

Assessment of Allelopathic Potentiality and Identification of Allelopathic Compounds on Korean Local Rice Varieties

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This study was conducted to assess the allelopathic potentiality of Korea traditional seventy-nine rice (*Oryza sativa* L.) cultivars on barnyardgrass (*Echinochloa crus-galli* P. Beauv. var. *oryzicola* Ohwi) and to identify possible allelopathic compounds from selected rice cultivars, such as Seogandodobyeo, Huadobyeo and Heugbalbyeo. In the straw mixture, Seogandodobyeo showed the greatest inhibition (67.07%) on total emergence percentage. The greatest inhibition on total seedling length and dry weight of barnyardgrass occurred in Huadobyeo (58.32%) and Heugbalbyeo (81.20%), respectively. An HPLC analysis with nine standard compounds showed that the concentrations and compositions of allelopathic compound depend upon the cultivars. Four compounds including *o*-coumaric acid in Seogandodobyeo extracts, four compounds including *p*-coumaric acid in Huadobyeo extracts, and seven compounds including *o*-coumaric acid in Heugbalbyeo were detected, respectively. *O*-coumaric acid (0.97 mg/g) in Seogandodobyeo, *p*-coumaric acid (0.92 mg/g) in Huadobyeo and *o*-coumaric acid (1.02 mg/g) in Heugbalbyeo was detected as the highest amounts, respectively. The preliminary identification by HPLC analysis resulted in peaks with retention times close to those of standards which were confirmed with EI/MS. The mass spectra of *p*-coumaric, and *o*-coumaric acids showed molecular ions (*m/z*) at 164 and 164, and their base peaks (*m/z*) at 164 and 118, respectively.

Keywords: rice, allelopathy, barnyardgrass, HPLC, EI/MS

Rice (1984) defined that allelopathy is "any direct or indirect harmful effect by one plant on another through production of chemical compounds that escape into the environment". The substances which are involved in allelopathy were termed allelochemicals which are usually secondary plant products or waste products of the main metabolic pathways in plants (Swain, 1977; Whittaker & Feeny, 1971).

Rice (*Oryza sativa* L.) has been cultivated for a long time

as one of our main food crops. Since rice competes with weeds for light, nutrients, water and other growth requirements, and direct-seeded rice and conservation tillage practices have increased in recent years, ecological weed control as an alternative strategy for weed management becomes necessary in rice production. Direct seeded rice has been extensively studied to reduce production costs. Especially, barnyardgrass (*Echinochloa crus-galli* P. Beauv. var. *oryzicola* Ohwi), one of the greatest yield limiting weeds in Korean irrigated rice systems, is expected to become a major weed in dry direct-seeded rice because it is better adapted for growth under dry rather than wet conditions (Im *et al.*, 1993). One of the most environmentally acceptable and sustainable approaches is to develop rice cultivar showing higher allelopathic activity.

Much attention has been paid to the positive aspects of allelopathy as an ecological control to select rice cultivars with higher allelopathic potentiality (Ahn & Chung, 2000; Chou & Lin, 1976; Chung *et al.*, 1997, 2000; Dilday *et al.*, 1994; Elmore, 1980; Friedman & Waller, 1983; Fujii, 1992; Hassin *et al.*, 1994; Kim *et al.*, 1999a, 1999b; Lin *et al.*, 1992; Olosofotter *et al.*, 1995; Tamak *et al.*, 1994). These researches are conducted on the evaluation of allelopathic potentiality against one or more weeds including barnyardgrass from the collection of rice germplasm.

Few investigations have identified the chemical structure of the allelopathic compounds of rice. *P*-hydroxybenzoic, vanillic, *p*-coumaric, and ferulic acids were reported as the most common putative allelopathic compounds in plant (Rice, 1987). Also, Chou and Lin (1976) identified five compounds such as *o*-hydroxyphenylacetic, *p*-hydroxybenzoic, vanillic, *p*-coumaric and ferulic acids. Recently, Chung *et al.* (2000) identified nine compounds like *p*-hydroxybenzoic from Juma 10 rice cultivar which showed the most inhibitory effect on the agronomic characteristics of barnyardgrass in the field study. Among the identified compounds, *p*-hydroxybenzoic acid caused the highest inhibitory effect on the germination percentage, seedling (shoot and root) length and the total seedling dry weight of barnyardgrass. These results demonstrated that the allelopathic

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compounds may serve as natural herbicides by inhibiting the germination and seedling growth of certain weed species.

