

石菖蒲根茎挥发油对稗、鳢肠和水稻的萌发与生长的影响^{*}

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摘 要: 在实验室和温室条件下, 用从石菖蒲根茎中提取的挥发油经减压蒸馏获得的组分四, 对稗草、鳢肠和水稻的萌发与生长进行了生物学效应测试。结果表明, 在低浓度时促进供试植物的萌发和生长, 而在高浓度时则抑制萌发和生长。实验室培养皿条件下, 50~ 400 mg/L 浓度抑制作用产生并逐渐增加, 400 mg/L 以上达到 90% 或更高。但是, 温室盆栽条件下, 3 000 mg/L 时才出现抑制作用, 在 1 000 mg/L 以下均表现促进作用。

关键词: 生物活性; 提取物; 石菖蒲; 稗; 鳢肠; 水稻

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Effect of Essential Oil Extracted from *Acorus tatarinowii* Rhizomes on Germination and Growth of Barnyardgrass, Eclipta and Rice (*Oryza sativa*)

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Abstract: The laboratory and glasshouse experiments were conducted to evaluate the effect of fraction 4 on seed germination and seedling growth of barnyardgrass (*Echinochloa crusgalli* (L.) Beauv.), eclipta (*Eclipta prostrata* (L.) L.) and rice (*Oryza sativa* L.), which was separated from the essential oil of rhizomes of grass-leaved sweetflag (*Acorus tatarinowii*) that was dephlegmated at low pressure. A common phenomenon was observed in both laboratory and greenhouse trials, that is, seed germination and seedling growth was inhibited at higher concentration but slightly increased at 50 mg/L or lower concentration. Under laboratory conditions, inhibition was observed at concentration from 50 ~ 400 mg/L and more than 90% inhibition with > 400 mg/L. However, under glasshouse conditions, inhibition occurred at 3 000 mg/L or higher while increase of the growth was observed at 1 000 mg/L or lower concentration.

Key words: Biological activity; Extract; *Acorus tatarinowii* Schott; *Echinochloa crusgalli* (L.) Beauv.; *Eclipta prostrata* (L.) L.; *Oryza sativa* L.

The extract of grass-leaved sweetflag (*Acorus tatarinowii*) has been thoroughly investigated for repelling against post-harvest insects and for fungi-

cide during storage of grains^[1,2]. If it could inhibit germination of seeds, the repellent may have an additional function in addition to prolonging sto-

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rage duration of grains. The effect of inhibition to germination of seeds and growth of plants may be applied to weed control. However, allelopathy of extract from *Acorus tatarinowii* to higher plants has not been reported so far although a simple study was conducted on inhibition of lettuce seed germination by sesquiterpenoids isolated from *Acorus calamus*^[3] and the extract or some components of *Acorus tatarinowii* and *A. calamus* had some phytotoxicity to some aquatic algae^[4,5]. The purpose of this study was to determine its biological activity of the extract to two main paddy weeds in China and rice.

1 Materials and Methods

1.1 Preparation of extract

The rhizomes of *Acorus tatarinowii* were collected in the hilly regions of Wuhu City, PR China and air-dried under the sun. The dried rhizomes were broken into small segments, 10 kg of which were distilled in a 50 L distilling still for 10 h one time. As a result, the mixture of essential oil and water were obtained. Essential oil was extracted from the mixture using the solvent ethyl ether. After the solvent was evaporated, essential oil was further dried with Na_2SO_4 . Finally, light-yellow oil extract was obtained with the follow s

The index of physical characters of essential oil was given below:

$$D^{21} = 1.009, n_D^{24} = 1.551, [\alpha]^{21} = 1.1076.$$

Essential oil was dephlegmated through a dephlegmator at low pressure. Four fractions were obtained under the following conditions (Table 1).

