# Zalihe i promjene zaliha ugljika u neživoj organskoj tvari i tlu hrvatskih šuma

Bitunjac, Doroteja

**Doctoral thesis / Doktorski rad** 

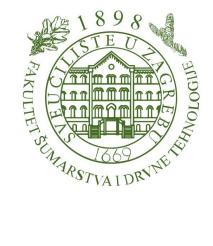
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### FACULTY OF FORESTRY AND WOOD TECHNOLOGY

Doroteja Bitunjac

## STOCKS AND STOCK CHANGES OF CARBON IN DEAD ORGANIC MATTER AND SOIL IN CROATIA'S FORESTS

**DOCTORAL THESIS** 



## Sveučilište u Zagrebu

## FAKULTET ŠUMARSTVA I DRVNE TEHNOLOGIJE

Doroteja Bitunjac

## ZALIHE I PROMJENE ZALIHA UGLJIKA U NEŽIVOJ ORGANSKOJ TVARI I TLU HRVATSKIH ŠUMA

**DOKTORSKI RAD** 

Zagreb, 2024.



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Supervisors: Prof Darko Bakšić, PhD Maša Zorana Ostrogović Sever, PhD

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**DOKTORSKI RAD** 

Mentori:

Prof. dr. sc. Darko Bakšić Dr. sc. Maša Zorana Ostrogović Sever

Zagreb, 2024.

## **BASIC DOCUMENTATION CARD**

TI (Title)	Stocks and stock changes of carbon in dead organic matter and soil in Croatia's
	forests
AU (Author)	Doroteja Bitunjac
AD (Address of Author)	Trg grada Vukovara 5, 10410 Velika Gorica; email: doroteja@sumins.hr
SO (Source)	Library of Faculty of Forestry and Wood Technology, University of Zagreb,
	Svetošimunska cesta 23, 10000 Zagreb; Library of Croatian Forest Research
	Institute, Cvjetno naselje 41, 10450 Jastrebarsko
PY (Publication Year)	2024
LA (Language of text)	English
LS (Language of Summary)	English
DE (Descriptors, key words)	Carbon, dead wood, forest floor, soil organic carbon, national greenhouse gas
	inventory reporting, Biome-BGCMuSo model
GE (Geo Headings)	Republic of Croatia
PT (Publication Type)	Doctoral Thesis
VO (Volume)	I-XIX + 133 pg + 26 tables + 27 figures + 192 references
AB (Abstract)	The Republic of Croatia, as a signatory country of the United Nations
	Framework Convention on Climate Change (UNFCCC), Kyoto Protocol and
	Paris Agreement, has committed to annually report on greenhouse gas (GHG)
	emissions and removals in its National GHG Inventory Report (NIR). In the
	forest ecosystems, carbon stock changes need to be accounted for under the
	Land Use, Land-Use Change and Forestry (LULUCF) sector for five ecosystem carbon pools: above- and below-ground live biomass, dead wood (DW), forest
	floor and soil organic matter (SOM). The estimation of carbon stock changes in
	DW, forest floor and SOM pools is often omitted due to a lack of available
	activity data and carbon stock change factors. The main aim of this research is
	to facilitate the improvement of estimates of carbon stocks in DW and forest
	floor, as well as carbon stocks and carbon stock changes in SOM in forest
	ecosystems in Croatia.
	This research includes the implementation of a field experiment on DW for
	ten forest tree species distributed across three biogeographical regions in
	Croatia; a compilation of a new national dataset on forest floor; a validation of
	the process-based model Biome-BGCMuSo (BBGCMuSo) version 4.0 for
	estimating national soil organic carbon stocks down to 30 cm depth (SOC <sub>30</sub> ),
	and a third measurement of SOC <sub>30</sub> in pedunculate oak chronosequence that will
	be used, with previous two measurements, for validation of the newest model
	version BBGCMuSo v.6.2 for estimating SOC <sub>30</sub> changes.
	The estimate of carbon stock in the DW pool from this study is lower by
	11–27% compared to the value currently used in the Croatian NIR. A newly
	compiled national database on forest floor carbon stocks can facilitate the
	increase of net CO <sub>2</sub> removals for 5% under this carbon pool in the Croatian NIR.
	BBGCMuSo v.4.0 model was shown to be suitable for the estimation of the
	overall mean of SOC <sub>30</sub> for deciduous and coniferous Forest land strata reported in the Croatian NIR, but disaggregation of the results with respect to
	biogeographical region decreased model accuracy. Calibration of the BBGCMuSo model v.6.2 highlights the importance of using different temporal
	resolution datasets in calibration of process-based models. In the pedunculate
	oak forest, represented by the chronosequence experiment, although there is no
	disagreement in trends between the measured and modelled SOC <sub>30</sub> , the trends
	were divergent (negative for measured and positive for modelled $SOC_{30}$ ). The
	obtained results have the potential to contribute to the improvement of
	calculations in Croatian NIR.
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## TEMELJNA DOKUMENTACIJSKA KARTICA

	T =
TI (naslov)	Zalihe i promjene zaliha ugljika u neživoj organskoj tvari i tlu hrvatskih šuma
OT (izvorni naslov)	Stocks and stock changes of carbon in dead organic matter and soil in Croatia's
	forests
AU (autor)	Doroteja Bitunjac
AD (adresa)	Trg grada Vukovara 5, 10410 Velika Gorica; email: doroteja@sumins.hr
SO (izvor)	Knjižnica Fakulteta šumarstva i drvne tehnologije Sveučilišta u Zagrebu,
	Svetošimunska cesta 23, 10000 Zagreb; Knjižnica Hrvatskog šumarskog
DV/ I' I'	instituta, Cvjetno naselje 41, 10450 Jastrebarsko
PY (godina objave)	2024.
LA (izvorni jezik)	Engleski
LS (jezik sažetka)	Engleski
DE (ključne riječi)	Ugljik, mrtvo drvo, šumska prostirka, organski ugljik u mineralnom dijelu tla,
	Nacionalno izvješće o inventaru stakleničkih plinova, Biome-BGCMuSo
GE ( 1: 1: )	model
GE (zemlja objave)	Republika Hrvatska
PT (vrsta objave)	Doktorski rad
VO (obujam)	I-XIX + 133 str. + 26 tablica + 27 slika + 192 citirane literature
AB (sažetak)	Republika Hrvatska potpisnica je Okvirne konvencije Ujedinjenih naroda
	o promjeni klime, Kyotskog protokola i Pariškog sporazuma prema kojima se
	obvezala jednom godišnje izraditi i dostaviti Nacionalno izvješće o inventaru
	stakleničkih plinova (engl. National Greenhouse Gas Inventory Report, NIR).
	Promjene zaliha ugljika u šumskim ekosustavima obračunavaju se u sektoru
	"Korištenje zemljišta, prenamjena zemljišta i šumarstvo" (engl. Land Use,
	Land-Use Change and Forestry, LULUCF) za pet pohraništa ugljika:
	nadzemna i podzemna živa biomasa, mrtvo drvo, šumska prostirka i organska
	tvar tla. Procjena promjena zaliha ugljika u mrtvom drvu, šumskoj prostirci i
	tlu često je izostavljena zbog nedostatka dostupnih podataka i faktora promjene
	zaliha ugljika. Glavni cilj ovog istraživanja je omogućiti unaprjeđenje procjene
	zaliha u mrtvom drvu i šumskoj prostirci te procjene zaliha i promjena zaliha
	ugljika u mineralnom dijelu tla u šumama Republike Hrvatske.
	Ovo istraživanje uključuje postavljanje terenskog istraživanja za mrtvo
	drvo za deset vrsta drveća u tri biogeografske regije u Republici Hrvatskoj;
	sastavljanje nove nacionalne baze podataka o šumskoj prostirci; validaciju
	procesnog modela Biome-BGCMuSo (BBGCMuSo) verzija 4.0 za procjenu
	nacionalnih zaliha ugljika u mineralnom dijelu tla do dubine od $30 \text{ cm (SOC}_{30)}$ ,
	i treću izmjeru SOC <sub>30</sub> -a u pokusu kronosekvence hrasta lužnjaka, koja je
	zajedno sa dvije prethodne izmjere, poslužila za validaciju novije verzije
	modela BBGCMuSo v.6.2 za procjenu promjena zaliha SOC <sub>30</sub> -a.
	Zaliha ugljika u mrtvom drvu u Republici Hrvatskoj, procijenjena u ovom
	radu, manja je za 11–27 % u odnosu na vrijednost koja se trenutno koristi u
	NIR-u Republike Hrvatske. Korištenje nove nacionalne baza podataka o
	zalihama ugljika u šumskoj prostirci može povećati odlive CO <sub>2</sub> iz ovog
	pohraništa ugljika za 5 % u odnosu na vrijednost koja se trenutno koristi u
	NIR-u Republike Hrvatske. Model BBGCMuSo može uspješno reproducirati
	prosječne zalihe SOC <sub>30</sub> -a za stratume šumskog zemljišta koji se koriste u NIR
	u Republike Hrvatske, šume listača i šume četinjača, ali pri detaljnijoj
	stratifikaciji rezultata na razinu stratuma šumskog zemljišta × biogeografska
	regija smanjena je točnost modela. Kalibracija BBGCMuSo modela v.6.2
	naglašava važnost korištenja skupova podataka različite vremenske rezolucije
	u kalibraciji procesnih modela. U šumi hrasta lužnjaka, predstavljenoj
	pokusom kronosekvence, trendovi izmjerenog i modeliranog SOC <sub>30</sub> -a, iako
	statistički neznačajni, suprotnog su smjera (negativan za izmjereni i pozitivan
	za modelirani SOC <sub>30</sub> ). Dobiveni rezultati potencijalno mogu pridonijeti
	unaprjeđenju izračuna u NIR-u Republike Hrvatske.

#### **INFORMATION ABOUT MENTORS**

The thesis was made under the supervision of Prof Dr Darko Bakšić, and Dr Maša Zorana Ostrogović Sever, as part of the doctoral study of Forestry and Wood Technology at the Institute of Ecology and Silviculture, Faculty of Forestry and Wood Technology, University of Zagreb.

Prof Dr Bakšić was born on the 21<sup>st</sup> of October 1971 in Banská Štiavnica, Slovakia. In 1997 he graduated from the Faculty of Forestry, University of Zagreb. In the same year, he was employed as a junior assistant at the Institute of Ecology and Silviculture, Faculty of Forestry, University of Zagreb. In 2006, he defended his PhD thesis under the title "Pedophysiographic relationships in forest communities of sessile oak (Quercus petraea (Matt.) Liebl.) and common beech (Fagus sylvatica L.) on Mt. Bilogora at the Faculty of Forestry, University of Zagreb. Since 2018 he has been a full professor at the Institute of Ecology and Silviculture, Faculty of Forestry (now Faculty of Forestry and Wood Technology), University of Zagreb. In his scientific work, he is interested in forest soil chemical and physical characteristics, genesis, management and protection, forest floor accumulation, carbon cycle and nutrients, and its potential as a fuel and carbon source in forest fires. As a professor at the Faculty of Forestry and Wood Technology, he holds classes in the subjects "Soil science" in two undergraduate studies, "Forest ecosystem soil management", "Soil science", and "Remediation of degraded land" in graduate studies and "Soil classification systems", "Soil colloids and chemistry", and "Soil organic matter and carbon cycle" at the doctoral studies. In his scientific research, he published, as the first author or co-author, more than 50 scientific papers in the field of forestry, of which 30 papers are in the a1 category of scientific papers, one university classbook, one university manual, two scientific books and chapters in five scientific books. Furthermore, he has been involved in 15 national scientific and professional projects, and two projects financed by the EU. He is a member of the Editorial Board of two scientific journals, Šumarski list (since 2019) and Croatian Journal of Forest Engineering (since 2020).

Dr Ostrogović Sever was born on the 24<sup>th</sup> of July 1982 in Zagreb, Croatia. In 2007 she graduated from the Faculty of Forestry, University of Zagreb. In the same year, she was employed as a research assistant at the Croatian Forest Research Institute, Division for Forest Management and Forestry Economics. In 2013, she defended her PhD thesis titled "Carbon stocks and carbon balance of an even-aged pedunculate oak (*Quercus robur* L.) forest in Kupa river basin" at the Faculty of Forestry, University of Zagreb. Since 2022 she has been a Senior Scientific Associate at the Croatian Forest Research Institute. In her scientific research, she investigates carbon

stocks and carbon stock changes in forest ecosystems and models forest ecosystems using the process-based model Biome-BGCMuSo. By now, she published, as the first author or co-author, 20 scientific papers, a chapter in one scientific book, and nine professional elaborates. She has been involved in 24 national and international scientific and professional projects. Since 2020, she has been a co-convener and co-chairperson at the European Geoscience Union conference (Vienna, Austria), organising sessions on the topics of estimation of carbon stocks and fluxes in the ecosystems, as a support for the National GHG Inventory Reports under the UNFCCC.

#### PREFACE AND ACKNOWLEDGMENTS

This thesis was written within the framework of the project "Modelling Forest Carbon Stocks, Fluxes and Forest Risks under Future Climate Scenarios – MODFLUX", funded by the Croatian Science Foundation (HRZZ IP-2019-04-6325). Dr Hrvoje Marjanović from the Division for Forest Management and Forestry Economics, Croatian Forest Research Institute (CFRI), was the project leader.

The thesis includes research objectives of a wider range and involves forest ecosystem modelling using a process-based model, which is still a novel method in Croatia. Therefore, it was a logical choice to have two mentors who would, with their expertise, complement all research objectives.

Firstly, I would like to greatly thank my mentor and a dear colleague, Dr Maša Zorana Ostrogović Sever for her unconditional support, great transfer of knowledge, priceless guidance, and for encouraging me to always grow as a researcher. I would also like to thank my mentor Prof Dr Darko Bakšić for his support, sharing knowledge and many advices during the research and writing of this thesis.

Furthermore, I would like to thank the MODFLUX project leader and a dear colleague, Dr Hrvoje Marjanović, for numerous scientific lessons and discussions, great statistical support and his help in overcoming obstacles when needed.

Also, great thanks to Dr Katarína Merganičová from the Institute of Landscape Ecology, Slovak Academy of Sciences (Slovakia) and Faculty of Forestry and Wood Sciences, Czech University of Life Sciences Prague (the Czech Republic) for hosting me in Zvolen, Slovakia, and generously guiding me on forest ecosystem modelling and for reviewing the modelling part of this thesis.

Moreover, I would like to thank the Scientific Council of CFRI, Dr Sanja Perić, Director General of the CFRI, and Dr Ivan Balenović, head of the Division for Forest Management and Forestry Economics, for enabling me to participate in many scientific conferences, trainings and schools, abroad and in Croatia. Also, I would like to thank my colleagues at the Division for Forest Management and Forestry Economics for always being positive and supportive.

Many thanks go to my colleagues Nikolina Milanović and Goran Tijan for their assistance in the challenging field sampling and to my colleague Monika Hlebić for undertaking numerous laboratory analyses. I would also like to thank my colleague Mia Marušić, for her emotional support, advices and motivation while writing this thesis.

And finally, my deepest thank you goes to you, my family and friends, for your comfort, understanding and unconditional support. To H, thank you for being my pillar. To V, thank you for always believing in me. To Lj, thank you for your warmth and smiles. To G, thank you for love.

I devote this thesis to all PhD students. Your research matters.

Doroteja

#### **ABSTRACT**

The Republic of Croatia, as a signatory country of the United Nations Framework Convention on Climate Change (UNFCCC), Kyoto Protocol and Paris Agreement, has committed to annually report on greenhouse gas (GHG) emissions and removals in its National GHG Inventory Report (NIR). In the forest ecosystems, carbon stock changes need to be accounted for under the Land Use, Land-Use Change and Forestry (LULUCF) sector for five ecosystem carbon pools: above- and below-ground live biomass, dead wood (DW), forest floor and soil organic matter (SOM). The estimation of carbon stock changes in DW, forest floor and SOM pools is often omitted due to a lack of available activity data and carbon stock change factors. The main aim of this research is to facilitate the improvement of estimates of carbon stocks in DW and forest floor, as well as carbon stocks and carbon stock changes in SOM in forest ecosystems in Croatia.

This research includes the implementation of a field experiment on DW for ten forest tree species distributed across three biogeographical regions in Croatia; a compilation of a new national dataset on forest floor; a validation of the process-based model Biome-BGCMuSo (BBGCMuSo) version 4.0 for estimating national soil organic carbon stocks down to 30 cm depth (SOC<sub>30</sub>), and a third measurement of SOC<sub>30</sub> in pedunculate oak chronosequence that will be used, with previous two measurements, for validation of the newest model version BBGCMuSo v.6.2 for estimating SOC<sub>30</sub> changes.

The estimate of carbon stock in the DW pool from this study is lower by 11–27% compared to the value currently used in the Croatian NIR. A newly compiled national database on forest floor carbon stocks can facilitate the increase of net CO<sub>2</sub> removals for 5% under this carbon pool in the Croatian NIR. BBGCMuSo v.4.0 model was shown to be suitable for the estimation of the overall mean of SOC<sub>30</sub> for deciduous and coniferous Forest land strata reported in the Croatian NIR, but disaggregation of the results with respect to biogeographical region decreased model accuracy. Calibration of the BBGCMuSo model v.6.2 highlights the importance of using different temporal resolution datasets in calibration of process-based models. In the pedunculate oak forest, represented by the chronosequence experiment, although there is no disagreement in trends between the measured and modelled SOC<sub>30</sub>, the trends were divergent (negative for measured and positive for modelled SOC<sub>30</sub>). The obtained results have the potential to contribute to the improvement of calculations in Croatian NIR.

Keywords: carbon, dead wood, forest floor, soil organic carbon, national greenhouse gas inventory reporting, Biome-BGCMuSo model

#### **EXTENDED ABSTRACT (IN CROATIAN)**

#### Zalihe i promjene zaliha ugljika u neživoj organskoj tvari i tlu hrvatskih šuma

Republika Hrvatska (RH) potpisnica je Okvirne konvencije Ujedinjenih naroda o promjeni klime (engl. United Nations Framework Convention on Climate Change, UNFCCC), Kyotskog protokola i Pariškog sporazuma prema kojima se obvezala jednom godišnje izraditi i dostaviti Nacionalno izvješće o inventaru stakleničkih plinova (engl. National Greenhouse Gas *Inventory Report*, NIR). Šumsko zemljište jedno je od šest kategorija zemljišta u NIR sektoru "Korištenje zemljišta, prenamjena zemljišta i šumarstvo" (engl. Land Use, Land-Use Change and Forestry, LULUCF) za koje se obračunavaju emisije i odlivi stakleničkih plinova (IPCC 2006). Svaka kategorija zemljišta podijeljena je u podkategorije "Zemljište koje ostaje u istoj kategoriji zemljišta" i "Zemljište koje je pretvoreno u drugu kategoriju zemljišta". Slijedno tome, Šumsko zemljište podijeljeno je u podkategorije: "Zemljište pretvoreno u šumsko zemljište" i "Šumsko zemljište koje ostaje šumsko". Za navedene podkategorije zemljišta emisije i odlivi ugljikovog dioksida (CO<sub>2</sub>) obračunavaju se za pet pohraništa ugljika: nadzemna i podzemna živa biomasa, mrtvo drvo i šumska prostirka (pod zajedničkim nazivom neživa organska tvar) i organska tvar tla (IPCC 2006). Organski ugljik u tlu (engl. Soil Organic Carbon, SOC) odnosi se na ugljik koji je organskog podrijetla, a nalazi se u mineralnom dijelu tla. Kontinuirana praćenja pohraništa ugljika u šumskom ekosustavu najčešće su dostupna iz nacionalnih inventura šuma (Tomppo i sur. 2010), pogotovo za živu biomasu koja je ekonomski najvažnije pohranište ugljika. S druge strane, važnost pohraništa ugljika mrtvo drvo, šumska prostirka i SOC temeljila se uglavnom na njihovoj ekološkoj ulozi (Olson 1963, Martinović 1973, Harmon i sur. 1986) te su se ta pohraništa ugljika pretežno pratila za lokacije od posebnog interesa (Hauβmann i Fischer 2004). Međutim, zahtjevi UNFCCC-a za što točnijim obračunom emisija/odliva stakleničkih plinova iz/u mrtvo drvo, šumsku prostirku i SOC potiču subjekte koji izrađuju NIR i znanstvenu zajednicu na kontinuirano praćenje i istraživanje ovih pohraništa ugljika. Ova disertacija bavi se pitanjima relevantnim za procjenu zaliha i promjena zaliha ugljika u mrtvom drvu, šumskoj prostirci i SOC-u u RH, a čije rješavanje može unaprijediti nacionalnu procjenu emisija/odliva ugljika u LULUCF sektoru NIR-a.

U NIR-u RH u podkategoriji Šumskog zemljišta "Zemljište pretvoreno u šumsko zemljište", izvješćuje se o promjenama zaliha ugljika u pohraništima mrtvo drvo, šumska prostirka i SOC koristeći pristup 2 (engl. *Tier* 2, HR NIR 2023). Za izračun zaliha ugljika u mrtvom drvu koristi se volumen mrtvog drva iz prve nacionalne inventure šuma (Čavlović 2010) i faktori konverzije

volumena drva, gustoća svježeg drva i sadržaja ugljika u drvu od 50 % (HR NIR 2023). Međutim, poznato je da se faktori konverzije volumena drva mijenjaju po stupnjevima raspadanja mrtvog drva (Harmon i sur. 2008, Sandström i sur. 2007). Razumno je za očekivati da bi upotreba faktora konverzije volumena mrtvog drva prema stupnjevima raspadanja dala niže vrijednosti zaliha ugljika u mrtvom drvu, u odnosu na izračun u kojem se koriste vrijednosti faktora za svježe drvo (Merganičová i Merganič 2010). Međutim, nacionalne vrijednosti za gustoću i sadržaj ugljika u mrtvom drvu prema stupnjevima raspadanja nedostaju. Uzimajući navedeno u obzir, a kako bi se unaprijedila procjena nacionalnih zaliha ugljika u mrtvom drvu, potrebni su nacionalni faktori konverzije volumena mrtvog drva po stupnjevima raspadanja. Nadalje, za izračun promjena zaliha ugljika u šumskoj prostirci koriste se podaci iz baze nacionalne inventure tla (Miko i sur. 2017). U navedenoj bazi o šumskoj prostirci mediteranska biogeografska regija (BGR) podzastupljena je u odnosu na kontinentalnu i alpinsku BGR. S obzirom da nacionalne studije pokazuju da zalihe ugljika u šumskoj prostirci u mediteranskoj BGR mogu imati značajno veće vrijednosti od trenutnog nacionalnog prosjeka (Bakšić i Bakšić 2017, Bakšić i Bakšić 2023), za očekivati je da su zalihe ugljika u ovom pohraništu podcijenjene. Za unaprjeđenje procjene zaliha ugljika u ovom pohraništu, potrebna je reprezentativnija nacionalna baza podataka o zalihama ugljika u šumskoj prostirci.

U podkategoriji Šumskog zemljišta "Šumsko zemljište koje ostaje šumsko", RH u svom NIR-u koristi pristup 1 za obračun zaliha ugljika u pohraništima mrtvo drvo, šumska prostirka i SOC, odnosno pretpostavlja se da su zalihe ugljika u ovim pohraništima stabilne (HR NIR 2023). Međutim, SOC i njegova dinamika potencijalno bi mogli biti izmijenjeni uslijed klimatskih promjena (Boisvenue i Running 2006). Jedan od načina predviđanja razvoja SOC-a u različitim klimatskim uvjetima je primjenom procesnih modela. U RH je trenutno operativan jedan procesni model koji može simulirati zalihe i promjene zaliha ugljika u ovom pohraništu, Biome-BGCMuSo (Hidy i sur. 2016a, 2022).

Glavni cilj ovog istraživanja je omogućiti unaprjeđenje procjene zaliha u mrtvom drvu i šumskoj prostirci te procjene zaliha i promjena zaliha ugljika u mineralnom dijelu tla (SOC) u šumama RH. Definirana su tri specifična cilja istraživanja (C) i četiri hipoteze (H):

C1) pružiti nacionalne konverzijske faktore volumena mrtvog drva po stupnjevima raspadanja za korištenje u izračunu zaliha ugljika u mrtvom drvu; H1: faktori konverzije volumena mrtvog drva za određene grupe drveća razlikuju se između biogeografskih regija,

- C2) pružiti reprezentativniju nacionalnu vrijednost zaliha ugljika u šumskoj prostirci; H2: zalihe ugljika u šumskoj prostirci za određene grupe drveća razlikuju se između biogeografskih regija,
- C3) istražiti primjenjivost procesnog modela Biome-BGCMuSo za procjenu zaliha i promjena zaliha ugljika u organskoj tvari tla; H3: mogućnost modela Biome-BGCMuSo da reproducira zalihe organskog ugljika u mineralnom dijelu tlu ograničena je veličinom stratuma; H4: model Biome-BGCMuSo može reproducirati izmjerene promjene zaliha organskog ugljika u mineralnom dijelu tla u šumi hrasta lužnjaka u razdoblju 2012. 2022.

Ovo istraživanje provedeno je kroz tri studije adresirajući sljedeća pohraništa ugljika: mrtvo drvo, šumska prostirka i SOC. Kako bi se testirala prva hipoteza, proveden je eksperiment za deset vrsta drveća u tri BGR: hrast lužnjak, crnu johu, obični grab i poljski jasen u kontinentalnoj BGR; običnu bukvu, običnu jelu i običnu smreku u alpinskoj BGR; crni bor, primorski bor i hrast crniku u mediteranskoj BGR. Za klasifikaciju mrtvog drva u stupnjeve raspadanja korišten je klasifikacijski sustav s četiri stupnja raspadanja drva. Ovaj klasifikacijski sustav odgovara prva četiri stupnja raspadanja drva u često korištenom klasifikacijskom sustavu s pet stupnja raspadanja drva prema Harmonu i sur. (1995). Za svaku vrstu drveća i svaki stupanj raspadanja (1 – 4) prikupljena su po tri uzorka ležećeg mrtvog drva u tri debljinska razreda (5 – 10, 10 – 20, i 20 – 30 cm). Ukupno je prikupljeno i analizirano 360 uzorka mrtvog drva (3 × 10 vrsta drveća × 3 debljinska razreda × 4 stupnja raspadanja drva). Volumen uzoraka mrtvog drva utvrđen je primjenom Arhimedovog zakona na temelju težine istisnute vode (Hughes 2005), zatim su uzorci prosušeni, izvagani i njihovim poduzorcima analiziran je sadržaj ugljika. Svakom uzorku mrtvog drva utvrđena je gustoća drva i gustoća ugljika, kao umnožak gustoće drva i sadržaja ugljika. Faktori konverzije volumena mrtvog drva, dobiveni iz ove studije korišteni su, zajedno s podacima o volumenu mrtvog drva iz prve nacionalne inventure šuma (Čavlović 2010) za izračun zaliha ugljika u mrtvom drvu u RH prema stratifikaciji šumskog zemljišta u RH NIR-u (šume listača, šume četinjača i šume makija i šikara). Nacionalna zaliha ugljika u mrtvom drvu izračunata u ovoj studiji uspoređena je sa zalihom ugljika u mrtvom drvu koja se trenutno izvješćuje u NIR-u (HR NIR 2023).

