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Mycoflora of Activated Sludge with MBRs in Berlin, Germany

Mohamed F. Awad[a]* & M. Kraume[a]

***Abstract*—**Thirty six samples from each (aerobic and anoxic) activated sludge were collected from two wastewater treatment plants with MBRs in Berlin, Germany. The samples were prepared for count and definition of fungal isolates; these isolates were purified by conventional techniques and identified by microscopic examination. Sixty tow species belonging to 28 genera were isolated from activated sludge samples under aerobic conditions (28 genera and 58 species) and anoxic conditions (26 genera and 52 species). The obtained data show that, *Aspergillus* was found at 94.4% followed by *Penicillium* 61.1 %, *Fusarium* (61.1 %), *Trichoderma* (44.4 %) and *Geotrichum candidum* (41.6 %) species were the most prevalent in all activated sludge samples. The study confirmed that fungi can thrive in activated sludge and sporulation, but isolated in different numbers depending on the effect of aeration system. Some fungal species in our study are saprophytic, and other a pathogenic to plants and animals.

***Keywords*—**Activated sludge, membrane bioreactors, aerobic, anoxic conditions, fungi

I. Introduction

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. The number and type of fungi in sludge depends on various factors namely, the wastewater source, the type of treatment plant, and other environmental factors such as the biological medium offered by the sewage sludge [1].Among the organic substances present in activated sludge are carbohydrates, lignin, fats, soaps, synthetic detergents, proteins and their decomposition products, as well as various natural and synthetic organic chemicals from process industries. Activated sludge (wastewater) with their high organic content is a suitable medium for a large number of microorganisms including some species of fungi [2]. Sewage sludge is valuable source of mineral substance and could be used as fertilizer in agriculture. In the case of natural utilization, knowledge related to mycoflora inhabiting sewage sludge seems to very important, with regard to possibility of introduction to soil not only bacteria and parasites, but also fungi pathogenic for human and animals or even plants. At fertilization or irrigation pathogenic fungi may include in food chain and stay in environment for long time [3].

Mohamed F. Awad* & M. Kraume, Technical University, Department of Process Engineering, Chair of Chemical & Process Engineering MA 5-7, Raum 578, Straße des 17. Juni 135, 10623 Berlin, Germany. (phone: +49 3031472622; fax: +49 3031472756; e-mail: mo_fadl2004@yahoo.com).

There are many conditions, which may increase the health risk of wastewater reuse in agriculture. The first of these conditions is survival time of pathogenic microorganisms. The natural survival time of pathogenic organisms depends on the carrying medium and the environment. The survival time is a time during which pathogens are capable of causing diseases if they came into contact with a host under favorable condition. The second of these conditions are pathogenic bacteria, viruses, protozoa, nematodes and fungi capable of causing diseases which can be found in foods contaminated with sewage water [4], [5]. On the other hand Pathogenic microorganisms can be transferred from raw sewage and secondary effluent during the irrigation process, directly or in directly to the plants, animal and human, also make various infectious diseases. Thus, the present study is conducted on the composition, numbers and incidence of various species of fungi inhabiting activated sludge and effect of aerobic and anoxic conditions on the prevalence of mycoflora.

II. Materials and Methods

Thirty six samples from each aerobic and anoxic activated sludge were taken from wastewater treatment plants with MBRs during the period of nine months (from August/08 to April/09) from tow places of Berlin (Vera in Wedding, and Berliner Wasserbetriebe (BWB) in Margaretenhöhe, Berlin). Samples were put in clean and sterile Boatels sealed and transferred to the laboratory and stored at 4^{o}C, where fugal analysis was made. The media were used for isolation of fungi from activated sludge was 50 % Sucrose Czapek-Dox agar. The media composition was a modified (Sucrose 20.0 g/L; sodium nitrate, 3.0 g/L; potassium chloride, 0.5 g/L; magnesium sulphate, 0.5 g/L; ferrous sulphate, 0.01 g/L; potassium dihydrogen phosphate, 1.0 g/L; agar, 15.0 g/L and distilled water 1000 ml) and Chloramphenicol 50 mg/L, which was used as bacteriostatic agent. All compositions of isolation media were added prior to autoclaving at 121 ^{o}C for 20 minutes, except chloramphenicol, which was sterilized and added to the media after autoclaving. After wards, aliquots of 0.1 ml homogenized activated sludge [6] was put into Petri-dish followed by 20 mL 50 % Sucrose Czapek-Dox agar media (3 replicates). Plates were incubated at 30 ^{o}C for 1-2 weeks to allow for development of pigment on colonies to facilitate complete differentiation of fungal types. Repeated sub-culturing on PDA medium was necessary to obtain pure cultures. Sporulation was induced by subjecting cultures to cultures to ultraviolet light. Isolates were characterized according to morphological features, cultural characteristics such as pigmentation of the mycelium and direction of growth of the hypha, whether aerial or lateral, microscopic

