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## IMPACT OF CLIMATE CHANGE ON THE APPEARANCE OF FIRE BLIGHT DISEASE IN A NEW AREA IN EGYPT

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**ABSTRACT:** Fire blight disease was first recorded in Egypt in 1962 on pear in an area between Alexandria and Dammanhour. The disease spread throughout the governorates of the Nile Delta until it reached the pear orchards in the surrounding governorates. Fire blight was never observed in the extended desert of the Faiyum governorate in northern Upper Egypt. Symptoms of fire blight were observed on pear orchards in March 2019 at Sawl Village, Atfih center, in the south of the Giza governorate. Blossom blight, leaf and shoot blight, branch cankers symptoms were observed on 15-20-year-old pear trees. The Global Positioning System (GPS) coordinates of the diseased pear orchards are 29°22'42.7"N 31°14'19.1"E. Selected five isolates produced on MS medium from infected pear samples were pathogenic on immature pear fruitlets. Morphological, physiological, and biochemical characteristics of the obtained isolates were conforming to the characteristics of *Erwinia amylovora*. The primers AMSJ14258 and AMSK14892c generated an amplicon of the expected size (600 bp) for all five isolates. The percentage of visibly diseased trees in the affected orchard was 100%. While the percentage of visibly diseased blossom clusters of the trees of the affected orchard was varied, in a range between 42.5 to 78.1%. As far as we know, this is the first recorded outbreak of fire blight disease in this area, south of 30 ° N latitude and near latitude 29 ° N in Egypt. Climatic data were studied during the winter and spring in the season of 2018/2019 and those of the previous three seasons, especially during blooming and early fruiting (March, and April), to build a relationship between the climatic change and fire blight disease outbreaks in pear orchards at this area. The results showed that despite the uncommon events in climate conditions in 2019, compared to the previous three years, the unusual increase in the intensity of rains in March 2019 with the availability of the other climatic conditions could be the most important reason of fire blight disease outbreaks in this region of Egypt recently.

**Key words:** *Erwinia amylovora*, south of the Giza governorate, maximum temperature, minimum temperature, rainfall, relative humidity (RH).

### INTRODUCTION

Pear (*Pyrus communis* L.) is one of the most important pome fruit crops in the world. The cultivation of pear spread in Egypt after the Ministry of Agriculture imported some varieties and cultivated them, nearly about the beginning of the second half of the twentieth century. According to the statistics of the Ministry of Agriculture and Land Reclamation (Egypt), the area cultivated with pear in Egypt in 1960 was 2490 feddans, which is estimated about 40% of the area of deciduous Fruit. This area reached 3571 feddans in 1970 and then 10436 feddans in 1980, which is estimated at 36.1% of the area of deciduous fruit. This area reached 18286 feddans in 1988. Then it started gradually decreasing until it

reached 15400 feddans in 1994, which is estimated at 8.4% of the area of deciduous fruit and then in 2000 to 8500 feddans, which is estimated at 5% of the area of deciduous fruit (**El-Shall et al., 2001**). Since 2000, cultivated areas have not significantly increased. The decline in the area cultivated with pear is due to the expansion of the problem of fire blight since the severe outbreak of the disease in 1982.

Fire blight caused by *E. amylovora* poses a major threat to apple and pear producers in many countries worldwide. Fire blight disease was first recorded on pear in Egypt in 1962 in an area between Alexandria and Dammanhour called Mamal El-Kezaz (**El-Helaly et al., 1964**). The removal of diseased pear orchards helped in the absence of symptoms of the fire blight

disease in Egypt until 1981. During 1982 and 1983 seasons, a severe outbreak of fire blight was observed on pears at Alexandria and Beheira Governorates in Egypt (Abo El-Dahab *et al.*, 1983). Since then, the disease spread rapidly through most governorates of northern Egypt. The disease spread throughout the governorates of the Nile Delta until it reached the pear orchards in the surrounding governorates of Nile Delta (Mtruh, Ismailia and Qalyubia), Resulting in a decrease in pear cultivation areas and serious economic losses to pear growers (van der Zwet and Mikhail, 1984; van der Zwet, 1986; El-Zayat *et al.*, 1986; Tawfik *et al.*, 2006; Ashmawy *et al.*, 2015 and Shoeib *et al.*, 2016). By 1988, eighty percent of all pear cultivation area was affected and fifty percent of all trees had been eradicated. The disease was never observed in the extended desert of the Faiyum governorate in northern Upper Egypt (Bonn and van der Zwet, 2000).

Some studies raise questions and hypotheses about the epidemiology of the occurrence of fire blight disease in Egypt, based on that the disease outbreaks require specific conditions of rainfall and temperature (Balabel *et al.*, 2018). Several studies have confirmed that temperature, rain rates, and relative humidity are favorable to the outbreak of fire blight in Egypt during March and April, especially in the northern governorates, although the disease severity is highly variable from one year to another mainly depending on climatic factors (Abol Maatey *et al.*, 2002). Fire blight is more common and severe in regions with warm, humid climates. The northern half of Europe faces a much lower problem with the disease than the southern half. Countries with fairly warm winter temperatures, such as Egypt, experience a higher primary inoculum level in the spring (van der Zwet, 1996). *E. amylovora* is a psychrotrophic bacterial species that can grow in temperatures from 4°C to 37°C, with an optimum temperature of 28°C. Temperatures above 18°C are required for blossom blight outbreaks under field conditions. Blossom blight of fire blight was unlikely to occur when the temperature during bloom was quite cool. *E. amylovora* has a particular potential of adaptations that enable it to maintain its pathogenicity even at low growth temperatures. The pathogenic potential and survival in moderate and low weather temperatures of *E. amylovora* have probably contributed to its successful spread to countries with different climates (Santander and Biosca, 2017). Climate change and future climate conditions can play a role in changing the pathological map of many plant diseases in Egypt (Hesham *et al.*, 2016).

