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LEAF RUST ASSESSMENT IN FOUR EGYPTIAN BREAD WHEAT CULTIVARS WITH THEIR SUBSEQUENT PHYSIO-HISTOLOGICAL CORRELATIONS

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ABSTRACT: *Leaf rust caused by Puccinia triticina Eriks is considered one of the most three important worldwide disease of wheat causing regular and significant losses over large geographical areas. Four bread wheat cultivars i.e. Gemmiza-7, Misr-1, Sids-1 and Sids-12 were assessed for their disease parameters, yield measurements, physiological and histological features. Assessments were carried out during 2012/13 and 2013/14 growing seasons, under realistic field conditions of Kafr El-Hamam Agricultural Research Station in El-Sharkia governorate, ARC, Egypt. According to their infection responses, Misr-1 and Sids-12 were considered as resistant cultivars. Meanwhile, Gemmiza-7 and Sids-1 were susceptible. Different correlations between disease parameters and some physio-histological features were performed. Resistant wheat cultivars recorded the lowest values of disease percentages, average coefficient of infection (ACI) and area under disease progress curve (AUDPC) values. These results were related to the free phenol contents and average number of stomatae/mm^2 by a direct correlation. These relations were reversed in total and conjugated phenols, peroxidase activity, chlorophyll contents, distance between stomatal rows, schlerenchyma thickness, and laminar bundles parameters. All the aforementioned correlations were completely reversed in the susceptible wheat cultivars. Also, a correlation between chlorophyll values and yield/plot was observed. The resistant Sids-12 wheat cultivar recorded the highest values of chlorophyll content with the subsequent highest values of yield/plot which compatible for the lowest percentage of yield loss (0.32 %), during 2012/13 growing season. While, the highest percentage of yield loss (23.73%) was recorded by the susceptible Sids-1 wheat cultivar, during 2012/13 growing season. Selected parameters used in the present work can be considered as a base for future investigations of leaf rust status on some commercial Egyptian bread wheat cultivars.*

Key words: *Wheat; Leaf rust; Losses; Physio-histological features.*

INTRODUCTION

Wheat plant (*Triticum aestivum* L.) is one of the first domesticated food crops, cultivated since the start of settled agriculture 10,000 years ago. Today, wheat is unrivalled in its range of cultivation. It is grown annually on roughly 200 million hectares with an average production of 600 million tons, providing 16% of total dietary calories consumed globally (Dixon *et al*., 2009). The cultivated area of wheat in Egypt is about 1.3 million hectares (3.1 million feddan) with an average production of 8 million tons according to the Ministry of Agric. statistics (Anon., 2012). Plant diseases affect 55% of the global wheat growing area, causing an estimated loss of 20 million tons of wheat per year (Kosina *et al*., 2007). Among the wide array of microorganisms causing diseases on wheat, the leaf rust fungal disease, caused by *Puccinia triticina*, is considered among these pathogens. *P. triticina* has a wide virulence range and is broadly adapted to diverse climatic conditions, leading to regular and significant yield losses over large geographical areas. Many research workers have reported reductions in grain yield and quality due to the primary effects of leaf and stem rust infections (Keed and White, 1972; Huerta *et al*., 1993; Sayre *et al*., 1998). Periodic leaf rust epidemics have occurred in most decades of the last century (Espino *et al*., 2011). Thus leaf rust is considered an important worldwide disease of wheat. Egypt is a part of the epidemiological zone of leaf rust (Saari and Prescott, 1985). In Egypt, yield losses of wheat due to leaf rust could reach 50% (Abdel-Hak *et al*., 1980). Such disease eliminated many commercial wheat cultivars, cultivated in Egypt, such as Giza-139, Chenab-70, Super-X, Giza-158 and Giza-160 (Nazim *et al*., 1983) with the subsequent commercial release of other cvs. such as Misr-1, Sids-12, Gemmiza-7 and Sids-1 in 2011, 2007, 1999 and 1995 years, respectively (Abdelbacki *et al*., 2013).

The final intensity of disease infection was a better criterion to select the resistant cultivars measured as area under disease progress curve (Gulati *et al*., 1985). This parameter value can be measured in the field through recording disease severity at weekly intervals (Wilcoxson *et al*., 1975). Physiological studies help the researchers in better understanding the correlation between resistance and biochemical component of the plants such as peroxidase, total, free and conjugated phenols as well as chlorophyll contents. Various studies suggested that, involvement of plant peroxidase not only in biosynthetic processes related to wall development, but also in resistance against infection by pathogens (Young *et al*., 1995 and Chittoor *et al*., 1997). Also, phenolics are well-known antifungal, antibacterial and antiviral compounds occurring in plants (Sivaprakasan and Vidhyasekaran, 1993). According to Matern and Kneusal (1988), the first step of the defense mechanism in plants involves a rapid accumulation of phenols at the infection site, which restricts or slows the growth of the pathogen. In recent years, there has been growing interest in the outermost layers of the plant as well as the internal tissue patterns. The structural features of epidermis and internal tissue patterns may greatly affect the ability of the pathogen to invade plant. Resistance mechanism act against the infection or the subsequent spread of the pathogen in the plant (Gelalcha and Hanchinal, 2003). Surface structures of a plant might play a role as mechanical barriers. In this respect, stomata on the leaf surfaces serve as entrance pathways of the urediospores of rust fungi. In addition, there are various types of hairs originating from the epidermal cells which usually protrude from the leaf and/or the stem (Lazniewska *et al*., 2012).

