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SOIL SOLARIZATION AND THE COMPOSITION OF SOIL FUNGAL COMMUNITY IN UPPER EGYPT

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ABSTRACT :

In Upper Egypt, 6 weeks solarization of soil resulted in elevating soil temperature to ranges considered to be lethal or sublethal to many soil fungi.

The composition of soil fungal community was altered in solarized soil. Both total count and number of fungal species detected on PDA medium at 28 ± 2°C were greatly reduced in solarized soil as compared to unmulched soil. On the other hand, number of fungal genera was not significantly affected by soil solarization throughout the sampling period (0 – 13 months). At the end of solarization period, several fungi re-colonized solarized soil and the total count of soil fungi was significantly higher than that of unmulched soil. Counts of thermophilic/ thermotolerant fungi isolated on YpSs were significantly reduced at the end of solarization period (40 days). Number of thermophilic/thermotolerant genera and species was not significantly affected by soil solarization.

INTRODUCTION:

Soil solarization is a term used to describe hydro/thermal soil heating accomplished by covering moist soil with clear polyethylene tarps during the summer months (Stapleton and De Vay, 1982). Several workers reported the success of this treatment in reducing plant diseases caused by soil-borne pathogens (Elad *et al.*, 1980; Pullman *et al.*, 1981 a,b; Katan *et al.*, 1983; Abdel-Rahim *et al.*, 1988; Abu-Gharbieh *et al.*, 1990 a,b; Tjamos *et al.*, 1991; Gamliel and Stapleton, 1993; Keinath, 1995, 1996 and Blok *et al.*, 2000).

Most of the previous studies reporting reduction in population densities of soil-borne pathogens were confined to target organisms and did not determine the effect of solarization on a broader range of soil microbiota, including those which may be antagonistic to plant pathogens. On the other hand, few studies undertaken to investigate the effect of solarization on soil microbiota in general (Stapleton and De Vay, 1982, 1984; El-Zayat *et al.*, 1991; Gamliel and Katan, 1991 and Botross *et al.*, 2000). In the present study, population densities of several genera, species and species varieties of common soil fungi in solarized, nonsolarized and shaded soils were periodically estimated for 13 months to determine the initial and residual effects of soil solarization on their survival and ability to colonize previously solarized soil.

MATERIALS AND METHODS:

Experimental design :

An experiment consisted of three treatments namely, solarized (mulched with 40 μm thick transparent polyethylene sheets), unmulched (exposed to direct sun-light), and shaded (cultivated with maize) was conducted in a field in Bani-Ady, Manfalout, Assiut, Upper Egypt. The experimental design was completely randomized block design with three replications (plots) for each treatment. Each plot consisted of six rows, 0.5 m apart and 8 m long. The soil was ploughed twice, listed to form raised beds and flood irrigated the day before polyethylene tarps (sheets) were placed on soil.

Soil solarization:

Soil solarization was accomplished by covering moist soil with 40 μm thick transparent polyethylene tarps on 5th July 2002, and plots of the unmulched soil were left exposed to direct sun light. Plots of the soil were cultivated with maize one month before starting the solarization period and soil in this case was considered as shaded soil. Edges of the polyethylene tarps were buried in furrows between beds. Special care was taken to minimize the distance between the tarps and soil to prevent the formation of air pockets that retard the soil heating process. All plots were supplemently irrigated with 10-15 cm flood irrigation every two weeks until the polyethylene traps were removed on 17th August 2002.

Monitoring of soil temperature :

Minimum and maximum soil temperatures were daily monitored for mulched, unmulched and shaded soils. Soil temperatures were monitored throughout the solarization period by thermometers fixed at 5-, 10-, 15- and 20 cm soil depths in one plot of each treatment. The minimum and maximum soil temperatures were recorded at 4 am and 3 pm, respectively according to El-Shami *et al.* (1990).

Soil sampling:

Soil samples were taken from the upper 20 cm of the soil profile with a sampling tube 2.5 cm inside diameter. Five soil samples were collected at random from each plot. The soil of each tube was divided into 0-10 and 10-20 cm depth. Tube halves related to the same soil layer were bulked for each treatment and kept in plastic bags to form composite samples according to the method of Johnson *et al.* (1959). Soil samples were collected at eight different sampling periods.