Za testiranje druge hipoteze, dostupni podaci o zalihama ugljika u šumskoj prostirci objedinjeni su iz dva nacionalna projekta u jedinstvenu bazu *Forest ecosystem database* (ForecoDB). ForecoDB uključuje podatke o šumskoj prostirci, mineralnom dijelu tla te sastojinskim i ekološkim varijablama za 274 plohe raspoređene u tri BGR RH. Iz ForecoDB analizirane su razlike u zalihama ugljika u šumskoj prostirci za grupe vrsta drveća (listače i četinjače) između

BGR i razlike između navedenih grupa drveća unutar svake BGR. Također su utvrđene linearne regresije zaliha ugljika u šumskoj prostirci i ekološkim varijablama (nadmorska visina, srednja godišnja temperatura zraka i srednja godišnja količina oborina). Na kraju, izračunata je nova procjena nacionalnih zaliha ugljika u šumskoj prostirci kao težinski prosjek s obzirom na udio šumske površine po BGR u ukupnoj površini RH.

Za testiranje treće i četvrte hipoteze korišten je procesni model Biome-BGCMuSo (BBGCMuSo, Hidy i sur. 2016a, 2022) i provedeno je terensko uzorkovanje mineralnog dijela tla do dubine od 40 cm u pokusu kronosekvence u g.j. Jastrebarski lugovi (Ostrogović Sever i sur. 2019). Model BBGCMuSo simulira tokove i zalihe ugljika, dušika i vode u sustavu tlobiljka-atmosfera. Simulacija procesa je na dnevnoj rezoluciji, a model se pokreće meteorološkim varijablama, ekofiziološkim parametrima sastojine i stanišnim karakteristikama. Rad s modelom uključivao je sljedeće faze: 1) validacija BBGCMuSo modela vezija 4.0 za procjenu zaliha SOC-a u mineralnom dijelu tla do dubine od 30 cm u tri BGR RH, 2) analiza osjetljivosti i kalibracija BBGCMuSo modela verzija 6.2 za šume hrasta lužnjaka, i 3) validacija kalibriranog modela BBGCMuSo verzija 6.2 za procjenu promjene zaliha ugljika u mineralnom dijelu tla do dubine od 30 cm u šumi hrasta lužnjaka.

Za validaciju BBGCMuSo modela verzija 4.0 modeliran je SOC do dubine od 30 cm (SOC<sub>30</sub>) za različite tipove šuma (hrast, bukva, jela/smreka i bor) na 243 plohe iz ForecoDB baze podataka raspoređene u tri BGR RH. Za usporedbu modeliranih i izmjerenih podataka, šume su grupirane prema stratifikaciji šumskog zemljišta u NIR-u RH (šume listača i šume četinjača). Za testiranje treće hipoteze ovog istraživanja, provedena je evaluacija modela za procjenu SOC<sub>30</sub> na temelju usporedbe rezultata modela s izmjerenim podacima tri razine: stratum šumskog zemljišta, stratum šumskog zemljišta × BGR i ploha.

Nadalje, analizom osjetljivosti varijabli modela BBGCMuSo verzija 6.2 ispitan je utjecaj ekofizioloških parametara modela na sljedeće izlazne varijable modela za šumu hrasta lužnjaka:
izmjena CO<sub>2</sub> između atmosfere i ekosustava (engl. *Net Ecosystem Exchange*, NEE), zaliha
ugljika u nadzemnoj živoj biomasi, šumskoj prostirci i SOC<sub>30</sub>. Kalibracija modela temeljila se
na GLUE metodi (engl. *Generalized Likelihood Uncertainity Method*, Beven i Benley 2014) uz
post-procesiranje podataka o izlaznim varijablama modela za 10,000 *Monte Carlo* iteracija. Za
usporedbu modeliranih rezultata s izmjerenim podacima korišteni su: 1) podaci o tokovima
ugljika (dnevni i kulumativni NEE) s mjerne stanice za praćenje tokova CO<sub>2</sub> (engl. *eddy covariance*) u šumi hrasta lužnjaka u g.j. Jastrebarski lugovi (Anić i sur. 2018) i 2) podaci o
zalihama ugljika (nadzemna živa biomasa (Anić i sur. 2018), šumska prostirka i SOC<sub>30</sub>)
izmjerene na trajnim pokusnim plohama koje se nalaze na području pokrivanja mjerne stanice

(engl. *footprint*). Optimizacija parametara provedena je za tri različita skupa varijabli različitih frekvencija izmjere: tokovi ugljika, zalihe ugljika, i tokovi i zalihe ugljika zajedno.

Validacija modela BBGCMuSo verzija 6.2 provedena je usporedbom modeliranog i izmjerenog SOC<sub>30</sub> i promjena SOC<sub>30</sub> za šest odabranih sastojina iz kronosekvence hrasta lužnjaka u g.j. Jastrebarski lugovi (Ostrogović 2013). Za testiranje četvrte hipoteze ovog istraživanja izračunat je trend modeliranog i izmjerenog SOC<sub>30</sub> za kratkoročno razdoblje 2012. – 2022., a naknadno se analizirao i trend za dugoročno razdoblje (ophodnja šume hrasta lužnjaka). Validaciji modela prethodilo je uzorkovanje mineralnog dijela tla do dubine od 40 cm, koje je ujedno i treće uzorkovanje tla u pokusu kronosekvence (godine 2012., 2017. i 2022.). Tlo je uzorkovano sondom za uzorkovanje tla (Eijkelkamp, Giesbeek, Nizozemska). Uzorci tla podijeljeni su u četiri geometrijska horizonta: 0 – 5, 5 – 10, 10 – 20 i 20 – 40 cm. Ukupno je prikupljeno 544 uzoraka tla u osam sastojina pokusa kronosekvence. Iz uzoraka tla izdvojio se sitni i krupni korijen, a uzorci mineralnog sloja tla su prosušeni i vagani te je iz kompozitnih uzoraka utvrđen sadržaj ugljika. Zalihe ugljika izračunate su do dubine od 30 cm kako bi bile usporedive s modeliranim zalihama ugljika do iste dubine tla.

Zaliha ugljika u mrtvom drvu u RH, procijenjena korištenjem faktora dobivenih u ovom radu, manja je za 26,6 %, 16,8 % i 11,1 % za šume listača, šume četinjača i šume šikara i makija, redom, u usporedbi s vrijednošću koja se trenutno koristi u RH NIR-u, a koja je procijenjena primjenom gustoće svježeg drva i sadržaja ugljika u drvu od 50 %. Ovaj rezultat naglašava važnost korištenja nacionalnih faktora konverzije volumena mrtvog drva prema stupnjevima raspadanja. Gustoća mrtvog drva pokazala je očekivani opadajući trend s porastom stupnja raspadanja mrtvog drva za većinu vrsta drveća i u prosjeku za obje grupe drveća, listače i četinjače. S druge strane, sadržaj ugljika u mrtvom drvu nije pokazao značajan trend sa stupnjem raspadanja mrtvog drva za većinu vrsta drveća i u prosjeku za listače, dok je za običnu smreku i u prosjeku za četinjače uočen pozitivan trend sadržaja ugljika sa porastom stupnja raspadanja mrtvog drva. Detaljnija stratifikacija rezultata na razinu BGR otkrila je dodatne razlike u gustoći mrtvog drva i sadržaju ugljika unutar pojedine grupe vrsta drveća, čime je potvrđena prva hipoteza ovog istraživanja. Utvrđena je značajno veća gustoća mrtvog drva u mediteranskoj BGR u odnosu na druge BGR u 1. i 2. stupnju raspadanja kod listača i u stupnjevima raspadanja 2 – 4 kod četinjača. Sadržaj ugljika u mrtvom drvu kod listača je značajno viši u alpinskoj BGR u odnosu na druge BGR za većinu stupnjeva raspadanja, dok kod četinjača nisu uočene značajne razlike u sadržaju ugljika između BGR.

Zalihe ugljika u šumskoj prostirci razlikuju se unutar pojedine grupe vrste drveća (listače i četinjače) između BGR, čime je potvrđena druga hipoteza ovog istraživanja. Zalihe ugljika u šumskoj prostirci listača značajno su više u alpinskoj BGR u odnosu na kontinentalnu BGR, dok su kod četinjača značajno veće zalihe ugljika u šumskoj prostirci u mediteranskoj BGR u odnosu na alpinsku BGR. Osim navedenog, u mediteranskoj BGR zalihe ugljika u šumskoj prostirci značajno su veće kod četinjača u odnosu na listače. Nova procjena zaliha ugljika u šumskoj prostirci za područje RH iznosi 4,81 t C ha<sup>-1</sup>, što je za ~5 % više u odnosu na vrijednost od 4,57 t C ha<sup>-1</sup> koja se trenutno koristi u NIR-u RH (HR NIR 2023).

Prosječne zalihe ugljika u mineralnom dijelu tla do dubine 30 cm (SOC<sub>30</sub>) za oba stratuma šumskog zemljišta, šume listača i šume četinjača, mogu se uspješno modelirati na razini države procesnim modelom BBGCMuSo (verzija 4.0), premda sa smanjenom varijabilnošću podataka u odnosu na izmjerene podatke. Pri detaljnijoj stratifikaciji rezultata na razinu stratum šumskog zemljišta × BGR, model je pokazao da i dalje dobro reproducira zalihe ugljika u ovom pohraništu ugljika, premda sa smanjenom točnošću. Također, podjelom rezultata s obzirom na BGR, uočena je tendencija modela da podcjenjuje SOC<sub>30</sub> u šumi listača u mediteranskoj BGR i precjenjuje vrijednosti u šumi četinjača u Alpinskoj BGR. Navedeno ukazuje na potrebu za daljnjim razvojem logike modela, kao i dodatnom kalibracijom parametara modela s obzirom na vrstu drveća i BGR. Na razini plohe, za oba stratuma šumskog zemljišta nije pronađena korelacija između modeliranog i izmjerenog SOC<sub>30</sub>. Dakle, stratifikacijom rezultata na niže razine (stratum šumskog zemljišta > stratum šumskog zemljišta × BGR > ploha) povećava se neslaganje modeliranih i izmjerenih zaliha ugljika u SOC<sub>30</sub>, čime je potvrđena treća hipoteza ovog istraživanja.

Analizom osjetljivosti odabranih izlaznih varijabli modela BBGCMuSo (verzija 6.2), otkriveni su sljedeći utjecajni parametri: fiksacija dušika (Nfix), potrošnja ugljika staničnim disanjem po jedinici dušika (MRperN), specifična lisna površina (SLA), omjer utrošenog ugljika i pohranjenog ugljika kroz proces rasta (GRC), teoretski maksimum omjera nestrukturiranih i strukturiranih ugljikohidrata (NSCvsSCmax) i maksimalna stomatalna provodljivost (MSC). U kalibraciji modela BBGCMuSo (verzija 6.2) vrijednosti optimiziranih parametara ovise o tipu podataka koji se koriste. Naime, optimizirane vrijednosti parametara dobivene korištenjem podataka o zalihama ugljika različite su od optimiziranih vrijednosti parametara dobivenih kada su u kalibraciji korišteni podaci o tokovima ugljika ili oboje. Ovo naglašava važnost korištenja skupova podataka različite vremenske rezolucije u kalibraciji procesnih modela.

Modeliran i izmjeren SOC<sub>30</sub> nije pokazao značajan trend tijekom istraživanog razdoblja od 2012. do 2022. godine u šumi hrasta lužnjaka. Međutim, iako statistički neznačajni, trendovi su

imali suprotan smjer, odnosno smanjenje izmjerenog SOC $_{30}$  za -0,474 t C ha $^{-1}$  godina $^{-1}$  (R $^2$  = 0,02, p = 0,61) i porast modeliranog SOC $_{30}$  od 0,146 t C ha $^{-1}$  godina $^{-1}$  (R $^2$  = 0,02, p = 0,57).

Ključne riječi: ugljik, mrtvo drvo, šumska prostirka, organski ugljik u mineralnom dijelu tla, Nacionalno izvješće o inventaru stakleničkih plinova, Biome-BGCMuSo model

#### LIST OF ABBREVIATIONS

AAT All-At-a-Time Sensitivity Analysis
AGC<sub>w</sub> Above-Ground Live Wood Carbon

BBGCMuSo Biome-BGCMuSo

BGR Biogeographical Region

Baylo Bosic Density of Fresh We

BWD Basic Density of Fresh Wood

C Carbon

CD Carbon Density
CF Carbon Fraction

CFS Coniferous Forest Stratum

CroNFI Croatian National Forest Inventory

CSC Carbon Stock Change
DFS Deciduous Forest Stratum

DW Dead Wood

DWCS Dead Wood Carbon Stock

EC Eddy Covariance
EPC Ecophysiological file

FF Forest Floor

FFC Forest Floor Carbon

FFCS Forest Floor Carbon Stock

FL-FL Forest Land Remaining Forest Land

FOOYS Forest Out of Yield Stratum

ForecoDB Forest Ecosystem Variables Database

GHG Greenhouse Gas

GLUE Generalized Likelihood Uncertainty Estimation Method

GPP Gross Primary Productivity

GWT Groundwater Table

ICP International Co-operative Programme on Assessment and Monitoring of

Air Pollution Effects on Forests

INI Initialization File

IPCC Intergovernmental Panel on Climate Change

LC-FL Land Converted to Forest Land LDW Lying Dead Wood Volume

LH Likelihood Function

LULUCF Land Use, Land-Use Change and Forestry

MAP Mean Annual Precipitation
MAT Mean Annual Temperature

MET Meteorological File MGM Management File

N Nitrogen

NEE Net Ecosystem Exchange NFI National Forest Inventory NIR National Greenhouse Gas Inventory Report

OAT One-At-a-Time Sensitivity Analysis

OL Litter Layer of Forest Floor

OFH Fermentation and Humus Layers of Forest Floor

SA Sensitivity Analysis

SDW Standing Dead Wood Volume

SOC Soil Organic Carbon

-SOC<sub>30</sub> Top 30 cm of Mineral Soil Layer

SOI Soil Properties File SOM Soil Organic Matter SWC Soil Water Content

TDW Total Dead Wood Volume

UNFCCC United Nations Framework Convention on Climate Change

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#### 1. INTRODUCTION

The Republic of Croatia is a signatory Party of the United Nations Framework Convention on Climate Change (UNFCCC, 1992, NN 2/1996) and its parent treaties, the Kyoto Protocol (1997, NN 5/2007) and the Paris Agreement (2015, NN 3/2017). As a country listed in Annex I of the UNFCCC, Croatia has committed to annually monitor and report on greenhouse gas (GHG) emissions and removals in its National GHG Inventory Report (NIR). In the NIR, GHG CO<sub>2</sub> is in focus, as its emissions from anthropogenic sources, i.e. industry and burning of fossil fuels, contribute the most to the total GHG emissions increase (IPCC 2014). As an offset to CO<sub>2</sub> emissions, parties are encouraged to enhance CO<sub>2</sub> sequestration, i.e. removal, for which nature-based solutions are relevant (Council of the European Union 2019). The only sector in NIR that accounts for carbon removals by natural processes is the Land Use, Land-Use Change and Forestry (LULUCF) sector. LULUCF has been recognized, in some regions, as crucial in reaching the goal of climate neutrality stated in the European Green Deal (EC 2019).

Forest ecosystems have a key role in climate change mitigation as they store ~45% of total terrestrial carbon (IPCC 2000) by sequestering large amounts of carbon into their woody and leaf biomass, and soil (Dixon et al. 1994, Pan et al. 2011). In the forest ecosystem, carbon is stored in five different carbon pools: above- and below-ground live biomass, dead wood and forest floor (combined referred to as dead organic matter – DOM), and soil organic matter – SOM (IPCC 2006). Carbon is continuously accumulated or lost during the natural processes of growth and decomposition in the forest ecosystem carbon pools, or due to mortality after natural disturbances or logging. These emissions and removals of carbon are known as carbon fluxes, and depending on their balance, forests can serve as a carbon sink or carbon source. On a global, as well as European scale, forests currently act as carbon sinks (Ceccherini et al. 2020, Harris et al. 2021).

Long-term observations of carbon pools are generally obtained from the National Forest Inventories (NFIs) (Tomppo et al. 2010). The first NFIs in Europe were carried out in the early 1920s (Spiecker 1999) and were primarily intended for economic purposes, i.e. for estimating growing stock and stemwood increments. Therefore, NFIs usually include measurements of basic forest variables, such are tree diameter at breast height and tree height, and lack on forest floor and soil data (Latte et al. 2013). Forest floor and soil, together with dead wood, were mainly seen through their ecological role (Olson 1963, Martinović 1973, Harmon et al. 1986) and have usually been monitored only for locations of specific interest (Hauβmann and Fischer

2004). National-scale long-term monitoring of these pools was, in general, affordable only for the most developed countries (Wolff and Riek 1997). Until recently, about 30% of Annex I parties of the UNFCCC have still not been reporting on carbon emissions and removals from the dead wood and forest floor and only 55% have been reporting estimates for carbon changes in the mineral soil at higher *Tiers* (Didion et al. 2016) meaning that value other than zero was reported. As the countries strive to improve their GHG reporting under the LULUCF sector, the NFIs have been broadened with new survey objectives in the last decades, such as estimates of dead wood and forest floor (Tomppo et al. 2010, Domke et al. 2016). Also, for the sound estimation of national carbon stocks in soil, national soil inventories with high density of plots are being carried out, although, in some of them, the forest floor data is still lacking (López-Senespleda et al. 2021).

In Croatia, dead wood has been recognized as an important carbon pool and therefore included and measured in the first Croatian National Forest Inventory (CroNFI) (Čavlović 2010), while carbon stocks in forest floor and soil have been estimated within the National scientific soil survey carried out in 2015/2016 (Miko et al. 2017). Until today repeated measurements of carbon stocks in these pools have not been performed at the national level, which makes a straightforward estimate of carbon stock changes in these pools impossible and poses a strong challenge to the Croatian GHG Inventory subjects to compile a complete report under the LULUCF sector. This research addresses issues relevant to specific carbon pools whose resolving can enhance the estimate of carbon emissions and removals in the national LULUCF sector.

#### 1.1. Dead organic matter and soil carbon pools in the forest ecosystem

#### 1.1.1. Dead wood

Dead wood (DW) constitutes ~1% of the carbon reservoir in temperate forest ecosystems (both managed and protected forest areas) in Europe (Pan et al. 2011) and in Croatia (Čavlović 2010). It contributes to the structural diversity of the forest ecosystem with a share of 5.4% in Europe's (excl. Russian Federation) stock of woody biomass and necromass combined (FAO 2020). Moreover, DW has a significant role in nutrient cycling (Harmon et al. 1986, Laiho and Prescott 2004, Błońska et al. 2017, Harmon et al. 2020) and in promoting ecosystem biodiversity (Humphrey et al. 2004, Seibold et al. 2015, Muys et al. 2022) by providing a special habitat for

many species (mammals, birds, insects, and other species) (Nordén et al. 2004, Müller and Bütler 2010, Stokland et al. 2012).

DW is usually divided into coarse (CWD) and fine (FWD) woody debris. According to the IPCC Guidelines (IPCC 2006), DW carbon pool consists of different fractions larger than a country-defined diameter: woody debris lying on the soil surface, standing dead trees (snags), dead roots and stumps, and other dead material not included in the forest floor or the soil. The minimum diameter limit for CWD varies from 0 to 35 cm among countries, although the most common limit is 10 cm (Cienciala et al. 2008), which corresponds to the IPCC Guidelines' definition (IPCC 2006).

For the calculation of DW carbon stocks, DW volume and volume-to-carbon conversion factors, i.e. basic density of DW and carbon fraction (CF), are needed. Data on DW volume is usually obtained from NFIs (Lawrence et al. 2010), while national DW basic densities and CFs often lack (Woodall et al. 2008) and are commonly used from the literature and applied uniformly for a variety of tree species and different areas.

The basic density of fresh wood (BWD) is a plant functional trait and is defined as the ratio of the dry weight mass and the volume of fresh wood. In wood-decay-related research, it usually corresponds to the wood density in decay class 0, which is a common label denoting fresh wood (Olajuyigbe et al. 2011). It varies between tree species as well as within individuals (Zanne et al. 2009) and is usually higher in deciduous than in evergreen tree species, and in tropical compared to either temperate or boreal tree species (IPCC 2003). The carbon fraction in fresh wood of tree species in temperate and boreal regions varies from 47% to 55% (IPCC 2006), and on average it is higher for gymnosperms compared to angiosperms (Harmon et al. 2013, Martin et al. 2018). Dead wood basic density and CF vary in decomposition processes in DW (Merganičová and Merganič 2010, Di Cosmo et al. 2013, Neumann et al. 2023). Decomposition processes can be represented robustly with decay classes where DW pieces are grouped according to a predefined DW classification scheme (Harmon et al. 1995, Sandström et al. 2007) with the number of decay classes spanning from three to eight (Holeksa 2001, Sandström et al. 2007, Čavlović 2010). Its application is rather simple and useful for reporting on DW pool (DE NIR 2023).

As the wood decomposes, its basic density decreases with increasing decay class (Yatskov et al. 2003, Harmon et al. 2008). In contrast to DW basic density, CF can slightly increase with the increasing decay class (Sandström et al. 2007, Di Cosmo et al. 2013), and for some tree species, this increase is significant (Stakenas et al. 2020). According to a recent review (Martin et al. 2021) CF in dead wood is on average around 48.5%. Therefore, using the old IPCC's

default value of 50% (IPCC 2003), which is sometimes still applied, can result in the overestimation of carbon stocks in DW pool (Martin et al. 2021).

Finally, using carbon density (CD), i.e. the product of DW basic density and CF, in the calculation of DW carbon stocks could reduce uncertainty considering the contrasting trends of DW basic density and CF with the increasing decay class (DW basic density decreases and CF increases with decay) (Stakenas et al. 2020).

#### 1.1.2. Forest floor

Forest floor accounts for ~8% of total carbon stocks in the temperate forests in Europe (Pan et al. 2011) and ~6% in Croatia (Čavlović 2010). Although forest floor carbon pool size is substantially smaller than that of other forest ecosystem carbon pools (e.g. biomass or soil), forest floor has a key role in forest ecosystems. It serves as a protection cover to the mineral soil layer and mitigates possible negative effects, such as erosion effects due to heavy rainfall (Miyata et al. 2009, Li et al. 2014). Similarly, the forest floor serves as an important source of carbon for the soil where, after fragmentation by fauna and weather, small organic particles, which have not been fully oxidised, gradually migrate, thus becoming integral constituents of the mineral part of the soil. The dynamics of forest floor carbon stocks, as a result of its production and decay rates, reflect the underlying energy flow and nutrient cycling in the forest ecosystem (Olson 1963).

The forest floor corresponds to the litter carbon pool used in the context of GHG reporting (IPCC 2006). The issue of ambiguous nomenclature is well-recognized and, in this research, the term 'litter', used in the Croatian NIR, is substituted with the term 'forest floor'.

The forest floor, i.e. O horizon of the soil profile, is generally divided into three layers: undecomposed leaf organic layer (OL or Oi), fragmented (OF or Oe) and humified (OH or Oa) organic layers (Zanella et al. 2011). According to the IPCC (IPCC 2006), forest floor includes all non-living biomass in various states of decomposition above or within the mineral layer of organic soil, with a diameter less than the minimum diameter for dead wood and greater than the maximum limit for soil organic matter. Live fine roots (with a diameter less than the chosen limit for below-ground biomass) are included in the forest floor where they cannot be distinguished from it empirically.

For the calculation of forest floor carbon stocks, forest floor mass and carbon fraction (CF) are needed.

Forest floor mass, and therefore its carbon stock, is highly variable with respect to climate region (Liski et al. 2003), forest type (Domke et al. 2016), and various site and stand characteristics (Qin et al. 2020). At the European scale, forest floor carbon stock varies in terrestrial humus forms between 8 to 22.5 t C ha<sup>-1\*</sup> depending on the humus type (De Vos 2015). On the global scale, forest floor carbon stocks typically range from 2.1 to 55 t C ha<sup>-1</sup> depending on climate region and forest type, with the highest values reported for boreal coniferous forests and lowest values for tropical deciduous forests (IPCC 2006). In general, forest floor carbon stocks are found to be higher in coniferous than in deciduous forests (Domke et al. 2016, Lee et al. 2020, López-Senespleda et al. 2021). This could be attributed to different forest floor chemical and physical properties that affect decomposition processes (Vesterdal and Raulund-Rasmussen 1998, Berg 2000). Needles have a higher share of substances that contribute to slower decomposition (e.g. waxy coating and higher lignin content) than leaves (Pernar 2017). When looking at the regional differences in forest floor carbon stocks, in the Mediterranean BGR, the forest floor tends to have higher carbon stocks, in comparison to the Continental BGR (Ostrogović Sever et al. 2019, Bakšić and Bakšić 2023).

Accounting for the diversity of forest floor mass, and therefore its carbon stock when calculating national carbon stocks is relevant.

#### 1.1.3. Soil organic matter and soil organic carbon

The organic matter within the mineral part of the soil, referred to as the soil organic matter (SOM), is the largest carbon pool in the forest ecosystems globally (44%, Pan et al. 2011) and in Croatia (50%, Čavlović 2010). SOM consists of fragments and particles of dead organic matter within the soil matrix, as well as live and dead fine roots having a diameter that is less than the minimum diameter limit for coarse roots if they cannot be distinguished from it empirically (IPCC 2006). SOM dynamics play an important role in forest productivity, and nutrient and hydrologic cycles (Grigal and Vance 2000, Masi et al. 2020).

The largest constituent of the SOM is carbon. The carbon in mineral soils which is of organic origin (unlike inorganic carbon in e.g. CaCO<sub>3</sub>) is referred to as soil organic carbon (SOC). Reporting on the changes in SOC in a country's NIR is most commonly done for a mineral soil layer from the top to a depth of 30 cm, although 100 cm depth is also an option (IPCC 2006).

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<sup>\*</sup> Carbon stocks in forest carbon pools are presented in measurement units t C ha<sup>-1</sup> to be in line with National Greenhouse Gas Inventory Report and IPCC Guidelines (IPCC 2006).

Change in SOC is determined either by calculating the difference in SOC stocks for a period between two measurements or by estimating the balance between carbon input by litterfall and rhizodeposition, and the loss of carbon through leaching, decomposition and erosion of organic matter (Jandl 2007).

For the calculation of SOC stocks, data on soil bulk density, carbon fraction, stone content and soil depth are needed. All these variables vary with space and have different associated measurement errors (Schrumpf et al. 2011).

