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The Allelopathy of the Invasive Plant Species *Ludwigia decurrens* against Rice and Paddy Weeds

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Abstract: *Ludwigia decurrens* Walter, belonging to the Onagraceae family, is native to southeastern and southcentral USA and has spread quickly, colonizing wetlands in South and Central America, East and South Asia and Africa. *Ludwigia decurrens* also infests rice paddy fields and causes serious damage to rice production. The phenomenon of allelopathy is often observed in invasive plant species and contributes to their invasive characteristics. However, no information has been made available on the allelopathy of *L. decurrens* against wetland species, including *Oryza sativa* (rice), or on its allelochemicals. Aqueous methanol extracts of whole *L. decurrens* plants showed allelopathic activity against *O. sativa* and paddy weeds, *Echinochloa crus-galli* and *Monochoria vaginalis* in an extract-concentration-dependent manner. Two allelochemicals, loliolide and dehydrololiolide, were isolated from the *L. decurrens* extracts through a bioassay-guided separation process using *O. sativa* as the test plant species. Loliolide and dehydrololiolide also suppressed the growth of *O. sativa* and *E. crus-galli* in a concentration-dependent manner. Thus, loliolide and dehydrololiolide may be involved in the allelopathy of *L. decurrens* and may provide a competitive advantage for *L. decurrens* due to their growth inhibitory activity. However, the inhibitory activity of loliolide was about 3-fold greater than that of dehydrololiolide. These investigations suggest the allelopathy of *L. decurrens* may play an important role in the invasion of the species into rice paddy fields and other wetlands. This is the first report on the allelopathy of *L. decurrens* against wetland species and the isolation and identification of the allelochemicals in *L. decurrens*.

Keywords: allelopathy; allelochemical; invasive species; rice; paddy weeds; wetland

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1. Introduction

The characteristics of the life history of invasive plant species, such as their high growth and high reproduction potential, and their phenotypic plasticity are considered to be important for the naturalization of these plants into their introduced ranges [1–4]. The interactions between these invasive plants and their natural enemies, such as pathogens and herbivores in their introduced ranges, are very critical for their naturalization [5–8]. In addition, the interactions between invasive plants and native plants are also one essential factor. Most plants come into conflict with other plants nearby for resources such as light, water and nutrients. Many invasive plant species have high allelopathic activity and contain a large amount of allelochemicals [9–11]. These allelochemicals are biosynthesized and stored in certain plant parts. Some of these allelochemicals are released into the neighboring environments, either by volatilization, root exudation, rainfall leachates, or the decomposition processes of plant residues [12–16]. These released allelochemicals can inhibit the germination, growth, establishment and regeneration process of native plant species, offering them an advantage in the acquisition of resources. Therefore, allelochemicals provide an advantage for invasive plants over native plant species in terms of resource competition in their introduced ranges [17,18].

Ludwigia decurrens Walter (syn. *Jussiaea decurrens* Walter D.C.), belonging to the Onagraceae family, thrives in wetlands such as riverbanks, slow-moving streams, swamps and

rice paddy fields [19,20]. *Ludwigia decurrens* develops rhizomes with secondary aerenchyma well, which allow it to adapt to aquatic environments [21]. *Ludwigia decurrens* is native to southeastern and southcentral USA and was introduced unintentionally and/or accidentally into wetlands in South and Central America, East and South Asia and Africa. The species has spread quickly in its introduced ranges and is considered an invasive plant species [19,22–29].

The species makes a large number of seeds and spreads rapidly using these seeds and stem fragments, which float on water due to their buoyancy [19,30–33]. The *Ludwigia* species was documented to spread around 120 km through plant fragments and seeds on flowing water [34]. The *Ludwigia* species alters the water flow in wetlands and accelerates sedimentation, which induces low-oxygen conditions. This alteration causes significant ecological impacts on fauna and flora [20,25,32]. Many invasive plant species cause significant reductions in biodiversity in their introduced ranges [35–37].

Ludwigia decurrens often infests rice paddy fields and disturbs water management and other cultural practices within rice production. In addition, this species is highly competitive with *Oryza sativa* (rice) because of its fast-growing potential and similar lifestyle to *O. sativa*. *Ludwigia decurrens* suppresses the development of the leaves, tillers and panicles of *O. sativa* plants [38]. It was recorded to reduce the grain yield of *O. sativa* by 30%–80% [29,38,39]. The risk of an environmental impact is ranked as high, as *L. decurrens* infests the water system and disturbs the wetland community, resulting in lower biodiversity, and its economic impact is also high, as the species reduces crop yields and quality [19–29].

