



Species diversity of Soybean Sudden Death Syndrome pathogens in Kansas

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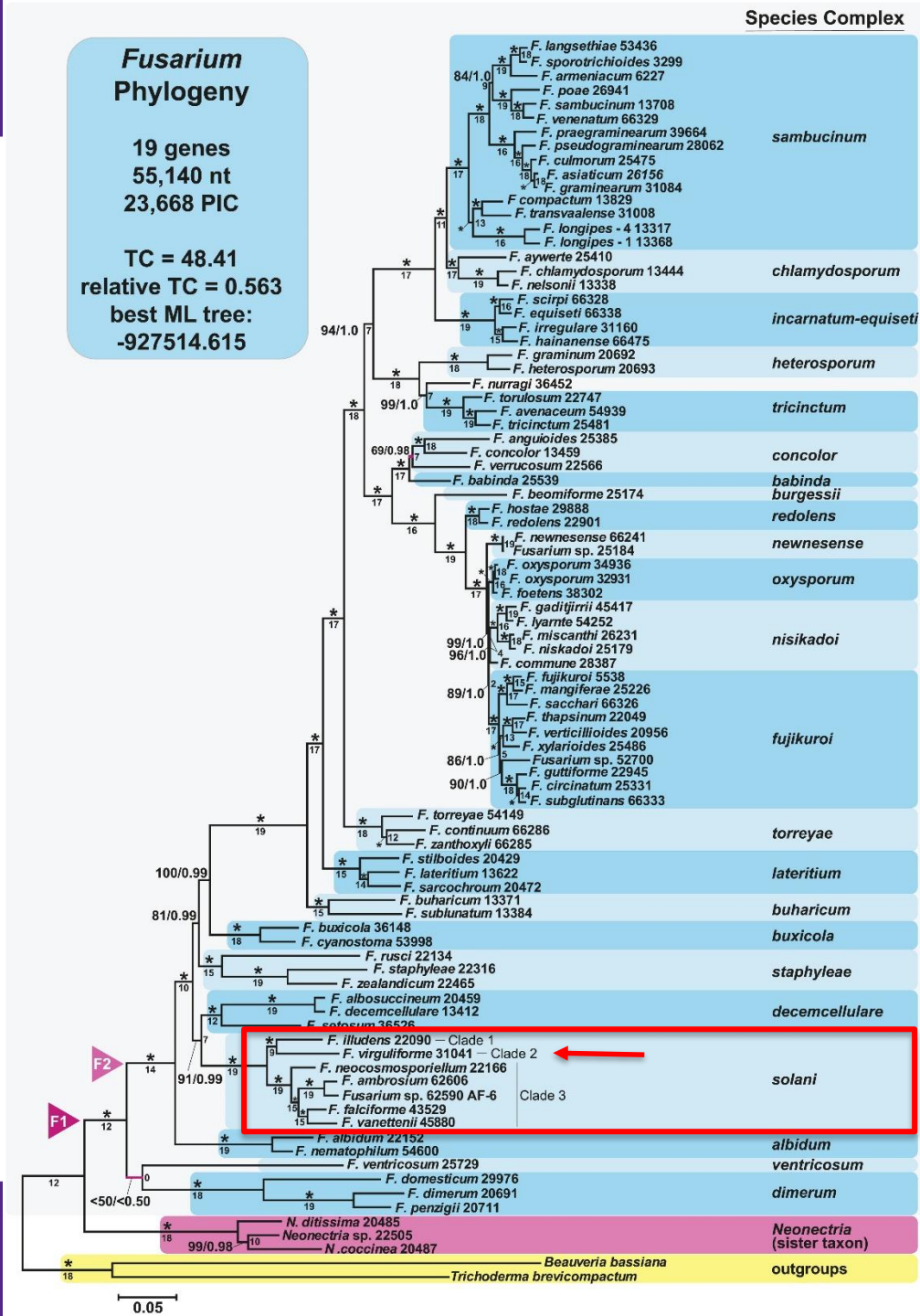
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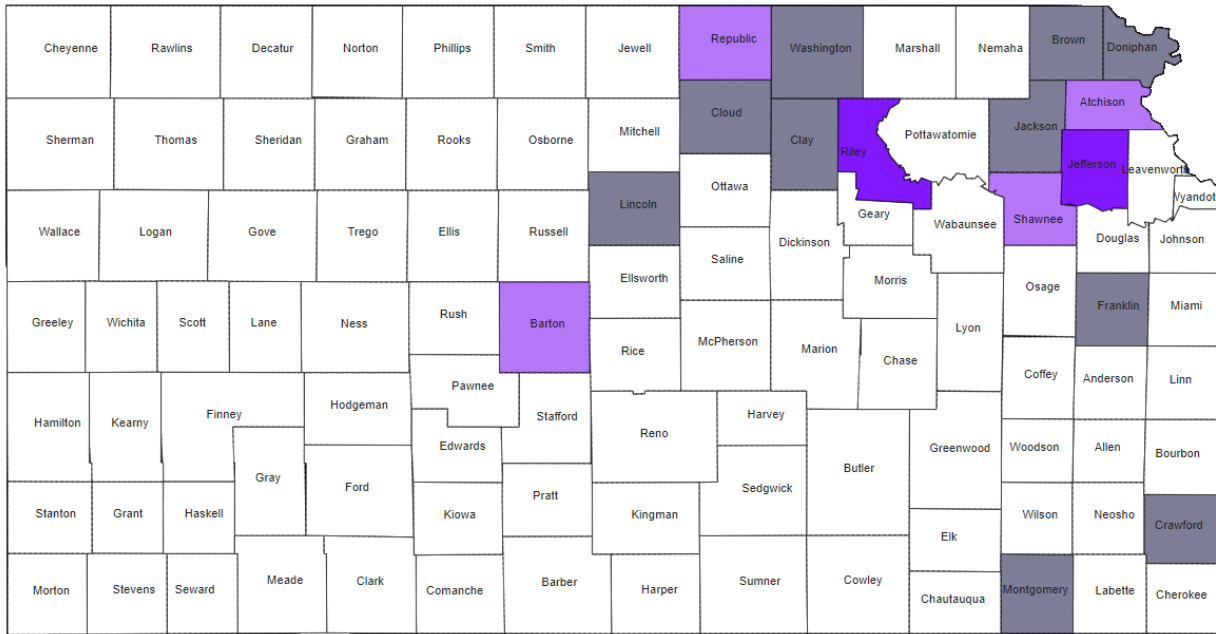
Background

