Arab Univ. J. Agric. Sci., Ain Shams Univ., Cairo, Egypt
27(4), 2115-2126, 2019
Website: http://ajs.journals.ekb.eg

OCCUREENCE OF LEAF SPOT DISEASE CAUSED BY *Xanthmonas campastris* AND *Pseudomonas cichorii* UNDER AQUAPONICS SYSTEM IN EGYPT

[168]

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Received 4 October, 2019 Accepted 1 December, 2019

ABSTRACT

Some plants grown in aquaponic system are affected by bacterial leaf spot disease, with equal adverse effect on market value. This research was conducted to evaluate leaf spot disease and the bacterial pathogens causative of the disease in aquaponic systems in Egypt. A leaf spot symptom consentient with bacterial leaf spot disease was observed on several plants grown in six different aquaponic farms in Egypt during 2016 and disease incidence (%) was recorded. Viable Count (log_{10} cfu/g) of pathogenic bacteria in cultivated water tank, infected plants and grow bed in six different aquaponic farms in Egypt was determined. In Al-Manashy farm, cultivated water tank and grow bed didn't included any pathogenic bacteria whereas these bacteria involved in different habitats of other tested farms. *Pseudomonas* sp. disappeared from grow bed of all tested farms except Al-Wahat and Al-Thawra Al-Khadra farm. The distribution of pathogenic bacteria, *Xanthomonas* and *Pseudomonas,* on infected plants was assessed in Al-Thawara Al-Khadra farm. Based on morphological and biochemical characterization and disease symptoms, the bacterial isolates belonged to the genera *Xanthomonas* and *Pseudomonas*. Pathogenicity test and host range of these isolates were determined on twelve different plants (six hybrids of lettuce, calendula, cherry tomato, basil, purple kale, hot pepper and spring onion). All the tested *Xanthomonas* isolates showed pathogenicity on all tested plants except calendula and spring onion, when leaves were sprayed with 1×10^8 cfu/ml of bacterial suspension. Out of nine, two *Pseudomonas* isolates, namely Ps1 and Ps 2, recorded the highest values of disease incidence on some tested plants. More virulence pathogenic bacteria of *Xanthomonas* and *Pseudomonas* were identified using 16S rRNA sequencing giving high identities with *Xanthomonas campestris campestris* and *Pseudomonas cichorii* respectively.

Keywords: Aquaponic system, Bacterial leaf spot, *Xanthomonas campestris*, *Pseudomonas cichorii*, 16S rRNA sequence

1. INTRODUCTION

Aquaponics is an intensive production system producing multiple crops with reduced water and fertilizer inputs and is highly suited for small farm producers targeting local markets and agritourism opportunities. Aquaponic systems are more complex than plant- or fish-only systems because the optimal conditions for each group of organisms; plants, fish, and the nitrifying bacteria **(Richard and Eric, 2017).** High-value vegetable crops that are commonly grown in hydroponic systems such as tomato, cucumber, pepper, lettuce, and mixed herbs are recommended for successful commercial aquaponic production (**Tyson et al 2013).**

Bacterial leaf spot or blight disease are important diseases of foliage and flowering ornamental plants as well as vegetables plants. These bacterial diseases may destroy leaves, petioles and stems rendering infected plants unsightly and unsalable. The disease is characterized by circular, gray to black, water-soaked lesions on the leaves. The lesions coalesce, become irregular in shape,

dry and dark brown with age **(Burgess et al 1986).** Among them, bacterial spot is the most problematic devastating disease in most of the tomato growing regions around the world **(Sharma and Bhattarai, 2019).** The incidence of bacterial disease ranged from 48 to 95% on lettuce in Turkey **(Ozyilmaz and Benlioglu, 2018).** Bacterial leaf spot of onion (*Allium cepa* L.) was observed in fields of Korea with incidence varying from 95 to 100%. Symptoms on leaves included leaf blight and white and brown spots on the leaf surface **(Myung et al 2011).** Two bacterial types were consistently isolated from the lesions of leaf spot. One type was off-white produced a green florescent pigment on King's B medium, identified as *Pseudomonas cichorii* **Burgess et al (1986)** and *Pseudomonas viridiflava* were isolated from the diseased marigold leaf tissues **(Moretti et al 2012).**

The second type produced yellow colony, it appears to be a species of *Xanthomonas* (**Burgess et al 1986).** The plant pathogenic bacterium *Xanthomonas campestris* is the causal agent of the serious diseases that denominated Xanthomonas-leaf spot which has been recorded on many araliaceous plants worldwide **(Chase, 1984).** The pathogen affects entire leaf surfaces and the typical disease symptoms include tiny yellowish lesions which are seen on the lower leaf surface as an initially symptom following stomatal infection **(Chase, 1984).** Bacterial spot, caused by a group of xanthomonads (*Xanthomonas* spp.), is a serious disease of tomato and pepper worldwide (**Sun et al 2002).** Bacterial leaf blight has been recorded on basil caused by *Xanthomonas campestris* **(Atit and Ranjan, 2015 ab)** and Pseudomonas *cichorii* (**Mustafa et al 2011).**

Therefore, the objectives of this study were to survey bacterial leaf spot, isolation and identification of the bacterial causal pathogens to determine its host range under aquaponic systems.

