รายงานวิจัยฉบับสมบูรณ์

ศักยภาพของข้าวไร่ในการลดการแข่งขันของวัชพืช

โดย

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30 มิถุนายน 2543

FINAL REPORT

ALLELOPATHIC POTENTIAL OF UPLAND RICE TO REDUCE WEED COMPETITION

BY

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30 JNUE 2000

สัญญาเลขที่ PDF/ 47/ 2541

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สนับสนุนโครงการโดยสำนักงานกองทุนสนับสนุนการวิจัย

ชุดโครงการทุนวิจัยหลังปริญญาเอก

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ผู้วิจัยขอขอบคุณ สำนักงานกองทุนสนับสนุนการวิจัยเป็นอย่างสูง ที่สนับสนุนด้านการเงินสำหรับงานวิจัยหลังปริญญาเอกในครั้งนี้

ขอบคุณ กองพฤกษศาสตร์และวัชพืช กรมวิชาการเกษตร ที่ให้ความอนุเคราะห์สถานที่ห้องปฏิบัติการ เรือนทดลอง และเครื่องมือวิทยาศาสตร์

ขอบคุณ คุณฉวีวรรณ วุฒิญาโณ ธนาคารเชื้อพันธุ์ข้าว ศูนย์วิจัยข้าวปทุมธานี ที่เอื้อเฟื้อเมล็ดพันธุ์ข้าวไร่สำหรับงานวิจัย

ขอบคุณ ดร. พูนพิภพ เกษมทรัพย์ มหาวิทยาลัยเกษตรศาสตร์ที่แนะนำเทคนิคการใช้เครื่องมือวัดการสังเคราะห์แสง

ท้ายที่สุด ขอขอบคุณ ศ. ดร. รังสิต สุวรรณเขตนิคม และ ศ. ดร.เบญจวรรณ ฤกษ์เกษม ที่ให้คำปรึกษาและคำแนะนำในการวิจัยมาโดยตลอด

บทคัดย่อ

สัญญาเลขที่: PDF/ 47/ 2541

ชื่อโครงการ: ศักยภาพของข้าวไร่ในการลดการแข่งขันของวัชพืช

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วัตถุประสงค์:

เพื่อศึกษาแนวทางการควบคุมวัชพืชในข้าวไร่โดยชีววิธี (allelopathy)

ขอบเขตการวิจัย:

คัดเลือกสายพันธุ์ข้าวไร่ จำนวน 569 สายพันธุ์ จากธนาคารเชื้อพันธุ์ข้าว กรมวิชาการเกษตร เพื่อทดสอบศักยภาพในการลดการเจริญเติบโตของวัชพืชเมื่อปลูกร่วมกัน นำสายพันธุ์ที่มีศักยภาพในการลดการเจริญเติบโตของวัชพืชในระดับต่างกัน มาทดสอบด้วยวัชพืชสำคัญที่พบทั่วไปในระบบการปลูกข้าวไร่ ทดสอบศักยภาพในการลดการเจริญเติบโตของวัชพืชเมื่อปลูกร่วมกับข้าวที่มีอายุต่างๆกัน และศึกษาความแตกต่างทางด้านการสังเคราะห์แสงของสายพันธุ์ข้าวไร่ที่คัดเลือก

ผลที่ได้รับ:

พบว่าสายพันธุ์ข้าวไร่ 23 สายพันธุ์ มีศักยภาพในการลดการเจริญเติบโตของพืชทดสอบได้ และสายพันธุ์ที่คัดเลือกไว้ 2 สายพันธุ์ (BWSD 16 และ BWSD 19) สามารถลดการเจริญเติบโตของวัชพืชใบแคบและใบกว้างได้ในสภาพเรือนทดลอง

สรุปสาระสำคัญของผลที่ได้จากการวิจัย:

การควบคุมวัชพืชในข้าวไร่โดยชีววิธี (allelopathy) สามารถกระทำได้โดยใช้สายพันธุ์ข้าวที่มีศักยภาพ ในการลดการเจริญเติบโตของวัชพืช แต่จำเป็นต้องศึกษาต่อไปในระบบการปลูกข้าวไร่ ซึ่งมีปัจจัยของสภาพแวดล้อมเข้ามาเกี่ยวข้องด้วย

ข้อเสนอแนะสำหรับงานวิจัยในอนาคต:

นำสายพันธุ์ข้าวไร่ที่มีศักยภาพในการลดการเจริญเติบโตของวัชพืชไปทดสอบในสภาพไร่ ซึ่งมีความหลากหลายของชนิดวัชพืช และมีสภาพแวดล้อมที่แตกต่างจากการทดลองในห้องปฏิบัติการหรือเรือนทดลอง หากพบว่าสายพันธุ์ดังกล่าวยังคงมีศักยภาพในการลดการแข่งขันของวัชพืชในสภาพไร่ได้ จะได้นำสายพันธุ์นั้นไปใช้ในการปรับปรุงพันธุ์ข้าวไร่ เพื่อนำไปสู่เป้าหมายลดการใช้สารกำจัดวัชพืชในการปลูกข้าวไร่ต่อไป

คำหลัก: ข้าวไร่ การควบคุมวัชพืชโดยชีววิธี การแข่งขันของวัชพืช

Abstract

Project Code: PDF/ 47/ 2541

Project Title: Allelopathic potential of upland rice to reduce weed competition

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Project Period: 1 JULY 1998- 30 JUNE 2000

Objectives: To use allelopathy as a potential strategy for biological rice weed control.

Methodology: Allelopathic potential was investigated in 569 rice accessions under laboratory condition. Four selected accessions were tested for allelopathic potential at different growth stages of rice. Selectivity of allelopathic rice on weed species was also determined. Photosynthesis of allopathic and non-allopathic accessions was investigated.

Results: Twenty three rice accessions decreased total dry weight of *Echinochloa crusgalli and Lactuca sativa* by>50% when grown together under laboratory condition. Two selected accessions (BWSD 16 and BWSD 19) exhibited the strongest allopathic effect on two monocots and four dicots weed species under glasshouse condition.

Discussion & Conclusion: It is plausible to utilize allopathic rice accessions to reduce weed competition in the field. However, the allopathic activity of those accessions should be further determined in the upland rice system with various environmental factors.

Suggestion/ Further Implication/ Implementation: The further investigation of allopathic potential in selected accessions (BWSD 16, BWSD 19, BWSD 22 and BWSD 25) should be established in upland rice system. These rice accessions then can be used as genetic resources to improve the allopathic potential in upland rice resulting in the reduction of herbicide application.

Key words: allelopathy, upland rice, weed competition

Executive Summary

Weeds are the greatest yield-limiting constraint in upland rice. Allelopathy is a potential strategy for biological weed control in rice. Allopathic potential in upland rice (*Oryza sativa* L.) was investigated in 569 accessions under laboratory condition. Results indicated that genotypic variation for allopathic potential exists in Thai upland rice germplasm. Twenty three rice accessions decreased total dry weight of *Echinochloa crusgalli* and *Lactuca sativa* by more than 50% when grown together. Most accessions with allelopathic potential were collected from the Northern Thailand. Four selected accessions (BWSD 16, BWSD 19, BWSD 22 and BWSD 25) were selected and tested in glasshouse with two monocots (*E. colona* and *E. crusgalli* and four dicots (*Euphorbia geniculata, Mimosa invisa, M. pudica* and *L. sativa)* weed species. Variation in allelopathic effect on the test species was measured. BWSD 16 and BWSD 19 exhibited the strongest allelopathic effect. These two accessions suppressed the growth of two monocots by 49-63% and four dicots weeds by 30-73%. In contrast, BWSD 22 had a slight allopathic effect only on some of test species. BWSD 16 and BWSD 19 clearly exhibited the allelopathic activity when weed species *M. invisa* was sown at 0 and 5 days after rice germination. In addition, photosynthesis rates and chlorophyll content in both allelopathic and non-allelopathic accessions were similar. Hence, allelopathic activity does exist in selected accessions, BWSD 16 and BWSD 19.

These findings indicate that allelopathy in upland rice might be used as an alternative method for controlling weeds in upland rice system to avoid harmful effects caused by herbicides on human health and the environment.

1. INTRODUCTION

1.1 Allelopathy

The term 'allelopathy' means inhibitory or stimulatory effects by root exudates, leaf leachates, crop residues and volatile chemical emitted from plants. It can be used as a weed management tool in three different ways: by planting an allelopathic crop or cultivar; planting an allelopathic non crop species associated with the crop; or through allelopathic residues left in the soil from a previous crop (Gliessman, 1982). Weston (1996) proposed that allelopathy may offer potential for biorational weed control through the production and release of allelochemicals from leaves, flowers, seeds, stems and roots of living or decomposing plant materials. In addition, under appropriate conditions, allelochemicals may be released in quantities suppressive to developing weed seedlings. Among these methods, allelochemical exudates which is naturally produced by plant can be merely potential herbicides (Duke, 1985; Worsham, 1989).

1.2 Allelopathy in plants

The phenomena of allelopathy have been found in numerous crops. For instances, allelopathic lines of the following crops have shown inhibition of weed growth: beets (*Beta vulgaris*), lupin (*Lupinus spp.*), maize (*Zea mays*), wheat (*Triticum aestivum*), oats (*Avena spp.*), peas (*Pisum sativum*), barley (*Hordeum vulgarae*), rye (*Secale cereale)* and cucumber (*Cucumis sativus*) (Rice, 1984). Putnam and Duke (1974) found some of 526 accessions of cucumber, originating from 41 countries, exhibiting powerful growth inhibition of *Panicum miliaceum* and *Brassica hirta*. Interestingly, one of these accessions showed 87% growth inhibition of the tested species and 25 other accessions had a 50% growth inhibitory effect. Later, a field study indicated that allelopathic activity could only be obtained under certain conditions. Total weed population was reduced by 54% in one accession but inhibition did not occur consistently and rainfall could suppressed this allelopathic effect

(Lockerman and Putnam, 1979).

