Received: 15 November 2023 | Revised: 29 February 2024 | Accepted: 12 March 2024

DOI: 10.1111/aab.12908

Annals of Applied Biology
An international journal of the aab

WILEY

ORIGINAL ARTICLE

Changes in the prevalence of fungal species causing post-harvest diseases of carrot in Finland

Satu Latvala[1] | Minna Haapalainen[2] | Petteri Karisto[1] | Pirjo Kivijärvi[3] | Oona Jääskeläinen[2] | Terhi Suojala-Ahlfors[4]

[1]Natural Resources, Natural Resources Institute Finland (Luke), Jokioinen, Finland

[2]Department of Agricultural Sciences, University of Helsinki, Helsinki, Finland

[3]Production Systems, Natural Resources Institute Finland (Luke), Mikkeli, Finland

[4]Production Systems, Natural Resources Institute Finland (Luke), Piikkiö, Finland

Correspondence
Satu Latvala, Natural Resources, Natural Resources Institute Finland (Luke), Jokioinen, Finland.
Email: satu.latvala@luke.fi

Present addresses
Minna Haapalainen, Natural Resources, Natural Resources Institute Finland (Luke), Helsinki, Finland; and Oona Jääskeläinen, Verdera/Lallemand Plant Care, Espoo, Finland.

Funding information
the European Agricultural Fund for Rural Development, Grant/Award Number: 15923

Abstract

Post-harvest diseases cause significant economic losses in the carrot production chain. In this study, storage losses and fungal pathogens causing them were analysed in the carrot yield from 52 different field plots in four areas in Finland in 3 years (2016–2018). Over 30,000 carrots were sampled and analysed at three time points during cold storage at 0–1°C. In March, after 5–6 months' storage, the average loss due to diseases was 20%–21% every year. Decay of the root tip was the most common disease symptom, followed by pits on the side and black rot in the crown, detected in 69.2%, 15.0% and 9.0% of the symptomatic samples, respectively. Both intensive carrot cultivation practice and early timing of harvest increased storage losses. Pathogens in 3057 symptomatic carrot tissue samples were isolated by culturing, and fungal species were identified. The most common fungal species detected were *Mycocentrospora acerina*, *Botrytis cinerea* and *Fusarium* spp., especially *F. avenaceum*. However, the frequency of different pathogens varied between the different years and time points during storage. Species-specific PCR tests revealed that *M. acerina* and *F. avenaceum* were present in many early time-point samples where they could not yet be detected by the culturing method. In Finland, this study on carrot post-harvest diseases is the first large-scale survey in which the fungal pathogens were isolated and identified by laboratory tests. In comparison with the previous studies, *Fusarium* spp. were detected more frequently in this study, while grey mould and Sclerotinia rot were less frequent.

KEYWORDS
crop rotation, culturing method, *Daucus carota*, fungal diseases, *Fusarium*, harvest time, PCR

1 | INTRODUCTION

Carrot (*Daucus carota*) is the most important outdoor vegetable in Finland with 76,000 tonnes production and 1726 ha cultivated area in 2021 (OSF: Natural Resources Institute Finland, Horticultural Statistics). This production volume is approximately 10% of the production in each of the major carrot producer countries in Europe: Germany, Poland and the United Kingdom (EUROSTAT, online data code APRO_CPSH1). However, in relation to the population in each country, the carrot production volume in Finland is large. Because the growing season is short in the Nordic countries, including Finland, most of the carrot yield needs to be stored in the cold store

(temperature 0–1°C) for several months before sale. During the long storage, post-harvest diseases cause significant losses, the level of which differs between years and field plots (Suojala, 1999; Suojala-Ahlfors & Laamanen, 2014). Hermansen and Amundsen (1995) reported that in Norway typical losses due to diseases were 20%–50% during 5–6 months' storage. In Finland, the average proportion of diseased carrots in March, after 5–6 months' storage, was 36% and 25% in the years 2005 and 2006, respectively, while the proportion of diseased carrots in different lots varied between 0 and 100% (Vanhala et al., 2008).

Post-harvest diseases of carrot are mainly caused by different soil-borne fungi and bacteria (Davis & Raid, 2002; Papoutsis & Edelenbos, 2021; Snowdon, 1992). Infection can occur either during the growing season in the field, during the harvesting process, or during the storage. The pathogens present in the field soil, contaminated surfaces, or diseased carrots in the same harvest lot can enter the carrot root through wounds caused by harvesting process and cause disease later during the storage (Davis & Raid, 2002; Kora et al., 2005a; Papoutsis & Edelenbos, 2021).

Globally, the most damaging post-harvest diseases of carrot are the fungal diseases Sclerotinia rot (*Sclerotinia sclerotiorum* (Lib.) de Bary) and grey mould (*Botrytis cinerea* Pers. ex Fr.), which cause losses especially after growing seasons with high precipitation (Davis & Raid, 2002; Li et al., 2018; Kora et al., 2005b; Papoutsis & Edelenbos, 2021). Both *B. cinerea* and *S. sclerotiorum* have a wide host range and these fungi can overwinter as sclerotia in dead or living plants or in the soil (Davis & Raid, 2002). Sclerotinia rot is a significant problem in both production and storage, and it can spoil the carrots already at the beginning of the storage period (Davis & Raid, 2002; Papoutsis & Edelenbos, 2021). *Botrytis cinerea* can survive on various surfaces in storage and packing sheds. In the modern refrigerated packing and storage facilities, grey mould is generally a minor problem (Davis & Raid, 2002). Losses due to grey mould can, however, increase with the duration of storage, as desiccation makes the carrots more susceptible to infections (Davis & Raid, 2002).

In Northern Europe, liquorice rot is one of the major post-harvest diseases of carrot (Hermansen et al., 2012). This disease is mainly caused by *Mycocentrospora acerina*, although other fungal species in the genera *Fusarium* and *Alternaria* can cause similar symptoms (Mohamad, 2021). Plant roots usually get infected by *M. acerina* already during the growing season via ascospores in the soil but can also get infected during the harvesting process. During storage, the disease can spread in the infected carrots, even at 0°C (Snowdon, 1992). Yet the spread of *M. acerina* from diseased to healthy roots is limited, since intact plant tissues are resistant to the fungus (Davis & Raid, 2002). *Mycocentrospora acerina* is a polyphagous fungus, having several weeds as alternate hosts that can maintain and increase the fungus in the field soil, even in the years when carrot is not grown (Hermansen, 1992). In Finland, liquorice rot became common in the 1980s (Tahvonen, 1989). Later, Suojala (1999) reported that based on the disease symptoms liquorice rot and grey mould were the most common storage diseases of carrot. Parikka (2008) found that liquorice rot was the main disease detected in carrots stored for more than 3 months, yet grey mould and Sclerotinia rot also caused significant storage losses.

Fusarium spp. have also been associated with carrot storage diseases. When occurring alone, they can cause brown leatherlike rot on carrot surface and crown (Snowdon, 1992). *Fusarium* spp. are often considered as secondary pathogens which utilize the tissue already damaged by other pathogens, thus increasing the damage. Especially, *Fusarium avenaceum* has been reported to cause spoilage of carrots (e.g. Pascouau et al., 2023).

Various fungal species and oomycetes can cause pits and cavities which spoil the appearance and marketability of carrots (Hiltunen & White, 2002; Kastelein et al., 2007). Some pits and cavities can already be seen at harvest in the field, for example, several *Pythium* species can cause carrot cavity spot in the field (Hiltunen & White, 2002). However, these oomycetes have a minor role in causing post-harvest diseases that spoil carrots during the cold storage (Davis & Raid, 2002). The majority of visible damages appearing during a long cold storage are caused by fungal pathogens. In the Netherlands *Rhexocercosporidium carotae* causing significant pit disease problems was reported (Kastelein et al., 2007), and in Norway *Athelia arachnoidea* (synonyms *Rhizoctonia carotae*, *Fibulorhizoctonia carotae*), causing crater rot, was reported as an important post-harvest pathogen (Hermansen et al., 2012). *Alternaria radicina* is a seedborne pathogen that occurs in most carrot production areas of the world and causes black rot, appearing as dark pits and cavities (Davis & Raid, 2002; Farrar et al., 2004; Kastelein et al., 2007; Papoutsis & Edelenbos, 2021). Also, black root rot, caused by *Berkeleyomyces basicola* (synonym *Thielaviopsis basicola*) and *B. rouxiae*, is an important fungal disease affecting carrot quality worldwide (Nel et al., 2019; Papoutsis & Edelenbos, 2021). *B. basicola* infection develops in storage after handling of carrots in the packaging station, especially if the storage temperature and humidity are too high, and symptoms lead to vast decay of the carrots packaged in plastic bags (Papoutsis & Edelenbos, 2021).

