

Fungal Species, Go-T Enzyme and Mycotoxins Isolated from Decayed Sugarcane (*Saccharrum Officinarum*) from Qena Governorate, Egypt

الأنواع الفطرية والإنزيمات الناقلة والسموم الفطرية المعزولة من نباتات القصب المتحللة من محافظة قنا، مصر

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Abstract: Seventy-three fungal species belonging to 43 genera were isolated from 40 samples of *Saccharrum officinarum* (collected from Naage-Hamadi canal in Qena Governorate) on 10 g/l glucose (20 species, 15 genera) in Winter and (23/15) in Summer and on 40 g/l sucrose (42/28) in Winter and (27/18) in Summer on Czapek's agar media at 25°C. *Aspergillus*, *Trichoderma*, *Mucor* and *Pythium* were the most common genera on the two isolation media. The dominant species of *Aspergillus* were *A. niger*, *A. flavus*, *A. ustus*, *A. terreus* and *A. wentii*. Some species were dominant on 40 g/l sucrose such as *Aspergillus niger*, *A. flavus*, *Emericella nidulans*, *Trichoderma viride*, *Torula herbarum* and *Mamaria echinoeotryoides*, while the dominant species on 10 g/l glucose were *Mucor circinelloides*, *Aspergillus niger*, *Torula herbarum* and *Trichoderma viride*. Mycotoxins including aflatoxins B1, B2, G1 and G2, zearalenone and diacetoxyscirpenol were detected in the examined samples of *Saccharrum officinarum*. The mycelial growth of *Aspergillus flavus*, *A. niger*, *Fusarium moniliforme* and *Torula harbarum* decreased with the increase in Dimethoate concentrations, although 25 ppm was less effective than the higher levels of the insecticide (75 - 200 ppm). Dimethoate stimulated the activity of Go-T in *Aspergillus niger*, *Fusarium moniliforme* and *Torula harbarum*, while the Go-T activity inhibited in *A. flavus* with the Dimethoate treatments.

Keywords: Egypt, Qena, Sugarcane, Fungi, aquatic and terrestrial fungi, *Saccharrum officinarum*, mycotoxins, enzymes.

المستخلص: تم عزل ثلاثة وسبعين نوع من الفطريات تابعة لثلاثة وأربعين جنس ، من 40 عينة قصب تم جمعها من قناة نجع حمادي بمحافظة قنا على بيئة شابكس المحتوية على 10 جم/لتر جلوكوز 20 نوع ، 15 جنس منها في الشتاء و23 نوع ، 15 جنس في الصيف والباقي على بيئة شابكس المحتوية على 40 جم/لتر سكروز 42 نوع ، 28 جنس في الشتاء و27 نوع ، 18 جنس في الصيف. كما وجد أن اسبرجلس ، ترايكوديرما ، ميوكور، بيثيوم اكثر الأجناس شيوعا على كلا بيئتي العزل. وأن الأنواع السائدة من هذا الجنس كانت اسبرجلس فلافس ، اسبرجلس استس ، اسبرجلس تيريس ، اسبرجلس ونتاجي. كما وجد أن بعض الفطريات كانت سائدة على 40 جم/لتر سكروز مثل اسبرجلس نيجر ، اسبرجلس فلافس ، اميرسيلا نديولانس ، ترايكوديرما فيريدي ، توريولا هربارم ، ماماريا ايشينوتريودز. بينما كانت الأنواع الفطرية ميوكور سيرسينولويدز ، اسبرجلس نيجر ، توريولا هربارم ، ترايكوديرما فيريدي سائدة على بيئة شابكس المحتوية على 10 جم/لتر جلوكوز. وأظهرت الدراسة وجود السموم الفطرية افلاتوكسين ب1 ، ب2 ، ج1 ، ج2 ، زيرالينون ، داي اسيتو اوكسيسكيريبيول في مستخلص العينات المختبرة من قصب السكر. وجد أيضا أن نمو ميسليوم اسبرجلس فلافس ، اسبرجلس نيجر ، فيوزاريم مونيليفورم ، توريولا هربارم ينخفض بزيادة تركيزات المبيد الحشري دايميثويت ، علي الرغم من أن التركيز 25 جزء في المليون كان أقل تأثيرا من التركيزات الأعلى من المبيد (75 - 200 جزء في المليون). وجد أن المبيد زددايميثويتز حفز نشاط أنزيم Go-T في فطريات ، اسبرجلس نيجر ، فيوزاريم مونيليفورم ، توريولا هربارم. بينما أدت معاملات المبيد إلى تثبيط نشاط أنزيم Go-T في فطره اسبرجلس فلافس.

