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Physiologic Races of *Helminthosporium turcicum* in Taiwan[1]

C. C. Yeh and A. H. Tsai[2]

Abstract: Experiments were conducted to identify the races of *Helminthosporium turcicum*, the northern corn leaf blight fungus, and their distribution in Taiwan, and to explore their reaction in the currently grown maize cultivars. Leaf samples of northern leaf blight were collected from various growing areas and varieties/lines in 1987. Inoculum of a single lesion origin was considered an isolate. Several isolates were obtained from each collection site and/or variety/line. According to the reactions of each isolate on the differential varieties, three races of *H. turcicum*, namely race 1, 2, and 3, the same as in the U. S., were reported for the first time in Taiwan. Race 1 existed in Hualien and Taitung Counties (eastern Taiwan), race 2 in County (southern Taiwan), and race 3 in Taichung Chiayi (central Taiwan), Varieties/lines within a collection site did not influence the distribution pattern of race within that particular area. The grown cultivars Tainan 5, Tainan Selection 10, Tainan 11 and Tainan 16 were all susceptible to the three races. Sweet corn cultivar Honey 236 was susceptible to races 2 and 3 but resistant to some isolates of race 1. Tainung 351, the most resistant cultivar now, is resistant to racs 1 and 3 and only susceptible to a few isolates of race 2 from Chiayi County.

Northern leaf blight is one of the most destructive disease of maize (*Zea mays* L.) in most areas of the world where maize is grown [7,12,17]. The disease is caused by *Helminthosporium turcicum* Pass. (Syn. *Exserohilum turcica* (Pass.) Shoemaker, *Drechslera turcica* (Pass.) Subram. & Jain., perfect state: *Trichometasphaeria turcica* Luttrell =*Setosphaeria turcica* (Luttrell) Leonard & Suggs [9,11,13,14,17]. Young seedling, when repeatedly infected, can be killed . When the pathogen attacks older plants under favorable conditions it can reduce yields by two-thirds or more [3,6,7]. In Taiwan, heavy infections had been recorded in the 1960s when the susceptible cultivars Tainan 5 and Tainan 11 were extensively grown [19]. Fungicides maneb and zineb were recommended to the farmers for the control of this disease since then [1].

Three physiologic races of *H. turcicum* have been reported in the U. S. [2,15,18]. Race

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2. Plant Pathologist and Assistant, respectively, Department of Plant Pathology, TARI, Wufeng 41301, Taichung, Taiwan, the Republic of China.

1, avirulent to maize plants having gene Ht, is the most common race in the U. S. corn belt[15]. In 1972 an isolate of *H. turcicum* virulent to plants having Ht was reported in Hawaii[2]. Race 3 of *H. turcicum* was collected in South Carolina in 1976[81]; ths race is virulent to plants having genes Ht2 and Ht3 but avirulent to plants having Ht. Up to now in Taiwan, no information of races of *H. turcicum* is available.

The objectives of this study were: (i) to identify, if any, the races of *H. turcicum* and their distribution pattern, and (ii) to evaluate their reaction in the currently grown maize cultivars in Taiwan.

Materials and Methods

Sample collection:

Northern leaf blight-infected maize leaves with typical symptoms of elliptic grayish-green lesions were collected in the spring crop season (May) of 1987 from different varieties/lines at Peipu of Hualien County, Kuanshan and Zueiho of Taitung County, Shueishang, Taipau, and Putzu of Chiayi County, and the Taiwan Agricultural Research Institute of Taichung County as shown in Table 2. The samples were dried in the laboratory at room temperatures until the water in the leaf tissue also was fairly removed when the leaves turned to gray color. They were, then, placed in paper bags sealed in plastic bags and stored at 4°C for use.

Purification and increase of inoculum:

Several single lesios from each collection site and variety/line were cut seperately and then put the single lesion on the top of a filter paper saturated with sterile distilled water in petri dish. The petri dishes were kept in an incubator (Hotpack-Model 332620) programmed for 8-hrs of light/day at 16°C. Abundant sporulation on the lesion surfaces was observed after two days[20]. If necessary, 1 to 3 times of repeat inoculations, following the methods described later in this report, could be conducted by inoculating the freshly-produced spores to the leaves of the susceptible cultivar Tainan 5 until the inoculum for each isolate was sufficient for race identification studies on the differential varieties and inoculation tests on the currently-grown cultivars.

