KINETICS OF BACTERIAL MORTALITY IN GRANULAR BED WETLANDS

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ABSTRACT
Faecal coliform and faecal *streptococci* die-off was investigated in the surface flow granular bed constructed wetland cell. The granular bed cell of effective plan area of 7.5 m × 1.5 m and 0.7 m deep was filled with aggregate size 19 mm up to a depth of 0.35 m. The remaining 0.35 m depth was filled with water. To assess the performance of the granular bed, samples were collected at the influent and effluent of the cell for calibration of a mathematical model. The analysis of the model suggests that bacterial mortality rate constant was largely influenced by sunlight intensity, which was responsible for 72.6% of removal. The influence of other parameters such as interception, filtration, dissolved oxygen, temperature, sedimentation on bacterial mortality rate were less significant. For instance, dissolved oxygen and pH contributed 7% and sedimentation only 0.44%. The ratio for the rate constant term to light attenuation coefficient (k_s/K) was 1.281 × 10^{-3} m cm^2/cal and the model efficiency was 0.80.

INTRODUCTION

Granular-based subsurface flow beds have been adopted for polishing of variety of wastewater in numerous countries of the world (Billore *et al*., 1998). These beds consist of shallow, water filled basin with a porous media through which water flows. The use of granular beds for wastewater is emerging as a viable alternative, especially for small sized communities and isolated areas (Swanson and Williamson, 1980). These systems are attractive because of the relative low cost in terms of capital investment, operation and maintenance costs compared to other techniques which are used for upgrading effluents from waste stabilization ponds (Stutz-McDonald and Williamson, 1979; Billore *et al*., 1998). Moreover, the use of granular beds provides a relatively simple and inexpensive solution for treatment of polluted water from small communities, industries, storm water, and agricultural runoff (Vymazal *et al*., 1998).

Granular bed constructed wetlands are effective in treating biochemical oxygen demand, suspended solids, nitrogen and phosphorus, as well as for reducing metals and pathogens (Adock, 2002). Effective granular bed performance depends on adequate pretreatment, conservation constituents, hydraulic loading rates, collection of monitoring information to assess system performance and knowledge of successful operation strategies (Vymazal *et al*., 1998). Researches conducted in the past have shown that granular beds are effective in removing pathogens and other pollutants from wastewater (Adock, 2002). The survival, fate and the distribution of microorganism in constructed wetlands depend on the type of the bed and associated phenomena that influence the death, other losses and growth.

One of the prime functions of constructed wetlands is the removal of pathogenic organisms, but factors affecting their decay have not been adequately modeled. Because of the similarity between the functioning of surface flow gravel bed constructed wetlands and other natural systems such as waste stabilization pond and filtration systems, it is reasonable to hypothesize that the mechanisms of pathogens removal in these systems are similar. A comprehensive model required should take into consideration the effects of temperature (Marais, 1974), solar radiation (Mayo, 1995), sedimentation (Auer and Niehaus, 1993), hydraulic dispersion (Polprasert and Bhattarai, 1985), pH (Parhad and Rao, 1974), dissolved oxygen concentration (Curtis *et al*., 1992), depth of water above gravel (Mayo, 1995), gravel bed particle size (McDowell-Boyer *et al*., 1993), adsorption and filtration (O'Melia, 1995).

The main objective of this study is to develop a model describing kinetics of coliform mortality removal in free water surface constructed wetlands. The specific objectives are to determine factors influencing bacterial mortality rate in surface flow constructed wetland, and to develop and determine model parameter governing equation.

