Accepted Manuscript

Factors driving low oxygen conditions in integrated rice-shrimp ponds

Luu Duc Dien, Le Huu Hiep, Stephen John Faggotter, Chengrong Chen, Jesmond Sammut, Michele Astrid Burford



PII: S0044-8486(19)30390-4

DOI: https://doi.org/10.1016/j.aquaculture.2019.734315

Article Number: 734315

Reference: AQUA 734315

To appear in: aquaculture

Received date: 21 February 2019
Revised date: 11 June 2019
Accepted date: 16 July 2019

Please cite this article as: L.D. Dien, L.H. Hiep, S.J. Faggotter, et al., Factors driving low oxygen conditions in integrated rice-shrimp ponds, aquaculture, https://doi.org/10.1016/j.aquaculture.2019.734315

This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

FACTORS DRIVING LOW OXYGEN CONDITIONS IN INTEGRATED RICE-SHRIMP PONDS

Luu Duc Dien^{a,b,1}, Le Huu Hiep^b, Stephen John Faggotter^a, Chengrong Chen^a, Jesmond Sammut^c, Michele Astrid Burford^a

Abstract

Integrated rice-shrimp ponds (IRSPs) are common in areas of Southeast Asia where saltwater intrudes into rice fields in the dry season, enabling rice production in the wet season, and shrimp farming in the dry season or throughout the year. Previous research has highlighted that IRSPs have periods of low dissolved oxygen concentrations which may have a critical effect on shrimp survival. To understand the causes of low dissolved oxygen, this study examined oxygen fluxes at two IRSPs in Ca Mau Province, Vietnam during a two-year period (two wet seasons and two dry seasons). Sediment oxygen demand (SOD) incubations and whole-system oxygen flux measurements were conducted and compared with a range of water and sediment parameters to explain drivers for low oxygen concentrations. A high percentage of oxygen demand at a whole pond scale was from the sediment; hence SOD drove low oxygen concentrations in the water column. SOD rates were significantly positively correlated with chlorophyll a concentrations in the water column. These findings suggested that algal production in the water column, rather than benthic algal production, or other organic loading, provided an organic carbon source driving SOD. Oxygen demand was much higher than oxygen production within the IRSPs, indicating high bacterial activity and low algal production. This study has shed new light

^a Australian Rivers Institute and School of Environment & Science, Griffith University, 170 Kessels Road, Nathan, Queensland 4111, Australia

^b Research Institute for Aquaculture No.2, 116 Nguyen Dinh Chieu Street, District 1, Ho Chi Minh City, Vietnam

^c Centre for Ecosystem Science, The School of Biological, Earth & Environmental Sciences, The University of New South Wales, Sydney 2052, Australia

¹ Corresponding author. Griffith University, 170 Kessels Road, Nathan, Queensland 4111, Australia. Tel.: +61-73735-7711; E-mail address:ducdien.luu@griffithuni.edu.au

on the importance of SOD in driving oxygen drawdown in IRSPs and improving shrimp survival requires new management approaches to reduce the negative effect of SOD.

Key words: chlorophyll *a*; dissolved oxygen; organic carbon; oxygen production; sediment oxygen demand.

1 Introduction

In the coastal zone of the Mekong Delta, Vietnam, where saline water intrusion in the dry season is a major constraint to agricultural production, many farmers farm rice and shrimp in integrated rice-shrimp ponds (IRSPs). This practice has occurred for over 40 years (Hung, 1992). Farming shrimp in shallow ponds, canals, ditches and rice fields has provided traditional rice farmers with an extra income during the dry season. The rice with shrimp rotation is believed to increase environmental sustainability and contributes to food security (Be et al., 2003). Shrimp metabolites are thought to benefit the rice crop through increased nutrient availability. Rice cultivated after the shrimp harvest utilizes and absorbs any excessive organic loads that may affect the shrimp negatively (Burford et al., 2004; Nhan et al., 2011). Additionally, the low salinity may destroy shrimp pathogens (Phong et al., 2003).

During the dry season, rice stubble provides a substrate for natural food for the shrimp. The integration of shrimp aquaculture with rice cultivation has been proposed as an environmentally-benign way to boost aquaculture production (Frei and Becker, 2005). However, the productivity of rice-shrimp ponds is generally low and unreliable, i.e. the average shrimp productivity of the rice-shrimp system ranges from 0.25 to 0.30 t ha⁻¹ crop⁻¹ with a stocking density of one to two post larvae m⁻² (Brennan et al., 2002; Preston et al., 2003). This limits the wealth creation for farmers. Hence, many questions have been raised about the environmental sustainability of the rice-shrimp model and solutions are needed to improve the socio-economics of rice-shrimp farmers in areas along the coast (Be et al., 1999).

Sediment oxygen demand (SOD) is the sum of the dissolved oxygen removed from the water column by the respiration of organisms living in the sediment, decomposition of organic matter, and the oxidation of reduced chemicals found in the sediment (Abhilash et

al., 2012; Liu et al., 2009; Sommaruga, 1991). SOD is often considered an indicator of the health of the system because sediment structure remains relatively stable while the overlying water conditions can be transient (Chen et al., 2012; Park and Jaffa, 1999; Todd et al., 2009). Therefore, SOD measurement is an important part of water quality investigations of waterbodies (such as riverine systems, aquaculture ponds), and is an important parameter that must be integrated into numerical simulations of water quality (Akomeah and Lindenschmidt, 2017; Liu and Chen, 2012).

Previous studies in IRSPs have demonstrated periods of high nutrient loads and low DO concentration (Dien et al., 2018; Leigh et al., 2017). The high frequency of occurrence of low DO concentration in aquaculture ponds generally and, particular in rice-shrimp ponds during the growing season, is directly related to the high rates of respiratory demand for oxygen. The major losses of oxygen in a static water pond system are due to respiration by shrimp, plankton, sediment microbes and benthic animals (Burford and Longmore, 2001; Casillas-Hernández et al., 2007; Zhong et al., 2015). However, the processes driving low oxygen concentrations in the IRSPs are not well-understood. This information is needed to guide management actions.

Therefore, the objective of this study was to determine oxygen fluxes in IRSPs and the key controlling factors. This is a critical step in developing management strategies to improve water quality and shrimp production.

2 Materials and methods

2.1 Study area

This study focused on two IRSPs, TB1 and TB2, in Thoi Binh District, Ca Mau province, Vietnam (Fig. 1). Rice-shrimp ponds have a platform (80% of area) for rice growing, and a surrounding ditch (20% of area) for water management and shrimp farming. IRSPs ranged in size from 1.5 to 2.0 ha, with water depth in the ditch typically 1.0–1.4 m, and for the platform 0.1–0.4 m.