The objectives of the present study are to correlate the leaf rust disease parameters of four Egyptian bread wheat cultivars with yield measurements, physiological and histological features, under realistic field weather conditions during two successive growing seasons (2012/13 and 2013/14).

MATERIALS AND METHODS

The current investigation was carried out in El-Sharkia governorate at Kafr El-Hamam Agricultural Research Station, ARC, Giza, Egypt during 2012/13 and 2013/14 growing seasons. Four cultivars (Gemmiza-7, Misr-1, Sids-1 and Sids-12) of bread wheat were adopted. Grains were obtained from Field Crop Research Institute, Giza, Egypt. The urediospores of *Puccinia triticina* were kindly provided by Wheat Dis. Res. Dep., Plant Pathol. Res. Inst., ARC.

Area of experiment was divided into two parts. All tested cultivars were cultivated in the two parts. One part was infected by the leaf rust pathogen urediospores while the other part was used as protected one. Plots were kept protected almost free from leaf rust using the fungicide Sumi-eight [(E)-(Rs)-1-(2,4 dichlorophenyl)-4,4- dimethyl-2- (1H-1,2,4- triazol-1-yl)- Rent-1- en-3-ol (IUPAC)] obtained from Sumitomo Chemi-Co at 0.35 ml/l. Wheat cultivars were grown in a randomized complete block design with three replicates. Plot size was 2.0 × 2.0 m, each plot contained six rows with 2 m long and 30 cm between rows. Plots were surrounded by spreader area planted with a highly leaf rust susceptible wheat genotype (Morocco) to spread inoculum. Artificial inoculation with a mixture of freshly collected urediospores of the most prevalent leaf rust physiologic races and a talcum powder at a ratio of 1/20 (v/v) was carried out at 70 days old plants using baby cyclones to assure equal deposition of spores on all spread plants and to generate an epidemic (Tervet and Cassel, 1951).

I. Disease parameters

1. Disease severity (DS)

Disease severity was estimated using the modified Cobb's scale adopted by Peterson *et al*. (1948).

2. Infection type (IT)

Infection type was detected using the adopted type scale of Saari and Wilcoxson (1974) where resistant = R, moderately resistant = MR, moderately susceptible = MS and susceptible = S.

3. Average coefficient of infection (ACI)

Average coefficient of infection was calculated by multiplying the percentage of the disease severity with infection type using scale of Saari and Wilcoxson (1974).

4. Area under disease progress curve (AUDPC)

Area under disease progress curve (AUDPC) was calculated for each cultivar using the equation of Pandey *et al*. (1989):

$$AUDPC = D\left[\frac{1}{2}(Y_1 + Y_K) + (Y_2 + Y_3 + \ldots Y_{K-1})\right]$$

Where:

D = days between two consecutive recordings (time intervals),

Y_1 + Y_K = sum of the first and the last disease scores and

Y_2 + Y_3 + . . . + Y_{K-1} = sum of all in between scores.

II. Yield measurements:

At the harvest stage, Yield measurements (weight of 1000 grains (g) and yield/plot (kg)) were weighted after harvesting from random samples of three plants for each replicate. The influence of leaf rust severities on yield was determined by comparing the yield of infected and protected cultivars. Yield loss was estimated using the simple equation as follows:

$$Loss\,\% = \left(1 - \frac{Y_i}{Y_p}\right) \times 100$$

suggested by (Colpauzos *et al*., 1976)

Where: Y_i = yield of infected plants.

Y_p = yield of protected plants.

III. Physiological analysis:

1- Determination of phenolic contents:

Free and total phenols were extracted from fresh blades flag leaves of wheat plants infected by *P. triticina* and those of wheat plants with the fungicide Sumi-eight after five days from inoculation. Phenols were determined and expressed as mg/g fresh weight according to the method described by Snell and Snell (1953), during 2013/14 growing season. In this method, Folin Denis reagent was used as described by Gutfinger (1981), and the color density was read in spekal -spectrocolorimeter (Carlzeizs jena) at 520 nm. Concentration of conjugated phenols was determined from the difference between total and free phenols.

2. Peroxidase activity:

Enzymatic activity of peroxidase (PO) was determined from fresh blades flag leaves of wheat plants infected by *P. triticina* and protected wheat plants using the fungicide Sumi-eight after five days from inoculation, during 2013/14 growing season. Peroxidase was determined as described by Allam and Hollis (1972). One unit of PO activity was expressed as the change in absorbance at 425 nm/minute/g fresh weight.

