KERNEL SMUT OF BARNYARD GRASS (ECHINOCLOA CRUS-GALLI) IN EGYPT

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Abstract

Echinocloa crus-galli (Barnyard grass) is considered one of the most serious weeds in rice fields. Three years ago some smutted kernels of E.crus-galli. were observed. The causal fungus was isolated in Egypt and identified as *Ustilago trichophoron* (Link) Konicke det by Dr. J.E.M. Mordue (International Mycological Institute, U.K). The fungus produces sori in scattered flowers and on stems as bullate bodies from a few mm in diameter to 10 cm long swellings covered by a hispid membrane composed of an inner fungal layer and an outer layer of host origin. This study included different methods of inoculation to find the techniques to control or to minimize the seed residuals of this kind of weed in the fields and to avoid using herbicide as possible.

In the greenhouse, seeds of *E.crus-galli* were soaked either in suspensions of ustilospores or sporidia or incorporated directly with dry spores prior to seeding. Low disease incidence and severity of infection were observed which did not exceed 3.0 and 3.5% respectively. In case of injection at booting stage or spray the sporidial suspension just after heading (flowering) or one week later, the highest disease severities of 47.5%, 56% and 31 respectively were observed. However, no smutted kernels appeared in case of spraying spore suspension at either tillering or maturity stages.

Under field conditions injected plants at booting or plants sprayed at flowering stage resulted in a disease severity of 25.5% and 44% respectively. No symptoms were observed on the tested four rice cultivars namely, Giza 171, Giza 176, Giza 177 and Giza 181 subjected to the same treatments, indicating the specificity of the pathogen to this weed.

INTRODUCTION

Weeds are one of the most serious constraints to rice production in Egypt as well as in other countries. Among the weed species that affect rice is barnyard grass (*Echinochloa crus-galli*). It grows under varied climates and in many agronomic cropping systems and is one of the worst weeds in the world (Holm et al., 1977). Singh et al., (1985) reported that 1 and 2 barnyardgrass seedlings/hill reduced paddy yield by 20.2 and 37.5% respectively. The increase in the number of barnyardgrass from 0 to 12 plants/m^2 resulted in gradual and significant decrease in height,

number of tillers/hill, leaf area index and panicle weight of rice (Hassan and Rao, 1994). Hand weeding is becoming increasingly expensive and rural labors are unavailable when needed most. This often delays weeding and the crop suffers.

It has been found that *Echinochloa* spp. is attacked by a smut disease. The causal fungus was identified as *Ustilago trichophoron* and recorded in some countries (Fullerton & Longdon, 1969; Duran, 1973 & 1987 and Vanky, 1994). Fullerton & Langdon (1969) in demonstrating structural and developmental similarities in sori of *Ustilago* species attacking *Echinochloa* spp. concluded that a number of *Ustilago* on *Echinochloa* could be referred to as one species, namely *Ustilago trichophora*. They also indicate the need for further studies emphasizing soral ontogeny in the classification of the smut fungi. This study was carried out to identify the causal fungus of kernel smut of *E.crus-galli* and to obtain more information about the best inoculation method in order to use it as a mycoherbicide to eradicate or at least minimize the seed production of *Echinochloa* spp. in the soil and to avoid pesticide pollution.

MATERIALS AND METHODS

Preparation of the inoculum:

Samples from smutted seeds (sori) were collected, sterilized and opened under aseptic conditions. The ustilospores (Fig. 1-B) were distributed on the surface of Petridishes containing potato dextrose agar (PDA). The inoculated dishes were left in the incubator at 28°C for one week. Mass of secondary sporidia (Fig. 2A) were collected by brushing in tap water and a sporidial suspension was prepared for artificial inoculation ($5x10^6$ sporidia/ml). On the other hand, chlamydospores (ustilospores) were prepared by suspending them from sterilized sori in tap water ($2x10^5$ spores/ml) and used for artificial inoculation.

Artificial Inoculation Techniques.

1. Greenhouse Conditions:

Trays (30x20x12 cm) filled with loamy soil were used to grow treated seeds of barnyard grass as follows:

A. Seeds of barnyardgrass were soaked in ustilospore or sporidial suspension for 24 hrs, then transplanted after germination.

B. Barnyardgrass seeds were incorporated with powders of ustilopores or collected sporidial spores from Petri-dishes and planted immediately after incorporation.