The purpose of this study was to assess the allelopathic potentiality of old traditional rice cultivars and to identify possible compounds on the selected rice cultivars. This may provide the basic information on development of allelopathic rice.

MATERIALS AND METHOD

Seventy-nine rice cultivars were grown at an experimental farm of Konkuk University at October 1998, and then were harvested and separated. The separated part (leaves plus stem) were dried at room temperature (24°C), ground in a Wiley mill through a 40 mesh screen and then stored at 5°C until needed. Barnyardgrass seeds which were collected in October 1998 were stored until use in a bioassay test at -35°C in a cold chamber after trash was removed from the seeds by floating it in distilled water.

The effect of rice straw mixtures on the growth of barnyardgrass

A straw experiment was conducted in a greenhouse with an average temperature of 26°C. The extracts concentration of seventy-nine rice cultivar straw (leaves + stem) including an Agudobyeyo used in this study was 1% (w/w). The rice straw was mixed thoroughly with 500 g of sand in each pot. All pots were steam sterilized for 5 h before being used. Each pot was placed on a brown plastic saucer to prevent loss of water soluble toxic substances. Plastic plugs were placed in the bottom of each pot to prevent loss of sand through the holes in the bottom. One hundred surface sterilized barnyardgrass seeds were planted uniformly 1 cm deep in each pot at 4 weeks after the rice straw and sand was incorporated. The barnyardgrass seeds were surface sterilized in a 1 : 10 (v/v) dilution of commercial hypochlorite bleach for 10 min and rinsed several times with distilled water. The seeds were then allowed to be imbibed on moistened paper towels for 2 h. The emergence was defined as the coleoptile protrusion through the soil surface and was counted at 24 h intervals for 20 days after seeding. After emergence, the seedling numbers were thinned to 25 plants per pot. Eighty of Hoagland solution I (Hoagland & Arnon, 1950) was added to each brown plastic saucer pot every 4 d interval to maintain adequate moisture. All plants were harvested 20 days after planting. The barnyardgrass seedlings were rinsed with water to remove sand debris and measured. After measuring their total seedling length, the seedlings were dried at 60 for measuring dry weight. The untreated controls were grown in sand with no straw.

The inhibitory percentage was calculated from the following equation:

Inhibition percentage (%) = [(control-straw mixture)/control] × 100

This experiment was conducted with three times in a completely randomized design. The analysis of the variance for all data was accomplished using the general linear model procedure of the statistical analysis system program (SAS, 1988). The pooled mean values were separated on the basis of least significant difference (LSD) at the 0.05 probability level.

Analysis of allelochemical

Three rice cultivars such as Seogandodobyeyo, Huadobyeyo and Heugbalbyeyo which showed the highest inhibitory effect on the growth of barnyardgrass, were tested for detecting allelopathic compounds. Five g of each dried straw of rice cultivars was finely ground and was placed in 100 ml flask containing 100 ml of MeOH 80% and stirred for 24 h at room temperature. The extracts were filtered through Whatman no. 42 filter paper. The filtrate was taken to dryness on a rotatory evaporator at 40°C. The dried material was redissolved in 10 times of 80% HPLC grade MeOH.

Standard chemicals

The standard phenolic compounds used for HPLC analysis were benzoic, ferulic, *m*-coumaric, *o*-coumaric, *o*-hydroxyphenylacetic, *p*-coumaric, *p*-hydroxybenzoic, salicylic and syringic acids (Aldrich Chemical Co.). All of the chemicals were purchased as high purity standards. The used solvents were HPLC spectral grade. All solvents and distilled water were degassed before use. All solvent ratios were based on volume.