Culture media :

Two selective media were used to estimate and compare population densities of soil fungi. Potato-dextrose agar (PDA) (Riker and Riker, 1936) was used for determination of mesophilic fungi (at $28^{o}\pm2^{o}$C). The medium was supplemented with rose-bengal (66 μg ml^{-1}) and streptomycin (30 μg ml^{-1}) as bacteriostatic agents (Martin, 1950). Yeast-starch (YpSs) agar medium+rose-bengal and streptomycin as above, was used for thermophilic/ thermotolerant fungi (at $45^{o}\pm2^{o}$C).

Assay procedure for counts of soil fungi:

The dilution plate method described by Dhingra and Sinclair (1995) was used for determination of soil fungi.

Statistical analysis :

All data were subject to analysis of variance (ANOVA) and LSD compared means or Duncan's multiple range tests. All analyses were performed with the M Stat program.

RESULTS AND DISCUSSION:

Monitoring of soil temperature:

The success of soil solarization depends on the soil temperatures reached during the process (De Vay, 1990 a). Daily records of soil temperatures during the period of solarization were taken for mulched, unmulched and shaded plots (Table 1). Results indicate that, soil temperatures elevated remarkably by mulching with transparent polyethylene tarps during the period from 5 July to 17 August 2002. Mulching increased average maximum soil temperature than unmulched one by 10°, 7.75°, 7.25° and 5.75°C at 5,10,15 and 20 cm depths, respectively. This result is almost in line with those obtained by Chen and Katan (1980); Stapleton and De Vay (1982); El-Shami *et al*. (1990); Mohamed (1990); Abdel-Rahim *et al*., (1988). On the other hand, shading reduced average maximum soil temperature by 6.4°, 4.25°, 4° and 3.25°C than unmulched soil at 5, 10,15 and 20 cm depths, respectively. Maximum temperatures obtained at the layer 5-15 cm of the mulched soil (57°-47.5°C) were in the range considered by many workers to be lethal to many soil fungi. Katan *et al*. (1976); Pullman *et al*. (1981 b); De Vay (1990 a); Stapleton (1990) and Keinath (1995) reported that temperatures at 47°C or higher are lethal to many mesophilic fungi. Maximum temperatures obtained at the layer 20-5 cm of unmulched soil (37°-47°C) were situated in the sub-lethal temperature range. A period of time ranging from 2-4 weeks may be required for mesophilic fungi to be killed within this temperature range as it was stated by Pullman *et al*. (1981b) and De Vay (1990 a). At the same time averages of minimum temperatures ranged from 33.5° to 38.5°, 30.75° to 34.5° and 27.75° to 31°C at 5 and 20 cm depths of mulched, unmulched and shaded soils, respectively. The average minimum temperatures were generally higher in mulched than unmulched soil by about 3-4°C. The temperature fluctuation amplitude, calculated as the difference between means of maximum and minimum temperatures of soil, was relatively high at the top layer (5 cm) of mulched soil then decreased sharply at lower depths. This result indicates that mulching enhanced the increase of soil temperature, but, mulched soil lost more heat during night. This is due to the greenhouse effect. Stapleton (1990) reported that covering soil with transparent polyethylene produces a "greenhouse effect" which raises soil temperature to levels that are lethal or injurious to many plant pathogens and pests. The greenhouse effect produced in solarized soil was also reported by Katan *et al*. (1976) and El-Shami *et al*. (1990).

Table (1): Diurnal variations of soil temperatures at different depths over the mulching period (5 July–17 August 2002), using 40µm thick transparent polyethylene

Treatment	Soil Depth (Cm)	Total averages of soil temperatures °C				
		Minimum	Maximum	Total	Mean	Temp. fluct. amplitude
Mulched soil	0	30	64.5	94.5	47.25	34.5
	5	33.5	57.0	90.50	45.25	23.5
	10	35.75	50.25	86.0	43.0	14.0
	15	37.5	47.5	85.0	42.5	10.0
	20	38.5	43.75	82.25	41.25	5.0
Unmulched soil	0	27.25	54.9	82.15	41.8	27.65
	5	30.75	47.0	77.75	38.88	16.25
	10	32.75	42.5	75.25	37.35	9.75
	15	34.0	40.25	74.25	37.13	6.25
	20	34.5	38.0	72.4	36.2	3.5
Shaded soil	0	26.75	42.75	69.5	74.75	16.0
	10	29.0	38.25	67.25	33.65	9.25
	15	30.0	36.2	66.2	33.1	6.2
	20	31.0	34.75	65.75	32.88	3.75

