

RST – Scientific report regarding the implementation of the "NATivE – Potential climate chaNge mediated recovery of AuTochthonous broadleaf species to the detrIment of allochthonous planted conifErs" project (code: PN-III-P1-1.1-PD-2016-0583; Financing Contract for Project Execution NO. 41 / 2018) within the 02/05/2018 – 30/06/2020 period

I. INTRODUCTION – THE INTERNATIONAL AND NATIONAL SCIENTIFIC CONTEXT

Forests occupy about 30% of the Earth's surface, being ecosystems that play a crucial role in regulating climate at the local, regional, and global levels (Bonan et al. 2016). In turn, the climate determines the distribution of the different species of trees that form forests. However, climate change seems to severely affect forest ecosystems. This is due to the fact that the lifespan of trees is very long, an aspect that prevents them from adapting quickly to new climatic conditions and makes them particularly vulnerable to climate change (Lindner et al. 2010). In this regard, severe droughts, associated with climate change, more common in recent decades (IPCC 2013), have affected large areas of forest (i.e., decline and mortality) worldwide (Allen et al., 2010; Allen et al. al. 2015; Hartmann et al. 2018).

However, the response of trees and forests to the climate does not necessarily have to be understood as a direct one. Most of the time, severe droughts are only the last determining factor that kill weakened trees over time by other factors (Manion 1991). Such a factor is represented by the management measures that have been taken over time and that determine the current structure of the forests. However, the link between these management measures and severe droughts associated with climate change, which have the potential to affect large areas of forests (Allen et al., 2010; Allen et al. 2015; Hartmann et al. 2018), is poorly studied despite its in determining the future of the forests (Camarero et al. 2011; Vilà-Cabrera et al. 2011; Sánchez-Salguero et al. 2013).

Romania has an estimated forest area of 7 million ha, according to the last National Forest Inventory (IFN 2018). Although characterized by a temperate-continental climate, Romania will also register more severe and frequent droughts in the future (Lindner et al. 2010). In fact, the effects of such severe climatic events, associated with climate change, are already being felt as in Romania there are already significant events of tree decline and mortality that affect different species (Barbu and Popa 2001; Curiel Yuste et al. 2019; Sidor et al. 2019; Hereş et al. *under review*). Recent studies

conducted in a mountainous area of the Braşov region indicate that conifers planted outside their natural distribution range are among the most targeted species, i.e., Scots pine (*Pinus sylvestris* L.) and Scots pine (*Pinus nigra* Arn.) (Photo 1; Curiel Yuste et al. 2019). These two species, which were planted in the past on land originally occupied by native deciduous species, recorded in 2012 significant events of decline and mortality. According to recent studies, their mortality could be associated with severe droughts recorded in previous years (Curiel Yuste et al. 2019; Hereş et al. *under review*). At the same time, deciduous species (i.e., European beech, *Fagus sylvatica* L.; sessile oak, *Quercus petraea* Matt. Liebl.), that grow on the same slopes and that probably represent the original native species, did not register such decline and mortality events, indicating that they are probably better adapted than planted conifers to cope with severe droughts and therefore climate change (Petrea Ştefan master thesis). These results highlight the fact that management measures significantly influence the way tree species, which form (native vs. non-native) our forests, respond and cope with severe droughts associated with climate change (Hereş et al. *under review*).



Photo 1 – Scots pine affected by decline and mortality in the vicinity of the city of Braşov (*photo: Ana-Maria Hereş*)

Severe droughts associated with climate change therefore have the ability to severely affect forest ecosystems, and their effect also seems to depend on management measures, a less studied aspect. The consequences of these events of tree decline and

mortality are difficult to estimate, as they are complex and affect the ecosystem services provided by forests locally, regionally, and globally (i.e., timber production, social services, climate change mitigation, etc.; Anderegg et al. 2013). It is therefore critical to try to understand the interaction between climate change (i.e., severe droughts) and management measures because this interaction determines how trees and therefore forests respond to such challenges and will also determine the presence and permanence of forests and therefore the essential ecosystem services they provide for us. In this sense, the NATivE project ("*Potential climate chaNge mediated recovery of AuTochthonous broadleaf species to the detriLment of allochthonous planted conifErs*") was meant to connect the effects of severe droughts and those of management measures on trees and forests. Specifically, the NATivE project concentrated on the mixed coniferous and deciduous forests from the Braşov region. These forests have been affected by decline and mortality, but only planted conifer species (i.e., Scots pine and Black pine) have been affected by high rates of decline and mortality, while native deciduous species (i.e., sessile oak and European beech) do not seem to be affected in this regard.

The results of the NATivE project highlight the fact that the past is key in understanding the current and future dynamics of different tree species, these dynamics obviously depending on their native or non-native condition. Not many studies have addressed these aspects, and in Romania it is the first project of this type.