Measuring SOC changes is challenging due to the high spatial variability of soil carbon and the slow process of soil carbon accumulation or loss (Jandl et al. 2007), i.e. changes in SOC with time are small depending on the size of the SOC pool (Conant et al. 2003). To detect relevant changes in SOC and to reduce the uncertainty of the estimates, a high sampling density is required (Saby et al. 2008), which poses a noteworthy cost challenge (Mäkipää et al. 2008). Hence, only a minority of countries have carried out repeated national soil inventories (Saby et al. 2008), e.g. Denmark, Germany and Sweden (Grüneberg et al. 2014). When there are no repeated soil measurements, the alternative and cost-effective method to estimate SOC changes is a modelling approach. Modelling of SOC is already in use for national GHG inventory reporting, e.g. Yasso model (AT NIR 2023).

There is a vast number of literature on SOC models (Campbell and Paustian 2015) that vary in complexity and input data requirements (Manzoni and Porporato 2009). These models differ in type (empirical or process-based) and modelled systems (soil or ecosystem). Empirical models are built based on the observed statistical relationships between specific stand or ecosystem variables. An example of such model is a tree height growth model with a diameter at breast height as an independent variable. In contrast, process-based models are mathematical representations of main biological processes (e.g. photosynthesis, decomposition, and respiration) driven by climate variables, the basis of which is the circulation of the main ecosystem elements, most often carbon, nitrogen, and water. Soil carbon models (e.g. Yasso20, Viskari et al. 2022) simulate processes in soil, while ecosystem carbon models (e.g. CENTURY (Parton et al. 1994) and Biome-BGCMuSo (Hidy et al. 2016a)) simulate processes throughout the entire soil–plant–atmosphere system.

Process-based models are complex, with a high number of input parameters and variables, and are under continuous development (Hidy et al. 2016a). To address uncertainty in ecosystem modelling, models are calibrated and validated by verifying the results of the model with the results from field observations (Hararuk et al. 2014, Tupek et al. 2019). For this, wide-scale and/or long-term field measurements of SOC are necessary.

#### 1.2. Reporting on carbon stock change in the LULUCF sector

Within the LULUCF sector, there are six Land-use categories: Forest land, Cropland, Grassland, Wetlands, Settlement, and Other land (IPCC 2006). Each category is partitioned into two subcategories, i.e. land remaining in the same category and land converted to other land-use categories. Forest land is therefore partitioned into subcategories *Forest Land Remaining Forest Land* (FL-FL) and *Land Converted to Forest land* (LC-FL). Subcategories can further be stratified regarding climate zone, ecological zone, management system, etc., while LC-FL is also divided in a way to express the change from each of the non-forest land-use categories into *Forest land*. The land is converted to Forest land by afforestation and reforestation, either by natural succession or anthropogenic conversion (e.g. the establishment of plantation on non-forest land use). According to the IPCC Guidelines (IPCC 2006), the default reporting and accounting period for a land conversion is set to be 20 years. Although for most forest ecosystems, a longer period is needed to reach the stable level of carbon stocks as in the undisturbed state (Bárcena et al. 2014), the default 20-year period is suggested to capture the establishment of forest ecosystems.

Carbon stock changes can occur in both main Forest land subcategories, either due to carbon stock change with time within the FL-FL or due to the land-use change (Figure 1.1, left and right panels, respectively). Here it should be noted that the carbon stock change due to deforestation is particularly important as it can lead to significant carbon loss from the soil. However, deforestation is not reported and accounted for under the Forest land category (as the deforested area is no longer a forest), but under the new land-use category into which that Forest land has been converted.



Forest land Remaining Forest land

Land Converted to Forest land

**Figure 1.1.** Forest land subcategories in the National GHG Inventory Report. At the right panel, 20 years indicates a period for land to be converted to another land-use subcategory.

The capacity of countries to assess all GHG emissions and removals varies. Therefore, the IPCC Guideline (IPCC 2006) offers three tiers of reporting (Tier 1–3) regarding the data requirements

and methodological complexity, with Tier 1 being the simplest one. Data needed for reporting are activity data (national data on the area under specific land-use category and subcategory) and the carbon stock change (CSC) factors to which activity data is to be multiplied. CSC factors are equal to carbon stock change that occurred within the land-use category and subcategory for a certain carbon pool. Carbon stock changes can be estimated using two methodologies: 1) the Gain-Loss Method, which uses a mass balance of inputs and losses to and from a certain pool over a specified period, and 2) the Stock-Difference Method, which accounts for changes in carbon stocks measured at two points in time (IPCC 2006).

# 1.3. National reporting on carbon stock change in dead organic matter and soil in the LULUCF sector

Under *Forest Land Remaining Forest Land* (FL-FL) Croatia uses the Tier 1 approach (IPCC 2006) for the calculation of carbon stock change in dead wood, forest floor and soil carbon pools, assuming no changes in carbon stocks under these pools. The main reason for using Tier 1 is the absence of two consecutive national forest inventories that account for these pools. If no carbon stock change data is available for these pools, enhancement of reporting under this land-use subcategory could only be reached using the modelling approach.

Under *Land Converted to Forest land* (LC-FL), Croatia reports annual carbon stock changes in dead wood, forest floor and soil carbon pools using the Tier 2 approach (IPCC 2006), which means that national CSC factors are used. National CSC factors for soil and forest floor are calculated as a difference in average carbon stocks between Forest land and any other non-forest land from which conversion to forest occurred, and are divided by 20 to obtain a yearly carbon stock change rate (assuming a 20-year conversion period). For the dead wood pool, the CSC factor is calculated from DW volume in the afforested area using volume-to-carbon conversion factors for fresh wood. Enhancement of reporting under this land-use subcategory would imply a more improved estimate of carbon stocks, out of which CSC factors will be calculated.

#### 1.3.1. Dead wood carbon stocks

Currently in the Croatian NIR, DW carbon stocks are calculated using: 1) total DW volume published in the first Croatian National Forest Inventory (CroNFI) (Čavlović 2010), but

disregarding decay classes; 2) national values for basic wood density of fresh wood; and 3) default IPCC values for carbon fraction of fresh wood (IPCC 2003).

Disregarding decay classes in DW carbon stock calculation may lead to under/overestimation in the emissions/removals reported in NIR, as it is reasonable to expect that the use of decay class-specific DW volume-to-carbon conversion factors would yield lower DW carbon stock estimates, compared to the ones based on the values for fresh wood (Merganičová and Merganič 2010). However, national DW basic density and carbon fraction by decay class do not exist. Having in mind the high biogeographical diversity of Croatia, to improve the estimate of DW carbon stocks at a national level a country-specific DW volume-to-carbon conversion factors for specific tree species group under different biogeographical regions (BGRs) would be desirable.

#### 1.3.2. Forest floor carbon stocks

Forest floor carbon (FFC) stocks, currently used in the Croatian NIR, are calculated from the forest floor database collected within the National scientific soil survey carried out in 2015/2016 (Miko et al. 2017). The current forest floor database is underrepresented for Mediterranean BGR. Nevertheless, an overall country mean value of 4.57 t C ha<sup>-1</sup> is used for the estimate of the CSC factor within this carbon pool.

National studies indicate that FFC stocks in Mediterranean forest ecosystems can have very high values (Bakšić and Bakšić 2017, Bakšić and Bakšić 2023). Therefore, it is reasonable to assume that FFC stocks, currently reported in the Croatian NIR, are underestimated.

To improve the estimate of national FFC stocks, a more representative database on FFC stocks would be desirable.

#### 1.3.3. Soil organic carbon stocks

SOC under different land-use categories is estimated from the soil database collected within the National scientific soil survey carried out in 2015/2016 (Miko et al. 2017). The current soil database is very well distributed across Croatia, which is why no significant need for improvement under the LC-FL subcategory was noted. However, under the FL-FL subcategory, SOC stock changes in the Croatian NIR are assumed to be stable (Tier 1 approach). Although this can be justified with the commonly used shelterwood system for which it is assumed to preserve stable SOC stocks (Ostrogović Sever et al. 2019), due to the ongoing changes in

climate, SOC stocks and SOC dynamics could potentially be altered (Boisvenue and Running 2006).

This issue can be addressed by simulating SOC stock changes with a climate-driven process-based model. The application of a process-based biogeochemical model for simulating the development of carbon stocks in forest ecosystems in Croatia is slowly growing (Hidy et al. 2016a, Ostrogović Sever et al. 2017, Ostrogović Sever et al. 2021). Currently, there is only one operational biogeochemical model in Croatia that can simulate SOC stocks and fluxes, and that is Biome-BGCMuSo (Hidy et al. 2016a, 2022). For the model to be used for reporting on SOC changes, first its performance regarding simulating SOC stocks at the national level needs to be evaluated. Moreover, since the model is continuously under development, a calibration of the most recent model version, as well as its validation with field measurements of SOC dynamics are required.

#### 1.4. Research aim

The main aim of this research is to facilitate the improvement of estimates of carbon stocks in dead wood and forest floor, as well as carbon stock changes in soil organic carbon pools in forest ecosystems in Croatia. Based on the national needs regarding these carbon pools identified in the previous section, three specific research aims (A) and four hypotheses (H) were defined:

A1) to provide national volume-to-carbon conversion factors by decay classes for use in the calculation of dead wood carbon stock, with the hypothesis

H1: Dead wood volume-to-carbon conversion factors of a specific tree species group differ between biogeographical regions;

A2) to provide a more representative national forest floor carbon stock mean, with the hypothesis

H2: Forest floor carbon stocks for a specific tree species group differ between biogeographical regions; and

A3) to investigate the applicability of the process-based model Biome-BGCMuSo for estimating carbon stocks and carbon stock changes in the soil organic matter, with the hypotheses

H3: The ability of the model Biome-BGCMuSo to reproduce measured forest soil organic carbon of the mineral soil layer is determined by the size of strata;

H4: The Biome-BGCMuSo model can reproduce measured soil organic carbon changes in the mineral soil layer of the pedunculate oak forest during the period 2012-2022.

#### 2. MATERIALS AND METHODS

#### 2.1. Research design

This research is divided into three studies addressing the following carbon pools: dead wood, forest floor and soil organic carbon. For each study, a different research design was applied at a different spatial scale.

**Dead wood study** was conducted at the country level and consisted of field experiment on ten forest tree species in four locations distributed across three biogeographical regions in Croatia. **Forest floor study** was also conducted at the country level and consisted of a compilation of available data sources on national forest floor carbon stocks.

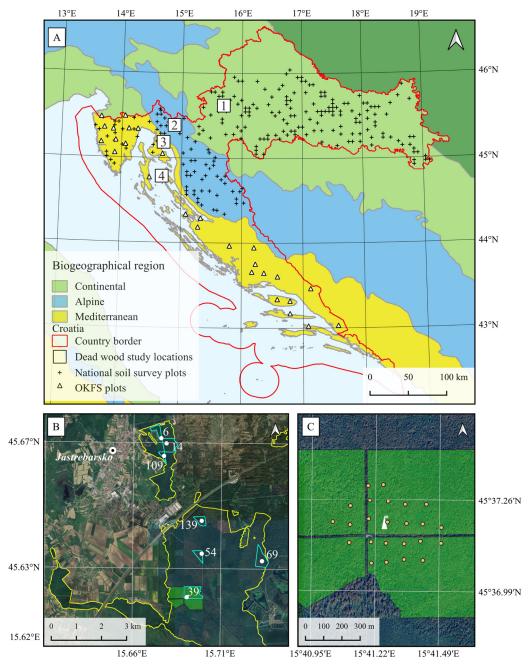
Soil organic carbon study was conducted at three levels: country, forest, and forest stand. At the country level, a validation of the Biome-BGCMuSo model for estimating national SOC stock in the top 30 cm of the soil mineral layer was performed across three BGRs in Croatia. At the forest stand level, sensitivity analysis and calibration of the latest Biome-BGCMuSo model version were performed for a pedunculate oak forest stand at the eddy-covariance site in Jastrebarsko. Finally, at the forest level, a validation of the calibrated Biome-BGCMuSo model for estimating SOC change in the top 30 cm of the soil mineral layer was performed using a chronosequence experiment in a pedunculate oak forest in Jastrebarsko.

#### 2.2. Study area

The research was conducted across three biogeographical regions (BGRs) of Croatia: Continental, Alpine and Mediterranean (EEA 2016) (Figure 2.1).

The Continental BGR covers the northern and central parts of Croatia and is characterized by a temperate rainy climate with lowest amount of precipitation in the month of the cold part of the year and two precipitation maxima, classified as Cfwbx" according to the Köppen classification (Zaninović et al. 2008). The dominant mean annual temperature (MAT) is about 11°C and the mean annual precipitation (MAP) ranges from approximately 600 to 1,200 mm following an east-west gradient, with the greater amount of precipitation of up to 1,500 mm at higher altitudes (Zaninović et al. 2008, data for the period 1961–1990). The soil parent material is mainly alluvial deposits (lowlands) and silicate, or occasionally carbonate, rock (hills and

mountains) (Bogunović et al. 1997, Bašić et al. 2007), developed on a parent material consisting of magmatic, clastic and metamorphic rocks (Halamić and Miko 2009, Velić and Vlahović 2009).



**Figure 2.1.** The panel A) represents three biogeographical regions in Croatia and geographical locations of sampling sites in the dead wood study (white squares; 1) Jastrebarsko, 2) Zalesina, 3) Crikvenica, 4) Rab), National scientific soil survey plots (black pluses) and OKFŠ project plots (white triangles). The panel B) represents the geographical locations of the chronosequence experiment plots (white circles; 6, 14, 39, 54, 69, 109 and 139-year-old stands in the year 2012) located within the management unit Jastrebarski lugovi (delineated with yellow border) and forest compartments (delineated with cyan borders). The panel C) represents the geographical location of the eddy covariance (EC) site with the EC tower (specific white symbol) and permanent plots (pastel orange circles) located within the management unit Jastrebarski lugovi and forest compartments (filled with green colour).

The Alpine BGR is situated between the Continental and Mediterranean BGRs and has mainly a Cfsbx" climate and at the mountain peaks (higher than 1,200 m a.s.l.) a Dfsbx" climate according to Köppen classification, with the highest amount of precipitation occurring in the cold part of the year (Zaninović et al. 2008). The Alpine BGR has great altitude differences and substantial variation in MAT and MAP across the region. At low altitudes (Gospić, 564 m a.s.l.) MAT is 8.7°C and MAP is 1,365.9 mm, while at higher altitudes (Zavižan, 1,594 m a.s.l.) MAT is 3.8°C and MAP is 1,973.4 mm (564 m a.s.l.) (Zaninović et al. 2008, data for the period 1971–2000). Gorski kotar and peak areas of Velebit can receive even up to 3,500 mm MAP (Zaninović et al. 2008, data for the period 1961–1990). The soil bedrock is mainly limestone and dolomite (Bogunović et al. 1997, Bašić et al. 2007), developed on the parent material consisting dominantly of carbonate Mesozoic rocks (Halamić and Miko 2009, Velić and Vlahović 2009).

The Mediterranean BGR covers Istria and Dalmatia, coastal areas and islands in the Adriatic Sea. According to the Köppen classification, the climate is classified as Cfsax'' in Istria and Kvarner Bay (Seletković and Katušin 1992) and Csa and Csax'' on the islands and in the coastal area of the Middle and Southern Dalmatia (Zaninović 2008). The Mediterranean BGR also greatly varies in MAT and MAP. MAT ranges from 8°C (Učka Mountain) to 17°C on the islands of Middle and South Dalmatia, while MAP ranges from 800 mm up to 2,500 mm at the Učka Mountain (Zaninović et al. 2008, data for the period 1961–1990). The soil parent material is mainly carbonate, predominantly limestone.

**The dead wood study** was performed on four sampling locations across three BGRs in Croatia (Figure 2.1, panel A, square symbols; Table 2.1).

In the Continental BGR, the sampling site was located near the town of Jastrebarsko in the management unit Jastrebarski lugovi in a pedunculate oak forest managed with an even-aged management system (Anić et al. 2018). The dominant tree species in the basin is pedunculate oak (*Quercus robur* L.) with a volume share of 61%, followed by a 17% share of black alder (*Alnus glutinosa* (L.) Geartn.), 13% of common hornbeam (*Carpinus betulus* L.), and 9% of narrow-leaved ash (*Fraxinus angustifolia* Vahl) (Anić et al. 2018).

The sampling site in the Alpine BGR was located in management units Belevina, Kupjački vrh and Sungerski lug within the educational and experimental forest "Zalesina" of the Faculty of Forestry and Wood Technology, University of Zagreb. The "Zalesina" forest is managed using an uneven-aged management system. In the management units Belevina and Kupjački vrh the dominant tree species is silver fir (*Abies alba* Mill.) with a volume share of 73% and 57%,

respectively, followed by common beech (*Fagus sylvatica* L.) with a volume share of 23% and 30%, respectively, and a low volume share of Norway spruce (*Picea abies* (L.) Karst.) (<4%). In the management unit Sungerski lug silver fir is the dominant tree species with a volume share of 65%, while Norway spruce and common beech contribute with 29% and 6%, respectively. In the Mediterranean BGR, two sampling locations were selected, one near the coastal town of Crikvenica in the management unit Kotor planina in the even-aged managed, 120 years old, planted forest of Austrian pine (*Pinus nigra* Arnold) and one on the island of Rab in the management unit Kalifront within the educational and experimental forest "Rab" of the Faculty of Forestry and Wood Technology, University of Zagreb. The "Rab" forest is an even-aged managed holm oak (*Quercus ilex* L.) and maritime pine (*Pinus pinaster* Aiton) forest with the species volume shares of 64% (holm oak), 31% (pines), and 5% (other tree species).

**Table 2.1.** Location description of dead wood experimental study.

Location	Biogeographical region	Y	X	Climate type <sup>1</sup>	MAT <sup>2</sup> (°C)	MAP <sup>2</sup> (mm)
Jastrebarsko	Continental	45°37'N	15°41'E	Cfwbx"	10.6	962
Zalesina	Alpine	45°23'N	14°52'E	Cfsbx"	8.1	2,063
Crikvenica	Mediterranean	45°10'N	14°41'E	Cfsax"	14.8	1,242
Rab	Mediterranean	44°46'N	14°40'E	Cfsax"	15.6	1,087

NOTE: MAT – Mean annual temperature; MAP – Mean annual precipitation; <sup>1</sup>According to the Köppen classification; <sup>2</sup>Data from the nearest meteorological station (Jastrebarsko, Delnice, Crikvenica and Rab) for the period 1981–2010.

The forest floor study includes a compilation of data from 274 locations distributed across three BGRs in Croatia. Data were collected in the period 2015–2016 within the National scientific soil survey, project "Soil carbon stock changes and calculation of soil organic carbon, total soil nitrogen and C:N trends" (10-14-1442/79, Croatian Environment Agency) (Figure 2.1, panel A, plus symbols) and in the period 2017–2018 within the OKFŠ project "Adaptive capacity of Croatian Mediterranean forests to environmental pressures" (Ministry of Agriculture) (Figure 2.1, panel A, triangle symbols).

Within the **Soil organic carbon study**, SOC data were used from three datasets: 1) 274 locations across three BGRs in Croatia, described above for the forest floor study (Figure 2.1, panel A, plus and triangle symbols), 2) the chronosequence experiment (Figure 2.1, panel B), and 3) the eddy covariance site (Figure 2.1, panel C).

The chronosequence experiment and eddy covariance site are located near the town of Jastrebarsko in the managed forests of the Kupa river basin, with climate conditions described

in Table 2.1 for the Jastrebarsko location. At the Kupa river basin, the soils are hydromorphic and, according to the FAO World Reference Base for Soil Resources (IUSS Working Group WRB 2015), they are classified as gleysol and luvic stagnosol on parent material that consists mainly of a clay fraction (Mayer 1996). During winter and early spring, parts of the forest are waterlogged or flooded with stagnating water, while during summer the soil dries out. The average groundwater table depth in the Kupa river basin ranges from -60 to -200 cm during the vegetation period (Mayer 1996).

# 2.3. Dead wood study

# 2.3.1. Field sampling

A field experiment was performed on ten tree species: pedunculate oak, black alder, common hornbeam and narrow-leaved ash in the Continental BGR; silver fir, common beech and Norway spruce in the Alpine BGR; holm oak, maritime pine and Austrian pine in Mediterranean BGR.

A four-class decay classification scheme was used (Table 2.2, Figure 2.2). This classification scheme corresponds to the first four decay classes in the commonly used five-class decay classification scheme presented by Harmon et al. (1995). The fifth decay class was not assessed in this study since in this decay class wood integrity is lost and the accurate volume estimation with the method used in this research would be difficult. Also, distinguishing tree species at this stage of decomposition is challenging.

**Table 2.2.** Description of the five-class decay classification scheme – modified from Harmon et al. (1995) and Di Cosmo et al. (2013).

Decay class	Class description
1	Solid dead wood (bark still attached, penetration depth with a knife or similar tool indistinguishable from that of fresh wood, wood integrity preserved)
2	Weakly decayed wood (partially no bark, resistance to penetration still considerable, wood integrity mostly preserved)
3	Decayed wood (no bark, easy to partially penetrate, wood integrity still partly preserved, parts disintegrate when stepped upon, signs of losing the original shape)
4	Very decayed wood (rotten, penetration very easy and deep, wood integrity mostly compromised, original shape visibly changed)
5*	Almost decomposed (wood integrity lost – dust, wood scattered across the soil surface)

<sup>\*</sup>not analysed in this study.



Figure 2.2. Silver fir (Abies alba (Mill.)) lying dead wood from first (left) to fourth decay class (right).

Lying DW was randomly sampled within the selected management units at each sampling location until the sufficient number of samples per tree species and decay class was collected. For each of the selected tree species and each of the decay classes (1–4), three DW discs were sampled from three different DW logs and within each of three predefined diameter classes (5–10, 10–20, and 20–30 cm), resulting in nine samples per tree species and decay class. Dead wood volume-to-carbon conversion factors were analysed relative to the DW diameter and decay classes. DW logs thicker than 30 cm were not sampled since the experiment was performed in managed forests where larger dying trees are removed during thinning, and since obtaining a sufficient number of samples with a diameter thicker than 30 cm for all decay classes was not feasible.

For each sampled DW log two perpendicular diameters were measured at the position where the DW disc would be sampled, with a calliper at 1 mm precision. Also, a knife penetration test was performed using a 5 mm Philips screwdriver below or above the position a disc was cut from. From each sampled DW log, a 3 cm-wide wood disc was cut at the position of diameter measurements with a chainsaw. Plastic bags were placed around the cut of the very decayed DW samples (decay class 4) to ensure all DW pieces of the sample were collected during the cut. The samples were placed in plastic bags, stored in a cool place and transported to the laboratory for further analyses within 3 days.

In total, 360 disc samples of DW were collected and analysed. The samples represented individual tree species, decay and diameter classes, and were subsequently grouped according to different tree species groups and biogeographical regions (Table A.1).

#### 2.3.2. Laboratory analysis

The volume of each dead wood disc sample was estimated using the suspension water displacement method (Hughes 2005). Before the volume measurements, samples were immersed in the water and left there until saturation (Figure 2.3, left panel). Complete wood discs were then taken and suspended in the water (Figure 2.3, right panel). Very decayed wood samples were placed in the net fabrics to ensure all parts of the wood were retained during the volume measurement. After the volume measurements, samples were first air-dried, then ovendried at 105°C for 48 h and weighed.



**Figure 2.3.** Dead wood samples immersed in the water and left there until saturation (left panel) and volume analysis of the dead wood sample using the suspension water displacement method (right panel) (Hughes 2005).

From each DW sample, a subsample was taken for the elemental analysis. Subsamples were taken as 1-2 mm thick slices (Figure 2.4, left panel) across the entire cross-section of a wood disc (including bark). Keeping in mind that bark and wood differ in CF during decomposition (Romashkin et al. 2021), the bark was included in the subsample proportionally to its share in the wood sample. Subsamples were ground (Figure 2.4, right panel) in a laboratory mill (Retsch ZM 200), homogenised and analysed for carbon fraction with a CNS-2000 Elemental Analyser (LECO 2000).



**Figure 2.4.** Dead wood slices from a cross-section of a wood disc (left panel) and grounded samples of dead wood prepared for analysis for carbon fraction (right panel).

#### 2.3.3. Basic density and carbon density calculation

To test the first hypothesis, volume-to-carbon conversion factors, basic density and carbon density (CD) were calculated for every sample from the sample's oven-dry mass  $(m_{sample, d.m.})$ , volume  $(v_{sample})$  and measured carbon fraction (CF) (Eq. 2.1 and Eq. 2.2).

$$Basic\ density_{sample} = m_{sample,d.m.}/v_{sample}$$
 (2.1)

$$CD_{sample} = Basic\ density_{sample} \cdot CF_{sample}$$
 (2.2)

The mean and standard error (s.e.) of basic density, CF and CD were calculated for every tree species by each decay class separately.

#### 2.3.4. Recalculation of national dead wood carbon stock

The obtained volume-to-carbon conversion factors from this study, together with DW volume data from the first Croatian NFI (CroNFI), were used for the recalculation of national DW carbon stock.

To evaluate the effect of using national decay class-specific volume-to-carbon conversion factors on GHG inventory results, carbon stocks in the DW pool (DWCS) were compared using two calculation methods.

The <u>first calculation method</u> represents the approach currently used in the Croatian NIR (HR NIR 2023), where for a given Forest land stratum (i) *DWCS* is calculated as follows:

$$DWCS1_i = TDW_i \cdot BWD_i \cdot CF \tag{2.3}$$

$$TDW_i = LDW_i + SDW_i \tag{2.4}$$

$$SDW_i = SDW_i^M \cdot BEF2_i \cdot (1 + RS_i)$$
 (2.5)

where  $DWCSI_i$  is dead wood carbon stock, in t C ha<sup>-1</sup>;  $TDW_i$  is total dead wood volume (lying and standing dead wood including branches and roots of stumps and snags), in m<sup>3</sup> ha<sup>-1</sup>;  $BWD_i$  is basic density of fresh wood, in  $t_{dry \, matter \, (d.m.)}$  m<sup>-3</sup>; CF is a default value for the carbon fraction of fresh wood (IPCC 2003), in t C  $t_{d.m.}$  <sup>-1</sup>;  $LDW_i$  and  $SDW_i$  are lying and standing dead wood volumes, in m<sup>3</sup> ha<sup>-1</sup>;  $SDW^M_i$  is merchantable standing dead wood volume, in m<sup>3</sup> ha<sup>-1</sup>; and  $BEF2_i$  and  $RS_i$  are default values for the biomass expansion factor and root-to-shoot ratio, respectively (IPCC 2006).

In this calculation method, volume-to-carbon conversion factors are used regardless of the decay class. Values and factors currently used in the Croatian NIR for estimating DW carbon stock are presented in Table 2.3 by Forest land stratification: deciduous forests stratum (DFS), coniferous forests stratum (CFS) and forests out of yield stratum (FOOYS, i.e. maquis and shrubs) (HR NIR 2023).

