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Incidence of Common Bunt (*Tilletia* spp.) in Syria

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ABSTRACT

Field surveys were carried out in main wheat growing governorates during 2005 and 2006, covering major wheat growing areas in several Syrian provinces. Wheat fields were randomly observed and data relative to disease incidence recorded. Results showed that common bunt (*Tilletia* spp.) occurred in all wheat fields surveyed, but low variations in disease incidence were recorded. The highest rate was shown in the first year in Idleb province, while the lowest rate appeared in Daraa province. In the second year, the incidence was recorded in Aleppo and Hassakeh provinces. Results showed that causal agents of common bunt disease were both *T .carries* and *T. foetida,* with different proportions depending upon crop species and field geographical site. The frequencies of *T. caries*: *T. foetida* teliospores were 89.6: 10.4 in durum wheat and 6.3: 93.7 in bread wheat. Pathogenicity of 29 combinations (1:1w) of different Syrian isolates of pathogen species was tested against four durum wheat (*Triticum durum)* and four bread wheat *(Triticum aestivum*) lines which differ in reaction to the disease. A slight difference in spikes' infection rate (SIR) was observed among the tested combinations, where SIR varied between 0 and 56% in durum and between 0 and 80% in bread wheat lines. The collected combinations during 2005 are divided according to their pathogenicity into three groups: Group I= weakly virulent: includes 9 combinations, Group II= moderately virulent: includes 13 combinations and Group III= highly virulent: includes 7 combinations.

Keywords: Survey, Common bunt, Incidence, *Tilletia* spp., Host specificity.

INTRODUCTION

Common bunt, also known as covered smut, is a seed and soil borne wheat disease, caused by *Tilletia caries* (DC.) Tul. and *Tilletia foetida* (Wallr.) Liro. It is potentially one of the most important plant diseases; it occurs in all wheat-growing regions of the world (Hart, 2000; Lipps *et al.*, 2000; Watkins and Prentice, 1997), mainly in West Asia and North Africa (WANA) (Mamluk, 1993; Mamluk and Zahour, 1993). The disease was recorded in Syria, for the first time, in 1958 (Mulder, 1958). Reports showed that 50% of wheat fields were affected with common bunt during the period 1984-1988, with over 60% of infected plants (Mamluk *et al*., 1990). The annual crop losses due to common bunt in north eastern Syria ranged from 5% to 7% in the 1980s (Mamluk *et al*., 1989; Mamluk *et al*., 1990) and 83% of the grain samples have been found polluted by the teliospores of the pathogen (Williams, 1983). In 2001, yield production of the susceptible durum wheat

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cultivar ACSAD 299 has reached up to 41% when infected with 1:1 teliospores mixture of *T.foetida* and *T.caries* (Al-Chaabi and Matrod, 2005a). It was reported that bunt balls are the basic source of the primary infection in Syria (Mamluk *et al*., 1989). Bunt spores could maintain their vigor in the soil for over two years (Goates, 1996; Hoffman, 1982). The use of resistant varieties is the most feasible approach for managing the disease (Wilcoxon and Saari, 1996). The wheat cultivar, Taro3, was found to be resistant under artificial inculcation in Syria (Al-Chaabi and Matrod, 2005b).

The objectives of the study were 1: to assess the current situation of CB in the Syrian wheat fields, 2: to characterize fungal isolates and 3: to assess their virulence spectrum of *Tilletia* spp. against *Triticum* species and cultivars.

