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Exploratory study on allelopathic effect of selected Malaysian rice varieties and rice field weed species

(Kajian kesan alelopati varieti padi Malaysia terpilih dan spesies rumpai di sawah padi)

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Key words: allelopathy, rice variety, weed species

Abstrak

Pembentukan alelopati padi mungkin menjadi alternatif kepada amalan penggunaan kimia dan amalan kultur yang lain bagi mengurangkan serangan rumpai dalam tanaman padi tabur terus. Kajian saringan terhadap sejumlah varieti padi Malaysia untuk aktiviti alelopati telah dijalankan di Stesen MARDI, Seberang Perai dengan menggunakan kaedah *plant box*. Melalui kaedah ini, kesan alelopati rembesan akar telah didapati dengan membiarkan tumbuhan yang diuji bersama-sama kultur campuran dalam medium agar. Salad telah digunakan sebagai tumbuhan penunjuk dan padi sebagai tumbuhan yang diuji. Kesan alelopati varieti padi dinilai dengan meneliti pertumbuhan radikel anak benih salad. Beberapa varieti tradisional seperti Siam Er 54 Jambok, dan Wangi serta varieti moden iaitu MR 77 dan MR 84 telah menunjukkan aktiviti alelopati yang kuat terhadap anak benih salad. Ujikaji diulang dengan menggantikan salad dengan spesies rumpai sawah padi iaitu rumput sambau [*Echinochloa crus-galli* (L.) Beauv]. Begitu juga, beberapa varieti padi telah dikesan mempunyai aktiviti alelopati yang kuat terhadap anak benih *E. crus-galli*. Varieti tradisional seperti Padang Gelap, Acheh Puteh, Pasir, Singgora, Merah Isi, Chatek Kuning, Anak Naga dan Anak Didek 3 mempunyai kesan alelopati yang kuat terhadap anak benih *E. crus-galli*.

Kajian kedua dijalankan dengan menggunakan kaedah bioassei, dikenali sebagai kaedah *sandwich* untuk mengesan pengaruh alelopati pada daun spesies rumpai sawah padi yang gugur. Beberapa spesies rumpai sawah padi menunjukkan tindakan membantutkan pertumbuhan anak benih salad. Spesies rumpai tersebut ialah *Pennisetum purpureum* Schumach, *Isachne globosa* (Thunb.) O. K., *Cyperus rotundus* L., *Sagittaria guyanensis* H. B. K. dan *Fimbristylis miliacea* (L.) Vahl.

Abstract

Development of allelopathic rice would be an alternative to chemicals and other cultural practices in reducing the occurrence of weed infestation in direct-seeded rice. A study to screen Malaysian indigenous and improved rice varieties for allelopathic activity was conducted in MARDI Research Station, Seberang Perai using *plant box* method. This method detects the allelopathic effect of root exudates by growing test plant in mixed culture in agar media. Lettuce was used

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as acceptor-indicator (test plant) and rice plant as a donor. The allelopathic effect of rice varieties was assessed by observing radicle growth of lettuce seedlings. Several traditional varieties like Siam Er 54, Jambok, Wangi and modern varieties viz. MR 77 and MR 84 were found to have strong allelopathic activity on lettuce seedlings. The tests were repeated by replacing lettuce with a selected rice field weed species namely *Echinochloa crus-galli* (L.) Beauv. Similarly, some rice varieties were detected to have strong allelopathic effect on *E. crus-galli* seedlings. Traditional varieties such as Padang Gelap, Acheh Puteh, Pasir, Singgora, Merah Isi, Chatek Kuning, Anak Naga and Anak Didek 3 were found to have strongest allelopathic activity on *E. crus-galli* seedlings.

A second study was conducted using bioassay method known as *sandwich* method to assess the effect of leaf and debris taken from rice field weed species. Again, several rice field weed species showed strong inhibitory action on the growth of lettuce seedlings. The weed species were *Pennisetum purpureum* Schumach, *Isachne globosa* (Thunb.) O. K., *Cyperus rotundus* L., *Sagittaria guyanensis* H. B. K. and *Fimbristylis miliacea* (L.) Vahl.

Introduction

Allelopathy, the direct or indirect effect of one particular plant on another through the production of chemical compounds that are released into the root environment may provide an alternative weed control strategy. This approach may lead to less dependence on the use of herbicides in agricultural production. Since plants are known to self-regulate their densities and distribution in nature through allelopathic interactions, attention is now being given to the possibility of exploiting this phenomenon to aid in placing crops in a more favourable competitive position over weeds (Dilday et al. 1998). Herbicide use moves the agro-ecosystem to low species diversity with new weeds appearing, so that there is a need for an ecological approach to weed control instead of relying totally on chemical control methods (Moody 1992). One of the methods is by exploring allelopathic potential in crop varieties.

Allelopathic potential can be utilized for weed control in four ways: (1) plant associated allelopathic plants to provide weed control, (2) use allelopathic plants in crop rotation and obtain allelopathic activity from plant residues in soil, (3) identify and isolate potent chemicals and use these as herbicides and, (4) optimise the allelopathic potential in the crop by developing allelopathic cultivars (Gliessman 1982; Duke 1985). All of these methods have some constraints that need to be considered.

In Japan, Fujii (1993) reported that about 5% of 189 rice strains which were screened possessed allelopathic activity. Improved Japonica cultivars showed little allelopathic activity where traditional Javanica cultivars and red rice strains showed strong activity. The allelopathic activity of *Oryza glaberrima* was also strong. Dilday et al. (1991) reported that 347 accessions from the USDA/ARS rice germplasm collection exhibited allelopathic activity to *Heteranthera limosa* (Sw.) Wild. Furthermore, Lin et al. (1992) found that six allelopathic rice lines reduced the dry weight of aquatic weeds from 93% to 99% compared to Rexmont, a cultivar without allelopathic activity.

Development of allelopathic crop cultivars is probably the best solution for many parts of the world, both in terms of environment safety and accessibility. However, it is also the solution that requires much research on both genetic control of allelopathic potential and allelochemicals (Olofsdotter and Navarez 1995).