3. Chlorophyll content:

Flag leaves of the tested four cultivars (either infected by *Puccinia triticina* or protected by Sumi-eight) were used to estimate the total chlorophyll contents using SPAD-502 apparatus (Castelli *et al*., 1996), during 2013/14 growing season. The SPAD-502 meter is a hand-held device that is widely used for the rapid, accurate and non-destructive measurement of leaf chlorophyll concentrations. It has been employed extensively in both research and agricultural applications, with a range of different plant species.

IV. Histological structure:

1. Number of stomata:

To calculate number of stomata, specimens from the blades of flag leaves on the main stems obtained from four tested wheat cultivars at booting stage (70 day) were examined during 2013/14 growing season. The upper epidermis was prepared by scraping lower and mesophyll tissue till we had obtained the upper epidermis. Then they were examined by using light microscope (Olympus) with digital camera (Canon power shot S80) connected to computer. The photographs were taken by Zoom Browser Ex Program. Number of stomata per one square millimeter calculated as average of 10 reading from 5 slides using Image J 1.47v program.

2. Leaf blade cross section:

Samples of four tested cultivars of bread wheat were collected from the blades of flag leaves on the main stems. These samples were obtained at booting stage through the second growing season (2013/14). Samples were cut into pieces of 1.0 cm length. These pieces were fixed in standard FAA fixation solution (5 ml of formalin, 20 ml of 70% alcohol and 5 ml of acetic acid) for 24 hrs. The selected materials were washed in 70% alcohol, dehydrated in a normal butyl alcohol series, embedded in paraffin wax of melting point of 54-56°C, sectioned to a thickness of 12 µm size with EPMA rotary microtom and double stained with Safranin and Fast-green (Nassar and El-Sahhar, 1998). The stained sections were examined and photographed using light microscopy as previously mentioned in number of stomata.

Statistical analysis:

Obtained data were analyzed in combined analysis of Randomized Complete Block Design (R.C.B.D.) to study the interactions between tested cultivars (Snedecor and Cochran, 1980). Mean values were compared at $P< 0.05$ using the least significant different (LSD).

RESULTS AND DISCUSSION

Gemmiza-7, Misr-1, Sids-1 and Sids-12 wheat bread cultivars were screened for adult plant response against leaf rust caused by *Puccinia triticina*, under field conditions. This screening was carried out including disease parameters, yield measurements, physiological analysis and anatomical features of wheat cvs.

I. Disease parameters

Leaf rust parameters severity (%), infection type (IT), average coefficient of infection (ACI) and area under disease progress curve (AUDPC) were determined. Data in Table (1) categorized the four tested bread wheat cultivars into two groups according to their infection responses. The first group included Misr-1 and Sids-12 representing the resistant wheat cultivars. The second group included Gemmiza-7 and Sids-1 representing the susceptible wheat cultivars. The most pronounced observation in the first group was that Misr-1 showed complete resistance for both infected and protected treatments, during the two growing seasons. Meanwhile the infected adult plants of Gemmiza-7 and Sids-1 recorded a high disease severity with S infection type, during the two growing seasons. Such reaction was improved to MS with lower disease severity, during the two growing seasons, when adult plants were protected by Sumi-8 fungicide.

Regarding the values of ACI and AUDPC, data in Table (1) also indicated that Misr-1 recorded the lowest values of such two parameters. The complete resistance behavior in Misr-1 cultivar may be due to the presence of *Lr*19 gene (Abdelbacki *et al*., 2013) which is the most widely distributed gene for resistance to *P. triticina* (McIntosh *et al*., 1995 and Winzeler *et al*., 2000).

Statistically analyzed data which presented in Table (1) revealed that years (Y), cultivars (C) and treatments (T) were significant sources of variation in percentage of DS, ACI and AUDPC. Also, these parameters were significantly affected by interaction between years of application, cultivars used and treatments applied.

Table (1): Leaf rust parameters of four adult wheat cultivars either infected by *Puccinia triticina* or protected by Sumi-8, during 2012/13 and 2013/14 growing seasons.

Cultivar	Treatment	DS (%)		IT		ACI		AUDPC	
		2012/13	2013/14	2012/13	2013/14	2012/13	2013/14	2012/13	2013/14
Misr-1	Infected	0.0	0.0	0	0	0.0	0.0	0.0	0.0
	Protected	0.0	0.0	0	0	0.0	0.0	0.0	0.0
Sids-12	Infected	0.0	3.0	0	MR	0.0	1.2	0.0	27.0
	Protected	0.0	0.0	0	0	0.0	0.0	0.0	0.0
Gemmiza-7	Infected	70	70.0	S	S	70.0	70.0	764.0	868.33
	Protected	5.0	10.0	MS	MS	4.0	8.0	73.50	86.50
Sids-1	Infected	90.0	80.0	S	S	90.0	80.0	1060.0	660.67
	Protected	10.0	10.0	MS	S	8.0	10.0	138.25	150.0
L.S.D. at 5%:									
	Year (Y)	1.117				1.124		24.72	
	Cultivar (C)	1.580				1.589		34.95	
	Treatment (T)	1.117				1.124		24.72	
	Y × C	2.235				2.247		49.43	
	Y × T	1.580				1.589		34.95	
	T × C	2.235				2.247		49.43	
	Y × C × T	3.160				3.178		69.91	

