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Fungal Flora and Mycotoxins Associated with Onion (*Allium cepa* L.) in Egypt

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양파에 관련된 균독소 및 균프로라에 관한 연구

ABSTRACT: Seven genera and 15 species of fungi were isolated from 50 samples of then different steps of dried onion (5 samples of each step) collected from an onion factory in Sohag Governorate, Egypt, and grown on glucose-Czapek's agar (7 genera and 15 species) and 10% NaCl glucose-Czapek's (2 genera and 6 species). The average total counts of fungi were gradually decreased throughout the different steps of drying from 2090 to zero and 152 to zero colonies/g on glucose-Czapek's agar and 10% NaCl glucose-Czapek's agar media, respectively. *Aspergillus* was the most common genus on the two types of media used. The dominant species were *Aspergillus niger, A. flavus, A. terreus, Penicillium chrysogenum* and *Fusarium oxysporum* on glucose-Czapek's agar and *A. terreus* and *A. niger* on 10% NaCl glucose-Czapek's agar. The chloroform extracts of different samples were tested for the presence of mycotoxins using thin layer chromatographic analysis. The results indicated that aflatoxin was present at concentrations decreased throughout the different steps of the drying from step No. 1, onion bulbs, 120 μg/kg; to step No. 8, standard A, 20 μg/kg while step Nos. 9 & 10 (completely dry powdered onion) were free from aflatoxin. Citrinin was also present in the first three steps at concentrations gradually decreased from 30 to 10 mg/kg.

KEYWORDS: Fungal flora, mycotoxins, onion.

Onion (*Allium cepa* L.) is one of the most important crops in Egypt. It ranks as the third exportable field crop in Egypt after cotton and rice crops. Onions are cultivated on 26 thousand hectares in Egypt with an average yield of 550 thousand metric tons (about 21154 kg/ha) (FAO, 1991). Egyptian onion is famous for its high grade and keeping quality characteristics. The foreign markets depends on Egyptian onion, since they mature early.

Fungal contamination could be a deleterious not only for causing damage to the onion yield, but also in contaminating the soil making it unsuitable for growing onion for several years. Also many of the fungal species contaminating onion bulbs produce mycotoxins. Some of these fungal toxins are highly toxic to mammals.

Mycotoxins are fungal metabolites contaminating many food products, particularly stored products such as peanut, grains and cereals. Many moulds of the genera *Aspergillus, Penicillium* and *Fusarium* produce 80% of the total toxic substances (Hesseltine, 1974; Ciegler, 1979). The most important mycotoxins occurring in feed-and foodstuffs are aflatoxins, patulin, ochratoxins, citrinin, sterigmatocystin rubratoxin, T-2 toxin diacetoxyscirpenol and zearlaenone. The natural occurrence of these mycotoxins (as hepatotoxic, mutagenic, carcinogenic and teratogenic agents) in manufactured dried onion has become of increasing interest because of the world-wide use of dried onion.

Although, the mycotoxins and mycoflora of many agricultural commodities in Egypt were intensively studied in this laboratory (El-Kady *et al.*, 1982, 1991; El-Maraghy & El-Magraby, 1986, 1988; El-Magraby & El-Maraghy, 1987; El-Maraghy, 1989; Mazen *et al.*, 1990) none of these studies concentrated on the mycoflora or mycotoxins of dried onion. For this study, 50 samples repre-

senting the ten different steps of production of dried onion collected from an onion factory in Sohag Governorate, Egypt.

Materials and Methods

Collection of onion samples: Fifty samples representing ten different steps of production (Table 3) of dried onion (five, 500g, samples of each step of production) were collected from an onion factory at Sohag Governorate, Egypt. Onion was dried in onion Factory by hydrostatic drying.