observation of structures involved in asexual reproduction e.g., conidia or spores, and in sexual reproduction, and the presence of fruiting bodies. Light photomicrographs were made mostly from slide cultures. Slide cultures were made by removing a small cylinder of the agar medium by a cork borer, and inserting it on the surface of the same agar inside a Petri-dish. The top cylinder is inoculated with the fungus and covered with a sterilized cover slip. After few days, the fungus growing on the cover slip is gently stained with cotton blue and mounted in lactophenol. Identification was accomplished using appropriate taxonomic techniques, [7]; [8]; [9]; [10]; [11]; [12].

III. Results

A. Fungi recovered from aerobic activated sludge samples

Fifty-eight species representing 28 genera were collected from 36 aerobic samples on 50 % Sucrose Czapek-Dox agar at 30 oC for 1-2 weeks (Table 1). The total count of fungi in aerobic activated sludge ranged between 17-62 colonies/ml activated sludge and the highest count was estimated in sample No. 23. Also, the data in Table 3.6 indicated the *Aspergillus* was the most common genus and was recovered in high frequency of occurrence 94.4 % of samples constituting 21.1 % of total fungi. The count of *Aspergillus* ranged between 1-16 colonies/ ml activated sludge. It was represented by 11 species of which *A. fumigatus* was isolated in high frequency and *A. niger* was isolated in moderate frequency. They emerged in 55.5 % and 41.6 % of samples matching 58.8 % and 44.1 % of total *Aspergillus* and 6.0 % and 4.6 % of total fungi, respectively. *A. flavus* var. *columnaris*, *A. flavus* var. *flavus*, *A. alulaceus*, *A. carneus*, *A. nidulans* (*Emericella nidulan*), *A. oryzae*, *A. terreus* var. *africanus*, *A. terreus* var. *terreus*, and *A. ustus* were isolated in moderated, low or rare frequency. They emerged in 19.4 %, 30.5 %, 5.5 %, 2.7 %, 5.5 %, 11.1 %, 8.3, 2.7, and 5.5 % of samples matching 20.6 %, 32.4 %, 5.9 %, 2.9 %, 5.9 %, 11.7 %, 8.8 %, 2.9 %, and 5.9 % of total *Aspergillus*, respectively.

Data in Table 1 showed that *Fusarium* occupied the second place in the number of cases of isolation and was recovered in high frequency of occurrence 61.1 % of samples constituting 6.2 % of total fungi. Its counts ranged between 1-6 colonies/ml activated sludge. *Fusarium* was represented by 4 species of which *F. dimerum*, *F. oxysporum*, *F. solani*, and *F. roseum*, were isolated in moderate and low frequency and emerged in 27.7 %, 13.8 %, 11.1 %, and 8.3 % of samples matching 45.4 %, 22.7 %, 18.2 %, and 13.63 % of total *Fusarium* and 3.3 %, 1.8 %, 1.0 %, and 0.95 % of total fungi, respectively. *Penicillium* was also common and ranked third according to their total counts. It was encountered in 55.5 % of samples constituting 12.0 % of total fungi. The genus counts ranged between 1-9 colonies/ml activated sludge giving maximum in sample No. 9 (9 colonies). It was represented by 6 species of which *P. chrysogenum* and *P. citrinum*, were isolated in low frequency and emerged in 19.4 % and 22.2 % of samples matching 35.0 % and 40.0 % of total *Penicillium*, respectively. *Penicillium brevicompactum*, *P. corylophilum*, *P. oxalicum*, and *P. roqueforti* were isolated in rare frequency. They emerged in 8.0 %, 2.0 %, 8.0 %, and 2.0 % of samples and 15.0 %, 5.0 %, 5.0 %, and 15.0 % of total *Penicillium*, respectively (Table 1).