MATERIALS AND METHODS

2.1. Survey and isolation of bacterial leaf spot disease of plants grown under aquaponics systems in Egypt

Survey of leaf spot disease of six hybrids of lettuce (Buterhead, Oakleaf, Rocket, Red Baby Leaf, Watercress, Pock Choi), Purple Kale, Hot Pepper, basil, Calendula, Cherry tomato and Spring Onion plants were conducted in six aquaponic farms at different locations in Egypt, that is Al-Thawra Al Khadra, Al-Manashy, Al-Wahat, Al-Ismaiellia, Abo sultan and Al-Sadat. The number of plants showing leaf spot symptoms were recorded and the disease incidence of plants in each farm was calculated. The collected samples from different habitats of examined aquaponic farms (cultivated water tank, infected plants which have typical symptoms of leaf spot and grow bed) were brought to the laboratory for Isolation, identification and counting of the causal agents of leaf spot disease on CKTM **(Sijam et al 1991)** and King's B media (KB medium) (**King et al 1954**) for *Xanthomonas* spp. and fluorescent pseudomonades, respectively, by adopting the standard procedures to investigate the occurrence of these pathogens in each farm.

2.2. Assessment and isolation of causal bacterium of leaf spot disease

In Al-Thawra Al-Khadra farm, the cultivated plants in this farm were investigated for leaf spot disease. The diseased plants were collected to isolate the pathogenic bacteria; *Xanthomonas* and *Pseudomonas,* from young developing lesions to calculate the percentage of incidence for each pathogenic bacterium individually on tested plants.

2.3. Identification of collected isolates

The bacterial isolates were identified according to morphological and biochemical characteristics, as described by **Lelliot and Stead, (1987**), and **Klement et al (1990)** using the following procedures; staining the bacterial cells by Gram's staining, potassium hydroxide solubility for gram reaction, fluorescent pigmentation on KB, levan formation on nutrient agar plus 5 % sucrose, oxidase reaction, arginine dehydratase activity, catalase production, oxidative metabolism of glucose, reduction of nitrate to nitrites, gelatin liquefication, colony appearance on king's B, nutrient agar and YDC media, acid production from arabinose, glucose, mannose, lactose, maltose, sorbitol, sucrose as carbon sources, H_2S production, growth on 5%NaCl, relation to O_2 and hypersensitive on pepper leaves.

2.4. Pathogenicity tests and determination the host range of bacterial pathogens

Pathogenicity test and host range of the selected isolates were performed on several healthy plants, that is Butterhead, Red Baby Leaf, Oakleaf, Water cress, Pock Choi, Rocket, Calendula, Cherry tomato, basil, Purple Kale, Hot Pepper, and

Spring Onion, in Al-Thawra Al-Khadra farm. Inocula were prepared in liquid culture by growing *Xanthomonas* spp. on CKTM broth medium and *Pseudomonas* sp. on King's B broth medium for 48 h at 28°C which were adjusted to approximately equivalent to 1×10^8 cfu/ml.

Suspension of the bacterial culture was sprayed on the abaxial and adaxial side of plant leaves then plants were maintained in a mist chamber (>90% relative humidity) for 3-5 days at 25-28°C (**Klement et al 1990).** Five seedlings of tested plants (2 weeks old) were used for each tested isolate. For negative control, five seedlings were inoculated by sterile distilled water only. The seedlings were grown in plastic pots (10 cm) containing coco peat (7.5 gm) under greenhouse, plants were misted twice daily to keep high relative humidity, the symptoms were monitored daily and disease incidence was calculated after five days post inoculation. Re-isolation was performed from the leaf of the inoculated plants after the symptoms had appeared. Disease assessment was expressed as disease incidence using the following formula:

Disease incidence (%) =
(No. of infected plants / total plants) × 100.

2.5. Phylogenetic analysis

At the sub-species level, identification of *Xanthomonas* and *Pseudomonas* isolates were determined by polymerase chain reaction (PCR) amplification with specific probes and primers. The PCR amplification for RAPD analysis was performed according to **Williams, (1980).** The sequences of 16S specific primers and probe used for *Xanthomonas* sp. Were 16SF-CGGGCGTCCTCACAA ATTAC and 16SR-CCCCAAGTTGCCGGTTCC CC, and for *Pseudomonas* sp. were GACGGGT GAGTAATGCCTA and CACTGGTGTTCCTTCC-TATA

3. RESULTS

3.1. Survey of bacterial leaf spot disease of plants grown under aquaponics systems in Egypt

Six aquaponic farms in Egypt, cultivated with Lettuce Butterhead, Oakleaf Lettuce, Pock Choi, Rocket, basil, Hot Pepper, Spring Onion, Watercress, Cherry tomato, Calendula, Red Baby Leaf and Purple Kale plants, were surveyed for leaf spot disease incidence of those plants during 2016. The disease incidence percent ranged from 1- 17.6 % on tested plants. The highest values of percentage of leaf spot incidence were recorded on Lettuce Butterhead, Oakleaf Lettuce, Pock Choi, basil, Hot Pepper and Spring Onion at Al-Mnashy farm. In Al-Wahat farm, highest percentage of disease incidence were recorded in Red Baby Leaf, Lettuce Butterhead and Watercress at. While, Cherry tomato and Calendula recorded the highest of disease incidence at Al-Ismailia farm. Additionally, Rocket and Purple Kale recorded the highest disease incidence at Al-Sadat farm. Moreover, Basil recorded high value of disease incidence at Al-Thawra Al Khadra, Al-Mnashy and Al-Sadat **(Table 1).** It was observed that infected basil with the highest value were recorded in all tested farms.

