

ALGAL TURF SCRUBBER® (ATS™) HISTORY AND APPLICATION FOR LOW NUTRIENT APPLICATIONS

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Periphytic and epiphytic algal communities have for some time been recognized as ecologically important, both for their contributions as primary producers, and for their modulating influences regarding energy and nutrient flux within low nutrient systems. In his classic study on Florida's Silver River Odum (1955) discusses this role of the epiphytes (aufwuchs) that develop upon the submerged vascular plant *Vallisneria sp.* (eel grass) in stabilizing and distributing the energy and material flows within this freshwater spring system.

These types of algal communities also serve a critical role in nutrient management within the oligotrophic Everglades Ecosystems. Browder et al. (1994) note that "the assemblage of microalgae that live in shallow, submerged substrates, ---referred collectively as periphyton, aufwuchs, or the algae mat", is the most widely distributed plant community in the Florida Everglades. Adey and Loveland (1998) referred to this community as "Algal Turf". It is recognized that the use of "plant community" to describe algae may not be taxonomically correct, but it does correctly identify the community as being composed primarily of photoautotrophic organisms.

The evolution of the Algal Turf Scrubber® (ATS™) found genesis in studies by researchers with the Smithsonian Institution working on marine systems. Adey (1978) was among the first of these researchers to disclose the role of periphytic and epiphytic algae in the maintenance of very low nutrient levels within coral reef systems, noting the ability of these organisms to sustain relatively high levels of productivity under oligotrophic conditions, when supported by the energy associated with tidal movement and oscillatory waves. He later elaborated upon this dynamic in developing the Algal Turf Scrubber® (Adey and Goertmiller, 1987; Adey and Hackney, 1989; Adey, 1998).

Carpenter et al. (1991) examined the influence of boundary layer disruption through increased flow velocity upon algal productivity in low nutrient seawater within the laboratory, noting significant increases in productivity of marine periphytic algae as the flow velocity increased from zero cm/sec to over 22 cm/sec. The point at which improved production was no longer aided by velocity varied with the species. For all species studies, velocities over 22 cm/sec (about 0.75 ft/sec) did not solicit improved production. However, oscillatory waves did further stimulate higher production when compared to steady flow.

From the early works by Adey and his colleagues evolved the concept of a structured approach to promote the growth of periphytic algae in association with treating waters attendant with mesocosms, including coral reef mesocosms. This structured approach was patented as an Algal Turf Scrubber® (ATS™) by Adey in subsequent U.S. Patents: 4,333,263 -1982; 4,966,096-1990; 5,097,795—1992: and 5,851,398—1997. The central theme within these patents is the cultivation of periphytic algae communities or algal turf upon a constructed substrate, typically sloped, and the surging or pulsing of water across the substrate, with the periodic harvesting of accumulated biomass. In addition, the latest patents include (i) the purposeful management of the periphyton system to optimize phosphorus recovery through

precipitation of phosphorus upon or within the algal cell walls (U.S. Patent No. 5,851,398), and (ii) the efficient recovery of biomass (U.S. Patent No. 6,572,770).

As noted, early work on the ATS™ concept was applied to very low nutrient conditions related to coral reef systems, and the effectiveness of the ATS™ concept helped facilitate successful cultivation of corals within aquaria, and larger cultivation tanks (Luckett et al. 1996). Considering the potential of the ATS™ to provide wholesale removal of nutrient pollutants from wastewaters, and polluted freshwater as well as saltwater surface waters, Adey provided oversight to two ATS™ demonstrations—the first being in Patterson, California (Craggs et al. 1996) the second in the Everglades Agricultural Area (EAA) in South Florida (Adey et al. 1993). Subsequently, the ATS™ was applied in two large-scale recirculating fish production facilities—one in Fall River, Texas, the other in Okeechobee County, Florida. The latter facility was designed and operated by HydroMentia, Inc. The flows within this facility approached 30 MGD of recycled flow. The ATS™ unit removed an average of 9.5 lb-P/acre-day (389 g-P/m²-yr) within this facility (Stewart, 2000), with algal productivity above 20 dry-g/m²-day. Because this facility included recirculation of water from a high intensity fish cultivation operation, nutrient levels were very high. Low nutrient ATS™ systems, which are more germane to ATS™ application for impaired surface waters and Everglades's restoration, are discussed in some detail in the following subsection.

KEY PARAMETERS RELATED TO LOW NUTRIENT ATS™ DESIGN AND OPERATION

In 2003, as noted previously, HydroMentia commenced operation of a Managed Aquatic Plant System (MAPS) prototype project financed jointly by the District, the Florida Department of Environmental Protection (FDEP), the Florida Department of Agriculture and Consumer Services (FDACS). This project, which included the use of Water Hyacinth Scrubbers (WHS™) and Algal Turf Scrubbers (ATS™), was known as the S-154 MAPS Prototype, located within the Lake Okeechobee Watershed (LOW) along one of the more nutrient rich canals discharging into Lake Okeechobee—L-62—which drains a priority basin labeled S-154. The operation and monitoring of this facility has provided important information related to the performance of MAPS facilities receiving soft, highly colored, agriculture impacted surface waters.

Recent work was authorized through an extension to the primary S-154 contract to investigate three (3) independent ATS™ flowways receiving feedwater directly from L-62. Data from these flowways has provided clear indication of the influence of increased flow velocity upon ATS™ total phosphorus uptake rate, as expressed by total phosphorus removal rate as g-P/m²-yr, as shown. Within these flowways, the areal removal rate was considerably higher when compared to the ATS™ units receiving lower hydraulic loading rates and WHS™ effluent as the feedwater.

As water for the ATS™ flowways was delivered from the same source, by the same pumping system, with environmental conditions surrounding the flowways essentially identical, the sole influencing variable was the flow velocity parameter - linear hydraulic loading rate or LHLR, expressed as gallon per minute (gpm) per linear foot of ATS™ width - gallon/minute-ft. The extent and nature of this influence is demonstrated with somewhat greater clarity in Figure 1.

Shown in Figure 2 are the ranges of total phosphorus concentrations associated with these flowways. Within this figure, the best-fit lines are extended beyond the data point scatter. It needs to be recognized that this was developed for a flowway of only 300 foot length. The trends shown in this figure provide implication that with an extended length, at high LHLR, very low total phosphorus concentrations are likely to be achieved. Actual fieldwork at lower total phosphorus concentrations would be the only reliable means of evaluating system behavior within low nutrient, high alkalinity feedwaters.

