Short communication. *Fusarium graminearum* Schwabe, a maize root and stalk rot pathogen isolated from lodged plants in northwest Spain

J. L. Andrés Ares*, R. C. Alonso Ferro, L. Campo Ramírez and J. Moreno González

Centro de Investigaciones Agrarias de Mabegondo. Ctra. Betanzos-Santiago, km 7.5. 15318 Abegondo (A Coruña). Spain

Abstract

On September 2001 a survey of lodged maize (Zea mays L.) plants to identify the main root rot pathogens related to maize lodging was carried out in 23 cornfields of Abegondo (A Coruña, Spain). From 328 maize plants showing lodging, 33 isolates of the following potential maize pathogens were collected: *Fusarium semitectum* Berk & Rav., *F. graminearum* Schwabe, *F. culmorum* (W. Smith) Sacc., *F. solani* Appel & Wall and *F. moniliforme* Sheldon. The inoculation of four strains of *F. moniliforme*, three of *F. graminearum*, one of *F. culmorum* and one of *F. semitectum* on two maize inbreds with different susceptibility to lodging —EC23D and EC136 as susceptible and resistant inbreds respectively— showed *F. graminearum* Schwabe as the most pathogenic fungus considering either root rot symptom or seedling growth reduction. The importance of considering the susceptibility to root rot pathogens in maize breeding programs for resistance to lodging is also discussed.

Key words: Zea mays, root rot, root lodging, Galicia.

Resumen

Fusarium graminearum Schwabe, patógeno causante de podredumbre de tallo y raíz aislado de plantas de maíz con encamado en el noroeste español

En septiembre de 2001 se realizó un muestreo de plantas con encamado en 23 campos de la comarca de Abegondo (A Coruña), con la finalidad de identificar los principales hongos patógenos relacionados con el encamado del maíz. De 328 plantas de maíz con síntomas se obtuvieron 33 aislados de los siguientes patógenos potenciales: *Fusarium semitectum* Berk & Rav., *F. graminearum* Schwabe, *F. culmorum* (W. Smith) Sacc., *F. solani* Appel & Wall y *F. moniliforme* Sheldon. La inoculación de cuatro cepas de *F. moniliforme*, tres de *F. graminearum*, una de *F. culmorum* y una de *F. semitectum* sobre dos variedades de maíz con diferente susceptibilidad a encamado —EC23D como susceptible y EC136 como resistente— mostró a *Fusarium graminearum* Schwabe como el hongo de mayor poder patógeno, considerando tanto los síntomas como la reducción de desarrollo de la plántula. Se discute la importancia de considerar la susceptibilidad a patógenos productores de podredumbre de raíz en los programas de mejora de la resistencia al encamado en maíz.

Palabras clave: Zea mays, podredumbre radicular, encamado raíz, Galicia.

Lodging is a serious problem in maize that makes mechanical harvest difficult and reduces dry matter assimilation. Evaluation of lodging resistance under natural conditions is difficult because lodging varies with meteorological conditions and growing environments, and significant lodging may not occur every year. Therefore, methods for evaluating maize lodging resistance based on mechanical or morphological traits have been proposed by several authors (Ishige *et al.*, 1983; Melchinger *et al.*, 1986; Koinuma *et al.*, 1990; Hebert *et al.*, 1992). With the general aim of studying and comparing different methods of evaluation of maize lodging resistance the present work was carried out to specifically study the relationship between root rot pathogens and lodging.

Root rots are among the least studied diseases of corn. Fungi recovered from roots include seedling blight and stalk rot pathogens as well as secondary invaders. *Fusarium* spp. are commonly isolated from corn roots,

^{*} Corresponding author: jose.luis.andres.ares@xunta.es Received: 06-10-03; Accepted: 03-02-04.

being *F. oxysporum* Schlectend and *F. solani* Sacc. the most commonly reported. Other pathogens including *F.* graminearum Schwabe, *F. acuminatum* Ellis & Everh. and *F. equiseti* (Corda) Sacc. *F. moniliforme* J. Sheld., *F. proliferatum* (Matsushima) Nirenberg, and *F. sub*glutinans (Wollenweb. & Reinking) tend to be associated with root rots in young plants (White, 2000). When symptoms occur, they range from a very slight brownredish colour to dark black discoloration of completely rotted roots. Roots infected by *F. graminearum* are often red or pink. *Fusarium* spp. can also be isolated from symptomless roots (White, 2000).