Ludwigia species such as *L. adscendens* L., *L. grandiflora* (Michaux), *L. hexapetala* Hook and *L. peploides* (Kunth) Raven have been reported to possess allelopathic activity against wetland plant species, including phytoplankton [40–47]. Quercitrin, prunin and myricitrin have been identified within *L. hexapetala* leaf extracts as allelochemicals [43,44]. The allelochemicals of these *Ludwigia* species may be released into nearby environments, including the rhizosphere, soil and surrounding water, and may contribute to their invasive characteristics [40,42,43,45,46]. It is possible that *L. decurrens* also possesses allelopathic potential and contains allelochemicals, which may contribute to the invasive characteristics of *L. decurrens*. In fact, root exudates of *L. decurrens* suppressed the growth of *Corchorus olitorius* L. and increased its mortality [47]. Unfortunately, *C. olitorius* is not a wetland plant species, and it is important to evaluate whether the allelopathy of *L. decurrens* impacts wetland plant species in order to determine its invasive characteristics. In addition, the allelochemicals of *L. decurrens* have not yet been documented. The objective of our investigation was the determination of the allelopathic activity of *L. decurrens* against wetland plant species and the isolation and identification of the allelochemicals involved in its allelopathy. Therefore, we evaluated the allelopathic activity of *L. decurrens* against *O. sativa* and the paddy weed species *Echinochloa crus-galli* and *Monochoria vaginalis* and isolated and characterized the allelochemicals from *L. decurrens*.

2. Materials and Methods

2.1. Plant Materials

Whole plants of *Ludwigia decurrens* Walter, including the belowground parts, were collected from paddy fields in Miki, Kagawa, Japan (E134°8′13.08″ N34°15′54.02″), in July 2021, which are located on a flattish plain and in a temperate climate. The plant samples were then dried in shade conditions. *Oryza sativa* L. cv. Nipponbare (rice) seeds were provided by University Farm, Faculty of Agriculture, Kagawa University. The seeds of the paddy weed species *Echinochloa crus-galli* (L.) P.Beauv. and *Monochoria vaginalis* (Burm.f) Kunth were collected from the paddy fields in Miki, Kagawa, in September 2021. *E. crus-galli* and *M. vaginalis* were selected as the test plants because these plants are also wetland weed species and are often found together with *L. decurrens* in the same paddy fields. The *E. crus-galli* and *M. vaginalis* seeds were kept in a refrigerator at 5 °C for 3 months to break their dormancy [48,49].

The seeds of *Oryza sativa*, *E. crus-galli* and *M. vaginalis* were sterilized in an aqueous solution of sodium hypochlorite (25 mM) for 10 min and rinsed in distilled water five times. Then, the seeds of *O. sativa* and *E. crus-galli* were germinated on moist filter paper (No 1, Advantec-Toyo Ltd., Tokyo, Japan) under a 12 h light and 12 h dark cycle at 30 °C for 48 h. The seeds of *M. vaginalis* were kept in cold water (5 °C) for 24 h to break their dormancy and germinated on moist filter paper under the same conditions as *O. sativa* and *E. crus-galli* [48].

2.2. Evaluation of the Allelopathic Activity of L. decurrens

The whole plants, including the belowground parts, of *L. decurrens* (100 g dry weight) were cut into small pieces and extracted by soaking them in aqueous methanol (80% *v*/*v*, 500 mL) for 48 h. After filtration using filter paper (No. 2; Advantec-Toyo Ltd.), the resulting residue was soaked again in methanol (500 mL) for 24 h and filtered. Both filtrates were combined and concentrated in vacuo at 40 °C.

The extract of *L. decurrens* was then dissolved in methanol (100 mL), and an aliquot of the methanol solution of the extract (0.001, 0.03, 0.1, 0.3 or 0.1 mL) was applied to filter paper (No. 2) in a 3 cm Petri dish. The methanol solutions of 0.001, 0.03, 0.1, 0.3 and 0.1 mL were equivalent to extracts obtained from 1, 3, 10, 30 and 100 mg dry weight of the *L. decurrens* plants, respectively. Tween 20 solution (1 mL, 0.05% *w*/*v*, polyoxyethylene-sorbitan monolaurate, Nacalai, Kyoto, Japan) was applied onto the filter paper after the methanol in the Petri dishes was evaporated in a fume hood. Therefore, each Petri dishes contains the substances obtained from 1, 3, 10, 30 or 100 mg dry weight of *L. decurrens* in 1 mL solution of Tween 20. These concentrations were selected because the inhibition caused by these concentrations ranged roughly between 0 and 100%.

Ten germinated seeds of *O. sativa*, *E. crus-galli* or *M. vaginalis* were transferred onto the filter paper in the Petri dishes. After 48 h of incubation in darkness at 25 °C, the length of the coleoptiles and roots of these plant species was determined. The percentage length of the coleoptiles and roots of these plant species was calculated against the respective control coleoptile and root length. Control seeds of *O. sativa*, *E. crus-galli* and *M. vaginalis* were grown following exactly the same process with the exception of the *L. decurrens* extracts. The experiment was independently repeated four times with 10 germinated seeds for each determination.

