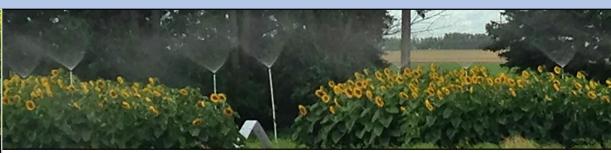


Preliminary testing – Oilseed sunflowers, bumble-vectored Clonostachys rosea

Non-replicated studies. Sunflowers exposed to bees were spatially separated from identically managed sunflowers not exposed to bees.

Langdon, ND (2016, 2017)





Langdon 2016 NuSun '306' Langdon 2017 NuSun '306'

Sclerotinia head rot incidence (% of plants)

no bees 39

exposed to bees 26

Sunflowers were inoculated twice:

- Once at approx. R5.4-R5.6
- Once at approx. R5.5-R5.9

To each head, 15,000 ascospores were applied per head per inoculation (delivered with a hand-held spray bottle calibrated to deliver 5,000 spores per spray).

Sunflower yield (pounds/acre)

bagged heads 1880

unbagged heads 2053

1981

Preliminary testing – Non-oil sunflowers, honeybee-vectored *Clonostachys rosea*

Replicated studies (4-5 reps). Bees were excluded from sunflower heads in the non-treated control by placing perforated pollination bags over heads. Heads were bagged from bloom initiation to R7. **Carrington, ND (2017, 2019)**





Carrington 2018
NuSeed 'Jaguar'

Carrington 2019 NuSeed 'Jaguar'

Sclerotinia head rot incidence (% of plants)

bagged heads 70 b 93 a unbagged heads 34 a 69 a CV: 16.0 CV: 9.4

INOCULATIONS:

- Sunflowers were inoculated twice in 2018 (at R5.5 and R5.8-R5.9) and once in 2019 (at R5.7-R5.9).
- To each head, 15,000 ascospores were applied per head per inoculation (delivered with hand-held spray bottle calibrated to deliver 5,000 spores / spray).

POLLINATION BAGS:

18 x 16 inch (length x width) pollination bags made of fine mesh fabric with 1 mm x 1 mm holes (Lawson Bags; Northfield, IL)

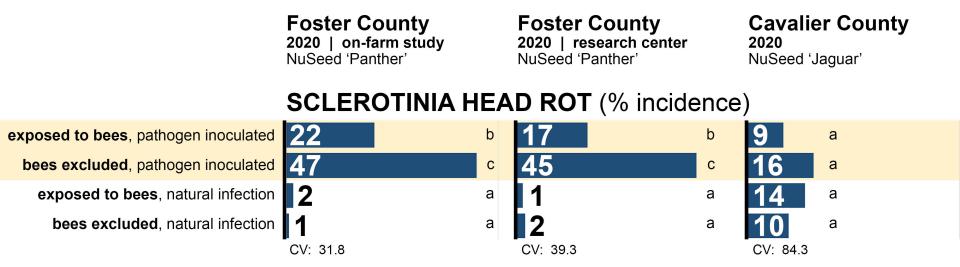
Sunflower yield (pounds/acre)

bagged heads 635
unbagged heads 1607

NO DATA

Replicated studies (3 reps). Sunflowers were established in a strip 60 to 110 feet wide by half-mile long. Bee hives were placed at one end and two-thirds the distance along the strip.

Year #1: Foster and Cavalier Counties, ND (2020)



PLOT SIZE: Approx. 100 plants/plot across 3 or 4 rows. HIVES: 4 hives at each location, 100% of hives equipped with dispensers of the biological control agent at studies conducted in Foster County; 25% of hives equipped with dispensers at the study conducted in Cavalier County.

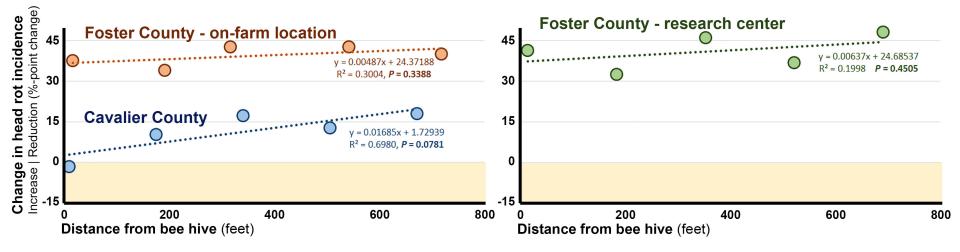
INOCULATIONS: Sunflowers were inoculated once at approx. R5.4-R5.8. To each head, 15,000 ascospores were applied per head.