METHODS AND MATERIALS

Description of the study area

Samples were collected from a pilot constructed wetland of dimensions of 7.5 x 1.5 x 0.7 m and a bottom slope of 1%. To achieve plug flow conditions, length to width ratio of 5 was adopted. This plant, which was constructed downstream of University of Dar es Salaam primary facultative ponds, was filled with aggregate of 19mm diameter and porosity of 35% up to a depth of 0.35 m and the depth of wastewater was varied up to a depth of 0.7 m (Fig. 1). The wastewater was received from a primary facultative pond treating wastewater of largely domestic characteristics with a small portion of chemicals from laboratories, the health center and workshops. The hydraulic detention time was obtained from the conductivity of the bed and the flow rate and the volume of water in the bed.

Sampling and sample examination

To obtain reliable results from any bacteriological analysis the standard procedure laid down for their collection, dispatch to the laboratory, storage and sample examination was strictly followed. All examinations were carried out in accordance with the standard methods as laid in the *Standard methods for the examination of water and wastewater* (1992). Temperature and pH were measured in the field using a pH meter (Metrohm, model 704). Dissolved oxygen was measured *in-situ* using a dissolved oxygen meter (TOA Electronics Limited, model DO-1B). Solar intensity records were obtained from a nearby computerized solar intensity recorder at the Department of Physics, University of Dar es Salaam.

The enumeration of faecal coliform and faecal *streptococci* closely followed procedure laid in the *Standard methods* (1992). The mortality rate constants of faecal coliform and faecal *streptococci* were calculated based on plug flow model (equation 1).

$$N_e = N_{ie}^{-kt} \quad (1)$$

In which N_e = Effluent bacteria number per 100 ml,

N_i = Influent bacteria number per 100 ml,

k = bacterial mortality rate (d^{-1}),

and t = retention time (d).