In *Avena* spp., 3,000 accessions were tested for their ability to exude scopoletin, a compound known to inhibit root growth (Fay and Duke, 1977). It was found that four accessions exuded three times of scopoletin and inhibited the growth of wild mustard in the sand culture. The phytotoxic symptoms of tester plants were chlorotic, stunted and/or twisted, indicating that inhibition was caused by chemical rather than by competition.

Barley (*Hordeum vulgarae*) has been known as a "smother" crop because of its ability to suppress weeds. Overland (1966) reported that barley produced and released phytotoxic substances in concentartions that inhibited the growth of several common weeds. To date, none of allelopathic cultivar has been commercialized for any of the crops mentioned, however, these results indicated that the transfer of allelopathic genes is relevant to weed control research (Burke, 1987).

1.3 Allelopathy in rice

Allelopathy in rice was firstly observed in seed production plots which were naturally infested with ducksalad (*Hetheranthera limosa*) at Arkansas Rice research Institute, USA (Dilday *et. al.*, 1989a and 1991). During 1988-1989, 10,000 accessions of rice were evaluated for allelopathic potential in ducksalad. Of these, 3.5% were categorized as allelopathic when they had a weed free radius at the base of rice plant >10 cm. The screening experiment was also conducted for allelopathic activity in 5,000 rice accessions against some weed species such as *Ammania coccinea*, *Echinochloa crusgalli*, *Brachiaria platyphylla* and *Heteranthera limosa*. Two hundred (3.9%) of these accessions from 27 countries demonstrated allelopathic potential against one or more of these weed species (Dilday *et. al.*, 1989b). A follow-up study using a strongly allelopathic accession planted in a weed mixture showed 72-95% control of *H. limosa, A. coccinea* and *Bacopa rodundifolia* (Lin *et. al.*, 1992a,b). Later, 12,000 rice accessions were evaluated for allelopathic potential against ducksalad and

5,000 accessions against redstem (Purple ammania). The results showed that 412 rice accessions were allelopathic to redstem, 145 accessions to ducksalad and 16 accessions to both species (Dilday *et. al.*, 1997 and 1998).

In Japan, 189 rice strains were tested for allelopathic potential in the laboratory using the 'Plant Box Method' (Fujii, 1993). Diversity of rice strains were Japonica type (72 accessions), Indica type (18), Tropical Japonica type (32), Chinese type (29), African type (4) and 34 unidentified strains. Each strain was planted together with lettuce (*Lactuca sativa*). The result showed that twenty four strains had more than 75% inhibition on radicle growth of lettuce. It was noted that Tropical Japonica types had stronger allelopathic potential than the improved cultivars.

In Egypt, field screening for allelopathic potential of 1,000 rice accessions was conducted. A natural infestation of target weed species, *E. crusgalli* and *Cyperus difformis* occurred in the field. Thirty accessions of those showed promising allelopathic activity (50-90% weed reduction) against *E. crusgalli* and 15 were allelopathic (30-75% weed reduction) against *C. difformis*. Five accessions showed strong allelopathic potential for both weed species (Hassan *et. al.*, 1997 and 1998).

At International Rice Research Institute, IRRI, a laboratory screening procedure for allelopathic rice had been established (Navarez and Olofsdotter, 1996). Seeds of *E. crusgalli* were placed in the petri-dish with 7-day-old rice seedlings. The plants were grown together for 10 days before measuring the root length of tester plant. Allelopathic rice caused strong inhibition of root growth of weed. The result was confirmed in the field in 1995 and 1996. It was found that 19 of 111 rice accessions was capable to reduce growth of *E. crusgalli* by >40% and 7 accessions suppressed *E. crusgalli* dry weight by>50%. In addition, two rice accessions had a strong inhibition of both grass weed *E. crusgalli* and broadleaved weed *Trianthema portulacastrum* (Olofsdotter *et. al.*, 1997).

It is likely that allelopathy is widespread in rice germplasm since those allelopathic rice accessions were originated in 37 countries around the world. In Thailand, preliminary laboratory study among 50 upland rice genotypes confirmed that allelopathic rice genotypes exist (Maneechote and Krasaesindhu, 1996). As more than 1,000 accessions of upland rice in Rice Germplasm Bank of Thailand are still needed to be screened for the stronger allelophatic effects.

1.4 Allelopathic chemical in root exudates

With a few exceptions, the allelopathic chemicals reported from higher plants are secondary compounds that arise from either the acetate or shikimate pathway (Einhellig, 1995). Higher plants regularly release organic compounds by volatilization from their surfaces, and through leaf leachates and root exudates. Root exudates comprise both high and low-molecular weight solutes released or secreted by roots. The most important components of the high molecular weight (HMW) solutes are mucilage and ectoenzyme and those of the low-molecular weight (LMW) fractions are organic acids, sugars, phenolic acids and amino acids (Marschner, 1995). So far, no breakthroughs have been achieved in trying to characterize chemicals involved in rice allelopathy (Olofsdotter *et. al.*, 1997). Recently, some allelopathic compounds were identified in water from allelopathic rice cultivars as 3-hydroxybenzoic acid (3HBA), 4-hydroxybenzoic acid (4HBA), 4-hydroxyhydrocinnamic acid (4HHCA), 3,4-dihydroxyhydrocinnamic acid (3,4DHHCA) and tentatively identified 4-hydroxyphenylacetic acid (4HPAA) when compared with water from non-allelopathic rice cultivars (Mattice *et. al.*, 1997 and 1998).

Unfortunately, chemical approaches in allelopathy were often performed under arbitrary condition. Rather than collecting the responsible extracellular inhibitors from the intact, living donor plants, tissue extracts from either fresh or dried plant materials were commonly used (Rice, 1984). Compounds identified in this manner, however, were not necessarily responsible for the observed allelopathic interactions. Collection of allelopathic chemicals from undisturbed plant root system is difficult because of their low concentrations and the high level of contaminants in growth media such as soil

(Tang and Young, 1982).

1.5 Plant growth stages and allelopathic potential

Plants produce different allelochemicals at different development stages. Dekker and Meggit (1983) found that most allelochemicals were released during germination and early growth. In barley, the highest amount of allelopathic alkaloids were released at 36 days after germination at the rate of less than 2g plant-1day-1 when grown in hydroponic culture (Liu and Lovett, 1993). In rice, the different stages of donors plants were used in various experiments. For examples, rice plants at 1-2 months-old (Fujii, 1993), rice seedlings at 7 days-old (Olofsdotter *et. al.*, 1997) and pregerminated rice at day 0 (Maneechote and Krasaesindhu, 1996). Nevertheless, weed control research should focus on allelopathic activity during early development stages. During this period, weeds which germinate and establish faster than crops will lead to the severe weed problems (Zimdahl, 1980). In upland rice cultivation where rice and weeds germinate at the same time, the identification of allelopathic activity in rice at the early stage is required.

1.6 Mechanisms of action of alellochemicals

Mechanisms of inhibitory effects of allelochemicals have not been adequately researched since the allelopathic compounds are secondary compounds. Allelochemicals active against higher plants are typically characterized as suppressing seed germination, causing injury to root growth and other meristems or inhibiting seedling growth (Einhellig, 1995). They can also affect mineral uptake by altering membrane function in plant roots, as when phenolic acids depolarise electrical membrane potential and thereby inhibit the absorption of mineral ions into plants (Balke, 1985).

1.7 Factors affecting quantities of allelochemicals produced by plants

The amount of allelochemicals in plants are influenced by many factors. The factors which affect the photosynthesis play an important role on allelochemical production. The light intensity, daylength, mineral deficiencies, water stress and temperature may affect the quantity of those chemicals in plants (Rice, 1974).

1.8 Screening procedures for allelopathy

In allelopathy research, it is essential for all studies to test compounds released by the living donor plant on growth and development of another living receiver plant. To prove that the effects were allelopathic, careful experimental design was used to differentiate them from the influences of competition for light, water and nutrients. Bioassays are helpful techniques to screen the large number of plant species in order to search for the allelopathic plants. Many procedues were proposed to be used for screening steps. For examples, filter papers can be used as a never ending resource of water so that two species can be germinated in the same petridish without competing for water (Liu and Lovett, 1993). Agar can also be used as a germination medium as it allows the use of very small amounts of allelochemicals (Fujii, 1993; Maneechote and Krasaesindhu, 1996) and therefore it is very useful in the fractional and extrating phase of allelopathy research (Pederson, 1986).

1.9 Tester plants

As weed seeds germinate unevenly resulting in high variation of germination rates and radicle elongation measurements. Research on allelopathic activity in rice has used different tester species. Barnyardgrass (*Echinochloa crusgalli*) is a common grass weed found in rice field and is widely used as tester plant in numerous experiments (Dilday *et. al.*, 1989a,1989b; Hassan *et. al.*, 1994; Maneechote and Krasaesindhu, 1996; Olofsdotter *et. al.*, 1997). Lettuce (*Lactuca sativa*) is quite versatile to be represent the dicot weed species because germination rate is high and uniform (Fujii, 1993; Maneechote and Krasaesindhu, 1996). The other dicots weeds, *Ammania coccinea*, *Heteranthera limosa*, and sedge, *Cyperus difformis*, were also tested for allelopathic activity in rice (Dilday *et. al.*, 1989a and 1989b). In Thailand, the major weeds found in upland rice are *Ageratum conyzoides*, *Biden pilosa*, *Mimosa invisa*, *Digitaria* spp., *Dactyloctenium aegyptium* and *Eleusine indica* (Vongsaroj, 1997).

1.10 Objectives of this project

The ultimate goal of this project was to utilize allelopathy to reduce herbicide uses and increase yield in upland rice. To reach this goal, the following objectives were addressed in the project.

