Sains Malaysiana 55(5)(2026): 802-811
http://doi.org/10.17576/jsm-2026-5505-03

Allelopathic Potential of Selected Invasive Plants on Early Seedling Growth of Weedy Rice (*Oryza* spp.)

(Potensi Alelopati Tumbuhan Invasif Terpilih terhadap Pertumbuhan Anak Benih Awal Padi Rumpai (*Oryza* spp.))

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Received: 2 November 2025/Accepted: 22 April 2026

ABSTRACT

Weedy rice is a major constraint in rice cultivation, particularly in direct-seeded systems causing significant yield reduction. Current management practices often lack long-term sustainability and environmental safety. Allelopathy has emerged as a potential eco-friendly strategy for the control of weedy rice. This study aimed to evaluate the allelopathic potential of four invasive plant species namely *Mimosa pigra*, *Dicranopteris linearis*, *Saccharum arundinaceum*, and *Leucaena leucocephala* against the early seedling growth of weedy rice. Dried leaf tissues of the four invasive species were tested using two standard bioassay techniques: dish pack and sandwich methods. Both methods were designed to assess the inhibitory effects of leaf volatiles and water-soluble compounds, respectively. The experiments were conducted under controlled conditions and included multiple weedy rice biotypes. All four invasive species exhibited allelopathic effects, significantly suppressing the shoot and root growth of weedy rice seedlings but limited effect on cultivated rice, MR220CL2. However, the magnitude of inhibition differed among weedy rice biotypes and between bioassay methods suggesting variation in allelochemical potency and modes of action. The tested invasive species possess allelopathic properties that may be explored for developing environmentally sustainable strategies for weedy rice management.

Keywords: Agroecological approach; bioherbicide development; eco-friendly weed management; pre-emergence control; sustainable agriculture

ABSTRAK

Padi angin merupakan salah satu kekangan utama dalam penanaman padi, khususnya dalam sistem tabur terus yang menyebabkan pengurangan hasil yang ketara. Amalan pengurusan semasa sering kali tidak mempunyai kelestarian jangka panjang dan keselamatan alam sekitar. Alelopati telah muncul sebagai satu strategi mesra alam yang berpotensi untuk kawalan padi angin. Penyelidikan ini dijalankan bagi menilai potensi alelopati empat spesies tumbuhan invasif iaitu *Mimosa pigra*, *Dicranopteris linearis*, *Saccharum arundinaceum* dan *Leucaena leucocephala* terhadap pertumbuhan awal anak benih padi angin. Tisu daun yang dikeringkan daripada keempat-empat spesies invasif ini diuji menggunakan dua teknik bioasai piawai iaitu kaedah pek piring dan kaedah sandwic. Kedua-dua kaedah ini direka untuk menilai kesan perencatan oleh sebatian meruap dan sebatian larut air daripada daun. Uji kaji dijalankan di bawah keadaan terkawal dan melibatkan beberapa biotip padi angin. Semua empat spesies invasif menunjukkan kesan alelopati yang ketara dengan merencat pertumbuhan pucuk dan akar anak benih padi angin, namun memberi kesan yang terhad terhadap varieti padi tanaman, MR220CL2. Walau bagaimanapun, tahap perencatan berbeza antara biotip padi angin dan antara kaedah bioasai, menunjukkan adanya variasi dalam keamatan alelokimia dan mekanisme tindakan alelokimia. Spesies invasif yang diuji mempunyai sifat alelopati yang boleh diteroka dalam pembangunan strategi pengurusan padi angin yang lestari dan mesra alam.

Kata kunci: Kawalan pra cambah; pembangunan bioherbisid; pendekatan agroekologi; pengurusan rumpai mesra alam; pertanian lestari

INTRODUCTION

Weedy rice (*Oryza* spp.) represents a formidable challenge in rice agroecosystems worldwide, exhibiting aggressive growth and fierce competition with cultivated rice varieties, leading to reduced crop yields and increased production costs (Baki & Shakirin 2010; Chauhan 2013). Managing weedy rice in infested agricultural fields is arduous due to the striking morphological similarity between weedy and cultivated rice, making it difficult to curb the infestation (Mispan et al. 2019).

The imidazolinone (IMI) herbicide tolerant rice variety has been suggested as an effective herbicide-tolerant rice technology to control weedy rice in Malaysia, especially in direct-seeding systems (Azmi et al. 2012). Although successful in managing weedy rice infestations and enhancing rice production (Sudianto et al. 2013), concerns about sustainability have been raised (Bzour, Zuki & Mispan 2018). These include weedy rice developing resistance to IMI herbicides (Jaafar et al. 2014), hybridization with the herbicide tolerant rice in close proximity (Engku et al. 2016), and residue carryover impacting non-rice crops in rotational systems (Bahm, Barnes & Jensen 2011) and non-IMI rice cultivars (Bzour, Zuki & Mispan 2018).