Fuerst and Putnam (1983) published a protocol which can be used to demonstrate allelopathy using suitable control procedures, to isolate and characterize allelochemicals and assay the isolates on sensitive plants, to obtain toxicity and similar symptoms with pure chemicals added to the growth medium and to monitor the release of allelochemicals from donor plants. Later, to detect them in the environment and ideally find them in a receiver plant.

The objective of this study was to screen selected Malaysian indigenous rice varieties for allelopathic activity. Varieties possessing allelopathic activity will be identified as potential gene pool for future research notably in the improvement for allelopathic modern rice varieties.

Materials and methods

Plant box method

The plant box method for measuring allelopathy in root exudates which was developed by Fujii and Shibuya (1991) was adopted. The procedure involved several steps beginning with the preparation of donor plants. Firstly, the rice seeds were seeded in a plastic pot (7.5 cm x 7.5 cm x 10 cm) containing river sand of 3 mm in size. Hoagland's solution was added in progression from 1/4 to 1/2 strength to full strength at alternate days. The plant was considered ready when its root dry weight reached 200 g. Secondly, a semi-solid agar medium was prepared by dissolving 5 g of 0.5% (w/v) agar. The preparation was autoclaved at 115 °C for 15 min and let to cool until 45 °C before it was ready for use.

Thirdly, a plant box named as *magenta box* measuring 6.5 cm x 6.5 cm x 10 cm was used to accommodate 250 mL of agar i.e. to a level of 5.5 cm. Fourthly, a root zoning tube was prepared using a vinyl chloride tube. The outer portion of 32 mm diameter and inner portion of 25 mm diameter was cut into 65 mm lengths leaving the side wall of about 1/4 intact. An open window with upper and under ridge of

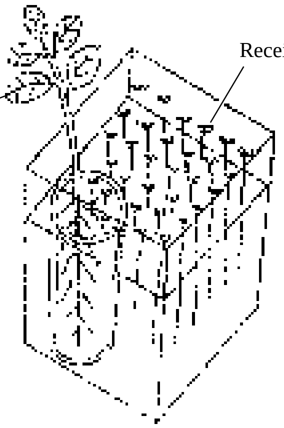

Figure 1. The plant box method

5 mm was made at 3/4 portion of the tube. The bottom of the tube was sealed with the same vinyl chloride plate of 2 mm thickness. A nylon gauge was attached to the windows using adhesive agent. The suitable rice plants were positioned at one corner of the plant box *(Figure 1)*. By using transparent tape, the basal part of plants were anchored to the wall of the plant box.

Agar medium which was incubated at 45 °C was allowed to flow into the plant box, and immediately it was cooled by ice cubes. Gelatinization was completed after 30–60 minutes. Lettuce seeds were sowed into agar with its rooting side underneath by a distance approximately 1 cm apart, as guided by the marks on the side walls of plant box. Upon completion, the plant box was wrapped with vinylidene chloride film to avoid airborne microorganism. The same procedure was repeated for *E. crus-galli* seeds.

Percentage of radicle means the percentage of the root radicles (by length) of the target plants present in the root zone of the rice (based on a calculation of radicle length within the root zone controlled by the rice). On the other hand, trend means the trend of linear regression curves, indicating the migration speed of substances exuded from the rice root through agar medium.

Hence, low figures for both parameters indicate strong activity by rice root exudates.

For each variety, the distances of root zone separating tube and radicle length were plotted, and the linear regression curve was fitted using computer calculation using Microsoft Excel software. From this curve, radicle length at the point of the surface of rice root (distance equal to zero) was compared to the length of lettuce seedlings of control treatment.

Sandwich method

A sandwich method which was introduced by Fujii et al. (1999) was used in this study. The leaf specimens were collected among the basal leaves of the matured plants. Only dried leaves were collected for use in the required assay. A multi-dish plate with six-well was used for this experiment. Fifty milligrams of defoliated leaves were spread in a 10 mm^2 petri dish. To mimic the natural condition, cutting of leaves was restricted to the minimum. A 0.5% agar (w/v) was autoclaved in 115 °C for 15 min, and later incubated at 45 °C. Five mL of agar was added to each dish by a calibrated pipette such as pipette-man. The leaves were floated on the surface of the agar gels. After 1–2 hr, 5 mL more of agar was added to make up the sandwich. After the gelatinization of the agar, 5 seeds of lettuce (var. Great Lakes 366), were put on the surface of the agar. The same procedure was followed for assessing allelopathic activity of selected species of rice field weeds.

Results and discussion

Plant box method

The plant box is transparent on both sides. One could easily see the condition of the growing roots as affected by the exudates released by the roots of donor plant. As expected, radicle length of acceptor plants closer to the donor plant were shorter and the overall reduction was greater for rice varieties possessing stronger allelopathic activity. This length showed the inhibitory effect of allelochemicals emitted from the root of donor plant. The results on allelopathic effect of rice varieties against germinating lettuce and *E. crus-galli* were as shown in *Table 1, Table 2, Table 3, Table 4* and *Table 5*. The decline of each regression equation showed the migration speed of allelochemicals in agar medium (*Figure 2* and *Figure 3*).

Allelopathic effect of rice varieties on germinating lettuce seeds The results from allelophathic screening of modern rice varieties (MR series), rice varieties from foreign countries and *padi angin* variants and traditional varieties are as shown in *Table 1, Table 2* and *Table 3* respectively. Different rice varieties showed variation in allelopathic effect on lettuce root growth. There was a wide variation in the level of root exudate activity among rice varieties tested. Some varieties showed no activity at all, while others showed slight to strong activity such that in some cases the tip of lettuce radicle became discoloured.

Allelopathic effect of MR rice varieties A total of 36 MR lines were tested for allelopathic effect against lettuce. Of all the MR lines tested, MR 77 and MR 84 were found to have strong allelopathic activity on lettuce seedlings. They recorded a 27.72% and 29.78% lettuce radicle growth respectively *(Table 1).*

Allelopathic effect of foreign rice varieties and *padi angin* variants A total of 52 accessions of foreign varieties and five *padi angin* variants were tested against lettuce for allelopathic activity *(Table 2).* Most of the varieties had slight to moderate allelopathic activity as shown by higher percentage of radicle growth. The strongest allelopathic activity among foreign accessions was Sujeong, where it recorded 31.51% radicle growth. This was followed by Taebaeg (32.05%), Raekyeong (32.72%) and HR1619 (32.87%).