This study aimed to isolate and identify *E. amylovora*, the causal organism of fire blight disease,

from a new area south of 30 ° N latitude and near latitude 29 ° N in Egypt after the first outbreak of the disease in this area. Study the change in some climatic conditions in this region and its effect on the occurrence of the disease for the first time in this region.

## MATERIALS AND METHODS

### Sampling

Diseased pear flowers, leaves, fruitlets, and branches with fire blight symptoms were collected from Sawl Village, Atfih center, in the south of the Giza governorate on the eastern bank of the Nile, in March 2019. Collected samples were kept in plastic bags in iceboxes and transferred to the laboratory of Bacterial Diseases Research Department, Plant Pathology Research Institute, Agricultural Research Center, Giza, Egypt.

### Isolation

Diseased samples were washed in sterile distilled water. Pieces of diseased tissues were macerated in sterile distilled water in Petri-dish and left for 15-30 min. A loopful of the resulting suspension was streaked on the surface of the MS medium (Miller and Schroth, 1972). The inoculated Petri-dishes were incubated at 28°C for 3 days. Single colonies of growing bacteria were transferred to slants of King's medium B (King *et al.*, 1954) to use in subsequent tests.

### Pathogenicity test

Bacterial cultures recovered from diseased samples were tested for the ability to induce disease symptoms on green immature pear fruitlets (Sobiczewski and Millikan, 1985 and Jones and Geider, 2001). The fruitlets were disinfected by dipping in 70% ethanol for 2 min. and rinsed three times with sterile distilled water. These fruitlets were distributed in sterilized Petri plates and inoculated by stabbing with a needle laden with the culture (24 h. old) of the bacteria (one loopful for each fruitlets). Each plate contained four inoculated fruitlets in which high humidity was maintained with wet cotton pieces and incubated at 28°C for 5 days. Control fruitlets were inoculated with sterile distilled water.

### Identification of the pathogen by morphological, physiological and biochemical characteristics

Pathogenic isolates were identified using morphological, physiological, and biochemical characteristics as suggested by Schaad and Hildebrand (1980) and Jones and Geider (2001).

### Verification of identity by the polymerase chain reaction (PCR)

Two primers AMSJ14258: 5' TTACTGCAGACGTGCTC 3' and AMSK14892c: 5'ATCTTCTCCGCCGGACA 3' (Bio Basic Inc., Markham, Canada) designed from the chromosomal *ams* region were used (Mohammadi *et al.*, 2009). The suspension ( $10^9$  CFU/mL) of bacteria were diluted 1:100 in 0.1% Tween 20 and lysed for 15 min at 65°C. The lysate was centrifuged for 1 min at 14,000xg and 3 µl of the aqueous phase was used as a template for each 25 µl of PCR reaction mixture. The following amounts of reagents were used: 12.5 µL of MyTaq RedMix, 2x (Bioline Reagents Ltd, UK) + 7.5 µL molecular grade water + 2 µL primers (10 µM each) + 3 µL DNA template. Amplification was performed in a thermal cycler (Applied Biosystems, 2720, Life Technologies Holdings Pte Ltd, Singapore). The PCR conditions were as follows: initial denaturation at 94°C for 5 min.; followed by 35 cycles at 94°C for denaturation (30 s), 52°C for annealing (30 s) and primer extension at 72°C for 30 s; followed by final extension at 72°C for 10 min. and 4°C (hold temperature). PCR products were separated on agarose gel electrophoresis using 1.5% agarose in 1x TBE buffer, stained with nucleic acid staining solution (RedSafe™, 20000x, iNtRON Biotechnology, Inc.) and visualized on a UV-trans-illuminator.

### Disease readings

Disease incidence refers to the number of plant units (whole plant, limbs, shoots, blossom clusters) that are visibly diseased relative to the total number (Momol *et al.*, 1996). Three orchards were randomly selected to estimate the percentage of fire blight in trees and blossom clusters. For the percentage of fire blight in trees, five rows were randomly chosen in each orchard, the number of infected and healthy trees in each row was calculated. The percentage of visibly diseased trees was calculated as follows:

Visibly diseased trees % = (number of infected trees in the row / the total number of trees in the row) x 100.

For the percentage of fire blight in blossom clusters, five trees were randomly chosen in each orchard, the number of infected and healthy blossom clusters were calculated in each tree. This is done by calculating the number of infected and healthy blossom clusters in three branches, and then

calculating the average of infected and healthy blossom clusters in one branch and subsequently in the whole tree. The percentage of visibly diseased blossom clusters was calculated as follows:

Visibly diseased blossom clusters % = (number of infected blossom clusters in the tree / the total number of blossom clusters in the tree) x 100.

### Climate data

Meteorological data for Sawl Village, Atfih center, in the south of the Giza governorate during the winter and spring months (from December to June) in seasons 2015/2016 to 2018/2019 obtained from the Central Laboratory for Agricultural Climate (CLAC). The obtained data consist of maximum temperatures, minimum temperatures, relative humidity, and rainfall.