*** Temperature fluctuation amplitude is the difference between averages of minimum and maximum daily temperatures.**

1-Assay of Mesophilic fungi:

a-At zero time:

Immediately before starting soil solarization (at zero time), total count (Fig.1a), number of genera (Fig. 1b), number of species (Fig. 1c) and density levels of species (Table 2) of soil fungi did not show any significant difference between mulched, unmulched and shaded soils at 0-10 and 10-20 cm depths. This result indicating homogeneity of the native mycocommunity present in the tested field.

b- During solarization period :

Population densities of total fungi (Fig. 1a and tables 2, 3) were greatly reduced in solarized soil. Most reduction of total count occurred in the first 20 days at 0–10 and 10–20 cm depths.. Reduction of total count of soil fungi in solarized soil was previously reported by many workers (Stapleton and De Vay 1982 and 1984; El-Zayat *et al*., 1990; Gamliel and Katan, 1991; Keinath, 1995; Abdellah *et al*., 1998 and Botross *et al*., 2000). Reduction in total count of fungi in mulched soil compared to unmulched one were 46, 42 and 18% at 0-10 cm and 18.6, 10.5 and 8.7% at 10-20 cm depth of soil after 20, 30 and 40 days, respectively. In this respect, soil solarization was much effective at the upper 10 cm of the mulched soil (pullman *et al*., 1981a, b; Katan *et al*., 1983; Greenberger *et al*., 1987; Mohamed 1990). Ahmed *et al*. (2000) reported that, in tarped soil, populations of *Rhizoctonia solani, Macrophomina phaseolina* and *Fusarium oxysporum* f. sp. *vasinfectum* the causal pathogens of root-rots and wilt diseases in cotton were greately decreased at depth of 0-10 cm more than that of 10-20 cm of soil. Reduction in population densities of *Emericella* spp., *Fusarium* spp., and *Rhizopus stolonifer* was mainly responsible for the reduced population densities of total fungi isolated from solarized soil. In agreement with the above results, several workers reported significant reduction in population densities of *Fusarium* spp. in solarized soil (Katan *et al*., 1976 and 1983; Katan, 1981; Freeman and Katan, 1988; Abu-Gharbieh *et al*., 1990a; Mohamed, 1990; Sarhan, 1990; Keinath 1995; and Wadi, 1999). Ioannou (2000) reported that soil solarization reduced population density of *Fusarium* spp. in solarized soil by 91-98%. 20 days solarization resulted in reduction of *Fusarium* spp. (*F. chlamydosporum*, *F. dimerum* and *F. oxysporum*) to undetectable levels. Abd-El Razik *et al*. (1990) reported that *Fusarium* was a soil thermolabile fungus. Botross *et al*. (2000) recorded reduced population density of *Rhizopus* spp. in solarized soil (50% at 2 months solarization and undetectable levels at 4 and 6 months solarization). Results of the present work show that 20 days solarization resulted in reduction of *Rhizopous stolonifer* to undetectable level (Table 2). The reduced population densities recorded for *Emericella* spp. obtained from solarized soil during the present investigation disagree with the findings of Dwivedi (1998) who reported increasing counts of *Emericella nidulans* in mulched soil (about 50% increase over nonsolarized soil).

At 40 days, total count of soil fungi showed no significant difference between mulched, unmulched and shaded plots. (Table 3). However, the total count of fungi at 40 days, was significantly increased in mulched soil than at the previous periods (20 and 30 days). This increase was due to the high population of *Aspergillus* spp., which represented 69 and 53.6% of total count of fungi at 0–10 and 10–20 cm depths, respectively. *A. flavus*, *A. fumigatus* and *A.niger* represented 8.25%, 37.11% and 23.71% of total count of fungi at 0–10 cm and 3.19%, 37.23% and 14.49% of total count of fungi at 10–20 cm depth, respectively. Abu-

Gharbieh *et al*. (1990a, b) reported about 44% increase in count of *Aspergillus* spp. over the wet control at the end of 11 weeks solarization period. This result agrees with the findings of Tjamos *et al.* (1991);Dwivedi (1998) and Botross *et al*. (2000).

Regardless of sampling time, there were insignificant differences among mulched, unmulched and shaded soils concerning the number of fungal genera at 0-10 and 10-20 cm depths (Fig. 1b).