1. To identify the upland rice genotypes with strong allelopathic potential.

2. To study allelopathic potential at different growth stages of rice.

3. To determine the selectivity of allelopathic rice on weed species.

2. MATERIALS AND METHODS

2.1 Plant materials

Upland rice germplasm

A total of 569 accessions of upland rice (*Oryza sativa* L.) from Rice Germplasm Bank of Rice Research Institute, Department of Agriculture, Thailand were evaluated for allelopathic activity. The origin of upland rice accession is shown in Table 2.1 and local name of accessions are in Annex 1.

Table 2.1 Number of upland rice accessions in different regions of Thailand.

Region	No. of accessions
North	270
Northeast	12
East	8
South	56
Central	29
Unknown	194
Total	**569**

Tester species

Barnyardgrass (*Echnichloa crusgalli* L. Beauv.)

Jungle rice (*Echnichloa colona* L. Link),

Painted spurge (*Euphorbia geniculata* Ort.)

Sensitive plant (*Mimosa invisa* Mart.)

Sensitive plant (*M. pudica* L.)

Lettuce cv. Grand Rapids (*Lactuca sativa* L.)

2.2 Laboratory procedure

Agar (0.5% w/v) was placed in 250-ml glass bottles with 50 mm in diameter (25 ml per bottle) and allowed to solidify. Rice and tester species were sown together in each bottle. Treatments consisted of five seeds of rice and ten seeds of a tester species. Rice seeds were placed as a single row on the agar surface and tester species sown in two rows parallel to rice row. Then the lid was closed to prevent the moisture loss. The bottles were placed in a growth room illuminated with fluorescent lights (photoperiod,12h: light intensity, approximately 50-60 μmol m^{-2} s^{-1}; temperature 27°C dark/light). Four replicates of each accession were made for each tester species using randomized block design. Experiments were carried out separately for each tester species and fifty rice accessions were assayed at a time. The tester species sown without rice was retained as controls.

Ten days after sowing, growth of tester plants were determined. Shoot length was measured as length of the longest leaf for monocots and as the combined length of hypocotyl for dicots. The dry weight of shoot and roots were taken after oven drying at 70°C for 48 h.

2.3 Glasshouse procedure

Ten-days old seedlings of rice germinated on agar were transplanted into 10 cm-diam plastic pot, containing 500 g Bangkhen clay soil (60% clay, 31% silt and 9% sand, pH 5.6, CEC 25.7 meq/100g soil). One day after transplanting, ten germinated seeds of each tester species were sown in the same manner as in the bottle. Plants were grown in the glasshouse with soil maintained at or near field capacity by daily watering. Rice accessions were arranged in four replicates using Completely Block Design. Growth of tester plants (plant height and number of leaves) was determined. Sixty days after sowing, tester plants were harvested and measured for leaf number, length and dry weight of shoot/root.

2.4 Identification of upland rice genotypes with strong allelopathic activity

Five hundred sixty nine accessions of upland rice from Rice Germplasm Bank in Thailand were tested for allelopathic activity. The experiments were conducted in the laboratory by using the agar method (Maneechote and Krasaesindhu, 1996). Fifty accessions of rice were tested at a time. Barnyardgrass (*Echinochloa crusgalli*) and lettuce (*Lactuca sativa* cv. Grand Rapids) represent the monocots and dicots tester species, respectively. The shoot and root length of tester plants were measured 10 days after planting. Dry weight of shoot and root of tester species were measured separately. Control check (without rice and with non-allelopathic rice) will be included in each bioassay.

Response of each tester species was calculated as relative growth as followed.

$$\textit{Relative Dry Weight} = \frac{\text{total dry weight of tester species grown with rice}}{\text{total dry weight of tester species grown without rice}} \times 100\%$$

Based on the relative dry weight of each tester species, the rice accessions were classified into four groups:

0-25% = highly allelopathic

26-50% = moderately allelopathic

51-75% = slightly allelopathic

76-100% = no allelopathic

This classification is useful for characterizing allelopathic potential present in upland rice accessions and the representative from each group will be used as plant materials in the second experiment.

2.5 Allelopathic activity in rice to major weed species

To evaluate rice genotypic differences in allelopathic potential against some common weed species in upland rice plantation, four accessions (BWSD 16 and BWSD 19, BWSD 22 and BWSD 25) were selected. Two accessions (BWSD 16 and BWSD 19) are moderately allelopathic giving the relative growth of *L. sativa* by 26-50%. BWSD 22 is slightly allelopathic with <25% inhibition on tester species. BWSD 25, which had no inhibitory effects on both species, was used as non-allelopathic check. Those selected four accessions were repeatedly bioassayed with *L. sativa*

Ten days later, rice seedlings were transplanted to the soil for seed multiplication. Rice seeds harvested from selected accessions were separately tested for allelopathic potential with six tester species in the laboratory. Consequently, five seedlings in each bottle were transplanted into 10 cm-diam plastic pot, containing 500g Bangkhen clay soil. One day after transplanting, ten germinated seeds of each tester species were sown in the same manner as in the bottle. Plants were grown in the glasshouse with soil maintained at or near field capacity by daily watering. Rice accessions were arranged in four replicates using Complete Block Design. Growth of tester plants (plant height and number of leaves) was weekly determined. Thirty days after sowing, tester plants were harvested. Leaf area, length and dry weight of shoot/root of tester plants were measured.

2.6 Allelopathic potential at different growth stages of rice.

To verify whether there were differences between allelopathic and non-allelopathic rice, in allelopathy at the different growth stages of rice. Four selected accessions of rice (BWSD 16, BWSD 19, BWSD 22 and BWSD 25) were planted in soil. Five germinated seeds of each accession were placed in 30 cm-diam plastic pot, containing 5 kg Bangkhen clay soil. At various growth stages of rice (at 0, 5, 10, 15, 20, 25, 30 and 35 days after rice germination), tester species was planted in the same manner as in the bottle. Plants were kept in the glasshouse with soil moisture maintained at or near field capacity by daily watering. Pots were arranged in four replicates using Complete Block Design. Growth of tester plants (plant height and number of leaves) was weekly determined. Sixty days after sowing rice seeds, rice and tester plants were harvested. Leaf area, length and dry weight of shoot/root of tester plants were measured. The growth parameters of both donors and receiver plants were evaluated.

2.7 Physiological characteristics of allelopathic upland rice

To evaluate the physiological differences between the allelopathic and non-allelopathic rice accessions, plants were grown in the 30 cm-diam plastic pots in the glasshouse. Three plants of each accession were grown in a pot. At the tillering stages, single leaf net photosynthesis in the 1^{st} and 2^{nd} leaf of each plant were collected using a portable photosynthesis system model LI-6400 (Li-Cor, Lincoln, NE) at 1,500 $\mu mol.m^{-2}.s^{-1}$ light intensity. Air flow to the chamber was adjusted to give approximately 50% humidity. Leaf temperature was kept at $28 \pm 1^{\circ}C$. Data was taken when photosynthesis was steady. Chlorophyll content of each accession was also determined by chlorophyll meter (Minolta, Japan).

For each accession, photosynthesis of flag leaf was measured at various light intensity (100, 200, 500, 1000, 1500, 2000 $\mu mol.m^{-2}.s^{-1}$) at the booting stage. Each plant was used as one replicate. Four plants were measured for each accession.

3. GENOTYPIC VARIATION OF ALLELOPATHIC ACTIVITY IN UPLAND RICE

3.1 Allelopathic activity on *Echinochloa crusgalli*

The responses of *E. crusgalli* to 569 accessions of upland rice were determined under laboratory condition. Shoot and root length of test species was inhibited when grown with allelopathic accessions (data not shown). Most of rice accessions (87.7%) tested did not contain allelopathic potential. None of the rice accessions showed 0-25% relative growth of tester species. Only one accession (0.2%) was grouped in the 26-50%. About 12% of 569 accessions could reduce relative growth of *E. crusgalli* by 25-50% (Table 3.1). It is obvious that allelopathic rice originated in the north where the hilltribes population are greater than any other part of Thailand.

In the north, upland rice accessions are mostly widespread in Chiangmai with 16 accessions were slightly allelopathic to *E. crusgalli* (Table 3.2). In northeast, only one accession from Nakonratchasima gave 51-75% relative growth of test species. Most of the slightly allelopathic rice (35 accessions) was from unknown origin (Table 3.2).

Table 3.1 Distribution of upland rice accessions based on relative growth of two test species *E. crusgalli*.

Region	Number of accessions (%)				Total
	Relative growth				
	0-25	26-50	51-75	76-100	
E. crusgalli					
North	0	0	4.6	26.9	31.5
Northeast	0	0	0.2	1.9	2.1
East	0	0	0	4.0	4.0
South	0	0	0.4	9.5	9.8
Central	0	0	0.9	17.6	18.5
Unknown	0	0.2	6.2	27.8	34.1
Total	**0**	**0.2**	**12.1**	**87.7**	**100.0**

Table 3.2 Responses of *Echinochloa crusgalli* to 569 accessions of upland rice from different regions of Thailand. Data are four replicates of relative growth of tester species germinated with rice compared to that of tester species germinated alone.