Table 1. Screening of rice varieties (MR series) for allelopathic potential using the plant box method

Rice variety	Accession No./Designation	[1]Radicle (%)	[2]Trend
MR 77	Acc 4559	27.72	0.3130
MR 71	Acc 4557	28.33	0.1944
MR 84	Acc 4633	29.78	0.6294
MR 106	Acc 7487	31.51	0.6482
MR 27	Acc 4554	32.42	0.8066
MR118	Acc 7881	32.08	0.2562
MR 73	Acc 4588	33.65	0.2835
MR 39	Acc 4593	33.83	0.3741
MR 43	Acc 4597	34.03	0.9120
MR 27	Acc 4554	36.50	–0.0477
MR 10	Acc 4533	38.69	–0.0616
MR 167	Acc 8646	39.42	0.4568
MR 159	Acc 8638	39.06	0.0985
MR 47	Acc 4555	42.75	0.8122
MR/Sri Malaysia I	Acc 2671	43.70	0.4689
MR77	Acc 4559	44.30	0.3434
MR/Sri Malaysia II	Acc 2672	45.00	1.3498
MR 103	Acc 7484	49.25	0.1614
MR 7	Acc 4552	49.82	0.7355
MR 47	Acc 4555	56.05	0.3043
MR 142	Acc 8621	56.29	0.1093
MR 144	Acc 8623	58.43	0.1534
MR 151	Acc 8630	59.40	0.1533
MR 123	Acc 7488	61.67	0.0149
MR 150	Acc 8629	66.40	0.0547
MR 114	Acc 7827	71.61	0.0032
MR 127	Acc 7489	72.26	0.3139
MR 148	Acc 8627	72.96	0.0712
MR 145	Acc 8624	73.31	0.1680
MR 119	Acc 7832	74.37	0.1858
MR 147	Acc 8626	76.42	0.3780
MR 146	Acc 8625	77.27	0.2550
MR 67	Acc 4619	79.12	0.5829
MR 143	Acc 8622	88.69	0.1365
MR 109	Acc 7740	94.38	0.0533
MR 149	Acc 8628	101.90	0.0966

[1]Lettuce radicle growth (%) = Percentage of lettuce roots growing within the root zone of the rice
[2]Trend = Gradient of regression line

Allelopathic effect of traditional rice varieties In general, the allelopathic activity of traditional varieties was strong compared to that of foreign varieties and MR lines. Out of 101 traditional varieties tested against lettuce for allelopathic effect, 25 varieties were found to have strong allelopathic activity. The recorded percentage of lettuce radicle growth ranged from 20% to 30% *(Table 3)*. Among the strongest allelopathic effect were Siam Er 54 (20.4%), Jambok (20.53%), Ketitir (20.64%) and Wangi (20.96%).

Allelopathic effect of rice varieties on germinating *E. crus-galli* A total of 85 rice accessions (i.e. 26 MR lines, 12 foreign varieties and 47 traditional varieties) were

Table 2. Screening of foreign rice varieties and *padi angin* (weedy rice) for allelopathic potential using the plant box method

Rice variety	Accession No./Designation	[1]Radicle (%)	[2]Trend
Sujeong	PM160	31.51	0.3495
Taebaeg	PM158	32.05	0.3318
Raekyeong	PM156	32.72	0.0908
HR1619	PM154	32.87	0.1906
Shinkwang	PM172	33.38	0.4870
Guar er ai 5	PM186	35.51	0.1561
Milyang 98	PM163	36.08	0.2204
HR 1888	PM155	37.28	0.3519
Ketan Nangka (Indonesian var.)	PM93	38.29	–0.1226
IR59601-301-3-6R	PM142	38.74	0.2414
BR827-35-2-1-1-1R	PM133	41.60	0.1880
CP231 (Philipino variety)	PM96	42.80	0.1650
Milyang 92	PM162	45.28	0.6081
IR-54791-19-2-3R	PM126	46.10	0.5420
IR52256-5-2-2-IR	PM137	46.54	0.3630
Samyang	PM157	46.84	0.2167
Lemont (American variety)	PM95	47.06	–0.015
IR42686-C2-118-6-2	PM123	47.72	0.7236
Namyeong	PM161	47.95	0.4613
Yeongpung	PM152	50.40	0.4164
Guang ye ai	PM185	50.58	0.0914
IR49461-128-3-3-3R	PM125	50.59	0.7102
IR26 (IRRI line)	PM97	51.19	0.1551
IR54869-41-2-2R	PM128	52.08	0.7158
IR25912-81-2-IR	PM119	52.23	–0.0766
Banten	PM94	52.60	0.2462
Jamuna (Indian line)	PM98	53.60	–0.0664
Jungweon	PM151	53.69	0.3650
Jia nong xian 43	PM195	54.06	0.3622
Er jiu ai 4	PM190	56.38	0.2304
Taichung Sen Yu 85	PM134	57.88	0.2224
IR39323-182-2-3-3	PM122	58.98	0.6475
Tei qiu	PM191	59.12	0.2310
IR58773-35-3-1-2R	PM130	59.36	0.3176
Dular	PM99	59.61	0.1720
Padi angin	PA1	61.76	0.2645
Gui chao 2	PM182	61.97	0.5071
IR37721-90-3-3-3-2	PM121	63.27	0.6217
Nang jing II	PM189	63.47	0.2971
Padi angin	PA9	65.53	0.0861
Kele	PM100	68.00	–0.2362
Hong yang ai	PM188	72.97	0.0616
Guang 104	PM184	74.02	0.4246
Padi angin	PA17	74.50	0.2381
Jian mei ai	PM183	76.79	0.568
Padi angin	PA19	77.94	–0.0988
Nipponbare	PM202	80.75	0.4955
Yumehikari	PM201	83.78	0.3265
Guang liu ai 4	PM187	84.95	0.2279

(cont.)