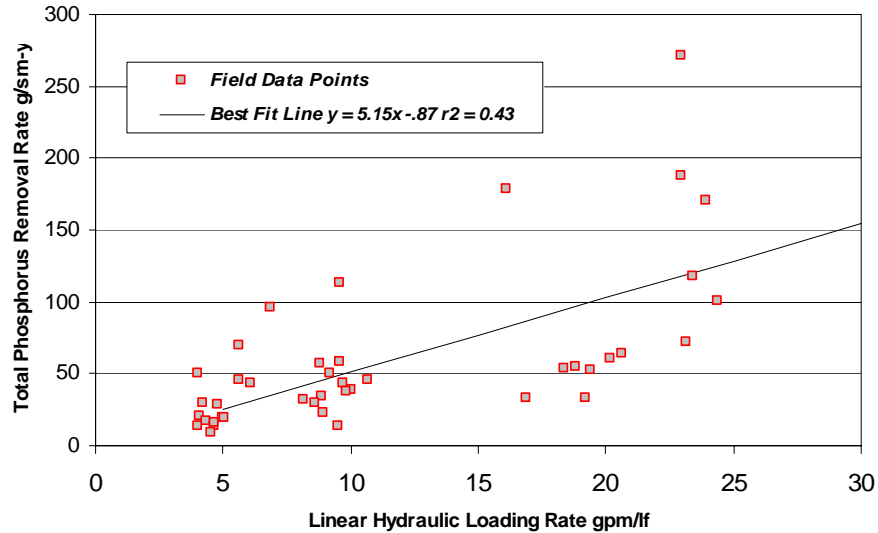


Figure 1: S-154 MAPS prototype independent ATSTTM flowways performance at different linear hydraulic loading rates.

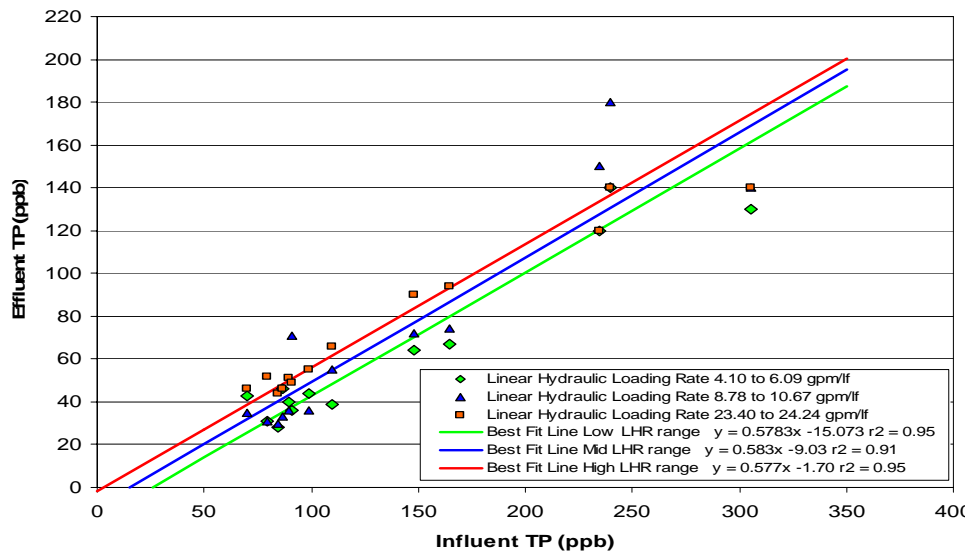


Figure 2: Total phosphorus concentrations of influent and effluent for S-154 MAPS prototype independent ATSTTM flowways.

Brezonik (1994) in a detailed discussion regarding the relative role of nutrient uptake within algae as influenced by both Monod dynamics and boundary layer transport through molecular diffusion, presents work done on models which include consideration of both phenomena. He notes that at high substrate [S] concentrations, boundary-layer diffusion control becomes negligible. At low concentrations, however, diffusion influences can overwhelm the Monod kinetics, and uptake projections based solely upon the Monod equation without inclusion of diffusion can be higher than observed. He identifies a factor $1/(1+P')$ as representative of the proportion of the total resistance to nutrient uptake caused by diffusion, where:

$$P' = a(14.4pD_s r_c K_s)/V$$

When a = shape factor applied to algal cell shape

D_s = Fick's diffusion coefficient as substrate changes per unit area per unit time

r_c = algal cell radius

K_s = Substrate concentration when uptake rate v is $1/2$ of maximum uptake rate V

V = Michaelis Menton substrate uptake rate moles per unit time

The Michaelis-Menton V may be seen in this case as analogous to the Monod m_{max} , therefore it is reasonable to express the equation as $P' = a(14.4pD_s r_c K_s/m_{max})$. Brezonik (1994) includes this P' into the Monod relationship at low concentrations of S , resulting in the equation:

$$m = m_{max} [P'/(P'+1)]S/ K_s$$

It is noted then, the smaller P' the greater the influence of diffusion on reaction rate or growth rate.

Observations regarding velocity influences relates to the general thickness of the boundary layer around the cell wall. Carpenter et al. (1991) discuss the influence water movement has upon the thickness of the boundary layer. This is consistent with discussions offered by Brezonik (1994) who notes that "turbulence increases nutrient uptake rates at low concentrations where diffusion limitations can occur". He generally observed that at low concentrations Monod dynamics can be influenced by boundary layer conditions, and uptake rates are lower than predicted by Monod kinetics. This is relevant when discussing the use of periphytic algae for reduction of total phosphorus to 10 ppb because passive systems such as PSTA which rely upon extensive areas and very low velocities, would be expected to be much more restrained by boundary layer thickness at low concentrations, which as noted by both Carpenter et al. (1991) and Brezonik (1994), is inversely related to the gradient through which diffusion occurs. The ATS™ system by adding the influence of flow and turbulence will substantially enhance the uptake rate and production of the algal mat.

Turbulence and water movement therefore serve to increase the rate of substrate transport, and hence decrease the importance of diffusion. This quite logically is why the use of high velocities and turbulence (e.g. oscillatory waves) enhances productivity (which is typically directly related to nutrient uptake). Brezonik (1994) notes that in low nutrient conditions there exists a minimum velocity (u_{min}) at which diffusion limitation of nutrient uptake is avoided. He defines this mathematically as:

$$u_{min} = (2D_s/r_c)\{(2/P')-1\}$$

This means that at $P' = 2$, $u_{min} = 0$, and u_{min} increases as P' decreases. Values for P' of some algae species are provided, ranging from 0.33 to 680, but there is no discussion offered for assessing the cumulative influence of an algal turf community upon the general role of diffusion or how u_{min} might be determined on the community level. Rather, empirical information such as that provided by Carpenter et al. (1991) can provide insight into the reaction of algal communities to velocity changes.

