African Journal of Biotechnology Vol. 11(5), pp. 1163-1168, 16 January, 2012
Available online at http://www.academicjournals.org/AJB
DOI: 10.5897/AJB11.2869
ISSN 1684–5315 © 2012 Academic Journals

Full Length Research Paper

Efficiency of some actinomycete isolates in biological treatment and removal of heavy metals from wastewater

Wael N. Hozzein[1,2]*, Mohammed Bastawy Ahmed[3] and Marzouka Shaban Abdel Tawab[4]

[1]Chair of Advanced Proteomics and Cytomics Research, Zoology Department, College of Science, King Saud University, Riyadh, Saudi Arabia.
[2]Botany Department, Faculty of Science, Beni-Suef University, Beni-Suef, Egypt.
[3]Biochemistry Department, Faculty of Science, Beni-Suef University, Beni-Suef, Egypt.
[4]Beni-Suef Wastewater Treatment Plant, Beni-Suef, Egypt.

Accepted 2 December, 2011

The main focus of studies and research in the field of wastewater treatment is treating wastewater without causing environmental hazards as well as getting benefits from the treated waste materials. In this regard, the aim of the current study was to isolate some actinomycete strains from Beni-Suef Wastewater Treatment Plant to study their capacities for biological treatment and for removal of heavy metals from the wastewater. For this purpose, 17 actinomycete strains were isolated from 3 wastewater samples representing the main stages of treatment in the plant, namely: the influent, the secondary sedimentation tank and the effluent. Then, 10 morphologically dissimilar isolates were selected. The 10 selected actinomycete isolates were characterized by their morphological characteristics and were found to belong to the genera *Nocardia*, *Streptomyces*, *Rhodococcus*, *Gordonia* and *Nocardiopsis*. The ability of the selected actinomycetes for biological treatment of the wastewater was evaluated by measuring the biochemical oxygen demand (BOD), the chemical oxygen demand (COD) and the total suspended solids (TSS); and their ability for removal of some heavy metals (Cu, Fe, Mn, Pb and Zn) was also determined. The results showed that most of the selected actinomycetes were effective in the biological treatment of the wastewater and have the ability to decrease the values of BOD, COD and TSS; and also to reduce the concentrations of the tested heavy metals (Cu, Fe, Mn, Pb and Zn) markedly. The *Streptomyces* strain C11 was found to be the most efficient organism in respect to biological treatment of the wastewater and removal of the heavy metals from the raw wastewater.

Key words: Actinomycetes, wastewater, biological treatment, heavy metals removal.

INTRODUCTION

The increased awareness of the role of microorganisms in diseases led to an enhanced demand for wastewater treatment. This has resulted in the passage of pieces of legislation that encouraged the construction of wastewater treatment plants (Guest, 1987). The practice of wastewater treatment started at the beginning of the twentieth century. The British Royal Commission on Sewage Disposal proposed that the goal of wastewater treatment should be to produce a final effluent of 30 mg/l of suspended solids and 20 mg/l of biochemical oxygen demand (BOD) (Sterritt and Lester, 1988). Research has focused on microbial populations in the wastewater environment for their importance in biological treatment (Chan et al., 2009; Sim et al., 2010). Actinomycetes in the wastewater treatment plants are able to use several

*Corresponding author. E-mail: hozzein29@yahoo.com.

Abbreviations: BOD, Biochemical oxygen demand; **COD,** chemical oxygen demand; **TSS,** total suspended solids.

growth substrates varying from sugars to high molecular weight polysaccharides, proteins and aromatic compounds (Lemner and Kroppenstedt, 1984; Lemmer, 1986). Other advantages of actinomycetes over other wastewater bacteria are their higher resistance to desiccation and ultraviolet irradiation; and their ability to store poly-phosphates and poly-β-hydroxybutyric acid (Lemmer and Baumann, 1988). Recently, actinomycetes in activated sludge and wastewater have become the focus of research because they are believed to play an important role in sludge bulking and foaming in activated sludge plants (Davenport et al., 2000; Madoni et al., 2000; Heard et al., 2008). Apart from these negative roles, actinomycetes were found to be active in the decomposition of organic materials in the wastewater bioreactors (McCarthy, 1987).

