Evaluation Method of Weed Suppression by Rice Plant

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ABSTRACT: Rice and weed interference in the paddy field caused by resource competition and allelopathy. Evaluation method of active weed suppressive behavior of rice to weed was developed by eliminating light competition at soil condition. Twenty eight days old rice seedlings (6 - 7 leaf stage) which was grown at saturated, no drainage pot were clipped above $3 \sim 4$ cm from the soil surface. Weeds seeded around clipped rice stem, named ratoon seeding screening method, showed varietal suppressive difference to Echinochloa crus-galli, Echinochloa crusgalli var. praticola and Monochoria vaginalis. Potential allelopathic rice varieties, Sathi, AC1423 and PI312777 showed better suppressive activity to weed seedling growth than Nonganbyeo and Keumobyeo. Weed suppression of one plant of rice cultivars could be evaluated by the cell size of 2.5×2.5 cm at rice clipping of seedling 29 days after rice seeding.

Keywords: rice, allelopathy, weed suppression, rice-weed competition, ratoon seeding screening, *Echinochloa crusgalli*, *Monochoria vaginalis*

A Iternative technologies for weed control are necessary to combat the rising cost of labor and herbicides. Weed management tools that can reduce chemical inputs will also decrease environmental degradation caused by intensive herbicide application. Enhancement of weed competitive ability of rice through breeding is one of the roads that can be followed to achieve the goal of sustainable weed management (Rice, 1995; Bhowmik, 2003).

Allelopathy is defined as "any direct or indirect harmful or beneficial effect by one plant on another through the production of chemical compounds that escape into the environment" (Rice, 1984). Over the last decade several studies have shown that some crop cultivars are allelopathic and that their inhibitory effects on weeds do play a role under field conditions (Dilday et al., 1991; Fujii, 1993; Kim et al., 1996; Churg, 1997; Olofsdotter et al., 1995, 1999; Wu et al., 1999). Recently, crop allelopathy research also includes identification of the responsible chemicals, released from crops, that are responsible for the realized weed suppression (Rimando et al., 2001; Kato-Noguchi & Ino, 2003; Wu et

al, 2000; Chung et al., 2003), and genetical identification of allelopathy in rice (Jensen et al, 2001; Ebana et al., 2001) and in wheat (Wu et al, 2003).

Various methods have been developed for efficient and consistent mass screening to identify varietal differences with weed suppressing ability that will serve as a basis for future breeding programs (Dilday et al, 1991; Fujii et al, 1993; Navarez & Olofsdotter, 1996; Wu et al., 1999; Lee et al., 2003). These techniques concentrated to separate the effect of allelopathy from competition or to find potential allelochemicals, which cannot be accomplished in field experiment. To obtain entire understanding of allelopathy it is therefore important to make the experimental system more and more complex and it goes to more "nature like". It is therefore important to also have methods to study allelopathy in a soil based experimental protocol to ensure that laboratory methods are reflecting allelopathy in field. Such experimentation can be used for bridging gaps, such as understanding of weed suppression mechanisms, between laboratory and field.

This study aims to develop a soil based pot screening method for verifying rice cultivars with weed suppressing ability, particularly potential allelopathy.

MATERIALS AND METHODS

Greenhouse experiments were conducted to develop a screening method for determining the suppressing effects of ten rice cultivars, AC1423, Chungmyungbyeo, Daelipbyeol, Hwajinbyeo, IAC165, Keumobyeo, Nonganbyeo, PI312777, Sathı, and Taichung Native 1, on the three weed species, *Echinochloa crus-galli, Echinochloa crus-galli* var. *praticola* and *Monochoria vaginalis* at National Institute of Crop Science and International Rice Research Institute.

Ratoon seeding screening method

The plastic pots, with a diameter of 30 cm and no draining hole, were filled with finely sieved paddy soil. Nitrogen was applied to a field level of 100 kg/ha. The pots were watered to saturation and conserved at 25 ± 5 °C greenhouse and removed unwanted germinated weeds during 15 days. Seven days old rice seedlings were transplanted on the pots arranged in three rows, 3 seedlings on the outer rows and 5

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seedlings on the center row. Rice shoots were cut $3 \sim 4$ cm above soil surface 28 days after transplanting. Pre-soaked seeds of *Echinochloa crus-galli* were seeded in the same pots on the day of rice clipping. Five weed seeds were sown around each rice hill and fifteen weed seeds between the two rice rows (between 1st and center row and between center row and 3rd row). Planting distance were about 7.5 cm in row and 6 cm between plants. The pots were irrigated to flooded condition (1 cm) when the weeds had grown to about 3 cm. Pots of weeds alone and unclipped rice served as the control treatment.