Replicated studies (3 reps). Sunflowers were established in a strip 60 to 110 feet wide by half-mile long. Bee hives were placed at one end and two-thirds the distance along the strip.

Year #1: Foster and Cavalier Counties, ND (2020)

Impact of bee-vectored *C. rosea* relative to distance from bee hive: reduction in Sclerotinia head rot incidence

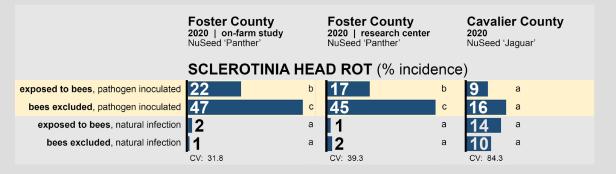
Sunflowers inoculated with laboratory-grown ascospores of *S. sclerotiorum*. Honeybees excluded from sunflowers in the control treatment with perforated pollination bags



PLOT SIZE: ~100 plants/plot across 3 or 4 rows.

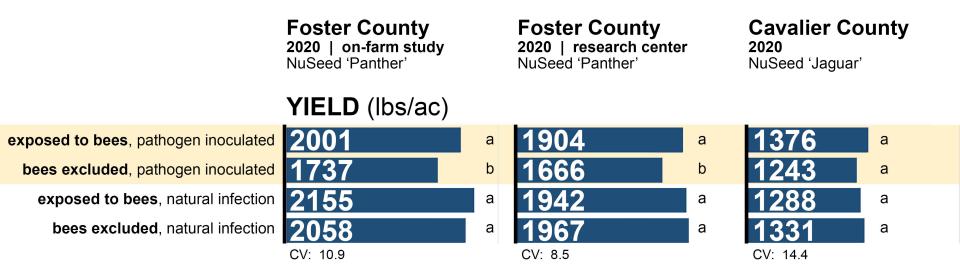
HIVES: 4 hives at each location, 100% of hives equipped with dispensers of the biological control agent at studies conducted in Foster County; 25% of hives equipped with dispensers at the study conducted in Cavalier County.

INOCULATIONS: Sunflowers were inoculated once at approx. R5.4-R5.8. To each head, 15,000 ascospores were applied per head.



Replicated studies (3 reps). Sunflowers were established in a strip 60 to 110 feet wide by half-mile long. Bee hives were placed at one end and two-thirds the distance along the strip.

Year #1: Foster and Cavalier Counties, ND (2020)



PLOT SIZE: Approx. 100 plants/plot across 3 or 4 rows. HIVES: 4 hives at each location, 100% of hives equipped with dispensers of the biological control agent at studies conducted in Foster County; 25% of hives equipped with dispensers at the study conducted in Cavalier County.

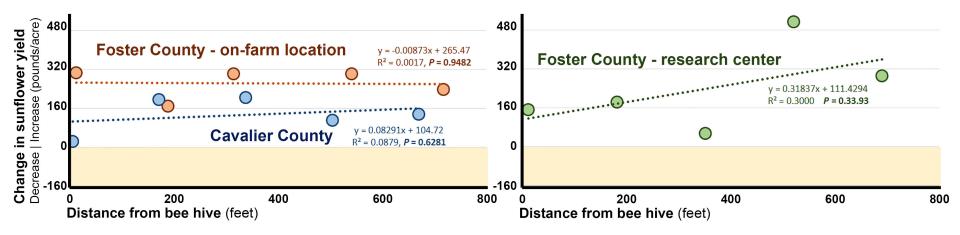
INOCULATIONS: Sunflowers were inoculated once at approx. R5.4-R5.8. To each head, 15,000 ascospores were applied per head.

Replicated studies (3 reps). Sunflowers were established in a strip 60 to 110 feet wide by half-mile long. Bee hives were placed at one end and two-thirds the distance along the strip.

