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Understanding the Causes of Sludge Bulking and Foaming Phenomena at Al-Bireh Wastewater Treatment Plant

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The findings, interpretations and conclusions expressed in this study do not necessarily express the views of Birzeit University, the views of the individual members of the MS.c committee or the views of their respective employers.

DEDICATION

To my mother and my father for continuous support and encouragement

To all my brothers and sisters

Eng. Hitham, Dr. Ayman and Eias for support

Dr. Abood and Eng. Mohammad in Russia

To Anas in the United States

To Mo'ath, Shatha and Leen

To Dr. Joman Obaid

To all my friends

To my supervisor, Dr. Rashed Al-Sa'ed

To Al-Bireh wastewater treatment plant operator, Nayif Tummaleh

With all my love and respect

Eman Hasan

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ABSTRACT

Filamentous bulking and foaming are considered the major problems in the operation of activated sludge systems (ASS), that consists of slow settling and poor compaction of solids in the clarifier causing high total suspended solid (TSS) content in the effluent with adverse impacts on process performance. The goal of the study was to present the main reasons that cause the sludge bulking and foaming in Al-Bireh Wastewater Treatment Plant (AWWTP) by analyzing the physical, chemical and biological parameters of both aeration and sedimentation tanks (1) and (2). Also the goal was to verify the process performance and sustainability of the ASS with special emphasis on process operation relationship and their implications for effluent reuse. This study was conducted to the main reasons that caused sludge bulking and foaming in AWWTP by monitoring of the sludge volume index (SVI) of both aeration tanks (1) and (2) over a period of ten months. The SPSS software was used to make ANOVA and regression analysis to differentiate between the critical parameters that are responsible for increased SVI. The decrease of the temperature, the dissolved oxygen (DO) concentration and the increase of fat, oil and grease (FOG) level and the F/M ratio were the main reasons that lead to the increase of the SVI especially in aeration tank (1).

The results showed that the bulking and foaming was mainly caused by the excessive propagation of some filamentous bacteria. The dominant

filamentous bacteria identified from mixed liquor and foam samples included a long branched form of *Nocardia, Microthrix parvicella, Type 1701, and Sphaerotilus natans*. *Microthrix parvicella,* was dominant in the cold winter and spring period while *Nocardia* was dominant in warm weather.

All filamentous bacteria identified were found in both samples that came from aeration tanks and scum throughout the study period. It is concluded that specific filamentous bacterial population in mixed liquor and foaming activated sludge was constant and not dependent on variability of seasons.

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LIST OF ABBREVIATIONS

APHA	American Public Health Association
ANOVA	Analysis of Variance
ASS	Activated Sludge System
AWWTP	Al- Bireh Wastewater Treatment Plant
BOD_5	Biological Oxygen Demand
CFU	Colony Forming Unit
DGGE	Denaturing Gradient Gel Electrophoresis
DNA	Deoxyribonucleic Acid
COD	Chemical Oxygen Demand
DO	Dissolved Oxygen
DSVI	Diluted Sludge Volume Index
EPA	Environmental Protection Agency
FC	Fecal Coliform
FISH	Fluorescent *In Situ* Hybridization
F/M	Food to Microorganisms Ratio
FOG	Fat, Oil, and Grease
HRT	Hydraulic Retention Time
IEWS	Institute of Environmental and Water Studies
MCRT	Mean Cell Retention Time
MLSS	Mixed Liquor Suspended Solid
MSA	Mannitol Salt Agar
NA-agar	Nutrient Agar

NTU	Nephelometric Turbidity Units
PCR	Polymerase Chain Reaction
PE	Population Equivalent
RAS line	Return Activated Sludge line
RNA	Ribonucleic Acid
SPSS	Statistical Package for the Social Sciences
SV_{30}	Sludge Volume after 30 minutes
SVI	Sludge Volume Index
TC	Total Coliform
TKN	Total Kjeldahl Nitrogen
TSS	Total Suspended Solids
UASB	Up flow Anaerobic Sludge Blanket
UV	Ultra Violate

CHAPTER ONE

Background and problem definition

1.1 Introduction

The activated sludge system (ASS) is the most commonly used technology for biological wastewater treatment. It consists of two stages, a biochemical stage (aeration tank) and a physical stage (secondary clarifier). In the aeration tank, organic carbon, ammonium and phosphate are removed from the waste water by the activated sludge. Worldwide, ASS is considered an accepted technology with over thousands of systems treating domestic, urban and industrial sewage. The activated sludge process is a reliable and adequate technology for developing countries where financial and human resources are secured. Another benefit is the effluent quality and sludge stabilization, which is different from the centralized energy-intensive schemes promoted for urban centers and few rural areas (Cooper and Green, 1995; Griffen and Upton, 1999). However, the concept of sustainability is crucial with regard to economic and social development of Palestinian rural communities; and attention must be drawn to treatment processes which attain the criteria of environmental impact on a local scale. In the context of municipal wastewater treatment, the ideal process uses minimal land area and is feasible in construction and operation, whilst providing effective treatment to meet standards which protect the environment at the lowest possible economic and social cost.

Filamentous bulking and foaming (Figures 1 and 2) are considered the major and serious problem in the operation of activated sludge processes, that consists of slow settling and poor compaction of solids in the clarifier causing high TSS content in the effluent with adverse impacts on process performance (Kappeler and Gujer, 1994).

Figure 1. The foam in aeration tank (2) in AWWTP (March, 2010).

Figure 2. The foam in aeration tank (1) in AWWTP (April, 2010).

Considerable efforts have been made to identify the type of filaments in actual activated sludge processes and to understand their microbiological characteristics (Parker *et al*., 2004).

Currently, the ASS, which serves a population equivalent (PE) of about 45,000, is having particularly difficult influent conditions to meet, with a variable daily flow rate and varying loads exacerbated by large point sources such as domestic septage and industrial discharges. Loads vary dramatically throughout the year - especially for septage and industry. The ideal urban sewage works must be able to deal with varying diurnal flows and loads; and resist excessive loss of treatment capability during wet weather flow periods.

Full treatment capability must be rapidly achieved after an extended period and the process should be robust - giving effective and reliable treatment throughout the year to the desired standard. The ability of well-designed, properly operated and maintained ASS with few process malfunctions to achieve the required quality standards (i.e. BOD, SS, TN, pathogens) has been extensively reported (Mino, 1995; Orizo et al., 1997).

In the growing number of conflicts between agricultural and domestic use of scarce water resources in Palestine, an increased use of treated wastewater for irrigation purposes is vital. However, it is crucial that wastewater treatment systems should comply with prescribed national and regional effluent hygienic standards to reduce water borne disease and produce an acceptable effluent quality.

Since 2000, the AWWTP, a single stage activated sludge process achieving simultaneous nitrification and denitrification with aerobic sludge stabilization was put into operation for the treatment of municipal wastewater in Al-Bireh city and a few refugee camps within the Al-Bireh-Ramallah district. This study will try to evaluate the efficiency of this treatment plant when sludge bulking and foaming phenomena appear.

CHAPTER TWO

2.1. Literature review

The activated-sludge process is a biological method of wastewater treatment that is performed by a variable and mixed community of microorganisms in an aerobic aquatic environment. These microorganisms derive energy from carbonaceous organic matter in aerated wastewater for the production of new cells in a process known as synthesis, while simultaneously releasing energy through the conversion of this organic matter into compounds that contain lower energy, such as carbon dioxide and water, in a process called respiration. Also, a variable number of microorganisms in the system obtain energy by converting ammonia nitrogen to nitrate nitrogen in a process termed nitrification. This consortium of microorganisms, the biological component of the process, is known collectively as activated sludge (Albertson, 1987; Wanner, 1994; Casey *et al*., 1995).

The biological component of the activated sludge system is comprised of microorganisms. The diversity of the biological community is very large, containing many species of viruses, bacteria, protozoa, fungi, metazoan and algae. The microorganisms that are of greatest numerical importance in activated sludge are bacteria. Some bacteria are strict aerobes (they can only live in the presence of oxygen); whereas others are anaerobes (they are active only in the absence of oxygen). The preponderance of bacteria living in activated sludge are facultative-able to live in either the presence or absence of oxygen, an important factor in the survival of activated sludge when dissolved oxygen concentrations are low or approaching depletion. The former predominate bacteria in activated sludge system are heterotrophic and autotrophic bacteria.

Heterotrophic bacteria (floc-formers), obtain energy from carbonaceous organic matter for the synthesis of new cells such as *Pseudomonas*, *Zoogloea*, *Achromobacter*, *Citromonas*, *Flavobacterium*, and *Arthrobacter* (Eikelboom, 2000; Wagner *et al*., 2002; Jenkins, *et al*., 2004; Tsang *et al*., 2008). Autotrophic bacteria obtain their energy by oxidizing ammonia nitrogen to nitrate nitrogen in a two-stage conversion process known as nitrification. Two types of nitrifying bacteria are *Nitrobacter* and *Nitrosomonas* (Clayton *et al*., 1991; EPA, 2002). Nitrifying bacteria have a slower rate of reproduction than heterotrophic and there are representing as a small percentage of the total population of microorganisms in activated sludge.

$$\mathbf{NH_3 + O_2 \rightarrow NO_2^- + 3H^+}$$

Nitrosomonas

$$\mathbf{NO_2^- + H_2O \rightarrow NO_3^- + 2H^+}$$

Nitrobacter

The objective of the activated sludge process is to remove soluble and insoluble organics from the wastewater and to convert this material into a flocculent microbial suspension that settles well in a conventional gravity clarifier (Orhon and Artan, 1994; Eckenfelder and Musterman, 1995; Ramothokang *et al*., 2003). Floc-forming bacteria kept in the activated sludge process under the correct environmental conditions efficiently remove the organic material and nutrients from wastewater (Tixier *et al*., 2003; Sponza, 2003). However, not all bacteria in the activated sludge process are floc-formers. Many different types of filamentous bacteria have been identified in activated sludge and play important roles in wastewater treatment. Filamentous bacteria directly affect sludge settling as they make provision for the rigid support network or backbone upon which floc-forming bacteria can adhere and grow into suitable activated sludge flocs (Richard, 1989; Ramothokang *et al*., 2003; Rossetti *et al*., 2005). The excessive growth of filamentous microorganisms will cause problems in activated sludge system.

An important feature of the activated sludge process is the formation of settleable solids by the balanced growth of filamentous and floc-forming organisms which are subsequently removed by gravity and settle in secondary settlers (Flores-Alsina, 2009). A good separation (settling) and compaction (thickening) of activated sludge in the secondary clarifier is a necessary condition to guarantee a good effluent quality from the activated sludge process (Alleman and Prakasam, 1983; Albertson, 1987). Many problems can develop in an activated sludge operation that adversely affects effluent quality such as:

1. Dispersed growth

Dispersed growth occurs when bacteria that are not associated with the flocs are consumed by protozoa. This phenomenon is present in high numbers as dispersed cells result in a turbid effluent. Dispersed growth is associated with the failure of flocs forming bacteria to bioflocculate (Jenkins *et al*., 2004).

2. Pinpoint flocs

This phenomenon occurs when a very small, weak floc is formed in activated sludge that can easily pass as flotation in the final clarifier leading to a turbid effluent. These small flocs, termed pin floc, consist only of floc-forming bacteria without a filament backbone and usually are <50 µm in diameter. Pin floc occurs most commonly at starvation conditions, a very low F/M and long sludge age. Chronic toxicity can also cause a pin floc condition (Richard, 2003).

3. Rising sludge

Rising sludge is the result of excess denitrification, which results from an anoxic condition in the settling tank. This phenomenon appears as sludge particles attached to rising nitrogen bubbles, form a sludge blanket at the surface of the clarifier and cause a turbid effluent with an increased BOD_5. According to

Richard (2003), denitrification problems are more prevalent during the warmer times of the year and can be more severe when a filamentous sludge present due to more extensive entrapment of the nitrogen gas bubbles by a filamentous sludge.

4. Foaming and scum formation

Foams are dispersions of gas in a liquid. Their formation is a physical process, which is divided into three phases according to Linke and Berger (2010).

a. The initial phase is the generation of gas bubbles into the lower part of a liquid phase by bubbling gas. The smaller the pore size, the more homogeneous is the bubble swarm.

b. The gas bubbles rise up through the liquid phase due to the density difference, while surface-active compounds such as proteins or enzymes adsorb to the gas–liquid interface. This decreases the surface tension of the liquid, and the formation of foam is initiated.

c. Finally, the gas bubbles leave the liquid phase and cover its surface. The foam column grows.

Biological foaming in activated sludge is a common problem encountered in many wastewater treatment plants around the world (Jenkins *et al.*, 2004). It can be described as the formation of a scum layer on the surfaces of aeration basins and secondary clarifiers due to the presence of large quantities of hydrophobic filamentous and possibly non-filamentous microorganisms (Davenport and Curtis, 2002; Frigona *et al*., 2006). Foaming in activated sludge plants has been attributed to the combination of the presence of surfactants (detergents), biosurfactants (substances produced during the metabolic activity of microorganisms) and the presence of two groups of filamentous bacteria, *Nocardia* sp. and *Microthrix parvicella* (Ganidi, *et al*., 2009). According to Richard (2003), the description and causes of activated sludge foams can be classified into six groups Table 2.1.

Table 2.1. Description and Causes of Activated Sludge Foams (Richard, 2003)

Foam Description	**Cause(s)**
Thin, white to grey foam.	Low cell residence time or "young" sludge.
White, frothy, billowing foam.	Once common due to nonbiodegradable detergents (uncommon).
Pumice-like, grey foam (ashing).	Excessive fines recycle from other processes (e.g. anaerobic digesters).
Thick sludge blanket on the final clarifier(s).	Denitrification.
Thick, pasty or slimy, greyish foam (Industrial systems only).	Nutrient-deficient foam; foam consists of polysaccharide material released from the floc.
Thick, brown, stable foam enriched in filaments.	Filament-induced foaming, caused by *Nocardia*, *Microthrix* or type 1863.

Thick, brown foam is always common in the most activated sludge system (De los Reyes, 2002; Richard 2003; Ganidi, *et al*., 2009). *Nocardia* and *Microthrix parvicella* are believed to be the main causative organisms of filamentous foaming.

Nocardial foaming appears to be the most common of activated sludge plants in the world. This phenomenon appears as thick, stable and brown foam inches to many feet thick on aeration basin and final clarifier surfaces. This foam consists of activated sludge flocs containing large amounts of *Nocardia* filaments growing from their surface and is quite stable. These foams are easy to diagnose microscopically using simple Gram stain. The most common *Nocardia* species found in such foams are *Nocardia amarae* (*N. amarae*). These organisms were able to utilize the fatty acids with a different length of carbon chain for their growth. They had lower maximum specific growth rates and smaller saturation constants, and can survive on a low concentration of fatty acids (Tsang *et al*., 2008). *N. amarae* are not pathogenic to laboratory animals; however, other less frequently isolated actinomycete strains are known opportunistic human pathogens (e.g. *N. caviae*, *N. brasiliensis*, *N. asteroides* and strains of *Mycobacterium*), but no actual infection has been documented (Richard, 2003).

Microthrix parvicella (*M. parvicella*) was linked with foaming in the activated sludge system (Ganidi, *et al.*, 2009). It is also hydrophobic and utilizes long chain fatty acids as carbon source. *M. parvicella* can store excess long chain fatty acids in large globules and has an advantage over other bacteria for water-insoluble fats and lipids due to its hydrophobicity.

These foams also occur equally in domestic and industrial plants. Industrial wastes promoting *Nocardia* growth (and foaming) include dairy, meat, food processing, pharmaceutical, and any others that contain a significant amount of grease, oil or fat.

Severe foaming causes a number of operational problems. These include blockages of gas mixing devices, foam binding of sludge recirculation pumps, fouling of gas collection pipes due to entrapped foam solids, foam penetration between floating covers and digester walls and tipping of floating covers during foam expansion and collapse. Other problems include aesthetics, odors; and safety hazards if they overflow basins to cover walkways and handrails. In cold weather these foams can freeze; and would need "pick and shovel" removal. Foam may escape to the effluent, increasing effluent suspended solids and compromising disinfection.

5. Sludge bulking

Sludge bulking has been one of the major problems affecting biological waste treatment in activated sludge process. There are two distinct types of thought that attribute the causes of bulking to the type of microbial population in the activated sludge (Schuler and Jassby, 2007). These are:

i. Non filamentous bulking

Non filamentous bulking is caused by excess production of exopolysacharides by a special type of bacteria called *Zooglea ramigera*. This phenomenon is sometimes called zoogleal bulking (Hossain, 2004). The Zoogleal bulking might surprisingly occur when a system is modified to improve setting by incorporating plug-flow characteristics (Novak., *et al*, 1993). *Zoogloea* occurs at high F/M conditions and when specific organic acids and alcohols are high in amount due to a septic condition or low oxygen conditions (Richard, 2003).

ii. Filamentous bulking

Filamentous bacteria are normal permanent residents of activated sludge with single cell unit and are not dominant under normal conditions. There are forms of filaments under certain conditions that cause a problem when numerically dominant (Eikelboom, 2000; Jenkins *et al*., 2004; Schuler and Jassby, 2007; Li *et al*., 2008; Guo, 2010). Filamentous bacteria have been known for decades now, to be the main cause of activated sludge bulking in wastewater treatment worldwide (Eikelboom, 1982; Kappeler and Gujer, 1994; Krhutkov'a *et al.*, 2002; Schuler and Jassby, 2007). Filamentous bulking is a problem that consists of slow settling and poor compaction of solids in the clarifier of activated sludge system. This phenomenon is usually caused by excessive growth of filamentous microorganisms.

A bulking sludge can result in the loss of sludge inventory to the final effluent, causing environmental damage and effluent violations. In severe cases, loss of the sludge inventory can lead to a loss of the plant's treatment capacity and failure of the process. Additionally, disinfection of the treated wastewater can become compromised by the excess solids present during bulking. In less severe cases, bulking leads to excessive return sludge recycle rates and problems in waste activated sludge disposal. Many problems in waste sludge thickening are really filamentous bulking problems (Casey *et al*., 1995).

Sometimes the presence of some filaments may serve to catch and hold small particles during sludge settling, yielding a lower turbidity effluent. It is only when filaments grow in large amounts (approximately 107 µm filaments per gram of activated sludge) that hindrance in sludge settling and compaction occurs (Casey *et al*., 1995; Richard, 2003).

Filamentous bulking is the number one cause of effluent noncompliance today in the world. There are several survey about the sludge bulking and foaming phenomenon and the appearances of various filamentous microorganisms in activated sludge that have been made in the world.

According to Faheem and Khan (2009), the dominant filamentous bacteria identified from mixed liquor and foam samples in Dubai included: actinomycetes species, *Thiothrix*, Type 021N, *Sphaerotilus natans*, *Beggiatoa* and *Nostocoida limicola.*

In South Africa, Blackbeard *et al*; (1986) reported that *Microthrix parvicella* and Types 1851, 0041, 0675 and 0914 were the main filamentous microorganisms that were dominant in activated sludge system. Mino, (1999; 1995) and Antonio; *et al*., (2004) reported that *S. natans*, Types 021N, 0041, 0675, and *Thiothrix* sp were common in Japan, while; Types 021N, 1701, 0092, 0041, and 0675 were dominant in Thailand.

In Europe, *M. parvicella*, Type 0092 and Types 0041 were common in Denmark, Germany, Greece and the Netherlands (Kristensen *et al*., 1994; Eikelboom *et al*., 1998; Krhutkov'a *et al*., 2002; Xie *et al*., 2007; Fia'kowska, and Pajdak-Sto, 2008). While in France, Germany, Italy, Netherlands and United Kingdom *M. parvicella*, Types 0041, 0675, 0092, 021N, *H. hydrossis* and *N. limicola*, were apparently the major morphotype filaments, mainly responsible for the bulking that was observed in biological nutrient removal activated sludge systems (Foot, 1992; Eikelboom, 1994; Kruit *et al*., 1994; Kunst and Reins, 1994; Pujol and Canler, 1994; Rossetti *et al*., 1994; Madoni *et al*., 2000 and; Lavender *et al*., 2003).

In North America; Types 1701, 021N, 0092, 0041, 0675, and *M. parvicella* were dominant in the most activated sludge systems (Switzenbaum *et al*., 1992). Also in South America, Type 1701, *S. natans*, *M. parvicella*, and Type 0041, 0675 were common (Di Marzio, 2002). According to (Seviour *et al*., 1994), *M. parvicella*, Types 0041, 0675, 0092, and *H. hydrossis* were the most common filamentous found in Australia (Antonio *et al*., 2004; Fia'kowska, and Pajdak-Sto, 2008).

Earlier studies were focused on finding effective methods to control and prevent filamentous bulking sludge since employing activated sludge process in wastewater treatment plants (Contreras *et al*., 2004; Martins *et al*., 2004; Comas *et al*., 2008; Fialkowska and PajdakStós, 2008; Al-Mutairi, 2009). The dominant approaches to eliminate bulking sludge mainly include non-specific methods such as chlorination, ozonation, metal salts and polymer. Specific strategies to control filamentous bacteria are by finding relationships between the dominant filamentous bacteria and the operational conditions (Caravelli *et al*., 2004; Martins *et al*., 2004; Juang, 2005 and; Agridiotis *et al*., 2007).

Acording to Nakhla and Lugowski (2003) and Al-Mutairi, (2007), the most widespread engineering tools that were employed to control filamentous bulking are bio-selectors including aerobic, anoxic and anaerobic selectors. Anaerobic selectors showed the best performance to combat filamentous bacteria for municipal and industrial wastewater (Jolis *et al*., 2007).

Filamentous bulking and foaming are common and serious problems in activated sludge operation that affect most activated sludge plants at one time or another.

To date, there has been a limited report on sludge bulking and foaming phenomena in an activated sludge system in Palestine especially since the

AWWTP is the only activated sludge plant in all West Bank. This study will try to find the main reasons that cause the sludge bulking and foaming in AWWTP.