II. THE OBJECTIVES OF THE NATivE PROJECT

The main objective of the NATivE project was to study the forests from the Braşov region affected by important tree decline and mortality events. Specifically, these type of events were recorded in mixed forests of conifer and deciduous species, forests where Scots pine and Black pine have been planted (i.e., they are non-native species) while deciduous trees appear naturally, being native (Figure 1, Petrea Ştefan master thesis). It should be noted that the two species of conifers (i.e., Scots pine and Black pine) suffered significant rates of decline and mortality due to severe droughts registered in the region, while the predominant deciduous trees (sessile oak and European beech) were not affected by such climatic events. The NATivE project studied the growth (through annual tree-rings) of the four tree species (i.e., Scots pine, Black pine, European beech, and sessile oak), how they face to severe drought events, and how they regenerate.

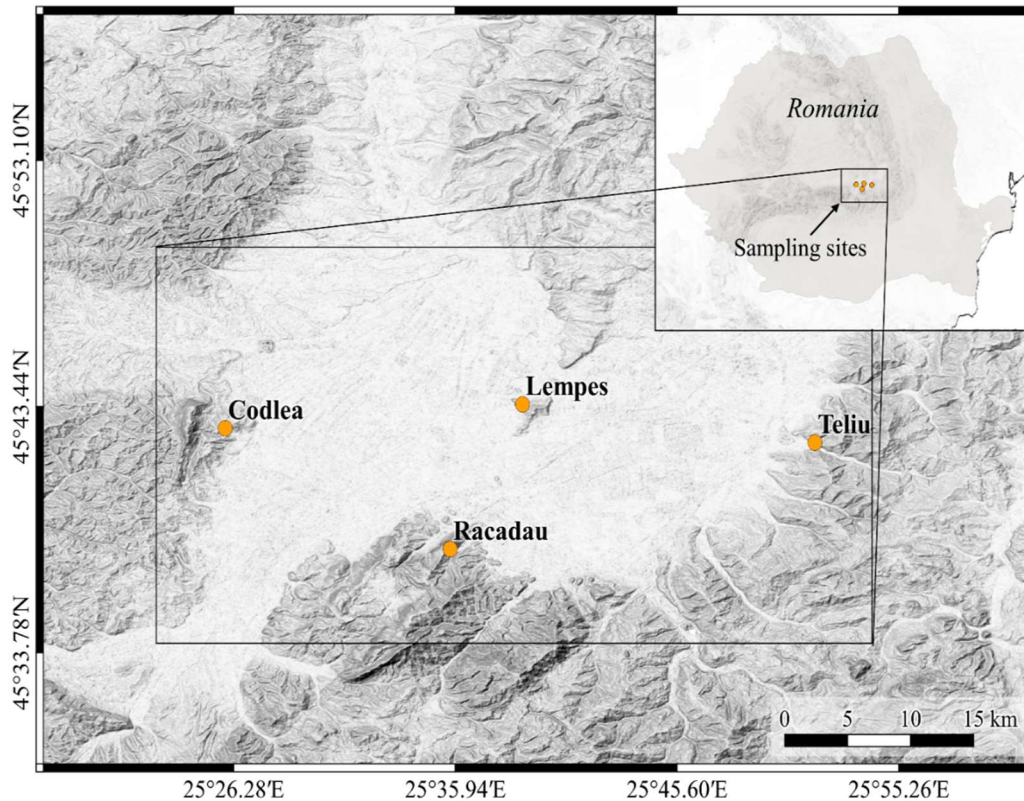


Figure 1 – The location of the four mixed forests in the vicinity of the city of Brașov: Codlea (Scots pine with sessile oak), Lempes (Black pine with sessile oak), Răcădău (Black pine with European beech) și Teliu (Scots pine with European beech)
(map made by: Petrea Ștefan; master thesis)

The specific objectives of the NATivE project were:

1). To study the extent to which drought could act as an inciting or contribution factor to the observed mortality of Scots pine and Black pine. By studying the annual tree-rings, we can go back in time and estimate how the trees grew when they were dealing with severe drought events and how they managed to recover or not after such events. In addition, estimates on how trees will be able to respond to drought events in the future, can be made.

2). To study the historical growth of the four species of native and non-native trees. The growth trend of the trees is a clear indicator of how these trees are doing. Specifically, growth trends can be used to determine in advance whether some trees are more likely to die when facing severe drought events (negative growth trends) or not (positive growth trends).

3). To study the resilience of the four tree species after they have been exposed to severe drought events. Such estimates are made by comparing growth trends before and after droughts. Trees that fail to return to previous-to-drought growth rates are more prone to die than those that do manage to return to previous-to-drought growth trends and are thus more resilient.

