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Lentinan Promotes the Root of *Brassica Campestris* L.

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ABSTRACT

The aim of this work was to study the effect of lentinan on Brassica campestris L (rape). Spraying on the leaves of lentinan B. campestris L. at 0.05×10^{-6} g m l^{-1} concentration significantly promoted the root elongation (P < 0.05). The results for the first time showed that lentinan could prolongate roots as a new plant hormone.

Key words: Lentinan, oilseed rape, promoting root

INTRODUCTION

Chihara et al. (1969) had first extracted lentinan and demonstrated that lentinan exhibited antitumor bioactivities. Maeda et al. (1971) and Suzuki et al. (1982) reported the weight-average molecular weight and number-average molecular weight of lentinan as $9.5 \times 10^5 \sim 10.5 \times 10^5$ and $3 \times 10^5 \sim 8 \times 10^5$, respectively. Structyrlly, lentinan contains a β - (1 - 3) - D - glucan backbone with many β - (1 - 6) connection of glucose. It has been reported that lentinan exists as triple-helical chains in aqueous solution, which is related to its bioactivity It may turn into a single random coils in water/dimethyl sulfoxide (DMSO) mixtures or in sodium hydroxide solution or at 140°C (Zhang et al. 2002; 2005; Wang et al. 2008). Lentinan has been used in the preventions and treatments of many diseases originated from cancers. Studies have found that the intakes of lentinan through different ways could combine to monocytes and exhibit anticancer bioactivitiy (Masaaki et al. 1996). Lentinan stimulates different receptors, such as natural killer cells, T cells, B cells and macrophages to strengthen the body's immune system, so as to achieve its antitumor, antiviral antibacterial bioactivities (Ren et al. 2013). In addition, lentinan can stimulate plants to close off the virus infection sites, so as to inhibit the viral infection of the plants and induce systemic acquired resistance of the plants to suppress virus proliferation (Wang et al. 2011). The aims of this study was evaluate the effect of lentinan on rape.

MATERIAL AND METHODS

Lentinan Fermentation and Structure Identification

The lentinan was isolated and purified from the fresh mycelia of *Lentinula edodes* (Cao et al. 2012). The chemical groups of lentinan were identified by IR.

Bioactivity of the Rape Seeds Identification

The seeds of *B. campestris* L. were dissolved in 1% NaClO solution for 5 min, and put into a Petri dish covered by filter paper. Distilled water was added into the dish to maintain the seeds as wet. The germination potential and germination rate of

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the seeds was measured after 3 and 7 days, respectively.

Experimental Design

Twenty rape seeds were sowed to plastic crates $(30\times20~\text{cm}^2)$, with perlite as matrix. The crates were placed in greenhouse with 14 hours sunlight every day and the temperature between $25 \sim 40^{\circ}\text{C}$. A 1/5 MS culture (Murashige et al. 1962) was sprayed on the seeds after germination every week $(100~\text{mL}~\text{per}~600~\text{cm}^2)$. Two weeks later, lentinan solution was sprayed on the seedlings every week $(50~\text{mL}~\text{per}~600~\text{cm}^2)$. The concentrations of lentinan solution were set as 5×10^{-6} , 0.5×10^{-6} and $0.05\times10^{-6}~\text{g mL}^{-1}$. The plants were harvested after five weeks and root length, hypocotyl length, dry weight of root and hypocotyl of each plant were measured (Fig. 1A).

RESULTS AND DISCUSSIONS

During the submerged fermentation of lentinan, the biomass of hypha could reached 2.75±0.98 g mL⁻¹ and the concentration of exopolysaccharides (EPS) was 1.72±0.23 g mL⁻¹. The purified lentinan was analysed by infrared spectrum, which showed that the constituents of lentinan were complex, and lentinan had multiple of functional groups (Table 1), hence, it could possibly possess several biological functions.