Table 2. *(cont.)*

Rice variety	Accession No./Designation	[1]Radicle (%)	[2]Trend
Hitomebore	PM197	86.37	0.5322
IR48725-B-B-141-2	PM124	87.06	0.2257
Koshihikari	PM199	89.20	0.4231
Padi angin	PA10	89.25	0.1198
Jia nong xian 41	PM194	91.75	0.3722
Niigatawase	PM198	91.91	0.3499
Reimei	PM203	99.24	0.4285
Shinkei8546/Milyang (H91-23)	PM196	115.20	0.1479

[1]Lettuce radicle growth (%) = Percentage of lettuce roots growing within the root zone of the rice
[2]Trend = Gradient of regression line

Table 3. Screening of traditional rice varieties (landraces) for allelopathic potential using the plant box method

Rice variety	Accession No./Designation	[1]Radicle (%)	[2]Trend
Siam Er 54	Acc 1687	20.40	0.3156
Jambok	Acc 6212	20.53	0.2681
Ketitir	Acc 3955	20.64	0.2186
Wangi	Acc 4151	20.96	0.1723
ADT 10	Acc 15	21.44	0.4030
Siam 48	Acc 1661	21.83	0.2391
Pulut Manis	Acc 4092	21.98	0.6688
Merah isi	Acc 1007	22.17	0.2859
Gading	Acc 449	22.25	0.7825
Anak Singgora	Acc 131	22.67	0.6574
Mayang Gerbi	Acc 888	23.72	0.6035
Tangkai Rambai 170	Acc 1772	23.79	0.4055
Jintan Kuning	Acc 600	24.00	0.4555
Bujang Kelsom	Acc 988	24.42	0.3979
Jintan Puteh	Acc 620	24.57	0.5754
Pangsi 2	Acc 4132	25.38	0.2697
Chatek Kuning	Acc 320	26.16	0.7239
Panji Kuning	Acc 4187	26.39	0.7170
Payaw A	Acc 3592	26.52	0.2271
Merak Sepilai Kecil	Acc 1015	26.70	0.3540
Janda	Acc 565	27.19	0.5452
Mayang Segumpal	Acc 956	27.96	0.6998
Tega	Acc 4053	28.03	0.6491
Muar Kuning 1818	Acc 1023	28.87	0.2746
Anak Limbat	Acc 101	29.03	0.8346
Parjugan C	Acc 1201	29.26	0.3515
Radin Pahang	Acc 1423	29.73	0.4613
Chempaka	Acc 334	29.87	0.3383
Bodong	Acc 223	30.70	0.3678
Padi Cina	Acc 3689	30.72	0.3995
Mayang Segumpal	Acc 956	31.00	0.2253
Anak Gajah	Acc 47	31.20	0.4377
Batas	Acc 184	31.20	0.2547

(cont.)

Table 3. *(cont.)*

Rice variety	Accession No./Designation	[1]Radicle (%)	[2]Trend
Mayang Gerbi	Acc 101	31.24	0.2302
Tok Suan 57	Acc 1811	31.31	0.5737
Lembut Ketam	Acc 8076	31.36	0.6996
Batas	Acc 184	31.20	0.2547
Mahsuri	Acc 826	32.66	0.0338
Anak Ikan Cina	Acc 56	32.98	0.7610
Anak Naga	Acc 113	32.99	0.6544
Sambug	Acc 4161	33.09	0.2378
Bahagia	Acc 167	33.93	0.5555
Anak Ikan Tinggi	Acc 8	34.85	0.7388
Aceh Putih	Acc 11	34.90	–0.0477
Anak Didek 3	Acc 38	35.88	0.5704
Acheh Puteh	Acc 8	36.20	0.7133
Ketumbar	Acc 7899	36.54	0.2570
Batikan	Acc 187	36.91	0.2883
Mayang Pasir	Acc 909	36.98	0.4799
Anak Ikan Kelubi	Acc 66	36.99	0.7691
Janda Berhias	Acc 566	38.92	0.5247
Melor	Acc 1001	39.36	0.9414
Kedah	Acc 672	40.33	0.4911
Malinja	Acc 839	41.40	0.8323
Musang B	Acc 1044	41.82	0.4762
Anak Naga	Acc 114	41.84	0.6892
Lembu Basah	Acc 777	43.83	0.1967
Mek Bujang Kelsom	Acc 988	45.00	0.2093
Anak Puteh	Acc 125	46.02	0.5543
Radin Ebos 36	Acc 1352	46.89	–0.2127
Anak Cina	Acc 32	46.96	0.7263
Anak Siam	Acc 129	47.06	0.1484
Anak Nalong	Acc 120	47.67	0.2072
Dulitik	Acc 412	49.52	0.6571
Burok Bakul	Acc 271	51.26	–0.1158
Morak Sepilai Kecil	Acc 1015	52.27	0.7257
Nyandal	Acc 1126	53.28	0.2678
Radin	Acc 1162	55.12	–0.0105
Bisbang A	Acc 213	55.21	0.1557
Sait A	Acc 1501	56.13	0.5295
Radin Pulau Pinang 146	Acc 1425	55.54	0.5341
Rambai	Acc 1437	56.61	–0.2030
Mayang Mandin	Acc 904	57.20	–0.1294
Che Ali Puteh	Acc 323	58.59	–2302
Anak Ulat	Acc 134	58.39	0.6411
Anak Ikan Cina	Acc 57	59.29	0.0009
Koncho Kecil 58	Acc 712	60.02	0.2514
Pandasan	Acc 1195	62.06	0.1499
Lautit	Acc 755	62.41	0.2561
Anak Ikan Tinggi	Acc 89	62.66	0.4944
Arohan C	Acc 148	62.83	0.0223
Rambai 28	Acc 1439	63.11	–0.4816
Rambutan	Acc 1444	63.29	0.1274

(cont.)