## RESULTS

### Location and symptomatology

Symptoms of fire blight were observed on pear orchards (Fig., 1a) during March 2019 at Sawl Village, Atfih center, in the south of the Giza governorate, on the eastern bank of the Nile (Fig., 1b). The flowering of pear trees in this region begins with the early days of March. Blossom blight, leaf shoot blight, branch canker symptoms were observed on 15-20-year-old plants. A single flower or entire cluster wilted and turned brownish to black (blossom blight) and the whole diseased leaves dried out and their edges curled to the inside and turned to black (Fig., 1c). Cankers were also observed on the infected branches (Fig., 1d). The latitude of the diseased pear orchards is 29.3785383, and the longitude is 31.238625 with the Global Positioning System (GPS) coordinates of 29°22'42.7"N 31°14'19.1"E. This is the first recorded outbreak of fire blight disease in the south of latitude 30° N and near latitude 29 ° N in Egypt.

### Isolation and pathogenicity

Five reddish-orange colonies with deep orange centers produced on MS medium from infected pear samples collected from Sawl Village, Atfih center, south of the Giza governorate were selected. These five isolates were pathogenic on immature pear fruitlets and showed blackened affected areas with drops of bacterial ooze (Fig., 2). There were no notable differences in pathological ability between isolates.



Fig. 1. Symptoms of pear fire blight observed on pear orchards (A) in March 2019 at Sawl Village, Atfih center, in the south of the Giza governorate (B). A single flower or entire cluster wilted and turned brownish to black and whole diseased leaves dried out and their edges curled to inside (C), cankers were observed on branches (D).

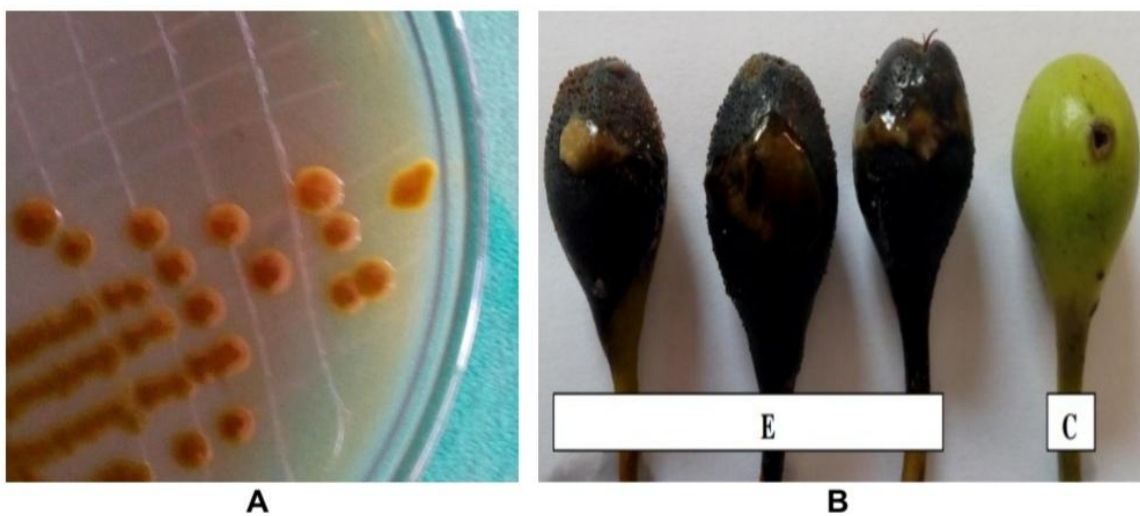


Fig. 2. Isolates with the typical shape of *Erwinia amylovora* produced on MS medium (A) were pathogenic on immature pear fruitlets and showed blackened affected areas with drops of bacterial ooze (BE) and no symptoms appear on immature pear fruitlets inoculated by sterile distilled water (BC).

**Morphological, physiological and biochemical characteristics**

Results in Table (1) show the characteristics of pathogenic isolates isolated from infected pear tissues collected from Atfih center, south of Giza the governorate. All isolates were gram-negative, rods, positive for KOH 3%, and gave a positive reaction with catalase. These isolates gave a negative reaction

with oxidase, urease production, and H<sub>2</sub>S production from cysteine. Also, these isolates grow on MS medium, don't grow at 36°C, don't produce fluorescent pigment on KB agar medium, and don't give pink pigment on the YDC medium. Whereas, these isolates can't produce acid from salicin and α-methyl glucoside. These isolates can produce acid from L (+) arabinose.

**Table 1. Characterization of five pathogenic bacterial isolates obtained from infected pear samples collected from Sawl Village, Atfih center, south of the Giza governorate**