2.3. Isolation of Allelochemicals from L. decurrens

Extraction and purification were followed by the procedure described in the published paper [50]. The whole plants of *L. decurrens* (1 kg dry weight) were cut into small pieces and extracted with aqueous methanol (80% *v*/*v*, 5 L). After filtration, the resulting residue was re-extracted with methanol (5 L), as described in Section 2.2. Then, two filtrates were mixed and evaporated to obtain an aqueous residue. The aqueous residue was regulated to a pH of 7.0 using 1 M phosphate buffer and partitioned against ethyl acetate to separate the aqueous and ethyl acetate fractions. The allelopathic activity of both fractions was determined using the *O. sativa* bioassay, as described in Section 2.2.

After evaporation of the ethyl acetate fraction, the resulting residue was separated by a silica gel column (100 g, silica gel 60, Nacalai) and divided into 9 fractions using a stepwise solvent system with *n*-hexane containing increasing amounts of ethyl acetate (10% per step, *v*/*v*, 200 mL per step) from 20% ethyl acetate. The allelopathic activity of these separated fractions was determined using the *O. sativa* bioassay, and allelopathic activity was detected in fraction 6 (70% ethyl acetate in *n*-hexane).

Active fraction 6 was evaporated to dryness, and the resulting residue was divided into 9 fractions by column chromatography with a Sephadex LH-20 (40 g, Sigma-Aldrich, Burlington, VT, USA) using the stepwise solvent system with water containing increasing amounts of methanol (10% per step, *v*/*v*, 100 mL per step) from 20% methanol. The allelopathic activity was detected in fraction 4 (40% methanol). After evaporation of fraction 4, the resulting residue was divided using reverse-phase Octa Decyl Silyl (ODS) disposal

cartridges (SPE; YMC Ltd., Kyoto, Japan). The cartridge was eluted with water containing increasing amounts of methanol (10% per step, *v*/*v*, 30 mL per step) from 20% methanol. Fraction 4 showed allelopathic activity and was evaporated to dryness. The resulting residue of fraction 4 was finally purified using a reverse-phase HPLC system (column; Inertsil ODS-3, 4.6 mm i.d. x 250 mm, GL Science Inc., Kyoto, Japan) eluted at a flow rate of 0.8 mL per min with 20% (*v*/*v*) aqueous methanol at 220 nm detection. Two active compounds, **1** and **2**, were isolated from the fractions eluted between 38 and 40 min and 56 and 58 min, respectively (Figure 1), and their chemical structures were determined using spectrum data of its HRESIMS, ^{1}H-NMR ($CDCl_3$, 400 MHz), and optical rotation.

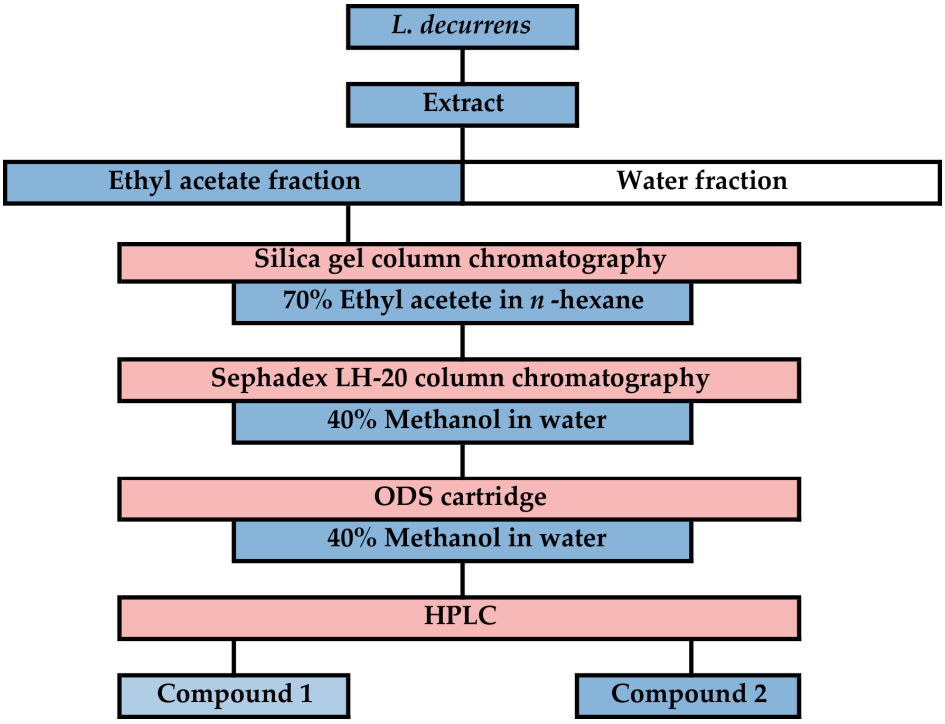

Figure 1. Separation process of compounds **1** and **2** from whole plants of *L. decurrens*.

2.4. Allelopathic Activity of the Isolated Compounds

The allelopathic activity of compounds **1** and **2** isolated from *L. decurrens* was determined using the bioassay of *O. sativa* and *E. crus-galli*, as described in Section 2.2. The concentrations of loliolide and dehydrololioide in the bioassay solution were 0.003, 0.01 0.03, 0.1, 0.3 and 1 mM. The bioassay was repeated four times with 10 germinated seeds for each bioassay.