Table 3. *(cont.)*

Rice variety	Accession No./Designation	[1]Radicle (%)	[2]Trend
Jintan Besut	Acc 3782	64.05	0.7742
Seraup	Acc 1537	64.45	–0.0969
Anak Didik	Acc 36	65.11	–0.0394
Amur	Acc 26	65.11	–0.0394
Bok Soi 40	Acc 227	67.00	0.3114
Mayang Seroi 70	Acc 974	68.40	0.0187
Anak Limbot	Acc 100	69.43	0.3593
Kedinga B	Acc 677	72.36	0.5318
Morak Sepilai Kecil	Acc 1015	74.75	0.7309
Acheh Puteh	Acc 11	76.45	0.8138
Anak Ikan Sombong	Acc 73	76.74	0.3649
Jarum Emas	Acc 569	77.98	0.2044
Radin Goi 29	Acc 1394	80.30	0.3089
Pulut Karau	Acc 1277	80.57	0.1205
Radin Siak 24	Acc 1427	80.98	0.1235
Jenalek	Acc 587	85.40	0.5703
Batas	Acc 185	86.18	0.2203
Chatek	Acc 304	92.19	0.3133

[1]Lettuce radicle growth (%) = Percentage of lettuce roots growing within the root zone of the rice
[2]Trend = Gradient of regression line

Table 4. Screening of rice varieties for allelopathic potential against *Echinochloa crus-galli* using the plant box method

Rice variety	Accession No./Designation	[1]Radicle (%)	[2]Trend
Chatek Kuning	Acc 320	15.79	0.1770
Merah Isi	Acc 1007	19.95	0.1646
Singgora	Acc 1712	24.79	0.3642
Anak Didek 3	Acc 38	25.37	0.4901
Acheh Puteh	Acc 8	25.97	0.3684
Pasir	Acc 1202	26.95	0.2927
Anak Naga	Acc 113	28.31	0.2053
Padang Gelap	Acc 3986	28.52	0.4815
Pahit B	Acc 1181	29.14	0.1037
Chempaka 173	Acc 334	30.28	0.0928
Batikan	Acc 187	30.47	0.2265
Pulut Manis	Acc 4092	31.13	0.4838
Anak Gajah	Acc 47	31.30	0.2398
Tangkai Rambai 170	Acc 1772	31.76	0.2678
Siam 48	Acc 1661	31.81	0.0983
Anak Ikan Tinggi	Acc 88	32.54	0.3110
Radin Ebos 59	Acc 1371	32.80	0.1348
Panji Kuning	Acc 4187	33.09	0.3220
Jintan Puteh	Acc 620	33.44	0.1107
Anak Limbat	Acc 101	34.06	0.1160
Muar Kuning 1818	Acc 1023	34.50	0.1510
Landak	Acc 749	34.88	0.3835
Anak Ikan Cina	Acc 56	35.14	0.3826

(cont.)

Table 4. *(cont.)*

Rice variety	Accession No./Designation	[1]Radicle (%)	[2]Trend
Mayang Gerbi 101	Acc 888	35.67	0.2532
Anak Singgora	Acc 131	35.75	0.1392
Byatgele	Acc 273	37.50	0.2794
Melor	Acc 1001	37.80	0.1663
Payaw A	Acc 3592	38.00	0.2011
Siam ER 54	Acc 1687	38.18	0.2422
Gading	Acc 449	38.73	0.3905
Lembut Pandan 36	Acc 789	38.92	0.1422
Wangi	Acc 4151	39.09	0.4240
Ketitir	Acc 3905	39.61	0.3346
Radin Pahang	Acc 1423	41.15	0.2178
Kalimoch	Acc 4281	42.14	0.2439
Nyandal	Acc 1127	42.67	0.1341
Jambok	Acc 6212	43.48	0.2828
Pangsi 2	Acc 4132	44.33	0.2580
Capek	Acc 4119	44.56	0.2588
Ketitir	Acc 3955	46.60	0.3794
MR 105	Acc 7737	46.87	0.4203
Janda	Acc 565	47.33	0.3328
Anak Puteh	Acc 125	47.42	0.2985
Anak Ikan Kelubi	Acc 66	48.38	0.5103
Bunga Melor	Acc 248	49.44	0.3784
Seriap	Acc 1631	52.49	0.1923
Rambai 48	Acc 1442	52.57	0.3361
MR 76	Acc 4626	56.74	0.3828
MR 66	Acc 4618	57.20	0.0958
Che Lawi 16	Acc 325	57.78	0.4533
805A/IR44699-21-1-3-4	96C-HT4	57.88	0.2945
841-1A/BR27-35-2-1-1-1R	96C-HT13	57.91	0.2262
MR 65	Acc 4617	58.19	0.0857
841-2A/BR736-20-3-1	96C-HT27	60.53	0.2267
Tega	Acc 4053	60.56	0.4640
MR 84	Acc 4633	60.59	0.2925
MR 106	Acc 7487	61.68	0.4967
841-1A/IR54689-41-2-2	96C-HT22	61.88	0.4091
MR84/BR827-35-2-1-1-1R	96-C-HT74	62.92	0.0964
Sarawak	Acc 4150	63.26	0.3885
MR 100	Acc 7733	63.30	0.5396
MR 62	Acc 4614	64.07	0.2479
MR 5	Acc 4563	65.25	0.1059
MR 49	Acc 4602	65.26	0.1383
MR 95	Acc 7728	65.54	0.5129
MR 82	Acc 4631	66.43	0.1799
MR 20	Acc 4575	66.79	0.1233
MR 86	Acc 4635	67.19	0.4008
MR 40	Acc 4594	69.08	0.4786
MR 45	Acc 4559	69.73	0.6844
Muda 2	Acc 8476	70.50	0.1459
MR 59	Acc 4611	70.94	0.2427
MR 149	Acc 8628	73.00	0.0284

(cont.)

Table 4. *(cont.)*

Rice variety	Accession No./Designation	1Radicle (%)	2Trend
MR 148	Acc 8627	77.07	0.0583
MR 119	Acc 7832	77.75	0.1187
MR 167	Acc 8646	79.24	0.3439
IR58025A/NR163	96C-HT50	82.48	0.2841
MR 145	Acc 8624	82.75	0.1336
MR 103	Acc 7484	82.88	0.1118
MR 109	Acc 7740	84.69	–0.3587
IR62829A/MR159	96C-HT54	85.50	0.4566
841-1A/H90-69	96C-HT16	86.27	0.1606
MR 117	Acc 7870	90.07	0.3529
MR 45	Acc 4599	96.83	0.3969
MR76/IR34686-179-1-2-IR	96C-HT69	101.13	0.4577
MR85/BR736-20-3-1	96C-HT77	102.80	0.4728
805B	96PL90,mix	103.00	0.2534
Parjugan C	Acc 1201	104.00	0.4175
841-1A/MR163	96C/HT26	106.00	0.1211

1Radicle (%) = Percentage of *Echinochloa crus-galli* roots growing within the root zone of the rice
2Trend = gradient of regression line