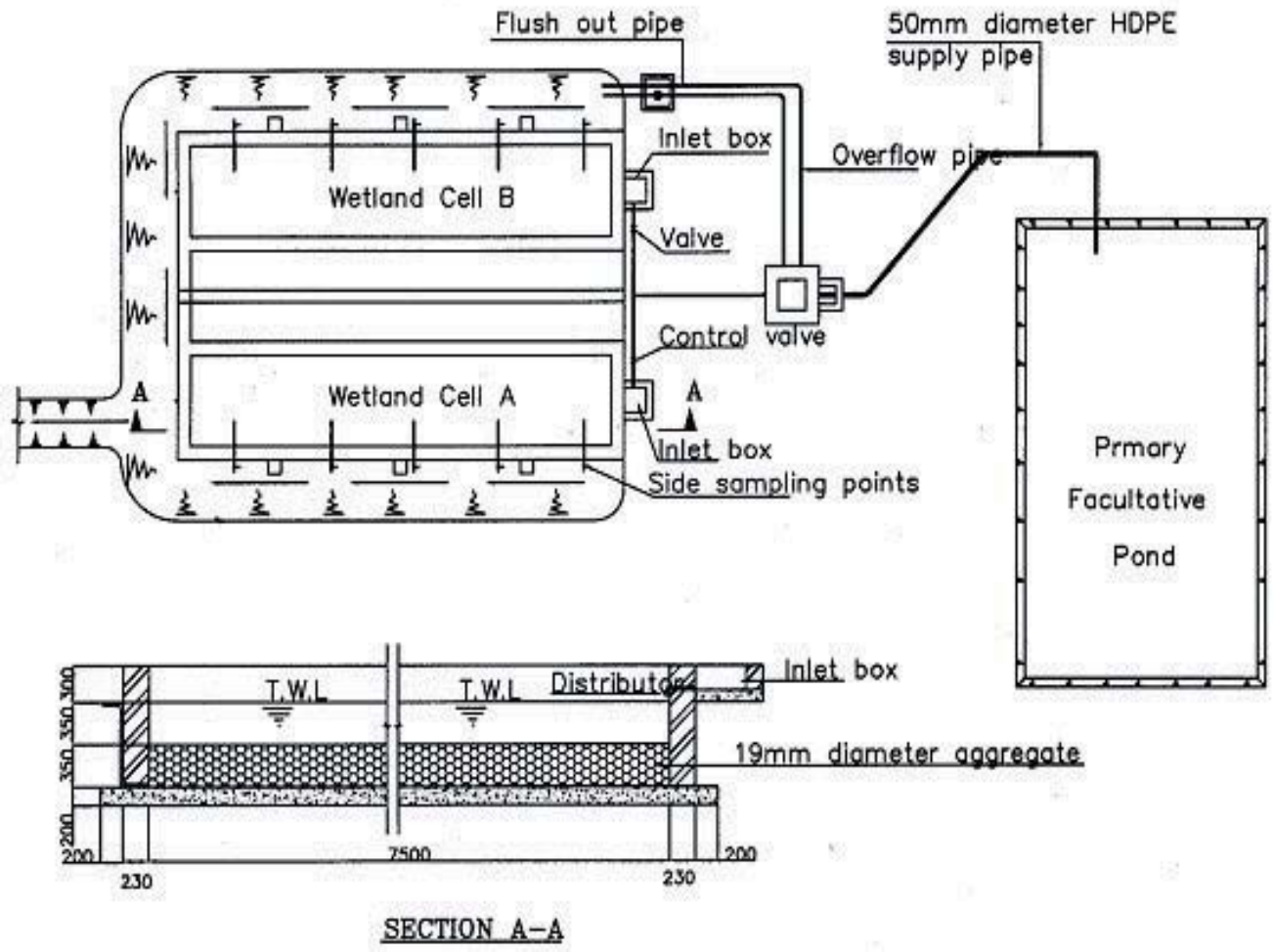


Fig. 1: Layout and cross section of experimental gravel bed wetlands

RESULTS AND DISCUSSION

Model development

A model was developed incorporating major mechanisms influencing coliform mortality in surface flow granular bed. The model assumed that the removal of bacteria in wastewater layer above the granular bed is influenced largely by solar radiation and other environmental factors such as dissolved oxygen, temperature and pH. The granular layer contributes through filtration, adsorption and sedimentation. The proposed model is shown by equation (2).

$$k = k_{20}\theta^{(T-20)}\left(\frac{k_s}{KH}\right)S_o^{\ a}\left(1-e^{-KH}\right)(pH)^b(DO)^c + \frac{3}{2}\alpha\eta\left(\frac{1-n}{d_c}\right) \quad (2)$$

in which k_{20} is the first-order bacterial mortality rate at 20°C, θ is the temperature coefficient, T is water temperature (°C), K is the light attenuation coefficient (m^{-1}), k_s is the rate constant for light mortality term (cm^2/cal), S_o is solar intensity received at the surface of granular bed (cal/cm^2/d), H is the depth of water column above the granular bed (m), DO is the concentration of dissolved oxygen, α is the sticking coefficient, n is the porosity of the bed, and d_c is the diameter of the collector, and a, b, and c are coefficients of the model.

Temperature is one of the major factors governing mortality of bacteria in wastewater (Marais, 1974). Unfortunately, inclusion of the temperature effect in the model was considered unrealistic because field water temperature was practically constant (28.0 ~ 30.9 °C) during the research period. To confirm if temperature had significant effect on mortality of bacteria within the observed narrow temperature range, temperature was plotted against mortality rates of faecal coliform and faecal *streptococci*. The resulting regression coefficients (R^2) for faecal coliform and faecal *streptococci* were 0.04 and 0.00, respectively. This poor correlation confirms similar observations made elsewhere (Auer and Niehaus, 1993; Mayo, 1995). Sieracki (1980) reported that the death of *Escherichia coli* was unrelated to temperature over the temperature range of 9 ~ 20°C. Mitchell and Chamberlin (1978) cited work on Ohio River by Frost and Streeter (1924), which demonstrated that total coliform decay rates were identical between 5°C and 20°C. In a study of mortality of total coliform in waste stabilization ponds, Moeller and Calkins (1980) observed no temperature-related differences in the mortality rate coefficient among five temperature groups. Therefore the temperature term was removed from equation (2) and the resulting model is shown by equation (3).