Year #1: Foster and Cavalier Counties, ND (2020)

Impact of bee-vectored *C. rosea* relative to distance from bee hive: increase in sunflower yield (lbs/ac)

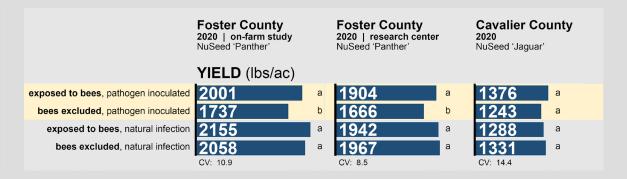
Sunflowers inoculated with laboratory-grown ascospores of *S. sclerotiorum*. Honeybees excluded from sunflowers in the control treatment with perforated pollination bags



PLOT SIZE: ~100 plants/plot across 3 or 4 rows.

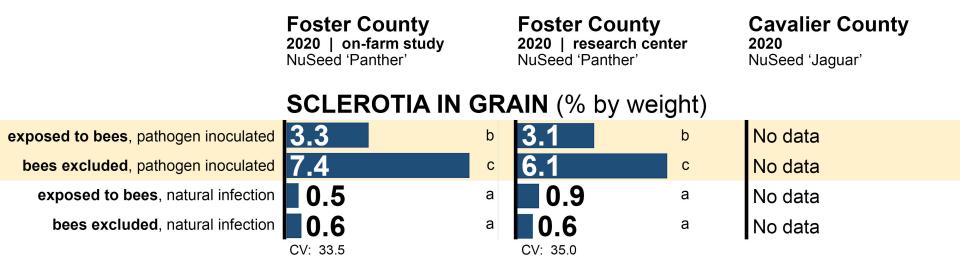
HIVES: 4 hives at each location, 100% of hives equipped with dispensers of the biological control agent at studies conducted in Foster County; 25% of hives equipped with dispensers at the study conducted in Cavalier County.

INOCULATIONS: Sunflowers were inoculated once at approx. R5.4-R5.8. To each head, 15,000 ascospores were applied per head.



Replicated studies (3 reps). Sunflowers were established in a strip 60 to 110 feet wide by half-mile long. Bee hives were placed at one end and two-thirds the distance along the strip.

Year #1: Foster and Cavalier Counties, ND (2020)



PLOT SIZE: Approx. 100 plants/plot across 3 or 4 rows. HIVES: 4 hives at each location, 100% of hives equipped with dispensers of the biological control agent at studies conducted in Foster County; 25% of hives equipped with dispensers at the study conducted in Cavalier County.

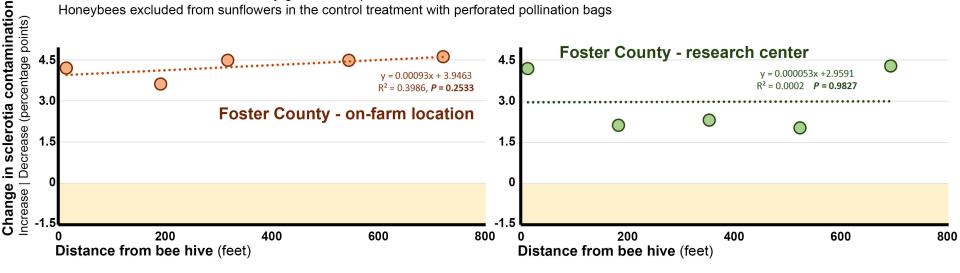
INOCULATIONS: Sunflowers were inoculated once at approx. R5.4-R5.8. To each head, 15,000 ascospores were applied per head.

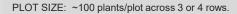
Replicated studies (3 reps). Sunflowers were established in a strip 60 to 110 feet wide by half-mile long. Bee hives were placed at one end and two-thirds the distance along the strip.

Year #1: Foster and Cavalier Counties, ND (2020)

Impact of bee-vectored *C. rosea* relative to distance from bee hive: reduction in sclerotia contamination of the grain (percent by weight)

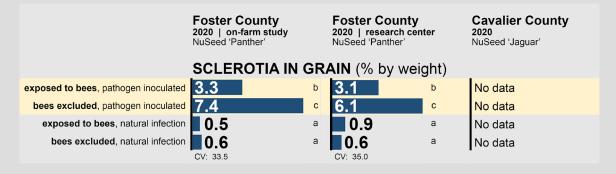
Sunflowers inoculated with laboratory-grown ascospores of *S. sclerotiorum*. Honeybees excluded from sunflowers in the control treatment with perforated pollination bags





HIVES: 4 hives at each location, 100% of hives equipped with dispensers of the biological control agent at studies conducted in Foster County; 25% of hives equipped with dispensers at the study conducted in Cavalier County.