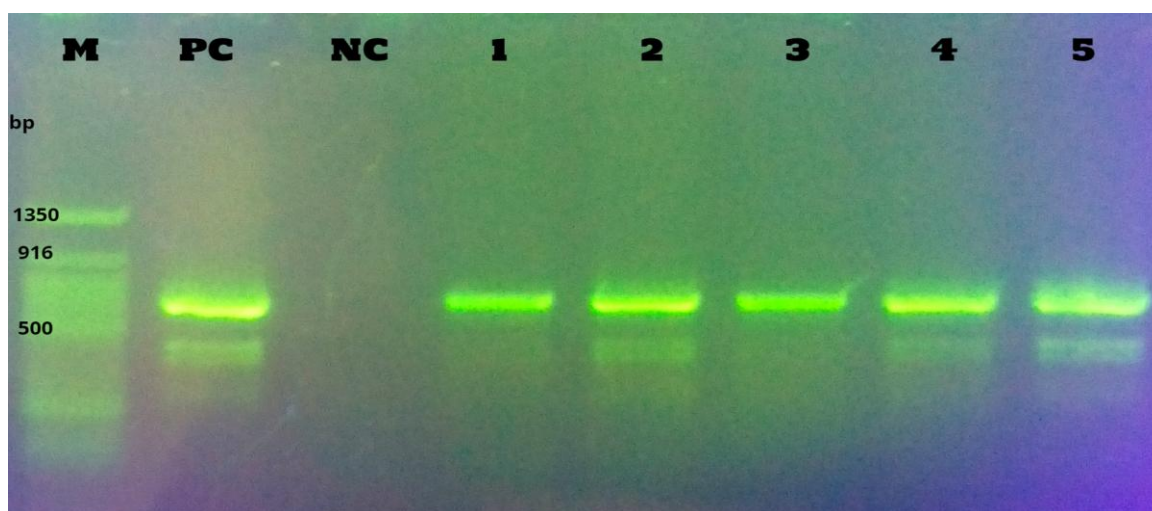
Character	Isolate					
	SEa1	SEa2	SEa4	SEa7	SEa8	Reference isolate
Gram stain	-	-	-	-	-	-
Cell shape	Rod	Rod	Rod	Rod	Rod	Rod
KOH 3%	+	+	+	+	+	+
Catalase	+	+	+	+	+	+
Oxidase	-	-	-	-	-	-
Urease	-	-	-	-	-	-
H <sub>2</sub> S from cysteine	-	-	-	-	-	-
Growth on MS medium	+	+	+	+	+	+
Growth at 36°C	-	-	-	-	-	-
Fluorescent pigment on KB agar	-	-	-	-	-	-
Pink pigment on YDC	-	-	-	-	-	-
Acid production from:						
Salicin	-	-	-	-	-	-
α-methyl glucoside	-	-	-	-	-	-
L (+) arabinose	+	+	+	+	+	+

- = Negative reaction, + = Positive reaction

**Polymerase chain reaction (PCR).**

Polymerase Chain Reaction (PCR) was used for further confirmation of the identity of *E. amylovora* isolates. Primers AMSJ14258 and AMSK14892c

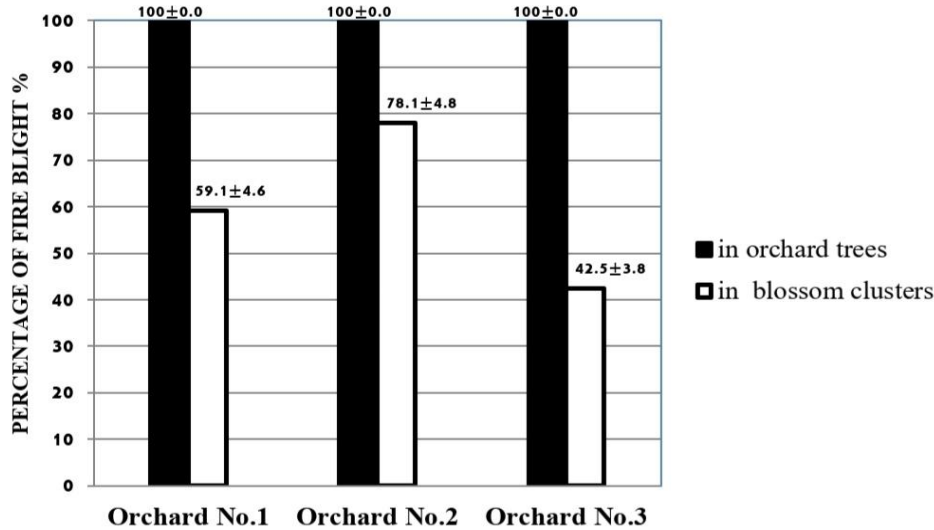
designed from the chromosomal *ams* region and produced a PCR fragment of 0.6 kb, specific for *E. amylovora* were used. All *E. amylovora* Sawl Village isolates showed a band at expected amplicon sizes 600 bp (Fig., 3).



**Fig. 3. PCR products are based on AMSJ14258 and AMSK14892c primers for *Erwinia amylovora* Sawl Village isolates. Lane M: 50bp DNA marker ladder. Lane PC: Reference isolate. Lane NC: Negative (water) control. Lanes 1-5: Sawl Village pear isolates (SEa1, SEa2, SEa4, SEa7, SEa8, respectively). a band at the expected amplicon size was generated (600 bp) for all isolates.**

Three orchards were randomly selected to estimate the percentage of fire blight in trees and blossom clusters. No differences in the percentage of visibly diseased trees in the three orchards. The percentage of visibly diseased trees in the affected

orchard was 100 %. While the percentage of visibly diseased blossom clusters of the trees of these three orchards was varied, in a range between 42.5 to 78.1% (Fig., 4).



**Fig. 4. The percentage of fire blight in trees and blossom clusters on pear orchards in March 2019 at Sawl Village, Atfih center, in the south of the Giza governorate. Means of five replicates; values expressed as mean ± Standard Deviation (SD).**

**Climate conditions**

Climatic conditions were studied during the winter and spring, especially during blooming and early fruiting (March, and April) in the season of 2018/2019 and those of the previous three seasons to extrapolate the possible relationship between the climatic change and fire blight disease outbreaks in pear orchards in March 2019 at Sawl Village, Atfih center, in the south of the Giza governorate.

**Maximum temperature**

A decrease in the average maximum temperatures occurred in March and April 2019 compared to those of the previous three years was recognized. The average maximum temperatures in March and April 2019 were 23.76±3.22 and 28.29±4.11 °C, respectively. Whereas the average maximum temperatures in March ranged between 24.39±2.67 and 28.96±4.19 °C, and in April between 30±3.88 and 33.8±4 °C in 2016, 2017 and 2018 (Fig., 5).