2.5. Statistics

All the tested samples were subjected to bioassays with four independent replications using a randomized design. Significant differences in the allelopathic activity of the tested samples were examined by multi-comparison using a one-way ANOVA (SPSS, version 16.0) and post hoc analysis with Tukey's HSD test at the $p < 0.05$ level. The concentration causing 50% growth inhibition (IC_{50} value) was determined using GraphPad Prism 6.0 based on the respective bioassay data.

3. Results

3.1. Allelopathic Activity of L. decurrens

The *L. decurrens* extracts significantly suppressed the coleoptile and root growth of *O. sativa*, *E. crus-galli* and *M. vaginalis* at concentrations $\geq$ 10 mg equivalent extract per mL at the $p < 0.05$ level (Figure 2). The IC_{50} values for the coleoptile growth were 9.8 mg, 34.5 mg and 13.4 mg equivalent of extract per mL for *O. sativa*, *E. crus-galli* and *M. vaginalis*,

respectively, and the IC_{50} values of the extracts for the root growth were 9.1 mg, 31.5 mg and 11.7 mg equivalent extract per mL, respectively.

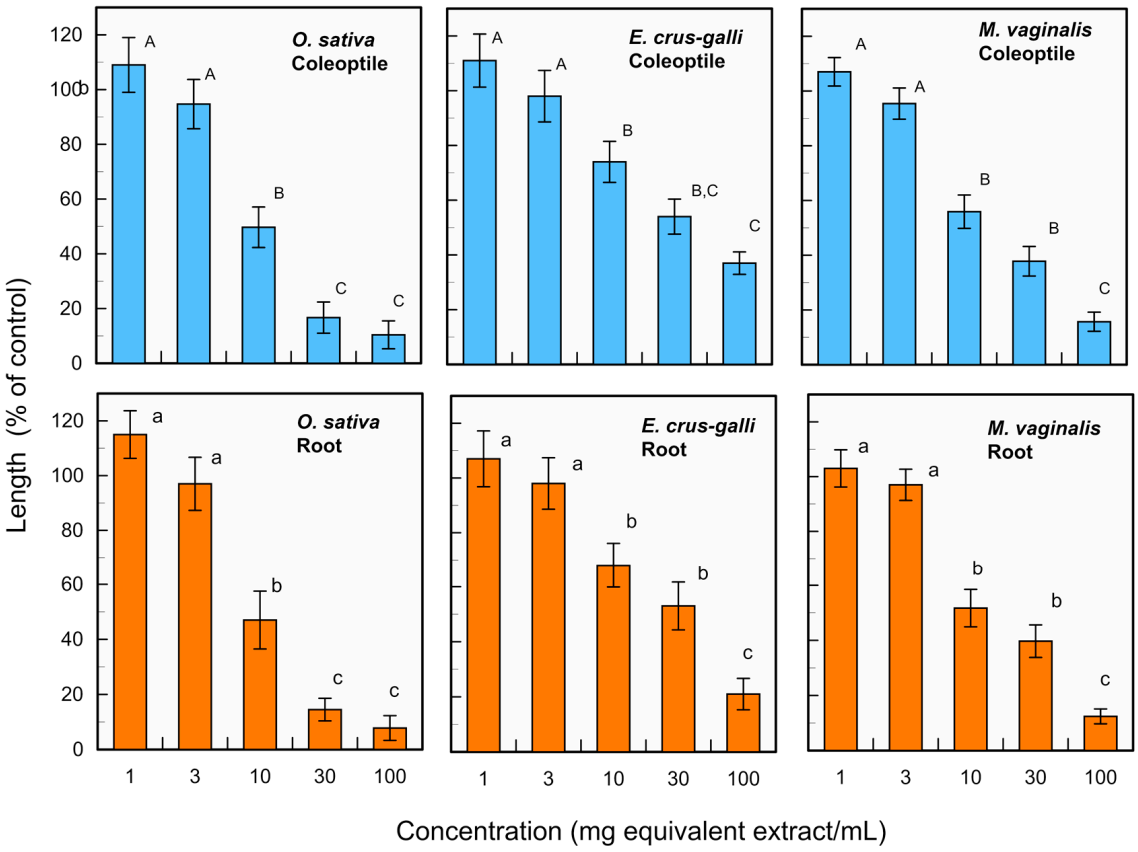

Figure 2. Effects of the extracts of *L. decurrens* on the coleoptile and root growth of *O. sativa*, *E. crus-galli* and *M. vaginalis*. The concentrations on the X-axis indicate the substances obtained from 1, 3, 10, 30 and 100 mg dry weight of *L. decurrens* in 1 mL of the assay solution. The coleoptile and root length of these test plant species was compared with the respective coleoptile and root length of the control plants, and the percentage length compared to the controls was determined. Means ± SE were calculated from 4 independent experiments with 10 seedlings for each determination. Different letters on bars in the same panels indicate significant differences at the $p < 0.05$ level.