3.2. Distribution and isolation of leaf spot bacterial pathogens under aquaponic systems in Egypt

Data in **Table (2)** shows the cell densities ($\log_{10}$ cfu /g) of Xanthomonas and Pseudomonas bacteria in three different habitats i.e., cultivated water tank, infected plants and grow bed of six aquaponic farms on selective media. In Al-Mnashy farm, cultivated water tank and grow bed didn't includ any pathogenic bacteria whereas these bacteria were detected in different habitats of other tested farms. *Pseudomonas* sp. were not found in grow bed of all tested farms except Al-Wahat and Al-Thawra Al-Khadra farm. In Al-Wahat farm, it was observed that *Xanthomonas* was recorded in infected plant samples. In Abo-Sultan farm, cultivated water tank, grow bed and infected plant recorded high densities of *Xanthomonas* whereas high population of *Xanthomonas* was recorded with infected plant only. The *Pseudomonas* was observed in cultivated water only in Al-Ismailia and Al-Sadat farm whereas other habitats didn't contain it.

Data in **(Fig. 1)** shows the percentage of infected plant (leaf spot disease) caused by Pseudomonas or *Xanthomonas* individually on some plants cultivated in an aquaponic farm in Egypt. Presence of *Xanthomonas* and *Pseudomonas* species on selective media was recorded from eleven plants (have typical leaf spot disease symptoms. Twenty percent of infected plants of lettuce butterhead, red baby leaf, water cress and hot pepper caused by *Xanthomonas* whereas the other infected plants recorded 10 % except calendula and spring onion which didn't infected. The percentage of infected plants by *Pseudomonas* ranged between 10-30% whereas hot pepper, purple kale and spring onion plants didn't have *pseudomonas* as a causal agent, none of pathogenic bacteria were recorded on spring onion plants.

Table 1. Survey of occurrence of bacterial leaf spot of plants grown under different aquaponic farms in Egypt during 2016 expressed as disease incidence (%).

Location of farm	Host plant											
	Hybrid of lettuce											
	Butterhead	Red Baby Leaf	Oak leaf	Water cress	Pock Choi	Rocket	Calendula	Cherry tomato	Basil	Purple Kale	Hot Pepper	Spring Onion
Al Thawra Al Khadra	5.0	2.1	4.4	1.8	3.2	5.5	4.5	2.1	8.8	12.1	10.0	1.0
Al-Mnashy	17.6	3.5	14.5	5.9	14.5	3.9	3.1	4.8	8.2	3.8	13.0	8.0
Al-Wahat	8.8	9.9	6.2	7.6	3.4	4.1	4.1	3.5	4.1	2.2	3.4	3.6
Al-Ismaiellia	1.2	3.3	5.6	4.6	5.6	5.9	7.4	15.0	6.5	3.8	2.3	3.0
Abo sultan	2.1	2.5	2.6	6.0	4.7	2.3	4.3	4.2	6.0	4.7	3.3	1.8
Al-Sadat	2.1	3.6	2.0	4.9	2.3	8.8	4.0	4.0	8.9	15.0	2.8	3.4

Table 2. Counting of pathogenic bacteria in different habitats (viable count $\log_{10}$ cfu /g) of tested aquaponic systems in Egypt.

Location	Xanthomonas count			Pseudomonas count		
	Cultivated Water tank	Infected plants	Grow bed	Cultivated Water tank	Infected plants	Grow bed
Al-Thawra Al-Khadra	3.0	0	2.8	3.2	3.0	6.0
Al-Mnashy	0	3.1	0	0	6.2	0
Al-Wahat	0	5.8	0	3.3	0	6.9
Al-Ismaiellia	3.2	3.4	3.0	3.5	0	0
Abo sultan	6.0	6.2	6.1	3.3	3.0	0
Al-Sadat	3.0	3.1	6.6	2.9	0	0

