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Detection of Soil-Borne Bacterial Diseases in Potato Cultivation in Niğde Province

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Abstract

Potato (*Solanum tuberosum L.*) is a strategically important crop worldwide, and Türkiye's Niğde province has the largest potato cultivation area in the country. However, soil-borne bacterial diseases including blackleg, soft rot, bacterial wilt, ring rot and common scab cause substantial yield and quality losses. Direct detection of these pathogens from field soils has been rarely performed. This study aimed to isolate and characterize bacterial pathogens from soil samples collected from potato production fields in Niğde. Ten soil samples were collected from the vicinities of symptomatic plants in Hasaköy, Alay and Altunhisar districts. The serial dilution method was used to inoculate samples onto Nutrient Agar. Morphologically distinct bacterial isolates were obtained. Pathogenicity tests were performed on 21 isolates, and ten isolates tested positive in both the potato disc test and the tobacco hypersensitivity test. Genomic DNA was extracted from these ten isolates, followed by PCR amplification of the 16S rRNA gene region and Sanger sequencing. BLAST analysis identified isolates as members of bacterial genera commonly associated with potato diseases, including *Streptomyces* spp., *Bacillus* spp., *Chryseobacterium* spp., and *Pectobacterium* spp. These results demonstrate that diverse soil-borne bacterial pathogens are present in Niğde's potato production areas. Therefore, regular soil monitoring and integrated pest management are essential to reduce disease risks and sustain potato production in the region.

Keywords: *Potato; Soil-borne diseases; Bacterial pathogens; Isolation and characterization; Pathogenicity test; Niğde province*

Introduction

Potato (*Solanum tuberosum L.*) is a strategic crop from the *Solanaceae* family and stands as one of the world's primary food sources. Owing to its high yield potential and relatively low production costs, it contributes significantly to global food supply and ranks fourth among staple foods, after cereals (Yılmaz et al., 2006; Wang et al., 2021). In 2024, potato is cultivated in approximately 153 countries worldwide, with a total production of 390.4 million tonnes. The major producing countries include China, India, Ukraine, and the United States of America. In Türkiye, total potato production amounted to 6.9 million tonnes in 2024, cultivated across a harvested area of 195,767 hectares, with an average yield of approximately 35.3 t ha^{-1} (FAO, 2025). Production is heavily concentrated in the provinces of Niğde, Konya, Afyonkarahisar, İzmir, and Nevşehir. According to 2025 data, Niğde ranked first with 884,023 tonnes, followed by Konya with 837,272 tonnes, Afyonkarahisar with 648,029 tonnes, İzmir with 339,920 tonnes, and Nevşehir with 295,089 tonnes (TÜİK, 2025)

Potato yield is strongly limited by both biotic and abiotic factors (Agrios, 2005). Among biotic factors, bacterial, fungal and viral diseases cause annual yield losses of up to about 22% worldwide (Czajkowski et al., 2011). Within these, soil-borne bacterial pathogens are particularly important because they severely affect both yield and quality. The main bacterial pathogens found in potato production include *Pectobacterium* spp. and *Dickeya* spp., which cause blackleg and soft rot, as well as *Ralstonia solanacearum*, the agent of bacterial wilt, *Clavibacter michiganensis* subsp. *sepedonicus*, the cause of bacterial ring rot, and *Streptomyces* spp., which cause common scab (Ünlü & Elçi, 2022).

