



Assessment of the resistance of modern apple varieties to the bacterial blight pathogen *Erwinia amylovora*

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Abstract. Modern requirements for new apple varieties include not only high yield and improved fruit quality, but also resistance to the most dangerous diseases, including bacterial canker caused by *Erwinia amylovora*. The aim of this study was to evaluate the resistance of modern apple varieties common in Kyrgyzstan to the causative agent of fire blight under artificial infection conditions. For the experiment, a pure culture of the phytopathogenic bacterium *E. amylovora* (strain ZH-2) isolated from infected branches of the 'Talgarka' pear variety was used. Infection was carried out *in vitro* by applying a bacterial suspension with a concentration of 10^9 CFU/ml to the wound surfaces of shoots and leaves of two-year-old seedlings. The degree of damage was assessed visually 1, 6, 12 and 30 days after inoculation on a five-point scale, taking into account the percentage of damaged tissue. The results of the study showed significant differences in resistance between the eleven apple varieties studied. The most resistant varieties were 'Red Chief', 'Bishkek', and 'Golden', with damage levels ranging from 6.3 to 9.5%, indicating strong defensive responses and low levels of necrosis symptoms. The 'Fuji' variety showed an average level of susceptibility (about 16-18%), characterised by moderate damage to shoot tissues. The most susceptible to infection were the varieties 'Idared' (48%), 'Gala' (43%) and 'Ligolina' (37%), which showed rapid development of necrotic spots and wilting of shoots. The results obtained allow for identifying varieties that are promising for further propagation and breeding work, resistant to bacterial burn, and also give an idea of the range of susceptibility of modern apple varieties in the conditions of Kyrgyzstan

Keywords: artificial infection; degree of damage; phytopathogenic bacteria; tissue necrosis; *in vitro*

Introduction

The domestic apple tree (*Malus domestica* Borkh.) is one of the most important fruit crops. According to FAO (n.d.), annual global apple production exceeds 87.2 million tonnes; China is the largest producer

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(41.39 million tonnes), followed by the United States and other countries. Kyrgyzstan ranks 45th in the world in apple production: approximately 147,593 tonnes. Therefore, controlling pests and diseases of apple trees is crucial to ensuring crop stability, improving product quality and expanding export potential. According to R. Yuvarani *et al.* (2025), the development of intensive horticulture, the introduction of disease-resistant varieties, the use of modern plant protection methods and the monitoring of the phytosanitary condition of plantations can significantly reduce losses from dangerous infections such as bacterial burn. At the same time, strengthening the Kyrgyz Republic's position in the global fruit and vegetable market and meeting the domestic market's demand for high-quality fruit products is closely linked to the effective protection of apple orchards. Bacterial fire blight of fruit crops is one of the most dangerous diseases of apple trees and other plants of the Rosaceae family, caused by the bacterium *Erwinia amylovora*. It is a quarantine disease and poses a serious threat to fruit plantations (Aćimović *et al.*, 2023; Moskalets *et al.*, 2023). The disease causes necrosis of all plant organs, leading to significant crop losses and tree death. The main strategy for combating the disease traditionally remains the destruction of infected trees, treatment with copper-containing preparations and other pesticides, and in some countries, the use of antibiotic solutions. However, the widespread use of such preparations causes the emergence of drug-resistant bacteria. For example, a study by A. Jimenez Madrid & M. Lewis Ivey (2023) revealed the strain resistance of *E. amylovora* to streptomycin and other antibiotics.

In the context of new developments in the field of protecting apple trees from bacterial blight, the following areas are particularly relevant. Firstly, genotypic and phenotypic assessment of the resistance of varieties and rootstocks. G. Kairova *et al.* (2023a), based on the results of a study of 59 apple varieties in Kazakhstan, found that in all genotypes studied, the markers of resistance to *E. amylovora* (e.g., AE10-375 and GE-8019) were identical in all genotypes studied, indicating limited existing genetic variability for resistance. Similarly, a study of 11 apple rootstocks in Kazakhstan revealed

that the own-rooted rootstock lines G.41, G.16 and “62-396” show the best resistance to fire blight (Kairova *et al.*, 2023b). Secondly, a study of the wild apple tree *Malus sieversii*, the ancestral species of the domestic apple tree, revealed the presence of genotypes with high resistance to *E. amylovora* (Tegtmeier *et al.*, 2025). Thirdly, genomic and associative mapping of resistance to fire blight allow the identification of key regions and genes. R. Thapa *et al.* (2021) identified 23 and 38 quantitative trait loci (QTLs) significantly associated with resistance to shoot and flower blight, respectively. Finally, alternative environmentally friendly control approaches are emerging: for example, the isolation and characterisation of three bacteriophages infecting *E. amylovora* as a potential biological control agent (Hassan *et al.*, 2023). Thus, the relevance of the study lies in the need to assess the resistance of modern apple varieties and rootstocks to the bacterial blight pathogen *E. amylovora* in the conditions of the Kyrgyz Republic, with the aim of identifying the most promising ones for industrial and amateur horticulture and their rapid introduction into practice. In the context of climate change, increased international trade and stricter quarantine requirements, this approach is becoming particularly important.

The aim of this study was to assess the resistance of modern apple varieties to the bacterial blight pathogen (*Erwinia amylovora*) by conducting artificial infection of plants and analysing the severity of external symptoms of the disease in order to identify the most resistant varieties that are promising for cultivation in the Kyrgyz Republic.