HPLC instrumentation

The HPLC system used the Young-Lin company and the column employed YMC AM-303 (ODS, 4.6 × 250 mm) and employed a Micromeritics 725 Autoinjector with a 20 µl sample loop. An analysis of extracts was conducted based on the method of Chung *et al.* (2000). The mobile phase (solvents A and B) consisted of 98% water and 2% glacial acetic acid in 0.018 M ammonium acetate, and 68% water, 25% methanol, 5% butanol and 2% glacial acetic acid in 0.018 M ammonium acetate, respectively. Both extracts and standard compounds were used in the following gradient system: (a) 0.0 to 1.0 min isocratic at 10% B; (b) 1.0 to 21.0 min linear gradient from 10 to 25% B; (c) 21.0 to 36.0 min linear gradient from 25 to 45% B; (d) 36.0 to 56.0 min linear gradient from 45 to 100% B; (e) 50.0 to 50.15 min flow

increased to 1.20 ml/min; (f) 82.0 to 82.15 min linear gradient from 100 to 10% B; (g) 92.0 to 92.15 min flow decreased to 1.00 ml/min; (h) at 99.0 min sample loop rinsed and gradient repeated. The UV detector wavelength was set at 280. Standard compounds were chromatographed alone and as mixtures. Retention times for the standard compounds and the major peaks in the extract were recorded.

Quantification of allelopathic compounds by HPLC

The analysis of the compounds was performed based on the method of Chung *et al.* (2000). Phenolic compounds such as benzoic, ferulic, *m*-coumaric, *o*-coumaric, *o*-hydroxyphenylacetic, *p*-coumaric, *p*-hydroxybenzoic, salicylic and syringic acids were identified by retention times or standard addition, and contents were calculated by comparing peak areas with those of standards.

Mass spectra (MS)

To confirm the allelopathic compounds detected by HPLC analysis, an MS analysis was performed according to the method of Chung *et al.* (2000). Mass spectrometry was carried out on a VG 70-VSEQ Mass Spectrometer (VG ANALYTICAL, UK) and the spectra of the samples was compared with those of standard. The source temperature was 250°C, and the nominal potential, 70 eV with an acceleration voltage 8 KV. The UV spectrum of each collected portion of eluant was obtained following the freeze-drying and subsequent dissolving of the freeze-dried solid material in methanol. The solid material was analyzed by EI/MS. The collected eluant was frozen, and placed in a freeze-drier to remove the methanol/water solvent. The remaining dried material was redissolved in methanol solutions. A portion of each solution was evaporated to dryness with nitrogen. The dried material was then analyzed by direct probe electron impact on the EI/MS system.

RESULTS AND DISCUSSION

Rice cultivar straw mixture

All of the mixtures of rice cultivar straw with sand significantly inhibited the total emergence and seedling length and dry weight of barnyard grass (Table 1). The influence of the incorporation of rice straw on inhibition growth and development of barnyardgrass suggested that toxic compounds are released from rice straws. Especially, the inhibition rate based on the dry weight of the seedlings was higher than that of the emergence and seedling length of barnyardgrass. In terms of emergence, Seogandodobyeyo showed the highest inhibition (67.07%) and Gancheogbyeyo 1 was the lowest (15.61%).

Table 1. The inhibitory percentage of total emergence, total seedling length and dry weight on barnyardgrass.