Many recent studies reported that more applied research is still needed and useful in this field (Fattaa and Anayiotoub, 2007; Yang et al., 2011). Therefore, the objective of the present study was to examine the ability of the actinomycete strains isolated from wastewater samples for biological treatment and to study their ability in the removal of heavy metals from the wastewater to make better use of the microbial resources in this interesting environment.

MATERIALS AND METHODS

Sampling

The Wastewater Treatment Plant (WWTP) in Beni-Suef is working by the extended activated sludge system, where the raw sewage is received in the inlet chamber (influent) as explained in details elsewhere (Hozzein et al., 2008). Samples for isolation were collected manually in sterile 500 ml non-reactive borosilicate glass from the three sites representing the main different stages in the wastewater plant; the influent, the secondary sedimentation tank and the effluent.

Isolation of the wastewater actinomycetes

Isolation of the actinomycetes from the wastewater samples was carried out by the spread plate technique (Johnson et al., 1959) on yeast extract-malt extract agar (Pridham et al., 1956, 1957). The isolation plates were then incubated for 14 days at 30°C. After incubation, 17 isolates were selected from the plates and repeatedly streaked onto the same medium to obtain pure cultures. The pure isolates were preserved as stock suspensions in 20% glycerol at -20°C. Then, 10 morphologically different isolates were selected for further studies.

Characterization of the wastewater actinomycetes

The 10 selected actinomycete isolates were recognized and initially characterized by their morphological characteristics. The coverslip culture technique (Kawato and Shinobu, 1959) was used to prepare the cultures for the morphological examinations. Each coverslip culture was examined microscopically to investigate the mycelial structures, spore chain morphology and spore surface of the isolated actinomycetes; and photographed at the suitable magnification using light and scanning electron microscopes. A good review on the examination of morphology of actinomycetes was given by Williams and Cross (1971). Characterization of the isolates was based on the guide described in Bergey's manual of determinative bacteriology (Holt et al., 1994).

Efficiency of the actinomycete strains in biological treatment and removal of heavy metals

A 4 L sample from the influent waste (raw waste) was collected from Beni-Suef WWTP. Some physico-chemical characteristics of this untreated sample were measured; namely: biochemical oxygen demand (BOD), chemical oxygen demand (COD) and total suspended solids (TSS) according to Standard Methods for the Examination of Water and Wastewater (APHA, 1998). Also, the heavy metals (Cu, Fe, Mn, Pb and Zn) were measured in the untreated sample after digestion in concentrated HNO_3 and analyzed using the direct flame emission photometric method by atomic absorption spectrophotometer (ZEENIT 700) according to the manufacturer's operating manual. 1 L of the sample was divided into 10 flasks and each flask (contained 100 ml) was inoculated with a loopful of a young culture of the selected actinomycete strains. The flasks were incubated in a shaking incubator at 30°C for 5 days. The same physico-chemical parameters and heavy metals were measured again after the incubation period to evaluate the efficiency of the actinomycete strains in treatment of the wastewater sample. The treatment experiment was run in triplicates.