Materials and Methods

Modern apple varieties (*Malus domestica* Borkh.), mainly imported from other countries and widely used by gardeners planting trees on their plots in Kyrgyzstan, were used as research objects. The selection of varieties was based on their popularity and official recommendations. The study was conducted at the experimental stationary site of the Kyrgyz-Turkish Manas University, Faculty of Agriculture (Bishkek, Kyrgyz Republic). For the experiment, two-year-old seedlings of 11 different apple varieties were planted in individual pots. A brief description of the varieties is given in Table 1.

Table 1. Characteristics of apple varieties grown in the Kyrgyz Republic

No.	Variety name	Origin	Brief description	Included in the state register
1	'Starkrimson'	American selection (Genus Star Delicious)	American apple tree variety. Ripening period: autumn-winter; apple taste: sweet and sour; average weight: medium (100-200 g); fruit colour: greenish-red, dark red; flesh colour: light yellow; transportability: good; frost resistance: weak; disease resistance: powdery mildew.	Introduced, regionalised in Kyrgyzstan and registered in 1980
2	'Red Chief'	American selection (Genus Red Delicious)	American apple tree. Ripening period: autumn-winter; apple taste: sweet; average weight: large (over 200 g); fruit colour: deep red; flesh: red-yellow, dense, medium juiciness; transportability: good; winter hardiness: high; drought resistance: average; disease resistance: good resistance to powdery mildew, bacterial burn.	Introduced and regionalised in Kyrgyzstan in 2015-2017, registered in 2019

Table 1. Continued

No.	Variety name	Origin	Brief description	Included in the state register
3	'Bellefleur'	American selection	American apple tree variety. Ripening period: autumn; apple taste: sweet and sour with a spicy note; average weight: medium-large (200 g); fruit colour: light yellow; flesh: white, juicy, fine-grained; transportability: excellent; resistance to low temperatures: weak; disease resistance: low, especially to scab.	Introduced and regionalised in Kyrgyzstan in 1938-1940
4	'Golden'	American selection ('Grimes Golden' x 'Golden Reinnet')	American apple tree variety. Ripening period: winter; taste: dessert, with a rich aroma; average weight: large (140-160 g); fruit colour: light green at the beginning of ripening, then gradually acquires a golden-yellow colour, with a pink blush appearing on the sunny side; flesh: white with a slight greenish tint, dense; transportability: excellent, long-term preservation of marketable appearance; drought resistance: poor; disease resistance: low resistance to brown spot and powdery mildew.	Introduced and regionalised in Kyrgyzstan in 1980-1982
5	'Bishkek'	Kyrgyz selection	Apple tree of Kyrgyz selection. Ripening period: winter; taste: sweet and sour with a strong aroma; average weight: large, uniform (140-170 g); fruit colour: light green, creamy white with a bright crimson blush; flesh: juicy, greenish-white; transportability: excellent, can be stored for a long time in a cold room; disease resistance: increased resistance to scab, powdery mildew, apple fruit moth, highly resistant to bacterial burn.	Local variety, bred in Kyrgyzstan in 1990, registered in 2018
6	'Niedzwetzky'	Wild	A rare species of apple tree (red, Chinese), endemic, is a valuable material for breeding work to develop new varieties. I. Michurin used this species to obtain the varieties 'Red Bellefleur', 'Red Standard', etc. It grows in Kyrgyzstan (in the Chatkal Valley, in the Sary-Chelek Nature Reserve, in the Kara-Suu and Aflatun tracts, in the foothill forests of Kara-Alma and Ak-Terek-Kaba). It is found in the mountain ecosystems of Kazakhstan and Uzbekistan. It also grows in western China (Xinjiang Western District). Fruit: spherical, purple-red; average weight: small, below average; leaves: rounded-elongated, rich green-red colour.	Endemic
7	'Idared'	American selection ('Wagner' x 'Jonathan')	American apple tree variety. Ripening period: winter or late winter. On medium-sized rootstocks, trees begin bearing fruit in 5-6 years. Regular fruiting. The advantages of this variety are high ecological adaptability, yield, and high marketability of fruit. Susceptible to scab and powdery mildew.	In Kyrgyzstan, it was entered in the register in 2006
8	'Ligolina'	Polish selection	Polish apple tree variety. Late winter variety, high-yielding, resistant to scab and powdery mildew. Medium-sized trees. Fruits are above average in size, cylindrical. The main colour is yellow-green, with a blurred red blush covering almost the entire surface of the fruit. The quality of the fruit does not change during storage.	Not registered in the Kyrgyzstan registry
9	'Early Geneva'	American selection ('Quinty' x 'Julyred')	American apple tree variety. An early-ripening apple tree variety that differs from other varieties in its regular harvest, high yield and excellent taste. The variety is very early-bearing (1 year after planting). Resistant to scab and powdery mildew.	Registered in the Kyrgyzstan registry in 2019
10	'Gala'	New Zealand selection ('Golden Delicious' x 'Kidd's Orange Red')	A late autumn apple variety bred in 1962 in New Zealand by breeder J. Kidd as a result of crossing two varieties – 'Golden Delicious' x 'Kidd's Orange Red'. The new variety quickly gained worldwide popularity (2 nd place in the world in terms of apple yield), and almost immediately after its development, orchards were planted in the USA, Canada, Brazil and Europe.	It was registered in the Kyrgyzstan registry in 2019
11	'Fuji'	Japanese selection ('Red Delicious' x 'Virginian Rale Janet')	The variety was obtained by crossing two American apple varieties – 'Red Delicious' and 'Virginian Rale Janet'. It was first obtained at the Japanese research station in Tohoku (Fujisaki village, Minamitsugaru district, Aomori prefecture) in the late 1930s and introduced to the market in 1962. Fuji apples are usually round and range in size from large to very large, with an average diameter of 75 mm. They contain 9 to 11% sugar by weight and have firm, crisp flesh that tastes sweeter than other varieties.	Registered in the Kyrgyzstan registry in 2025

Source: compiled by the authors

The full cycle of the experiment was conducted from early spring to the end of June 2025, which ensured an active phase of shoot and leaf growth for inoculation. During the adaptation phase, the plants were kept without the use of fertilisers; only standard soil substrate was used. Before infection, the plants were examined, and all specimens with signs of other diseases were excluded from the experiment. Ten seedlings were used from each variety. All experiments with plants were conducted in strict accordance with the principles of CBD (1992) and CITES (1973).