Number of fungal species in mulched soil was significantly reduced through the solarization period than in the unmulched and shaded soils. The highest reduction in the number of fungal species occurred after the first 20 days (Fig. 1c). Variation of species number among mulched, unmulched and shaded soils can be attributed to reduction undetectable levels of some fungal species in solarized soil. *Alternaria alternata, Cochliobolus spicifer, Emericella nidulans v. dentata, E. nidulans v. lata, E. nidulans v. nidulans* and *E. rugulosa* were reduced to undetectable level in mulched soil at 0-10 cm depth, after 20 days soil solarization. While *Rhizopus stolinifer* was eradicated from 0-20 cm of mulched soil at the same period. In addition, *Fusarium chlamydosporum*, *F. dimerum* and *Gibberella fujikuroi* var. *fujikuroi* (anamorph) were eradicated at 0-20 cm depth after 30 days soil solarization. While, solarizing soil for 40 days eradicated *Cochliobolus sativus, Gliocladium roseum, Melanospora zamiae and Nectria heamatococca* (anamorph) in addition to the species eradicated after 30 days. It is clear that, solarizing soil for 40 days was more effective in eradicating some fungal species than solarizing it for 30 or 20 days. This result is in harmony with the result of Ahmed *et al*. (2000) who found that, mulching soil (in Assiut) for 30 days was more effective than mulching it for 15 days in reducing populations of *Rhizoctonia Solani, Macrophomina Phaseolina* and *Fusarium oxysporum f. sp. vasinfectum*. There are indications from field study that, soil solarization resulted in eradication of certain target species of fungi (Katan *et al*., 1976; Elad *et al.*, 1980; pullman *et al*., 1981a, b and Gamliel and Katan, 1991). They reported that soil solarization resulted in eradication of *Verticillium dahlia* and *Fusarium oxyspoum* f.sp. *lycopersici* at 0-15 cm depth, *Sclerotium rolfisii, pythium* spp., *Rhizoctonia solani* and *V. dahlia* at 0- 20 cm depth, *Thielaviopsis basicola, Fusarium* spp. and *pythium* spp., at 0-46 cm, *Penicillium pinophilum* and *pythium* spp. at 0-90 cm.

The shift in microbial balance is the basis on which soil solarization operates and causing control of soil-borne pathogens, suppressiveness of solarized soil to introduced pathogens and increased growth responses in solarized soil (pullman *et al*., 1981a; Stapleton and De Vay 1982, 1983 and 1984; Katan *et al*., 1983; Greenberger 1987; Abdel-Rahim *et al*., 1988; and Gamliel and Katan, 1991).

The substrate made available by soil solarization was rapidly occupied by the surviving organisms. Populations of these species were increased, as compared with their populations in unmulched and shaded soils. Population densities of *Aspergillus* spp., (especially *A. fumigatus, A. flavus* and *A. niger*) were significantly increased in mulched soil 20-40 days after starting soil solarization. The higher count values of these species in mulched soil compared to unmulched one indicate their greater competition capacity for available niches. This result is in line with those reported by Stapleton and De Vay (1982, 1984), Greenberger *et al*. (1987) and Stapleton (1990).