Cultivars	TE [†]	TSL [‡]	TSDW [§]
-----Inhibition (%)-----			
Agudobyeyo	41.00	25.22	43.20
Agukdobyeyo	31.39	49.97	59.67
Arongbyeyo	44.43	32.51	46.89
Badolbyeyo	34.82	37.75	32.89
Baekgwangokbyeyo	28.65	39.60	32.24
Baekhaedalbyeyo	38.25	32.53	66.35
Baekjobyeyo	25.22	39.19	68.75
Baekkyeongjobyeyo	36.88	35.56	42.51
Baekmangjobyeyo	49.92	33.54	57.20
Baeknabyeyo	32.76	37.37	45.73
Baekseokbyeyo	38.94	33.84	72.05
Bakkyebyeyo	40.31	39.24	63.49
Banchonjobyeyo	38.94	36.42	43.59
Bandalbyeyoyeo	47.86	40.16	65.31
Baramdungkuribyeyo	43.74	34.00	70.56
Boribyeyo	47.86	38.78	73.48
Buldobyeyo	45.11	34.46	66.41
Chanarakbyeyo	47.17	33.41	71.79
Cheonggunbyeyo	35.51	32.96	25.39
Dabaegjobyeyo	40.31	32.86	74.39
Dadajobyeyo	45.11	35.37	69.78
Daegoldobyeyo	43.74	40.46	49.68
Daegudobyeyo	23.16	43.55	56.62
Daigolbyeyo	38.25	33.70	44.36
Damagunbyeyo	43.74	20.14	73.35
Deokjeokjobyeyo	43.74	39.17	50.27
Dongobyeyoyeo	39.62	35.24	35.55
Dongsanjobyeyo	45.80	30.07	58.57
Donnabyeyo	51.29	53.01	74.26
Doraebyeoyeo	39.62	36.76	46.63
Duchungjongbyeyo	32.76	36.33	51.89
Dudobyeyo	38.25	33.79	72.70
Eumseonbyeyo	52.66	28.48	42.29
Eunjobyeyo	45.11	38.76	43.65
F3-220	41.00	37.71	58.76
Gaegjodobyeyo	46.48	37.14	74.39
Gancheog 1	15.61	40.31	53.25
Gancheongdobyeyo	27.27	37.04	70.43
Gangreungdobyeyo	34.14	41.57	51.56

The cultivars were classified into six groups based on the degree of allelopathic potentiality by inhibitory emergence percentage of barnyardgrass. One group, Seogandodobyeyo, was over 60%. The second group over 50% is 3 cultivars including Sangpungbyeyo. The third group over 40% is 33 cultivars including Baekmangjobyeyo. The fourth group over 30% is 30 cultivars including Dongobyeyo. The fifth group over 20% is 9 cultivars including Baekjobyeyo. The sixth group below 20% is 3 cultivars including Sanchaengdobyeyo.

The inhibition rate on the total seedling length and dry weight of barnyardgrass was ranged from 20.14% to 58.32% and from 25.39% to 81.20%, respectively. Huado-

Table 1. Continued.

Cultivars	TE [†]	TSL [‡]	TSDW [§]
Gangweonnabyeo	27.27	37.04	70.43
Geumchangdobyeyo	32.08	39.56	68.75
Geumjeomdobyeyo	21.10	32.61	59.54
Guandobyeyo	33.45	32.52	60.19
Hambureubbyeo	41.68	37.79	41.12
Heugbalbyeo	34.82	43.70	81.20
Heugsaekdobyeyo	34.82	44.07	74.32
Heunbebyeo	25.22	40.88	54.22
Hochokjindbyeo	42.37	33.95	58.63
Hongcheonbyeo	33.45	34.94	41.77
Hongdodobyeyo	31.39	43.19	62.00
Huadobyeyo	43.05	58.32	41.97
Hwanghaedbyeo	28.65	38.03	43.20
Hwangjobyeyo	43.05	32.06	45.34
Hwangjubyeyo	49.23	42.71	74.13
Hwangtodobyeyo	43.74	40.69	53.25
IRI 233	43.05	34.62	43.91
IRI 301	45.11	37.35	74.52
IRI 268	37.57	42.59	72.96
Jaeraejongnbyeo	40.31	38.78	49.68
Jangwangbyeo	47.17	41.64	46.25
Jeonabyeo	37.57	40.13	55.78
Jeongdaldobyeyo	31.39	36.33	42.42
Jeosaekdobyeyo	43.74	41.51	51.95
Jinhwabyeo	41.68	39.34	62.52
Joseokjobyeyo	27.96	39.92	37.30
Namkangbaekjobyeyo	38.25	48.06	77.63
Namseon 1	38.94	46.60	79.44
Noindaribyeyo	35.51	38.16	74.26
Noindobyeyo	44.43	44.56	67.77
Nokdudobyeyo	39.62	33.20	56.43
Pungujobyeyo	35.51	39.08	57.59
Pyeongyangbyeo	38.94	37.23	59.99
Sancheongdobyeyo	19.04	34.85	63.69
Sangpungbyeo	54.03	39.10	49.28
Sanjobyeyo	41.68	22.12	58.95
Seogandodobyeyo	67.07	36.27	25.30
Siga-chata mochibyeyo	17.67	34.34	72.57
Sinbaegswogbyeo	42.21	32.11	42.10
PI 274471	37.57	40.48	38.01
CV(%)	33.6	15.6	38.1
LSD(0.05)	25.88	11.61	13.45