Isolation of fungi: The fungal flora of the samples was determined using the dilution plate technique as described by Johnson *et al.* (1959), but with some modifications as employed by Moubasher *et al.* (1972). Ten g of each sample were washed by shaking mechanically in 100 m*l* of sterile distilled water. One m*l* of the water suspension was transferred to a sterile petri-dish, and poured with melted but cooled agar medium. Two types of media were used; glucose-Czapek's agar in which glucose (10g/*l*) replaced sucrose and glucose-Czapek's agar medium fortified by 100g/*l* of sodium chloride. Streptomycin (20 μ/m*l*) and rose-bengal (30 ppm) were applied to suppress bacterial growth (Smith & Dawson, 1944; Al-Doory, 1980). Ten plates were used for each sample (5 plates for each type of medium). The plates were incubated at 28± 2℃ for 1-2 weeks during which the developing colonies were counted, identified and the numbers were calculated per g of each sample.

Identifiecation: Purified fungal isolates were identified whenever possible in the original petri-dish culture. When this was not possible, fungi were subcultured and stored for later identification, according to Raper & Thom, 1949; Gilman, 1957; Raper & Fennell, 1965; Ellis, 1971, 1976; Pitt, 1979, 1985; Samson, 1979; Domsch *et al.*, 1980; Onions *et al.*, 1981; Ramirez, 1982 and Sivanesan, 1984.

Mycotoxin analysis: Twenty g of each sample were defatted by extraction with cyclohexane for 10 h using a Soxhlet-type extractor. The defatted residue was extracted for another 10 h with chloroform. The chloroform extract was dried over anhydrous sodium sulphate, filtered and then distilled under vacuum to near dryness. The residue was diluted with chloroform to 1 m*l*.

Chromatographic analysis of the chloroform extracts was achieved on precoated silica gel plate type 60 F 254 (Merck) for the presence of aflatoxins B_1, B_2, G_1 & G_2, citrinin, ochratoxin A, patulin, sterigmatocystin, T-2 toxin, diacetoxyscirpenol and zearalenone according to Scott *et al.* (1970) and Roberts & Patterson (1975).

Aflatoxins: quantitative determination of aflatoxins was made according to the methods discribed by Jones (1972) and Preybylski (1975). The aflatoxins concentration were estimated visualy in the chloroform extracts by quantitative thin layer chromatography by comprison with aflatoxins B_1, B_2, G_1 & G_2 standards. The practical limit is about 5 μg/kg for aflatoxin B_1 and total flatoxins and the recovery of aflatoxins is about 80%.

Citrinin; quantitative determination of citrinin was made according to the methods discribed by Scott *et al.* (1972) and Hald & Krogh (1973). Extraction of acidified (6 N Hcl to pH 2-3) substrate with chloroform was followed by estimation of citrinin concentration visualy using thin layer chromatography by comprison with citrinin standard. The potential lower limit of visual detection is about 0.2 ppm (=200 μg/kg) and the recovery of citrinin is about 71%.

Source of mycotoxin standard: All of mycotoxin standards used throughout this study were purchased from Makor Chemical Ltd. Jersualem, Israel and kindly provided by Prof. Dr. I. A. El-Kady, Botany Dept., Faculty of Science, Assiut University, Egypt.

Results and Discussion

Mycoflora of onion samples: Seven genera and 15 species of fungi were isolated and identified. Total counts of the fungi gradually decreased through the different drying steps from 2090 to zero and 152 to zero colonies/g on the two different media, respectively. The gross total counts of glucophilic and xerophilic fungi in all samples tested

were 8876 and 871 colonies/g samples, respectively (Tables 1-2). All of these fungi were previously recovered from the rhizosphere and phyllosphere of some Egyptian plants (Montasir *et al.*, 1959; Moubasher *et al.*, 1971; Tolba & Ali, 1972; Abdel-Wahab, 1975; Abdel-Fattah *et al.*, 1977; El-Hissy *et al.*, 1980; Abdulla, 1981; Mazen *et al.*, 1985, 1988; Ismail *et al.*, 1989; Salama *et al.*, 1989; Abdel-Hafez *et al.*, 1990).

The samples of the final product (step No. 10, onion powder) were completely fungal free, whilst the samples of step No. 9 (prefinal product) were contaminated with three species of *Aspergillus* only.