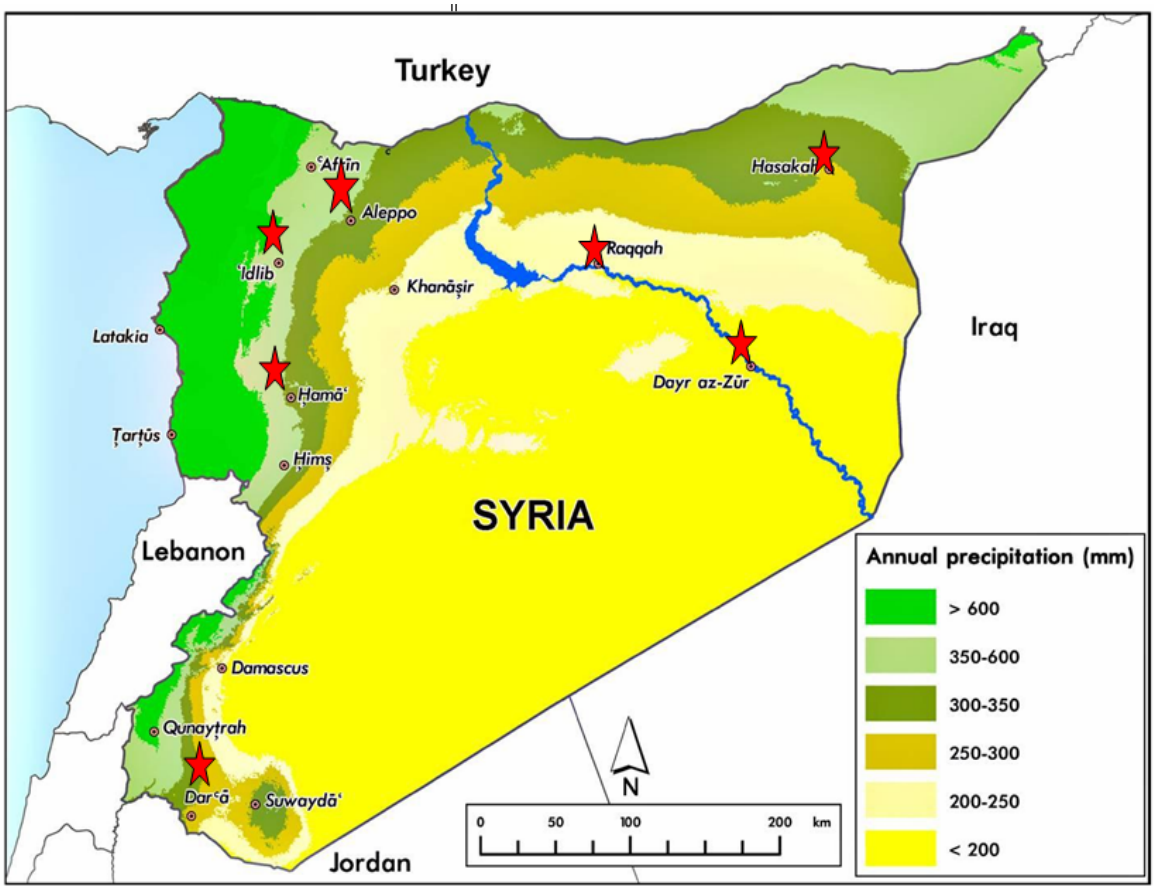

Figure 1. A survey of common bunt disease in major wheat growing provinces in Syria (★ : source).

The survey covered major wheat growing provinces in Syria and was carried out during late May – early June of 2005 and 2006, at maturity stage. Locations (longitudes and latitudes) of fields were projected on a map of Syria utilizing GPS. Disease samples were collected from three points along the field diameter, where a 1x1m wooden frame was used to assess bunted spikes and associated pathogens. The disease incidence was randomly observed as spike infection rate (SIR) in bread and durum wheat. Results were analyzed and the

least significant difference (LSD) was calculated at the level of 0.05 using Genstat 7. Ten randomly infected spikes were collected from each field and tested in the laboratory to identify the pathogen species in each single spike. The bunt balls in each spike were grounded and examined under light microscopy for the determination of *Tilletia* species.