A pure culture of *Erwinia amylovora* bacteria was used for artificial infection. Artificial inoculation was carried out in late May – early June, after the appearance of stable shoot growth and full physiological development of the leaves. At the preliminary stage of the study, pure cultures of the phytopathogenic bacterium *Erwinia amylovora* were isolated from various infected branches and leaves of apple and pear trees. A total of 18 strains were obtained, which were subsequently tested for pathogenicity. The White test was used to assess pathogenicity: immature apple fruits were inoculated with a suspension of cells in physiological solution at a concentration of 10^9 cells/ml ($OP_{620} = 1.0$). Ten microlitres of the suspension were injected into the fruit tissue using a sterile syringe, after which the fruit was incubated in a humid chamber in Petri dishes at a temperature of 25°C for 5 days. The appearance of tissue necrosis and milky-white bacterial exudate in the inoculation area was considered a positive test result. Based on the test results, the *E. amylovora* culture with the laboratory designation ZH-2, isolated from infected branches of the 'Talgarka' pear variety collected in the Zheti-Oguz district of the Issyk-Kul region, was selected for further experiments. This strain was used for subsequent artificial infection of apple seedlings (Fig. 1).

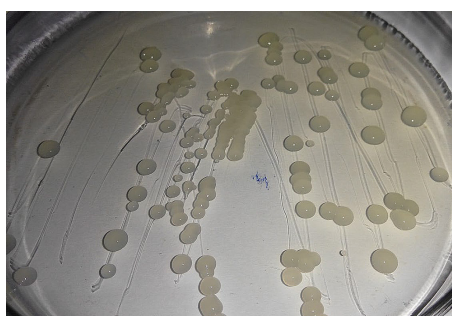


Figure 1. Laboratory strain ZH-2 – the causative agent of bacterial blight *Erwinia amylovora*

Source: authors' photo

To obtain biomass, the culture was grown at 27°C for 24-36 hours on a solid Levan nutrient medium with the following composition: yeast extract – 2 g, peptone – 5 g, NaCl – 5 g, sucrose – 50 g, agar – 20 g. To determine the number of live *Erwinia amylovora* cells,

the colony-forming unit (CFU) counting method was used on a nutrient medium. From a 24-hour liquid culture, a series of tenfold serial dilutions (from 10^{-1} to 10^{-8}) in sterile phosphate-buffered saline (PBS). From each dilution, 0.1 ml of the suspension was seeded onto the surface of Levan nutrient medium, evenly distributed with a sterile glass spatula, and incubated at 27°C for 48 hours. After incubation, the number of colonies grown on plates containing 30-300 colonies was counted, and the number of CFU in 1 ml of the initial suspension was calculated using formula (1):

$$N = \frac{a \times 10^n}{V}, \quad (1)$$

where N is the number of CFU/ml; a is the average number of colonies per dish; 10^n is the dilution factor; V is the inoculum volume, ml.

The concentration of the suspension for inoculation was set at 10^9 CFU/ml. After artificial infection of apple seedlings and the appearance of characteristic symptoms of bacterial blight, leaf samples of equal weight were taken from the affected plants. The samples obtained were homogenised and sown on Levan medium for the purpose of re-isolating *E. amylovora* bacteria and determining the level of bacterial biomass on infected plants. The intensity of pathogen reproduction was assessed based on the number of colonies that grew, which made it possible to determine the degree of bacterial load on the seedlings after infection. To confirm the presence of the pathogen in infected tissues, rapid diagnostics were additionally performed using AgriStrip immunoassays (Bioreba, Switzerland), which allow the detection of specific *Erwinia amylovora* antigens. The positive reactions obtained served as preliminary confirmation of infection before bacteriological isolation on Levan's medium.

After inoculation, the plants were kept for 30 days under a protective canopy providing diffused natural light and protection from direct precipitation. The average air temperature during the observation period was 22-25°C, with relative humidity of 60-70%. To assess the resistance of varieties to *Erwinia amylovora*, a method of artificial infection of plants with a bacterial culture suspension at a concentration of 10^9 cells/ml was used. Artificial infection was carried out *in vitro* on young actively growing shoots. A sterile scalpel was used to create the wound. A small transverse incision 3-5 mm long was made on each shoot, located 5 mm below the shoot apex. Small punctures were made on the leaves near the central vein using a sterile injection needle (diameter 0.6 mm). Ten to twenty microlitres of *E. amylovora* bacterial suspension was applied to each wound surface using a micropipette. The suspension was evenly distributed over the wound area, ensuring that it completely wetted the damaged area. Infection was carried out on the shoots and leaves of the lower, middle and upper tiers of each plant. Control plants were

treated in a similar manner, but instead of the bacterial suspension, a sterile buffer solution was applied. The development of the infection was recorded visually 1, 6, 12 and 30 days after inoculation. The degree of damage was determined on a five-point scale: 0 – no symptoms, 1 – damage to up to 25% of the shoot, 2 – damage to 26-50%, 3 – damage to 51-75%, 4 – damage to more than 75% of the shoot or its death. The results obtained were statistically processed using analysis of variance (ANOVA) methods. The reliability of differences between varieties was assessed using Duncan/Tukey's test at a significance level of $p < 0.05$. All calculations were performed using IBM SPSS Statistics software, version 26.0.