**Table 2.3.** Standing (SDW), lying (LDW) and total dead wood (TDW) volume and dead wood carbon stocks per hectare (DWCS1) according to Forest land stratification (DFS – deciduous forests stratum, CFS – coniferous forests stratum and FOOYS – forests out of yield stratum) reported in the Croatian NIR, and IPCC's parameters (IPCC 2003, 2006) for biomass expansion factors (BEF2) for conversion of merchantable volume (SDW<sup>M</sup>) to aboveground tree biomass, root-to-shoot ratio (RS) and carbon fraction (CF) and basic density of fresh wood (BWD) (HR NIR 2023).

	DFS	CFS	FOOYS
$SDW^{M}$ (m <sup>3</sup> ha <sup>-1</sup> )	5.84	5.16	0.58
BEF2	1.197	1.039	1.15
RS	0.23	0.29	0.46
$SDW (m^3 ha^{-1})$	8.60	6.92	0.97
$\mathbf{LDW}$ (m <sup>3</sup> ha <sup>-1</sup> )	7.28	10.32	0.36
$TDW (m^3 ha^{-1})$	15.88	17.24	1.33
$\boldsymbol{BWD}\;(t_{d.m.}\;m^{-3})$	0.558	0.395	0.68
$\mathbf{CF}$ (t C $t_{d.m.}^{-1}$ )	0.50	0.50	0.50
DWCS1 (t C ha <sup>-1</sup> )	4.43	3.40	0.45

<sup>\*</sup>d.m. – dry matter.

The <u>second calculation method</u> uses decay class-specific dead wood carbon density conversion factors estimated within this research. Firstly, an estimate of total DW volume by stratum and decay class was needed. In the Croatian NIR, DW volume data are reported as total DW volumes in each Forest land stratum, regardless of decay classes (HR NIR 2023). For the estimation of DW volume by Forest land stratum and decay class, an assessment of the share of each decay class in every stratum was needed. This was made by applying the relative shares of DW volume according to the decay classes from the first CroNFI (Čavlović 2010). In the first CroNFI, standing DW volume refers only to decay class 1, while lying DW volume is distributed among 3 decay classes with the following shares: 12.5%, 34.2% and 53.3% for decay class 1–3, respectively.

The dead wood carbon stock (DWCS) for a given stratum (i) and a decay class (j) was calculated as follows:

$$DWCS2_{ij} = TDW_{ij} \cdot CD_{ij} \tag{2.6}$$

$$CD_{ij} = BD_{ij} \cdot CF_{ij} \tag{2.7}$$

where  $DWCS2_{ij}$  is dead wood carbon stock, in t C ha<sup>-1</sup>;  $TDW_{ij}$  is total dead wood volume, in m<sup>3</sup> ha<sup>-1</sup>;  $CD_{ij}$  is dead wood carbon density, in t C m<sup>-3</sup>;  $BD_{ij}$  is dead wood basic density, in t<sub>d.m.</sub> m<sup>-3</sup>; and  $CF_{ij}$  is dead wood carbon fraction, in t C t<sub>d.m.</sub> -1.

Furthermore, to enable the application of carbon density conversion factors by Forest land stratum, tree species from this study were categorized in a way to correspond with the Forest land stratification used in NIR, namely: deciduous broadleaves (black alder, common beech, common hornbeam, narrow-leaved ash, pedunculate oak) – DFS; conifers (Austrian pine, maritime pine, Norway spruce, silver fir) – CFS; evergreen broadleaves (holm oak) – FOOYS. Also, in the first CroNFI, a three-class decay classification scheme was defined as follows: 1 – solid wood with no signs of decomposition, 2 – solid wood with the visible start of decomposition (< 50% of wood is decomposed), and 3 – rotten wood (> 50% of wood is decomposed) (Čavlović 2010). To enable the straightforward use of carbon density factors from this research, a link between decay classification schemes used in this research and CroNFI was created:

Decay class (this study)	Decay clas (first CroNF		
1	$\leftrightarrow$	1	
2, 3	$\leftrightarrow$	2	
4	$\leftrightarrow$	3	

#### 2.3.5. Statistical analysis

Statistical analyses were performed using R Statistical Software (v4.1.1; R Core Team 2021). The significance level (alpha value) in statistical testing was set to 0.05. Dead wood samples were grouped according to tree species, tree species groups (broadleaves and conifers), biogeographical regions (Continental, Alpine and Mediterranean) and Forest land strata (deciduous forests, coniferous forests and forests out of yield). The normal distribution of the investigated traits (basic density, CF and CD) were checked with the Shapiro-Wilk normality

test performed on data grouped according to tree species. The hypothesis of a normal distribution of investigated variables had to be rejected (p < 0.05) for the majority of the tree species. Therefore, the differences in traits between different decay classes, tree species and tree species groups were tested using a nonparametric Kruskal-Wallis equality-of-populations rank test with post-hoc Dunn's test of multiple comparisons using rank sums with the Holm-Bonferroni method for the adjustment of the family-wise error rate (Dinno 2015).

# 2.4. Forest floor study

#### 2.4.1. Foreco database

To test the second hypothesis, two data sources on forest floor carbon stocks were compiled into a Forest ecosystem database (ForecoDB). In this way, data on the forest floor in ForecoDB more adequately represents all three BGRs in Croatia and enables the comparison of forest floor carbon stocks by tree species groups (broadleaves and conifers) across the three BGRs.

ForecoDB comprises data collected within two national projects, the National scientific soil survey\*\* – "Soil carbon stock changes and calculation of soil organic carbon, total soil nitrogen and C:N trends" (10-14-1442/79, Croatian Environment Agency) and OKFŠ – "Adaptive capacity of Croatian Mediterranean forests to environmental pressures" (Ministry of Agriculture). The database includes 274 sample plots distributed among various forest ecosystems and three BGRs, i.e. Continental, Alpine and Mediterranean (Table A.2).

ForecoDB is a spatially-explicit database on forest ecosystem variables; forest floor, soil and stand. Forest floor data includes depth, dry mass, bulk density, carbon stock and C:N in OL and OFH forest floor layers. Soil data includes soil organic carbon (top 30 cm of the soil mineral layer), soil texture and bulk density. Stand variables include main tree species, stand basal area and tree density. Additionally, the database includes information on mean annual temperature (MAT), mean annual precipitation (MAP) and elevation at the plot level.

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<sup>\*\*</sup> The author collaborated on field sampling and laboratory analysis in this project.

#### 2.4.2. Recalculation of national forest floor carbon stocks

The current forest floor carbon stock (FFCS1) value used in the calculation of the CSC factor under the Forest land subcategory LC-FL is 4.57 t C ha<sup>-1</sup> (HR NIR 2023). It is estimated using forest floor data collected at 249 plots of the National scientific soil survey using the following equation:

$$\overline{FFCS1} = \frac{1}{N} \sum_{i=1}^{N} \left( \overline{CF_{OL}} \cdot \frac{m_{OL,i}}{A} + CF_{OFH,i} \cdot \frac{m_{OFH,i}}{A} \right)$$
 (2.8)

where *i* is a sampling plot;  $\overline{FFCS1}$  is total average forest floor carbon stock, in t C ha<sup>-1</sup>;  $\overline{CF_{OL}}$  is an average carbon fraction in OL forest floor layer, in t C t<sub>d.m.</sub> <sup>-1</sup>;  $CF_{OFH}$  is a carbon fraction in OFH forest floor layer, in t C t<sub>d.m.</sub> <sup>-1</sup>;  $m_{OL}$  and  $m_{OFH}$  are the oven-dry mass of OL and OFH forest layers, respectively, in g; and A is forest floor sampling area ( $A = 625 \text{ cm}^2$ ).

In the National scientific soil survey, CF was analysed for all collected OFH forest floor samples and for 40 OL forest floor samples. Therefore, the carbon stock in OL forest floor layer was calculated using the average value of CF for the OL forest floor layer.

To facilitate the accuracy of a new estimation of national FFC stocks (FFCS2), two plots used in the calculation of the FFCS1 were excluded in the compilation of the ForecoDB due to incomplete data, i.e. missing data on the OL forest floor layer. Field and laboratory methods used in the OKFŠ project were in line with methods used in the National scientific soil survey, with the exception that for all OL forest floor samples CF was determined.

FFC stocks at each sampling location (i) were calculated as follows:

$$FFCS2_i = \overline{CF_{OL}} \cdot \frac{m_{OL,i}}{A} + CF_{OFH,i} \cdot \frac{m_{OFH,i}}{A}, (samples\ from\ National\ soil\ survey) \tag{2.9}$$

$$FFCS2_i = CF_{OL,i} \cdot \frac{m_{OL,i}}{A} + CF_{OFH,i} \cdot \frac{m_{OFH,i}}{A}, (samples \ from \ OKF\S \ project)$$
 (2.10)

where  $FFCS2_i$  are carbon stocks in the forest floor at sampling plot i, in t C ha<sup>-1</sup>;  $\overline{CF_{OL}}$  is an average carbon fraction in OL forest floor layer, in t C td.m. <sup>-1</sup>;  $CF_{OFH}$  is a carbon fraction in OFH forest floor layer, in t C td.m. <sup>-1</sup>;  $m_{OL}$  and  $m_{OFH}$  are oven-dry mass of OL and OFH forest floor layers, respectively, in g;  $CF_{OL}$  is a plot-specific carbon fraction in OL forest floor, in t C td.m. <sup>-1</sup>; A is forest floor sampling area ( $A = 625 \text{ cm}^2$ ).

Forest floor carbon stock country mean (FFCS2<sub>national</sub>) was estimated as a weighted mean of FFCS2 within forest area in each specific biogeographical region, i.e. Continental (CON), Alpine (ALP) and Mediterranean (MED), using the following equation:

$$\overline{FFCS2}_{national} = \frac{\overline{FFCS2}_{CON} \cdot A_{CON} + \overline{FFCS2}_{ALP} \cdot A_{ALP} + \overline{FFCS2}_{MED} \cdot A_{MED}}{A_{TOTAL}}$$
(2.11)

where  $\overline{FFCS2_{national}}$  is the average carbon stock on forest floor, in t C ha<sup>-1</sup>, and  $A_{CON}$ ,  $A_{ALP}$ ,  $A_{MED}$ , and  $A_{TOTAL}$  are forest areas in Continental, Alpine, and Mediterranean biogeographical regions, and the total forest area in ha, respectively.

Furthermore, to facilitate more options for the use of new FFCS estimates, aside new area-weighted country mean, FFCS were calculated with respect to Forest land stratification used in the Croatian NIR. Carbon stock change (CSC) factors by Forest land strata were calculated as FFCS / 20, where 20 denotes default 20-year period for a land conversion (IPCC 2006).

# 2.4.3. Statistical analysis

Statistical analyses were performed using R Statistical Software (v4.3.2; R Core Team 2023) with the significance level (alpha value) in statistical testing set to 0.05. The normal distribution and the homogeneity of variance of the FFCS on data grouped according to tree species group (broadleaves and conifers), tree species group × BGR and Forest land strata were checked with the Shapiro-Wilk normality test and Barlett's test, respectively. The hypothesis of a normal distribution of investigated variable was accepted (p > 0.05) for both tree species groups and tree species group × BGRs, while the hypothesis of homogeneity of variance of investigated variable had to be rejected (p < 0.05). Therefore, the differences in FFCS within tree species group between BGRs, in BGR between tree species groups, and between Forest land strata were tested using a nonparametric Kruskal-Wallis equality-of-populations rank test with post-hoc Dunn's test of multiple comparisons using rank sums with the Holm-Bonferroni method for the adjustment of the family-wise error rate (Dinno 2015). The analysis of linear regressions between the FFCS and environmental variables, namely site elevation, MAT and MAP, was performed for tree species groups.

# 2.5. Soil organic carbon study

# 2.5.1. General model description

The Biome-BGCMuso model (BBGCMuSo, Hidy et al. 2012, 2016a, 2022) is an improved version of a well-known biogeochemical model Biome-BGC (Running and Hunt 1993, Thornton 2000) used for the simulation of terrestrial ecosystems worldwide. Biome-BGC is a big-leaf process-based model that simulates the storage and fluxes of carbon, nitrogen and water in a soil-plant-atmosphere system. The represented processes include canopy radiation, photosynthesis, stomatal conductance, evapotranspiration, allocation, respiration, litterfall, and decomposition (Thornton et al. 2002). The main parts of a modelled ecosystem are defined as plant (leaf, stem, roots), litter, soil and coarse woody debris. The model uses a daily time step and is driven by meteorological variables (such as daily maximum and minimum air temperature and daily total precipitation), plant's ecophysiological parameters (such as C:N ratio of a specific plant parts, specific leaf area and maximum stomatal conductance) and site properties (e.g. soil texture, soil bulk density, elevation, long-term mean annual air temperature). The model calculations apply to a unit ground area that is considered to be spatially homogeneous.

The main improvement of BBGCMuSo, in comparison to the original model, is the implementation of a multilayer soil submodel (MuSo refers to **Mu**ltilayer **So**il Module) including substantial soil- and plant-related developments (Hidy et al. 2016a, 2022) that allow a more realistic simulation of carbon and water fluxes across the soil profiles. BBGCMuSo improvements also include the implementation of management modules, enabling modelling ecosystems under different management practices (e.g. simulation of different forest thinning rates and frequencies).

Model simulation typically has three phases: spinup, transient and normal run. The first phase, spinup, starts from a 1 g C m<sup>-2</sup> in plant parts and bare ground with zero SOC levels and usually lasts for several hundred to several thousand years until the steady state condition in soil organic matter is reached, using long-term local meteorological data (Thornton 2000) and constant preindustrial values for CO<sub>2</sub> concentration and nitrogen deposition. Transient run is used to mitigate the sharp changes in the environmental conditions between the spinup and normal runs using varying data on CO<sub>2</sub> concentration, nitrogen deposition, and management practices. The normal run simulates the ecosystem development using current meteorology, CO<sub>2</sub>

concentration, and nitrogen deposition values and management practices for a time period of interest.

The user can select from more than 2,000 output variables representing carbon, nitrogen and water stocks and fluxes in different pools in the ecosystems, on a daily, monthly or annual resolution.

# 2.5.2. Modelling workflow

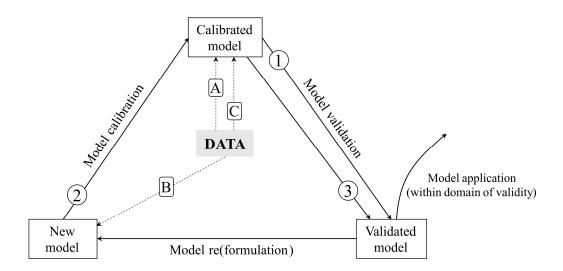
The simplified workflow with the BBGCMuSo model is presented in Figure 2.5. The workflow was based on a concept of model-data fusion (Williams et al. 2009, Guiot et al. 2014), which is an iterative and interactive approach that implies a comparison of model results with measured data, with continuous adjustment of model parameters and/or model processes to obtain satisfactory agreement. When the agreement is found, the model can be considered validated and can be applied.

Process-based models, such as BBGCMuSo, are continuously evolving and are, more or less frequently, being updated with new versions. In this thesis, two versions of the model have been used, namely BBGCMuSo v.4.0 (Hidy et al. 2016a), and BBGCMuSo v.6.2 (Hidy et al. 2022). The main reason for using BBGCMuSo version 4.0 was the fact that at the beginning of the research for this thesis, version 4.0 was published in a peer-reviewed journal, as well as successfully calibrated for a site in Croatia. As such, it could serve as the benchmark version for testing the BBGCMuSo model in a wider forest area in Croatia. The subsequent major model version release (BBGCMuSo v.6.2, Hidy et al. 2022) became available after the beginning of the work on this thesis while, at that time, the publication in a peer-reviewed journal of the description article for the new model version was still pending. The new model version again requires calibration as well as validation for use in forests in Croatia.

The first task in the model workflow (Figure 2.5) was the validation of BBGCMuSo v.4.0 (Hidy et al. 2016a) for estimating SOC in different forest ecosystems spatially distributed across three BGRs in Croatia. This task was performed to test the third hypothesis of this study. In the second and third tasks in the model workflow (Figure 2.5), a new, improved version of the model BBGCMuSo v.6.2 (Hidy et al. 2022) was calibrated for pedunculate oak forest and validated for the estimation of SOC change in a pedunculate oak forest in a ten-year period, respectively. These tasks were performed to test the fourth hypothesis of this study. In each task, model results were compared with the observed data, taking into account the independency

between the observation datasets for calibration and validation phases. Three independent observation datasets were used in the modelling tasks (Figure 2.5):

- (A) Forest ecosystem database (ForecoDB) for task 1,
- (B) Data from the eddy covariance site for task 2,
- (C) Data from the chronosequence experiment for task 3.



**Figure 2.5.** The simplified methodological workflow with the Biome-BGCMuSo model (adjusted from conceptual diagram of the model-data fusion process, Williams et al. 2009). Solid arrows refer to modelling tasks and dashed arrows refer to inflow of dataset. Numbers in circles refer to modelling tasks (1 – validation of Biome-BGCMuSo v.4.0 for estimating soil organic carbon (SOC) stocks in different forest ecosystems spatially distributed across three BGRs in Croatia, 2 – calibration of Biome-BGCMuSo v.6.2 for pedunculate oak forest, 3 – validation of Biome-BGCMuSo v.6.2 for estimating SOC stock change in a pedunculate oak forest during ten years). Letters in squares refer to the observational datasets (A – forest ecosystem database, B – data from the eddy covariance site, C – data from the chronosequence experiment). For each modelling task, the corresponding dataset was used as follows: 1–A, 2–B, 3–C.

#### 2.5.3. Model input files

The model is driven with required input files: initialization (INI), meteorological data (MET), soil properties (SOI), and ecophysiological constants (EPC), and optional input files: management (MGM), groundwater table (GWT), and annually varying values of carbon-dioxide concentration in the atmosphere, nitrogen deposition and mortality.

## Initialization (INI) file

This is the main file that contains general information needed for a single model run. It provides file paths for all necessary input files, a description of the time frame of the simulation, sitespecific parameters, fixed CO<sub>2</sub> and nitrogen deposition values (or optionally paths to the files containing varying CO<sub>2</sub> and nitrogen deposition values), various simulation control flags that indicate specific model routine that will be used for certain processes, the initial state of carbon, nitrogen and water in the simulated ecosystem, and codes of the desired daily and annual output variables. The user needs to provide INI file for each run, one for normal and one for spinup and transient model run together.

INI file in model v.4.0 includes soil properties and management data (Table A.8 and Table A.9), while in model v.6.2, these data are handled separately in SOI (Table A.5) and MGM files.

#### Meteorological data (MET) file

Meteorological data needed for running the model include daily maximum, daily minimum air temperature and average daytime air temperature (in  $^{\circ}$ C), daily precipitation amount (in cm), daylight mean global radiation (in W m<sup>-2</sup>), average daylight vapour pressure deficit (in Pa), and day length, i.e. the length of the light part of the day (in s). Spatially explicit data that fully match these requirements are available for Central Europe within the FORESEE database (Kern et al. 2024). FORESEE v4.0 (Kern et al. 2024) is an open-access gridded meteorological database with a spatial resolution of  $0.1^{\circ} \times 0.1^{\circ}$ , spanning between  $41^{\circ}30'-51^{\circ}30'N$  and  $9^{\circ}-30^{\circ}E$ , and covering the period 1951-2020 based on observations, and the period 2021-2100 based on climate model projections. Meteorological data from FORESEE v4.0 for studied locations was used for creating time-series for all model simulations in this study. For the period before the year 1951, replicating of meteorological data from the period 1951-1970 was performed.

# Ecophysiological constants (EPC) file

EPC file includes parameters grouped into several sections titled: plant functioning, crop-specific, stress and senescence, growing season and phenological (allocation) parameters. EPC files for model versions 4.0 and 6.2 are shown in Table A.3 and Table A.4, respectively.

A major re-arrangement of the EPC file was implemented in BBGCMuSo v.6.2 relative to the earlier versions entailing a substantial number of new parameters. Also, the format of allocation parameters in this model version was changed in comparison to model version 4.0. For the conversion of allocation parameter values from model v.4.0 to values used in model v.6.2, the Microsoft Excel tool (available at <a href="https://nimbus.elte.hu/bbgc/download.html">https://nimbus.elte.hu/bbgc/download.html</a>) was used.

When setting up the EPC files in each modelling task in the study, special attention was given to species-specific C:N ratios in plant parts: leaves, leaf litter after re-translocation and fruit. Within this research, the selected traits were investigated for *Quercus*, *Fagus*, *Pinus*, *Abies* and

Picea genera. The research included the collection of already available C:N data for fresh leaves, laboratory analysis of the collected samples of plant parts, and field sampling and laboratory analysis of plant parts collected in the field for the investigated tree genera. Data on C:N ratios of fresh leaves was obtained for pedunculate oak, pubescent oak (Quercus pubescens Willd.), common beech, Aleppo pine (Pinus halepensis Mill.) and silver fir from ICP plots in Croatia (Jastrebarski lugovi, Lividraga, Poreč, Sljeme, Vrana, Vrbanja) for the period 2011–2019. Litterfall samples of the same tree species specified above, excluding Aleppo pine, were obtained from the samples archive of the Croatian Forest Research Institute (collected with litter traps at ICP plots Jastrebarski lugovi, Poreč, Sljeme and Vrbanja during the year 2020). Fruit samples of common beech, oak tree species (pedunculate oak, pubescent oak, holm oak and sessile oak (Quercus petraea (Matt.) Liebl)), pine tree species (Aleppo pine, Austrian pine, maritime pine), silver fir and Norway spruce were obtained from a personal archive of Prof. Igor Poljak from the Faculty of Forestry and Wood Technology, University of Zagreb, and from a seed archive of the Croatian Forest Research Institute. To increase the C:N dataset, fresh leaves, litter and fruits of some tree species were additionally collected in the field. All provided and collected samples were oven-dried at 80°C and 105°C for 8 and 16 hours, respectively, weighed and analysed on carbon and nitrogen concentrations with CNS-2000 Elemental Analyser (LECO 2000).

EPC files included in the model workflow were based on available literature for a specific forest ecosystem (White et al. 2000, Pietsch et al. 2005, Cienciala and Tatarinov 2006, and Hidy et al. 2016a).

# Soil properties (SOI) file

One of the most important novelties in BBGCMuSo v.6.2, compared to its previous versions, is the extended and detailed soil parameter set, presented in a separate SOI file.

SOI file used in the calibration of the BBGCMuSo v.6.2 is presented in Table A.5. For the majority of soil parameters, proposed values from Hidy et al. (2021) were used. Parameters "maximum height of pond water" and "rate constant scalar of physical fragmentation of coarse woody debris" were modified based on expert judgment.

SOI files used in the validation of BBGCMuSo v.6.2 differ from the SOI file used in the model calibration only in soil texture and pH, which were site-specific (Table A.6).

Site-specific soil texture and pH were set based on field measurements at the eddy covariance site and the chronosequence experiment, conducted in the year 2012. The soil texture was determined following the method by Škorić (1982) by which the soil samples are prepared using

sodium pyrophosphate decahydrate ( $Na_4P_2O_7 \times 10\,H_2O$ ). The soil pH was determined in a solution of 1 mol  $I^{-1}$  potassium chloride (KCl) following the ISO 10390 protocol (ISO 10390 1995). The soil characteristics need to be defined for all soil layers in the model. The soil layer depths in the model v.6.2 (0–3, 3–10, 10–30, 30–60, 60–90, 90–120, 120–150, 150–200, 200–400, 400–1,000 cm) do not exactly correspond to the ones used in the field measurements (0–5, 5–10, 10–20, 20–40 cm), hence the following assumptions and estimations were applied. Site-specific averaged pH values from all measured soil layers were applied to all soil layers in the model equally. For the soil texture values, an assumption was made that the first and the second soil layers in the model corresponded to the first and second soil layers in field measurements, respectively. The soil texture in the third soil layer in the model was calculated as an average of the third and fourth soil depths used in field measurements. Finally, for all deeper soil layers in the model, data for the fourth soil layer from the field measurements were used.

#### Management (MGM) file

Of the optional input files, the management file is the most important one, as it allows the simulation of managed forest ecosystems under different management practices. The user can define the date of the management activity, the rate of thinning, and the amount of removed cut biomass from the ecosystem. MGM files are also used to define the year of the establishment of the forest stand by simulating the final cut at the specific year.

For the calibration and validation of BBGCMuSo v.6.2, the management file was reconstructed using available information on the stand age in each forest compartment (eddy covariance site and chronosequence experiment). The year of the final cut was set as the first year of new (regenerated) stand development, and from that year onward thinning events were set at every 10 years. Thinning rates were estimated as an averaged value from yield tables for pedunculate oak, site yield class II, and for pedunculate oak forest with common hornbeam, site yield class I (Meštrović and Fabijanić 1995). Site yield classes were selected based on available data on growing stock in chronosequence stands including eddy covariance site (Ostrogović 2013). Calculated thinning rates ranged from 27% at the beginning of the stand development to 9% approaching the end of the rotation period of the stand at the age of 140 years. The stand regeneration was simulated as two regeneration cuts, with rates of 50% and 99% for the first (i.e. seed) felling and the second (final) felling, respectively. The first regeneration felling was set five years before the second felling.

#### Groundwater table (GWT) file

GWT file requires daily data. Considering that daily information on the groundwater table was available only for the eddy covariance (EC) site, the GWT file was used only in the calibration of BBGCMuSo v6.2. At the EC site, since 2008 onward, groundwater depth has been measured in piezometers on a weekly or monthly scale (mostly during vegetation season). Together with GWT measurements, soil water content (SWC) is measured at the site with water content reflectometers (CS616, Campbell Sci. Inc., Logan, UT, USA, period 2013–2017) at half-hour intervals. GWT daily time-series was constructed from: 1) measured GWT data during the vegetation period, a gap filled by applying linear interpolation between two consecutive GWT measured values, and 2) estimated GWT values during the period out of vegetation, based on a linear regression between GWT and daily SWC. The obtained GWT series of daily data for the period 2008–2017 were used for the normal run, while daily-averaged GWT depths of all observation years were used for the spinup run.

## Varying atmospheric carbon dioxide concentration and nitrogen deposition files

In transient and normal runs of the model simulation, it is possible to use annual varying CO<sub>2</sub> concentration in the atmosphere and/or nitrogen deposition. Annual time-series for a period specific for each modelling task constructed using yearly atmospheric CO<sub>2</sub> concentration data were used from Mauna Loa Observatory (Mauna Loa Observatory 2023) and ice cores (Etheridge et al. 1996), and yearly atmospheric nitrogen deposition data were estimated from the literature (Churkina et al. 2009).

## 2.5.4. Validation of BBGCMuSo v.4.0 (forest SOC stocks at country level)

## 2.5.4.1. Validation dataset

For the comparison of model results with the measured values in the validation of Biome-BGCMuSo v.4.0, the Forest ecosystem database (ForecoDB) was used.