Bacterial soft rot can also cause spoilage of carrots, both in the field, at warm temperatures and in case of waterlogging, and during storage, under low oxygen (poorly ventilated) conditions (Farrar et al., 2000; Lampert et al., 2017). Pectolytic bacteria identified as causal agents of carrot soft rot include *Pectobacterium*, *Erwinia* and *Pseudomonas* spp. (Kahala et al., 2012), and more recently Leuconostoc mesenteroides was found as a causal agent of bacterial oozing (Lampert et al., 2017).

Carrot is a cool climate crop, sensitive to high temperatures, and thus the global climate warming can affect the yield and quality of this vegetable. Both thermal stress and elevated CO_2 level have been experimentally shown to reduce the yield and quality of carrot (Azam et al., 2013; Ibrahim et al., 2006). As different fungal pathogens have different optimal growth temperatures, the elevated temperatures may also affect the pathogen populations, favouring some species and suppressing others. The climate change has already altered the crop growing conditions, which may affect the plants' susceptibility to diseases and the occurrence of pathogens. Hence, knowledge of the current pathogen incidence is needed for effective disease control. This

study is the first large-scale survey on carrot post-harvest diseases in Finland where the fungal pathogens were isolated and identified by laboratory tests, in addition to identifying the disease symptoms. The specific aims of this study were (i) to determine the level of post-harvest losses due to the pathogens in the current climate conditions and with the agronomical practices used in Finland, (ii) to identify the main fungal pathogens spoiling carrots in cold storage at different time points, and (iii) to compare two methods, fungal culturing and PCR detection for identification of the main post-harvest pathogens.

2 | MATERIALS AND METHODS

2.1 | Carrot sampling

Carrot samples were collected manually from 52 field plots from four regions in Finland—Southwest Finland, Häme, South Savo and North Savo—at the end of the growing season in 2016, 2017 and 2018. The samples were collected in September–October, as close to the farmer's harvest time as possible, between 8 September and 4 October in 2016, from 14 September to 9 October in 2017 and from 19 September to 9 October in 2018. Due to crop rotation, the fields used for carrot growing were different in different years. From each field, approximately 12 kg of carrots were collected as samples from five different points, to form a 60-kg lot per field. The leaves were manually removed on the field.

The number of collected lots was 16 in 2016, 15 in 2017 and 21 in 2018. In 2017, one lot was obtained from the farmer's storage, while the other lots were collected directly from the field. The main cultivars were Maestro (26 lots), Romance (eight lots) and Natalja (seven lots). 21 lots were grown in organic soils and 31 lots in mineral soils. Background information of farming practices and pre-plants grown on the survey plots was collected by interviewing the farmers. However, all these data were not obtained for all the fields, and thus only the frequency of carrot growing was utilized in the data analysis.

2.2 | Storage conditions and processing of carrot samples

All carrot samples were taken to storage facilities (Luke Piikkiö, Kaarina, Finland) where each 60-kg lot was divided into seven equal subsamples, weighing approximately 8 kg each, and packed in woven polypropylene bags. One of the subsamples was stored at 10°C for 6 weeks to predict the storability of the lot (data not shown). Six subsamples were taken to the cold store (temperature 0–1°C) for storage experiment. The storage samples were inspected at three time points, with 6-week intervals, in December, January, and March. Two parallel storage samples (a and b) were analysed at each time point, and their weight was measured before and after storage. The number of stored carrots in each year and at each inspection time point is presented in Data S1.

The inspection was always started by separating the healthy looking and diseased carrots. The number of carrots in both groups was counted and the carrots were weighed. Storage loss due to diseases was calculated both per number and per weight (the total weight of carrots after storage).

2.3 | Analysis of the diseased carrots

The diseased carrots were washed and divided into four categories based on visible (macroscopical) symptoms. If there was more than one type of symptom present on a root, the same carrot was recorded into several categories accordingly. The percentage of carrots in different symptom categories was counted per total number of carrots in each lot. The symptom categories were:

1. Decay (starting) in the root tip.
2. Pits and cavities on the side of the root.
3. Black or dark brown rot in the crown of the root.
4. Carrots spoilt by grey mould or Sclerotinia rot, and the pathogen identifiable by sclerotia. The disease was recognized as grey mould if the split sclerotia were brown inside and as Sclerotinia rot when they were white inside.

In addition to identifying the diseases based on the visible symptoms, samples were taken from maximum 30 carrots per lot in each symptom category to identify the fungal pathogens by culturing method. No samples from the carrots severely spoiled by grey mould or Sclerotinia rot were plated on agar for fungal identification. In total, 3057 samples were taken for fungal identification tests (Data S1).

2.4 | Isolation and identification of fungi by culturing method

Since pentachloronitrobenzene (PCNB) agar has been reported to be semi-selective for *Fusarium* spp. (Nelson et al., 1983), we tested whether it would be applicable for isolating the other fungal pathogens commonly affecting carrots. For comparison, different filamental fungi isolated from symptomatic carrot roots were grown in parallel on PCNB and potato dextrose agar (PDA) media. Two isolates of each of the species *F. avenaceum*, *M. acerina* and *B. cinerea*, and two isolates representing the *Cylindrocarpon* spp. complex (one *Neonectria* and one *Ilyonectria*) were chosen for the experiment. Eight agar plugs (diameter 5 mm) were cut with a sterile cork bore from each 1-week-old fungal culture on PDA. Four of these plugs were transferred onto PCNB medium and the other four plugs onto PDA medium, one plug per plate. These 90 mm plates were incubated at room temperature (20–22°C) in darkness. The fungal cultures were photographed, and their diameter was measured after 2, 5, and 7 days of incubation.

To isolate and identify fungal species from carrots classified in different symptom categories, a cubic piece with approximately 5 mm sides was cut with a sterile scalpel at the edge between injured and healthy-looking tissue. These tissue pieces were surface-sterilized by soaking in 70% ethanol for 1 min, rinsed twice with deionized water

and dried on sterile paper towel in laminar flow cabinet for 30 mins. The pieces were then transferred onto PCNB agar medium (Nelson et al., 1983), and incubated at room temperature (20–22°C) in darkness for 5 days. Tips of the fungal hyphae growing out of the plant tissue pieces on the PCNB agar were subsequently transferred onto standard PDA medium (Difco™ Potato Dextrose Agar, Becton, Dickinson and Company). If different fungi grew from a single piece, they were all transferred separately onto PDA. The pure cultures were incubated in darkness at room temperature for 3–4 weeks. The fungal species were tentatively identified by the culture features and spore morphology according to the systematics by Gerlach and Nirenberg (1982).

2.5 | Identification by molecular methods

To compare the results of the culturing method to PCR detection of pathogens, we used both methods on a subset of samples. From the 2018 yield, 25 symptomatic carrots from the disease prediction samples, 10 from the December storage samples, 20 from the January storage samples and 20 from the March storage samples were tested by species-specific PCR in parallel with the culturing method (75 samples in total).

For this subset of samples, a piece of symptomatic tissue from each carrot was split into two halves. One of them was surface-sterilized and placed on a selective medium as described above. The other half was frozen at −80°C prior to DNA extraction. Then 0.1 g of tissue was ground using FastPrep-24 tissue and cell homogenizer (MP Biomedicals) in tubes containing Lysing matrix A (MP Biomedicals). The total DNA was extracted using DNeasy Plant Mini Kit (Qiagen) according to the manufacturer's instructions.

The DNA samples were tested for the presence of *M. acerina*, *F. avenaceum* and *B. cinerea* by single PCR reactions using species-specific primers. The primers were KLO15F (CGGTCGGACTCCATTCAAAC) and KLO20R (GCTCCGAAGCAAGTACGCCG) for *M. acerina* (Hermansen et al., 2012), FA-F1 (AACATACCTTAATGTTGCCTCGG) and FA-R1 (ATCCCCAACACCAAACCCGAG) for *F. avenaceum* (Mishra et al., 2003), and Bc-F (CAGGAAACACTTTTGGGGATA) and Bc-R (GAGGGACAAGAAAATCGACTAA) for *B. cinerea* (Fan et al., 2015). The sizes of the expected PCR products were 314 bp, 314 bp and 354 bp, respectively.

PCR was run using 0.4 µL Phire Hot Start II DNA polymerase with its 1× reaction buffer (Thermo Scientific), 200 µM dNTPs, primers at 250 nM concentration, and 20 ng template DNA in 20 µL reaction volume. The programme was as follows: 98°C for 30 s; 35 cycles of 98°C for 1\ 0 s, annealing for 10 s, extension at 72°C for 30 s; followed by a final extension at 72°C for 5 min. Annealing temperatures were 63°C for *M. acerina* and *F. avenaceum*, and 60°C for *B. cinerea*. The PCR products were analysed by electrophoresis on 1.5% agarose gels in Tris-borate-EDTA buffer. Gels were stained with ethidium bromide and visualized under a UV-transilluminator (SCIE-PLAS).