There are two main seasons in the region: the dry season from December to April, and the wet (rainy) season from May/June to October/November (Leigh et al, 2017). The hottest period is typically between April and May, while the wettest period is from September to

October/November. During the dry season, water salinities range between brackish and seawater conditions, while in the rainy season, salinity typically drops gradually from around 30 to 2, and may become fresh (ADB, 2013; Tho et al., 2006). The air temperature ranges from 24 to 34°C, and the mean monthly rainfall ranges from 0 mm in the dry season to 250 mm in the wet season, with an annual rainfall of around 2300 mm (ADB, 2013).

2.2 Pond management and harvesting

The first step in preparation for the IRSP for this field trial involved cleaning and reinforcing the pond bank, discharging the water in the ditch through a sluice gate, and transferring accumulated sludge from the ditch to the platform. After that, lime was added to the pond (using calcium oxide, a dose of 1 t ha⁻¹ ditch⁻¹ and 500 kg ha⁻¹ platform⁻¹), then the platform was dried for 5–7 days. Approximately 15–20 days later, water was added from the canal, via a filter bag to remove unwanted organisms, provided the salinity of the supply channel was suitable (> 10). Unwanted fish were killed using Rotenone, and water was disinfected using iodine added at 0.5 mg L⁻¹ (Boyd, 1995).

IRSPs were stocked with *P. monodon* with a mean density of 1.5 to 2.0 post larvae m⁻². Stocking occurred every 3–4 months during the two-year period over both the wet and dry season. Mud crabs (*Scylla paramamosain*) were also stocked in the IRSPs at an estimated density of 0.5 crab m⁻². No formulated feed was added. Rice were planted at a density of 4.0 kg 1000 m⁻². Rice were grown through to the end of the dry season and harvested in December with short-term varieties (OM429, OM18) or January with long-term variety (Lun Kien Giang).

Trap nets with a large mesh size (12 mm) were used for partial harvests frequently throughout the year to ensure that only shrimp greater than 20 g were caught and removed from the ponds. At the end of the year, the ponds were drained, and additional shrimp were harvested using trap or seine nets. In addition to the shrimp, mud crabs were occasionally harvested.

2.3 Water sampling and analysis

Water temperature and salinity in the two IRSPs were measured daily throughout each year

using calibrated loggers (Thermocron ibuttons, Odyssey conductivity probes), and logging at two-hour intervals. Physico-chemical parameters, i.e. temperature, salinity, pH and DO, were measured in the ditch (around 0.5 m from the surface) throughout 2016 and 2017 by using a calibrated Hydrolab Quanta multi-parameter probe. Light attenuation was measured through the water column using a Licor LI-250A with a 4 pi-sensor. This was used to calculate euphotic depth.

During the two-year period from 2016 to 2017, water samples for nutrients and other parameters were collected, while SOD incubations were undertaken twice each year (in both dry and wet seasons). For nutrient analyses, water samples from the surface were taken at five different locations in the ditch using a 1.0 L bottle. Each bottle of water was poured into one bucket to make a composite sample (three replicates) from which subsamples for various analyses were taken. For chlorophyll *a* analyses, known volumes of water were filtered through glass fibre filters (Whatman GF/F with nominal pore size 0.7 µm, 2.5 cm diameter). Filters were stored on ice until frozen at -20°C before being analyzed. A similar sampling method was applied for ash free dry weight (AFDW) and total suspended solid (TSS) parameters.

Two replicate water samples were taken and frozen at -20° C for measurements of total organic carbon (TOC), total nitrogen (TN), total phosphorus (TP) and sulfide. For ammonium nitrogen (NH₄-N), nitrite nitrogen (NO₂-N), nitrate nitrogen (NO₃-N), soluble reactive phosphate (PO₄-P), total dissolved nitrogen (TDN) and total dissolved phosphorus (TDP) analyses, a subsample was taken and filtered through a 0.45 μ m membrane filter (Sartorius, 2.5 cm diameter), then the filtrate was frozen until analyzed.

NH₄-N was analyzed using the phenate method; NO₂-N, NO₃-N using cadmium reduction and sulphanilamide method; PO₄-P using the ascorbic acid method, and TP, TDP, TN and TDN using the persulfate method followed by colourimetric analyses (APHA, 2005). Concentrations of chlorophyll *a* were determined by acetone extraction of the glass fibre filters and spectrophotometric measurements following APHA methods (2005). TOC was determined following APHA methods (2005) using the chromic acid rapid titration method. Sulfide concentrations were measured through the methylene blue method using a UV-VIS Thermo Spectronic Helios Alpha spectrophotometer (APHA, 2005). For AFDW and TSS samples, filters were defrosted and dried in an oven at 60°C for 24 h (APHA,

2005).

2.4 Sediment nutrient and carbon sampling and analysis

Sediment samples were taken twice in 2016 (once in the dry season and once in the wet season) by collecting five cores (up to 15 cm depth) in the ditch, and five cores in the platform from each pond. All sediment samples were oven-dried at 60° C for 24 h, pulverized to pass through a 0.25 mm mesh screen, and analyzed for TN, TP and TOC (APHA, 2005). The method for sediment chlorophyll a was the same as for water column chlorophyll a.

2.5 Sediment oxygen demand

During a two-year period from 2016 to 2017, SOD experiments were conducted pond side twice each year (both in the dry season and wet season).

Intact cores from both ditch and platform were collected by wading onto the platform or from a boat (in the ditch) with the pole corer. For each sediment incubation, 12 cores were taken from two ponds ([three replicates in the ditch + three replicates in the platform] x two ponds). The incubations were conducted using the method of Kaiser et al. (2015). Overlying water was removed to avoid sediment resuspension during sampling and cores were transported to an incubation site adjacent to the ponds. Prior to running the incubations, all cores were equilibrated for about 8 hours with around 450 mL of filtered rice-shrimp pond water to ensure water saturation of the pore space.

Prior to commencing measurements, cores were completely filled with rice-shrimp pond water above the cores to remove any gas space and sealed without disturbance of the sediment surface. The cores were incubated in a container (a nally bin) with constantly flowing pond water to maintain the same temperature as the pond. The water velocity within cores was controlled by a 12-V pump at 2–3 cm sec⁻¹, the lowest velocity that resulted in a linear decline in DO concentration over time (Miskewitz et al., 2010; Murphy and Hicks, 1986). This maintained aerobic conditions at the sediment-water interface but did not suspend fine clay particles (Renaud et al., 2007). Sediment cores were kept at an *in situ* temperature of 30°C.