2.2. Measurements of sludge settleability

Many previous studies have sought to determine the relationship between filament content and settleability (Tsang *et al*., 2006; Schuler and Jassby, 2007). Settlebility of sludge may be measured as sludge volume (SV) in a 1000 ml measuring cylinder (height of 36 cm) after 30 minutes sedimentation and is expressed for a known initial sludge concentration as sludge volume index (SVI) (Hultman *et al*., 1991; Löwén and Piirtola, 1998). In some parts of the world, the diluted sludge volume index (DSVI) is now preferred. This involves settling one liter of MLSS and a series of diluted samples of MLSS. The sample used to determine the DSVI is that which gives a settled volume after 30 minutes of less than 25% (Lee *et al*., 1983; Forster, 2003; Jenkins *et al*., 2004). According to (Hultman *et al*., 1991; Wiley and Sons, 1997; Bye and Dold, 1998; Löwén and Piirtola, 1998), the measurements of DSVI were determined by a series of diluted samples to obtain SV_{30} smaller than 200 ml after 30 minutes. The DSVI test has been found to be better correlated with filament content than the undiluted SVI test or the stirred test (Lee *et al*., 1983; Schuler and Jassby, 2007).

$$\mathbf{SVI = SV_{30}/SS_i}$$

$$\mathbf{DSVI = (SV_{30} * 2^n) / SS_i \quad for\ SV_{30} < 200\ (ml/l)}$$

SVI=sludge volume index (ml/g).

DSVI=diluted sludge volume index (ml/g).

SV_{30} =sludge volume after 30 minutes of settling (ml).

SS_i = initial suspended solids concentration of the mixed liquor (g/L).

n= number of two-fold dilutions required to achieve SV_{30} less than 200 ml.

An operational definition often using a high SVI (more than 150 ml/g) indicates bulking conditions whereas an SVI below 70 ml/g indicates the predominance of pin flocs (EPA, 1987a). Thus, a bulking sludge may or may not lead to a bulking problem, depending on the specific treatment plant's ability to contain the sludge within the clarifier (Casey *et al*., 1995; Richard, 2003).

2.3. Type of filamentous microorganisms

Ttwenty six different morphological types of Filamentous organisms can be found in activated sludge system, which were distinguished and grouped into seven assemblages Table 2.2 (Eikelboom, 1975; Eikelboom and van Buijsen, 1981; Kampfer, 1997; Motta *et al*., 2001; Jenkins *et al*., 2004). This is important as many of the filaments found in activated sludge have not been isolated in pure culture and hence their identity remains unknown (Richard, 2003).

Table 2.2 Filamentous organisms in activated sludge grouped and listed according to (Eikelboom and van Buijsen, 1981; Kampfer, 1997; Motta *et al*., 2001).

Group Number	Filamentous organisms
Group I	**Sheath-forming, Gram-negative bacteria** 1.Sphaerotilus natans 2.Type 1701 3.Type 1702 4.Haliscomenobacter hydrossis 5.Type 0321
Group II	**Sheath-forming, Gram-positive bacteria** 6.Type 0041 7.Type 0675 8.Type 1851
Group III	**Sheathless curled, multicellular bacteria resembling blue green algae** 9.Type 021N 10.Nostocoida limicola 11.Cyanophycae
Group IV	**Slender, coiled bacteria** 12.Microthrix Parvicella * 13.Type 0581 14.Type 0192
Group V	**Straight, multicelluler, Gram-negative bacteria** 15.Type 0803 16.Type 1091 17.Type 0092 18.Type 0961
Group VI	**Filamentous bacteria motile by gliding** 19.Type 0914 20. Beggiatoa spp. 21.Type 1111 22.Type 1501
Group VII	**Additional Types** 23.Type 1863 ** 24.Type 0411 25.Fungi 26. Nocardia spp. **

* This filament causes both bulking and foaming.

** These filaments cause foaming only.

2.4. Isolation and identification of filamentous microorganisms

Filamentous Identification should be used as a tool to monitor the health of this biomass when a filament problem is suspected. Filamentous Identification is used to determine the type of filaments present so that, a cause can be found and corrections can be made to the system to alleviate future problems.

Filamentous microorganisms can be determined by different separate approaches. One of these approaches is the isolation of the number of filaments in pure culture. This approach has been successful for *M. parvicella*, *S. natans,* type 1701, *H. hydrossis*, type 021N, and *Thiothrix*. Their competitive growth abilities were examined in laboratory studies. Many of these studies are summarized by Jenkins *et al*. (1993; 2004). Another approach to identify filamentous bacteria in activated sludge system is by a direct microscopic examination using simple Neisser and Gram staining (Eikelboom, 2000; Jenkins *et al*., 2004).

Molecular methods based on DNA and RNA analyses were introduced to biological wastewater treatment plants. These methods allow a correct identification of the filamentous bacteria population. Therefore, it is advisable to apply specific gene probes (Amann *et al*., 1995). For activated sludge, two methods that are presently commonly used are Denaturing Gradient Gel Electrophoresis (DGGE) and Fluorescence *In Situ* Hybridization (FISH) combined with polymerase chain reaction (PCR) (Wagner *et al*., 1994; De los Reyes *et al*., 1998; Hussein, 2006).

All filamentous bacteria usually have a process control variation associated with the type of filament present that can be implemented to change the environment present and select out for floc forming bacteria instead.

2.5. Some factors causing filamentous bulking

There are six growth conditions that cause the overgrowth of filaments in activated sludge. Four of these occur in municipal wastewater systems while all six occur in industrial wastewater systems, with two specific only to industrial systems (low nutrients and low pH). Many of the filaments have been associated with other causes in the past, but recent work has indicated the causes given in Table 2.3 as the primary reason for their growth. Some filaments have more than one cause and the combination of conditions listed may favor bulking by a particular filament more so than any single condition.

Table 2.3. Causes of Filament Growth in Activated Sludge system

Causative Condition	Filament Types
1. Low Dissolved Oxygen Concentration	*Sphaerotilus natans* Type 1701 *Haliscomenobacte hydrossis*
2. Low F/M	Type 0041 Type 0675 Type 1851 Type 0803
3. Septicity	Type 021N *Thiothrix* I and II *Nostocoida limicola* I,II,III * Type 0914* Type 0411 Type 0961* Type 0581* Type 0092*
4. Grease and Oil	*Nocardia* spp. *Microthrix parvicella* Type 1863
5. Nutrient Deficiency a. Nitrogen: b. Phosphorus:	Type 021N *Thiothrix* I and II *Nostocoida limicola* III *Haliscomenobacter hydrossis* *Sphaerotilus natans*
6. Low pH	Fungi

Note that some filaments occur at several conditions. * These filaments occur at lower F/M at septic conditions.

2.5.1. Dissolved oxygen level

Low aeration basin dissolved oxygen (DO) concentration for the applied organic loading (F/M) leads to filamentous bulking by several filaments see Table 2.3. The rate of BOD removal is near maximum at 1.0 mg/L DO concentration, while the rate of nitrification is near maximum at 2.0 mg/L DO. The actual DO concentration within the biological flocs is less than that measured in the bulk solution around the flocs, due to oxygen use as it penetrates into the flocs (Richard, 1993; Martins *et al*., 2003; Guo, 2010). The DO concentration in the bulk solution around the flocs has to be high enough to maintain an aerobic floc interior. Since oxygen moves into the floc by diffusion, its bulk concentration needs to be high enough to reach the floc centers before becoming depleted (Palm *et al*., 1980; Richard *et al*., 1993).

The effective method to avoid filamentous bulking due to low DO is increasing DO concentration, usually up to approximate 2 mg/L or even more (Gaval and Pernelle, 2003; Huang and Ju, 2007; Guo, 2010).

2.5.2. Substrate concentration (F/M Ratio)

Four filaments type Table 2.3, are specifically caused by low F/M conditions. Low substrate concentration (low F/M ratio) appears to be the most prevalent cause of filamentous bulking. At low substrate concentration filamentous microorganisms have a higher substrate removal rate than that of floc-formers, which prevail at high substrate concentrations.

2.5.3. Septisity

Influent wastewater septicity is usually indicated by odours "rotten egg", H_2S smell and a dark colour to the wastewater, caused by precipitated ferric sulfide. Septic wastes contain elevated amounts of sulfides and low molecular weight organic acids (such as acetic and butyric acids), both of which encourage the growth of certain filaments (see Table 1.3). Observation of these filaments with intracellular sulfure granules is a tip-off of a septicity problem. Septicity is more common in systems in warmer climates and in those with large wastewater collection systems that have lift stations and force mains.

2.5.4. Fat, Oil and Grease (FOG)

The three filaments that cause foaming all grown on grease and oil, and these can become a problem when grease and oil are high in amount in the influent wastewater. Systems that lack primary clarification (the main grease and oil removal mechanism) appear to suffer more foaming problems.

2.5.5. Nutrients deficiency

Microorganisms require certain nutrients for growth. Nitrogen and phosphorus can be growth limiting if not present in sufficient amounts in influent wastewater, a problem with industrial wastes and not domestic wastes. In general, a BOD_5: N: P weight ratio in the wastewater of 100:5:1 is needed for complete BOD removal. Other nutrients such as iron or sulfur have been reported as limiting to activated sludge, but this is not common.

When proper nutrients are not available, the metabolism fails and a kind of bacterial exopolysaccharide (slime) will begin to accumulate around the cell. The cell slows down in activity because it cannot produce enough enzymes and because needed nutrients cannot penetrate the slime layer as they should. The sludge will not settle and BOD removal slows down.

2.5.6. Low pH

The aeration basin pH should be maintained in the range 6.5 to 8.5. Low pH, <6.5, otherwise, it may cause the growth of fungi and fungal bulking. The aeration basin pH can be adjusted using caustic, lime or magnesium hydroxide.

There are another two conditions that are responsible in the overgrowth of filaments in activated sludge such as.

2.5.7. Sludge loading and sludge age

The relationship between sludge loading and sludge age depends on whether the reactor is a completely mixed or plug flow system. In completely mixed systems, increasing sludge loading leads to a decrease of SVI and thus to a decrease of filamentous microorganisms. Some filamentous microorganisms occur over a wide range of sludge age (MCRT: mean cell retention time) values while others occur only at low or high values.

2.5.8. Low Temperature

Low temperature also causes the excessive growth of *M. pravicela*. These microorganisms maintain a higher growth rate than floc-forming microorganisms under low temperature.

2.6. Research Goal and Objectives

Proponents of activated sludge systems and official regulators such as the Palestinian Water Authority have expressed interest in more efficient and applicable guidelines for the development, design and permitting of such systems. However, the scientific knowledge on the occurrence, the causes of sludge bulking and foaming phenomena in Al-Bireh wastewater treatment plant and understanding the impacts of unit operations stability through the sludge bulking and foaming occurrence are still limited (Hussein, 2006).

The main goal of this research study is to present the main reasons that cause the sludge bulking and foaming in Al-Bireh wastewater treatment plant by analyzing the physical, chemical and biological parameters. Also, the goal is to verify the process performance and sustainability of the ASS process established in Al-Bireh with special emphasis on process operation relationship and their implications for effluent reuse.

Objectives:

i. The specific objectives are formulated in the following research question; does the process offer a reasonable performance to meet the local effluent quality guidelines and protect the public health and aquatic environment?

ii. To understand the relationship and correlation between pathogens and sludge bulking and foaming phenomena.

iii. Recommendation of ways to co ntrol these phenomena that may causes deterioration of the effluent and operational problems.

The finding of this research study will provide guidance for environmental performance, especially for projects which are intended to provide water reuse. Insight gained in this study will be helpful in updating the national guidelines for the use of treated effluent for agriculture purposes. Finally, the results gained will enhance the scientific knowledge and understanding of the relationship between the process malfunction as sludge bulking, foaming phenomenon and pathogens fate.

CHAPTER THREE

Materials and Methods

3.1. Site description and design data

The City of Al-Bireh is located in the center of West Bank, neighboring Ramallah in the middle of a series of mountains 15 km north of Jerusalem (Figure .3) and, in the center of the central mountains at approximately 860 m above sea level. Eastwards the mountains slope steeply towards the Jordan valley and westwards to the Mediterranean Sea about 48 km far from the Mediterranean coast .It has an area about 29,000 donums and has a population of over 45,000 peoples (Al-Bireh municipality, 2009).

Al-Bireh has a Mediterranean climate. The climate is classified into a wet season in the winter and a dry season in the summer. In general the climate is moderate and sub-humid with north-easterly winds in the summer and; autumn seasons and south-westerly winds in the winter, and spring seasons. The average temperature in the summer is 22^{o}C and the humidity percentage reaches 55%. The annual average temperature is 16^{o}C and it decreases in winter to 8.5^{o}C (Palestinian Statistics Centre, 2009).

The annual average precipitation in Al-Bireh is between 500 mm to 600 mm; and the average daily amount of wastewater discharge from Al-Bireh is 5000 m3/day (Al-Bireh municipality, 2009).

Al-Bireh has a geographical importance relating to wastewater where a system of wadis drains the area in a south-eastern direction. Al-Bireh wastewater treatment plant for the treatment of domestic and industrial wastewater has been constructed in the El-Ain wadi 2 km south of the town centre. It contributes to the improvement of environmental conditions and prevents further groundwater pollution. Due to the scarcity of water in the West Bank, the effluent will be reused to irrigate suitable agricultural crops and woodland. Plant design was chosen to meet BOD levels below 20 mg/l and total suspended solids below 30 mg/l.

Figure 3.The location of Al-Bireh City in the West Bank (Eman, 2010)

The wastewater treatment plant, put a new second oxidation ditch into operation, and it, is currently serving a population equivalent of about 38,000 capita, where the municipal sewage is reaching the plant by gravity (Figures. 4a and 4b). The main of operation consists of a preliminary treatment stage (screening and aerated grit chambers), two oxidation ditches, 2 secondary settling tanks, a UV disinfecting unit (out of order); and an effluent regulation tank. The sludge line consists of a gravity thickener for the excess sludge and 2 filter presses for biosolids dewatering. For more details on technical design and process dimensioning refer to published literature elsewhere (Al-Sa`ed, 2007; Albireh Municipality, 2000).

Figure 4a. Overview of AWWTP

Figure 4b. Biosolids dewatering (Filter presses)

Disinfection is usually the final step before the treated effluent is discharged into Wadi Al-Ain, a small seasonal wadi flowing towards the Jericho district passing a few pristine freshwater springs. The effluent is planned to be used for agricultural irrigation in the Deir Dibwan area. With increased pollution levels, disinfection is considered as the final barrier within AWWTP fence.

Currently, the treated effluent is discharged into the nearby wadi without being disinfected as the planned reuse is still pending due to technical and social problems. The stabilized biosolids are disposed of at the municipal landfill in the district without being used for agricultural purposes.

3.2. Sampling sites

In order to monitor performance, sampling points were set up at (a) the influent to the screening units, (b) the effluent from the secondary settling tanks 1&2, (c) the aeration tank 1&2, (Figure. 5).

In this study, the samples were taken from the influent, the aeration tank 1&2 and the effluent from secondary settling tank 1&2 once a week, every Saturday at 11.00 a.m. The samples were taken in wet and dry weather for four seasons (autumn, winter, spring and summer) starting from October 2009, to July 2010 through sterile bottles (APHA, 1995) ;and transported within half an hour to Birzeit University laboratory using an icebox then analyzed within 24 h. Also samples were taken from the foam (scum) layer of the aeration tank through sterile bottles in an ice box. The parameters like temperature, pH and dissolve oxygen (on surface and at a depth of 3.5 m) were measured onsite.

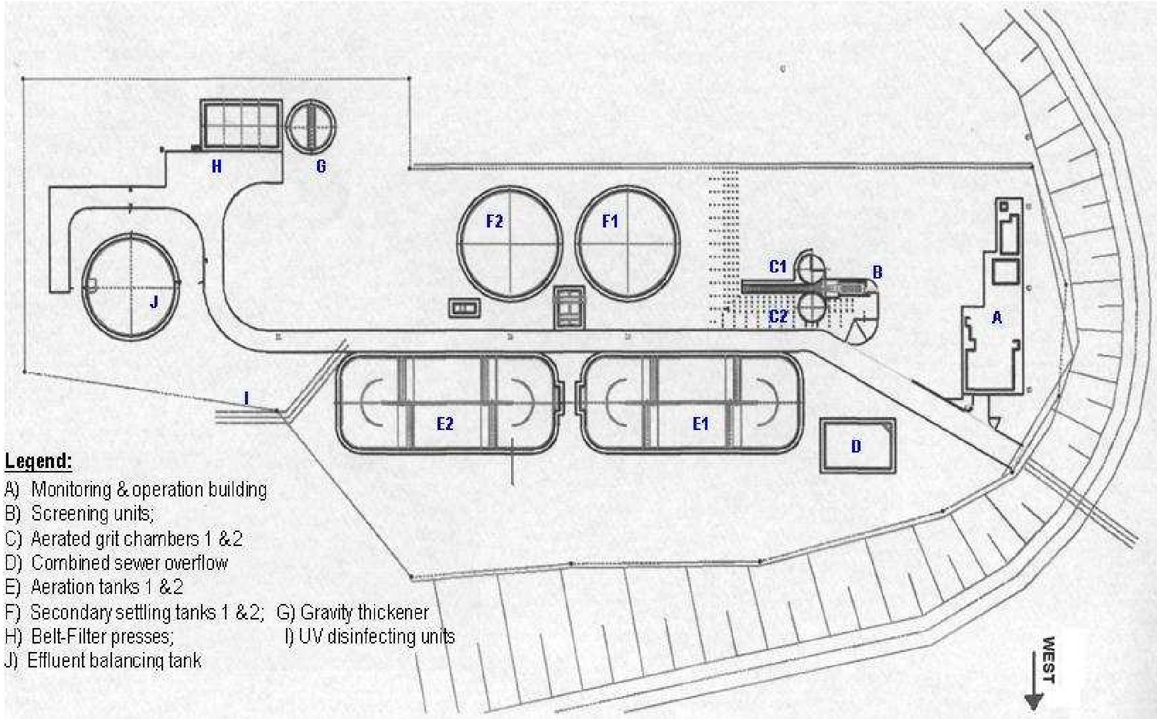

Figure 5. Sampling points for sludge bulking and foaming in liquid lines (Al-Bireh municipality, 2001)

3.3. Analytical methods

3.3.1 Physical tests

Temperature and pH tests

The temperature and pH value tests were measured onsite for influent, aeration tanks 1&2; and Sedimentation tanks 1&2 by HANNA (HI 991001 pH/Temperature Meter) Instrument (Figure. 6).

Figure 6.The measurements of T and pH value onsite in aeration tanks

Dissolve oxygen test (DO)

The Dissolve oxygen was determined onsite for influent, aeration tanks 1&2; and sedimentation tanks 1&2 by HANNA (HI 9142) Dissolved Oxygen METERS (Figure. 7). All measurements of dissolved oxygen were on surface but in aeration tanks 1&2, the measurements were taken from the surface to a depth of 3.5 meters.

Figure. 7. The measurements of (DO) onsite to depth of 3.5m in aeration tanks

Turbidity

Turbidity is measured in NTU: Nephelometric Turbidity Units. The instrument used for measuring is called (2100P Turbidimeter), which measures the intensity of light scattered at 90 degrees as a beam of light passes through a water sample (Figure. 8).

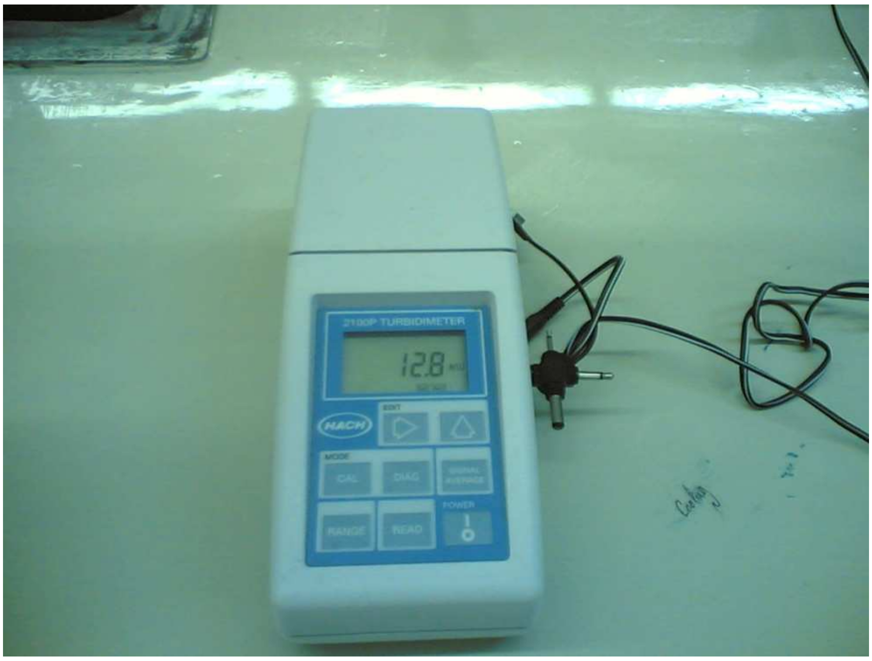

Figure. 8. Turbidity measurements in NTU unit

Sludge volume index (SVI)

Sludge's ability to settle is determined by measuring the sludge volume index (SVI), which is the measurement of volume of one gram TSS after 30 minutes settling in one liter graduated cylinder (Figure. 9). The SVI is given by the volume of settled sludge after 30 min in (ml/l) divided by mixed-liquor suspended solids MLSS (mg/l) (APHA, 1995).

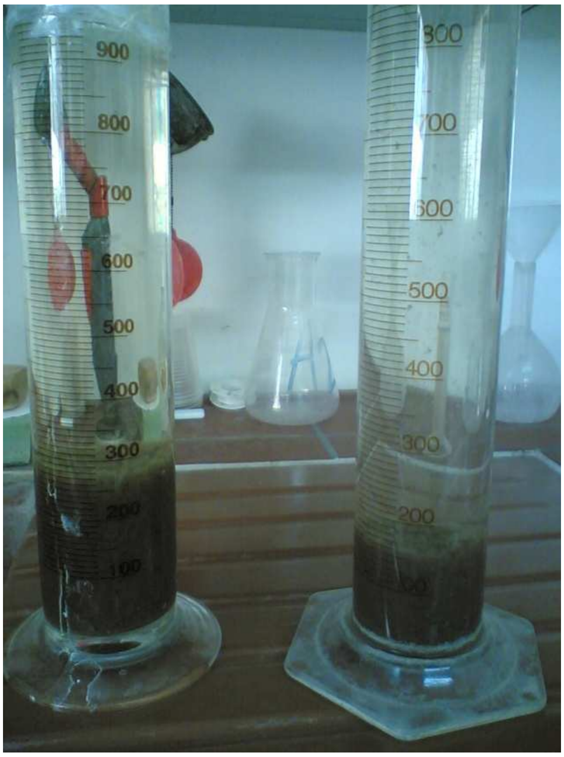

Figure. 9. Measurement of SVI in summer season

Diluted Sludge Volume Index (DSVI)

Acording to Lee et al., 1983; Bye and Dold 1998, samples were diluted to obtain SV_{30} smaller than 200 ml after 30 minutes, when the settling of sludge in one liter graduated cylinder is above 500 ml after 30 minutes.