كلمات مدخلة: مصر، قنا، قصب السكر، فطريات، إنزيمات ناقلة، سموم

Introduction

Sugarcane plant (*Saccharum officinarum*) is the predominant crop used in sugar production in Upper Egypt. Sugar present in the stem of *Saccharum officinarum* represents the main source for fungal growth. Accordingly, the fallen stems in canal or irrigation water during the harvest, become a source of pollution for the water and other cultivated plants. The pollution effects depend on the mycoflora grown on the stem and on some environmental factors such as temperature. Mohawed, *et al.* (2001) studied the seasonal fluctuations of soil and root

surface fungi of sugarcane (*Saccharum officinarum*) in Upper Egypt. They isolated 73 species and 5 varieties representing 33 genera using glucose-, cellulose- and sucrose-Czapek's agar media. Abdelraheem (1999) examined the presence of fresh-water ascomycetes in various decayed plant parts of *Eucalyptus rostrata*, *Phragmites australis* and *Phoenix dactylifera*, collected from the River Nile in Egypt. In addition, El-Sharouny, *et al.* (1999) studied the biodiversity and distribution of fungi on submerged wood in the River Nile and irrigation canals in Upper Egypt.

Some moulds can produce toxic metabolites (mycotoxins) and therefore proliferation of the organisms represents a potential health hazard (Northolt, *et al.* 1995). Therefore, detection of fungal contaminants is essential to ensure safe and high quality food (Bullerman, 1979).

On the other hand, insecticides have been used to control insects and play a significant role in increasing crop production. They commonly affect some of the non-target organisms such as microbial population ranging from inhibitory to stimulatory effects. Dimethoate is a broad-spectrum insecticide commonly used on sugarcane to control the white fly (Anonymous, 1989). The effect of insecticides on fungal growth and enzyme activity has been studied by different workers (Audus, 1960; El-Hissy and Abdel Kader, 1980 and Abd-Elaah, 1993).

Materials and Methods

Forty samples of sugarcane stems (*Saccharum officinarum*) were collected during two seasons (Winter and Summer 2002) from the Naage-Hamadi canal in Qena Governorate in Upper Egypt. Each sample was represented by 10 decayed stem parts. The samples were transferred directly to the laboratory for fungal isolation and toxin analyses.

Isolation of fungi from Saccharum officinarum

The stem-samples were washed with sterilized distilled water. Each stem was cut into segments (ca. 0.5 cm long) by knife and then each segment was cut into four equal parts. These segments were placed on the surface of two solidified media, glucose (10 g/l) and sucrose (40 g/l) Czapek's agar to which chloromphenicol was added as a bacteriostatic agent (Smith and Dawson, 1944). Five Petri plates of tested medium were used for each stem sample. Plates were incubated at 25°C for one week. For the recovery of aquatic fungi, stem segments from the collected samples were placed in Petri dishes 12 cm in diameter (6 replicates). The segments in each Petri-dish were then covered with sterile distilled water (20 ml) and 12 sterilized sesame seeds were introduced into each Petri-dish, as employed by El-Nagdy (1986).

The growing fungi were identified, counted as numbers per segment. The identification of fungal genera and species was performed according to Raper and Thom (1949), Gilman (1957), Raper and Fennell (1965), Domsch and Gams (1972), Booth (1971), Pratt and Heather (1973) and Lund (1978). Fungal species recovered from stem samples were

purified on suitable media such as glucose-peptone-agar, malt-extract-agar, potato-dextrose-agar, potato-dextrose-yeast-agar, sabouraud's-dextrose-agar and Czapek's-medium.

Extraction of toxins from Saccharum officinarum

Fifty grams of decayed stems of each sugarcane sample were transferred to 500 ml Erlenmeyer flasks containing 150 ml of chloroform each and placed in a shaker (200 r.p.m.) for 16 hours, then filtered through filter paper (Whatman No.1). The chloroform extract was dried over anhydrous sodium sulphate. The remaining stem samples were dried at 50°C over night, followed by re-extraction by 150 ml of 90% methanol-water.