4). To study the regeneration processes of the four species of native and non-native trees. The seedlings that grow under the adult trees represent the future of the forest. Such data therefore helps to make estimations on the future composition of forest ecosystems.

III. STAGES AND ACTIVITIES

In order to achieve the main objective and the specific ones, within the NATIVE project several stages and activities were established. These activities took place over different periods of time throughout the project (**02/05/2018 - 30/06 / 2020**).

A first stage took place between **02/05/2018 and 31/12/2018**. Its purpose was to study the extent to which droughts act as inciting or contribution factor to the observed mortality of Scots pine and Black pine and to compare the historical growth trends of non-native conifer species with those of native deciduous species.

The activities related to this first stage were:

Activity 1.1: Selection of sites and tree species (non-native conifers and native deciduous trees), sampling (extraction of wood cores), as well conducting a first inventory of seedlings to estimate the regeneration of these tree species.

Activity 1.2: Measurements of annual tree-rings using specific dendrochronological methods and programs, activity that will be continued during stage II.

A second stage took place between **01/01/2019 and 31/12/2019**. Its purpose was to study the resilience of the trees after facing severe drought events.

The activities related to this first stage were:

Activity 2.1: The measurements of the tree-rings will be continued. For this, specific dendrochronological methods and software will be used. This activity started in the I stage.

Activity 2.2: After evaluating the field seedlings inventory made in the I stage, a second one will be conducted, is necessary.

Activity 2.3: Carrying out statistical analyses and writing manuscripts in order to disseminate (international conference) the preliminary results of the NATivE project and publish them in specialized journals. This activity will continue in stage III.

A third stage took place between **01/01/2020 and 30/06/2020**. Its purpose was to study regeneration processes in forests affected by tree decline and mortality.

Activitățile aferente acestei a treia etape, au fost:

Activity 3.1: Carrying out statistical analyzes and writing articles in order to publish them in specialized ISI journals, an activity that continues from stage II.

Activity 3.2: Dissemination (international conference) of the final results of the NATivE project.

IV. THE RESULTS OF THE NATivE PROJECT

I stage (02/05/2018 – 31/12/2018):

A total of four study sites (Table 1) were identified and selected for the implementation of the NATivE project. These study sites are represented by four mixed forests (conifer with deciduous species): Codlea, Lempeș, Răcădău, and Teliu (Figure 1, Table 1). All four study sites were characterized by high rates of decline and mortality among planted conifers (i.e., Scots pine and Black pine). Deciduous species (i.e., European beech and sessile oak), on the other hand, did not show such symptoms, although they grow on the same slopes with the conifers and share therefore similar conditions in all aspects.

Table 1: Study sites and conifer (Scots pine and Black pine) and deciduous (European beech and sessile oak) species used for the implementation of the NATIVE project

Study sites	Geographic coordinates	Species	No. of trees	Age (years)
Codlea	45°42'35.22"N	Scots pine	30	117
	25°25'54.93"E	Sessile oak	30	134
Teliu	45°42'1.66"N	Scots pine	30	117
	25°51'36.30"E	European beech	30	50
Lempeş	45°43'31.57"N	Black pine	30	105
	25°38'52.11"E	Sessile oak	30	94
Răcădău	45°37'50.77"N	Black pine	30	99
	25°35'43.07"E	European beech	30	57

A total of 240 trees were selected, marked, and georeferenced (Table 1). Two wood cores were extracted from each of them using Pressler increment borers (5 mm) (Photo 2). Both wood cores sampling and processing were done following standard dendrochronological procedures (Fritts 1976). All trees used to implement the NATIVE project were selected to be adult and dominant. At the same time, they had to have: similar diameters, as similar as possible microclimatic conditions, crowns without severe defoliation (i.e., <10 - 20%; this visual evaluation is accepted as a good indicator of the health of a tree and was always made by the same person to ensure the consistency of the data), and not to show signs of infections (i.e., fungi, insects). This selection was made in order to avoid the introduction of unwanted noise in statistical analyses.

A total of 480 wood cores were extracted during field campaigns that were conducted in October and November 2018. The wood cores were extracted in late autumn so that the last tree-ring, corresponding to 2018, was complete. These wood cores were subsequently processed in the laboratory (i.e., dried, mounted on permanent wood supports, and cleaned), scanned, measured (with an accuracy of 0.01 mm) using CooRecorder and Cdendro software (Cybis Elektronik & Data, Saltsjöbaden, Sweden), and checked (COFECHA, Holmes 1983). All chronologies obtained from both conifers (i.e., Scots pine and Black pine) and deciduous (i.e., European beech and sessile oak)

species met all quality standards from a dendrochronological point of view (Grissino-Mayer 2001). Therefore, these chronologies are scientifically valuable both for the NATivE project and for further studies, representing an important database for the Romanian forests. It should be mentioned that chronologies were obtained at the tree, species, and study sites levels. Along with the measurements of the annual tree-rings, the age of the studied trees was also estimated (i.e., the total number of tree-rings counted at 1.3 m height) (Table 1).



