SUMMARY
Phosphorus (P) is one of the major essential macronutrients for biological growth and development whose concentration determines the productivity in aquatic ecosystems. Most agricultural soil as well as wetland sediment contains large reserves of phosphorus. Mineral forms of phosphorus are represented in soil or sediment by primary minerals, such as apatite, hydroxyapatite or oxyapatite and their principal property is their insolvency. Mineral phosphate can also be found associated with the surface of hydrated oxides of Fe, Al and Mn, which are poorly soluble and assimilable. Thus despite presence of sufficiently in sediment some fresh water environments may remain deficient of P. P fertilizers are sometimes used to combat the situation, however, a large portion of inorganic phosphate applied as chemical fertilizer is rapidly immobilized and becomes unavailable to primary producers. The phenomenon of fixation and precipitation of P in sediment is dependent on pH and sediment type. Besides P sorption, the sediment may also act as source of P for the overlying water. This dissolved P is used by the biotic community, starting from bacteria to organisms of higher trophic level. Internal P loading by sediment release plays an important role in the phosphorus status of lakes, and thereby supports the trophic status of water bodies. The P cycle in the biosphere can be described as ‘open’ or ‘sedimentary’, because there is no interchange with the atmosphere. The classical P chemistry described the aquatic P cycle in relation to iron chemistry: oxygenated sediment retains P by fixing it to iron (III), while reduced sediment releases P by reduction of iron and subsequent dissolution of Fe-P. Besides inorganic binding, microorganisms play a critical role in P cycle through cyclic oxidation and reduction of phosphorus compounds, where electron transfer reactions between oxidation stages ranges from phosphine (-3) to phosphate (+5). However, the

**Isolation of Bacteria Capable of Releasing Phosphorus from Bound Forms in Wetland Ecosystems**

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insoluble or otherwise unavailable P compounds can also be solubilized or mineralized by microorganisms making P available for other organisms. These phosphorus-releasing microorganisms may provide one strategy for enhancing P availability through regeneration of sediment P in an ecologically benign manner for better productivity management. These groups of microorganisms are classified as P solubilizing bacteria (PSB) or P mineralizing bacteria.

In this thesis attempt was made to examine the P dynamics in terms of P forms and their mobilization in freshwater ecosystems, and to evaluate the role of microorganisms (P solubilizing and mineralizing, or in other words P releasing) in this P mobilization, thus identifying a few microbial strains as good candidate for biofertilization application for aquatic productivity management.

For this, two floodplain wetlands (Akaipur and Bhomra wetland, West Bengal, India) and a small perennial Churni river spanning both India and Bangladesh was studied. The level and distribution of P fractions were evaluated using a sequential chemical extraction to examine potential sorption or mobilization of sediment P in these water bodies. Five P fractions were quantified: loosely sorbed P (NH\(_4\)Cl-P), Al-P (NH\(_4\)F-P), Fe-P (NaOH-P), Ca-P (H\(_2\)SO\(_4\)-P), and the residual P (organic and refractory P). Exceedingly high levels of total P (mean: 5468.00 ± 363.45, 6037.04 ± 344.45 and 3118.02 ± 117.83 mg kg\(^{-1}\) sediment), consisting largely of organic and refractory fraction (83.77 – 89.5%), followed by Ca-P (mean: 142.67 ± 8.42, 584.21 ± 31.26 and 420.15 ± 47.07 mg kg\(^{-1}\) sediment) and Fe-P (mean: 91.02 ± 7.68, 107.55 ± 10.10 and 87.54 ± 28.18 mg kg\(^{-1}\) sediment) were recorded respectively in Akaipur and Bhomra wetlands and Indian part of Churni river. The inorganic P, comprising the loosely sorbed P and all the mineral bound forms contributed only about 6.07 -17.08 % of total P indicating their less significance in P sorption or desorption in these tropical wetlands. Although the
loosely sorbed P (Avl. P/ Avail-P) was in moderate level (1.54 ± 0.53, 2.69 ± 0.69 and 1.11 ± 0.76 mg kg\(^{-1}\) sediment), which was also reflected in low to moderate water dissolved P (in Akaipur 0.033 - 0.298 mg l\(^{-1}\); mean: 0.16 ± 0.02 mg l\(^{-1}\), in Bhomra 0.25 – 1.81 mg l\(^{-1}\); mean: 1.08 ± 0.12 mg l\(^{-1}\) and in Churni 0.057 – 0.132 mg l\(^{-1}\); mean: 1.63 ± 0.26 mg l\(^{-1}\)), which, however, sometimes remained in traces due to uptake by massive macrophyte production in these tropical water bodies. The study reflected sediment P accumulation in form of organic matter in these aquatic systems, as well as, a potentiality of its release through microbial processes under appropriate condition for productivity management.