**Minimum temperature**

A decrease in the average minimum temperatures occurred in March and April 2019 compared to those of the previous three years was recognized. The average minimum temperatures in March and April 2019 were 8.88±2.55 and 12.94±3.37 °C, respectively. Whereas the average maximum temperatures in March ranged between 10.08±2.43

and 12.7±2.62 °C, and in April between 13.87±2.68 and 16.14±3.49 °C in 2016, 2017, and 2018, respectively (Fig., 6).

**Average daily temperature**

A decrease in the highest average daily temperature that occurred in April 2019 compared to those of the previous three years was recognized. The highest average daily temperature in April 2019 was 27.3 °C. Whereas the highest average daily temperature in April 2016, 2017, and 2018 was 30.62, 29.08, and 28.73°C, respectively (Fig.,7). There was a decrease in the lowest average daily temperature in March, April, and May 2019 compared to the previous three years. The lowest average daily temperature in March was 15.31, 13.74, 16.43, and 12.24 °C in 2016, 2017, 2018, and 2019, respectively. Whereas the lowest average daily temperature in April was 19.29, 16.87, 17.61, and 14.3 °C in 2016, 2017, 2018, and 2019, respectively. The lowest average daily temperature in May was 21.31, 22.68, 22.77, and 19.44 °C in 2016, 2017, 2018, and 2019, respectively (Fig.,8). The number of days in March with an average daily temperature ≥15.6 °C was 30, 24, 31, and 18 in 2016, 2017, 2018, and 2019, respectively. Also, the number of days in April with an average daily temperature ≥15.6 °C was 30, 30, 30, and 28 in 2016, 2017, 2018, and 2019, respectively (Table, 2).

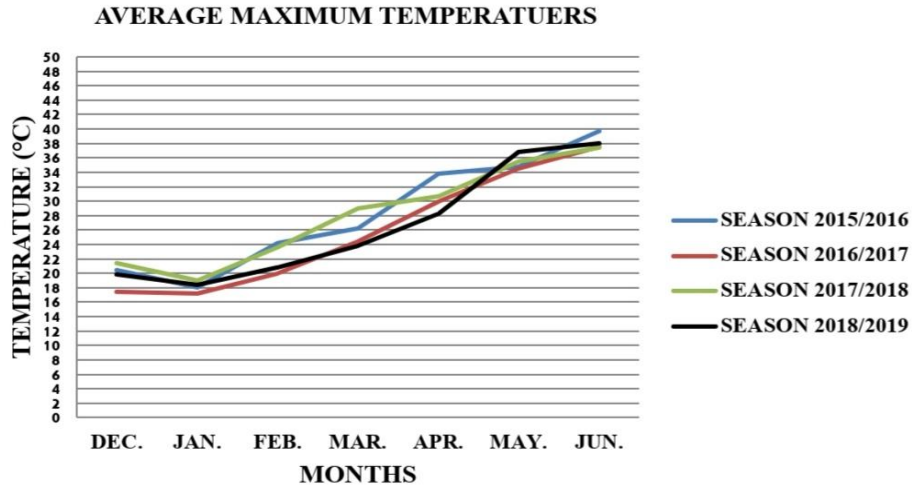


Fig. 5. The average maximum temperatures in the winter and spring months during seasons from 2015/2016 to 2018/2019. The standard deviation of data was calculated.

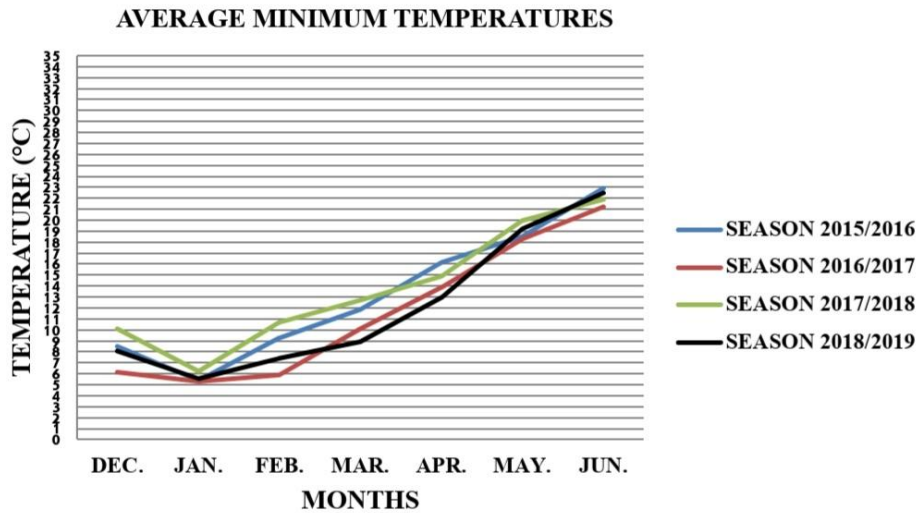


Fig. 6. The average minimum temperatures in the winter and spring months during seasons from 2015/2016 to 2018/2019. The standard deviation of data was calculated.