Photo 2 – Wood cores extractions from sessile oak (Lempeș) (*photo: Jorge Curiel Yuste*)

II stage (01/01/2019 – 31/12/2019):

In the second stage of the project implementation, the measurements of the wood cores extracted in the first stage was completed. This process of measuring annual tree-rings is extremely elaborate and therefore a lot of time is needed in order be correctly done and to meet all quality standards from a dendrochronological point of view (Grissino-Mayer 2001). At the same time, the number of wood cores extracted from the 240 trees (i.e., Scots pine, Black pine, European beech, and sessile oak) was high (i.e., 480 wood cores).

In this second stage of implementation of the NATivE project, the measurement of the wood cores was carried out with the support of a master student (Petrea Ștefan; "Silvicultură multifuncțională") from the Transilvania University of Brașov. Based on the measurements made on deciduous wood cores (i.e., European beech and sessile

oak), Ștefan could complete his master's thesis. This master's thesis will be defended on June 30, 2020 and was carried out under the supervision of dr. Hereș Ana-Maria (NATivE project director) and dr. Petritan Ion Catalin (professor of Transilvania University of Brașov).

In the second stage of implementation of the NATivE project, the seedling inventory was also made in order to estimate the regeneration rates. This inventory was made in June and July in order to have a more accurate image of the individuals that survived the winter and the new ones. Simultaneously with the regeneration estimation, an estimation of the competition, to which the trees studied within the NATivE project are subjected to, was also made. Both the regeneration and the competition surveys were made considering a circle with a radius of 5 m (Photo 3) around each Scots pine, Black pine, European beech, and sessile oak (i.e., the 240 trees from which wood cores were extracted in 2018). In order to ensure the quality and consistency of the data and to minimize possible errors, these estimates were always made by three people who contrasted the data obtained for validation. Thus, in order to estimate regeneration, all seedlings (diameter <10 cm) were counted and identified at the species level. At the same time, in order to estimate the competition, all the trees with a diameter > 10 cm were counted, identified at the species level, and measured.



Photo 3 – Surveys mode around a sessile oak (Codlea) (*photo:Ana-Maria Hereș*)

In 2019, the statistical analysis of the data and their dissemination began, according to the proposed working plan. A first ISI article was published (Curiel Yuste et al. 2019), in which the soil part of the study sites was studied. This article is a basis for the NATivE project because it tried to understand the processes that take place at soil level

when Scots pine and Black pine trees register decline and mortality. Specifically, this article highlights the fact that tree mortality results in increased soil respiration (i.e., significant CO₂ emissions) which causes forests to become carbon sources and diminish their ability to absorb carbon and thus combat climate change. These results are therefore of particular importance given that CO₂ emissions contribute to exacerbate climate change (Curiel Yuste et al. 2019).

Also during 2019, intense work was done on a second article in which only conifers were considered (Hereş et al. *under review*). Specifically, in this article we tried to understand the mortality of Sild pine and Black pine from a dendrochronological point of view. The results of this article show that the two planted pine species have suffered significant negative growth trends (Figure 2). At the same time, it was possible to establish a direct relationship between severe drought events, such as the one registered in 2011 (Ionita et al. 2016) and the mortality of the trees. Both Scots pine and Black pine are very affected by severe drought events, their recovery being difficult (i.e., they are not resilient species). In addition, neither Scots pine nor Black pine regenerate successfully, their number of seedlings being very low compared to that of deciduous trees that do regenerate well (Figure 3). These results indicate that species planted outside their natural distribution range (i.e., non-native), such as Scots pine and Black pine in the region of Braşov, do not seem to have the capacity to cope with climate change because they show significant negative growth trends (Cailleret et al. 2017), are not drought resilient (DeSoto et al. 2020), and do not regenerate (Zlatanov et al. 2010). This article under second revision in the STOTEN (Science of the Total Environment) journal.