Growth suppression of *E. crus-galli*, *E.c* var. *praticola* and *M. vaginalis*

Rice were established in the same way as the rice clipping experiment. *E crus-galli*, *E crus-galli* var. *praticola* and *M vaginalis* seeds sowed around rice stalk used to know weed species difference of suppression ability on the above selected rice cultivars. Twenty seeds of *E crus-galli* and *E c* var. *praticola* seeded and managed at water saturated condition and *M vaginalis* seeded 0.01 g (about 150 seeds) of flooding condition of the pot. During all experiments, a number of non-destructive measures were recorded such as plant height, and tiller number for rice and germination and plant height were measured for all the weeds. At the end of the experiment root length, root dry weight and shoot dry weight were measured for both rice and the weeds. Dry weight of the shoots and leaves cut from rice were also measured.

Percent growth reduction (PGR) in the tested weed spe-

cies were computed using the formula: $PGR = 1 - (WR/WC) \times 100$, where WC is the growth of weeds in pots with no rice and WR is the growth of weeds in pots with rice. The formula was used for computing weed height reduction, weed root length reduction, and weed shoot and root biomass reduction.

Evaluation one rice plant to weed suppression

Rice were established one plant per pot and minimized the pot size to per 2.5×2.5 cm square, 2.5 cm depth, at seedling nursing tray. The pots were irrigated through the bottom of the tray. Sathi, AC1423, PI312777, Nonganbyeo, Keumobyeo, Chungmyungbyeo and Daeanbyeo were used in this experiment. Rice clipping dates were 15, 22, 29 and 37 days after rice seeding. Twelve *Echnochola crus-galli* soaked seeds were seeded at $1 \sim 2$ cm distance around the rice stalk on the clipping date. Weed shoot and root growth and biomass were measured 10 days after seeding.

Experimental design and statistical analysis

The experiments used the split plot design with rice cultivars as the main plot, the weeds as the sub plot. All treatments were established in four 4 replications. All experiments were repeated once over time.

The SAS software was used in the data analysis (SAS 9.1 for windows). Analysis of variance (ANOVA) and general linear models (GLM) were the procedures used for generating tables showing the sources of variation from the treatments and their F probability values, means and Duncan

Table 1. Growth of E	crus-galli which grown with clipped and no-clipped rice cultivars.
Table I. Olowill of L	crus-gam which grown with chipped and no-chipped rice cultivals.

	E crus-galli								
Cutivars	Height	(cm)	Root leng	gth (cm)	Dry weight (mg/10plants)				
	No-clipped	Clipped	No-clipped	Clipped	No-clipped	Clipped			
AC1423	8.8e	12.6°	3 4°	5.3°	0.4 ^d	0.7°			
Sathı	9.6 ^e	12 4°	4.1°	6 7 ^{bc}	0.4^{d}	$0.6^{\rm c}$			
PI312777	13.8 ^{bcd}	17 7 ^{abc}	6.1 ^{abc}	6 7 ^{bc}	0.8^{cd}	1.2 ^{bc}			
TN1	12.3 ^{cde}	14.4 ^{bc}	4 9 ^{bc}	6.9 ^{bc}	$0.7^{\rm cd}$	0.7^{c}			
Nonganbyeo	17.0 ^b	19 1 ^{abc}	8.2 ^{ab}	10 0 ^a	1.5 ^b	1.7 ^{ab}			
Chungmyungbyeo	11 9 ^{ed}	19.3 ^{abc}	4 4 ^{bc}	6.7 ^{bc}	$0.7^{\rm cd}$	1.4 ^{bc}			
Daelipbyeo	15.9 ^{bc}	21.1 ^{ab}	5 9 ^{abc}	8 9 ^{ab}	1.6 ^b	1.9 ^{ab}			
Hwajinbyeo	14.7 ^{bcd}	21.1 ^{ab}	8 2 ^{ab}	9.3 ^{ab}	1.1 ^{bc}	1.8 ^{ab}			
IAC165	13.3 ^{ed}	17.6 ^{abc}	4 8 ^{bc}	7.9 ^{abc}	$0.8^{\rm cd}$	1.3 ^{bc}			
Keumobyeo	21 9ª	23.3ª	8.6^{a}	9 9 ^{ab}	2.2ª	2.5ª			
Mean	13 9	17.9	5 8	5.3	1.0	1.4			
Control (no-rice)	33 1		15 3		6.9				

multiple range test.