$$k = \left(\frac{k_s}{KH}\right)S_o^{\ a}\left(1-e^{-KH}\right)(pH)^b(DO)^c + \frac{3}{2}\alpha\eta\left(\frac{1-n}{d_c}\right) \quad (3)$$

The analytical determination of η involves some assumptions concerning flow velocity, steady state, and, where appropriate, the diffusion alone has been developed by Levich (1962). Methodology for considering sedimentation and interception alone is presented elsewhere (Yao, 1968). The following results were obtained:

$$\eta_D = 4Pe^{2/3} = 0.9\left(\frac{kT}{\mu d_p d_c V_o}\right)^{2/3} \quad (4)$$

$$\eta_I = \frac{3}{2}\left(\frac{d_p}{d_c}\right)^2 \quad (5)$$

$$\eta_G = \frac{(\rho_{V-}\rho)gd^2}{18\mu V_o} \quad (6)$$

The value h_D, h_I and h_G represent theoretical values for the single collector efficiency when the sole transport mechanisms are diffusion, interception, or sedimentation, respectively, and P_e is the Peclet number. The single collector efficiency can be numerically approximated by the sum of the analytical expressions. In other words

$$h = h_D + h_I + h_G \quad (7)$$

The discrepancies between the Yao model and observed removal rates led Yao *et al*. (1971) to propose incorporating a correction factor A_s derived by Happel (1958) in the collisions generated by diffusion. The Happel correction factor is defined by equation (8).

$$A_S = \frac{2(1-p^5)}{2-3p+3p^5-2p^6} \quad (8)$$

Where $p = (1-n)^{1/3}$. Including this term transforms (7) into equation (9).

$$h = 4A_S^{1/3}N_{pe}^{-2/3} + \frac{3}{2}N_R^2 + N_G \quad (9)$$

Where:

$$N_{pe} = \frac{ud_c}{D_p}$$

is the peclet number,

$$N_G = \frac{g(r_p - r)d_p^2}{18m}$$

$$N_R = \frac{d_p}{d_c}$$

is the particle settling velocity, is the interception number, g = the gravitational constant m/s^2, d_p = particle diameter in m, d_c = collector diameter in m, r_p = the density of particles in kg/m^3, r = the fluid density in kg/m^3, m = fluid coefficient of dynamic viscosity in Ns/m^2, and D_p = is the particle diffusion coefficient.

Model parameters

Model parameters used were obtained from the literature and some from the field. Table 1 shows the parameters used in the model and their sources. Fluid coefficient of dynamic viscosity, μ, was obtained from equation (10).

$$Log\left(\frac{\mu}{\mu_{20}}\right) = \frac{1.3272(20-T) - 0.001053(T-20)^2}{T+105} \quad (10)$$

The correlation matrix was developed to see how different physical factors affected the survival of fecal coliform in the hybrid gravel bed system.

The major contributing factor to fecal coliform removal in this study was solar intensity, which gave a correlation coefficient of 0.852. Other factors like pH, dissolved oxygen, temperature gave a poor correlation. In the model developed solar intensity, pH and dissolved oxygen were considered. Temperature was ignored because of the poor correlation.

The mortality rate due to diffusion, adsorption and filtration was obtained from equation (11).

$$k_f = \frac{3}{2}\eta\alpha\frac{V(1-n)}{d_c} \quad (11)$$

In which η = single collector efficiency; α = sticking efficiency; and V = velocity of flow, m/day. The value of η = was obtained from the expression given by O'Melia (1985) shown by equation (12).

$$\eta = 0.9A^{1/3}\left[\frac{K_B T}{\mu d_c d_p V}\right]^{2/3} + \frac{2}{3}A_S\left[\frac{d_p}{d_c}\right] + \frac{(\rho_{p-\rho})gd_p^2}{18\mu V} \quad (12)$$

