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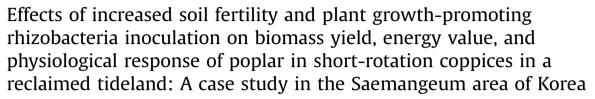
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Research paper





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ABSTRACT

This study aims to investigate the physiological response, biomass yield and its bio-energy value to charcoal mineral fertilizer and Bacillus subtilis strain JS, a plant growth-promoting rhizobacteria, under nine different soil-enrichment procedures, and to provide a reference for suitable management strategies for short-rotation coppices on the marginal soils of reclaimed tidelands in Saemangeum area of Korea. The treatments used in this experiment were as follows: 1) 1:100 (volume basis) diluted rhizobacteria inoculation {B1, (B. subtilis: double-distilled water, v/v)}, 2) 1:50 diluted rhizobacteria inoculation (B2), 3) mineral fertilization {F1, (charcoal 200 kg ha⁻¹)}, 4) additional mineral fertilization {F2, (charcoal 300 kg ha⁻¹)}, 5) F1+B1, 6) F2+B1, 7) F1+B2, 8) F2+B2, and 9) control plot with no soil enrichment (C). The plant species used in this study is Populus euramericana. Net photosynthetic assimilation using leaves acclimated for 2 min at 1500 μ mol m⁻² s⁻¹ was the highest in F1+B2, followed by B2+F2 and B2. Total biomass yield in F2+B2 was the highest by a significant margin among the nine soil-enrichment procedures, whereas that in the control soil was lowest. Plants in all soil conditions provided similar net calorific values of dry mass, and they all fell within the third grade of wood pellet quality criteria of the National Institute of Forest Science standard specification (Korea), within the premium category in the Pellet Fuels Institute standard specification (US), and within the A1 category in the EN-Plus standard specification (EU). Moreover, it was confirmed that the poplar was suited to produce wood pellets for heating energy.

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1. Introduction

Wood biomass in the form of wood pellets is a form of energy with a high potential to economically offset greenhouse gases. However, the wood biomass potential from forests is limited by technical constraints, ecological restrictions, and the sustainability principles of forest management [1,2]. Woody crops have an

important role to play, particularly in short-rotation coppices (SRCs), because of their numerous ecological benefits: they positively affect biodiversity, nutrient capture, and carbon circulation in the soil-plant-atmosphere system, especially in marginal areas. Moreover, they protect the soil from water and wind erosion [3,4], and the use of marginal areas for their production might contribute to a sustainable energy supply [2].

Biomass has always been the most important renewable source for heat generation [5]. Among the many ways to produce wood biomass, SRC culture offers high productivity. Fast-growing trees, such as poplars, are planted with high areal density and harvested in 3–5 year rotations. Unlike natural forests, SRCs can produce a large amount of biomass in a short-cycle harvest term [6,7]. Other advantages of SRCs are their: (1) adaptability to a new

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environment, (2) rapid growth, (3) high biomass and energy yield, (4) ability to remediate contaminated soil, (5) wide rhizosphere and high transpiration capability from fertilizer contamination, (6) flourishing rootlet development for absorbing moisture and ingredients, and (7) ability to enhance the quality of soil by providing an environment conducive to soil microbes [8.9]. Therefore, wood biomass, especially poplar, is being produced on short-rotation as a feedstock for sustainable, renewable, and carbon-neutral energy generation in Europe, USA, Canada, Japan, and Korea [7,10]. Since the adoption of the Renewable Energy Portfolio Standard began in Korea in 2012, the demand will exceed the renewable energy production capacity because the mandatory rate of renewable energy proposed to 4% by 2017, with further increase to 6% by 2020 and 10% by 2024 [11]. Moreover, the production capacity for wood pellets in Korea is predicted to be unable to meet demand after 2020 [12]. Therefore, the development of sustainable biomass energy sources is urgently needed, and interest in renewable resources has been increasing. However, planting SRCs is constrained by land availability, especially in Korea. Therefore, the Korea Forest Service established a plan to develop SRC culture on reclaimed land in Saemangeum. During the early stage of this plan, the reclaimed tideland area contained high levels of soluble salts and exchangeable sodium that hindered plant growth and reduced fertility [13]. Plantation growth will require the lands to be purified through a sodium elimination process, such as abundant rain or mixing soils. Currently, the area of Korean reclaimed tidelands is 135,100 ha; of which 44.415 ha is under development, and 90.685 ha have completed the reclamation process [14]. Because the developed area is large enough to be used, the free reclaimed tideland area will increase the production of biomass. However, current research is limited to promoting the growth of crops, and its development in fostering growth using rhizobacteria and mineral fertilizer remains insignificant. Therefore, applying inorganic or organic nutrients that can foster the growth of the rhizosphere system could increase the production of woody biomass on poor marginal land, such as that in the Saemangeum area. The results of our research support the production of wood biomass through increased soil fertility from plant growth-promoting rhizobacteria and charcoal mineral fertilizer.