For this purpose, plots in the ForecoDB were grouped into five forest types: Oak, Beech, Pine, Fir/Spruce, and Forests out of yield by three BGRs (Table A.2). Considering that FOOY is a heterogeneous forest type that includes maquis and shrubs of various tree species, it would be challenging to define suitable ecophysiological parameters to be used in the model, which is why this forest type was excluded from the validation. Also, coniferous forests in Continental BGR had only three plots and were excluded as the representativeness of this stratum would be

highly questionable. Finally, 243 sample plots from the ForecoDB were selected for the model-data comparison.

# 2.5.4.2. Input files

INI files used in the BBGCMuSo v.4.0 model validation with the information about simulations and their time-frames are given in Tables A.8 and A.9, for spinup and normal run, respectively. Site-specific data in the INI file were estimated from field observations (soil texture and maximum rooting depth), as well as from FORESEE v.4.0 (Kern et al. 2024) (mean annual temperature and temperature range), ancillary data (site elevation and latitude), and expert judgment (shortwave albedo). Soil texture was estimated from site-specific National scientific soil survey measurements. As in model v.6.2, the soil layer depths in model v.4.0 (0–10, 10–30, 30–60, 60–100, 100–200, 200–300, 300–1,000 cm) do not exactly correspond to the ones used in the National scientific soil survey (0–10, 10–20, 20–30 cm), the following assumptions and estimations were applied. Soil texture in the first soil layer in the model corresponded to values of the first soil layer in field measurements. Soil texture in the second and all deeper soil layers in the model was estimated as an average of the second and third soil layers from field measurements. Management practices were estimated based on national regulations and statistical yearbooks (Table A.7).

Lists of EPC files for specific forest types, used in BBGCMuSo v.4.0 model validation, are presented in Table A.3. For the Oak forest type, the EPC file was based on the EPC list for the Oak forest published in Hidy et al. (2016a). EPC files for Beech, Pine and Fir/Spruce forest ecosystems were based on the EPC lists from Cienciala and Tatarinov (2006) for the associated genus, together with the EPC list for Oak forest (Hidy et al. 2016a). A further distinction between specific forest types was based on White et al. (2000) and Pietsch et al. (2005).

## 2.5.4.3. Modelling of soil organic carbon (SOC)

Spatial modelling of SOC to the top 30 cm of mineral soil layer (SOC<sub>30</sub>) was performed for 243 sample plots across three BGRs in Croatia and four forest types, i.e., Oak, Beech, Pine and Fir/Spruce using the runMuso function within the RBBGCMuso package (Hollós et al. 2023) in R Statistical Software (v4.1.1; R Core Team 2021).

The spinup phase was simulated for 6,000 years using repeating meteorological time-series created for the period 1900–1999 (see subchapter 2.5.3) together with fixed pre-industrial atmospheric CO<sub>2</sub> concentration of 290 ppm, and N deposition of 0.0002 kg N m<sup>-2</sup> year<sup>-1</sup> for

each year of the simulation. The annual fire mortality rate was set to 0.002 (Ostrogović Sever et al. 2021). The transient run was simulated for 100 years, from 1900 to 1999, using the same meteorology as in the spinup phase, varying yearly atmospheric CO<sub>2</sub> concentration from estimates (1900–1957) (Etheridge et al. 1996) and records (1958–1999) (Mauna Loa Observatory 2023), as well as varying atmospheric nitrogen deposition (Churkina et al. 2009) and forest type-specific management activities (Table A.7). The normal run was simulated for the period 2000-2016 using the observed meteorology for a given period, varying atmospheric CO<sub>2</sub> concentrations (Mauna Loa Observatory 2023) and atmospheric nitrogen deposition (Churkina et al. 2009), and forest type-specific management activities (Table A.7).

A total of 214 successful plot-level simulations for use in the model-data comparison were achieved. For 29 plots the model collapsed, meaning that model runs resulted in zero live biomass at some point during the simulation. One of the possible reasons for the model to collapse could be model sensitivity to nitrogen resulting in the ecosystem's unsuccessful growth during the spinup run due to high vegetation demand for nitrogen and low nitrogen availability at the specific location.

For more details on the technical components of the model and its simulation, please see the User's Guide (Hidy et al. 2016b).

#### 2.5.4.4. Model evaluation

For the evaluation of the BBGCMuSo model for estimating national SOC<sub>30</sub>, forest types were categorised according to Forest land stratification currently used in NIR, i.e. oak and beech forests into deciduous forests stratum, and pine and fir/spruce forests into coniferous forests stratum. Aside from the mandatory land-use stratification, IPCC Guidelines (IPCC 2006) recommend additional land stratification concerning climate region and site characteristics. Therefore, additional stratification, Forest land stratum × BGR was implemented.

A comparison of modelled and measured  $SOC_{30}$  data was performed at three strata: Forest land stratum, Forest land stratum × BGR, and plot using R Statistical Software (v4.1.1; R Core Team 2021).

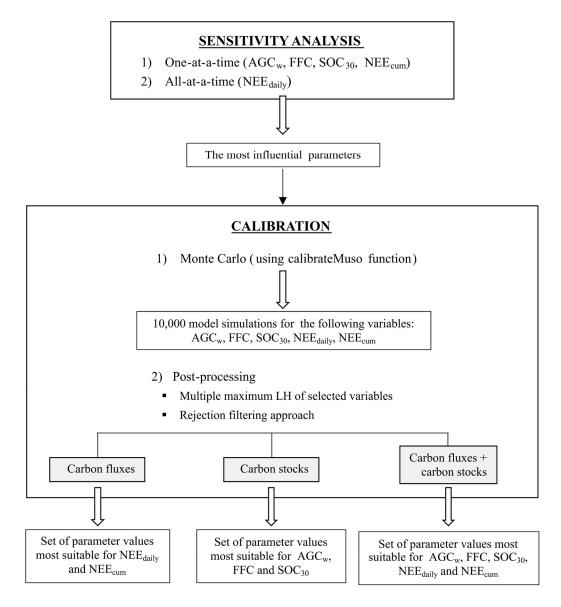
For each Forest land stratum and Forest land stratum  $\times$  BGR, the difference between the measured and modelled SOC<sub>30</sub> was assessed with a paired t-test. The normal distribution of the measured and modelled SOC<sub>30</sub> was checked with the Shapiro-Wilk normality test performed on data grouped according to Forest land stratum. The hypothesis of a normal distribution of measured and modelled SOC<sub>30</sub> had to be rejected (p < 0.05) for all Forest land strata. Therefore, the differences in measured and modelled SOC<sub>30</sub> between different Forest land strata and Forest

land strata  $\times$  BGR were tested using a nonparametric Kruskal-Wallis equality-of-populations rank test with post-hoc Dunn's test of multiple comparisons using rank sums with the Holm-Bonferroni method for the adjustment of the family-wise error rate (Dinno 2015). Furthermore, evaluation of model results was performed using quantitative measures, namely coefficient of determination ( $R^2$ ) of the linear regression (not performed for the Forest land stratum level), mean absolute error (MAE), root mean square error (RMSE) and Nash–Sutcliffe model efficiency (NSE). Finally, analysis of residuals, estimated as the difference between measured and modelled SOC<sub>30</sub> data, was performed at the plot level.

## 2.5.5. Calibration of BBGCMuSo v.6.2 (forest stand carbon stocks and fluxes)

Before the BBGCMuSo v.6.2 calibration, the sensitivity analysis of the model output variables to variations in parameters was performed to support and complement model calibration by revealing the most influential ecophysiological parameters for the selected output variables. The sensitivity analyses and calibration of the Biome-BGC model and its developed versions (including BBGCMuSo) have been primarily focused on high-frequency data, i.e. main ecosystem carbon fluxes (White et al. 2000, Bond-Lamberty et al. 2007, Ren et al. 2022). Nevertheless, the use of long-term carbon stocks of various ecosystem variables has also been applied, but more rarely in comparison to the carbon fluxes (Tatarinov and Cienciala 2006, Hlásny et al. 2014, Merganičová et al. 2024). In this study, a multi-variable approach using datasets with a different temporal resolution was used, namely carbon fluxes and carbon stocks. With this approach, the intention was to capture influential parameters on both carbon fluxes and carbon stocks in the ecosystem.

The simplified methodological workflow of the sensitivity analysis and calibration processes is presented in Figure 2.6.



**Figure 2.6.** Simplified methodological workflow of Biome-BGCMuSo v.6.2 sensitivity analysis and calibration conducted in this study.  $AGC_w$  – Above-ground live wood carbon; FFC – Forest floor carbon;  $SOC_{30}$  – Soil organic carbon in the mineral soil layer down to 30 cm depth;  $NEE_{cum}$  and  $NEE_{daily}$  – Cumulative and daily net ecosystem exchange, respectively; LH – likelihood.

#### 2.5.5.1. *Input files*

The same input files were used in SA and calibration.

INI files are given in Tables A.10 and A.11, for spinup and normal run, respectively.

Site-specific data in the INI file were estimated from Croatian base map (site elevation), meteorological station Jastrebarsko (data for the period 1981–2010) (mean annual air temperature and temperature range), and were based on expert judgment (shortwave albedo).

A priori, i.e. the initial values of the parameters in the EPC file were set. Parameters were based on the EPC list for Oak forest ecosystems (Hidy et al. 2016a), proposed values from a generic

EPC file for deciduous broadleaves forest (available at <a href="http://nimbus.elte.hu/bbgc/index.html">http://nimbus.elte.hu/bbgc/index.html</a>), from Hidy et al. (2021), or on the best expert knowledge and field measurements (Table A.4). SOI file is presented in Table A.5, and a description of MGM and GWT files is given in subchapter 2.5.3.

# 2.5.5.2. Sensitivity analysis

In the Sensitivity Analysis (SA), the attribution of changes in the selected output variables to variations in input ecophysiological parameters was explored.

Out of 122 EPC parameters, 50 parameters related to forest ecosystem were selected to be used in the SA. Those include fully operational parameters (parameters with abbreviation in Table A.4), excluding parameters that are still under development, simulation control flags and flag-related parameters. Additionally, parameters for the allocation of carbon into a specific plant part were excluded from the analysis due to their large intra-seasonal variability.

Parameter ranges were defined based on the previous modelling studies (Hidy et al. 2016a, Merganičová et al. 2024), literature review and model logic (e.g. parameter C:N of leaf litter should be equal to or greater than C:N of fresh leaves) (Table A.4). Additionally, some parameter ranges were narrowed to avoid the model from collapsing, as this would lead to unrealistically high sensitivity index (Table A.4). Parameter ranges defined here were also applied in the next task, model calibration.

SA was performed using two methods: One-[parameter]-At-a-Time (OAT) and All-[parameters]-At-a-Time (AAT) (Pianosi et al. 2016). In the OAT method, the influence of a single parameter on variations in output variables is tested by repeatedly varying only one parameter at a time, while other parameters are kept fixed. On the other hand, in the AAT method, the influence of multiple parameters on variations in output variables is tested by varying all selected parameters at the time. OAT method was used for a quick screening of influential parameters to be selected for the use in AAT.

Modelling settings and *a priori* values of the parameters and their predefined plausible ranges were the same in both SA methods. The spinup phase was simulated for 6,000 years, with repeating meteorology from the period 1900–2017 (see the subchapter 2.5.3), the constant pre-industrial atmospheric CO<sub>2</sub> concentration of 290 ppm, and nitrogen deposition values of 0.0002 kg N m<sup>-2</sup> year<sup>-1</sup> applied for each year. The annual fire mortality rate was set to zero (Hidy et al. 2016). The transient run was simulated for 108 years, from 1900 to 2007, using the same meteorology as in the spinup phase, varying atmospheric CO<sub>2</sub> concentration from estimates (1900–1978) (Etheridge et al. 1996) and records (1979–2007) (Mauna Loa

Observatory 2023), as well as varying atmospheric nitrogen deposition (Churkina et al. 2009) and stand-specific management (see subchapter 2.5.3). The normal run was simulated for the period 2008–2017 using the observed meteorology for a given period, with varying atmospheric CO<sub>2</sub> concentrations (Mauna Loa Observatory 2023) and atmospheric nitrogen deposition (Churkina et al. 2009) and stand-specific management (see subchapter 2.5.3).

# **One-At-a-Time (OAT) Sensitivity Analysis**

The influence of the changes in each of the selected 50 individual parameters was tested on the following output variables: yearly carbon stocks (AGC<sub>w</sub>, FFC, SOC<sub>30</sub>) and carbon flux (NEE<sub>cum</sub>). NEE stands for the Net Ecosystem Exchange and presents the carbon balance between the atmosphere and the ecosystem.

The analysis was performed in R Statistical Software (v4.3.2; R Core Team 2023) using the runMuso function within the RBBGCMuso package to run the simulations in R (Hollós et al. 2023). For each selected parameter, ten model iterations were performed, with the value of the selected parameter being gradually increased in each iteration between the minimum (first iteration) and the maximum (tenth iteration) value defined by plausible ranges. An iteration represents a model simulation including all three phases, i.e. spin up, transient and normal run, with a unique set of parameter values, except, of course, for the selected parameter, for which the value is predetermined for each of the ten iterations.

The sensitivity index (SI) for a parameter (p) was calculated by the contribution of a given parameter to the variability of the output variable, following the equation by Hoffman and Gardner (1983):

$$SI_{p} = \frac{|V_{max} - V_{min}|}{Max\{|V_{min}|, |V_{max}|\}}$$
 (2.12)

where  $V_{max}$  and  $V_{min}$  are the maximum and minimum values of the simulated output variable in the associated measurement unit of the output variable, respectively.

Finally, parameters with a sensitivity index greater than 20% for at least one of the output variables were identified. Out of 21 identified parameters, nine parameters were selected for AAT sensitivity analysis.

#### All-At-a-Time (AAT) Sensitivity Analysis

Aside from selected parameters from OAT, two additional parameters from the phenological (allocation) block, which could not be tested in OAT due to model logic, were included in AAT: Specific leaf area (SLA) and Current growth proportion (CGP).

AAT analysis was performed using the musoMonte and musoSensi functions (Hollós and Barcza 2020) from the RBBGCMuso package (Hollós et al. 2023) in R Statistical Software (v4.3.2; R Core Team 2023) following the approach by Verbeeck et al. (2006). Ten thousand (10,000) Monte Carlo iterations were used to uniformly sample the parameter space within a user-defined range of parameters. Then, BBGCMuSo simulations were performed using generated parameter combinations. Because of the computationally demanding AAT analysis, the AAT was performed for only one variable, daily NEE. This variable was chosen over the carbon stock variables considering that the model uses a daily time step and daily NEE is a high frequency data. The regression-based sensitivity analysis was performed with the musoSensi function by evaluating the outputs of Monte Carlo simulations. The results of this analysis revealed the most influential parameters within the group and these parameters were selected to be used in the model calibration.

#### 2.5.5.3. Calibration dataset

For the comparison of model results with the measured values in the model calibration, data on daily and annual carbon fluxes (NEE<sub>daily</sub> and NEE<sub>cum</sub>) and annual carbon stocks (AGC<sub>w</sub>, FFC and SOC<sub>30</sub>) from the eddy covariance site were used.

A measurement station for monitoring of CO<sub>2</sub> fluxes, an eddy covariance site (EC site), was set up in the year 2007 in a pedunculate oak forest stand in the management unit Jastrebarski lugovi, and since then it has continuously provided meteorological and EC measurements (Marjanović et al. 2011, Anić et al. 2018). Daily and annual NEE data, calculated from EC raw measurement data for the period 2008–2017 (Anić et al. 2018) were used for the model calibration.

Around the EC tower, a network of permanent circular plots was set up in the years 2007 and 2008, out of which 24 plots are in the footprint of the EC tower (Anić et al. 2018). At permanent plots, the diameter at breast height (DBH) and the height of all trees (with DBH > 2 cm) are measured at the end of every growing season and wood volume data is expressed per tree species (Anić et al. 2018). Within this study, tree AGC<sub>w</sub> was calculated by multiplying wood volume with the basic wood density of a given tree species and a carbon fraction of 0.5. Total AGC<sub>w</sub> at the site at the end of each year during the period 2007–2017 was calculated as the sum of all trees at the site.

At the EC site, the forest floor and mineral soil layer (top 30 cm) are sampled in five-year intervals starting in the year 2012. EC site is at the location of one of the permanent plots from the chronosequence experiment (see subchapter 2.5.6.1). The calibration and validation datasets need to be independent, therefore, data on SOC<sub>30</sub> from this plot were excluded from the validation dataset.

#### 2.5.5.4. Calibration

For the model calibration, a Generalized Likelihood Uncertainty Estimation method was used (GLUE; Beven and Binley 2014). GLUE is implemented in the RBBGCMuSo package (Hollós et al. 2023) in R Statistical Software and can be performed with the function calibrateMuso that uses the Monte Carlo technique. With the GLUE method, measured and modelled data for a model output variable and model iteration are compared using the likelihood function that should be defined by the user.

In this study, calibrateMuso function was performed with 10,000 model iterations and for the growing season (80<sup>th</sup>–310<sup>th</sup> day of the year). The winter period was excluded to avoid high disagreement in carbon fluxes estimated from EC measurements and simulated with the model, as the model assumes no Gross Primary Production (GPP) out of the defined growing season (due to a complete lack of photosynthetic capacity, i.e. no leaves), while EC measurements sometimes detected GPP in the winter period, as some of the forest's components are still productive, such as moss. Furthermore, calibrateMuso function, by default, can optimize the model parameters for only one output variable when the spinup phase is included in the process. However, it provides (and stores) Monte Carlo simulation results for all selected output variables with their related unique set of parameter values for each model iteration. This allows the user to optimise the model parameters for any of the selected output variables.

Modelling settings and *a priori* values of the parameters and their predefined plausible ranges were the same as in the sensitivity analysis (see subchapter 2.5.5.2). Therefore, spinup run was always included in simulations, consequently, for initializing calibrateMuso function daily NEE was selected and other variables of interest (AGC<sub>w</sub>, FFC, SOC<sub>30</sub>, and NEE<sub>cum</sub> together with daily NEE) were included in the post-processing.

Likelihood function (LH) for each selected variable (v) and model iteration (i) was defined as follows:

$$LH_{v,i} = e^{-NRMSE_{v,i}} (2.13)$$

$$NRMSE_{v,i} = \frac{_{RMSE_{v,i}}}{\overline{v_{obs_i}}} \tag{2.14}$$

where e is the base of the natural logarithm; NRMSE is normalised root mean square error; RMSE is root mean square error; and  $\overline{v_{obs}}$  is mean of the measured (observed) variable.

The introduction of NRMSE (Eq. 2.13, 2.14 and 2.15) was done to equally account for errors in all selected variables which should provide the best estimate of parameter values considering all selected variables. Post-processing of the data included the calculation of **multiple LH** (multiLH) for different groups (g) of variables (v) that reflect different times-scales: 1) short-term (i.e. daily) and cumulative carbon fluxes, 2) long-term (annual) carbon stocks, and 3) combined daily and cumulative carbon fluxes and annual carbon stocks (Figure 2.6, Eq. 2.15). **Maximum of multiple LH** (max(multiLH)) was calculated for each group (g) of variables (v) and each model iteration (i) as follows:

$$multiLH_{g_{k},i} = \prod_{j=1}^{N_{g_{k}}} LH_{v_{g_{k}},j} = e^{-(\sum_{j=1}^{N_{g_{k}}} NRMSE_{v_{g_{k}},j})}$$

$$max(multiLH_{g_{k},i}), i = 1..10,000$$

$$g_{k} = \{v_{g_{k},1}, ..., v_{g_{k},N_{g_{k}}}\}; N_{g_{k}} = number\ of\ variables\ in\ group\ g_{k}$$

$$k = 1, ..., N_{g}; N_{g} = number\ of\ groups$$

$$g_{1} = \{NEE_{daily}, NEE_{cum}\}$$

$$g_{2} = \{AGC_{w}, FFC, SOC_{30}\}$$

$$g_{3} = \{NEE_{daily}, NEE_{cum}, AGC_{w}, FFC, SOC_{30}\}$$

where LH is a likelihood function, e is the base of the natural logarithm, and NRMSE is a normalized root mean square error.

Furthermore, within the post-processing a **rejection filtering approach** was applied to identify simulations with values of output variables that are within the predefined, reasonable range for variables NEE<sub>daily</sub>, NEE<sub>cum</sub>, AGC<sub>w</sub>, FFC and SOC<sub>30</sub>. To determine plausible ranges in NEE<sub>daily</sub>, NEE<sub>cum</sub> and AGC<sub>w</sub>, measured data from Anić et al. (2018) were used, and for SOC<sub>30</sub> and FFC,

field observations from the pedunculate oak stand at the EC site were used. For daily and cumulative NEE, 5% of the minimum and maximum measured values were calculated and added/subtracted to the corresponding minimum or maximum value of variable. For AGC<sub>w</sub>, due to the age trend, the standard deviation of the minimum and maximum values was added/subtracted from the corresponding minimum or maximum value. For SOC<sub>30</sub> and FFC, two standard deviations of the averaged value from measurements from four subplots and the years 2012 and 2017 were calculated and added/subtracted from the mean value. The following constraints were applied: AGC<sub>w</sub> should be within the range from 5.7 to 11.9 kg C m<sup>-2</sup>; FFC should vary between 0.16 and 0.6 kg C m<sup>-2</sup>; SOC<sub>30</sub> within the range from 6.2 to 10.5 kg C m<sup>-2</sup>;  $NEE_{daily}$  should vary between -0.0105 and 0.0068 kg C  $m^{-2}$  day<sup>-1</sup> and  $NEE_{cum}$  should be between -0.71 and -0.28 kg C m<sup>-2</sup> year<sup>-1</sup>. All 10,000 simulations were identified as feasible (1) or unfeasible (0) from the point of the simulated values of individual output variables, i.e. NEE<sub>daily</sub>, NEE<sub>cum</sub>, AGC<sub>w</sub>, FFC and SOC<sub>30</sub>. The feasible simulation for each output variable is the one where the selected variable has simulated values within its plausible ranges. Furthermore, when looking at the groups of variables (carbon fluxes, carbon stocks, carbon fluxes and carbon stocks combined) model simulation was feasible if all examined variables within a defined group have simulated values within their plausible ranges. Finally, feasible simulations in the groups of variables were sorted by their corresponding multiLHs. Simulation with max multiLH was selected as the best fit for a selected group.

The selected parameter sets were evaluated on measured data for the variables NEE<sub>daily</sub>, NEE<sub>cum</sub>, AGC<sub>w</sub>, SOC<sub>30</sub>, and FFC. Evaluation of model results was performed using quantitative measures, namely coefficient of determination (R<sup>2</sup>) of the linear regression, mean absolute error (MAE), root mean square error (RMSE), the arithmetic mean of differences between modelled and observed values of the respective variable (BIAS) and Nash-Sutcliffe model efficiency (NSE) in R Statistical Software (v4.3.2; R Core Team 2023).

A corresponding parameter set that included both carbon stocks and carbon fluxes was selected as a calibrated parameter set to be used in the validation of BBGCMuSo 6.2.

## 2.5.6. Validation of BBGCMuSo 6.2 (forest stand SOC stocks and SOC stock changes)

# 2.5.6.1. Validation dataset

For the comparison of model results with the measured values in the validation of Biome-BGCMuSo v.6.2, data from the chronosequence experiment were used.

The chronosequence approach (space-for-time-substitution) has been widely used for studying processes in the forest ecosystem (Peltoniemi et al. 2004, Bruckman et al. 2011, De Simon et al. 2012) in which sites of different ages represent points in time. For this study, the chronosequence experiment was primarily used to reveal SOC<sub>30</sub> temporal dynamics and not to resolve its dynamics by stand age. The chronosequence experiment in the management unit Jastrebarski lugovi was established in the year 2010 and consists of eight pedunculate oak forest stands of different ages spanning from 6 to 169 in the year 2012 (Ostrogović 2013, Ostrogović Sever et al. 2019). Two stands were excluded from the model validation: stand 201 located at the eddy covariance site as it is in use in the model calibration (see subchapter 2.5.5.3.) and stand 801, which was declared as a forest of special purpose for scientific research (Ministry of Agriculture, UP/I-321-01/11-01/1, 01/02/2012 in Zagreb) in the year 2012 by which it is protected and excluded from the regular forest management anymore. Also, stand 801 is 181 years old (in the year 2024) and was under different management practices in comparison to the other chronosequence stands, i.e. for acorn feed of wild game (pannage). This management practice resulted in the different habitus of oak trees (wider canopy, lower heights and thicker trunks) in comparison to the trees that are managed for timber production in other chronosequence stands.

In each stand, four permanent circular plots were set up in line with the Terrestrial Carbon Observation (TCO) protocol (GTOS 2008) (Figure 2.7). On permanent plots, above-ground biomass is measured yearly and the forest floor and a mineral layer of the soil (top 40 cm) are sampled in 5-year intervals.

# Field sampling

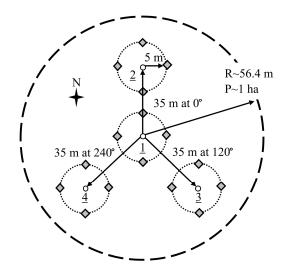
Within this research, sampling of the mineral layer of the soil was conducted in spring 2022 as the third measurement soil sampling campaign in the chronosequence experiment (sampling years: 2012, 2017\*\*\*, 2022).

The mineral soil layer was sampled at four positions on each permanent subplot, 5 m away from the plot centre in the directions North, East, South, and West (Figure 2.7). Sampling was performed with a split tube sampler (Eijkelkamp, Giesbeek, The Netherlands) and each soil core was cut into four samples according to predefined depths (0–5, 5–10, 10–20, and 20–40 cm) (Figure 2.8), to be consistent with previous sampling campaigns. Exceptionally, on north positions, the soil core was cut into five samples with depths 0–5, 5–10, 10–20, 20–30,

<sup>\*\*\*</sup> The author also collaborated on field sampling and laboratory analysis in this sampling year.

and 30–40 cm to improve the accuracy of SOC estimates in line with IPCC (2006), which stipulated reporting for the top 30 cm of the mineral soil layer.

Volume correction, regarding the sample compression that occurred during the soil sampling with the instrument, was performed in the field. Compression (in cm) of the soil core was estimated by subtracting the measured length of the sampled soil core (in cm) from the measured soil pit after the soil core was extracted (in cm). For example, if the length of the extracted soil core was 39 cm and the depth of the corresponding soil pit was 40 cm, the compression of the soil sample (soil core) was estimated to be 1 cm. The assumption is that the compression mostly happens in the upper, less dense soil layers. Therefore, the observed compression was attributed equally to the first two soil layers and the position of the soil core cutting was adjusted accordingly. Consequently, volume correction, due to sample compression, was done for layers 0–5 cm and 5–10 cm depths. Based on the above example where soil core compression was 1 cm, the cut of the samples representing 0–5 cm and 5–10 cm soil layers were made at 4.5 and 9 cm from the top of the sampled soil core, respectively. The soil samples for the layers 10–20 cm and 20–40 cm (or 20–30 cm and 30–40 cm and the north position) were cut after the second layer in corresponding lengths of 10 or 20 cm.





