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# A New Type of *Acinetobacter* spp. *Attacking Ziziphus spina-christi* (L.) Willd Tree in Iraq

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**Abstract.** The principal objective of this experiment was to determine the type of bacterial associated with *Ziziphus spina-christi* (L.) tree showed individual colonies of *Acinetobacter* spp. and Molecularly diagnosed by using molecular diagnostic technology ( PCR) polymerase chain reaction by using some biochemical tests. It is the first recording of *Acinetobacter Ziziphus spina-christi* (L.) tree in Iraq.

**Keywords**: *Acinetobacter* spp., PCR. *Ziziphus spina-christi* (L.)willd , Iraq.

## Introduction

The *Ziziphus spina-christi* (Rhamnaceae) is a perennial tree , its grows in middle and southern Iraq. It is cultivated for timber, fruit, fodder for livestock, a dune stabilizer, because *Ziziphus spina-christi* has deep roots which is spreading lateral roots and is used as traditional medicine(AL-Dawoody,1979 and Saied and others ,2008).

The *Acinetobacter* spp. dates back to the 20th century when described from the Dutch microbiologist Beijernick (Kay and others , 2002). *Acinetobacter* spp. was widely distributed in nature and commonly occurs in water and soil as well as *Acinetobacter* spp. strains was isolated from fruits and vegetables (Berlau and others,1999). The *Acinetobacter* spp. have been isolated from different plants (Kuan and others, 2016). There are more than fifty species which are mostly non-pathogenic environmental organisms(Wong and others,2017).

The current study aimed at isolating and molecularly characterizing the bacterial associated with *Ziziphus spina-christi* trees.

## Materials and Methods

Isolation

Samples were brought from the stem of the *Ziziphus spina-christi* (L.) trees which were appeared with symptoms . isolated bacterial were cultured in nutrient agar (AN) after purification for twenty four hours at 30 ±2°C.

Biochemical tests of *Acinetobacter* spp.

Biochemical characteristics of the *Acinetobacter* spp. were examined by using some methods which include:-

Schaad (1988) and Holt and others (1994) and Goszczynska and others (2000) & Winn and others (2006). growth at 37°C , Levin formation, grow on 2% NaCl, Catalase and Gram stain.

Identification of *Acinetobacter* spp. PCR

The bacteria were diagnosed by using polymerase chain reaction technique in center- Asco-learning,Baghdad,Iraq.

The extracted DNA (rDNA) pieces were amplified employment types primers were used (F968) & R1401 which targeting 16S rDNA gene (Nübel and others, 1996). In Macrogen_ Inc, Seoul South Korea the PCR outcome was sequenced.

The results of nucleotide sequencing were compared at GenBank, NCBI with other sequences of bacterial applying for the (BLAST) program Basic- Local- Alignment Search Tool (Zheng and others, 2000).

## Results and discussion

Biochemical tests of *Acinetobacter* spp.

Introductory identification of isolatethe genus *Acinetobacter* spp. in depending on the cultural , morphological and some biochemical characteristics. The Acinetobacter spp. was rod shaped , motile, Gram-negative, positive Catalase and negative growth at 37°C, positive Levin formation and positive growth on 2% NaCl (Table 1) .These results are consistent with Lee and others (2017).

**Table 1**. Biochemical tests for *Acinetobacter* spp.

| S. | Biochemical tests | Result |
|---|---|---|
| 1 | growth at 37°C | - |
| 2 | Catalase test | + |
| 3 | Levin formation | + |
| 4 | Gram stain | - |
| 5 | growth on 2% NaCl | + |

**Fig 1**. The natural infection of the Ziziphus spina-christi tree.

Identification of Acinetobacter spp. bacterial by PCR technique

The isolation was diagnosed after 16S rDNA sequence analysis as Acinetobacter spp.the sequence was put in the Genbank National Center of Biotechnology Information) NBCI) which on record at the serial number MW82538.1, its the first record on Ziziphus spina-christi trees in Iraq.

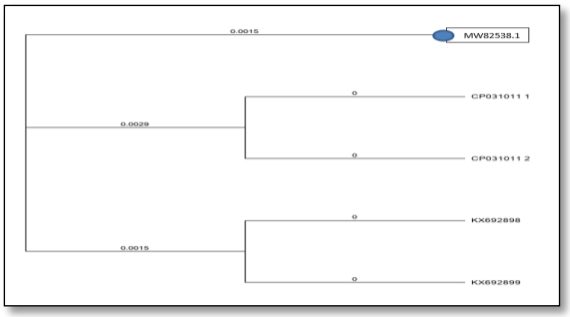


**Fig 2**. Genetic tree of Acinetobacter spp.

The technique of polymerase chain reaction (PCR) used in former experiments because its high precision in the diagnosis of different organisms which include bacterial such as *Pseudomonas grimontii* , *Agrobacterium tumefaciens* (Sawada and others ,2020 and Al-Tememe and others,2020).

## Conclusions

The results showed that the the first reported occurrence of Acinetobacter spp . in Iraq, there are no studies abaut this bacterial accompanying Ziziphus spina-christi trees in Iraq.

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