Alternaria isolated in moderate frequency and was recovered 36.1 % of samples and represented by 2 species, *A. alternata* and *A. chlamydospora* were recovered from 8.3 %

Table I
Numbers of cases of isolation (out of 36), percentage frequency and occurrence remarks of fungal genera and species recovered from aerobic activated sludge with MBRs on 50 % Sucrose Czapek-Dox agar media at 30°C

Genera and Species	Aerobic activated sludge		
	NCI	%F	OR
Acremonium	6	16.6	L
A. curvulum W. Gams	3	8.3	R
A. strictum W. Gams	4	11.1	R
Alternaria	13	36.1	M
A. alternata (Fr.) Keissl.	3	8.3	R
A. chlamydospora Mouch.	10	27.7	M
Aspergillus	34	94.4	H
A. alutaceus var. alutaceus Berk. & M.A. Curtis	2	5.5	R
A. carneus Blochwitz	1	2.7	R
A. flavus Raper & Fennell var. *columnaris*	7	19.4	R
A. flavus Link var. *flavus*	11	30.5	M
A. fumigatus Fresen.	20	55.5	H
A. nidulans (Emericella nidulan) (Eidam) G. Winter	2	5.5	R
A. niger sensu auct. pro parte, pre	15	41.6	M
A. oryzae (Ahlb.) E. Cohn	4	11.1	R
A. terreus var. *africanus* Fennell & Raper	3	8.3	R
A. terreus var. *terreus* Thom	1	2.7	R
A. ustus (Bainier) Thom & Church	2	5.5	R
Aurobasidium pullulans (de Bary) Arnaud	1	2.7	R
Botryodiplodia theobromae Pat.	1	2.7	R
Chaetomum	6	16.6	L
C. cochliodes Palliser	4	11.1	R
C. globosum Kunze	2	5.5	R
Chrysosporium	5	13.8	L
C. georgii (Vasravsky&Ajello) Oorschot	4	11.1	R
C. tropicum J.W. Carmich.	2	5.5	R
Cladosporium	9	25.0	M
C. cladosporioides (Fresenius) de Vries	4	11.1	R
C. herbarum (Pers.) Link	2	5.5	R
C. oxysporum Berk. & M.A. Curtis	4	11.1	R
Cochliobolus lunatus R.R. Nelson & F.A. Haasis	3	8.3	R
Doratomyces stemonitis (Pers.) F.J. Morton & G. Sm	8	22.2	L
Fusarium	22	61.1	H
F. dimerum Penz.	10	27.7	M
F. oxysporum Schltdl.	5	13.8	L
F. roseum Link	3	8.3	R
F. solani (Mart.) Sacc.	4	11.1	R
Geosmithia lavendula (Raper & Fennell) Pitt	3	8.3	R
Geotrichum candidum Link	5	13.8	L

NCI = Number of cases of isolation (out of 36), % F = Percentage frequency of occurrence (calculated per 36 samples), OR = Occurrence remarks: [H= High occurrence, isolated more than 18 cases (out of 36 samples), M = Moderate occurrence, from 9 to 18 cases, L = Low occurrence, from 5 to 8