**Figure 2.7.** Field plot layout of subplots (circle with dotted line) and positions of soil sampling (diamonds) in chronosequence stand (dashed circle), based on Terrestrial Carbon Observation (TCO) protocol (GTOS 2008).

**Figure 2.8.** Soil core sample with defined depths of soil samples.

In total, 544 samples were collected in the 2022 sampling campaign (8 stands  $\times$  4 subplots  $\times$  (3 positions  $\times$  4 layers + 1 position (North)  $\times$  5 layers)). Samples were placed in plastic bags and stored in a cool place on the same day.

#### Laboratory analysis

Fine roots ( $d \le 2$  mm) and coarse roots (d > 2 mm) were separated from the soil samples using a 2 mm sieve, oven-dried at 80°C and 105°C for 8 and 16 hours, respectively, and then weighed. It should be noted that the soil at the experimental site did not contain coarse fragments (stones or rocks) in the sampled soil layers in the majority of stands with the exception of the forest stand 801. The soil samples were then ground using a laboratory mill, oven-dried at 40°C for 48 hours and weighed.

From the total of 68 samples per chronosequence stand, the carbon fraction was analysed for 24 samples, 16 composite and 8 original ones. For soil layers, 0–5 cm, 5–10 cm, and 10–20 cm composite samples were formed from all sampling positions (North, East, South, West), while for soil layers 20–40 cm composite samples were formed from East, South and West positions. Original samples were used for soil layers 20–30 cm and 30–40 cm from North positions. Carbon fraction was analysed with a CNS-2000 Elemental Analyser (LECO 2000).

# **Calculation of soil organic carbon stocks**

Soil organic carbon (SOC) for every horizon (i) in the mineral soil layer was calculated as follows:

$$SOC_i = CF_i \cdot BD_i \cdot D_i \cdot (1 - VP_{cf,i}) \tag{2.16}$$

where  $SOC_i$  is the soil organic carbon, in t C ha<sup>-1</sup>;  $CF_i$  is a carbon fraction of the soil, in %;  $BD_i$  is soil bulk density, in g cm<sup>-3</sup>; and  $D_i$  is the depth of the soil layer, in cm;  $VP_{cf}$  is the volume proportion of coarse fragments, in m<sup>3</sup> m<sup>-3</sup>.

For each subplot,  $SOC_{40}$  (0–40 cm) was calculated cumulatively by soil layer depths, 0–5 cm, 0–10 cm, 0–20 cm and 0–40 cm. For each stand,  $SOC_{40}$  was calculated as an average value of four subplots.

To be comparable with modelled SOC<sub>30</sub> (0-30 cm), cumulative SOC<sub>30</sub> was calculated as an average of cumulative values of SOC in cumulative soil layers 0–20 cm and 0–40 cm in each stand. SOC stock change was calculated for a ten-year period (2012–2022) for six stands, excluding the stand at the eddy covariance site (201) and the old-growth oak stand (801).

## 2.5.6.2. Input files

Site-specific data in the INI file were the same as the ones used in the INI file in the model calibration (for details see subchapter 2.5.5.1).

The EPC file was based on the optimized EPC list obtained within the calibration phase in this research (Table A.4).

A description of SOI and MGM files is given in subchapter 2.5.3.

# 2.5.6.3. Modelling of soil organic carbon (SOC) change

Modelling of SOC to the top 30 cm of mineral soil layer (SOC<sub>30</sub>) was performed for six pedunculate oak stands in the chronosequence experiment using the runMuso function within the RBBGCMuso package (Hollós et al. 2023) in R Statistical Software (v4.3.2; R Core Team 2023). The spinup phase was simulated for 6,000 years using the repeating meteorology from the period 1820–2011 (see subchapter 2.5.3), together with a constant pre-industrial atmospheric CO2 concentration of 290 ppm, and nitrogen deposition of 0.0002 kg N m<sup>-2</sup> year<sup>-1</sup>. The annual fire mortality rate was set to zero (Hidy et al. 2016). The transient run was simulated for 192 years, from 1820 to 2011, using the same meteorology as in the spinup phase, varying yearly atmospheric CO<sub>2</sub> concentration from estimates (1820–1978) (Etheridge et al. 1996) and records (1978–2011) (Mauna Loa Observatory 2023), varying atmospheric nitrogen deposition (Churkina et al. 2009) and stand-specific management activities (see subchapter 2.5.3). The normal run was simulated for a ten-year period (2012–2022) using the observed meteorology for a given period, varying atmospheric CO<sub>2</sub> concentrations (Mauna Loa Observatory 2023), atmospheric nitrogen deposition (Churkina et al. 2009) and stand-specific management activities (see subchapter 2.5.3).

#### 2.5.6.4. Model evaluation

Statistical analyses were performed in R Statistical Software (v4.3.2; R Core Team 2023) with the significance level (alpha value) in statistical testing set to 0.05. The analyses were performed for SOC<sub>30</sub> and SOC<sub>30</sub> changes. Firstly, the normal distribution and the homogeneity of variance of measured SOC<sub>30</sub> stocks by forest stands and sampling years were checked with the Shapiro-Wilk normality test and Barlett's test, respectively. The hypotheses of both tests were accepted (p > 0.05) for all forest stands. Considering that the soil sampling was performed at specific location at each plot in three consecutive years, the soil samples collected in three measurement years were considered dependent. Therefore, the analysis of the difference in measured SOC<sub>30</sub> in a specific forest stand between the sampling years (2012, 2017, 2022) was performed using the repeated ANOVA measures with a post-hoc pairwise t-test with the Bonferroni adjustment method. A comparison of measured and modelled SOC<sub>30</sub> at the stand level and sampling year

was assessed with a paired t-test. To test the fourth hypothesis of this study, short-term (period 2012–2022) trends in modelled and measured SOC<sub>30</sub> at the forest level (all chronosequence stands combined) were investigated. Additionally, long-term (the rotation period of the pedunculate oak stand in Croatia, 140 years) trends in modelled and measured SOC<sub>30</sub> at the forest level were investigated in two ways: 1) a single year chronosequence approach by observing the data from each sampling year separately (2012, 2017, 2022) and 2) repeated chronosequence approach by combining the data from all three sampling years.

# 3. RESULTS

# 3.1. Dead wood study

## 3.1.1. Species-specific dead wood volume-to-carbon conversion factors

Dead wood basic density for investigated tree species by decay class is presented in Figure 3.1.

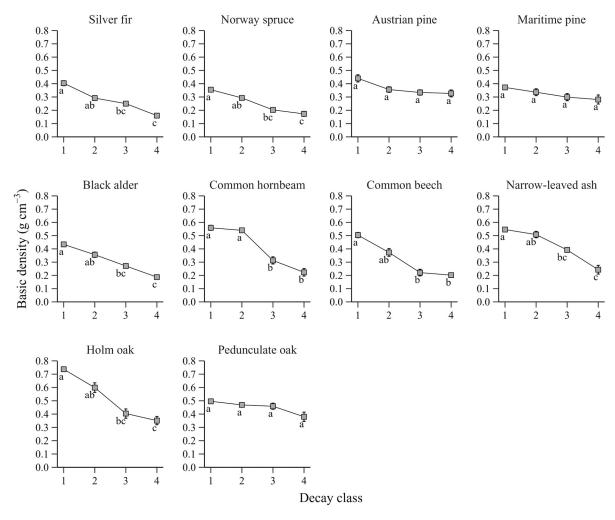


Figure 3.1. Dead wood basic density (mean  $\pm$  s.e.) by decay class for silver fir (*Abies alba* Mill.), Norway spruce (*Picea abies* (L.) Karst.), Austrian pine (*Pinus nigra* Arnold), maritime pine (*Pinus pinaster* Aiton), black alder (*Alnus glutinosa* (L.) Gaertn.), common hornbeam (*Carpinus betulus* L.), common beech (*Fagus sylvatica* L.), narrow-leaved ash (*Fraxinus angustifolia* Vahl), holm oak (*Quercus ilex* L.) and pedunculate oak (*Quercus robur* L.). Different lowercase letters next to data points indicate a statistically significant difference (p < 0.05) of dead wood basic density between different decay classes for a given tree species. The number of samples per tree species and decay class is nine.

For the majority of tree species, DW basic density exhibited a decreasing trend with the increasing decay class. The exceptions were Austrian pine, maritime pine and pedunculate oak, for which no difference in basic density between decay classes was observed (Figure 3.1). Overall, the highest DW basic density (mean  $\pm$  s.e., for the average of decay classes 1 to 4) was observed for holm oak  $(0.523 \pm 0.03 \text{ g cm}^{-3})$  and the lowest one was reported for Norway spruce  $(0.256 \pm 0.014 \text{ g cm}^{-3})$ .

When looking at the tree species groups, in both broadleaves and conifers, a decreasing trend of DW basic density with increasing decay class was observed (Table 3.1). Conifers, on average, had significantly lower basic density than broadleaves for decay classes 1–3, while in decay class 4, no statistically significant difference was observed between these two groups (Table 3.1).

**Table 3.1.** Basic density, carbon fraction (CF) and carbon density (CD) of dead wood by decay class for different tree species groups, broadleaves and conifers (mean  $\pm$  s.e.).

Decay		BROADLEAVES				CONIFERS			
class	N	Basic density (g cm <sup>-3</sup> )	<b>CF</b> (%)	<b>CD</b> (t C m <sup>-3</sup> )	N	Basic density (g cm <sup>-3</sup> )	<b>CF</b> (%)	<b>CD</b> (t C m <sup>-3</sup> )	
1	54	$0.546 \pm 0.014^{a\pi}$	$47.78 \pm 0.16^{a\pi}$	$0.260 \pm 0.006^{a\pi}$	36	$0.393 \pm 0.010^{ap}$	$50.76 \pm 0.71^{ap}$	$0.199 \pm 0.006^{ap}$	
2	54	$0.474 \pm 0.015^{\mathrm{b}\pi}$	$47.42 \pm 0.20^{a\pi}$	$0.224 \pm 0.007^{\mathrm{b}\pi}$	36	$0.319 \pm 0.011^{\mathrm{bp}}$	$50.23 \pm 0.32^{abp}$	$0.161 \pm 0.00^{\mathrm{bp}}$	
3	54	$0.343 \pm 0.015^{c\pi}$	$47.56 \pm 0.20^{a\pi}$	$0.163 \pm 0.007^{c\pi}$	36	$0.272 \pm 0.013^{\rm bcp}$	$50.42 \pm 0.28^{abp}$	$0.137 \pm 0.006^{bc\rho}$	
4	54	$0.264 \pm 0.015^{\mathrm{d}\pi}$	$47.82 \pm 0.33^{a\pi}$	$0.127 \pm 0.007^{\mathrm{d}\pi}$	36	$0.235 \pm 0.016^{c\pi}$	$51.26 \pm 0.40^{bp}$	$0.121 \pm 0.09^{c\pi}$	

NOTE: Different lowercase Latin letters in columns indicate a statistically significant difference (p < 0.05) in a given trait between decay classes of a given tree species group. Different lowercase Greek letters in rows indicate a statistically significant difference (p < 0.05) for a given trait between tree species groups in a given decay class.

Carbon fraction, on the other hand, showed no trend along the decay class for the majority of tree species (Figure 3.2), except for Norway spruce for which CF slightly increased with the increasing decay class (slope  $\pm$  s.e. was  $1.1\% \pm 0.2\%$  per decay class, p < 0.001). Within the conifers tree species group, no significant difference in CF (mean  $\pm$  s.e., for the average of decay classes 1 to 4) was observed between different tree species, while within the broadleaves group, the highest CF was observed in common beech (49.00%  $\pm$  0.12%) and the lowest CF in holm oak (46.25%  $\pm$  0.44%) DW samples. Furthermore, CF in broadleaves showed no trend along the decay class, while in conifers, an increasing trend with the increasing decay class was observed (Table 3.1), probably led with the same pattern observed in the Norway spruce (Figure 3.2). Moreover, CF was found to be consistently higher across all decay classes in the conifers compared to the broadleaves (Table 3.1), with the average (mean  $\pm$  s.e., decay classes 1–4) CF of 50.67%  $\pm$  0.23% and 47.65%  $\pm$  0.12% for conifers and broadleaves, respectively.

Carbon density, as a product of basic density and CF, showed the same trend as basic density, considering that CF in general showed no trend with decay class (Figure A.1, Table 3.1).

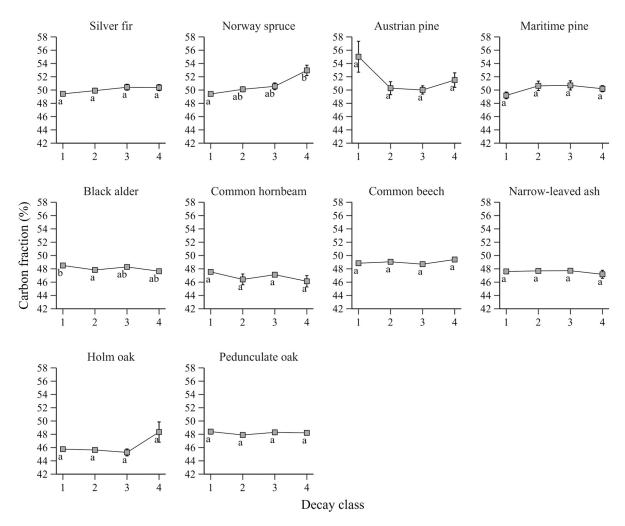
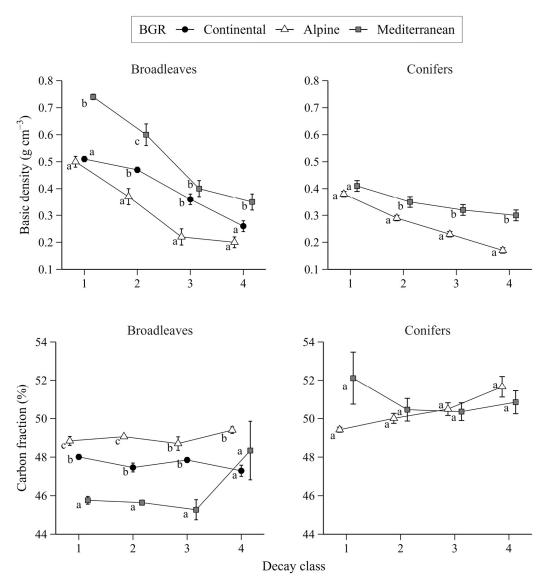


Figure 3.2. Carbon fraction of dead wood (mean  $\pm$  s.e.) by decay class for silver fir (*Abies alba* Mill.), Norway spruce (*Picea abies* (L.) Karst.), Austrian pine (*Pinus nigra* Arnold), maritime pine (*Pinus pinaster* Aiton), black alder (*Alnus glutinosa* (L.) Gaertn.), common hornbeam (*Carpinus betulus* L.), common beech (*Fagus sylvatica* L.), narrow-leaved ash (*Fraxinus angustifolia* Vahl), holm oak (*Quercus ilex* L.) and pedunculate oak (*Quercus robur* L.). Different lowercase letters next to data points indicate a statistically significant difference (p < 0.05) in dead wood carbon fraction between different decay classes for a given tree species. The number of samples per tree species and decay class is nine.

The analysis of volume-to-carbon conversion factors per tree species and decay class in general showed no significant difference (p < 0.05) between diameter classes. The exception for basic density is in common beech in decay class 2 in which the difference between diameter classes 10-20 cm and 20-30 cm was observed (p < 0.05). Similarly, CF differed solely in pedunculate oak DW samples (decay class 3) between diameter classes 5-10 cm and 10-20 cm and in silver fir (decay class 4) between diameter classes 10-20 cm and 20-30 cm (p < 0.05).

Further disaggregation of DW basic density and carbon fraction data according to biogeographical regions and testing for differences (hypothesis H1) confirmed the existence of significant differences (p < 0.05) in these traits between BGRs for some tree species groups and decay classes (Figure 3.3).



**Figure 3.3.** Dead wood basic density and carbon fraction by decay classes, for broadleaves and conifers in different biogeographical regions (BGRs) (mean  $\pm$  s.e.). Different lowercase letters next to data points indicate a statistically significant difference (p < 0.05) in a given trait between different biogeographical regions for a given tree species group and decay class.

The highest DW basic density was observed in the Mediterranean BGR for both broadleaves and conifers for the majority of decay classes (Figure 3.3). The exceptions are in the broadleaves for decay class 3 in which basic density in DW samples from the Mediterranean BGR did not significantly differ from the Continental BGR (Figure 3.3) and in the conifers for decay class 1 in which no significant difference in basic density between different BGRs was

observed (Figure 3.3). Moreover, the DW basic density of broadleaves was significantly higher in the Continental BGR than in the Alpine BGR in decay classes 2 and 3 (Figure 3.3). Carbon fraction in DW samples of broadleaves was significantly higher in the Alpine BGR in the majority of decay classes in comparison to the other BGRs (Figure 3.3), with the exception of decay class 3 in which no significant difference was observed between the Alpine and Continental BGRs (Figure 3.3). In DW samples of the coniferous tree species, no significant difference was observed in CF in any of the decay classes between different BGRs (Figure 3.3).

# 3.1.2. Implications of the use of dead wood carbon densities by decay class for the estimation of national dead wood carbon stocks

The first aim of this thesis was to provide national DW volume-to-carbon conversion factors by decay classes for use in the estimation of national DW carbon stocks, and in this chapter the potential for their use in the compilation of the country's NIR will be presented.

Carbon density conversion factors by tree species groups that correspond to Forest land stratification in the Croatian NIR and by decay classes compatible with CroNFI (for details see subchapter 2.3.4) are presented in Table 3.2.

When we look at the individual decay class level, carbon density differed between all three Forest land strata except for decay class 3<sub>NFI</sub> where only CD of FOOYS differed from CD of the other two groups (Table 3.2). Furthermore, at the individual strata level, CD was significantly different between all decay classes in the Forest land strata DFS and CFS, while in FOOY only CD of decay classes 1<sub>NFI</sub> significantly differed from CD in higher decay classes (Table 3.2).

**Table 3.2.** Carbon density (CD) of dead wood by decay classification scheme used in CroNFI for different Forest land strata (DFS – deciduous forests stratum, CFS – coniferous forests stratum, FOOYS – forests out of yield stratum) (mean  $\pm$  s.e.).

CroNFI	Deciduous broadleaves (DFS)			Conifers (CFS)	Holm oak (FOOYS)		
decay class	N	<b>CD</b> (t C m <sup>-3</sup> )	N	<b>CD</b> (t C m <sup>-3</sup> )	N	<b>CD</b> (t C m <sup>-3</sup> )	
$1_{ m NFI}$	45	$0.244 \pm 0.004^{a\pi}$	36	$0.199 \pm 0.006^{ap}$	9	$0.337 \pm 0.004^{a\tau}$	
$2_{NFI}$	90	$0.187 \pm 0.006^{b\pi}$	72	$0.149 \pm 0.005^{\mathrm{bp}}$	18	$0.229 \pm 0.016^{b\tau}$	
$3_{NFI}$	45	$0.118 \pm 0.008^{c\pi}$	36	$0.121 \pm 0.009^{c\pi}$	9	$0.171 \pm 0.018^{bp}$	

NOTE: Different lowercase Latin letters in columns indicate statistically significant differences (p < 0.05) in CD between decay classes of given strata. Different lowercase Greek letters in rows indicate statistically significant differences (p < 0.05) in CD between strata in a given decay class.

The total DW volume by Forest land stratum and decay class is presented in Table 3.3. It is important to keep in mind that the highest values of the total DW volume observed in decay class 1<sub>NFI</sub> are a result of the fact that standing DW volume in the first CroNFI refers only to decay class 1, therefore all SDW volume is included under decay class 1<sub>NFI</sub>. The CD values from Table 3.2 were used together with total DW volumes for a new estimate of the national DW carbon stocks (DWSC2) presented in Table 3.3. The comparison of DW carbon stocks estimated in this study (DWCS2), with those currently reported in the Croatian NIR (DWCS1, Table 2.3) revealed that the DW carbon stocks in the Croatian NIR are overestimated by 26.6%, 16.8% and 11.1% for deciduous, coniferous and forests out of yield strata, respectively.

**Table 3.3.** Total dead wood (TDW) volume and dead wood carbon stocks per hectare calculated using total dead wood volume and carbon densities from this study and by decay classes used in CroNFI (DWCS2), according to Forest land stratification (DFS – deciduous forest stratum, CFS – coniferous forest stratum and FOOYS – forests out of yield stratum).

	CroNFI	Forest lan	d stratum u	sed in NIR
	decay class	DFS	CFS	FOOYS
<b>TDW</b> (m <sup>3</sup> ha <sup>-1</sup> )	$1_{ m NFI}$	9.51	8.21	1.01
	$2_{NFI}$	2.49	3.53	0.12
	$3_{NFI}$	3.88	5.50	0.19
DWCS2 (t C ha <sup>-1</sup> )	$1_{ m NFI}$	2.32	1.63	0.34
	$2_{NFI}$	0.47	0.53	0.03
	$3_{NFI}$	0.46	0.67	0.03
	All	3.25	2.83	0.40

## 3.2. Forest floor study

#### 3.2.1. Tree species group-specific forest floor carbon stocks

Analysis of forest floor carbon stocks (FFCS) from a new Foreco database regarding tree species groups revealed that FFCS are significantly higher (p < 0.05) for conifers in comparison to the broadleaves, but with similar variability (Table 3.4).

At the BGR scale and for the broadleaves and conifers combined, FFCS showed high variability (CV > 30%), with the Mediterranean BGR having the highest (CV of 45%) and the Alpine BGR having the lowest (CV of 32%) variability. Average FFCS (with s.e.) were  $4.36 \pm 0.13$  t C ha<sup>-1</sup>,  $5.08 \pm 0.22$  t C ha<sup>-1</sup> and  $5.25 \pm 0.31$  t C ha<sup>-1</sup> for the Continental, Alpine and Mediterranean

BGR, respectively. The significantly lower (p < 0.05) FFCS were found in the Continental BGR, in comparison to the other two BGRs.

Separation of FFCS data according to tree species groups and testing for differences between biogeographical regions (hypothesis H2) confirmed the existence of significant difference (p < 0.05) in FFCS between BGRs (Table 3.4). At the tree species group level, FFCS in broadleaves were significantly higher in the Alpine BGR in comparison to the Continental BGR, while in conifers, a significantly higher FFCS were observed in the Mediterranean BGR in comparison to the Alpine BGR (Table 3.4). When looking at a specific BGR, a significant difference in FFCS between tree species groups was observed only in the Mediterranean BGR (Table 3.4), with higher FFCS observed in conifers than in broadleaves (Table 3.4). Overall, conifers in the Mediterranean BGR have the highest (7.92 t C ha<sup>-1</sup>) and broadleaves in Continental BGR the lowest (4.32 t C ha<sup>-1</sup>) FFCS (Table 3.4).

**Table 3.4.** Comparison of forest floor carbon stocks (FFCS) (mean  $\pm$  s.e.) and coefficient of variance (CV) by biogeographical regions for tree species groups.

Biogeographical		Broadleaves			Conifers			
region	N	FFCS (t C ha <sup>-1</sup> )	CV (%)	N	FFCS (t C ha <sup>-1</sup> )	<b>CV</b> (%)		
Continental	156	$4.32 \pm 0.13^{a\pi}$	38	4	$6.05 \pm 1.04^{\mathrm{ab}\pi}$	34		
Alpine	32	$5.19 \pm 0.29^{\mathrm{b}\pi}$	32	24	$4.93 \pm 0.33^{a\pi}$	32		
Mediterranean	44	$4.40 \pm 0.27^{ab\pi}$	41	14	$7.92 \pm 0.56^{bp}$	26		
Total	232	$\textbf{4.45} \pm \textbf{0.11}^{\Pi}$	38	42	$6.03 \pm 0.35^{P}$	37		

NOTE: Different lowercase Latin letters in columns indicate statistically significant differences (p < 0.05) in FFCS between biogeographical regions of a given tree species group. Different lowercase Greek letters in rows indicate a statistically significant difference (p < 0.05) in FFCS between tree species groups in a given biogeographical region. Different uppercase Greek letters in rows indicate a statistically significant difference (p < 0.05) in total FFCS between tree species groups. CV = coefficient of variance. Data source: National scientific soil survey and OKFŠ project (see subchapter 2.4.1).

For both tree species groups FFCS showed a significant correlation with elevation, MAT and MAP (Table 3.5). Overall, the correlations were higher in conifers than in broadleaves, with the highest correlation observed in FFCS with the MAT for conifers (Table 3.5).

**Table 3.5**. The response of forest floor carbon stocks to the selected environmental variables in a given tree species group.

Variable		Broadle	eaves	Conifers			
Variable	N R <sup>2</sup> Coefficient		N	$\mathbb{R}^2$	Coefficient		
Elevation (m a.s.l.)		0.10***	Positive		0.17**	Negative	
MAT (°C)	232	0.05***	Negative	42	0.35***	Positive	
MAP (mm)		0.03**	Positive		0.20**	Negative	

NOTE: \*p < 0.05, \*\*p < 0.01, \*\*\* p < 0.001. Data source: National scientific soil survey and OKFŠ project (see subchapter 2.4.1).

## 3.2.2. Implication of the new forest floor carbon stocks estimates for the use in the Croatian NIR

The results from this study can facilitate direct (immediate) and potential (future) enhancement of net CO<sub>2</sub> emission/removal estimates from forest floor carbon pool under LC-FL subcategory of Forest land within the Croatian NIR. A new estimate of national FFCS (FFCS2) for the total area of the Republic of Croatia is 4.81 t C ha<sup>-1</sup>, estimated as a weighted mean of FFCS2 within the forest area in each specific BGR, is 5% higher in comparison to the value of 4.57 t C ha<sup>-1</sup> currently used in the Croatian NIR (HR NIR 2023). The use of new carbon stock change (CSC) factor of 0.2405 t C ha<sup>-1</sup> year<sup>-1</sup>, compared to the current one of 0.2285 t C ha<sup>-1</sup> year<sup>-1</sup> (HR NIR 2023), would increase the reported net CO<sub>2</sub> removal from this pool under subcategory LC-FL.

Apart from this direct enhancement, which can immediately be applied into the calculation of the next Croatian NIR, new CSC factors by Forest land stratification used in the Croatian NIR (Table 3.6) can potentially be used in the future NIR calculations, after the spatially-explicit land use-change matrix within the Croatian NIR will be available. The use of new CSC factors (Table 3.6) would increase the estimate of the current net CO<sub>2</sub> removal if land use conversion would occur in the coniferous forest stratum (CFS). On the other hand, it would decrease the reported net CO<sub>2</sub> removal if land use conversion would occur in the deciduous forests or forests out of yield strata (DFS and FOOYS, respectively).