Test plants:

Seeds of the differential varieties for race identification as shown in Table 1 were provided by Dr. H. S. Lu of the Agronomy Department, TARI from the University of Illinois. Seeds of the currently-grown cultivars as shown in Table 3 were purchased from Taiwan Seed Service or from local markets except Tainung 351 which was obtained from our breeders. Plants were grown in the greenhouse in black disposable plastic pots (diam. 6-in), 3 plants/pot, containing clay loams. In order to get plants with normal growth, one to two applications, depending upon the appearance of the plants, of chemical fertilizer (Taiwan Fertilizer Co. No. 39) were necessary. Five to 6-week-old plants (approximately 6—7 leaves/plant) were used for inoculations throughout the experiments.

Inoculations:

Inocula were prepared from the freshly produced spores, as described previously,

by agitating the sporulating lesions/leaves in a small amount of distilled water containing 100ug/ml of Tween 80. The concentrated spore suspension then were diluted to ca. 10,000~20,000 spores/ml for inoculations. Greenhouse-grown plants of the differential varieties for race studies were seperately sprayed with the spore suspension of each isolate using hand-drived plastic atomizers in floriculture until the leaf surfaces were completely wet [20]. Because there was only one race identified in each collection site, the isolates or lesions of the same place from different varieties/lines were mixed in the preparation of inoculum for testing the reaction of the grown cultivars to the three identified races of *H. turcicum*. Inoculated plants were covered with transparent plastic bags to keep high moist for infection and incubated in a local-made walk-in growth chamber programmed for 24/20°C (day/night) with 12-hrs of light illumination (11,000 lux at the top of the plants) per day [20]. Plastic covers were removed after 24 hours. Inoculated plants were either remained in the same growth chamber for observation or returned to the greenhouse if the temperatures were not higher than 28°C and lower than 18°C.

Host reaction :

Lesion types were recorded two weeks after inoculation. The susceptible (S) reactions were large, elliptic grayish-green lesions ranging from 3 to 5 cm or larger whereas the resistant (R) reactions were chlorotic type of lesions, and sometimes necrosis might be observed in the center of the lesions. However, they were much smaller than the susceptible ones [4,5,8] (Fig. 1).

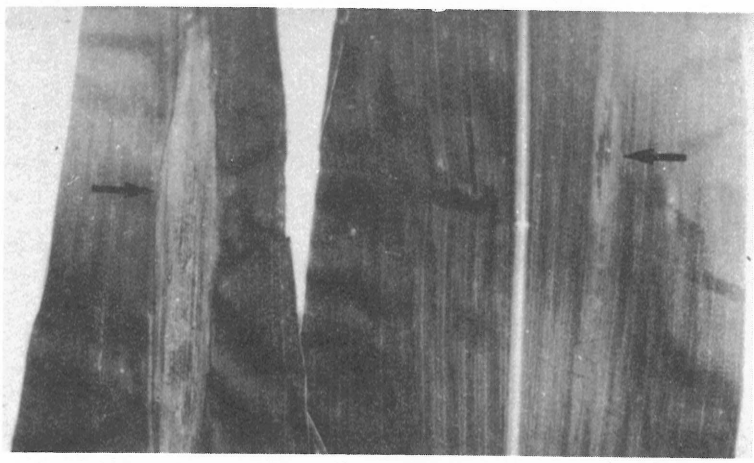

Fig 1. Susceptible (left) and resistant (right) reactions (arrows indicated) of northern leaf blight in maize leaves.

Race identification :

The reactions of the three U. S. reported races of *H. turcicum* in the differential varieties were summarized in Table 1 [2,8,10,15,16,18]. Race 1 produced susceptible (S) lesions on W153R, B37, and A619 and resistant (R) lesions on the rest of the differential varieties. Race 2 produced S lesions on W153R, B37, A619 and W 153 RHt1 A, and R lesions on the others. Race 3, on the contrary to race 2, was resistant on W153R, B37,

A619, W153RHt1A and susceptible on the others. By comparing the reactions of the isolates in this study with the reactions in Table 1, the presence of the races of *H. turcicum* in Taiwan was identified.

Table 1. Reaction of the three races of *H. turcicum* reported in the U. S. in the maize differential varieties [2,15,18]

Differential variety	Reaction of *H. turcicum**		
	Race 1	Race 2	Race 3
W153R, B37, A619	S	S	R
W153RHt1A	R	S	R
W153RHt2, B37Ht2, A619Ht	R	R	S
W153RHt3, B37Ht3, A619Ht3	R	R	S

* R : resistant reaction or small chlorotic type of lesions.
S : susceptible reaction or large elliptic grayish-green type of lesions.