[†]Total Emergence [‡]Total Seedling Length [§]Total Seedling Dry Weight

byeo showed the highest inhibition (58.32%) of total seedling length and Damagunbyeo showed the lowest inhibition (20.14%). In seedling dry weight, Heugbalbyeo showed the highest (81.20%) inhibition and Cheonggunbyeo showed the lowest (5.39%).

This result was similar to that of Chung *et al.* (2000) and Kim *et al.* (1999a, 1999b) who reported that some cultivars including foreign cultivar extract exhibited a strong inhibitory effect on the germination and growth of barnyardgrass. This study demonstrated the inhibition of germination and growth on barnyardgrass by allelochemicals released from

rice straw. This data also shows the genetic difference in allelopathic potentiality among rice cultivars and demonstrates that it may be possible to develop allelopathic rice.

One limitation of this experiment was that the concentration of toxic substances was greater than that which would be found in nature. Another is that the toxic substances contained in the straw or released from their decomposition may not exist for long under field conditions. Therefore, the several factors which are involved in allelopathic activity must be studied more carefully.

Analysis of allelopathic compounds

This study was conducted to detect which allelopathic compounds showed the highest inhibition percentage in the rice cultivar straw mixture. The results of the HPLC analysis clearly demonstrate that rice residue contains allelopathic compounds and the concentrations and compositions depend upon the cultivars (Table 2). In Seogandodobyeyo cultivar extracts which showed the most inhibitory effect in total emergence percentage, benzoic acid (0.01 mg/g), ferulic acid (0.004 mg/g), *m*-coumaric acid (0.47 mg/g) and *o*-coumaric acid (0.97 mg/g) were detected. In Huadobyeyo cultivar extracts, which showed the most inhibitory effect on total seedling length, salicylic acid (0.56 mg/g), benzoic acid (0.06 mg/g), *p*-coumaric acid (0.92 mg/g) and *o*-hydroxyphenylacetic acid (0.39 mg/g) were detected. In Heugbalbyeo cultivar extracts (Fig. 1) which showed the most inhibitory effect on total dry weight, salicylic acid (0.62 mg/g), *p*-coumaric acid (0.98 mg/g), syringic acid (0.41 mg/g), ferulic acid (0.005 mg/g), *m*-coumaric acid (0.10 mg/g), *p*-hydroxybenzoic acid (0.11 mg/g) and *o*-coumaric acid (1.02 mg/g) were detected. *O*-coumaric acid in Seogandodobyeyo and Heugbalbyeo and *p*-coumaric acid in Huadobyeyo exhibited the highest concentrations. The results of this analysis were similar to that of Chung *et al.* (2000) and Chou and Lin (1976) who isolated phenolic acid including ferulic acid in rice straw. These results suggest that the allelopathic effect on barnyardgrass may be partly due to the presence of these compounds.

Also, the results of the HPLC analysis indicated the presence of a large number of unknown peaks as compared to standard compound mixtures. The chromatogram (did not shown) suggested that rice straw extracts contain a complicated mixture of other allelopathic compounds other than those identified compounds. These unknown compounds need to be identified in the future.