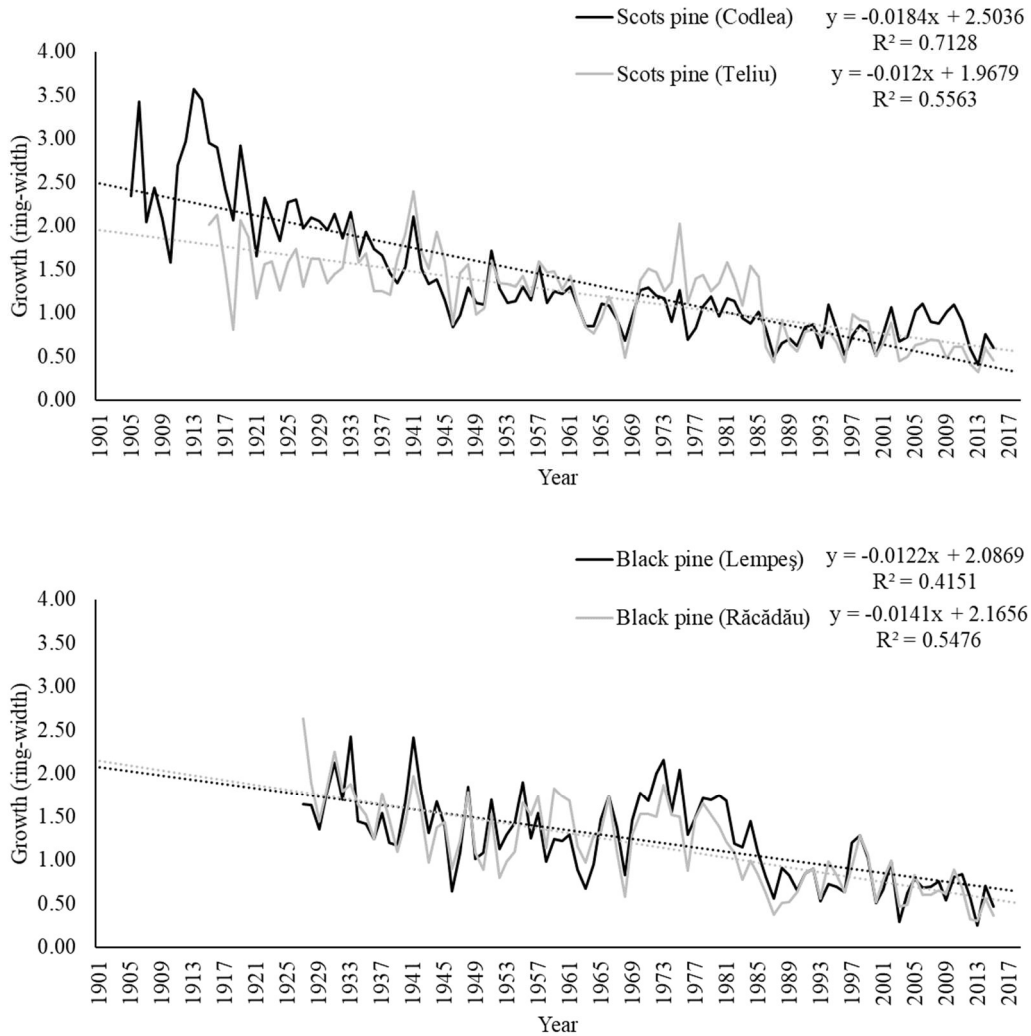


Figure 2 – Growth (i.e., ring-width) trends of Scots pine and Black pine species
(Hereș et al. *under review*)

The results obtained during 2019 were presented at the EGU General Assembly 2019 conference held in Vienna (Austria). During this conference, the project director presented preliminary results of the NATIVe project in a poster (Title: "*How does drought-related mortality affect conifer species? The role of historical management practices on the current response of trees to climate*").