RESULTS AND DISCUSSION

More efficient and safer wastewater treatment methods or systems with minimum requirements and low costs are still needed to overcome problems or hazards of conventional methods currently used (Vymazal, 2002; Bécares, 2006; Puigagut et al., 2007). Isolation of microorganisms from unexploited environments holds tremendous promise as unexploited environments may yield novel organisms with new more interesting biotechnological applications that hopefully, will have greater impact in the future (Ōmura, 1986; Steele and Stowers, 1991; Hozzein and Goodfellow, 2007; Hozzein et al., 2011). In this study, the focus of the work was to isolate some actinomycetes from the wastewater environment which thought to cause the foaming and bulking problems in wastewater tanks and rather study their capabilities for biological treatment and removal of heavy metals from the wastewater. For this purpose, 17 actinomycete strains were isolated from the collected wastewater samples. Then, 10 isolates were selected for further studies based on differences in their cultural appearance. The ten selected wastewater actino-mycete strains, designated A11, A13, B21, B22, B31, C11, C12, C13, M1 and M13 were characterized by studying their morphological characteristics. The observations revealed that strains A11, B21 and B31 showed abundant growth with aerial mycelium color varied from white to yellow and substrate mycelium color varied from pale yellow to yellowish brown. Fragmentation of the substrate mycelium was observed and short spore chains were formed on the aerial mycelium. These characteristics are similar to members of genus *Nocardia*.

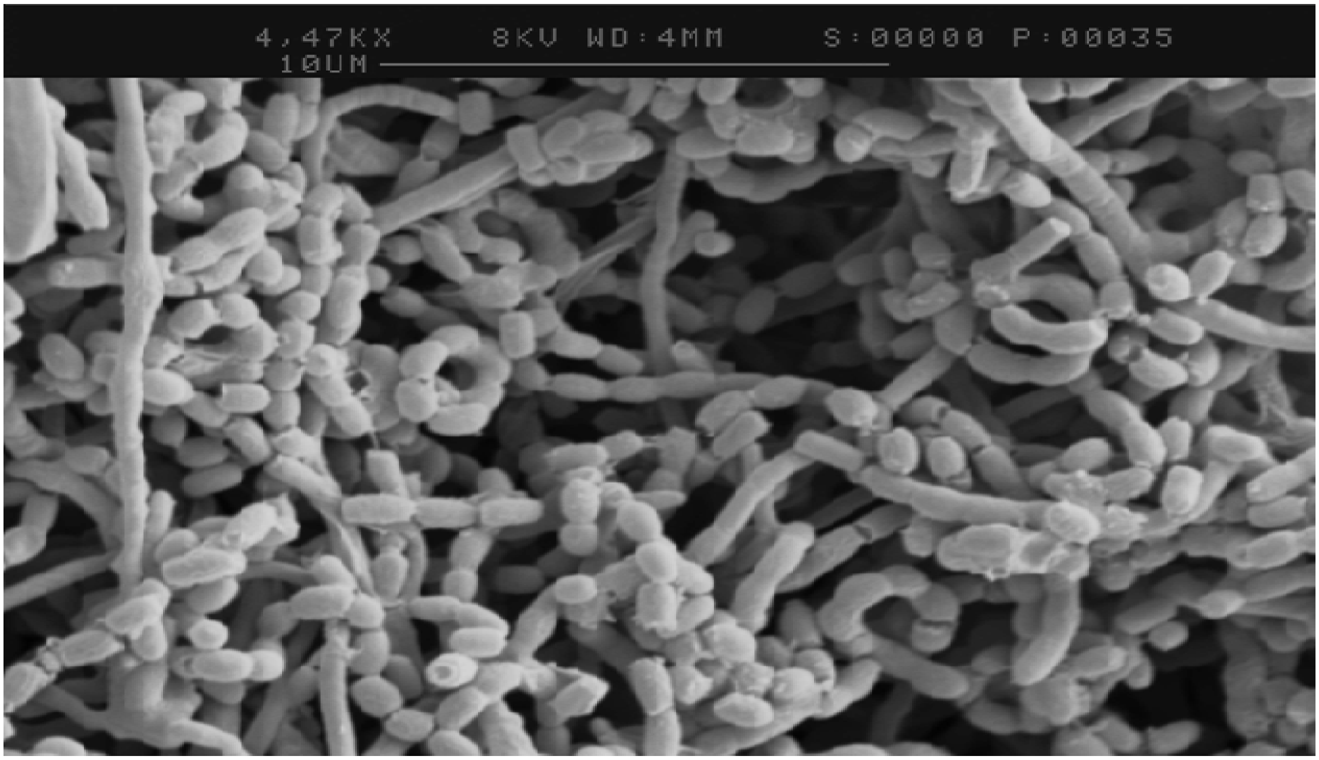

Figure 1. The electron micrograph of strain C11 showing long spore chains forming coil ends with smooth-surfaced spores.