A fragment of the translation elongation factor 1-alpha gene (*TEF*) of a few *Fusarium*-positive fungal and carrot tissue samples was amplified and sequenced as described in Haapalainen et al. (2016). Primers ITS1 and ITS4 (White et al., 1990) were used to amplify a fragment of ITS region of *Pythium* spp. by PCR, using the same protocol as described above, with the annealing temperature of 60°C. Prior to Sanger sequencing, the PCR products of approximately 1 kb in size were purified using QIAquick PCR Purification kit (Qiagen). The *TEF* and ITS sequences obtained were compared to the sequences available in the GenBank, by using blastn tool (NCBI).

2.6 | Weather conditions

Weather data were obtained from the observation stations of Finnish Meteorological Institute (https://fmi.fi/en). Mean temperature and precipitation sum in May–September from four observation stations, situated nearby the carrot fields, were compared to the long-term averages in the period 1991–2020 (Data S2). In 2016, the mean temperature in May–September was 0.7–1.3°C higher than the long-term average, whereas in 2017 the mean temperature was 0.3–1.3°C lower than the long-term average. The growing season in 2018 was the warmest, and the mean temperature was 2.0–2.9°C higher than the long-term average.

In Southwest Finland, the precipitation sum in the growing season was lower than the long-term average in all the 3 years. In the other regions the precipitation was close to the average in 2016 and 2017, whereas in 2018 the precipitation sum was lower than the long-term average.

2.7 | Statistical analyses

To compare the growth of different fungal species on solid PCNB and PDA media, the measurement data of the two isolates representing the same species or species complex (*Cylindrocarpon*) were combined to obtain groups with $N = 8$ for each species. Comparisons between these species groups were performed using Welch two sample *t*-test in R (version 4.2.1). Difference between the species was considered significant when the *p*-value of two-sided test was less than .05.

Significance of the effect of crop rotation and harvest time on storage losses due to diseases was tested by Welch two sample *t*-test in R. The harvest times of different carrot lots were classified into three categories. The "average" harvest time ($N = 14$) was defined as ±1 week from the median harvest date in each year. Harvest dates earlier than the "average" were classified to "early" harvest category ($N = 21$) and harvest dates later than the "average" were classified to "late" harvest category ($N = 17$). Significance of differences in the relative amounts of different fungal pathogens detected by culturing method in the carrot lots with different crop rotation background or different harvest time was tested by paired *t*-test in R. The significance of differences in the frequency of different fungal pathogens detected at different time points during the cold storage was tested by Pearson's Chi-squared test in R. The *p*-values less than .05 were considered significant.

3 | RESULTS

3.1 | Storage losses due to diseases

Over 30,000 carrots collected from 52 different field plots in four areas in Finland in 3 years (2016–2018) were sampled and analysed at three time points during the cold storage at 0–1°C. The average storage loss of carrots was roughly similar in all the 3 years. In December the average proportion of diseased carrots was 4%–10%, in January 11–12% and in March, in the end of the storage period, 20%–21% of the weight (Figure 1). However, in the individual lots the storage losses varied between 0% and 76%. The carrot lots with the best storability remained healthy up to the end of the storage period, whereas in the poorest lots the storage loss was already high in December. No significant differences were detected in the amount of storage loss between the lots from different carrot cultivation regions (data not shown).

3.2 | Disease symptoms

The most common symptom in carrots was softening and darkening of the root tip (Figure 2a,b), comprising on average 69.2% of all the visible symptoms developed during the cold storage. The appearance of the root tip symptoms varied in different samples from slight darkening of the end of the root tip to fully softened black tissue over 2–3 cm from the tip and even beyond that. 45% of the root tip symptoms were severe, having darkened and softened tissue more than 1 cm up from the tip (Figure 2b), and 55% of the symptoms were milder (Figure 2a). In the December samples the symptoms were still mild, but later the disease progress was rapid in some lots. In March, identifiable mycelium and sclerotia of *B. cinerea* were observed in some symptoms started from the root tip (Figure 2c). In total, the proportion of carrots visibly spoilt by grey mould or Sclerotinia rot was 6.8%. In March 2018, four sample lots from the parallel set b were discarded due to an uncontrollable spread of Sclerotinia rot. Deep and/or large pits and cavities, and small and clearly defined pits on the side of the root were also detected (Figure 2d,e). On average, different pits constituted 15.0% and black rot in the crown (Figure 2f) constituted 9.0% of the visible symptoms. During the cold storage all the symptoms caused by pathogens increased; however, the proportions of different symptom types increased at slightly different rates, and thus the relative proportion of pit symptom decreased by time. The variation between individual carrot lots in the amount of diseased carrots was large (Figure 1) and the proportions of different symptom types were also different in different lots.

3.3 | Applicability of the PCNB-PDA culturing method for different fungal pathogens

The post-harvest fungal pathogens commonly found on carrot in Finland—*M. acerina*, *B. cinerea*, *F. avenaceum*, *Ilyonectria* spp. and *Neonectria* spp. (*Cylindrocarpon* spp. complex)—were all able to grow well on PCNB medium (Table 1; Data S3; S4). However, some differences in the growth rate were detected. During the first 2 days on PCNB medium the *B. cinerea* isolates grew significantly less (*t*-test, $p < .05$) than the other fungi, which did not significantly differ from each other. In 5 days, *M. acerina* isolates had grown significantly more than the other species (Table 1). The difference between *B. cinerea* and *F. avenaceum* was also significant ($p = .0119$), while neither of these fungi showed significant difference from the *Cylindrocarpon* species. In 7 days, *M. acerina* isolates had grown significantly more than the other species, while there were no significant differences between the other species. On PDA medium, all the fungal species differed from each other significantly in their growth during the first 2 days. In 5 days, there was no significant difference between *F. avenaceum* and the *Cylindrocarpon* isolates, while *M. acerina* and *B. cinerea* significantly differed from them and from each other (Table 1). Both of the *B. cinerea* isolates completely filled the PDA plates within 5 days. After 7 days, *M. acerina* isolates had grown significantly more than *F. avenaceum* and *Cylindrocarpon* isolates, which did not differ significantly from each other (Data S3; S4).

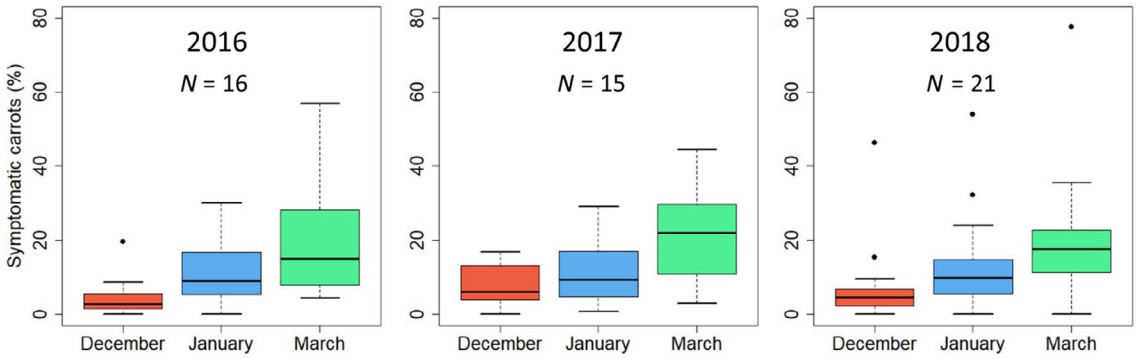

FIGURE 1 Proportion of symptomatic carrots at three time points during the cold storage. *N* is the number of stored lots inspected at each time point. In the box plots the line inside each box indicates the median value.

FIGURE 2 Different symptoms observed in carrots. (a) and (b) softening and darkening of the root tip, (c) identifiable mycelium and sclerotia of *B. cinerea*, (d) small and clearly defined pits on the side of the root, (e) deep and/or large pits and holes on the side of the root, and (f) black rot in the crown.

3.4 | Pathogens in symptomatic carrots identified by the culturing method

Altogether 3057 symptomatic samples taken from the cold-stored carrots were tested by the culturing method for pathogen identification by morphological characteristics (Data S1). Of these sample pieces 72.6% were of the root tip symptoms, 17.1% of pits on the side of the root and 10.3% of the crown black rot. The most common fungal species detected in the symptomatic tissue samples were *M. acerina*, *B. cinerea* and *Fusarium* spp., of which 65% were identified as *F. avenaceum*, 22% as *F. sambucinum*, and the rest as other species, including *F. culmorum* and *F. sporotrichioides*.

