

# Virulence and Genetic Diversity of *Phakopsora pachyrhizi* in Nigeria

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## Introduction

Soybean rust, caused by *Phakopsora pachyrhizi* (Fig. 1), is a major disease in many soybean-producing areas of the world. Nigeria (Fig. 2), located in the West Africa region, is the largest soybean producer in Africa. Between 1994 and 1998, soybean production increased by 250%. The screening of newly released cultivars developed by IITA in many locations (2) revealed some considerable variations in rust resistance among soybean genotypes at different sites pointing to variability in the pathogen population. Understanding the virulence and genetic diversity of the pathogen may help with the deployment of soybean rust resistance genes.



Fig. 1. Soybean rust (A) leaf heavily infected, (B) urediniospores, (C) urediniospore germination, (D) uredinia and (E) mass of spores on infected leaves

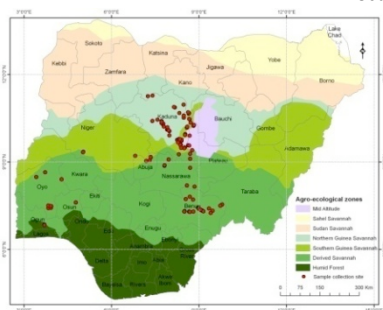


Fig. 2. Nigeria map with sample collection sites

## Objective

- To examine the variation of *Phakopsora pachyrhizi* populations collected from soybean growing areas in Nigeria.

## Materials and methods

- A total of 116 purified isolates established from infected leaves arbitrarily collected from soybean fields in four agroecological zones (AEZ) were used for the pathogenic variation study. Eight soybean genotypes including four accessions containing *Rpp1* (PI 200492), *Rpp2* (PI 230970), *Rpp3* (PI 462312), and *Rpp4* (PI 459025B) genes, two highly resistant and two highly susceptible accessions were used as a host differential set.
- In addition to the isolates used in the pathogenic variation study, another 146 isolates collected from infected plants in two fields (73 isolates in each field) located 292 km apart were used for the genetic diversity study.
- Principal component and cluster analyses were used to assign isolates into pathotypes (1). Shannon's index was used to estimate diversity in the pathogen population at the selected areas.
- Eighteen simple sequence repeat (SSR) markers were used to study the genetic variability among the isolates. POPGENE, Arlequin version 3.11, GenAlEx6 and PAUP version 4.0b10 were used for population genetic differentiation and relationship using binary data generated after scoring peaks from SSR amplifications detected by capillary electrophoresis on an ABI3130x1 Analyzer. Peak calls were made with GeneMarker v1.51 software.

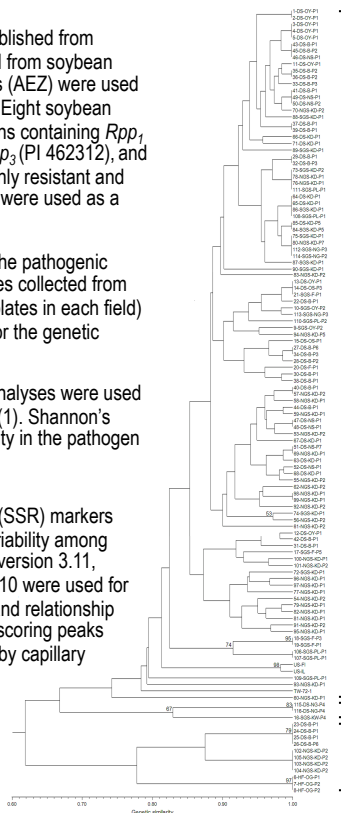


Fig. 4. Genetic similarity dendrogram of 116 *P. pachyrhizi* isolates collected from soybeans in four agroecological zones in Nigeria, two *P. pachyrhizi* isolates from U.S., and one from Taiwan, based on unweighted pair group method with arithmetic means clustering method. Numbers along the nodes are bootstrap values > 50% in 1000 replicas. Isolate names are composed by four parts: (i) number of isolates (1 to 116), (ii) AEZ (DS: Derived Savanna, SGS: Southern Guinea Savanna, NGS: Northern Guinea Savanna and HF: Humid Forest), (iii) state of origin (OY: Oyo, B: Benue, NS: Nasarawa, KD: Kaduna, OS: Osun, NG: Niger, PL: Plateau, OG: Ogun, KW: Kwara, F: Federal Capital Territory), and (iv) pathotype number 1 to 7.