- Sudden Death Syndrome (SDS) of soybeans:
 - *Fusarium solani* species complex (FSSC) clade 2.
 - *Fusarium virguliforme**, *F. tucumaniae*, *F. brasiliense* and *F. crassistipitatum*:
 - 1993: First report of SDS in Kansas.
 - 2019: First report of *F. brasiliense* present in Michigan (2014 and 2016).
- Soilborne disease;
- Primary inoculum:
 - Infested soil with chlamydospores;
 - Soybeans and corn crop residue.



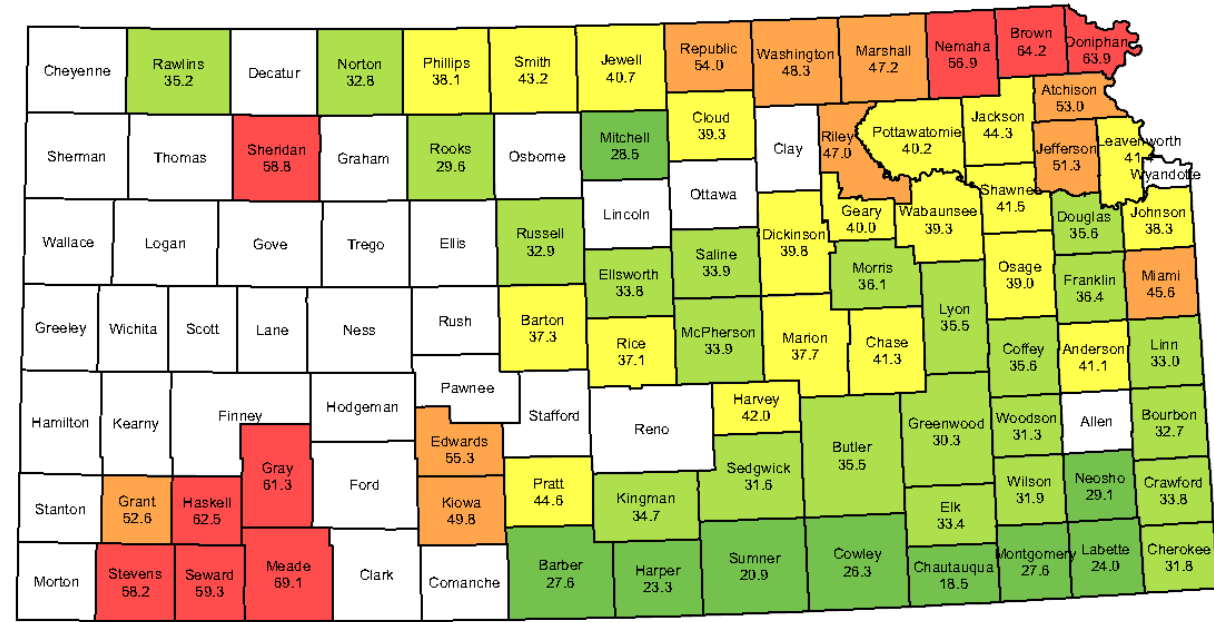
Samples collection

Kansas counties where samples were collected.



2022 Season – 6 counties
 2023 Season – 12 counties
 2022 and 2023 Season

2021 Kansas Soybean Production



Soybean Yield
 38.5 - 44.6
 44.7 - 55.3
 55.4 - 69.1
 Not Published

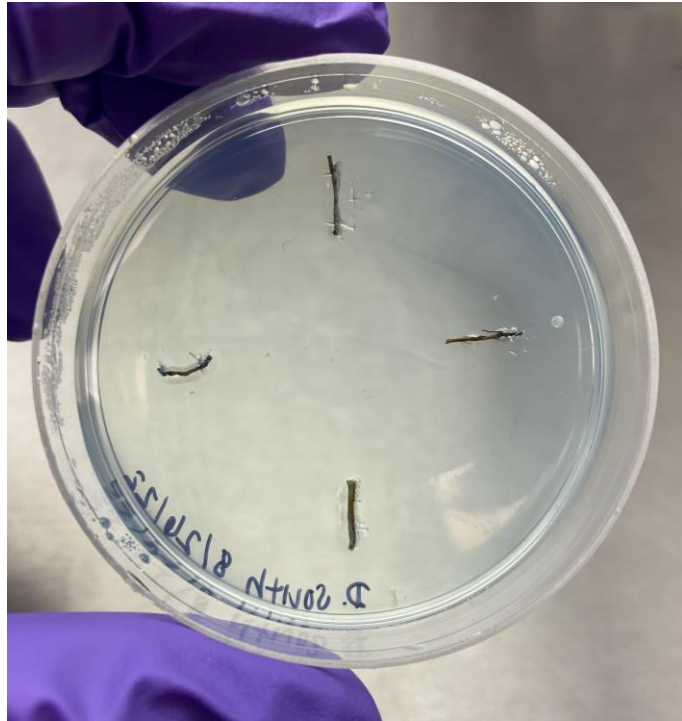
Bushels Per Acre
 18.5 - 29.1
 29.2 - 36.4