From equation (8) the value of A_s was found to be 52.527. Substituting the values from Table 1 equation (12) becomes

$$\eta = 3.4577\times10^{-11}\left[\frac{TT}{\mu_T}\right]^{2/3}(d_p)^{-2/3} + 2.173\times10^5(d_p)^2 + 2.47065\times10^4\left(\frac{\rho_{p-995.7}}{\mu_T}\right)d_p^2 \quad (13)$$

Where TT = T°C + 273

From the results above the collection efficiency by Brownian

diffusion is given by equation (14).

$$\eta_D = 3.4577 \times 10^{-11} \left(\frac{TT}{\mu_T}\right)^{2/3} (d_p)^{-2/3} \quad (14)$$

This value η_D was 1.54 x 10^{-4}, which is small even under worst condition when the maximum diameter of particle was assumed (d_p = 40 μm and T = 30.9°C, μ_T = 0.7825, α = 0.009).

The mortality rate due to sedimentation is given by equation (15). In the model developed the mortality rate due to diffusion was ignored because of the poor results.

$$k_D = 97.54 \eta_D \alpha = 1.37 \times 10^{-4} \quad (15)$$

The efficiency by interception is given by equation (16).

$$\eta_I = 2.1734 \times 10^5 (d_p)^2 \quad (16)$$

Assuming the worst condition that is d_p = 40 μm, the $h_I = 3.47744 \times 10^{-4}$. The mortality rate due to interception is shown by equation (17) (α = 0.009)

$$k_I = 97.54 \alpha \eta_I = 0.0003053 \quad (17)$$

The mortality rate due to interception was insignificant and was therefore rejected in the developed model. The final model was developed considering solar intensity, pH, dissolved oxygen and sedimentation.

$$k_{pred} = \left(\frac{k_s}{KH}\right) S_o^{\ a} \left(1 - e^{-8H}\right) (pH)^b (DO)^c + 2.409 \times 10^6 \left(\frac{\rho_{p-\rho}}{\mu_T}\right) d_p^{\ 2} \quad (18)$$

The efficiency criterion, which is widely accepted for testing of the model accuracy, was used to determine the efficiency of the developed model. The model efficiency R^2, was defined analogously to the coefficient of determination in linear regression, as the proportion of the initial variance accounted for by the sum of square of differences between the observed and estimated mortality rates.

The Fortran program was written to determine the parameter and the efficiency of the model. The resulting model after running the program is as shown in equation (18). The model gave R^2 of 0.80. The inclusion of sedimentation, in the model didn't improve the efficiency of the model significantly because large particles settled in the primary facultative pond. The diameter of particle of 2 μm obtained in this study was relatively small compared to diameters of up to 40 μm obtained in literature (Metcalf and Eddy, 1995). This is because the gravel bed received wastewater from the primary facultative pond, which allowed particles to settle before being discharged to the secondary facultative ponds. Since the intake was near the effluent of the primary facultative pond, it is obvious that only few particles were able to reach the intake of the unit. At the intake the screen was placed to prevent large particles from entering the pipe to reduce the possibility of clogging the system. This contributed to less number of particles of large sizes reaching the cell. Sedimentation was also taking place in the distribution chamber built to achieve continuous flow of wastewater to the unit resulting into further reduction of density of particles reaching the unit.

The parameter in the model were found to be $\frac{k_s}{KH}$ = 0.00366, b = 0.762, c = 0.329, d = 0.088, ρ_p=1100 kg/m^3, d_p = 2 μm, H = 0.35 m, K = 8 m^{-1}, k_s = 0.01025 cm^2/cal (Table 1).

$$k_{pred} = 0.00366 S_o^{\ 0.762} (1 - e^{-8H}) (pH)^{0.329} (DO)^{0.088} + \frac{0.00905}{\mu_T} \quad (19)$$

m_T = Viscosity of water in centipose.