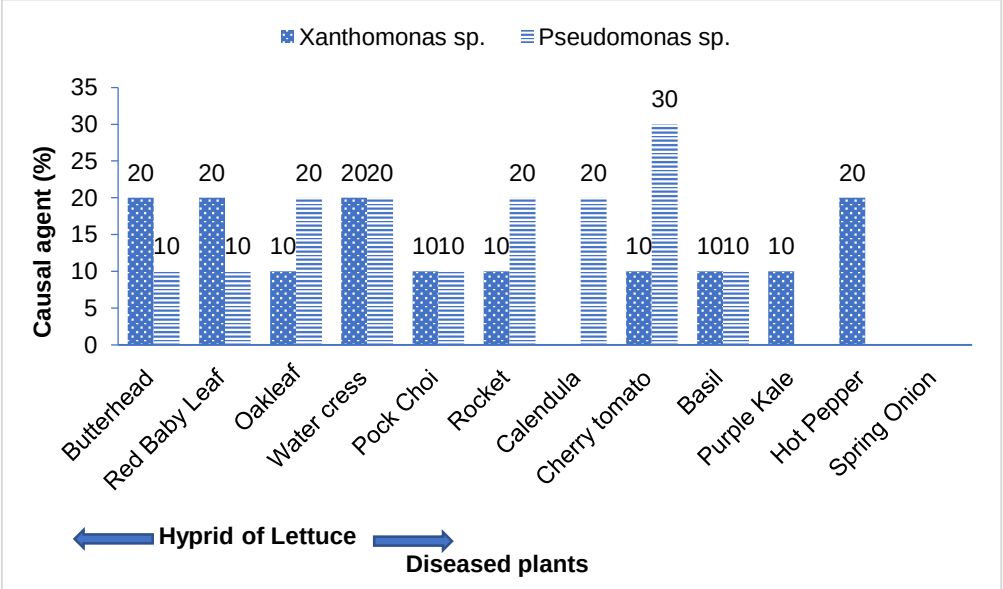

Fig. 1. Percentage incidence of leaf spot disease caused by *Xanthomonas* sp. and *Pseudomonas* sp. individually on diseased plants cultivate in aquaponic farm at Al-Thawra Al-Khadra in Egypt

3.3. Identification of pathogenic bacteria

According to the variations in morphological features, the isolated bacteria from naturally infected leaves of tested plants were found to belong to two genera; *Xanthomonas* and *Pseudomonas.*

3.3.1. Morphological and biochemical features

Ten *Xanthomonas* isolates (X1, X2, X3, X4, X5, X6, X7, X8, X9 and X10) were obtained. After 72 h of inoculation, the colony of *Xanthomonas* isolates on nutrient agar and YDC appeared as pale yellow, while on potato dextrose agar and yeast extract glucose chalk agar were empire yellow and yellow respectively. The colony shape on previous media was circular with entire smooth margin and no diffusible pigments. Convex elevation was observed on YDC, yeast extract glucose chalk and nutrient agar media but flat on potato dextrose agar. The morphological shape of cell was short rod, mostly singly and rarely in chains and gram negative, uniflagellate, motile and non-spore forming. These results were agreement with **(Viana 2006 and Tolba 2017).**

On nutrient agar and King's B media, the colonies of *Pseudomonas* isolates appeared straight rod, smooth margin, convex elevation, and greenish yellow diffusible fluorescent pigment. The color of colonies was green yellow with high diffusible fluorescent pigment (++) but on Nutrient agar medium colonies were white with little diffusible fluorescent pigment. The cells of Pseudomonas isolates were short rod shape, Gram negative, aerobic, non-spore forming and motile.

Morphological tests showed that these bacterial isolates could be classified as *Xanthomonas*, these isolates were short rod shape, motile, Gram negative

The biochemical tests confirmed that these bacterial isolates were classified as *Xanthomonas campestris,* where these isolates were aerobic, and gave positive reaction with KOH 3%, starch hydrolysis, catalase-positive, H_2S production, arginine dehydrolase, acid production from glucose and mannose, levan formation, growth on 5% NaCl and hyper sensitive on pepper. Negative reactions were recorded with gelatin liquification, oxidase activity, nitrate reduction and acid production from arabinose. According Bergey's manual of systematic bacteriology **(2000)** confirmed our results of identification of *Xanthomonas campestris.*

Results presented in **Table (3)** revealed that isolates could be classified as genus *Pseudomonas*. The biochemical tests of the second group isolates were classified as *Pseudomonas cichorii* where, these isolates were aerobic, and gave positive reaction with hypersensitive on pepper, oxidase activity, catalase activity and KOH 3%. But the negative reactions were recorded with starch hydrolysis, levan formation, gelatin liquification, H2S production, arginine dihydrolase, nitrate reduction, growth on 5% NaCl and acid production from glucose, arabinose and mannose according to **Lelliott and Stead, (1987).** The present results are in harmony with those reported by **Mustafa et al (2011) and Jagdale et al (2018).**

