

Allelopathic impact of some dominants in clean cuttings of Scots pine forest under climate change conditions

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The objective of the study was to evaluate allelopathic effect of selected plant species (dominants which appear during the first year following clean cuttings of Scots pine (*Vacciniosa*) forest) on Scots pine (*Pinus sylvestris* L.) seed germination and seedlings development as well as to reveal whether the effect is dependent on temperature (day / night) regime conditions. Aqueous extracts of *Pleurozium schreberi*, *Vaccinium vitis-idaea* and *Calluna vulgaris* were tested. The effect of ground parts and roots was analysed.

The extracts exerted stronger phytotoxicity on germination of Scots pine seeds at 20 °C/29 °C than at 18/27 °C temperature regime. The inhibitory effect of the plant species and plant parts tested was stronger on radicle growth than on hypocotyls growth both at 18/27 °C and 20 °C/29 °C temperature regime.

Key words: *Pinus sylvestris* L., allelopathy, dominants, regeneration, climate change

INTRODUCTION

A large number of plants possess inhibitory effects on the germination and growth of neighbouring or successional plants by releasing allelopathic chemicals into the soil either as exudates / leachates from living tissues or by decomposition of plant residues (Putnam, Tang, 1986; Fernandez et al., 2008; Gallet, Pellissier, 2002; Inderjit, 2006). The early plant recruitment including germination, seedling growth and establishment is particularly important in plant communities' development and in forest regeneration as well (Einhellig, 1989; Nathan, Neeman, 2004; Lalljee et al., 2000). It is suggested that allelopathic activity of plants should be the major factor enabling implementation of growth management between neighbours and defence mechanisms developed during long co-evolution with their competitors and enemies (Donnelly et al., 2008; Gradeckas, Malinauskas, 2005; Luis et al., 2007; Utsugi et al., 2006).

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Allelopathy which is defined as a process of plant-plant chemical interaction with either positive or negative effects (Djurdjević et al., 2005; Carlini, Sa, 2002; Donnelly et al., 2008; Inderjit, Dakshini, 2000). On one hand, donor plants under stressful conditions usually increase both production and release of secondary metabolites (Baležentienė, Šėžienė, 2010; Grodzinsky, 1990; Meng et al., 2009), thereby resulting in higher concentration of allelochemicals in the environment. Besides allelochemicals, abiotic factors such as temperature, moisture, light, and nutrients, as well as biotic factors such as insects, herbivores, and pathogens may influence synergically plant regeneration. However, only a few studies deal with plant settlement and regeneration taking into account the concomitant effect of several biotic and abiotic parameters (Anaya, 1999; Foy, 1999). One of these factors is temperature.

The objective of the present study was to determine allelopathic effect of several herbaceous plant species on Scots pine (*Pinus sylvestris* L.) seed germination and seedlings growth depending on two

temperature regimes. Laboratory test was carried out in order to evaluate and to compare the allelopathic activity of the aqueous extracts produced from both ground parts and roots of dominant plant species which occur during the first year after clean cuttings of pine forests (*Vacciniosa*) at a different temperature regime. The allelopathic potential of some dominants of clean cuttings of pine forests (*Vaccinio-myrtillo Pinetum*): European blueberry, *Vaccinium vitis-idaea* L., common heather, *Calluna vulgaris* (L.) Hull, and feather moss, *Pleurozium schreberi* (Brid.) Mitt. at different growth stages (Gradeckas, Malinauskas, 2005; Jurelionis, Karazija, 1975) was demonstrated. Thus new information will be helpful for understanding the common allelochemical potential of dominant species of clean cuttings of forests, and also their impact on reforestation as well as on management of forest ecosystems under climate change conditions.

MATERIALS AND METHODS

To evaluate biological activity and impact of different donor-species and plant parts on acceptor-species Scots pine, *Pinus sylvestris* seed germination and seedling growth under a different temperature regime laboratory trials were carried out in Ecology Laboratory at the Institute of Forestry, Lithuanian Research Centre for Agriculture and Forestry. Plants and their parts (roots and shoots) were collected in three localities of clear cuttings of pine forest in Žėronys forestry (Vilnius district, Lithuania) and aqueous extracts for bioassay were produced as indicated below.

As typical dominant species (Baležentienė, Šėžienė, personal communication) in clean cuttings of pine forests (*Vaccinio-myrtillo Pinetum*) were admitted: European blueberry, *Vaccinium vitis-idaea* L., common heather, *Calluna vulgaris* (L.) Hull, and feather moss, *Pleurozium schreberi* (Brid.) Mitt. Laboratory trials were carried

out at Environment Laboratory of Lithuanian University of Agriculture. These species intensively grow and spread thus they become dominants, their impact on pine seed germination and forest regeneration is relevant to investigate in clear cuttings (Karazija, 1988). Plants and their parts (roots and shoots) were collected at flowering stage in clear cuttings of pine forest in Kačerginė (Kaunas district, Lithuania) for producing and bioassay of aqueous extracts (Table). BBCH (*Biologische Bundesanstalt, Bundessortenamt and Chemical Industry*) scale was chosen for determining plant growth stages (Meier, 2001).