TABLE I
CONTINUED

Genera and Species	Aerobic activated sludge		
	NCI	%F	OR
Gibberella fujikuroi var. *fujikuroi* (sawada) Wollenweber.	6	16.6	L
Gliocladium roseum Bainier	3	8.3	L
Mucor circinelloides Tiegh.	7	19.4	L
Paecilomyces	6	16.6	L
P. lilacinus (Thom) Samson	3	8.3	R
P. marquandii (Massee) S. Hughes	-	-	-
P. variotii Bainier	4	11.1	R
Penicillium	20	55.5	H
P. brevicompactum Dierckx	3	8.3	R
P. chrysogenum Thom	7	19.4	L
P. citrinum Thom	8	22.2	L
P. corylophilum Dierckx	1	2.7	R
P. oxalicum Currie & Thom	1	2.7	R
P. roqueforti Thom	3	8.3	R
Phialophora verrucosa Medlar	1	2.7	R
Rhizopus	7	19.4	L
R. arrhizus Fischer	5	13.8	L
R. oryzae Went & Prinsen-Geerligs	2	5.5	R
Scopulariopsis	13	36.1	M
S. asperula (Sacc.) Hughes	7	19.4	L
S. brevicaulis (saccardo) Bainier	6	16.6	L
Setosphaeria rostrata Leonard	1	2.7	R
Stachybotrys	4	11.1	R
S. chartarum (Ehrenberg) Hughes	3	5.5	R
S. elegans (Pidopl.) W. Gams	1	2.7	R
Syncephalastrum racemosum Cohn ex Schöter	3	8.3	R
Trichoderma	12	33.3	M
T. hamatum (Bonorden) Bainier	4	11.1	R
T. koningii Oudemans	5	13.8	L
T. viride Persoon	4	11.1	R
Trichophyton	1	2.7	R
T. ajelloi (Vanbreuseghem)Ajello var. *ajelloi*	1	2.7	R
*Ulocladium chartarum (*Preuss) Simmons	4	11.1	R
Yeasts	10	27.7	M
Number of genera = 28	28		
Number of species = 62	58		

and 27.7 % of samples matching and 23.1 % and 76.9 % of total *Alternaria*, respectively. *Geotrichum candidum* was recovered 36.1 % of samples and 4.9 % of total fungi. *Scopulariopsis* was isolated in moderate frequency and emerged in 36.1 % of the samples and 5.48 % of total fungi and represented by *S. asperula* and *S. brevicaulis* and were recovered from 19.4 % and 8.3 % of samples matching 53.8 % and 46.2 % of total *Scopulariopsis* and 3.1 % and 2.3 % of total fungi, respectively.

Trichoderma was isolated in moderate frequency and emerged in 33.3 % of sample and 6.5 % of total fungi. Three species were identified *T. hamatum* and *T. koningii*, *T. viride* and were recovered from 11.1 %, 13.8 %, and 11.1 % of samples matching 46.6 %, 40.0 % and 26.6 % of total Trichoderma and 1.7 %, 2.0 % and 2.9 % of total fungi, respectively. Unidentified yeasts were recovered from 27.7 % of samples and 6.8 % of total fungi. *Cladosporium* was isolated in moderate frequency and emerged in 25.0 % of samples and 3.39 % of total fungi. Three species were identified *C. cladosporioides*; *C. herbarium* and *C. oxysporum* and were recovered from 11.1 %, 11.1 %, and 5.5 % of samples matching 44.4 %, 44.4 %, and 22.2 % of total *Cladosporium* and 0.5 %, 1.6 %, and 1.3 % of total fungi, respectively.

Doratomyces stemonitis was isolated in low frequency and emerged in 22.2 % of samples and 3.1 % of total fungi. *Rhizopus* was isolated in low frequency and emerged in 19.4 % of samples and 3.1 % of total fungi and represented by *R. arrhizus* and *R. oryzae* were recovered from 13.8 % and 5.5 % of samples matching 80.0 % and 60.0 % of total *Rhizopus*, and 2.3 % and 0.7 % of total fungi, respectively. *Mucor* was represented by *M. circinelloides* and recovered from 19.4 % of samples and 2.6 % of total fungi.