3.4. Virulence test and host rang assay

The host range of *Xanthomonas* and *Pseudomonas* isolates was determined on several plants belonging to different families i.e. six hybrid of lettuce (buttered, pock choi, red baby leaf, oakleaf, watercress, rocket), calendula, hot Pepper, basil, purple kale, cherry tomato and spring onion plants. As in naturally infected plants, similar leaf spot symptoms were observed evidently on the inoculated leaves. Within five days after inoculation, small water-soaked lesions were developed on the lower surfaces of inoculated leaves. These lesions coalesced and gradually turned chlorotic and the centers of lesions became brown and surrounded by greenish brown water-soaked, irregular margins. At last, the margins were raised, dried out then became corky **(Fig. 2).** The leaves of control plants remained healthy. The bacterium was readily re-isolated from symptomatic leaves. Data in **Fig. (3)** illustrated that, all the tested *Xanthomonas* isolates were pathogenic on all tested plants except on calendula and spring onion when leaves were sprayed with 1×10^8 cfu/ml of bacterial suspension. Three *Xanthomonas* isolates of ten, i.e., X1, X2, X3 were highly pathogenic where they recorded the highest disease incidence values. X1, X2 and X3 isolates were more virulence on cherry tomato and basil plants that they recorded 40-50 % disease incidence (%). X6 isolate caused 50 % as disease incidence on three hybrids of lettuce (water cress, pock choi and rocket). Three isolates namely (X4 X5 and X10) recorded the lowest values of disease incidence.

Table 3. Biochemical characteristics for classification of collected bacterial pathogens

Identification Tests	Xanthomonas isolates	Fluorescent Pseudomonads isolates
Levan formation	+	-
Starch hydrolysis	+	-
Gelatin liquefaction	-	-
hypersensitive reaction on pepper leaves	+	+
H2S production	+	-
Oxidase activity	-	+
Motility	+	+
Catalase activity	+	+
Arginin dihydrolase	+	-
Nitrate reduction	+	-
Relation to O_2	A	A
potassium hydroxide solubility	+	+
Growth on 5 % NaCl	+	-
acid production from arabinose	-	-
acid production from glucose	+	-
acid production from mannose,	+	-
Bacterial genera	*Xanthomonas campestris*	*Pseudomonas cichorii*

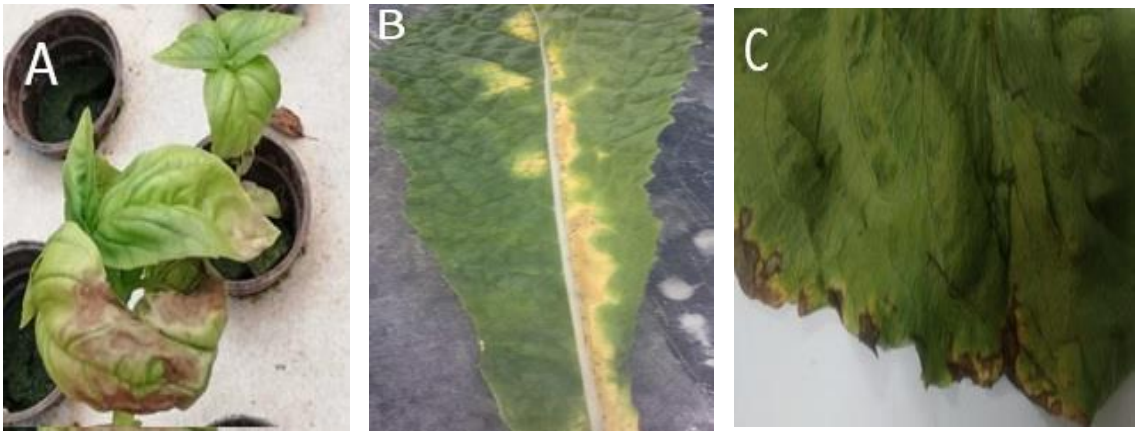

Fig. 2 Bacterial leaf spot symptoms on some plants (A: Basil, B: Purple Kale, C: Lettuce) grown in aquaponic system at Al Thawra Al Khadra.

Results in **Fig. (4)** indicated that, all the tested *Pseudomonas* isolates were pathogenic on all tested plants except spring onion, hot pepper and purple kale when leaves were sprayed with 1×10^8 cfu/ml of bacterial suspension. The leaves of control plants remained healthy. The bacterium was readily re-isolated from symptomatic leaves. Two *Pseudomonas* isolates of nine i.e., Ps1 and Ps2 recorded the highest values of disease incidence on some tested plants where they induce clear bacterial spot on leaves plants. While, the other isolates recorded low values of disease incidence ranged between 10-30 %. Calendula is highly sensitive for ps1, ps2, ps3 and ps4 pathogenic isolates recorded 50 % disease incidence. The obtained data could be interpreting in light of the findings of **Mustafa et al (2011)** who found that *Pseudomonas cichorii* produced symptoms on plants (tomato, lettuce & dwarf umbrella tree), but when the strains were inoculated to the original host, they were more aggressive than on the other species.