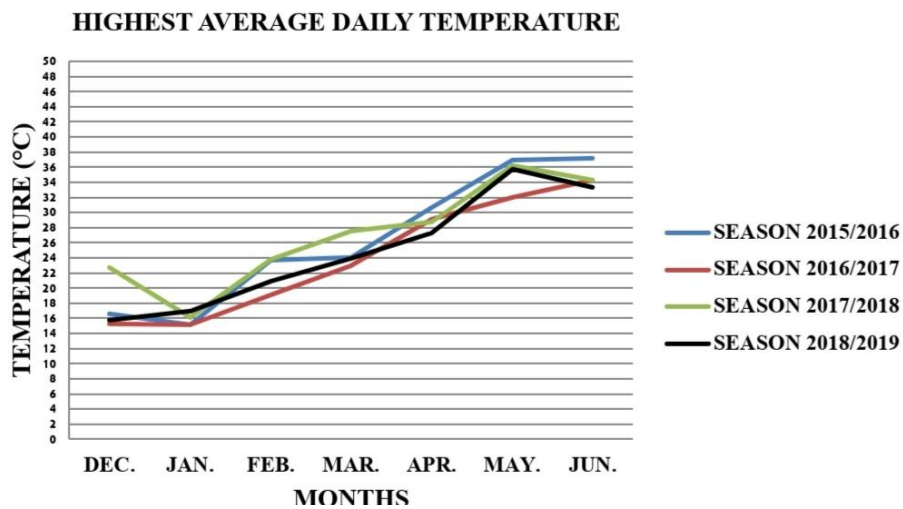


Fig. 7. The highest average daily temperature in the winter and spring months during seasons from 2015/2016 to 2018/2019.

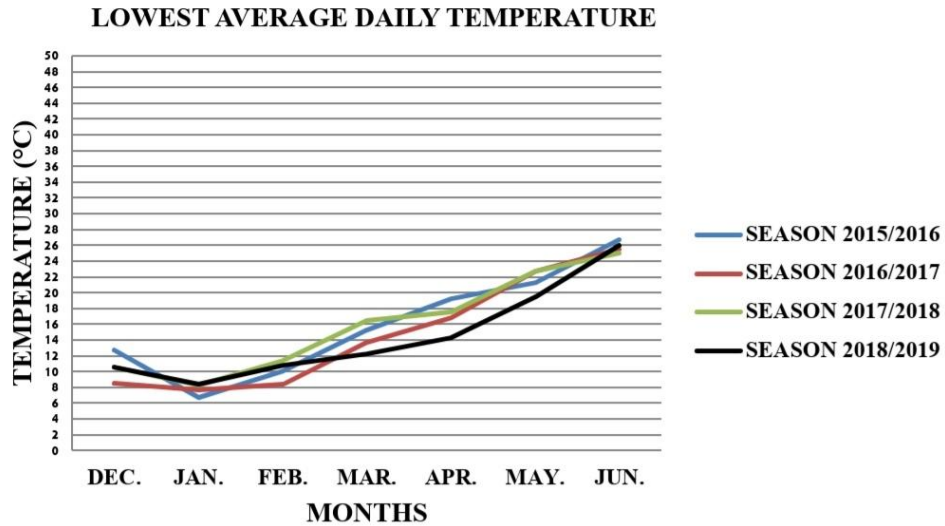


Fig. 8. The lowest average daily temperature in the winter and spring months during seasons from 2015/2016 to 2018/2019.

**Relative humidity (RH).**

There was an increase in the average relative humidity in February, March, and April 2019 compared to those of the previous three years except when comparing February and March 2019 with February and March 2017. The average relative humidity in February was  $45.26 \pm 15.62$ ,  $58.04 \pm 11.05$ ,  $46.52 \pm 14.69$ , and  $47.47 \pm 10.46\%$  in 2016, 2017, 2018, and 2019, respectively. Whereas the average relative humidity in March was  $38.08 \pm 12.60$ ,  $45.64 \pm 10.84$ ,  $32.81 \pm 11.08$ , and  $43.21 \pm 9.91\%$  in

2016, 2017, 2018, and 2019, respectively. The average relative humidity in April was  $26.7 \pm 10.47$ ,  $33.96 \pm 10.08$ ,  $34.34 \pm 10.47$ , and  $35.06 \pm 8.60\%$  in 2016, 2017, 2018, and 2019, respectively (Fig.,9). The number of days in March 2019 in which relative humidity was  $\geq 50\%$  was 12 days, which is a large number of days compared to the previous three years except March 2017. The number of days in March with relative humidity  $\geq 50\%$  was 5, 13, and 1 in 2016, 2017, and 2018, respectively (Table, 2).

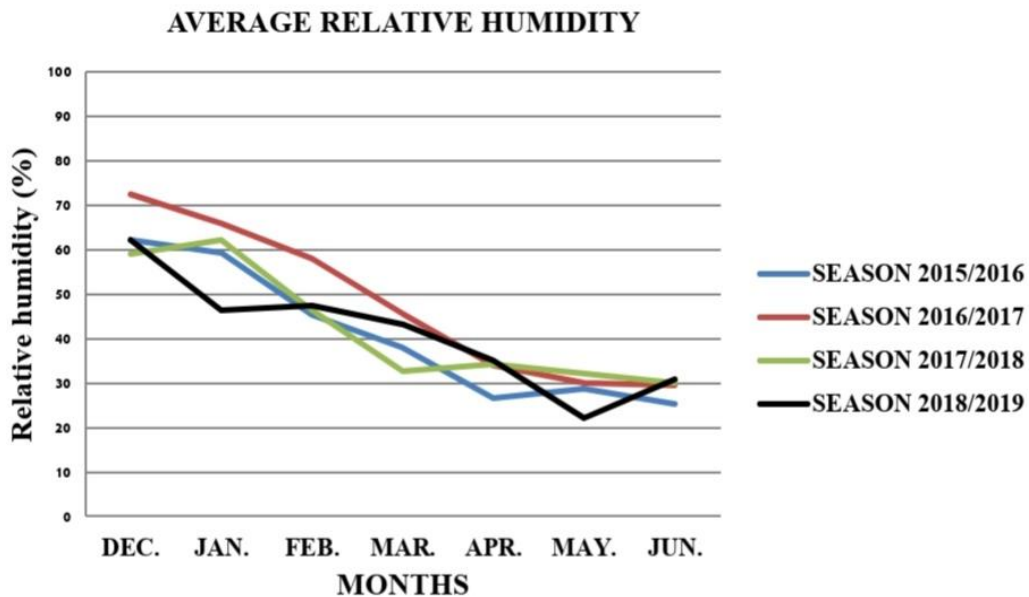


Fig. 9. Average relative humidity in the winter and spring months during seasons from 2015/2016 to 2018/2019. The standard deviation of data was calculated.