Chemical detection of mycotoxins

Thin layer chromatography technique (TLC) was carried out on plates using precoated with Silica Gel type 60, F254 (MERCK, Germany). Aflatoxins B1, B2, G1 & G2; ochratoxins A & B; sterigmatocystin; citrinin; T-2 toxin; diacetoxyscirpenol (DAS); zearalenone; moniliformin and fusarin C were used as standards. The developing solvent systems used were methanol-chloroform (v/v 3:97), ethyl acetate-hexane(v/v, 70:30), ethanol-chloroform (v/v, 5:95) and toluene-acetone-methanol (v/v/v, 50:30:20). The developed plates were then viewed under UV light (254 and/or 366nm) and sprayed with reagents for identification according to Gimeno (1976) and Vesonder (1986).

Determination the effect of Dimethoate insecticide on mycelial growth and Go-T activity

The effect of the insecticide Dimethoate (O,O-dimethylS-methylcarbamoymethyl phosphorodithioate, IUPAC) on mycelial growth and Go-T (glutamic-oxaloacetic transaminase) activity were studied, using the most commonly occurring fungal species, namely *Aspergillus flavus*, *A. niger*, *Fusarium moniliforme* and *Torula harbarum*.

Fifteen Erlenmeyer flasks (250 ml) each containing 50 ml Czapek's-Dox liquid medium were used for each fungus. Triplicate flasks served as control, in which media were amended with 25, 75, 150 or 200 ppm of the insecticide. Each flask was inoculated with 1 ml of the spore suspension obtained from seven day old cultures (Czapek's-Dox medium) of the required fungus. The flasks were then incubated at 27°C for seven days, after

which the produced mycelial filtrates were collected from the flasks by Buchner filtration using hardened filter papers, washed several times with sterile distilled water and weighed.

Determination of Go-T: ten milligrams of the fresh mycelium were mixed and homogenized with 1.0 ml phosphate buffer. The extracts were clarified by centrifugation for 15 min at 8000 Xg and then analyzed for Go-T.

Results and Discussion

The total fungi isolates obtained from 40 decayed sugarcane (*Saccharum officinarum*) samples on 10g/l glucose and 40g/l sucrose Czapek's agar media were 1151 and 1116 colonies for all samples, respectively (Table 1). These results showed that counts of fungi were greater on 10 g/l glucose medium than 40 g/l sucrose medium. However, the number of species isolated on sucrose medium was greater than that recovered on glucose medium. In this respect, 26 species belonging to 18 genera were collected from the 40 samples of sugarcane on glucose-medium, while thirty-eight species belonging to twenty-six genera were collected from all tested samples on the sucrose-medium.

A total of 48 fungal species belonging to 32 genera were isolated from the 40 samples of sugarcane on both glucose- and sucrose-media during Winter and Summer seasons (Table 1). Sixteen out of these species were isolated on both tested media while 10 species were isolated only on glucose medium and 22 species were isolated only on sucrose medium. The dominant genera on the two types of media were *Aspergillus*, *Trichoderma*, *Mucor*, *Mammaria*, *Torula* and *Cephalosporium*

Fungi recovered on glucose-agar medium

Twenty-six species belonging to eighteen genera were recovered from 40 decayed sugarcane samples from canal water on glucose-Czapek's agar at 25°C (Table 1).

Aspergillus was the dominant genus representing 95% of the samples constituting 45.78% of the total count of fungi. It was represented by 7 species of which *A. flavus* (8.78), *A. niger* (30.41), and *A. ustus* (4.42) were of high occurrence. These *Aspergillus* species were also recovered from sugarcane leaves, stem, bagasse and juice by Higgy, *et al.* (1977), Sandhu and Sidhu (1980), Sandhu, *et al.* (1980), Olufolaji (1986), Sivanesan and Waller (1986), Muhsin and Abdel-Kader (1995), and Abdel-Hafez, *et al.* (1995).

The remaining *Aspergillus* species were of moderate to rare occurrence on 20-5% of the samples. These species namely, *A. awamori* (1.12% of the total count of fungi), *A. terreus* (0.56%), *A. wentii* (0.35%) and *A. japonicus* (0.14%).

Trichoderma, *Emericella*, and *Torula* were second in occurrence and frequency. These were recovered from 85%, 75% and 90% of the tested samples and represented by the high occurrence of 19.38%, 11.87% and 7.94% of the total fungal counts, respectively. Each was represented by one species, namely *Trichoderma viride*, *Emericella nidulans* and *Torula herbarum*.