0 15 30 60 90 120 Miles

Source: USDA National Agricultural Statistics Service
February 25, 2022

Fusarium isolation

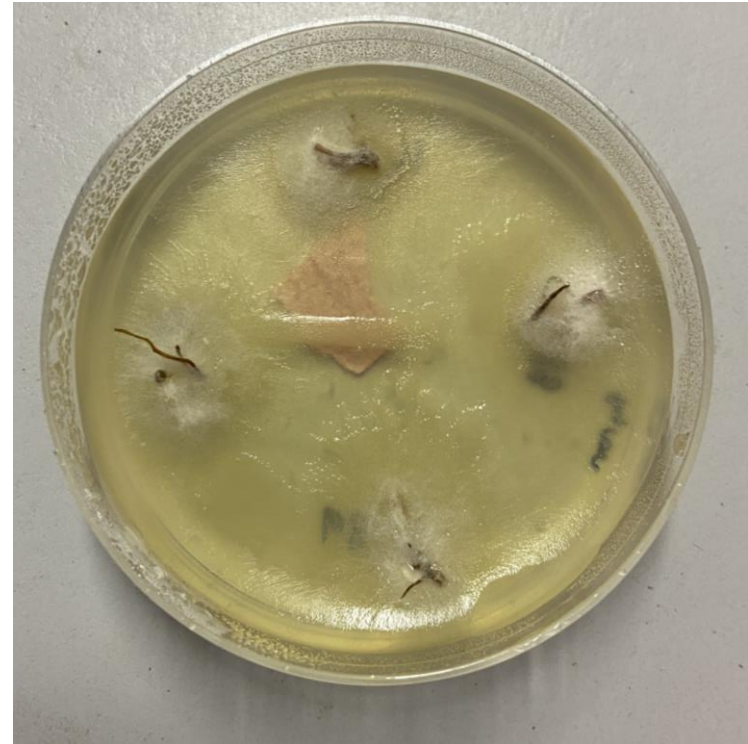
- SDS-symptomatic soybean plants collected across Kansas:



