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Reevaluation of *Phomopsis* species affecting sunflowers in the United States

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Phomopsis on sunflowers

- Phomopsis stem canker (PSC) frequently causes serious economic damage on sunflowers in other countries.

- In the U.S., very low incidence and limited damage occurred before 2010.
- In 2010 and 2011, PSC continued to increase in the North Central States

Phomopsis on sunflowers

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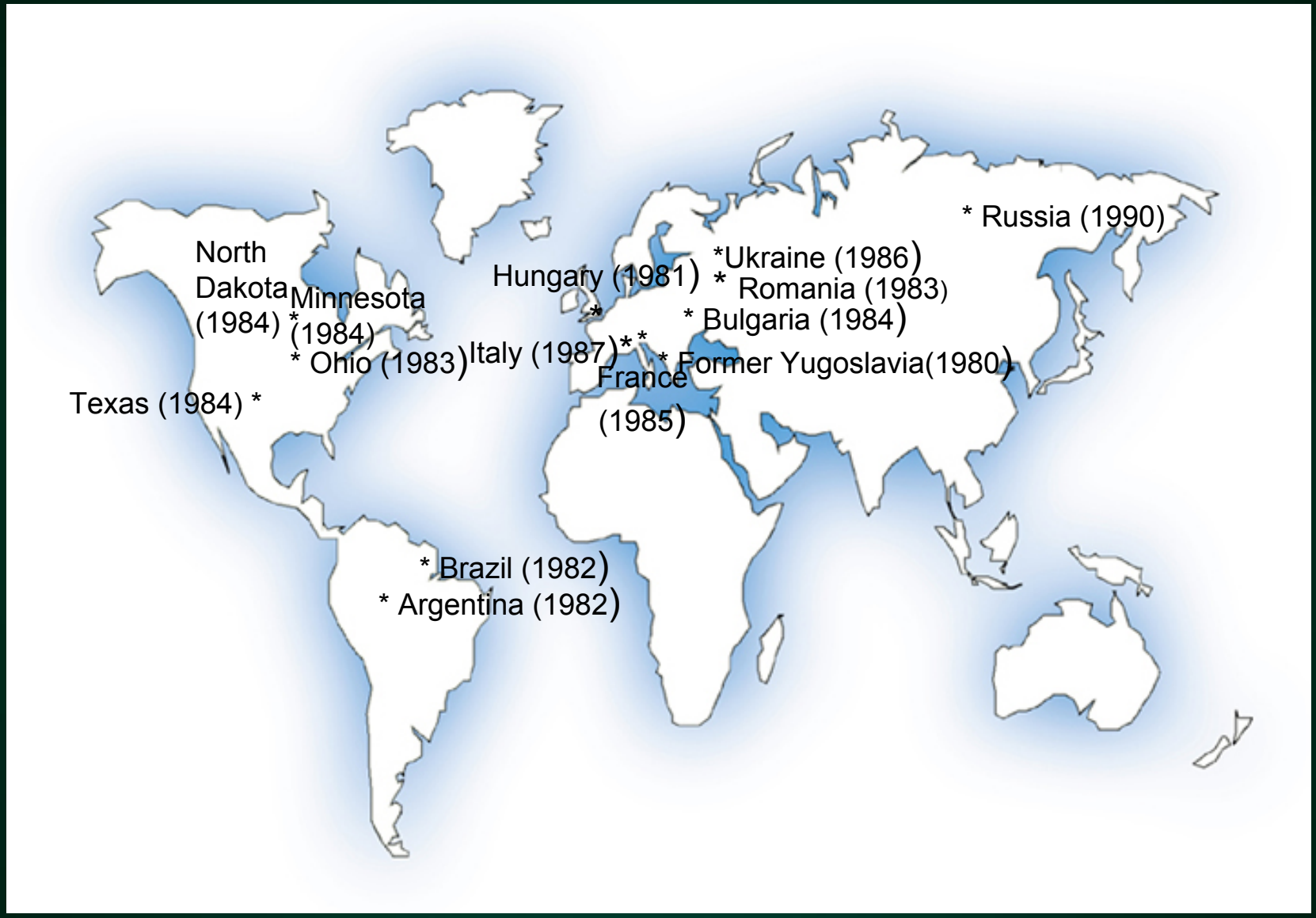
Tracing Phomopsis

