical miles per hour.

Conversely, the near bottom flow was weaker, as is often expected but almost a mirror reverse image for direction, to the west-northwest. It is possible to produce these profiles for different time periods and the outcomes show some variation but center around the above illustrated patterns. Figure 6 illustrates the current vector rose from the same 1 year time period but a different depth of 15 meter below the surface. Note that the dominant currents have become southerly (SSE) oriented and the appearance of a significant current to the NNW is now also apparent, not as shown above in Figure 6.

Water currents at various depths in inshore locations that are tidally controlled tend to flow in similar directions and decrease in velocity slightly with depth, particularly near the sea bottom. There are of course many exceptions, particularly in strong velocity channels and passages. Offshore waters of the Southern California Bight are replete with several stationary or slowly migrating mesoscale current gyres (eddies), although most of them are further north than the project site. We observed the current roses as they were constructed by the model and the beginning (e.g., one month elapsed time) and the end of the one year



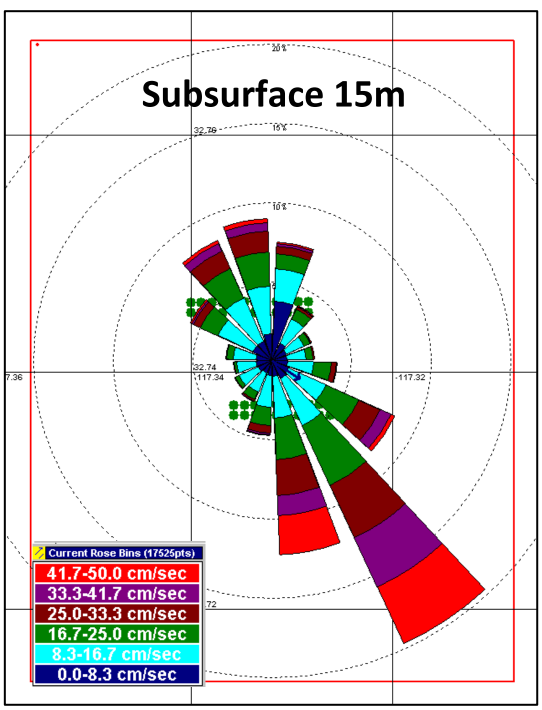


Figure 6. Water current vector rose diagrams from near surface 6m bin and near bottom 90 m bin (above) and for subsurface depth bin of 15 m (below) for the RCF project site.

Growth and Physiology Submodel of California yellowtail

Concurrently, a fish physiology submodel for *S. dorsalis* was constructed using the Australian data that had quite similar accompanying water temperatures. The *S. dorsalis* physiology submodel was then run within *AquaModel* to insure that the growth was similar to that observed in Australia, using the water temperature data where the growth data was collected. The results were close, but not perfect as show in Figure 7. These are quadratic equations so it is sometimes not possible to fit them exactly. See Rensel et al. (2015) for another *Seriola* fish species with a fit that was nearly identical to observed.

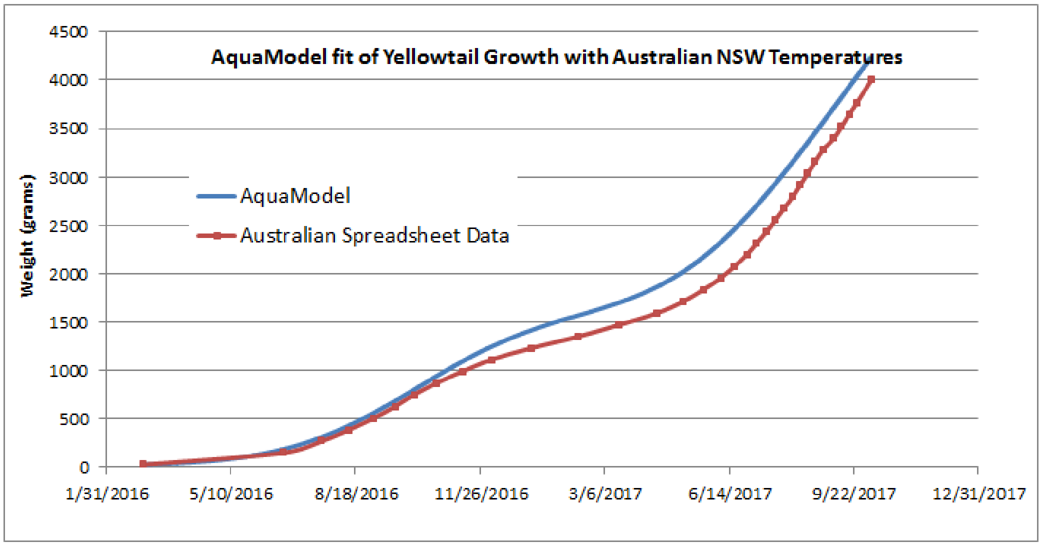


Figure 7. *AquaModel Seriola* growth performance using Australian water temperatures versus actual data from the culture area near Port Lincoln.

The fish growth and food assimilation factors were recorded from an entire culture cycle to assure that FCR values obtained were similar to those expected. This ensured that waste production would be relatively accurate. Finally, the fish submodel was assessed for growth and FCR using the slightly cooler Southern California water temperatures. There are no *S. dorsalis* data from the SCB region to compare to from open ocean pens, and the model can easily be adjusted later when actual data may be available. The average biological FCR for the California yellowtail averaged 1.4 during their growth from 30 grams at stocking to harvest size of about 4 kg.

Shoreline and Bathymetry Data

Shoreline and bathymetric data from the proposed site and surrounding region were provided by NOAA National Ocean Survey. There were 99,985 data points in a format of latitude, longitude and depth relative to MLLW. The data were read into an *AquaModel* wizard and the graphic output was checked for missing area and inconsistencies including unexpectedly deep or shallow point or areas.

Figures 8 shows the color bathymetry imagery with a transect plot from near shore out to 200 meters depth. The slope is very gradual from inshore to offshore and steepening with distance offshore onto the continental shelf. The imagery also shows some of the GIS data for the region, that was brought into the *AquaModel* GIS system.