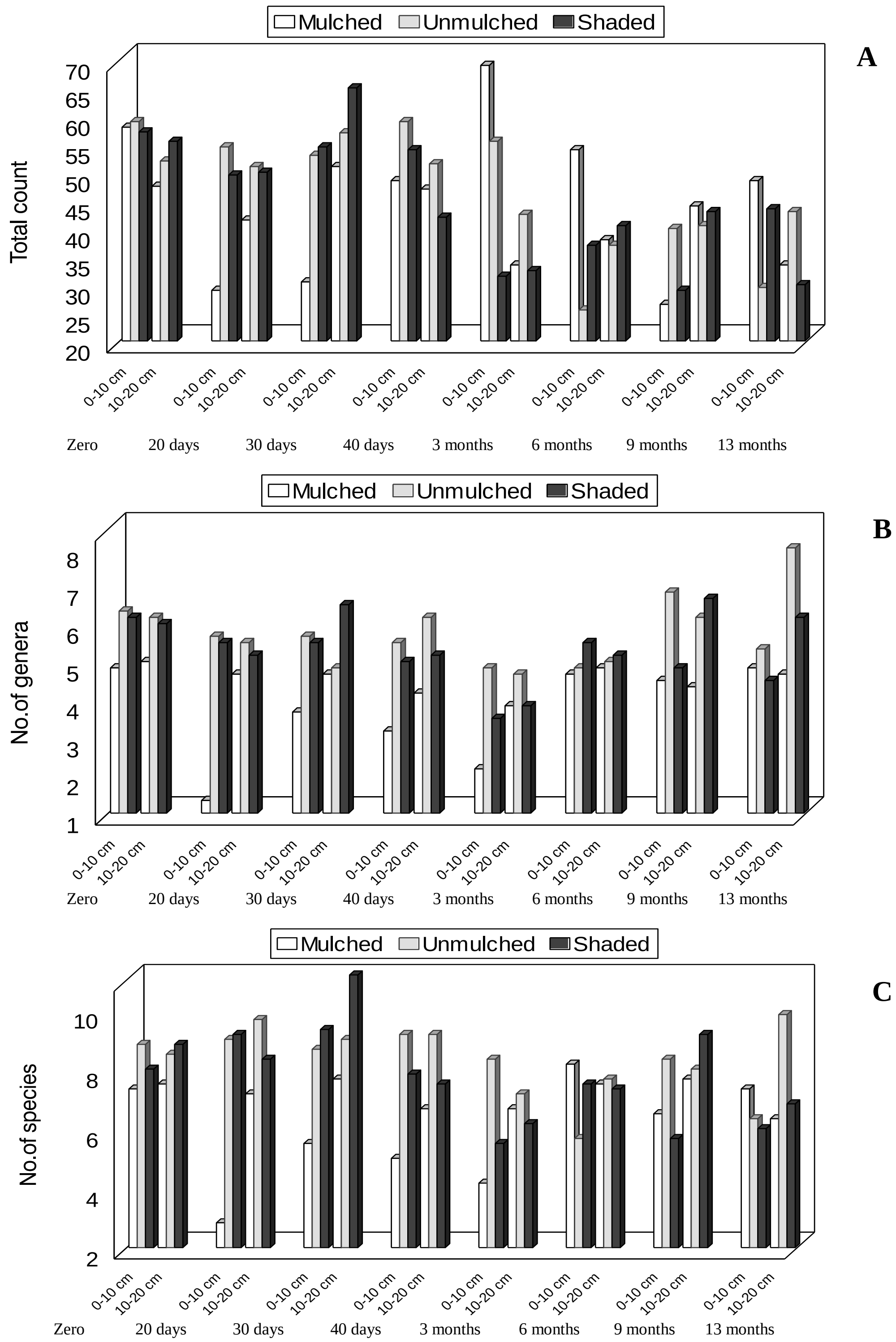

Figure (1) Effect of soil solarization on total count number (A), of genera (B) and number of species (C) of common soil fungi isolated on PDA medium at 28º ± 2º C

c- At the end of solarization period:

In solarized soil, the population density of total fungi was initially (during solarization period) depressed but it rapidly increased at the end of solarization period (Tables 4 and 5). After 13 months, the difference in total count between mulched, unmulched and shaded soils was insignificant. But after 3, 6 and 9 months the significantly higher counts of total fungi were recorded in mulched soil at 0-10 cm. This significance can be attributed to the relatively high population density of *Aspergillus* spp., comprising 86.23, 36.36 and 24.74% of total count of fungi, respectively (Fig. 1a). This result disagrees with the findings of El-Zayat *et al*. (1990) and Stapleton and De Vay (1982).

Over the period 3-13 months, number of genera was not significantly affected in solarized soil at 0–20 cm. But the number of species was significantly reduced in mulched soil (0-20 cm) 3 months after the end of solarization period. However insignificant differences in species number were detected between mulched, unmulched and shaded soils over the period 6-13 months after the end of solarization. due to recolonization of solarized soil by some species such as *Cochliobolus spicifer, Emericella nidulans v. dentata, E. nidulans v. nidulans, E.quadrilineata, E. rugulosa, Melanospora Zamiae and Rhizopus stolonifer* that invaded solarized soil (0-10 cm) at 6 months; *Fusarium dimerum, F.oxysporum* and *Nectria heamatococca* (anamorph) (10-20 cm) at 9 months; and *F. chlamydosporum* (0-10 cm) at 13 months. This variation can be attributed to variation in the degree of injury caused by soil solarization. The injured propagules of a species require a time to recover and germinate. This time varies according to the degree of injury. In agreement with the above explanation pullman *et al*. (1981b) reported that solarization may cause delays in propagules germination that varies with temperature and the duration of exposure.

