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The XXI International Grassland Congress / VIII International Rangeland Congress took place in Hohhot, China from June 29 through July 5, 2008.

Proceedings edited by Organizing Committee of 2008 IGC/IRC Conference

Published by Guangdong People's Publishing House

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Genetic diversity analysis of *Colletotrichum gloeosporioides* from Stylo and Anthracnose resistant breeding

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Keywords : Stylosanthes , Colletotorichum gloeosporioides , RAPD , AFLP , virulence , epidemiology , resistant breeding

Introduction Stylosanthes spp is the most important and commercial forage legume in tropical area in China . It is widely used as forage, green manure, cover crops, erosion control, hay and meal commercial production. However, stylo anthracnose, mainly caused by a fungus Colletotrichum gloeosporioides, is the most severe disease in stylo production which has resulted in serious damage in China in the past 20 years. More than 3 productive varieties have been given up because of anthracnose.

Materials and methods A field survey on stylo anthracnose was carried out widely in all the stylo production sites in China in the last ten years and samples of infested plant materials were collected for isolation by pure-culture and single sporing .338 isolates of *Colletotrichum gloeosporioides* were selected randomly for RAPD and AFLP analysis from the isolates bank in CATAS and 104 isolates were selected for race typing based on virulence and pathogenic variation identification by using a set of host differientials. Disease development with time was also observed regularly for epidemiological research. Base on the results above , anthracnose resistant breeding for new varieties by different methods was implemented .

Results and discussions

1. Total 703 isolates of *Colletotrichum gloeosporioides* have been collected . 114 isolates were selected for RAPD analysis and 181 isolates for AFLP. In addition , 43 for RAPD which were clustered and compared with 276 isolates abroad. The isolates collected in China were grouped in clusters II , III and VI based on six clusters of isolates from South America and other countries. However, both of 114 isolates for RAPD and 181 siolate for AFLP were grouped in four clusters .-Two mollecular markers showed a significant DNA polymorphism and genetic diversity among the tested isolates from China .

2. The results showed different virulence in different isolates. Total 47 of type A isolates were grouped in 3 races and 57 of type B in 2 races. The results from race typing also showed that a wide pathegenic variation existed in type B. Thus more attention should be paid to the monitoring and evaluation on pathogens.

3 . An on-station observation on epidemyology for three years showed the severity and incidence of stylo anthracnose were significantly correlated with air humidity (r=0.44037, Pr=0.0457). There were two peaks of disease incidence in a year. The main peak was in August and September, the other in Jannuary which followed the rainfall.

4 . 36 new accessions were evaluated and compared based on the anthracnose resistance from 2001 to 2003 . 5 lines from S. *guianensis* and 3 accessions from S. *Seabrana* and Seca were selected with higher anthracnose resistance and good yield performance.

5 . Seeds from seven stylo species were treated by 60 Co- γ ray at 8 dose levels for suitable radioactive dose determination and new lines selection with high anthracnose resistance . 10 .32 \sim 15 .48C/kg was the suitable dose for stylo . 128 lines with highest disease resistance were selected by inoculation .

Conclusion It is concluded that the genetic diversity, race types and field epidemiology of Colletotrichum gloeosporioides from stylo in China have been clear initially and tens of new lines with high anthracnose resistance selected from this research have potential for new cultivars in the future.

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