INOCULATIONS: Sunflowers were inoculated once at approx. R5.4-R5.8. To each head, 15,000 ascospores were applied per head.



Replicated studies (3 reps). Sunflowers were established in a strip 60 to 110 feet wide by half-mile long. Bee hives were placed at one end and two-thirds the distance along the strip. **Year #2: Foster, Wells and Cavalier Counties, ND (2021)**

Facilitating Sclerotinia head rot pressure under severe drought:

- Water was hauled to site in 1200-gal tank and pumped through mist system.
- Sunflowers inoculated in morning.
- Irrigation delivered continuously until dark.



Replicated studies (3 reps). Sunflowers were established in a strip 60 to 110 feet wide by half-mile long. Bee hives were placed at one end and two-thirds the distance along the strip. **Year #2: Foster, Wells and Cavalier Counties, ND (2021)**

Wells County 2021 NuSeed 'Panther' Foster County 2021
NuSeed 'Panther'

Cavalier County 2021 NuSeed 'LD5009'

SCLEROTINIA HEAD ROT (% incidence)

exposed to bees, pathogen inoculated
bees excluded, pathogen inoculated
exposed to bees, natural infection
bees excluded, natural infection

			<u>′</u>	
23	b	4 b	27	b
70	С	8 b	41	b
0	а	0 a	4	а
0	а	0 a	0	а
CV: 12.9	'	CV: 64.0	CV: 32.7	



PLOT SIZE: Approx. 100 plants/plot across 3 or 4 rows.

HIVES: 4 or 6 hives at each location, 50 to 100% of hives equipped with dispensers of the biological control.

INOCULATIONS: Sunflowers were inoculated once at approx. R5.4-R5.9. To each head, 15,000 ascospores were applied per head.

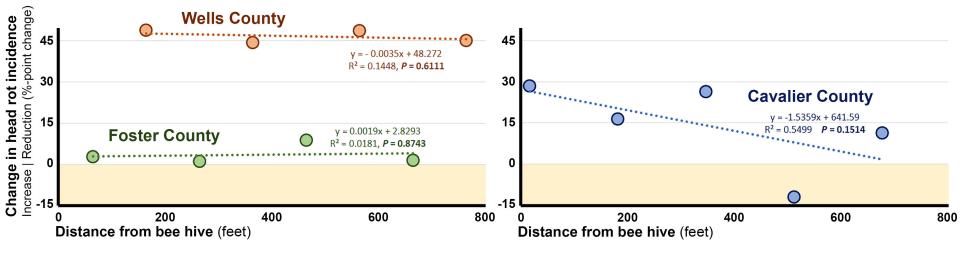
POLLINATION BAGS: Rep 3 of Wells County study: 18 x 16 inch (length x width), fine mesh fabric with 1 mm x 1 mm holes (Lawson Bags; Northfield, IL)

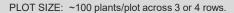
Remainder of studies: 16x18" (40.64 x 45.72 cm) HDPE plastic with 336-micron pore diameter; 24% of surface open (Midco Global; Kirkwood, MO)

Replicated studies (3 reps). Sunflowers were established in a strip 60 to 110 feet wide by half-mile long. Bee hives were placed at one end and two-thirds the distance along the strip. **Year #2: Foster, Wells and Cavalier Counties, ND (2021)**

Impact of bee-vectored *C. rosea* relative to distance from bee hive: reduction in Sclerotinia head rot incidence

Sunflowers inoculated with laboratory-grown ascospores of *S. sclerotiorum*. Honeybees excluded from sunflowers in the control treatment with perforated pollination bags.





HIVES: 4 or 6 hives at each location, 50 to 100% of hives equipped with dispensers of the biological control.

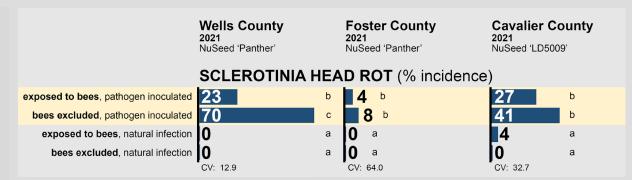
INOCULATIONS: Sunflowers were inoculated once at approx. R5.4-R5.8. To each head, 15,000 ascospores were applied per head.