Region	Number of accessions				Total
	Relative growth (%)				
	0-25	26-50	51-75	76-100	
NORTH					
Chiangrai	0	0	0	1	1
Chiangmai	0	0	16	49	65
Kamphaengphet	0	0	0	37	37
Lumpoon	0	0	0	7	7
Maehongsorn	0	0	0	7	7
Nan	0	0	6	7	13
Pichit	0	0	0	10	10
Pitsanuloke	0	0	2	6	8
Prae	0	0	0	4	4
Sukhothai	0	0	0	7	7
Utaithani	0	0	4	87	91
Uttradit	0	0	2	18	20
Total	0	*0*	*30*	*240*	***270***
NORTHEAST					
Khonkaen	0	0	0	2	2
Loei	0	0	0	5	5
Mookdaharn	0	0	0	1	1
Nakonpanom	0	0	0	1	1
Nakonratchasima	0	0	1	1	2
Nongkhai	0	0	0	1	1
Total	*0*	*0*	*1*	*11*	***12***
EAST					
Chantaburi	0	0	0	2	2
Kanchanaburi	0	0	0	15	15
Rayong	0	0	0	3	3
Trat	0	0	0	3	3
Total	*0*	*0*	*0*	*23*	***23***
SOUTH					
Chumporn	0	0	0	2	2
Naratiwat	0	0	0	3	3
Pattaloong	0	0	2	44	46
Phangha	0	0	0	5	5
Total	*0*	*0*	*2*	*54*	***56***
CENTRAL					
Patumthani	0	0	0	1	1
Nakornpathom	0	0	1	2	3
Ratchaburi	0	0	0	1	1
Saraburi	0	0	0	9	9
Total	0	0	1	13	***14***
UNKNOWN	*0*	*1*	*35*	*158*	***194***
TOTAL	*0*	*1*	*69*	*499*	**569**

3.2 allelopathic activity on *Lactuca sativa*

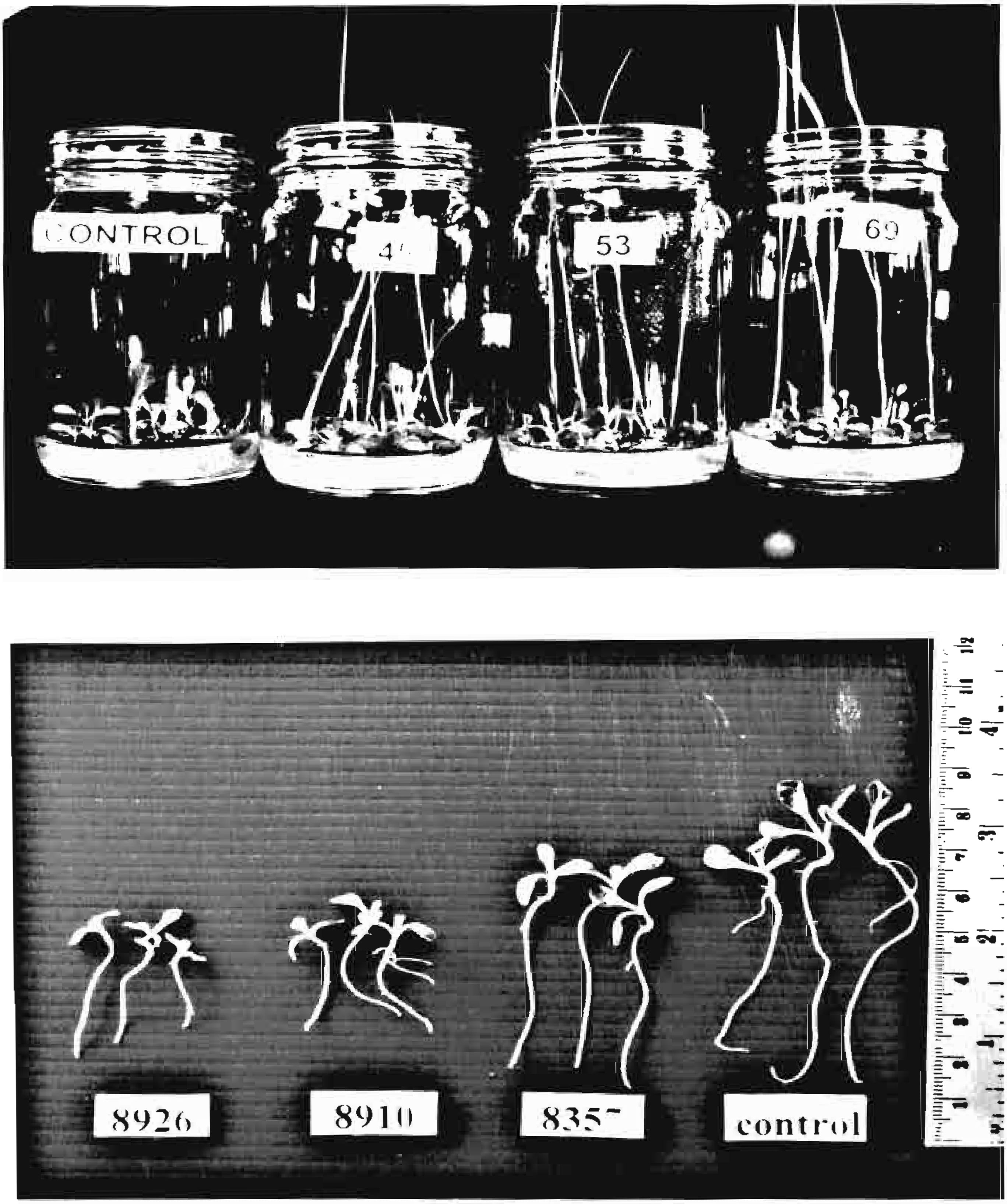
The responses *L. sativa* to 569 accessions of upland rice were determined. Ten days after germination, shoot and root length of test species were inhibited when grown with allelopathic rice accessions (Picture 3.1). None of the rice accessions showed 0-25% relative dry weight of tester species (Table 3.3). The relative growth of tester species, was ranged in 26-100%. When tested with *L. sativa,* 23 accessions were ranked into 26-50%, 82 into 51-75% and 464 into 76-100% relative dry weight of the tester. Only 4% of the 569 accessions were moderately allelopathic to the test species (Table 3.3).

Most of moderately allelopathic rice (16 accessions) originated in the Northern Thailand, particularly in Chiangmai and Utaithani (Table 3.4). It is interesting to note that allelopathic rice was not found in the other regions of Thailand. About 464 accessions of 569 accessions were found to be non-allelopathic.

Total dry weight of test species, *L. sativa* and *E. crusgalli*, was depressed by allelopathic rice accessions. The relative growth of test species was reduced by 50-75% when grown with allelopathic accession (Table 3.2 and 3.3). Shoot and root length of two test species were also inhibited (data not shown). A genotypic variation of allelopathic acitvity in Thai upland rice germplasm is suggested by these results. About 4% of germplasm were allelopathic against L. sativa. Similarly, in USDA-ARS collection, about 3.75% of rice germplasm had allelopathic effects against test species (Dildays *et. al.*, 1998). In addition, The inhibitory effect of allelopathic rice depended upon the plant species. *L. sativa* was more sensitive to allelopathic activity than *E. crusgalli.* It may be due to the differences in species. *L. sativa* is dicots crop plants which is more sensitive than the *E. crusgalli*, moncots grass weed. However, the laboratory and field study in Philippines showed that 17% of rice accessions suppressed the growth of *E. crusgalli* by more than 40% (IRRI, 1998). Differences in inhibitory activity of allelopathic rice may be due to the screening procedure and the stage of rice plant since plants produce different allelochemicals at different development stages. Dekker and Meggit (1983) found that most allelochemicals were released during germination and early growth.

The degree of inhibition was greater with the increasing growth stages of rice. Fujii (1993) used plant box method with 30-60 days-old rice plants. Olofsdotter *et. al.*, (1997) assayed rice seedlings at 7 days-old. In this study, rice and *E. crusgalli* germinated at the same time. Among those procedures, plant box method by Fujii (19993) had greater effects on test species than any other procedures. Nevertheless, weed control research should focus on allelopathic activity during early development stages. During this period, weeds which germinate and establish faster than crops will lead to the severe weed problems (Zimdahl, 1980). In upland rice cultivation where rice and weeds germinate at the same time, the identification of allelopathic activity in rice at the early stage is required.

Although allelopathic rice could suppress the growth of test species when grown on agar, the allelopathic activity of these accessions need to be further investigated when grown in the soil. Selectivity of allelopathic activity on various weed species is also required.



Picture 3.1 Shoot and root length of *L. sativa* were inhibited when grown with allelopathic rice accessions.

Table 3.3 Distribution of upland rice accessions based on relative growth of two test species, *L. sativa*.

Region	**Number of accessions (%)**				**Total**
	Relative growth				
	0-25	**26-50**	**51-75**	**76-100**	
L. sativa					
North	0	0.5	3.0	27.9	31.5
Northeast	0	0.2	0.4	1.6	2.1
East	0	0	0.2	3.9	4.0
South	0	0	0	9.8	9.8
Central	0	2.3	7.0	9.1	18.5
Unknown	0	1.1	3.9	29.2	34.1
Total	**0**	**4**	**14.4**	**81.6**	**100.0**

Table 3.3 Responses of *Lactuca sativa* to 569 accessions of upland rice from different regions in Thailand. Data are four replicates of relative growth of tester species.