Optical oxygen sensors (Presens) were used to measure DO concentrations over time to identify trends. DO concentrations were read every one or two hours during the daytime, and every three hours during night time over 24 h.

We also measured the whole pond oxygen demand (i.e. total oxygen demand – TOD) using calibrated YSI multi-parameter probes (YSI ProODO Optical) placed in the ditch (around 0.4 m from the water surface) and platform (0.1 m from the surface) of each pond. DO was logged at 15-minute intervals over time (24 h). The results were calculated by linear regression of concentrations over time for both night and day times to determine oxygen demand and production. In the final results, SOD rates at 30°C were adjusted to SOD rates at 20°C (Rong et al., 2016).

2.6 Data analysis

Data were analyzed using R Studio and SPSS 20.0 for Windows. The SOD and TOD rates were compared between years (2016 and 2017) and seasons (dry and wet) using analysis of variance (ANOVA) followed by Duncan's multiple range tests for post hoc comparisons to determine the significant differences. The normal distribution of the data and homogeneity of variances among the treatments were verified before conducting ANOVA. Relationships between SOD and parameters in the water column (eg. salinity, chlorophyll a, DO), and sediment (chlorophyll a, TOC, TN, TP), were examined using linear regression models. The strength of the correlations was established using Pearson's correlation coefficient (r) and their p-values.

3 Results

3.1 Production

Overall, shrimp survival was less than 50%, with shrimp survival in 2016 having a mean of 21.4% compared with 41.1% in 2017. Shrimp yield in 2017 was approximately two times higher than that of 2016 (536 and 296 kg ha⁻¹, respectively) (Table 1). In addition to the shrimp harvest, there was also a significant crab harvest from these two IRSPs, where mean crab yield ranged from 87 (in 2016) to 166 (in 2017) kg ha⁻¹. Rice harvests (only on

the platform) ranged from 1550 to 3040 kg ha⁻¹.

3.2 Physico-chemical parameters

Water temperatures typically ranged from 29 to 35°C in the dry season, while from 26 to 32°C in the wet season. Salinity ranged from 2 to 25. The water temperature nearly reached 40°C over the period of April to May 2016. The salinity was more than 35 during April to May 2016, and for a few days, the salinity was more than 40 (Fig. 2). The mean salinity in 2016 was nearly three times higher than that in 2017 (22.4 compared with 8.9, Table 2).

The mean ammonium concentration during the experiments in the dry season was 0.148 mg L^{-1} , this was approximately half of the wet season mean during experiments (0.287 mg L^{-1}) (Table 2). The concentration of nitrite in the wet season was approximately four times higher than that of the dry season (0.039 and 0.011 mg L^{-1} respectively). In contrast, the mean concentration of nitrate in the dry season was 0.232 mg L^{-1} , more than three times higher than that in the wet season (0.067 mg L^{-1}). The mean concentration of phosphate was approximately 0.115 mg L^{-1} in both the dry and the wet season. However, despite the differences in values, the high variability meant that there were no major differences (p > 0.05) in concentrations in nutrient parameters in the wet and dry seasons.

The euphotic depth ranged from 1.2 to 1.6 m in the dry season, and between 0.6 and 0.8 m in the wet season (Table 2). There were no major differences (p > 0.05) in concentrations of a range of nutrient parameters in the wet and dry seasons. This was because concentrations varied widely between ponds, sampling sites, and sampling occasions.

Mean chlorophyll a concentration in the water column in the dry season was 12.3 μ g L⁻¹, which was only 25% of the wet season value (55.8 μ g L⁻¹) (Table 2). This was the opposite result to the benthic chlorophyll a concentrations which were 2.95 \pm 1.54 mg m⁻² in the dry season, substantially higher than that in the wet season (0.16 \pm 0.09 mg m⁻²) (Table 3).

Sediment nutrients and organic carbon concentrations in the sediment were also very variable with no obvious differences (p > 0.05) between sites within ponds or between seasons (Table 3).

3.3 Sediment oxygen demand

The DO concentrations in sediment cores typically decreased during the experimental period, from around 9.0 mg L⁻¹ at 10:00:00 h (10 am) to close to zero during the night before increasing again during the morning as algal oxygen production (OP) increased (Fig. 3).

The calculated SOD rate in the dry season was statistically lower (p < 0.05) than that in the wet season (i.e. 1.42 and 2.33 g m⁻² d⁻¹ respectively, Fig. 4). The SOD rates for the dry and wet seasons accounted for around 60% and 80% of the TOD, respectively (Fig. 4). The mean OP rate in the dry season was 0.63 g m⁻² d⁻¹, which was approximately half than that in the wet season (1.19 g m⁻² d⁻¹), and the OP rates were only 45–50% of the SOD rates (Fig. 4). SOD rates were higher in 2016 (p < 0.05) than that of 2017 (2.21 and 1.48 g m⁻² d⁻¹, respectively) (Fig. 5). Across years and ponds, the ditch and platform were not significantly different (p > 0.05) (Table 4).

3.4 Factors driving SOD

SOD rates and a range of parameters in the water column (Table 5) and sediment were correlated (Table 6). SOD increased with chlorophyll a concentrations (0.875, p = 0.002), and AFDW (0.656, p = 0.039), and decreased with mean salinity (-0.692, p = 0.028) (Table 5).

There was a significant negative correlation between SOD and both TN and TP in the sediment (p < 0.05) (Table 6).

4 Discussion

This study was the first to determine oxygen fluxes in IRSPs and showed that SOD was the dominant oxygen flux process in the system, accounting for around 70% of whole pond oxygen demand (i.e. TOD). SOD can play a primary role in the water quality of a stream system, including affecting biota via low oxygen conditions (Matlock et al., 2003; Rutherford et al., 1991; Wallace et al., 2016). IRSPs have been shown to have periods of low oxygen with detrimental effects on shrimp production (Leigh et al., 2017). Our study has identified the role of sediment in driving oxygen concentrations in the water column. Oxygen production, indicative of primary production, was low by comparison.