Aspergillus was the dominant genus, recorded in 90% of the samples using both isolation media. It was represented by 6 species. *Aspergillus niger* and *A. terreus* were the commonest species on both media used. *A. flavus* was isolated with high and low frequencies of occurrence on glucose-Czapek's and 10% NaCl glucose-Czapek's agar media, respectively. *A. tamarii* was recorded with moderate frequency while *A. ochraceus* and *A. sydowii* were isolated with less frequencies on glucose-Czapek's agar. On 10% NaCl glucose-Czapek's agar, *A. sydowii* and *A. tamarii* were only isola-

Table 1. Average total counts (calculated per g fresh weight in each & all sample steps), number of cases of isolation (NCI, out of 10 steps) and occurrence remarks (OR) of fungal genera and species isolated from different steps of production of dried onion on glucose-Czapek's agar medium at 28± 2℃.

Average total counts / Genera & species	Step numbers										Total counts	NCI & OR*
	1	2	3	4	5	6	7	8	9	10		
Aspergillus	250	1220	1140	928	136	320	220	120	458	0	4792	9H
A. niger van Tieghem	130	1120	1020	888	56	200	176	112	428	0	4130	9H
A. flavus Link	120	10	10	8	40	64	28	0	30	0	310	8H
A. terreus Thom	0	90	100	32	24	48	8	0	0	0	302	6H
A. tamarii Kita	0	0	10	0	16	8	0	0	0	0	34	3M
A. ochraceus Wilhelm	0	0	0	0	0	0	0	8	0	0	8	1R
A. sydowii (Bain. & Sart.) Thom & Church	0	0	0	0	0	0	8	0	0	0	8	1R
Penicillium	1550	50	30	40	672	432	384	24	0	0	3182	8H
P. chrysogenum Thom	1550	40	30	32	320	336	384	24	0	0	2716	8H
P. citrinum Thom	0	10	0	0	0	40	0	0	0	0	50	2L
P. funiculosum Thom	0	0	0	8	0	56	0	0	0	0	64	2L
P. oxalicum Currie & Thom	0	0	0	0	352	0	0	0	0	0	352	1R
Fusarium oxysporum Schlecht	290	0	60	0	0	0	4	464	0	0	818	4M
Cladosporium cladosporioides (Fres.) de Vries	0	50	0	0	0	8	0	0	0	0	58	2L
Epicoccum nigrum Link	0	10	0	0	0	0	4	0	0	0	14	2L
Cochliobolus spicifer Nelson	0	0	0	0	0	0	4	0	0	0	4	1R
Ulocladium atrum Preuss	0	0	0	8	0	0	0	0	0	0	8	1R
Total count	2090	1330	1230	976	808	760	616	608	458	0	8876	
Number of genera	3	4	3	3	2	3	5	3	1	0	7	
Number of species	4	7	6	6	6	8	8	4	2	0	15	

OR*: Occurrence of remarks, H: High occurrence; between 5-10 cases, M: Moderate occurrence; 3 or 4 cases, L: Low occurrence; 2 cases. R: Rare occurrence: one case.

Table 2. Average total counts (calculated per g fresh weight in each & all sample steps), number of cases of isolation (NCI, out of 10 steps) and occurrence remarks (OR) of fungal genera and species isolated from different steps of production of dried onion on 10% NaCl glucose-Czapek's agar medium at 28± 2℃.

Average total counts	Step numbers										Total	NCI &
Genera & species	1	2	3	4	5	6	7	8	9	10	counts	OR*
Aspergillus	152	140	126	110	96	84	74	58	27	0	867	9H
A. terreus Thom	152	88	92	98	34	28	32	28	18	0	570	9H
A. niger van Tieghem	0	52	32	10	59	56	30	30	9	0	278	8H
A. flavus Link	0	0	2	0	0	0	12	0	0	0	14	2L
A. sydowii (Bain. & Sart.) Thom & Church	0	0	0	2	0	0	0	0	0	0	2	1R
A. tamarii Kita	0	0	0	0	3	0	0	0	0	0	3	1R
Penicillium citrinum Thom	0	0	4	0	0	0	0	0	0	0	4	1R
Total count	152	140	130	110	96	84	74	58	27	0	871	
Number of genera	1	1	2	1	1	1	1	1	1	0	2	
Number of species	1	2	4	3	3	2	3	2	2	0	6	

OR*: Occurrence of remarks, H: High occurrence; between 5-10 cases, M: Moderate occurrence; 3 or 4 cases, L: Low occurrence; 2 cases, R: Rare occurrence; one case.

ted from one out of ten production steps.