Pectobacterium and *Dickeya* species are Gram-negative bacteria. They secrete pectinolytic enzymes, producing symptoms characterized by soft rot in plant tissues and blackening of the stem (Czajkowski et al., 2011; Pérombelon & Kelman, 1980). *Ralstonia solanacearum*, the causal agent of bacterial wilt, is a Gram-negative, soil-borne pathogen with a broad host range. It enters plants mainly through the roots, colonizes the vascular system, and disrupts water transport, leading to wilting, chlorosis, and eventual plant death (Ahmed et al., 2013). *Clavibacter michiganensis* subsp. *sepedonicus*, the causal agent of bacterial ring rot, is another highly significant pathogen, which causes typical symptoms including vascular tissue rot, stunting (dwarfing), leaf curling, and necrosis in infected plants (Whitworth et al., 2019). Pathogenic species belonging to the genus *Streptomyces*, which are commonly found in soil, cause common scab. This disease lowers the commercial value of the crop by forming superficial or deep lesions, especially on tubers. The severity of symptoms varies depending on pathogen density, environmental conditions and variety susceptibility (Bouchek-Mechiche et al., 2006; Charkowski et al., 2020; Uysal et al., 2025). *Chryseobacterium* spp. are yellow-pigmented bacteria, and some species have been reported to promote plant growth and suppress pathogens through various mechanisms (Morohoshi et al., 2014; Jeong et al., 2017). However, certain species within this genus act as opportunistic pathogens and have been associated with potato diseases; for instance, *Chryseobacterium mulctrae* has been reported to cause soft rot in potato tubers (Shrestha, 2025). *Bacillus* spp. are among the plant growth-promoting rhizobacteria (PGPR) and are

known to suppress plant pathogens through various mechanisms (Khan et al., 2022). However, some *Bacillus* species have also been reported to act as plant pathogens under certain conditions; for instance, *Bacillus amyloliquefaciens* has been reported to cause soft rot in potato tubers (Wang et al., 2017). These findings indicate that both *Bacillus* spp. and *Chryseobacterium* spp. may exhibit both beneficial and potentially pathogenic characteristics.

In the literature, studies on potato bacterial diseases have largely relied on isolations from diseased plant samples. However, with respect to the epidemiology of soil-borne pathogens, differing hypotheses exist as to whether the main source of inoculum is infected seed tubers or the soil itself (Czajkowski et al., 2011; De Boer, 2004). Although it has been proposed that the role of the soil may be more dominant, especially for pathogens such as *Streptomyces*, which are naturally present and persistent in the soil as saprophytic microorganisms, comprehensive studies on the prevalence of soil-borne inoculum in intensive production regions remain limited (Lerat et al., 2009).

This study was carried out in Niğde Province, a major potato production center in Türkiye. Soil samples collected from potato-growing areas were used for the isolation, identification, and characterization of bacterial populations associated with potato fields. The study focuses on determining the presence and diversity of soil-borne bacterial microorganisms in the region, with particular attention to potential plant pathogenic species.

Materials and Methods

Sampling and Soil Collection

A total of ten soil samples were taken from around symptomatic potato plants in Hasaköy, Alay and Altunhisar districts of Niğde province. The samples were collected carefully without disturbing the root zone of the plants. They were kept at +4°C until analysis. Reference pathogen isolates were obtained from the culture collection of the Plant Pathology Laboratory at Niğde Ömer Halisdemir University.

Bacterial Isolation

One gram of each soil sample was homogenized in 10 ml of sterile distilled water. Serial dilutions were made from 10^{-1} to 10^{-5}. From the 10^{-3} and 10^{-5} dilutions, 0.1 ml was spread onto Nutrient Agar and incubated at 29.5°C for 24-48 hours. Colonies showing different morphologies were purified and grown on Nutrient Agar and then transferred to Nutrient Broth. The isolates were finally kept at -80°C in 50% glycerol.

Pathogenicity Tests

Potato Disc Test

Healthy potato tubers were surface-sterilized by soaking in 1% sodium hypochlorite for 3 minutes. They were then rinsed with sterile distilled water and cut into discs about 1 cm thick. The discs were placed in petri dishes. Bacterial cultures grown on NA for 24-48 hours were taken with a sterile toothpick and inoculated onto the disc surfaces. One milliliter of sterile water was added to each dish, and the dishes were sealed with parafilm before incubation. A reference isolate was used as a positive control, and sterile distilled water served as the negative control.

Hypersensitivity (HR) Test

Bacterial suspensions were injected into wounds made on tobacco leaves (*Nicotiana tabacum*) with a sterile needle. Positive and negative controls were applied in the same way. The leaves were then observed for necrosis. Isolates that caused necrosis were recorded as HR positive.

