四种苔藓植物提取液对二种蓼科植物种子萌发和幼苗生长的影响

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摘 要:研究卵叶泥炭藓(*Sphagnum ovatum*),卷叶凤尾藓(*Fissidens cristatus*),弯叶灰藓(*Hypnum callichroum*)和大金发藓(*Polytrichum commune*)的水粗提取液对维管植物虎杖和皱叶酸模种子萌发和幼苗生长的影响。结果显示:四种苔藓植物的粗提取液对两种植物的种子萌发率并没有显著影响。然而,弯叶灰藓,、卵叶泥炭藓和卷叶凤尾藓的提取液抑制了皱叶酸模种子的活力指数。除此之外,卷叶凤尾藓还显著地抑制了它幼苗的干重。四种藓类的提取液对虎杖的种子活力指数和干重,但是卷叶凤尾藓的提取液对幼苗的生长有显著的影响。苔藓植物粗提取液对种子萌发和幼苗生长的影响的机制并不清楚,有待进一步研究。

关键词:苔藓植物;皱叶酸模;虎杖;种子萌发;幼苗生长

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Effects of aqueous extract of four mosses on seed germination and early seedling growth of two Polygonaceae plants

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Abstract: In this paper, the possible influences of the aqueous extracts of four bryophytes(*Sphagnum ovatum*, *Fissidens cristatus*, *Hypnum callichroum* and *Polytrichum commune*)on the seed germination and young seedling growth of two vascular plants(*Rumex crispus*, *Polygonum cuspidatum*) were investigated. The results showed that the aqueous extracts of these four mosses didn't affect seed germination. However, the crude aqueous extracts of *H. callichroum*, *S. ovatum* and *F. cristatus* inhibited the vigor indices of *R. crispus*. Furthermore, the crude aqueous extract of *F. cristatus* significantly enhanced its dry seedling mass, and *F. cristatus* and *S. ovatum* significantly inhibited its seedling elongation. For *P. cuspidatum*, the crude aqueous extracts of four mosses had little effects on its vigor indices and dry mass, but the aqueous extract of *F. cristatu* had significantly negative influences on its seedling elongation. The possible mechanism that the aqueous extracts of these four bryophytes influencing seed germination and seedling growth of these two vascular plants was not determined. More studies are needed.

Key words: Bryophyte; *Rumex crispus*; *Polygonum cuspidatum*; seed germination; seedling growth

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1 Introduction

Bryophytes are small plants without true stems, leaves, roots and vascular system, so they have not been attracted importance for a long time. However, as the main ground cover of many ecosystems, bryophytes play an important role in the population regeneration of some vascular plants (Lin *et al.*, 2006; Willam *et al.*, 1997). Studies about the influences of bryophytes on the seed germination and/or early seedling growth of vascular plants were documented in various plant communities as well as greenhouse experiments (Manuela, 2000; Ekaterina & Joan, 2003; Merit & Martin, 2004; Aimeedelach & Robin, 2002). Most of them commonly investigated the non-chemical interactions including the influence of microenvironment caused by bryophyte mat. There is increasing information about the chemical interactions between bryophytes and vascular plants. Some secondary compounds produced by bryophytes have been proved to affect seed germination and seedling growth (Lou, 2006; Lin *et al.*, 2006). The flavonoids from some mosses could inhibit the spore germination, protonemal development of other bryophyte species, as well as the root growth and even cause the morphological alterations of some vascular plants (Adriana *et al.*, 2003; Asakawa, 1981, 1998).

In the Beishan Mountain of Jinhua, Zhejiang Province, China, two species of Polygonaceae, *R. crispus* and *P. cuspidatum* often grow with some mosses such as *H. callichroum*, *F. cristatus*, *S. ovatum*, *P. commune*. On the basis of the available information regarding the chemical interaction of bryophytes on the seed germination and seedling growth of other plants, we guessed that these mosses might have some influences on populations of *R. crispus* and *P. cuspidatum*, but the evidences are lacking. The objective of this study was therefore to elucidate the possible influences of the aqueous exacts of these four mosses on the seed germination and seedling growth of two species of Polygonaceae.