3.2. Isolation and Identification of Allelochemicals in L. decurrens

The *L. decurrens* extract was divided into aqueous and ethyl acetate fractions by the partition process (Figure 1). The ethyl acetate fraction had higher allelopathic activity than that of the aqueous fraction. Thus, the ethyl acetate fraction was separated using silica gel column chromatography, and the allelopathic activity of all the separated fractions was determined using *O. sativa* as a test plant species. The allelopathic activity was detected in fraction 6 at the $p < 0.05$ level (Figure 3). Fraction 6 was further separated using Sephadex LH-20 and ODS cartridges. Two compounds, **1** and **2,** with allelopathic activity were finally isolated using HPLC (Figure 1).

The molecular formula of compound **1** was found to be $C_{11}H_{14}O_3$ by HRESIMS analysis. The ^{1}H NMR spectrum as measured in $CDCl_3$ showed the presence of three methyl proton signals at δ_H 1.59 (3H, s), 1.43 (3H, s) and 1.30 (3H, s); an olefinic proton signal at δ_H 5.93 (1H, s); and four methylene proton signals at δ_H 2.97 (1H, d, J = 13.8, 1.9), 2.67 (1H, d, J = 13.8), 2.48 (1H, dd, J = 14.2, 1.9) and 2.42 (1H, d, J = 14.2). The optical rotation was $[\alpha]_D^{24}$ = +136.3 (c 0.07, CH_2Cl_2). Based on comparison of its spectrum data with the published spectrum data [51], compound **1** was determined to be dehydrololiolide (Figure 4)

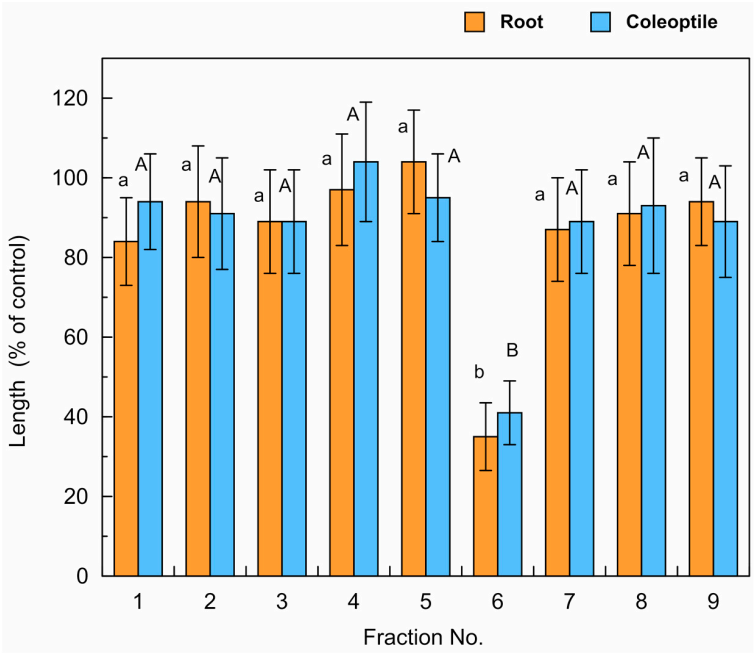

Figure 3. Effects of the fractions separated by silica gel column chromatography on root and coleoptile growth of *O. sativa*. Concentration of tested samples corresponded to the extract obtained from 30 mg of *L. decurrens* per mL. The other conditions were the same as described in Figure 2.

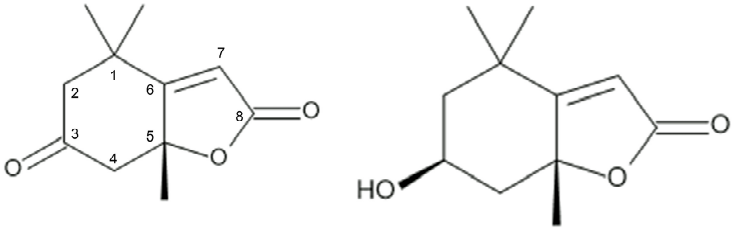

Figure 4. Chemical structures of dehydrololiolide (compound **1**) and loliolide (compound **2**).

The molecular formula of compound **2** was found to be $C_{11}H_{16}O_3$ by HRESIMS analysis. The ^{1}H NMR spectrum as measured in $CDCl_3$ showed the presence of three methyl proton signals at δ_H 1.76 (3H, s), 1.47 (3H, s) and 1.28 (3H, s); an olefinic proton signal at δ_H 5.75 (1H, s); one methine proton signal at δ_H 4.22 (1H, m); and four methylene proton signals at δ_H 2.42 (1H, dt, J = 13.8, 2.7), 1.99 (1H, dt, J = 14.4, 2.7), 1.75 (1H, dd, J = 13.8, 4.1) and 1.53 (1H, dd, J = 14.4, 3.7). Its optical rotation was $[\alpha]_D^{24} = -96.6$ (c 0.25, MeOH). Based on comparison of its spectrum data with the published spectrum data [52,53], compound **2** was determined to be loliolide (Figure 4).