C. Healthy seeds were soaked in water for 24 hrs and transplanted after germination, then the following inoculation methods were carried out using sporidia or ustilospore suspension as follows:

a) Sprayed at tillering stage using plastic sprayer.

b) Injected at booting stage by a syringe.

c) Sprayed just after complete heading at anthesis.

d) Sprayed one week after heading.

e) Sprayed after complete heading (ripening).

f) Plants were left free from any inoculation as control.

Four rice cultivars, namely, Giza 171, Giza 176, Giza 177 and Giza 181 were tested as host plants using the same above mentioned inoculation methods under the same environmental conditions and the same period of incubation.

II. Field Experiment:

Four plots (each 4 x 5 m) were transplanted with E.crus-galli, while four other plots were transplanted with the rice cultivar Giza 177 alternatively. Labelled plants in the first plot of *E.crus-galli* were injected at booting stage with sporidial suspension, while ustilospore suspension was injected in the plants of the second plot. In the third plot, the inoculation were carried out at the evening in the middle two square meters kept under polyethylene cover, during spraying the sporidial suspensions, just after panicle emergence. The fourth plot was sprayed with ustilospore suspension. The same inoculation methods were adopted in case of rice cultivar Giza 177 at both booting and just after panicle emergence. Two plots were left free from inoculation as control, one for E.crus-galli, aua the other for the rice variety Giza 177.

The infection was estimated as a percentage representing the number of smutted panicles compared with the healthy ones, whereas the severity of infection was estimated as a number of smutted grains/panicle.

RESULTS AND DISCUSSION

The causal fungus of smutted kernels of E.crus-galli was isolated and identified as *Ustilago trichophoron* (Link) Kornicke det by Dr. J.E.M. Mordue. The ustilospores are single, globose to subglobose or ovoid 7-14 x 6-12 (mean 9.2 x 8.4) µm

The same trend was clear when plants were injected at booting stage or sprayed just after heading. However, percentage or the severity of infection were lower in the field than in the greenhouse. This may be due to the higher relative humidity in the greenhouse than in the field. Neither infected panicles nor other mentioned symptoms were observed on the four tested rice cultivars. These result indicated that the fungus *Ustilago trichophoron* is specific to *Echinocloa* spp. and support the findings of Duran (1973).

It is obvious that infection is affected by the time and method of inoculation. Under field conditions, spraying inoculum atbooting stage and just after heading led to an infection that may reach 20%. Therefore, this could be utilized as a biocontrol method of barnyard grass in providing some effect against the reproduction of the weed and reducing the number of seeds produced.

However, the presence of spores mixed with rice grains and its subsequent efficiency in controlling the weed in the field depends on its survival and other factors which need to be identified and studied further. Trials employing soil infestation with smut spores be may be useful.

Table 1. Effect of different methods of inoculation with sporidia or ustil or ustilospores of Ustilago trichophoron on Echinochloa crus-galli under greenhouse and field conditions.

Treatments	Sporidial spore suspension					Utilospore suspension				
	No.of healthy panicles	No.of smutted panicles	No.of distorted panicles	% of infection	Severity of infection	No.of healthy panicles	No.of smutted panicles	No.of distorted panicles	% of infection	Severity of infection
I. Greenhouse:										
Seed were soaked for 24 hrs	115	115	115	115	115	115	115	115	115	115
Seeds were incorporated before growing	127	127	127	127	127	127	127	127	127	127
Plants were sprayed at tillering stage	136	136	136	136	136	136	136	136	136	136
Plants were injected at booting stage	157	157	157	157	157	157	157	157	157	157
Plants were sprayed just after heading	116	116	116	116	116	116	116	116	116	116
Plants were sprayed one week from heading	130	130	130	130	130	130	130	130	130	130
Plants were sprayed after complete heading	118	118	118	118	118	118	118	118	118	118
Control (free from inoculation)	150	150	150	150	150	150	150	150	150	150
II. Field:										
Plants were injected at booting stage	220	220	220	220	220	220	220	220	220	220
Plants were sprayed just after heading	190	190	190	190	190	190	190	190	190	190
Control (free from inoculation)	200	200	200	200	200	200	200	200	200	200