SOD rates in our study were comparable with other aquaculture systems. The mean SOD rate of our study was similar to that of shrimp ponds in China (2.09 g m $^{-2}$ d $^{-1}$) (Yao et al., 2000). Similar SOD was reported in *Penaeus vannamei* ponds, i.e. 24.18 to 202.69 mmol m $^{-2}$ d $^{-1}$ (or 0.91 to 6.46 g m $^{-2}$ d $^{-1}$) (Zhong et al., 2015). In contrast, the SOD in settlement ponds of a small-scale recirculating shrimp farm (*Penaeus monodon*) in rural Thailand was around 0.67 ± 0.21 g m $^{-2}$ d $^{-1}$ (Erler et al., 2007) which was a tenth of the values in this study. Conversely, rates in semi-intensive, brackish-water shrimp ponds in the Philippines (12.31 g m $^{-2}$ d $^{-1}$) (Fast et al., 1988) and commercial channel catfish ponds were higher than in our study (4.43 and 8.61 g m $^{-2}$ d $^{-1}$ respectively, Berthelson et al.,1996; Steeby et al., 2004). These differences were due to many factors such as the stocking density, the technique and management, and feeding regime.

Carbon availability from phytoplankton appeared to drive SOD, based on the positive correlation between water column chlorophyll a and SOD. The mechanism is likely to be that senescing and settling phytoplankton provide carbon to the benthic microbial community (Golde et al., 1994; Wang et al., 2008). When microalgae, containing chlorophyll a, die and settle on the pond bottom, they are broken down by bacteria (Burford et al., 2004). The chlorophyll a concentration in 2017 was significantly lower than that in 2016 (p < 0.05), at the time of doing the study, and was likely to have reduced the available carbon. Therefore, the major higher chlorophyll a concentrations in 2016 may have led to the higher SOD in 2016 (compared to those in 2017).

Light had the potential to be limiting primary production at some times when the euphotic depth in the ditch was less than the water depth in the ditch (Havens, 2003). In the dry season, light availability was higher, but OP, as a measure of primary production, was lower than the wet season. This suggests that light was not the driving factor for primary production rates. In the case of the platforms, the water was very shallow (around 20–30 cm), hence light availability was very unlikely to be a controlling factor.

The correlation of SOD with salinity suggests salinity changes between the wet and dry seasons are likely to disrupt biogeochemical processes in the system. This may directly affect microbial activity, depending on the organisms present and their salinity tolerances, or indirectly via limits to algal production (Gantzer et al., 2009; Nhan et al., 2011). Most algal species are not adapted to growth in both low and high salinity water, and this is

likely to negatively affect productivity within the whole system (Liu et al., 2007; Pal et al., 2011).

Shrimp survival and shrimp yield in 2017 were approximately two times higher than those of 2016. The main reasons for differences in shrimp survival and yield were likely to be high water temperature, high salinity and SOD rates in 2016. In 2016, very high water temperatures and salinity prevailed in the dry season creating extreme conditions for shrimp farming. Dry season rainfall was at a record low, causing a severe drought and salt water intrusion surpassing that seen in the past 100 years. This was mainly due to the 2015–2016 El Niño Southern Oscillation (ENSO) phenomenon, the second most severe since 1965 (Thirumalai et al., 2017). These conditions were known to be a major stress factor for shrimp (Leigh et al., 2017). Additionally, SOD was also likely to have contributed to low shrimp survival since high SOD rates reduced water column DO concentrations.

Similarly, the rice production in 2016 was just as a half as that of 2017. High salinity was considered the primary risk factor for rice (Leigh et al., 2017; Nhan et al., 2011). Therefore, the higher salinity in IRSPs in 2016 resulted in the failure of the rice-crop.

Oxygen fluxes, particularly SOD, appear to be a cause of low shrimp survival in IRSPs, hence for the sustainability of the system. Accordingly, to improve conditions for shrimp, interventions must address the SOD problem. For example, changes to farm design and water management, and application of probiotics to increase oxygen concentrations in the water column could be investigated. Other strategies may involve the reduction of nutrients, for example, by removing sludge from the bottom layer of the ditch (Dien et al., 2018). Managing salinity changes, especially during extreme climatic conditions, in the system remains a significant challenge and could be improved by using reservoirs as buffers to variable salinity. This option might be impractical due to the need to sacrifice or acquire additional land, and the pressure on existing shared freshwater resources in a delta.

5 Conclusions

This study found that a high percentage of oxygen demand at a whole pond scale was from the sediment; hence SOD drove low oxygen concentrations in the water column. SOD rates

were significantly positively correlated with chlorophyll *a* concentrations in the water column. These findings suggested that algal production in the water column, rather than benthic algal production, or other organic loading, provided an organic carbon source driving SOD. Additionally, oxygen demand was much higher than oxygen production within the IRSPs, indicating high bacterial activity and low algal production. The measured OP rate in the dry season was approximately half of that in the wet season, and accounted for approximately 44% and 51% of the SOD, respectively. Likewise, the mean SOD rate in the dry season was statistically lower than that of the wet season, and accounted for around 60% and 80% of the TOD. Low oxygen concentrations can have a major effect on the growth and survival of shrimp, hence improving shrimp survival requires management strategies focusing on reducing SOD, particularly in the wet season.

Acknowledgements

Luu Duc Dien's PhD research was supported by an ACIAR-funded John Allwright Fellowship and Griffith University. The other authors were supported by an Australian Centre for International Agricultural Research (ACIAR) grant (SMCN/2010/083). We would like to thank Graeme Curwen for producing Figure 1, Tran Minh Thien, Nguyen Minh Duong, Le Van Truc for field and laboratory assistance, and the farmers in Thoi Binh District, Ca Mau province, Vietnam for their generously allowing access onto their integrated rice-shrimp ponds.