Results and Discussion

Bacterial blight of fruit crops, caused by the bacterium *Erwinia amylovora*, is one of the most dangerous diseases affecting apple trees and other members of the Rosaceae family. The disease leads to significant economic losses, reduced yields, deterioration in fruit quality and, in particularly severe cases, the death of trees. The first signs of bacterial blight are darkening of inflorescences and young leaves, which turn brownish-black, as if burnt. According to A. Pedroncelli & G. Puopolo (2024), as the disease progresses, shoots wilt, gummosis occurs, and branches and trunks are affected, often resulting in the death of the tree. The disease spreads mainly through pollinating insects, raindrops, wind, and agricultural practices (EFSA PLH, 2014). Bacterial blight was first recorded in North America, but over time the disease spread to Europe, Asia, and Central Asian countries, including Kyrgyzstan (Wang *et al.*, 2025). In the Kyrgyz Republic, the development of the disease is facilitated by hot and humid weather during the apple blossoming period, as well as insufficient phytosanitary control. Agrotechnical, chemical and biological methods are used to combat bacterial blight. As noted by R. Mendes *et al.* (2024), among agrotechnical measures, sanitary pruning of affected branches, removal of plant debris, disinfection of tools and ensuring optimal water regime play an important role. Chemical protection involves the use of copper-based preparations, antibiotics (streptomycin), and bacteriophages, but their effectiveness varies depending on climatic conditions and the stage of infection development.

The most promising approach to combating fire blight is to cultivate disease-resistant varieties. Varieties with high resistance to *Erwinia amylovora* have been identified worldwide, including Enterprise, Florina, Liberty, and others. However, as noted by B. García-Fernández *et al.* (2023), the adaptation of these varieties to the conditions of Central Asia, including Kyrgyzstan, requires further research. Modern genotyping methods allow the identification of genes for resistance to fire blight, such as the FB_MR5 gene, which is associated with resistance to *Erwinia amylovora*. Genomic technologies and molecular selection

contribute to the creation of new apple varieties with improved resistance characteristics. Studying the resistance of modern apple varieties to *Erwinia amylovora* bacterial blight is a pressing task, especially in the Kyrgyz Republic, where horticulture plays a significant role in the agricultural economy. Research in this area will enable the introduction of varieties adapted to local conditions, which will ensure stable production and improve fruit quality. In Kyrgyzstan, fire blight appeared around 2008-2009. According to T. Doolotkeldieva *et al.* (2021), the first signs were noticed in the Chüy region, then spread to the Issyk-Kul region and in subsequent years covered the entire republic, even reaching wild forest areas. Figure 2 shows a tree infected with bacterial blight.



Figure 2. Bacterial blight (*Erwinia amylovora*) infection of a 'Talgarka' pear tree

Note: sample collected during an expedition to the Issyk-Kul region of the Kyrgyz Republic

Source: authors' photo

In 2011, bacterial blight was registered as a quarantine disease by the Ministry of Water Resources, Agriculture and Processing Industry of the Kyrgyz Republic. Employees of the Department of Plant Protection and Quarantine surveyed more than 5,000 hectares of fruit plantations, and it is now known that 37 genera and 129 species of the Rosaceae family are affected: these include pears, quince, apples, peaches, apricots, raspberries, roses, persimmons, walnuts, etc. (Ministry of Water Resources..., n.d.). Figure 3 shows the results of a laboratory experiment to assess the damage to apple fruit caused by bacterial canker, the causative agent of which is the bacterium *Erwinia amylovora*. The bacterium, penetrating through mechanical damage or natural stomata, causes tissue necrosis and exudate secretion, which is typical of bacterial canker. The presence of exudate indicates the active phase of the disease and a high level of infection. Fruits with less pronounced symptoms are strains with low pathogenicity for infecting plants with bacterial canker.

As shown in Figure 3, not all strains are capable of causing disease. Petri dishes show that some of the fruits contain pronounced signs of bacterial damage: darkening of tissues, necrotic spots, exudate secretion (whitish or yellowish drops of bacterial fluid), which is

characteristic of *Erwinia amylovora* infection. In the dishes, however, some fruits show only superficial necrotic spots without bacterial exudate, which may indicate a weaker reaction of the strain. Thus, this experiment allows for visual differentiation of the pathogenic strain, which was subsequently selected for further experiments to determine the experimental variants for the resistance and susceptibility of seedlings of various

apple varieties to the causative agent of bacterial blight. To visually represent the differences in the response of varieties to artificial inoculation, photographic monitoring of the manifestation of infection symptoms was carried out throughout the experiment. Figure 4 shows two-year-old seedlings of all studied apple varieties after treatment with *Erwinia amylovora* suspension, demonstrating characteristic manifestations of bacterial blight.

