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Determination of Some Vitamins in Different Matrixes in a Municipal Wastewater Treatment Plant

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Abstract: This paper presents various vitamin levels determined by using High Performance Liquid Chromatography (HPLC) in the influent and effluent of municipal wastewater treatment plant and its some sludge fractions. A, E and C vitamin levels in the influent were found to be 0.11, 0.84 and 5.16 mg/l, respectively. The levels of A vitamin of the microorganisms in the aeration tank and the effluent were determined to be 0.39 mg/kg and 0.17 mg/l, respectively. E vitamin levels in the aeration tank and effluent were also determined to be 6.61 mg/kg and 1.32 mg/l, respectively. C vitamin levels in aeration tank and effluent were 15.7 mg/kg and 7.35 mg/l, respectively. A, E and C vitamin levels of the sludge cake in the dried beds were found to be 0.44 mg/kg, 20.95 mg/kg and 11.39 mg/kg, respectively. A, E and C vitamin levels in the sludge cake disposed for a long time were also assigned to be 0.09, 4.76 and 1.93 mg/kg.

Key words: Municipal wastewater, treatment plant, vitamin

Introduction

Activated sludge treatment of wastewater is used to treat a wide of range of wastewater worldwide (Clark and Stephenson, 1998). The process is an aeration oxidation system. The process performance is based on the mixture level of influent with microbial population under aerobic conditions. It takes place the removal of organic matter associated with the microbial growth as the compounds in the influent are oxidized by microorganisms in sludge medium. The microbial mass and the oxidation products occur at the consequence of that. Biomass kind of the occurred products is based on the operation method, the influent characteristics and the microorganism kind in the system. The efficiency of a biological wastewater treatment plant depends upon the environment conditions such as the level of dissolved oxygen and nutrient, the addition of trace elements and vitamin, the mixture regime and physical conditions. Not only are vitamins necessary for normal activities of human and animals, but they are also necessary for matter change of high vegetables and microorganisms. Many researchers suggested vitamins having a catalytic affect to overcome the deficiency of micronutrients in the wastewater treatment and thus they stated that the microbial activity and treatment efficiency increased (Lemmer and Nitschke, 1994; Lemmer *et al*., 1998; Burgess *et al*., 2000).

The aims of this paper are to determine the contents of A, E and C vitamin at influent and effluent of treatment plant and at sludge fractions of various processes and to present their behaviours through overall system.

Materials and Methods

Samples were supplied weekly from Elazig-City Wastewater Treatment Plant as composite for two hours. Wastewater Treatment Plant is a conventional activated sludge process treating municipal wastewater. The current plant consisted of the main units of screen, grift chamber, primary settling tank, activated sludge (aeration) tank, final settling tank, sludge thickening, digestion tank and dried beds. Solid samples were collected at depth of 15 cm in drying beds and disposal area.

0.5 g of solid samples and 1.5 ml of water samples for each samples were taken for analysis. Samples for analysis of A and E vitamins were centrifuged by adding 2 ml ethanol. After that, it was separated by centrifuge, followed by the addition of 0.3 ml hexane, and mixed well, and then vitamins were extracted. This step was repeated again, and n-hexane phase added to the first one. Under nitrogen atmosphere, n-hexane was evaporated to dryness. Then 0.2 ml methanol was added to solve the residue followed by injection to HPLC. Mobil phase used in HPLC was methanol, acetonitrile and chloroform (47:42:11 v/v). The quantification was according to Catignani (1983); Miller *et al*. (1984) utilizing absorption spectra of 326 and 296 for vitamin A and E respectively, HPLC separations were accomplished at room temperature with a Cecil liquid chromatography system (Series 1100) consisting a sample injection valve (Cotati 7125) with a 20 µl sample loop, an ultra-violet (UV) spectrophotometric detector (Cecil 68174), integrator (HP 3395) and a Techsphere ODS-2 packed (5 µm particle and 80 ^{0}A pore size) column (250 x 4, 6 ID) with a methanol: acetonitrile: chloroform (47: 42: 11, v/v) mobile phase at 1 ml min^{-1} flow rate.

