EPPO Datasheet: Dickeya dianthicola

Last updated: 2023-01-20

IDENTITY

Preferred name: Dickeya dianthicola

Authority: Samson, Legendre, Christen, Fischer-Le Saux, Achouak

& Gardan

Taxonomic position: Bacteria: Proteobacteria:

Gammaproteobacteria: Enterobacterales: Pectobacteriaceae **Other scientific names:** Erwinia carotovora f. sp. dianthicola (Hellmers) Bakker, Erwinia chrysanthemi pv. dianthi Alivizatos, Erwinia chrysanthemi pv. dianthicola (Hellmers) Dickey, Erwinia parthenii var. dianthicola (Hellmers) Bakker & Scholten, Pectobacterium carotovorum var. dianthicola Hellmers, Pectobacterium parthenii var. dianthicola Hellmers

Common names: bacterial stunt of carnation, bacterial wilt of carnation, bacterial wilt of dahlia, blackleg of potato, slow wilt of

carnation

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EPPO Code: ERWICD



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Notes on taxonomy and nomenclature

In 2005, Pectobacterium chrysanthemi was reclassified into the new genus Dickeya based on 16S rRNA sequencing, DNA-DNA hybridisation and biochemical properties (Samson et al., 2005). Dickeya spp. are closely related to the genus Pectobacterium. Both genera comprise soft rotting pathogens that cause plant disease by producing large quantities of plant cell wall-degrading enzymes. The genus Dickeya includes several validly named species: In addition to D. dianthicola these are D. chrysanthemi, D. dadantii subsp. dadantii, D. dadantii, subsp. dieffenbachia, D. paradisiaca, D. zeae, Dickeya solani, Dickeya aquatica, Dickeya fangzhongdai, Dickeya lacustris, Dickeya undicola and D. oryzae (Samson et al., 2005; Brady et al., 2012; van der Wolf et al., 2014; Parkinson et al., 2014; Tian et al., 2016; Hugouvieux-Cotte-Pattat et al., 2019; Oulghazi et al., 2019; Wang et al., 2020). D. dianthicola contains strains formerly classified as Pectobacterium chrysanthemi pv. dianthicola. Phylogenetic analysis of 16S rRNA, recA and dnaX gene sequences show that D. dianthicola is most closely related to D. dadantii and exhibits little diversity between strains, with no obvious delineation between isolates from different host plants (Samson et al ., 2005; Parkinson et al., 2009; S?awiak et al., 2009).

HOSTS

The earliest reference isolates of what is now classified as Dickeya dianthicola were collected in the 1950s from Dianthus caryophyllus (carnation) in Denmark, the Netherlands and the United Kingdom (Hellmers, 1958) and from Chrysanthemum originating from the USA (Parkinson et al., 2009). Initial high economic losses in several European countries resulted in the adoption of quarantine measures and voluntary certification schemes by carnation producers. As a consequence, very few findings of D. dianthicola have been reported on carnation in recent years (EFSA, 2013). Occasional reports on other ornamental hosts include Begonia, Dahlia, Hyacinthus, Kalanchoe, members of the Crassulaceae (stonecrop family e.g. Hylotelephium) and most recently on Impatiens hawkeri (New Guinea Impatiens).

Due to apparent spread from ornamentals, the bacterium is now included among several Pectobacterium and Dickeya species that cause blackleg and soft rot diseases of potato. It was first reported on European potato in the 1970s (Maas Geesteranus, 1972) with more recent findings on potato in Australia (Wright et al., 2018), Pakistan (Sarfraz et al

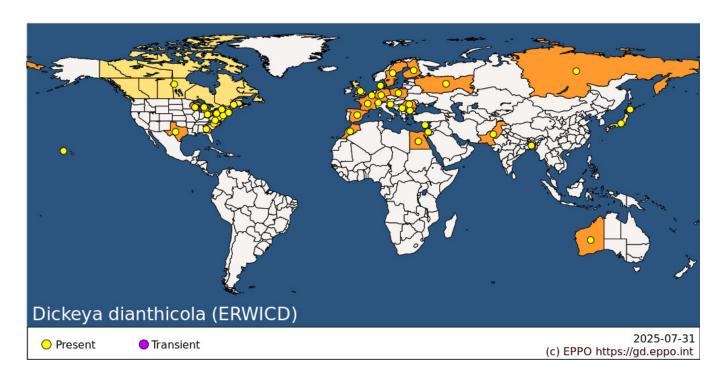
., 2020) and North-Eastern USA (Curland *et al.*, 2021). *D. dianthicola* has also been found to cause occasional disease in crops of *Cichorium intybus* (chicory) (Lan *et al.*, 2013), *Cynara scolymus* (artichoke) (Samson *et al.*, 2005), *Daucus carota* (carrot) (Farrar *et al.*, 2000), *Ipomoea batatas* (sweet potato) (Dickey, 1979; Curland *et al.*, 2021) and *Solanum lycopersicum* (tomato) (Samson *et al.*, 2005).