As specified in objective, a major part of the study concentrated on identifying the bacterial community responsible for inorganic phosphate solubilization in these tropical water courses. Seventy two phosphate solubilizing bacteria were isolated from bottom soil, water, and fish gut and examined for solubilization of tricalcium phosphate. Results revealed aquatic PSB to be low to moderately capable in P solubilization (range: 6.3 - 68.87 mg P l\(^{-1}\)), and bacteria from sediment were more effective in P release than those from water and fish gut. The important P solubilizers in freshwater environments were identified in diverse genera, viz. *Bacillus*, *Brevibacillus*, *Enterobacter*, *Agrobacterium*, *Pseudomonas*, *Acinetobacter*, *Microbacterium*, *Curtobacterium*, *Stenotrophomonas*, *Novosphingobium*, *Pantoea*, *Bacillus*, *Agrobacterium*, *Rhizobium*, *Enterobacter* and *Acinetobacter*; with dominance of genus *Bacillus* in sediment. Genus-wise, *Bacillus* had the highest mean PS activity, followed by *Enterobacter* and *Agrobacterium*, and *Pseudomonas* had the lowest activity.

Study of isolation and identification of phytate mineralizing bacteria (PMB) was done from fish gut, water and sediment with aim to isolate potential PMB from different niches and different ecological significance. Phytic acid (myo-inositol 1, 2, 3, 4, 5, 6-
hexakis-dihydrogen phosphate), or phytate, a major constituents of P in grains or other plant feedstuffs is not digested by fish due to lack of intestinal phytase. This excreted phytate which increase the sedimented P load could have become a long term source of P if properly managed. The study indicated presence of weak phytase producer in these tropical water bodies and many of them also had P solubilization activity. The strains were identified to belong to genus *Bacillus, Arthrobacter, Klebsiella, Methylobacterium,* and *Fictibacillus.*

To examine activity and significance of the isolates in nature, the strains were studied in simulated sediment microcosm, along with the study of natural mobilization of Ca-P in sediment. Round-the-year monitoring of sediment Ca–P fraction revealed a marked seasonal variation which gradually increased during rainy and winter seasons reaching a peak at the end of winter, followed by a sharp fall in summer with the concomitant change in pH, elucidating probable role of microorganisms in sediment P status regulation. About 43% of the isolated strains were effective in enhancing available P concentration in interstitial water in laboratory sediment microcosms indicating their P release potential. Study with cyanobacteria (*Aphanocapsa* sp. and *Oscillatoria* sp.) as productivity indicator for two strains CPSM8 and Aph1 (those showed hight P solubilization and phytate mineralization activity and not characterized in details. by 16s rDNA method) showed profuse growth of cyanobacteria for CPSM8 and moderate growth for Aph1.

Whole genome study of these two strains identified both of them as *Bacillus* sp. with close similarities of CPSM8 with *Bacillus licheniformis* and Aph1 with *Bacillus megaterium.* Harvesting of their genetic information revealed presence of various genes for P solubilization (*viz.* glucose dehydrogenase, citrate synthase, and lactate dehydrogenase) and mineralization (*viz.* phytase, acid and alkaline phosphatase, 5′-
nucleotidase genes), along with some other genes related to organic matter mineralization (viz. exo- and endo-glucanases, xylanase, glucosidases, galactosidases, peptidases, esterase) and biofertilization (viz. hemolysin, bacillibactin), indicating their ability to be a potential candidate for bio-fertilizer application in aquatic productivity management.