It is noteworthy that at low nutrient concentrations, adapted algae species would likely be characterized by a low K_s value. This is validated by Brezonik (1994), who notes the difficulty in determining the controlling influence of nutrients upon algae production at low nutrient levels, as “ K_s may be below analytical detection limits—making it difficult to define the m vs. $[S]$ curve.” He includes some of the documented K_s values for several algae species associated with low nutrients. Phosphate appears as a limiting nutrient in several cases, with K_s values as low as 0.03 mM as PO_4 , or about 3 ppb as PO_4 , or just less than 1 ppb as phosphorus. As K_s is directly proportional to P' , then it would not be unexpected that at low nutrient levels, P' would be comparatively small, and hence u_{min} comparatively large—the implication being that elimination of diffusion influence becomes very important, and hence flow velocity becomes an important design parameter. As noted, Kadlec and Walker (2003) made reference to the influence of flow velocity upon the efficacy of PSTA systems. With velocities orders of magnitude greater within ATS™ systems, it becomes an even more essential design component with ATS™. The inclusion of higher velocities and oscillatory motion within the ATS™ operational protocol allows contemplation of much higher algae productions, and higher phosphorus uptake rates, which has broad economic implications as noted previously.

Based upon literature review and field observations, it is likely that algae productivity and nutrient removal rates are impacted by more than one parameter, particularly at low concentrations. Brezonik (1994) includes in his discussions related to Monod and diffusion algal growth dynamics this recognition that more than one controlling factor may be involved, and that the Monod relationship may need to reflect this within the model, as noted in the following equation form:

$$m = m_{max} \cdot \{[P]/(K_p+[P])\} \cdot \{[N]/(K_n+[N])\} \cdot \{[CO_2]/(K_c+[CO_2])\}$$

In higher nutrient concentrations, it is more likely that one controlling factor can be used to model growth rates.

The general theme of this discussion related to substrate concentration and diffusion influences indicates two potential operational scenarios related to periphytic algae systems:

1. At high concentration of S, Monod kinetics dominates, and diffusion influence is negligible.
2. At low concentrations of S, diffusion becomes a controlling factor in nutrient uptake (hence productivity). Diffusion influences can be avoided by increasing the velocity beyond some value u_{min} .

The distinction however between low and high substrate concentration will vary, but it is reasonable to consider the total phosphorus concentration of 10 ppb as low concentration. Hence, system design efforts should be as attentive to velocities and turbulence as they are to phosphorus concentrations.

In the case of an ATSTTM, the velocity parameter is expressed as gal/minute-ft of ATSTTM width, also known as the Linear Hydraulic Loading Rate or LHLR, as presented previously. The LHLR converts to flow by multiplying by the ATSTTM width. Within the ATSTTM velocity can be estimated using the Manning's Equation as previously presented ($V = (1.49/n)r^{2/3}s^{1/2}$). However, the Manning's coefficient "n" will vary as the algal turf develops, and is harvested, and in addition, surging will create a predictable change in flow velocity from zero to the something greater than u_{min} during the siphon release. Actual velocity variations are best determined from field observations under different conditions (e.g. high standing biomass, pre-surge, post surge, etc.)

As applied to an ATSTTM, we can simplify the Manning Equation by first multiplying both sides of the equation by the flow area A, which is equal to the flow depth (d) in feet times the ATSTTM width (w) in feet, or:

$$Q_{cfs} = Vdw = (1.49/n)dw r^{2/3} s^{1/2}$$

As the hydraulic radius r is flow area (A) over the wetted perimeter, then:

$$r = dw/(w+2d)$$

With flow $Q_{cfs} = 0.00223(\text{LHLR})w$

when LHLR is gallons/minute-ft. If w is set at 1 ft, then

$$\text{LHLR} = \{0.00332[d^{5/3}/(2d+1)]s^{1/2}\}/[n(2d+1)^{2/3}]$$

This allows for the flow depths to be established for specific Manning's "n" values and slopes, and accordingly, velocity can be estimated. These relationships are noted in Figure 3 and 4.

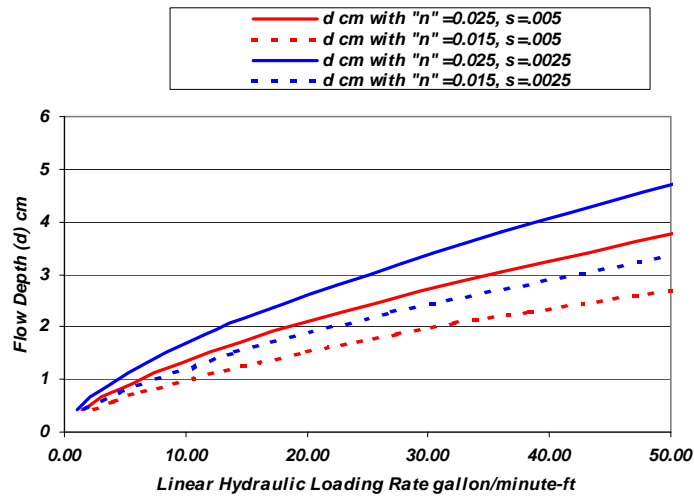


Figure 3: Comparative depth across ATS™ at varying slopes and Manning's "n"

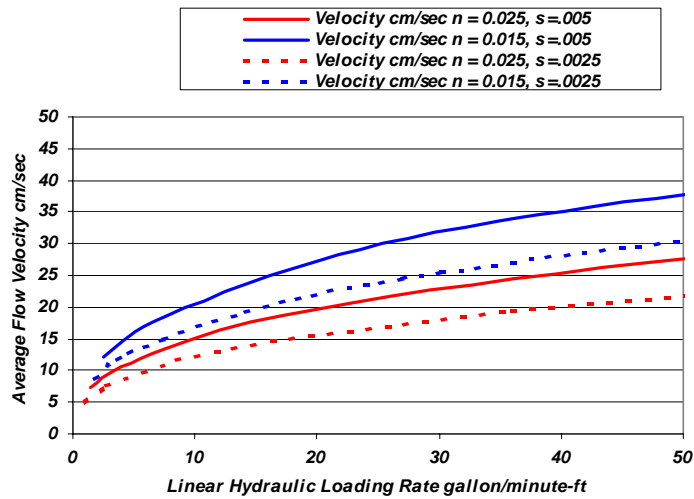


Figure 4: Comparative velocity across ATS™ at varying slopes and Manning's "n"

As noted from these graphs, the flow velocity approximates the optimal velocity identified by Carpenter et al. (1991) of 22 cm/sec at an LHLR = 20 gallons/minute-ft at the slope of 0.005. The velocity drops below this level at a slope of 0.0025.