All of the above *Aspergillus* species have been previously isolated from the rhizospheres and the surrounding soils of onion plants as well as from the phyllosphere of some Egyptian plants (Abdel-Wahab, 1975; Abdel-Fattah *et al.*, 1977; Abdel-Gawad, 1978, 1984; Mazen *et al.*, 1985; Ismail *et al.*, 1989). El-Dohlob (1972) found that *A. niger, A. terreus* and *A. carneus* were the common *Aspergilli* species in the rhizosphere of onion. Recently, Salama *et al.* (1989) reported that *Aspergillus* was the common genus in the rhizosphere and the surrounding soils of onion plants. They isolated 16 *Aspergilli* species and found that *A. flavus, A. terreus* and *A. niger* were the most common.

Penicillium came second in prevalence and was recovered from 80% of the samples, being 35.85% of the total fungi on glucose-Czapek's agar medium, but only isolated once on 10% NaCl glucose-Czapek's agar (Tables 1-2). It was represented by four species on glucose-Czapek's agar of which *P. chrysogenum* was the most prevalent, present in 80% of the samples comprising 85.36% of total *Penicillia* and 30.60% of total fungi (Table 1). *P. citrinum* was isolated on both media used while *P. funiculosum* and *P. oxalicum* were isolated only on glucose-Czapek's agar with low or rare frequency as listed in Tables 1 and 2. These four species were recovered from the rhizospheres and the surrounding soils of onion plants in Egypt (El-Dohlob, 1972; Ismail *et al.*, 1989; Salama *et al.*, 1989). In addition, they were isolated from the rhizosphere and phyllosphere of some other Egyptian plants (Moubasher *et al.* 1971; Abdel-Fattah *et al.*, 1977; El-Hissy *et al.*, 1980; Mazen *et al.*, 1985, 1988; Abdel-Hafez *et al.*, 1990).

Fusarium (represented by *F. oxysporum*) ranked third, growing only on glucose-Czapek's agar and recovered from 40% of the samples yielding 9.2% of total fungi. El-Dohlob (1972) and Salama *et al.* (1989) isolated *F. oxysporum* from the rhizosphere and the surrounding soil of onion plants.

Cladosporium cladosporioides, Epicoccum nigrum; Cochliobolus spicifer and *Ulocladium atrum* were of low occurrence and represented in 10-20% of the samples accounting 0.06-0.66% of total fungi on glucose-Czapek's agar only (Table 1). Ismail *et al.* (1989) and Salama *et al.* (1989) isolated these genera from the rhizosphere and the surrounding soil of onion plants.

Table 3. Natural occurrence of mycotoxins in the different onion samples (5 samples represented each step) of different steps of manufactured dry onion.

Step No.	Different steps of onion manufacturing	aflatoxins (μg/kg)						Citrinin (mg/kg)
		B_1	B_2	G_1	G_2	Total	%AFB_1/TAFS**	
1	Onions (bulbs)	55	25	30	10	120	46	30
2	Onions without scaly leaves	50	20	20	10	100	50	25
3	Washed onion	45	20	12	8	53	10	
4	Glutted onion	40	15	12	8	75	53	--
5	1st step of drying (≈80% m.c.*)	20	10	10	--	40	50	--
6	2nd step of drying (≈40% m.c.)	25	10	--	--	35	71	--
7	3rd step of drying (≈5% m.c.)	20	10	--	--	30	67	--
8	Standard A (shreded dry onion after sieving)	20	--	--	--	20	100	--
9	Prefinal product	--	--	--	--	--	--	--
10	Final product (dry powdered onion)	--	--	--	--	--	--	--

*m.c.=Moisture contents. ** % AFB_1/TAFS=% (Aflatoxin B_1/Total Aflatoxins).

