Yield losses in blackleg of canola and pyraclostrobin sensitivity in populations of *Leptosphaeria maculans*
Introduction

• Canola is one of the world's most important oilseed crops.

• Blackleg disease, caused by *Leptosphaeria maculans*, is an important disease in Canada and worldwide.
Symptoms

- Basal stem cankers, lesions on leaves, and root rot
(1 & 2) Ascospores infect canola seedlings. (3) Greyish lesions develop on leaves. Pycnidiospores are released. (4) The fungus grows inside the vascular tissues. (5 & 6) Stem cankers develop, leading to girdling and lodging. (7) Infected pods and seeds may give rise to infected seedlings.

(Fraser et al., 2016)
Introduction

• In Canada, blackleg has been managed largely by the deployment of resistant cultivars.

• To establish the relationship between blackleg disease severity and yield of canola hybrids in western Canada is necessary.
Materials and Methods

• The blackleg moderately resistant cultivars ‘1950RR’ and ‘73-15RR’ were planted in two inoculated research plots near Edmonton, Alberta, in 2017 and 2018.

• A 1-m² area of each plot was hand-harvested.

• The plants were sorted according to disease severity on a 0-5 scale.

• Pod numbers and seed yield per plant were recorded and calculated.
The mean disease severity over two site-years

P = 0.3163
Average pods number of 73-15RR

\[ y = -6.977x^2 + 7.1194x + 158.91 \]
\[ R^2 = 0.9758 \]

Average pods number of 1950RR

\[ y = -9.9525x^2 + 16.106x + 186.78 \]
\[ R^2 = 0.9446 \]
Seed yield per plant of 73-15RR

\[ y = -0.559x^2 + 0.4369x + 11.995 \]

\[ R^2 = 0.9629 \]

Seed yield per plant of 1950RR

\[ y = -0.5265x^2 - 0.1836x + 13.947 \]

\[ R^2 = 0.9591 \]
Relationship between blackleg severity and yield loss in the canola hybrids “73-15RR” and “1950RR”

yield losses of “73-15RR”: $y = 5.3784x^2 - 9.0118x + 0.4464; R^2 = 0.8064$

yield losses of "1950RR": $y = 3.2262x^2 + 2.1284x - 5.9863; R^2 = 0.8669$
Conclusion

• Canola plants with very mild symptoms of blackleg do not suffer negative effects on seed yield per plant.

• However, even in moderately resistant hybrid canola cultivars, seed yield per plant decreases greatly as a result of severe *L. maculans* infection.

• Therefore, alternative disease management strategies are required to mitigate yield losses due to blackleg.
Fungicide Sensitivity

• Pyraclostrobin is a strobilurin fungicide, also known as a quinone outside inhibitor (QoI) (Balba, 2007).

• The strobilurin fungicide is single-site mode-of-action fungicide (Bartlett 2002), inhibiting mitochondrial respiration (Gisi et al. 2002).

• Pyraclostrobin has been used on various crops since 2003 as the foliar treatment Headline EC (BASF Canada Inc., Mississauga, On).
Previous study

• In 2011, 13 isolates of *L. maculans* were obtained from infected canola stubble from five fields in Alberta were used to determine the effective concentration of pyraclostrobin needed to inhibit mycelial growth by 50% (EC$_{50}$), which serve as the sensitivity baseline (Fraser et al., 2017).

• With the conventional mycelial growth plate assay, the mean EC$_{50}$ for 13 isolates was 0.09 mg L$^{-1}$ (Fraser et al., 2017).
Conventional agar plate

• 33 isolates collected from eighteen counties in Alberta in 2016 were used to determine EC$_{50}$ as well as 12 baseline isolates from 2011.

• Ten treatments were arranged in a completely randomized design, with five replicates per isolates.

• The mean EC$_{50}$ for 2011 isolates was 0.07 mg L$^{-1}$. A two-sided pairwise t-test was used to compare isolates results in 2017 and 2011 (p = 0.3305).
• The mean EC$_{50}$ for 2016 isolates was 0.28 mg L$^{-1}$.

• Two discriminatory doses, 1X EC$_{50}$ of 2016 isolates (0.28 mg L$^{-1}$) and 50X EC$_{50}$ of 2011 baseline isolates (3.5 mg L$^{-1}$), were used to examine the fungicide sensitivity of 251 isolates were obtained in 2016.

• The treatments were arranged in a completely randomized design, with four replicates per isolates.
Frequency distribution of the inhibition of mycelial growth in 251 *L. maculans* isolates in response to inclusion of pyraclostrobin (0.28 mg L$^{-1}$) in V8 growth medium.
Frequency distribution of the inhibition of mycelial growth in 251 L. maculans isolates in response to inclusion of pyraclostrobin (3.5 mg L$^{-1}$) in V8 growth medium.
Microtiter bioassay

• 38 isolates collected in 2016 and 12 baseline isolates from 2011 were used to determine EC_{50}.

• Twelve treatments were arranged in a completely randomized design, with four replicates per isolates.

• The mean EC_{50} for 2011 isolates was 0.0015 mg L^{-1}.
• The mean EC_{50} for 2016 isolates was 0.0049 mg L^{-1}.

• Two discriminatory doses, 4X EC_{50} of 2011 isolates (0.006 mg L^{-1}) and 50X EC_{50} of 2011 baseline isolates (0.075 mg L^{-1}), were used examine the fungicide sensitivity of 251 isolates were obtained in 2016.
Frequency distribution of the inhibition of mycelial growth in 251 *L. maculans* isolates in response to inclusion of pyraclostrobin (0.006 mg L⁻¹) in YBA liquid medium.
Frequency distribution of the inhibition of mycelial growth in 251 *L. maculans* isolates in response to inclusion of pyraclostrobin (0.075 mg L\(^{-1}\)) in YBA liquid medium.
Conclusion

• The result suggests an increased insensitivity among the *L. maculans* isolates over time while all isolates were still sensitive to high dose of pyraclostrobin.

• It is essential for future monitoring of changes in pyraclostrobin sensitivity and proper fungicide stewardship is warranted.
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