Therefore, the objectives in this study were 1) to identify the effects of rhizobacteria and charcoal mineral fertilizer on biomass yield and physiological characteristics and 2) to evaluate the wood quality of poplar (*P. euramericana*) produced on reclaimed tideland.

2. Materials and methods

2.1. Site description and study design

The poplar plantation SRCs are situated in the Saemangeum reclaimed tideland, which is located in Gimje City, Jeollabuk-do Province in western Korea (35°52′ N, 126°47′ E). The climatic conditions are continental, with cold, snowy winters and hot, humid summers. The monthly rainfall and mean air temperature from June to October 2014 were 143.2 mm and 21.08 °C, respectively. The monthly mean duration of sunshine per day is 5.78 h for 5 months. The precipitation had a marked seasonal concentration in August. The maximum rainfall period was from July 10 until August 10. The next year, precipitation at the site was affected by drought. The monthly precipitation and monthly mean temperature from May to September 2015 were 67.8 mm and 21.69 °C, respectively (Fig. 1).

The field experiment and investigation were conducted on 1 ha from June 2014 until the end of August 2015. The experimental factor was fertilization method, and the following nine treatments were applied: 1) 1:100 diluted rhizobacteria inoculation {B1, (*B. subtilis*: double-distilled water, v/v)}, 2) 1:50 diluted

rhizobacteria inoculation {B2, (*B. subtilis*: double-distilled water, v/v)}, 3) mineral fertilization {F1, (charcoal 200 kg ha $^{-1}$)}, 4) additional mineral fertilization {F2, (charcoal 300 kg ha $^{-1}$)}, 5) F1+B1, 6) F2+B1, 7) F1+B2, 8) F2+B2, and 9) a control plot with no soil enrichment (C). The experiment was set up with three replications of each treatment using 5 m × 5 m plots. Each plot had 30 trees, and a total of nine sampling plots were laid down. The sample size and design are shown in Fig. 2. The soil at this site is silt-loam soil with low levels of organic matter, nitrogen, and phosphorus (Table 1). The soil of this site was considered poor, but the soil salinity was 0.001–0.01%, which was considerably lower than is common in reclaimed tidelands (0.72–1.24%) [15] because desalinization in the SRC of Saemangeum reclaimed land has been well conducting through water and soil management by blocking of capillary and planting halophytes, gradually [16].

2.2. Plant materials

The species selected for the experiment was Italian poplar (*Populus euramericana*), which is generally known for having the best biomass production in SRCs in Korea [19]. Many poplars were planted on the marginal lands of the Saemangeum reclaimed tideland in Gimje City, Jeollabuk-do Province in Korea on March 17, 2014. On our site, poplar seedlings (2-year-old) were grown for the production of biomass in about 1 ha during the experimental period. A total of 3000 un-rooted 1 m long poplar cuttings were planted manually at a density of 10000 ha $^{-1}$ (1 m \times 1 m spacing). Each cutting slip was replicated 3 times on elemental plots (5 m \times 5 m spacing per plot), among which 810 seedlings (30 seedlings \times 27 plots) were chosen for this study.

2.3. Soil fertilization

SRCs are often established on poor marginal soils (dry, damp, landfill, reclaimed tideland, or with poor location) or contaminated soils (abandoned mine areas). When woody crops are grown on such soils, the type and dosage of fertilizers are of great importance.

2.3.1. Chemical properties of charcoal fertilizer

Fertilizer treatment was performed before planting the poplar SRCs. The type of fertilizer used in this study was charcoal mineral fertilizer, which is made by wood vinegar of *Quercus* spp., and affects the rhizosphere (National Environment-friendly Organic Agro-Materials Classification Code: 07-OM-3-007, National Agricultural Products Quality Management Service, Korea). The topsoil was tilled and plowed with a cultivator, and charcoal mineral 200 kg ha⁻¹ (F1) and 300 kg ha⁻¹ (F2) were applied, respectively. The fertilizer was applied separately for each plot with a different concentration and was then covered with soil. The chemical characteristics of the fertilizer used in the experimental site are given in Table 2.

2.3.2. Rhizobacteria Bacillus subtilis JS culture

The rhizobacteria, *Bacillus subtilis* JS (National Patent Classification Code: KR1020140028777), was obtained by reproducing fungi isolated from the rhizosphere of flame grass (*Miscanthus sinensis* var. *purpurascens*) grown in Korea. This strain was chosen because it helped lettuce and tobacco plants survive in harsh conditions of abiotic stress. The isolates and inoculation used in this study followed previously published procedures. The *Bacillus subtilis* JS gram-positive bacterial strain identified by Song et al. [22] was used during the experiment. This bacterium was cultured by the following process. Bacterial strains were streaked onto nutrient agar medium (NA, Junsei Chemical, Japan) and cultured at 28 °C. A single colony on NA was transferred to 30 cm³ of nutrient broth

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