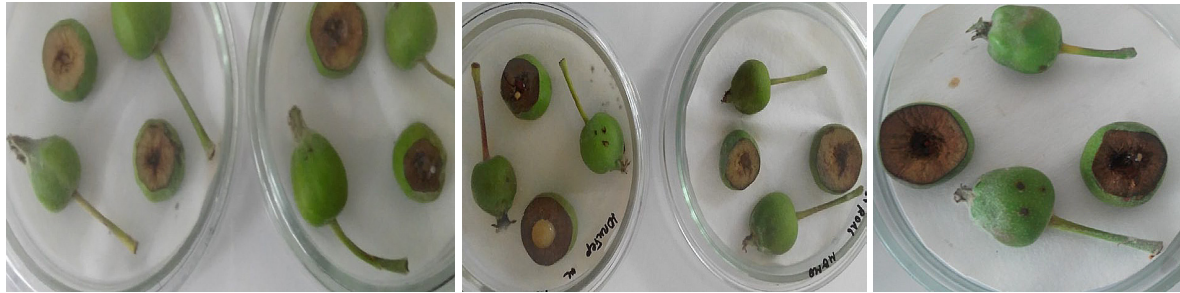


Figure 3. Manifestation of *Erwinia amylovora* pathogenicity on unripe apple fruits

Source: authors' photo



Figure 4. Artificial infection of two-year-old apple seedlings with *Erwinia amylovora*, the causative agent of fire blight

Note: a – ‘Starkrimson’ variety, b – ‘Red Chief’, c – ‘Bellefleur’, d – ‘Bishkek’, e – ‘Golden’, f – ‘Idared’, g – ‘Niedzwetzky’, h – ‘Ligolina’, i – ‘Early Geneva’, j – ‘Gala’, l – ‘Fuji’

Source: authors' photos

Figure 4 shows the external manifestations of infection in different varieties 30 days after inoculation. It can be seen that susceptible varieties ('Idared', 'Gala', 'Ligolina') show pronounced blackening of young shoots, drying of the tops and the appearance of necrotic areas on the leaves. Moderately resistant varieties ('Starkrimson', 'Fuji', 'Early Geneva') show localised necrosis on leaf blades and partial wilting of shoots.

The most resistant varieties ('Red Chief', 'Bishkek', 'Golden'), as well as the species 'Niedzwetzky', retain their green leaf colour and minimal damage, limited to isolated necrotic spots at the inoculation sites. Figure 4 reflects the visual differences in the degree of damage to the studied genotypes and confirms the results of the quantitative assessment. Figure 5 shows the average degree of damage to apple seedlings.

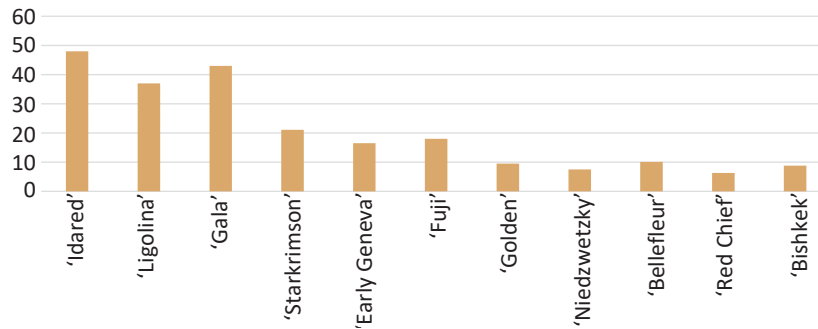


Figure 5. Average degree of damage to apple seedlings by bacterial blight (%)

Source: compiled by the authors

As shown in Figure 5, the following results were obtained after artificial infection over a period of one month. The 'Idared' variety showed 48% damage to seedlings from bacterial blight – a variety with a high degree of damage. The variety itself is winter-hardy but susceptible to disease. The 'Gala' variety had an average infection rate of 43% – it is a popular variety with good taste qualities, but has moderate resistance to various diseases. The 'Ligolina' variety (37% infection rate) is relatively vulnerable to infection compared to other varieties. The 'Starkrimson' variety showed 21.1% damage from bacterial blight over the course of a month – a beautiful red variety that is moderately susceptible to disease. The 'Fuji' variety (16.5%) is moderately resistant. The 'Early Geneva' variety (18%) is an early variety that is resistant to certain diseases. The 'Bellefleur' variety (10.1%) is an old variety with good shelf life and low susceptibility to disease. The 'Golden' variety (9.5%) is a famous yellow variety with high shelf life and resistance. The 'Bishkek' variety (8.8%) is a local variety with good adaptability and low susceptibility to disease. The

species 'Niedzwetzky' (7.5%) is a decorative apple tree variety that is resistant to disease. The 'Red Chief' variety (6.3%) is one of the most moderately resistant varieties and has high marketability. In addition, it has been noted that the climatic conditions of Kyrgyzstan contribute to the active spread of the disease. High temperatures and humidity during the growing season create optimal conditions for infection, which requires a more detailed study of the influence of climatic factors on the development of the disease. The identified resistant varieties, such as 'Red Chief', 'Bishkek' and 'Golden', are of particular interest for further introduction into production. Their use can significantly reduce crop losses and reduce the need for chemical protection. Diagnostic and bacteriological studies were conducted to confirm the presence of the bacterial blight pathogen in infected tissues and to quantitatively assess the bacterial load. Figures 6 and 7 show the results of rapid diagnosis using an immunostrip test and the isolation of *Erwinia amylovora* colony-forming units on a selective nutrient medium.