Colletotrichum dematium was of high occurrence, appearing in 45% of the sugarcane samples and constituting 3.93% of the total fungal counts; this agrees with Brinker and Seigler, (1991).

Eurotium chevaliere, *Mucor heimalis*, *Pythium intermedium* and *Pilobolus* sp. were of moderate occurrence (40%, 40%, 30% and 35% of the samples) and constituted 1.47%, 1.40%, 1.33% and 1.83% of the total fungi, respectively. Each of *Gliomastix cerealis* and *Melanospora fallax* were of low occurrence (20% of the samples), matching 0.98% and 0.63% of total fungi, respectively.

The remaining species were of rare occurrence (5 -10% of the samples) constituting 0.70-0.14% of the total fungi. These species were *Apodachlya brachynema*, *Achlya megasperma*, *A. Americana*, *Allomyces macrogynous*, *Curvularia tetramera*, *Cunninghamella elegans*, *Monciliium mucidum*, *Synecephalostrum racemosum* and *Rhizopous stolonifer*. Several researches have reported that some strains of these fungi produced several toxic metabolites (Debeaupuis and La-Font, 1985; Charles, *et al.* 1979; Stinson, 1985; Megalla, *et al.* 1985 and Leitao, *et al.* 1989).

Fungal genera and species recovered on 40 g/l sucrose agar

Thirty-eight species belonging to twenty-six genera were recovered from 40 sugarcane samples on 40g/l sucrose-Czapek's agar at 25°C (Table 1). Abdel-Hafez, *et al.* (1995) isolated 46 species and 2 varieties belonging to 20 genera from 50 sugarcane juice samples on glucose-, sucrose- and cellulose-Czapek's agar at 28°C. The dominant genera were *Aspergillus* (8 species), *Trichoderma* (1 species), *Mucor* (4 species), *Torula* (2 species) and *Cladosporium* (1 species). They occurred in samples at rates of 0.5-85% of the total samples investigated. In this respect, Abdel-Sater and Sabah Saber (1999) recorded that *Aspergillus*, *Eurotium* and *Penicillium* were the most common genera in

Table (1): Fungi isolated from sugarcane on 10 g/l glucose and 40 g/l sucrose Czapeck's agar medium, total counts (TC), percentage of total counts (TC%), frequency of occurrence (F%), and season.