It is tempting to evaluate the general influence of LHLR on uptake rate by treating it as a controlling factor (S) within the Monod relationship, and accordingly, identify some LHLR analogous to K_s where uptake rate of phosphorus is $\frac{1}{2}$ of the maximum uptake rate potential. This can be done through the method developed by Lineweaver and Burke (1934) to

identify the value for K_s as the negative invert of the x-intercept and V or m_{max} as the invert of the y-intercept. Using the recent data collected on the individual flowways in S-154, this results in a good fit as noted in Figure 5. But with x intercept = -0.012 and y intercept = 0.0019 indicating a $K_s = 83$ gal/minute-ft, and a maximum uptake potential of 526 g-P/m²-yr., the values appear unrealistically high. While the fit is good for this range, it needs to be remembered that Carpenter et al. (1991) observed an upper level influence for velocity, while Brezonik (1994) notes that there is an “on-off” type influence of velocity regarding negation of diffusion influences for individual species, and therefore the relationship would possibly not be linear much beyond the range shown, and the K_s and V values may well be erroneously high.

Nonetheless, based upon the data collected to date, velocity does appear to have a modulating influence upon removal rate, indicating that there is more involved than a simple “off-on” phenomenon at the community level. This could be due to the interference of Monod kinetics as concentrations decline, or to the community influence (multiple values of u_{min}) with higher velocities soliciting increased production in a larger number of contributing algae species.

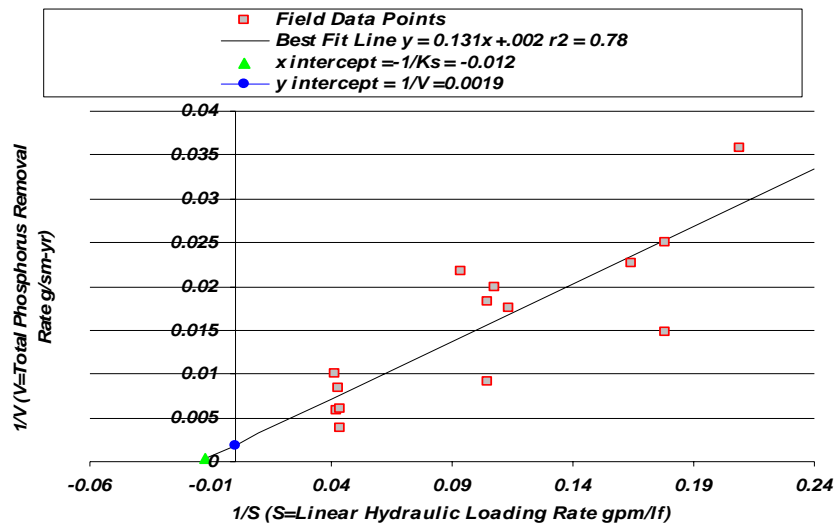


Figure 5: Lineweaver-Burke plot with LHLR as rate controlling factor.

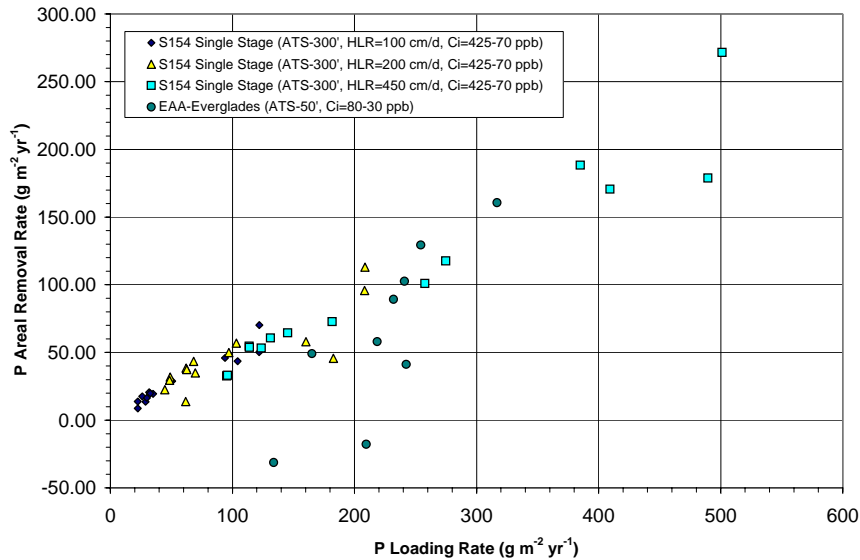
While there may remain some question regarding the nature of LHLR influence on production and nutrient uptake, particularly as total phosphorus levels approach 10 ppb, there is little question the LHLR does show a dramatic influence upon removal rate.

Additional insight regarding algal turf productivity and removal rates within the EPA is noted in a study conducted in the Everglades region by Adey et al. (1993). They designed and operated a small scale ATS™ for a period of about 8 months. The flowway unit of 12 m² was loaded at an LHLR of about 9 gallons/minute-ft. across a width of 2.6 ft, and a length of only 50 ft. This resulted in an average reduction of total phosphorus from 53 ppb to 44 ppb, or a total phosphorus removal rate of about 41 g/m²-yr, although removal rates as high as 160 g/m²-yr were recorded. The lower average is related to 2 data points in which the effluent

concentration exceeded influent concentrations. Effluent concentrations ranged from 26 ppb to 51 ppb. Algae production was reported at 21.2 dry-g/ m²-yr, with a tissue phosphorus content of 0.336% on a dry weight basis.

When the LHLR from this study is applied to the best-fit equation noted in Figure 1, the projected total phosphorus removal rate for this system is 65 g/m²-yr. The difference between observed and projected in this case could well be related to the two negative values, as well as the influence of Monod kinetics and temperature influences, as the Adey et al. (1993) work was conducted during the cooler months. Considering these influences, the projection appears reasonably close to the field value.

It is noteworthy that the productivity and the total phosphorus removal rates documented by Adey et al. (1993) and by the S-154 MAPS research are considerably higher than the values projected in the ATSDM model runs. These runs are based upon available carbon as S, and do not include a factor associated with LHLR. Performance scattergrams that include both the S-154 MAPS individual ATS™ flowway data and the Adey et al. (1993) data that are noted in Figures 6 through 9.