In France, *D. dianthicola* has been isolated from certain annual weeds (*Urtica dioica*, *Solanum nigrum*, *Mercurialis annua* and *Persicaria maculosa* (synonym *Polygonum persicaria*)) growing within a diseased potato crop, but only during the growing season and not after harvest of the crop (EFSA, 2013). The pathogen has also been isolated from *Solanum dulcamara* growing around waterways in France. The potential role of weed species in survival of the pathogen and disease epidemiology is not yet understood.

Host list: Begonia x intermedia, Chrysanthemum x morifolium, Cichorium intybus, Cynara scolymus, Dahlia sp., Daucus carota, Dianthus barbatus, Dianthus caryophyllus, Hyacinthus sp., Hylotelephium spectabile, Impatiens hawkeri, Ipomoea batatas, Kalanchoe blossfeldiana, Solanum lycopersicum, Solanum tuberosum

GEOGRAPHICAL DISTRIBUTION

The geographic distribution of *D. dianthicola* is likely to be wider than reported since many worldwide reports of *Erwinia chrysanthemi* prior to reclassification of the pathogen (Samson *et al.*, 2005) have not been followed up to determine the species of *Dickeya* involved.



EPPO Region: Belgium, Bulgaria, Cyprus, Denmark, Finland, France (mainland), Germany, Israel, Morocco, Poland, Romania, Russian Federation (the) (Central Russia, Eastern Siberia), Serbia, Slovenia, Spain (mainland), Sweden, Switzerland, United Kingdom

Africa: Egypt, Morocco

Asia: Bangladesh, Israel, Japan (Hokkaido), Pakistan

North America: Canada, United States of America (Delaware, Georgia, Hawaii, Maine, Maryland, Massachusetts, Michigan, New Jersey, New York, North Carolina, Ohio, Pennsylvania, Rhode Island, Texas, Virginia, Wisconsin)

Oceania: Australia (Western Australia)

BIOLOGY

D. dianthicola causes soft rot of fleshy plant roots, tubers, stems and leaves. Mainly biotrophic, it multiplies and survives long periods only in association with a host plant. D. dianthicola seeded to field soils persisted only for a

maximum of 3 weeks, irrespective of soil type, temperature or humidity (van der Wolf *et al.*, 2007). However, a maximum survival period of 12 months has been previously reported for *Dickeya* in potting media in glasshouses (Haygood *et al.*, 1982), whilst Garibaldi (1972) showed that isolates from *Dianthus* could not survive in plant-free soil for more than 6 months. *D. dianthicola* isolated from river water in Finland (Laurila *et al.*, 2010) and England (Elphinstone *et al.*, 2014), and from pond water in Pennsylvania and Maine (Curland *et al.*, 2021) was able to cause disease on potato.

The bacteria enter plants through wounds or natural openings (e.g. lenticels, stomata, or growth cracks formed during lateral root development). The pathogen can then colonise the entire plant systemically, where it can survive long periods of time even in the absence of initial disease development (so-called latent infection). In this form it can overwinter in a host plant or survive low temperature storage of vegetative propagules (e.g. plant cuttings, tubers etc.). Symptoms develop when temperature and high humidity allow systemic bacterial populations to multiply and exceed a threshold (around 10^4 cfu per g plant tissue) where production of cell wall degrading enzymes is triggered through quorum sensing (Whitehead *et al.*, 2002). The optimum *in vitro* growth temperature of *D. dianthicola* is around 27 °C, although it can still multiple below 10 °C and up to 35 °C. Waterlogging, resulting in oxygen depletion, increases plant stress, reduces host resistance, and encourages multiplication of the facultatively anaerobic bacteria.