Cylindrocarpon spp. were also frequently detected. *Acremonium* spp., *Mucor* spp., and *Phoma* spp. were identified as minor species, as well as some oomycetes, tentatively identified as *Pythium* spp. in 3% of the tested samples, from all the symptom types. ITS sequencing was performed for 13 of the isolates identified as *Pythium*, and based on the sequence four isolates were confirmed as *P. intermedium*, one as *P. sylvaticum* and another as *P. irregulare*, whereas the other seven isolates remained as undefined *Pythium* isolates.

During the cold storage the frequency of different pathogens detected in the symptomatic tissue samples changed in the course of time. *Fusarium* spp. were typically detected early in the storage period, whereas *M. acerina* and *B. cinerea* became more prevalent later in January and March (Figure 3). *Fusarium* spp. and *M. acerina* were the most frequent fungal species detected in all the three symptom types. *Acremonium* spp., *Mucor* spp., and *Phoma* spp. and oomycetes that were detected to a minor extent in the symptomatic tissues, are included in the "other microbes" in Figure 3.

The proportions of different pathogens also varied between the years. In 2016, *B. cinerea* was more common than in 2017 and 2018. In 2017, *M. acerina* was found to be the most common fungus, and *Cylindrocarpon* spp. were also detected from many of the symptomatic root tip samples. In 2018, fungal isolates were only obtained from 30% of the symptomatic root tips and from 50% of the black crown samples or large cavities on the side. Instead, bacteria were growing from many samples on the culture medium (Figure 3).

TABLE 1 Growth of different fungal isolates on the semi-selective PCNB agar and the non-selective potato dextrose agar.

Fungal species[a]	Isolate	Diameter of fungal growth on PCNB agar medium (mm)			5 days *t*-test[b]	Diameter of fungal growth on potato dextrose agar medium (mm)			5 days *t*-test
		2 days average (SD)	5 days average (SD)	7 days average (SD)		2 days average (SD)	5 days average (SD)	7 days average (SD)	
Neonectria sp.	OJ16	8.00 (0.00)	12.25 (1.09)	16.00 (1.22)		11.75 (0.83)	23.50 (0.87)	34.50 (0.50)	
Ilyonectria sp.	OJ25	9.75 (1.09)	20.25 (1.64)	26.00 (1.58)	AB	13.50 (2.06)	29.75 (2.38)	41.00 (2.92)	A
Fusarium avenaceum	OJ17	9.25 (0.83)	17.25 (0.43)	22.00 (1.41)		13.25 (0.83)	25.75 (0.43)	33.50 (1.12)	
Fusarium avenaceum	FAV11	10.00 (0.00)	21.75 (3.42)	29.75 (3.42)	A	18.75 (2.59)	38.25 (3.70)	47.25 (3.96)	B
Mycocentrospora acerina	MAC041003	13.50 (1.12)	25.75 (0.43)	34.00 (0.71)		22.50 (1.50)	50.50 (3.20)	66.00 (2.35)	
Mycocentrospora acerina	MAC041065	7.75 (0.83)	25.00 (2.24)	35.25 (0.43)	C	18.75 (0.43)	44.75 (0.43)	64.75 (0.43)	C
Botrytis cinerea	OJ3	7.00 (0.00)	17.00 (1.73)	24.00 (3.00)		39.00 (2.83)	90.00 (0.00)	90.00 (0.00)	
Botrytis cinerea	BC4	7.00 (0.00)	12.75 (1.09)	18.00 (2.12)	B	37.75 (1.79)	90.00 (0.00)	90.00 (0.00)	D

[a]Fungal species were confirmed by ITS sequence and for *Fusarium* spp. and *Cylindrocarpon* spp. (*Neonectria* and *Ilyonectria*) also by *TEF* gene sequence.
[b]The fungal genera (*Cylindrocarpon*, *Fusarium*, *Mycocentrospora* and *Botrytis*) whose growth significantly (*t*-test $p < .05$) differed from the other genera are designated with different capital letters.

3.5 | PCR detection in comparison to the culturing method

In total, 75 symptomatic samples from different time points within the storage period were tested by species-specific PCR in parallel with the culturing method. Based on the identification by the culturing method, samples having mainly one of the following fungi: *F. avenaceum*, *M. acerina* or *B. cinerea*, or no culturable fungi at all, were selected. Three of the chosen samples contained both *F. avenaceum* and *M. acerina*. When DNA extracted from these samples was tested for *F. avenaceum*, *M. acerina* and *B. cinerea* by species-specific PCR, more than one of the three fungi were detected in most of the samples. *Mycocentrospora acerina* was detected in 66 samples, *F. avenaceum* in 54 and *B. cinerea* in 20 samples.

In the disease prediction samples taken in November, *M. acerina* and *B. cinerea* could already be detected by PCR, but not by the culturing method. PCR tests also revealed that many storage samples contained all the three fungi—*B. cinerea*, *M. acerina* and *F. avenaceum*—even though only *B. cinerea* was growing on the culture plates. While the two methods agreed quite well in the detection of *B. cinerea*, many more positive results were obtained for *M. acerina* and *F. avenaceum* by PCR than by culturing (Figure 4). In 30% of the samples tested, none of these three fungal species was found by the culturing method, whereas in species-specific PCR only one sample was negative for all three, suggesting that the detection limits of the PCR methods were lower. By PCR, two different fungal species were detected in 66.7% of the samples and all the three species were detected in 10.7% of the samples.

Based on the *TEF* gene sequences, obtained from a few *Fusarium*-positive samples from the culture plates and symptomatic carrot tissue samples positive in FA-F1/FA-R1 PCR test, the main *Fusarium* species was confirmed as *F. avenaceum*. Two of these sequences, representing the two different sequence types detected, were deposited to GenBank (accession numbers PP357886 and PP357887).

3.6 | Effect of pre-crop and harvest time on storage losses and pathogen community

On those fields in which carrot had also been grown during the previous 2 years, the proportion of symptomatic carrots was on average twice as high as on the fields where carrot had not been grown. The difference was statistically significant ($p < .05$) at all the three sampling time points during the storage (Data S5). When classified by the time of harvest, 21 carrot lots were harvested earlier and 17 later than the average time and 14 lots were harvested at the average time (median ± 1 week). The early harvested lots had significantly ($p < .05$) higher proportion of symptomatic carrots than the lots harvested at the average time or later (Data S5). However, when the proportions of symptomatic carrots were compared between sample lots grouped by both the harvest time and pre-crop (carrot or no carrot), it was the combination of early harvest and intensive carrot culturing that significantly increased the disease incidence (Figure 5). In December and