Similarly,the strains A13 and B22 showed abundant growth with orange aerial mycelium color and brownish orange sub-strate mycelium color. Sparse aerial mycelium was formed and rod shaped and branched spores were observed. On the basis of these features, the strains A13 and B22 were initially characterized as members of the genus *Rhodococcus*.

On the other hand, the strains C11, C12 and C13 were very close to each other and showed abundant growth with aerial mycelium color varied from grayish white to gray and substrate mycelium color varied from yellowish white to grayish brown. Aerial mycelium with short to long chains of spores with coiled ends and smooth surfaces were formed (Figure 1), while fragmentation of the extensively branched substrate mycelium or other distinguished structures were not observed. These characteristics are typical for members of genus *Streptomyces*.

Strain M1 formed pink to reddish pink colonies. It produced short rod-like and cocci-like elements but aerial hyphae were not produced. These characteristics are similar to those of genus *Gordonia*. While, strain M13 showed moderate growth, yellowish white aerial mycelium and yellowish brown substrate mycelium. Long chains of spores were formed on the aerial hyphae which was zig-zag shaped before maturation and a sort of fragmentation of the substrate mycelium was observed. These characteristics are typical to members of genus *Nocardiopsis.*

These results are similar to the results of El-Shatoury et al. (2004) who reported that *Streptomyces*, *Nocardia* and *Nocardiopsis* were of the most dominant genera in the bed of a system constructed wetland for industrial waste-water treatment. In a recent study on the actinomycetes community in a starch factory wastewater, Saenna et al. (2011) indicated that out of 30 isolated actinomycete strains, 28 were classified as *Streptomyces* spp. and the other 2 as *Nocardia* spp. The ten characterized waste-water actinomycete strains were then tested for biological treatment and removal of heavy metals from the raw wastewater.

Capabilities of the actinomycete strains for wastewater treatment

Biological treatment is one of the most widely used treatment methods of wastewater. Efficiency of wastewater treatment is measured mainly in terms of removal percentage of biochemical oxygen demand (BOD), chemical oxygen demand (COD) and total suspended solids (TSS) (Roesler and Wise, 1974). The capabilities of the selected ten actinomycete isolates for treatment of the wastewater sample collected from Beni-Suef WWTP are summarized in Table 1. It was found that the starting values of the biochemical oxygen demand (BOD), chemical oxygen demand (COD) and total suspended solids (TSS) in the sample (raw waste before treatment) were 283, 498 and 316 mg/l, respectively. The results in Table 1 revealed

Table 1. The capabilities of the actinomycete strains for treatment of wastewater.

Organism	BOD (mg/l)	BOD removal (%)	COD (mg/l)	COD removal (%)	TSS (mg/l)	TSS removal (%)
Raw water	283	--	498	--	316	--
A11	45	84.0	200	59.8	70	83.1
A13	85	69.9	264	46.9	80	74.6
B21	22	92.2	115	76.9	14	95.6
B22	100	64.6	260	47.7	90	71.5
B31	25	91.1	163	67.2	30	90.5
C11	13	95.4	76	84.7	20	93.7
C12	20	92.9	100	79.9	25	92.1
C13	25	91.1	120	75.9	27	91.4
M1	90	68.1	164	67.0	86	72.7
M13	18	93.6	110	77.9	24	92.4

BOD, Biochemical oxygen demand; COD, chemical oxygen demand; TSS, total suspended solid.