F. graminearum has been isolated from maize root rots by some authors (Hulea *et al.*, 1968; Mohamed *et al.*, 1968; Palmer and Kommedahl, 1969; Kommedahl *et al.*, 1987) and from maize root and stem rots by others (Ramsey, 1990). This fungus has also been reported as pathogenic on maize in inoculation tests (Palmer and Kommedahl, 1969; Kommedahl *et al.*, 1987); and has also been referenced as a collar rot pathogen in Andalucía (south of Spain) and Cataluña (northeastern Spain) (De Andrés *et al.*, 1998), and as a stem rot pathogen in Cataluña (Marín *et al.*, 1992).

F. moniliforme J. Sheld is also responsible for root rots in maize (Paul, 1975; Kommedahl *et al.*, 1987), but seems to be less pathogenic than *F. graminearum* (Kommedahl *et al.*, 1987). In Spain, *F. moniliforme* has only been reported as a maize stem pathogen in Cataluña (Marín *et al.*, 1975).

To identify the main root rot pathogens related to lodging, 328 lodged plants were sampled from 23 cornfields in Abegondo (A Coruña) one month after pollination in 2001. Fragments of the stem and root basis in the affected plants were prepared for fungi isolation. The surface of these fragments was disinfected with 0.6% sodium hypochlorite for 4 min and plated on PDA (potato dextrose agar) (Rapilly, 1968). The fungi were grown under laboratory conditions and microscope observations were carried out every 24 h during one week. *Fusaria* isolates were classified according to Nelson *et al.* (1983).

The maize inbreds EC23D and EC136, susceptible and resistant to lodging respectively, were used in the experiment. The inoculation test was run in a greenhouse at 16-22°C. Plants were grown in plastic trays containing a mixture of peat and sand previously sterilised at 120°C for 45 min (1:1, w:w). A randomized complete block design with three replications per isolate-variety combination and 10 plants per plot were used. Non inoculated controls of each inbred were also included. The isolates specified on Table 1 were grown on PDA medium (Rapilly, 1968) at 24°C for 7 days. The inoculum was prepared by shaking 100 ml of sterile water per petri dish (18 ml of medium) with the grown isolate. The suspension was shaken for 1 min and adjusted to 105 macroconidia -for F. graminearum, F. culmorum and F. semitectum- or microconidia ---for F. moniliforme--- per ml using a Burker camera. At the stage of four leaves, each plant was inoculated by placing 5 ml of the solution into the seedling collar with a sterile micropipette. Disease readings were registered 21 days after the inoculation. Roots of each plant were washed and the percentage of the root rotted area was assessed (Turner and van Alfen, 1983). The square root transformation was applied to the raw data, since it gave the best transformation for normality. Individual plants were also weighted after washing and drying the roots with a precision scale (0.01 g). The Waller-Duncan's multiple range test (at P = 0.05) was performed on the transformed disease severity and on the plant weight variable, using SAS 8.2, Cary, NC software.

The potential maize pathogens isolated from the lodged plants are listed on Table 1. *F. moniliforme* was the

Table 1. Potential maize pathogens isolated from collar and roots of maize lodged plants

Fusarium species	Number of isolates ¹	Potential pathogen	Reference
F. semitectum	4	Yes	White, 2000
F. graminearum	6	Yes	Palmer and Kommedahl, 1969
F. culmorum	2	Yes	Oswald, 1950
F. sambucinum	2	No	
F. lateritium	1	No	_
F. solani	1	Yes	Mohamed, 1990
F. moniliforme	17	Yes	Kommedahl et al., 1987
Total	33		_

¹ Strains isolated from 23 samples included 328 collar and root rotted plants.