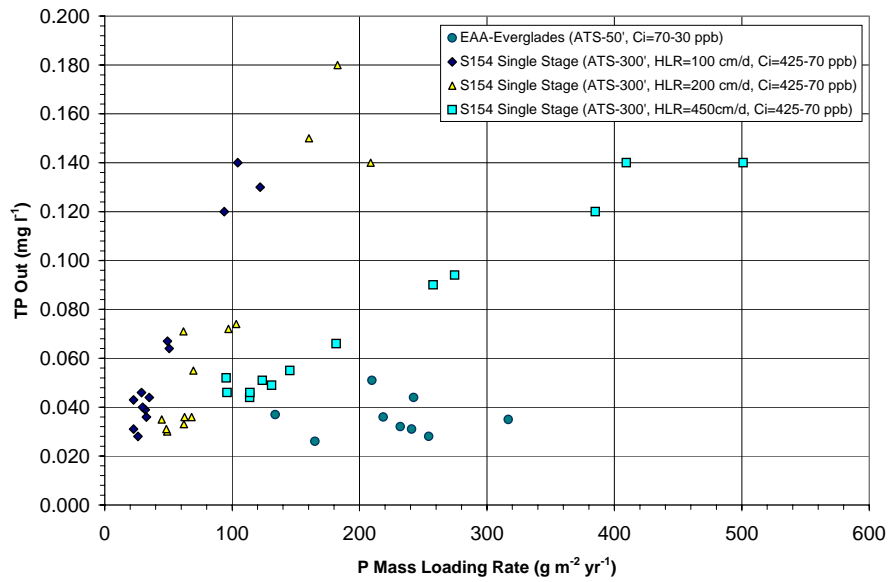


Figure 7: Phosphorus loading rate versus outflow phosphorus concentrations for four (4) ATSTM MAPS individual flowways with varying hydraulic loading rates and flowway lengths of 50 to 300 feet. Data set includes Adey et al. (1993) EAA data.

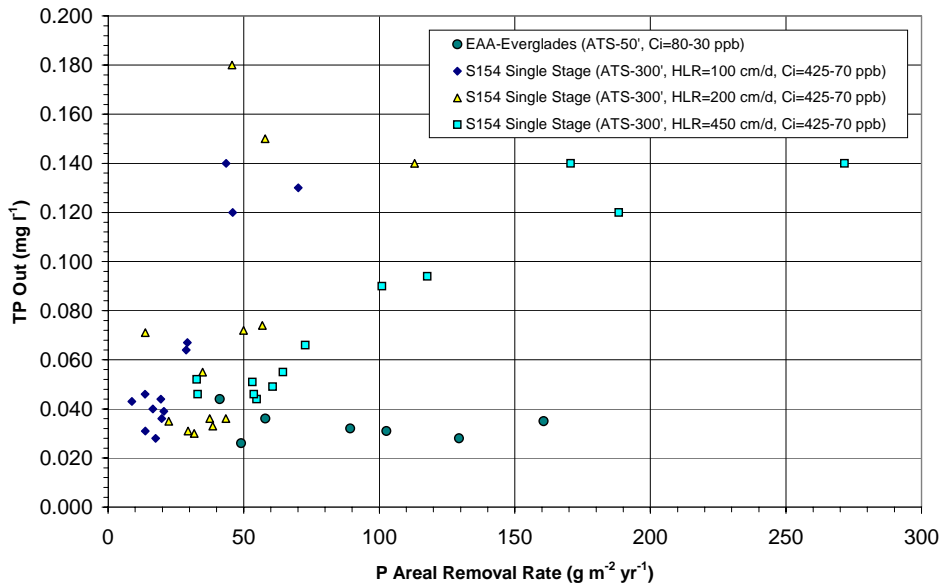


Figure 8: Phosphorus areal removal rate versus outflow phosphorus concentrations for four (4) ATSTM MAPS individual flowways with varying hydraulic loading rates and flowway lengths from 50 to 300 feet. Data set includes Adey et al. (1993) EAA data.

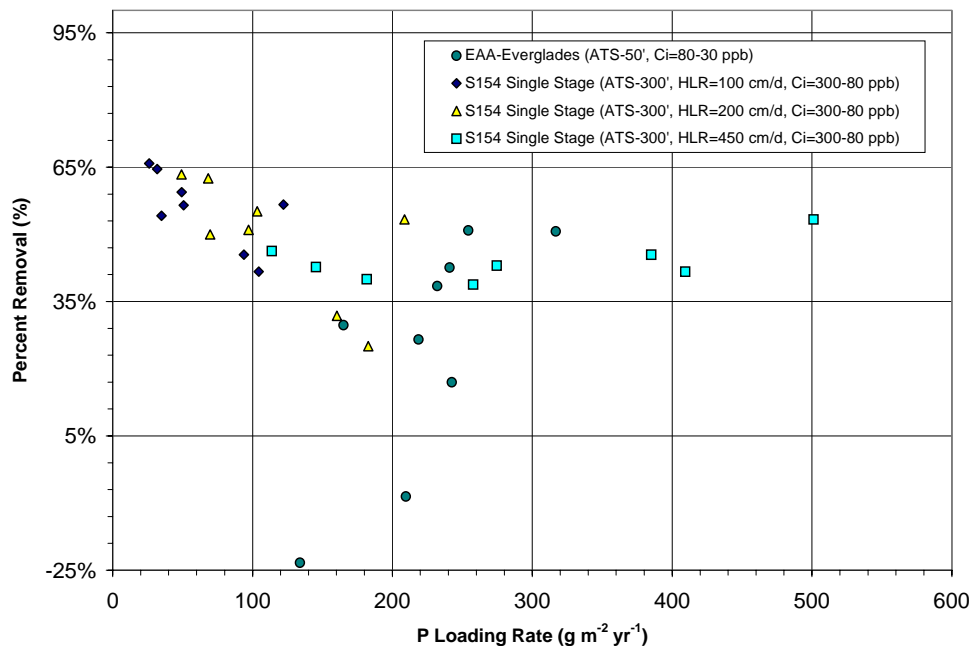


Figure 9: Phosphorus loading rate versus percent removal for four (4) ATSTM MAPS individual flowways with varying hydraulic loading rates and floway lengths from 50 to 300 feet. Data set includes Adey et al. (1993) EAA data.

While neither the individual ATSTM MAPS flowways nor the Adey et al. (1993) flowways were specifically designed to achieve an effluent of 10 ppb, if the trend shown in these figures as well as Figure 2 are extended it appears the 10 ppb target is achievable.