POLLINATION BAGS:

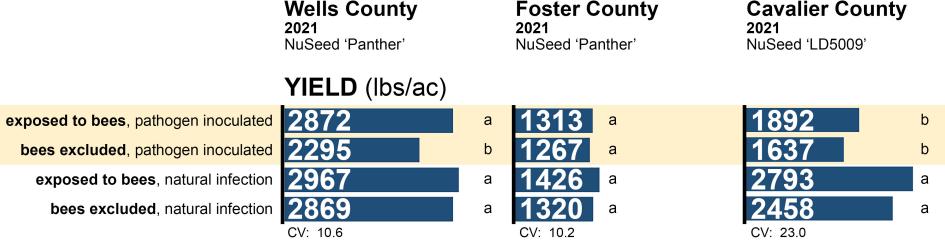
Rep 3 of Wells County 2021: 18 x 16 inch (length x width), fine mesh fabric with 1 mm x 1 mm holes (Lawson Bags; Northfield, IL)

Remainder of studies: 16x18" (40.64 x 45.72 cm) HDPE plastic with

Remainder of studies: 16x18" (40.64 x 45.72 cm) HDPE plastic wit 336-micron pore diameter; 24% of the surface open (Midco Global; Kirkwood, MO)



Replicated studies (3 reps). Sunflowers were established in a strip 60 to 110 feet wide by half-mile long. Bee hives were placed at one end and two-thirds the distance along the strip. **Year #2: Foster, Wells and Cavalier Counties, ND (2021)**





PLOT SIZE: Approx. 100 plants/plot across 3 or 4 rows.

HIVES: 4 or 6 hives at each location, 50 to 100% of hives equipped with dispensers of the biological control.

INOCULATIONS: Sunflowers were inoculated once at approx. R5.4-R5.9. To each head, 15,000 ascospores were applied per head.

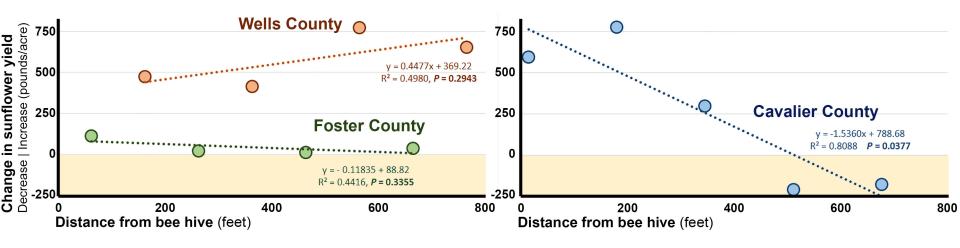
POLLINATION BAGS: Rep 3 of Wells County study: 18 x 16 inch (length x width), fine mesh fabric with 1 mm x 1 mm holes (Lawson Bags; Northfield, IL)

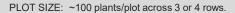
Remainder of studies: 16x18" (40.64 x 45.72 cm) HDPE plastic with 336-micron pore diameter; 24% of surface open (Midco Global; Kirkwood, MO)

Replicated studies (3 reps). Sunflowers were established in a strip 60 to 110 feet wide by half-mile long. Bee hives were placed at one end and two-thirds the distance along the strip. **Year #2: Foster, Wells and Cavalier Counties, ND (2021)**

Impact of bee-vectored *C. rosea* relative to distance from bee hive: **change in sunflower yield (lbs/ac)**

Sunflowers inoculated with laboratory-grown ascospores of *S. sclerotiorum*. Honeybees excluded from sunflowers in the control treatment with perforated pollination bags





HIVES: 4 or 6 hives at each location, 50 to 100% of hives equipped with dispensers of the biological control.

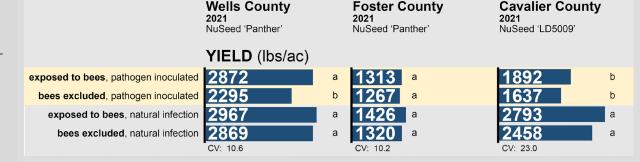
INOCULATIONS: Sunflowers were inoculated once at approx. R5.4-R5.8. To each head, 15,000 ascospores were applied per head.