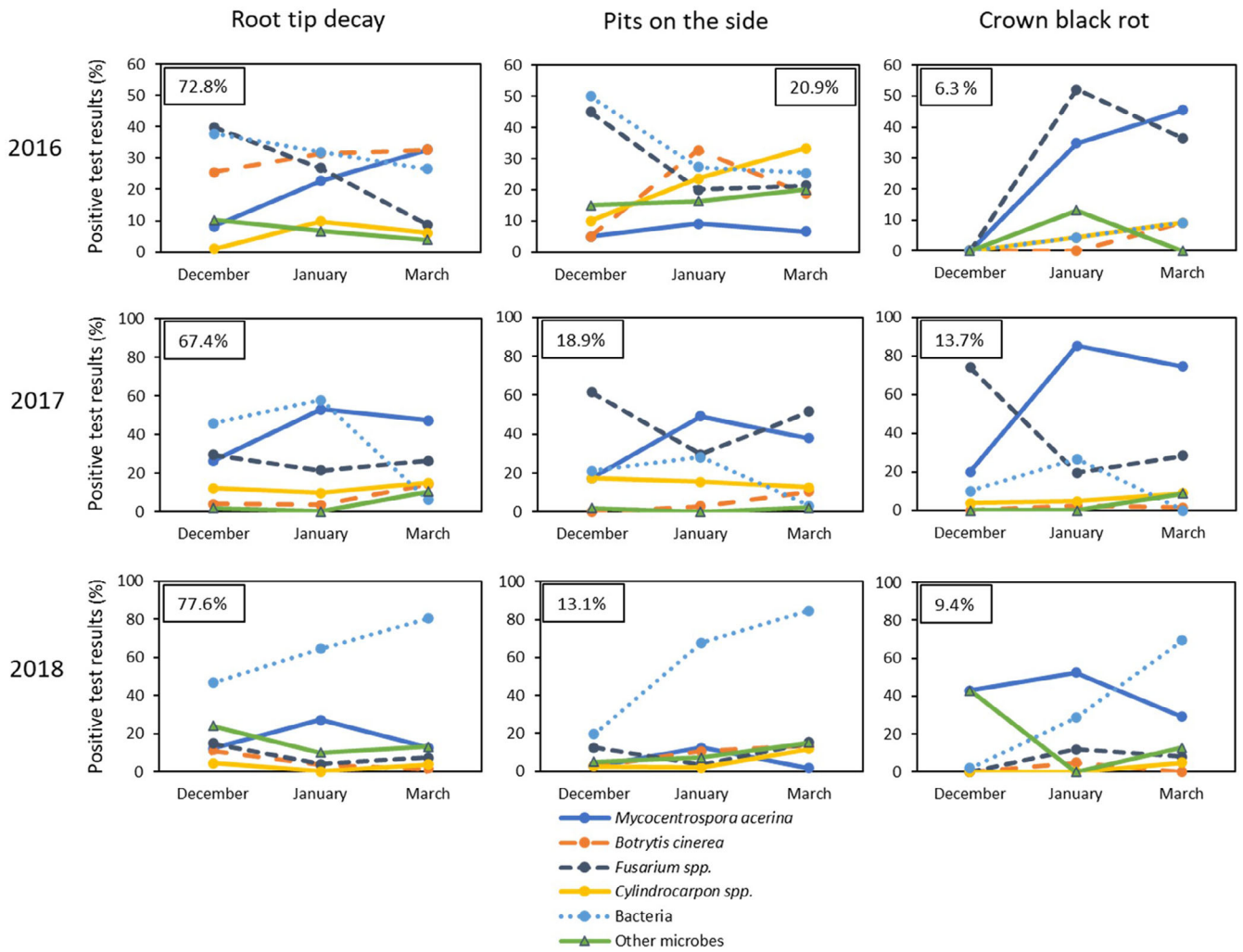

FIGURE 3 Fungal pathogens and other microbes identified from symptomatic carrot samples by the culturing method. Samples were taken from three different types of symptoms: root tip decay, pits on the side and crown black rot. The percentage of each sample type of all the tested samples in each year is shown in a box inside each panel of the figure.

January, the group "Early harvest & Carrot grown in previous 2 years" had significantly ($p < .05$) higher disease incidence than the other groups, except for the group "Early harvest & No data on pre-crop". In March, the difference between the groups "Early harvest & Carrot grown in previous 2 years" and "Average or late harvest & Carrot grown in previous 2 years" was only marginally significant ($p = .0682$).

The relative amounts of different fungal pathogens detected by the culturing method were significantly different between the plots where carrot had been or had not been grown during the last 2 years (Chi-squared test, df = 3, in December $\chi^2 = 15.4$ and $p = .0015$, in January $\chi^2 = 34.6$ and $p < .001$, in March $\chi^2 = 24.7$ and $p < .001$). Especially, *Fusarium* spp. were significantly more frequent in the storage lots with intensive carrot culture background ($p < .05$, paired *t*-test), whereas *Cylindrocarpon* spp. were more frequently detected in the samples from fields where carrot had not been grown during the last 2 years ($p = .0092$; Figure 6a). In December and January, *B. cinerea* was significantly more frequent in the samples with intensive carrot cultivation background ($p = .0136$), whereas in March there was no significant difference. The frequency of *M. acerina* was higher in January and March in the samples from fields with intensive carrot cultivation background, however, the difference was not statistically significant. The effect of early harvest time on the pathogens was similar to the effect of intensive carrot cultivation, except for *Fusarium* spp. (Figure 6b). At all the time points, the proportions of tests positive for *M. acerina*, *B. cinerea*, *Fusarium* spp. and *Cylindrocarpon* spp. were different between the early harvest samples and the late or average harvest time samples ($p < .001$, Chi-squared test). Especially, *Cylindrocarpon* spp. were more frequent in the late or average harvest time samples ($p < .01$, paired *t*-test). In contrast, *M. acerina* was significantly more frequent in the early harvest samples than in the samples from lots harvested at late or average time in 2017 ($p < .05$, paired *t*-test), whereas for the samples harvested in 2016 and 2018 the effect of harvest time was not significant. When pre-crop and harvest time were viewed together, *B. cinerea* was significantly more frequent in the early harvest samples when

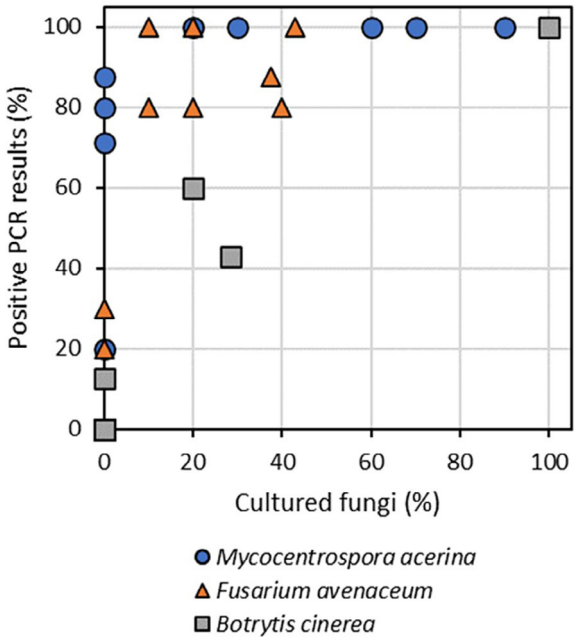

FIGURE 4 Comparison of two different methods in detection of fungal pathogens. Symptomatic carrot samples from nine sample lots, taken either before or during the cold storage, were tested for three major pathogens by both the culturing method and by species-specific PCR. For each sample lot, the proportion of positive culturing results is shown on the x-axis and the proportion of positive species-specific PCR tests is shown on the y-axis. The PCR tests were more sensitive, yielding more positive results.

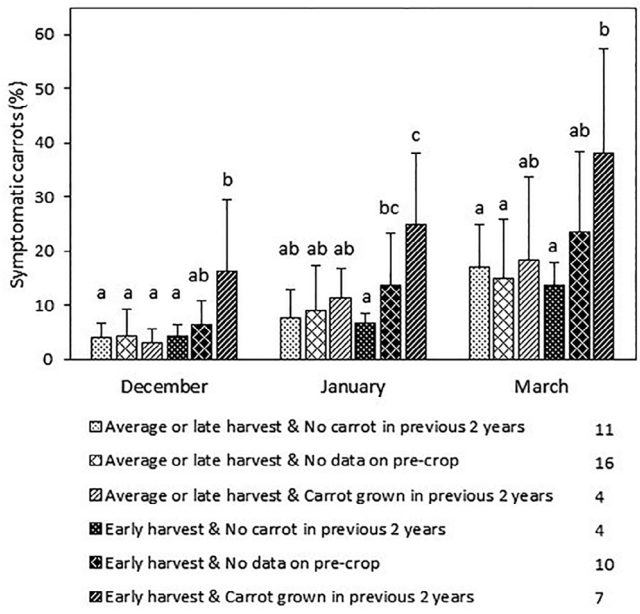

FIGURE 5 Effect of harvest time and pre-crop on the proportion of symptomatic carrots at different time points during storage. At all the time points the group "Early harvest & Carrot grown in previous 2 years" showed a higher proportion of symptomatic carrots than most other groups. The groups significantly ($p < .05$) different from each other are marked with different letters. Group means are shown, and the error bars indicate standard deviation. The numbers on the right side of the legend denote the number of sample lots in each group.

carrot had been grown in the previous 2 years ($p < .05$, paired t-test). In contrast, significantly more bacteria were detected in the early harvest samples when carrot had not been grown in the previous 2 years.

4 | DISCUSSION

In the carrot production chain different post-harvest diseases cause significant economic losses and decline of quality (Davis & Raid, 2002; Papoutsis & Edelenbos, 2021). In this study, the average loss due to post-harvest diseases after 5–6 months' storage period was 20%–21% each year (2016–2018), even though the weather conditions were different during the growing season in the different years. This is slightly less than the losses during storage reported in the earlier Nordic studies (Hermansen & Amundsen, 1995; Vanhala et al., 2008). However, there was a large variation in quality between the individual storage lots. As the storage time increased, the number of diseased carrots increased, and the symptoms became more severe. The most common symptom was softening or decaying of the root tip, while pits and cavities on the side of the carrot and black rot in the crown were minor symptom types. The pit symptom increased at a lower rate than the other symptom types and thus its relative proportion of all the symptoms decreased during the storage.