Total Suspended Solids (TSS)

A well-mixed sample is filtered through a weighed standard glass-fiber filter and the residue retained on the filter is dried to constant weight at 105°C. The increase of the weight on the filter represents the total suspended solids.

3.3.2 Chemical tests

Chemical Oxygen Demand (COD)

Measurements were performed by using the closed reflux, colorimetric method. Then the absorbance was measured by spectrophotometer at 600 nm wavelength.

Biological Oxygen Demand (BOD_5)[20]

The BOD_5 test was determined by the use of the five-day BOD method by filling 300 ml diluted and seeded sample to overflowing; an airtight bottle of specified size and incubating it in the dark at 20 °C for five days. Dissolved oxygen was measured initially and after incubation by Dissolved Oxygen METER (INOLAND TERMINAL 740) (Figure.10).

Figure. 10. The measurements of BOD_5 by Dissolved Oxygen METER

Ammonia (NH_4-N)

Ammonia was determined by Nesslerization spectrophotometer; and then the absorbance was measured by UV-VIS spectrophotometer at 425 nm wavelength.

Nitrate (NO_3 -N)

The method of Nitrate analysis was determined by Standard Methods for the Examination of Water and Wastewater 4500-NO_3. The absorbance was measured by UV-VIS spectrophotometer at 420 nm wavelength.

Total Kjeldahl Nitrogen (TKN-N)

Total Kjeldahl Nitrogen was determined by using the macro-kjeldahl method for three steps: digestion, distillation and titration.

Phosphorous (Orthophosphate) $(PO_4)^{-3}$

Phosphorous was measured by the automated ascorbic acid reduction method by a filtered wastewater sample through 4.4 µm filters; and then measuring absorbance by UV-VIS spectrophotometer at 880 nm wavelengths.

Oil, Fat and Grease (FOG)

Oil, fat and grease were measured by the liquid-liquid, partition-gravimetric method using a separation funnel by two steps: extraction and condensation (APHA, 1995).

3.3.3 Biological tests

Total Coliform test

Total coliform densities was determined by the Membrane Filter (MF) procedure (Figure. 11) using M-endo broth medium with 1% (v/v) (ethanol 99% N), a sterilized membranes filter, absorbent pads and Vacuum Filtering Apparatus and then incubated for 22 to 24 hours at 35±0.5 °C (APHA, 1995).

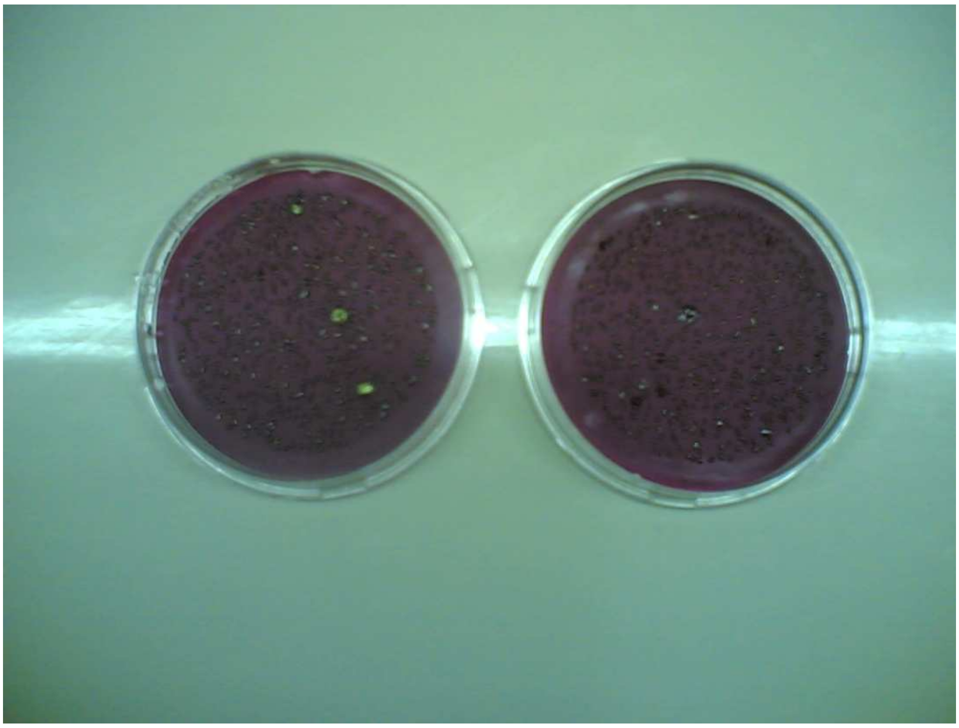

Figure. 11. Total coilform test using M-endo broth medium

Fecal Coliform test

Based on (APHA, 1995), Fecal coliform was determined by the Membrane Filter (MF) procedure using M-FC broth medium with 1% (v/v) Rosalic acid. The incubation period was 24±2 hours at 44.5±0.2 °C (Figure. 12).

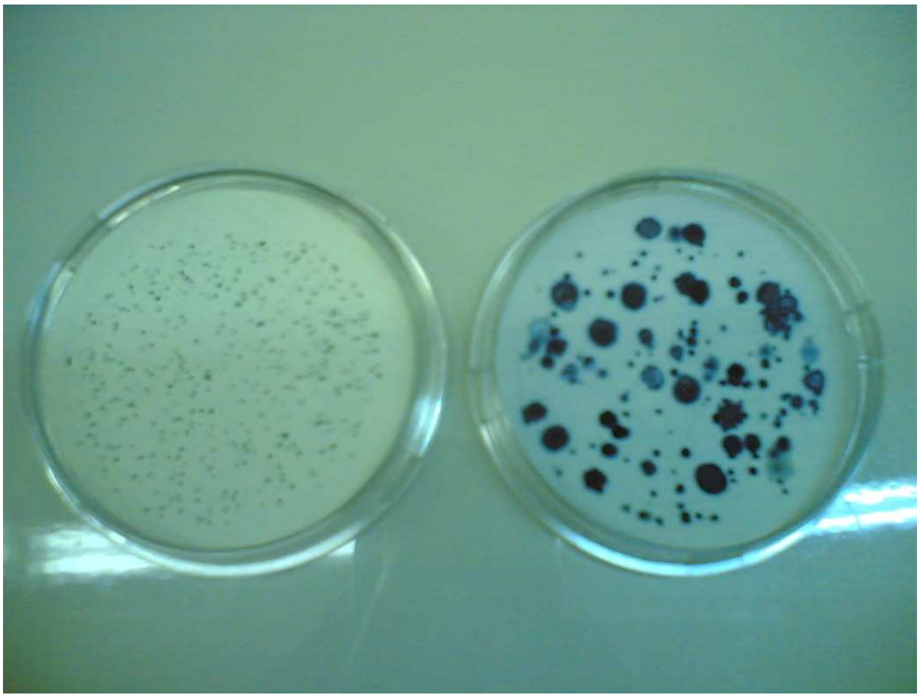

Figure 12. Fecal coliform test using M-FC broth medium

All methods of analysis were determined by Standard Methods for the Examination of Water and Wastewater (American Public Health Association 1995) shown in Table 3.1.

Table 3.1: Methods used for the determination of chemical, physical and biological parameters.

Parameters measurement	Instrumental used for analysis	Methods of analysis	Location of analysis	References
Temperature (T)	HANNA (HI 991001 pH/Temperature Meter).	2550A	AWWTP (onsite)	APHA 1995,21st ed
pH value	HANNA (HI 991001 pH/Temperature Meter).	4500-H^+ A	AWWTP (onsite)	APHA 1995,21st ed
Dissolve Oxygen (DO)	HANNA (HI 9142) Dissolved Oxygen METERS.	4500A	AWWTP (onsite)	APHA 1995,21st ed
Total Suspended Solids (TSS)	HOT AIR OVEN YCO-NO1	2540D	IEWS LAB	APHA 1995,21st ed
Sludge Volume Index (SVI)	One Liter Cylinder	2710D	AWWTP Lab	APHA 1995,21st ed
Diluted Sludge Volume Index (DSVI)	One Liter Cylinder	Sample dilution to obtain SV_{30} smaller than 200 ml after 30 min	AWWTP Lab	Lee et al., 1983; Bye and Dold, 1998.
Turbidity	2100P TURBIDIMETER	2130B	IEWS LAB	APHA 1995,21st ed
Chemical Oxygen Demand (COD)	COD REACTOR	5220D	IEWS LAB	APHA 1995,21st ed
Biochemical Oxygen Demand (BOD)	BOD_5 Bottles	5210B	IEWS LAB	APHA 1995,21st ed
Ammonia (NH_4-N)	UV-VIS spectrophotometer	4500C	IEWS LAB	APHA 1995,21st ed
Nitrate (NO_3 -N)	UV-VIS Spectrophotometer	4500-NO3	IEWS LAB	APHA 1995, 21st ed
Total Kjeldahl Nitrogen (TKN-N)	Gerhardt, kjeldatherm Gerhardt, Vapodest 30.	4500B	IEWS LAB	APHA 1995,21st ed
Orthophosphate $(PO_4)^{-3}$	UV-VIS spectrophotometer	4500E	IEWS LAB	APHA 1995, 21st ed
Oil, Fat and Grease (FOG)	BUCHI Waterbath B-480 BUCHI Rotavapor R-114	5520B	IEWS LAB	APHA 1995, 21st ed
Total Coliform (T.C)	Vacuum Filtering Apparatus	9222B	IEWS LAB	APHA 1995, 21st ed
Fecal Coliform (F.C)	Vacuum Filtering Apparatus	9222D	IEWS LAB	APHA 1995, 21st ed

3.4. Statistical analysis

In order to minimize the effect of systematic error, all physical, chemical and biological parameters measurements were replicated three times, and all data in this study represents the arithmetic mean of three repetition using software Excel 2010.

To find the relationship between SVI of the aeration tank 1&2 and other parameters, all the data were analyzed by statistical analysis (ANOVA and Regression) using software SPSS 17.

3.5. Microscopic examination and filamentous bacteria identification in activated sludge system

The identification of filamentous bacteria in the Al-Bireh activated sludge system was by morphology observation using Gram staining methods (Guo, 2010; Eikelboom, 2000; David, 2000). Gram staining is an indispensable aid when identifying bacteria. This staining first colours the bacteria blue using carbol gentian violet. The cells are then washed with an alcohol solution. The cells of some bacterial strains re-release the absorbed blue dye during this process. These bacteria are known as Gram negative. In the case of Gram positive bacteria, the absorbed carbol gentian violet cannot be removed by washing with alcohol. The colourless Gram negative bacteria are subsequently restained with safranine, which gives them a red colour. Gram staining was performed to the mixed liquor samples from aeration tanks (1&2) and from foam weekly in the autumn, winter, spring and summer. Microscopic examination was used to evaluate the abundance of filamentous bacteria present in the samples (Eikelboom, 2000). The pictures have been taken by a direct microscope under 100X magnification using immersion oil in Palestinian Water Authority (PWA) lab.

CHAPTER FOUR

Results and Discussion

4.1. Physical, chemical and biological parameters of Al-Bireh wastewater treatment plant

After data collection and analysis in the laboratory, the data was analyzed in Excel and SPSS software. This section will present the main reasons that caused sludge bulking and foaming in Al-Bireh wastewater treatment plant, and find the relationship between the increase of SVI value of aeration tanks (1&2) and the other parameters that may cause filament growth in activated sludge system. In this research, SVI in aeration tanks (1&2) started to increase from 6. Feb. 2010 to 15. May. 2010. After that, large amounts of sludge were pumped out of the plant to the valley (Figures 13 and 14). A very heavy rainy day was in 27. Feb. 2010 where it made dilution in the aeration tanks.

SPSS software is used to make ANOVA and regression analysis for this section in order to find (P-value) of all the parameters.

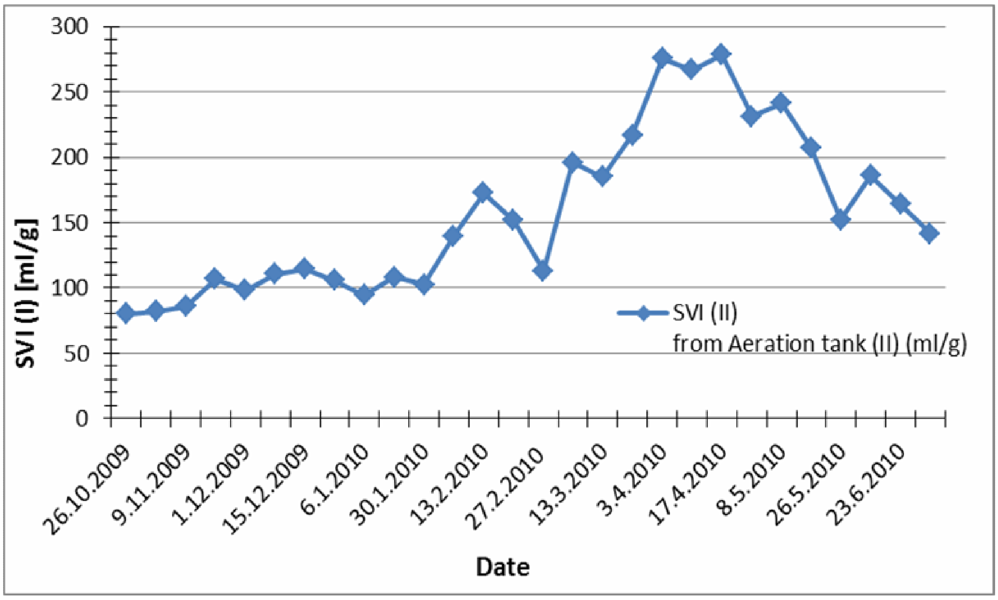

Figure 13. The relationship between sludge volume index (SVI 1) and Date in Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

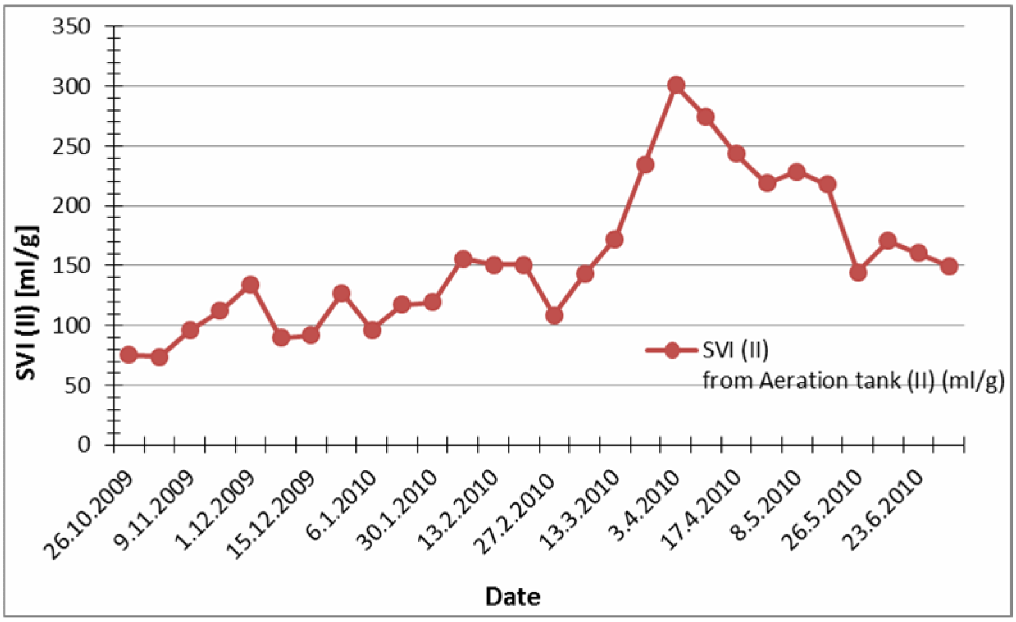

Figure 14. The relationship between sludge volume index (SVI 1) and Date in Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

4.1.1 Physical parameters

4.1.1.1 Temperature

Temperature is an important parameter that affects the sludge bulking and foaming phenomena. Sludge bulking and foaming are more likely to appear in low temperature conditions.

The results of this study showed that low temperature conditions can help induce sludge bulking and foaming especially at the beginning of winter to the end of spring and threre was an increase in SVI value. The increase started in February when the temperature was the lowest tepmereture of the year. In 27.Feb.2010 a critical decrease of temperature occured and it continued into the next week making it the coldest week of the year. After this week a critical increase of SVI occurred when the thickness of foam reached about 40 cm. This means the decrease of temperature in February was the critical point to increase the SVI in two aeration tanks and thus cuase sludge bulking and foaming in this research.

As shown in (Figures 15 and 16), the P-value is more than 0.05. This can be attributed to a temperature fluctuation every week in the study area. Therefore it can be concluded that another parameter can have a significant relationship with the SVI.

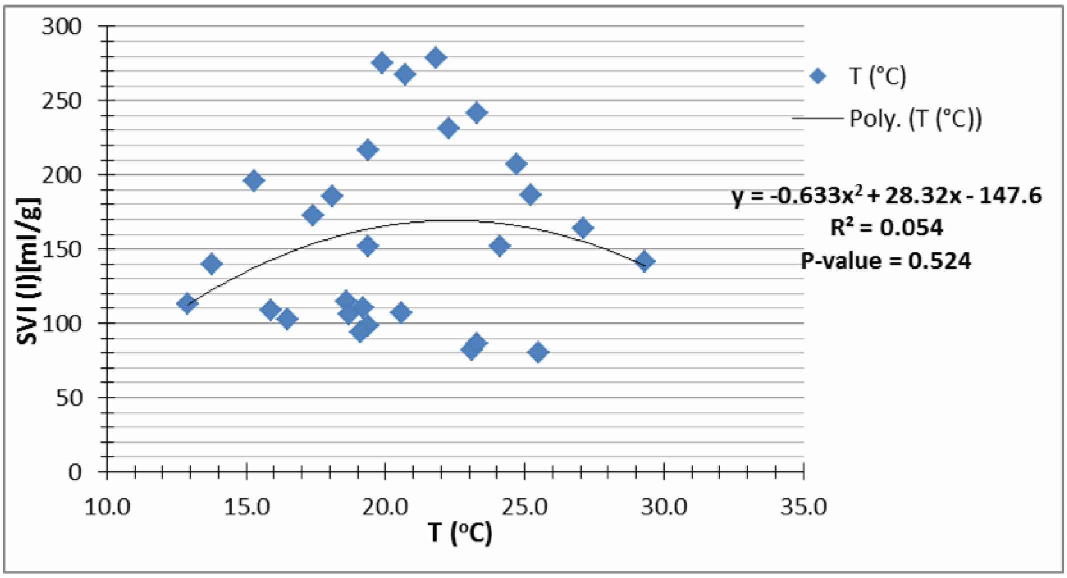

Figure 15. The relationship between sludge volume index (SVI 1) and temperature in sedimentation tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

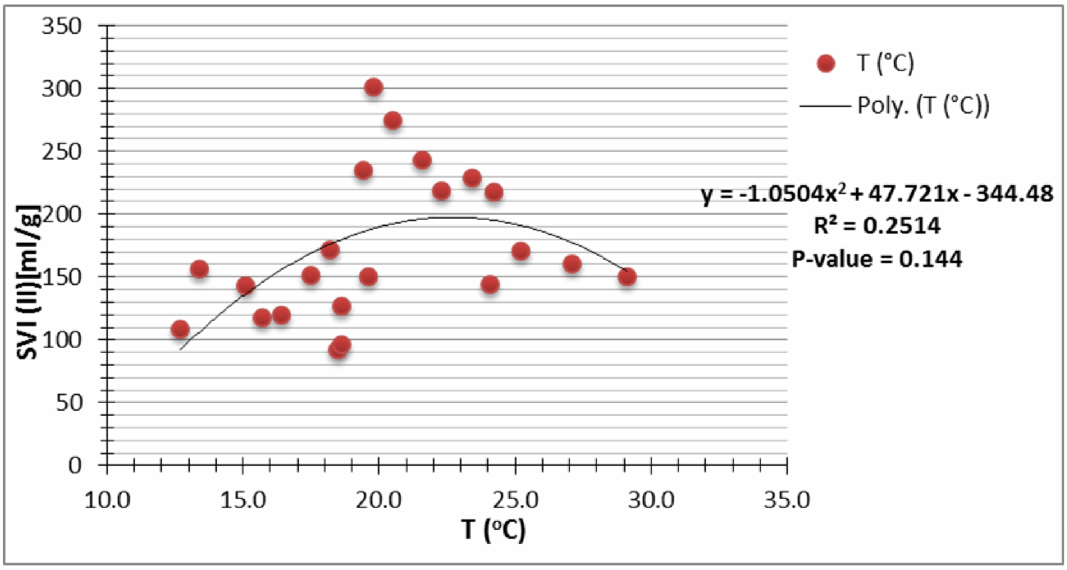

Figure 16. The relationship between sludge volume index (SVI 2) and temperature in sedimentation tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

4.1.1.2 pH value

The aeration basin pH should be maintained in the range of 6.5 to 8.5 (Richard, 2003). Low pH, <6.5, may cause the growth of fungi and fungal bulking. In this study as shown in (Figures 17 and 18) there is no important relationship between the increase of SVI (I and II) and pH value. The P-value is more than 0.05 in aeration tanks (1&2) and it close to one, especially in aeration tank (2). As a result, there is no effect of the pH value on the appearance of sludge bulking and foaming of two aeration tanks (1&2) in Al-Bireh WWTP.