Results

Races of *H. turcicum* and their distribution in Taiwan :

This is the first report on the races of *H. turcicum* in Taiwan. Based on the lesion types on the differential varieties, three physiologic races of *H. turcicum* were identified. All isolates collected from varieties/lines in the eastern parts of Taiwan including Peipu of Hualien County, and Kuanshan and Zueiho of Taitung County were all belonged to race 1. Isolates from cultivars Tainan 5 and Tainan 16 at Shueishang, Taipau and Putzu of Chiayi County were all belonged to race 2 and those collected from cultivar Tainan 5 at the TARI of Taichung County were all belonged to race 3 (Table 2, Fig. 2).

Table 2. Reaction of the differential varieties to the infection by the isolates of *H. turcicum* collected from different maize cultivars/lines in different maize growing areas of Taiwan, 1987

Collection site	Variety/line	Isolates collected	Reaction on differential varieties*				Race designation
			W153R B37A619	W153R Ht1A	W153RHt2 B37Ht2 A619Ht2	W153RHt3 B37Ht3 A619Ht2	
Peipu, Hualien	PS 1	10	S	R	R	R	Race 1
Peipu, Hualien	Tainan 16	11	S	R	R	R	Race 1
Kuanshan, Taitung	F 12	6	S	R	R	R	Race 1
Kuanshan, Taitung	PS 1	10	S	R	R	R	Race 1
Kuanshan, Taitung	Tainan 16	7	S	R	R	R	Race 1
Zueiho, Taitung	Mexico	9	S	R	R	R	Race 1
Shueishang, Chiayi	Tainan 16	15	S	S	R	R	Race 2
Taipau, Chiayi	Tainan 5	15	S	S	R	R	Race 2
Putzu, Chiayi	Tainan 5	15	S	S	R	R	Race 2
TARI, Taichung	Tainan 5	12	R	R	S	S	Race 3

*R : resistant reaction or small chlorotic type of lesions.
S : susceptible reaction or large elliptic grayish-green type of lesions.

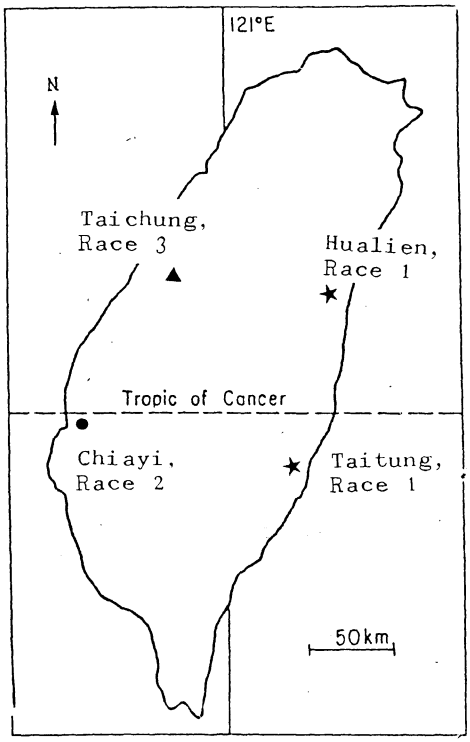

Fig 2. Distribution of races 1, 2, and 3 of *H. turcicum* in Taiwan, 1937.

Reaction of the currently grown cultivars :

Cultivars Tainan 5, Tainan Selection 10, Tainan 11, and Tainan 16 were susceptible to all the three races. Sweet corn cultivar Honey 236 was susceptible to race 2 and 3, but resistant to race 1 collected at Peipu of Hualien County and Zueiho of Taitung County. However, it is susceptible to the race 1 collected at Kuanshan of Taitung County. Cultivar Tainung 351 was resistant to all races except some isolates of race 2 collected at Shueishang in Chiayi (Table 3).

Table 3. Reaction of the currently grown maize cultvars in Taiwan to the infection by the three identified races of *H. turcicum*

Race	Source	Reaction of the cultivar*					
		Tainan 5	Tainan Sel. 10	Tainan 11	Tainan 16	Honey 236	Tainung 351
1	Peipu, Hualien	S	S	S	S	R	R
1	Kuanshan, Taitung	S	S	S	S	S	R
1	Zueiho, Taitung	S	S	S	S	R	R
2	Shueishang, Chiayi	S	S	S	S	S	S, R
2	Taipau, Chiayi	S	S	S	S	S	R
2	Putzu, Chiayi	S	S	S	S	S	R
3	TARI, Taichung	S	S	S	S	S	R

*R : resistant reaction or small chlorotic type of lesions.
S : susceptible reaction or large elliptic grayish-green type of lesions.