RESULTS AND DISCUSSION

Ratoon seeding screening method

No-clipped rice plants caused the larger reduction on weed height, weed root length and weed dry weight than those of clipped rice (Table 1). Growth average of weeds grown with clipped rice cultivars were 17.9 cm, 7.8 cm and 1.4 mg but no clipped rice were 13.9 cm, 5.8 cm and 1.0 mg in weed height, root length and dry weight. Apparently, growth suppression of weeds by unclipped rice included not only competition effect but also allelopathy. Consequently, clipped rice were significantly divided 5 to 6 groups but no-clipped rice showed 3 to 4 groups at growth differences of weed height and dry weight. Dalipbyeo1, Hwajinbyeo, Keumobyeo, Nonganbyeo were the lowest suppressive and AC1423, Sathi and TN1 were the highest suppressive culti-

vars. PI312777 and Chungmyungbyeo were medium suppressive cultivars.

It is important to note here that eliminating the shading effect is one of the prime considerations in conducting field and greenhouse experiments for allelopathy screening research.

On the other hand, rice clipping significantly eliminated the effect of shading on the growth of the *Echinochloa* weed species. There were observed significant differences on the growth of weeds planted with the different rice cultivars. However, rice clipping may caused a great reduction in the observed root length and biomass of rice. This effect is due to the fact that clipping the rice shoot could lead to the rapid decomposition of the rice roots. Further, it was difficult to explain the cultivar differences without invoking several mechanisms involving roots. One could be that we were looking at decomposition products by chemical residues of roots that were shed in response to stem clipping and these products are stunting weed seedling. Another could be that

Table 2. Effect of several rice cultivars on height, root length, root biomass, and shoot biomass of weeds.

Rice cultivars	Weeds	Plant Gowth Reduction(%)							
Rice cultivals	weeds –	Height	Root length	Root biomass	Shoot biomass				
	E c	62	55	96	98				
Sathı	Ep	71	65	94	97				
Saun	Mv	75	50	-	93				
	mean	69	57	95	96				
	E c	54	54	93	93				
AC1423	Ep	39	33	75	85				
AC1423	Mv	70	38	-	84				
	mean	54	42	84	87				
	Ec	43	38	86	91				
DI212777	Ep	61	50	85	95				
P1312///	Mv	63	39	-	85				
	Mv 63 39 mean 56 42	42	86	90					
	E c	23	13	65	87				
TN1	Ep	31	21	64	78				
IINI	Mv	78	54	-	90				
	mean	44	29	65	85				
	E c	12	3	57	66				
Nonconbuco	Ep	14	6	52	63				
Nonganbyeo	Mv	57	19	-	72				
	mean	28	9	55	67				
	var	**	**	**	**				
F-value	weed	**	**	ns	**				
	var. x weed	ns	ns	ns	ns				

^{*}E c Echinochloa crus-galli, E p Echinochloa crus-galli var. praticola, M v Monochoria vaginalis

some cultivars have produced more root biomass than others before clipping, which dies back in response to clipping and in the chemical decomposition sufficiently increases the BOD/changes the redox etc in the soil/water to reduce weed seedling growth. Thirdly, that a specific response to the clipping by some cultivars is active allelochemical production and exudation through the roots to cause the suppression

Growth suppression of E. crus-galli, E.c var. praticola and M. vaginalis

Rice cultivars AC1423, PI312777 and TN1 had been reported in their high ability to suppress weed growth in the field and laboratory screening for allelopathy (Navarez & Olofsdotter, 1996; Dilday *et al.*, 1991). The potential allelopathic varieties were well in accordance with the weeds suppressive findings (Table 2).

All rice cultivars showed suppression on the growth *E crus-galli*, *E c* var. *platicola* and Monochoria vaginalis weed species. AC1423, PI312777, TN1 and Sathi were significantly reduced in the height, root length, root and shoot biomass of the weeds planted with these rice cultivars compared with the weeds of control. Nonganbyeo showed relatively less significant reduction for height and shoot biomass of the weeds.