DS = disease severity, IT = infection type, ACI = average coefficient of infection, AUDPC = area under disease progress curve

2. Yield measurements

Data in Table (2) show the effect of leaf rust disease incidence on yield measurements of infected and sprayed adult wheat plants with Sumi-8, during two growing seasons, expressed as weight (g) of thousand grain and yield/plot (kg). Results indicated that protected plants of all tested cultivars recorded the highest weight of 1000 grains and yield/plot during the two growing seasons compared to the infected plants. Protected plants of Sids-12 recorded the highest weight of 1000 grains during the two growing seasons being 45.82 and 45.39 g, respectively, along with the highest values of yield/plot (3.134 and 3.066 kg, respectively). In contrast, the 2012/13 growing season gave the lowest weight values of 1000 grains and yield/plot by infected plants of Gemmiza-7 (41.23 g) and Sids-1 (1.784 kg), respectively. During this growing season, the loss percentage in the weight of 10^3 kernel (g) was ranged from 0.70% (Misr-1 cv.) to 3.97 % (Sids-1 cv.). While during 2013/14 the loss in the weight of 10^3 kernel (g) was ranged from 0.74 % (Misr-1 cv.) to 4.63 % (Gemmiza-7 cv.). The loss percentage of yield/plot (kg) during 2012/13 growing season was ranged from 0.32% (Sids-12 cv.) to 23.73% (Sids-1 cv.). While the loss of yield/plot (kg) during 2013/14 growing season was ranged from 1.70% (Sids-12 cv.) to 19.90% (Gemmiza-7 cv.). The trend of yield loss was in harmony with losses reported by Nazim *et al*. (1983) who reported that, in Egypt the losses in wheat grain yield due to leaf rust may reach 23%. Ochoa and Parlevliet (2007) reported that yield loss was correlated strongly with area under disease progress curve, which means that high levels of resistance are needed to prevent significant yield loss.

Statistical analysis revealed that growing seasons had non-significant differences on kernel weight and yield/plot. Meanwhile, cultivars and treatments (infected and protected) had significant differences on yield measurements. Also, yield measurements were significantly affected by interaction between years, cultivars and treatments.

Table (2): Weight of thousand grains (g) and yield/plot (kg) of four adult wheat cultivars either infected by *Puccinia triticina* or protected by Sumi-8, during 2012/13 and 2013/14 growing seasons.

Cultivar	Treatment	Weight of 10^3 grain (g)			Yield/plot (kg)		
		Infected	Protected	Loss %	Infected	Protected	Loss %
Misr-1	2012/13	45.33	45.65	0.70	2.599	2.648	1.85
	2013/14	44.56	44.89	0.73	2.600	2.712	4.13
Sids-12	2012/13	44.79	45.82	2.25	3.124	3.134	0.32
	2013/14	44.26	45.39	2.49	3.014	3.066	1.70
Gemmiza-7	2012/13	41.23	42.71	3.46	2.082	2.650	21.44
	2013/14	42.22	44.27	4.63	2.427	3.030	19.90
Sids-1	2012/13	42.37	44.12	3.97	1.784	2.339	23.73
	2013/14	43.46	45.14	3.72	1.905	2.309	17.50
L.S.D. at 5% for							
	Year (Y)	n.s.			n.s.		
	Cultivar (C)	0.5566			146.5		
	Treatment (T)	0.3936			103.6		
	Y × C	0.7872			207.0		
	Y × T	0.5566			146.5		
	T × C	0.7872			207.2		
	Y × C × T	1.113			293.1		