There is a growing interest to utilize invasive-derived allelochemicals as an alternative, sustainable, and eco-friendly approach to control weed infestations (Farooq et al. 2020; Jabran 2017). Plant allelopathy utilizes natural allelochemical compounds produced by certain plants to inhibit weed growth, offering various application methods such as mulching (Schulz et al. 2013), intercropping (Khamare, Chen & Marble 2022; Rad et al. 2020; Saudy 2015), or crop extracts (Ihsan et al. 2015), thereby reducing reliance on synthetic chemical herbicides.

Invasive plants often succeed in nature because of their capability to compete for resources with native plant communities (Kolar & Lodge 2001; Sun et al. 2018). Allelopathy occurs in a substantial proportion of invasive plant species as one of their biochemical strategies contributing to invasion success (Kalisz, Kivlin & Bialic-Murphy 2021). Allelopathic compounds released by the plants generally can suppress seed germination, inhibit plant growth, reduce nutrient uptake, and interfere with photosynthesis (Brilli, Loreto & Baccelli 2019; Kong et al. 2019; Sothearith et al. 2021).

Allelopathy can be a possible alternative approach to the current weedy rice problem. If the selected invasive species possess allelopathic properties and can suppress the growth of weedy rice seedlings, these plant species can be used to manage the infestation of weedy rice in an eco-friendly manner without jeopardizing the environmental health. Therefore, this study aims to explore the allelopathic potential of selected invasive species as potential candidates for controlling weedy rice by assessing the allelopathic effects of these invasive plants on weedy rice seedling growth.

MATERIALS AND METHODS

INVASIVE PLANT SAMPLES

Saccharum arundinaceum, *Dicranopteris linearis*, *Leucaena leucocephala*, and *Mimosa pigra* - also locally known as *teberau, paku resam, petai belalang* and *semalu gajah*, respectively - were chosen for this study due to their allelochemical toxicity in previous reports (Motmainna et al. 2021) (Table 1). The fresh leaves of these invasive species were collected within the vicinity of Glami Lemi Biotechnology Research Centre, Jelebu, Negeri Sembilan (3.05°N 102.06°E). The plants were carefully washed under running water and air-dried for two weeks. The leaves for each invasive plant were further oven-dried at 70 °C for 3 days to reduce the effect of external contaminants. The dried samples were kept separately in zip lock bags subjected to further experiments.

WEEDY RICE SEED SAMPLES

The seed samples of different weedy rice biotypes were collected from rice fields at eight locations in Selangor, Kedah, Perlis, and Kelantan (Table 2). All collected weedy rice biotypes exhibited straw-coloured hulls, red pericarps, and varying degrees of seed shattering. The seed samples were cleaned, separated from extraneous materials, and air-dried in the greenhouse under normal condition (34/25 °C for day/night) for 3 days. Subsequently, the seeds were placed in the sealed plastic bags with silica gel desiccants and stored in a dry cabinet at room temperature for ~100 days to remove seed dormancy. The local variety MR220CL2 was chosen as a control for this study to compare the effects of allelopathy with the weedy rice. The rice seeds were procured from Loji Benih Padi in Kota Bharu, Kelantan (6°4'37.00513"N 102° 18'1.95424"E). Likewise, these seeds were stored in a dry cabinet at room temperature for later use.

ALLELOPATHIC SCREENING OF SELECTED INVASIVE PLANTS

To evaluate the allelopathic effect of the test plants, the dish pack and sandwich methods (Fujii, 2001) were employed. Lettuce (*Lactuca sativa*) was used as a test plant material in bioassay due to its reliability in germination and its susceptibility to inhibitory and stimulatory chemicals (Fujii et al. 2003).

Volatile allelopathic activity was assessed using a dish-pack setup modified from Fujii et al. (2005). In this method, six-well plates were arranged with 200 mg dried leaf tissues placed in the central well, while the surrounding wells were lined with filter papers moistened with 0.75 mL of distilled water and sown with seven lettuce seeds each. Plates without plant material served as controls. The dishes were sealed with cellophane tape, wrapped in aluminium foil to exclude light, and incubated at 30 °C for 3 days. Radicle and hypocotyl lengths of the germinated seeds were then measured to determine growth inhibition relative to distance from the source well.