It is worthy to mention that, the final population of fungi present in solarized soil was nearly the same as the initial population before starting solarization. The gradual return to the initial population composition of the mycocommunity may indicate that the resulting mycocommunity did not represent a climax state. The same indication was previously reported by Stapleton and De Vay (1982).

2-Assay of thermophilic/thermotolerant fungi:

The results in Fig. (2a) show that the total count of thermophilic/thermotolerant fungi was significantly reduced after 40 days of soil solarization. The reduction in total count of these fungi reached 43 and 60% at 0-10 and 10-20 cm depths, respectively as compared with the counts in unmulched soil. This result agrees in part with the findings of Stapleton and De Vay (1982) who studied the effect of soil solarization on population density of soil microorganisms in two sites in California, USA. They found that at one site (Davis site) thermophilic/ thermotolerant fungi showed decreases in population, while they showed increases in populations at the Hickman site. Gamliel and Katan (1991) reported that, thermotolerant fungi and bacteria were reduced at the end of 35-55 days soil solarization period. The reduction in total counts of thermophilic/ thermotolerant fungi obtained after 40 days soil solarization was due to the very reduced count of some fungi. The count of *Emericella* spp. in mulched soil was zero and 11 for *Emericella nidulans v. lata, E. nidulans v. nidulans* and *E. rugulosa.*