Vitamin C was determined by the following procedure: 0.5 ml of 0.5 M perchloric acid was added to 0,5 g sewage sludge sample Cerhata *et al*. (1994) and then the volume of sample brought to 3 ml, after that it was

centrifuged for ten minutes at 4000 rpm, The supernatant was filtered and the vitamin C level was determined using the method of Tavazzi *et al.* (1992) by HPLC utilizing a column (250 x 3,9 ID) packed with Tecopak C18 reversed-phase material (10 µm particle size) with for Vitamin C mobile phase (3,7 mM phosphate buffer, pH 4,0), flow rate was 1.0 ml/min; absorption wavelength was 246 nm. All chemicals and reagents used were of analytical grade and were purchased from Merck Results for the groups are expressed as means±S,D.

Results and Discussion

A, E and C vitamin levels in the influent of Elazig City Municipal Wastewater Treatment were found to be 0.11, 0.84 and 5.16 mg/kg, respectively. The organic, nutrient and trace matters in the wastewater are consumed by the mixed microorganism culture for the growth of new viability generations. To realize the relationship between the microorganisms in the activated sludge and vitamins in the wastewater, the vitamin levels were either determined in the activated sludge or the effluent of the treatment plant. The vitamin level in the activated sludge means the amount of vitamin absorbed by microorganism bodies. While A vitamin level of the microorganisms in the aeration tank increased to 0.39 mg/kg. After the microorganisms were separated from the treated water through final settling tank, A vitamin level in the effluent was found to be 0.17 mg/kg. While a part of activated sludge (microorganisms) settled in the final settling tank is given to the dried beds after thickening and digesting for the sludge disposal, other part is also returned to aeration tank, which the ratio in the current plant was 0.5. A vitamin level of the sludge cake in the dried beds was found to be 0.44 mg/kg.

While E vitamin level of activated sludge in the aeration tank increased up to 6.61 mg/kg, the level in the effluent decreased to 1.32 mg/kg. Then, this value increased up to 20.95 mg/kg in the sludge cake of the dried beds.

While C vitamin of activated sludge in aeration tank increased up to 15.7 mg/kg, the level in the effluent decreased to 7.35 mg/kg, and again increased up to 11.39 mg/kg in the sludge cake of the dried beds.

A, E and C vitamin levels in the sludge cake disposed for six months were also assigned to be 0.09, 4.76 and 1.93 mg/kg. The results indicated that the resistances of vitamin against the degradation were 22.7, 20.5 and 16.2 % for E, A and C at six months period, respectively. The results indicated that E vitamin was absorbed successfully by the microorganism in the activated sludge systems and even could be produced, A vitamin level didn't change too much and only accumulated in the microorganism structure, C vitamin was wasted from microorganism structure at a short time although it could be easily absorbed by their mass. However, it was shown that these vitamins decreased in the sludge cake with time due to environmental conditions and C vitamin was also the most unsteady one.

Conclusion: Vitamins are specific organic compounds required for the growth oh organisms and the synthesis of enzymes. It is known that some vitamins are produced by bacteria, fungus and yeast. There are important differences between microorganism species according to vitamin demand. As autotrophy organisms synthesize the vitamins required for their activity, heterotrophy organisms consume vitamins. Lemmer and Nitschke (1994) reported that vitamin demands of aerobic and anaerobic bacteria were in ppb levels. In this study, it was shown that vitamin levels in all compartment of treatment plant were higher than ppb. Leclerc *et al.* (1991) and Warman and Havard (1997) found that content of vitamin and mineral matter in crops increased with organic manuring. When treatment sludge and treated water are used in the agricultural fields, A, E and C vitamins increase the biological activity of soil and thus crop yield by developing plant growth.

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