Strain	Fusarium species	Root area showing symptoms (%)		
		Inbred EC23D*	Inbred EC136*	
221 Fm	F. moniliforme	18.3 b	3.8 b	
243 Fm	F. moniliforme	6.2 cd	2.5 b	
244 Fm1	F. moniliforme	13.7 bc	2.3 b	
244 Fr1	F. graminearum	12.2 bc	6.8 b	
219 Fr1	F. culmorum	9.8 c	4.5 b	
241 Fr1	F. graminearum	55.8 a	26.3 a	
241 Fm1	F. moniliforme	6.4 cd	4.2 b	
218 Fr1	F. semitectum	6.5 cd	6.5 b	
221 Fr1	F. graminearum	20.0 b	5.3 b	
Control	Not inoculated	1.0 d	2.0 b	

Table 2. Pathogenicity of nine strains isolated from collar and roots of maize lodged plants, on two maize inbreds with different resistance to root lodging, 21 days after inoculation

Inbreds EC23D (susceptible to lodging) and EC136 (resistant). * Means of the same column followed by the same letter are not significantly different based on Waller-Duncan's multiple range test at P = 0.05.

most frequently isolated species, followed by *F. gra*minearum and *F. semitectum*.

The pathogenicity of nine selected potential pathogenic isolates on each of the two maize inbreds is shown on Table 2. *F. graminearum* (241 Fr1 and 221 Fr1) and F. moniliforme (221 Fm) were the two fungi that induced significant disease symptoms on the susceptible inbreds, 241 Fr1 induced significant disease symptoms on the resistant inbred, but the disease level was lower than that registered on the susceptible line. Symptoms induced by the other strains did not differ significantly from the controls.

Growth reduction induced by the disease is presented in Table 3. Four of the nine isolates produced sig-

nificant growth reduction on the susceptible maize seedlings. Two of them were *F. graminearum* isolates, which had also induced severe root rot symptoms (241 Fr1 and 221 Fr1); the other two were *F. moniliforme* and *F. culmorum* isolates, which did not induce severe root rots symptoms. The most pathogenic isolate, 241 Fr1—*F. graminearum*—, also produced the highest seedling growth reduction. Plant weights of the resistant inbred, after inoculation of all the strains, did not differ from the control.

Though root rot diseases, much like stalk rot diseases, result from the activities of numerous organisms including different fungi, bacteria, nematodes and root-feeding insects, in this study root rot was

Table 3. Plant weight average of two maize inbreds with different resistance to lodging after being inoculated with nine strains of different potential pathogens isolated from collar and roots of maize lodging plants

Strain	Fusarium species	Plant weight (g)		
		Inbred EC23D*	Inbred EC136*	
221 Fm	F. moniliforme	25.58 a	25.82 a	
243 Fm	F. moniliforme	17.55 bc	25.80 a	
244 Fm1	F. moniliforme	21.52 ab	23.73 ab	
244 Fr1	F. graminearum	21.85 ab	12.89 c	
219 Fr1	F. culmorum	17.07 bc	25.84 a	
241 Fr1	F. graminearum	11.94 с	21.77 ab	
241 Fm1	F. moniliforme	21.44 ab	21.94 ab	
218 Fr1	F. semitectum	22.76 ab	15.27 bc	
221 Fr1	F. graminearum	18.07 b	20.5 ab	
Control	Not inoculated	26.8 a	18.62 b	

Plant weight average of 30 plants (three replications of 10 plants) of the inbreds EC23D (susceptible to lodging) and EC136 (resistant). * Means of the same column followed by the same letter are not significantly different based on Waller-Duncan's multiple range test at P = 0.05.

mainly produced by the action of a *Fusarium* species: *F. graminearum*. This fungus was also mentioned as a root rot pathogen isolated from lodged plants by several authors (Hulea *et al.*, 1968; Ramsey, 1990). The term «stalk rot« is often used to include stalk breakage, stalk lodging, premature death of plants, and occasionally even lodging (White, 2000); this is specially important in the case of this study because *F. graminearum* is a stalk rot pathogen as well as a root rot fungus.

Root rots may be associated with growth reduction of maize seedlings, however, these symptoms are more related to seedling blights produced more frequently by *F. moniliforme* (Futrel and Kilgore, 1969) and, to a lesser extent, by *F. graminearum* (White, 2000), which differs from the data discussed in this work. Two of the four isolates which significantly reduced maize seedling weight were *F. graminearum* and only one of them was classified as *F. moniliforme* Sheldon. These two *F. graminearum* also produced the most clear root rot symptoms on EC23D.