The pathogenicity test of isolated *Tilletia* species was carried out using 4 bread wheat lines (Table 1): Kapsw/Smuha-17**,** Star's/Florkwa-2, Cham-6/Tui's, Cham-4/Guhrab-1 and 4 durum wheat lines (Table 1): Bigost-1, Ammar, Azeghar N2 and Ombit; representing different reaction types to common bunt (Mamluk and Zahour, 1993). These lines were screened under artificial inoculation in pots (25 cm diameter) containing a mixture of soil: beat moss (1:3 v). The inoculum consisted of a mixture (1:1w) of teliospores of both pathogenic fungi (Table 1). The amount of used inoculum was in the rate of 1g of teliospores' mixture per 100g seeds (Gaudet *et al.*, 1989), where seeds and inoculum were then rubbed by hand for one minute. Inoculated seeds were sown in pots in the rate of 4 seeds/ cultivar and 4 cultivars/pot, in three replicates. Healthy seeds of tested cultivars were also sown as checks. Results were recorded as SIR and pathogen species.

Table 1. Weight of 100 kernels of tested wheat cultivars and rate of applied inoculum from each pathogen (*T. caries* and *T. foetida*).

Wheat cultivars	Variety reaction to common bunt	100 kernels weight (g)	Inoculum weight for each species (g)
Durum wheat			
1. Bigost-1	Very Susceptible	4.2	0.025
2. Ammar	Susceptible	4.5	0.027
3. Azeghar N2	Moderate resistant	4.8	0.028
4. Ombit	Resistant	5.1	0.030
Bread wheat			
5. Kapsw/Smuha-17	Very Susceptible	3.3	0.019
6. Star's/Florkwa-2	Susceptible	3.0	0.018
7. Cham-6/Tui's	Moderate resistant	3.8	0.022
8. Cham-4/Guhrab-1	Resistant	3.7	0.022

RESULTS AND DISCUSSION

Field Survey

The survey covered 339 and 231 wheat fields in 2005 and 2006, respectively; distributed as 220 and 151 durum wheat fields, 119 and 80 bread wheat fields, in both seasons, respectively (Table 2). Results showed that common bunt was found in all studied provinces. In 2005, 54 fields were found infested (Aleppo 5, Idleb 4, Hama 4, Hasakeh 6, Deir Ezzor 2, Raqqa 4, Daraa 29), but in 2006 this number was reduced to 27 fields only (10, 0, 5, 8, 0, 0, 4, respectively) as common bunt was not recorded. Different incidence levels were associated with wheat species and variety assessed in the survey.

Significant infection levels were detected. The

highest infection rate was recorded in the first season in Idlib on durum (14.5%) and bread wheat (32.2%), whereas the lowest infection rate was obtained in Daraa (3.8% and 8.8% in durum wheat and bread wheat, respectively) (Table 3). In the second season, a highest infection rate was recorded in Aleppo (15.5% and 25.1% on durum wheat and bread wheat, respectively), while the lowest infection was recorded in Hasakeh, on durum wheat (3.7%) (Table 3).

Table 2. Survey of common bunt disease in major wheat growing provinces in Syria during 2005 and 2006 growing seasons.

Province	Season	Total of fields surveyed	Durum wheat	Bread wheat	No. of infected fields	Durum wheat	Bread wheat
Aleppo	2005	91	55	36	5	3	2
	2006	60	39	21	10	4	6
Idleb	2005	68	49	19	4	2	2
	2006	25	17	8	0	0	0
Hama	2005	43	25	18	4	2	2
	2006	45	40	5	5	5	0
Hasakeh	2005	33	25	8	6	6	0
	2006	32	11	21	8	3	5
Deir Ezzor	2005	28	23	5	2	2	0
	2006	17	13	4	0	0	0
Raqqa	2005	37	23	14	4	2	2
	2006	22	14	8	0	0	0
Daraa	2005	39	20	19	29	19	10
	2006	30	17	13	4	3	1

Table 3. Average spike infection rate (SIR) in wheat common bunt in different provinces during the cropping seasons 2005 and 2006 in Syria.