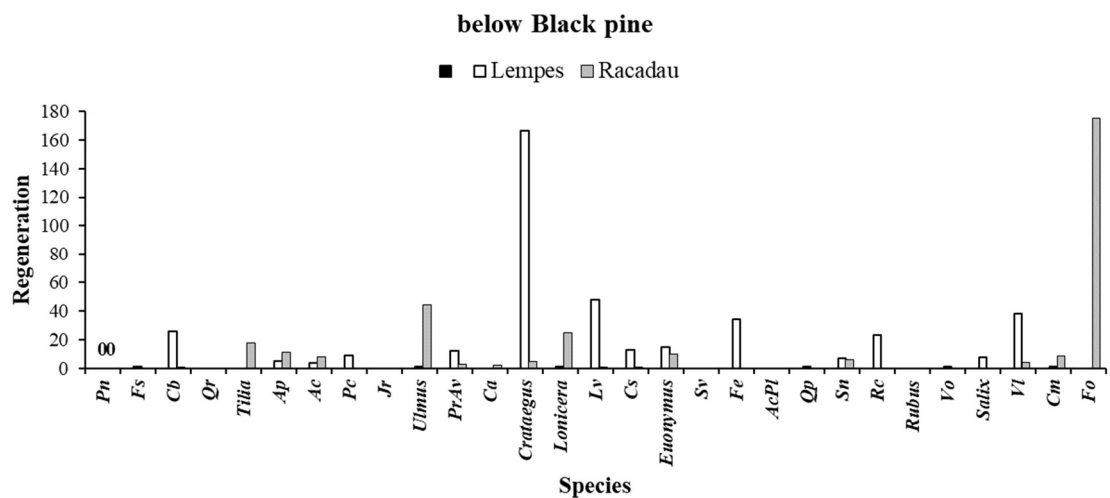
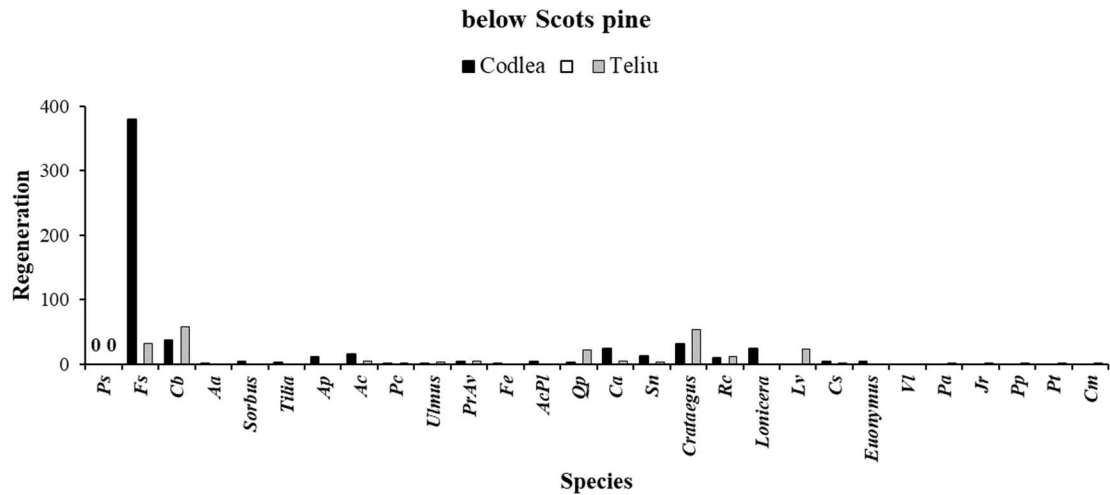


Figure 3 – Regeneration below Scots pine and Black pine. Where, **Aa** = *Abies alba* Mill.; **Fs** = *Fagus sylvatica* L.; **Cb** = *Carpinus betulus* L.; **Pa** = *Picea abies* Karst.; **Sorbus** = *Sorbus* sp. L.; **Ca** = *Corylus avellana* L.; **Sn** = *Sambucus nigra* L.; **Crataegus** = *Crataegus* sp. L.; **Rc** = *Rosa canina* L.; **Lonicera** = *Lonicera* sp. L.; **Pn** = *Pinus nigra* Arn.; **Qr** = *Quercus robur* L.; **Tilia** = *Tilia* sp. L.; **Ap** = *Acer pseudoplatanus* L.; **Ac** = *Acer campestre* L.; **Pc** = *Prunus cerasifera* Ehrh.; **Jr** = *Juglans regia* L.; **Ulmus** = *Ulmus* sp. L.; **PrAv** = *Prunus avium* L.; **Lv** = *Ligustrum vulgare* L.; **Cs** = *Cornus sanguinea* L.; **Euonymus** = *Euonymus* sp. L.; **Sv** = *Syringa vulgaris* L.; **Fe** = *Fraxinus excelsior* L.; **AcPl** = *Acer platanoides* L.; **Qp** = *Quercus petraea* (Matt.) Liebl.; **Rubus** = *Rubus* sp. L.; **Vo** = *Viburnum opulus* L.; **Salix** = *Salix* sp. L.; **VI** = *Viburnum lantana* L.; **Cm** = *Cornus mas* L.; **Fo** = *Fraxinus ornus* L.; **Ps** = *Pinus sylvestris* L.; **Pp** = *Pyrus pyraeaster* L.; **Pt** = *Populus tremula* L. (Hereş et al. under review)

III stage (01/01/2020 – 30/06/2020):

During the third stage, the activities statistical analysis of data and dissemination of results were continued. In this sense, the statistical analysis on deciduous species (i.e., European beech and sessile oak) was completed. More specifically, the growth data obtained from the measurement of annual tree-rings were analyzed. These data were used to see how these two species have grown over time and how they behave when dealing with severe droughts. The results of this study were used to make a master's thesis. This thesis was written by Petrea Ștefan under the guidance of dr. Hereș Ana-Maria (project director of NATIVE) and dr. Petritan Ion Catalin (professor of Transilvania University of Brașov). The results of this study showed that both European beech and sessile oak registered significant positive growth trends. In addition, both species are resilient to severe drought events, although they have different strategies in this regard. Specifically, the European beech is adapted to withstand severe droughts without significantly reducing its growth when dealing with such events (Figure 4). On the other hand, the sessile oak, even if during severe droughts it is forced to significantly reduce its growth, it then manages to return to the level of pre-drought growths, its recovery being achieved in a period of maximum two years after the severe drought events (Figure 4). The results of this study indicate that the two native deciduous species (i.e., European beech and sessile oak) have the ability to successfully cope with drought events, being thus also better adapted to cope with climate change than the non-native species of planted conifers (i.e., Scots pine and Black pine).