Lodging is a great problem in maize fields in northwest Spain and effective breeding programs are not easy to carry out, due to the difficulty of the evaluation of lodging resistance. If further research confirm that lodging is related to susceptibility to *F. graminearum* the assessment of this character would also be important for breeding programs.

Acknowledgements

This work has been financed by the Spanish Ministry of Science and Technology (project RTA01-140).

References

- DE ANDRÉS M.F., GARCÍA ARENAL F., LÓPEZ M.M., MELGAREJO P. (eds), 1998. Patógenos de plantas descritos en España. SEF-MAPA. 301 pp.
- HÉBERT Y., BARRIERE Y., BERTHOLEAU J.C., 1992. Root lodging resistance in forage maize: Genetic variability of root system and aerial part. Maydica 37, 173-183.
- HULEA A., BUNESCU S., SANDRU I., TIRCOMNICU M., PITICAS G., SCHMIDT.R., 1968. Investigations of stalk and root rot of maize under the environmental conditions in Romania. Analele Institutului de Cercetari pentru Protectia Plantelor 6, 25-55.

- ISHIGE T., YAMADA M., SHIGA T., 1983. Screening for resistance to lodging based upon discriminant function value in maize and biometrical analysis for general components. Bulletin of the National Institute of Agricultural Sciences Series D., 3-1-1 Kannondai, Yatabe, Tsukuba, Ibaraki, Japan 35, 125-152.
- KATO A., KOINUMA K., 1999. Environmental effects on two non-destructive root lodging related indicators and correlations between hybrids and parental inbred lines in maize. Maydica 42, 167-174.
- KOINUMA K., INOUE Y., KATO A., 1990. Evaluation of lodging resistance of maize (*Zea mays* L.) by the measurement of the horizontal pull resistance. Bulletin of the National Grassland Research Institute, Nishinasuno, Japan, 43, 23-29.
- KOMMEDAHL T., SABET K.K., BURNES P.M., WINDELS C.E., 1987. Occurrence and pathogenicity of *Fusarium proliferatum* on corn in Minnesota. Plant Dis 71, 281.
- MARIN J.P., SEGARRA J., ALMACELLAS J., 1992. Enfermedades de los cereales en Cataluña durante 1988-90. Invest Agr: Prod Prot Veg 7(2), 261-275.
- MELCHINGER A.E., GEIGER H.H., SCHMIDT G.A., 1986. Vertical root pull resistance and its relationship to root lodging and forage traits in early maturing European inbred lines and F1 hybrids of maize. Maydica 31, 335-348.
- MOHAMED H.A., ASHOUR W.E., SIRRY A.R., FATHI S.M. 1968. Fungi causing seedling blight of corn in the United Arab Republic. Plant Dis Rep 52, 84-86.
- NELSON P.E., TOUSSOUN T.A., MARASAS W.H.O., 1983. Fusarium species. An illustrated manual for identification. Pennsylvania State University Press, 193 pp.
- OSWALD J.W., 1950. Etiology of cereal root rots in California. Hilgardia 19(15), 447-462.
- PALMER L.T., KOMMEDAHL T., 1969. Root-infecting Fusarium species in relation to rootworm infestations in corn. Phytopathology 59, 1613-1617.
- PAUL V., 1975. Root rot of maize caused by *Fusarium moniliforme* Sheldon. Proc Third EPPO Conference on Pathological Organisms in Cereal Monocultures. EPPO Bulletin 5, 415-417.
- RAMSEY M.D., 1990. Etiology of root and stalk rots of maize in north Queensland. Disease development and associated fungi. Australas Plant Path 19, 2-12.
- RAPILLY.F., 1968. Les téchniques de mycologie en Pathologie Végétale. Ann Epiphyties 19, 102 pp.
- SANGUINETTI M.C., GIULIANI M.M., GOVI G., TU-BEROSA R., LANDI P., 1998. Root and shoot traits of maize inbred lines grown in the field and in hydroponic culture and their relationships with root lodging. Maydica 43, 211-216.
- TURNER V., VAN ALFEN N.K., 1983. Crown rot of alfalfa in Utah. Phytopathology 73, 1333-1337.
- WHITE D.G., 2000. Root rots. Compendium of corn diseases. Third edition. APS Press. 78 pp.