Table 5. Screening of rice varieties for allelopathic potential against lettuce (var. Great Lakes 366) using the plant box method, tested in Allelopathy Laboratory*, Tsukuba, Japan

Rice variety	Accession No./Designation	1Radicle (%)	2Trend
Acheh Puteh	Acc 8	4	0.937
MR 58	Acc 4610	4	0.933
MR 127	Acc 7489	4	0.862
MR 85	Acc 4634	5	0.910
Haji Harun 10	Acc 502	8	0.927
Chempaka	Acc 737	8	0.924
MR 159	Acc 8638	8	0.957
MR 14	Acc 4569	9	0.922
MR 106	Acc 7487	10	0.876
Intan Merah	Acc 541	11	0.914
Muda	Acc 4557	12	0.957
MR 67	Acc 4619	12	0.787
Anak Nalong	Acc 120	13	0.916
Manik	Acc 4556	13	0.875
Seberang	Acc 4559	13	0.926
MR 15	Acc 4570	13	0.660
Kedah	Acc 672	14	0.828
Pulu Siding	Acc 4555	14	0.933
Haji Haron	Acc 498	16	0.901
Jintan Kuning	Acc 600	16	0.897
MR 39	Acc 4593	16	0.781
MR 55	Acc 4607	16	0.827
MR 103	Acc 7486	16	0.819
Anak Ikan	Acc 73	17	0.875

(cont.)

Table 5. *(cont.)*

Rice variety	Accession No./Designation	[1]Radicle (%)	[2]Trend
Anak Naga	Acc 114	17	0.943
MR 123	Acc 7488	17	0.832
MR 167	Acc 8646	18	0.754
Anak Ikan	Acc 53	19	0.900
Jarom Emas	Acc 569	19	0.886
Anak Ikan Cina	Acc 56	20	0.826
Che Mek Molek	Acc 327	21	0.936
MR 43	Acc 4597	21	0.627
Lembu Basah	Acc 777	23	0.865
MR 10 (Sekembang)	Acc 4553	23	0.735
Lembu Basah	Acc 826	24	0.810
Anak Ikan Tinggi	Acc 82	25	0.800
Makmur	Acc 4558	26	0.829
Merak Sepilai Kecil	Acc 1015	27	0.821
Bahagia	Acc 167	29	0.931
Mayang Ebos	Acc 877	29	0.780
Mayang Segumpal	Acc 956	32	0.239
Anak Limbat	Acc 100	33	0.869
Ketitir	Acc 694	33	0.574
Janda Berhias	Acc 566	35	0.884
Malinja	Acc 839	35	0.872
Mayang Gerbi	Acc 888	38	0.499
Mayang Pasir	Acc 909	39	0.780
Batas	Acc 184	46	0.841
MR 7 (Sekencang)	Acc 4552	46	–0.220
Kadaria	Acc 4554	47	0.870
Mek Bujang Kelsom	Acc 988	54	0.795
Melor	Acc 1001	60	0.678

[1]Lettuce radicle growth (%) = Percentage of lettuce roots growing within the root zone of the rice
[2]Trend = gradient of regression line
*Allelopathy Laboratory, Department of Environment Biology, National Institute of Agro-Environmental Sciences, Tsukuba, Ibaraki, 305 Japan

screened in greenhouse to identify those with allelopathic effects on *E. crus-galli* var. *crus-galli*. This is the most troublesome weed in direct-seeded rice. Forty varieties showed 50% suppression on *E. crus-galli* seedlings *(Table 4)*. Varieties such as Chatek Kuning, Merah Isi, Singgora, Anak Didek 3, Acheh Puteh, Pasir, Anak Naga and Padang Gelap, were found to have strong allelopathic activity on *E. crus-galli* seedlings. They recorded less than 30% radicle growth of *E. crus-galli*.

Allelopathic test on Malaysian indigenous rice varieties in Allelopathy Laboratory, Tsukuba, Japan Some of the rice varieties from rice germplasm were sent to Allelopathy Laboratory in Japan for further verification on allelopathic activity. A total of 52 rice varieties were screened for allelopathic activity in the laboratory. It was observed that some Malaysian rice varieties showed significant allelopathic activity against germinated lettuce. Eight varieties recorded lower percentage radicle growth of lettuce seedlings at less than 10% namely Acheh Puteh, MR 58, MR 127, MR 85, Haji Harun 10, Chempaka, MR 159 and MR 14,

Table 6. Assessment of allelopathic activity of rice varieties using sandwich method

Rice variety	Accession No./Designation	Germination (%)*	Radicle (%)*	Hypocotyl (%)*
Jambok	Acc 6212	100.0	31	88
Badshabhong	Acc 166	83.3	31	52
Putih Perak	–	77.8	32	72
Sarawak	Acc 4150	66.7	38	90
Anak Cina	Acc 32	100.0	39	69
IR 3351-38-3-1	Acc 3905	100.0	40	89
Che Lawi 16	Acc 325	83.30	42	76
Comisena	Acc 363	100.0	43	93
Anak Singgora	Acc 131	100.0	46	79
Rambai 48	Acc 1442	100.0	47	69
Landak	Acc 749	88.9	50	90
Anak ikan tinggi	Acc 88	77.7	50	53
MR 84	Acc 4633	100	51	81
Raja Muda 4	Acc 1432	72.2	52	68
Ayam	Acc 161	100.0	54	79
Mek Bujang Kelsom	Acc 988	22.2	54	78
Payaw (A)	Acc 3592	83.3	56	87
Muar Kuning 1818	Acc 1023	77.8	56	87
Anak Puteh	Acc 125	94.4	59	88
Badong	Acc 223	83.3	61	100
Anak Limbat	Acc 101	55.6	61	100
Pangsi	Acc 4132	77.8	62	91
Anak ikan kelubi	Acc 66	77.8	62	94
Cempaka 173	Acc 334	100.0	62	81
Seri Raja	Acc 1612	50.0	65	86
Padi Siam	Acc 7102	100.0	67	69
MR 185	Acc 8455	100.0	67	91
Siam 48	Acc 1661	100.0	69	87
MR 167	Acc 8646	100.0	70	93
Padang Gelap	Acc 3986	88.9.0	71	99
Siam 48	Acc 1661	44.4	71	86
Jintan Kuning	Acc 600	94.4	74	96
Singgora	Acc 1712	83.3	78	100
Muar Kuning	Acc 1023	100.0	78	100
Sesat Liar	Acc 4123	66.7	80	89
Kalimoch	Acc 4281	94.4	84	100
Seri Jaya	Acc 1624	100.0	87	100
Bunga Melor	Acc 248	100.0	89	100
Batikan	Acc 187	77.8	90	100

*Growth % to the control (lettuce)

(Table 5). In this study, low temperature gelatinizing agar (Nakarai Tesque, Kyoto, Japan) and lettuce (*Lactuna sativa*,var. Great Lakes 366) were used.