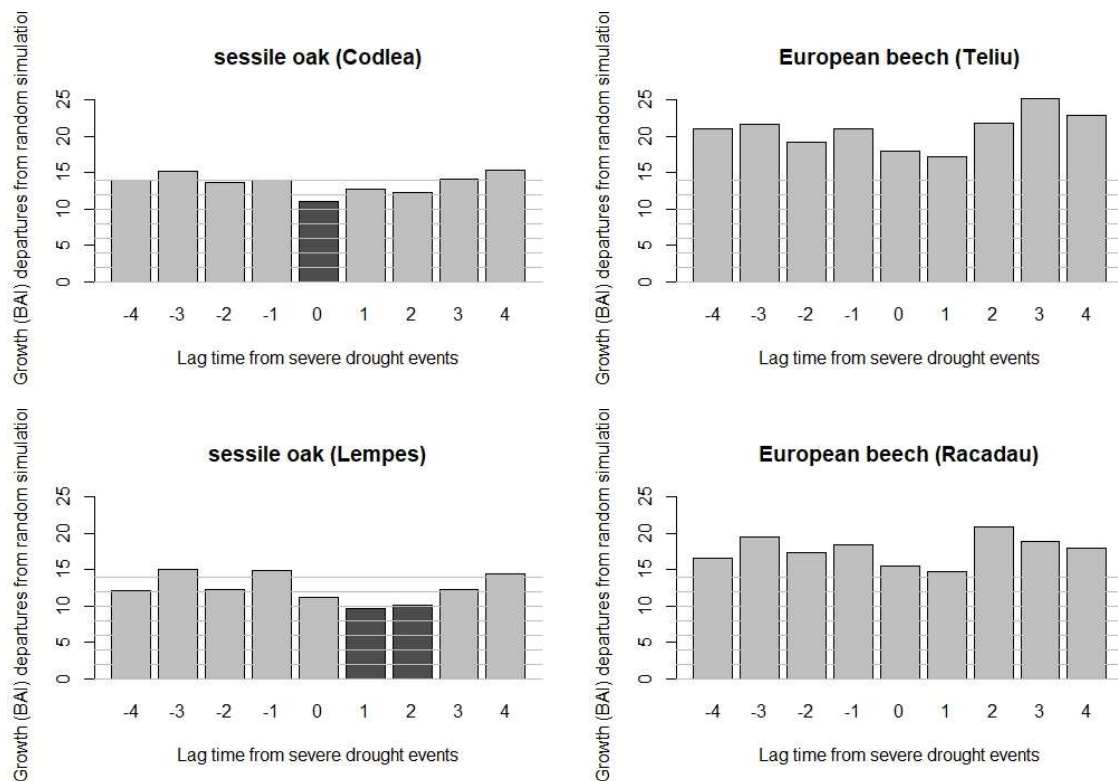


Figure 4 – Results of SEA (Superposed Epoch Analyses) analyses: growth (BAI = basal area increment) departures from the mean considering s series of events: four years before severe drought events (-4, -3, -2, -1), during severe drought events (0; 1976, 1987, 2002, 2012), and after severe drought events (1, 2, 3, 4). Different colours indicate non-significant (grey) or significant (black) growth departures considering random simulations (*figure: Petrea Ștefan; master thesis*)

In parallel with Petrea Ștefan's master's thesis, work began on an article in which the two species of conifers (i.e., Scots pine and Black pine) will be directly compared with the two species of deciduous trees (i.e., European beech and sessile oak) (Hereș et al. *in prep*). This article will analyze the growth (Figure 5) of the four species taking into account severe droughts and the management measures to which these species have been subjected to along time. Preliminary results indicate that deciduous species are indeed more resilient to drought than the two coniferous species. It should be noted that such studies are not common and that management measures, which have contributed to the current structure of forests and are therefore critical to better understand the processes of decline and mortality, are often ignored. The effect of environmental factors such as climate (i.e., severe droughts) on trees and therefore forests must be studied and understood taking into account important aspects, such as management

measures, which can be decisive in terms of how trees respond to stressful conditions. The NATivE project therefore fills a knowledge gap and brings important information about how different species may or may not successfully cope with severe drought events depending on their native or non-native status.