DETECTION AND IDENTIFICATION

Symptoms

In general, symptoms caused by *D. dianthicola* resemble those caused by other non-host specific pectolytic *Dickeya* and *Pectobacterium* species and may vary with prevailing climatic conditions. It is therefore not possible to identify the species of bacterium from symptoms alone. General symptoms include soft rotting of stems, roots, storage organs and foliage; wilting, stunting and discolouration of foliage; vascular discolouration and bacterial slime oozing from cut vessels; collapse or desiccation and premature death of entire plants. Latent infections are also common when conditions favour slow bacterial multiplication and host colonisation without inducing symptoms.

On carnation: Slow wilt development due to D. dianthicola is more prolonged compared with rapid wilting caused by Burkholderia caryophylli. Severe stem infections with D. dianthicola show a brownish-yellow discoloration, excessive slime formation, and rotting at the stem base, together with general wilting of the plant (Helmers, 1958). Minor cracks may appear on the stem surface and roots may fail to form or, in older plants, roots completely rot away releasing the bacteria into the soil. The disease tends to spread more readily in older plants. In less severe cases, roots and shoots may continue to form but young shoots become short and thick and leaves narrow (hence the disease is also known as bacterial stunt). Roots of younger infected plants remain largely healthy but brown discoloration can be found in the xylem vessels when the stem is cut. Mother plants often rot and die before cuttings can be taken. However, in less severe cases (symptomless plants or those showing some stunting), the bacteria may reach the upper parts of the plant and be transferred via cuttings.

On potato: Rotting of the mother tuber in the field either leads to stem soft rot or to necrosis and hollowing of the stem. Under warm dry conditions, disease symptoms begin with wilting of the top leaves with desiccation at the margins, progressing to the entire leaf. Symptoms then become visible in the lower leaves, accompanied by wilting, eventually resulting in desiccation of the whole plant (Lumb et al., 1986). In warm but wetter conditions, symptoms include black/brown soft rotting from the base of the stem together with chlorosis and wilting of the foliage more resembling blackleg symptoms caused by Pectobacterium and other Dickeya spp. (Toth et al., 2011, van der Wolf and De Boer, 2007). Soft rot of potato tubers, in the field or during storage, initiates when a film of water induces anaerobic conditions within respiring tissues, both impairing host resistance and encouraging multiplication of the facultative anaerobic bacteria. Hence, blackleg is often observed in poorly drained patches of the field. Condensation on stored potatoes favours soft rot development by stimulating multiplication of the bacteria latently present in vascular tissues, lenticels or wounds. The risk of soft rot is therefore increased when potato tubers are stored wet and poorly ventilated or exposed to condensation induced by fluctuating storage temperatures.

On other ornamental hosts: D. dianthicola causes Dahlia tubers to swell and burst due to production of gases during the rotting process, sometimes before emergence of the plant (van Leeuwen et al., 2012). On Kalanchoe, Dickeya spp. cause plants to become dull and limp, leading to secondary fungal infections and plant death. On stonecrops, the

entire plant becomes infected, water-soaked spots develop on leaves and soft rot spreads along the stem. Combined infection by *Phoma* and *Dickeya* can result in a purple-blue discoloration of the upper parts of the plant (van der Helm, 2009). Characteristic symptoms on hyacinth, *Impatiens* and *Begonia* have also been described (Jafra *et al.* 2009; Liu *et al.*, 2021; Saaltink and Kamerman, 1971).

On other crops: Dickeya spp. typically cause stem wilt of tomato, water-soaked areas on stems, hollowing of stems, pith necrosis and maceration of stems and fruits (Alivizatos, 1985; Wick and Shrier, 1990; Aysan et al., 2005). Core rot of carrot, caused by Dickeya sp., was reported as a soft rotting of the carrot root with pale orange, circular, soft rot lesions with multiple lesions per root and coalescence of some lesions to form large, irregular rotted areas. On sweet potato, Dickeya initially causes the leaves turn yellow, and a black, water-soaked rot occurs on the bottom of the stems that gradually extends to the top of the stems. Finally, the entire plant collapses and dies (Huang et al., 2010). On chicory, infected plants in fields were discoloured, had wilted foliage, and black necrosis of petioles near the crown. Wilted leaves subsequently collapsed and died, forming a dry, brown or black rosette. The root and crown became partially or wholly affected by soft-rot. Slimy masses on infected areas of roots, turn dark brown or black (Lan et al., 2013). Dickeya spp. also cause bacterial crown rot of globe artichoke, which is characterised by stunted plant growth, wilted leaves at high temperatures and plant collapse. New leaves do not expand and turn brown and dry, the crown tissue becomes soft and rots and black discoloration of vascular tissues can be seen on cross-sections of the stem (CABI, 2011).