From data available to date, the implication then is that while the algal communities associated with total phosphorus levels of 10 ppb may lose some efficiency in phosphorus uptake, possibly because the available soluble phosphorus is at or near depletion, there is ample evidence that under proper environmental conditions there exist algal species which can reduce total phosphorus to below 10 ppb. It is fairly certain that as total phosphorus declines, the dissolved organic fraction (DOP) is likely to become predominant. Because most photoautotrophs and precipitation processes require phosphorus to be as soluble PO_4^{-3} form (also known as ortho phosphorus or soluble reactive phosphorus or SRP), it is critical that within these low nutrient communities there exist a mechanism for conversion of phosphate attached to an organic complex, generally referred to as organic P. If this organic P is in a particulate form, removal can be facilitated by either settling or entrapment. Consequently, by the time the water has moved through a series of water retention devices such as reservoirs, STA's, SAV's and FEB's, it is a reasonable assumption that any organic P introduced to the ATSTM will be in a dissolved, or colloidal form. Dissolved organic phosphorus then becomes a major challenge for the system when the effluent target is 10 ppb total phosphorus. There are of course enzymes in nature that can break the bonding between an organic complex and phosphate. These have been shown to be produced by specialized bacteria and algae species.

Wright and Reddy (2001) noted that Alkaline Phosphatase was inversely related to phosphorus loading within Everglade's soils. This would be expected, as there would be no selective advantage to organisms capable of producing Alkaline Phosphatase if soluble phosphate was relatively abundant—a phenomenon known as feedback inhibition.

DB Environmental (2002) determined the susceptibility of organic phosphorus to both Alkaline Phosphatase and Phosphodiesterase (PDEase) within the influent and effluent to an SAV cell (Cell 4). They noted that the organic P retained by a 0.4 mm filter was the most recalcitrant, with only 27% hydrolysable by PDEase. For all fractions, about 71% of the organic P was hydrolysable by PDEase. Alkaline Phosphatase was not nearly as effective in conversion of Organic P, indicating that most of the organic P was as diesterase-hydrolysable phosphorus.

While it is reasonable to expect PDEase producing organisms to become favored in a low phosphorus environment, and to expect a significant portion of the total phosphorus to be PDEase hydrolysable, there remains some question related to the ability to promote and sustain these organisms within an ATSTM environment. Of concern would be the removal of such organisms with harvesting, and the issue of whether sufficient residual standing crop is left behind. One could also argue that within an ATSTM type operation in which there is minimal P storage, that the only available target for PDEase would be the transient organic P within the water column, whereas sediment held organic P might be an additional target within PSTA and other passive systems in which P storage is the mechanism for removal from the water column. This would be advantageous to the ATSTM system, as enzymatic regeneration of SRP from sediment stores would not be an issue of concern.

In an effort to assess the rate of organic P conversion or capture within an ATSTM, HydroMentia conducted studies in which water samples were taken at intervals down an active flowway at the S-154 MAPS facility. Noted in Figure 10 are these trends. From this data, it appears that organic P removal equals, and typically exceeds ortho P removal within the ATSTM. The implication is that organic P hydrolysis, which generates ortho P, is occurring at a rate equal to or greater than ortho P uptake or precipitation. This provides strong indication that enzymatic activity is extensive within the ATSTM. As this particular flowway is harvested once weekly, it also appears that the enzyme-producing organisms are sustained on the actively managed flowway. It is of course necessary to recognize that perhaps not all of the organic P removal may be associated with enzymatic activity, as particulate capture could also be involved. However, if there were no hydrolysis of organic P, it would be expected that ortho-P, not being replenished, would be reduced noticeably down the flowway. This is not the case however, providing support to the probability that enzymatic activity is prevalent.

Included within the S-154 data review is a summary of algal community changes down the ATSTM flowways. As a general trend, it has been noted that the prevalence of filamentous green algal fades with distance from the influent. As would be expected, the production rates appear to also decline, as noted by a visually observed reduction in biomass. Desmids and diatoms appear to be much more competitive in the downstream regions of the flowway. This is similar to what Browder et al. (1994) reported for soft oligotrophic waters within the Everglades. The oligotrophic blue-green algae are virtually absent throughout all of the flowways. While oligotrophic blue-greens such as *Schizothrix calacicola* might be expected to develop in very low nutrient applications, it is noteworthy that Adey et al. (1993) observed development of green algae and diatoms even within the high alkalinity, calcareous waters of the EAA with total phosphorus concentrations of 26-70 ppb. He also noted a paucity of blue-

green algae.