Provinces	2005 (2006) Infection average %	
	Durum wheat	Bread wheat
Aleppo	6.7^{de} (15.5^{a})	18.3^{c} (25.1^{a})
Idleb	14.5^{a} (0.0)	32.2^{a} (0.0)
Hama	12.0^{ab} (11.1^{b})	28.3^{b} (0.0)
Hasakeh	10.9^{bc} (3.7^{d})	0.0 (12.3^{c})
Deir zzor	8.5^{cd} (0.0)	0.0 (0.0)
Raqqa	6.7^{de} (0.0)	15.3^{cd} (0.0)
Daraa	3.8^{ef} (8.0^{c})	8.8^{e} (18.3^{b})
LSD(0.05)	0.38 (0.81)	0.66 (0.41)

In the 2005 cropping season, the highest SIR was recorded in Idleb, where the average prevailing soil temperature during December was 6.3 ^{0}C (Table 4a), which was accompanied by 87 mm rainfall. This environment appears to be the favorite or most appropriate [high soil moisture and low temperature (5-10 ^{0}C)] for teliospores' germination, bazidiospores' development and host infection (Wilcoxson *et a*l., 1996). The lowest SIR was recorded in Daraa, where the temperature in December (11.3^{0}C) was also favorable for pathogen development and wheat seedling infection, but soil moisture was low in this province and most probably seeds as well as teliospores did not germinate. The spikes' infection rate (SIR) variation between and within provinces could be attributed to many factors, such as soil temperature and soil moisture (rainfall amount) during seed germination, which affect the rate of infection (Veisz *et al.*, 1997). This is also associated with: wheat species, grown variety, pathogen species, its virulence and availability in the soil, seed sanitation and depth of planting. In Syria, wheat sowing occurs during November–December– early January, where the soil-temperature during the two seasons of the study ranged between 5.4 and 16.7^{0}C (Table 4a). With the exception of some sites, the temperature of locations was however suitable for infection. As for rainfall, it was generally much lower in the second season compared with the first growing cycle, particularly in Idleb, Raqqa and Deir Ezzor (Table 4b) which could have had negative effects on host growth and infection, pathogen development and consequently the value of SIR, which was 0% in these three provinces (Table 3).

Table 4a. Monthly average of air temperatures (^{0}C) of the Syrian provinces covered by the common bunt survey during 2005 and 2006 crop seasons.

Province	Season	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	June
Aleppo	2005	22.2	13.31	5.4	6.3	7.4	11.6	16.3	20.9	25.6
	2006	18.2	11.6	9.3	7.7	9.3	12.6	17.1	17.1	22.3
Hama	2005	23.5	14.5	6.7	7.9	9	14.8	19.3	24.4	28.8
	2006	20.2	12.3	9.8	8.6	10.6	14.7	18.9	18.9	25
Hasakeh	2005	22.7	12.7	5.7	6.0	7.7	13	19.4	22.7	30.5
	2006	21.1	12.3	10.5	7.7	9.6	14.2	17.9	17.9	26.2
Daraa	2005	21.3	16.7	11.3	9.5	10.4	14	16.7	20.4	32
	2006	21	10.2	10.2	7.9	9	11.6	15	15	20.7
Idleb	2005	22.7	15.5	6.3	7.1	7.9	12.7	17.2	21.8	26.1
	2006	19.5	13.7	8.7	8.8	9.1	13.6	16.5	19.5	24.3
Raqqa	2005	21.4	13	5.4	5.6	8	13.2	18.3	22.5	27.3
	2006	20.5	11.9	5.1	5.3	7.7	12.8	17.9	21.8	26.4
Deir Ezzor	2005	22.4	13.1	5.9	6.8	8.8	13.7	19.8	24.2	29.3
	2006	21.5	12.7	5.5	6.4	8.3	13.4	19.6	23.9	28.6

Table 4b. Monthly average rainfall (mm) of the Syrian provinces surveyed during 2005 and 2006 crop seasons.