**Rainfall and duration**

The rainfall increased in March 2019 compared to those of the previous three years (Fig., 10). The rainfall in March 2019 was about 9.29 mm while it was 3.93, 0, and 0.71 mm in 2016, 2017, and 2018 respectively. Rainfall occurred in December, January, February, and March during the 2018/2019 season and in a range between 1.48 and 9.29 mm. While the rainfall in February 2016 (season 2015/2016) was

relatively little and it was 0.95 mm. Also, during the 2016/2017 season, there was no rain in March 2017 and it was relatively few in January 2017 (0.6 mm). Furthermore, during season 2017/2018 there was little rainfall in March 2018 (0.71 mm). The number of days in March 2019 in which rainfall  $\geq 0.25$  mm was 5 days. while the number of days in March with rainfall of  $\geq 0.25$  mm was 1, 0, and 1 in 2016, 2017, and 2018, respectively (Table, 2).

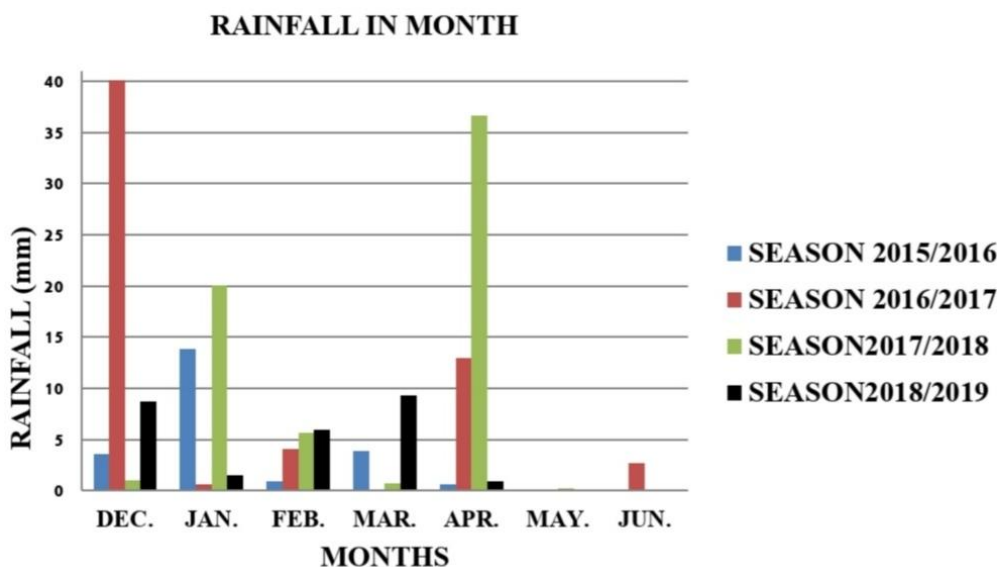


Fig. 10. The intensity of rains in the winter and spring months during seasons from 2015/2016 to 2018/2019.

Table 2. The number of days when climatic conditions were suitable for fire blight infection during blooming and early fruiting (March and April) in 2019 and the previous three years

Year	Month	The number of days		
		average daily temperature $\geq 15.6$ °C	RH $\geq 50$ %	Rainfall $\geq 0.25$ mm
2016	Mar.	30	5	1
	Apr.	30	1	0
2017	Mar.	24	13	0
	Apr.	30	2	4
2018	Mar.	31	1	1
	Apr.	30	1	3
2019	Mar.	18	12	5
	Apr.	28	0	1

**DISCUSSION**

Fire blight bacteria overwinter in cankers or invisible infection on twigs, branches, or trunks of host trees. Nurseries make efforts to only use clean budwood for propagation, but *E. amylovora* may be present in trees that appear to have no apparent fire blight symptoms at the time of collection. The use of infected budwood can be a potential reason for fire

blight transmission over long distances (Tancos *et al.*, 2017). In Cheonan, typical fire blight symptoms (Leaf and shoot blight) were observed on 10-year-old plants of Chinese quince (*Chaenomeles sinensis*) in a commercial nursery (36°54'54.10" N, 127°8'21.84" E) in July 2015, this city is located 12 km from an orchard (36°57'31.15" N, 127°15'55.15" E) where the apple fire blight occurred in Anseong, South Korea (Myung *et al.*, 2016). Most infected pear plants in