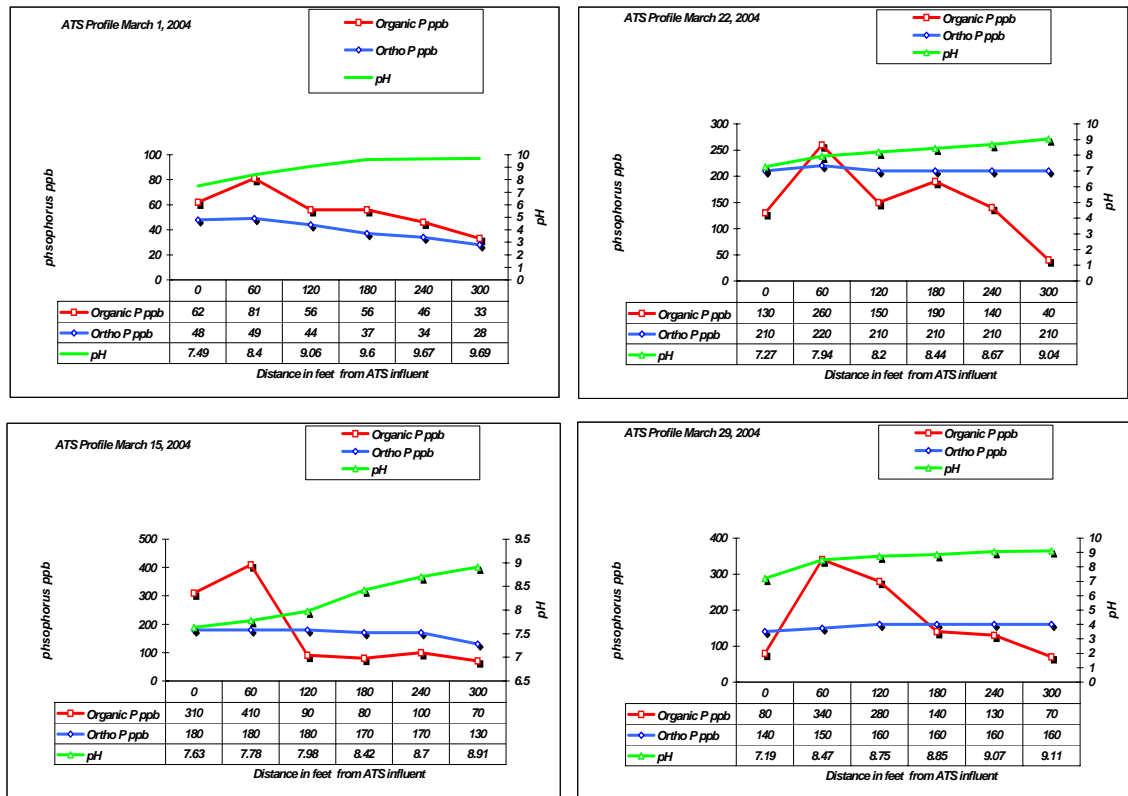


Figure 10: Sequential removals of ortho and organic phosphorus down ATS™ flowway S-154 MAPS facility.

The phosphorus content of the algal biomass is another critical parameter. Typically, it would be thought that the relative abundance of phosphorus within aquatic plant tissue would be a function of the water column concentration. This was discussed by Gopal (1987) in a review on water hyacinths. He cites a study by Sato and Kondo (1981) in which the phosphorus content of water hyacinths was expressed by the hyperbolic equation:

$$1/X_p = 6.545/f + 0.8668$$

Where X_p = tissue phosphorus
 f = phosphorus concentration in the water column.

In general, HydroMentia has noted this trend in water hyacinth, although as observed by Reddy and Tucker (1983), phosphorus uptake appears more closely related to nitrogen uptake—as nitrogen is recognized typically as the growth controlling agent. For the S-154 MAPS facility for example the best correlation for water hyacinth tissue phosphorus ($r^2 = 0.18$) is noted with total nitrogen concentration, as seen in Figure 11.

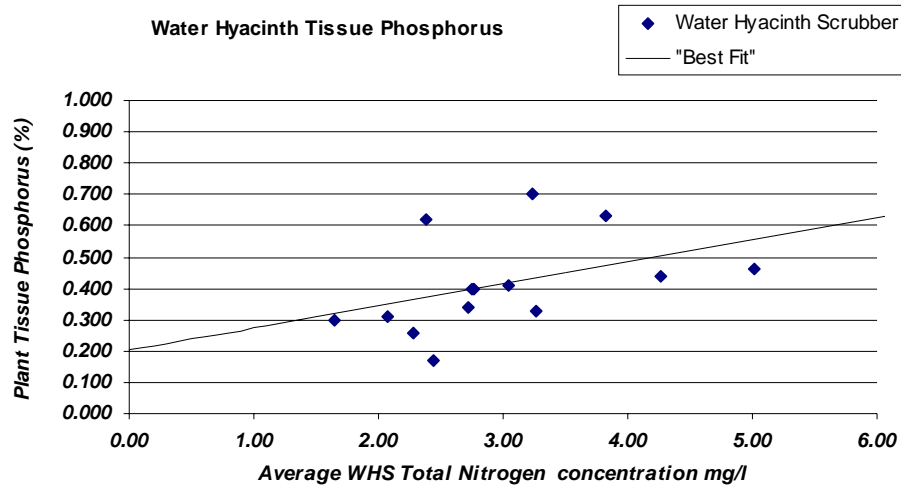


Figure 11: Tissue phosphorus content versus total nitrogen concentration for S-154 MAPS water hyacinth scrubber (WHS™).

With the algae biomass however no meaningful correlations could be found regarding tissue phosphorus content and nitrogen, phosphorus or alkalinity concentrations. This is noted in within the scattergram shown in Figure 12 as related to total phosphorus concentration.

For the ATST™ associated with the WHS™ - ATST™ system at the S-154 MAPS prototype, for 37 data points, the tissue phosphorus within the algal tissue averaged 0.51% with a standard deviation of 0.12%. Considering the wide degree of variation in other parameters, this is rather tight data pattern. Adey et al. (1993) noted a similar trend, with the tissue phosphorus content for 26 samples averaging 0.37%, with a standard deviation of 0.097%, which is also a tight pattern. For the three independent S-154 Maps ATST™ flowways, early tissue analysis indicate tissue phosphorus content from 0.50% to 0.60%. In work by Craggs et al. (1996) on an ATST™ unit operated at high nutrient levels (average TP = 2,400 ppb), the tissue phosphorus averaged 2.07% with a standard deviation of 0.16% (n = ca.100), which again is a rather concentrated data set, considering the environmental variability associated with a year long operational period. If the data sets from the S-154 MAPS operations, and the Adey et al. (1993) study, as well as the Craggs et al. (1996) study are plotted with average total phosphorus concentration across the ATST™ as the x-axis, and tissue phosphorus content as the y-axis, the resulting best fit equation is a straight line, indicating linear rather than hyperbolic relationship among a wide range of total phosphorus concentrations—see Figure 13. If this equation were applicable, it might be projected that at total phosphorus of 10 ppb, the tissue phosphorus would be just above 0.40%. However, this correlation analysis is on very limited data, and field work, e.g. an ATST™ Demonstration Project, would be necessary to confirm what changes occur in tissue phosphorus at lower water column total phosphorus concentrations. It is also quite possible that tissue phosphorus levels may respond directly to LHLR, an issue that will continue to be evaluated at the S-154 MAPS Prototype, and which needs close scrutiny in any ATST™ Demonstration Project.

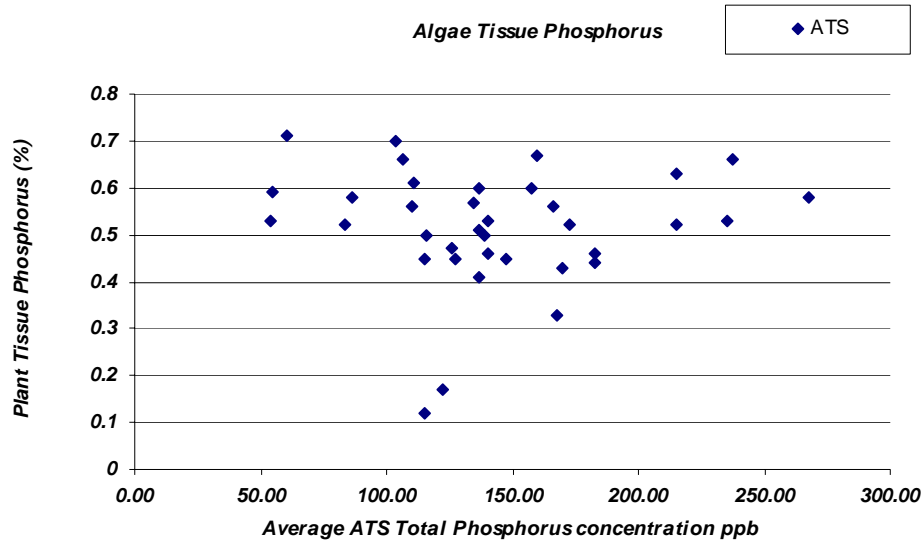


Figure 12: Algal tissue phosphorus content versus total phosphorus concentration for S-154 MAPS ATS™.