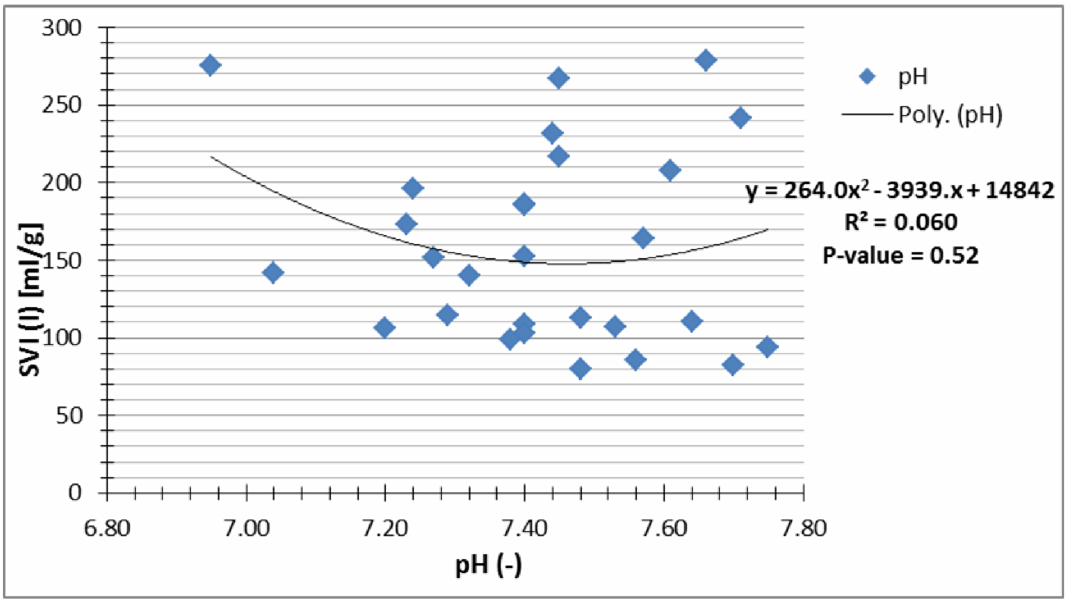

Figure 17. The relationship between sludge volume index (SVI 1) and pH in aeration tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

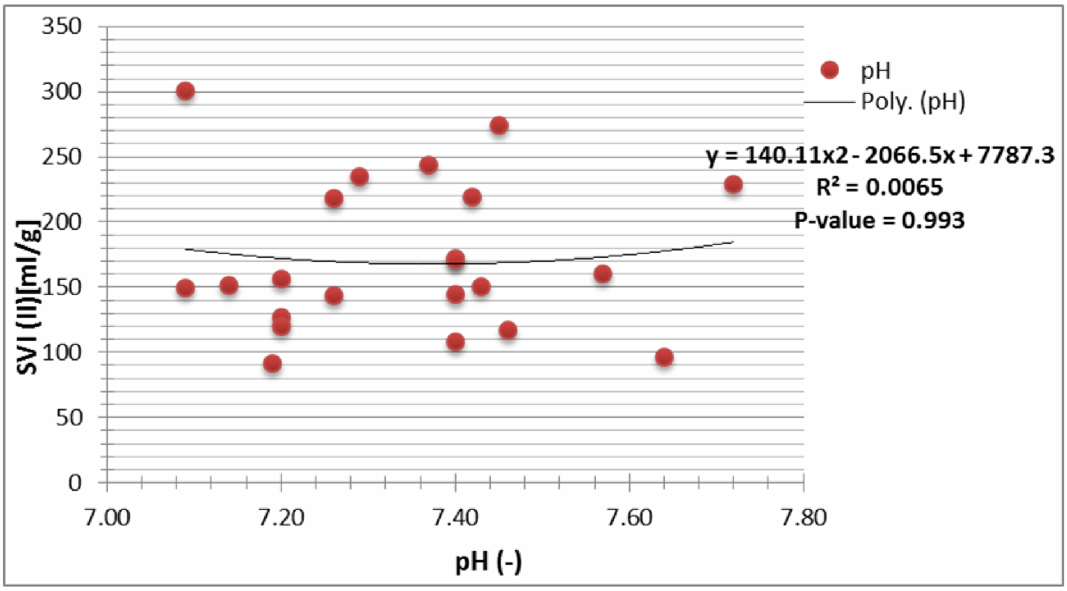

Figure 18. The relationship between sludge volume index (SVI 2) and pH in aeration tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

4.1.1.3 Dissolved oxygen

Dissolved oxygen is an important parameter in ASS to present the efficiency and sustainability of the treatment plant. Low aeration tank dissolved oxygen (DO) concentration leads to the increase of SVI and causes filamentous bulking. In this study there is a strong significant relationship between the decrease of DO and the increase of SVI (I) (Figure. 19). The P-value is much lower than 0.05 where P-value = 4.3^{-5} and R^2 = 0.571. The decrease of the DO in sedimentation tank (1) is the main factor that cause increases in SVI (I) in aeration tank (1), and it has the main effect on the appearance of sludge bulking and foaming phenomena.

In (Figure. 20), there is no significant relationship between the decrease of DO and the increase of SVI (II) and the P-value is more than 0.05.

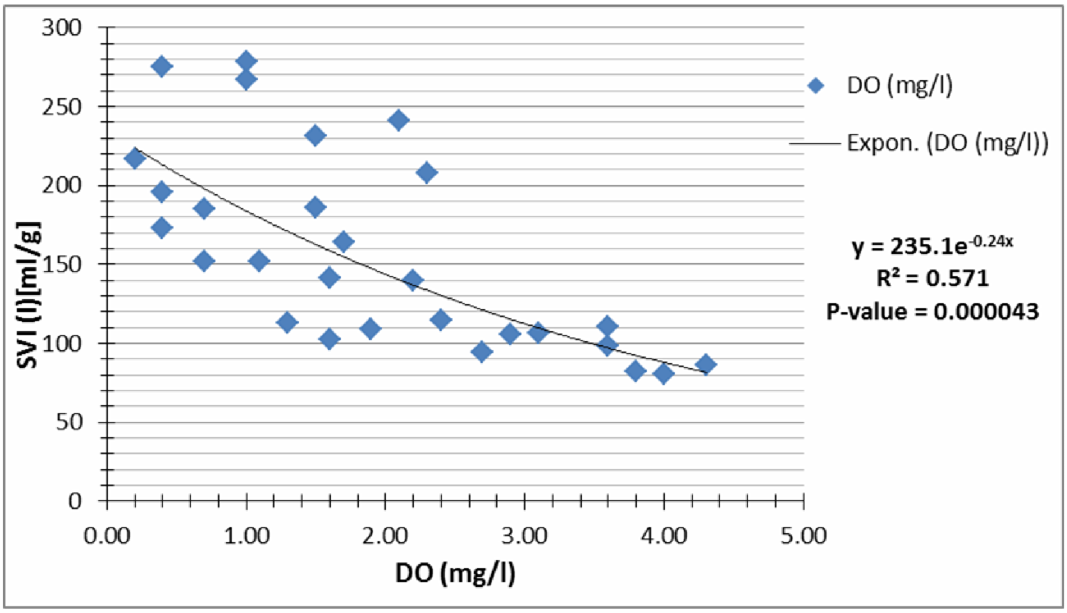

Figure 19. The relationship between sludge volume index (SVI 1) and Dissolved Oxygen (DO) in sedimentation tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

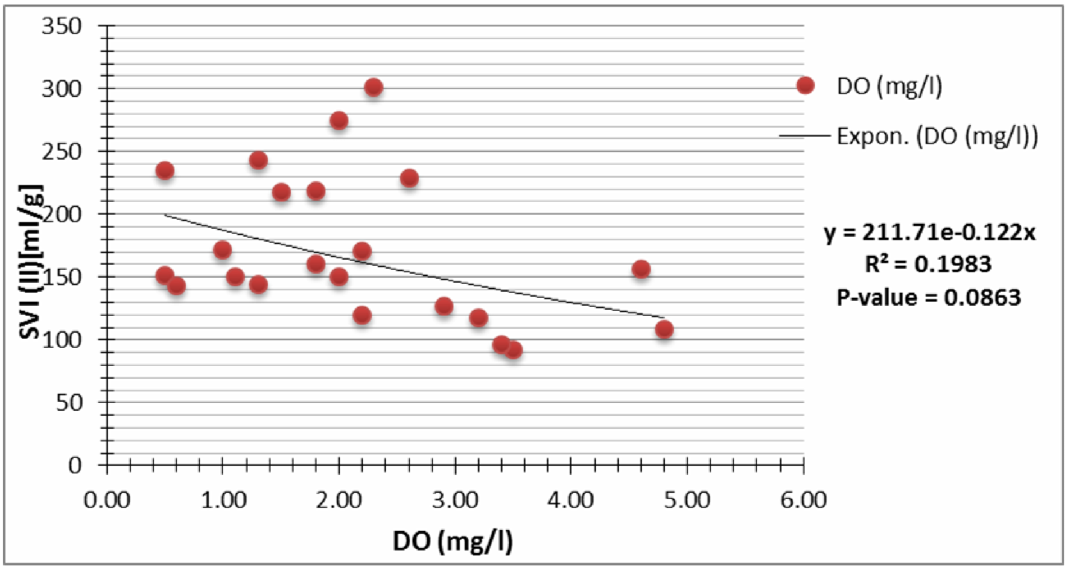

Figure 20. The relationship between sludge volume index (SVI 2) and Dissolved Oxygen (DO) in sedimentation tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

The decrease of DO concentration in the aeration tanks leads to the increase the opportunity of filamentous bacteria to overgrowth and multiply. There are special types of filamentous bacteria that multiply when the limitation of oxygen occurs.

The foam layers were dominating in March and April at a depth of 30 to 40 cm. This phenomenon resulted from some types of filamentous bacteria that survive when DO is limited. The foam layers made a cover on the surface of the treatment plant and prevented oxygen from penetrating below the thick layer which also caused oxygen deficiency below the surface.

In this research the DO concentration was measured weekly from the surface to a depth of 3.5 meters of both aeration tanks (1&2). When the DO was measured at increasing depths, there was a significant decrease in the amount of DO. It decreased steadily every meter, however on a few occasions it has been recorded as zero from 2.5 to 3.5 m, and even zero throughout the entire column. The decrease of the DO concentration in both aeration tanks is the main reason that leads to the increase of SVI and cause of sludge bulking and foaming in AWWTP. The aeration tank (1) is more affected with the DO deficiency than aeration tank (2) because the amount of the sludge in aeration tank (2) is lower than that found in aeration tank (1).

Almost always DO concentrations were below 2 mg/l especially in aeration tank (1) and during the occurrence of sludge bulking and foaming phenomena.

4.1.2 Chemical parameters

4.1.2.1 Organic nitrogen and Orthophosphate $(PO_4)^{-3}$

Nitrogen and phosphorus can be growth-limiting elements if they are not present in sufficient amounts in the influent wastewater. In this study, there was an increasing amount of the organic nitrogen that entered to the plant into two aeration tanks during the increase of SVI (I and II) (Figures 21 and 22). The relationship between the increase of organic nitrogen and the increase of SVI is very strong. The P-value has a much lower value than 0.05 where it equals 4.19×10^{-5} and 0.0012 during the relationship with SVI (I) and SVI (II) respectively.

Also there is an increase in the amount of orthophosphate from the influent during the increase of SVI especially with (SVI 1) (Figures 23 and 24). There was a weak relationship between the increase of sludge volume index (SVI 1) and the increase of orthophosphate from the influent Figure (23), where the P-value equals 0.0463.

In this research, there was no effect of the nutrient deficiency on the appearance of sludge bulking and foaming in the two aeration tanks (1&2) in Al-Bireh WWTP.

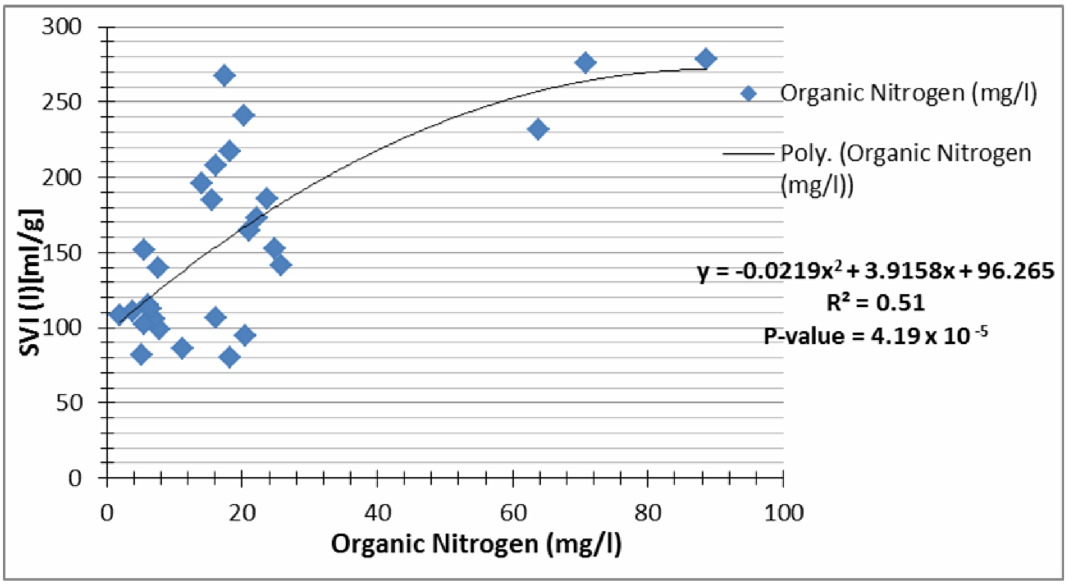

Figure 21. The relationship between sludge volume index (SVI 1) and organic nitrogen from the influent of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

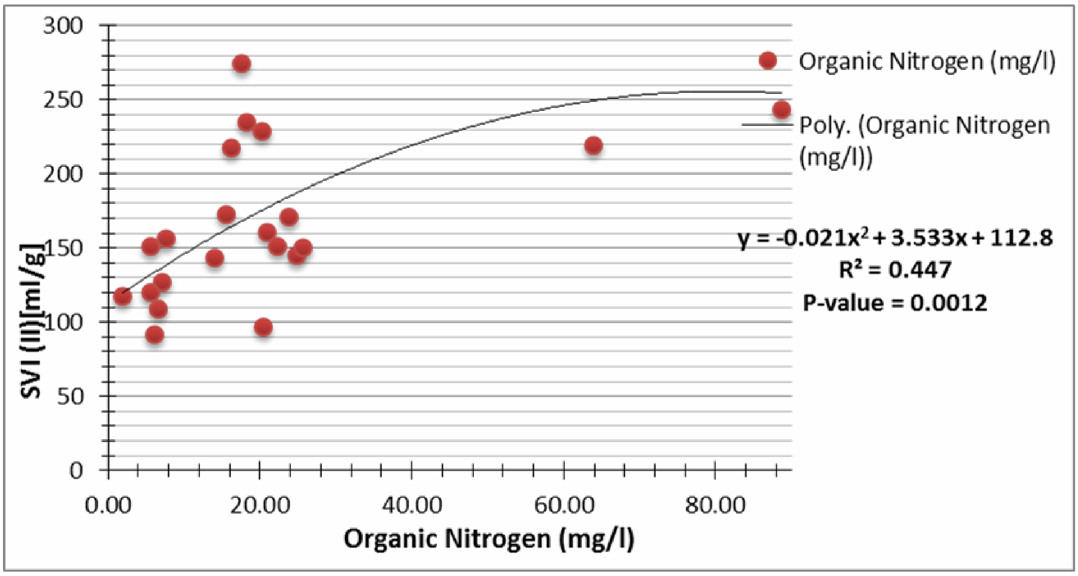

Figure 22. The relationship between sludge volume index (SVI 2) and organic nitrogen from the influent of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

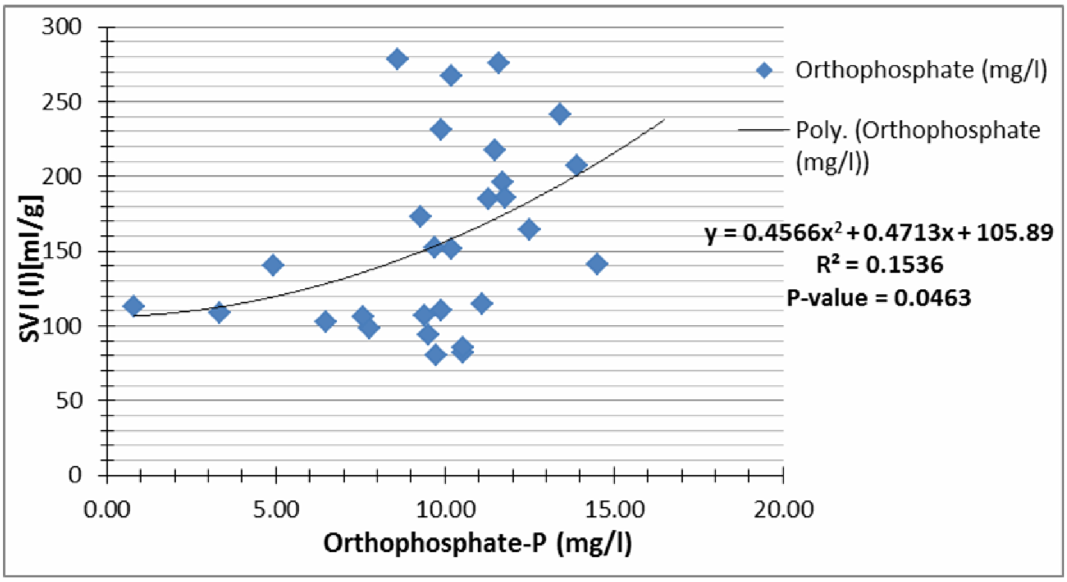

Figure 23. The relationship between sludge volume index (SVI 1) and orthophosphate from the influent of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

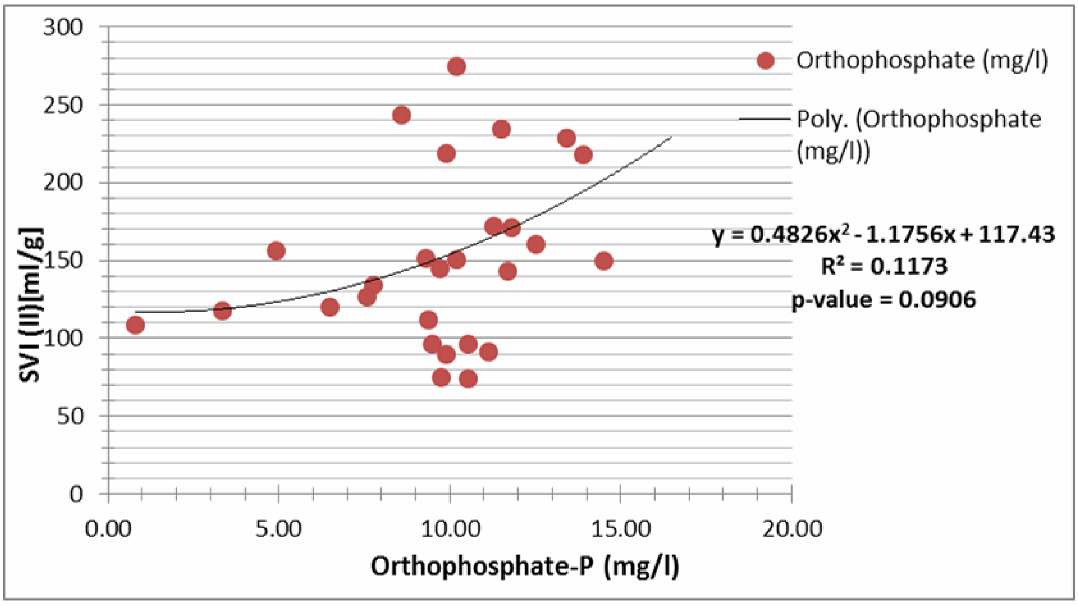

Figure 24. The relationship between sludge volume index (SVI 2) and orthophosphate from the influent of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

4.1.2.2 Fat oil, and grease (FOG)

Fat, oil and grease (FOG) that pass to the treatment plant are effective to cause foaming in activated sludge system and help the growth of the filamentous organisms. As shown in (Figures 25 and 26) below there is a significant relationship between the increase of FOG level and the increase of (SVI 1). This trend was also clear after making regression analysis when P-value is equal to 0.021 and it is less than 0.05. There was no effect of the (FOG) on the (SVI 2) and the P-value is equal to 0.277 (Figure 26).

This means FOG was the second parameter responsible for the increase of SVI in aeration tanks (1). The increase of FOG caused the increased amount of foam in aeration tank (1).