Most of the fungal genera (5 out of 7 genera) and species (9 out of the 15 species)were recovered on glucose-Czapek's agar only. Xerophilic fungi were represented by *Aspergillus* (5 species)in 90% of the samples and *Penicillium citrinum* in one sample only. The total count of fungi were gradually decreased throughout the different steps of dried onion production. The final product (onion powder) was completely fungus free.

Natural occurrence of mycotoxin in onion samples: Thin-layer chromatographic analysis was used for the detection of mycotoxins. Results in Table 3 indicate that aflatoxin was present in decreasing concentrations for the different steps of onion drying, from step No. 1(onion bulbs, 120 μg/kg) to step No. 8 (standard A, 20 μg/kg). Steps 9 and 10 were completely free from aflatoxin and Ca 5 μg/kg. Citrinin was also present in the first three steps only of dried onion production, at concentrations gradually decreasing from 30 to 10 mg/kg and Ca 200 μg/kg.

Natural occurrence of citrinin in different substrates was previously found in corn and silage at concentrations of 2-3 ppm (=2000-3000 μg/kg) (Chalam and Stahr, 1979) and in cereals used as feed for swine in districts of Denmark at 2 ppm. Scott *et al.* (1972) isolated citrinin from 13/29 heated Canadian grains including samples of wheat, oats, barley and rye at concentrations ranged between 0.07 and 80 ppm. Subrahmanjan and Rao (1974) obtained citrinin in peanut pods at concentrations ranged from a trace to 1200 mg/kg.

Patulin, ochratoxins, sterigmatocystin, rubratoxin, T-2 toxin, diacetoxyscirpenol and zearalenone were not detected in any sample of onion tested.

Since no detectable amounts of any mycotoxins were recorded in the final products (dried onion), it can be concluded that the subsequent refining operations adopted in the onion factory, Sohag Governorate, Egypt completely eliminated mycotoxins.

References

Abdel-Fattah, H. M., Moubasher, A. H. and Abdel-Haffez, S. I. 1977. Fungus flora of root and leaf surface of broad bean cultivated in Oasis, Egypt. *Naturalia Monspel. Sér. Bot.* **27**: 167-171.

Abdel-Gawad, K. M. 1978. Studies on the phyllosphere and phylloplane mycoflora of some plants M. Sc. Thesis. *Bot. Dept. Fac. Sci., Assiut Univ.,*

Assiut, Egypt 207 p.

Abdel-Gawad, K. M. 1984. Further studies on the fungal flora of the phyllosphere and phylloplane of some plants *Ph. D. Thesis, Bot. Dept. Fac. Sci., Assiut Univ.,* Assiut, Egypt 271 p.

Abdel-Hafez, S. I., Mazen, M. B. and Gehan, M., Shaban. 1990. Seasonal fluctuations of rhizosphere and rhizoplane fungi of Egyptian wheat plant. *Bull. Fac. Sci., Assiut Univ.* **19**: 173-184.

Abdel-Wahab, A. M. 1975. Phyllosphere microflora of some Egyptian plants. *Folia Microbiol.* **20**: 236-245.

Abdulla, M. E. 1981. Responses of phyllosphere mycoflora of *Eucalyptus* plant to volatile substances emanated from its leaves. *Annual. Rev. Univ., Fac. Education, Ain-Shams Univ.* **4**: 377-387.

Al-Doory, Y. 1980. Laboratory medical mycology. *London, Lea and Febiger* 410 p.

Chalam, R. V. and Stahr, H. M. 1979. Thin layer chromatographic determination of citrinin. *J. Assoc. Off. Analytical Chem.* **62**: 570-572.

Ciegler, A. 1979. *Mycopathologia. 65: 5. Cited in Lutomski J. and Kedzia B. 1980. Mycoflora of crude drugs. Estimation of mould contaminations and their toxicity. Planta Medica* **40**: 212-217.

Domsch, K. H., Gams, W. and Anderson, T. E. 1980. Compendium of soil fungi *New York, Academic Press,* 859 p.