Molecular Identification

Genomic DNA was extracted from pathogenic bacterial isolates using the Zymo Quick-DNA™ Fungal/Bacterial Microprep Kit (D-6007) and stored at −20°C until further use. Molecular identification of the isolates was performed by amplification of the 16S rRNA gene using the universal primers 27F and 1492R. Polymerase chain reaction (PCR) was carried out under appropriate reaction conditions using a PCR Master Mix. PCR products were visualized by agarose gel electrophoresis. Subsequently, amplified DNA fragments were subjected to Sanger sequencing, and the obtained sequences were compared with those available in the NCBI-BLAST nucleotide database for species-level identification.

Results

Bacterial isolation was performed using the serial dilution method, and colonies showing different morphological characteristics were purified. A total of 27 isolates were re-evaluated based on colony morphology, and after removing duplicates with similar features, 21 isolates were selected for further analyses. Images of petri dishes of some isolates are shown in Figure 1.

In the potato disc test, 10 isolates induced tissue degradation, whereas 17 isolates caused necrosis on tobacco leaves. Ultimately, the 10 isolates that exhibited positive reactions in both tests were confirmed as pathogenic. No symptoms were observed in the negative controls, while the positive control reference strains yielded the expected symptoms.

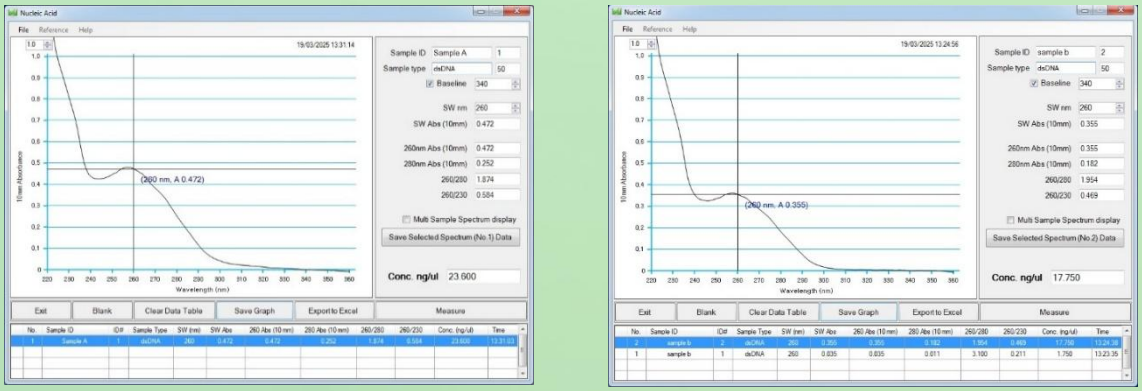
Genomic DNA was extracted from 10 bacterial isolates identified as potential pathogens. The quality and concentration of the obtained DNA samples were evaluated using spectrophotometric measurements and found to be suitable for molecular analyses (Figure 2). The extracted DNA samples were subsequently used for PCR amplification and molecular identification of the bacterial isolates.

For molecular identification, the 16S rRNA gene region of bacterial isolates was amplified by PCR using genomic DNA and universal primers 27F/1492R. The PCR products were visualized by agarose gel electrophoresis, and the expected bands were observed (1568 bp). The selected PCR products were sent for Sanger sequencing analysis.

The obtained 16S rRNA sequence data were compared in the NCBI-BLAST database, and the isolates were identified at the genus level. The analysis revealed *Streptomyces* spp., *Bacillus* spp., *Chryseobacterium* spp., and *Pectobacterium* spp. BLAST results for *Chryseobacterium* spp. and *Pectobacterium* spp. isolates are presented in Figure 3.