References

- Abhilash, K.R., Raveendran, T.V., Limna Mol, V.P., Deepak, M.P., 2012. Sediment Oxygen Demand in Cochin Backwaters, a tropical estuarine system in the southwest coast of India. Mar. Environ. Res. 79, 160–166.
- ADB, 2013. Climate Risk in the Mekong Delta: Ca Mau and Kien Giang Provinces of Vietnam. Asian Development Bank, 145 pp.
- Akomeah, E., Lindenschmidt, K.E., 2017. Seasonal Variation in Sediment Oxygen Demand in a Northern Chained River-Lake System. Water 9(4), 254. doi:10.3390/w9040254
- APHA, 2005. Standard Methods For The Examination Of Water And Wastewater, 21st Ed. American Public Health Association, Washington D.C., USA.
- Be, T.T., Clayton, H., Brennan, D., 2003. Socioeconomic characteristics of rice-shrimp farms in the study region. In: Preston, N., Clayton, H. (Eds), Rice-shrimp farming in the Mekong Delta: biophysical and socioeconomic issues. ACIAR Technical Reports No. 52e, pp. 15–26.
- Be, T.T., Dung, L.C., Brennan, D., 1999. Environmental costs of shrimp culture in the rice growing regions of the Mekong Delta. Aquac. Econ. Manag. 3(1), 31–42.
- Berthelson, C.R., Cathcart, T.P., Pote, J.W., 1996. *In situ* measurement of oxygen demand in catfish ponds. Aquac. Eng. 15, 261–271.
- Boyd, C.E., 1995. Bottom Soils, Sediment and Pond Aquaculture. Chapman & Hall, New York, USA. 348 pp.
- Brennan, D., Preston, N., Clayton H., Be, T.T., 2002. An Evaluation of Rice-Shrimp Farming Systems in the Mekong Delta. In: Report Prepared Under the World Bank, NACA, WWF and FAO Consortium Program on Shrimp Farming and the Environment. Work in Progress for Public Discussion. Published by the Consortium. 10 pp.
- Burford, M.A., Longmore, A.R., 2001. High ammonium production from sediments in hypereutrophic shrimp ponds. Mar. Ecol. Prog. Ser. 224, 187–195.
- Burford, M.A., Preston, N.P., Minh, T.H., Hoa, T.T.T., Bunn, S.E., Fry, V.M., 2004.

 Dominant sources of dietary carbon and nitrogen for shrimp reared in extensive rice-shrimp ponds. Aquac. Res. 35, 194–203.

- Casillas-Hernández, R., Nolasco-Soria, H., García-Galano, T., Carrillo-Farnes, O., Páez-Osuna, F., 2007. Water quality, chemical fluxes and production in semi-intensive Pacific white shrimp (Litopenaeus vannamei) culture ponds utilizing two different feeding strategies. Aquac. Eng. 36(2), 105–114. doi:10.1016/j.aquaeng.2006.09.001
- Chen, W., Liu, W., Huang, L., 2012. Measurement of sediment oxygen demand for modeling the dissolved oxygen distribution in a Subalpine lake. Int. J. Phys. Sci. 7(27), 5036–5048. doi:10.5897/ijps12.412
- Dien, L.D., Hiep, L.H., Hao, N.V., Sammut, J., Burford, M.A., 2018. Comparing nutrient budgets in integrated rice-shrimp ponds and shrimp grow-out ponds. Aquaculture, 484, 250–258. doi:10.1016/j.aquaculture.2017.11.037
- Erler, D., Songsangjinda, P., Keawtawee, T., Chaiyakam, K., 2007. Nitrogen dynamics in the settlement ponds of a small-scale recirculating shrimp farm (*Penaeus monodon*) in rural Thailand. Aquac. Int. 15, 55–66. doi:10.1007/s10499-006-9068-8
- Fast, A.W., Carpenter, K.E., Estilo, V.J., Gonzales, H. J., 1988. Effects of water depth and artificial mixing on dynamics of Phillippines brackishwater shrimp ponds. Aquac. Eng. 7, 349–361.
- Frei, M., Becker, K., 2005. Integrated rice-fish culture: Coupled production saves resources. Nat. Resour. Forum 29, 135–143.
- Gantzer, P.A., Bryant, L.D., Little, J.C., 2009. Effect of hypolimnetic oxygenation on oxygen depletion rates in two water-supply reservoirs. Water Res. 43(6), 1700–1710. doi:10.1016/j.watres.2008.12.053
- Golde, H.M., Crosby, M.P., Lovell, C.R., Stancyk, S.E., 1994. Reduction of marine sediment oxygen uptake: pre-treatment for the determination of infaunal respiration rates. Mar. Ecol. Prog. Ser. 111, 203–207.
- Havens, K.E., 2003. Submerged aquatic vegetation correlations with depth and light attenuating materials in a shallow subtropical lake. Hydrobiologia, 493, 173–186.
- Hung, L.T., 1992. Integration of crustacean aquaculture with coastal rice farming in Vietnam. *Naga* ICLARM Q. 15(2), 27–29.
- Kaiser, D., Kowalski, N., Böttcher, M.E., Yan, B., Unger, D., 2015. Benthic Nutrient Fluxes from Mangrove Sediments of an Anthropogenically Impacted Estuary in Southern China. J. Mar. Sci. Eng. 3, 466–491. doi:10.3390/jmse3020466.
- Leigh, C., Hiep, L.H., Stewart-Koster, B., Vien, D.M., Condon, J., Sang. N.V., Sammut, J.,

- Burford, M.A., 2017. Concurrent rice-shrimp-crab farming systems in the Mekong Delta: Are conditions (sub)optimal for crop production and survival? Aquac. Res. 00, 1–12. doi: 10.1111/are.13338.
- Liu, W., Au, D.W.T., Anderson, D.M, Lam, P.K.S., Wu, R.S.S., 2007. Effects of nutrients, salinity, pH and light: dark cycle on the production of reactive oxygen species in the alga *Chattonella marina*. J. Exp. Mar. Biol. Ecol. 346, 76–86.
- Liu, W., Chen, W., 2012. Monitoring sediment oxygen demand for assessment of dissolved oxygen distribution in river. Environ. Monit. Assess. 184, 5589–5599.
- Liu, W., Chen, W., Kimura, N., 2009. Measurement of Sediment Oxygen Demand to Simulate Dissolved Oxygen Distribution: Case Study in the Main Danshuei River Estuary. Environ. Eng. Sci. 26(12), 1701–1711. doi:10.1089/ees.2009.0132.
- Matlock, M.D., Kasprzak, K.R., Osborn, G.S., 2003. Sediment Oxygen Demand in the Arroyo Colorado River. J. Am. Water Resour. Assoc. 39, 267–275.
- Miskewitz, R.J., Francisco, K.L., Uchrin, C.G., 2010. Comparison of a novel profile method to standard chamber methods for measurement of sediment oxygen demand. J. Environ. Sci. Health A Tox. Hazard. Subst. Environ. Eng. 45(7), 795–802. doi:10.1080/10934521003708919
- Murphy, P.J., Hicks, D.B., 1986. *In-situ* method for measuring sediment oxygen demand. In: Hatcher, K.J. (Ed.), Sediment oxygen demand: Processes, Modelling, and Measurement. Institute of Natural Resources, University of Georgia, Athens, Georgia, pp. 307–322.
- Nhan, D.K., Trung, N.H., Sanh, N.V., 2011. The impact of weather variability on rice and aquaculture production in the Mekong Delta. In: Stewart, M.A., Coclanis, P.A. (Eds.), Environmental Change and Agricultural Sustainability in the Mekong Delta. Springer Advances in Global Change Research, 45, pp. 437–451.
- Pal, D. Khozin-Goldberg, I., Cohen, Z., Boussiba, S., 2011. The effect of light, salinity, and nitrogen availability on lipid production by *Nannochloropsis* sp. Appl. Microbiol. Biotechnol. 90, 1429–1441. doi: 10.1007/s00253-011-3170-1.
- Park, S.S., Jaffa, P.R., 1999. A Numerical Model to Estimate Sediment Oxygen Levels and Demand. J. Environ. Qual. 28, 1219–1226.
- Phong, N.D, My, T.V, Nang, N.D, Tuong, T.P., Phuoc, T.N, Trung, N.H, 2003. Salinity dynamics and its implications for cropping patterns and rice performance in rice—