Genera and species	10 g/l Glucose-medium				40 g/l Sucrose-medium			
	TC	TC %	F %	Season*	TC	TC %	F %	Season*
<i>Aspergillus</i>								
<i>A. awamori</i> Nakazawa (Usami)	16	1.12	20.0	W&S	28	2.50	40.0	W&S
<i>A. niger</i> Van Tieghem	433	30.41	95.0	W&S	286	25.58	85.0	W&S
<i>A. ustus</i> (Bain.) Thom & Church	63	4.42	70.0	W&S	8	0.72	20.0	W
<i>A. flavus</i> Link	125	8.78	80.0	W&S	61	5.46	60.0	W
<i>A. versicolor</i> (vuill.) Tiraboschi					19	1.70	20.0	W&S
<i>A. oryzae</i> (Ahlb) Cohn					4	0.36	10.0	W
<i>A. terreus</i> Thom	8	0.56	15.0	W	6	0.54	20.0	W
<i>A. terricola</i> Marchal					2	0.18	5.0	W
<i>A. wentii</i> wehmer	5	0.35	10.0	W				
<i>A. japonicus</i> Saito	2	0.14	5.0	S				
<i>Apodachlya brachynema</i> (Hildebrand)	9	0.63	10.0	W				
<i>Achlya megasperma</i> (Humphrey)	4	0.28	10.0	S	2	0.18	5.0	S
<i>Achlya americana</i> (Humphrey)	2	0.14	5.0	S				
<i>Allomyces macrogynous</i> (Emerson & Willson)	5	0.35	10.0	S				
<i>Acremonium furcatum</i> F.et V.Moreau					2	0.18	5.0	W
<i>Curvularia tetramera</i> (Mckinney) Boedijn	2	0.14	5.0	S	21	1.88	15.0	W&S
<i>Cunninghamella elegans</i> Lendner	8	0.56	10.0	W	17	1.52	30.0	S
<i>Cladosporium herbarum</i> Link ex Fr.		2			8	2.50	30.0	S
<i>Colletotrichum dematium</i> (Pers.ex Fr.)	56	3.93	45.0	S	6	0.54	10.0	W
<i>Cephalosporium curtipes</i> (Saccardo)					16	1.43	25.0	W&S
<i>Emericella nidulans</i> (Edam.) Vuill	169	11.87	75.0	W&S	28	2.50	25.0	W&S
<i>Eurotium chevaliere</i> Mangin	21	1.47	40.0	W				
<i>Fusarium moniliforme</i> Shled.					14	1.25	25.0	W
<i>Mammaria echinoeotryoides</i> Cesati		2			9	2.59	40.0	W
<i>Mortierella polycephala</i> Coemans			-		2	0.18	5.0	W&S
<i>Melanospora fallax</i> Zukal	9	0.63	20.0	W	4	0.36	5.0	W
<i>Moncillium mucidum</i> W. Gams	6	0.42	10.0	S				
<i>Mucor circinelloides</i> Van Tiegh					23	2.06	20.0	S
<i>M. heimalis</i> Wehmer	20	1.40	40.0	W&S	83	7.42	50.0	S
<i>M. plumbeus</i> Bon.					2	0.18	5.0	W
<i>M. racemosus</i> Fres.					85	7.60	70.0	S
<i>Gliomastix cerealis</i> (Kart.) Dickinson	14	0.98	20.0	W				
<i>Gymnoascus reessii</i> Baran.					2	0.18	5.0	W
<i>Torula herbarum</i> Link ex Fr.	113	7.94	90.0	W&S	55	4.92	20.0	W
<i>T. grisea</i> Szilvinyi					7	0.63	10.0	W
<i>Trichoderma viride</i> Pers.ex Fr.	276	19.38	85.0	W&S	180	16.10	80.0	W&S
<i>Stemphylium piriforme</i> Wallroth		1			0	0.89	10.0	S
<i>Syenecephalostrum racemosum</i> (Schroeter)	10	0.70	10.0	W				
<i>Saccharomyces</i> spp.					39	3.49	30.0	W&S
<i>Penicillium luteum</i> (Zukal)	1				2	1.07	20.0	S
<i>Pilobolus</i> sp Van Tieghem.	26	1.83	35.0	W&S				
<i>Pythium intermedium</i> (de Bary)	19	1.33	30.0	W&S	16	1.43	20.0	W
<i>Py. aphanidermatum</i> (Drechsler & water house)					2	0.18	5.0	W
<i>Verticillium tenerum</i> Link					4	0.36	10.0	S
<i>Rhizopous stolonifer</i> (Fhrenb) Lindat	3	0.21	5.0	W	5	0.45	10.0	W
<i>Humicola fusco-atra</i> Traaen					4	0.36	5.0	W
<i>H. grisea</i> Traaen					4	0.36	5.0	W
<i>Helminthosporium sativum</i> (Pammel, King and Bakke					2	0.18	5.0	S

* S = summer and W = winter.

Table (2): Visual estimation of mycotoxins in sugarcane samples.

Fungal species	Toxin production					
	Aflatoxins				Zearalenon	Diacetoxy-scripenol
	B1	B2	G1	G2		
<i>Fusarium moniliforme</i>	-	-	-	-	+	+
<i>Aspergillus flavus</i>	+	+	+	+	-	-
<i>A. niger</i>	-	-	-	-	-	-
<i>A. ustus</i>	-	-	-	-	-	-
<i>Emericella nidulans</i>	-	-	-	-	-	-
<i>Colletotrichum dematium</i>	-	-	-	-	-	-

dried raisins, dates and figs, using 20% sucrose-Czapek's agar at 28°C. These results nearly agree with the findings of Abdel-Sater and Ismail (1993), Megalla, *et al.* (1985), Ismail (1993) and Aran and Eke (1987) who noted that *Aspergillus* and *Penicillium* were the most common in Egyptian and Turkish foodstuffs, respectively. Of the *Aspergillus*, the most dominant species were *Aspergillus awamori*, *A. niger*, *A. ustus*, *A. flavus*, *A. versicolor*, *A. oryzae* and *A. terreus*.