- *Phomopsis* (Sacc.) Bubák, 1905 (teleomorph *Diaporthe* Nitschke, 1870) was first applied to anamorphs of nectriaceous fungi
- Difficult to distinguish morphologically
- Host association was the basis for species identification in *Diaporthe* and *Phomopsis* (van Rensburg *et al.* 2006)



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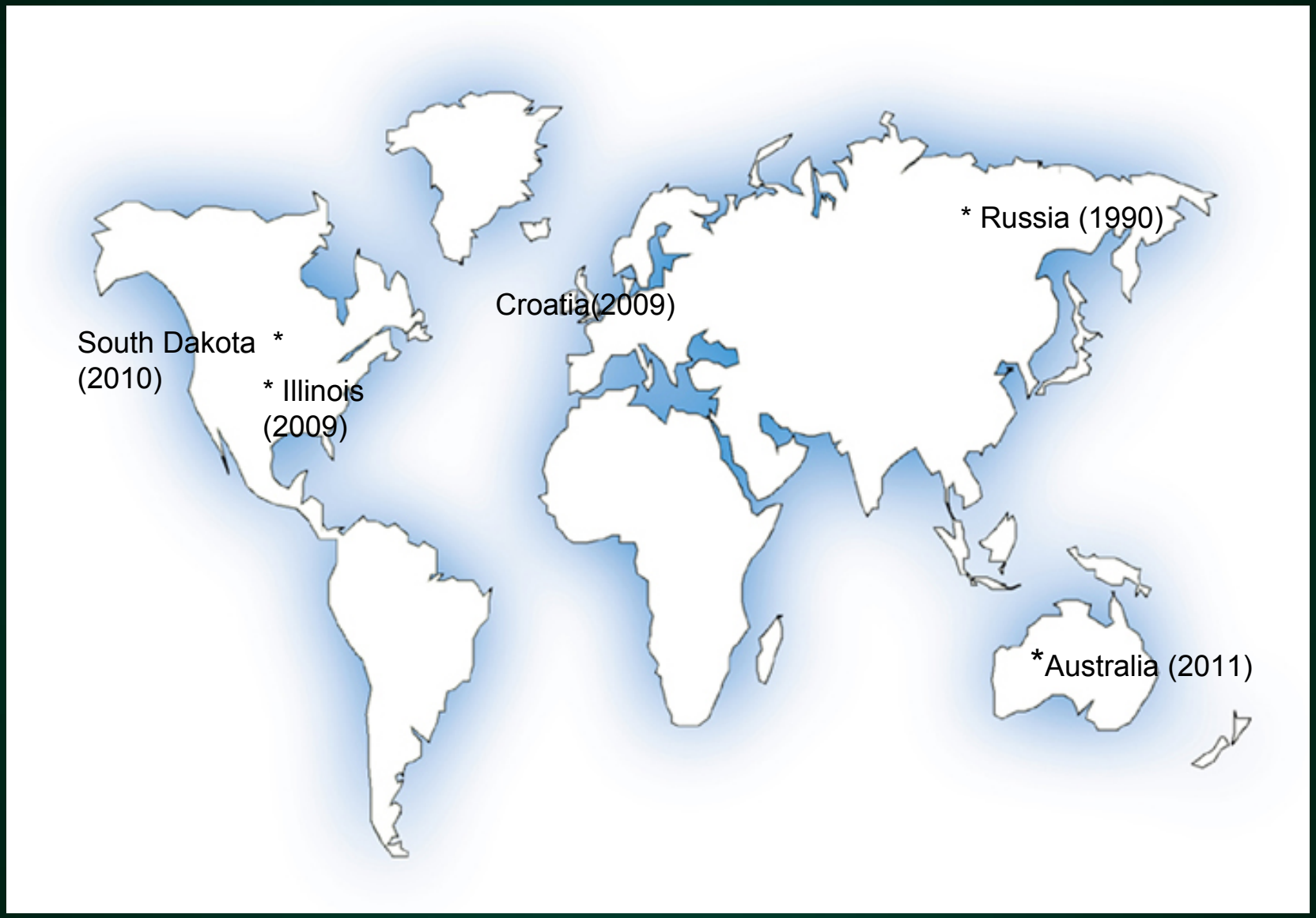
Diaporthe helianthi