3.3. Allelopathic Activity of the Isolated Compounds

Loliolide showed growth suppression (at the $p < 0.05$ level) of *O. sativa*'s coleoptiles and roots at concentrations $\geq$ 0.01 mM and of *E. crus-galli*'s coleoptiles and roots at concentrations $\geq$ 0.03 mM (Figure 5). The IC_{50} values of loliolide for *O. sativa*'s coleoptile and root growth were 0.025 mM and 0.022 mM, respectively, and the IC_{50} values for *E. crus-galli*'s coleoptile and root growth were 0.081 mM and 0.064 mM, respectively.

Dehydrololiolide showed growth suppression ($p < 0.05$ level) of the coleoptiles and roots of *O. sativa* and *E. crus-galli* at concentrations $\geq$ 0.03 mM (Figure 6). The IC_{50} values of dehydrololiolide for *O. sativa*'s coleoptile and root growth were 0.097 mM and 0.083 mM, respectively, and the IC_{50} values for *E. crus-galli*'s coleoptile and root growth were 0.297 mM and 0.255 mM, respectively.

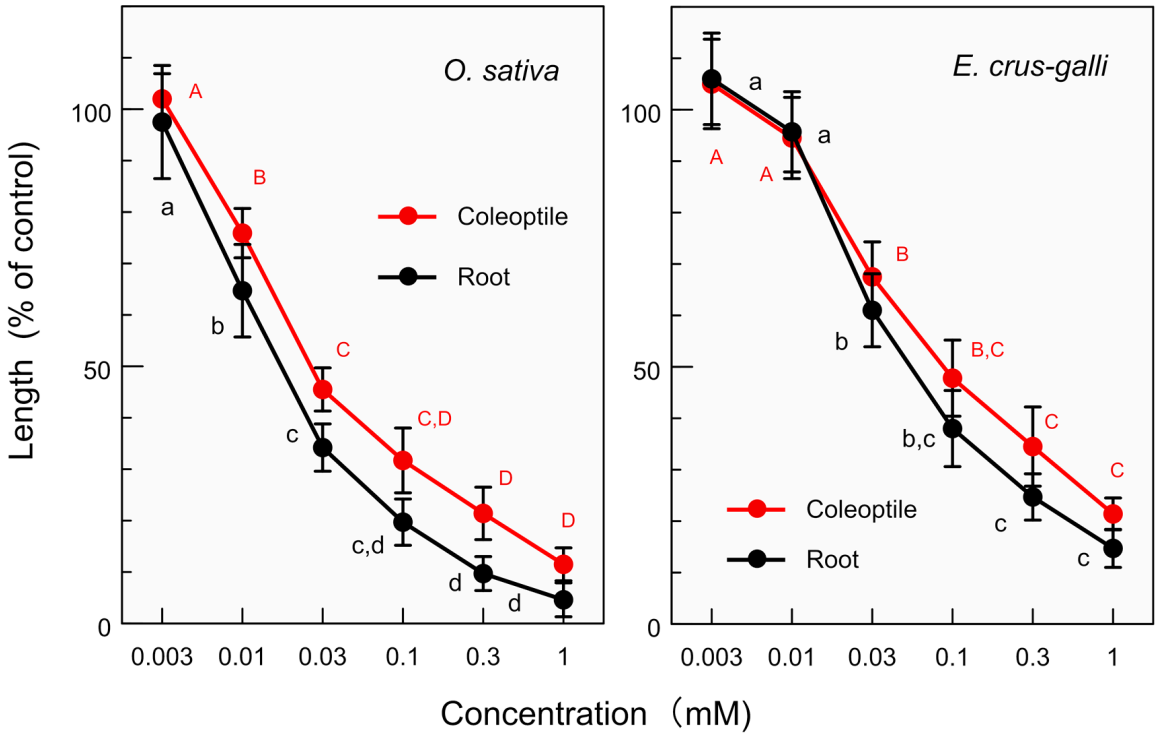

Figure 5. Effects of loliolide on the coleoptile and root growth of *O. sativa* and *E. crus-galli*. The coleoptile and root length of these test plant species was compared with the respective coleoptile and root length of the control plants, and the percentage length against the controls was determined. Means ± SE were calculated from 4 independent experiments with 10 germinated seeds for each determination. Different letters in the same panels indicate significant differences at the $p < 0.05$ level.