The presented data in Table 1 show that, *Acremonium*; *Chaetomum*; *Gibberella* and *Paecilomyces* were recovered from 16.6 % of samples and 2.3 %, 3.7 %, 2.3 %, and 2.1 % of total fungi, respectively. *Acremonium* was represented by *A. curvulum* and *A. strictum* were recovered from 8.3 % and 11.1 % of samples matching 50.0 % and 66.6 % of total *Acremonium* and 1.3 % and 1.0 % of total fungi, respectively; *Chaetomum* was represented by *C. cochliodes* and *C. globosum*, were recovered from 11.1 % and 5.5 % of all samples matching 66.6 % and 33.3 % of total *Chaetomum*, 2.3 % and 1.3 % of total fungi, respectively, *Gibberella* was represented by *G. fujikuroi* var. *fujikuroi*, and *Paecilomyces* was represented by *P. lilacinus* and *P. variotii* were recovered from 8.3 % and 11.1 % of samples. *Chrysosporium* was recovered from 13.8 % of samples and represented by *C. georgii* and *C. tropicum* and were recovered from 11.1 % and 5.5 % of samples matching 80.0 % and 40.0 % of total *Chrysosporium* and 1.8 % of total fungi, respectively. *Stachybotrys* and *Ulocladium* ware isolated in rare frequency and emerged in 11.1 % of samples and 1.5 % of total fungi; *Stachybotrys* was represented by *S. chartarum* and *S. elegans* and were recovered from 5.5 % and 2.7 % of samples and *Ulocladium* was represented by *U. chartarum.*

Cochliobolus lunatus, *Geosmithia lavendula*, *Gliocladium roseum* and *Syncephalastrum racemosum* and were isolated in rare frequency of occurrence matching collectively 8.3 % of all samples. *Aurobasidium pullulans*; *Botryodiplodia theobromae*; *Phialophora verrucosa*; *Setosphaeria rostrata* and *Trichophyton ajelloi* var. *ajelloi* were isolated in rare frequency of occurrence matching collectively 2.7 % of all samples (Table I).

B. Fungi recovered from anoxic activated smples

Fifty-two species representing 26 genera were collected from 36 samples of each anoxic activated sludge on 50 % Sucrose Czapek-Dox agar at 30 ^{o}C for 1-2 weeks as presented in Table II. The total count of fungi in anoxic activated sludge ranged between 12-58 colonies/ml activated sludge and the highest count was estimated in sample No. 18.

Data in Table 2 illustrated the *Aspergillus* was the most common genus and was recovered in high frequency of occurrence 77.7 % of samples constituting 20.98 % of total fungi. The count of *Aspergillus* ranged between 1-14 colonies/ ml activated sludge. It was represented by 9 species of which *A. niger* was isolated in moderate frequency and *A. flavus* var. *flavus* was isolated in low frequency and were emerged in 27.7 % and 19.5 % of samples matching 35.7 % and 25 % of

TABLE II
NUMBERS OF CASES OF ISOLATION (OUT OF 36), PERCENTAGE FREQUENCY AND OCCURRENCE REMARKS OF FUNGAL GENERA AND SPECIES RECOVERED FROM ANOXIC ACTIVATED SLUDGE WITH MBRS FOR 50 % SUCROSE CZAPEK-DOX AGAR MEDIA AT 30 °C

Genera and Species	Anoxic activated sludge		
	NCI	%F	OR
Acremonium	2	5.5	R
A. curvulum W. Gams	1	2.7	R
A. strictum W. Gams	1	2.7	R
Alternaria	5	13.8	L
A. alternata (Fr.) Keissl.	1	2.7	R
A. brassicae (Berk.) Sacc.	2	5.5	R
A. chlamydospora Mouch.	5	13.8	L
Aspergillus	28	77.7	H
A. fischerianus Samson & W. Gams	1	2.7	R
A. flavus var. *columnaris* Raper & Fennell	5	13.8	L
A. flavus var. *flavus* Link	7	19.5	L
A. fumigatus Fresen.	5	13.8	L
A. niger sensu auct. pro parte, pre	10	27.7	M
A. oryzae (Ahlb.) E. Cohn	1	2.7	R
A. terreus var. *africanus* Fennell & Raper	1	2.7	R
A. terreus var. *terreus* Thom	1	2.7	R
Botryodiplodia theobromae Pat.	2	5.5	R
Chaetomum	3	8.3	R
C. cochliodes Palliser	3	8.3	R
Chrysosporium	4	11.1	R
C. georgii (Vasravsky&Ajello) Oorschot	2	5.5	R
C. tropicum J.W. Carmich.	3	8.3	R
Cladosporium	4	11.1	R
C. cladosporioides (Fresenius) de Vries	3	8.3	R
C. oxysporum Berk. & M.A. Curtis	1	2.7	R
Cochliobolus lunatus R.R. Nelson & F.A. Haasis	1	2.7	R
Doratomyces stemonitis (Pers.) F.J. Morton & G. Sm	5	13.8	L
Fusarium	20	55.5	H
F. dimerum Penz.	7	19.4	L
F. oxysporum Schltdl.	11	30.5	M
F. solani (Mart.) Sacc.	4	11.1	R
Geosmithia lavendula (Raper & Fennell) Pitt	1	2.7	R
Geotrichum candidum Link	7	19.4	L