Discussion

This study revealed that all the three physiologic races of *H. turcicum* reported in the U. S. were all existed in Taiwan (Table 1, 2). Growing varieties did not influnce the distribution pattern of race within a certain area and there was only one race was identified within each area such as eastern, southern or central Taiwan (Table 2, Fig. 2). This may due to the small sample size. More observations or samplings from different varieties/lines in various years and seasons are needed. Cultivar Honey 236 was resistant to some isolates but susceptible to the other isolates of race 1. Tainung 351 also was observed with two types of lesions when it was inoculated with race 2 collected from cv. Tainan 16 at Shueishang of Chiayi County. Therefore, if Honey 236 and Tainung 351 were used as differentials more races of *H. turcicum* might be identified.

Cultivars Tainan 5, Tainan Selection 10 and Tainan 11 were all susceptible to the three races. Therefore. the prevalence of northern leaf blight in the 1960s were due to the lack of resistance in the growing cultivars during that time. When the resistant cultuvar Tainung 351 was released and extensively grown in the past few years the importance of northern leaf blight decreased since then. At the time of sampling for this study in Taitung area, northern leaf blight was very severe in a nickname variety Mexico. Therefore, if the acreage of this variety increased with time the importance of northern leaf blight might be increased. The cultivation of resistant variety Tainung 351 in the past few years also proved that breeding and growing resistant varieties is the most effective and economic strategy among the various disease control measures.

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臺灣玉米煤紋病菌之生理小種[1]

葉忠川　蔡阿輝[2]

摘　要

玉米煤紋病（northern leaf blight）是玉米重要的葉部病害，玉米煤紋病菌（*Helminthosporium turcicum*）在美國報告有三種生理小種（physiologic race），但本省則未有研究，因此本試驗之目的在探討本省之玉米煤紋病菌的生理小種及其分佈做爲抗病育種之指標，並瞭解目前推廣之玉米栽培品種對各生理小種之抵抗性。於1987年春作，從花蓮之北埔、臺東之關山及瑞和、嘉義之水上、太保與朴子及臺中本所等地採得煤紋病病葉，將不同地點及不同品種／系上之各單一病斑分別處理當做一個菌株，病斑洗淨並陰乾後保存在 4°C 恒溫箱備用。若再將病葉重新在 16°C 保持濕潤二天後，病斑表面即會產生大量之分生胞子，以供調配接種源之用。源自單一病斑的每一菌株，可採用本試驗之接種方法在感病品種臺南五號上重複接種數次以增加接種源之量，直到有足量之接種源爲止。供試植物包括表一或表二中所列之判別品種及表三所列之栽培種，均在溫室以盆缽盛土培養至六至七個葉片（約四～五週）時，以花卉栽培用之小型指壓塑膠瓶式噴霧器噴洒各菌株之胞子懸浮液（以含 100μg/ml Tween 80 之蒸餾水配製，濃度爲一萬至二萬個胞子／ml），直到葉片表面完全溼潤爲止，接種後之植株在日溫 24±1°C 夜溫 20±1°C 之生長箱（白天光照12小時，11,000lux）以透明塑膠袋覆蓋保溼24小時後除去，以後植株可留置在生長箱或者當溫室內之氣溫在 28～18°C 之間時也可放在溫室內，接種後二週調查及記載病斑形態，大型紡錘形青枯病斑者爲感病（S）反應，小型黃化形病斑者爲抗病（R）反應。表一所列爲文獻上所記載在判別品種上各生理小種之反應，表二爲本研究接種試驗所得之結果，兩者比對即可知本省玉米煤紋病菌之生理小種。結果顯示美國所報告的三種生理小種本省均有存在，而且生理小種之分佈有地域性，本省東部包括花蓮之北埔及臺東之關山與瑞和所採集者均爲 Race 1，嘉義之水上、太保及朴子者均爲 Race 2，而於本所所採集者均爲 Race 3，但試驗中所採集之品種／系並沒有影響生理小種在同一地區內之分佈，亦即在同一地區之不同的品種／系上所獲得之菌株均爲同一生理小種。目前之栽培品種包括臺南五號、臺南選十號、臺南十一號及臺南十六號對三個生理小種均無抵抗性。超甜玉米236號僅抗來自北埔及瑞和之 Race 1的某些菌株，對 Race 2 及 3 則無抵抗性。臺農351號是目前最具抵抗性之品種，可抗 Race 1 及 3，僅對來自嘉義水上之 Race 2 的某些菌株表現感病反應。本研究顯示過去本省煤紋病猖獗，乃因栽培品種缺乏抵抗性所致，同時證明栽培抗病品種如臺農351號爲病害防治最經濟而且有效之方法。

1.臺灣省農業試驗所　研究報告第 1391 號。本研究部份經費承行政院農業委員會補助，謹此致謝。

2.本所植物病理系研究員及助理。臺灣省　臺中縣　霧峰鄉。