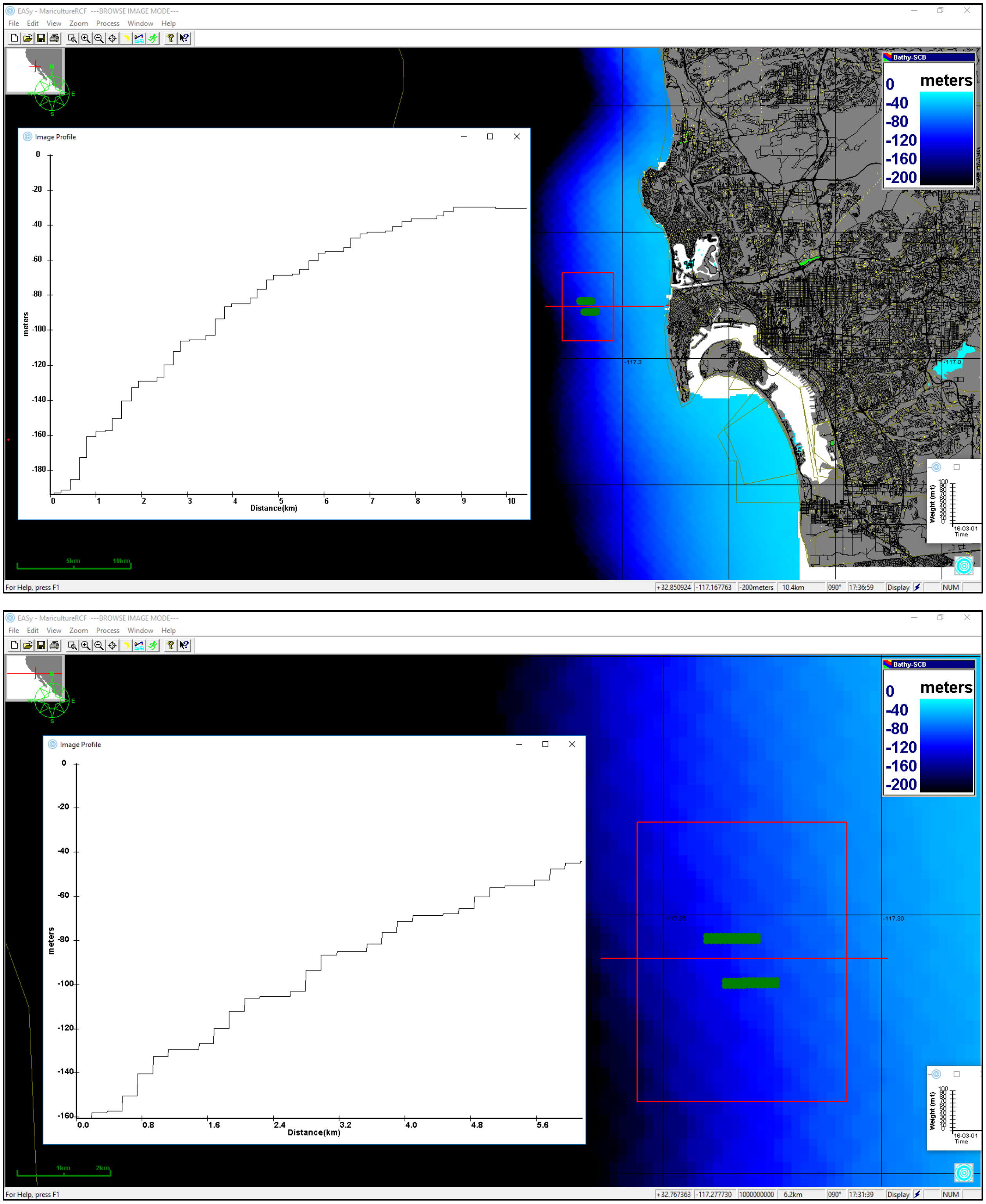


Figure 8. *AquaModel* bathymetry for modeling domain (the red rectangle) and surrounding areas with a profile plot showing depth in meters zoomed out (above) and zoomed in (below).

Water Quality Data

The Southern California Bight is replete with water quality data from many locations. We selected data from the most appropriate geographic locations and timing as discussed below.

The CalCOFI (California Cooperative Oceanographic Fisheries Investigations) hydrographic and plankton data bases were consulted for this project and yielded some general information. However, their sampling cruise tracks were located north of the project site and instead, we accessed data collected and summarized by the Point Loma Ocean Outfall (PLOO, City of San Diego 2015) for the year 2014.

Year 2014 was selected because an analysis of the past 20 years of data indicated that water quality parameters of major interest were the most normal during this year, as shown below in Figure 9. Water temperature and dissolved oxygen anomalies appear less than normal in recent years in this image, but sampling frequency was less than initially since mid-2004, giving the impression of less change in recent years. This decision was made in concert with Dr. Yi Chao, physical oceanographer and formerly of the NASA JPL laboratory in Los Angeles and well-known circulation modeler for the SCB region.

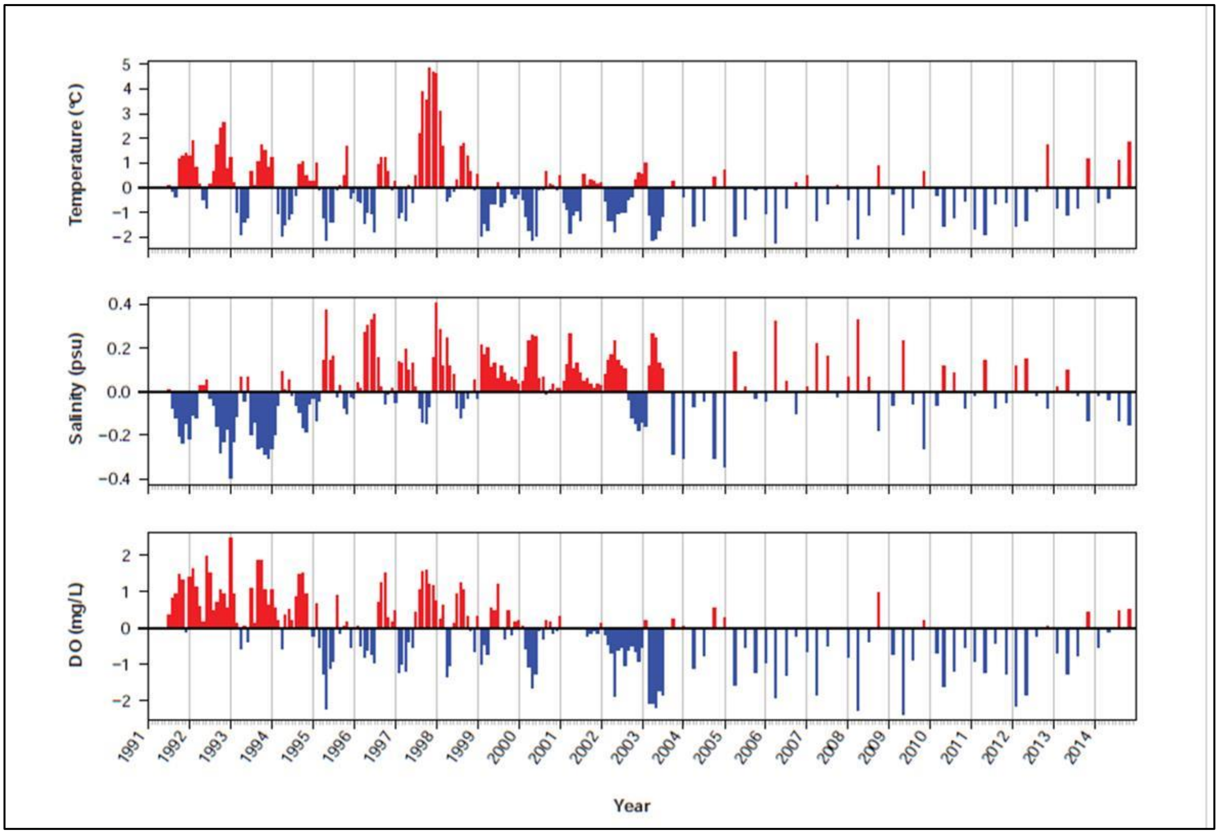


Figure 9. Time series of water temperature, salinity and dissolved oxygen (DO) anomalies from 1991 through 2014 at 11 outfall depth stations sampled in the Point Loma Ocean Outfall (PLOO) region with all depth combined (from PLOO 2015).

Dissolved oxygen anomalies to lower concentrations were prevalent in recent years, but over the past 60+ years have cycled through positive and negative anomaly periods, as explained by McClatchie et al. (2010) and as discussed herein. Water temperatures were very slightly warmer than the average in 2014, but not enough to greatly influence fish growth and waste production algorithms in the model.

Seabottom Sediment Data

To calibrate *AquaModel* for seabottom effects, a single input parameter is required based on average sediment total organic carbon (TOC) concentration. Cores are taken of the top 2 cm of the seabottom and analyzed in a laboratory after treating with acid to remove carbonates such as shell. It is a routine analysis conducted by many laboratories, and in this case was performed at the University of Washington Routine Chemistry Laboratory of the School of Oceanography. This laboratory has advanced equipment and is utilized by many scientists due to its expertise and quality assurance program. Using the TOC average value, the model is calibrated to maintain steady state TOC concentrations in reference, unaffected areas in the modeling array but distant from the fish farm. This method accounts for existing current flows that are put into the model as explained by Rensel et al. (2015) that would otherwise wash TOC away out of the modeling array as has happened with other net pen models (Chamberlain and Stucchi 2007). If an initial model run indicates that a fish farm effects will elevate anaerobic bacteria populations, usually directly beneath the cages, then a second step is performed to calibrate the model for both aerobic and anaerobic benthic community carbon factors.