total *Aspergillus* and 8.2 % and 3.9 % of total fungi, respectively.

Penicillium occupied the second place in the number of cases of isolation and was recovered from 61.1 % of samples constituting 13.7 % of total fungi. *Penicillium* was represented by 5 species of which *P. chrysogenum*, *P. citrinum*, and *P. oxalicum* were isolated in low frequency and emerged in 19.4 % and 13.8% of anoxic samples matching 31.8 % and 22.7 % of total *Penicillium* and 3.6 %, 5.2 % and 2.3 % of total fungi, respectively (Table 2).

Fusarium occupied the third place in the number of cases of isolation and was recovered in high frequency of occurrence 55.5 % of samples constituting 10.2 % of total fungi. It was represented by 3 species of which *F. dimerum*, *F. oxysporum*, and *F. solani* were isolated in moderate and low frequency and emerged in 19.4 %, 30.5 % and 11.1 % of samples matching 35.0 %, 55.0 % and 20.0 % of total *Fusarium* and 3.6 %, 4.2 % and 2.3 % of total fungi, respectively.

Also, Data in Table 2 indicated the *Trichoderma* occupied the fourth place in the number of cases of isolation and was recovered from 44.4 % of samples constituting 8.8 % of total fungi. *Trichoderma* was represented by 2 species *T. koningii* and *T. viride* were recovered from 22.2 % and 25.0 % of all anoxic samples matching 50.0 % and 56.0 % of total Trichoderma and 3.6 % and 5.2 % of total fungi, respectively. Unidentified *yeasts* were isolated in moderate frequency and recovered 25.0 % of samples and 5.6 % of total fungi.

Gibberella fujikuroi var. *fujikuroi* was isolated in low frequency and emerged in 22.2 % of samples constituting 4.2 % of total fungi. *Geotrichum candidum* was isolated in low frequency and emerged in 19.4 % of samples constituting 5.9 % of total fungi.

Paecilomyces was recovered 16.6 % of samples constituting 3.2 % of total fungi. It was represented by *P. lilacinus*,*P. marquandii*, and *P. variotii* were recovered from 5.5 %, 5.5 %, and 13.8 % of samples matching 22.2 %, 22.2 %, and 55.5 % of total *Paecilomyces* and 0.6 %, 0.9 % and 1.6 % of total fungi, respectively. *Alternaria* (represented by *A. alternata*, *A. brassice*, and *A. chlamydospora*), *Doratomyces stemonitis*, *Mucor circinelloides* and *Rhizopus* (represented by *R. arrhizus* and *R. oryzae*) ware isolated in low frequency of occurrence matching collectively 13.8 % of samples and 3.0 %, 2.3 %, 2.3 % and 3.0 % of total fungi, respectively.