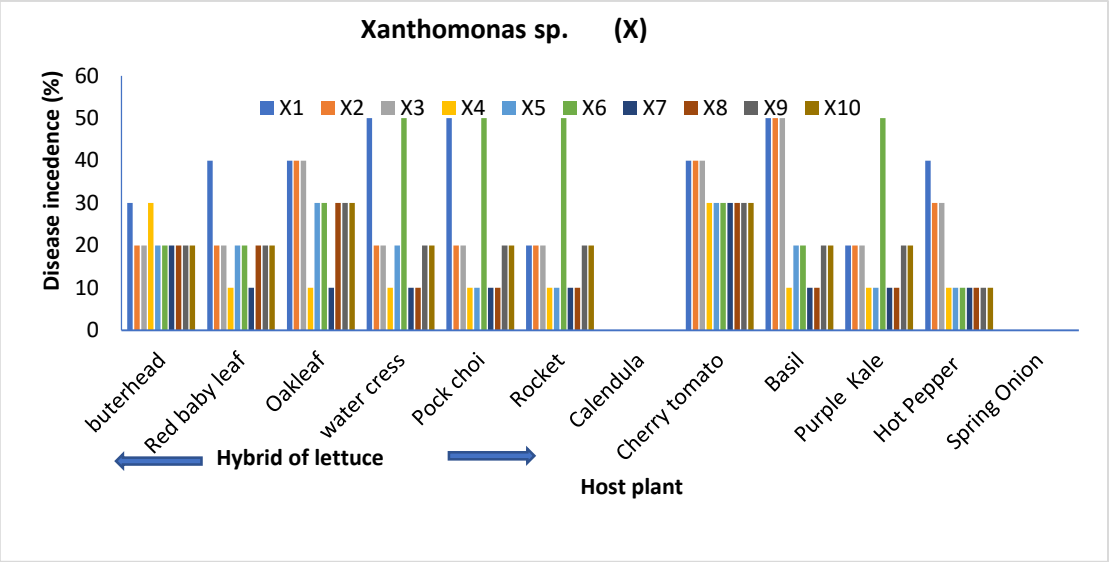

Fig. 3. Pathogenicity test of ten strains of *Xanthomonas* sp. isolates on several plants expressed as percentage of disease incidence under greenhouse conditions

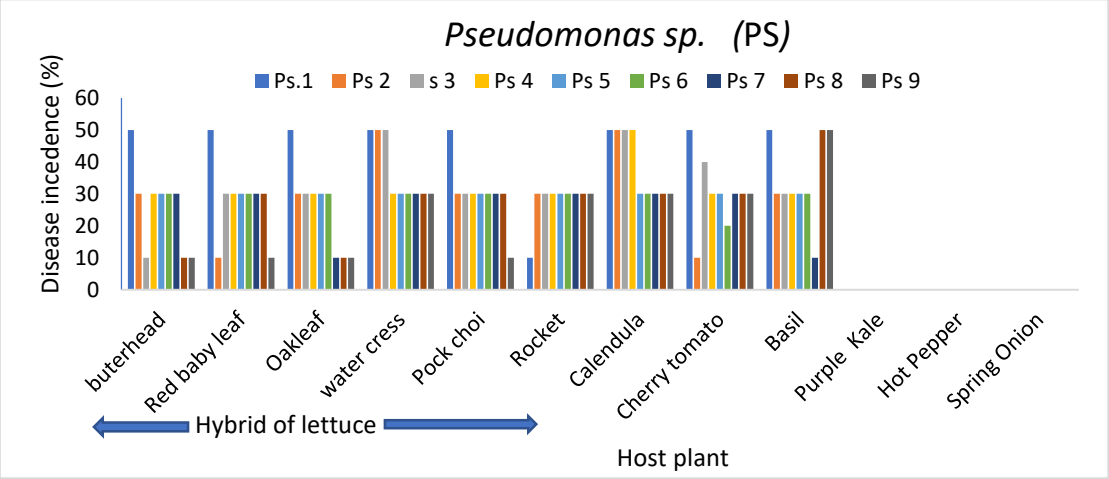

Fig. 4. Pathogenicity test of nine isolates of *Pseudomonas* sp. on several plants expressed as percentage of disease incidence under greenhouse conditions

3.3. Sequence analysis of 16S rRNA

The more virulent pathogenic bacterial isolates of *xanthomonas* and *pseudomonas* were identified by amplifying and sequencing the 16Sr RNA techniques. The results showed that 16S rRNA of X1, X2, X3, X6, X7, X8 and X9 isolates had 99% identities with *Xanthomonas campestris pv. campestris* and the results showed that 16S rRNA of ps1, ps2, ps8 and ps9 isolates had 98% identities with *pseudomonas cichorii* **(Fig. 5).**