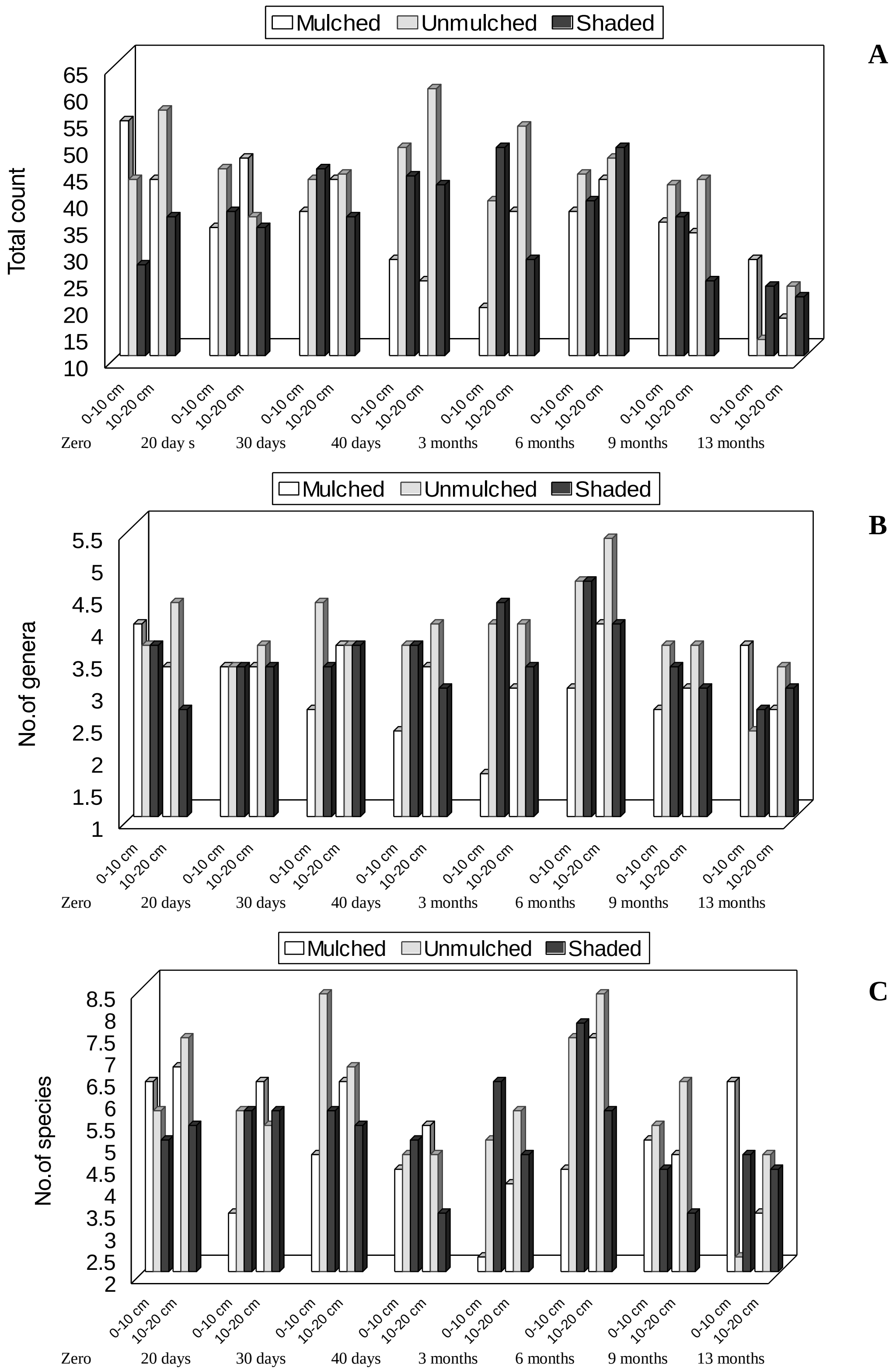

Figure (2) : Effect of soil solarization on total count (A), number of genera (B) and number of species (C) of common soil fungi isolated on YPSs medium at 45º±2º C

Total count of thermophilic/ thermotolerant fungi was still reduced in mulched soil after 3 months (Fig. 2a). This reduction reached 51.3 and 30.2% compared with the corresponding count at 0-10 and 10-20 cm of the unmulched soil, respectively. This reduction was mainly due to reduction of *Emericella* spp. to undetectable count level in mulched soil. Six months after the end of solarization period, the total count of thermophilic/thermotolerant fungi was increased so that differences among mulched, unmulched and shaded soil were insignificant. This result can be attributed to the increase in count of *Aspergillus* spp. (especially *A.fumigatus, A.niger* and *A.terreus*) which represent 51.4 and 41.9% of total count of thermophilic/ thermotolerant fungi at 0-10 and 10-20 cm of mulched soil, respectively.

Number of fungal genera of the thermophilic/thermotolerant fungi (Fig. 2b) was not significantly affected by soil solarization during the period 0-13 months. This was due to *Emericella* was the only genus which was eradicated as a result of soil solarization (after 30 and 40 days and 3 months). In most cases this genus was compensated by appearance of some species which were restricted to solarized soil. *Corynascus sepedonium* appeared at 0-10 cm of mulched soil at 30 and 40 days, and *Thermomyces lanuginosus* appeared at 0-10 cm of mulched soil at 30 days after the end of solarization period.

Number of species of thermophilic/ thermotolerant fungi (Fig. 2c) fluctuated between treatments and depths with a random pattern that its variation can not be attributed to any of these variables.

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التشميس ومكونات المجتمع الفطرى فى تربة بصعيد مصر

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أدى تشميس التربة لمدة ٦ أسابيع فى صعيد مصر إلى ارفع درجة حرارة التربة إلى معدلات تعتبر قاتلة أو مثبطة لمعظم فطريات التربة، وكان نتيجة لذلك أن تغير تركيب المجتمع الفطرى تغيراً ملحوظاً. وكان هذا التغير واضحاً فى التعداد الكلى للفطريات، وكذلك فى عدد الأنواع الفطرية المعزولة على بيئة PDA والتحضين عند ٢٨+٢ °م، وعلى النقيض فإن عدد الأجناس الفطرية المعزولة على نفس البيئة لم تتأثر تأثراً معنوياً بعملية التشميس خلال الفترة ٠-١٣ شهراً.

كما زاد التعداد الكلى للفطريات زيادة معنوية بعد انتهاء فترة التشميس، وأصبح التعداد الكلى للفطريات فى التربة المشمسة أعلى منه فى التربة غير المشمسة أو المظللة، وذلك بعد ٣ شهور من انتهاء فترة التشميس.

كما أدى تشميس التربة إلى اختزال التعداد الكلى للفطريات المحبة أو المقاومة للحرارة والمعزولة على الوسط الغذائى YpSs ، وذلك بعد ٤٠ يوماً من التشميس، كما لم يتأثر تعداد الأجناس والأنواع الفطرية تأثراً معنوياً بعملية التشميس.