that high BOD removal efficiency was achieved by strain C11 followed by the strains M13, C12 and B21. The BOD concentration decreased from 283 mg/l in the raw waste to 13 mg/l in the effluent after the treatment with strain C11 which means a BOD removal efficiency of 95.4%. Also, the highest efficiency of COD removal was achieved by strain C11 which decreased the COD concentration from 498 mg/l in the raw waste to 76 mg/l in the effluent, corresponding to 84.7% removal efficiency. The remaining COD is the amount of hard and/or non-biodegradable materials (Fritsche and Hofrichter, 2005). This result is in agreement with the results of Buzzini et al. (2006) who reported that the average COD removal efficiency is 80 to 86%. On the other hand, the highest TSS removal efficiency was achieved by strain B21 followed by strain C11. The two strains showed TSS removal efficiencies of 95.6 and 93.7%, respectively. In general, high quality effluent was delivered by treatment with the *Streptomyces* strains C11, C12 and C13, the *Nocardia* strain B21; and the *Nocardiopsis* strain M13 with respect to the measured three parameters (BOD, COD and TSS). Similar results of good effluent after biological treatment were reported by various researchers (Al-Shahwani and Horan, 1991; Martin-Cereceda et al., 1996; Chen et al., 2004; Lee et al., 2004).

The ultimate function of the biological treatment is to remove the organic matter to meet the correspon-ding discharge standards set by the local government.

According to the local standards applied for Beni-Suef WWTP, the final effluent should have less than 60 mg/l of BOD, 80 mg/l of COD and 50 mg/l of TSS (law 48/82, discharge into non potable surface water, EEAA and EPAP, 2002). Therefore, the *Streptomyces* strain C11 was found to be the most efficient organism in the biological treatment and the only organism met the local standards applied for the WWTP under study as it produced a final effluent with 13 mg/l of BOD, 76 mg/l of COD and 20 mg/l of TSS. This means that the *Streptomyces* strain C11 is highly efficient in the removal of BOD and TSS, but less efficient in removal of COD, although it was the only strain that met the local standards for COD.

Capabilities of the actinomycete strains for the removal of heavy metals

Waste containing metals is directly or indirectly discharged into the environment, especially in developing countries, having brought serious environmental pollution and threatens biolife (Bishop, 2002; Wang, 2002). Although, the heavy metal contents in urban drainage systems generally do not reach the proportions found in industrial effluents, the problems caused by their presence, particularly in areas with dense population are of public concern. Many recent studies reported that the use of biological processes in the treatment of wastewater containing high concentrations of metals can overcome some of the limitation of physical and chemical treat-ments and provide a cost effective removal of metals (Veglio and Beolchini, 1997; Sag et al., 1998, 2000; Puranik et al., 1999; Volesky, 2001; Ahluwalia and Goyal, 2007; Das, 2010). The results in Table 2 indicated that a significant decrease in the concentrations of the five tested heavy metals (Cu, Fe, Mn, Pb and Zn) was achie-ved by the actinomycete strains under study. The highest Cu, Mn and Zn removal efficiencies (77.5, 90.9 and 80.7%) were achieved by the *Streptomyces* strain C11. On the other hand, the highest Fe removal efficiency (95.5%) was achieved by the *Nocardia* strain B21, while the highest Pb removal efficiency (56.5%) was achieved by the *Nocardiopsis* strain M13. Among the 10 actinomy-cete strains, the *Streptomyces* strain C11 also was the most efficient organism in respect to removal of three of the studied five heavy metals from the wastewater raw sample.

Some previous studies indicated also the biosorption

Table 2. The capabilities of the actinomycete strains for removal of heavy metals from the wastewater.