Diaporthe phaseolorum



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Diaporthe helianthi
Diaporthe stewartii

Diaporthe phaseolorum

Diaporthe gulyae
Diaporthe kochmanii
Diaporthe kongii

Objective

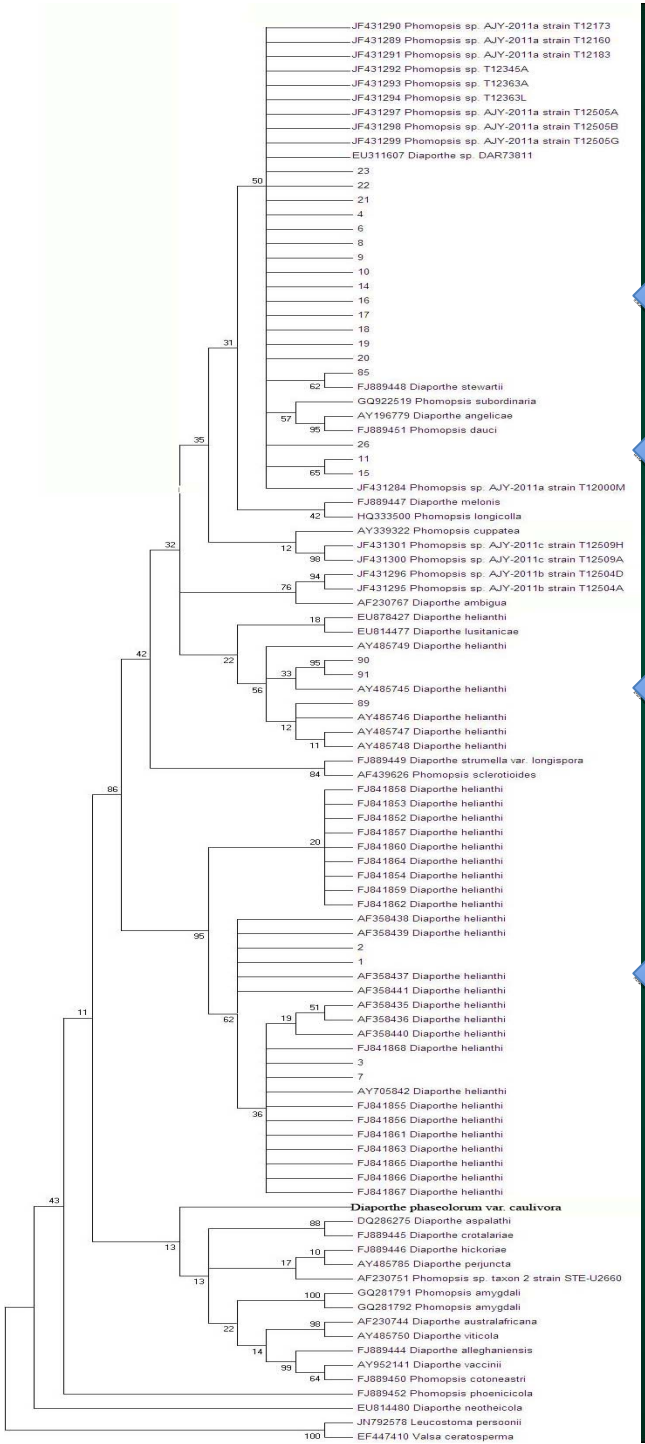
Characterization of the species and their prevalence causing *Phomopsis* on sunflowers in the United States

Lisa Castlebury

- Systematicist –
  USDA Beltsville, MD

Materials & Methods: 2010

- 1150 stalks were chopped, sterilized, and plated on potato dextrose agar (PDA) for 7-10 d.
- Phomopsis isolates were hyphal tipped
- Several dozen isolates recovered
- The rDNA-ITS region was amplified and sequenced with primers ITS4 and ITS5 (White *et al*., 1990)
- Analysis was performed using BLASTN via the NCBI database (www.ncbi.nlm.nih.gov).