Trichoderma (80% of the samples) was second to *Aspergillus* and was represented by one species namely, *T. viride* which constitutes 16.10% of the total count of the isolates. *Mucor* came third and it was represented by four species namely, *M. circinelloides*, *M. heimalis*, *M. plumbeus*, and *M. racemosus* constituting 0.18-7.60% of the total fungi recovered (Table 1). *Torula* was represented by two species namely, *T. herbarum* and *T. grisea* constituting 4.93% and 0.63% of the total fungi recovered, respectively. *Cladosporium herbarum*, *Cunninghamella elegans*, *Saccaromyces* spp., *Fusarium moniliforme* and *Cephalosporium curtipes* were of moderate occurrence, they comprised 1.25-3.49% of the total fungi recovered. The following genera, namely *Curvularia tetramera*, *Mammaria echinoeotryoides*, *Penicillium luteum* and *Pythium intermedium* were represented by low occurrence, and constituting 1.08-2.30% of the total fungi recovered. The remaining genera and species were represented by rare occurrence (5-10%); with a frequency of 0.07-0.83%. *Saccharomyces* spp. appeared only on the sucrose medium.

Seasonal fluctuation of the fungal species

The results given in Table 1 showed that 26 species appeared on glucose medium in both Summer and Winter seasons. The seasonal fluctuation of these species on glucose medium revealed that 9 species appeared in Winter, 7 species

in Summer and 10 species in both seasons. On the other hand, 38 fungal species were recovered on sucrose medium in both Summer and Winter seasons. 19 of these were isolated in Winter, 10 species in Summer and 9 species were isolated in both seasons on sucrose medium. Generally, 48 fungal species were recovered from the two seasons on the two tested media. Nineteen out of these species were isolated only in Winter and 12 species were isolated only in Summer, while 17 species were isolated in both seasons. Seasonal fluctuations of fungi were also studied by El-Hissy (1979); El-Hissy, *et al.* (1982) and Steciow (1998).

Mycotoxins

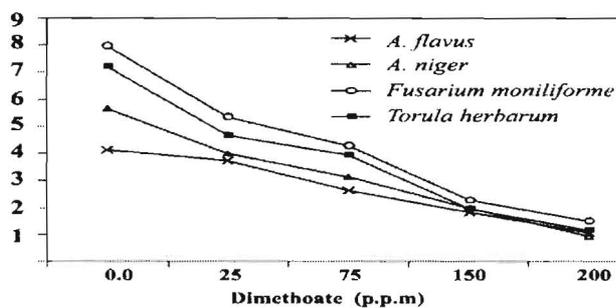
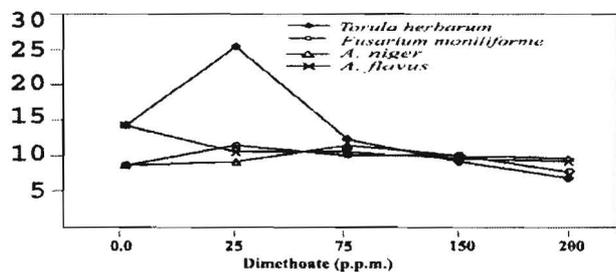
Fusarium moniliforme produced zearalenone and diacetoxyscirpenol toxins (Table 2). These results are in agreement with those of Basch and Mircua (1992). *Fusarium* has been recorded as zearalenone producer in Egypt (El-Maraghy, 1984, El-Kady & El-Maraghy, 1982; El-Maghraby & El-Maraghy, 1988; El-Maghraby, *et al.* 1995).

Thin layer chromatographic analysis revealed the significant amounts of aflatoxins B1, B2, G1 and G2 in the tested samples produced by *A. flavus* (Table 2). These aflatoxins, particularly B1, are associated with acute poisoning of animals and humans (Jukes, 1978), causing lack of appetite, weight loss, unthriftiness, neurological abnormalities, jaundice of mucous membrane, convulsions and death (Harwig & Munro, 1975), causing damage of chromosomes (El-Zawahri *et al.*, 1977) and being carcinogenic for the human liver (Smith & Moss, 1985).

Table (3): Mycelial fresh weight (mg) and GO-T of *Aspergillus flavus*, *A. niger*, *Fusarium moniliforme* and *Torula herbarum* in the presence of different concentrations of the pesticide Dimethoate.