Fig. 2 shows the variation of influent and effluent fecal coliform in the hybrid gravel bed wetland unit. From this figure it was observed that the values of the observed effluent fecal coliform were fitting very well with the predicted ones. However, after 27 days of experiment the performance of the system was poor because of the growth of plant species identified as *Lemna Aequinolictialis Welm*, a minute floating aquatic flowering plant without distinct stems or real leaves, but consisting of usually floating, leaf-like, ovate, elliptic or more or less orbicular thalli, 1- several millimeters in diameter. These plants formed a scum on the gravel bed unplanted wetland that inhibited the penetration of light.

Some of the coefficients in the developed model compare very well with coefficients found in the literature. The value of k_d obtained in this work was 0.156 d^{-1}, which is within the range reported in the earlier work on Onondaga lake (0.97 ~ 1.87 d^{-1}, Stearns and Wheler, 1979) and waste stabilization ponds (k_d = 0.081~144 d^{-1}) (Sarikaya and Saatci, 1987; Mayo, 1989; Mayo, 1995). Light attenuation coefficient K of 8 m^{-1} observed in this study falls within a typical range observed in waste stabilization ponds (7.8 ~ 16 m^{-1}) (Sarikaya *et al*., 1987; Calkins *et al*., 1976). The rate constant for the light mortality term k_s = 0.01 cm^2/cal observed in this study was considerably higher than the k_s of 8.24x 10^{-4} cm^2/cal from a lake water sample (Auer and Niehaus, 1993).

Application of the model

To illustrate the use of the proposed die-off model in the design, assuming that solar intensity S_o (cal/cm^2/d), pH and dissolved oxygen (mg/l) are known, then the faecal coliform mortality rate can then be calculated from equation (19). The value of k determined will then be substituted in plug flow model to determine the retention time of the system, based on the desired effluent quality. Equation (20) can be used to determine the density of faecal coliform in the gravel bed if the density of faecal coliforms in the influent is known.

$$N_e = N_i \exp[0.00366 S_o^{\ 0.762} (1 - e^{-8H}) (pH)^{0.329} (DO)^{0.088} + \frac{0.00905}{\mu_T}] \quad (20)$$

CONCLUSIONS

The effects of pH, temperature, solar intensity, dissolved oxygen; filtration, adsorption and sedimentation on faecal coliform mortality were studied. It was found out that the mortality rate k, was directly proportional to solar intensity. Other factors such as pH, dissolved oxygen, temperature, filtration, adsorption and sedimentation did not alter the value of the mortality rate significantly due to their insignificant effects.

Table 1: Parameters used in the model

Coefficient	Units	Range	Source
Light mortality constant (k_s)	cm^2/cal	0 . 0 0 8 2 4 - 0.011	Auer and Niehaus (1993), Mayo (1995)
Light attenuation coefficient (K)	m^{-1}	7.8 - 25	Moeller and Calkins (1980); Sarikaya *et al*. (1987); Polprasert and Khatiwada (1998)
Boltzman constant (K_B)	J/K	1.38×10^{-23}	Weast (1981)
Diameter of particle (d_p)	μm	0.5 - 40	Metcalf and Eddy (1995)
Density of water ρ	Kg/m^3	995.7	Weast (1981)
Density of particle ρ_p	Kg/m^3	1050 - 1500	Metcalf and Eddy (1995)
Flow velocity (V)	m/s	2.20×10^{-5}	From the field
Porosity (n)		0.35	From the field
Diameter of collector (d_c)	m	0.01905	From the field
Sticking coefficient (α)	-	0.005 - 0.03	Polprasert and Khatiwada (1998)
Wetland depth (H)	m	0.35	From the field.