III. Physiological analysis

1. Total, free and conjugated phenols content as well as peroxidase activity

Results in Table (3) show that the total and conjugated phenols as well as peroxidase activity is higher in resistant wheat cultivars (Misr-1 and Sids-12) than in susceptible ones (Gemmiza-7 and Sids-1). This observation was reversed concerning the free phenol content where the susceptible wheat cultivars were showed higher concentrations of free phenols than resistant ones. Furthermore, resistant protected treatments recorded lower values of total and conjugated phenols than resistant infected cultivars, and vice versa. Moerschbacher *et al*. (1989) reported that there is a correlation between the phenol status of wheat with host resistance to a variety of diseases, including rusts. Results tabulated in Table (3) indicated that the infected resistant Misr-1 cv. was characterized by the highest levels of total phenolics and peroxidase activity (2.97 mg/g and 4.00 µmol H_2O_2/mg, respectively). While the infected and protected susceptible Sids-1 cv. recorded the lowest levels of both total phenolics and peroxidase activity (1.87 and 0.67, respectively). Such results might be explained according to the following explanations. According to Matern and Kneusal (1988), the first step of the defense mechanism in plant involves a rapid accumulation of phenols at the infection site, which restricts or slows down the growth of the pathogen. This reinforces our results, wherefore phenolic contents were higher in resistant cultivars (Misr-1 and Sids-12). Peroxidase is one of the major enzymes contributing role in disease resistance especially in host–pathogen interactions (Thirupathiraja *et al*., 2004). Also, Invasion of the pathogen leads to development of oxidative stress due to an imbalance between the formations of reactive oxygen species and their detoxification (Smirnoff, 1998; Mika and Luthje 2003). The excessive accumulation of reactive oxygen species is potentially damaging to plant cells unless effectively detoxified by an antioxidative system (Suzuki and Mittler, 2006; Stoilova *et al*., 2009). Peroxidase participates in a variety of plant defense mechanisms in which H_2O_2 is often supplied by an oxidative burst, a common event in defense responses (Montalbini *et al*., 1995). Furthermore, peroxidase, a versatile and ubiquitous group of enzymes that can oxidize phenols to form more toxic quinones, was also reported to play an integral part in disease resistance in wheat against leaf rust (Johnson and Lee, 1978).

2. Chlorophyll content

Chlorophyll contents in the examined four wheat cultivars were given in Table (3). The highest values of SPAD 502 chlorophyll units were recorded by the resistant Sids-12 cv., either in infected or protected status (39.30 and 43.40, respectively). While the lowest values were recorded by the susceptible Sids-1 cv., either in infected or protected status (24.00 and 27.10, respectively). In addition, protected plants of all tested cultivars recorded the highest values of chlorophyll content compared to the infected ones. Decrease in these photosynthetic pigments by increasing the susceptibility behavior of the examined wheat cultivars was explained by McGrath and Pennypacker (1990). They stated that the rust severity was associated with the decrease in photosynthetic rates. Such decrease in values of SPAD 502 chlorophyll units in our results might be due to increase chlorosis around the pustules and a general yellowing of the leaf blades of infected treatments. However, Keutgen *et al*. (1995) and Keutgen & Roeb (1996) found that, declined photosynthesis might be due to the formation of appressoria and the growth of infection tubes into the stomata, leading to a decrease in gas exchange.

IV. Histological structures

Constitutive, pre-formed defenses are thought to comprise a major part and first step in resistance against pathogens. They include the stomata, epidermal reinforcements such as the epidermal leaf hairs (trichomes), as well as sclerenchyma tissue and leaf bundles (Lipka *et al*., 2005). These pre-formed barriers are believed to delay the initial colonization by microbes, allowing the plant sufficient time to mount effective inducible defense responses.

Table (3): Phenolic contents, peroxidase activity and SPAD 502 chlorophyll units of four adult wheat cultivars either infected by *Puccinia triticina* or protected by Sumi-8, during 2013/14 growing season.

Cultivar	Treatment	**Total phenols** (mg/g fresh weight)	**Free phenols** (mg/g fresh weight)	**Conjugated phenols** (mg/g fresh weight)	**Peroxidase** (µmol H_2O_2/mg fresh weight/min)	**SPAD 502 Chlorophyll units**
Misr-1	**Infected**	2.97	0.93	2.04	4.00	33.13
	Protected	2.92	0.98	1.94	3.86	34.93
Sids-12	**Infected**	2.60	0.89	1.71	3.19	39.30
	Protected	2.48	0.92	1.56	2.93	43.40
Gemmiza-7	**Infected**	2.13	1.37	0.76	1.59	32.70
	Protected	2.18	1.32	0.86	1.40	37.66
Sids-1	**Infected**	1.87	1.17	0.70	0.82	24.00
	Protected	1.93	1.08	0.85	0.67	27.10
L.S.D. at 5% 0.05		**0.15**	**0.08**	**0.19**	**0.22**	**3.81**

Resistance to wheat leaf rust was determined through the analysis of many histological structures in the flag leaf of the adult tested wheat cvs. transections were made in flag leaf blade at booting stage through the second growing season (2013/2014).

1. Stomata

Stomata on the leaf surfaces serve as entrance pathways for the majority of the urediospores of rust fungi. The emerging germ tubes should adhere first to the leaf surface, and next they will grow in a direction where they may encounter a stoma. This is called directional growth (Niks and Rubiales, 2002). Therefore, in the present study it is important to examine the stomata of wheat flag leaf in terms of average number/mm^2 and distance between stomatal rows. Figure (1) shows the stomatal features on the upper surface of flag leaf of adult plants of Misr-1, Sids-12, Gemmiza-7 and Sids-1 wheat cultivars, during 2013/14 growing season. According to the observed features in Figure (1) and data in Table (4), the studied wheat cultivars can be divided into two groups. The first group (Fig. 1A&B) included the resistant cvs. (Misr-1 and Sids-12) which characterized by the lowest average numbers of stomata (33.33 and 36.32/mm^2, respectively) with the highest distances between stamatal rows (193.78 and 173.23 µm, respectively), compared to the second group (Fig. 1C&D) included the susceptible cvs. (Sids-1 and Gemmiza-7) which characterized by the highest average numbers of stomata (51.52 and 42.62/mm^2, respectively) with the lowest distances between stamatal rows (107.11 and 148.37 µm, respectively).