## Results

- Principal component and Cluster analyses separated Nigerian rust isolates into seven pathotype clusters. Isolates in cluster III were the most virulent, while those in cluster IV were the least virulent (Table 1, Fig. 3). All seven pathotype clusters were present in the Derived Savanna unlike the other geographical zones where only some pathotypes occurred. Shannon diversity index indicated considerable variation in the population of *P. pachyrhizi*, with diversity being highest in Derived Savanna and Southern Guinea Savanna zones ( $h = 1.21$  and  $1.08$ , respectively).

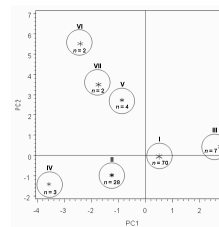


Fig. 3. Results of the principal component and cluster analyses. Sample size ( $n$ ) shown within clusters (circles) represent the number of isolates, plotted in a plane defined by the first two principal components (PC1\*PC2). Roman numerals adjacent to each cluster are the pathotype cluster groupings.

Table 1. Mean and standard deviation (in parenthesis) of rust severity on a set of eight differential genotypes of soybean for 116 Nigerian field isolates of *Phakopsora pachyrhizi* in seven groupings derived by cluster analysis

Pathotype cluster	Number of isolates	Soybean rust severity (number of uredinia per cm <sup>2</sup> ) on differential							
		TGx 1485-1D	TGx 1844-4F	PI 459025B	PI 200492	PI 230970	PI 462312	UG-5	PI 594538A
1	69	58.7 (6.3)	38.2 (5.3)	13.1 (2.2)	13.0 (3.2)	9.2 (2.1)	7.2 (3.2)	3.2 (2.4)	0.0
2	28	39.1 (6.0)	27.6 (6.5)	10.7 (2.6)	9.8 (3.7)	5.6 (3.6)	4.5 (3.7)	0.3 (0.9)	0.0
3	7	64.2 (7.5)	50.9 (2.7)	13.6 (1.5)	13.9 (2.1)	10.0 (2.1)	10.4 (2.9)	5.2 (1.3)	0.0
4	3	32.7 (8.8)	16.8 (0.3)	8.1 (5.4)	11.2 (1.6)	5.8 (5.1)	4.4 (3.8)	0.0 (0.0)	0.0
5	4	56.1 (6.2)	40.3 (4.0)	13.1 (2.8)	21.4 (4.8)	11.1 (3.7)	7.7 (2.1)	4.6 (2.0)	0.0
6	3	59.0 (1.9)	31.8 (7.4)	19.9 (0.2)	22.1 (2.3)	13.1 (1.9)	7.7 (0.0)	4.5 (0.9)	0.0
7	2	34.9 (8.2)	32.3 (8.8)	18.4 (0.9)	11.0 (1.0)	8.4 (2.3)	4.3 (3.7)	0.0 (0.0)	0.0

- Among 116 isolates from three geographical zones, 84 distinct SSR marker genotypes were identified and 48 distinct SSR marker genotypes were identified among 146 isolates from the two fields. Nei's average genetic diversity across geographical zones was 0.22 while for both fields was 0.09.
- The majority (> 90%) of the genetic diversity was distributed within a soybean field, while almost 6% was distributed among fields within geographic regions. The phylogenetic analysis showed three groups in Nigerian rust populations with one major group comprising more than 90% of the isolates (Fig. 4). The genetic similarity clustering of 116 isolates did not follow expectations based on virulence, indicating a poor correlation between pathogenic and molecular variation.

## Conclusion

Knowledge on soybean rust pathogen variation and the population distribution is useful in breeding and disease management by facilitating the deployment of rust-resistant cultivars. Similar studies have been initiated on US *P. pachyrhizi* isolates.

## References

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