Sandwich method

Allelopathic activities of matured dry (let to rot naturally) leaves of 39 rice varieties were evaluated using the sandwich method. Varieties like Jambok, Badshabhong and Putih Perak showed strong allelopathic activity on germinated lettuce *(Table 6).* They recorded lower radicle percentages of lettuce i.e. 31%, 31% and 32% respectively.

Fallen leaves of more than 150 samples of rice field weed species were collected for

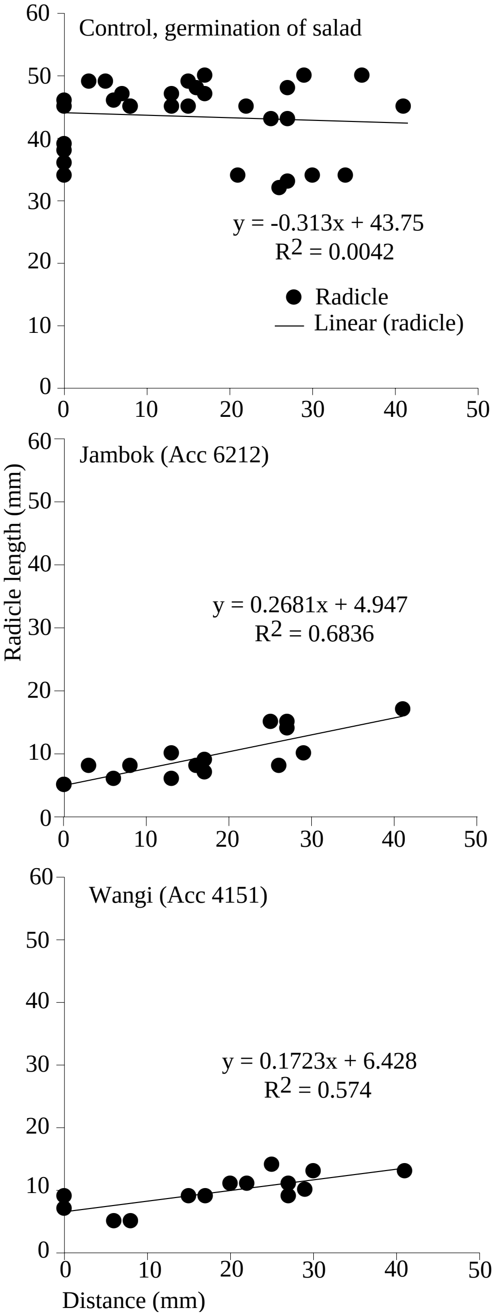

Figure 2. Assessment of allelopathy in two rice varieties using plant box method. {Distance from the membrane versus growth of acceptor lettuce plants. Intercept (radicle) and slope of regression line (trend) represents the level of activity. Jambok (middle) and Wangi (below) showed strongest inhibitory activity compared to control (top)}

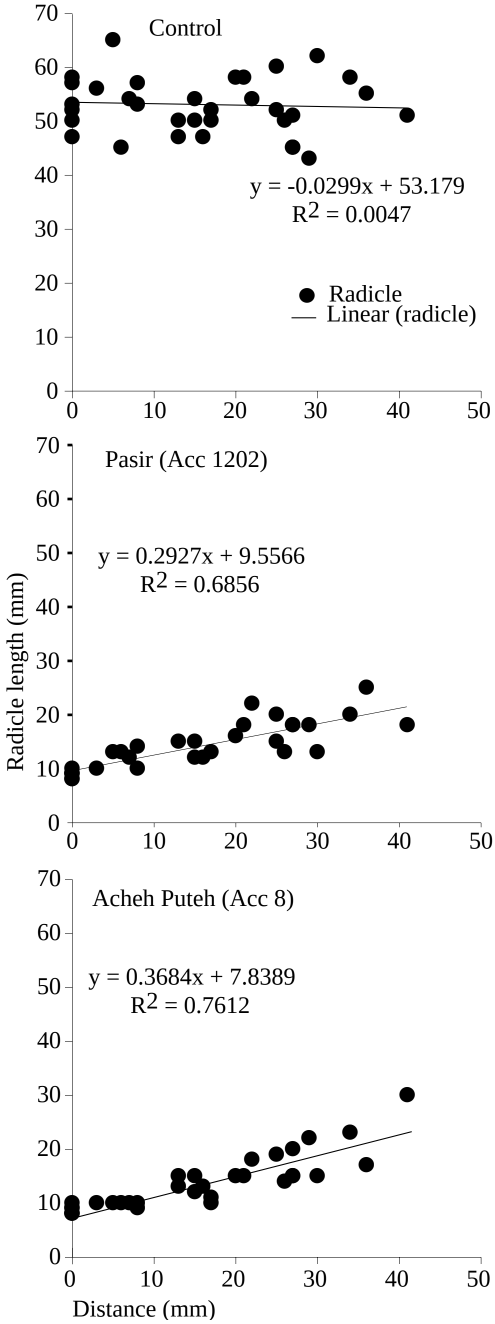

*Figure 3. Assessment of allelopthy in two rice varieties using plant box method {Distance from the membrane versus growth of acceptor **E. crus-galli** plants. Intercept (radicle) represents the level of activity. Pasir (middle) and Acheh Puteh (below) showed strongest activity compared to control (top)}*

the study. Leaves were collected mainly from rice weed species in Seberang Perai rice area. Several weed plants such as *P. purpureum*, *I. globosa* and *C. rotundus* showed strong inhibitory activity *(Table 7)*. They recorded lower percentages of lettuce radicle growth at 4%, 18% and 23% respectively.