**Table 3.6.** Forest floor carbon stocks (FFCS) and carbon stock change (CSC) factors (20-year conversion period) (mean  $\pm$  s.e.) with respect to Forest land strata used in the Croatian NIR.

Forest land stratum used in NIR	N	<b>FFCS</b> (t C ha <sup>-1</sup> )	CSC factor (t C ha <sup>-1</sup> year <sup>-1</sup> )
Deciduous forests	207	$4.50 \pm 0.11^{a}$	$0.2251 \pm 0.0057^{a}$
Coniferous forests	42	$6.03 \pm 0.35^{b}$	$0.3016 \pm 0.0173^{b}$
Forests out of yield	25	$4.04 \pm 0.40^{a}$	$0.2020 \pm 0.0200^{a}$

NOTE: Different lowercase letters in columns indicate statistically significant differences (p < 0.05) in FFCS and CSC factor between different Forest land strata. Data source: National soil survey and OKFŠ project (see subchapter 2.4.1).

In addition, new CSC factors (Table 3.6) could also facilitate the reporting of net carbon stock change in FFC pool under FL-FL subcategory, as soon as activity data (area) for this subcategory would be reported. FFCS significantly differed between CFS and other two Forest land strata, i.e. DFS and FOOYS (Table 3.6). The most probable conversions between three forest strata are: from CFS to DFS and from FOOYS to CFS or DFS. CSC factor that could be used for conversion of CFS to DFS would be -0.0765 t C ha<sup>-1</sup> year<sup>-1</sup>, and it would result with

the increase in net CO<sub>2</sub> emissions from FFC pool. On the other hand, for the conversion of FOOYS to CFS or DFS, CSC factors that could be used would be 0.0996 t C ha<sup>-1</sup> year<sup>-1</sup> and 0.0231 t C ha<sup>-1</sup> year<sup>-1</sup>, respectively, and they would both result with the increase in the reported net CO<sub>2</sub> removals from FFC pool.

### 3.3. Soil organic carbon study

#### 3.3.1. Validated BBGCMuSo v.4.0 (forest SOC<sub>30</sub> stocks at the country level)

At the Forest land strata level, the model showed good performance (Table 3.9). For both deciduous and coniferous forests, no significant difference (p < 0.05) between modelled and measured SOC<sub>30</sub> stocks was observed (Table 3.7). Nevertheless, high coefficient of variance was observed for the measured and modelled SOC<sub>30</sub> stocks, with the measured SOC<sub>30</sub> stocks having higher variability for both Forest land strata in comparison to the modelled SOC<sub>30</sub> stocks (Table 3.7). Overall, in both modelled results and measured data, the coniferous forest stratum had significantly higher SOC<sub>30</sub> stocks (p < 0.05) in comparison to the deciduous forest stratum (Table 3.7).

**Table 3.7.** Variability in measured and modelled soil organic carbon stocks in the mineral soil layer down to 30 cm depth ( $SOC_{30}$ ) across different Forest land strata used in the Croatian NIR.

Forest land		Measured SO	C30	Modelled SOC	30
stratum used in NIR	Ν -	Mean $\pm$ s.e. (t C ha <sup>-1</sup> )	CV (%)	Mean $\pm$ s.e. (t C ha <sup>-1</sup> )	<b>CV</b> (%)
Deciduous forests	176	$65.34 \pm 1.96^{a\pi}$	40	$63.20 \pm 1.22^{a\pi}$	26
Coniferous forests	38	$77.99 \pm 5.73^{\mathrm{b}\pi}$	45	$80.86 \pm 3.13^{\mathrm{b}\pi}$	24

NOTE: Different lowercase Latin letters in columns indicate statistically significant differences (p < 0.05) in measured and modelled SOC<sub>30</sub> between Forest land strata. Different lowercase Greek letters in rows indicate a statistically significant difference (p < 0.05) between measured and modelled SOC<sub>30</sub> in a given Forest land stratum. CV = coefficient of variance. Data source for measured SOC<sub>30</sub>: National scientific soil survey and OKFŠ project (see subchapters 2.4.1 and 2.5.4.1).

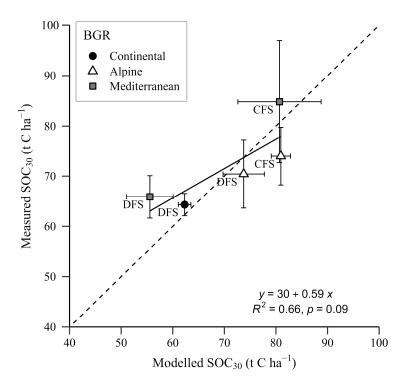
Further disaggregation of the results by Forest land strata  $\times$  BGR still revealed a good agreement between the measured and the modelled data (Figure 3.4, Table 3.8), although not statistically significant (Figure 3.4). For all Forest land strata  $\times$  BGR, there was no significant difference (p < 0.05) between the modelled and the measured SOC<sub>30</sub> stocks (Table 3.8), although there are indications that the model is underestimating SOC<sub>30</sub> stocks in the Mediterranean BGR and overestimating SOC<sub>30</sub> in the Alpine BGR (Figure 3.4). Overall, measured SOC<sub>30</sub> stocks again showed very high variability (28–53%) and no significant difference (p < 0.05) in specific Forest land strata between different BGRs (Table 3.8).

Modelled SOC<sub>30</sub> stocks showed lower variability (11–37%) compared to the measured SOC<sub>30</sub>, and a significant difference (p < 0.05) was observed for deciduous forests in the Alpine BGR, having higher SOC<sub>30</sub> stocks in comparison to Continental and Mediterranean BGRs (Table 3.8). The range of SOC<sub>30</sub> stocks was slightly higher in the modelled data compared to the measured data,  $56-81 \text{ t C ha}^{-1}$  and  $64-85 \text{ t C ha}^{-1}$ , respectively.

**Table 3.8.** Variability in measured and modelled soil organic carbon stocks in the mineral soil layer down to 30 cm depth ( $SOC_{30}$ ) across different Forest land strata in NIR and biogeographical regions.

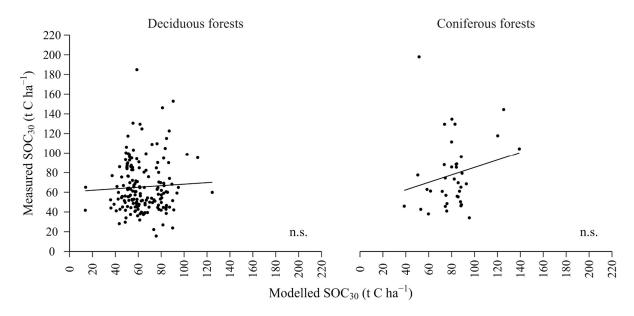
Forest land	Biogeographical		Measured SO	C30	Modelled SOC <sub>30</sub>	
stratum used in NIR	region	N -	<b>Mean</b> $\pm$ <b>s.e.</b> (t C ha <sup>-1</sup> )	<b>CV</b> (%)	<b>Mean ± s.e.</b> (t C ha <sup>-1</sup> )	<b>CV</b> (%)
	Continental	132	$64.30 \pm 2.20^{a\pi}$	39	$62.30 \pm 1.22^{a\pi}$	22
Deciduous forests	Alpine	25	$70.42 \pm 6.78^{a\pi}$	48	$73.75 \pm 4.00^{b\pi}$	27
	Mediterranean	19	$65.87 \pm 4.22^{a\pi}$	28	$55.57 \pm 4.51^{a\pi}$	35
Coniferous forests	Alpine	24	$74.00 \pm 5.77^{a\pi}$	38	$80.95 \pm 1.86^{a\pi}$	11
	Mediterranean	14	$84.83 \pm 12.13^{a\pi}$	53	$80.69 \pm 8.07^{a\pi}$	37

NOTE: Different lowercase Latin letters in columns indicate statistically significant differences (p < 0.05) in measured and modelled  $SOC_{30}$  between biogeographical regions of a given Forest land stratum. Different lowercase Greek letters in rows indicate a statistically significant difference (p < 0.05) between measured and modelled  $SOC_{30}$  in a given Forest land stratum × BGR. CV = coefficient of variance. Data source for measured  $SOC_{30}$ : National scientific soil survey and OKFŠ project (see subchapters 2.4.1 and 2.5.4.1).



**Figure 3.4.** Comparison of the measured and modelled soil organic carbon (mean  $\pm$  s.e.) in the mineral soil layer down to 30 cm depth (SOC<sub>30</sub>) at the forest land stratum (DFS – deciduous forests stratum, CFS – coniferous forests stratum)  $\times$  biogeographical region (BGR) level. The dashed line is a 1:1 line. NOTE: Data source for measured SOC<sub>30</sub>: National scientific soil survey and OKFŠ project (see subchapters 2.4.1 and 2.5.4.1).

Comparison at the plot level revealed no correlation between the modelled and measured SOC<sub>30</sub> stocks for both deciduous and coniferous forests (Figure 3.5), and increased discrepancies between measurements and modelling (Table 3.9).



**Figure 3.5.** Comparison of the measured and modelled soil organic carbon in the mineral soil layer down to 30 cm depth ( $SOC_{30}$ ) at the plot level for different Forest land strata. NOTE: Data source for measured  $SOC_{30}$ : National scientific soil survey and OKFŠ project (see subchapters 2.4.1 and 2.5.4.1).

Residual analysis (Figure A.2) revealed that the smaller values of the modelled  $SOC_{30}$  stocks are more likely to underestimate the true values (positive residuals), whereas the larger model values are likely to overestimate the true values. The bias was higher for deciduous forests  $(R^2 = 0.25, p < 0.01)$  in comparison to the coniferous forests  $(R^2 = 0.11, p = 0.05)$  (Figure A.2). Summary results of the model evaluation at different levels of stratification are shown in Table 3.9. Although at the Forest land strata level, the model performance is very good, due to only two data points these results should be taken with caution.

When disaggregating results to lower levels, an increase in discrepancies between the measured and modelled SOC<sub>30</sub> data is observed, which suggests that the ability of the Biome-BGCMuSo model to reproduce measured forest soil organic carbon of the mineral soil layer declines with increase in level of stratification (hypothesis H3).

**Table 3.9.** Summary of the statistics model evaluation regarding different stratification levels for estimating soil organic carbon in the mineral soil layer down to 30 cm depth ( $SOC_{30}$ , in t C  $ha^{-1}$ ).

Stratification level	N	R <sup>2</sup>	MAE	RMSE	NSE*
Forest land stratum	2		2.505	2.531	0.840
Forest land stratum × biogeographical region	5	0.66	5.344	6.109	0.303
Plot	214	0.02	24.004	31.158	-0.227

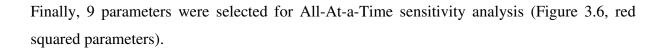
NOTE: MAE = mean absolute error, RMSE = root mean square error, NSE = Nash-Sutcliffe model efficiency coefficient. \* Acceptable levels of model performance when  $0 < NSE \le 1$  (Moriasi et al. 2007).

### 3.3.2. Calibrated BBGCMuSo v.6.2 (forest stand carbon stocks and carbon fluxes)

#### 3.3.2.1. Sensitivity analysis

One-At-a-Time sensitivity analysis resulted in 21 parameters with sensitivity higher than 20% for at least one of the selected output variables (AGC<sub>w</sub>, FFC, SOC<sub>30</sub>, NEE<sub>cum</sub>) (Figure 3.6). The analysis revealed different sensitivities of the selected output variables to changes in a single ecophysiological parameter (Figure 3.6). For example, mortality parameters (fire mortality fraction – FM, and whole-plant mortality fraction – WPM) were highly influential on AGC<sub>w</sub> and NEE<sub>cum</sub> (sensitivity > 40%) and at the same time showed lower influence on SOC<sub>30</sub> and FFC (sensitivity < 20%). Similarly, senescence parameters (SMCA and SMCB) had a strong influence only on FFC (sensitivity > 80%), while for the other three investigated variables sensitivity was less than 15% (Figure 3.6). Also, the parameter nitrogen fixation (Nfix) showed the highest influence on SOC<sub>30</sub> (sensitivity > 45%) in comparison to other parameters, but with a sensitivity of less than 20% for FFC and NEE<sub>cum</sub>. On the contrary, for some parameters, a similar sensitivity of all investigated variables (or at least three) was observed, as is the case of C:N of fine roots (CN\_ro).

Considering the computational limitations of All-At-a-Time sensitivity analysis, not all parameters presented in Figure 3.6 could be selected for further analysis. Therefore, some of the influential parameters were excluded for various reasons. As an example, for the WPM parameter, an estimated value from field observations in the pedunculate oak forest in Jastrebarsko is available (Hidy et al. 2016a), therefore changing this parameter would not be justified. Likewise, in Jastrebarsko oak forest no fire events were recorded in the study period, therefore the parameter of annual fire mortality fraction (FM), although influential on AGC<sub>w</sub> and cumulative NEE, was not modified. Also, the parameter of C:N of roots (CN\_ro) was not selected since this parameter can be measured with standard laboratory techniques.



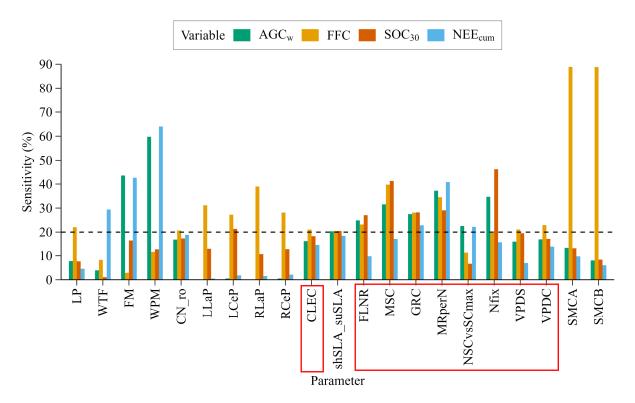
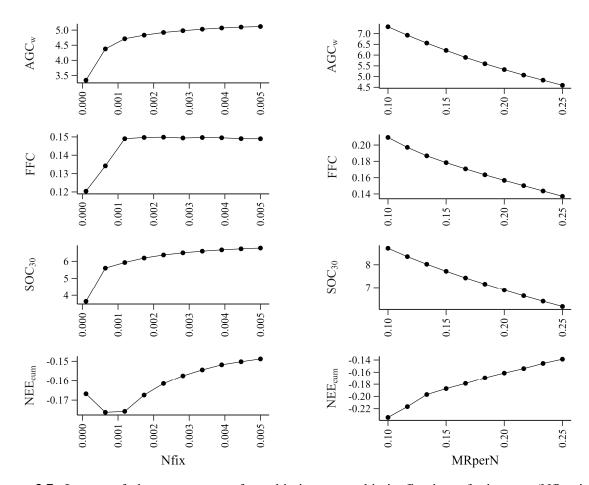


Figure 3.6. Results of One-At-a-Time sensitivity analysis of simulated output variables: carbon stock in above-ground live wood (AGC<sub>w</sub>), forest floor (FFC) and soil organic carbon in the mineral soil layer down to 30 cm depth (SOC<sub>30</sub>), and carbon flux cumulative net ecosystem exchange (NEE<sub>cum</sub>), to the changes in the ecophysiological parameters of the Biome-BGCMuSo model. The horizontal dashed line indicates a 20% threshold. The red squares indicate parameters selected for use in All-At-a-Time sensitivity analysis. LP – litterfall as fraction of the growing season; WTF – annual live wood turnover fraction; FM - annual fire mortality fraction; WPM - whole-plant mortality fraction in the vegetation period; CN\_ro - C:N of fine roots; LLaP - leaf litter labile proportion; LCeP - leaf litter cellulose proportion; RLaP – fine root labile proportion; RCeP – fine root cellulose proportion; CLEC – canopy light extinction coefficient; shSLA\_suSLA - ratio of shaded SLA:sunlit SLA; FLNR - fraction of leaf N in Rubisco; MSC – maximum stomatal conductance (projected area basis); GRC – growth resp per unit of C grown; MRperN – maintenance respiration in kg C day<sup>-1</sup> per kg of tissue N; NSCvsSCmax – theoretical maximum prop. of non-structural and structural carbohydrates; Nfix – symbiotic+asymbiotic fixation of N; VPDS – vapor pressure deficit: start of conductance reduction; VPDC – vapor pressure deficit: complete conductance reduction; SMCA - maximum senescence mortality coefficient of aboveground plant material; SMCB - maximum senescence mortality coefficient of belowground plant material.

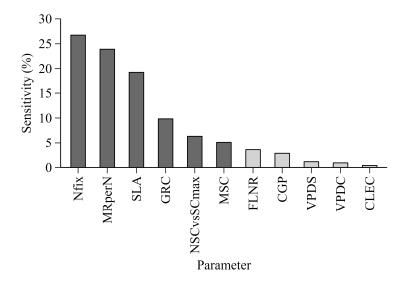
The selected parameters had different impacts on the examined output variables that varied along the tested parameter range (Figure 3.7). As expected, some parameters had the opposite impact, e.g. Nfix resulted in an increase, while maintenance respiration per tissue of nitrogen (MRperN) caused a decrease of carbon stocks in above-ground biomass, soil and forest floor.

Also, the increase of the parameter Nfix resulted in a rapid increase of carbon stocks in the above-ground biomass, soil and forest floor at the beginning of the parameter range, after which the carbon stocks in the examined variables were stable through the whole range of the parameter (Figure 3.7). On the contrary, for cumulative NEE, the increase of Nfix resulted in an initial decrease of carbon flux and was then followed by a gradual increase (Figure 3.7).



**Figure 3.7.** Impact of the parameters of symbiotic + asymbiotic fixation of nitrogen (Nfix, in kg N m<sup>-2</sup> year<sup>-1</sup>, left panel) and maintenance respiration per tissue of nitrogen (MRperN, in kg C kg N<sup>-1</sup> day<sup>-1</sup>, right panel) on carbon stocks in above-ground live wood, forest floor and soil organic carbon in the mineral soil layer down to 30 cm depth (AGC<sub>w</sub>, FFC and SOC<sub>30</sub>, respectively) and cumulative carbon flux net ecosystem exchange (NEE<sub>cum</sub>); all in kg C m<sup>-2</sup>, along the tested parameters' range in One-At-a-Time sensitivity analysis.

<u>All-At-a-Time sensitivity analysis</u> revealed that within the group of the selected parameters, Nfix had the greatest influence on the variable daily NEE, i.e. 26.73% of the total group sensitivity (Figure 3.8). Six parameters had sensitivity greater than 5%: Nfix, MRperN, SLA, GRC, NSCvsSCmax and MSC (Figure 3.8, dark grey bars) and were selected for the next step, model calibration.



**Figure 3.8.** Results of All-At-a-Time sensitivity analysis of simulated variable daily net ecosystem exchange (NEE<sub>daily</sub>) to the changes in the group of ecophysiological parameters of the Biome-BGCMuSo model. Dark grey bars indicate parameters selected for use in the model calibration (sensitivity > 5%). Nfix – symbiotic + asymbiotic fixation of N; MRperN – maintenance respiration in kg C day<sup>-1</sup> per kg of tissue N; SLA – canopy average specific leaf area (projected area basis); GRC – growth resp per unit of C grown; NSCvsSCmax – theoretical maximum prop. of non-structural and structural carbohydrates; MSC – maximum stomatal conductance (projected area basis); FLNR – fraction of leaf N in Rubisco; CGP – current growth proportion; VPDS – vapor pressure deficit: start of conductance reduction; VPDC – vapor pressure deficit: complete conductance reduction; CLEC – canopy light extinction coefficient.

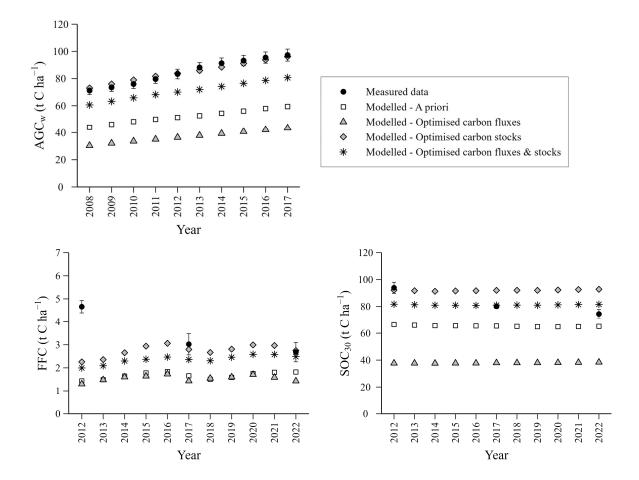
#### 3.3.2.2. Model calibration

The values of six optimised parameter values obtained using carbon flux data, carbon stock data, and carbon flux and carbon stock data combined differed from the *a priori* values as well as between each other (Table 3.10). In all three sets of optimised parameter values, maximum stomatal conductance (MSC) and theoretical maximum prop. of non-structural and structural carbohydrates (NSCvsSCmax) increased, while MRperN and Nfix decreased compared to the *a priori* values. The most substantial magnitude of change relative to the *a priori* values was observed in NSCvsSCmax (average increase of 78%), followed by Nfix (average decrease of 51%).

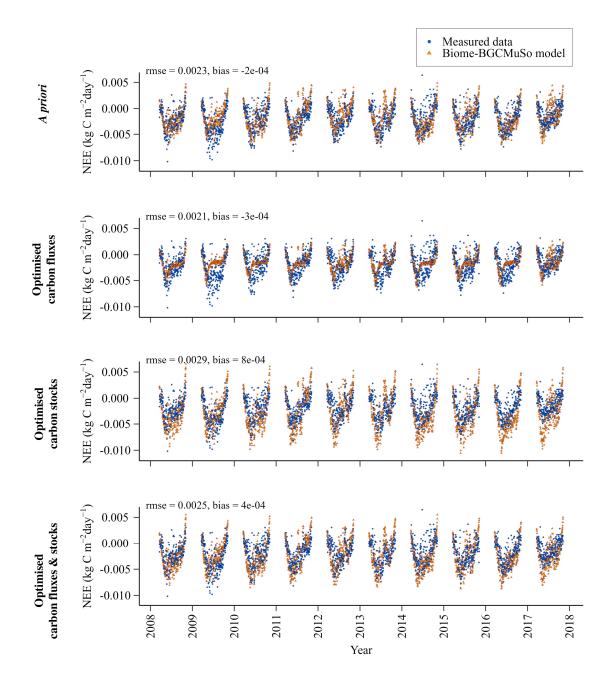
**Table 3.10**. Parameters MSC – maximum stomatal conductance (projected area basis); GRC – growth resp per unit of carbon grown; MRperN – maintenance respiration in kg C day<sup>-1</sup> per kg of tissue nitrogen; NSCvsSCmax – theoretical maximum prop. of non-structural and structural carbohydrates; Nfix – symbiotic+asymbiotic fixation of nitrogen; SLA – canopy average specific leaf area (projected area basis) values in different parameter sets. Min and Max refer to the minimum and maximum range of parameter values used in the Biome-BGCMuSo v6.2 calibration. "A priori" refers to the initial parameter set before the model calibration, "Optimised carbon fluxes" refers to the parameter set obtained from the calibration for carbon fluxes daily and annual net ecosystem exchange (NEE<sub>daily</sub> and NEE<sub>cum</sub>), "Optimised carbon stocks" refers to the parameter set obtained from the calibration for carbon stock in above-ground live wood (AGC<sub>w</sub>), forest floor (FFC) and soil organic carbon in the mineral soil layer down to 30 cm depth (SOC<sub>30</sub>), and "Optimised carbon fluxes & stocks" refers to the parameter set obtained from the calibration for all examined carbon fluxes and carbon stocks.

				Parameter set				
Parameter	Unit	Min	Max	A priori	Optimised carbon fluxes	Optimised carbon stocks	Optimised carbon fluxes & stocks	
MSC	$(m s^{-1})$	0.002	0.01	0.0024	0.0029	0.0029	0.003	
GRC	(prop.)	0.1	0.4	0.3	0.11	0.132	0.345	
MRperN	$(kg\;C\;kg\;N^{-1}\;day^{-1})$	0.1	0.25	0.218	0.132	0.104	0.138	
NSCvsSCmax	(ratio)	0.07	0.3	0.1	0.158	0.224	0.152	
Nfix	$(kgN m^{-2} yr^{-1})$	0.0001	0.005	0.0036	0.0001	0.0035	0.0017	
SLA	$(m^2 kgC^{-1})$	16	48	34.5	23.51	23.54	36.95	

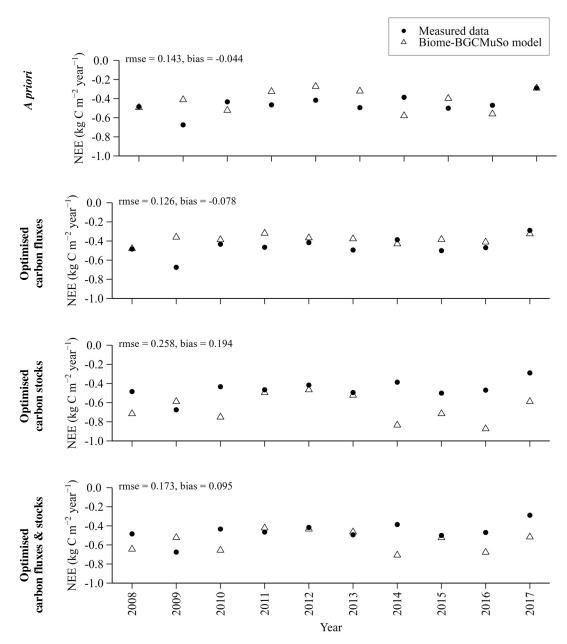
Optimizing model performance using high-frequency data, namely carbon fluxes (daily NEE and its annual sums, cumulative NEE), resulted in a better agreement between measured and modelled daily and cumulative NEE in comparison to the use of the *a priori* parameter set (Figure 3.10, Figure 3.11, Table 3.11). However, parameters optimised in this way resulted in reduced intra-seasonal variability in daily NEE (Figure 3.10) and an increased discrepancy between measured and modelled carbon stocks (Figure 3.9, Table 3.11). Correspondingly, calibration using solely carbon stocks resulted in a better agreement between measured and modelled carbon stocks in above-ground live wood, forest floor and SOC<sub>30</sub> in comparison to the case when *a priori* parameter set is used, with the greatest improvement observed for the AGC<sub>w</sub> (Figure 3.9, Table 3.11). However, by using this set of parameters, the discrepancy between measured and modelled carbon fluxes has increased (Figure 3.10, Figure 3.11, Table 3.11). Finally, for SOC<sub>30</sub> stock minimised error was obtained when using an optimised set of parameters for both carbon fluxes and carbon stocks combined (Table 3.11).