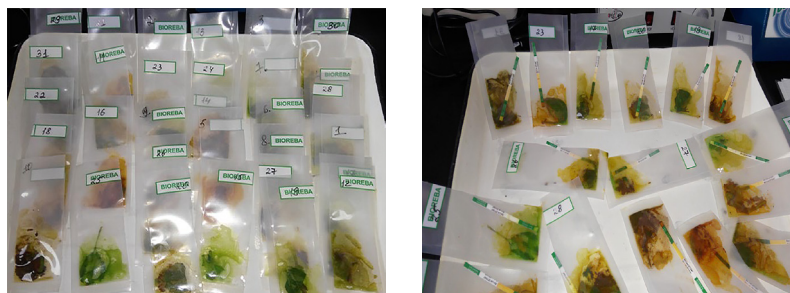


Figure 6. Immunoassay strips for the detection of *Erwinia amylovora* bacteria

Source: authors' photo

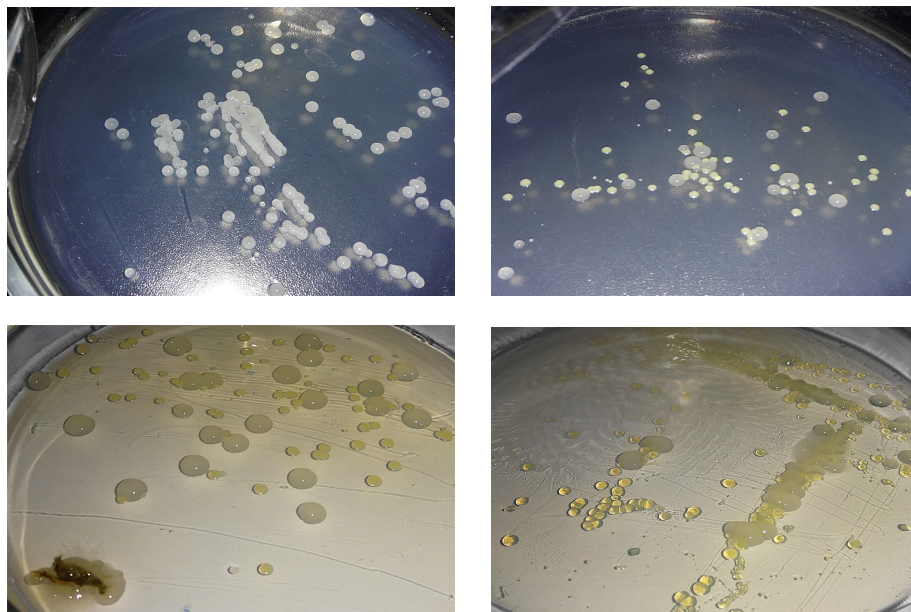


Figure 7. Colony-forming unit of *Erwinia amylovora* bacteria on Levan's nutrient medium

Source: authors' photo

Figures 6 and 7 demonstrate laboratory confirmation of apple tree seedlings infected with *Erwinia amylovora*, the causative agent of fire blight. Figure 6 shows a positive result of an immunoassay strip test, which allows rapid detection of specific pathogen antigens in the tissue samples under investigation. Figure 7 shows *E. amylovora* colonies grown on Levan's nutrient medium; the growth intensity and number of colonies formed reflect differences in bacterial load between samples of different varieties. The combination of visual diagnosis and bacteriological isolation confirms both the fact of infection and the varying degrees of pathogen reproduction in the tissues of the

varieties, which is consistent with the data obtained on the level of damage.

The results of bacteriological analysis showed the presence of *Erwinia amylovora* bacteria in all analysed samples. However, the number of CFU varied significantly depending on the degree of tissue damage. The highest number of CFU was observed in severely affected samples, while in tissues with visually less pronounced symptoms, the number of bacteria was significantly lower (Table 2). The data obtained indicate different susceptibility of seedlings to infection, which may be due to their individual physiological characteristics or resistance to the pathogen.

Table 2. Colony-forming unit of *Erwinia amylovora* bacteria

No.	Name of varieties	<i>Erwinia amylovora</i>	CFU/ml
1	'Idared'	+	35.8 * 10 ⁶
2	'Early Geneva'	+	19.3 * 10 ⁶
3	'Ligolina'	+	23.4 * 10 ⁶
4	'Starkrimson'	+	25.7 * 10 ⁶
5	'Red Chief'	+	21.3 * 10 ⁶
6	'Bellefleur'	+	9.3 * 10 ⁶
7	'Golden'	+	8.7 * 10 ⁶
8	'Bishkek'	+	12.2 * 10 ⁶
9	'Niedzwetzky'	+	26.1 * 10 ⁶
10	'Gala'	+	30.4 * 10 ⁶
11	'Fuji'	+	24.7 * 10 ⁶

Source: compiled by the authors

Table 2 presents data on the number of colony-forming units of *Erwinia amylovora* isolated from infected tissues of various apple varieties after artificial inoculation. It can be seen that the highest CFU values are observed in highly susceptible varieties ('Idared',

'Gala', 'Ligolina'), which corresponds to their high degree of infection, while in resistant varieties ('Golden', 'Bishkek', 'Red Chief') and in the 'Bellefleur' variety, the number of pathogen cells isolated is significantly lower. The data presented confirm the differences in

bacterial load between varieties and are consistent with visual assessments of resistance. Figure 8 presents summarised data from the quantitative analysis

of bacterial load, showing the comparative distribution of *Erwinia amylovora* colony-forming units among the apple varieties studied.

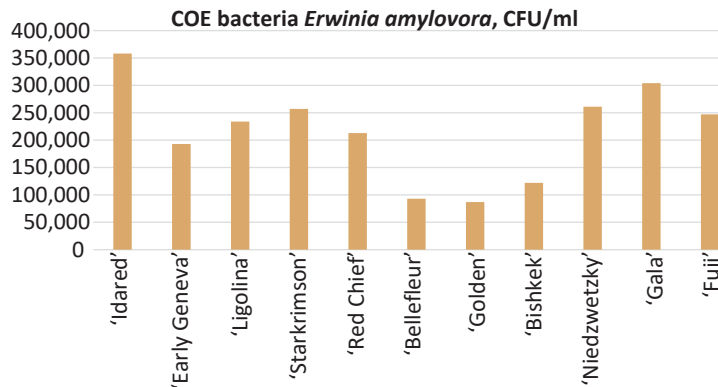


Figure 8. CFU of *Erwinia amylovora* bacteria

Source: compiled by the authors

The highest number of *Erwinia amylovora* bacteria cells was isolated from the affected tissue of the 'Idared' variety – a total of 358,000 cells/ml. The lowest number of cells was isolated from the affected tissues of the 'Golden' variety – 80,000 cells/ml. An interesting feature is the high level of CFU in the

species 'Niedzwetzky', despite the low visual degree of damage, which may indicate the role of the variety as a possible carrier or reservoir of bacteria in the absence of pronounced symptoms. Table 3 shows the results of multiple comparisons of the degree of damage to apple fruit.