- shrimp farming systems in My Xuyen and Gia Rai. In: Preston, N., Clayton, H. (Eds), Rice-shrimp farming in the Mekong Delta: biophysical and socioeconomic issues. ACIAR Technical Reports No. 52e, pp. 70–89.
- Preston, N., Brennan, D., Clayton, H., 2003. An overview of the project research. In:

 Preston, N., Clayton, H. (Eds), Rice-shrimp farming in the Mekong Delta:

 biophysical and socioeconomic issues. ACIAR Technical Reports No. 52e, pp. 7–

 14.
- Renaud, P.E., Riedel, A., Michel, C., Morata, N., Gosselin, M., Juul-Pedersen, T., Chiuchiolo, A., 2007. Seasonal variation in benthic community oxygen demand: A response to an ice algal bloom in the Beaufort Sea, Canadian Arctic? J. Marine Syst. 67(1–2), 1–12. doi:10.1016/j.jmarsys.2006.07.006
- Rong, N., Shan, B., Wang, C., 2016. Determination of Sediment Oxygen Demand in the Ziya River Watershed, China: Based on Laboratory Core Incubation and Microelectrode Measurements. Int. J. Environ. Res. Public Health 13, 232. doi:10.3390/ijerph13020232
- Rutherford, J.C., Wilcock, R.J., Hickey, C.W., 1991. Deoxygenation in a Mobile-Bed River: I. Field Studies. Water Res. 25, 1487–1497.
- Sommaruga, R., 1991. Sediment oxygen demand in man-made Lake Ton-Ton (Uruguay). Hydrobiologia, 215, 215–221.
- Steeby, J.A., Hargreaves, J.A., Tucker, C., 2004. Factors Affecting Sediment Oxygen Demand in Commercial Channel Catfish Ponds. J. World Aquacult. Soc. 35(3), 322–334.
- Thirumalai, K., DiNezio, P.N., Okumura, Y., Deser, C., 2017. Extreme temperatures in Southeast Asia caused by El Nino and worsened by global warming. Nat. Commun. 15531, 1–8. doi: 10.1038/ncomms15531.
- Tho, N., Vromant, N., Hung, N. T., Hens, L., 2006. Organic Pollution and Salt Intrusion in Cai Nuoc District, Ca Mau Province, Vietnam. Water Environ. Res. 78(7), 716–723. doi:10.2175/106143006x101755
- Todd, M.J., Vellidis, G., Lowrance, R.R., Pringle, C.M., 2009. High Sediment Oxygen Demand Within an Instream Swamp in Southern Georgia: Implications for Low Dissolved Oxygen Levels in Coastal Blackwater Streams. J. Am. Water Resour. Assoc. 45(6), 1493–1507. doi: 10.1111/j.1752-1688.2009.00380.x

- Wallace, T.A., Ganf, G.G., Brookes, J.D., 2016. Sediment oxygen demand in a constructed lake in south-eastern Australia. J. Environ. Manage. 181, 449–454. doi:10.1016/j.jenvman.2016.07.008
- Wang, Y.X., Li, X.Y., Lee, J.H., 2008. Influence of the biomass content in sediment on the sediment nutrient flux for a pulsed organic load. Mar. Pollut. Bull. 57(6–12), 681–688. doi:10.1016/j.marpolbul.2007.12.016
- Yao, S., Jufa, C., Li, Y., Yun-li, S., 2000. *In-situ* determination of sediment oxygen demand in cultivation ponds. Chin. J. Oceanol. Limnol. 18(2), 157–161.
- Zhong, D., Wang, F., Dong, S., Li, L., 2015. Impact of Litopenaeus vannamei bioturbation on nitrogen dynamics and benthic fluxes at the sediment—water interface in pond aquaculture. Aquac. Int. 23(4), 967–980. doi:10.1007/s10499-014-9855-6.

Figure captions

Figure 1. Study site layout showing two integrated rice-shrimp ponds (symbolled TB1 and TB2) in Tan Bang Ward, Thoi Binh District, Ca Mau province, Vietnam. The total area of TB1 is 1.5 ha, and that is 2.0 ha of TB2. D = ditch, P = platform.

Figure 2. Mean (\pm SD) water temperature (°C) (a) and salinity (b) across two integrated rice-shrimp ponds in 2016 and 2017

Figure 3. Mean (\pm SD) DO concentrations (mg L⁻¹) in sediment cores from two integrated rice-shrimp ponds from experiments conducted in 2016 and 2017. DO concentrations were recorded every one or two hours during the daytime, and every three hours during night time over 24 h (n = 12). Night time is from approximately 17:45:00 (5:45 pm) to around 05:45:00 (5:45 am).

Figure 4. Comparison of oxygen production (OP), sediment oxygen demand (SOD) and total oxygen demand (TOD) rates (g m⁻² d⁻¹) between the dry and wet seasons. Different letters indicate significant differences between seasons (p < 0.05).

Figure 5. Comparison of oxygen production (OP), sediment oxygen demand (SOD) and total oxygen demand (TOD) rates (g m⁻² d⁻¹) between 2016 and 2017. Different letters indicate significant differences between years (p < 0.05).

Tables

Table 1. Production and related data of two integrated rice-shrimp ponds in 2016 and 2017. Shrimp (*P. monodon*) were stocked with a mean density of 4.5 to 5.0 post larvae m⁻², but should be noted that densities in the ponds were unlikely to be this high as shrimp were harvested semi-continuously.

