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# ***Interactive comment on “Relationship between N : P : Si ratio and phytoplankton community composition in a tropical estuarine mangrove ecosystem” by A. K. Choudhury and P. Bhadury***

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We thank reviewer for the in depth analysis and review of our manuscript. The observations made by reviewer were significant and was helpful in improving our knowledge on the concept of Redfield ratio and nutrient limitation. We hope to improve our manuscript significantly in the revised version, keeping in view the comments and concerns raised by reviewers.

Even though there have been different publications on the phytoplankton population of this area, due to the vastness of the area every study represent some new findings that allow us to further enrich our knowledge on this unique ecosystem. The Sundar-

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bans ecoregion faces several influences like high localised anthropogenic pressure, seasonal precipitation, proximity of freshwater as well as marine water inflow from the Bay of Bengal. However, the range and severity of these influences are not similar all throughout Sundarbans as the level of human settlements as well as flora and fauna are spatially different. Accordingly, there cannot be a single consensus view of the Sundarbans ecosystem including for phytoplankton biology and nutrient chemistry. Continuing with this idea, we focused in an area where there are significant localised anthropogenic influences as well as prevalent mixing conditions between freshwater sources and marine water. Thus the primary objective of the present work was to understand the spatial and temporal variations in phytoplankton population as a response to nutrient ratio in a eutrophic mangrove ecosystem located at the confluence of a freshwater tidal creek and marine water. Moreover, we were also interested to find out if the concept of species specific traits (largely used in experimental conditions) were applicable in a natural mangrove ecosystem as well. We agree with the comments of reviewer and hope to modify our manuscript in accordance with the suggestions made by reviewer in the revised version. The concept of N limitation due to high phosphate loadings relative to nitrogen concentrations will be taken into consideration while rewriting the Abstract and Discussion part of the present manuscript. Tidal fluctuation data and correlation results will be included in the manuscript. Likewise, correlation between phytoplankton biovolume and nutrient molar ratios will be developed and appropriate reference will be included as suggested by reviewer. Discussion about the role of silicate in diatom growth and possible outcome of silicate deficiency will be included as well.

Sample collection: Samples were mainly collected during periods of high tide. A tidal chart will be included in the manuscript. Air temperature data were included to establish the high correlation with water temperature. However, we agree that air temperature data can be removed as we have also included water temperature. We would like to clarify that during nutrient analysis, for each individual parameter 125 mL of sample was collected and results were determined in triplicates. May be the represen-

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tation in the manuscript was not clear which will be rewritten in the revised version of the manuscript. Even though filtration was used for other parameters, no filtration was done in case of nitrogen estimation. Ambient nitrogen contents were directly measured spectrophotometrically in triplicates. The present group is involved with phytoplankton research for the past several years. We take appropriate precautions while cleaning our sampling containers. There are specific set of containers that are used for specific parameters. Before each sample collection, these containers are cleaned with Extran<sup>®</sup> (a product of EMD Milipore) to minimise phosphate contamination of detergents, preceded by rinsing with 1N HCl. Each of the containers is then oven dried to remove excess water. Temperature, salinity and pH were measured in situ using hand held instruments immediately after sample collection and were not measured under lab conditions. All hand held instruments were calibrated before sample collection on each occasion.

Results: As rightly pointed out by reviewer, our sampling area is divided into three stations and is designated as Stn 1, Stn 2 and Stn 3 respectively. The phytoplankton diversity of all the sampling stations using a molecular approach has already been worked out by other authors of the present group (Samanta and Bhadury, 2014). However, Stn 1 is located within the creek whereas Stn 3 is further downstream with close proximity with the Bay of Bengal. Since the primary objective of the present work was to understand the effects of nutrient molar ratio on phytoplankton population, we segregated the stations on the basis of freshwater and marine water influences respectively. As Stn 2 occupies a more intermediate position between the freshwater and marine sources, datasets of Stn 2 may not allow the authors to make significant demarcation in the nutrient molar ratio between stations. Accordingly, to keep it simpler and analytically more relevant, datasets for Stn 1 and Stn 3 were taken into consideration. Tables 3 and 4 will be rectified in accordance with the suggestions made by reviewer. Font size and other typos will be rectified and resolutions will be improved in the revised version of the manuscript. The inclusions of physicochemical data in the figure

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of study area was done on the initial comments and suggestions made by Handling Editor. They can be separated in the revised version of the manuscript as per the discretion of the Editor and the Reviewers. Monthly variation in oxygen concentration was measured and can be included in the revised version of the manuscript. In an attempt to reduce the number of graphs, this was not included in the present version of the manuscript. Both salinity and pH were measured in situ in triplicates as mentioned previously. The same stands true for oxygen concentrations as well. We would say that may be due to localised anthropogenic influences from adjacent agricultural and aquaculture farms that may have enhanced inputs of combined forms of nitrogen. This may also account for high levels of ammonium in the habitat as pointed out by reviewer. We have used Milipore<sup>®</sup> water for preparation of all reagents used for spectrophotometric estimations. The phytoplankton cell count were also included initially which were later removed in view of the suggestions made by handling Editor. Similarly correlations were also prepared which we can definitely be included in the revised version of the manuscript. Likewise, Chlorophyll a concentrations are also available with the authors. With regard to phytoplankton cell carbon content estimates, we agree with reviewer's comment about using POC/PON elemental analyser. However, due to infrastructural constrains we had to take a more mathematical approach for representation of our data. Accordingly, we reported the carbon content of the dominant species only. This was done due to their high abundance where at least 5 individual cells of the same species with different cell sizes were taken into consideration. Subsequently, mean values of 5 cells of each species were used to calculate the carbon contents to make a statistically correct representation. We sincerely hope that the reviewer will find our clarifications to be justified for the different concerns that have been raised. We feel that much of the manuscript can be revised and improved to make it suitable for publication. Many of the data like Chlorophyll content, tidal chart, oxygen concentrations are already with us. We can definitely include those data to make our manuscript more acceptable. Accordingly, we would request the reviewer as well as Editor to provide us with an opportunity to further work on the manuscript and revise it significantly to make

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it more scientifically important for the readers. Since this work is from an ecologically vulnerable unique ecosystem, a revised form of the manuscript can be worth reading not only for the scientific community only but for other audiences as well.

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