El-Dohlob, S. M., 1972. The effect of some soil condition on the composition of soil fungus flora. *Ph. D. Thesis. Bot. Dept. Fac. Sci. Assiut Univ.* Assiut, Egypt.

El-Hissy, F. T., Abdel-Hafez, S. I. and Abdel-Kader, M. I. 1980. Rhizosphere of five plants in Egypt. *Z. Allg. Mikrobiol.* **20**: 177-184.

El-Kady, I. A., Abdel-Hafez, S. I. and El-Maraghy, S. S. 1982. Contribution to the fungal flora of cereal grains in Egypt. *Mycopathologia* **77**: 103-109.

El-Kady, I. A., El-Maraghy, S. S. and Zohri, A. A. 1991. Mycotoxin production on different cultivars and lines of broad bean (*Vicia faba* L.) seeds in Egypt. *Mycopathologia* **113**: 165-169.

Ellis, M. B. 1971. Dematiaceous Hyphomycetes. *Kew, England, CMI,* 608 p.

Ellis, M. B. 1976. More Dematiaceous Hyphomycetes. *Kew, England, CMI,* 507 p.

El-Magraby, O. M. O. and El-Maraghy, S. S. 1987. Mycoflora and mycotoxins of peanut (*Arachis hypogaea* L.) seeds in Egypt. 1. Sugar fungi and natural occurrence of mycotoxins. *Mycopathologia* **98**: 165-170.

El-Maraghy, S. S. M. and El-Magraby, O. M. O. 1987. Mycoflora and mycotoxins of sunflower (*Helianthus annuus* L.) seeds in Egypt. 1. Sugar fungi and natural occurrence of mycotoxins. *Bull. Fac. Sci., Qatar Univ.* **6**: 107-121.

El-Maraghy, S. S. M. and El-Magraby, O. M. O. 1988. Mycotoxins production on different varieties of four cereal grains cultivated in Egypt. *Proc. Int. Congress of Plant Physiology, New Delhi, India, Feb.* 15-20, 1988, **1**: 127-130.

El-Maraghy, S. S. M. 1989. The fungal flora and natural occurrence of mycotoxins of some leguminous varieties and hybrids seed *Bull. Fac. Sci., Assiut Univ.* **18**: 63-75.

Fao. 1991. Production Yearbook, Food and Agriculture Organization of the United Nations, Rome. *Statistics series* **44**(99): 141-142.

Gilman, J. C. 1957. A manual of soil fungi. *Iowa, USA. State Univ. Press,* 450 p.

Hald, B. and Krogh, P. 1973. Analysis and chemical confirmation of citrinin in barley. *J. Assoc. Off. Analytical Chem.* **56**: 1440-1443.

Hesseltine, C. W. 1974. Natural occurrence of mycotoxins in cereals. *Mycopathol. Mycol. Appl.* **53**: 141-153.

Ismail, I. M. K., Salama, A. M., Ali, M. I. A. and Ouf, S. A. 1989. Effect of onion root treatment with *Aspergillus terreus* and *Bacillus pumilus* culture filtrates on rhizosphere microflora.. 7th *Conf. of Microbiology, Nov.* 19-22, Cairo Egypt.

Johnson, L. F., Curl, E. A., Bond, J. H. and Friboury, H. A. 1959. Method for studying soil microflora, plant disease relationships. *Minneapolis, Burgers Publ. Co.*

Jones, B. D. 1972. Method of aflatoxin analysis G. 70. London, *Tropical Products Institute.* 58 p.

Mazen, M. B., Abdel-Hafez, S. I. and Gehan, M. Shaban. 1985. Seasonal fluctuations of phyllosphere and phylloplane fungi of Egyptian wheat. *Acta Mycol.* **21**: 109-116.

Mazen, M. B., Moubashear, A. H. and El-Sharouny, H. M. 1988. Studies on the genus *Pythium* in Egypt. IV-Seasonal occur-rence of rhizosphere and rhizoplane fungi of healthy and damped-off seedlings of five economic plants. *Bull. Fac. Sci. Assiut Univ.* **17**: 29-42.