Pond	Pond area (ha)	Whole season mean shrimp stocking density (PL m ⁻²)	Whole year survival (%)	Mean harvest size (shrimp kg ⁻¹)	Shrimp yield (kg ha ⁻¹)	Crab yield (kg ha ⁻¹)	Rice yield (kg ha ⁻¹)
TB1-2016	1.5	4.5	25.5	40	333	65	1600
TB1-2017	1.5	5.0	46.2	38	568	248	3330
TB2-2016	2.0	4.5	17.3	35	260	109	1500
TB2-2017	2.0	5.0	36.0	39	504	85	2750
Mean (± SD) 2016	1.75 ± 0.35	4.5 ± 0.0	21.4 ± 5.8	37.5 ± 3.5	296 ± 51	87 ± 31	1550 ± 70
Mean (± SD) 2017	1.75 ± 0.35	5.0 ± 0.0	41.1 ± 7.2	38.5 ± 0.7	536 ± 45	166 ± 115	3040 ± 410

Table 2. Physico-chemical parameters in the water column (mean \pm SD). DO = dissolved oxygen. AFDW = ash free dry weight. DON = dissolved organic nitrogen. DOP = dissolved organic phosphorus.

Damamatana	Der	Wind	2016	2017
Parameters	Dry	Wet	2016	2017
Temperature (°C)	34.2 ± 3.4	30.5 ± 1.6	33.0 ± 4.4	31.8 ± 1.6
$DO (mg L^{-1})$	6.38 ± 2.14	5.08 ± 1.65	6.02 ± 2.20	5.44 ± 1.84
pН	7.92 ± 0.52	8.11 ± 0.56	7.74 ± 0.61	8.29 ± 0.21
Salinity	29.3 ± 14.9	2.9 ± 0.8	22.4 ± 21.8	8.9 ± 8.6
Euphotic depth (m)	1.4 ± 0.2	0.7 ± 0.1	1.0 ± 0.6	0.9 ± 0.3
TSS (mg L^{-1})	80.6 ± 7.5	94.5 ± 30.1	76.0 ± 10.6	99.1 ± 25.0
AFDW (mg L^{-1})	27.7 ± 9.6	46.0 ± 4.5	33.8 ± 13.8	39.9 ± 11.0
NH_4 - $N (mg L^{-1})$	0.148 ± 0.056	0.287 ± 0.214	0.225 ± 0.203	0.210 ± 0.142
NO_3 -N (mg L ⁻¹)	0.232 ± 0.228	0.067 ± 0.034	0.239 ± 0.224	0.050 ± 0.010
NO_2 -N (mg L ⁻¹)	0.011 ± 0.008	0.039 ± 0.023	0.026 ± 0.023	0.027 ± 0.021
DON (mg L ⁻¹)	0.215 ± 0.126	0.382 ± 0.263	0.349 ± 0.315	0.248 ± 0.170
TN (mg L^{-1})	0.828 ± 0.196	1.059 ± 0.189	0.921 ± 0.087	0.966 ± 0.318
PO_4 - $P (mg L^{-1})$	0.115 ± 0.052	0.113 ± 0.034	0.134 ± 0.042	0.098 ± 0.028
DOP (mg L^{-1})	0.114 ± 0.084	0.133 ± 0.053	0.143 ± 0.061	0.104 ± 0.055
Total P (mg L ⁻¹)	0.255 ± 0.132	0.272 ± 0.109	0.298 ± 0.132	0.241 ± 0.115
Chlorophyll $a (\mu g L^{-1})$	12.3 ± 5.5	55.8 ± 34.8	45.2 ± 27.0	19.9 ± 6.9
Sulfide (mg L ⁻¹)	0.002 ± 0.001	0.005 ± 0.001	0.003 ± 0.001	0.004 ± 0.002
$TOC (mg L^{-1})$	15.1 ± 1.2	13.3 ± 1.5	15.0 ± 1.6	13.4 ± 1.3

Table 3. Background physico-chemical parameters in the sediment (mean \pm SD)

Parameters	Ditch	Platform	Dry	Wet
TOC (%)	3.16 ± 1.39	4.54 ± 2.31	5.27 ± 1.68	2.43 ± 0.58
TN (%)	0.28 ± 0.12	0.30 ± 0.11	0.38 ± 0.04	0.19 ± 0.03
TP (%)	0.03 ± 0.00	0.04 ± 0.00	0.04 ± 0.00	0.03 ± 0.01
Chlorophyll $a \text{ (mg m}^{-2}\text{)}$	0.85 ± 0.93	2.17 ± 2.43	2.95 ± 1.54	0.16 ± 0.09

Table 4. Mean $(\pm SD)$ oxygen production (OP), sediment oxygen demand (SOD) and total oxygen demand (TOD) rates $(g m^{-2} d^{-1})$ in the ditch and platform.

Parameter	Ditch	Platform	p value
OP (g m ⁻² d ⁻¹)	0.97 ± 0.21	1.18 ± 0.32	> 0.05
$SOD (g m^{-2} d^{-1})$	1.74 ± 0.32	2.21 ± 0.46	> 0.05
TOD (g $m^{-2} d^{-1}$)	2.57 ± 0.54	2.88 ± 0.69	> 0.05

Table 5. The Pearson's correlation between SOD and parameters in the water column, n = 48. TEMP = Temperature. DO = dissolved oxygen. SAL = Salinity. EUPH = Euphotic depth. AFDW = ash free dry weight. DON = dissolved organic nitrogen. DOP = dissolved organic phosphorus. CHL = Chlorophyll a.

Parameter	TEMP	DO	pН	SAL	EUPH	TSS	AFDW	NH ₄ ⁺	NO ₃	NO ₂	DON	TN	PO ₄ ³⁻	DOP	TP	CHL	TOC
Pearson's correlation	-0.603	-0.461	0.015	-0.692	-0.061	0.021	0.656	0.309	-0.286	0.584	-0.098	0.394	0.090	0.127	0.094	0.875	-0.478
Significance	0.057	0.140	0.486	0.028	0.123	0.480	0.039	0.228	0.246	0.064	0.409	0.167	0.416	0.382	0.412	0.002	0.095
(1-tailed)																	

Table 6. The Pearson's correlation between SOD and parameters in the sediment, n = 24

Parameter	Chlorophyll a	Total nitrogen	Total phosphorus	Total organic carbon
Pearson's correlation	-0.578	-0.867	-0.213	-0.631
Significance (1-tailed)	0.083	0.004	0.022	0.068

Figures

Figure 1.