POLLINATION BAGS:

Rep 3 of Wells County 2021: 18 x 16 inch (length x width), fine mesh fabric with 1 mm x 1 mm holes (Lawson Bags; Northfield, IL)

Remainder of studies: 16x18" (40.64 x 45.72 cm) HDPE plastic with

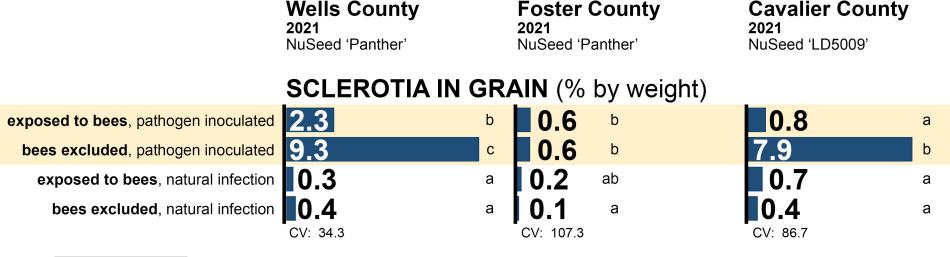
336-micron pore diameter; 24% of the surface open (Midco Global; Kirkwood, MO)



Replicated studies (3 reps). Sunflowers were established in a strip 60 to 110 feet wide by half-mile long. Bee hives were placed at one end and two-thirds the distance along the strip. Year #2: Foster, Wells and Cavalier Counties, ND (2021)

Foster County

Cavalier County





PLOT SIZE: Approx. 100 plants/plot across 3 or 4 rows.

HIVES: 4 or 6 hives at each location, 50 to 100% of hives equipped with dispensers of the biological control.

INOCULATIONS: Sunflowers were inoculated once at approx. R5.4-R5.9. To each head, 15,000 ascospores were applied per head.

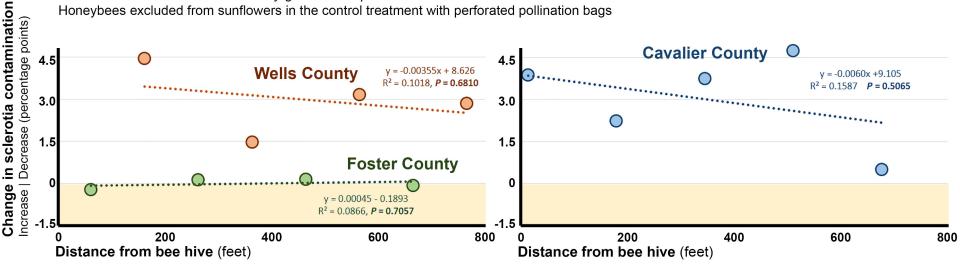
Rep 3 of Wells County study: 18 x 16 inch (length x width), fine mesh fabric with 1 mm x 1 mm holes (Lawson Bags; Northfield, IL) POLLINATION BAGS:

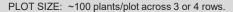
Remainder of studies: 16x18" (40.64 x 45.72 cm) HDPE plastic with 336-micron pore diameter; 24% of surface open (Midco Global; Kirkwood, MO)

Replicated studies (3 reps). Sunflowers were established in a strip 60 to 110 feet wide by half-mile long. Bee hives were placed at one end and two-thirds the distance along the strip. Year #2: Foster, Wells and Cavalier Counties, ND (2021)

Impact of bee-vectored *C. rosea* relative to distance from bee hive: reduction in sclerotia contamination of the grain (percent by weight)

Sunflowers inoculated with laboratory-grown ascospores of S. sclerotiorum. Honeybees excluded from sunflowers in the control treatment with perforated pollination bags



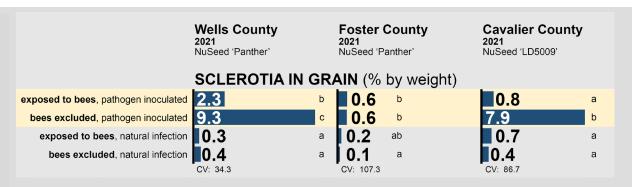


HIVES: 4 or 6 hives at each location, 50 to 100% of hives equipped with dispensers of the biological control.

INOCULATIONS: Sunflowers were inoculated once at approx. R5.4-R5.8. To each head, 15,000 ascospores were applied per head

POLLINATION BAGS:

Rep 3 of Wells County 2021: 18 x 16 inch (length x width), fine mesh fabric with 1 mm x 1 mm holes (Lawson Bags; Northfield, IL) Remainder of studies: 16x18" (40.64 x 45.72 cm) HDPE plastic with 336-micron pore diameter; 24% of the surface open (Midco Global; Kirkwood, MO)



Efficacy of honeybee-vectored *Clonostachys rosea* relative to sunflower hybrid

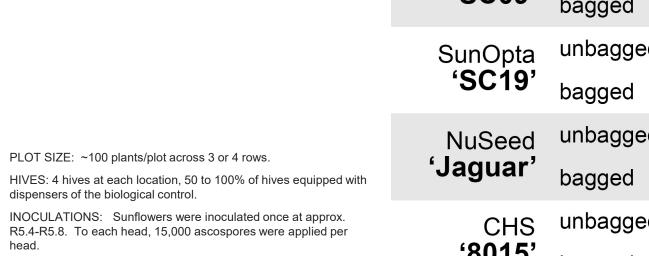
Replicated studies (3 reps).