2. Leaf hairs

Hairs are surface structures originating from the epidermal cells which usually protrude from the leaf and/or the stem. They act as physical barriers providing protection against various abiotic and biotic factors of the external environment, including pathogen attack. In recent years, there has been growing interest in the outermost layers of the plant (Lazniewska *et al*., 2012).

The epidermal hairs of the tested wheat cultivars were examined and illustrated in Fig. (2) and Table (4). Transections shown in Fig. (2a) and data in Table (4) indicated that Misr-1 wheat cultivar was characterized by the highest number of epidermal hairs. This result coincides with those obtained in the aforementioned disease assessment section where Misr-1 was the highly resistant cultivar among the studied ones.

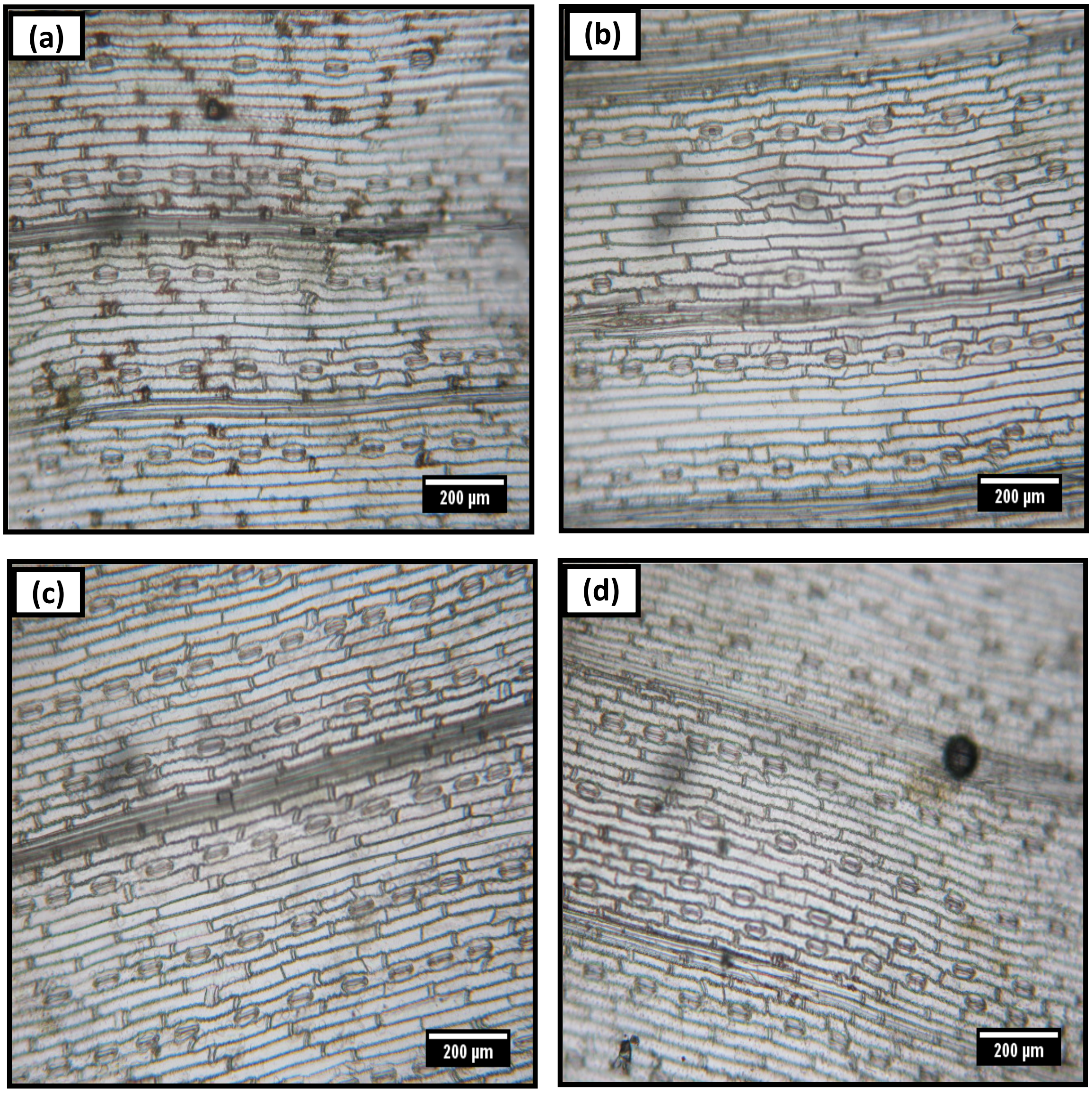

Fig. (1): Stomata distribution on the upper surface of flag leaf of adult plants of a) Misr-1, b) Sids-12, c) Gemmiza-7 and d) Sids-1 wheat cultivars, at the age of 70 days during 2013/14 growing season.