Morphology

D. dianthicola, like other Dickeya spp., is a Gram-negative, non-sporing, straight rod with rounded ends and is usually found alone or in pairs but occasionally in chains. It varies in size from 1.0 to 3.0 x 0.5–1.0 ?m and is motile with peritrichous flagella. On nutrient agar D. dianthicola forms round, convex, creamy-translucent colonies. On PDA, young colonies are either circular, convex, smooth and entire, or sculptured with irregular margins, depending on the moisture content of the growth medium. After 4-5 days, both colony types resemble a fried egg, with a pinkish, round, raised centre and lobed periphery, which later becomes feathery or almost coralloid. They form characteristic pits on pectate-based media such as crystal violet pectate agar. Detailed morphology is described by Lelliott and Stead (1987).

Detection and inspection methods

Host plants, growing and stored, should be routinely inspected for disease symptoms of blackleg and soft rot, as performed during the recommended certification scheme for carnation (EPPO, 2002) and potato (EPPO, 2017a; under revision).

Several methods are used to detect and identify *Dickeya* species, and to differentiate *D. dianthicola*. Generally, non-selective nutrient media can be used for isolation from symptomatic plant material. However, since secondary infections and saprophytic bacteria are often present when rotting symptoms are more advanced, dilution plating is required. Semi- selective CVP (crystal violet pectate) medium (Cuppels and Kelman, 1974; Hyman *et al.*, 2001; Bdliya and Langerfeld, 2005; Hélias *et al.*, 2012) is commonly used to isolate *Pectobacterium* and *Dickeya* from symptomatic plants and tubers and from environmental samples. Molecular tests for the identification of *Dickeya dianthicola* are conventional PCR (Pritchard *et al.*, 2013) followed by restriction fragment length polymorphism (RFLP); real-time PCR (Pritchard *et al.*, 2013; van der Wolf *et al.*, 2014; Karim *et al.*, 2019; Dobhal *et al.*, 2020); multiplex real-time PCR (Dobhal *et al.*, 2020) and LAMP test (Ocenar *et al.*, 2019). Sequenced PCR amplicons from selected gene loci allow accurate differentiation of *Pectobacterium* and *Dickeya* strains to species level. Multilocus sequence typing (MLST) has also been applied to differentiate isolates of *Pectobacterium* and *Dickeya* species and it is recognised as a useful technique for this purpose. Genomic fingerprinting tests, proteomic analysis based on MALDI-TOF mass spectrometry and fatty acid analysis can be used for the identification of *Dickeya* species.

Protocols for sample preparation have not been standardised nor validated through inter-laboratory comparisons. Information on detection and identification methods used in a number of laboratories in the EPPO region are provided in a PM 7 Standard on *Pectobacterium* spp. and *Dickeya* spp. (EPPO, 2023).

PATHWAYS FOR MOVEMENT

The main pathway for entry and spread of *D. dianthicola* in a country is considered to be via vegetatively propagated host plants with asymptomatic latent infections that escape visual surveillance inspections (EFSA, 2013).

It seems that *D. dianthicola* was initially introduced into the EPPO region and spread through carnation production in the 1950s (Hellmers, 1955). The pathogen is known to remain viable in ornamental plants and cuttings during long distance transport under refrigeration. The bacteria may also have been locally spread through drainage water, on pruning knives or by workers moving from plant to plant and crop to crop. However, the fact that the pathogen is no longer causing problems in carnation production, despite many producers currently importing cuttings produced in warm climates outside of the region, suggest that modern certification and hygiene programmes are effectively managing the risk with this pathway (EFSA, 2013).

Genetic similarity between isolates from all hosts within the region suggests a common initial source, although genetic divergence in some isolates from kalanchoe and *Impatiens* also suggests that there may have been separate introductions of the bacterium in these hosts (Pédron *et al.*, 2022). Since import of seed potato tubers are widely prohibited, it is suggested that initial findings on European potato crops in the 1970s were due to spread from ornamentals, possibly because of irrigation with surface water. A direct link between *D. zeae* found in surface water and on potato crops has been shown in Australia (Cother *et al.*, 1992).