Allelopathic effects mediated through aqueous leachates were evaluated using the sandwich method (Fujii 2001). In the sandwich bioassay, the release of water-soluble allelochemicals into the agar matrix occurs through

TABLE 1. List of reported allelochemicals of invasive species used in this research

Species[a]	Allelochemicals	References
Saccharum arundinaceum (Teberau)[b]	α-Linolenic acid; Linoleic acid	Hijano et al. (2021)
	Ferulic acid; Vanillic acid; Syringic acid	Sampietro et al. (2007)
Dicranopteris linearis (Paku resam)	Cinnamtannin B-1	Kato-Noguchi et al. (2024)
	Phenolic compounds	Othman et al. (2020)
Leucaena leucocephala (Petai belalang)	Phenolic acids; Flavonoids; Mimosine	Kato-Noguchi & Kurniadie (2022)
	p-hydroxybenzoic acid; Gallic acid; Vanillic acid; Protocatechuic acid; *p*-hydroxyphenilacetic acid; *p*-hydroxycinnamic acid; Caffeic acid; Ferulic acid; Gallocatechin	Kato-Noguchi & Kato (2025)
	Mimosine	Kato-Noguchi & Kurniadie (2022), Huang et al. (2025)
Mimosa pigra (Semalu gajah)	Flavonoids; Tannins; Phlobatannins; Alkaloids; Saponins	Koodkaew et al. (2018)
	Lupeol; Stigmastane-3,6-dione; Quercetin; Chrysoeriol; Methylgallate; Daucosterol	Khang et al. (2023)

[a]Local Malaysian name for respective invasive plant species is stated in bracket
[b]List of allelochemicals for *S. arundinaceum* were postulated from reported allelochemicals of *S. officinarum*

TABLE 2. Summary of location and phenotypic variations of weedy rice collected for dish pack and sandwich methods assessment against selected invasive species

Weedy rice biotypes	State	Sampling location	Traits			
			Shattering	Hull	Pericarp	Awn
WRM001	Selangor	3°40’54.7”N 100°59’26.7”E	Shattered	Straw	Red	Awnless
WRM002		3°42’58.5”N 101°01’22.7”E				Awned
WRM003	Kedah	5°53’54.9”N 100°21’59.7”E				Awned
WRM004		5°54’31.2”N 100°22’44.9”E				Awnless
WRM005	Perlis	6°32’34.9”N 100°12’59.8”E				Awned
WRM006		6°32’35.6”N 100°12’59.8”E				Awned
WRM007	Kelantan	6°03’55.0”N 102°15’33.0”E				Awned
WRM008		6°03’50.9”N 102°15’18.4”E				Awnless
Cultivar						
MR220CL2	Kelantan	6°04’37.0”N 102°18’1.9”E	Non-shattered	Straw	White	Awnless

passive diffusion from the embedded dried leaf tissues. Although no visual indicator is used to directly confirm leachate movement, the inhibitory responses observed in the test seedlings relative to the agar-only control provide indirect biological evidence of allelochemical diffusion into the surrounding medium. This approach follows the standard methodology described by Fujii (2001), where phytotoxicity is used as a proxy indicator of leachate release. Autoclaved agar (5 mL) was first solidified in each well of a six-well plate under sterile conditions. Dried plant material (10 or 50 mg) was placed on the agar surface and subsequently overlaid with another 5 mL agar layer, embedding the sample between two layers to enable the gradual release of leachates during the bioassay.

A total of five lettuce seeds were placed on top of the second agar medium and then the plate was sealed tightly with parafilm and kept in an incubator at 30 °C. For control, 10 mL of autoclaved agar was added followed by the lettuce seeds on top, with no plant materials added. The seedlings were left to grow in complete darkness.

The weight and length of the radicle and hypocotyl of the lettuce seeds were recorded on day 7. The experiment was conducted in 3 replicates.

ALLELOPATHIC BIOASSAY ON WEEDY RICE

The dish pack and sandwich methods were conducted to evaluate the allelopathic effects of the selected invasive plants on weedy rice seeds. Weedy rice sample seeds substituted lettuce seeds for both methods. Prior to allelopathy bioassays, the seeds were prepared by sterilizing them to eliminate any contaminant from the surface of the seeds that may affect the result. Commercial (Clorox®) 10% sodium hypochlorite was used to sterilize the seeds by soaking the seeds and swirling them for 2 min to remove air bubbles, enabling the sterilizing liquid to have better access to the seeds. Following that, the seeds were rinsed with distilled water to remove the remaining sodium hypochlorite. Finally, the seeds were disposed of onto a Whatman No 1 filter paper to dry them prior to each procedure.