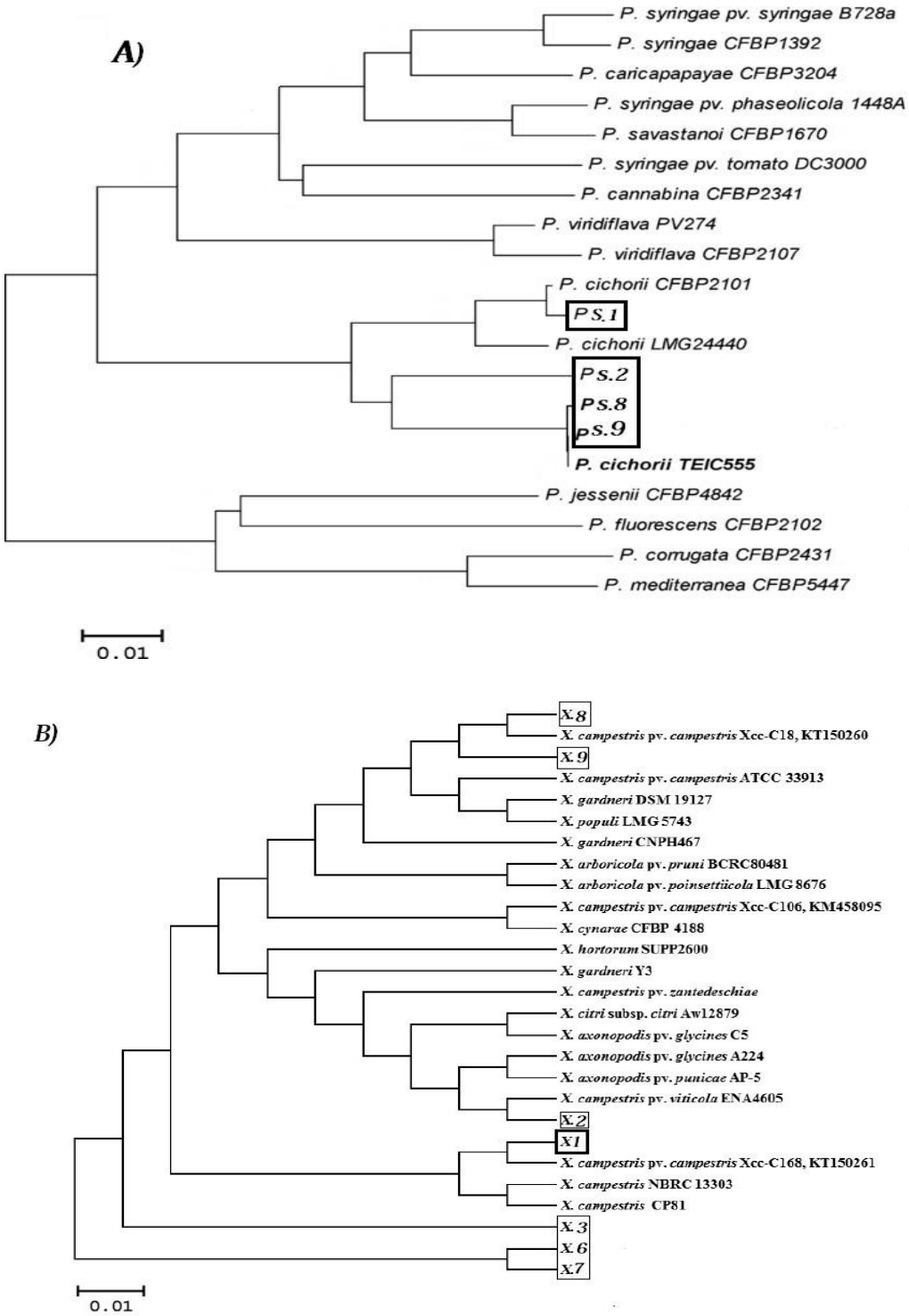

Fig. 5 Neighbour-joining phylogenetic tree, based on 16S rRNA gene sequences, showing the relationships between isolated bacteria of ps 1, ps 2, ps 8, ps 9 (A) and X1, X2, X3, X6, X7, X8, X9 (B) with related taxa**.**

4. DISCUSSION

Few reads assigned to the genus *Pseudomonas* were detected in the aquaponic systems. The number of assigned reads to the pseudomonads was in the same order of magnitude as previously observed by **Schmautz et al (2017).** *Pseudomonas* are usually found in close proximity to roots and are less prevalent in bulk soil **(Dennert et al 2018; Eck et al 2019).** The Oxalobacteraceae and Comamonadaceae families that can be found in water, soil and also in association with plants (**Baldani et al 2014; Schmautz et al 2017**) in aquaponic systems**. Eck et al (2019)** found xanthomonodaceae in the filter and water tank of some aquaponic system. According to the mentioned statement, our present study revealed to, the *Pseudomonas* and *Xanthomonas* were observed on plants, grow bed and water in aquabonic farm.

Appearance and the percentage of infected plant of (leaf spot disease) caused by pseudomonas or *xanthomonas* individually on some plants cultivated in an aquaponic farm in Egypt were recorded. These results could be discussed in light of the findings with **Wellman-Desbiens, (1998) and Fokkema, (1984**) who found that the dispersion of the bacteria in the greenhouse showed that many seedlings in greenhouse can be contaminated by overhead irrigation. *Xanthomonas* sp. can survive as an epiphyte on leaf surfaces before inducing symptoms. Several species of *Xanthomonas* cause bacterial leaf spot, disease that affects solanaceous crops worldwide **(Roach et al 2018). Moreira et al (2015)** proved that, leaf spot of basil is caused by the bacterium *Pseudomonas cichorii.* Morphological and biochemical features of Xanthomonas isolates were in the accordance of results published by **Viana, (2006) and Tolba, (2017)** who identify the isolates of *Xanthomonas campestris* pv. *hederae* obtained from symptomatic plants which were characterized based on physiological and biochemical tests, pathogenicity test. Compare the efficacy of some media used in the selective isolation of *Pseudomonas* species was studied by **Peekate et al (2018)** who found that Cetrimide agar is a better medium for the selective enumeration and isolation of greenish pigment producing *Pseudomonas* species.