Sediment quality at the proposed site was sampled by Hubbs SeaWorld Research Institute Staff and provided for this study (M. Shane, pers. comm. 14Mar2015). Regional data for the general locale and region were examined using recent data from the local waste water treatment and discharge authority (PLOO 2015). Sediment grain size data, particularly the percent silt and clay is typically positively correlated with TOC concentration and serves as a check on the validity of the TOC data, along with other common QAQC procedures such as sample splits, field replicates and laboratory spiking procedures. Table 3 summarizes the laboratory data. Separate samples were collected from separate grabs at the same locations and therefore are field duplicates and are expected to vary slightly.

Table 2. Sediment sampling results of 5 September 2014 at and near the proposed RCF project site.

Sampling Date	Sample ID	Relative Location	Time	Lat. (N)	Long. (W)	Depth in Feet MLLW	Depth in Meters MLLW	Total Organic Carbon	% Hydrogen	% Nitrogen
9/5/2014	W1	offshore	1147	32 44.492'	117 20.597'	305	93.0	0.710	0.475	0.097
9/5/2014	W2	offshore	1210	32 44.510'	117 20.619'	305	93.0	0.755	0.515	0.104
9/5/2014	M1	Center	1300	32 44.479'	117 19.906'	271	82.7	0.949	0.602	0.111
9/5/2014	M2	Center	1307	32 44.474'	117 19.886'	271	82.7	0.854	0.544	0.105
9/5/2014	E1	Inshore	1339	32 44.448'	117 19.300'	254	77.5	1.061	0.658	0.132
9/5/2014	E2	Inshore	1352	32 44.464'	117 19.303'	254	77.5	1.006	0.615	0.115

Sampling Date	Sample ID	Free Sulfide (µM)	Redox (mV)	Total Volatile Solids %	Copper ppm	Zinc ppm	% Gravel	% Sand	% Silt & Clay
9/5/2014	W1	0.0	11.5	2.14	5.5	24	0	44	56
9/5/2014	W2	24.4	-4.1	2.13	5.4	24	0	47	53
9/5/2014	M1	19.4	-9.3	2.50	7.0	29	0	38	62
9/5/2014	M2	0.0	83.2	2.23	6.2	28	0	38	62
9/5/2014	E1	6.5	33.7	2.69	7.6	47	0	30	70
9/5/2014	E2	0.0	34.0	2.78	8.0	85	0	22	78

The proposed RCF project site had the following characteristics:

- The mean sediment TOC concentration was 0.9 % (DW) with a SD of 0.1. This result is within the range of expected results for continental slope coastal waters, given the physical sediment grain size distribution.
- The percent silt and clay varied from an average of 54 to 74% and was 64% in the center of the proposed fish farm site.
- Unexpectedly higher TOC concentration occurred at the shallower, nearer shore station vice versa for the deeper, offshore station. This result is likely correct as the TOC results were strongly and positively correlated with percent silt and clay content ($r^2 = 0.86$).
- The TOC and percent silt and clay results are indicative of persistently higher currents in deeper water than shallower water or the center of the proposed fish farm site.
- If site realignment or shift is required for other reasons, moving slightly offshore may make a major difference in carrying capacity of site and yet further reduction of sediment effects
- PLOO (2015) data indicates heterogeneity of sediment TOC and grain size results over similar or larger spatial scales.
- Strong positive correlations were noted for several pairs of sediment data (Table 4) including hydrogen, copper and TVS. Increased heavy metal concentrations are typically positively correlated with coastal sediment TOC and percent silt and clay.
- Total volatile solid (TVS) results (i.e., organic carbon, inorganic carbon and other components such as oxides of nitrogen and salts) were very strongly correlated with sediment TOC ($r^2 = 0.95$), suggesting that the non-TOC fractions are minimal. In inshore waters of the U.S. Pacific Coast at net pen sites, typically the correlation coefficient is less, with a Pearson r^2 averaging about 0.8 (Rensel unpublished routine Puget Sound monitoring data). Despite strong TOC:TVS correlations for these baseline samples, use of TVS as a surrogate for net pen monitoring is not recommended as net pen depositions have a very high percent of labile organic carbon that is subject to fast decomposition versus the more refractile components of TOC within TVS (APHA 2013).
- Sulfide or redox to TOC ratio correlations were very weak as the samples were all from surficial layer of these baseline measurements that are influenced by overlying water column oxygen flux.

Table 3. Pearson correlation coefficients for pairs of sediment parameter from the proposed RCF project site. N = 6 for each parameter.

Comparison	Correlation Coefficients
TOC: Hydrogen	0.99
TOC: Copper	0.96
TOC:TVS	0.95
TOC:Nitrogen	0.92
TOC:Silt/Clay	0.86
TOC: Sand	-0.86
TOC: Zinc	0.67
TOC: Redox	0.20
TOC: Sulfide	-0.13

Sensitivity Analysis of Key Aquamodel Parameters

AquaModel, the modeling software used in this project, is a system of equations that describe growing fish inside of cages in the marine environment. The equations describe the transformation of food stuffs into fish flesh, the production of waste products, and the routes that the wastes move and are transformed, and subsequently assimilated by the food web of aquatic environments, all of which are based on first principals of physics and biology.

To adapt those equations to different environments and various cultured fish, model configuration parameters that are input settings are necessary to modify the rates of the key system equations. For example, waste solids deposited onto a seafloor comprised of hard, flat rock will erode or wash away much more easily than the same materials falling onto a seafloor with coarse and rugose surface, assuming the near-sea-bottom flow rates are the same. Therefore, the correct parameter settings are needed to adapt the model equations to either of those environments to reflect differing erosion rate properties.

Many of the model configuration parameters can be measured directly from *in situ* or laboratory observations, such as the water-column settling velocity of uneaten food or fecal materials. Other parameters are more difficult to measure such as the percentage of food that is missed by the fish and falls through the cage. Waste feed rate is commonly estimated by all large-scale fish growers by estimating the food conversion ratio or feed used to fish flesh grown. For modern salmon aquaculture it is typically in the range of 2% to 3% as widely discussed in the literature and is limited in most all cases by the use of underwater cameras in the cages that are monitored at all feeding times.

But what happens in the planning phase of a farm when only limited observations of the site environment have been measured or the farm operational measurements do not yet exist, and what is the relative sensitivity or influence of each parameter? Would there be large changes in the predicted output(s) for small changes in the parameters? How sensitive are the modeled responses to variations in the configuration parameters? These are the questions we address herein.

This sensitivity analysis quantifies the percent change in the predicted results of several output parameters for a 1 % change in select configuration or input parameters. A fundamental goal of modern, sustainable net pen aquaculture is to avoid exceeding the assimilation rate of waste materials by the aquatic food web. That goal is the overarching reason for using computer models to select sites and evaluate their effects. Confidence in these computer models is enhanced by exploring which configuration settings need to be set with the most precision and which configuration settings do not cause much deviation in the predictions.

Because there are no existing data for the effects of a proposed fish farm, before operation, sensitivity analysis uses a “base case” simulation as the reference point to compare all input settings. The base case is the most likely parameter settings found for similar, open ocean fish farm locations that have been assessed through *AquaModel* use (e.g., Rensel et al. 2015). The base case, best solution was configured using parameters from reviews of limited literature sources and from experience of *AquaModel* configurations of other open ocean sites that were actively sampled and validated.

To maintain comparable units of measurement across all of the different configuration and observed parameters, we ran the model with systematic percent changes in configuration settings then compiled the percent change of each key parameter relative to the base case settings and results.