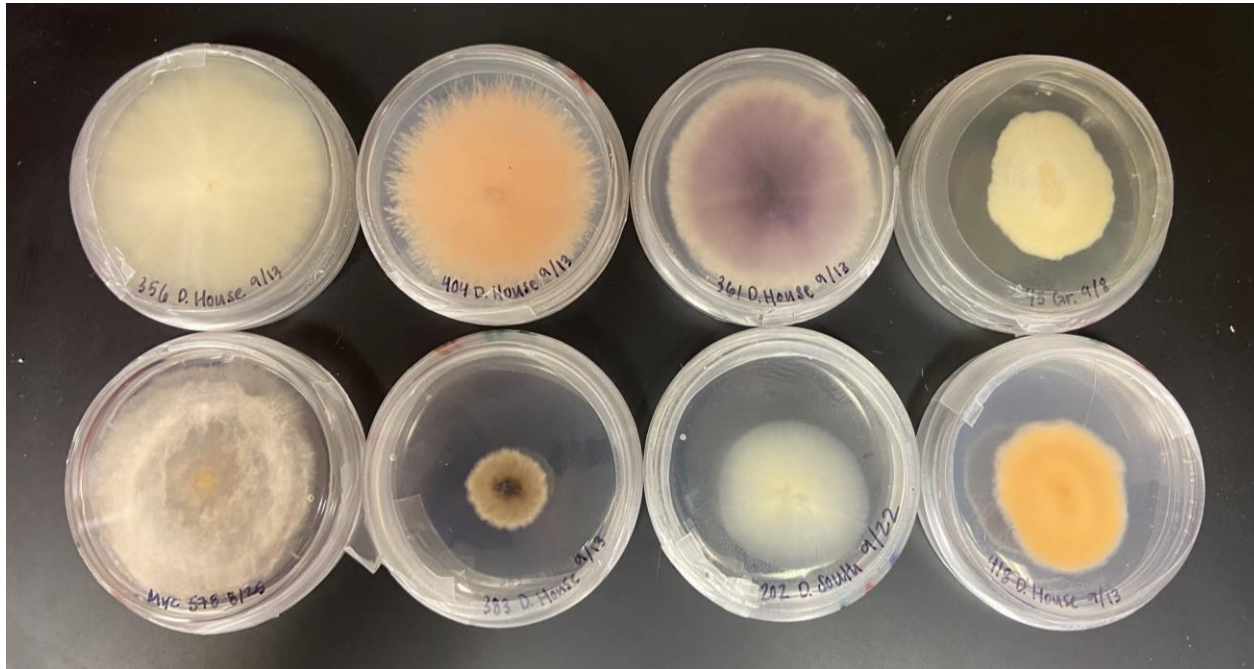
Malachite Green Agar



26 °C in the dark
for 4-5 days

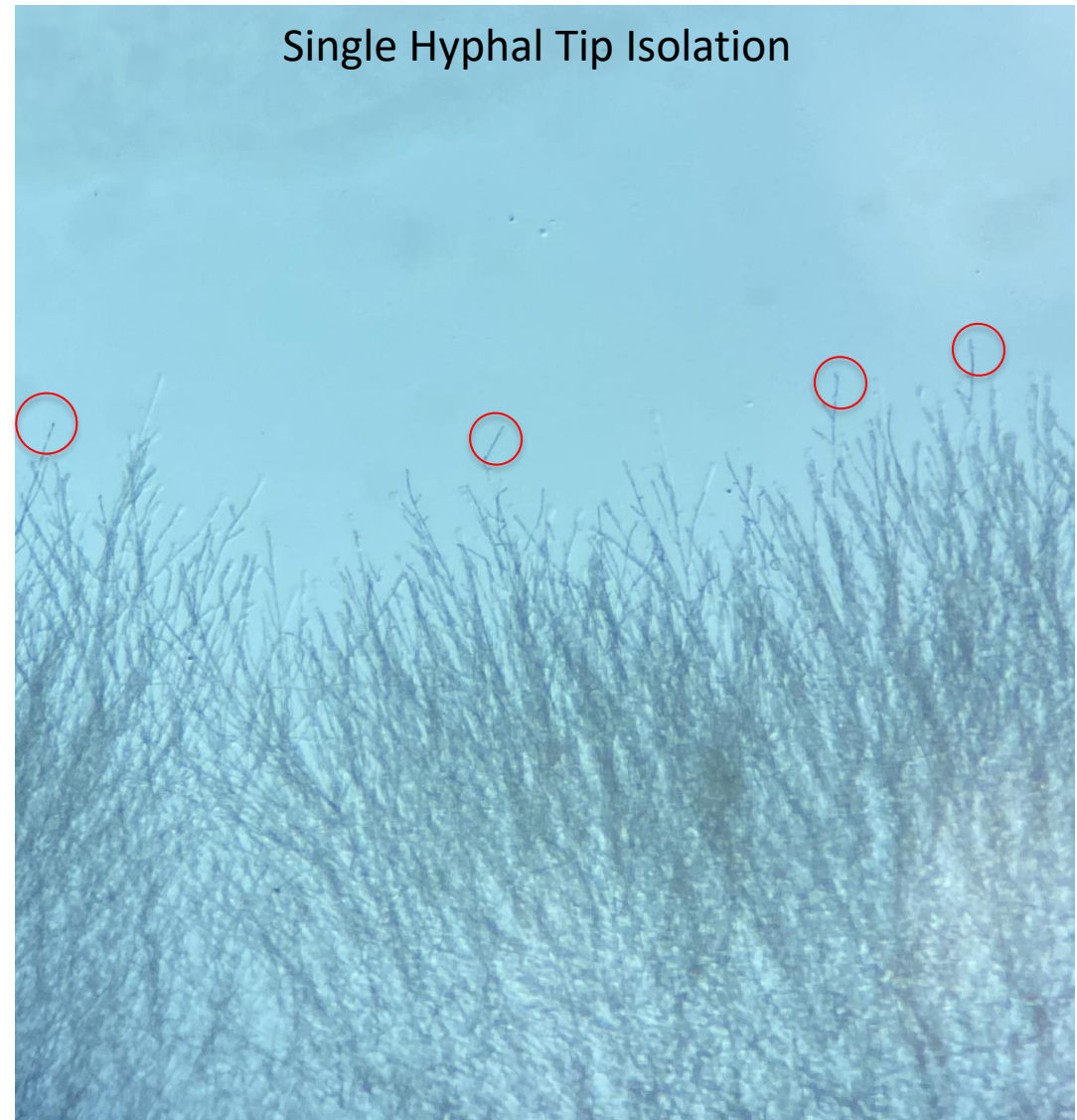


Fusarium – Pure culture



Potato Dextrose Agar

Single Hyphal Tip Isolation



2022 – Diversity Analysis of *Fusarium* isolates associate with SDS in Kansas

County	Total <i>Fusarium</i> -like Isolates #
Shawnee	6
Jefferson	97
Barton	11
Riley	12
Republic	2
Total	128

DNA Extraction and PCR Amplification

- 113 *Fusarium virguliforme*-like isolates and 15 *Fusarium*-like isolates:



- Target genes:
 - **Broad screening for *Fusarium* Species Complex and FSSC – clade 2:** Translation elongation factor-1 α (EF-1 α) for all *Fusarium* isolates.
 - **Screening for FSSC clade 2 - species:** 28S-18S ribosomal RNA intergenic spacer (IGS) only for FSSC species.