After the bioassay treatments, all the weedy rice seedlings were removed from the media and then gently separated from filter paper particles or any sticking agar. The seedlings were then carefully run through distilled water to remove any remaining agar sticking to them and then were carefully pat dry with tissue paper. The length of radicle and coleoptile was determined by measuring from the base to the tip of radicle/coleoptile. Lettuce hypocotyl corresponds functionally to rice coleoptile. Radicle and coleoptile weights were measured separately using an analytical balance (Mettler Toledo Dragon 204/S Balance Electronic Scale).

The inhibition percentage was calculated according to radicle or hypocotyl/coleoptile length and fresh weight using the following formula:

$$\text{Inhibition percentage (\%)} = 100 - \left(\frac{a-b}{a}\right) \times 100\%$$

where a is the Average growth measurement of the treatment (experimental group) and b is the Average growth measurement of the control group.

A positive inhibition percentage (IP) value indicates a suppressive effect of volatile or leachate compounds from the invasive plants on the target seeds, whereas a negative IP value indicates growth stimulation relative to the control.

STATISTICAL ANALYSIS

The elongation of coleoptile and radicle length, with the fresh weight of the coleoptile and radicle of weedy rice seedlings, were analyzed and compared to the control readings. The mean data of each parameter were compared between the allelopathic effect of selected species using Multivariate Analysis in a complete randomized design setup. Relationships were significant when $P<0.05$. The significant difference between treatments were analysed by using Tukey's method. All statistical analysis was conducted with IBM SPSS Statistics Version 27.0.

RESULTS AND DISCUSSION

PRELIMINARY TEST WITH LETTUCE SEEDS

Preliminary screening of the invasive species towards lettuce seeds in the dish pack and sandwich methods displayed statistically significant differences ($P < 0.001$) in all different groups on the overall set of dependent variables (Supplementary data not shown). These preliminary test results indicated that the leaf parts of *S. arundinaceum, L. leucocephala, M. pigra,* and *D. linearis* could significantly inhibit the growth of the seedlings of lettuce. These findings provide empirical evidence of the relationship between the variables, suggesting that the growth of lettuce seedlings was influenced by potential allelopathic components and has the potential to be used to test on the weedy rice seeds.

VOLATILE EFFECT ON THE SEEDLING GROWTH OF WEEDY RICE

The positive IP values from the dish pack method demonstrated that the leaves of *S. arundinaceum, D. linearis, L. leucocephala,* and *M. pigra* were all capable of inhibiting the radicle and coleoptile of weedy rice seedlings through their volatile allelochemicals (Supplementary data not shown). Various strengths of allelopathic effect were also notable from the different IP values of the leaves of the different invasive species on the radicle of weedy rice and cultivated rice seedlings (Figures 1 & 2).

S. arundinaceum exhibited the strongest inhibitory volatile allelopathic influence on the average radicle length of the eight weedy rice biotypes (16.23±21.59%), followed by *L. leucocephala* (14.62±16.73%), *M. pigra* (12.01±17.46%), and *D. linearis* (7.37±15.64%) (Figure 1(a)-1(d)). Similarly, *S. arundinaceum* also displayed the strongest average inhibition (26.19±24.29%) to radicle weight of weedy rice seed followed by *D. linearis* (23.45±16.86%), *M. pigra* (12.77±26.85%), and *L. leucocephala* (9.25±32.58%) (Figure 1(e)-1(h)). On the contrary, all invasive species on average showed negative IP value indicating the growth-promoting effects from the volatile allelopathy on coleoptile length and weight for all wells in the dish pack method (Figure 2). The IP displayed negative value on average of the coleoptile length for *S. arundinaceum* (-10.08±33.21%), *D. linearis* (-4.84±24.58%), *M. pigra* (-10.48±27.31%), and *L. leucocephala* (-8.08±17.83%) (Figure 2(a)-2(d)). For coleoptile weight, the average IP was recorded for *S. arundinaceum* (-7.60±43.81%), *D. linearis* (-7.30±36.09%), *M. pigra* (-13.36±57.58%), and *L. leucocephala* (-2.41±34.33%) (Figure 2(e)-2(h)).

On the other hand, negative IP values indicate increased of radicle length and weight of rice cultivar MR220CL2 seedlings were promoted or increased by the volatile allelopathy of all tested invasive species (Figure 1). For radicle length, *D. linearis* (-68.21±18.86%) has the highest growth-promoting influence followed by *L. leucocephala* (-44.27±30.28%), *M. pigra* (-27.01±74.72%), and *S. arundinaceum* (-21.32±27.49%). For radicle weight, *M. pigra* (-161.84±55.33%) has the highest growth-promoting influence followed by *D. linearis* (-133.41±66.13%), *S. arundinaceum* (-131.41±92.05%), and *L. leucocephala* (-55.76±35.94%).