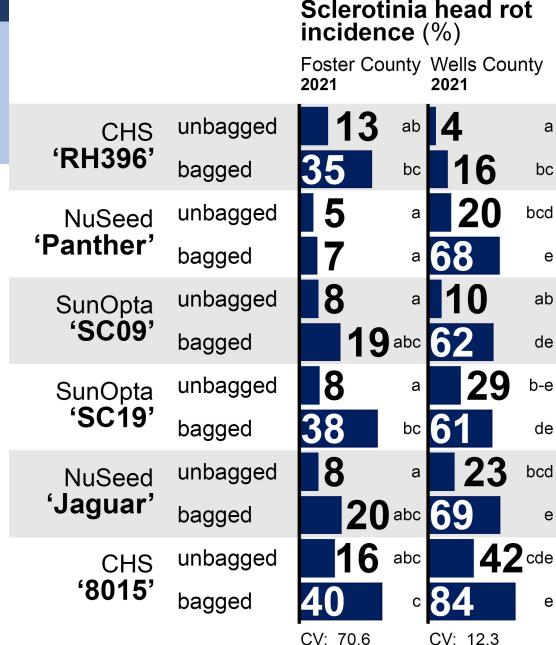
Sunflowers were established in plots 15 to 215 feet from each set of bee hives (Foster and Wells County study sites) and 615 to 815 from each set of bee hives (Foster County only).

POLLINATION BAGS: 16x18" (40.64 x 45.72 cm) HDPE plastic with 336-micron pore diameter; 24% of the surface open (Midco Global;

Kirkwood, MO)

Foster and Wells Counties, ND (2021)





Efficacy of honeybee-vectored *Clonostachys rosea* relative to sunflower hybrid

Replicated studies (3 reps).

Sunflowers were established in plots 15 to 215 feet from each set of bee hives (Foster and Wells County study sites) and 615 to 815 from each set of bee hives (Foster County only). Foster and Wells Counties, ND (2021)

PLOT SIZE: ~100 plants/plot across 3 or 4 rows. HIVES: 4 hives at each location, 50 to 100% of hives equipped with dispensers of the biological control.

head.

Kirkwood, MO)

INOCULATIONS: Sunflowers were inoculated once at approx. R5.4-R5.8. To each head, 15,000 ascospores were applied per

POLLINATION BAGS: 16x18" (40.64 x 45.72 cm) HDPE plastic with 336-micron pore diameter; 24% of the surface open (Midco Global;

NuSeed

SunOpta **'SC09'**

SunOpta 'SC19'

NuSeed

CHS

'8015'

'Jaguar'

'Panther'

unbagged CHS 'RH396'

bagged unbagged

bagged unbagged bagged

unbagged bagged

bagged

unbagged bagged unbagged

Sunflower

2021

975

1423

1450

1375

1306

yield (pounds/acre)