TABLE II
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Genera and Species	Anoxic activated sludge		
	NCI	%F	OR
Gibberella fujikuroi var. fujikuroi (sawada) Wollenweber.	8	22.2	L
Mucor circinelloides Tiegh.	5	13.8	L
Paecilomyces	9	25.0	M
P. lilacinus (Thom) Samson	2	5.5	R
P. marquandii (Massee) S. Hughes	2	5.5	R
P. variotii Bainier	5	13.8	L
Penicillium	22	61.1	H
P. brevicompactum Dierckx	2	5.5	R
P. chrysogenum Thom	7	19.4	L
P. citrinum Thom	7	19.4	L
P. corylophilum Dierckx	1	2.7	R
P. oxalicum Currie & Thom	5	13.8	L
Phialophora verrucosa Medlar	3	8.3	R
Rhizopus	5	13.8	L
R. arrhizus Fischer	3	8.3	R
R. oryzae Went & Prinsen-Geerligs	2	5.5	R
Scopulariopsis	7	19.4	L
S. asperula (Sacc.) Hughes	3	8.3	R
S. brevicaulis (saccardo) Bainier	4	11.1	R
Setosphaeria rostrata Leonard	3	8.3	R
Stachybotrys	3	8.3	R
S. chartarum (Ehrenberg) Hughes	3	8.3	R
S. elegans (Pidopl.) W. Gams	1	2.7	R
Syncephalastrum racemosum Cohn ex Schöter	1	2.7	R
Trichoderma	16	44.4	M
T. koningii Oudemans	8	22.2	L
T. viride Persoon	9	25.0	M
Trichophyton	3	8.3	R
T. ajelloi (Vanbreuseghem) Ajello var. *ajelloi*	1	2.7	R
T. terrestre Durie & Frey	2	5.5	R
Ulocladium chartarum (Preuss) Simmons	1	2.7	R
Yeasts	9	25.0	M
Number of genera = 28	26		
Number of species = 62	52		

Chrysosporium (represented by *C. tropicum* and *C. georgii*) and *Cladosporium* (represented by *C. cladosporioides* and *C. oxysporum*) were isolated in low frequency and emerged in 11.1 % of samples and 2.3 % and 1.6 % of total fungi, respectively.

Chaetomum cochliodes, *Phialophora verrucosa*, *Setosphaeria rostrata*, *Stachybotrys* (represented by *S. chartarum* and *S. elegans*) and *Trichophyton terrestre*, were isolated in rare frequency and emerged in 8.3 % of anoxic samples. *Acremonium* (represented by *A. curvulum* and *A. strictum*) and *Botryodiplodia theobromae* were isolated in rare frequency and emerged in 5.5 % of the anoxic samples.

Cochliobolus lunatus, *Geosthmithia lavendula*, *Syncephalastrum racemosum*, and *Ulocladium chartarum* were isolated in rare frequency and emerged in 2.7 % of anoxic samples (Table II).

IV. Discussion

The results indicate that activated sludge is habitat for the growth and sporulation of different groups of fungi, both saprophytic, pathogenic and some of these fungi also produce mycotoxins. A variety of types of filamentous fungi and like-yeasts was obtained from aerobic and anoxic activated sludge.The obtained data showed some different are found for most fungal spices among the aerobic and anoxic condition this depend on aeration system. Generally, the fungal diversity present in both types of activated sludge has been some similar, with different spore population. Although, the chance of presence fungal spore in aerobic was better than anoxic activated sludge. This trend could be explained by the continuous turning process of sludge and transfer it from the aerobic to anoxic tank. Some fungi were recovered only from aerobic activated sludge (*Alternaria brassicae*, *Aspergillus alulaceus*, *A. carneus*, *A. ustus*, *Aurobasidium pullulans*, *Chaetomum globosum*, *Cladosporium herbarium*, *Emericella nidulans var. nidulans*, *F. roseum*, *Gliocladium roseum* and *Penicillium roqueforti*). Also, *Aspergillus fischerianus*, *Paecilomyces marquandii* and *Trichophyton terrestre* were encountered only from anoxic activated sludge (Table 1 and 2). These results agree to some extent with the findings reported by [1]-[3]; [5]-[6]; [13]-[18]. Most of fungal isolates in our study are will know a pathogenic or potentially pathogenic [7]; [14]-[15]; [18]; [20]-[22].

V. Conclusions

Activated sludge produced from wastewater treatment plants with membrane bioreactors is rich in moulds and other dermatophyte. Most fungi recovered in the present investigation can be considered as potential pathogens and some of these fungi also produce mycotoxins such as *Alternaria*, *Aspergillus*, *Chrysosporium Fusarium*, *Geotrichum*, *Paecilomyces*, *Scopulariopsis*, *Stachybotrys* and *Trichophyton*. Therefore, all workers in the field of activated sludge process, wastewater treatment and farm operation should be careful to avoid mycotic infections and the productions must be adapted to control the spread of pathogenic fungi in the environment.

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