Coleoptile length of the cultivated rice (Figure 2) was inhibited by *M. pigra* (19.36±18.96%), *S. arundinaceum* (19.43±28.26%), and *L. leucocephala* (3.16±28.04%), while *D. linearis* (-4.83±27.13%) promoting the increase of length for the coleoptile. All invasive species promoted the growth of coleoptile weight of the cultivated rice with *D. linearis* (-137.86±29.36%) as the highest followed by *L. leucocephala* (-49.20±17.71%), *M. pigra* (-35.66±44.68%), and *S. arundinaceum* (-15.99±41.59%).

LEACHATE EFFECT ON THE SEEDLING GROWTH OF WEEDY RICE

The sandwich method showed that allelochemicals leachate released from the leaves of *S. arundinaceum*, *D. linearis*, *L. leucocephala*, and *M. pigra* could affect the growth of both radicles and coleoptiles in weedy rice seedlings (Supplementary data not shown). The degree of allelopathic impact varied among the invasive species, as indicated by the differing IP observed in the radicle development of both weedy and cultivated rice seedlings (Figure 3).

M. pigra (71.81±16.29%) displayed the highest average of inhibition towards the radicle length of weedy rice biotypes followed by *S. arundinaceum* (62.24±21.90%), *L. leucocephala* (58.34±24.85%), and *D. linearis* (17.17±34.84%) (Figure 3(a)-3(d)). *M. pigra* (53.02±19.91%) also showed the highest inhibition towards radicle weight of weedy rice seedlings followed by *S. arundinaceum* (37.73±37.63%), and *L. leucocephala* (37.16±44.61%) (Figure 3(e)-3(h)). However, *D. linearis* (-60.89±67.01%) exhibited growth-promoting effect on average for the radicle weight of the weedy rice seedlings especially on weedy rice WRM002 and WRM006 (Figure 3(f)).

All invasive species showed inhibiting effect on average towards coleoptile length of the weedy rice seedlings with *M. pigra* (21.27±27.71%) displayed the strongest inhibition to coleoptile length followed by *S. arundinaceum* (19.55±27.68%), *L. leucocephala* (16.79±24.09%), and *D. linearis* (8.62±42.38%) (Figure 3(i)-3(l)). On the other hand, only *M. pigra* (13.59±23.26%) and *S. arundinaceum* (7.60±45.18%) displayed inhibition effect towards coleoptile weight while *D. linearis* (-16.56±10.61%) and *L. leucocephala* (-7.11±41.87%) showed growth-promoting effect towards all weedy rice biotypes (Figure 3(m)-3(p)).

The length and weight of radicle of the cultivated rice MR220CL2 was inhibited by all invasive weeds in the sandwich method while *L. leucocephala* promoting the increase of both the length and weight of the coleoptile and *D. linearis* also promoted the growth of coleoptile length. The rest of the invasive species displayed inhibition towards the coleoptile in this sandwich method.

POTENTIAL APPLICATIONS OF INVASIVE PLANTS TO CONTROL WEEDY RICE

The selected invasive species demonstrated consistent allelopathic suppression of weedy rice seedlings across both volatile and leachate-mediated bioassays, supporting their potential as candidates for eco-friendly weedy rice management. Several application strategies could potentially be used to harness the allelopathic potential of these invasive weeds in weedy rice management including direct application of allelopathic extracts, usage as mulching material, or use as a source of development of bioherbicide (Cheng & Cheng 2015). However, due to their invasive nature, direct intercropping or crop rotation involving these species is not advisable, and their utilization should be restricted to controlled biomass harvesting and extract-based applications.

The use of extracts from the invasive species enables the farmers to have control over the timing of the application of allelochemicals. The perks of that are especially if the allelochemicals were meant for pre-emergence control or early post-emergence control. The allelochemicals listed in Table 1, particularly phenolic acids (ferulic acid, vanillic acid, caffeic acid), flavonoids, tannins, and mimosine, have been widely reported to interfere with seed germination, membrane integrity, enzyme activity, and hormonal balance in target species (Huang et al. 2025; Kato-Noguchi & Kato 2025; Sampietro et al. 2007). In flooded rice systems, the mobility and persistence of water-soluble phenolics may be enhanced due to continuous soil-water interaction, potentially increasing their contact with germinating weedy rice seeds located near the soil surface. However, microbial degradation and dilution in submerged soils may limit persistence and overall efficacy under field conditions. Since weedy rice emergence typically coincides with early crop establishment, application of invasive-derived allelochemical extracts during pre-sowing drainage or early flooding stages may selectively suppress weedy rice while minimizing prolonged soil accumulation. With the precise timing of application, the critical stages of weedy rice development can be targeted by farmers. In addition, the application of *L. leucocephala* under field conditions should also be done after the drainage stage, before sowing the rice seeds, as the water condition may affect the allelochemical concentration (Pudcharaporn 2019). Consequently, suppression of weedy rice may be enhanced while potentially minimizing effects on cultivated rice, although this requires validation under field conditions.