Province	Season	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	June	Total
Aleppo	2005	0.0	91.5	51.3	58.3	44.0	29.2	33.7	4.9	1.0	313.9
	2006	28.2	38.2	13.8	76.7	41.9	77.5	24.1	0.4	0.0	300.8
Hama	2005	11.5	106.4	29.3	70.1	84.6	20.5	16.5	0.9	0.0	339.8
	2006	23.9	43.9	56.4	115.0	38.6	23.7	37.8	1.9	0.0	341.2
Hasakeh	2005	2.1	97.8	4.9	30.1	63.1	16.3	4.7	10.9	1.6	231.5
	2006	10.5	16.9	53.3	57.9	4.6	41.6	0.0	2.1	0.0	186.9
Daraa	2005	3.2	79.6	2.9	39.2	143.8	8.0	6.2	0.0	0.0	282.9
	2006	19.2	25.0	98.4	42.6	72.1	39.8	6.6	2.3	0.0	306.0

Province	Season	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	June	Total
Idleb	2005	18.0	37.5	87.0	110.4	113.5	26.0	15.3	20.0	0.0	427.7
	2006	18.2	35	90.1	109.4	113.8	26.2	15.4	15	0.0	423.1
Raqqa	2005	25.0	35.8	17.0	21.3	23.4	7.6	9.4	3.3	0.0	143.3
	2006	26.4	40.2	19.8	22.9	30.1	10.2	11.3	7.1	0.0	168
Deir Ezzor	2005	5.0	56.9	7.5	21.8	40.3	11.2	2.9	11.2	0.0	151.8
	2006	8.2	65.3	8.9	22.5	40.7	11.7	3.1	7.6	0.0	168

Pathogens Frequency/ Host Specificity

A total of 290 spikes (out of 664 collected randomly from different locations) were microscopically examined. Results revealed the presence of both *Tilletia* species: *T. caries* and *T. foetida*, with variation in their geographical distribution and in host specificity as well (Table 5). In the Northern provinces (Aleppo and Idleb), both types of the causal agent were found on both hosts: durum wheat and bread wheat. *T. caries* was, however, more prevalent on durum wheat in Aleppo (92.5%) and Idleb (78.1%). In contrast, *T. foetida* was more common on bread wheat in both provinces (83.3% and 84.6%, respectively).

In the central province Hama, bunted spikes were sampled from durum wheat and bread wheat cultivars and *T. caries* was prevalent on durum wheat (86.6 %); whereas *T. foetida* prevailed on bread wheat (80%). In Hassakeh and Deir Ezzor provinces, samples were collected only from durum wheat where the prevalence of *T. caries* species varied from 80-100%. The microscopic examination of teliospores collected from durum wheat and bread wheat spikes in Raqqa and Darraa provinces revealed that *T. caries* was common on durum wheat at a rate of 92.5% and 93.7%; meanwhile *T. foetida* was prevalent on bread wheat at a rate of 87.5% and 69.2%, respectively. The distribution of *T.caries* and *T.foetida* was not only geographically restricted, but also was dependent on the host wheat species. In this study, *T.caries* was found to be the dominant pathogen on durum wheat, whereas *T.foetida* was the most prevalent pathogen on bread wheat, irrespective of the area where the respective wheat types were grown. These findings indicate that there is a specific host preference by the pathogens.

The study showed that *T.caries* in Northern, Central and Southern regions (Aleppo, Idleb, Hama, Raqqa and Daraa) was found to attack both durum wheat and bread wheat and this finding disagrees with (Mamluk, 1993) but it is in agreement with (Mamluk *et al*., 1990). However, Deir Ezzor results have conformed with Mamlouk and Zahour (1993), where *T. caries* was found to attack only durum wheat in many countries of the Middle East and North Africa. This study has concluded that *T. foetida* was mainly found on bread wheat and *T. caries* was prevalent on durum wheat; hence its distribution in Syria could be affected more by wheat species rather than by climatic/geographical factors.

Table 5. Frequency of geographical distribution / host specificity of wheat common bunt pathogens *T. caries* and *T. foetida*, in 2005 in Syria.