To identify the major pathogens in the disease symptoms, we chose to use two-step culturing method. Although the PCNB medium was earlier reported to be semi-selective for *Fusarium* spp. (Nelson et al., 1983), our culturing test results showed that the most common fungal pathogens causing post-harvest diseases in carrots in Finland were all able to grow well on the PCNB medium. Especially, *M. acerina*, a common species causing carrot liquorice rot, was found to grow faster on PCNB than *F. avenaceum*. As the *Cylindrocarpon* species also grew on PCNB as well as *F. avenaceum* and *B. cinerea* in 5 days, this medium did not seem to favour *Fusarium* over the other main pathogens of carrot. The applicability of PCNB medium for isolating other fungi than *Fusarium* spp. is supported by the observations of Castellá et al. (1997) who compared PCNB medium and malachite green medium for their selectivity for *Fusarium* spp. They reported that malachite green medium performed as a potent selective medium for *Fusarium* spp., whereas PCNB allowed the growth of many different fungal species, including *Zygomycetes* and yeasts. Thus, this medium was chosen as the first step medium for this study, to restrict the growth of soil bacteria and enable the isolation of fungi from carrots, followed by cultivation of the fungi as pure cultures on PDA medium. In general, when isolating fungi from plant roots, using a semi-selective medium is necessary at the first step. Without this step, if placing pieces of roots directly on a non-selective medium, bacteria would overtake the fungi. Some fast-growing fungi—like *B. cinerea*—also need to be restrained, to prevent them overtaking the other species on the culture plate.

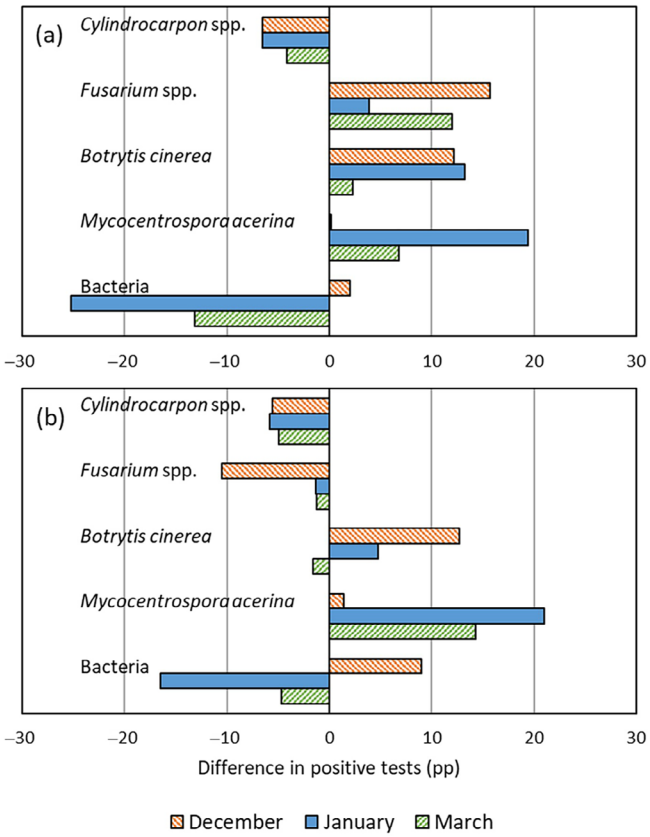

FIGURE 6 Effect of pre-crop (a) and harvest time (b) on the occurrence of different fungal pathogens and bacteria in the disease symptoms detected during storage. For each time point, the bars show how much more (or less) each type of microbe was detected in the symptomatic carrots in (a) samples from fields where carrot had been grown in the previous 2 years ($N = 802$) in comparison to samples from fields where carrot had not been grown in the previous 2 years ($N = 820$), or (b) samples from early harvest lots ($N = 1341$) in comparison to lots harvested late or at the average time ($N = 1714$). pp, percentage points.

By using this culturing method, *M. acerina*, *Fusarium* spp. and *B. cinerea* were the fungal pathogens most frequently detected from the symptomatic carrot tissues. Compared to the earlier studies in Finland (Mukula, 1957; Tahvonen, 1985), the proportion of *Fusarium* spp. was higher and the proportion of *B. cinerea* (grey mould) was lower in the symptomatic carrots in this study. Most strikingly, *S. sclerotiorum* that had earlier been a devastating post-harvest pathogen was only found in some single storage lots. This may be, at least partially, due to the improved storage conditions, as previously suggested (Geeson et al., 1988). Proper crop rotation and good knowledge of the host plants have also been found important in control of Sclerotinia rot: cultivation of oil seed rape or other hosts of *S. sclerotiorum* prior to carrot can cause quick spoilage of the crop already during the early stage of storage (Parikka, 2008).

When species-specific PCR methods were compared with the culturing method in identifying the three main fungal pathogens, *B. cinerea* was detected equally well by both methods, whereas more samples were found positive for *M. acerina* and *F. avenaceum* by PCR than by culturing. By PCR *M. acerina* could even be detected in the beginning of the storage period, when the amount of this fungus was still low, and it could be occluded on the culture medium by other more abundant fungi. The growth rate of different fungal species and different strains of the same species also differ, and thus the ones with a higher growth rate can supersede the others, even when they were equally abundant in the original sample. On the other hand, the culturing method allows identification of new pathogens and the main pathogen(s) in each sample and distinguishing between the viable and non-viable microbes in the plant tissue.

Cylindrocarpon spp. were also frequently detected from the symptomatic tissue in the root tip, pits and cavities. Few reports are available of the importance of *Cylindrocarpon* spp. as causal agents of carrot diseases (Mohamad, 2021; Mukula, 1957; Sweetingham, 1983). However, as *Cylindrocarpon* spp. are considered to cause pits and cavities on parsnip (Channon & Thomson, 1981) and potato (Choiseul et al., 2006), they may also have a role in the symptom development on carrot. *Alternaria radicina*, *Athelia arachnoidea* or *Rhexocercosporidium carotae* species, found causing pit symptoms in carrot in other countries in Europe (Årsvoll, 1969; Hermansen et al., 2012; Kastelein et al., 2007; Snowdon, 1992), were not detected in the samples from Finland. No symptoms characteristic of carrot infection by *Berkeleyomyces* spp. were detected in this study, neither were these fungi detected by culturing. Our results thus confirm the previous reports in that *Berkeleyomyces* spp. have not been found in Finland (Nel et al., 2019).

Although the focus of this study was on the fungal pathogens, the culture method used for detection also revealed the presence of some *Pythium* spp. in 3% of the carrots tested. Different *Pythium* spp. have been reported to cause carrot cavity spot, and species involved in the disease appear to vary according to geographical areas (Allain-Boulé et al., 2004; Howard et al., 1978). *P. irregulare* and *P. sylvaticum*, detected in this study, are fast-growing species which are often isolated from the older cavities, and Hiltunen and White (2002) suggested them to be secondary invaders that utilize the moisture and decaying organic matter. Although *Pythium* spp. are major pathogens affecting carrot during the growing season, they have a minor role in post-harvest diseases spoiling carrots in storage (Davis & Raid, 2002).

For the carrots grown in 2018, in a warm growing season, no fungal pathogens could be isolated from the majority of the symptomatic tissues but instead, bacteria were growing in these samples. The bacterial species were not identified in this study, however, Kahala et al. (2012) previously reported that different strains of the genera *Pseudomonas*, *Pectobacterium* and *Erwinia* were the primary bacteria causing decay of carrots in Finland during storage and marketing. These pectolytic bacteria can cause considerable financial losses, because they account for a large proportion of bacterial rot of fruits and vegetables during storage, transit, and marketing (Kahala et al., 2012).

The incidence of the main fungal pathogens also varied between the 3 years of this study. During storage time, water loss from the carrot root makes it susceptible to fungal pathogens, especially to grey mould infection. The infection often starts from the delicate root tip,

since it desiccates easily (Snowdon, 1992). The biggest weight loss during the storage was observed in the 2016 harvest, and this could, at least partially, explain the high incidence of grey mould that year. On the other hand, the highest incidence of *M. acerina* (liquorice rot) was detected in the 2017 harvest, after the growing season with the highest amount of precipitation. Previously, Hermansen et al. (2012) reported that in Norway the incidence of carrot crown rot and tip rot caused by *M. acerina* showed significant positive correlation with precipitation in July and September. In the presence of free water, the optimal germination temperature of *M. acerina* was reported to be from 14 to 22°C in vitro, however, the fungus did germinate even at 4°C (Yang et al., 2022). Thus, the weather during the growing season can significantly affect the build-up of *M. acerina* inoculum. Other factors, such as pre-crop, soil type, cropping systems and fertilization, as well as competition with other organisms can affect the development of disease (Ghorbani et al., 2008; Hermansen et al., 1997; Kastelein et al., 2007).