Region	Number of rice accessions				Total
	Relative growth (%)				
	0-25	26-50	51-75	76-100	
NORTH					
Chiangrai	0	0	0	1	1
Chiangmai	0	3	5	57	65
Kamphaengphet	0	0	3	34	37
Lumpoon	0	0	0	7	7
Maehongsorn	0	0	1	6	7
Nan	0	0	1	12	13
Pichit	0	0	1	9	10
Pitsanuloke	0	0	0	8	8
Prae	0	0	1	3	4
Sukhothai	0	0	0	7	7
Utaithani	0	13	40	38	91
Uttradit	0	0	5	15	20
Total	*0*	*16*	*57*	*197*	***270***
NORTHEAST					
Khonkaen	0	0	0	2	2
Loei	0	0	2	3	5
Mookdaharn	0	1	0	0	1
Nakonpanom	0	0	0	1	1
Nakonratchasima	0	0	0	2	2
Nongkhai	0	0	0	1	1
Total	*0*	*1*	*2*	*9*	***12***
EAST					
Chantaburi	0	0	1	1	2
Kanchanaburi	0	0	0	15	15
Rayong	0	0	0	3	3
Trat	0	0	0	3	3
Total	*0*	*0*	*1*	*22*	***23***
SOUTH					
Chumporn	0	0	0	2	2
Naratiwat	0	0	0	3	3
Pattaloong	0	0	0	46	46
Phangha	0	0	0	5	5
Total	*0*	*0*	*0*	*56*	***56***
CENTRAL					
Patumthani	0	0	0	1	1
Nakornpathom	0	0	0	3	3
Ratchaburi	0	0	0	1	1
Saraburi	0	0	0	9	9
Total	*0*	*0*	*0*	14	***14***
UNKNOWN	0	6	22	166	**194**
TOTAL	**0**	**23**	**82**	**464**	**569**

4. ALLELOPATHIC ACTIVITY OF SELECTED UPLAND RICE ACCESSIONS ON MAJOR WEED SPECIES

4.1 Effects on monocots species

4.1.1 *Echinochloa colona*

Shoot length of test species, *E. colona*, was significantly inhibited at 30 days after emergence compared with the control (data not shown). At harvesting, shoot dry weight per plant of test species was significantly affected by BWSD 16 and was slightly reduced by BWSD 19 (Table 4.1). Total dry weight per plant of test species was highly inhibited by BWSD 16 and BWSD 19 while that of test species was slightly affected by BWSD 22. Leaf area of test species was also minimized when grown with BWSD 16 and BWSD 19. In contrast, leaf area of test species was not affected by neither BWSD 22 nor BWSD 25 (Table 4.1). However, shoot length of test species was more susceptible to BWSD 16, BWSD 19 and BWSD 22 compared with the non-allelopathic check BWSD 25 (Figure 4.1). It is noticeable that allelopathic effects were consistent with leaf area, shoot length and total dry weight of *E. colona*. The color of leaves of test species in the treatment with BWSD 16 and BWSD 19 were yellowish green whereas that with BWSD 25 (check) was green (Picture 4.1).

4.1.2 *Echinochloa crusgalli*

Shoot length of *E. crusgalli* was moderately reduced with tested with BWSD 16 and BWSD 19. However, BWSD 22 and BWSD 25 did not inhibit shoot length of the test species. In addition, shoot and root growth of *E. crusgalli* was markedly reduced when grown together with BWSD 16 and BWSD 19 (Table 4.2). Leaf area of each test plant was reduced by 75, 60 and 45% when tested with BWSD 16, BWSD 19 and BWSD 22, respectively (Figure 4.2).

Table 4.1 Effects of four selected rice accessions on shoot length and dry weight of shoot and root of *Echinochloa colona L.* Link. Data are the mean of four replicates. Ten seedlings of test species were used as one replicate.

Rice accession	Shoot length (cm)	Shoot DW (mg)	Root DW (mg)	Total DW (mg)
BWSD 16	33.5 a[1]	628 a	215 a	843 a
BWSD 19	33.6 a	1017 ab	284 a	1301 ab
BWSD 22	37.5 a	1566 b	399 a	1965 bc
BWSD 25	46.3 b	1759 b	607 b	2366 c

[1] Means in column followed by the same letter are not significantly different by Duncan Multiple Range Test at $p < 0.05$

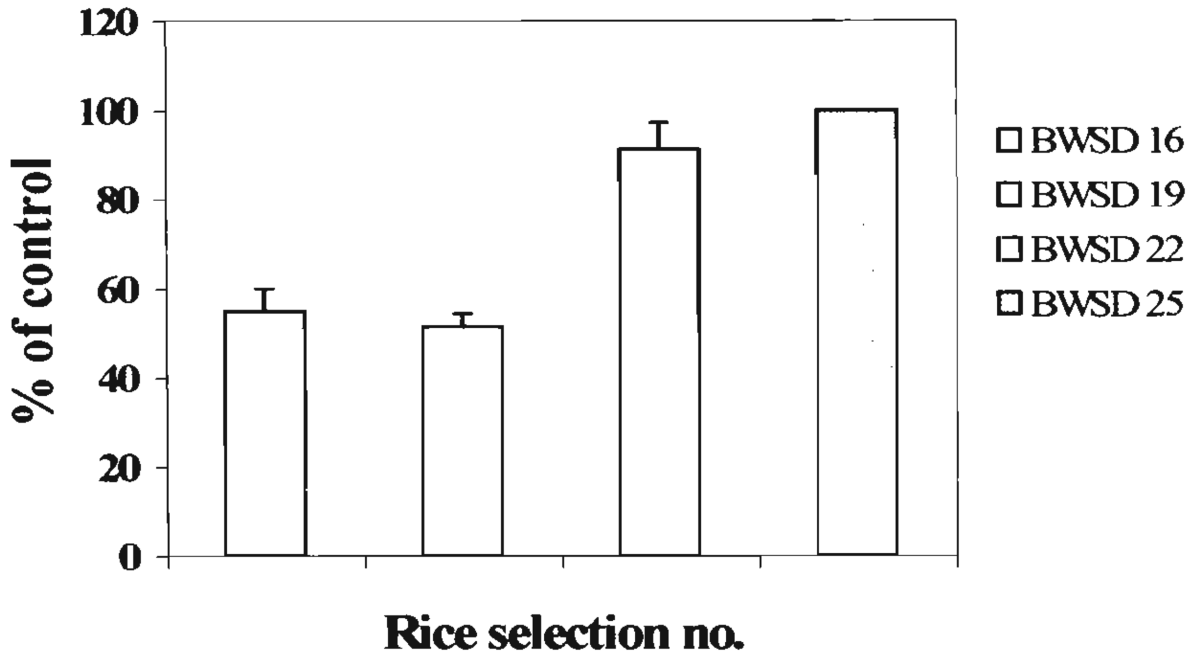
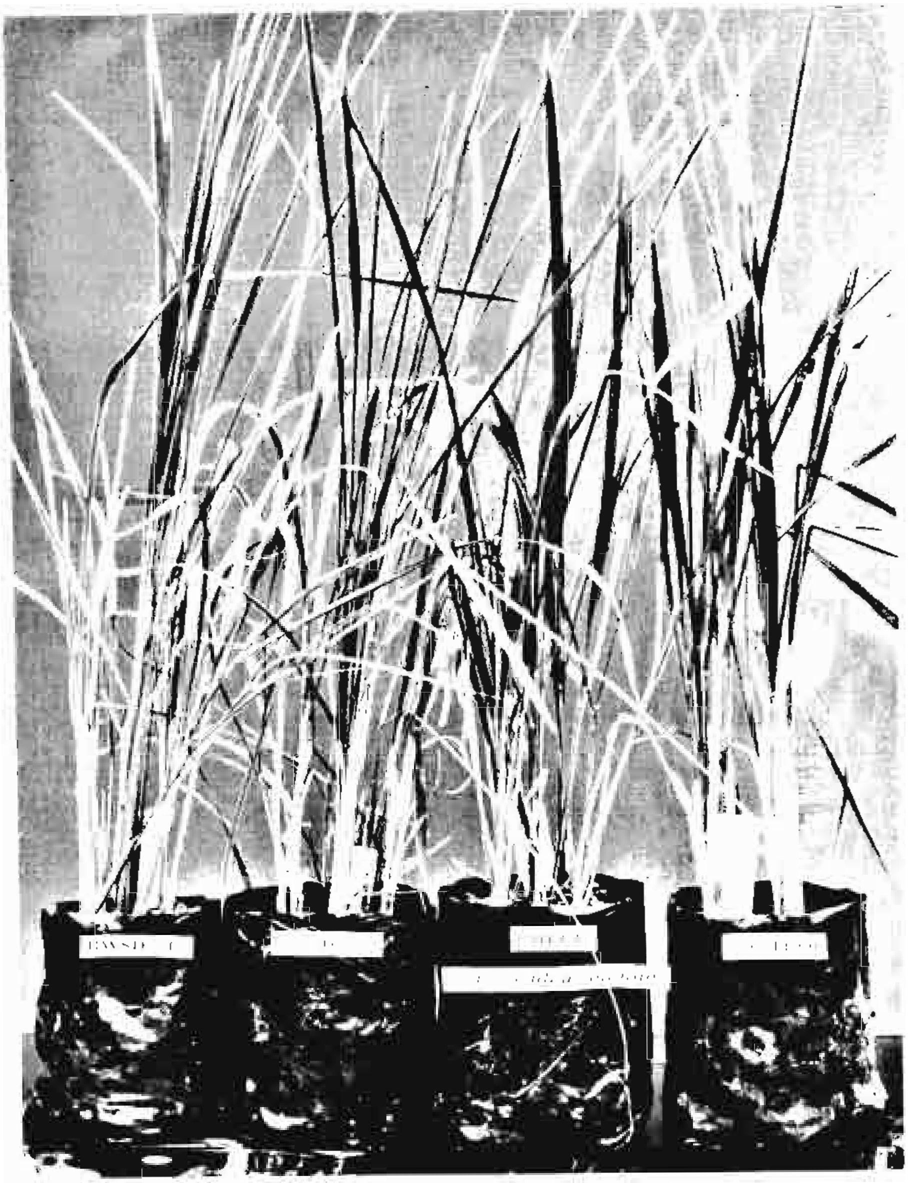


Figure 4.1 Leaf area of *Echinochloa colona* when grown with four selected rice accessions. Data are the mean of four replicates in percentage of non-allelopathic check (BWSD 25). Vertical bars represent the standard errors of the means.



Picture 4.1 Growth of test species *Echinochloa colona* was inhibited when grown with allelopathic rice accession, BWSD 16 or BWSD 19, in the soil.

Table 4.2 Effects of four selected rice accessions on shoot length and dry weight of shoot and root of *Echinochloa crusgalli* L. Beauv. Data are the mean of four replicates and ten seedlings were used in each replicate.