а

bc

bcd

Foster County Wells County

2021

3358

2790

2715

а

bc

bcd

е

ab

de

b-e

de

bcd

cde

е

2243 е ab de b-e de bcd

е

729

CV: 24.3

2184

CV: 20.0

1049

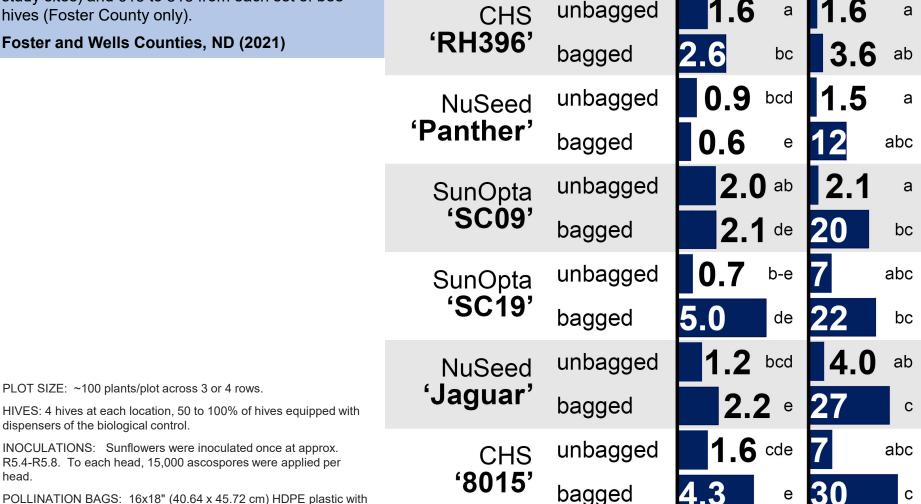
Efficacy of honeybee-vectored *Clonostachys rosea* relative to sunflower hybrid

Replicated studies (3 reps).

Sunflowers were established in plots 15 to 215 feet from each set of bee hives (Foster and Wells County study sites) and 615 to 815 from each set of bee hives (Foster County only).

336-micron pore diameter; 24% of the surface open (Midco Global;

Kirkwood, MO)



Sclerotia in grain

Foster County Wells County

a

2021

CV: 28.3

а

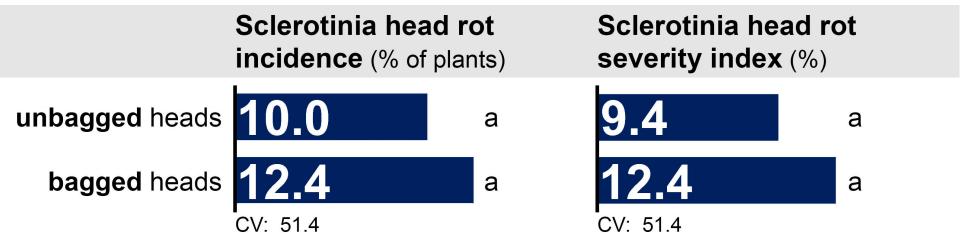
(% by weight)

2021

CV: 74.6

What impact do pollination bags have on Sclerotinia head rot disease pressure?

Replicated study (5 reps; average 34 plants/plot). **Foster County, ND (2021)**



- Pollination bags were associated with a modest, but not statistically significant, increase in head rot pressure
- Increase in head rot pressure was less than that observed in the beevectoring studies

PLOT SIZE: Average 34 plants/plot (max = 46 plants, min = 28 plants).

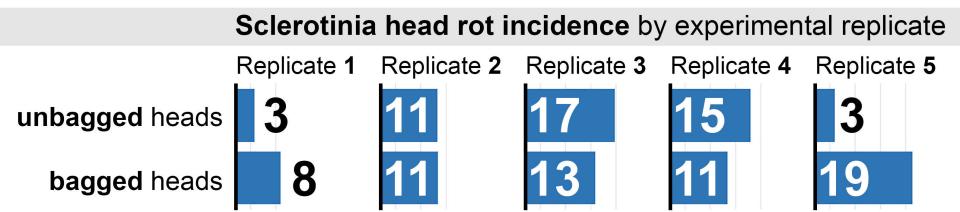
NO BEE HIVES. Study was conducted nearly 1 mile from the bee-vectoring study.

INOCULATIONS: Sunflowers were inoculated once at approx. R5.5-R5.8. To each head, 15,000 ascospores were applied per head.

POLLINATION BAGS: 18 x 16 inch (length x width), fine mesh fabric with 1 mm x 1 mm holes (Lawson Bags; Northfield, IL))

What impact do pollination bags have on Sclerotinia head rot disease pressure?

Replicated study (5 reps; average 34 plants/plot). **Foster County, ND (2021)**



Follow-up testing is needed.

- Very high variability in results was observed across replicates.
- Two brands of pollination bags were utilized, and the impact of the pollination bag on head rot may differ across brands.

PLOT SIZE: Average 34 plants/plot (max = 46 plants, min = 28 plants).

NO BEE HIVES. Study was conducted nearly 1 mile from the bee-vectoring study.

INOCULATIONS: Sunflowers were inoculated once at approx. R5.5-R5.8. To each head, 15,000 ascospores were applied per head.

POLLINATION BAGS: 18 x 16 inch (length x width), fine mesh fabric with 1 mm x 1 mm holes (Lawson Bags; Northfield, IL))