TEF tree – isolates distribution in different species complex

FSSC Clade 2

FSSC Clade 11

FSSC Clade 3

FSSC Clade 5

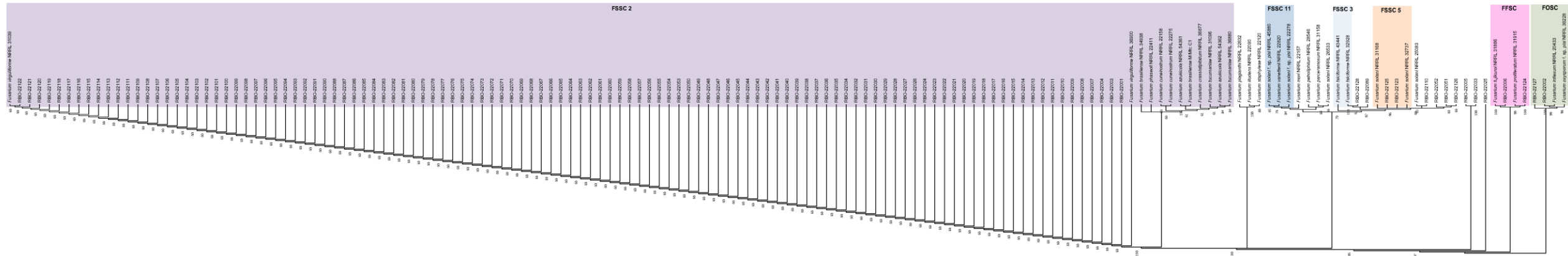
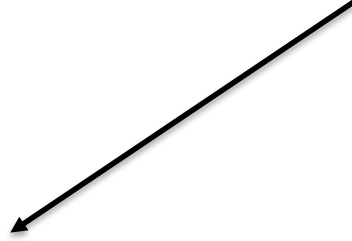
FFSC

FOSC

RBO: isolates from this work.

NRRL: USDA-ARS Culture Collection

Most of the isolates – FSSC Clade 2



The evolutionary history was inferred by using the Maximum Likelihood method and Kimura 2-parameter model. The bootstrap consensus tree inferred from 1000 replicates is taken to represent the evolutionary history of the taxa analyzed. Branches corresponding to partitions reproduced in less than 60% bootstrap replicates are collapsed. This tree is rooted on the sequences of *F. oxysporum* f. sp. *pisi* from the National Center for Biotechnology Information GenBank (NRRL 36228).