Isolates from South Dakota - 2010 collection, now called "*Diaporthe gulyae*"

A single isolate of *Diaporthe stewartii* from South Dakota - 2010 collection

Two isolates of *Diaporthe helianthi* from Texas - 2010 collection), A single isolate of *Diaporthe helianthi* from South Dakota - 2010 collection

Four isolates of *Diaporthe helianthi* from Minnesota and North Dakota- 2010 collection

2011

- 163 fields randomly sampled (ND, MN, SD).
- Additional samples (627) were also received through NSA survey and other contacts.
- Additionally, hierarchical sampling of 9 infected fields (3 MN, 3 ND and 3 SD) was completed; 405 stems

Results

- An approximate total of 900 isolates were retrieved from both sampling strategies
- 150 isolates from randomly sampled fields were sequenced.
  - 86 isolates (ND, MN, SD) were *D. helianthi*
  - 2 isolates (SD, ND) were *D. gulyae*

Results and Discussion

- There are *a total of six species* causing phomopsis on sunflowers in the world as of 2011
  - Three confirmed in the US.
    - *D. helianthi, D. gulyae and D. stewartii*
- Greenhouse trials to assess virulence in U.S.
- Gulya et al. evaluation of resistance in the field.

Fungicides

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Treatment/Rate	Timing		
Non-Treated	NA		
Headline 6 fl oz	V10		
Headline 6 fl oz	V10, R1		
Headline 6 fl oz	V10, R5		
Headline 6 fl oz	V10, R1, R5		
Headline 6 fl oz	R1		
Headline 6 fl oz	R1, R5		
Headline 6 fl oz	R5		

Treatment/Rate	Timing		
Non-Treated	NA		
Headline 6 fl oz	V10		
Headline 6 fl oz	V10, R1		
Headline 6 fl oz	V10, R5		
Headline 6 fl oz	V10, R1, R5		
Headline 6 fl oz	R1		
Headline 6 fl oz	R1, R5		
Headline 6 fl oz	R5		
Folicur / Propulse	R1, R5		
Headline / Priaxor	R1, R5		
Headline /Quash	R1, R5		
Tilt / Quadris	R1, R5		
Aproach /Vertisan	R1, R5		

Treatment/Rate	Timing	Rust R6	
Non-Treated	NA	3.73 a	
Headline 6 fl oz	V10	2.20 b	
Headline 6 fl oz	V10, R1	1.53 cde	
Headline 6 fl oz	V10, R5	1.55 cde	
Headline 6 fl oz	V10, R1, R5	0.64 fg	
Headline 6 fl oz	R1	2.43 b	
Headline 6 fl oz	R1, R5	0.86 efg	
Headline 6 fl oz	R5	1.65 cd	
Folicur / Propulse	R1, R5	0.44 g	
Headline / Priaxor	R1, R5	0.70 fg	
Headline /Quash	R1, R5	0.60 fg	
Tilt / Quadris	R1, R5	0.78 fg	
Aproach /Vertisan	R1, R5	1.09 defg	

Treatment/Rate	Timing	Rust R6	Phomopsis
Non-Treated	NA	3.73 a	2.95 a
Headline 6 fl oz	V10	2.20 b	2.1 b
Headline 6 fl oz	V10, R1	1.53 cde	1.08 def
Headline 6 fl oz	V10, R5	1.55 cde	1.9 bc
Headline 6 fl oz	V10, R1, R5	0.64 fg	0.43 f
Headline 6 fl oz	R1	2.43 b	1.63 bcd
Headline 6 fl oz	R1, R5	0.86 efg	0.65 ef
Headline 6 fl oz	R5	1.65 cd	2.0 bc
Folicur / Propulse	R1, R5	0.44 g	0.53 ef
Headline / Priaxor	R1, R5	0.70 fg	1.08 def
Headline /Quash	R1, R5	0.60 fg	0.90 def
Tilt / Quadris	R1, R5	0.78 fg	1.05 def
Aproach /Vertisan	R1, R5	1.09 defg	1.25 cde

Treatment/Rate	Timing		
Non-Treated	NA		
Headline 6 fl oz	V10		
Headline 6 fl oz	V10, R1		
Headline 6 fl oz	V10, R5		
Headline 6 fl oz	V10, R1, R5		
Headline 6 fl oz	R1		
Headline 6 fl oz	R1, R5		
Headline 6 fl oz	R5		
Folicur / Propulse	R1, R5		
Headline / Priaxor	R1, R5		
Headline /Quash	R1, R5		
Tilt / Quadris	R1, R5		
Aproach /Vertisan	R1, R5		