Table 1 The conditions under which four fractions were obtained through a dephlegmator

Fraction	Temperature ($^{\circ}\text{C}$)	Pressure (mmHg)	Diptr (20°C)
1	66- 123	5	1.5346
2	123- 128	5	1.5570
3	128- 131	5	1.5599
4	131- 137	5	1.5600

It was believed that fraction four contained the most abundant asarone so only this fraction was used for bioassay^[6].

1.2 Preparation of solution of the extract

Circular dextrin was selected and used as an emulsifier to dissolve the extract. Two mother solution with 5 000 mg/L and 1 000 mg/L was prepared as mother solution by mixing 5 mL extract with 2.5 g circular dextrin, 0.5 mL extract with 0.5 g circular dextrin and then added sterile distilled water to 500 mL. Then, the mother solution was diluted to 3 000 mg/L, 500 mg/L, 400 mg/L, 300 mg/L, 200 mg/L, 100 mg/L and 50 mg/L concentration separately.

As a control, the emulsifier circular dextrin solution was prepared with equal amount of the emulsifier in 50 mg/L, 100 mg/L, 200 mg/L, 300 mg/L, 400 mg/L, 500 mg/L, 1 000 mg/L, 3 000 mg/L and 5 000 mg/L.

1.3 Bioassay

The seeds of barnyardgrass (*Echinochloa crus-galli*), eclipta (*Eclipta prostrata*) and rice (*Oryza sativa*) were sterilized in 0.1% HgCl_2 for 15 min and then washing three times with distilled water. Twenty weed seeds for each species and ten rice seeds were counted and put on a piece of sterilized filter paper in a 9-cm-diameter petri dish. 10 mL of extract solution at each of concentrations of 50 mg/L, 100 mg/L, 200 mg/L, 300 mg/L, 400 mg/L and 500 mg/L solution groups in every three petri dishes group were added into the petri dish. Three replicates were used. Two sets of control were used, i.e. sterilized distilled water and circular dextrin. The circular dextrin control included six different concentrations in 50 mg/L, 100 mg/L, 200 mg/L, 300 mg/L, 400 mg/L and 500 mg/L.

All of treatments were placed in an illuminated incubator with a 14 h light ($1\,000\ \mu\text{mol E}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$)/10 h dark regime at 25°C . After 2 d incubation, germination of both weed and rice seeds was observed. After the plants were further incubated for additional 7 d, the length of the seedlings and number of the roots were measured and recorded.

A 13 cm-layer of paddy soil was put into twenty eight 45-cm-diameter pots with 15 cm depth in a glasshouse. 100 seeds of each weed

species and 50 rice seeds were sowed at the surface of soil. After 24 h, 200 mg/L, 300 mg/L, 400 mg/L, 500 mg/L, 1 000 mg/L, 3 000 mg/L, 5 000 mg/L extract solution and emulsifier were sprayed with a hand-sprayer. Dry weight of the seedlings was measured after 18 d of treatment.

ANOVA in Duncan's multiple comparison was employed to compare the difference between the treatments with Statistic Software SPSS 10.0 and Excel 97.

The experiments were conducted twice.

2 Results and Discussion

At the concentration of 50 mg/L, the extract promoted germination of the seeds of rice, barnyardgrass and eclipta. There were two peaks occurred at 50 mg/L and 300 mg/L in the germination curve of rice and eclipta respectively. The steep change occurred two times in the curves of all of three tested species between 50 mg/L and 100 mg/L and between 300 and 400 mg/L (Fig. 1). However, barnyardgrass only had one peak at 50 mg/L and as concentration of extract increased to greater than 50 mg/L, germination of the seeds was inhibited. There was a flat curve for barnyardgrass between 100 mg/L and 300 mg/L (Fig. 1). Extract solution at 500 mg/L reduced the rate of germination down to about 50% for all tested plants.