The germination potential and germination rate of rape seedling is 79% and 89% respectively, The rape plants were harvested and put together to find the differences between the groups sprayed by pure water and lentinan solution respectively (Fig. 1B, C). Which showed that the lentinan concentration of 0.05×10^{-6} g mL⁻¹ group resulted a higher root elongation than the distilled water group.

The variation trend of root length, hypocotyl length and ratio of root and hypocotyl are presented in Figure 2A. It showed after the treatment of 0.05×10^{-6} g mL⁻¹ of lentinan, resulted higher length of root than the control (distilled water). The promoting effect to the length of root was the best and was less as lentinan concentration increased. However, for hypocotyl, it showed a reverse trend.

The changes of dry weight of the root and hypocotyl are shown in Figure 2B. It showed that with different concentrations of lentinan, the dry weight of root was similar to the control. Evidently, the roots were elongated but had no change in total number of cells in the treatments at different concentrations of lentinan.

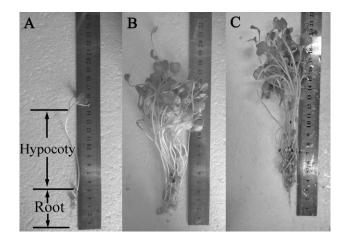
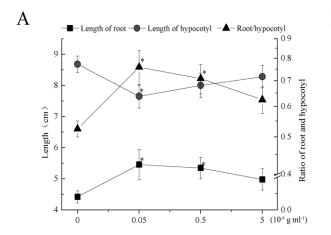


Figure 1 - Length measurement and the comparisons of 0.05×10^{-6} g mL⁻¹ group and control group. **A**) The length of root and hypocotyl. **B**) The control group which was sparyed by distilled water. **C**) The 0.05×10^{-6} g mL⁻¹ group which was sparyed by the concentration of 0.05×10^{-6} g mL⁻¹ lentinan solution.

Table 1 - Lentinan infrared absorption peak wave number and its corresponding relationship with functional groups.

| The absorption peak wave number of lentinan (cm ⁻¹) | Vibration type | Radical group |
|---|--|---------------------------------------|
| 3416 | O-H stretching vibration | Primary alcohol and secondary alcohol |
| 2926 | C-H stretching vibration | Equilibration C-H |
| 1631 | C=O stretching vibration or Hydration of vibration | -COOH (C-O) |
| 1385 | C-H Variable Angle vibration | $-CH_2$ |
| 1196 | C-O-C stretching vibration | cyclic ether |
| 1138 | C-O-C stretching vibration | Two alkyl ether |
| 924, 897 | C-O-C Asymmetric stretching vibration | Pyranose ring |
| 877, 835 | C-H Variable Angle vibration | The furanose ring |



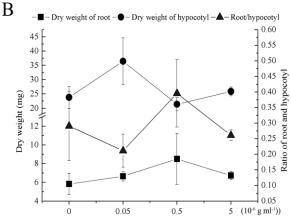


Figure 2 - The variance of root and hypocotyl between the control group and treatment groups. **A**) The variation trend of the length of root and hypocotyl. **B**) The variation trend of the dry weight of root and hypocotyl. (The "*" means the data of this group is significantly different from the data of control group).

This study revealed that, lentinan significant influence on the growth of root and hypocotyl of oilseed rape. Lentinan can promote the elongation of the roots but not the dry weight. Hence, it might promote the physiological activity of elongation region of root. It had no similarity with the known plant hormones. Since the concentration of lentinan used was low, it could be considered as plant growth regulator of sensibility. If the effect of inhibiting growth of hypocotyl and promoting elongation of root of lentinan also adapt to other plants, it could be used as one of broad spectrum of lodging-resistant materials, which would be of great significance in agricultural industry.

CONCLUSION

Based on the result that the lentinan efficiently enhanced root prolongation under very low concentration, it coulded be suggested that the lentinan might impact the plants as a new plant hormone.

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