Beginning with the base case for each input parameter in Table 1 as the starting point, 45 separate *AquaModel* simulations were executed and recorded while systematically changing the configuration parameters one at a time. Nine years of modeling involved 30 minute time steps of the model for a total of 175,200 increments and approximately 52 million internal (within the model) increments for each of the model grid partitions.

Model configuration (input) parameters were adjusted +/- 10 and 20% of the base case while recording the effect on the output parameter. In this way, a value for the sensitivity, or “gain” was calculated (Table 4). Output parameter gain was calculated by dividing the percent change in an observed parameter output by the percent change in the configuration input parameter. For example, if a +13% change of sediment total organic carbon (TOC) was recorded from a -10% change in the fecal erosion rate factor then the sensitivity gain would be -1.3. This tells us that for every +1% change in the fecal erosion rate factor, there would be a -1.3% change in the predicted Sediment TOC, if all other conditions and parameters remained unchanged.

Table 4. Schedule of sensitivity testing of key *AquaModel* calibration parameters illustrating the range of settings for each primary input parameter.

Configuration Parameter (Inputs)		Units	Description	Test Lower		Base Case	Test Upper	
			Model Setting +/- Base Case →	**-20%**	**-10%**	**--**	**10%**	**20%**
1	**Feed Waste Rate**	% of total food fed	Fish feed fed, but not eaten by the cultured fish[1/]	2.4	2.7	3	3.3	3.6
2	**Fish Fecal Waste Settling Velocity**	cm/s	Fish fecal settling velocity thru seawater towards the seabottom	2.4	2.7	3	3.3	3.6
3	**Waste Feed Waste Settling Velocity**	cm/s	Waste feed settling velocity thru seawater toward the seabottom	7.2	8.1	9	9.9	10.8
4	**Fecal TOC Sediment Consolidation Rate**	fraction/day	Fraction fish fecal TOC retained in seabottom sediments per day	0.008	0.009	0.01	0.011	0.012
5	**Feed TOC Sediment Consolidation Rate**	fraction/day	Fraction waste fish feed TOC retained in seabottom sediments per day	0.008	0.009	0.01	0.011	0.012
6	**Fish Fecal Deposition Threshold Velocity**	cm/s	Velocity below which material lands on sediment	4.8	5.4	6	6.6	7.2
7	**Waste Feed Deposition Threshold Velocity**	cm/s	Water velocity below which waste feed falls	6.4	7.2	8	8.8	9.6
8	**Fish Fecal Erosion Threshold Velocity**	cm/s	Velocity above which material is transported	6.4	7.2	8	8.8	9.6
9	**Waste Feed Erosion Threshold Velocity**	cm/s	Velocity above which material is transported	8	9	10	11	12
10	**Fish Fecal Erosion Rate Factor**	grams $C/m^2/day$	Grams carbon resuspended from the sediments when the threshold velocity is attained	0.4	0.45	0.5	0.55	0.6
11	**Waste Feed Erosion Rate Factor**	grams $C/m^2/day$	Grams sediment carbon resuspended above threshold velocity	0.4	0.45	0.5	0.55	0.6

Observation Locations

The entire simulated fish farm is comprised of a grid of 100 m square cells numbering 45 cells by 60 cells for 2,700 total cells. The simulation therefore covers an area of 4.5 km by 6 km. To record the 7 observed parameters in each of the 2,700 simulation cells plus recording at the 40 operational pen locations at each 1 hour reporting step, would have generated an output file approximately 19,140 recording cells wide and 87,000 rows long. This size of output file would have been unmanageable to manipulate or store. To reduce the volume of the step-wise captured data while preserving the results, 33 output recording locations were specified that would represent the effects of the fish farm on various water or sediment quality parameters, see Figure 11. The output locations were positioned in the center of the farm, directly under the corners of the farm cages, at concentric spacing at 500 m around the center of the farm, and at locations that could more closely illustrate the interactions of a fish farm and the surrounding environment.

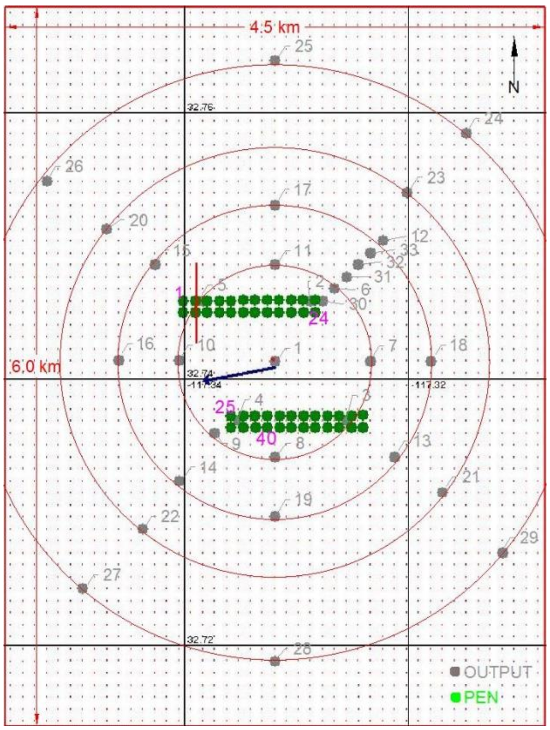


Figure 10. RCF AquaModel fish farm layout and data output locations.

The recording scheme of defining the 33 output locations was selected as representative since it compiled results from points directly under the pens, around the pens, and incrementally further away to far-field points. In this experiment, the summed or integrated values from each of the seven observed parameters, at all 33 output locations, with no set threshold, over the nine year simulation produced a single number that represented the simulation's predicted effect across the entire farm. For example, the value of Sediment Total Sulfide (STS) from all 33 output locations, at every one-hour step, for 24 hours per day, over nine years, were summed together to produce the single STS signal. This method produced a large "signal" at all scheduled input settings that changed relative to the configuration parameter settings. This single value result is therefore the "currency" of the simulation results for the sensitivity analysis. The accumulated signal was obtained for each of the seven observed parameters in the base solution and became the simulation values to which all subsequent simulation results were compared.

It is important to note that the sensitivity analysis was not a search for the most accurate configuration since the base case simulation remains the best solution to predicting the farm's interaction with the environment at this planning phase. The sensitivity analysis was conducted to discover which of the configuration settings would cause the largest change in the predicted output. A small change in a single parameter that would precipitate a large change in the results requires much more scrutiny and validation effort once real effects have been measured, than a parameter that has little effect on the results. As a consequence, we can be more confident in model settings that display a lower sensitivity gain than those that produced higher values.

Moreover, the sensitivity analysis is site specific. In a low-flow environment, unlike the RCF site, some settings that displayed high gains here would have little effect or lower gains at the other site, owing to the mechanics of water velocity calculated in *AquaModel*.

Sensitivity Analysis Results

The results of the sensitivity analysis were condensed as shown in Table 2. The values within each cell of Table 2 are the percent change in an observed parameter for a one (1) percent change in the configuration parameter. Values in red indicate the four highest values within in a column of output parameter gain.

Overall the sensitivity analysis indicates that changes in most fish *fecal* configuration parameters, the lower rows in the table, were generally more influential than the fish *feed* configuration parameters in their effects on the model output parameters. This conclusion is drawn by noticing that higher gain values were prevalent in many more categories of output for fish feces (lower 5 rows) compared to waste feed (upper 6 rows). The influence of the parameters describing fish fecal movement occurs at lower water velocities than for waste feed. As a result, changes in settings describing fecal movement will change predicted outputs more than those of waste feed and produce a higher response gain. The exception was for the sediment feed *loading rate*, but this measure is not nearly as important as sediment *TOC fraction* model results. As explained above, sediment feed or fish fecal loading rate is only the measure of the amount of waste falling to the bottom at a given location before the feed is resuspended and moved across the bottom for enhanced food web assimilation so that the sediment oxygen demand effects are not necessarily expressed at the point of impact.