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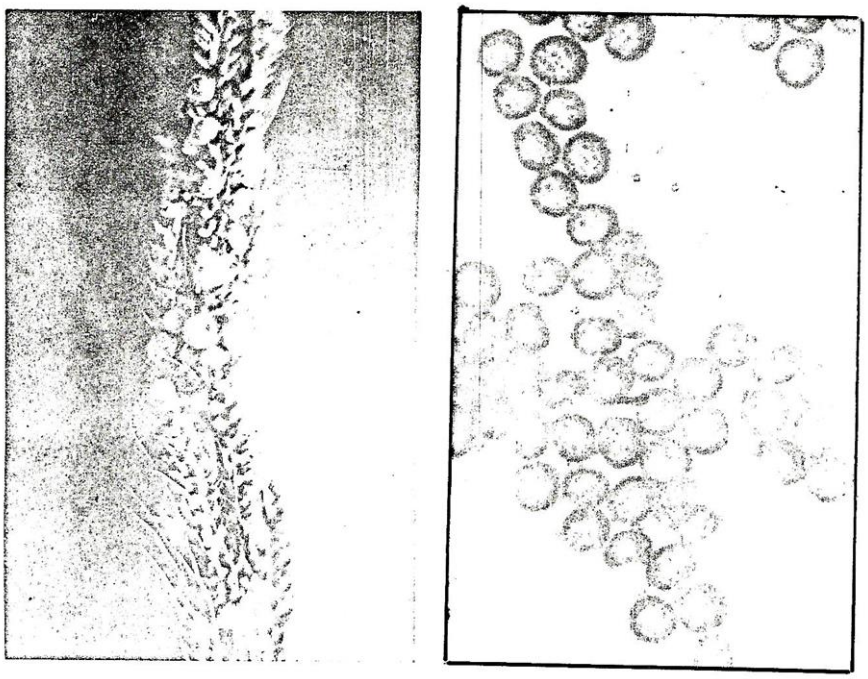

A B

Fig. 1. A: Smutted swolen seeds (Sori)

B: Ustilosspores (x 700)

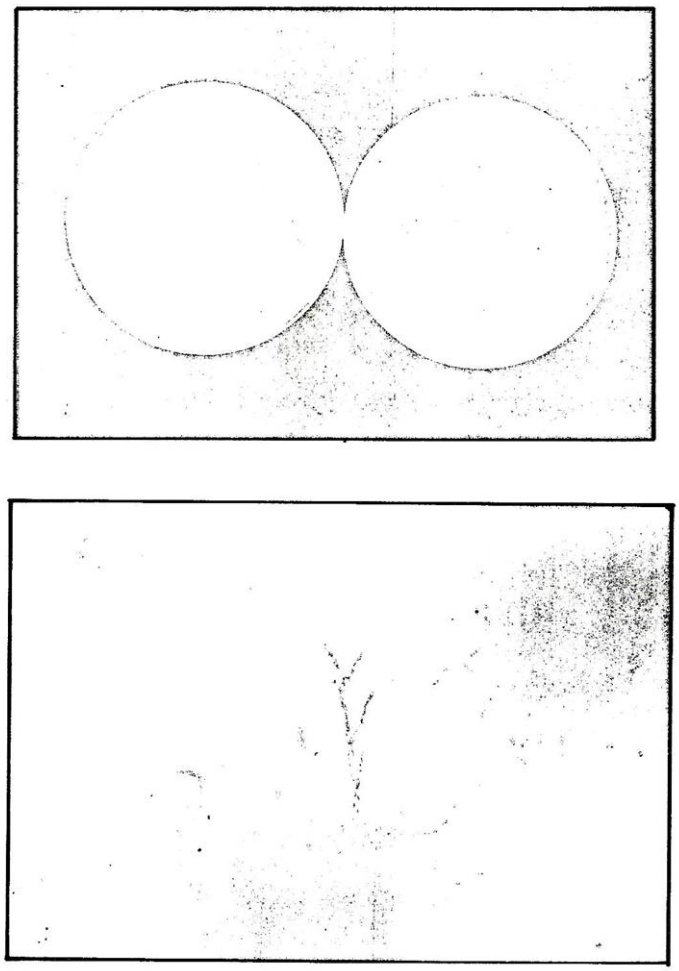

Fig. 2. A: Mass of secondary sporidia in Petri-dishes inoculated with ustilospores.

B: Secondary sporidia from Petri-dishes under the microscope (x 200).