From experiment observations, it was possible to develop a mathematical model of faecal coliform mortality in the surface constructed wetland for the tropical climate.

$$k_{predicted} = 0.00366 S_o^{0.762}\left(1-e^{-8H}\right)(pH)^{0.329}(DO)^{0.088} + \frac{0.00905}{\mu_T}$$

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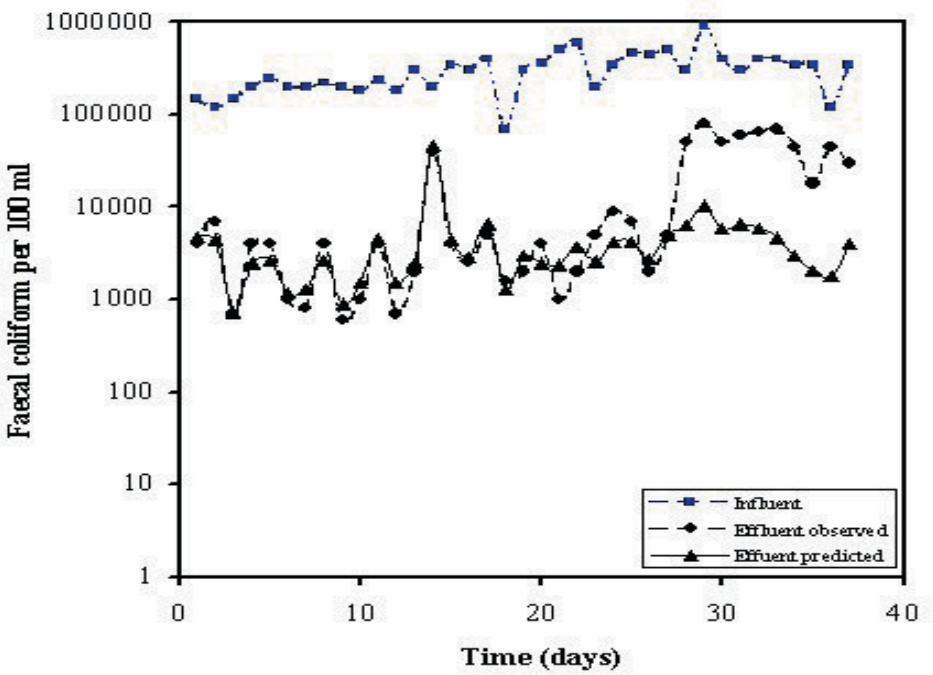


Fig. 2 : Variation of influent and effluent Fecal coliform density with time

REFERENCES

Adock, P. (2002) Subsurface gravel bed wetlands. Internet http://www.wetlands.com.an/gravelbeds.htm.

Auer M. T. and Niehaus S. I. (1993) Modeling fecal coliform bacteria (I). Field and laboratory determination of loss kinetics. *Wat. Res.* **27**(4), 693-701.

APHA (1992) *Standard methods for the examination of water and wastewater*, 19th edn. Am. Publ. Hlth Assoc., Am. Wat. Works Assoc. and Wat. Environ. Fed., Washington, D. C., 1992.

Billore S. K., Singh N., Sharma J. K., Dass P. and Nelson R. M. (1998) Horizontal flow subsurface constructed wetlands with *Phragmites karka* gravel bed treatment of municipal wastewater carried in ground - earth network in tropical central India. 6th *Internat. Conf. on Wetland Systems for Water pollut. Control*, Sao Paulo, Brazil.

Calkins J., Buckles J. D., and Moeller J. R. (1976) The role of solar ultraviolet radiation in natural water purification. *Photochem. photobiol.*, Pergamon Press, Elmsford. N. Y., 49-57.

Curtis T.P., Mara, D.D. and Silva S.A. (1992) The effect of sunlight on faecal coliforms in ponds: Implications for research and design. *Wat. Sci. Technol*., 26(7-8), 1729-1738.

Happel, J. (1958) J. of *Amer. Inst. Chem. Engrs.,* 4, 197-201.
Levich V. G. (1962) Physicochemical hydrodynamics. Prentice-Hall, Englewood Cliffs, N. J., 80-85.