Rice accession	Shoot length (cm)	Shoot DW (mg)	Root DW (mg)	Total DW (mg)
BWSD 16	30.0 a[1]	931 a	198 a	1129 a
BWSD 19	33.5 a	923 a	239 a	1162 a
BWSD 22	36.8 ab	1311 b	278 a	1589 b
BWSD 25	40.1 b	1832 b	513 b	2345 c

[1] Means in column followed by the same letter are not significantly different by Duncan Multiple Range Test at $p< 0.05$

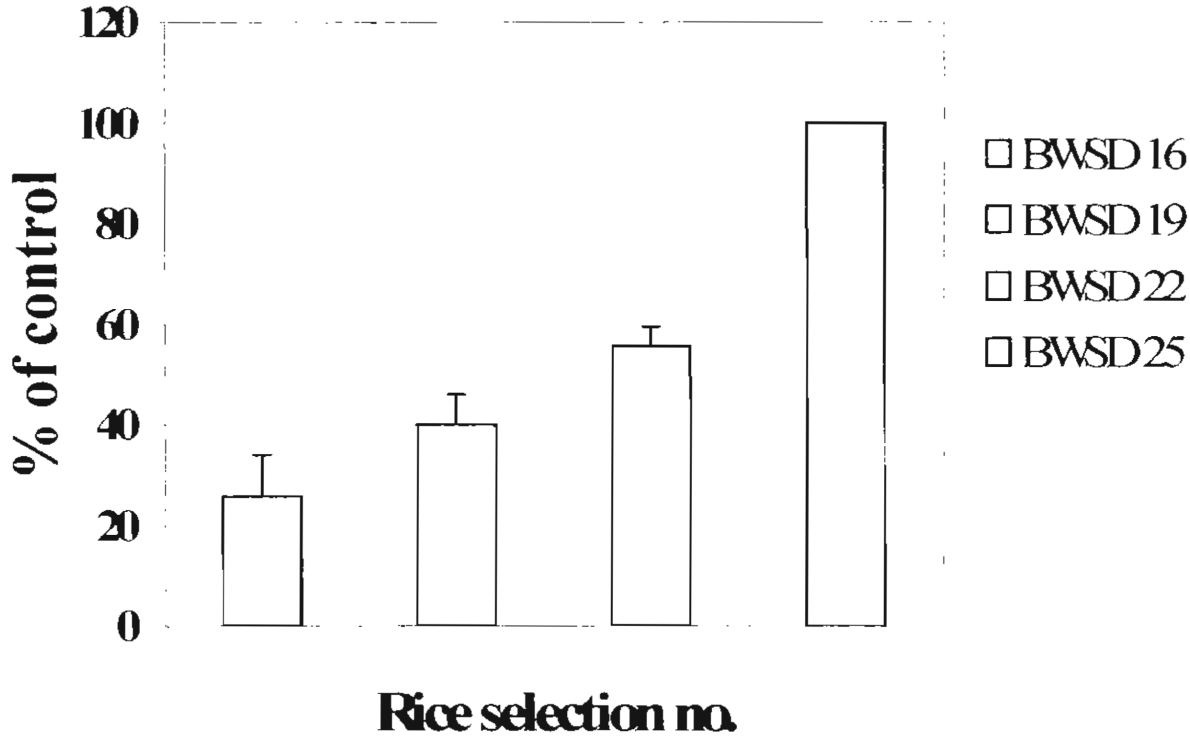


Figure 4.2 Leaf area of *Echinochloa crusgalli* when grown with four selected rice accessions. Data are the mean of four replicates in percentage of non-allelopathic check (BWSD 25). Vertical bars represent the standard errors of the means.

4.2 Effects on dicots species

4.2.1 *Euphorbia geniculata*

Shoot length of *E. geniculata* was moderately reduced when tested with BWSD 16 and was slightly reduced by BWSD 19 and BWSD 22. Similarly, shoot and root growth of *E. geniculata* was markedly reduced when grown together with BWSD 16 and BWSD 19 (Table 4.3). Leaf area of each test plant was reduced when tested with BWSD 16, BWSD 19 and BWSD 22 by 62, 50 and 39%, respectively (Figure 4.3).

4.2.2 *Mimosa pudica*

Shoot length of *M. pudica* was moderately reduced when tested with BWSD 16, BWSD 19 and BWSD 22. Shoot and root growth of *M. pudica* was markedly reduced when grown together with BWSD 16 and BWSD 19. However, BWSD 22 had a little effect on total dry weight of *M. pudica* (Table 4.4). Leaf area of each test plant was reduced when tested with BWSD 16, BWSD 19 and BWSD 22 by 65, 45 and 44%, respectively (Figure 4.4).

4.2.3 *Mimosa invisa*

Shoot length of *M. invisa* was inhibited when tested with BWSD 16 and BWSD 19 and was slightly reduced by BWSD 22. BWSD 19 had a strong allelopathic effect on shoot and root growth of *M. invisa.* In contrast, BWSD 16 and BWSD 22 were slightly allelopthic to test species (Table 4.5). Leaf area of each test plant was reduced more than 60% when tested with BWSD 16, BWSD 19 and BWSD 22 (Figure 4.5). When grown with BWSD 16, *M. pudica* plants became smaller and had less number of leaves with yellowish green (Picture 4.3A). At harvest, root length of test species was observed. When grown with BWSD 25, root length was longer than that with BWSD 19 (Picture 4.3B).

4.2.4 *Lactuca sativa*

For *L. sativa*, the growth was more sensitive to allelopathic rice accessions, BWSD 16, BWSD 19 and BWSD 22 (Table 4.6 and Figure 4.6). BWSD 16 and BWSD 19 depressed the total dry weight of *L. sativa* by three times compared with that in non-allelopathic check BWSD 25 (Table 4.6). Likewise, shoot and root dry weight was inhibited by those accessions. Leaf area of L. sativa was badly affected after grown with allelopathic rice for 30 days. BWSD 16, BWSD 19 and BWSD 22 reduced the leaf area per plant by 75, 83 and 60%, respectively (Figure 4.6).

The current result showed that the inhibitory activity of allelopathic rice depended on weed species (Figure 4.1-4.6). BWSD 16 and BWSD 19 gave the same pattern of test species inhibition as in the laboratory experiment. In contrast, BWSD 22 which was slightly allelopathic to two test species, *E. crusgalli* and *L. sativa*, in the laboratory exhibited the variation of allelopathic potential on different test species. For instances, It was moderately allelopathic to *E. crusgalli*, *Mimosa pudica, M. invisa* and *L. sativa* at different levels. In general, the growth of test species was markedly inhibited when grown with allelopathic accessions. This indicates that allelopathic accessions released allelochemicals into the soil resulting in inhibition of growth. However, the released allelochemicals in each accession might be different in both quantity and types of allelochemicals since trend of inhibition was not the same in all test species. Much of evidence indicates that several chemicals are released together and may exert toxicity in an additive or synergistic manner (Putnam and Tang, 1986). Thongma *et al.*, (1998) investigated the allelopathic activity of Mexican sunflower extract in soil and found that the degree of inhibitory activity was dependent on the test plant species.

To date, some allelopathic compounds were identified in water from allelopathic rice cultivars as 3-hydroxybenzoic acid (3HBA), 4-hydroxybenzoic acid (4HBA), 4-hydroxyhydrocinnamic acid (4HHCA), 3,4-dihydroxyhydrocinnamic acid (3,4DHHCA) and tentatively identified 4-hydroxyphenylacetic acid (4HPAA) (Mattice *et. al.*, 1997 and 1998).

Table 4.3 Effects of four selected rice accessions on shoot length and dry weight of shoot and root of *Euphorbia geniculata*. Data are the mean of four replicates and ten seedlings were used in each replicate.

Rice accession	Shoot length (cm)	Shoot DW (mg)	Root DW (mg)	Total DW (mg)
BWSD 16	10.3 a[1]	273.7 ab	53.1 ab	326.8 a
BWSD 19	12.8 b	245.0 a	43.8 a	288.8 a
BWSD 22	13.6 b	341.8 b	63.4 bc	405.2 ab
BWSD 25	16.1 c	415.8 ab	70.9 c	486.7 b

[1] Means in column followed by the same letter are not significantly different by Duncan Multiple Range Test at $p < 0.05$

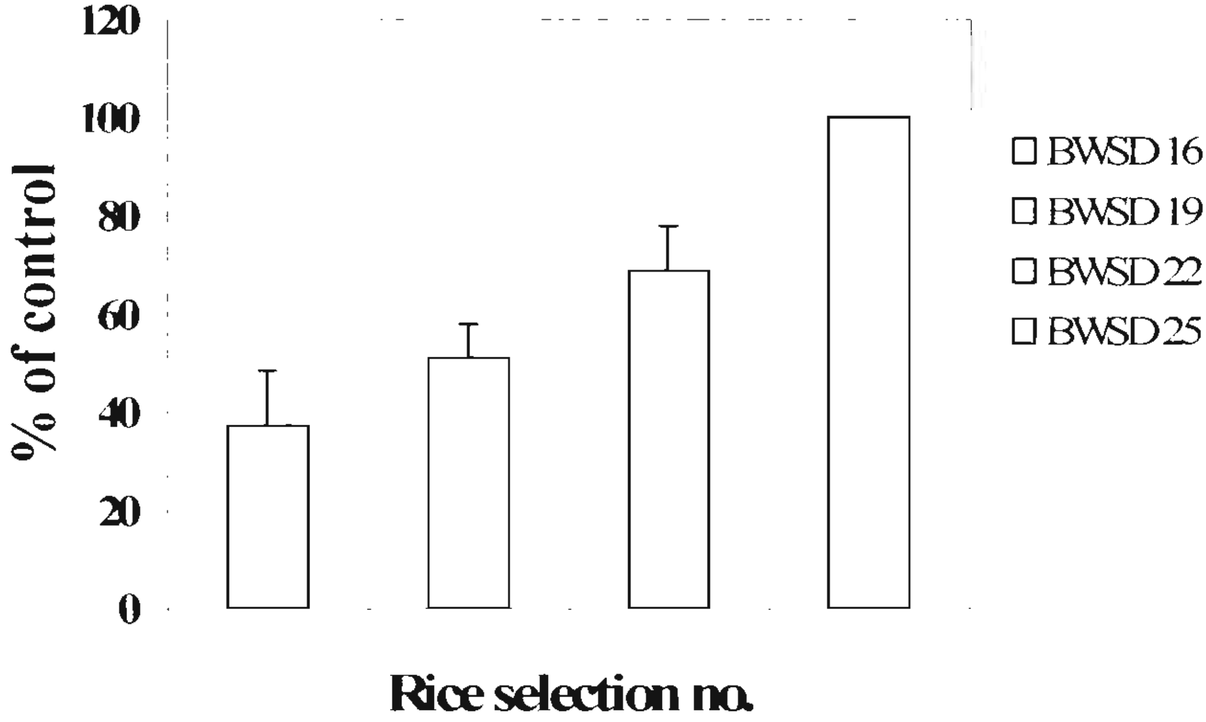


Figure 4.3 Leaf area of *Euphorbia geniculata* when grown with four selected rice accessions. Data are the mean of four replicates in percentage of non-allelopathic check (BWSD 25). Vertical bars represent the standard errors of the means.