Table 3. Results of multiple comparisons of the degree of damage to apple fruit by bacterial blight (Duncan/Tukey test, $p < 0.05$)

Variety	Average degree of damage (%)	Duncan group
'Idared'	48	a
'Ligolina'	37	ab
'Gala'	43	ab
'Starkrimson'	21.1	bc
'Early Geneva'	16.5	bc
'Fuji'	18	bc
'Golden'	9.5	c
'Niedzwetzky'	7.5	c
'Bellefleur'	10.1	c
'Red Chief'	6.3	c
'Bishkek'	8.8	c

Note: mean values marked with the same letters are not statistically different from each other according to Duncan/Tukey test ($p > 0.05$)

Source: compiled by the authors

The results of the study demonstrate significant differences in the resistance of modern apple varieties to bacterial blight caused by *Erwinia amylovora*. The 'Idared' variety proved to be the most susceptible to the disease, as confirmed by the high degree of infection (48%). This may be due to its genetic predisposition to infections, as well as insufficient resistance to pathogens, which is consistent with data on its susceptibility to other diseases, such as scab and powdery mildew. The 'Ligolina' and 'Gala' varieties also showed a relatively high degree of damage, indicating the need for further research to identify the factors contributing to

their vulnerability. In contrast, the 'Red Chief', 'Golden', 'Bishkek' varieties and the species 'Niedzwetzky' showed a low degree of infection, making them promising for cultivation in the Kyrgyz Republic. This is confirmed by the data from multiple comparisons presented in Table 2. The results of this study confirm significant differences in the resistance of modern apple varieties to bacterial canker caused by *Erwinia amylovora*. Similar data were obtained in the work of H. Aldwinckle & M. Malnoy (2009), which reports on the high resistance of some apple varieties to fire blight due to the presence of specific genetic markers. In particular, the

species 'Niedzwetzky' and the varieties 'Bishkek' and 'Golden' showed a low degree of damage, which is consistent with studies conducted in Central Asia and Europe. The high susceptibility of the 'Idared' and 'Gala' varieties confirms the findings of previous studies, which noted that these varieties have insufficient resistance to bacterial blight and require additional protective measures (Emeriewen *et al.*, 2021). This fact points to the need for breeding programmes aimed at improving the resistance of these varieties or finding alternative options adapted to local conditions. To optimise breeding programmes, it is critical to understand the molecular genetic mechanisms underlying resistance. At the molecular level, a study by S. Schröpfer *et al.* (2021) revealed the activation of genes involved in the formation of flavonoids and terpenoids, as well as a heat shock response in the susceptible response, indicating specific defence mechanisms in apple trees.

The use of artificial infection in this study made it possible to determine the degree of resistance of varieties under controlled experimental conditions. Studies have shown that commercial apple varieties have different levels of susceptibility to bacterial canker, and assessing the resistance of varieties and rootstocks is one of the key areas of research, which is also consistent with the conclusions of S. Kostick *et al.* (2019). In addition, this study confirmed that genotypes of the wild apple tree *Malus sieversii* can be a source of resistance, as previously reported by J. Harshman *et al.* (2017). Wild *Malus* species are of particular interest in breeding programmes, as they are sources of complex resistance not only to bacterial canker. A comprehensive analysis of *Malus* species for resistance to scab (*V. inaequalis*) and ornamental value showed that wild and hybrid species, in particular the berry apple, outperform the domestic apple, confirming their value as sources of resistance for breeding programmes (Moskalets *et al.*, 2024). The results obtained in the current study emphasise the importance of breeding resistant varieties as the main direction in the fight against bacterial blight, since chemical control methods, including the use of copper and antibiotic-containing preparations, have limited effectiveness and can lead to the formation of resistant strains of bacteria. Thus, this study confirms the importance of introducing resistant apple varieties into horticultural practice in Kyrgyzstan and other regions with similar phytopathological problems.

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Conclusions

The study conducted a comprehensive assessment of the resistance of modern apple varieties (*Malus domestica*) to the bacterial blight pathogen (*Erwinia amylovora*) in the Kyrgyz Republic. Through artificial infection with the phytopathogenic culture ZH-2, significant differentiation of varieties in terms of their susceptibility was established. The differences identified are critical for optimising plant protection strategies in the region, especially given the status of fire blight as a quarantine disease. The most resistant varieties were 'Red Chief', 'Bishkek' and 'Golden', with a degree of damage not exceeding 9.5% (while the susceptible variety 'Idared' suffered damage of up to 48%). These varieties, as well as the wild species 'Niedzwetzky', which showed a low percentage of damage, are the most promising for introduction into commercial and amateur horticulture in Kyrgyzstan. They can serve as a basis for creating sustainable orchards and reducing dependence on chemical control methods, including antibiotics, to which the pathogen is developing resistance. In contrast, the varieties 'Idared', 'Gala' and 'Ligolina' showed high or moderate susceptibility, requiring additional protective measures, including the use of more resistant rootstocks, when growing them.