Once introduced, the bacteria can survive cold storage periods in latently infected seed tubers and multiply and spread within crops when the tubers are handled (e.g. during harvesting, grading, store loading/emptying and replanting), effectively disseminating the bacteria from rotting to healthy tubers as rotting tissue smears over their surface. It is highly likely that spread of *D. dianthicola* since the 1970s has occurred during trade of latently infected seed potato so that the pathogen is now quite widespread across many European potato growing countries and has also been discovered in countries outside of Europe to which European seed potatoes have been exported, e.g. Egypt, Israel, Morocco, Pakistan and Bangladesh. Similar pathways of spread through trade in latently infected seed potato also appear to apply in recent *D. dianthicola* outbreaks in North-Eastern and North-Central USA (Curland *et al.*, 2021) and Western Australia (Wright *et al.*, 2018). *D. dianthicola* has been known to occur historically on ornamentals in the USA (Dickey, 1979) and has also been found in pond water in recently affected areas (Curland *et al.*, 2021). In Australia, *D. dianthicola* was found on dormant *Dahlia* tubers in addition to potato.

Airborne dissemination within crops, from diseased plants and cull piles, can occur in windblown aerosols generated by rainsplash. Infections with *Pectobacterium* in field crops have been shown to occur via insects (Kloepper *et al.*, 1981), and a related *Dickeya* sp. has been isolated from insects (Costechareyre *et al.*, 2012). However, a recent study (Insinga *et al.*, 2021) showed that *D. dianthicola* was not vectored to potato by two of its common insect pests; the green peach aphid (*Myzus persicae*) or the Colorado beetle (*Leptinotarsa decemlineata*).

PEST SIGNIFICANCE

Economic impact

On carnation, hardly any damage has been recorded in recent decades (EFSA, 2013), compared with the 1950s when it was estimated that for example 26.5 % of carnation stocks in Denmark were affected by bacterial stunt disease (Hellmers, 1958).

Until 2000, *D. dianthicola* was the only *Dickeya* sp. recorded on potato in the EU and may have caused up to 25% of blackleg disease in warmer seasons, the remainder being due to *Pectobacterium* (EFSA, 2013). Losses due to *Dickeya* are probably most significant in countries with warmer climates. In recent years, potato losses have increased generally in many European countries due to the emergence of other aggressive blackleg and soft rot pathogens *D. solani* (van der Wolf *et al.*, 2014) and *P. parmentieri* (Khayi *et al.*, 2016) in addition to the longer established *Pectobacterium atrosepticum*, *P. carotovorum* and *D. dianthicola*. Losses in ware potato can be 100% when prolonged wet conditions prevent harvesting and tubers rot in the field or where inadequate temperature control or ventilation is used during storage. Downgrading or rejection of potato seed crops due to blackleg is common, affecting both price and the reputation of the grower. Blackleg occurring later in the season has a significant effect on yield, since there is less time for the remaining healthy plants to compensate for the yield loss and high inoculum levels can be carried into the storage period. Soft rots can lead to supermarket product recalls in

some years and the high cost of this is ultimately passed back up the supply chain to packers and growers. Recent outbreaks of *D. dianthicola* and *P. parmentieri* on potato crops in North-Eastern and North Central USA (Ge *et al.*, 2021) have caused millions of USD in losses for the potato industry there. It was estimated that *Dickeya dianthicola* and *P. parmentieri* were found in 38.1% and 53.3% of all potato samples collected, respectively, and that 20.6% of samples contained both. Field trials showed that *D. dianthicola* was more virulent than *P. parmentieri*, but coinoculation of the two species resulted in increased disease severity compared to single-species inoculation with either pathogen.

In 1998, soft rot caused by *D. dianthicola* resulted in an estimated loss of 1633 tonnes of carrots in California (Farrar *et al.*, 2000). The disease appeared to be related to unusually high temperatures and excessive irrigation. Outbreaks of *D. dianthicola* on other hosts are infrequently reported and losses have not been quantified. Furthermore, it is often difficult to differentiate losses due to *D. dianthicola* and those caused by related pectolytic bacteria on the same hosts.

Control

In the absence of any curative chemical control methods, the control of *D. dianthicola* (and related pectolytic bacteria) largely relies on the availability of pathogen-free planting material, the application of strict hygiene measures during growing, storage and transport and the avoidance of growing and storage conditions that favour bacterial multiplication.

In *Dianthus* and other glasshouse ornamental production, *D. dianthicola* has been readily controlled owing to the widespread implementation of effective voluntary certification schemes within the industry. In nurseries, growing cuttings or plants on elevated benches, hygienic measures, pathogen-free water and the use of certified production systems for cuttings and mother plants are sufficient to prevent infection and spread (EFSA, 2013). Measures for the production of certified pathogen-tested material of carnation are recommended in EPPO Standard PM 4/2 (EPPO, 2002).