Conc. (ppm)	<i>A. flavus</i>		<i>A. niger</i>		<i>Fusarium moniliforme</i>		<i>Torula herbarum</i>	
	Fresh weight (mg)	GOT μ /l	Fresh weight (mg)	GOT μ /l	Fresh weight (mg)	GOT μ /l	Fresh weight (mg)	GOT μ /l
0.0	4.14	14.0	5.67	8.0	7.98	8.0	7.2	14.0
25	3.75	10.0**	4.01	8.5	5.35*	11.0**	4.69*	26.0**
75	2.62**	10.0**	3.14**	11.0**	4.30**	9.5*	3.95**	12.0
150	1.82**	9.0**	1.97**	9.5**	2.26**	9.5*	1.95**	8.5*
200	1.06**	8.5**	0.94**	9.0	1.52**	7.0	1.14**	6.0**
LSD 0.05	1.129	1.900	1.602	1.009	2.257	1.351	2.096	5.286
0.01	1.481	2.492	2.101	1.323	2.960	1.771	2.748	6.931

*, **: Significant and highly significant values as compared with the control treatment.

**Fig. (1):** The effect of Dimethoate on mycelial growth of *Aspergillus flavus*, *A. niger*, *Fusarium moniliforme* and *Torula herbarum*.**Fig. (2):** The effect of Dimethoate on Go-T continent of *Aspergillus flavus*, *A. niger*, *Fusarium moniliforme* and *Torula herbarum*.growth of *Aspergillus flavus*, *A. niger*, *Fusarium moniliforme* and *Torula herbarum*.

Based on visual estimation, when the sugarcane samples were subjected to aflatoxin screening, *Aspergillus niger*, *A. ustus* and *Emericella nidulans* were not a toxin producers. Similarly, the isolated *Colletotrichum dematium* was also not toxin

producer. Brinker and Seigler, (1991) isolated piceatannol as a phytoalexin from the infected sugarcane with *Colletotrichum falcatum* but not from healthy or wounded sugarcane.

Effect of Dimethoate insecticide on mycelial growth and Go-T activity:

Table 3 and Figs. 1 and 2 indicate the effect of different concentrations of the insecticide Dimethoate on mycelial growth and Go-T activity on *Aspergillus flavus*, *A. niger*, *Fusarium moniliforme* and *Torula herbarum*. Generally, the growth of mycelium decreased with the increase in Dimethoate concentrations in all tested fungi. Although 25 ppm was less effective than the higher levels of the insecticide, a highly significant decrease in mycelial fresh weight was observed at 75-200 ppm of Dimethoate in all studied fungi, as compared with the control treatment. At low level (25 ppm) of Dimethoate, mycelial growth decreased significantly in *F. moniliforme* and *T. herbarum* while the reduction in mycelial growth of *A. flavus* and *A. niger* did not statistically differ from the control treatment. The results revealed that the fungal growth decreased with the increase in pesticide concentrations, which comes in agreement with Abd-Elaah (1993) who found that *Dimethoate* sharply reduced the growth of *Saprolegnia ferax*, *Achlya proliferoides* and *Dictyochus sterilis*. The effect of insecticides on the inhibition of mycelial dry weight of *Aspergillus fumigatus* and *Fusarium moniliforme* was also observed by El-Hissy and Abdel Kader (1980). They reported the rate of

inhibition to be also influenced by the type of the fungus, age of the mycelium and concentration of the pesticides.

The results indicated that the Dimethoate treatments stimulated the Go-T activity in *A. niger* and inhibited it in *A. flavus*, as compared with the respective control values (Table 3 and Fig.2). The activity of Go-T in *F. moniliforme* was higher than that of the control at 25-150 ppm of the insecticide, while it was inhibited at the higher level (200 ppm). In *T. herbarum*, the lowest level of Dimethoate (25 ppm) greatly stimulated the Go-T activity, while its activity decreased with the increase of Dimethoate concentration.

The above results revealed that the insecticide Dimethoate stimulated the activity of Go-T in *Aspergillus niger*, *Fusarium moniliforme* and *Torula harbarum* especially at low doses. The inhibitory effect was prominent in case of *Aspergillus flavus*, indicating that this fungus was more sensitive to this insecticide than the other tested fungi. Audus (1960) suggested that microorganisms can develop the ability of degrade pesticides either by enzyme induction or by mutation. Abd-Elaah (1993) found that the activity of Go-T increased in *Saprolegnia ferax* and *Dictyuchus sterilis* by the application of the insecticide Dimethoate and the herbicide Basta.

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