Separate aqueous extracts of each plant species were prepared according to Grodzinsky. For details see Baležentienė, Sampietro, 2009; Baležentienė, Šėžienė, 2010. The germination was assessed according to International Seed Testing Association Technique (ISTA, 2009). As acceptor species-Scots pine 100 seeds were placed on filter paper in each 9 cm diameter Petri dishes 5 ml aqueous plant extract of 0.2% (m/v) concentration was added per Petri dish, 4 replications were used per treatments. Seeds placed in Petri dishes with distilled water served as a control. Petri dishes were kept (Immersion thermostats DC30, Roth, Germany) at natural 18 °C/27 °C (day / night) and 20 °C/29 °C (day / night) temperature for 21 day due to predicted rise of approx. 2 °C in mean global temperatures during the next century. Photoperiod used was 16 h/day. Germination was recorded after 7, 14 and 21 days. In order to evaluate extracts impact on seedling growth the length of radicles and hypocotyls was measured after 21 day. All the data are presented in relative numbers (%) compared to the control.

The confidence limits of the data were based on Wilkin's-λ test and Student's theoretical criterion (F) Standard error (SE) as well as statistical significant difference at the level of $p < 0.05$ was calculated.

Table. Donor plants' growth stage

| Donor species | Growth stage | Description BBCH-scales code |
|---|--------------|------------------------------|
| European blueberry, <i>Vaccinium vitis-idaea</i> L. | Flowering | 6/69 |
| Common heather, <i>Calluna vulgaris</i> (L.) Hull | Flowering | 6/67 |
| Feather moss, <i>Pleurozium schreberi</i> (Brid.) Mitt. | Vegetative | – |

RESULTS AND DISCUSSION

After the floristic assessment of plant community of selected clean cuttings of pine forest, it was assessed that *Pleurozium schreberi* and *Vaccinium vitis-idaea* have the dominating position. These plants were detected in 100% of the study plots of clean cuttings of pine forests, also the abundance of *Calluna vulgaris* was documented (Šežienė, Baležentienė, in preparation for publication). Feather mosses are an ever-present component of the boreal forest floor, thereafter their role in boreal forest ecosystems could be important (Gundale et al., 2011). While the ecology of these bryophytes within the boreal forest has been widely analysed by numerous researchers, the contribution of bryophytes and, in particular, feather mosses, to plant-plant interaction models was not clearly stated. Numerous studies (Kumarasamy et al., 2002; Orhan et al., 2007; Tokaloğlu, 2012) of natural productivity and medicine implications of *Vaccinium vitis-idaea* and *Calluna vulgaris* were done, but their allelopathic peculiarities in forest ecosystems had not been tested. Therefore, ongoing and proposed studies were designed to test the role of these dominant species on the pine seed germination.

During the experiment the impact of phytotoxicity of produced aqueous extracts on Scots pine seeds germination was evaluated depending on plant species, plant part, and a different temperature regime (Figure).

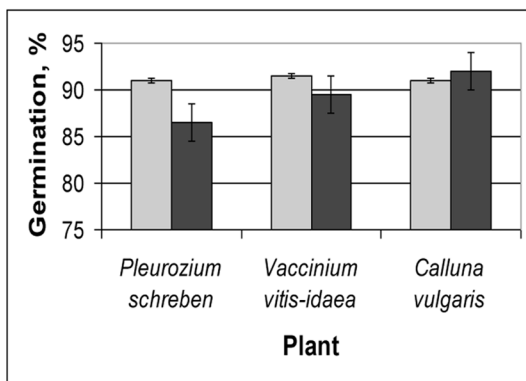
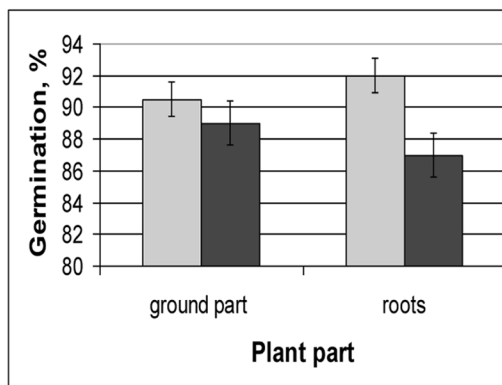
The aqueous extracts of *P. schreberi* showed the strongest phytotoxicity suppressing germination of Scots pine seeds within the limits from 8 to 13.5% at both 18/27 °C (mean seed germination 91%) and 20 °C/29 °C (mean seed germination 86.5%) temperature regime, respectively (Fig. a). The weakest suppressive effect by 8.5 and 8% was documented both for *V. vitis-idaea* extract (mean germination 91.5%) at 18/27 °C and that of *C. vulgaris* (92%) at 20 °C/29 °C, respectively.