Table 4.4 Effects of four selected rice accessions on shoot length and dry weight of shoot and root of *Mimosa pudica* L. Data are the mean of four replicates and ten seedlings were used in each replicate.

Rice accession	Shoot length (cm)	Shoot DW (mg)	Root DW (mg)	Total DW (mg)
BWSD 16	11.0 a[1]	577 a	189 a	766 a
BWSD 19	10.7 a	804 b	190 a	994 ab
BWSD 22	11.1 a	807 b	244 b	1051 b
BWSD 25	13.6 b	823 b	259 b	1082 b

[1] Means in column followed by the same letter are not significantly different by Duncan Multiple Range Test at $p< 0.05$

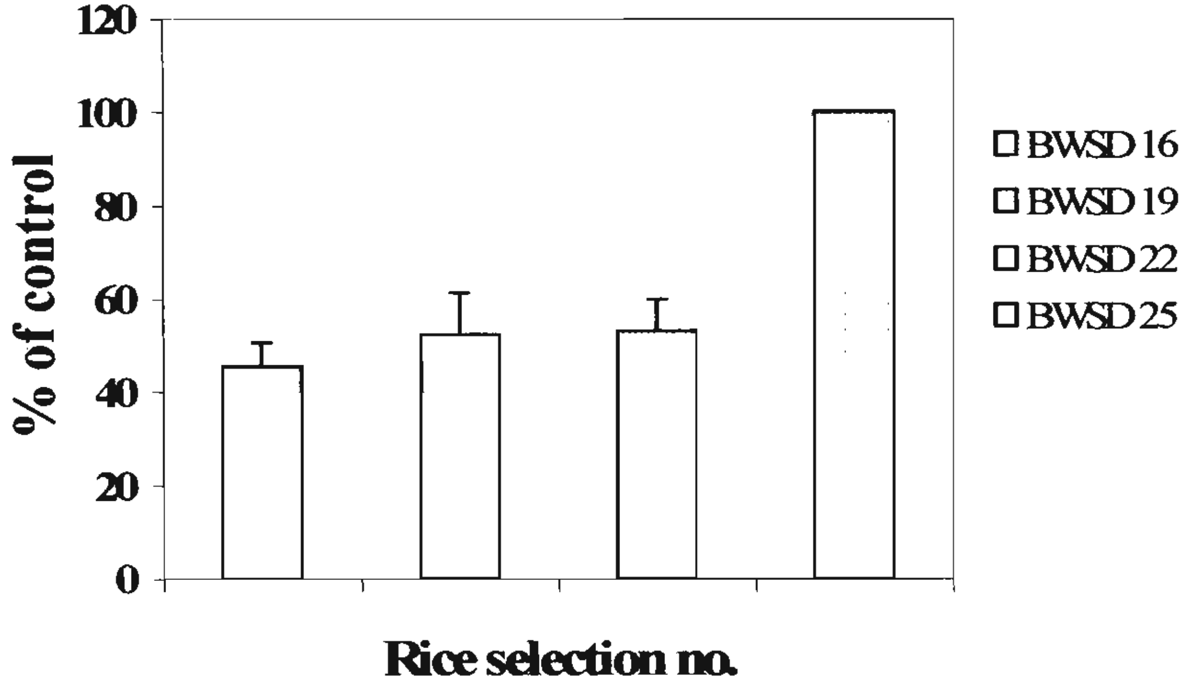


Figure 4.4 Shoot length of *Mimosa pudica* when grown with four selected rice accessions. Data are the mean of four replicates in percentage of non-allelopathic check (BWSD 25). Vertical bars represent the standard errors of the means.

Table 4.5 Effects of four selected rice accessions on shoot length and dry weight of shoot and root of *Mimosa invisa*. Data are the mean of four replicates and ten seedlings were used in each replicate.

Rice accession	**Shoot length (cm)**	**Shoot DW (mg)**	**Root DW (mg)**	**Total DW (mg)**
BWSD 16	7.8 a[1]	29.3 bc	11.3 b	40.6 bc
BWSD 19	7.9 a	18.5 a	7.1 a	25.6 a
BWSD 22	9.5 ab	27.1 b	11.1 b	38.2 ab
BWSD 25	10.5 b	36.0 c	11.4 b	47.4 c

[1] Means in column followed by the same letter are not significantly different by Duncan Multiple Range Test at $p < 0.05$

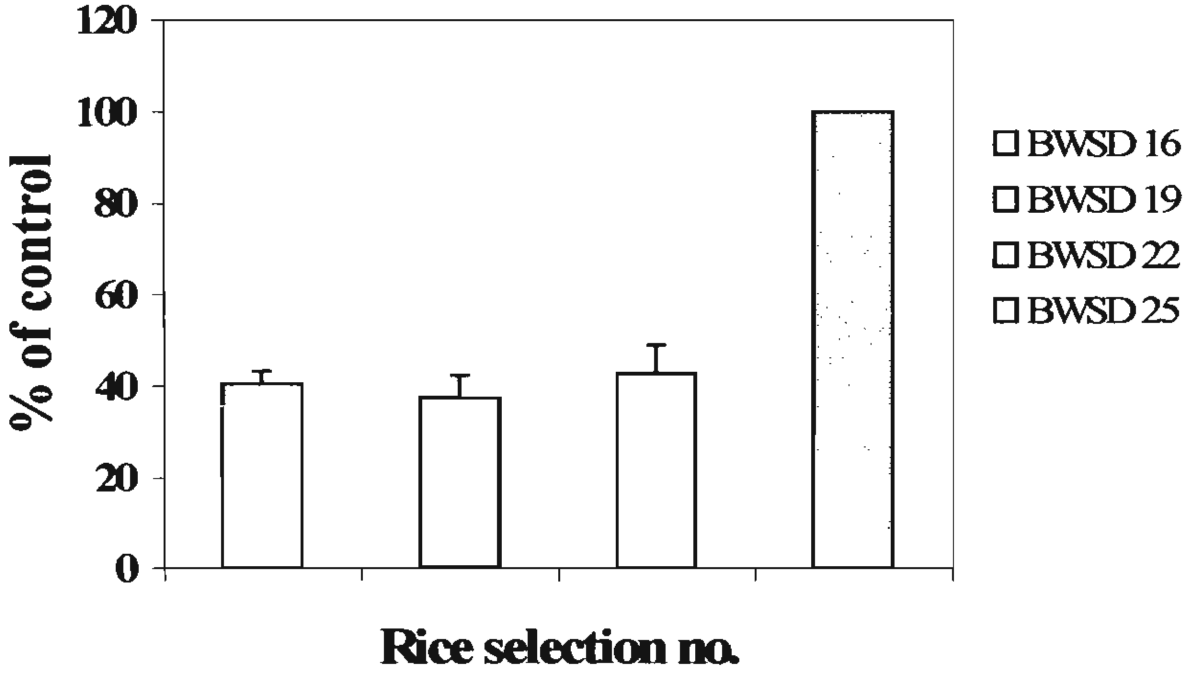
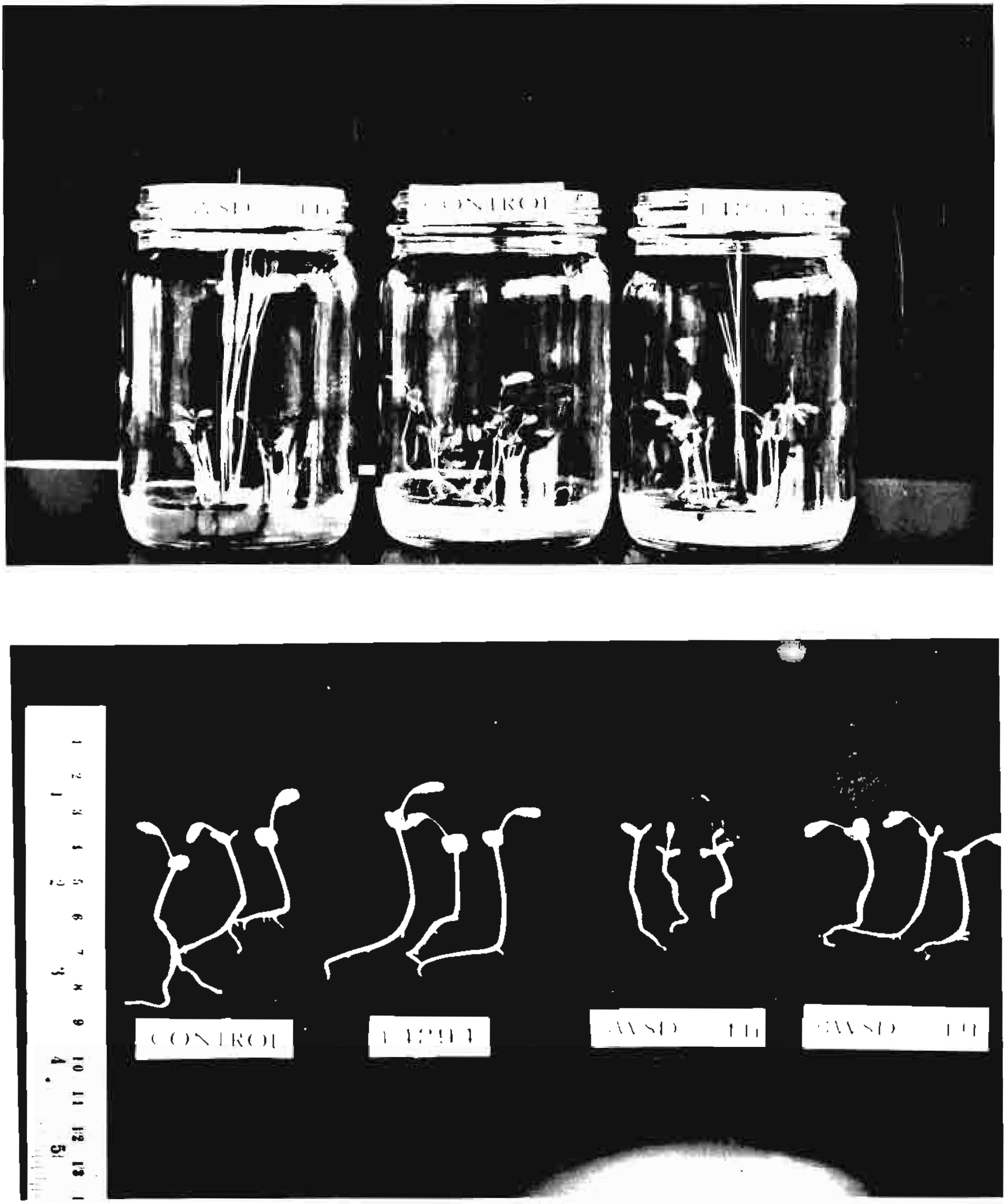