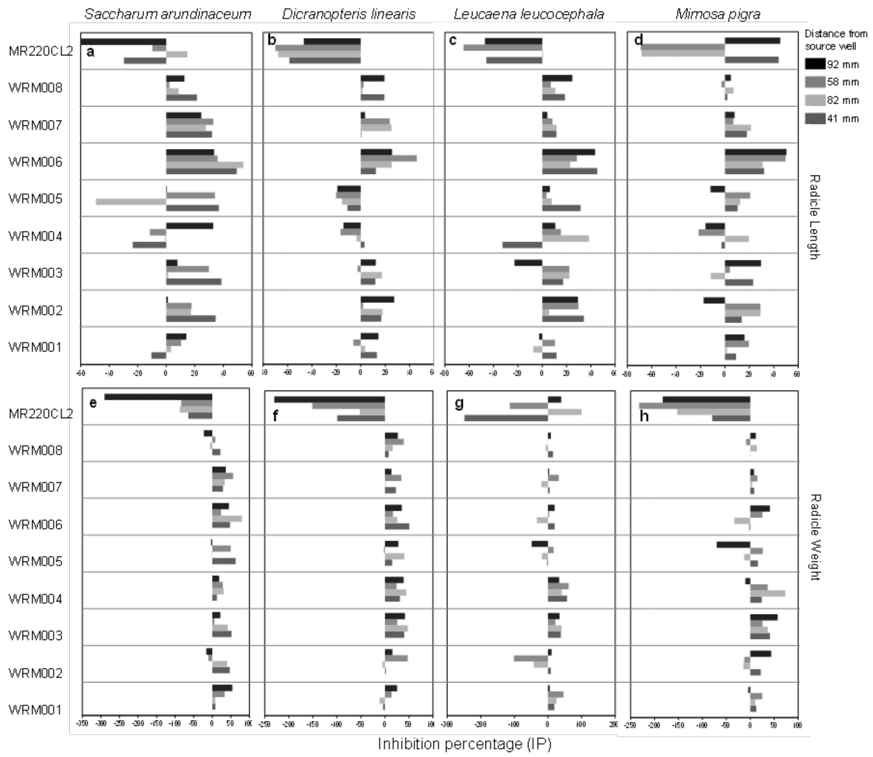

FIGURE 1. Summary of inhibition percentage (IP) by invasive species on the radicle growth of different weedy rice biotypes (WRM) and cultivated rice MR220CL2 at different source well distances in the dish pack method

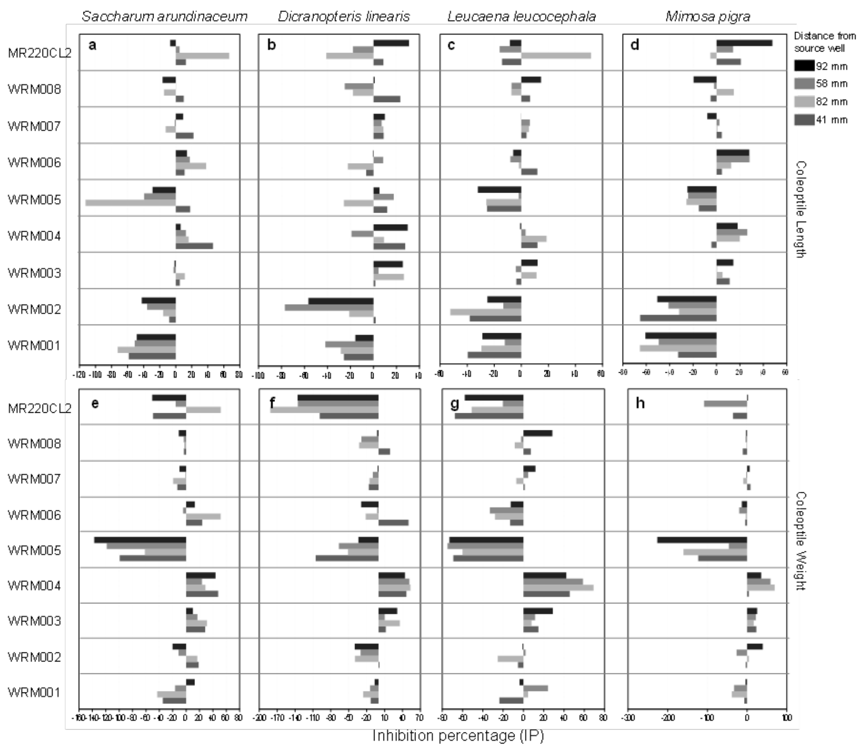

FIGURE 2. Summary of inhibition percentage (IP) by invasive species on the coleoptile growth of different weedy rice biotypes (WRM) and cultivated rice MR220CL2 at different source well distances in the dish pack method