The *p*-coumaric and *o*-coumaric acids identified by HPLC were confirmed using EI/MS. The freeze-dried solid materials were analyzed by EI/MS. The mass spectra (did not

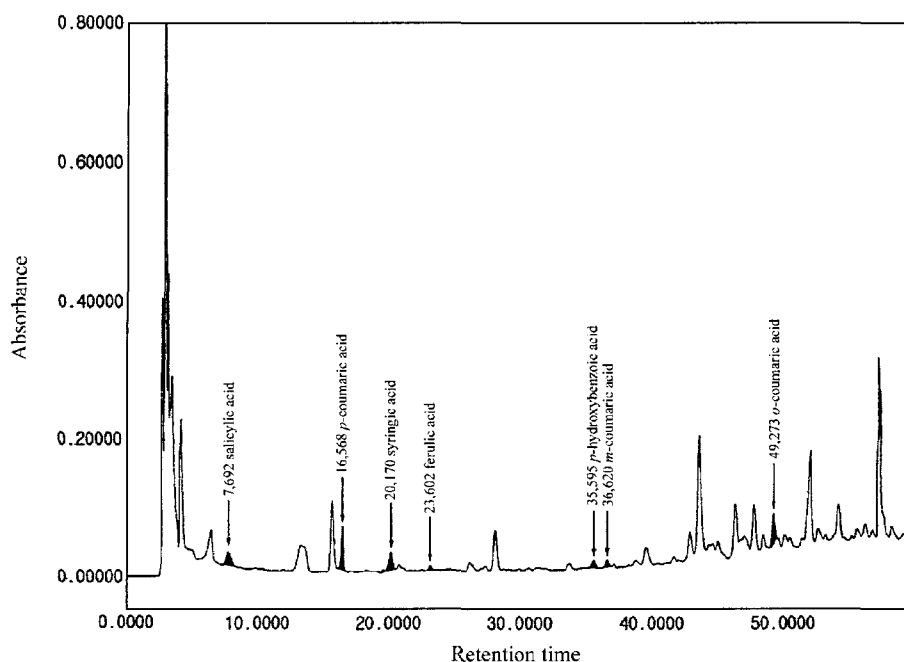


Fig. 1. HPLC Chromatogram of Heugbalbyeo extracts.

Table 2. The results of HPLC analysis of Seogandodobyeo, Huadobyeo and Heugbalbyeo cultivar extracts.

Chemicals	Standard retention time (min)	Retention time (min)			Concentration (mg/g)		
		A [†]	B [‡]	C [§]	A	B	C
salicylic acid	7.458	-	7.582	7.692	-	0.56	0.62*
<i>p</i> -coumaric acid	16.608	-	16.478	16.548	-	0.92	0.98
<i>o</i> -hydroxyphenylacetic acid	19.410	-	19.638	-	-	0.39	-
syringic acid	20.305	-	-	20.107	-	-	0.41
ferulic acid	23.873	23.462	-	23.602	0.004	-	0.005
benzoic acid	30.990	30.427	30.803	-	0.01	0.06	-
<i>p</i> -hydroxybenzoic acid	35.552	-	-	35.595	-	-	0.11
<i>m</i> -coumaric acid	36.902	36.667	-	36.620	0.47	-	0.10
<i>o</i> -coumaric acid	49.332	49.887	-	49.273	0.97	-	1.02

[†]Seogandodobyeo [‡]Huadobyeo [§]Heugbalbyeo

*average four measurements

shown) of *p*-coumaric acid and *o*-coumaric acid showed molecular ions (*m/z*) at 164 and 164, and their base peaks (*m/z*) at 164 and 118, respectively. These spectra were exactly identical to those obtained from authentic standards.

However, although these potential allelochemicals were detected by HPLC and confirmed by MS, a more detailed study is needed to investigate variety variation containing compounds and agronomic practices, environmental factors, temperature, light, plant age and the nutrient conditions of the growing season since these compounds are presented in plants.