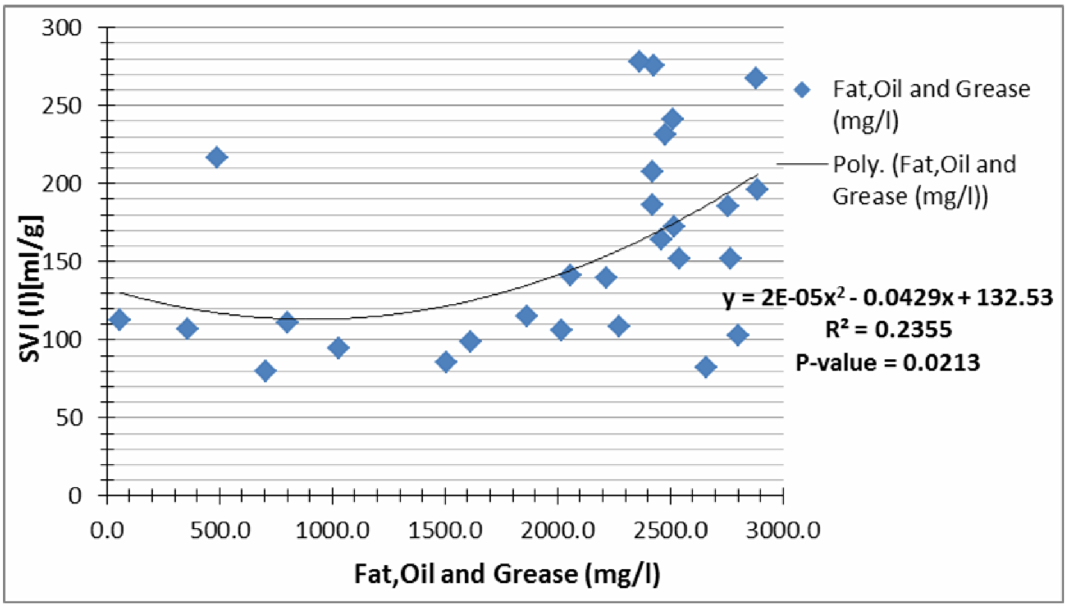

Figure 25. The relationship between sludge volume index (SVI 1) and (FOG) from the influent of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

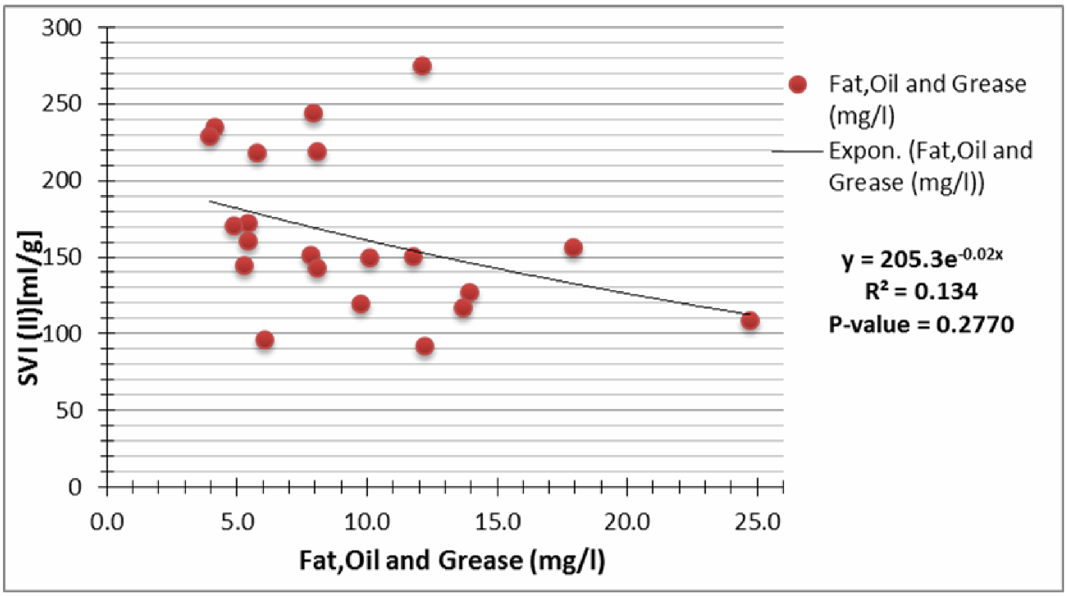

Figure 26. The relationship between sludge volume index (SVI 2) and (FOG) from the influent of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010

4.1.2.3 Sludge age and F/M ratio

As shown in (Figures 27 and 28), there is no important relationship between the increases of SVI value and sludge age. The P-value is more than 0.05 in both sedimentation tanks (1&2). Some filamentous microorganisms were found over a wide range of sludge age values while others occured only at low or high values.

According to Husain (2004), the high and low F/M ratio causes sludge bulking and foaming phenomena and leads to the overgrowth of special types of filamentous bacteria. In this study there was a weak significant relationship between the increase of F/M ratio and the increase of (SVI 1) where P-value is equal to 0.05 (Figure 29) and there is no relationship between F/M ratio and (SVI 2) (Figure 30).

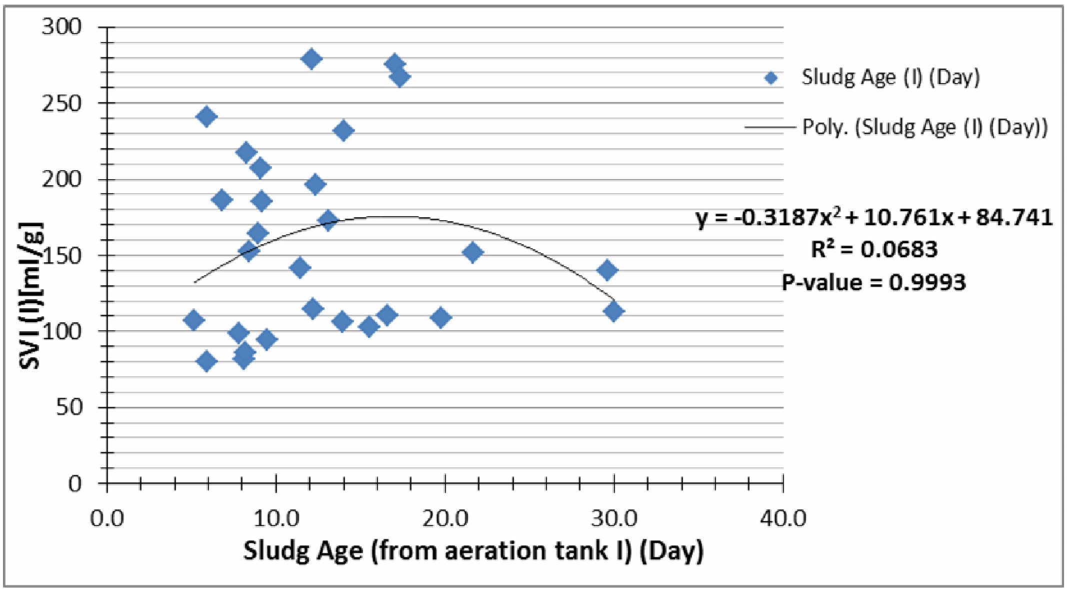

Figure 27. The relationship between sludge volume index (SVI 1) and sludge age in aeration tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010

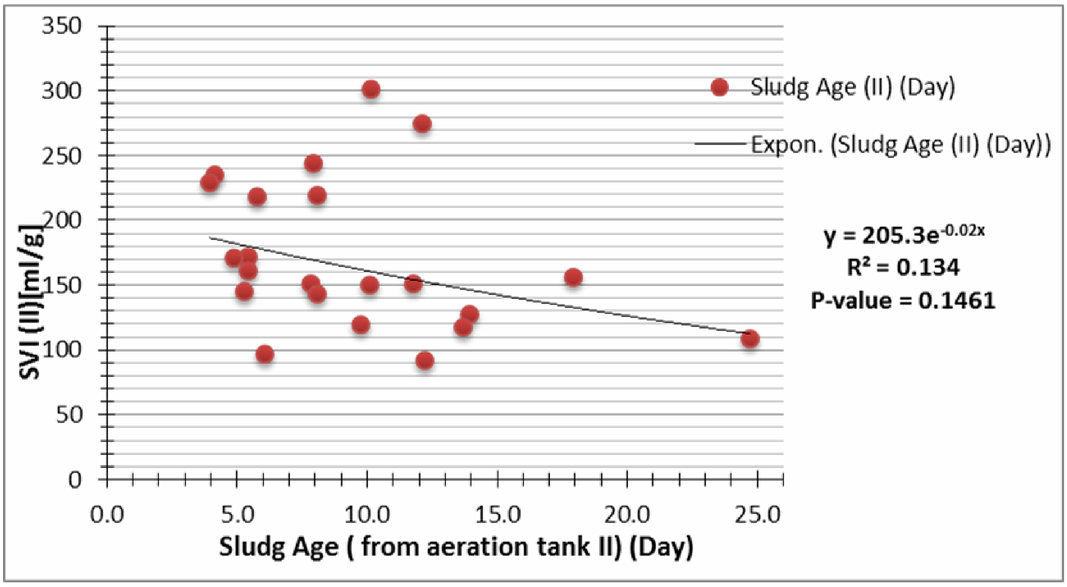

Figure 28. The relationship between sludge volume index (SVI 2) and sludge age in aeration tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010

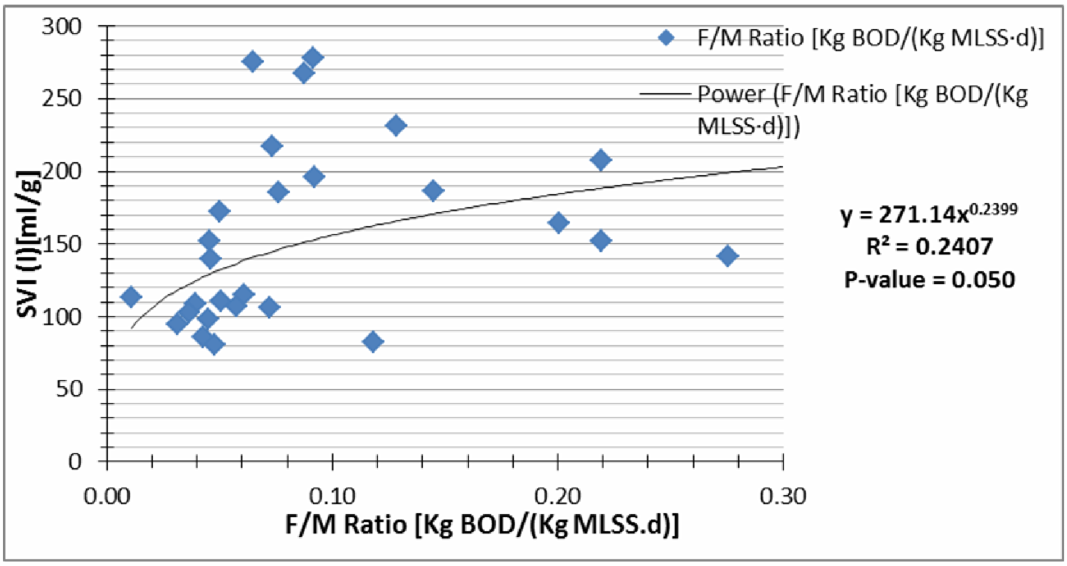

Figure 29. The relationship between sludge volume index (SVI 1) and F/M ratio in aeration tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010

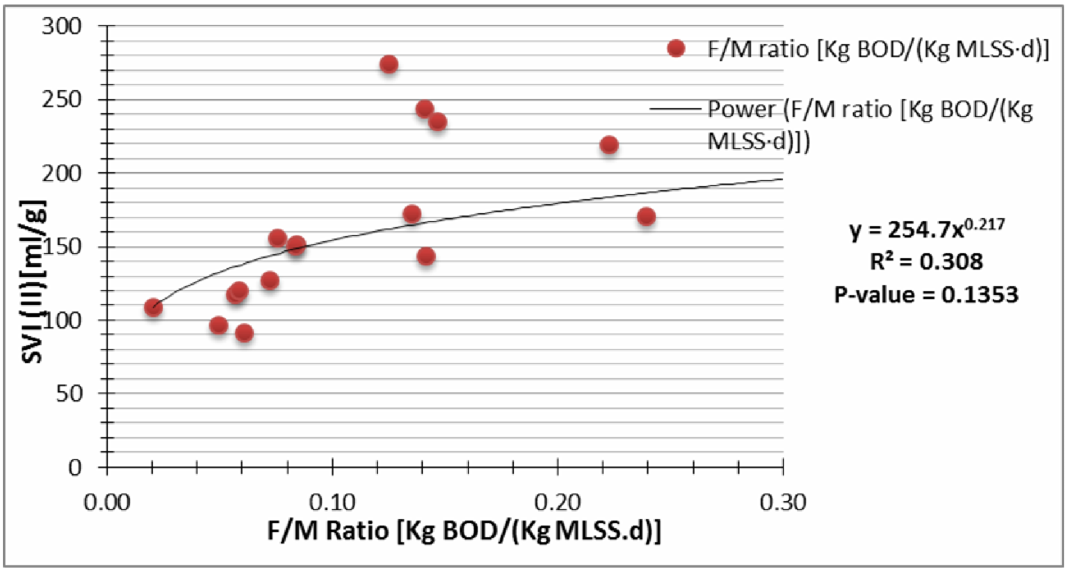

Figure 30. The relationship between sludge volume index (SVI 2) and F/M ratio in aeration tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010

4.2. Identification of filamentous microorganisms in aeration tanks of AWWTP.

The excess growth of filamentous microorganisms causes bulking that deteriorates the ability of sludge to settle, resulting in decreased sludge settling rate and a non-compact sludge blanket, usually indicated by an increased SVI. The foaming and bulking occurred seasonally and periodically, which caused serious operating problems and took a relatively long time to restore.

The cause of foaming and bulking in aeration tanks at Al-Bireh wastewater treatment plant (AWWTP) were investigated. The results showed that the foaming and bulking was mainly caused by the excessive propagation of *Microthrix parvicella, Nocardia, Type 1701, Sphaerotilus natans* and other filaments.

Figure 31. Gram-positive *Micothrix parvicella* from foam and mixed liquor of AWWTP, X100 magnification. April, 2010.

Microthrix parvicella (Figure. 31) has a Medium length, non-motile filaments with irregularly coiled filaments, no branching and no cell septa. Filaments are found in spaghetti-like tangles within the floc structure causing open, lacy, diffuse floc or in loose patches free in the bulk solution. The filament staining is strongly Gram positive when in the floc short clear spaces may occur in the filament. It doesn't have sulfur granules, attached growth or sheath present. It is a common cause of foaming phenomenon. This filament is usually found in environments where there is a low F/M ratio and with a long sludge age but with high grease. In this study it was found more commonly in the colder climates. The abundance of *M. parvicella,* was more frequent in winter and spring than in summer and autumn. High wastewater grease and fat content are usually the main reasons that cause the appearance of this filament.

Nocardia has relatively short, non-motile filaments, irregularly bent filaments with true branching and cell septa without constrictions (Figure. 32). Branched mycelium is often observed on this filament. Filaments are found within the floc structure causing open, lacy, diffuse floc or free in the bulk solution. The filament staining is strongly Gram positive and easy to identify due to its staining characteristics and branching.

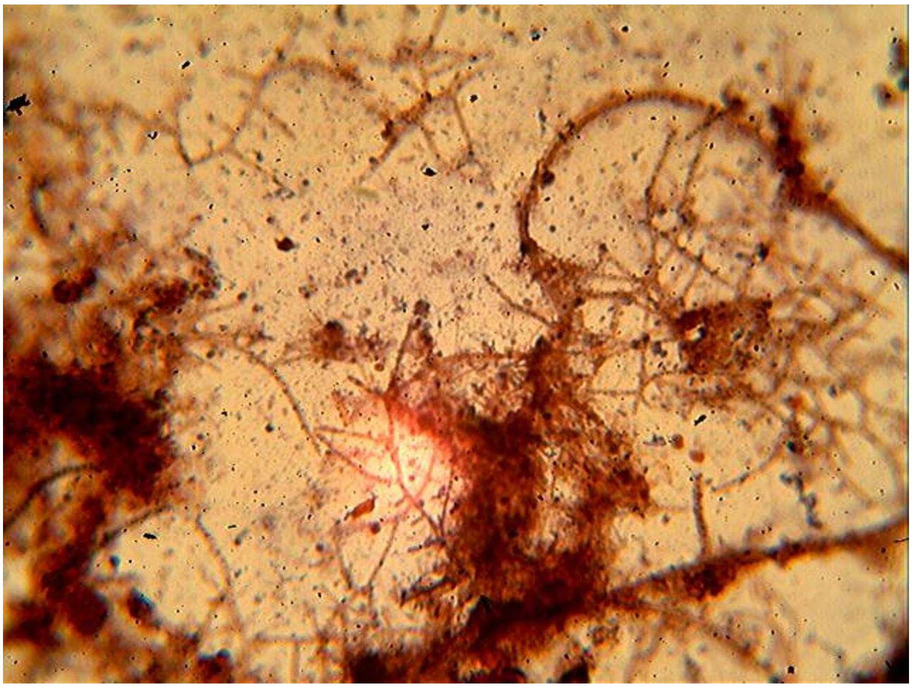

Figure 32. *Nocardia* from foam of AWWTP, X100 magnification. March, 2010.

There are no sulfur granules, attached growth or sheath present. It is the most common cause of foaming, not bulking and causes floating sludge. This filament is usually found in environments where there is a low F/M ratio, high sludge age and with high grease. In this study it is clearly present when (FOG) increased and it couples with the increase of SVI value. It found more commonly in the warmer temperatures in March and April.

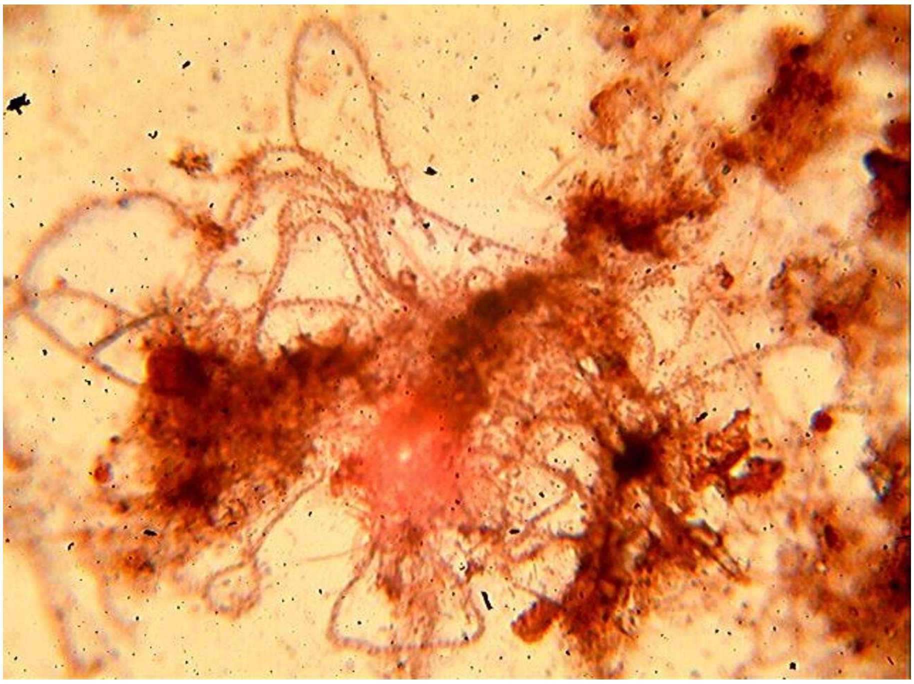

Figure 33. Gram-Negative *Type 1701* from mixed liquor samples of the aeration tank (2) in AWWTP, X100 magnification. April, 2010.

Type 1701 (Figure. 33) is non-motile filaments and curved with no branching. The cells are round-ended and rods shaped “sausages” and are contained in a clear, tightly fitting sheath. The cell septa are clear and easily observable with indentations at the septa. Filaments are usually found intertwined in the floc structures with only short filaments extending into the bulk solution and can cause sludge settling interference by open and diffuse floc. The filament is usually Gram negative and there are no sulfur granules. This filament is usually found in environments where there is low DO but reasonable F/M ratios. These conditions are the same conditions that found in the aeration tank (1) in this research. Also it can be caused by too long RAS lines or sludge being held too long in the secondary clarifier.

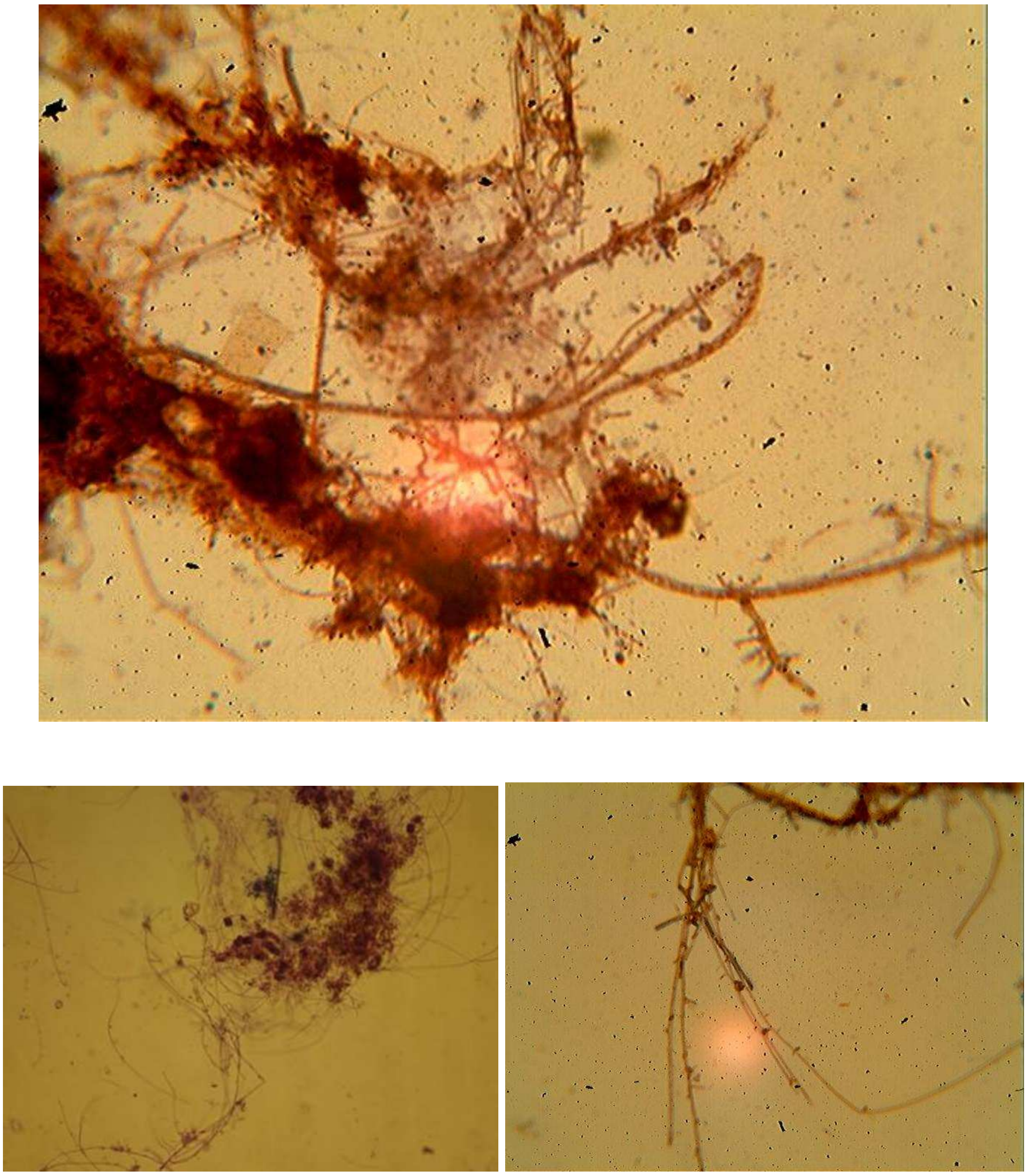

Figure 34. ***Sphaerotilus natans*** **from mixed liquor samples, X100 magnifications. March and April, 2010.**

Sphaerotilus natans (*S. natans*) is usually found in environments where there is low DO coupled with a high F/M ratio (Hossain, 2004). In this study, these environments were found clearly in aeration tank (1). *S. natans* (Figure. 34) is relatively long, with non-motile filaments, and is straight or smoothly curved with tree-like false branching. The cells are rod shaped and are contained in a clear, tightly fitting sheath. The cell septa are clear and easily observable with indentations. Filaments radiate outward from the floc surface into the bulk solution and can cause sludge settling interference by inter-floc bridging. The filament is usually Gram negative without sulfur granules. Poly-ß-hydroxybutric acid (PHB) is frequently observed as dark intracellular granules.