The prevalence of different fungal species detected in the symptomatic carrots also changed during the time in storage. *Fusarium* spp., especially *F. avenaceum*, which was a minor fungus in the old report (Tahvonen, 1985), were prevalent fungi in the December samples, whereas *M. acerina* was more prevalent in the January and March samples. This suggests that *M. acerina* infection develops slowly, which agrees with those earlier reports stating that the amount of *M. acerina* increased during the prolonged storage at low temperature (Årsvoll, 1969; Snowdon, 1992). In addition to *M. acerina* and *Fusarium* spp., *B. cinerea* was often detected in the wide shallow pits. Although *B. cinerea* has been earlier considered a major pathogen spoiling the crown of the carrot (Snowdon, 1992), in this study it was very rarely detected in the crown rot samples.

Fusarium species have not been commonly known as carrot post-harvest pathogens, however, recently carrot dry rot caused by *Fusarium* spp. has been reported in Europe (Le Moullec-Rieu et al., 2020; Stanković et al., 2015). The pathogen was identified as *F. avenaceum* in both Serbia and France, and in France also *F. tricinctum* was identified as a carrot pathogen (Le Moullec-Rieu et al., 2020). In this study the proportion of *Fusarium* fungi detected in the carrot pit symptoms was high compared to the previous reports in Nordic countries (Årsvoll, 1969; Hermansen et al., 2012; Kastelein et al., 2007; Mukula, 1957; Tahvonen, 1985), and especially *F. avenaceum* was frequently found, suggesting it has become an important pathogen of carrots in Finland. As the optimal growth temperatures for *Fusarium* spp. are relatively high, 20–25°C (Li et al., 2014; Moore, 1945), it is possible that these fungi could have benefited from the recent elevated temperatures during the growing season (Aalto et al., 2022).

The fungal pathogens detected from the carrot storage lots in this study differ from each other regarding the applicable disease control measures. While *M. acerina* is a soil-borne pathogen hosted by many weed plants, controlling *B. cinerea* with agronomical practices is generally challenging, as the airborne spores are abundant in the autumn (Snowdon, 1992). However, in this study grey mould was more common in those fields where carrot had been grown in the previous years, suggesting harmful effect of poor crop rotation on this disease as well. *Fusarium* spp. were also more common in the carrots from the plots with poorer crop rotation. An early harvest and/or carrot intensive crop rotation seemed to increase the incidence of both *B. cinerea* and *M. acerina*. Although the pre-crop (carrot or no carrot) alone was not a strong explanatory factor of the disease incidence level, together with the early harvest time it resulted in a significantly higher disease incidence. This result agrees with the previous studies in which poor crop rotation (Hermansen et al., 1997) and early harvest (Suojala, 1999) were reported to increase the risk of storage losses of carrot.

5 | CONCLUSIONS

In the 3 years of this study, the total amounts of symptomatic carrots were similar after 5 months in storage. However, the proportions of different pathogens detected from the symptoms were different each year. Of the most common fungal pathogens detected, *Fusarium* species were prevalent after 2 months of storage, whereas *M. acerina* and *B. cinerea* became more abundant later. From a large proportion of the symptoms no fungal pathogens could be isolated, and instead bacteria were detected. Thus, bacteria may also have an important role in spoiling carrots during storage, especially after a warm growing season like in the year 2018. Sclerotinia rot (*S. sclerotiorum*), which earlier was a major problem, was rarely detected in this study. Some of the fungal pathogens causing serious storage losses in other countries in Europe, like *Alternaria radicina*, *Athelia arachnoidea* or *Rhexocercosporidium carotae* were not detected in this study in Finland.

Altogether, controlling storage diseases is a complex challenge for which no single solution can be found. Our results imply the importance of optimal harvest time and proper crop rotation, which confirms the results of earlier studies. Information on the harvest time of each carrot lot could be more systematically used for planning the order in which the lots are sold to the customers, to avoid long storage of the early harvest lots. In addition, good storage conditions, with low temperature and high relative humidity, as well as avoiding mechanical damage are key factors in improved storability of carrot.

ACKNOWLEDGEMENTS

This work was funded by the European Agricultural Fund for Rural Development (project 15923). We are very grateful to our colleague Dr. Asko Hannukkala, who died in May 2020, for his precious contribution to this work. We thank Sirkka Jaakkola and Paula Rannikko from Häme University of Applied Sciences and Heikki Inkeroinen from ProAgria Eastern Finland for their help in collecting the samples. The technical assistance of Auli Kedonperä, Sanna Kulmala, Johanna Rihtilä, Senja Räsänen, Aila Siren and Senja Tuominen from Natural Resources Institute Finland (Luke) is greatly acknowledged. We also thank Anneli Virta from Luke Jokioinen for sequencing.

CONFLICT OF INTEREST STATEMENT

The authors declare that they have no conflict of interest.

ORCID

Satu Latvala https://orcid.org/0000-0002-6433-2798
Minna Haapalainen https://orcid.org/0000-0002-2497-4059
Petteri Karisto https://orcid.org/0000-0003-4807-0190
Terhi Suojala-Ahlfors https://orcid.org/0000-0001-7543-870X

REFERENCES

Aalto, J., Pirinen, P., Kauppi, P. E., Rantanen, M., Lussana, C., Lyytikäinen-Saarenmaa, P., & Gregow, H. (2022). High-resolution analysis of observed thermal growing season variability over northern Europe. *Climate Dynamics*, *58*, 1477–1493.

Allain-Boulé, N., Lévesque, C. A., Martinez, C., Bélanger, R. R., & Tweddell, R. J. (2004). Identification of *Pythium* species associated with cavity-spot lesions on carrots in eastern Quebec. *Canadian Journal of Plant Pathology*, *26*, 365–370.

Årsvoll, K. (1969). Pathogens on carrots in Norway. *Meldinger Fra Norges Landbrukshøgskole*, *48*(2), 52.

Azam, A., Khan, I., Mahmood, A., & Hameed, A. (2013). Yield, chemical composition and nutritional quality responses of carrot, radish and turnip to elevated atmospheric carbon dioxide. *Journal of the Science of Food and Agriculture*, *93*, 3237–3244.

Castellá, G., Bragulat, M. R., Rubiales, M. V., & Cabañes, F. J. (1997). Malachite green agar, a new selective medium for *Fusarium* spp. *Mycopathologia*, *137*, 173–178.

Channon, A. G., & Thomson, M. C. (1981). Parsnip canker caused by *Cylindrocarpon* destructans. *Plant Pathology*, *30*, 181.

Choiseul, J., Allen, L., & Carnegie, S. F. (2006). Fungi causing dry tuber rots of seed potatoes in storage in Scotland. *Potato Research*, *49*, 241–253.

Davis, R. M., & Raid, R. N. (Eds.). (2002). *Compendium of umbelliferous crop diseases*. American Phytopathological Society (APS Press).

Fan, X., Zhang, J., Yang, L., & Wu, M. (2015). Development of PCR-based assays for detecting and differentiating three species of *Botrytis* infecting broad bean. *Plant Disease*, *99*, 691–698.

Farrar, J. J., Nunez, J. J., & Davis, R. M. (2000). Influence of soil saturation and temperature on *Erwinia chrysanthemi* soft rot of carrot. *Plant Disease*, *84*, 665–668.

Farrar, J. J., Pryor, B. M., & Davis, R. M. (2004). Alternaria diseases of carrot. *Plant Disease*, *88*(8), 776–784.

Geeson, J. D., Browne, K. M., & Everson, H. P. (1988). Storage diseases of carrots in East Anglia 1978-82, and the effects of some pre- and post-harvest factors. *Annals of Applied Biology*, *112*, 503–514.

Gerlach, W., & Nirenberg, H. (1982). *The genus Fusarium: A pictorial atlas* (Vol. 209, pp. 1–406). Kommissionsverlag P. Parey.

Ghorbani, R., Wilcockson, S., Koocheki, A., & Leifert, C. (2008). Soil management for sustainable crop disease control: A review. *Environmental Chemistry Letters*, *6*, 149–162.

Haapalainen, M., Latvala, S., Kuivainen, E., Qiu, Y., Segerstedt, M., & Hannukkala, A. O. (2016). *Fusarium oxysporum*, *F. proliferatum* and *F. redolens* associated with basal rot of onion in Finland. *Plant Pathology*, *65*, 1310–1320.

Hermansen, A. (1992). Weeds as hosts of *Mycocentrospora acerina*. *Annals of Applied Biology*, *121*, 679–686.

Hermansen, A., & Amundsen, T. (1995). Two methods for the prediction of *Mycocentrospora acerina* infection on stored carrots. *Annals of Applied Biology*, *126*, 217–233.