McDowell-Boyer L. M., Hunt J. R. and Arnold R. G. (1993) Particle transport through porous media. *Wat. Resources Res*., 22(13), 1901-1921.

Marais G.v.R (1974) Faecal bacteria kinetics in stabilization ponds. *J. Env. Engrg*. *Div*., *Am. Soc. Civ. Engrs*, 100(1), 199-139.

Mayo A. W. (1989) Effect of pond depth on bacterial mortality. *J. Env. Engrg*, *Am. Soc. Civ. Engrs,* 115(5), 964-977.

Mayo A.W. (1995) Modeling coliform mortality in waste stabilization ponds. *J. Env.Engng., Am. Soc. Civ. Engrs*, 121(2), 140-152.

Metcalf and Eddy, Inc (1995) *Wastewater Engineering, Treatment, Disposal and Reuse*. McGraw-Hill Ltd, New York.

Mitchell R. and Chamberlin C. (1978) Survival of indicator organisms. In *Indicators of Viruses in Water and Food* (Edited by Berg G.), pp. 15-35. Ann Arbor Science, Ann Arbor, Mich.

Moeller J. R. and Calkins J. (1980) Bacteria agents in waste water lagoons and lagoons design. *J. Water pollut. Control Fed*., 52(10), 2442-2451.

O'Melia C. R. (1995) Particles, Pretreatment, and performance in water filtration. *J. Env Engng., Am. Soc. Civ. Engrs*, 116(6), 874-905.

Parhad N. M. and Rao N. U. (1974) Effect of pH on survival of *Escherichia coli*. *J. Water poll. Control Fed*, 46(5), 980-986.

Polprasert C. and Bhattarai K. K. (1985) Dispersion model for waste stabilization ponds. *J. Env. Engrg. Div., ASCE*, 111(1), 45-49.

Polprasert C. and Khatiwada N. R. (1998) Kinetics of fecal coliform removal in constructed Wetlands.

Sarikaya H. Z. and Saatci A. M. (1987) Bacteria die-off in waste stabilization ponds. *J. Env. Engrg*, *Am. Soc. Civ. Engrs*, 113(2), 366~382.

Sarikaya H. Z., Saatci A. M. and Abdulfattah A.F. (1987) Effect of pond depth on bacteria die-off. *J. Env. Engrg*, *Am. Soc. Civ. Engrs,* 113(6), 1350~1362.

Sierack M. (1980) Effects of exposures of natural sunlight on decay rates of enteric bacteria and coliphage in a simulated sewage outfall microcosm. M.Sc. thesis, University of Rhode Island, Kingston, R.I.

Stutz-McDonald S.E. and Williamson K.J. (1979) Settling rates of algae from wastewater lagoons. *J. Env. Engrg*, *Am. Soc. Civ. Engrs*, 105(2), 273~282.

Swanson, G.R. and Williamson, K.J. (1980) Upgrading lagoon effluents with rock filters. *J. Env. Engrg*, *Am. Soc. Civ. Engrs*, 106(6), 1111~1129.

Vymazal J., Brix H., Cooper P. F., Green M. B. and Habert R., (1998) Removal mechanisms in constructed wetlands. In: *Constructed Wetlands for Wastewater Treatment in Europe.* Backhuys publishers, Leiden, The Netherlands, pp 95-121.

Weast R. C. (1981) Handbook of chemistry and physics, 61st edition, CRC Press, Boca Raton, Florida, USA.

Yao K., Habibian M. T. and O'Melia C. R. (1971) Water and wastewater filtration: concepts and applications. *Envir. Sci. Technol*., 5(11), 1105-1112.

Yao K. M. (1968) Influence of Suspended Size on the Transport Aspect of Water Filtration. Ph.D. dissertation, University of North Carolina, Chapel Hill, N. C.