On the average, weed height reduction for *E crus-galli* ranged from about 12 to 62%, for *E c* var. *platicola* ranged from about 14 to 71%, for *M vaginalis* ranged from 57 to 75%. The weed root length showed the least reduction among the growth parameters. Nonganbyeo exhibited significant less root dry weight reduction on all the weed species. The growth reduction in root and shoot dry weight of weeds were as high as 98% for *E crus-galli*, 97% for *E c* var. *platicola* and 93% *M vaginalis* grown with the potential allelopathic cultivars.

Weed species specific growth reduction for specific rice cultivars, i.e the interaction of growth reduction between rice and weed species, did not recognized.

Apparently, the effect of suppressive rice cultivars on the tested weed species were not only on the observed parameters but also on the entire plant structure. It was observed that the stunting effect had lead to the dwarfing of the weeds. The suppressed weeds had small leaves and flowers. Although stunted and small, it had reached flowering stage at the same time with the weeds of control.

Evaluation of one rice plant to weed suppression

Development of one plant evaluation system could be necessary to adopt breeding program. The ration seeding screening method modified for one rice plant evaluation with minimized pot size and different clipping dates. The varietal difference of one rice plant to growth suppression showed on the plant height, root length and dry weight of weeds (Table 3). The plant height and dry weight difference of E crus-galli could be recognized by clipping of 37 days old rice seedling but root length difference was showed by clipping of 22 days old rice seeding. E crus-galli root length and dry weight showed $12 \sim 17$ cm and $6.3 \sim 5.5$ mg for the AC1423 and Sathi but Nongangbyeo and Keumobyeo showed $32 \sim 43$ cm and $11 \sim 13$ mg in the clipping of 37 days after rice seeding. On one plant of rice varieties, AC1423, Sathi and PI312777 shows higher weed growth suppression than that of Nonganbyeo, Keumobyeo, Daeanbyeo and Chungmyungbyeo.

The days difference to growth inhibition or necessary days for growth suppression effect of weeds could be thought that some amount of allelochemical should be accumulated at the pots to suppress the weeds.

However, the effects of other factors such as root decom-

Table 3. Growth of E crus-galli seeded around one rice plant at different rice clipping dates.

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Cultivars	*			<u></u>		E cru	s-galli					
	Height(cm)			Root length(cm)			Dry weight(mg)					
	15dat	22dat	29dat	37dat	15dat	22dat	29dat	37dat	15dat	22dat	29dat	37dat
AC1423	32.8	30.4	24.6	12.8 ^b	9.4	6.4	2 6 ^b	3 8 ^b	9.1	8.1	7.6	6.3 ^b
Sathı	29 3	32 0	25 3	12 1 ^b	78	9.2	$2.6^{\rm b}$	2.0^{b}	8.6	9.8	7 3	5.5 ^b
PI312777	25.6	36.8	25.3	16.9 ^b	5.0	7.8	2.6^{b}	2.4 ^b	7.0	11.0	8.6	9.1 ^{ab}
Nonganbyeo	24.5	36.0	27.4	31 6 ^a	52	11 0	7.8^{a}	11.4 ^a	7.3	92	8.8	13.4 ^a
Keumobyeo	27 0	36 6	28.0	42.9^{a}	7.0	10.0	6 4 ^a	13.3 ^a	8.5	10 7	9.4	11.4 ^a
Daeanbyeo	27.8	32.9	28.7	37.9 ^a	5.8	11.0	$6~0^{a}$	10 4 ^a	8.8	9.8	8.9	9.7^{ab}
Chungmyungbyeo	31.3	32.3	25.8	17.5 ^b	9.0	8.4	5 4 ^b	3.8^{b}	90	9.5	9.8	8.4 ^{ab}
F-value	ns	ns	ns	**	ns	*	**	***	ns	ns	ns	**

^{*}DAT : days after transplanting

position due to stem clipping were difficult to determine. There is a need to reiterate here that there should be a continuous effort to develop screening systems that will minimize the sources of variation other than allelopathy.

The late effect of growth reduction could be attributed to the increase in the concentration of rice allelochemicals in the latter growth stage of the rice plant or one may argue that this is partly due to nutrient competition.

In practice, screening systems should have inexpensive, rapid, space limited and reproducible for testing large amount of rice accessions. So, the method describe here would closely approximate actual soil conditions in the field. This criteria is very valuable in terms of considering the complicating effects of soil chemical reactions occurring at the field which is disregarded in the laboratory screen method set-up. In this one plant evaluation by ratoon seeding with proper space has the closeness of the set-up to actual field conditions and adaptable method to weed suppression breeding program.

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