Organism	Cu (mg/l)	Cu removal (%)	Fe (mg/l)	Fe removal (%)	Mn (mg/l)	Mn removal (%)	Pb (mg/l)	Pb removal (%)	Zn (mg/l)	Zn removal (%)
Raw water	0.12	--	5.75	--	0.331	--	0.6	--	0.26	--
A11	0.0897	25.25	1.6	72.1	0.1	69.7	0.586	2.3	0.14	46.1
A13	0.116	3.3	1.5	73.9	0.06	81.8	0.45	25	0.085	67.3
B21	0.06	50	0.255	95.5	0.04	87.9	0.262	56.3	0.074	71.5
B22	0.0288	76	2.8	51.3	0.114	65.5	0.303	49.5	0.12	53.8
B31	0.065	45.8	0.28	91.0	0.09	72.8	0.28	53.3	0.08	69.2
C11	0.027	77.5	0.4	93.0	0.03	90.9	0.303	49.5	0.05	80.7
C12	0.043	64.1	0.26	95.4	0.059	82.1	0.304	49.3	0.063	75.7
C13	0.0433	63.9	0.29	94.9	0.06	81.8	0.29	51.6	0.065	75
M1	0.081	32.5	2.3	60	0.096	70.9	0.325	45.8	0.137	47.3
M13	0.05	58.3	0.28	95.1	0.06	81.8	0.261	56.5	0.1	61.5

capacity of toxic metals by *Streptomyces* species. Mameri et al. (1999) studied zinc biosorption capacity of *Streptomyces rimosus* biomass in batch mode. Also, the lead biosorption capacity of *S. rimosus* biomass was reported in the batch mode (Selatnia et al., 2004). Moreover, *Streptomyces clavuligerus* biomass is used commercially in biosorption of Cd (Zouboulis et al., 2004). It is worth mentioning here that Das (2010) recently stated that among 19 tested actinomycete strains, *Streptomyces phaeochromogenes* HUT6013 showed maximum biosorption of gold.

These high percentages in heavy metals removal by actinomycetes can be attributed to a big surface area they offer for metal binding or due to the production of some extracellular polymers that have great affinity and can scavenge metals as suggested by Weiner (1997).

Conclusions

One of the remarkable findings in the present study is that the most efficient strains, C11, B21 and M13 in biological treatment of the wastewater raw sample were also the effective in heavy metals removal. In conclusion, the results draw the attention to the bright side of actinomycetes in the wastewater interesting environment and are encouraging to test other actinomycetes for wastewater treatment and to apply the most efficient strains at the batch or small plant treatment level.

ACKNOWLEDGEMENT

A grateful support from the Center of Excellence for Biodiversity Research, College of Science, King Saud University, Riyadh, Saudi Arabia is highly appreciated.

REFERENCES

Ahluwalia SS, Goyal D (2007). Microbial and plant derived biomass for removal of heavy metals from wastewater. Bioresour. Technol. 98: 2243-2257.

Al-Shahwani SM, Horan NJ (1991). The use of protozoa to indicate changes in the performance of activated sludge plants. Water Res. 25: 633-638.

APHA (1998). Standards Methods for the Examination of Water and Wastewater, 20th ed. American Public Health Association, Washington, DC.

Bécares E (2006). Limnology of natural systems for wastewater treatment. Ten years of experiences at the experimental field for low-cost sanitation in Mansilla de las Mulas (León, Spain). Limnetica, 25(1-2): 143-154.

Bishop PL (2002). Pollution prevention: fundamentals and practice. Tsinghua University Press, Beijing.

Buzzini AP, Sakamoto IK, Varesche MB, Pires EC (2006). Evaluation of the microbial diversity in an UASB reactor treating wastewater from an unbleached pulp plant. Process Biochem. 41: 168-176.

Chan YJ, Chong MF, Law CL, Hassell DG (2009). A review on anaerobic–aerobic treatment of industrial and municipal wastewater. Chem. Eng. J. 155: 1-18.

Chen S, Xu M, Cao H, Zhu J, Zhou K, Xu J, Yang X, Gan Y, Liu W, Zhai J, Shao Y (2004). The activated sludge fauna and performance of five sewage treatment plants in Beijing, China. Eur. J. Protistol. 40: 147-152.