According to earlier studies (Baležentienė, Šežienė, 2010; Lalljee et al., 2000; Louis et al., 2007; Mallik, 2003), seed germination and ecosystem (forest) regeneration after clear cutting are reliant on present plant species and plant parts. The ground part of all three donor-plant species tested indicated the lower inhibitory effect comparing with that caused by roots (Fig. b). Ground

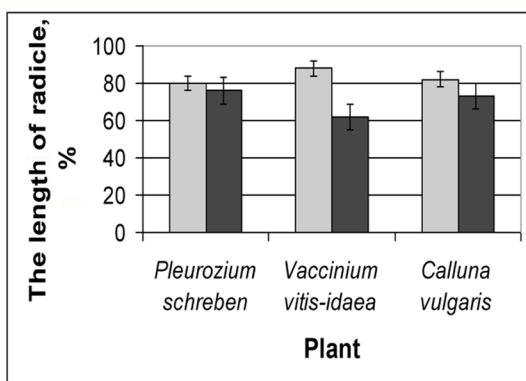
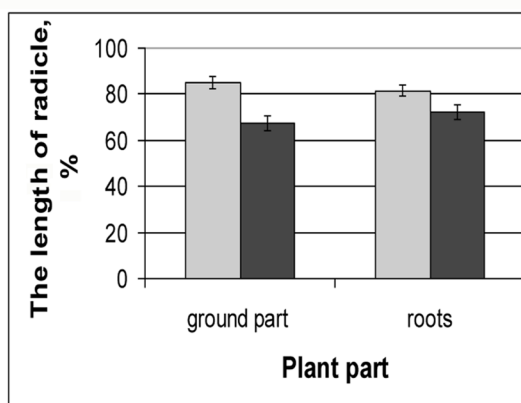
part decreased germination by 9.5% (mean relative germination 90.5%) at 18/27 °C and by 11% (mean germination 89%) at 20 °C/29 °C temperature regime. Roots inhibited germination by 8% (mean relative germination 92%) at 18/27 °C. Moreover, root phytotoxicity increased by 13% (mean germination 87%) at 20 °C/29 °C. Therefore, the temperature regime conditioned higher inhibition gradient and phytotoxicity of roots than those of ground part. This phenomenon of different impact of wooden plants (*Pinus* spp.) ground part and roots has been already observed in forest ecosystems by several researchers (Blanco, 2007; Fernandez, 2008).

Some studies have shown that the roots are more sensitive to allelochemicals than aerial parts of acceptor-seedlings (e. g. Djurdjević et al., 2005; Gallet, Pellissier, 2002). The results confirmed that the inhibitory effect on acceptor radicle growth of both different donor-plant (Fig. c, e) and their parts (Fig. d, f) was stronger than on growth of acceptor hypocotyls at both 18/27 °C and 20 °C/29 °C temperature regimes. *V. vitis-idaea* showed the weakest inhibitory effect on radicle growth at 18/27 °C (mean value 89% compared with control (100%)), but the strongest inhibitory effect at 20 °C/29 °C (mean value 62%). Consequently, *V. vitis-idaea* exhibited the highest thermo-sensitivity as its phytotoxicity gradient has changed in the broadest range between donor-species. However *P. schreberi* indicated the weakest inhibitory effect on acceptor-radicle growth at the higher temperature regime (mean value 75%) and the strongest at 18/27 °C (mean value 80%) (Fig. c). The radicle growth of pine seedlings was less affected by ground parts extracts of donor-species at 18/27 °C and by roots of plants at 20 °C/29 °C (mean value 85% and 72%) (Fig. d).