Mazen, M. B., El-Kady, I. A. and Sabah, M. Saber. 1990. Survey of the mycoflora and mycotoxins of

cotton seeds and cotton seed products in Egypt. *Mycopathologia* **110**: 133-138.

Montasir, A. H., Moustafa, M. I. and Youssef, Y. A. 1959. Studies on the peritrophic mycorrhiza between *Tropaeolum majus* L., and two associated rhizospheric fungi. I-Cultural studies of *Aspergillus awameri* and *A. terreus*. *Proc. Egypt Acad. Sci.* **14**: 16-21.

Moubasher, A. H., El-Naghy, M. A. and Abdel-Fattah, H. M. 1971. Citrus plantation fungi in Upper Egypt. *Trans. Brit. Mycol. Soc.* **57**: 289-294.

Moubasher, A. H., El-Naghy, M. A. and Abdel-Hafez, S. I. 1972. Fungal flora of three grains in Egypt. *Mycopathologia et Mycologia Applicata* **47**: 261-274.

Onions, A. H. S., Allsopp, D. and Eggins, H. O. W. 1981. Smith's introduction to industrial mycology. *London, E. Arnold Ltd.*, 398 p.

Pitt, J. I. 1979. The genus *Penicillium* and its teleomorphic states *Eupenicillium* and *Talaromyces*. *London, Academic Press,* 634 p.

Pitt, J. I. 1985. A laboratory guide to common *Penicillium* species, *Kew, England, CMI,* 184 p.

Przybylski, W. 1975. Formation of derivatives of aflatoxins on TLC plates. *J. Assoc. Off. Analytical Chem.* **58**: 163-164.

Ramirez, C. 1982. Manual and Atlas of *Penicillia. Amsterdam, Netherlands, Elsevier Biomedical Press,* 874 p.

Raper, K. B. and Thom, C. 1949. The Manual of the *Penicillium Baltimore, USA, Williams & Wilkins,* 875 p.

Raper, K. B. and Fennell, P. I. 1965. The genus *Aspergillus. Baltimore, USA, Williams & Wilkins,* 686 p.

Roberts, B. A. and Patterson, D. S. P. 1975. Detection of twelve mycotoxins in mixed animal feedstuffs, using a novel membrane cleanup procedure, *J. Assoc. Off. Analytical Chem.* **58**: 1178-1181.

Salama, A. M., Ismail, I. M. K., Ali, M. I. A. and Ouf, S. A. 1989. Rhizosphere microflora of onion in relation to soil amendement with *Eucalyptus rostrata* leaves and *Sclerotium cepivorum*. *Bull. Fac. Sci., Cairo Univ.* **57**: 577-605.

Samson, R. A. 1979. A compilation of the aspergilli described since 1965. *Stud. Mycol.* **18**: 1-39.

Scott, P. M., Lawrence, J. W. and Van Walbeak, W. 1970. Detection of mycotoxins by thin-layer chromatography. Application to screening of fungal extracts. *Appl. Microbiol.* **20**: 839-842.

Scott, P. M., Van Walbeak W., Kennedy, B. P. C. and Anyeti, D. 1972. Mycotoxins (ochratoxin A, citrinin and sterigmatocystin) and toxigenic fungi in grains and other agricultural products. *J. Agric. Food Chem.* **20**: 1103-1109.

Sivanesan, A. 1984. The bitunicate Ascomycetes and their anamorphs. *Hirschberg, Germany, Strauss & Gramer GmbH,* 701 p.

Smith, N. R. and Dawson, V. T. 1944. The bacteriostatic action of rosebengal in media used the plate count of soil fungi. *Soil Soc.* **58**: 467-471.

Subrahmanjan, P. and Rao, A. S. 1974. Occurrence of aflatoxins and citrinin in groundnut (*Arachis hypogaea* L.) at harvest in relation to pod condition and kernal moisture content. *Current Sci.* **43**: 707-710.

Tolba, M. K. and Ali, M. I. 1972. Studies on rhizosphere microflora of cotton plants in Egypt. *Egypt. J. Bot.* **15**: 23-43.

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