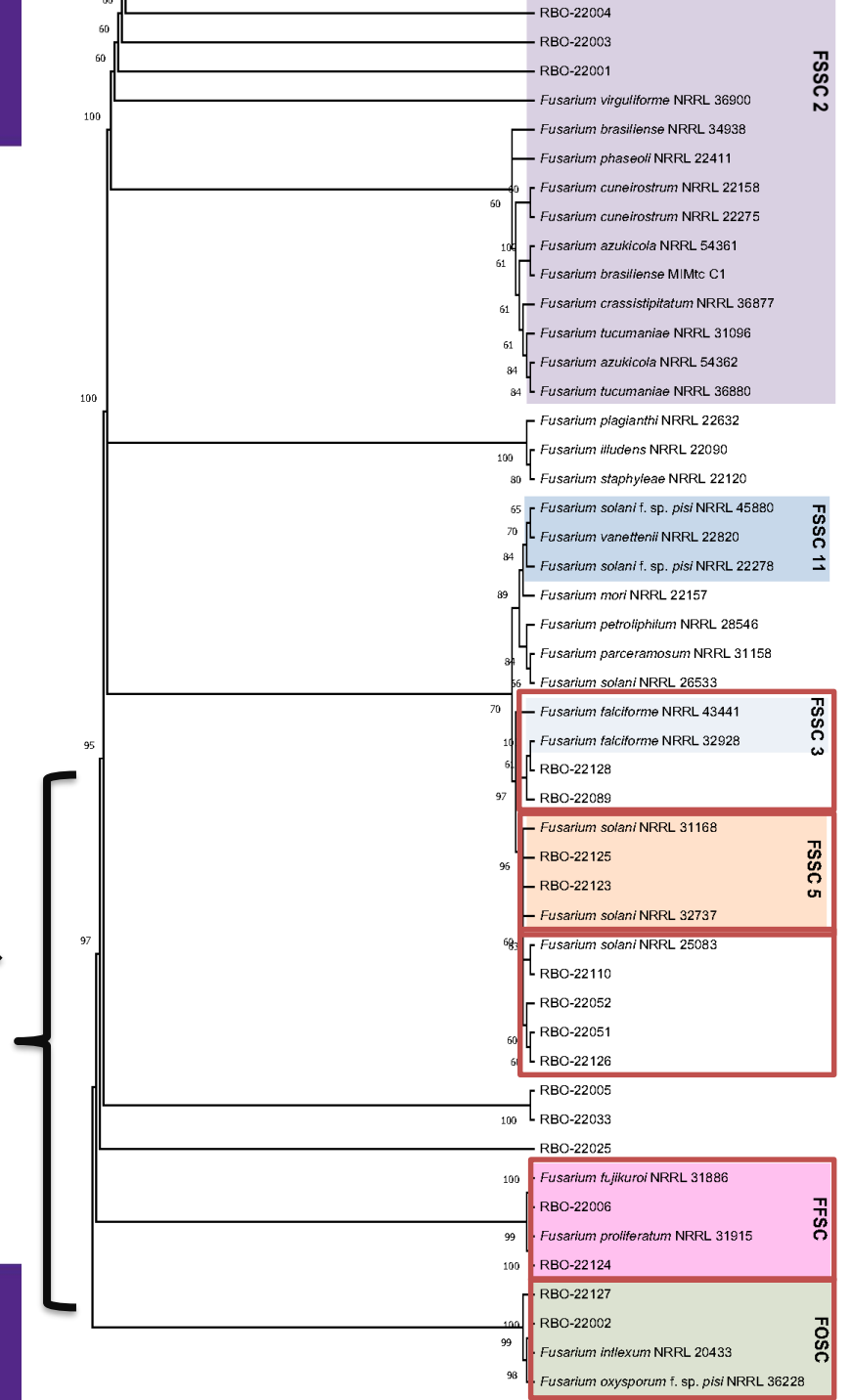
TEF tree – isolates distribution in different species complex