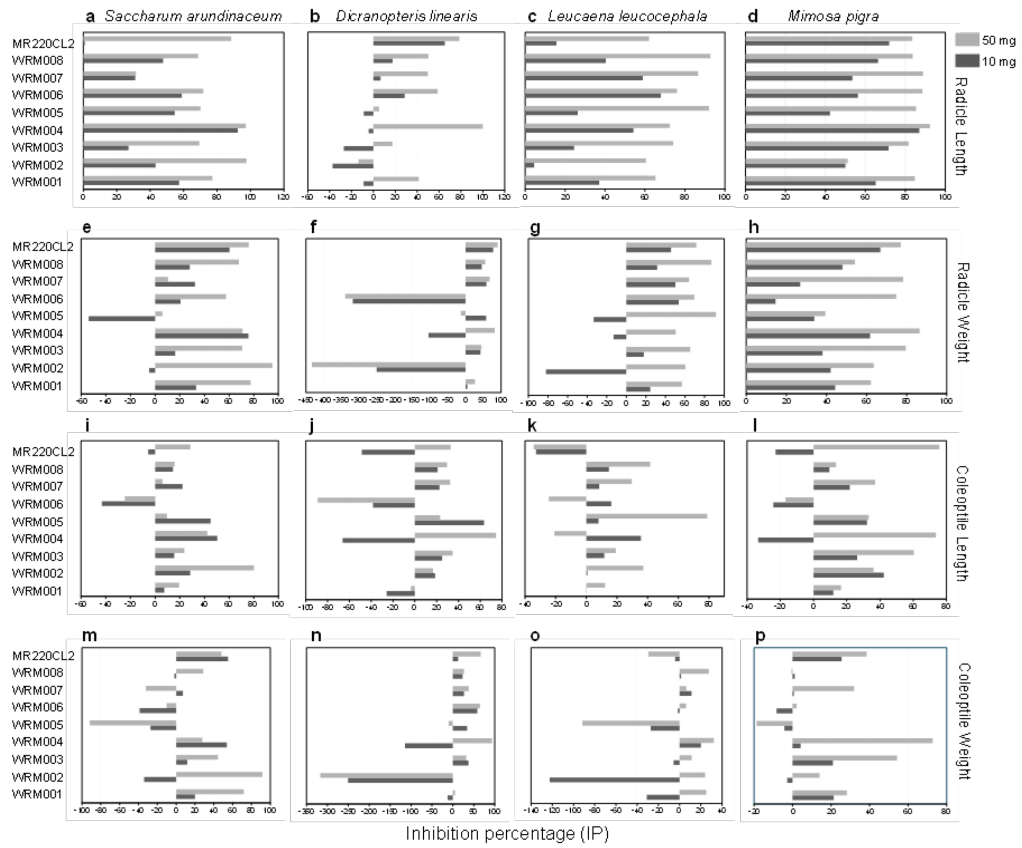

FIGURE 3. Summary of inhibition percentage (IP) by different leaf concentration (10 mg and 50 mg) of invasive species on the radicle (a-h) and coleoptile (i-p) growth of different weedy rice biotypes (WRM) and cultivated rice MR220CL2 used in the sandwich method

To enhance the efficacy of the extracts, a mixture of allelopathic extracts of different species may also be beneficial since the invasive species used in this study have different levels of efficacy on weedy rice. Phenolic acids such as ferulic, vanillic, and caffeic acids are known to disrupt cell division and impair root elongation, which corresponds with the strong radicle suppression recorded in both dish pack and sandwich assays. Mimosine, reported in both *L. leucocephala* and *M. pigra* is a well-documented allelopathic compound capable of interfering with DNA replication and amino acid metabolism (Kato-Noguchi & Kurniadie 2022; Khang et al. 2023; Koodkaew et al. 2018), potentially explaining the pronounced inhibition observed in the sandwich method. The presence of flavonoids and tannins may further contribute synergistically to oxidative stress and membrane destabilization in germinating weedy rice seeds. Selection and combining a desired effect from each of these invasive species could create a synergistic effect and further enhance the effect in the field. The combination of aqueous extracts from multiple allelopathic donors such as sorghum, sunflower, brassica, and mulberry has completely prevented the germination and suppressed seedling development of *Trianthema portulacastrum* L. weed (Mushtaq, Cheema & Khaliq 2010). Moreover, allelopathic effects are usually produced from the combined activity of multiple allelochemicals rather than by a single compound (Jabran et al. 2015).