Table 5. Sensitivity gains for waste fish feed and fish feces effects with the greatest four values in each column highlighted in red. Higher gain equates to higher sensitivity of the configuration inputs.

		MODEL PARAMETER GAIN (OUTPUTS)							
	MODEL CONFIGURATION PARAMETER (INPUTS)	Sediment Fecal Loading gC/m²/day	Sediment Feed Loading gC/m²/day	Sediment Oxygen mg/L	Sediment Total Sulfide µM	Sediment TOC fraction	Sediment TOC Rate gC/m²/day	Suspended Oxygen mg/L	Sum of Higher Rankings by Rows
WASTE FISH FEED	WASTE RATE (fraction of total)	-0.03	2.52	-0.003	1.48	0.001	0.169	0.000	2
	WASTE SETTLING VELOCITY (cm/s)	-0.01	0.87	-0.002	0.38	0.000	0.067	0.000	1
	TOC CONSOLIDATION RATE (fraction of total/day)	0.00	-0.50	-0.001	0.02	0.000	0.000	0.000	0
	DEPOSITION THRESHOLD (cm/s)	-0.04	3.20	-0.005	1.96	0.000	0.000	0.000	1
	EROSION THRESHOLD (cm/s)	-0.07	6.77	-0.012	2.80	0.001	0.000	0.000	1
	EROSION RATE FACTOR (g_C/m²/day)	0.02	-1.88	0.003	-0.87	0.000	0.000	0.000	1
FISH FECES	WASTE SETTLING VELOCITY (cm/s)	1.50	-0.12	-0.017	4.49	0.003	0.363	-0.001	6
	TOC CONSOLIDATION RATE (fraction of total/day)	-0.68	0.00	-0.004	-0.02	0.002	0.000	0.000	0
	DEPOSITION THRESHOLD (cm/s)	2.30	-0.14	-0.030	5.57	0.003	0.000	-0.002	5
	EROSION THRESHOLD (cm/s)	4.18	-0.18	-0.064	6.25	0.005	0.000	-0.004	5
	EROSION RATE FACTOR (g_C/m²/day)	-1.57	0.06	0.024	-2.34	-0.002	0.000	0.001	5

Sensitivity Analysis Interpretation

These results should not be interpreted to mean that fish feces are more important that waste feed on determination of fish farm effects on seabottom conditions. Chamberlain and Stucchi (2007) modeled and measured effects of both categories of wastes for a moderate sized salmon farm in British Columbia and demonstrated that near the farm, waste feed was a dominant contributor to sediment organic carbon and sulfides effects.

The sensitivity analysis herein indicates that fish fecal configuration settings have a larger relative effect on model output parameter gain than those for waste feed. This result gives guidance for future use of models and the need for additional laboratory or field studies to study fish fecal deposition and resuspension dynamics. Although there has been considerable study of fish fecal settling rates, there is only a scant amount of data concerning the fate of fish feces after it deposits on the sea bottom and more inference is drawn from other types of particulate wastes (e.g., Cromey et al. 2002a).

Waste feed settling velocities are well documented and published as a specification by various research efforts but fecal settling velocities can vary as the fish grow from fingerling to harvest size. The values used in the model prediction for fecal settling velocity are for adult fish with larger fecal pellet masses as a worse case. This may be an area where relatively little software coding can result in improved model performance in dynamic models like *AquaModel* that are frequently revised and updated, although the present approach provides a conservatively high effects estimate

The deposition and erosion threshold velocities and rates are less well understood or measured. Cromey et al. 2002 used mostly non-aquaculture references to choose values for their modeling with Depomod. The values we used have been found experimentally to produce better *AquaModel* simulation outcomes, but by no means are known to represent the exact values. Although it would be beneficial to conduct more applied research regarding these parameters, it is also a legitimate method to calibrate and tune a model based on modeled versus measured results, and then use these settings for other farms as long as the siting and benthic conditions are comparable.

Table 2 indicates that the most sensitive output parameter to the changes in the inputs was the prediction of sediment sulfide in units of micro-molarity (microgram atoms per liter, or simply µM). Sulfide production will generally increase in the presence of enhanced TOC when the oxygen carrying flow declines to relatively slow velocity. The faster the flow, the less likely a site is to experience elevated sulfide production. This observation is seen in the *AquaModel* interactive screen when stepping through a simulation playback and in the output data records of the sensitivity simulations. As water current decreases during neap tidal series, sulfide production would spike because the oxygen rich flow of water ceased and sediment conditions quickly became oxygen deprived. As soon as the flow returned, aerobic bacteria respond quickly and sulfide production ceased. Those low flow periods of high sulfide production responded to the increased benthic deposition of particulate organic wastes which were influenced by changes in the model settings. As a result sulfide production responded sharply as a result of the parameter settings, producing a higher sensitivity gain.

AquaModel has been deployed at various sites around the world and validated/calibrated through sampling and adjustment of model parameters. With a calibrated model the effects of operating the fish farm are more accurately forecast. The parameter values used in the base case of this sensitivity analysis were established though those validations at existing fish farm sites as well as corroborated in published literature when available. A similar sensitivity analysis using *AquaModel* was conducted at an open ocean site in Hawaii, completed in 2015 (Rensel et al. 2015). This RCF project analysis had the advantage of using model settings that had been refined at that similar site.

A valuable function of model sensitivity analysis at any proposed net-pen site is to enhance the ability of an analyst to know how much, on a percent basis, to change a configuration setting when calibrating or validating a model. If benthic sediment samples during fish farm operation indicate a difference from the predicted modeling output for a known set of circumstances, then the amount that the configuration settings have to be changed is known and this helps avoid expensive and time consuming trial and error. Such efforts are necessary and consistent with the *AquaModel* approach of calibrating and tuning to fit each major type of habitat within a specific ecoregion (Rensel et al. 2007, Rensel et al. 2015).

This sensitivity test suggests that for the RCF sites and similar locations in the Southern California Bight, that perhaps the most influential features of net cage fish farm modeling and siting are the settling rates of wastes, the near sea bottom water velocity threshold of deposition and the resuspension speeds. As mentioned above, this was more prevalent for fish feces than waste fish feed, as shown in Table 2, but if the farm had been shallower and in less flow we would have seen more effect on the waste feed output gain results.

Model Results and Discussion

Water and sediment quality effects data from many locations within the modeling grid were collected as discussed above in the *Sensitivity Analysis* section of this report. Here we report results using the best possible model settings, referred to as the "base settings".

Benthic Effects Model Results

Total organic carbon (TOC) in fish feces and waste feed is the principal fish farm waste constituent affecting the sea bottom nearby such facilities. With *AquaModel* use, benthic effects of a fish farm are gauged by several methods including tracking the change in concentration of sediment total organic carbon (TOC). Excess TOC produced by a fish farm may accumulate on the sea bottom from waste feed and fish feces and can cause an increase in oxygen demand as bacteria assimilate the wastes. Ideally, the fish farm waste TOC can be fully assimilated by the existing food web without any major perturbations; this is one of the objectives of the computer modeling process, to predict the benthic TOC concentrations. A fish farm can then be re-sized or reconfigured within the model until the ideal operation is achieved. In this way the fish farm is designed to minimize any adverse benthic effects.

TOC requires oxygen from the water column and sediments as it is respired by aerobic bacteria and other food web organism respiration. A minor amount of TOC flux to the sea bottom may have no disruptive effect on benthic species composition and may actually help increase species diversity and abundance (Pearson and Rosenberg 1978 and subsequent relevant publications). In some cases higher up components of the food web including fish and marine birds may benefit (Katz et al. 2002, Rensel and Forster 2007, Callier et al. 2013). Excessive rates of TOC deposition to the seabottom without subsequent resuspension can cause of shift to anaerobic bacteria dominance. These bacteria do not use oxygen as the electron receptor in organic matter respiration[1/].