Sawl Village, Atfih center are between 15 and 20 years old. Pear seedlings are brought from the commercial nurseries from the Nile Delta governorates, such as Beheira and Monufia, where the disease has previously been reported. The primary inoculum is therefore available but the disease has never been observed in the area before. Symptoms of fire blight (blossom blight, leaf and shoot blight, and branch cankers) were observed on pear orchards in March 2019 at Sawl Village (29°22'42.7"N 31°14'19.1"E), Atfih center, in the south of the Giza governorate. MS medium is a selective medium for *E. amylovora* inhibits the growth of most other microorganisms. The few pseudomonads and other bacteria that grow on the medium are blue or green in contrast to the red-to-orange hues characteristic of *Erwinia* sp. (Miller and Schroth, 1972). Five reddish-orange colonies with deep orange centers produced on MS medium from infected pear samples collected from Sawl Village were pathogenic on immature pear fruitlets and showed blackened affected areas with drops of bacterial ooze. Zhao *et al.* (2005) demonstrated that infection of immature pear fruitlets by *E. amylovora* required major pathogenicity factors. Isolates of *E. amylovora* were gram-negative, didn't produce fluorescent pigment on KB, and didn't grow at 36°C. The isolates gave a negative reaction with oxidase, urease, indole, H<sub>2</sub>S production but they gave a positive reaction with catalase and citrate tests. More than 87% of the isolates induced gelatin liquefaction, didn't reduce nitrate, and produced acid from sorbitol but not from inositol, raffinose, and salicin (Fatmi *et al.*, 2008). All Sawl Village isolates were gram-negative, rods, positive for KOH 3%, and gave a positive reaction with catalase. These isolates gave a negative reaction with oxidase, urease production, and H<sub>2</sub>S production from cysteine. Also, these isolates grow on MS medium, don't grow at 36 ° C, don't produce fluorescent pigment on KB agar medium, and don't give pink pigment on the YDC medium. Whereas, these isolates can't produce acid from salicin and α-methyl glucoside. These isolates can produce acid from L (+) arabinose. All *E. amylovora* Sawl Village isolates showed a band at expected amplicon sizes 600 bp with AMSJ14258 and AMSK14892c Primers. Primers AMSJ14258 and AMSK14892c produced a PCR fragment of 0.6 kb, specific for *E. amylovora* that was not detected in lysates from *Pseudomonas fluorescens*, *Pseudomonas graminis*, *Pantoea agglomerans*, *Pectobacterium atrosepticum*, or *Erwinia tasmaniensis* cells (Mohammadi *et al.*, 2009). The percentage of visibly diseased trees in the infected orchard was 100%. While the percentage of visibly diseased blossom clusters of the trees of affected orchards was varied, in a range between 42.5 to 78.1%. Tree age and succulence of tissues affect the severity of fire blight disease (Koski and Jacobi, 2014). Disease severity is highly variable from one year to another depending mainly on climatic factors

(Abol Maatey *et al.*, 2002). Fire blight was never observed in the large desert of the Faiyum governorate in northern Upper Egypt (Bonn and van der Zwet, 2000). As far as we know, this is the first recorded outbreak of fire blight disease in this area, south of 30 ° N latitude and near latitude 29 ° N in Egypt.

Fire blight disease is most dangerous when spring temperatures are warm during the pre-bloom and bloom stages. Warm rainy springs help the rapid spread of the pathogen, resulting in blossom blight (Koski and Jacobi, 2014). *E. amylovora* grow at temperatures ranging from 4 °C to 37 °C, with an optimum of 28 °C. Temperatures above 18 °C are required for blossom blight epidemics under field conditions, although *E. amylovora* was pathogenic at 4 °C, 14 °C, and 28 °C temperatures, with a slow-down of symptom development correlating with colder temperatures (Santander and Biosca, 2017). In the location of our study, the average maximum temperatures in March (month of flowering) and April 2019 (the year of the outbreak) were 23.76±3.22 and 28.29±4.11 °C, respectively. Whereas the average maximum temperatures in March ranged between 24.39±2.67 and 28.96±4.19 °C, and in April between 30±3.88 and 33.8±4 °C in 2016, 2017 and 2018. The average minimum temperatures in March and April 2019 were 8.88±2.55 and 12.94±3.37 °C, respectively. Whereas the average maximum temperatures in March ranged between 10.08±2.43 and 12.7±2.62 °C, and in April between 13.87±2.68 and 16.14±3.49 °C in 2016, 2017, and 2018, respectively. The lowest average daily temperature in March was 15.31, 13.74, 16.43, and 12.24 °C in 2016, 2017, 2018, and 2019, respectively. The number of days in March with average daily temperature ≥15.6 °C was 30, 24, 31, and 18 in 2016, 2017, 2018, and 2019, respectively. The occurrence of a blossom infection requires several factors, including precipitation event of either dew, ≥0.25 mm on the current day, and average daily temperature of ≥15.6 °C (Dewdney *et al.*, 2007). In detached crab apple flowers, *E. amylovora* multiplied on inoculated flower stigmas at between approximately 55 and 100% RH but not in hypanthium until the RH was higher than 80%. Flowers became diseased only with wetting, and incidence was high (77%) even when water application was immediately followed by a 52-min drying period (Pusey, 2000). Movement of bacteria in apples and pears from colonized stigmas to the nectarthodes occurs when 0.25 mm of rain (Turechek and Biggs, 2015). In the location of our study, there was an increase in the average relative humidity in February, March, and April 2019 compared to the previous three years except when comparing February and March 2019 with February and March 2017. The number of days in March 2019 in which relative humidity was ≥50% was 12 days, which is a large number of days compared to those of

the previous three years except March 2017. The rainfall increased in March 2019 compared to those of the previous three years. The number of days in March 2019 in which rainfall  $\geq 0.25$  mm was 5 days, while the number of days in March with rainfall of  $\geq 0.25$  mm was 1, 0, and 1 in 2016, 2017, and 2018, respectively. The results showed that despite the uncommon events in climate conditions in 2019, compared to the previous three years, the unusual increase in the intensity of rains in March 2019 with the availability of other climatic conditions could be the most important reason of fire blight disease outbreaks in this region of Egypt recently.

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