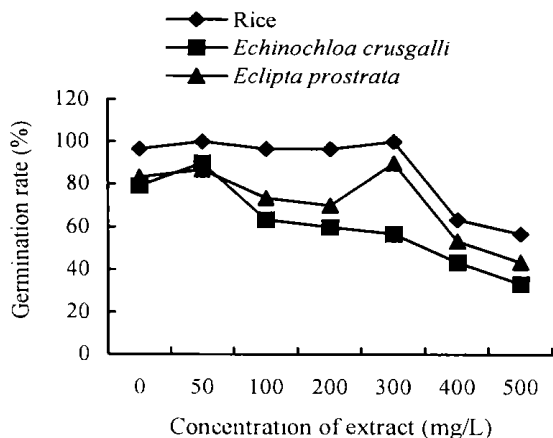


Fig. 1 The effect of extract on germination rate of the seeds of tested weeds and rice

However, there were differences in biological activity on germination of three species of seeds

Between 100 mg/L and 300 mg/L, the germination rate of rice seeds was not markedly influenced by the extract, while that of eclipta got high point at 300 mg/L.

Since the essential oil extracted from roots of *Acorus tatarinowii* can be used as repellent against stored-product insects, the biological activity of inhibition of seed germination could be beneficial to reduce consumption of grains during storing.

The inhibition of seedling growth for all three species increased with increasing concentration. When concentration was 500 mg/L growth was completely inhibited. The steep curves occurred between 50 mg/L and 100 mg/L. The small peak appeared at 50 mg/L in rice (Fig. 2). The extract retarded the root growth much more than stem and leaf growth (Fig. 2, Fig. 3 and Fig. 4). At 100 mg/L or more, the root growth was seriously inhibited ($P < 0.01$), and when the concentration

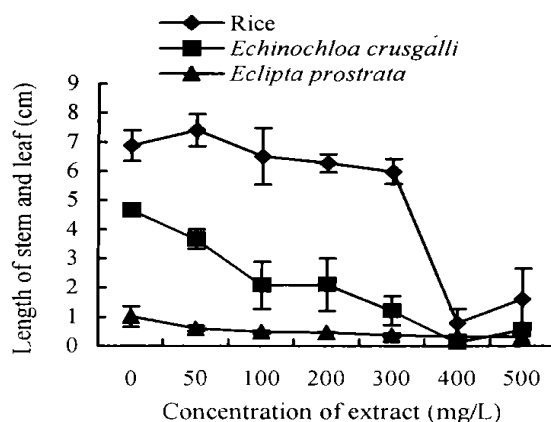


Fig. 2 The effect of extract on the growth of stem and leaf of tested weeds and rice

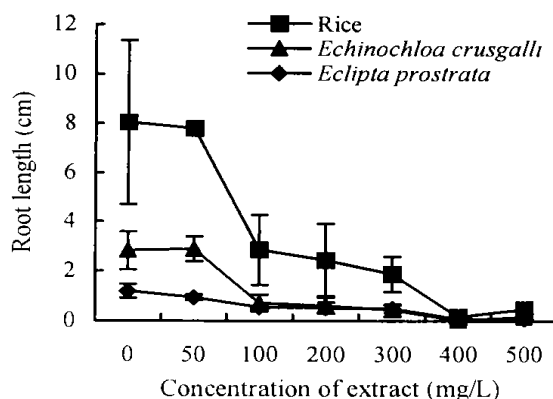


Fig. 3 The effect of extract on the growth of roots of tested weeds and rice

increased to 400 mg/L, the root growth nearly stopped (Fig 3). Therefore, the result showed that the inhibition of extract to root growth was more effective than leaf and stem growth, by which extract should be studied further to determine potential of biologically based herbicide to control weeds in paddy rice fields