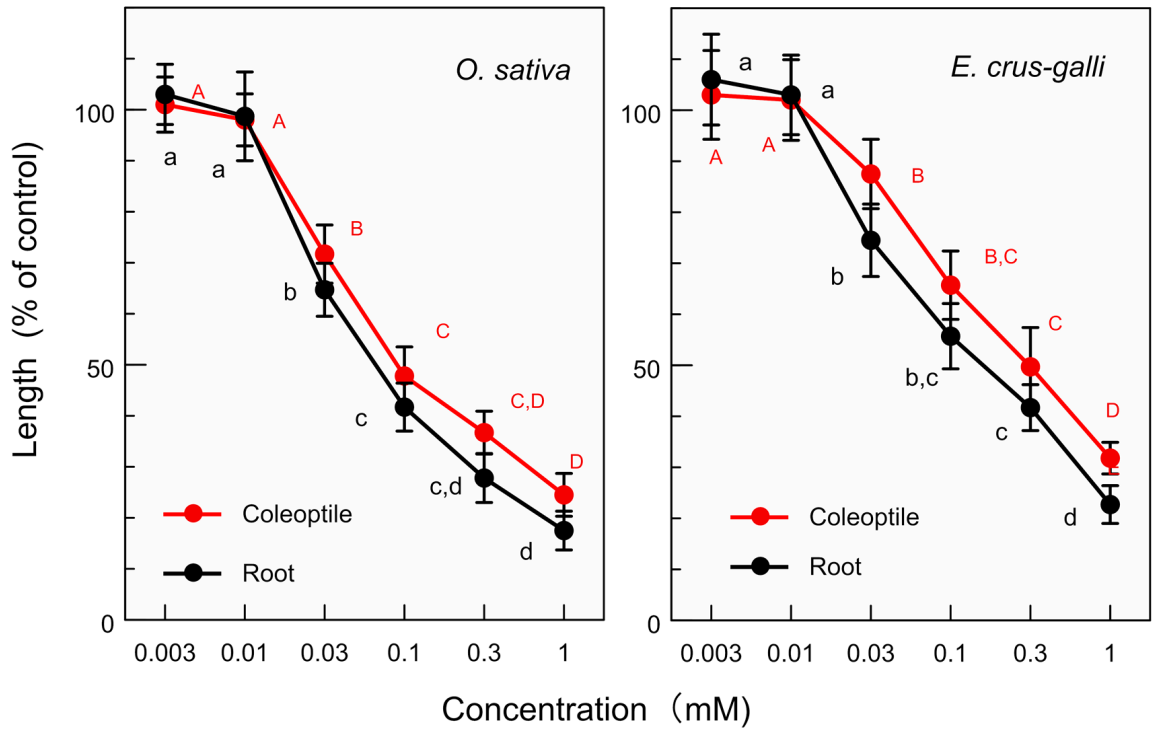

Figure 6. Effects of dehydrololiolide on the coleoptile and root growth of *O. sativa* and *E. crus-galli*. The other conditions were the same as those described in Figure 5.

4. Discussion

The extracts of whole plants of *L. decurrens* suppressed the coleoptile and root growth of *O. sativa* and the paddy weeds *E. crus-galli* and *M. vaginalis* in an extract-concentration-dependent manner (Figure 2). Comparing their IC_{50} values, the sensitivity of the *O. sativa* roots to the extracts was the highest, and that of the *E. crus-galli* coleoptiles was the lowest among the coleoptiles and roots of these test plant species. The average IC_{50} value for the coleoptiles and roots of each test plant species was 9.5 mg, 32.9 mg and 12.6 mg equivalent extract per mL for *O. sativa*, *E. crus-galli* and *M. vaginalis*, respectively. Thus, *O. sativa* was the most sensitive and *E. crus-galli* was the least sensitive to the extract among the three species. *O. sativa* and *M. vaginalis* showed a similar sensitivity to the extracts, but the sensitivity of *O. sativa* was 3-fold greater than that of *E. crus-galli.* Considering these results, the invasive plant *L. decurrens* may be allelopathic and contain some allelochemicals. These allelochemicals are extractable with a mixture of methanol and water. In addition, the effectiveness of these allelochemicals differed in the test plant species and their organs (coleoptiles and roots). Allelochemicals are synthesized, stored in certain plant tissues and released into the neighboring environment [12–18]. These released allelochemicals can suppress the germination and growth of plants nearby and may contribute to the invasive characteristics of *L. decurrens.* Many invasive plant species have been reported to have allelopathic activity [9–11]. Their extracts, plant parts (leaves, stems and roots) infiltrating the soil, root exudates and essential oil suppress the germination and growth of several other plant species, including native plant species in their introduced ranges [16,17]. Many secondary substances have also been identified in their extracts, residues, root exudates and essential oil. Some of these secondary substances were reported to act as allelochemicals [17,18].

Loliolide and dehydrololiolide were isolated from aqueous methanol extracts of *L. decurrens* as allelochemicals through the bioassay-guided separation process (Figure 1). Loliolide was isolated for the first time from *Lolium perenne* [54] and has been isolated from several other plant species [55], including two alga species, *Undaria pinnatifida* [56] and *Gracilaria lemaneiformis* [56], and a freshwater plant species, water hyacinth (*Eichhornia crassipes*) [57]. Water hyacinth is one of the most invasive plant species and has spread quickly in lakes, rivers and rice paddy fields in frost-free regions across the world [58,59]. Although loliolide has been isolated from these alga species and water hyacinth, its allelopathic activity was previously reported only against terrestrial plant species, such as *Lepidum sativum* and *Lolium multiflorum* [57,60]. The effectiveness of loliolide against wetland plant species has not yet been reported. Therefore, this is the first report demonstrating the effectiveness of loliolide against wetland species.