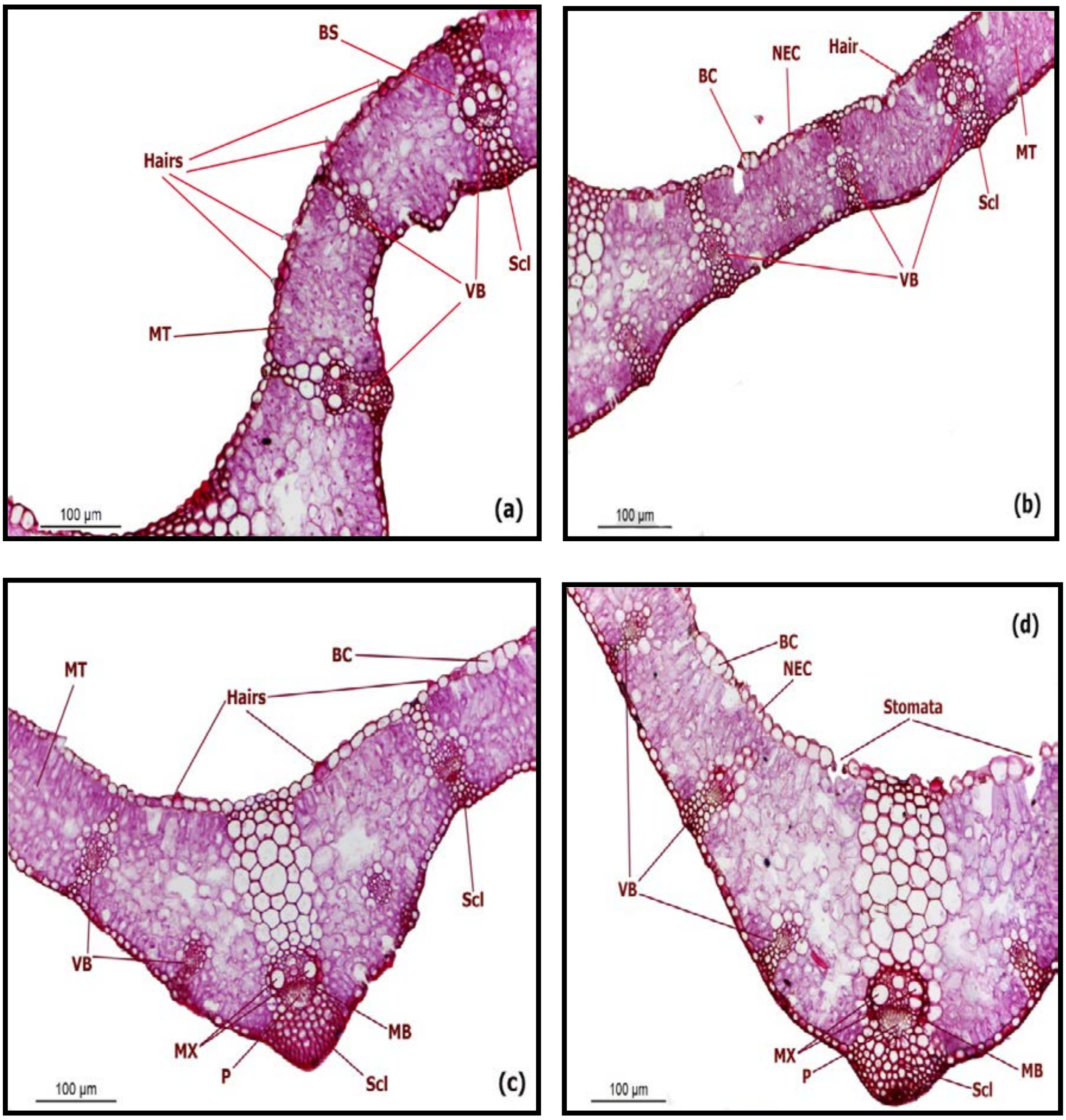

Fig. (2): Epidermal structural features and internal tissue patterns of flag leaf blade of wheat cultivars of a) Misr-1, b) Sids-12, c) Gemmiza-7 and d) Sids-1, at the age of 70 days during 2013/14 growing season.
(BC = Bulliform cell, BS = Bundle sheath, MB = Midvein bundle, MT = Mesophyll tissue, MX = Meta xylum, NEC = Normal epidermal cell, P = Phloum, Scl = Sclerenchyma tissue and VB = Veinlet (laminar or lateral) bundles.

Table (4): Flag leaf blade structural features (Stomata, hairs, Sclerenchyma tissue, Laminar bundles and bulliform cells) of four adult wheat cultivars, at the age of days during 2013/14 growing season.