Hermansen, A., Amundsen, T., Taksdal, G., Dragland, S., Synnevåg, G., Flønes, M., & Sundheim, L. (1997). *Mycocentrospora acerina* in carrots; effects of crop rotation on disease incidence. *Annals of Applied Biology*, *131*, 399–411.

Hermansen, A., Wanner, L., Naerstad, R., & Klemsdal, S. S. (2012). Detection and prediction of post harvest carrot diseases. *European Journal of Plant Pathology*, *133*, 211–228.

Hiltunen, L. H., & White, J. G. (2002). Cavity spot of carrot (*Daucus carota*). *Annals of Applied Biology*, *141*, 201–223.

Howard, R. J., Pratt, R. G., & Williams, P. H. (1978). Pathogenicity to carrots of *Pythium* species from organic soils of North America. *Phytopathology*, *68*, 1293–1296.

Ibrahim, M. A., Nissinen, A., Prozherina, N., Oksanen, E., & Holopainen, J. (2006). The influence of exogenous monoterpene treatment and elevated temperature on growth, physiology, chemical content and headspace volatiles of two carrot cultivars (*Daucus carota* L.). *Environmental and Experimental Botany*, *56*, 95–107.

Kahala, M., Blasco, L. T., & Joutsjoki, V. (2012). Molecular characterization of spoilage bacteria as a means to observe the microbiological quality of carrot. *Journal of Food Production*, *75*, 523–532.

Kastelein, P., Stilma, E. S. C., Elderson, J., & Köhl, J. (2007). Occurrence of *Rhexocercosporidium carotae* on cold stored carrot roots in The Netherlands. *European Journal of Plant Pathology*, *117*, 293–305.

Kora, C., McDonald, M. R., & Boland, G. J. (2005a). Occurrence of fungal pathogens of carrots on wooden boxes used for storage. *Plant Pathology*, *54*, 665–670.

Kora, C., McDonald, M. R., & Boland, G. J. (2005b). Epidemiology of sclerotinia rot of carrot caused by *Sclerotinia sclerotiorum*. *Canadian Journal of Plant Pathology*, *27*(2), 245–258.

Lampert, Y., Dror, B., Sela, N., Teper-Bamnolker, P., Daus, A., Sela (Saldinger), S., & Eshel, D. (2017). Emergence of *Leuconostoc mesenteroides* as a causative agent of oozing in carrots stored under non-ventilated conditions. *Microbial Biotechnology*, *10*(6), 1677–1689.

Le Moullec-Rieu, T., Chateau, C., Pascouau, C., Bastide, F., Hamon, B., Poupard, P., Laurent, E., Gombert, J., Morel, E., Sérandat, I., Guillermin, P.-L., Brandeis, P.-E., & Mallard, S. (2020). First report of root and collar rot caused by *Fusarium tricinctum* and *Fusarium avenaceum* on carrot in France. *Plant Disease*, *104*(2), 591.

Li, H., Chen, Y., Zhang, Z., Li, B., Qin, G., & Tian, S. (2018). Pathogenic mechanisms and control strategies of *Botrytis cinerea* causing post-harvest decay in fruits and vegetables. *Food Quality and Safety*, *3*, 111–119.

Li, F., Jiang, X., Sun, M., Xu, H.-I., Shi, L., Shan, W., & Feng, Y. (2014). Screening of culture conditions for pathogens of potato dry rot. *Acta Agriculturae Scandinavica*, *Section B—Soil & Plant Science*, *64*(8), 694–699.

Mishra, P. K., Fox, R. T. V., & Culham, A. (2003). Development of a PCR-based assay for rapid and reliable identification of pathogenic Fusaria. *FEMS Microbiology Letters*, *218*, 329–332.

Mohamad, R. A. M. (2021). *Identification of fungi that cause tip rot of carrot and determine effect of storage temperature and cultivar on tip rot development*. [MSc thesis. Faculty of Biosciences, Norwegian University of Life Sciences, Ås, Norway. 55 pp].

Moore, F. J. (1945). A comparison of *Fusarium avenaceum* and *Fusarium caeruleum* as causes of wastage in stored potato tubers. *Annals of Applied Biology*, *32*(4), 304–309.

Mukula, J. (1957). On the decay of stored carrots in Finland. *Acta Agriculturae Scandinaviae*, *Suppl. 2*, 132 Stockholm.

Nel, W. J., Duong, T. A., de Beer, Z. W., & Wingfield, M. J. (2019). Black root rot: A long known but little understood disease. *Plant Pathology*, *68*, 834–842.

Nelson, P. E., Toussoun, T. A., & Marasas, W. F. O. (1983). *Fusarium species: An illustrated manual for identification*. Pennsylvania State University Press.

Papoutsis, K., & Edelenbos, M. (2021). Postharvest environmentally and human-friendly pre-treatments to minimize carrot waste in the supply chain caused by physiological disorders and fungi. *Trends in Food Science & Technology*, *112*, 88–98.

Parikka, P. (2008). Porkkanan taudit. [Carrot diseases]. *Porkkanan kuluttajalaadun parantaminen. [Improving consumer quality of carrot]. Maa- ja elintarviketalous*, 128, pp. 48–54. Ed. P. Vanhala. Maa-ja elintarviketalouden

tutkimuskeskus. ISBN 978-952-487-185-3. http://urn.fi/URN:ISBN:978-952-487-185-3 [in Finnish]

Pascouau, C., Chateau, C., Bastide, F., le Moullecrieu, T., Guillemette, T., Hamon, B., Aligon, S., Cailleau, A., Sochard, D., Gombert, J., Morel, E., Laurent, E., Sérandat, I., Berruyer, R., & Poupard, P. (2023). Characterization and pathogenicity of *Fusarium* spp. isolates causing root and collar rot on carrot. *Canadian Journal of Plant Pathology*, *45*(1), 76–91.

Snowdon, S. (1992). *Color atlas of post-harvest diseases & disorders of fruits & vegetables* (Vol. 2, p. 416). BPCC Hazell Books. ISBN 0-8493-7734-X.

Stanković, I., Milojević, K., Vučurović, A., Nikolić, D., Krstić, B., & Bulajić, A. (2015). First report of Fusarium root rot of stored carrot caused by *Fusarium avenaceum* in Serbia. *Plant Disease*, *99*(2), 286.

Suojala, T. (1999). Effect of harvest time on the storage performance of carrot. *Journal of Horticultural Science & Biotechnology*, *74*, 484–492.

Suojala-Ahlfors, T., & Laamanen, T.-L. (2014). Effect of calcium amendment on the calcium content and storage quality of carrot (*Daucus carota* L.). *European Journal of Horticultural Science*, *79*, 278–282.

Sweetingham, M. W. (1983). *Studies on the nature of the pathogenicity of soil-borne* Cylindrocarpon *species*. [PhD thesis, University of Tasmania]. https://eprints.utas.edu.au/3136/

Tahvonen, R. (1985). The prevention of *Botrytis cinerea* and *Sclerotinia sclerotiorum* on carrots during storage by spraying the tops with fungicide before harvesting. *Annales Agriculturae Fenniae*, *24*, 89–95.

Tahvonen, R. (1989). Porkkanan taudit ongelmana. *Koetoiminta Ja Käytäntö*, *46*, 7 [in Finnish].

Vanhala, P., Kallela, M., Pitkänen, T., & Suojala-Ahlfors, T. (2008). Porkkanan varastokestävyys ja viljelytekijät [Storage quality and cultivation practices of carrot]. *Porkkanan kuluttajalaadun parantaminen. [Improving consumer quality of carrot]. Maa- ja elintarviketalous*, 128, pp. 36–47. Ed. P. Vanhala. Maa- ja elintarviketalouden tutkimuskeskus. ISBN 978-952-487-185-3. http://urn.fi/URN:ISBN:978-952-487-185-3 [in Finnish]

White, T. J., Bruns, T., Lee, S., & Taylor, J. (1990). Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In *PCR protocols: A guide to methods and applications* (Vol. 18, pp. 315–322). Academic Press Inc.

Yang, K., Wang, H. L., Ye, C., Wang, Z. H., Ye, K. H., Zhang, S., Huang, H. P., Wei, Z. X., Zhu, S. S., Zhu, Y. Y., Guo, L. W., & He, X. H. (2022). Infection characteristics and physical prevention strategy of *Panax notoginseng* round spot disease caused by *Mycocentrospora acerina*. *Plant Disease*, *106*, 2607–2617.

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Latvala, S., Haapalainen, M., Karisto, P., Kivijärvi, P., Jääskeläinen, O., & Suojala-Ahlfors, T. (2024). Changes in the prevalence of fungal species causing post-harvest diseases of carrot in Finland. *Annals of Applied Biology*, 1–13. https://doi.org/10.1111/aab.12908