Table 7. Assessment of allelopathic activity of rice field weed species using sandwich method

Weed species	Common name	Germination (%)	Radicle (%)*	Hypocotyl (%)*
Pennisetum purpureum	Rumput ekor kucing	54	4	0
Isachne globosa	Rumput minyak	100	18	49
Cyperus rotundus	Rumput cina lari	100	23	58
Sagittaria guyanensis	Keladi air	100	24	63
Fimbristylis miliacea	Rumput tahi kerbau	100	31	66
Chromolaena odorata	Rumput kapal terbang	100	33	42
Melastoma malabathricum	Sendudok	100	34	51
Ludwigia hyssopifolia	Jinaleh	100	36	75
Hydiotis corymbosa		100	39	49
Bacopa rotundifolia		100	39	89
Ipomea aquatica	Kangkung	100	42	96
Cyperus difformis	Para bulat	100	44	90
Cyperus acuminata		100	44	73
Sphenoclea zeylanica	Cempedak air	100	45	80
Leptochloa chinensis	Rumput miang	100	46	68
Impreta cylindrica	Lalang	74	47	44
Panicum repens	Rumput kerbau	100	47	57
Limnocharis flava	Paku rawan	100	48	78
Echinochloa crus-galli	Rumput sambau	100	48	82
Eclipta alba		100	55	98
Lindernia spp.	Kerak nasi	100	61	66
Marsilea crenata	Tapak itik	100	62	70
Oryza rufipogon	Acc 5007	100	63	96
Melochia corchorifolia		100	65	66
Ischaemum rugosum	Colok cina	100	67	85
Paspalium longifolium		100	68	77
Scirpus grossus	Menerung	100	69	83
Muelugo pentaphyla		100	70	83
Ageratum conyzoides		100	70	100
Oryza rufipogon	Acc 5009	100	71	93
Oryza rufipogon	Acc 50054	100	76	98
Oryza rufipogon	Acc 50083 (padi liar/hantu)	100	77	101
Cyperus haspan	Para	100	79	85
Oryza rufipogon	Acc 50114	100	79	88
Eichhornia crassipes	Keladi bunting	100	80	81
Conyza sumatrensis		100	80	80
Oryza rufipogon	Acc 50028	100	81	117
Paspalum vaginatum	Rumput masin	100	82	75
Oryza sativa (padi angin)	Dark grain	100	84	110
Oryza sativa (padi angin)	Grain like mahsuri	100	86	111
Hymenachne pseudointerrupta	Rumput colok	100	87	103
Gnaphalium indicum		100	88	95
Echinochloa stagnina	Sambau merah	100	89	105

(cont.)

Table 7. *(cont.)*

Weed species	Common name	Germination (%)	Radicle (%)*	Hypocotyl (%)*
Brachiara mutica	Rumput bulu	100	91	100
Eragrostis amabilis	Rumput minyak	100	93	100
Scirpus juncoides	Rumput bulat	100	95	113
Oryza sativa (padi angin)	Dark brown with awn	100	96	100
Oryza sativa (padi angin)	Dark brown small grain	100	97	97

*Growth % to the control (lettuce)

Conclusion

Allelopathic activity may provide an alternative control method to overcome the serious unfriendly ecological problems associated with herbicide use. Even though the present study indicated that allelopathic trait exists strongly in the traditional rice germplasm but relatively less in improved lines indicated that no linkage whatsoever to agronomically important features. Rice varieties with allelopathic potential have good plant suppressing ability against germinating lettuce and important rice weed namely *E. crus-galli*. They inhibited root development of lettuce and *E. crus-galli*. Further test in the rice field to ascertain allelopathic phenomenon need to be carried out in the future.

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References

Dilday, R. H., Natasi, P., Lin, J. and Smith, Jr. R. J. (1991). Allelopathic activity of rice (*Oryza sativa* L.) against ducksalad [*Heteranthera limosa* (Sw.) Willd]. *Proc. Symp. on sustainable agriculture research service*. (Hanson, J. D,. Shaffer, M. J., Ball, D. A., and Cole, C. V., eds.) p. 193–201 Washington D. C.: USDA, ARS, ARS-89

Dilday, R. H., Yan, W. G., Moldenhauer, K. A. K. and Gravois, K. A. (1998). Allelopathic activity in rice for controlling major aquatic weeds. In *Allelopathy in rice*. (Olofsdotter, M. ed.), p. 7–26. Manila: IRRI

Duke, S. O. (1985). Biosynthesis of phenolic compounds – Chemical manipulation in higher plants. In *The chemistry of allelopathy: Biochemical interactions among plants* (Thompson, A. C., ed.) p. 113–31. IRRI

Fuerst, E. P. and Putnam, A. R. (1983). Separating the competitive and allelopathic components of interference: Theoretical principles. *J. of Chem. Ecology*. ***9(8):*** 937–44

Fujii, Y. (1993). The allelopathic effect of some rice varieties. *Technical Bulletin No.* ***134.*** FFTC

Fujii, Y., Azmi, M. and Feng, C. F. (1999). Allelopathic effects of plants and rice in the Muda area in Malaysia. In *The management of biotic agents in direct seeded rice culture in Malaysia*. p.166–73. Tsukuba: JIRCAS

Fujii, Y. and Shibuya, T. (1991). A new bioassay for allelopathy with agar medium. II. Mixed culture of allelopathic candidates with acceptor plants in agar medium. *Weed Res. Japan (Suppl)****36:*** 152–3

Gliessman, S. R. (1982). Allelopathy and biological weed control in agroecosystem. *Proc. sem. on allelochemicals and pheromones*. p. 77–86

Lin, J., Smith, Jr. R. J. and Dilday, R. H. (1992). Allelopathic activity of rice germplasm on weeds. In *Proc. 45th annual meeting of the Southern Weed Science Society*, p. 99. Little Rock, Arkansas, U. S. A.

Moody, K. (1992). Efficient herbicide use in tropical crops. *Proc. 1st International Weed Control Congress, Volume 1*. p. 220–223. Melbourne: Weed Sci. Soc. of Victoria Inc.

Olofsdotter, M. and Navarez, D. (1995). Approaches in allelopathy research. *Proc. of 15th Asian-Pacific Weed Science Society Conference,* (24–28 July, 1995, p. 315–20. Tsukuba, Japan: East Japan Printing Co.

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