2 Material and Methords

The seeds of *P. cuspidatum* and *R. crispus* (Polygonaceae) were collected in September and October 2006 from wild plants growing in Beishan Mountain of Jinhua, Zhejiang Province, China (29° 13′N; 119°38′E, altitude 1 178 m). The seeds were then stored in plastic bags at 4 ℃ in a fridge until sowing. The seeds of *P. cuspidatum* have a high coat-imposed dormancy mainly due to water impermeability. Therefore, additional pretreatments (98% sulphuric acid for five minutes) have been conducted to overcome the coat dormancy.

Four mosses, *S. ovatum*, *F. cristatus*, *H. callichroum* and *P. commune* were collected in October and November 2006 in the Beishan of Jinhua, Zhejiang Province, China, nearby the place where the seeds of *P. cuspidatum* and *R. crispus* were collected. The materials were washed with running water to wipe off the impurity, then washed with distilled water and air-dried at 40 ℃ for 72 h, cut into 2-3 cm sections. The materials (10 g) were extracted with 250 mL of distilled water at 25 ℃ for 24 h in the darkness. The extracts were filtered and centrifuged at 1 500 rpm/min at 15 ℃ for 15 min. The supernatants were stored at 4 ℃ for one week till use.

The study was conducted at the department of botany research in Zhejiang Normal University. Healthy and mature seeds were selected through socking in water for five minutes. For both experiments, selected seeds were sterilized using 1% Javel water for 10 min followed by repeated washing (four times) with distilled water. The seeds of *R. crispus* were then socked in the distilled water for 24 h at(25±0.5) ℃ in the darkness, while the pretreated seeds of *P. cuspidatum* were washed with distilled water and then socked into distilled water for 2 h at(25±0.5) ℃ in the darkness. 50 seeds of *R. crispus* or 25 seeds of *P. cuspidatum* were

placed in 93 mm Petri dishes with two layers of filter paper moistened each with 5 mL of distilled water(control), or 5 mL aqueous extracts of the mosses. Each experiment was conducted with three replicates. To determine if the observed differences could be a consequence of an osmotic potential effect, we conducted an assay using solutions with an inert solute(NaCl)of similar osmotic potentials as the aqueous extracts. Seed germination tests were carried out at 30/20 ℃ under a photoperiod of 10 h light/14 h darkness. Seeds with radicles 2 mm long were counted as germinated. Germination rate was recorded at 24 h intervals until the cumulative number of germinated seeds became stable in two consecutive records or when 100% germination was achieved. The number of germinated seeds, length of root and shoot, dry weight of seedling was recorded. The root length included the total root and radicel elongation, while vigor index is the product of percentage germination and root length (Neeru & Johannes, 2006). Germination data were subjected to one-way analysis of variance(ANOVA)using SPSS 15.0.

3 Results

3.1 Osmotic potential effect of the crude aqueous extract on seed germination

The following observed differences were not resulted from an osmotic potential effect, because assays with solutions with electrical conductivity of 74.1, 95.2, 346, and 538 μSm^{-1} (NaCl solution), which were similar to those of the aqueous extracts of mosses, do not show significant difference(*P. cuspidatum*, $F=0.25$, $P=0.905$; R. crispus, $F=2.184$, $P=0.144$)in final germination percentage.

Table 1 Effects of bryophyte leachate on seed germination and seedling growth of *R. crispus* and *P. cuspidatum*