Province	Number of checked spikes	Durum wheat			Bread wheat		
		Number of spikes	*T. caries* %	*T. foetida* %	Number of spikes	*T. caries* %	*T. foetida* %
Aleppo	50	32	78.1	21.9	18	16.6	83.4
Idleb	40	27	92.6	7.4	13	15.4	84.6
Hama	40	30	86.6	13.4	10	20	80
Hasakeh	60	60	80	20	0	0	0
Deir Ezzor	20	20	100	0	0	0	0
Raqqa	40	32	93.7	6.3	8	12.5	87.5
Daraa	40	27	92.6	7.4	13	30.8	69.2

Pathogenicity

This experiment was carried out in 2005**,** using 29 combinations of the Syrian common bunt isolates (*T. caries* and *T. foetida*), tested for their pathogenicity on 4 durum wheat and 4 bread wheat cultivars, known for their reaction types (R, MR, S and VS). The isolates collected during 2005 were divided, according to their pathogenicity, into three groups: Group I= weakly virulent: includes 9 combinations, Group II= moderately virulent: includes 13 combinations and Group III= highly virulent: includes 7 combinations (Table 6a &6b). Slight infection was induced by each of K6 and R14 inoculum combinations on the resistant bread wheat variety, Cham-4/Ghurab-1 (SIR=5.9 and 5.3, respectively). The SIR ranged between 0 and 56 % in durum wheat and between 0 and 80 % in bread wheat cultivars. Results of this study showed that the variations in *Tilletia* species pathogenicity on durum wheat and bread wheat varieties contradict with some studies (Calvo, 1987) but were in agreement with others (Magnus and Storli, 1979). The study confirmed the possibility of infecting durum wheat and bread wheat germplasms by the two common bunt *Tilletia* species (Goates, 1996). Some studies showed the possibility of finding spores of both pathogenic fungi on the same plant (Nyvall, 1989), with a proportion of about 3% (Mamluk *et al*., 1990). A recent study showed that 90.8% of bunted balls (sori) randomly collected from durum wheat cultivar "ACSAD 299" artificially inoculated with a mixed inoculum of both pathogenic fungi *T. caries* and *T. foetida* (1:1) were infected with both pathogens, 6.5% of sori were infected with *T. caries*, whereas 2.7% of sori were infected with *T. foetida* (Al-Chaabi and Matrod, 2005a). *T. caries* and *T. foetida* have been hybridized and can produce a full range of morphological variants (Holton, 1942).

Table 6a. Average spike infection rate (SIR) on durum wheat and bread wheat cultivars inoculated artificially with a mixture of teliospores of *T.caries* and *T.foethda*, Syria, 2005/2006.

Source of isolates	Code of isolates of both pathogens	Average spike infection rate, SIR %							
		Durum wheat				Bread wheat			
		HS	S	MR	R	HS	S	MR	R
Hasakeh	K6 (*T. caries* + *T. foetida*)	55.6	50.0	30.2	0.0	73.0	63.0	32.0	5.9
Raqqa	R14 (*T. caries* + *T. foetida*)	40.0	33.3	28.6	0.0	78.0	64.0	33.0	5.3
	R15 (*T. caries* + *T. foetida*)	42.9	41.7	30.0	0.0	80.0	69.0	38.0	0.0
Idleb	I16 (*T. caries* + *T. foetida*)	35.3	29.4	25.0	0.0	62.0	56.0	36.0	0.0
Hama	H24 (*T. caries*+ *T. foetida*)	41.7	33.3	25.0	0.0	80.0	69.0	44.0	0.0
	H25 (*T. caries*+ *T. foetida*)	33.3	30.0	28.6	0.0	63.0	58.0	42.0	0.0
Daraa	D27 (*T. caries*+ *T. foetida*)	41.2	35.7	27.3	0.0	67.0	55.0	38.0	0.0
Check (Healthy control)		0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Standard error of mean		2.71	2.77	0.18	0.0	0.0	2.2	1.66	1.04

Durum wheat: HS = Bigost-1, S = Ammar, MR = Azeghar N2, R = Ombit.