Das N (2010). Recovery of precious metals through biosorption. Hydrometallurgy 103: 180-189.

Davenport RJ, Curtis TP, Goodfellow M, Stainsby FM, Bingley M (2000). Quantitative use of fluorescent in situ hybridization to examine relationships between mycolic acid-containing actinomycetes and foaming in activated sludge plants. Appl. Environ. Microbiol. 66: 115-1166.

EEAA, EPAP (2002). Industrial Wastewater Treatment Plants Inspection Procedures Manual. Egyptian Environmental Affairs Agency and Egyptian Pollution Abatement Project.

El-Shatoury S, Mitchell J, Bahgat M, Dewedar A (2004). Biodiversity of actinomycetes in a constructed wetland for industrial effluent treatment. Actinomycetologica, 18(1): 1- 7.

Fattaa D, Anayiotoub S (2007). Medaware project for wastewater reuse in the Mediterranean countries: An innovative compact biological wastewater treatment system for promoting wastewater reclamation in Cyprus. Desalination, 211: 34-47.

Fritsche W, Hofrichter M (2005). Aerobic degradation of recalcitrant organic compounds by microorganisms. In: Jördening HJ, Winter J (eds) Environmental biotechnology, Concepts and applications. Wiley-VCH, Weinheim, Germany, pp. 203-227.

Guest H (1987). The World of Microbes. Science Tech Publishers, Madison, WI.

Heard J, Harvey E, Johnson BB, Wells JD, Angove MJ (2008). The effect of filamentous bacteria on foam production and stability. Colloid. Surface, 63: 21-26.

Holt JG, Krieg NR, Sneath PHA, Staley JT, Williams ST (1994). Bergey's Manual of Determinative Bacteriology, 9th Edn. Williams and Wilkins, Baltimore, pp. 518-537.

Hozzein WN, Ahmed MB, Abdel Tawab MS (2008). Microbial community structure in a wastewater treatment plant in Beni-Suef City. New Egypt. J. Microb. 20: 189-201.

Hozzein WN, Goodfellow M (2007). *Nonomuraea aegyptia* sp. nov., a novel actinomycete isolated from a sand dune soil sample from Egypt. Antonie van Leeuwenhoek, 92: 165-171.

Hozzein WN, Rabie W, Ali MIA (2011). Screening the Egyptian desert actinomycetes as candidates for new antimicrobial compounds and identification of a new desert *Streptomyces* strain. Afr. J. Biotech. 10(12): 2295-2301.

Johnson LF, Curl EA, Bond JH, Fribourg HA (1959). Methods for studying soil microflora-plant disease relationships, Burgess, Minneapolis.

Kawato N, Shinobu R (1959). On *Streptomyces herbaricolor*, nov. sp. Supplement: A simple technique for the microscopic observation. Mem. Osaka Uni. Liberal Arts Edu. B., Nat. Sci. 8: 114- 119.

Lee S, Basu S, Tyler CW, Wei IW (2004). Ciliate populations as bio-indicators at Deer Island treatment plant. Adv. Env. Res. 8: 371- 378.

Lemmer H (1986). The ecology of scum causing actinomycetes in sewage treatment plants. Water Res. 20: 531-535.

Lemmer H, Baumann M (1988). Scum actinomycetes in sewage treatment plants. Part 3: Synergism with other sludge bacteria. Water Res. 22: 765-767.

Lemmer H, Kroppenstedt RM (1984). Chemotaxonomy and physiology of some actinomycetes isolated from scumming activated sludge. Syst. Appl. Microbiol. 5: 124-135.

Madoni P, Davol D, Gibin G (2000). Survey of filamentous microorganisms from bulking and foaming activated-sludge plants in Italy. Water Res. 34: 1767-1772.

Mameri N, Boudries N, Belhocine D, Lounici H, Grib H, Pauss A (1999). Batch zinc biosorption by a bacterial nonliving *Streptomyces rimosus* biomass. Water Res. 33: 1347-1354

Martin-Cereceda M, Serrano S, Guinea A (1996). A comparative study of ciliated protozoa communities inactivated sludge plants. FEMS Microbiol. Ecol. 21: 267-276.