In potato, disease management has mainly resulted from the use of limited generation seed multiplication from pathogen-free nuclear stocks coupled with low-temperature storage incorporating forced ventilation to facilitate drying and prevent condensation. Most countries rely on certification systems to monitor and maintain health of seed potato tuber stocks during their multiplication, as recommended in EPPO Standard PM 4/28 Certification scheme for seed potato (EPPO, 2017a; under revision). Visual inspections for disease symptoms in the field and store are used to grade seed stocks according to disease tolerance limits. Seed stocks with blackleg or soft rot levels exceeding tolerance limits are either downgraded or failed and removed from the seed system. Such schemes do not usually consider latent infections and do not specify the species of Dickeya or Pectobacterium causing blackleg or soft rot symptoms. Best agronomic practice includes planting of crops in well-drained soil, avoidance of excessive irrigation or flooding, control of weeds and feeding insects, harvesting in dry conditions, minimising damage during harvest and handling and cleaning with disinfection of machinery, graders, storage containers and stores. Most general disinfectants are effective against Dickeya spp. when exposed on clean surfaces but bacteria in systemic infections, healed wounds and suberized lenticels are usually protected from antibacterial activity. Chlorine dioxide is often added to wash water prior to packing of ware potatoes in well ventilated bags to prolong shelf-life during retail.

Phytosanitary risk

This pest is of particular concern in the EPPO region for potato, carnation, and to a lesser extent for other ornamentals.

Carnation is mainly cultivated under a protected cropping system with voluntary certification and strict sanitation processes that prevent infection from the surrounding environment or previous crops. EFSA (2013) conducted a pest risk assessment for *Dickeya dianthicola* on carnation for the EU in advance of revision of EU Plant Health Directive 2000/29/EC. This assessment concluded that, although the environmental conditions are more favourable to the pathogen in protected crops, removal of pre-existing specific quarantine measures would have minimal consequences for carnation since voluntary certification and sanitation practices in place are considered effective and no carnation crop losses had been reported in the previous 25 years.

The pest is not reported yet in some EPPO countries. If it turns out that it is really not present in these countries, it is likely to be able to establish: In the open field, suitable hosts are widespread, the environment is suitable and possibilities of removing inoculum from the environment are limited. It is able to cause disease at a wide range of temperatures and has been found in a range of different climates and locations (EFSA, 2013). The pest could also further spread in the field environment as multiple factors that are difficult to manage can be involved in spread of the pathogen.

Similar conclusions were made, from an express PRA conducted in Poland in 2018, that the probability of *D. dianthicola* spreading in potato crops was moderate but spread in decorative plants was less likely due to the controlled conditions for production of such crops (IOR, 2019).

PHYTOSANITARY MEASURES

EPPO countries where *D. dianthicola* does not occur or is not widespread may consider regulation. Visual inspections should be performed routinely upon export and import of known host plants for planting. Laboratory checks are necessary to detect asymptomatic (latent) infections.

EPPO Standard PM 8/1 recommends the phytosanitary measures which EPPO countries should use or require for seed and ware potatoes moving in international trade to prevent the introduction and spread of regulated pests (EPPO, 2017b). According to this Standard, only certified seed potato tubers produced according to EPPO Standard PM 4/28 (EPPO, 2017a; under revision) or its equivalent should be eligible for import. PM 3/21 Post entry quarantine for potato describes inspection and tests for the detection of pests (including pectolytic bacteria: Dickeya spp. and Pectobacterium spp.) infecting Solanum species or hybrids imported for germplasm conservation, breeding or research purposes, in post-entry quarantine.

Plants for planting of known host plants may be placed in post-entry quarantine to observe any symptoms and if relevant to test them to ensure their freedom from *D. dianthicola*.

Before starting the propagation and/or production of plants, it should be ensured that the planting material is free from *D. dianthicola* (the pest is included in certification schemes for potatoes and carnation, see Control section). During the EU Quality pest project, it was considered that in the EU, on carnation, it was not a problem thanks to voluntary certification schemes aimed at *Fusarium* prevention (Picard *et al.*, 2018). However, to implement a certification scheme was considered the only effective measure to prevent the presence of *Pectobacterium* and *Dickeya* over a certain threshold in seed potato.

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