Treatment/Rate	Timing	Yield	
Non-Treated	NA	1081 f	
Headline 6 fl oz	V10	1535 cdef	
Headline 6 fl oz	V10, R1	1831 abcde	
Headline 6 fl oz	V10, R5	1369 def	
Headline 6 fl oz	V10, R1, R5	2193 abc	
Headline 6 fl oz	R1	1666 bcdef	
Headline 6 fl oz	R1, R5	2133 abc	
Headline 6 fl oz	R5	1178 ef	
Folicur / Propulse	R1, R5	2486 a	
Headline / Priaxor	R1, R5	2045 abcd	
Headline /Quash	R1, R5	2342 ab	
Tilt / Quadris	R1, R5	1963 abcd	
Aproach /Vertisan	R1, R5	1936 abcd	

Treatment/Rate	Timing	Yield	Test Weight
Non-Treated	NA	1081 f	18.62 g
Headline 6 fl oz	V10	1535 cdef	19.97 ef
Headline 6 fl oz	V10, R1	1831 abcde	21.09 cde
Headline 6 fl oz	V10, R5	1369 def	20.42 ef
Headline 6 fl oz	V10, R1, R5	2193 abc	22.52 a
Headline 6 fl oz	R1	1666 bcdef	19.63 fg
Headline 6 fl oz	R1, R5	2133 abc	21.74 abcd
Headline 6 fl oz	R5	1178 ef	20.47 def
Folicur / Propulse	R1, R5	2486 a	22.64 a
Headline / Priaxor	R1, R5	2045 abcd	22.02 abc
Headline /Quash	R1, R5	2342 ab	22.41 ab
Tilt / Quadris	R1, R5	1963 abcd	21.76 abcd
Aproach /Vertisan	R1, R5	1936 abcd	20.89 cdef

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- NSA Surveyors

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Disease cycle of Phomopsis

- The disease is most severe under conditions of prolonged high temperatures and high humidity.
- The fungus overwinters as mycelium and perithecia on stem debris left on the soil.
- Spores released
  - Splash and wind dispersed
  - Ascospores germinate in the guttation drops at the leaf margin to initiate infection.
  - Leaf → petiole → stem (1 month)
  - The first resulting lesions on the stems carry pycnidia with pycnidiospores.

Thompson et al. 2010 2010 Summer Grains Conf. Qld Australia

Tracing Phomopsis on sunflowers

- *Phomopsis* sp. was identified on sunflower stems in 1980 in Yugoslavia.
  - Identity of causal agent was controversial.
- Differences noticed among Yugoslavian *Phomopsis* (Muntañola-Cvetkovic´ *et al.* 1985)
  - produced β-conidia only
  - Differences in symptoms on plants, responses of sunflower plants to inoculation under controlled conditions, etc.
  - Authors (Aćimović and Štraser, 1982) concluded sunflower is attacked by two *Phomopsis* species.

Tracing Phomopsis on sunflowers

- United States (Ohio) - 1983 (Ohio)
- Yang *et al.* (1984) published the first verified report of *D. helianthi* on the cultivated sunflower in Texas
- Biological differences between USA (α- and β-conidia or both) and Yugoslavian isolates (β-conidia)
  - Possibility of several pathogenic Phomopsis species or biotypes in USA and Europe (Gulya *et al.*, 1997)

Results - 2010

- 99 isolates recovered
  - 85 = SD, 7 = ND, = MN, 2 = TX
- ML tree generated from ITS of the nuclear ribosomal DNA (rDNA) of reference *Phomopsis* sequences of Thompson *et al.* (2011) and isolates collected in 2010.
- Reference sequences are indicated with GenBank accession numbers followed by the species.
- Three species identified and confirmed– *D. helianthi*, *D. gulyae* and *D. stewartii*

Tracing Phomopsis on sunflowers

- More than one Phomopsis species can occur on a single host (Mostert *et al.* 2001, Santos & Phillips 2009).
- A total of six species of *Phomopsis* on sunflowers across the world as of Dec 2011.
  - *Diaporthe helianthi* continues to be predominant on sunflowers (Mathew, unpublished)