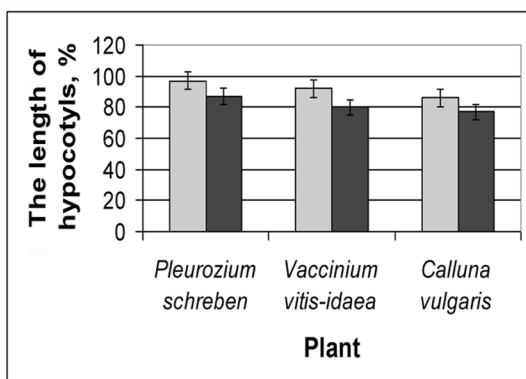
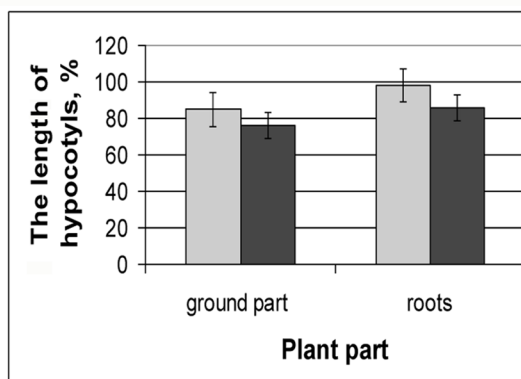
The length of hypocotyls of pine seedling was more affected by *C. vulgaris* aqueous extracts (85% and 78% of control) and less by *P. schreberi* aqueous extracts (98% and 87% of control) compared with *V. vitis-idaea* at both 18/27 °C and 20 °C/29 °C temperature regime (Fig. e). The ground parts of plants demonstrated the strongest inhibitory effect on the length of hypocotyls at both 18/27 °C (day / night) and 20 °C/29 °C temperature regime (85% and 75% of control) compared with roots (Fig. f).

**a****b**

($\lambda=0.81001, F=10.3277, p=0.0441$) ($\lambda=0.84790, F=6.9331, p=0.0070$)

**c****d**

($\lambda=0.77501, F=8.8986, p=0.01575$) ($\lambda=0.77854, F=1.60299, p=0.05075$)

**e****f**

($\lambda=0.71234, F=5.0828, p=0.00085$) ($\lambda=0.55949, F=21.652, p=0.0000$)

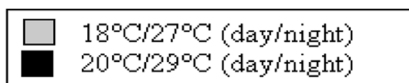


Figure. Phytotoxicity of donor-plant species and their part extracts on Scots pine germination (**a, b**), the length of radicles (**c, d**) and the length hypocotyls (**e, f**) at different temperature regime. Germination data presented as relative (% \pm SE) compared to the control. Vertical bars represent standard error (\pm SE)

This study demonstrated that the allelopathic activity could be affected by ambient temperature under changing climate conditions. Rise of ambient temperature by 2 °C conditioned increase of donor-species phytotoxicity on acceptor germination from 8 to 13.5%.

CONCLUSIONS

Inhibitory allelopathic effect of aqueous extracts produced from ground parts and roots of European blueberry, *Vaccinium vitis-idaea*; common heather, *Calluna vulgaris*; feather moss, *Pleurozium schreberi* (dominants of clean cuttings of pine forests) on Scots pine, *Pinus sylvestris* L., seed germination and seedling growth was documented. The effect depended on affecting plant species, plant part and temperature regime.

1. The aqueous extracts of *V. vitis-idaea*, *C. vulgaris* and *P. schreberi* of 1st year clean cuttings of pine forests exerted stronger phytotoxicity on germination of Scots pine seeds at 20 °C/29 °C than at 18/27 °C temperature regime.

2. The results confirmed that the inhibitory effect on acceptor radicle growth of *V. vitis-idaea*, *C. vulgaris* and *P. schreberi* and their parts was stronger than that on hypocotyl growth both at 18/27 °C and 20 °C/29 °C temperature regime.

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KAI KURIŲ DOMINANTŲ ALELOPATINIS POVEIKIS PLYNOSE PAPRASTOSIOS PUŠIES KIRTAVIETĖSE KINTANČIO KLIMATO SĄLYGOMIS

S a n t r a u k a

Šio darbo tikslas – nustatyti, ar kinta augalų alelopatinis poveikis paprastosios pušies (*Pinus sylvestris* L.) sėklų daigumui ir daigų vystymuisi priklausomai nuo pakitusių temperatūros režimo, atitinkančio prognozuojamą atšilimą. Laboratoriniu testu skirtinguose temperatūros režimuose palygintas alelopatinis aktyvumas vandeninių ekstraktų, paruoštų iš pirmųjų metų pušynų (*Vacciniosa*) kirtavietėse vyraujančių žolinių augalų *Pleurozium schreberi*, *Vaccinium vitis-idaea* ir *Calluna vulgaris* antžeminės dalies ir šaknų.

Bandymu buvo įvertintas vandeninių ekstraktų fitotoksiškumas priklausomai nuo augalo rūšies, augalo antžeminės (ūglių) ar požeminės (šaknų) dalies ir skirtingos temperatūros. Pirmųjų metų plynų kirtaviečių dominantų vandeniniai ekstraktai buvo labiau fitotoksiški pušų sėklų daigumui esant 20/29 °C negu 18/27 °C temperatūrai. Rezultatai rodo, kad esant tiek 18/27 °C, tiek ir 20 °C/29 °C temperatūrai, stipriau inhiuojamas šaknelių nei hipokotilio augimas.

Raktažodžiai: *Pinus sylvestris* L., alelopatija, dominantai, atsikūrimas, klimato kaita