Cultivar	Stomata		Hairs	Scl.	Laminar bundles	
	Average No./mm^2	Distance between stomata rows (µm)	Average No./ microscopic field		Average thickness (µm)	Average width (µm)
Mis-1	33.33	193.78	9	38.11	46.04	76.53
Sids-12	36.32	173.23	1	26.66	41.83	57.64
Gemmiza-7	42.62	148.37	3	13.96	40.05	50.47
Sids-1	51.52	107.11	1	9.22	36.94	47.71

Scl. = Sclerenchyma tissue thickness under upper epidermis and up lower epidermis (µm)

3. Sclerenchyma tissue and laminar bundles

Sclerenchyma cells are characterized by having uniformly thick lignified secondary walls. Loss of their protoplasm at maturity gave mechanical supporting to plants making them resistant to different pathogenic attacks. In susceptible examined wheat cultivars (Gemmiza-7 and Sids-1), the sclerenchyma tissue did not extended up the laminar (lateral or veinlets) vascular bundles (Fig.2c&d). Moreover, data in Table (4) indicated that leaves of Gemmiza-7 and Sids-1 wheat cvs. recorded the lowest values of sclerenchyma tissues thickness (13.96 and 9.22 µm, respectively), the lowest average values of laminar vascular bundles thickness (40.05 and 36.94 µm, respectively) and the lowest average values of laminar vascular bundles width (50.47 and 47.71 µm, respectively). Meanwhile leaves of resistant wheat cvs. (Misr-1 and Sids-12), recorded the highest values of sclerenchyma tissues thickness (38.11 and 26.66 µm, respectively), the highest values of average thickness of laminar vascular bundles (46.04 and 41.83 µm, respectively) and the highest average values of laminar vascular bundles width (76.53 and 57.64 µm, respectively). The microscopic examinations were supported by the hypothesis of Gelalcha and Hanchinal (2003) reported that, structural characteristics of wheat leaves in relation to resistance against leaf rust pathogen, showed increase number of sclerenchyma layers which significantly higher in resistant genotypes.

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تقدير مرض صدأ الأوراق وعلاقته بالخصائص الفسيولوجية والتشريحية لأربعة أصناف من أقماح الخبز المصرية

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الملخص العربي

يعتبر مرض صدأ الأوراق في القمح والذي يسببه الفطر بكسينيا ترتيسينا من أهم الأمراض التي تصيب نبات القمح مسببة له خسائر عالية في المحصول. تم في هذا البحث تقدير الصفات المرضية وتأثيرها على مكونات المحصول وعلاقتها بالخصائص الفسيولوجية والتشريحية لأربعة أصناف من أقماح الخبز المصرية وهي مصر–1 و سدس–12 و جميزة–7 و سدس–1. تم إجراء هذا العمل خلال موسمي النمو 13/2012 و 14/2013 تحت ظروف الحقل بمحافظة الشرقية بمحطة البحوث الزراعية بكفر الحمام التابعة لمركز البحوث الزراعية – مصر . أوضحت نتائج الأستجابة للعدوى مدى مقاومة الصنفين مصر–1 وسدس –12 لمرض صدأ الأوراق ، بينما تميز الصنفين جميزة –7 وسدس–1 بقابليتهما العالية للإصابة بهذا المرض . تم دراسة بعض الصفات الفسيولوجية والتشريحية وعلاقتها بمكونات المرض حيث أثبتت النتائج وجود علاقات وثيقة بينهم ، وفي هذا السياق سجلت الأصناف المقاومة أقل قيم لشدة الإصابة بالمرض ومتوسط معامل الإصابة والمساحة تحت المنحنى المرضي . ارتبطت هذه النتائج طرديا مع محتوى الفينولات الحرة وكذلك عدد الثغور/مم^2 ، وعلى العكس من ذلك زيادة محتوى الفينولات الكلية والمرتبطة ونشاط انزيم البيروكسيديز ومحتوى الكلوروفيل والمسافة بين صفوف الثغور وسمك النسيج الاسكلرنشيمي بالإضافة إلى صفات الحزم الوعائية. لوحظ وجود علاقة بين محتوى الكلوروفيل وانتاجية الحوض حيث تميز الصنف سدس –12 بأعلى قيم لمحتوى الكلوروفيل وكذلك أعلى قيم لإنتاجية الحوض . تم حساب النسبة المئوية للفاقد في المحصول وسجل الصنف سدس –12 أقل نسبة مئوية للفاقد في المحصول (0.32 %) خلال موسم 2013/2012 ، بينما سجل الصنف سدس –1 والذي تميز بقابليته للاصابة بالمرض أعلى نسبة مئوية للفاقد في المحصول (23.73 %) خلال نفس الموسم . يمثل العمل الحالي أساساً عملياً للدراسات المستقبلية لمرض صدأ الأوراق في بعض أصناف قمح الخبز المصرية .