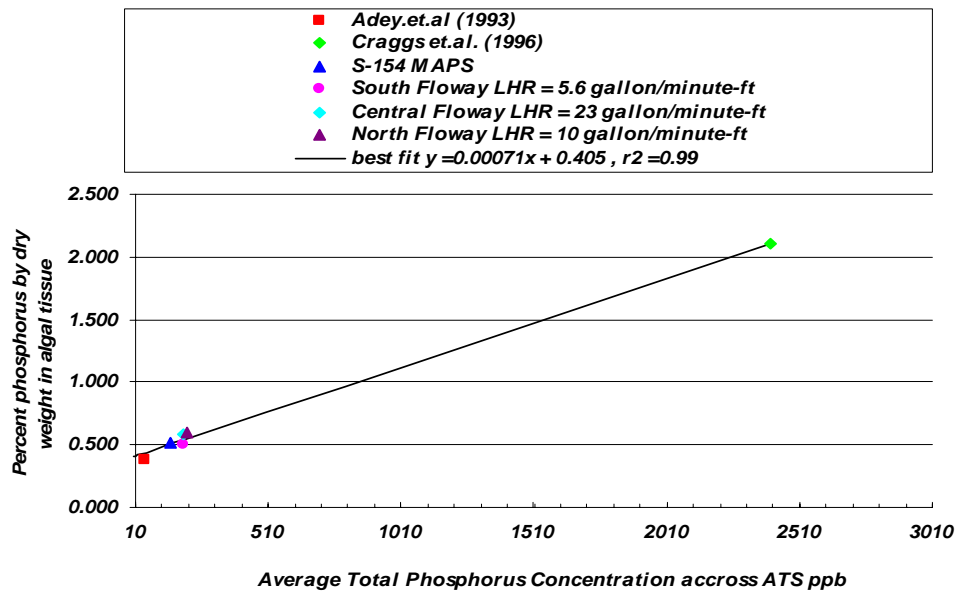


Figure 13: Average algal tissue concentrations versus average total phosphorus concentrations across the ATS™ floway for six separate floways.

LITERATURE CITED

- Adey, W.H. 1978. Coral reef morphogenesis: A multidimensional model. *Science* **202**:831-837.
- Adey, W.H. and T. Goertmiller. 1987. Coral reef algal turfs: master producers in nutrient poor seas. *Phycologia* **26**: 374-386.
- Adey, W.H. and J. Hackney. 1989. Harvest production of coral reef algal turfs. In *The Biology, Ecology and Mariculture of Mithrax spinosissimus Utilizing Cultured Algal Turfs*. W.H. Adey (Ed.) Mariculture Institute, Washington, DC.
- Adey, W.H., C. Luckett and K. Jensen. 1993. Phosphorus removal from natural waters using controlled algal production. *Restor. Ecol.* **1**: 29-39
- Adey, W.H. 1998. Coral Reefs: Algal-structured and mediated ecosystems in shallow, turbulent waters. *J. Phycology* **34**:135-148.
- Adey, W.H. and K. Loveland. 1998. *Dynamic Aquaria: Building Living Ecosystems*: 160-162, Academic Press, New York, NY USA ISBN 0-12-043792-9
- Brezonik, P.L., 1994. *Chemical Kinetics and Process Dynamics in Aquatic Systems*, 505-525, Lewis Publishers, Boca Raton, Florida, USA ISBN 0-87371-431-8
- Browder, Joan A., P.J. Gleason, D. R. Swift., 1994. Periphyton in the Everglades; Spatial variation environmental correlates and ecological implications. *Everglades the Ecosystem and its restoration*. Pages 379 – 418. Edited by Steven M Davis and John C. Ogden. St. Lucie Press. Del Ray, Florida.
- Carpenter, R.C., J.H. Hackney and W.H. Adey. 1991. Measurement of primary producer and nitrogenase activity of coral reef algae in a chamber incorporating oscillatory flow. *Limnol. Oceanogr.* **36**(1): 40-49.
- Craggs, R. W. Adey, W. Jessup and W. Oswald. 1996. A controlled stream mesocosm for tertiary and higher treatment of sewage. *Ecol. Eng.* **Vol. 6: 149-169**
- DB Environmental. 2002. *Demonstration of Submerged Aquatic Vegetation/Limerock Treatment Technology for Phosphorus Removal from Everglades Agricultural Area Waters: Follow-On Assessment prepared for the South Florida Water Management District and the Florida Department of Environmental Protection, West Palm Beach, Florida, USA.*
- Gopal, B., 1987. *Water Hyacinth* Elsevier, Amsterdam, ISBN 0-444-42706-6
- Kadlec, R. and W.W. Walker. 2003. *Dynamic Model for Stormwater Treatment for U.S. Department of Interior, Washington DC*
- Lineweaver, H. and D. Burke. 1934. The determination of enzyme disassociation constants. *J. Am. Chem. Soc.* **Vol 56:658**

Luckett, C., W.H. Adey, J. Morrissey, and D. Spoon. 1996. Coral reef mesocosms and microcosms – successes, problems, and the future of laboratory models. *Ecol. Eng.* **6**: 57-72

Odum, H.T., 1955. Trophic Structure and Productivity of Silver Springs, Florida. *Ecol. Monographs*, **27**, 55-112

Stewart, E.A., 2000. Estimates of Phosphorus and Nitrogen treatment Efficiency Aquatic Plant Based Wastewater Treatment System Phase 1A HydroMentia's Recirculating Aquaculture System, Okeechobee, Fl. Prepared for HydroMentia, Inc. Ocala, Florida, USA.

Wright, A.L and K.R. Reddy (2001) Phosphorus loading effects on extracellular enzymatic activity in Everglades wetland soils. *Soil.Sci. Am. J* **65**, 588-596