Bread wheat**:**HS = Kapsw/Smuha-17, S = Star's/Florkwa-2, MR = Cham-6/Tui's, R = Cham-4/Guhrab-1.

Table 6b. Number of infected spikes on durum wheat and bread wheat cultivars inoculated artificially with a mixture of teliospores of *T.caries* and *T.foethda*, Syria, 2005/2006.

Source of isolates	Code of isolates of both pathogens	Durum wheat			Bread wheat		
		No. of infected spikes tested	No. of spikes infected with		No. of infected spikes tested	No. of spikes infected with	
			T.caries	*T.foetida*		*T.caries*	*T. foetida*
Hasakeh	K6 (*T. caries* + *T. foetida*)	14	12	2	28	2	26
Raqqa	R14 (*T. caries* + *T. foetida*)	14	13	1	17	1	16
	R15 (*T. caries* + *T. foetida*)	14	14	0	20	3	18
Idleb	I16 (*T. caries* + *T. foetida*)	14	12	3	18	0	18
Hama	H24 (*T. caries*+ *T. foetida*)	13	11	2	21	2	19
	H25 (*T. caries*+ *T. foetida*)	12	12	0	22	0	22
Daraa	D27 (*T. caries*+ *T. foetida*)	16	14	2	19	1	18
Standard error of mean		0.46	0.43	0.43	1.38	0.45	1.27

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الإصابة بمرض التفحم المغطى في محصول القمح في سورية

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ملخـص

نفذ المسح الحقلي خلال عامي 2005 و 2006 ، حيث شمل المناطق الرئيسة الأكثر زراعة لهذا المحصول في المحافظات السورية. حددت الحقول بشكل عشوائي، وحسبت نسبة الإصابة في كل حقل على حدة.
أظهرت نتائج المسح الحقلي انتشار مرض التفحم المغطى (*Tilletia* spp.) في كافة المحافظات المشمولة بالدراسة، مع وجود تباين معنوي بينها في نسبة الإصابة. خلال الموسم الأول سجل أعلى متوسط نسبة إصابة في محافظة ادلب بينما كان أدنى متوسط في محافظة درعا، وفي الموسم الثاني سجل أعلى متوسط نسبة إصابة في محافظة حلب وأدنى متوسط في محافظة الحسكة.
وأظهرت النتائج انتشار الفطرين *T. caries* و *T. foetida* على القمح في كل المحافظات المدروسة، مع وجود اختلاف نسبي تبعاً للتفضيل العوائلي والتوزع الجغرافي لكل منهما. وبلغت نسبة تردد الأبواغ التيلية للفطرين خلال عامي الدراسة في القمح الصلب (89.6 - 10.4 %). أما في القمح الطري فبلغت (6.3 -93.7 %).
اختبرت القدرة الإمراضية لـ 29 تركيباً خليطاً من كلا الفطرين (1:1 وزن) جمعت من حقول المواقع المدروسة، وذلك إزاء 4 طرز لكل من نوعي القمح الصلب (*Triticum durum*) والطري (*Triticum aestivum*) تمثل ردود فعل متباينة (R، MR، S و VS)، وأظهرت النتائج تبايناً في نسبة الإصابة تبعاً لتراكيب اللقاح المعدي؛ إذ تراوحت على الصنف VS من القمح الصلب والطري بين 0 و 56% وبين 0 و 80%، على التوالي. وبناء على ذلك، تم تقسيم تراكيب الفطرين الممرضين إلى ثلاثة مستويات تبعاً لقدرتها الإمراضية:
المجموعة الأولى: ضعيفة الشراسة، ضمت 9 تراكيب.
المجموعة الثانية: متوسطة الشراسة، ضمت 13 تركيباً.
المجموعة الثالثة : عالية الشراسة، ضمت 7 تراكيب.

الكلمات الدالة: المسح الحقلي، مرض التفحم المغطى، التفضيل العوائلي، القمح، سورية.

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