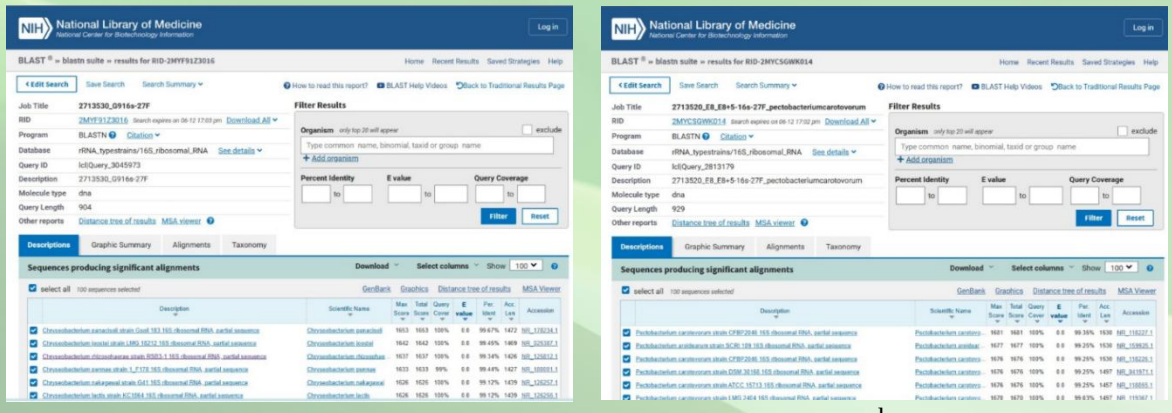
a b c

Figure 1. Images of petri dishes of some isolates (a, b, c).



a b

Figure 2. Concentration and purity analyses of genomic DNA from some bacterial isolates were performed using a NanoDrop spectrophotometer. DNA concentrations were determined as 23.6 ng/µL (a) and 17.75 ng/µL (b).



a b

Figure 3 NCBI-BLAST comparison results of 16S rRNA gene sequences from *Chryseobacterium* spp. (a) and Pectobacterium spp. (b) isolates.

Discussion

Bacterial diseases of potato are caused by several important pathogens in production areas in Türkiye. Among these, *Pectobacterium* spp. and *Dickeya* spp. are responsible for blackleg and soft rot diseases, *Streptomyces* spp. causes common scab, *Ralstonia solanacearum* causes bacterial wilt, and *Clavibacter michiganensis* subsp. *sepedonicus* is the causal agent of bacterial ring rot. In particular, *Pectobacterium* and *Dickeya* species are considered among the most economically important bacterial pathogens worldwide due to their significant impact on yield and tuber quality (Mansfield et al., 2012).

Previous studies conducted in the Central Anatolia Region have demonstrated the widespread occurrence of bacterial pathogens in potato fields. Benlioğlu (1991) reported blackleg and soft rot agents in Niğde and Nevşehir as *Erwinia carotovora* subspecies. Öztürk et al. (2018) identified *Pectobacterium carotovorum* subsp. *brasiliense*, *Pectobacterium parmentieri* and *Pectobacterium atrosepticum* in the Central Anatolia and Black Sea regions. In addition, Ünlü and Elçi (2022) confirmed the presence of *Streptomyces scabiei* as the causal agent of common scab in Niğde province at the molecular level. These findings indicate the long-term presence of bacterial pathogens in potato production areas of the region.

In the present study, *Pectobacterium* spp. (blackleg and soft rot), *Streptomyces* spp. (common scab), *Bacillus* spp., *Chryseobacterium* spp., and *Clavibacter michiganensis* subsp. *sepedonicus* were identified. The results are consistent with previous reports, particularly regarding the presence of *Pectobacterium* and *Streptomyces* species. Moreover, the detection of *Bacillus* spp. and *Chryseobacterium* spp. suggests that, in addition to well-known pathogenic groups, other bacterial groups are also present in the region's potato fields.

Conclusion

Bacteria belonging to the genera *Pectobacterium* spp., *Streptomyces* spp., *Bacillus* spp., and *Chryseobacterium* spp. were detected in soil samples collected from potato fields in Niğde Province. These findings highlight the presence and diversity of soil-borne bacterial communities associated with potato-growing areas in the region and suggest a potential risk for bacterial diseases under field conditions. Species-level identification is required to better understand the pathogenic potential of the isolates, as 16S rRNA gene analysis alone is insufficient for precise taxonomic resolution, and species-specific approaches are necessary for accurate characterization.

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