Sphaerotilus natans and *Type 1701* are lookalike sausage-shaped cells. Characteristics that are inherent to *S. natanas* are that occasionally the cells branch out from the main filament and it exhibits false branching which can be utilized in the identification of *S. natans* under the microscope. The cells are rod-shaped like *Type 1701* but are relatively bigger and better defined. *Type 1701* also often has attached growth. The cause for both is the same-low dissolved oxygen concentration. Both filaments were highly dominant in March and April when the increase of SVI occurred and the measurement of DO was close to zero even at 3.5m depth. Also these filaments were highly dominant when the foam layers covered most of the area of both aeration tanks at 30-40 cm depth.

The main reason of DO deficiency in both aeration tanks of AWWTP may be a result from the turning off of the Mammoth rotors sometimes for a long time. The turn off of some Mammoth rotors is the main cause of the foam formation that prevents the DO from penetrating to the wastewater surface and therefore the measurements of DO in the surface were close to zero.

4.3. Process performance and sustainability of the ASS process during the appearance of sludge bulking and foaming phenomena.

The study area in this research is considered a semi-arid area. It is located under the occupation. There is water conflict in this region; therefore, the resources of fresh water are very limited and rare. At the same time there is a large amount of treated water; near 5,000 m^3/day are discharged to the valley without reuse (Figure 35). Large amounts of fresh water are consumed in the agriculture sector. As a result, it will increase the stress and the demand on the freshwater resources. The Palestinian society has not accepted the reuse of treated wastewater for many reasons.

Figure 35. Treated effluent discharged to the Wadi Al-Ain nearby AWWTP

This study tried to check the performance of ASS in the four seasons especially during the appearance of sludge bulking and foaming to show the effect of these phenomena on the quality of effluent. The standards of water re-use in Palestine are used to make a comparison with these study results Table 4.1. The comparison is concentrated on the ability to reuse the treated effluent during the appearance of sludge bulking and foaming that started from 6.Feb.2010 to 15.May.2010.

Table 4.1. Israeli and Palestinian standards for effluent disposal in various applications (Samhan *et al.*, 2010).

Parameter	Unit	Israeli Standards 2002		Palestinian standards 2002	
		Unrestricted irrigation	Rivers	Unrestricted irrigation	Rivers
BOD	mg/l	10	10	20	–
TSS	mg/l	10	10	30	
COD	mg/l	100	70		200
Ammonia-N	mg/l	20	1.5	50	5
Total-N	mg/l	20	10		
Total-P/PO_4-P	mg/l	5	0.2	30	5
SO_4	mg/l	–	–	500	1,000
Chloride	mg/l	250	400	500	–
Sodium	mg/l	150	200	200	–
Fecal coliforms	CFU/100 ml	10	200	<200	<1,000
Boron	mg/l	0.4	–	0.7	2
Hydrocarbons	mg/l	–	1	0.002	1
Anionic detergents	mg/l	2	0.5	15	25
Total oil	mg/l	–	1	5	10
pH	[–]	6.5–8.5	7–8.5	6–9	6–9
Dissolved oxygen	mg/l	<0.5	<3	>0.5	>1

Some physical, chemical and biological parameters were measured from the effluent of two sedimentation tanks such as pH, DO, NH_4, orthophosphate, TKN, NO_3, COD, BOD, TSS, Turbidity, TC and FC.

The pH and DO are explained earlier in section 4.1. The pH value during the increase of SVI was within the range of Palestinian standard found in Table 4.1 however, DO was below this range especially that from the sedimentation tank (1). During sludge bulking and foaming phenomena there was no effect between SVI and the amount of NH_4 or orthophosphate from sedimentation tanks (1&2) Figures (36, 37, 38 and 39). The amount of NH_4 and orthophosphate from both sedimentation tanks (1&2) were within the range of the Palestinian standards.

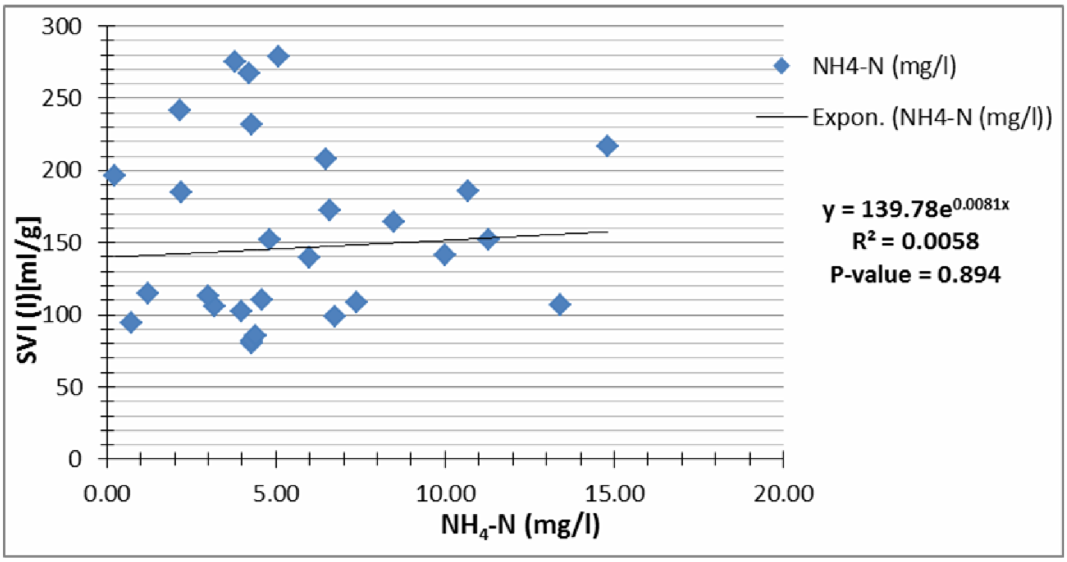

Figure 36. The relationship between sludge volume index (SVI 1) and NH_4-N in sedimentation tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

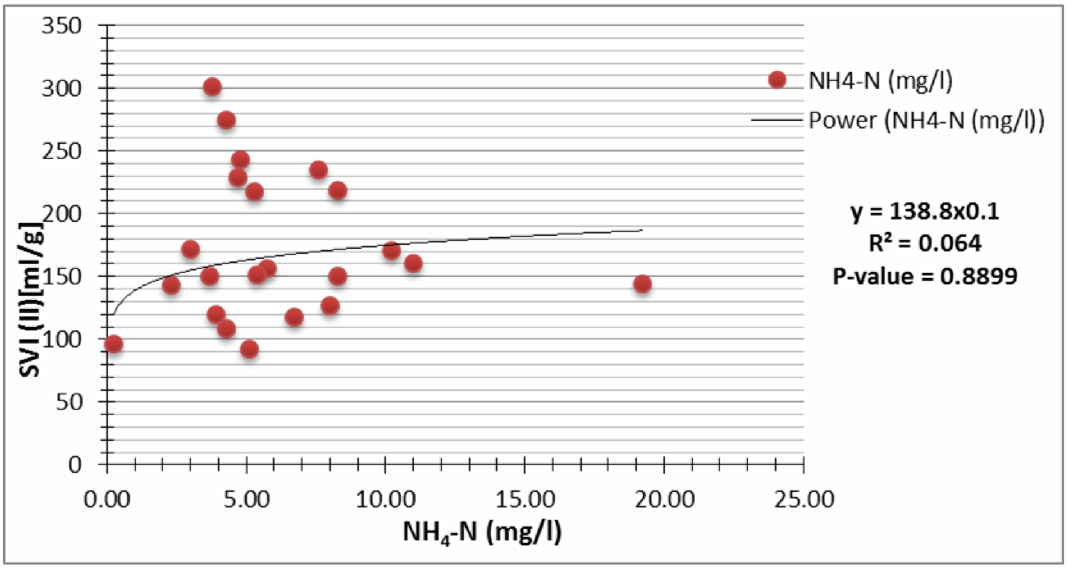

Figure 37. The relationship between sludge volume index (SVI 2) and NH_4-N in sedimentation tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

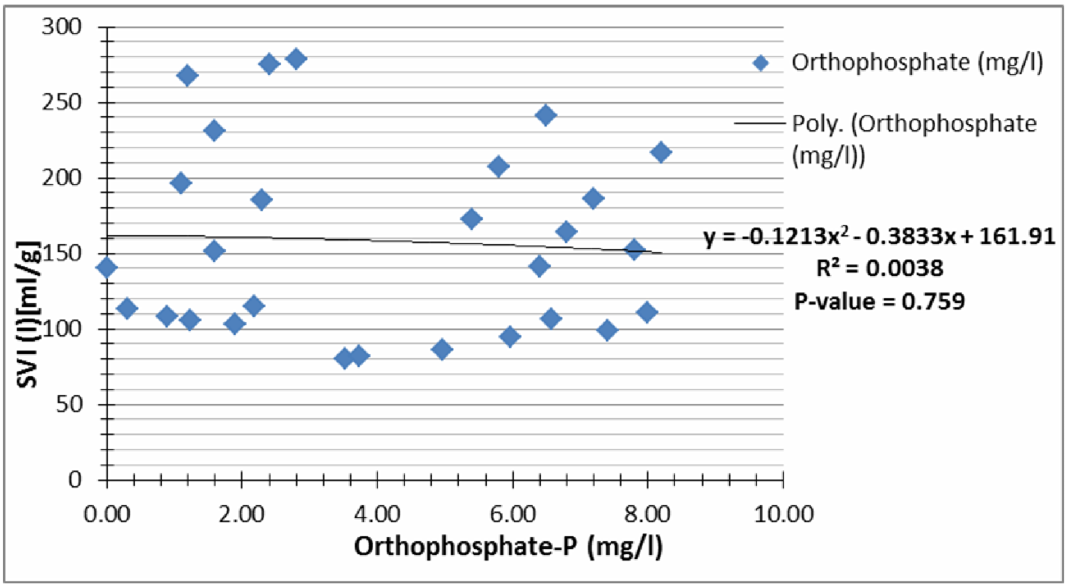

Figure 38. The relationship between sludge volume index (SVI 1) and orthophosphate in sedimentation tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

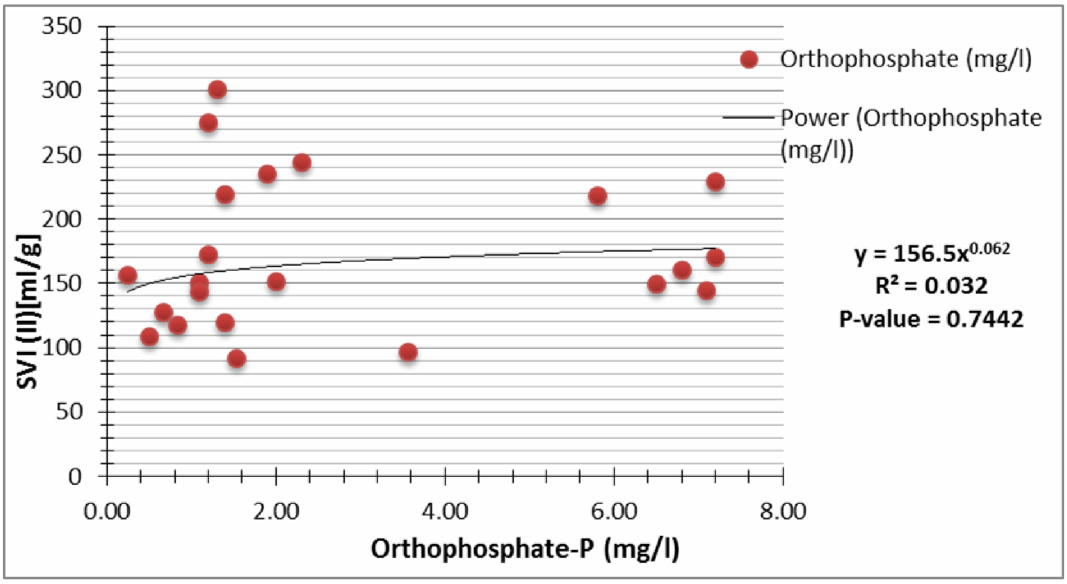

Figure 39. The relationship between sludge volume index (SVI 2) and orthophosphate in sedimentation tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

There is no effect between SVI and the amount of NO_3 from sedimentation tanks (1&2) (Figures 40 and 41) and there is no Palestinian standards for the amount of NO_3 Table 4.1.

In Figures (42 and 43) there is a stronger significant relationship between the increase of SVI and the amounts of TKN from sedimentation tanks (1&2) especially of the sedimentation tank (1). The P-value is much lower than 0.05 for both tanks. The amounts of the TKN from both sedimentation tanks (1&2) were higher than the suitable range of the water re-use standards during the appearance of sludge bulking and foaming phenomena.

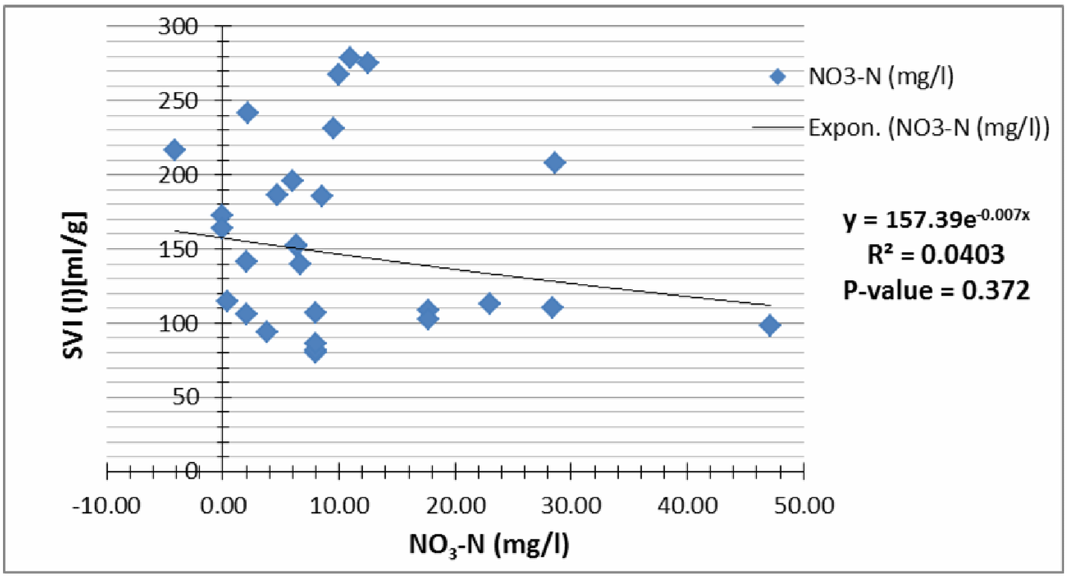

Figure 40. The relationship between sludge volume index (SVI 1) and NO_3-N in sedimentation tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

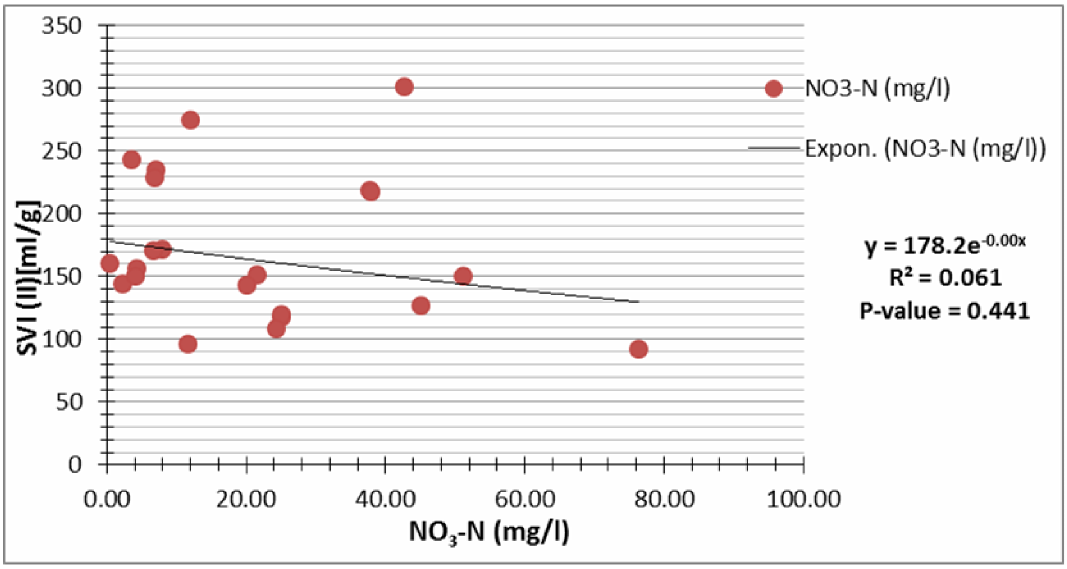

Figure 41. The relationship between sludge volume index (SVI 2) and NO_3-N in sedimentation tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

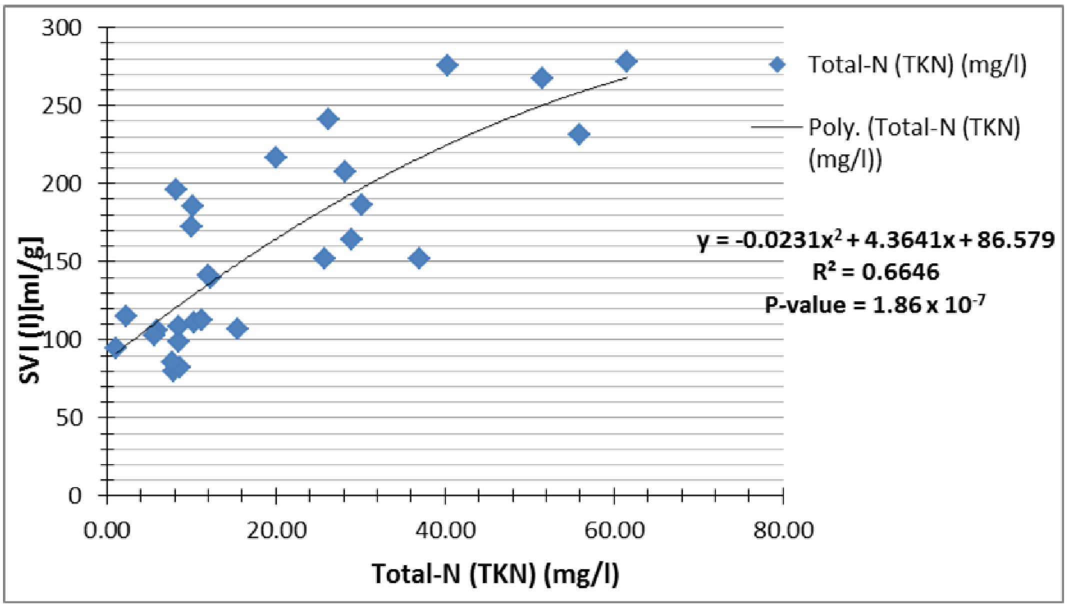

Figure 42. The relationship between sludge volume index (SVI 1) and total-N in sedimentation tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

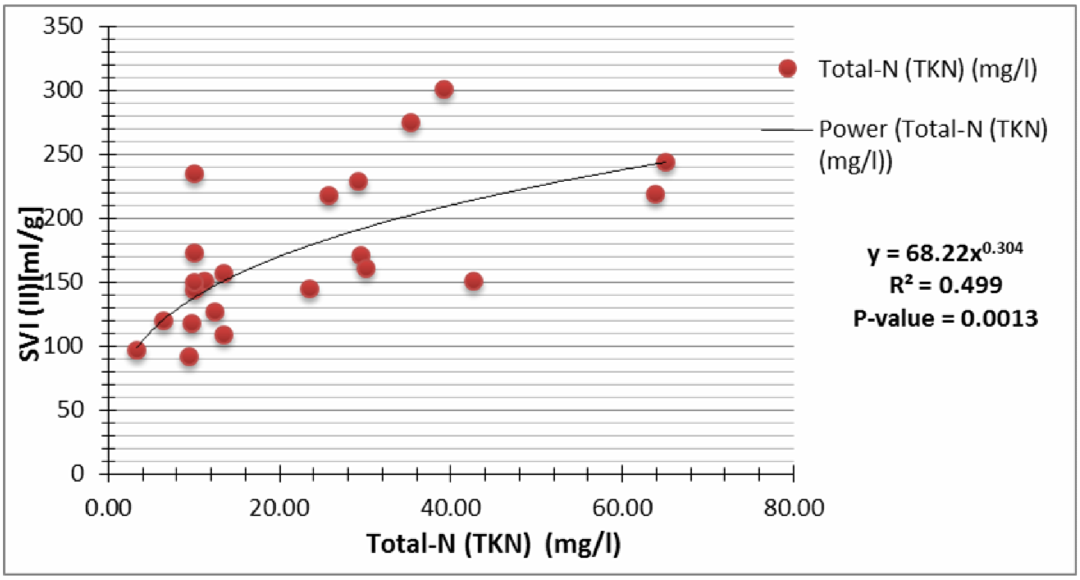

Figure 43. The relationship between sludge volume index (SVI 2) and total-N in sedimentation tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

In Figures (44 and 45) there is no relationship between the SVI and the COD values from both sedimentation tanks (1&2). There is no Palestinian standard of COD in the water re-use standards Table 4.1.