Atit and Ranjan, (2015 a,b) identified the *Xanthomonas campestris* pv. *campestris* (Xcc) based on Morphological, cultural, biochemical, and physiological characteristics of the pathogen, the bacterium were small, yellow, circular, entire, smooth and shining colonies in the culture medium.

All the tested *Xanthomonas* isolates were pathogenic on all tested plants except on calendula and spring onion and causal bacterial leaf spot and necrosis on leaves. These results are in agreement with those obtained by **Massomo et al (2003)** who stated that the pathogenic strains of *Xanthomonas campestris pv. campestris* caused black rot on smallholder cabbage (*Brassica oleracea* var. *capitata*). The symptoms of this pathogen on the plant *Schefflera actinophylla* were small greyish spots around the stomata followed by characteristic necrotic V-shaped lesions on the margins of leaves with chlorosis. Subsequently, vein blackening and wilting of leaves occurred **(Tolba, 2017).**

5. CONCLUSION

In the current investigation, we were interested in the presence of the bacteria, *Xanthomonas* and *Pseudomonas* causing bacterial leaf spot on six hybrids of lettuce (Butterhead, Oakleaf, Rocket, Red Baby Leaf, Water cress, Pock Choi), Purple Kale, Hot Pepper, basil, Calendula, Cherry tomato and Spring Onion plants which grown in aquaponic farm in Egypt, the study was to gain insight into the diversity of the bacterial pathogens communities in aquaponic systems

ACKNOWLEDGMENTS

The authors would like to thank the different partners that agreed to let us visit and sample their systems. The authors would like to acknowledge the Al-Thawra Al-Khadra farm for financial support.

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مجلد(27)، عدد(4)، 2115-2126، 2019
Website: http://ajs.journals.ekb.eg

تواجد مرض تبقع الاوراق المتسبب بواسطه الزانثوموناس كامبسترس والسيدوموناس كيشورى تحت نظام الاكوابونيك فى مصر

[168]

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Received 4 October, 2019 Accepted 1 December, 2019

الموجـــــــز

تتأثر بعض النباتات التي تزرع في نظام الاكوابونيك بمرض التبقع البكتيرى، مع تأثير سلبي مماثل على القيمة السوقية. أجري هذا البحث لتقييم مرض تبقع الأوراق ومسببات الأمراض البكتيرية المسببة لهذا المرض في النظم المائية في مصر. لوحظت أعراض التبقع الورقي الدالة على مرض التبقع البكتيري في العديد من النباتات التي نمت في ستة مزارع مائية مختلفة في مصر خلال عام 2016 وسجلت شدة الإصابة بالمرض (٪). تم تحديد أعداد البكتيريا المسببة للأمراض في خزان (log$_{10}$ cfu / g) المياه، والنباتات المصابة واصيص الزراعه في ست مزارع مائية مختلفة في مصر، في مزرعة المناشي، لم يشتمل خزان المياه المزروعة واصيص الزراعه على أي بكتيريا مسببة للأمراض، في حين أن هذه البكتيريا وجدت في بيئات مختلفة من المزارع الأخرى المختبرة. اختفت بكتيريا السيدوموناس من اصص الزراعه في جميع المزارع المختبرة بإستثناء مزرعة الواحات والثورة الخضراء. تم تقييم توزيع البكتيريـــــا المسببـــــة للأمـــــراض، الزانثوموناس والسيدوموناس على النباتات المصابة في مزرعة الثورة الخضراء. بناءً على التوصيف المورفولوجي والكيميائي الحيوي وأعراض المرض فإن العزلات البكتيرية تنتمي إلى الأجناس الزانثوموناس والسيدوموناس. تم تحديد اختبار الإمراضية والمدى العوائلى لهذه العزلات على اثني عشر نباتًا مختلفًا (ستة أنواع هجينة من الخس، الكالنديولا، طماطم شيرى، ريحان، اللفت الأرجواني، فلفل حار وبصل أخضر). أظهرت جميع عزلات Xanthomonas التي تم اختبار قدراتها المرضية على جميع النباتات التي تم اختبارها بإستثناء الكالنديولا والبصل الأخضر، عندما تم رش الأوراق بإستخدام 1 × 10^8 cfu/ml من المعلق البكتيري. من أصل تسعة، سجل اثنان من عزلات السيدوموناس، وهما Ps1 وps2، أعلى قيم لحدوث الأمراض في بعض النباتات التي تم اختبارها. تم تعريف البكتيريا المسببة للأمراض من Xanthomonas و Pseudomonas بإستخدام تسلسل 16 S rRNA لإعطاء نسب تشابه عالية مع *Xanthomonas campestris campestris* و *Pseudomonas cichorii* على التوالي .

الكلمات الداله: نظام الاكوابونيك، تبقع الاوراق البكتيرى، زانثوموناس كامبسترس، سيدوموناس كيشورى، 16s rRna

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