Figure 4.5 Leaf area of *Mimosa invisa* when grown with four selected rice accessions. Data are the mean of four replicates in percentage of non-allelopathic check (BWSD 25). Vertical bars represent the standard errors of the means.



Picture 4.2 Shoot length (A) and root length (B) of *Mimosa invisa* was inhibited when grown with allelopathic rice accession, BWSD 16 or BWSD 19, on 0.5% agar.

Picture 4.3 Growth of test species, *Mimosa invisa*, was suppressed when grown with allelopathic rice BWSD 16 and BWSD 19 (two on the right) in the soil under glasshouse condition.

Table 4.6 Effects of four selected rice accessions on shoot length and dry weight of shoot and root of *Lactuca sativa* L. Data are the mean of four replicates and ten seedlings were used in each replicate.

Rice accession	**Shoot length (cm)**	**Shoot DW (mg)**	**Root DW (mg)**	**Total DW (mg)**
BWSD 16	3.7 a[1]	11.5 a	7.9 ab	19.4 a
BWSD 19	2.8 a	13.3 a	3.3 a	16.6 a
BWSD 22	4.7 b	27.5 b	8.1 ab	35.6 ab
BWSD 25	5.9 c	42.8 c	13.6 b	56.4 b

[1] Means in column followed by the same letter are not significantly different by Duncan Multiple Range Test at $p< 0.05$

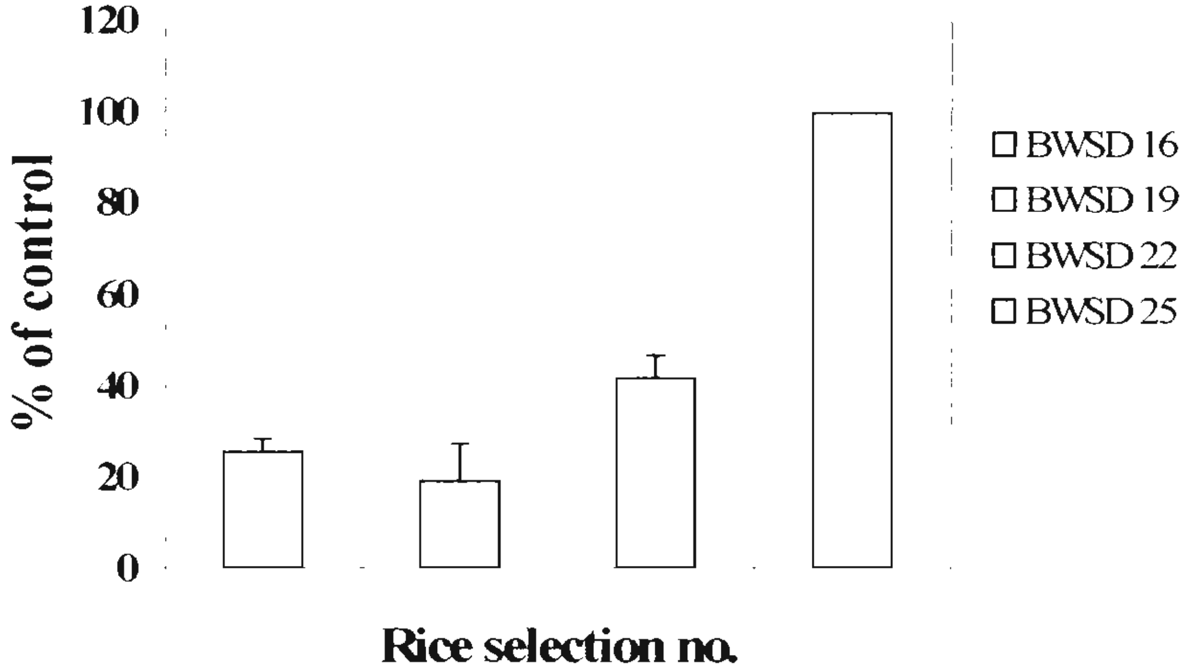


Figure 4.6 Shoot length of *Lactuca sativa* when grown with four selected rice accessions. Data are the mean of four replicates in percentage of non-allelopathic check (BWSD 25). Vertical bars represent the standard errors of the means.

5. ALLELOPATHIC POTENTIAL OF SELECTED UPLAND RICE ACCESSIONS AT DIFFERENT GROWTH STAGES

5.1 At 30 days after sowing

To evaluate whether allelopathic activity depends upon the growth stage of rice, *Mimosa invisa* was used as test species. At various growth stages of rice (0, 5, 10, 15, 20, 25, and 30 days), test plants were sown in the soil. At thirty days after sowing, shoot length of test species was measured. The results showed that at 0 day, no difference in shoot length was found in all four accessions. However, at 5-day, allelopathic accessions significantly reduced shoot length when compared with BWSD 25. After 10-day, the inhibitory effects were similar in all accessions (Table 5.1). Number of leaves was not significantly different among those accessions at all stages of rice. However, at 0-, 5- and 10-day, BWSD 16 and BWSD 19 were likely to reduce the number of *M. invisa* leaves when compared with BWSD 22 and BWSD 25 (Table 5.2).

5.2 At 60 days after sowing

When grown with BWSD 16, BWSD 19 and BWSD 22 at 0 and 5 days after rice germination, shoot length of *M. invisa* was markedly decreased compared with the non-allelopathic check BWSD 25. In contrast, all four accessions (BWSD 16, BWSD 19, BWSD 22 and BWSD 25) equally suppressed shoot length of test species when grown with rice at 10, 15, 20, 25 and 30-day old (Figure 5.1). The results indicate that allelopathic activity was interfered with competition effect when rice plants had larger canopy. Smaller plants perform less efficiently in the shade of the higher (Picture 5.1). Hence shading effects and competition for light and nutrient can obscure the allopathic activity at the beginning stage.

Similarly, total dry weight and number of leaves were inhibited by allopathic rice BWSD 16 and BWSD 19 when grown with 0-day and 5-days old rice (Figures 5.2 and 5.3). When *M. invisa* germinated at 10, 15, 20, 25 and 30 days later than rice, it would be very difficult to separate the allelopathy from competition effects. This may be due to the additive or synergistic effects between allelopathy and competition (Putnam and Tang, 1986). The results of this experiment indicated that BWSD 16, BWSD 19 and BWSD 22 were allelopathic to test species *M. inivisa* at the early stages (0 and 5 days)(Figures 5.2 and 5.3). It would be possible to use these allelopathic accessions as a biological tool to control weeds in upland rice system. However, field experiments have to be conducted to investigate the inhibitory effects of those allelopathic accessions prior to making use of them.

Table 5.1 Shoot length of test species *Mimosa invisa* grown with rice at various growth stages (0, 5, 10, 15, 20, 25 and 30 days). Data are the average of four replicates (first check).

Rice Accession	Shoot length (cm)						
	0*	5	10	15	20	25	30
BWSD 16	4.3a[1/]	4.4a	2.7a	1.5a	3.3a	1.9a	1.2a
BWSD 19	5.1a	5.6a	4.2b	2.7a	3.3a	2.3a	1.9a
BWSD 22	4.3a	5.9a	3.8b	2.5a	3.6a	2.6a	1.9a
BWSD 25	4.1a	7.8b	4.5b	5.3b	5.2b	2.8a	2.2a

* days after rice germination

[1/] Mean in column followed by the same letter are not significantly different by Duncan Multiple Range Test at $p<0.05$

Table 5.2 Number of leaves of test species *Mimosa invisa* grown with rice at various growth stages (0, 5, 10, 15, 20, 25 and 30 days). Data are the average of four replicates at 30 days (first check).

Rice Accession	Number of leaves						
	0*	5	10	15	20	25	30
BWSD 16	5	5	4	4	4	3	3
BWSD 19	6	6	5	5	5	3	3
BWSD 22	5	7	6	4	5	4	3
BWSD 25	6	7	6	5	5	4	3
	NS	NS	NS	NS	NS	NS	NS

* days after rice germination NS = not significantly different