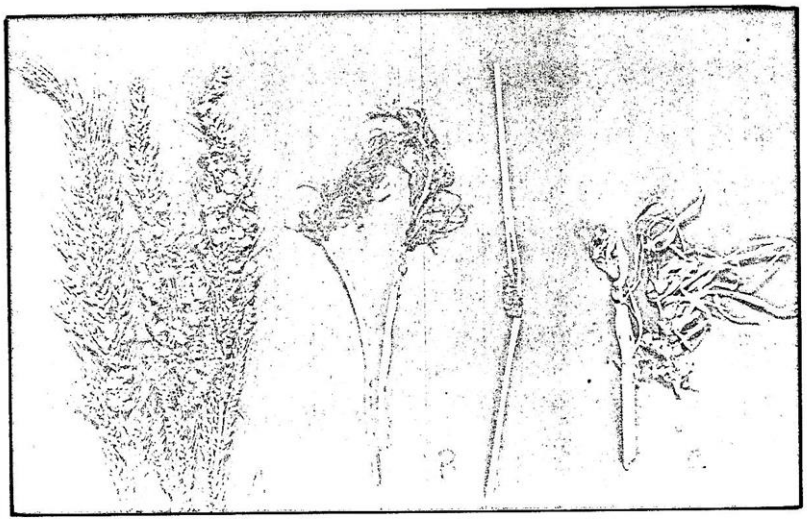

Fig. 3. A: Developed sori in seed
B: Distorted panicles
C : Swollen galls on culms and nodes

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تفحم الدنيبة فى مصر

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تعتبر حشيشة الدنيبه واحدة من أخطر الحشائش التى توجد فى حقول الأرز والتى تستخدم العديد من مبيدات الحشائش فى مقاومتها سنويا. منذ حوالى ثلاث سنوات لوحظ بعض التفحم فى الحبوب على سنابل الدنيبه حيث تم عزل الفطر المسبب وتنقيته، وتم تعريفه للمره الأولى فى مصر بواسطه معهد إنجلترا الدولى لتعريف الكائنات الدقيقة وهو فطر (يوستيلا جوتريكوفورون) والفطر يسبب تفحمات وتشوهات فى بعض الأزهار بالنوره، وعلى السيقان عادة ماتكون صغيره الحجم (عدة مليمترات فى القطر)، وقد تصل إلى ١٠ سم فى الطول. وهذه البثرات المتفحمه تكون مغطاه بغشاء داخلى من الفطر نفسه وغشاء خارجى يتكون من النبات العائل.

أجريت هذه الدراسة بغرض معرفة أكفأ طريقة فى إحداث العدوى لإستخدامها فى مقاومة حشيشة الدنيبة أو التقليل من كمية البذور الناتجة منها والتى تبقى فى الأرض للموسم القادم.

تمت الدراسة فى الصوبة بإستخدام نقع التقاوى فى معلق من الجراثيم التيلتيه أو الأسبوريدية أو خلط التقاوى مباشرة قبل الزراعة مع مسحوق كل من النوعين على حده. وقد كانت أعراض الإصابة بهذه الطرق قليلة الحدوث فقد تراوحت شدة الإصابة من صفر إلى ٣,٥٪ فقط. بينما فى حالة الحقن فى مرحلة قبل طرد السنابل مباشرة (Booting) أو الرش بالجراثيم الأسبوريديه مباشرة بعد الطرد فى مرحلة التزهير زو بعد أسبوع من الطرد، أظهرت النتائج أن شدة الإصابة عالية حيث وصلت إلى ٤٧,٥ ، ٥٦، ٣١٪ على التوالى. بينما لم تلاحظ أعراض تفحم فى حالة الرش بمعلق جراثيم النوعين على النباتات فى مرحلة التفريع أو مرحلة النضج الكامل.

كما أجريت دراسة حقليه حيث تبين أن حقن النباتات فى مرحلة قبل الطرد أو الرش فى مرحلة التزهير بالجراثيم الأسبوريديه قد أعطت إصابة تصل إلى ٢٥,٥ – ٤٤٪ على التوالى. لذا يتضح أن طريقة الرش بالجراثيم الأسبوريديه فى مرحلة التزهير أعطت أعلى إصابة يليها الحقن فى مرحلة قبل الطرد تحت ظروف كل من الصوبة والحقل.

كما تم إختبار أربعة أصناف من الأرز جيزة ١٧١ ، جيزة ١٧٦ ، جيزة ١٧٧ وجيزة ١٨١ حيث تم عدواها بنفس الطرق السابقة تحت نفس الظروف للتأكد من عدم قدرة الفطر على إصابة المحصول الرئيسى حيث لم يلاحظ أى تفحمات على أى جزء منها مما يدل على تخصص الفطر على حشيشة الدنيبه فقط