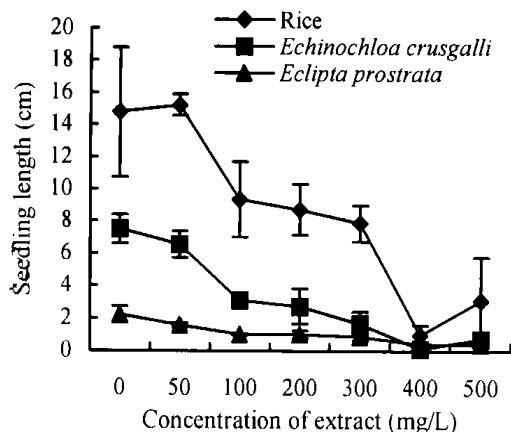


Fig. 4 The effect of extract on the growth of seedlings of tested weeds and rice

The number of roots was not significantly influenced at concentration of less than 400 mg/L ($P > 0.05$) (Fig 5). At 400 mg/L, the number of roots reduced to the lowest percentage. However, there were differences in bioactivity between those three species of plants since there were marked differences between rice and barnyard grass at concentration of between 400 mg/L or higher and 300 mg/L or lower for ($P < 0.01$). Although the germination rate of rice and barnyard grass seeds

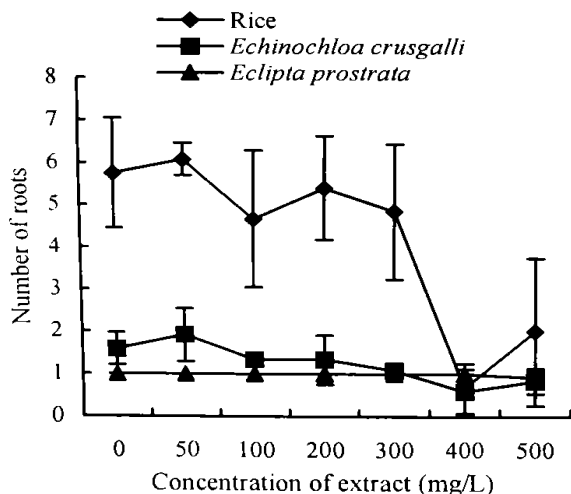


Fig. 5 The effect of extract on number of roots of tested weeds and rice

under the treatment of 300 mg/L extract were higher than at control, most of germinated seeds only had bud sheath out of seeds (Fig 5). It appears that bud sheath could not be inhibited by the extract. The elongation of bud sheath could be even promoted at 300 mg/L or higher concentration.

The growth of tested plants was promoted at lower than 3000 mg/L (Fig 6). At 3000 mg/L or higher, the growth of plants was inhibited. The inhibition effect of the extract on the growth of barnyardgrass and eclipta was greater than that on rice.

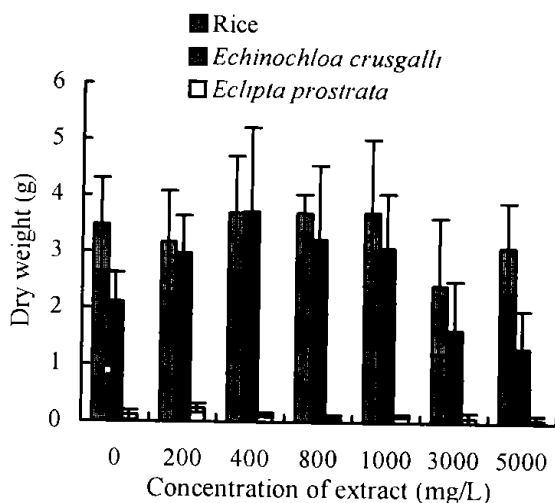


Fig. 6 The effect of extract on growth of the weeds and rice in a glasshouse

The inhibition effect of extract on two weed species was more than that on rice in the glasshouse experiment so that the extract could be used as a biological based herbicide for control of paddy weeds. In addition, promotive effect of the growth at low dosage of extract could be used as a plant growth regulator.

There were the marked differences in the effective dosage between the laboratory and glasshouse experiments since the former closing experimental system protected biological constituents from volatilizing while the latter opening one allowed some of them to volatilize.

Because asarones were major constituents of fraction four dephlegmated from essential oil, allelopathic effect of the extract on higher plants

may be due to asarones

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