Ietm	*R. crispus*				*P. cuspidatum*			
	Seed germination (%)	Vigor indices	Dry weight (mg)	Length of seedling (mm)	Seed germination (%)	Vigor indices	Dry weight (mg)	Length of seedling (mm)
Control	96.0±2.0a	392.3±10.5a	6.0±0.4a	21.9±0.5a	67.5±3.1a	171.0±9.2a	38.5±1.6a	27.7±0.8a
Hypnum callichroum	92.0±3.1a	356.5±12.5b	6.3±0.2ab	20.4±0.4ab	73.3±4.8a	199.4±2.7a	31.9±3.7a	29.2±1.2a
Polytrichum commune	93.3±1.8a	376.2±5.9a	6.8±0.4b	22.5±0.7ac	70.7±6.7a	170.9±21.0a	36.3±1.9a	26.9±0.8a
Fissidens cristatus	93.3±2.7a	187.5±5.8c	7.3±0.1bc	11.8±0.4d	74.7±1.3a	162.3±9.6a	41.1±2.4a	23.6±1.2b
Sphagnum ovatum	90.0±3.0a	237.2±8.1d	6.3±0.2a	14.9±0.5e	73.3±4.8a	179.9±9.4a	37.7±2.5a	25.8±1.6a

Note: Values represent mean ± standard error. Values were compared with the control($P\leqslant 0.05$). Means with the same letter do not differ significantly at the 5 % level.

3.2 Effect of the aqueous extracts of mosses on seed germination and seedling growth of *R. crispus*

For bryophyte species, no significant differences were observed in the influences of the aqueous extracts of four mosses on seed germination of *R. crispus* compared with the control(Table 1). However, the crude aqueous extract of *S. ovatum*, *F. cristatus* W and *H. callichroum* significantly decreased the seed vigor index. The lowest vigor index was recorded for the effects of *F. cristatus*. However, the crude aqueous extract of *P. commune* and *F. cristatus* promoted significantly seedling dry mass of *R. crispus*. Comparing to the positive effects of *P. commune*, the effects of *F. cristatus* was more positive. As for the seedling elongation, only *F. cristatus* and *S. ovatum* had negative effects(Table 1).

3.3 Effects of aqueous extract of mosses on seed germination and seedling growth of *P. cuspidatum*

There were no or little effects of aqueous extracts of four mosses on seed germination of *P. cuspidatum* as well as on vigor indices and dry weight. Only the significantly negative effects of the aqueous extract of *F. cristatus* could be reserved on the length of seedling.

4 Discussion

Some secondary compounds of bryophytes often have negative effects on other plants, especially on other bryophytes and vascular plants, because most bryophytes grow in the harsh environments, they not only

have to protect them from the invasion of bacteria, epiphytes, vermins and insects, but also compete with other plants for survival (Lou, 2006). Recently, the crude extracts of some mosses on the crop seed sprouting had been studied (Du, *et al.*, 2004). Lin *et al.* (2006) studied two bryophytes in dark coniferous forest of Changbai Mountains on three conifers seed germination and seedling growth. The effects varied with the moss and vascular plant species. Our results clearly indicated that there were no or little effects for the crude aqueous extract of the four mosses on the seed germination of two Polygonaceae plants. However, the effects of the crude extracts of four mosses on the seedling growth of the two vascular plants were significant. The effects varied with the bryophyte species and the testees. In the present paper, the effect of the crude extract of four mosses on seedling morphology of *R. crispus* was more significant than on seed growth of *P. cuspidatum*, which may be due to the size of the seeds.

Seed vigor index is an important physiological index for the ability of absorbing the nutrition and anabolism of root (Liu, 2003). In this paper, the effects of aqueous extracts of the mosses on seed vigor indices of the two Polygonaceae plants was all significantly negative if there were some effects. The results provided some evidences to support the relation between mosses and vascular vegetables, but such relation varied with moss species and vascular plants. There are two possible explanations of the effects of four mosses on seedling growth observed in this paper. First, seedling may be affected by the pH of the crude extract. It has been showed that seedling growth in most of species was markedly affected by pH, and the effects of pH on roots always affected the seedling elongation, the fresh and dry weight of seedling (Huang *et al.*, 2007). Second, the effects might be related with the difference of secondary compounds of mosses, which has been reported to regulate the plant growth. In spite of the fact that the aqueous extract of four mosses studied in this paper didn't affect the seed germination of two Polygonaceae plants, but those seedling with higher total seedling dry mass and longer root length had superiority in penetrating in soil for survival. The early mortality of seedling may attribute to lower total seedling dry mass and lower root length.

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