Dehydrololiolide was isolated for the first time from *Nicotiana tabacum* [61] and has been isolated from several other plant species, such as *Nephrolepis exaltata*, *Elaeocarpus floribundus* and *Fissistigma retusum* [62–64]. Although dehydrololiolide has been reported to have several biological activities, such as anti-neuroinflammatory activity [65], antioxidant activity, anti-inflammatory activity [66] and anticholinesterase activity [67], the allelopathic activity of dehydrololiolide has only been documented against a terrestrial plant, *Lepidum sativum* [64]. This is also the first report describing the effectiveness of dehydrololiolide against wetland plant species.

Loliolide and dehydrololiolide suppressed the growth of the wetland plant species *O. sativa* and *E. crus-galli's* coleoptiles and roots in a concentration-dependent manner (Figures 5 and 6). *O. sativa* and *E. crus-galli* were chosen as the test plant species because *O. sativa* was the most sensitive to the *L. decurrens* extract and *E. crus-galli* was the least sensitive (Figure 2). Comparing their IC_{50} values, the effectiveness of loliolide against *O. sativa*'s coleoptiles and roots was 3.2- and 2.9-fold greater than that against *E. crus-galli*'s coleoptiles and roots, respectively, and the effectiveness of dehydrololiolide against *O. sativa*'s coleoptiles and roots was 3.1- and 2.7-fold greater than that against *E. crus-galli*'s coleoptiles and roots, respectively. These values for the effectiveness of loliolide and dehydrololiolide against *O. sativa* and *E. crus-galli* are consistent with the values for the effectiveness of

the *L. decurrens* extracts against *O. sativa* and *E. crus-galli* as described above. In addition, *L. decurrens* may contain similar proportions of loliolide and dehydrololiolide based on HPLC. However, comparing the IC_{50} values, the inhibitory activity of loliolide was 3.9- and 3.8-fold greater than that of dehydrololiolide against *O. sativa*'s coleoptiles and roots, respectively, and 3.7- and 3.5-fold greater than that of dehydrololiolide against *E. crus-galli*'s coleoptiles and roots. Thus, a hydroxy group at the C-3 position in loliolide may increase its activity (Figure 4). Neither compound caused any apparent morphological alterations during the 2-day incubation compared to the respective control plants. However, the effectiveness of these compounds against the germination of these plant species should be evaluated in the future.

As an invasive plant species, *L. decurrens* has spread quickly in rice paddy fields and caused serious problems for the growth and grain yield of *O. sativa* [29,38,39]. *Ludwigia decurrens* can occupy more than 50% of the plant community in its introduced ranges and is recognized as one of the most aggressive weed species [20,25,32]. Aqueous extracts of *L. decurrens* showed allelopathic activity against *O. sativa* and two paddy weeds, *E. crus-galli* and *M. vaginalis* (Figure 2). Two allelochemicals, loliolide and dehydrololiolide, were isolated from the extracts through the bioassay-guided separation process using an *O. sativa* bioassay (Figures 1, 3 and 4). Loliolide and dehydrololiolide also suppressed the growth of *O. sativa* and *E. crus-galli* (Figures 5 and 6), suggesting that both compounds may be involved in the allelopathy of *L. decurrens.* After *L. decurrens* withers as an annual plant species, its plant residues, including the leaves, roots and rhizomes, accumulate in the underwater soil in wetland conditions [20,43]. Some allelochemicals of *L. decurrens* may be released into the soil during the residue decomposition process. Some other allelochemicals may be released into the surrounding water from *L. decurrens* plant bodies [12–17]. These allelochemicals may provide a competitive advantage in the acquisition of resources for *L. decurrens* over native plant species, including *O. sativa,* which may contribute to increasing the growth and population of *L. decurrens*. However, it is essential to clarify the release process and quantity of these allelochemicals in the wetland environment, including rice paddy fields, for an understanding of the involvement of *L. decurrens*' allelopathy in its invasiveness.

5. Conclusions

The invasive plant species *L. decurrens* often infests rice paddy fields and causes serious damage to rice production. Extracts of *L. decurrens* were allelopathic, and the extracts significantly suppressed the growth of *O. sativa* and the paddy weeds *E. crus-galli* and *M. vaginalis.* Two allelochemicals, loliolide and dehydrololiolide, were isolated from the extracts of *L. decurrens*. Loliolide and dehydrololiolide suppressed the growth of *O. sativa* and *E. crus-galli* in a concentration-dependent manner. Therefore, these allelochemicals may be involved in the allelopathy of *L. decurrens* and may provide a competitive advantage for resource acquisition. The allelopathic properties of *L. decurrens* may contribute to this species' invasiveness.

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