Different weedy rice accessions in this study showed variability in their response toward the allelopathic influence of the invasive weeds. This may suggest that these weedy rice accessions have different levels of susceptibility towards the different weed species. The different accessions of broad bean (*Vicia faba* L.) also exhibited varying degrees of responsiveness towards the allelopathic influence of *Sorghum halepense* extract where the accessions were differentiated into different groups of high-tolerant, medium-tolerant, low-tolerant, and sensitive accessions (Georgieva 2021). A study by Javaid et al. (2007) also demonstrated the tolerance difference between six rice genotypes in response to the allelopathic influence of purple nutsedge extract (*Cyperus rotundus* L.). Therefore, further investigation and study are needed to find a synergistic extract from *S. arundinaceum, D. linearis, L. leucocephala,* and *M. pigra* from this study to control a wider range of weedy rice populations.

The invasive weed leaves can also be used as an organic mulch material. The use of *D. linearis* leaves as organic mulching material has seen success in controlling weed infestation in pineapple fields (Chong, Nor Aris & Amarudin 2011). Seed germination can be obstructed through mulches that are spread over the soil surface which prevents light and moisture penetration to the seeds (Jordán, Zavala & Gil 2010; Petrikovszki et al. 2020). At the same time, their allelopathic properties will also be released into the soil as the leaves decompose or are leached off by water.

The variability in the responses of the different weedy rice accessions and the stimulatory effects seen in some bioassays emphasized the potential to increase the allelochemical's overall efficacy through a strategic combination of the different species of invasive weeds. The growth-promoting responses observed in certain treatments, particularly under volatile exposure, suggest that sub-lethal concentrations of allelochemicals may induce hormetic effects. Such stimulatory responses could be strategically exploited to induce synchronous germination of dormant weedy rice seeds prior to crop establishment, facilitating mechanical or chemical removal and reducing soil seedbank density. Encouraging pre-season germination of weedy rice seedlings may provide an opportunity for early removal, potentially contributing to seedbank reduction. By selecting and mixing extracts from different species to create the desired effects, farmers can deliberately maximize or minimize stimulatory or inhibitory effects to achieve their intended outcomes.

It is also well noted that all invasive species have limited effect on the cultivated rice MR220CL2. These differential responses between cultivated and weedy rice observed in this study (Figures 1-3) indicate that allelopathic interference is not uniformly phytotoxic and may offer a physiological window for selective suppression. Most treatments have no significant effect on the growth of cultivated rice (Figures 1-3) and in some cases, the invasive species can significantly promote the growth of the cultivar. This result indicates that the application of the invasive weeds to control weedy rice may have minimum effect on the current cultivars which can be neglected. These findings suggest potential selectivity under controlled conditions, indicating that invasive-derived allelochemicals may have limited impact on cultivated rice; however, field validation is required before confirming agronomic safety.

CONCLUSION

This study demonstrated that the selected invasive plant species i.e., *Mimosa pigra*, *Dicranopteris linearis*, *Saccharum arundinaceum*, and *Leucaena leucocephala* exhibit significant allelopathic effects on the early growth of weedy rice seedlings, supporting their potential use as alternative weed management methods. The inhibitory effects were consistent across bioassay methods, although varying in magnitude depending on the invasive species and weedy rice accessions. Importantly, these extracts had minimal or even positive effects on the cultivated rice MR220CL2 suggesting potential selective action under controlled conditions. The findings provide preliminary support for the potential of invasive plants as sources of natural allelochemicals for weedy rice management. The potential application of these species, either as allelopathic extracts or mulching material, provides a promising foundation for developing integrated and sustainable weed management strategies. Further research is recommended to optimize extract combinations and evaluate field-level efficacy across diverse weedy rice populations.

ACKNOWLEDGEMENTS

We would like to express our sincere gratitude to Mr. Mohd Shahmi Hakimi Mazlishah from Glami Lemi Biotechnology Research Center for his invaluable technical support throughout the research. This research was funded by Fundamental Research Grant Scheme from the Ministry of Higher Education, Malaysia, grant number FRGS/1/2019/WAB01/UM/02/4. All supplementary data is available upon requests from the corresponding author.

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