Sediment Sulfide

Sediment sulfide in surficial seabottom sediments of the baseline sampling for the proposed RCF project site were near zero where detected (Table 2). Considering the range of effects normally observed in poorly-located net pen sites, these values were within normal margin of error for the sulfide "probe" method that was used. See appendix A for an explanation of sulfide measurements and modeling and how the methodology used in *AquaModel* is highly conservative.

The RCF modeling results indicated a very slight increase in sulfide concentration of the surficial sediments during occasional low current flow conditions in areas underneath or adjacent to the fish farm. As current flows increased, sulfide concentrations quickly returned to zero when sea floor flow returned. Extreme maximum sulfide concentration was predicted to be 535 millimoles per m^3 (equivalent to µM units) sulfide, significantly below sediment performance standards for fish farms in other countries that use this metric as a performance standard. The presence of low but measurable concentration of sulfides in the sediment typically lasted for less than a day. Increase in sulfide concentrations were not observed beyond 100 meters from the cage array at any time. These results corroborate the results of the sediment TOC analysis to indicate only minor alteration of these sediment chemical parameters during full build out stage of the proposed project.

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[1] Anaerobic organisms respire using electrons that are shuttled to an electron transport chain with oxygen as the final electron acceptor. Oxygen is a strong oxidizing agent and efficient acceptor. When sediments shift to anaerobic bacterial dominance from increased TOC flux other less-efficient oxidizing substances such as sulfate (SO_4^{2-}), nitrate (NO_3^-), sulphur (S), or fumarate are used. These terminal electron acceptors have smaller reduction potentials than O_2, meaning that less energy is released per oxidized molecule. Anaerobic respiration is considered energetically less efficient than aerobic respiration.

Sediment Total Organic Carbon

The background, ambient concentration of TOC near the proposed RCF project is approximately 0.01% of the total dry weight mass of sediment materials. For the RCF proposed project using the optimum (base) case model settings, the sediment TOC concentration increased to a maximum of 0.012% of total mass directly beneath the fish farm, a 0.002 fractional increase. Such minimal TOC increases have been found to have no adverse biological effects on the benthos at other temperate water fish farms with modest flow velocities. The RCF study site is best characterized as having strong flow velocities throughout the water column and then do decline near the seabottom but cumulatively allow for wide waste particle dispersion. The maximum projected 0.002 fraction value increase is within normal, background variation found within 100m of the site at present, with no fish farm operating.

With *AquaModel* use, benthic effects of the fish farm are gauged by several methods including tracking the change in concentration of sediment total organic carbon (TOC). Excess TOC produced by a fish farm may accumulate on the sea bottom from waste feed and fish feces and can cause an increase in oxygen demand as bacteria assimilate the wastes. Ideally, the fish farm waste TOC can be fully assimilated by the existing food web without any major perturbations; this is one of the objectives of the computer modeling process, to predict the benthic TOC concentrations. A fish farm can then be re-sized or reconfigured within the model until the ideal operation is achieved. In this way the fish farm is designed to avoid or minimize adverse benthic effects.

As shown in Figure 12, near the end of nine years of operation the level of sediment TOC is projected to increase only one tenth of one percent beneath and immediately adjacent to the cages compared to background or baseline conditions. This is a very minimal fractional increase of 0.001, as shown in the Figure 12. Such changes will not adversely affect seabottom invertebrate infauna (i.e., bivalves, crustaceans, polychaete worms, etc.) but instead will likely increase the diversity and abundance of such organisms as discussed below.

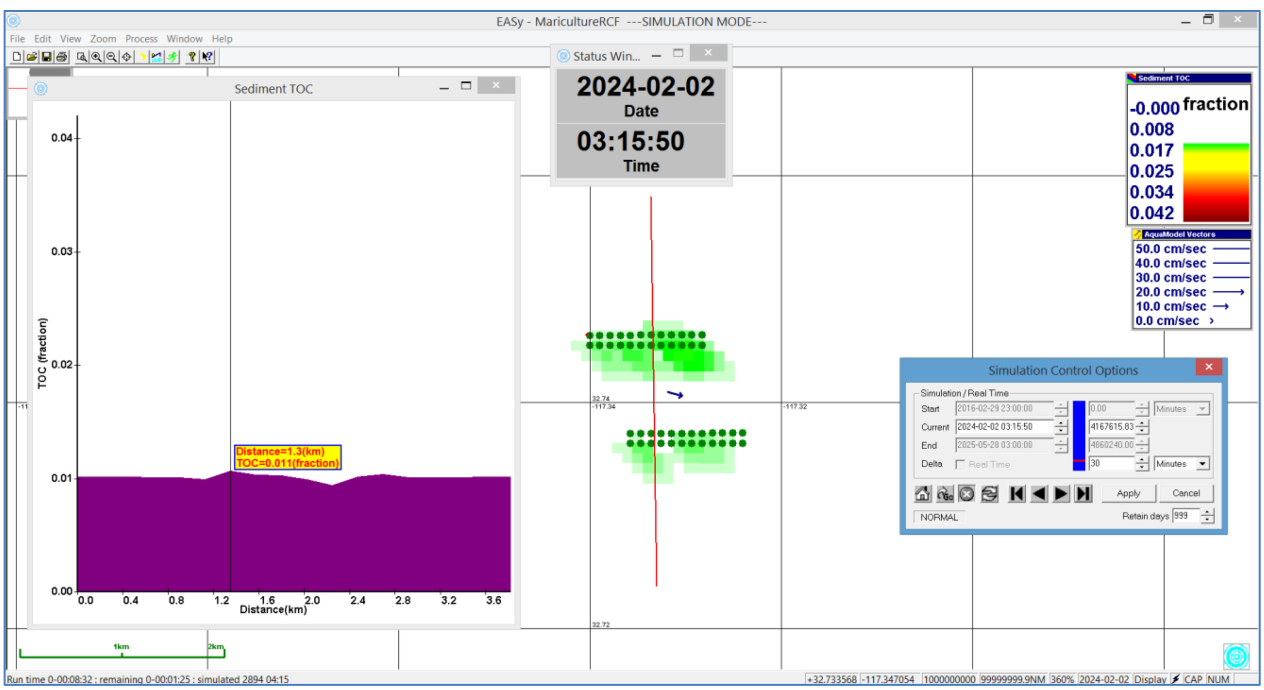

Figure 11. Projected fractional increase of total organic carbon on seabottom near proposed fish farm of 0.001 (1/10th percent) after nine years of operation.

Particulate organic carbon fish farm wastes are highly labile and processed relatively rapidly by sediment bacteria and other organisms up to a certain threshold loading rate. In other temperate water locations, a carbon flux rate threshold of 1 gram per square meter per day (g C m^{-2} d^{-1}) has been used as a general index of the maximum allowable loading rate to avoid sediment hypoxia and adverse changes in the benthic community composition (Hargrave 2008). In locations of weak currents, this threshold would occur much sooner than in high current locations for similarly sized farms operated with the same fish species and practices.

Sediment hypoxia causes a gradual shift from aerobic bacteria dominance with their use of oxygen and respiration of carbon dioxide to dominance by anaerobic bacteria and the concurrent respiration of sulfide that may form hydrogen sulfide gas that has a toxic fraction that increases principally with lower pH and with a lesser effect of different water temperature. Invertebrate infauna including crustaceans and filter (suspension) feeding molluscs are typically affected first and these species generally disappeared entirely when rates increase to >5 g C m^{-2} d^{-1} (Hargrave 2008). Some types of polychaete worms are more tolerant to the changes and a few opportunistic species thrive unless carbon loading rates are excessively high, when sediment anoxia and azoic conditions may occur. Processing of organic carbon and other wastes may be further impeded at high carbon loading rates as many or all of the burrowing invertebrate infauna is extirpated so that beneficial bioturbation of the bottom is reduced or eliminated (Pearson and Rosenberg 1978, Martinez-Garcia et al. 2015). These occurrences were recorded at some early net pen sites in the 1970s, but are rare or non-existent for large scale corporate aquaculture at the present time.