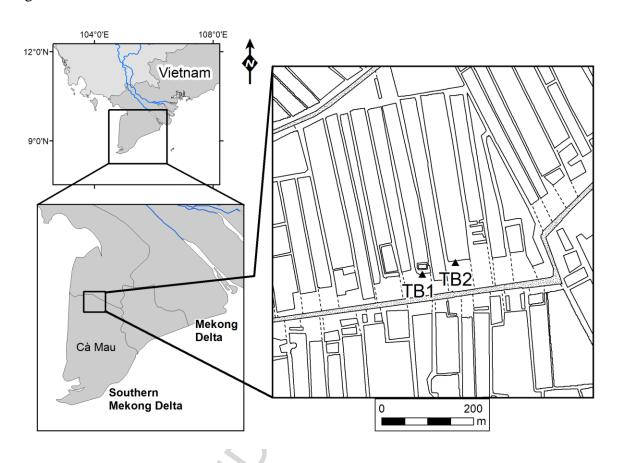
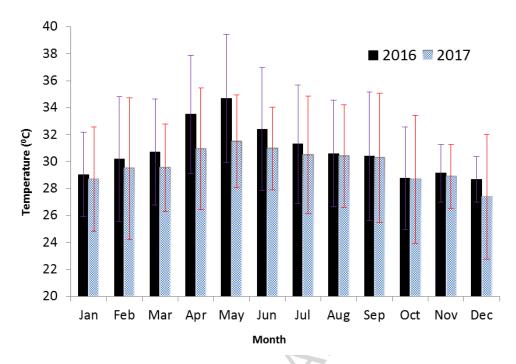


Figure 2.

(a)



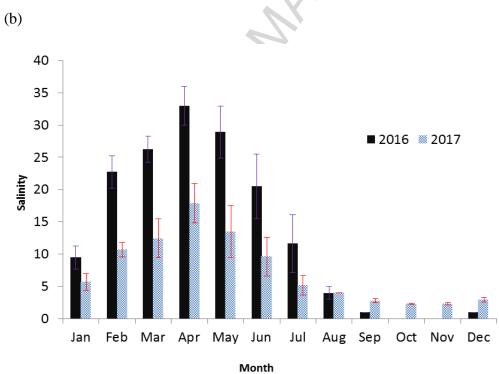
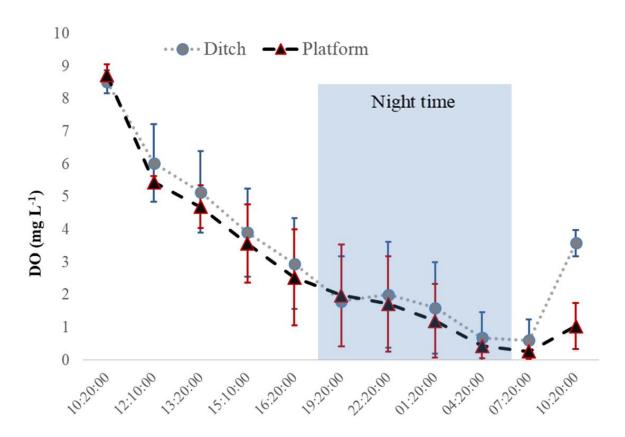


Figure 3.



Time (hh:mm:ss)

Figure 4.

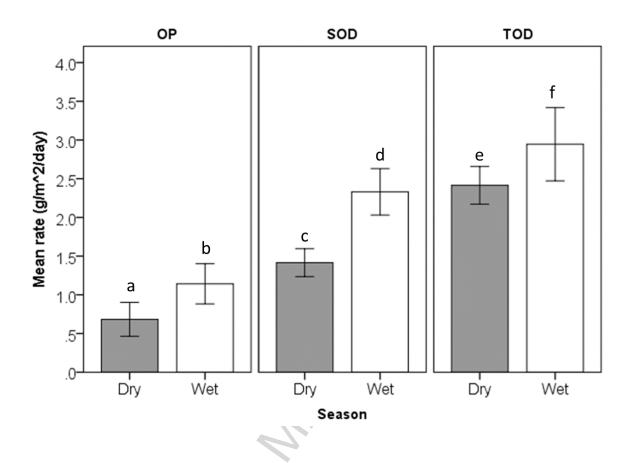
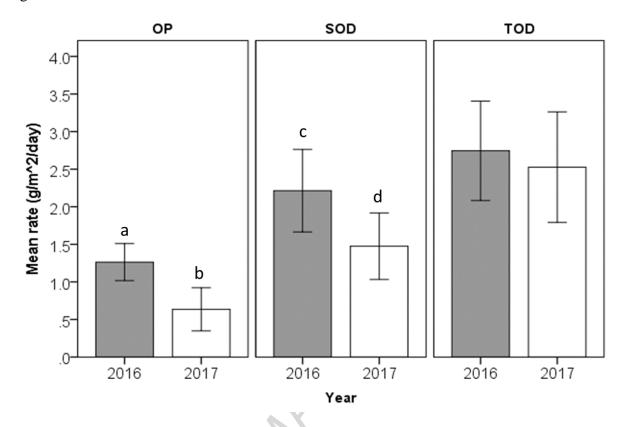


Figure 5.



Mr Luu Duc Dien

Australian Rivers Institute and School of Environment & Science
Griffith University
Nathan QLD 4111, Australia

Dr. Jian Qin Section Editor Aquaculture Journal

June 11th, 2019

Re: Submission of revised manuscript to Aquaculture journal

Dear Dr. Jian Qin,

We would like to express our sincere thanks to Dr. Jian Qin and two anonymous reviewers for the detailed comments to improve this manuscript.

The most significant revision of this revised manuscript is that we change the title. Comments mainly concentrated on the reasons why shrimp survival and yield in 2017 were significantly higher than those in 2016. We realize that our title may have confused readers as it was not our intention to look directly at shrimp survival, but to infer effects based on measures of oxygen cycling. Therefore, the title has been changed to "Factors driving low oxygen conditions in integrated rice-shrimp ponds". Please see all our edits in the "Response to Reviewers" file.

The main highlights of this study are:

- A high percentage of oxygen demand at a whole pond scale was from the sediment; hence SOD drove low oxygen concentrations in the water column.
- Algal production in the water column, rather than benthic algal production, or other organic loading, provided an organic carbon source driving SOD.
- Oxygen demand was much higher than oxygen production within the IRSPs, indicating high bacterial activity and low algal production.

We look forward to hearing from you at your earliest convenience and hope that our revised manuscript meets with your approval.

Yours sincerely,

(Iw

Luu Duc Dien