- FSSC Clade 2
- FSSC Clade 11
- FSSC Clade 3
- FSSC Clade 5
- FFSC
- FOSC

RBO: isolates from this work.
NRRL: USDA-ARS Culture Collection

Other *Fusarium* species are present in symptomatic plants

The evolutionary history was inferred by using the Maximum Likelihood method and Kimura 2-parameter model. The bootstrap consensus tree inferred from 1000 replicates is taken to represent the evolutionary history of the taxa analyzed. Branches corresponding to partitions reproduced in less than 60% bootstrap replicates are collapsed. This tree is rooted on the sequences of *F. oxysporum* f. sp. *pisi* from the National Center for Biotechnology Information GenBank (NRRL 36228).



IGS tree -

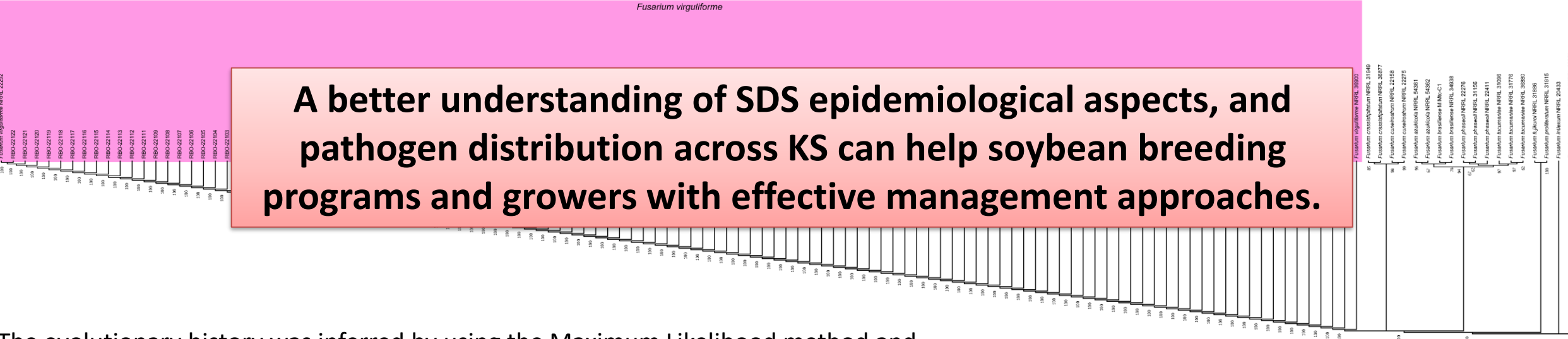
Fusarium virguliforme

RBO: isolates from this work.

NRRL: USDA-ARS Culture Collection

***F. virguliforme* is most likely causing SDS in Kansas**

All *F. virguliforme*-like strains had 100% identity with NRRL36900 and NRRL31039 (*F. virguliforme* NCBI deposit).



A better understanding of SDS epidemiological aspects, and pathogen distribution across KS can help soybean breeding programs and growers with effective management approaches.

The evolutionary history was inferred by using the Maximum Likelihood method and Hasegawa-Kishino-Yano model. The bootstrap consensus tree inferred from 1000 replicates is taken to represent the evolutionary history of the taxa analyzed. Branches corresponding to partitions reproduced in less than 60% bootstrap replicates are collapsed. This tree is rooted on the sequences of *F. oxysporum* f. sp. *pisi* from the National Center for Biotechnology Information GenBank (NRRL 36228).

2022 – Diversity Analysis of *Fusarium* isolates associate with SDS in Kansas

County	Total <i>Fusarium</i> -like Isolates #	<i>F. virguliforme</i> isolates #
Shawnee	6	3
Jefferson	97	90
Barton	11	9
Riley	12	11
Republic	2	0
Atchison	0	0
Total	128	113