The minimal sediment TOC change that are predicted by *AquaModel* for the RCF site is due to the relatively low loading rates of particulate waste carbon that is further enhanced by frequent seabottom particle resuspension. Together they should prevent a shift to sediment anaerobesis from the organic matter deposition and decomposition.

Other aquaculture models such as Depomod have been used to evaluate these effects but have been used with resuspension effects turned off as at that time that model was not capable of maintaining background TOC conditions (e.g., Chamberlain and Stucchi 2007, DFO 2012, Chang et al. 2014). For sites that have moderate to strong current flows, resuspension of waste particles has a profound effect and cannot be ignored. However, we avoid focusing on "TOC rate" (i.e., the rate of organic particles first hitting the seabottom) as the carbon will not be respired at any one location but is resuspended and further transported and aerated. The *AquaModel* sediment TOC concentration routine integrates three processes including carbon deposition, resuspension and bacterial processing of the wastes over time in order to predict expected concentration of organic carbon in the sediments. The process is truly highly dynamic and analysis of such processes is not achievable through use of any one or two of the individual modeling components separately. To do so would be to overstate the impact rates at significantly higher levels than actually occur.

Sediment Dissolved Oxygen

Sediment dissolved oxygen (DO) has rarely been measured in the past within sediment interstitial spaces during marine biological studies, particularly in the deep ocean, due to the fragility of the glass probes and other technical problems. More commonly, sediment respiration has been measured with enclosures placed over the seabottom and various treatments added to partition the component effects. With the recent introduction of optical fluorescence DO probes this is rapidly changing and the technique has shown to be much less variable than electrochemical probes such those used for sediment redox measurements (1.6% versus 9.1% standard deviation, respectively in Neill et al. 2014). Because *AquaModel* is a mass balance and dynamic model, it includes sediment oxygen flux estimates that involve diffusion of oxygen from the sediment/water interface and use of oxygen by an aerobic benthic community that competes with an aerobic benthic community.

Although not measured routinely, sediment DO is often modeled to assess the effects of other types of discharges, such as municipal waste treatment and discharge systems (EPA 2015). Surficial sediment oxygen is invariably less than overlying water column dissolved oxygen, even in substrates that are coarse sand with strong currents such as on the coastal shelf of the Island of Hawaii.

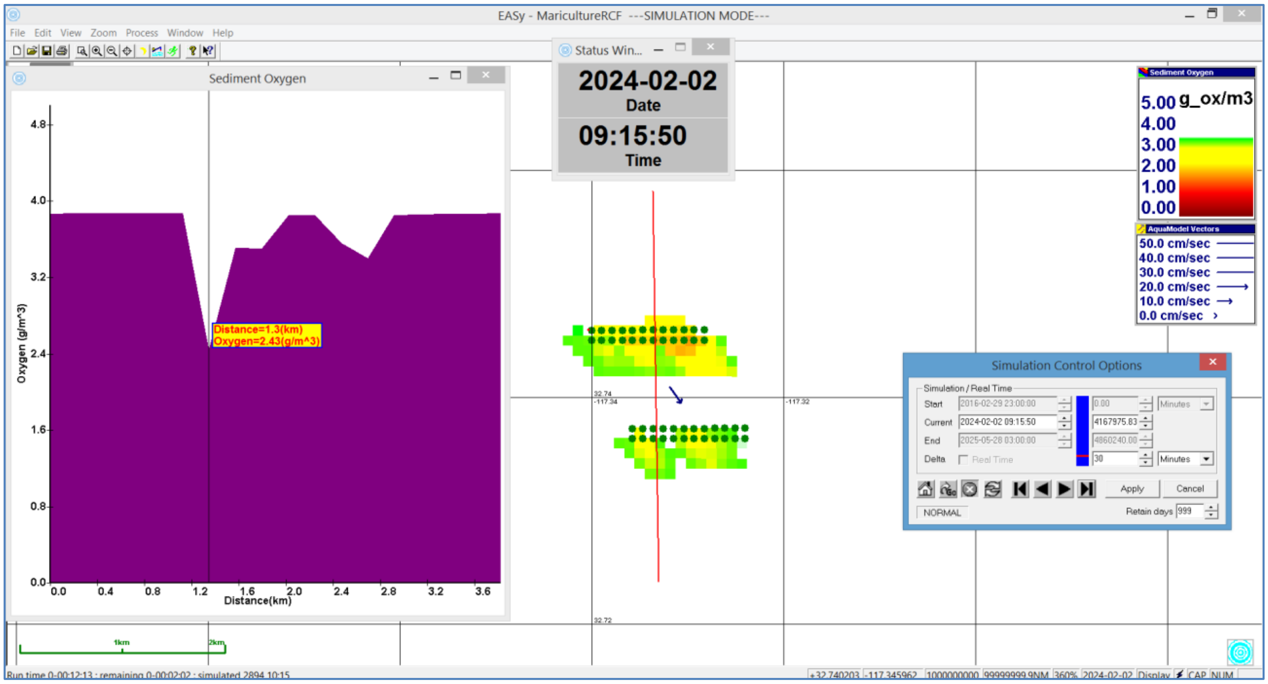

Figure 12. Typical projected sediment dissolved oxygen in seabottom near proposed fish farm during February after the fish farm reached full production and during the time of year when ambient dissolved oxygen is lowest.

Like the other sediment constituents, *AquaModel* focuses on the surficial layer where the abundance of benthic infauna invertebrates is often greatest. Ambient sediment dissolved oxygen (DO) in the RCF simulations typically ranged between 3.3 and 4.6 mg/L for background conditions, following the DO concentration and flux of ambient seawater directly above the seafloor. Ambient bottom oxygen was field measured at 3.8 mg/L in winter months. For fish farm affected sediment modeling, the lowest singular sediment DO was calculated as 1.86 mg/L at a bottom flow of 0.95 cm/s and lasted only 1 hour before the current increased and DO returned to average values. More typical later winter minimum values are shown in an *AquaModel* screen print (Figure 14).

Background concentrations of deepwater dissolved oxygen are seasonally low in the SCB, due to seasonally-extreme upwelling of deep ocean water. This affects interstitial sediment dissolved oxygen and there has been decadal scale oscillation of this factor in the past. There now appears to be noncyclical changes associated with climate variability affecting dissolved oxygen (DO) content of ocean waters across the Eastern Pacific according to leading scientists in peer-reviewed published studies. The trends and effects are complex but more pronounced in deeper waters (>600m) of the continental shelf trending from permanently hypoxic to anoxic (Chan et al. 2008) than at shallower depths of the proposed fish farm site (Booth et al. 2014). While DO concentrations in the Southern California bight are seasonally depressed every year, particularly in spring and summer, the benthic species composition has therefore been selected for such conditions, they must be adapted to these changes. The effects are more pronounced in deeper waters than at the depths of the proposed fish farm site.

Given the above and with the goal of reducing environmental impacts, no matter how slight, siting of open-ocean farms in the SCB is best performed by selecting sites with the largest possible distribution of particulate wastes to allow food web assimilation that limits further oxygen drawdown. In the present study there was a projected drawdown of dissolved oxygen in sediments underneath the cages shown by the model, but not in adjacent areas. The level of sediment dissolved oxygen in the model simulations mostly remained above hypoxic (2 mg/L) levels and is a seasonal, not annual condition and thus the effects, if any, will be muted. The model was purposely setup with conservative settings, including no use of waste particulate matter by zooplankton or wild fish species as the particles sink for long periods to the distant seabottom. In the Mediterranean Sea, these are significant factors in reducing waste loading of the bottom below fish cages (e.g., Vita et al. 2004) and it probably will occur in the SCB, based on what is known about wild fish fecal wastes in the SCB region. Stable isotope sampling of sediments near and away from fish farms in the Mediterranean Sea also suggest wild fish consumption could account for a wider than expected spread of (δN) nitrogen isotope signals, but current resuspension and transport of wastes are also a contributing factor (Sara et al. 2004).