Further research should focus on a more in-depth study of the molecular and physiological mechanisms of resistance in highly resistant varieties (e.g., 'Bishkek' and 'Red Chief'). This includes molecular genetic analysis to identify QTL loci and genes responsible for resistance to *E. amylovora*, which will accelerate the breeding process. It is also relevant to screen a larger sample of local wild *Malus* (in particular, *Malus sieversii*) as potential sources of new resistance genes for cross-breeding. Finally, it is advisable to conduct field trials of selected resistant varieties in different soil and climatic zones of Kyrgyzstan to confirm their ecological adaptability and commercial value in the long term.

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Conflict of Interest

None.

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Заманбап алма сортторунун *Erwinia amylovora* бактериялдык күйүк козгогучуна туруктуулугун баалоо

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Аннотация. Алманын жаңы сортторуна коюлган заманбап талаптарга жогорку түшүмдүүлүк менен мөмөнүн сапатын жакшыртуу гана эмес, ошондой эле эң коркунучтуу ооруларга, анын ичинде *Erwinia amylovora* бактериясы козгогон бактериялдык күйүккө туруктуулук да кирет. Бул изилдөөнүн максаты Кыргызстанда кеңири таралган заманбап алма сортторунун бактериялдык күйүктүн козгогучуна туруктуулугун жасалма жугузуу шартында баалоо болду. Эксперимент үчүн ‘Талгарка’ алмурут сортуна таандык жабыркаган бутактардан бөлүнүп алынган фитопатогендик *E. amylovora* бактериясынын таза (ZH-2) штаммы колдонулду. Жугузуу *in vitro* ыкмасы менен жүргүзүлүп, эки жаштагы көчөттөрдүн бүчүр жана жалбырак беттерине 10^9 КОЕ/мл концентрациядагы бактерия суспензиясы жаракатталган жерлерге сүртүлдү. Жугузгандан кийин 1, 6, 12 жана 30-күндөрү жабыркоонун деңгээли кыймылсыз (визуалдык) түрдө беш баллдык шкала боюнча, жабыркаган ткандардын пайызын эске алуу менен аныкталды. Изилдөөнүн жыйынтыктары каралган он бир алма сорту арасында туруктуулуктун олуттуу айырмачылыктарын көрсөттү. Эң туруктуу сорттор катары ‘Ред Чиф’, ‘Бишкек’ жана ‘Голден’ белгиленип, алардын жабыркоо деңгээли 6,3-9,5 %ди түзүп, өсүмдүктөрдүн коргонуу реакцияларынын күчтүү экенин жана некроз белгилеринин начар өнүккөнүн аныкталды. Орточо туруктуулук ‘Фуджи’ сортунда (16-18 %) байкалып, бүчүр ткандарынын бир аз жабыркашы менен мүнөздөлдү. Эң начар туруктуулукка ээ болгон сорттор ‘Айдаред’ (48 %), ‘Гала’ (43 %) жана ‘Лаголина’ (37 %) болуп, бул сорттордо некроздук тактардын тез өнүгүшү жана бүчүрлөрдүн солушу катталды. Алынган жыйынтыктар бактериялдык күйүккө туруктуу сортторду көбөйтүүдө жана селекциялык иштерде колдонууга мүмкүнчүлүк берип, ошондой эле Кыргызстан шартында заманбап алма сортторунун ооруларга болгон туруктуулук диапозону жөнүндө маалымат берет

Негизги сөздөр: жасалма жугузуу; жабыркоо даражасы; фитопатогендик бактериялар; ткандардын некрозу; *in vitro*

Оценка устойчивости современных сортов яблони к возбудителю бактериального ожога *Erwinia amylovora*

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Аннотация. Современные требования к новым сортам яблони включают не только высокую урожайность и улучшенное качество плодов, но и устойчивость к наиболее опасным заболеваниям, в том числе к бактериальному ожогу, вызываемому *Erwinia amylovora*. Целью данного исследования была оценка устойчивости современных сортов яблони, распространенных в Кыргызстане, к возбудителю бактериального ожога при искусственном заражении. Для эксперимента использовали чистую культуру фитопатогенной бактерии *E. amylovora* (штамм ZH-2), выделенную из пораженных ветвей груши сорта 'Talgarka'. Заражение проводили методом *in vitro*, нанося суспензию бактерий с концентрацией 10⁹ КОЕ/мл на раневые поверхности побегов и листьев двухлетних саженцев. Оценку степени поражения проводили визуально через 1, 6, 12 и 30 дней после инокуляции по пятибалльной шкале, учитывающей процент пораженной ткани. Результаты исследования показали значительные различия в устойчивости между изучаемыми одиннадцатью сортами яблони. Наиболее устойчивыми оказались сорта 'Red Chief', 'Bishkek' и 'Golden', у которых уровень поражения составил от 6,3 до 9,5 %, что свидетельствует о выраженных защитных реакциях растений и низкой степени развития симптомов некроза. Средний уровень восприимчивости проявил сорт 'Fuji' (около 16-18 %), характеризующийся умеренным поражением тканей побегов. Наиболее чувствительными к инфекции оказались сорта 'Idared' (48 %), 'Gala' (43 %) и 'Ligolina' (37 %), у которых наблюдалось быстрое развитие некротических пятен и увядание побегов. Полученные результаты позволяют выделить перспективные для дальнейшего размножения и селекционной работы сорта, устойчивые к бактериальному ожогу, а также дают представление о диапазоне восприимчивости современных сортов яблони в условиях Кыргызстана

Ключевые слова: искусственное заражение; степень поражения; фитопатогенные бактерии; некроз тканей; *in vitro*