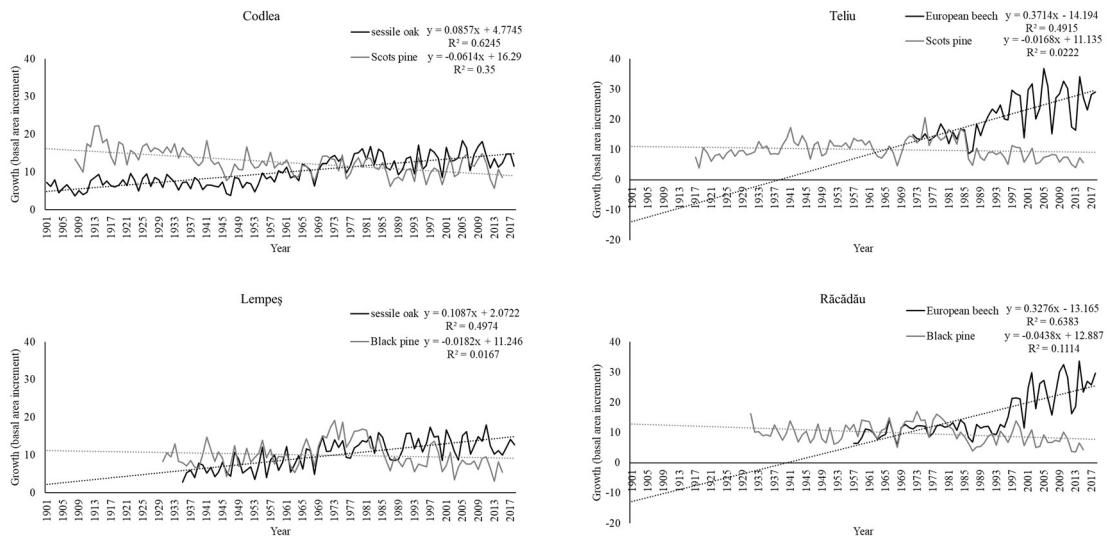


Figure 5 – Growth (basal area increment) trends of the four studied species (Scots pine, Black pine, European beech, and sessile oak) (Hereş et al. *in prep.*)

The results obtained in 2020 should have been presented at the Mixed Forests conference that would have taken place in Lund (Sweden) between 25-27 March 2020. At this conference the project director should have presented a poster entitled "*Mixed forests of allochthonous conifer and native broadleaf species from central Romania (Brasov)*". However, this was not possible due to the situation caused by the COVID-19 virus which affected the whole world.

V. CONCLUSIONS

All the activities proposed within the NATivE project were successfully carried out, according to the established working plan. Different species of non-native (i.e., Scots pine and Black pine) and native (European beech and sessile pine) trees were studied. The results show that severe droughts associated with climate change have the potential to severely affect trees and forests through decline and mortality, but in order to better understand such events, management measures need to be taken into account. The latter factor represents the background of current forest structures. In order to be able to

protect forests and adapt them to climate change, it is essential to take such factors into account in addition to climate.

The obtained results were also disseminated successfully:

ISI articles:

- Curiel Yuste J., Flores-Rentería D., García-Angulo D., **Hereş A.-M.**, Bragă C., Petritan A.-M., Petritan I.C., 2019, *Cascading effects associated with climate-change-induced conifer mortality in mountain temperate forests result in hot-spots of soil CO₂ emissions*. Soil Biology and Biochemistry 133:50-59.
- **Hereş A.-M.**, Polanco-Martínez J.M., Petritan I.C., Petritan A.-M., Curtu A.L., Rigling A., Bigler C., Curiel Yuste J., *under review in Science of the Total Environment, Management legacies determine current responses to severe drought events of conifer species in the Romanian Carpathians*.

International conferences:

- **2019, Hereş A.-M.**, Curiel Yuste J., Curtu A.L., Petritan A.M., Petritan I.C., *How does drought-related mortality affect conifer species? The role of historical management practices on the current response of trees to climate*. European Geosciences Union General Assembly; Vienna, Austria; poster and flash oral presentations
- **2020, Hereş A.-M.**, Petrea Ş., Curtu A.L., Petritan I.C., Curiel Yuste J., *Mixed forests of allochthonous conifer and native broadleaf species from central Romania (Brasov)*. Mixed Forests; Lund, Suedia; poster presentation; conferință anulată în cele din urmă datorită situației generate de către virusul COVID-19 care a afectat întreaga lume.

Master thesis:

- Petrea Ş., 2020, *Native sessile oak and European beech species are able to cope with severe drought events*. Master thesis supervised by dr. Hereş Ana-Maria (NATivE project director) and dr. Petritan Ion Catalin (professor of Transilvania University of Braşov).

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