Also there is no effect of the BOD values from both sedimentation tanks (1&2) on SVI Figures (46 and 47) and these values were within the range of the Palestinian standards of water re-use.

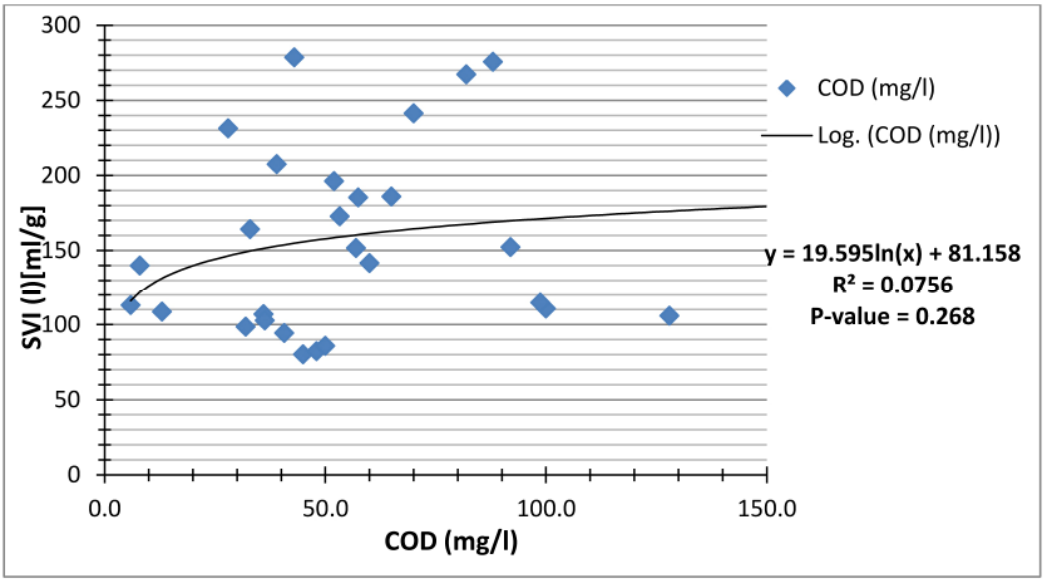

Figure 44. The relationship between sludge volume index (SVI 1) and COD in sedimentation tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

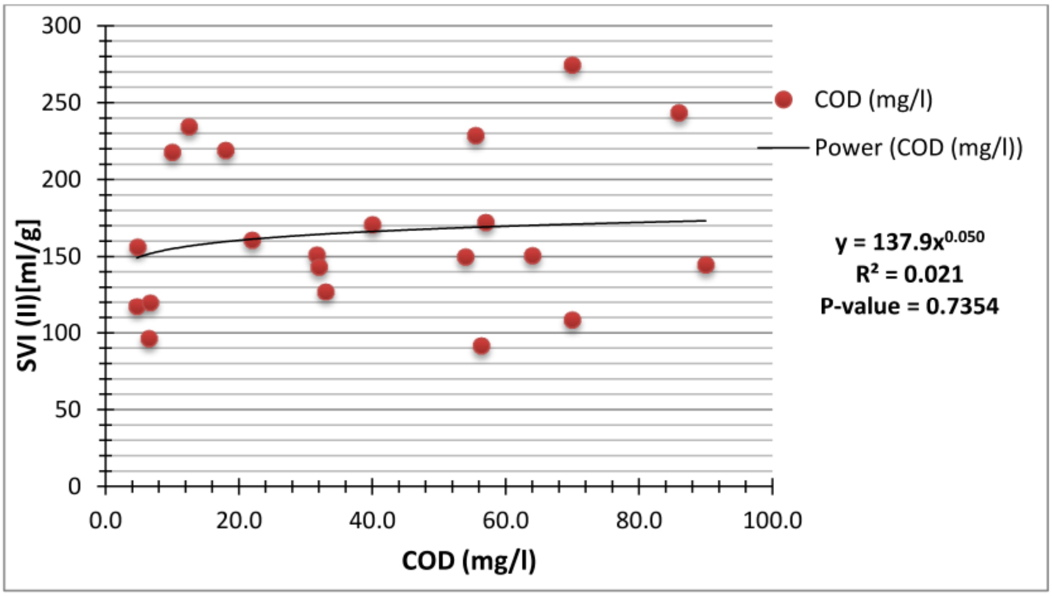

Figure 45. The relationship between sludge volume index (SVI 2) and COD in sedimentation tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

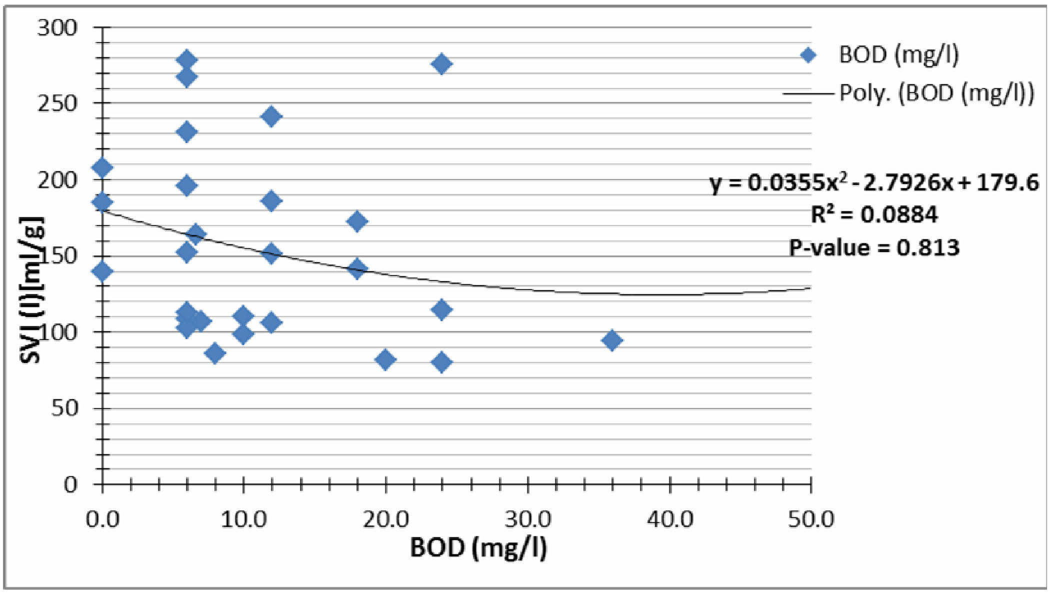

Figure 46. The relationship between sludge volume index (SVI 1) and BOD in sedimentation tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

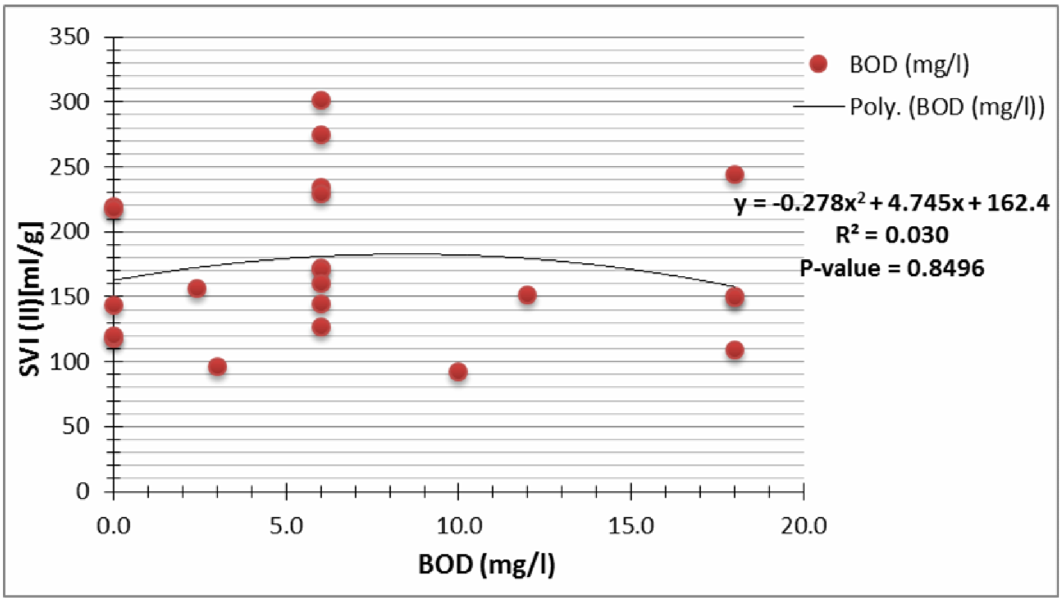

Figure 47. The relationship between sludge volume index (SVI 2) and BOD in sedimentation tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

In Figures (48 and 49) there is no important relationship between the SVI and the Fecal Coliform (FC) number from both sedimentation tanks (1&2). The number of the FC during the appearance of sludge bulking and foaming from both sedimentation tanks (1&2) were higher than the range of the water re-use standards Table 4.1.

As shown in Figures (50 and 51) there is a stronger significant relationship between the increase of the Total Coliform (TC) and the increase of SVI value in sedimentation tanks (2). There was a huge number of the TC from both sedimentation tanks during the appearance of sludge bulking and foaming phenomena.

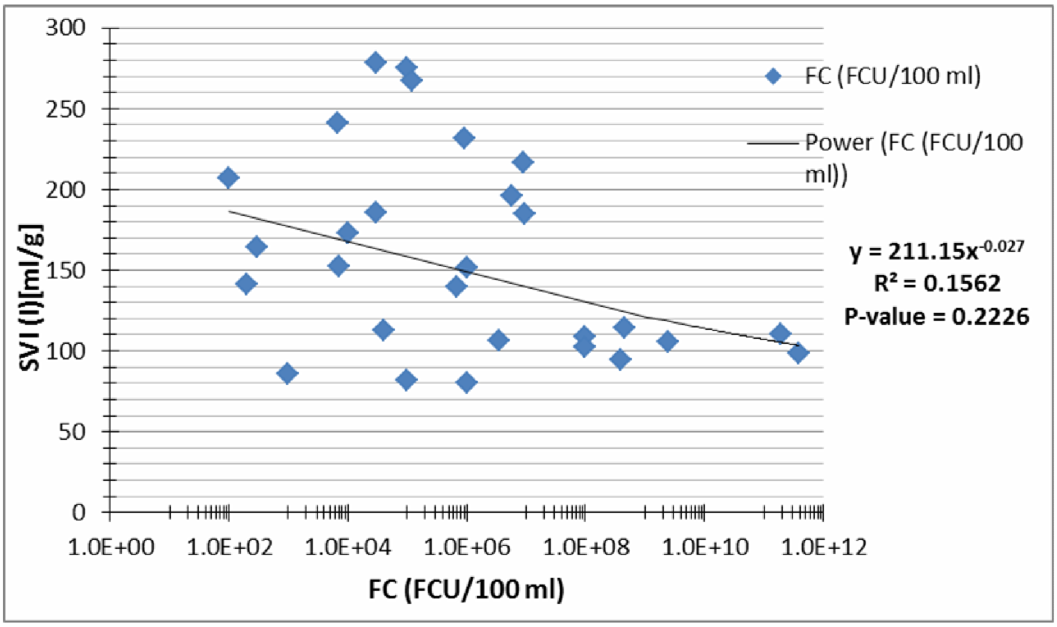

Figure 48. The relationship between sludge volume index (SVI 1) and Fecal Coliform (FC) in sedimentation tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

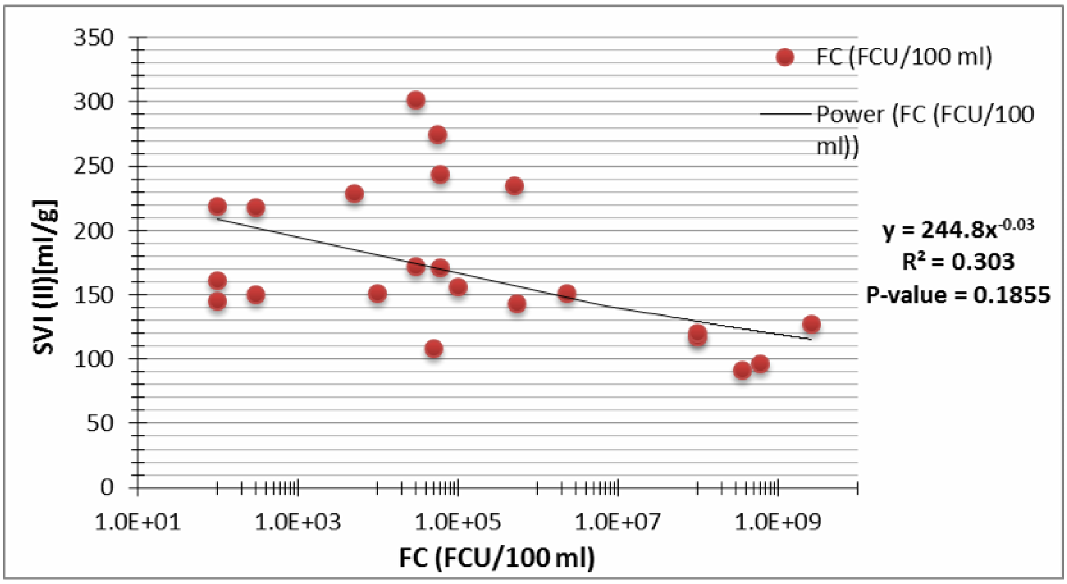

Figure 49. The relationship between sludge volume index (SVI 2) and Fecal Coliform (FC) in sedimentation tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

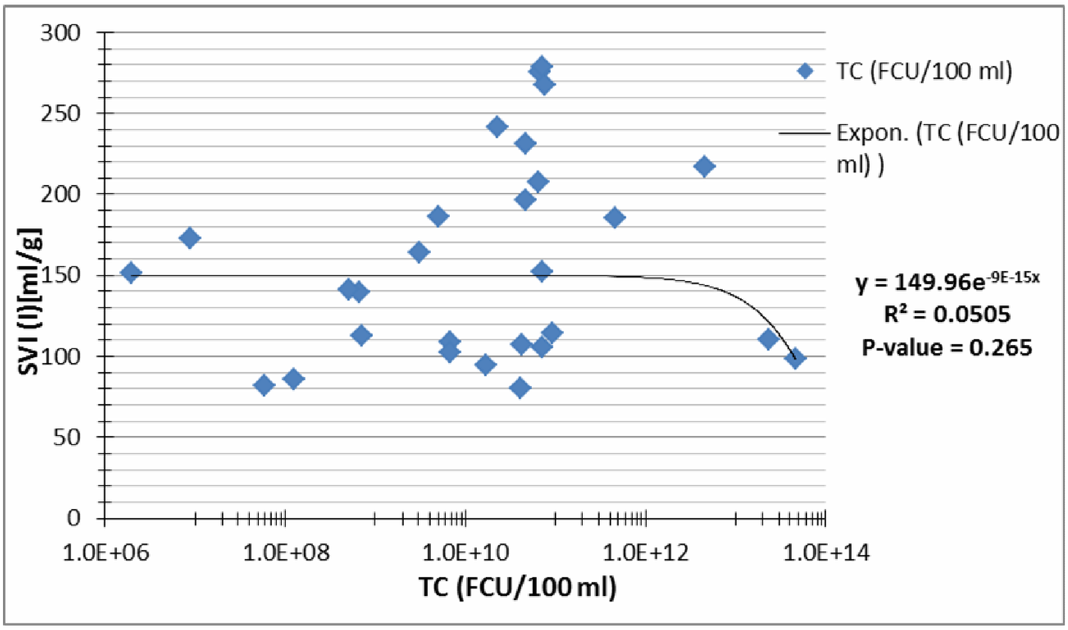

Figure 50. The relationship between sludge volume index (SVI 1) and Total Coliform (TC) in sedimentation tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

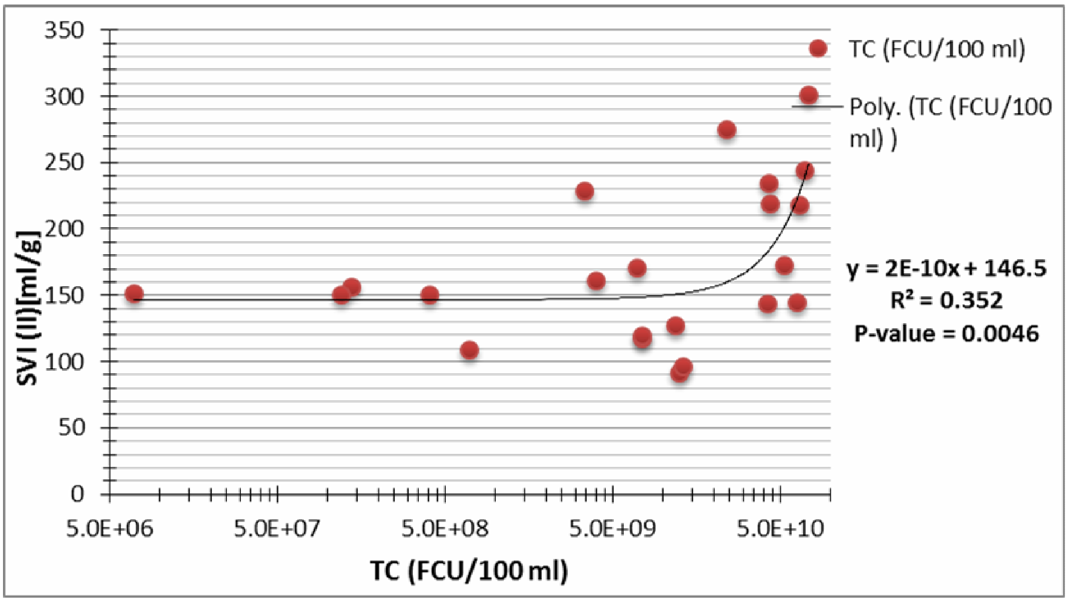

Figure 51. The relationship between sludge volume index (SVI 2) and Total Coliform (TC) in sedimentation tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

As shown in Figures (52 and 53) there is a significant relationship between the increase of the turbidity and the increase of SVI value in sedimentation tank (1). The P-value is less than 0.05 where it equal to 0.011.

The amount of the turbidity during the appearance of the sludge bulking and foaming from the sedimentation tank (1) was high and there is no Palestinian standards for the turbidity in water re-use standards.

In (Figures 54, 55) there is also significant relationship between the increase of the TSS and the increase of the SVI value just from the sedimentation tank (2). The amount of the TSS from both sedimentation tanks was higher than the range of the water re-use standards in Palestine.

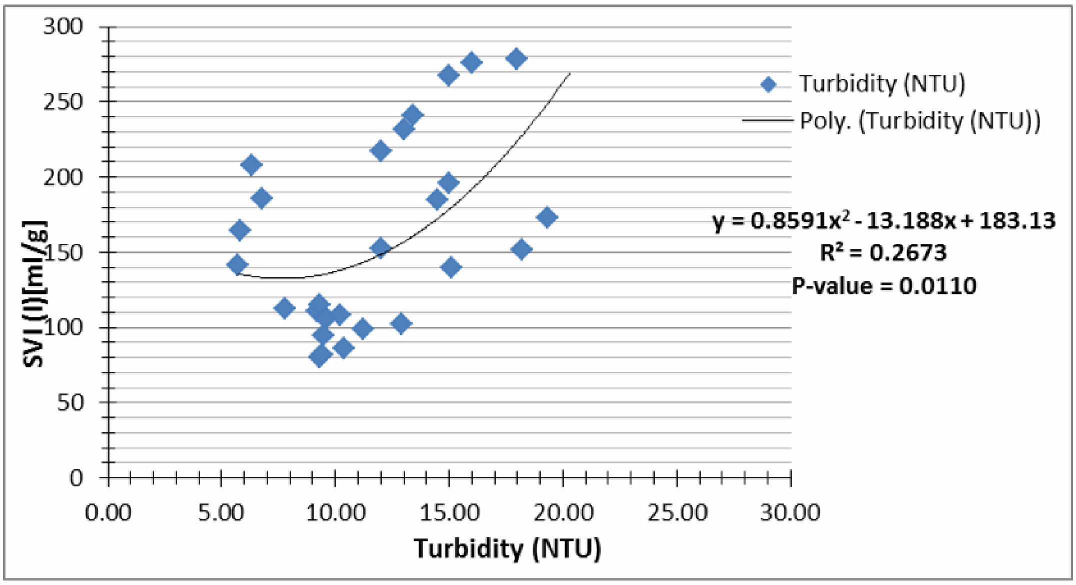

Figure 52. The relationship between sludge volume index (SVI 1) and turbidity in sedimentation tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

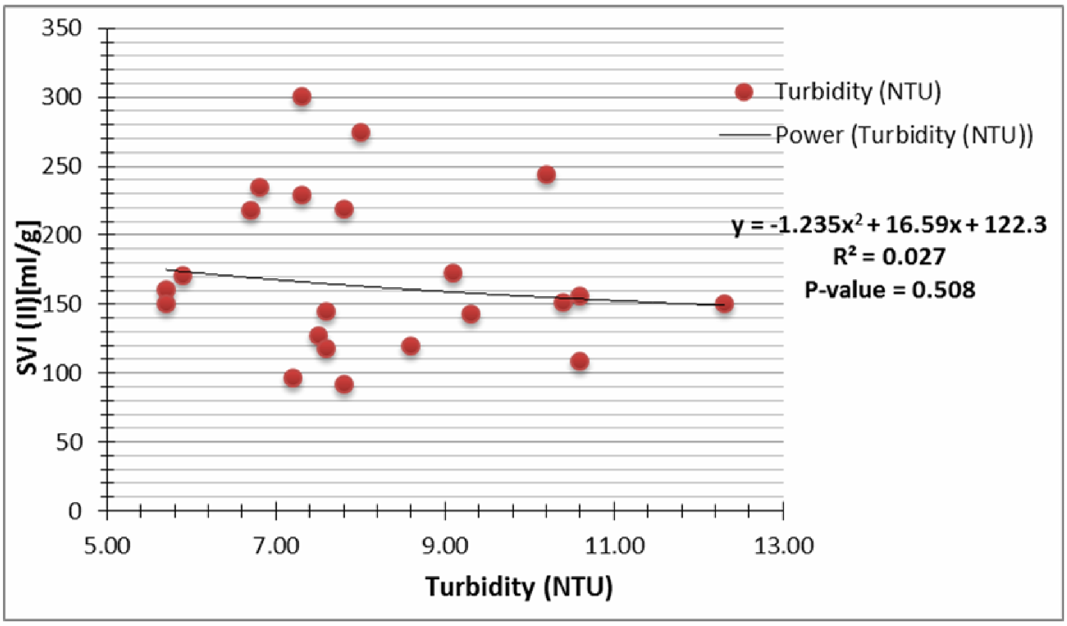

Figure 53. The relationship between sludge volume index (SVI 2) and turbidity in sedimentation tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