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REFERENCES

Ahn, J. K. and I. M. Chung. 2000. Assessment of allelopathic

- potential on germination and seedling growth of *Echinochloa crus-galli* using rice cultivars hulls. Weed. Sci. (Accepted)
- Chou, C. H. and H. J. Lin. 1976. Autotoxication mechanism of *Oryza sativa*. 1. Phytotoxic effects of decomposing rice residues in soils. J. Chem. Ecol. 2:353-367.
- Chung, I. M., J. K. Ahn, J. T. Kim, C. S. Kim, and Y. H. Lim. 2000. Identification of allelopathic compounds from rice (*Oryza sativa* L.) straw and test of its biological activity. J. Chem. Ecol. (accepted)
- Chung, I. M., K. H. Kim, J. K. Ahn, and H. J. Ju. 1997. Allelopathic potential evaluation of rice cultivars on *Echinochloa crus-galli*. K. J. Weed Sci. 17:52-58.
- Dilday, R. H., J. Lin, and W. Yan. 1994. Identification of allelopathy in the USDA-ARS rice germplasm collection. Aust. J. of Exp. Agr. 34:907-910.
- Elmore, C. D. 1980. Inhibition of turnip (*Brassica rapa*) seed germination by velvetleaf (*Abutilon theophrasti*) seed. Weed Sci. 28:658-660.
- Friedman, J. and G. R. Waller 1983. Seeds as allelopathic agents. J. Chem. Ecol. 9:1107-1117.
- Fujii, Y. 1992. The potential biological control of paddy weeds with allelopathy-allelopathic effect of some rice cultivars. In Proceedings of the International Symposium on Biological Control and Integrated Management of Paddy and Aquatic Weeds in Asia. pp. 305-320.
- Hassin, S. M., A. N. Rao, A. O. Bastawisi, and I. R. Aidy. 1994. Weed management in broadcast seeded rice in Egypt. In Proceeding from and International Workshop on: Constraints, Opportunities and Innovations for Wet-seeded Rice, 31 May-3 June 1994, Bangkok, Thailand.
- Hoagland D. R., and D. I. Arnon. 1950. The water culture method for growing plant without soil. Calif. Agric. Exp. Stn. Circ. No. 347. pp. 32.
- Im, I. B., O. J. Guh, and Y. J. Oh. 1993. Weed occurrence and competitive characteristic under different cultivation types of rice(*Oryza sativa* L.). 3. Difference in weed occurrence and rice growth under the competitive periods. K. J. Weed Sci. 13:114-121.
- Kim, K. U., D. H. Shin, H. Y. Kim, I. J. Lee, and M. Olofsdotter. 1999a. Study on rice allelopathy. Evaluation of allelopathic potential in rice germination. K. J. Weed Sci. 19:105-113.
- Kim, K. U., D. H. Shin, H. Y. Kim, I. J. Lee, J. H. Kim, and K. W. Kim. 1999b. Study on rice allelopathy. Factors affecting allelopathic potential of rice germination. K. J. Weed Sci. 19:114-120.
- Kim, K. W., K. U. Kim, D. H. Shin, H. Y. Kim, H. J. Jeong, G. Y. Chung, and G. S. Kwon. 1997. Isolation of allelopathic potentials produced by rice (*Oryza sativa* L.) cultivars. Bulletin of institute of agricultural science and technology. pp. 71-80.
- Lin, J., J. Jr. Smith, and R. H. Dilday. 1992. Comparison of allelopathy and herbicides for weed control in rice. In Proceedings of the 24th Rice Technical Working Group. pp. 127.
- Olofsdotter, M., D. Navarez, and K. Moody. 1995. Allelopathic potential in rice(*Oryza sativa* L.) germplasm. Ann. Appl. Biol. 127:543-560.
- Rice, E. L. 1984. Allelopathy, 2nd ed. Academic Press, Orlando, Florida. pp. 422.
- Rice, E. L. 1987. Allelopathy: An overview, pp.8-22, in G.R. Waller(ed.). Allelochemicals: Role in agriculture and forestry. ACS Symposium Series. American Chemical Society, Washington, D.C.
- SAS Institute. 1988. SAS/STAT User's Guide, 6.03 ed. SAS Institute, Cary, NC. pp. 108.
- Swain, J. 1977. Secondary compounds as protective agents. Ann. Rev. Plant Physiol. 28:479-501.
- Tamak, J. C., S. S. Narwal, L. Singh, and M. Ram. 1994. Effect of aqueous extract of rice stubbles and straw + stubbles on the germination and seedling growth of *Convolvulus arvensis*, *Avena ludoviciana* and *Phalaris minor*. Crop Research. 8:186-189.
- Whittaker, D. C. and P. P. Feeny. 1971. Allelochemicals:chemical interactions between species. Sci. 171:757-770.