Water Quality Model Results

Analysis of water quality results indicate that average surface (12 meters and shallower) water velocity of the 9 months current meter record from the proposed site ranged from 18 to 20 cm/sec. With a 26 m diameter cage, the total water volume is therefore exchanged completely every 2.6 minutes, allowing for a large supply of dissolved oxygen during average flow rates.

In fish culture the most significant consideration involves flow rates to maintain dissolved oxygen during minimal flows. The concentration of dissolved oxygen and thus the flux is more than sufficient to provide oxygen supply as the minimal flows were short term and infrequent with the 10% probability frequency of 5.4 cm/sec. Such a flow rate slightly exceed the threshold average rate of 5 cm/s previously recommended for salmon farms (SAIC 1986). The 1% probability of lowest flows was 0.5 cm/s, that suggests that stocking density will have to be within normally accepted ranges to avoid stressing the fish. Our AquaModel simulations indicated that dissolved oxygen concentrations in fish-occupied cages never became less than 6.3 mg/L in any of the cages in the 175,290 individual estimates calculated for this analysis over 10 years. These results suggest that the project site will not require aeration equipment that is increasingly common at many commercial net pen sites worldwide. Decreases in DO was not detectable during medium and high current flow periods and at no time will have an even a minor adverse effect on natural biota.

Excreted ammonia from the cultured fish will be rapidly diluted to immeasurable values near and inside the cage and will have no adverse effect on either the cultured fish or downstream biota. Ammonia is rapidly converted to nitrate in any oxygen rich aquatic environment by naturally occurring bacteria and other fauna. Ammonia toxicity to aquatic organisms increases with higher pH (more basic than occurs in the sea) and warmer water temperatures and is mostly a concern in warm freshwater habitats. These factors are well understood for the proposed project area both in the published literature (e.g., Eppley et al. 1979) and in technical analysis of deepwater municipal wastewater discharges (e.g., PLOO 2015)

The highest observed dissolved nitrogen concentration (i.e., nitrate + nitrite + ammonia) was calculated to be 1.1 mg-at/m^3 (0.015 mg/L). This is 0.008 mg/L above the background of 0.5 mg-at/m^3 (0.007 mg/L) observed during low surface flow periods inside of a cage. If we assume that all of the 0.008 mg/L is ammonia nitrogen, and the temperature is high at 20°C with a high pH of 8.2, then the toxic unionized fraction would be 3.36% or 0.0003 mg/L (Emerson et al. 1975), orders of magnitude below EPA 30 day limits of 0.69 mg/L or the 6 month limit of 0.60 mg/L for this area (Table T2, PLOO 2015). This increase in nitrogen concentration from the cages is diluted within a few tens of meters of the cage array depending on current direction and velocity.

Cumulative effects of all the cages at peak fish production times on phytoplankton production are not possible as the fish farm total nitrogen production is massively dwarfed by natural flux of nitrogen in seawater. Thus plankton flowing through the project area will not experience any biological perturbation from the project. This project included preparation of a far-field version of AquaModel that can be used to estimate the effects of numerous fish farms, should they be planned or occurred.

Sediment Conditions after Farm Removal

The model was also used to forecast how fast all traces of the fish farm would disappear from seabottom sediments if all the fish were harvested at one time, while allowing the modeling processes to continue normally. Sediment TOC and sulfide concentrations returned to background levels within two weeks of fish removal and there were no effects whatsoever forecast by the three week point. These forecasts are consistent with several recovery rate field studies conducted in British Columbia that illustrated that faster recovery for fish farms located in areas with minimal accumulation of sediment TOC and faster near-seabottom currents.

Multiple Fish Farm Model of Southern California Bight

A separate far-field version of *AquaModel* used elsewhere to assess cumulative water column effects of multiple fish farm sites was developed as part of this project.

This software involves the use of a sophisticated Regional Ocean Modeling System (ROMS) circulation system for the San Diego region of the Southern California Bight. The data was provided by Dr. Yi Chao of Remote Sensing Solutions, Inc. and consists of hourly time step data for 3 km resolution current vectors at several depths throughout the array. These data were processed and imported into Far-Field *AquaModel* to allow for the assessment of the transport of waste nitrogen, assimilation by phytoplankton and grazing by zooplankton over large geographic areas should additional fish farms ever be proposed. This version of *AquaModel* was designed to be used to evaluate eutrophication of coastal waters and has been used in several locations in different types of ecoregions from S.E. Asia, the Arabian Sea and in the waters of the Pacific Ocean near the Island of Hawaii.

Software coding was completed to utilize this circulation data so that multiple fish farms in the Southern California region could be assessed with regard to their effects on nitrogen, phytoplankton and zooplankton. The single fish farm modeled in this project would not have any measurable effect on these factors given the strong flux of water and nutrients in the California current, but the *AquaModel*3D program is available for assessment of cumulative effects of future fish farms and is being used overseas in other countries at present.

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Appendix A. Sediment Sulfide Details

Sediment sulfides measurements are often conducted for aquaculture monitoring but there has been considerable debate and controversy about it for several years. This appendix explains the importance of sulfides as an effect, but how *AquaModel* does not rely on this parameter as a fundamental component of its structure and operation.

There are two types of sediment sulfide analyses often conducted. More commonly the "probe" method is used as it is a relatively simple *in situ* analysis but is subject to large amounts of variation both in replicate samples and over short periods of time (weeks and days). It only measures "free sulfide" concentration, i.e., that fraction that is not tied up with metals. In aquaculture monitoring it is universally applied only to the shallowest surface strata of the sediments and involves processing of the samples that are a time and method-sensitive consideration. The variance is associated with the fact that free sulfides are highly volatile and that measurement techniques and the electrochemical probe itself have been shown to produce highly variable results. Ongoing controversy and concerns about accuracy of the standard analytical methods used in some countries exist that have been shown to produce unacceptably high bias of the results (e.g., orders of magnitude, Brown et al. 2011). More recent work in Atlantic Canada has shown that background concentrations vary as much as 20% over a single week at the same location (Chang et al. 2014) with the possibility of some significant spatial variation. Recent efforts have been made to improve the process through sampling and processing protocol improvement (Keimowitz et al. 2016).

The other method less commonly used is a laboratory method known at "total sulfide" that includes all forms of sulfide including free and complexed with iron and other metals. It is also known as "acid volatile sulfides" (AVS). Total sulfide sampling has shown complicated patterns for deeper sediment layers that vary significantly with depth within the sediment strata and with changes in TOC loading associated with particulate organic matter deposition as well as the sediment grain size distribution and metals present. Total sulfide measurement requires special sealed glass bottles and handling protocols and it is the free sulfide that is responsible for toxicity to infauna, not the complexed total sulfides.

AquaModel simulates the biologically-important upper 2 cm of the seabottom sediments and produces estimates of total sulfide. For background conditions for sandy-mud sites like the proposed RCF project site, it is probable that there would not be any detectable sulfide in the surficial sediments and that any that exists would be unbound by metals due to the availability of dissolved oxygen that fluxes from the water column into the sediments. Therefore, for aquaculture impacted sediments, *AquaModel* as presently structured will produce highly conservative sulfide results, i.e., total sulfides estimates that will be larger than free sulfide measurements that are carefully collected and processed immediately using the most accurate protocols available. *AquaModel*'s sediment modeling is based on organic carbon as the fundamental cause of sediment perturbation, not sulfides.