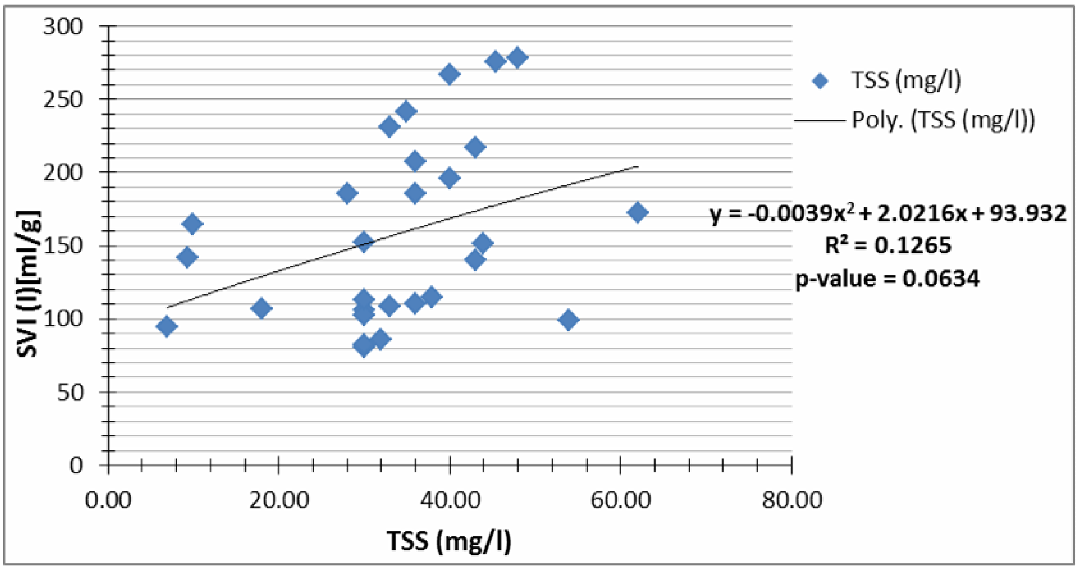

Figure 54. The relationship between sludge volume index (SVI 1) and Total suspended solid (TSS) in sedimentation tank (1) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

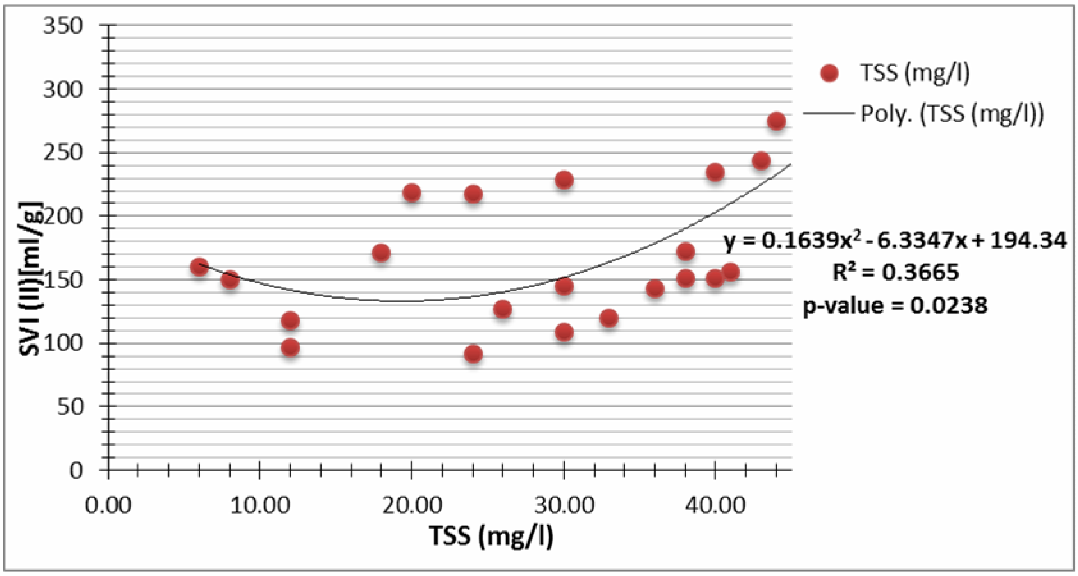

Figure 55. The relationship between sludge volume index (SVI 2) and Total suspended solid (TSS) in sedimentation tank (2) of Al-Bireh Wastewater Treatment Plant during the period from October, 2009 to July, 2010.

Figure 56. The aeration tank (1) in the spring (March, 2010)

Figure 57. The aeration tank (1) in the summer (July, 2010)

Foaming is a common operational problem in activated sludge processes that often adversely affects the quality of the treated effluent. In (Figure. 56) the foams layer covered the most area of the aeration tank in March when the all Mammoth rotors were turned off. Even when the Mammoth rotors were operating, the foam layer still appeared. The foams layers depth was more than 30-40 cm with a brown color and smell. This phenomenon is starting to appear in the beginning of the winter but it is predominant in the spring and completely disappears in summer and autumn, as in (Figure. 57).

Figure 58. The Sedimentation tank (1) in the spring (March, 2010)

As shown in (Figure. 58) high flotation scum appears on the surface of the sedimentation tank in the spring at the same time of the foam appearance in the aeration tank. The flotation of scum in the sedimentation tank affects the effluent quality, where it causes a high turbidity of the effluent and then it increases the TSS in the effluent especially from the sedimentation tank (2) Figures (59 and 60).

In the summer when the foams disappear in both aeration tanks, there is no flotation scum in sedimentation tanks, no turbidity in the effluent and the amount of the TSS was around 8 mg/l Figures (61 and 62).

Figure 59. Turbid effluents from the sedimentation tank (1) (April, 2010)

Figure 60. Turbid effluents from the sedimentation tank (2) (March, 2010)

Figure 61. The Sedimentation tank (1) in the summer (July, 2010)

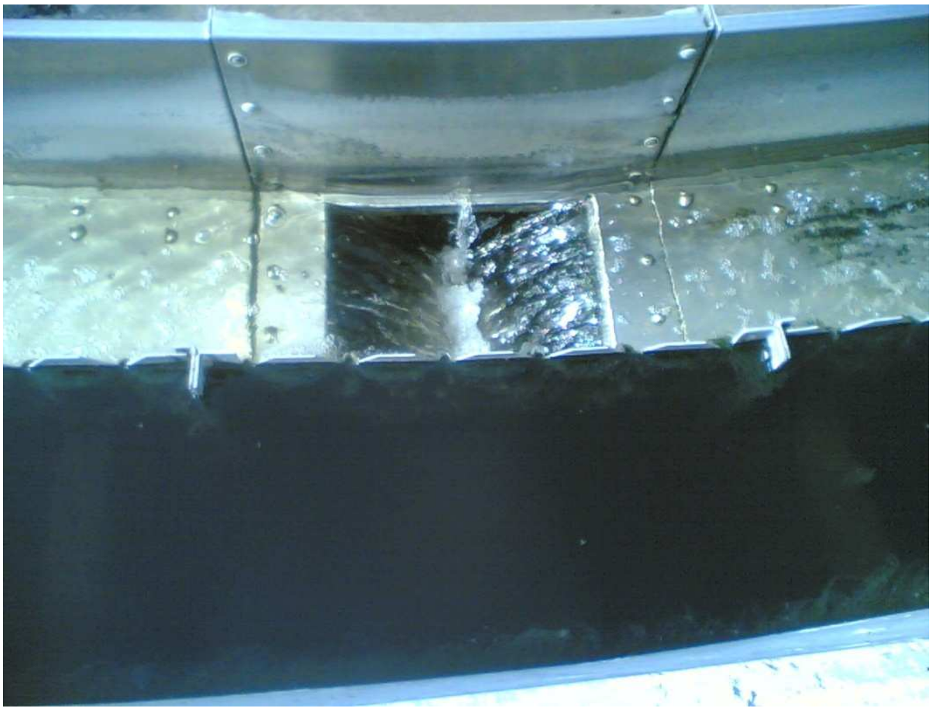

Figure 62. The effluent from the sedimentation tank (2) (June, 2010)

4.4. Microbial activities of foams in activated sludge system

This study tried to evaluate the presence of pathogens and microbial communities in the foam, especially after making the total coilform and fecal coliform tests for the foams during all four seasons, and a high number of coilform were observed all the time. The samples were taken in the winter and the spring. After the analysis of samples, the observation insures that the overgrowth of pathogens occurred in the spring more than in the winter. The components of the microbes in the activated sludge system are the same as the components that are present in the foam and that there are at least three types of pathogens present in the foam that can be dangerous when the handling of these foam.

Selective and Differential media were used to check the type of some pathogens that were found in the foams sample. For example Mannitol salt agar (MSA) is a selective media that has a pink color (Figure. 63) and it is used for the isolation of pathogenic staphylococci. *Staphylococcus* is Gram-positive, catalase-positive cocci. Catalase is the enzyme responsible for degradation of hydrogen peroxide. It was determined by adding a solution of dilute H_2O_2 to a bacterial smear on a glass slide. The formation of bubbles (O2) is evidence of catalase activity.

On MSA, pathogenic *Staphylococcus aureus* produces small colonies surrounded by yellow zones. The reason for this change in color is that *S. aureus* ferments the mannitol, producing an acid, which, in turn, changes the indicator from red to yellow. The growth of other types of bacteria is generally inhibited. *Staphylococcus aureus*, on the other hand, is associated with a wide variety of disease states, including impetigo, toxic shock syndrome and food poisoning.

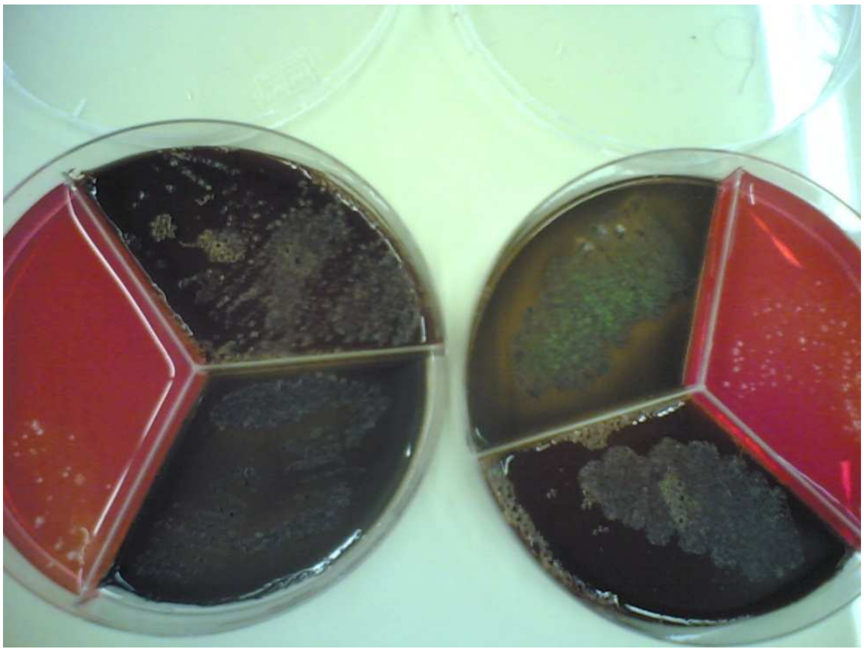

Figure 63. Foams samples using many types of media (Manitol salt Agar, MacCkonkeys Agar and Enrichment medium), (IEWS lab)

Other types of pathogens were determined when used the blood agar plates. As shown in (Figure. 64) three types of hemolytic reactions can be observed on blood agar plates which are alpha, Beta and gamma.

The pathogens are producing an exotoxin which causes the complete lysis of red blood cells. In (Figure. 64) when these pathogens are grown on the blood agar, their colonies are surrounded by a yellowish halo of complete clearing on a background of the bright red agar and called Beta hemolysins. The existence of these types of pathogens in foams is very dangerous, especially for the employees of the Al-Bireh wastewater treatment plant and other researchers if they do not follow the safety instructions.

There are also alpha hemolysins which are appearing as a few colonies in this Figure. It partially lyses the red blood cells and reduces the hemoglobin to methemoglobin which produces a green zone around the colony. In Gamma hemolysis there is no hemolysis or changes in the red blood cells occurred.

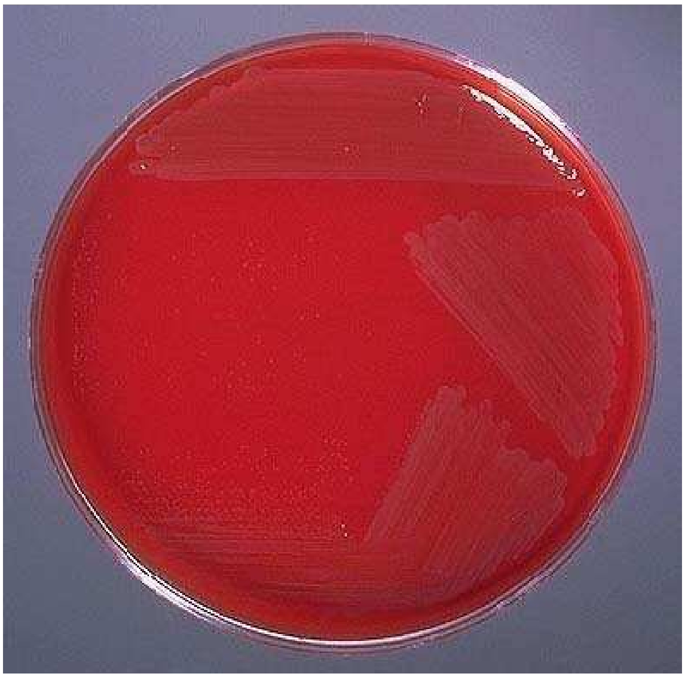

Figure 64. Foams sample using Blood Agar (Med Lab. Ramallah)

Pseudomonas aeruginosa are observed when using MacConkey agar, which has a brown color (Figure. 63), to identify the gram negative bacteria and then used selective media for *Pseudomonas aeruginosa.*

Also in this research observed at least three types of microorganisms that found in the foam. Acording to Al-Sa`ed, (1983) methodology, they can be analyzed starch, lipids, and proteins in winter and spring (Figure 65). Degradation was increased in the spring more than the winter.

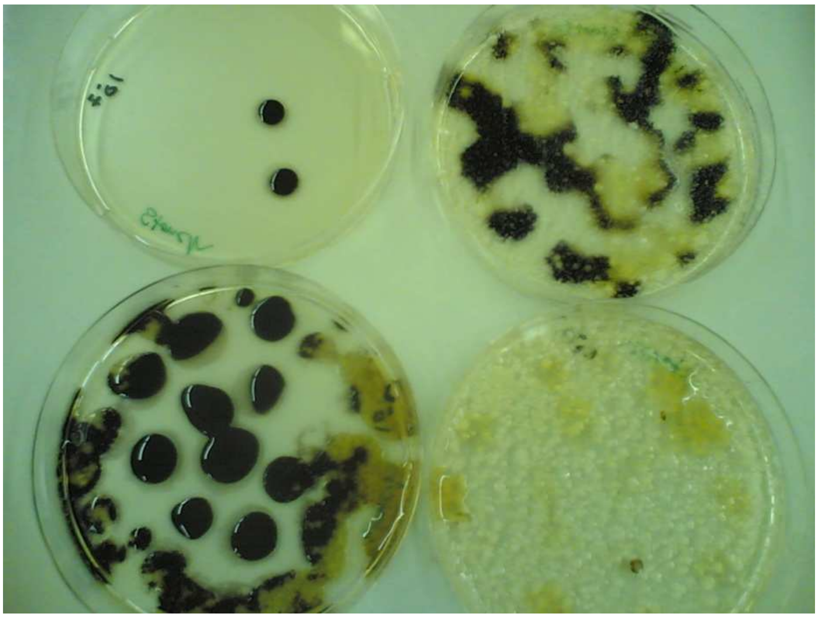

Figure 65. Biochemical test (Amylase test) of foam from AWWTP (IEWS lab).

CHAPTER FIVE

Conclusions and recommendations

5.1 Conclusions

- The decrease of the temperature is considered the critical point that is responsible for the increase of SVI in both aeration tanks and the cause of sludge bulking and foaming phenomena in Al-Bireh wastewater treatment plant.
- The significant decrease of the Dissolved Oxygen (DO) concentration and the increase of F/M ratio and (FOG) level were the main reasons that lead to the increase of the sludge volume index (SVI) especially in aeration tank (1) and the cause of the overgrowth of filamentous bacteria in the system.
- The dominant filamentous bacteria identified from both aeration tanks were *Microthrix parvicella, Type 1701, and Sphaerotilus natans* while the *Nocardia* were the most dominant filaments in the foam samples.
- The increase of the SVI in both aeration tanks caused an increase of the TKN in both sedimentation tanks (1&2), turbidity in sedimentation tank (1), TSS, and TC in sedimentation tank (2).
- The most parameters that were measured during the appearance of the sludge bulking and foaming phenomena were within the range of the Palestinian standards of water re-use except the DO in sedimentation tank (1), as well as the FC and TSS in both sedimentation tanks.

- The observation insures that the components of the microbes in the activated sludge system were the same as the components present in the foam. There were at least three types of pathogens found in the foam: analysis starch, lipids, and proteins in the spring more than the winter.
- The existence of these types of pathogens in foams is very dangerous, especially for the employees of the Al-Bireh wastewater treatment plant and other researchers if they do not follow the safety instructions.

5.2 Recommendations

- Find a suitable solution to increase the amount of the DO concentration especially for the aeration tank (1).
- Work to find the best solution to decrease the amount of the FOG that passes from the influent to the treatment plant through make primary sedimentation tank or increase the awareness to prevent oil discharge from the sinks.
- Make further studies about the presence of pathogens in the foam and the ability of the pathogens to transition from the foam to the aerosols.
- Evaluate the fate of pathogens in liquid and sludge lines at AWWTP during the appearance of sludge bulking and foaming phenomena.
- Make another research to find a suitable solution to decrease the TKN during the sludge bulking and foaming phenomena.

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الخلاصة

تعتبر ظاهرة الحمأة المنتفخه (sludge bulking) وظهور الرغوة بشكل كبير في خزانات التهويه من المشاكل الرئيسية التي تؤثر على تشغيل نظام تنشيط الحمأة. هذه الظاهرة تؤدي الى ابطاء عمليات الترسيب وتكثيف المواد الصلبة في حوض الترويق الثانوي. كل هذا من شأنه أن يؤدي الى ارتفاع تركيز العوالق في المياه المعالجة مع وجود اثار سلبية على اداء عملية المعالجة.

ان الهدف الرئيسي من هذه الدراسة هو معرفة الأسباب الرئيسية التي تسبب هاتين الظاهرتين (sludge bulking and foaming) في محطة البيرة لمعالجة مياه الصرف الصحي وذلك من خلال التحليل الفيزيائي والكيميائي والبيلوجي لعينات من خزانات التهوية والترسييب (1 و 2) . وكان الهدف أيضا من الدراسة هو التحقق من أداء عملية المعالجة واستدامة نظام تنشيط الحمأة اثناء ظهور هاتين الظاهرتين مع التركيز بشكل خاص على امكانية إعادة استخدام مياه الصرف الصحي.

لقد توصلت هذه الدراسة للاسباب المسببة لكلتا الظاهرتين عن طريق مراقبة المتغير(SVI) في خزانات التهوية خلال مدة زمنية قدرها عشرة أشهر. ولقد استخدم البرنامج الاحصائي(SPSS) لمعرفة العلاقة مابين المتغير(SVI) وباقي المتغيرات التي قيست.

ان الانخفاض في درجة الحرارة، والانخفاض في تركيز الأوكسجين المذاب وزيادة كمية الزيوت والدهون والشحوم الداخله الى المحطه وارتفاع نسبة الحمل العضوي للحمأة (F/M ratio) كل هذا يعد من الأسباب الرئيسية التي أدت إلى زيادة المتغير (SVI)لاسيما في خزان التهوية (1). وبالتالي أدت الى زيادة نمو انواع خاصة من البكتيريا الخيطية ونشوء تلك الظاهرتين .

من انواع البكتيريا الخيطية التي لوحظ ازدياد نموها اثناء ظهور هاتين الظاهرتين هي :

Nocardia, Microthrix parvicella, Type 1701, and Sphaerotilus natans. حيث وجدت بأعداد كبيرة في العينات التي أخذت من خزانات التهويه والرغوة. لقد تم ملاحظة سيادة البكتيريا الخيطية *Microthrix parvicella* أثناء انخفاض درجات الحرارة بينما سادت *Nocardia* أثناء درجات الحرارة الدافئة.

لقد لوحظ أيضا أن بعض أنواع البكتيريا الخيطية الموجودة في خزانات التهويه و الرغوة الطافية على خزانات التهويه كانت موجودة بشكل ثابت أثناء فترة الدراسة ولم تتأثر بتغير الفصول الأربعة وبالتالي لا يوجد لها أي تأثير على ظاهرتي (sludge bulking and foaming).

Annexes

Annex (1) Dissolved oxygen (DO) measurements at 3.5 m depth in aeration tank (1)

Annex (2) Dissolved oxygen (DO) measurements at 3.5 m depth in aeration tank (2)

Annex (3) all parameters that measured from the influent

Annex (4) all parameters that measured from the sedimentation tank (1)

Annex (5) all parameters that measured from the sedimentation tank (2)