Grower Summary

FV 382a

Internal browning of carrot: investigating a link with the viral diseases PYFV and CMD

Final 2013

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Before using all pesticides check the approval status and conditions of use.

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Further information

If you would like a copy of the full report, please email the HDC office (hdc@hdc.ahdb.org.uk), quoting your HDC number, alternatively contact the HDC at the address below.

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Headline

Internal necrosis of carrot is strongly linked to the aphid-transmitted Carrot Yellow Leaf Virus. This virus was first reported in the UK in 1979, but has not been previously linked to root symptoms. Specific diagnostic tests have been developed for this virus and other carrot viruses.

Background

The principal viruses affecting carrot crops in the UK are Parsnip Yellow Fleck Virus (PYFV) and the Carrot Motley Dwarf (CMD) complex consisting of Carrot Red Leaf Virus (CtRLV), Carrot Mottle Virus (CMoV) and Carrot Red Leaf-associated RNA (CtRLVaRNA). These viruses affect crops sporadically, but when they occur they can have devastating consequences.

The presence of viruses has been associated with internal necrosis around the root core extending from crown to tip; this has been observed in carrot crops for at least the last 10 years. The 2009 growing season saw some growers losing up to 10% of yield because of these symptoms. As the symptoms tend to be internal there are obvious practical issues in grading out affected carrots. For carrots destined for processing, this will lead to rejection of total consignments at processors and pack-houses.

Results of a limited survey in 2010 (HDC project FV 382, Fox, 2011) suggested a possible weak link between the presence of internal browning/root necrosis symptoms with viruses for the CMD complex. Detection of these viruses is carried out using conventional polymerase chain reaction (PCR) methods from assays developed following HDC project FV 228a (Morgan, 2004). Development of a suite of real-time RT-PCR diagnostic tests would bring advantages in sensitivity and turnaround time. In addition, the amenability of real-time PCR to automation would allow high throughput testing methods to be used, making epidemiological studies practical and affordable. Assays developed within this project have been used to ascertain which viruses are present within symptomatic carrots and to detect the presence of multiple viruses in carrot crops.

Traditionally, linking a pathogen with an expressed symptom requires demonstrating ‘Koch’s Postulates’, where a pathogen is isolated from a symptomatic host and then back-inoculated into the original host species to induce the original symptom. Where a complex of viruses may be affecting a host, or where there may be environmental or agronomic influences on symptom development (e.g. temperature, moisture, time from exposure, time in ground/crop

growth stage, etc.), trying to artificially induce symptoms can be challenging. A statistical model has been developed to establish a link based upon testing similar numbers of affected and unaffected carrots for the presence of multiple pathogens. We believe this model to have broad applications in plant pathology, where infection and disease are often anecdotally linked but lack statistical support.

Traditional diagnostic methods are targeted towards specific known pathogens. By using a metagenomic approach using novel sequencing technology, other possible causes of internal browning can be identified. This technology has already been used to describe previously unknown plant viruses such as Gayfeather Mild Mottle Virus (Adams *et al.,* 2009*)* and Watercress White Vein Virus (Harju *et al.*, 2012). This would ascertain whether other viruses/pathogens were present in necrosis-affected carrots.

Summary of the project and key findings

Carrot samples were collected from a crop exhibiting 3% necrosis symptoms. Approximately equal numbers of affected and unaffected carrots were included in the sample (102 affected: 99 unaffected). These samples were tested for the presence of the established carrot viruses Carrot Red Leaf Virus (CtRLV)*,* Carrot Mottle Virus (CMoV)*,* Carrot Red Leaf-associated Virus (CtRLVaRNA) and Parsnip Yellow Fleck Virus (PYFV). The presence of these viruses did not correlate to symptomatic carrot roots, either as single viruses or in combinations. Field weeds from around fields known to be associated with viral necrosis were tested, but few positive results were obtained for these established carrot viruses.

A sub-sample of 12 affected and 12 unaffected carrots of mixed virus status was subjected to non-targeted analysis (next-generation sequencing; NGS). The results from these tests showed an unexpected virus – Carrot Yellow Leaf Virus (CYLV) – to be associated to the presence of internal carrot necrosis in the sub-sample. A virus closely related to CYLV was also found to be present in the non-CYLV-infected symptomatic roots, but due to the low incidence of this virus in the sub-sample, no meaningful conclusions can be drawn on the importance of this finding. Additionally, several other novel plant viruses were found within the sub-sampled carrots. These were present in low numbers and did not appear to bear any relationship to the presence of necrosis in carrot roots.

Because of the high prevalence of CYLV in the sub-sampled symptomatic carrots, a specific diagnostic test was designed and the main field sample was tested for this virus. Of the affected (symptomatic) carrots, 98% were positive for CYLV, and of the unaffected (symptomless) carrots 22% were found to be positive for CYLV. This virus had the closest

association with necrosis across all carrots tested, either as a single virus, or in combination with other viruses. Because necrosis without the presence of CYLV is estimated to be rare in the original sampled population, if CYLV is the causative agent of necrosis, removal of this pathogen is estimated to potentially reduce necrosis by 96%. We believe this to be the first report to link CYLV to root necrosis.

Financial benefits

Unlike external root symptoms traditionally associated with PYFV (e.g. cigar shaped roots), internal necrosis cannot easily be graded out. However:

(i) The primary causal pathogen is now known. A better understanding of the relationship between virus infection and internal browning should lead to a reduced incidence of these symptoms through improved management strategies.

(ii) This, in turn, should lead to reduced waste from the industry both through reducing numbers of carrots rejected on the grading line and numbers of crops rejected at processors and pack-houses.

Advanced diagnostic assays have been designed and will be offered as a diagnostic service, helping UK carrot growers to save money through reduced waste and rejected produce. Additionally, the use of non-targeted diagnostic approaches to plant pathology applications have been successfully demonstrated and will hopefully be further developed and incorporated into a future diagnostic service; this will potentially reduce waste and further increase growers' income.

Action points

- CYLV can be transmitted by both willow-carrot aphid and parsnip aphid; growers need to monitor both species. At present, only willow-carrot aphid is shown on Rothamsted and HDC Pest Bulletin weekly reports.

- CYLV is carried semi-persistently in the aphid and is retained within an infectious individual for several days. Unlike PYFV, the virus can recycle within carrots leading to sequential transmission. Control programmes need to be extended even when low numbers of aphids are found, which may mean continuation of insecticide treatments beyond May and June (usually the peak time for willow-carrot aphid) and into July, August, September and even October.

- Yellow traps are the best approach to monitor local carrot aphid populations (both species) for farms in order to assess when programmes/treatments need to be started, maintained or stopped.