McCarthy AJ (1987). Lignocellulose-degrading actinomycetes. FEMS Microbiol. Rev. 46: 145-163.

Ōmura S (1986). Philosophy of new drug discovery. Microbiol. Rev. 50: 259-279.

Pridham TG, Anderson P, Foley C, Lindenfelser LA, Hesseltine CW, Benedict RG (1956-57). A selection of media for maintenance and taxonomic study of streptomycetes. Antibiotics Ann. pp. 947- 953.

Puigagut J, Villaseñor J, Salas JJ, Bécares E, García J (2007). Subsurface-flow constructed wetlands in Spain for the sanitation of small communities: a comparative study. Ecol. Eng. 30: 312-319.

Puranik PR, Modak JM, Paknikar KM (1999). A comparative study of the mass transfer kinetics of metal biosorption by microbial biomass. Hydrometallurgy, 52: 189-197.

Roesler JF, Wise RH (1974). Variables to be measured in wastewater treatment plant monitoring and control. J. Water Poll. Cont. Fed. 46(7): 1769-1775.

Saenna P, Gilbreath T, Onpan N, Panbangred W (2011). Actinomycetes community from starch factory wastewater. Res. J. Microbiol. 6: 534-542.

Sag Y, Aksu Z, Kutsal T (1998). A comparative study for the simultaneous biosorption of Cr and Fe on *C. volgaris* and *R. arrhizus*: application of the competitive adsorption models. Process Biochem. 33: 273-281.

Sag Y, Ataçoglu I, Kutsal T (2000). Equilibrium parameters for the single and multicomponent biosorption of Cr (VI) and Fe (III) ions on *R. arrhizus* in a packed column. Hydrometallurgy, 55: 165- 179.

Selatnia A, Boukazoula A, Kechid N, Bakhti MZ, Chergui A, Kerchich Y (2004). Biosorption of lead (II) from aqueous solution by a bacterial dead *Streptomyces rimosus* biomass. Biochem. Eng. J. 19(2): 127-135.

Sim WJ, Lee JW, Oh JE (2010). Occurrence and fate of pharmaceuticals in wastewater treatment plants and rivers in Korea. Environ. Poll. 158: 1938-1947.

Steele DB, Stowers MD (1991). Techniques for selection of industrially important microorganisms. Ann. Rev. Microbiol. 45: 89-106.

Sterrit RM, Lester JN (1988). Microbiology for environmental and public health engineers. E. and F.N. Spon, London.

Veglio F, Beolchini F (1997). Removal of metals by biosorption: a review. Hydrometallurgy, 44: 301- 316.

Volesky B (2001). Detoxification of metal-bearing effluents: biosorption for the next century. Hydrometallurgy, 59: 203-216.

Vymazal J (2002). The use of sub-surface constructed wetlands for wastewater treatment in the Czech Republic: 10 years' experience. Ecol. Eng. 18: 633-646.

Wang JL (2002). Biosorption of copper (II) by chemically modified biomass of *Saccharomyces cerevisiae*. Process Biochem. 37: 847-850.

Weiner RM (1997). Biopolymers from marine prokaryotes. Trends Biotechnol. 15: 390-394.

Williams ST, Cross T (1971). Actinomycetes. In: Booth C (ed.) Methods in microbiology. Academic Press, London, pp. 295-335.

Yang Q, Zhang W, Zhang H, Li Y, Li C (2011). Wastewater treatment by alkali bacteria and dynamics of microbial communities in two bioreactors. Bioresource Technol. 102(4): 3790-3798.

Zouboulis AI, Loukidou MX, Matis KA (2004). Biosorption of toxic metals from aqueous solutions by bacteria strains isolated from metal-polluted soils. Process Biochem. 39: 909-916.