**Figure 3.9.** Comparison of the measured (mean  $\pm$  s.e., black filled circles) and modelled (different symbols) carbon stocks in above-ground live wood (AGC<sub>w</sub>), forest floor (FFC) and in the mineral soil layer down to 30 cm depth (SOC<sub>30</sub>) using different sets of parameters. Modelled – *A priori* indicates the modelling results obtained using the "*A priori*" parameter set; Optimised carbon fluxes indicates the modelling results obtained using the "Optimised C Fluxes" parameter set; Optimised carbon stocks indicate the modelling results obtained using the "Optimised C stocks" parameter set; and Optimised carbon fluxes & stocks indicates the modelling results obtained using the "Optimised C fluxes and stocks" parameter set. NOTE: Data source for measured AGCw, FFC and SOC<sub>30</sub>: Anić et al. (2018) and Ostrogović (2013) (see subchapter 2.5.5.3).



**Figure 3.10.** Comparison of the measured (blue-filled circles) and modelled (yellow-filled triangles) daily net ecosystem exchange (NEE) using different sets of parameters. *A priori* indicates the modelling results obtained using the "*A priori*" parameter set, Optimised carbon fluxes indicate the modelling results obtained using the "Optimised carbon fluxes" parameter set, Optimised carbon stocks indicates the modelling results obtained using the "Optimised carbon stocks" parameter set, and Optimised carbon fluxes & stocks indicates the modelling results obtained using the "Optimised carbon fluxes and stocks" parameter set. NOTE: Data source for measured NEE: Anić et al. (2018) (see subchapter 2.5.5.3).



**Figure 3.11.** Comparison of the measured (black-filled circles) and modelled (white-filled triangles) cumulative net ecosystem exchange (NEE) using different sets of parameters. *A priori* indicates the modelling results obtained using the "*A priori*" parameter set, Optimised carbon fluxes indicate the modelling results obtained using the "Optimised carbon fluxes" parameter set, Optimised carbon stocks indicates the modelling results obtained using the "Optimised carbon stocks" parameter set, and Optimised carbon fluxes & stocks indicates the modelling results obtained using the "Optimised carbon fluxes and stocks" parameter set. NOTE: Data source for measured NEE: Anić et al. (2018) (see subchapter 2.5.5.3).

**Table 3.11.** Evaluation of model performance for eddy covariance site using different sets of parameters in comparison to the measured data for carbon fluxes – daily and cumulative net ecosystem exchange (NEE), in kg C  $\rm m^{-2}$ , and carbon stocks in above-ground biomass (AGC<sub>w</sub>), the mineral soil layer down to 30 cm depth (SOC<sub>30</sub>) and forest floor (FFC), in t C  $\rm ha^{-1}$ .

Variable	Parameter set	N	R <sup>2</sup>	MAE	RMSE	BIAS	NSE*
NEE <sub>daily</sub>	A priori	2310	0.282	0.0018	0.0023	-0.0002	-0.03
	Optimised carbon fluxes	2310	0.219	0.0016	0.0021	-0.0003	0.15
	Optimised carbon stocks	2310	0.288	0.0023	0.0029	0.0008	-0.67
	Optimised carbon fluxes & stocks	2310	0.293	0.0020	0.0025	0.0004	-0.25
NEE <sub>cum</sub>	A priori	10	0.011	0.121	0.143	-0.044	-1.35
	Optimised carbon fluxes	10	0.010	0.094	0.126	-0.078	-0.83
	Optimised carbon stocks	10	0.006	0.211	0.258	0.194	-6.66
	Optimised carbon fluxes & stocks	10	0.014	0.140	0.173	0.095	-2.45
$AGC_w$	A priori	10	0.977	32.95	33.24	32.95	-12.58
	Optimised carbon fluxes	10	0.987	47.83	48.09	47.83	-27.42
	Optimised carbon stocks	10	0.98	1.98	2.1	0.02	0.95
	Optimised carbon fluxes & stocks	10	0.982	13.95	14.24	13.95	-1.49
SOC <sub>30</sub>	A priori	3	0.987	17.25	18.85	17.25	-4.34
	Optimised carbon fluxes	3	0.871	44.83	45.62	44.83	-30.25
	Optimised carbon stocks	3	0.344	10.64	12.65	-9.49	-1.40
	Optimised carbon fluxes & stocks	3	0.126	6.74	8.17	1.37	0
FFC	A priori	3	0.926	1.82	2.08	1.82	-4.81
	Optimised carbon fluxes	3	0.969	2.07	2.26	2.07	-5.86
	Optimised carbon stocks	3	0.942	0.90	1.39	0.85	-1.60
	Optimised carbon fluxes & stocks	3	0.992	1.17	1.59	1.17	-2.38

NOTE: MAE = mean absolute error, RMSE = root mean square error, BIAS = arithmetic mean of differences between modelled and observed values of the respective variable, NSE = Nash-Sutcliffe model efficiency coefficient. \*Acceptable levels of model performance when  $0 < NSE \le 1$  (Moriasi et al. 2007).

#### 3.3.3. Validated BBGCMuSo 6.2 (forest SOC<sub>30</sub> stock changes)

Measured and modelled SOC<sub>30</sub> in the stands of the pedunculate oak chronosequence experiment were, in general, found to be stable over the investigated 10-year period (Table 3.12, Figure 3.14). In each chronosequence stand measured SOC<sub>30</sub> showed no significant trend over the investigated period and in the majority of the stands, no significant differences were observed in measured SOC<sub>30</sub> between the sampling years (Table 3.12). The exception is the oldest stand, 701, in which measured SOC<sub>30</sub> significantly decreased (p < 0.05) from 2017 to 2022 (Table 3.12, Figure 3.14). Also, for the stand 301, significant differences in measured SOC<sub>30</sub> were detected between sampling years (RM Anova, p < 0.05). However, the post-hoc test revealed no significant differences between the sampling years (p > 0.05) (Table 3.12). The substantially higher overall variability of SOC<sub>30</sub> (including all sampling years and all stands)

was observed in the measured data (CV of 23%) in comparison to the modelled data (CV of 6%).

When comparing measured and modelled SOC<sub>30</sub> at the stand level and within each sampling year, in the majority of the stands modelled SOC<sub>30</sub> significantly differed from the measured SOC<sub>30</sub> (Table 3.12), either overestimating (stands 102, 101, 601) or underestimating (stand 701) measured SOC<sub>30</sub>. In stands 301 and 401, modelled SOC<sub>30</sub> represented well the measured SOC<sub>30</sub> in all sampling years (Table 3.12, Figure 3.14).

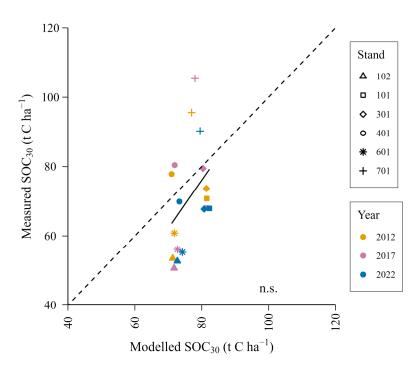
**Table 3.12.** Variability in the measured and modelled soil organic carbon stocks in the mineral soil layer down to 30 cm depth ( $SOC_{30}$ ) across different stands in the pedunculate oak chronosequence experiment during a ten-year period.

	Stand	Stand		Measured SOC <sub>30</sub>	Modelled SOC <sub>30</sub>		
Stand	age	Year	N	N Mean $\pm$ s.e. $(t C ha^{-1})$		N	<b>Mean</b> (t C ha <sup>-1</sup> )
	6	2012	4	$53.51 \pm 3.14^{a\pi}$	12	1	71.33 <sup>p</sup>
102	11	2017	4	$50.66 \pm 5.14^{a\pi}$	20	1	$71.75^{\rho}$
	16	2022	4	$52.70 \pm 2.45^{a\pi}$	9	1	$72.72^{\rho}$
	14	2012	4	$70.75 \pm 3.28^{a\pi}$	9	1	81.52°
101	19	2017	4	$68.00 \pm 3.23^{a\pi}$	10	1	81.41 <sup>p</sup>
	24	2022	4	$67.89 \pm 4.17^{a\pi}$	12	1	$82.32^{\rho}$
	54	2012	4	$73.66 \pm 3.46^{a\pi}$	9	1	$81.40^{\pi}$
301	59	2017	4	$79.43 \pm 4.17^{a\pi}$	10	1	$80.39^{\pi}$
	64	2022	4	$67.72 \pm 5.14^{a\pi}$	15	1	$80.73^{\pi}$
	69	2012	4	$77.81 \pm 6.81^{a\pi}$	18	1	$71.01^{\pi}$
401	74	2017	4	$80.41 \pm 4.30^{a\pi}$	11	1	$71.92^{\pi}$
	79	2022	4	$69.86 \pm 3.71^{a\pi}$	11	1	$73.33^{\pi}$
	109	2012	4	$60.82 \pm 3.11^{a\pi}$	10	1	71.82°
601	114	2017	4	$56.13 \pm 1.49^{a\pi}$	5	1	$72.75^{\rho}$
	119	2022	4	$55.29 \pm 2.21^{a\pi}$	8	1	$74.22^{\rho}$
	139	2012	4	$95.60 \pm 3.63^{\mathrm{ab}\pi}$	8	1	76.98 <sup>p</sup>
701	144	2017	4	$105.37 \pm 3.06^{b\pi}$	6	1	78.01°
	149	2022	4	$90.28 \pm 2.41^{a\pi}$	5	1	$79.52^{\circ}$

NOTE: Different lowercase Latin letters in columns indicate a statistically significant difference (p < 0.05) in SOC<sub>30</sub> in a given forest stand between the sampling years (analysed with post-hoc Tukey HSD test). Different Greek letters in rows indicate a statistically significant difference (p < 0.05, analysed with t-test) in SOC<sub>30</sub> in a given forest stand and sampling year between the measurements and Biome-BGCMuSo model. CV = coefficient of variance. NOTE: Data source for measured SOC<sub>30</sub>: Ostrogović (2013) and this study (see subchapter 2.5.6.1).

In the pedunculate oak forest (average for all chronosequence stands), measured  $SOC_{30}$  (mean  $\pm$  se) was  $72.02 \pm 5.95$ ,  $73.33 \pm 8.07$  and  $67.29 \pm 5.45$  t C ha<sup>-1</sup> for the years 2012, 2017, and 2022, respectively. Modelled  $SOC_{30}$  (mean  $\pm$  se) at the forest level for the years 2012, 2017

and 2022 was  $75.68 \pm 2.03$ ,  $76.04 \pm 1.81$ , and  $77.14 \pm 1.71$  t C ha<sup>-1</sup>, respectively. Again, measured SOC<sub>30</sub> showed greater spatial variability by sampling year (CV  $\geq$  20%) in comparison to the modelled SOC<sub>30</sub> variability (CV < 8%). At the forest level, a non-significant correlation between the measured and modelled carbon stocks in SOC<sub>30</sub> was revealed (Figure 3.12), although the slope of the linear trend is similar to the 1:1 line.

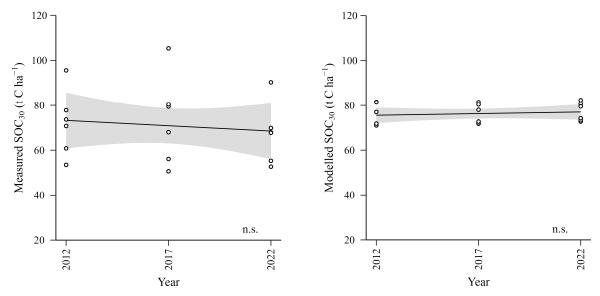


**Figure 3.12.** Comparison of the measured and modelled soil organic carbon in the mineral soil layer down to 30 cm depth (SOC<sub>30</sub>) at the forest stand and sampling year level in the pedunculate oak chronosequence experiment. The dashed line is a 1:1 line. The solid line is a regression line. NOTE: Data source for measured SOC<sub>30</sub>: Ostrogović (2013) and this study (see subchapter 2.5.6.1).

Forest carbon stock changes in SOC<sub>30</sub> were analysed for a short-term period of ten years (2012-2022) and for a long-term period of a rotation of a pedunculate oak forest in Croatia (140 years). The obtained trends in the measured and modelled SOC<sub>30</sub> during the investigated period from 2012 to 2022 were seemingly divergent, decreasing for measured SOC<sub>30</sub> ( $-0.474 \text{ t C ha}^{-1} \text{ year}^{-1}$ ;  $R^2 = 0.02$ , p = 0.61) and increasing for modelled SOC<sub>30</sub> ( $0.146 \text{ t C ha}^{-1} \text{ year}^{-1}$ ;  $R^2 = 0.02$ , p = 0.57), but neither of them was significant (p < 0.05; Figure 3.13) (hypothesis 4).

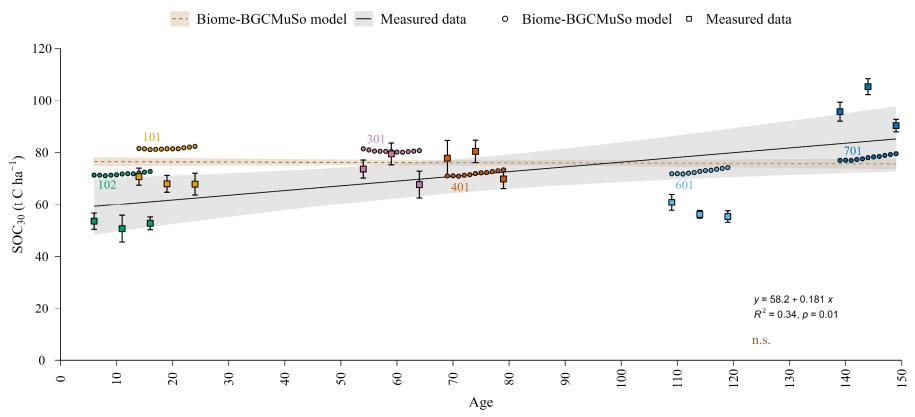
Additionally, when looking at the carbon stock change in  $SOC_{30}$  with the stand age using a single year chronosequence approach, no age-trend in measured nor modelled  $SOC_{30}$  was found in each sampling year (p < 0.05). By combining data from all three sampling years, a significant increasing trend of measured  $SOC_{30}$  with the stand age was observed (Figure 3.14). The average rate of change during the forest development (5–148 years old) was 0.181 t C ha<sup>-1</sup>. On the

contrary, modelled SOC<sub>30</sub> did not show a significant trend with stand age ( $R^2 < 0.01$ , p = 0.59), but it was possible to detect that the trend was divergent in comparison to the trend in measured SOC<sub>30</sub> with stand age (Figure 3.14).



**Figure 3.13.** Measured (left panel) and modelled (Biome-BGCMuSo model, right panel) soil organic carbon in the mineral soil layer down to 30 cm depth (SOC<sub>30</sub>) in the pedunculate oak forest (including six chronosequence stands, white filled circles) during a ten-year period with grey shading denoting 95% confidence intervals. NOTE: Data source for measured SOC<sub>30</sub>: Ostrogović (2013) and this study (see subchapter 2.5.6.1).

Soil texture was found to have a greater impact on the modelled SOC<sub>30</sub>, compared to the measured SOC<sub>30</sub> in the pedunculate oak chronosequence experiment in Jastrebarski lugovi (Figure 3.15). Modelled SOC<sub>30</sub> stocks showed higher correlation with clay and sand content in the soil (R<sup>2</sup> of 0.63 and 0.67 for clay and sand content, respectively), in comparison to the silt content ( $R^2 = 0.37$ ). The correlation of measured SOC<sub>30</sub> with the clay and silt content in the soil is not as strong as for modelled SOC<sub>30</sub>; however, it was significant (p < 0.05). Silt content in the soil had no significant correlation with measured SOC<sub>30</sub> (p < 0.05) (Figure 3.15). In both modelling and measurements, SOC<sub>30</sub> increased with the increasing content of clay in the soil and decreased with the increasing sand content (Figure 3.15). Also, modelled SOC<sub>30</sub> decreased with the increasing content of silt in the soil (Figure 3.15). Forest stands 101 and 301 have the highest clay content in the soil among all chronosequence stands (Figure 3.15), and were found to have higher modelled SOC<sub>30</sub> in comparison to the other stands (Figure 3.14, Table 3.12). Furthermore, the soil pH had no significant effect on the modelled SOC<sub>30</sub> stock (p < 0.05), while, on the contrary, measured SOC<sub>30</sub> significantly decreased with increasing pH (Figure 3.15). Forest stand 701 had the highest soil acidity (Figure 3.15) and in this stand measured SOC<sub>30</sub> stocks were found to be significantly higher in comparison to the modelled SOC<sub>30</sub> (Table 3.12, Figure 3.14).



**Figure 3.14.** Comparison of the measured (squares, mean  $\pm$  se; solid trendline with grey shading denoting 95% confidence intervals) and modelled (circles; dashed trendline with light-yellow shading denoting 95% confidence intervals) soil organic carbon in the mineral soil layer down to 30 cm depth (SOC<sub>30</sub>) for different stands in the chronosequence experiment (102, 101, 301, 401, 601, 701) and at different stand ages. Data points represent the year of the measurement for each stand; measured data years are 2012, 2017, and 2022, from left to right, and for modelled data, measured years range from 2012 to 2022, from left to right. NOTE: Data source for measured SOC<sub>30</sub>: Ostrogović (2013) and this study (see subchapter 2.5.6.1).

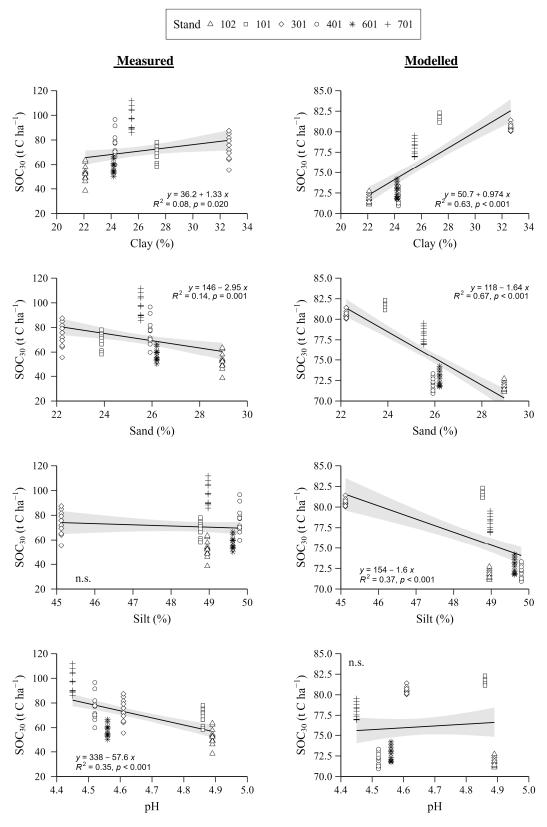


Figure 3.15. Linear regressions of pedunculate oak chronosequence stand-specific clay, silt, and sand content, and pH in the soil on the measured (left panel) and modelled (right panel) soil organic carbon in the mineral soil layer down to 30 cm depth (SOC<sub>30</sub>), with grey shading denoting 95% confidence intervals. Data points in the forest stand in measured data represent four sub-plots and sampling years (2012, 2017 and 2022) (N = 12), and in the modelled data modelled years (2012-2022) (N = 10). NOTE: clay content ( $< 0.002 \, \text{mm}$ ), silt content ( $< 0.002 \, \text{-0.02} \, \text{mm}$ ), pH, sampling year 2012 (Ostrogović 2013). Data source for measured SOC<sub>30</sub>: Ostrogović (2013) and this study (see subchapter 2.5.6.1).

#### 4. DISCUSSION

# 4.1. Variability and application of dead wood volume-to-carbon conversion factors

DW basic density is known to depend on tree species and tree species groups in early stages of DW decomposition (Adams and Owens 2001, Di Cosmo et al. 2013), and as DW decay progresses, the differences between tree species and tree species groups in the later decay classes are not significant (Di Cosmo et al. 2013, Herrmann et al. 2015). Within this research, accounting for different BGRs confirmed the pronounced differences in DW basic density in early decay classes (1–2) for broadleaves, and on the other hand, it revealed additional differences in later decay classes (2–4) for conifers (Figure 3.3).

In broadleaves, the most pronounced differences in DW basic density were found in decay classes 1 and 2, in which DW samples from the Mediterranean BGR had higher DW basic densities compared to other BGRs (Figure 3.3). In harsher environments, as it is commonly the case in the Mediterranean BGR, dry conditions repress radial growth (Corcuera et al. 2004), resulting in a denser fresh wood (Camarero et al. 2014, Zalloni et al. 2018). Basic density of fresh wood of holm oak, a representative of broadleaves in the Mediterranean BGR in this study, can range up to 0.96 g cm<sup>-3</sup> (Dilem 1995), which is considerably higher in comparison to the basic densities of tree species representing Continental BGR, 0.539 g cm<sup>-3</sup> (pedunculate oak; Tomczak et al. 2022) or 0.427 g cm<sup>-3</sup> (black alder; Johansson 2005). Consequently, high values of fresh wood basic density usually result in high DW basic density in subsequent stages of decomposition (Błońska et al. 2018).

DW samples of conifers had significantly higher DW basic densities in later decay classes (2–4), in the Mediterranean BGR (represented with pines) compared to the Alpine BGR (represented with fir and spruce) (Figure 3.3). This is an indication that DW samples of conifers from the Mediterranean BGR are likely to be assigned to advanced decay classes based on visual observations of the outer part of the sample, while having less decayed inner part of the wood. This could be attributed to the different decay resistance of sapwood and heartwood. Sapwood of nearly all species has no natural durability (Eslyn and Highley 1976), while regarding heartwood resistance tree species are categorized from very resistant to no-resistant to decay (Scheffer and Morrell 1998). Conifers are tree species which are moderately to

non-resistant to decay, although there is evidence of higher heartwood resistance in pines in comparison to spruce (Viitanen et al. 2006). Moreover, in very dry conditions, as it is commonly the case in the Mediterranean BGR, the activity of microbial community, which is usually hydrophilic (Bani et al. 2018), is temporarily obstructed limiting the DW decomposition (Edelmann et al. 2023). In such dry conditions, where only occasional raining periods supply the lying DW wood piece with water, it could be expected that decomposition would be more pronounced on the outer part of the DW wood piece, while inner dry parts would decompose much more slowly. This is also supported by the fact that in the most advanced stages of decomposition DW surface becomes more hydrophilic (Błońska et al. 2018).

Variability of DW CF is explained by many factors (including tree species group, tissue type and decay class), but mostly by biome (Martin et al. 2021) and site (Neumann et al. 2023). In the Mediterranean/Subtropical biome, DW CF is found to be significantly lower  $(46.24\% \pm 0.83\%)$  compared to DW CF in the Temperate biome  $(49.29\% \pm 0.74\%)$  (mean  $\pm$  se) (Martin et al. 2021), which corresponds to the results from this study for broadleaves (Figure 3.3). In fresh wood (decay class 0) CF is found to be negatively correlated with an increase of MAT (Paroshy et al. 2021). Considering the indications that live wood chemical traits have a deterministic role in driving DW C dynamics (Martin et al. 2021), the relationship between DW CF and MAT could potentially explain the observed variability of DW CF across BGRs from this study (Figure 3.3). Namely, DW CF in broadleaves, for decay classes 1 and 2, revealed a gradient with respect to BGRs, i.e. Alpine > Continental > Mediterranean (Figure 3.3), which is the opposite gradient of MAT at the study areas, i.e. Alpine < Continental < Mediterranean (Table 2.1). Furthermore, CF in fresh wood is found to be negatively correlated with wood basic density (Martin et al. 2018). This is another possible explanation of the results from this study. If we look at Figure 3.3, DW samples of broadleaves had the highest basic density and the lowest CF in the Mediterranean BGR, while in the Alpine BGR, the lowest basic density and highest CF were observed.

The results from this research indicate that DW basic density and CF are not only determined by specific wood properties that characterize tree species groups, but are also dependent on the environmental conditions under which trees were growing and DW logs were decomposed. However, it should be noted that both broadleaves and conifers are represented with different tree species in different BGRs, e.g. broadleaves in the Alpine BGR are represented with common beech, and in the Mediterranean BGR with holm oak. Therefore, the observed differences in DW traits between different BGRs can be a consequence of differences in environmental conditions and/or tree species characteristics. To distinguish the source of

variability, research on DW volume-to-carbon conversion factors for the same tree species in a different BGR would be desirable.

The importance of using the national DW volume-to-carbon conversion factors for the more accurate estimation of national DW carbon stocks has already been recognized (Di Cosmo et al. 2013, Stakėnas et al. 2020). Using decay class-specific DW basic density and CF instead of default values for fresh wood usually results in a substantial correction, i.e. a decrease in DW carbon stock estimate (Merganičová and Merganič 2010, Martin et al. 2021). National DW carbon stocks, calculated in this study, exhibited the largest correction in DFS (a decrease of 26.6%) and the smallest correction in FOOYS (a decrease of 11.1%) when using DW decay-class specific carbon density compared to the default values. The small variation in DW carbon stocks in FOOYS between two calculation methods was expected since in this stratum, there is almost three times more SDW volume compared to LDW (Table 2.3) and CD factor here was applied only for decay class 1 since all standing DW in CroNFI was classified as decay class 1. On the other hand, in the coniferous forest stratum there is almost two times more LDW volume compared to SDW (Table 2.3), indicating that in this stratum the highest difference between the DW carbon stock estimate is expected. Nevertheless, due to overestimated basic density and underestimated CF for conifers vs. the overestimation of both basic density and CF for broadleaves when estimating DW carbon stocks when using default values, a smaller correction was observed for the coniferous forest stratum compared to the deciduous forest stratum. This result suggests that the use of DW CD measure, which is the product of DW basic density and CF, for the calculation of DW carbon stocks, is a useful approach, as it encompass the possibly opposite trends of DW basic density and CF with the increasing decay class (Stakenas et al. 2020). Finally, it should be taken into account that in this study, only LDW was sampled, and there is evidence for differences in DW basic density and CF among different types of coarse woody debris (standing and lying) (Harmon et al. 2011, De Meo et al. 2018). Nevertheless, in decay class 1, no considerable difference in basic density and CF between standing and lying DW was found (Harmon et al. 2011, Harmon et al. 2013). In the first CroNFI, SDW volume refers only to decay class 1 (subchapter 2.3.4), therefore, we may argue that it is justified to use CD obtained from LDW for the conversion of SDW volume into national DW carbon stock. Finally, differences in DW basic density and CF between standing and lying DW are much smaller than the differences in basic density and CF between fresh and dead wood (Merganičová and Merganič 2010, Martin et al. 2021), which justifies the use of provided conversion factors even if they differ between standing and lying DW.

### 4.2. Variability and application of forest floor carbon stocks

Forest floor carbon stocks (FFCS) varied across BGRs for both broadleaves and conifers (Table 3.4). In broadleaves, significantly higher FFCS were observed in the Alpine BGR compared to the Continental BGR (Table 3.4). According to Gasparini and Di Cosmo (2015), FF is accumulated at high elevation sites, which could be attributed to cooler conditions limiting the microbial activity (Jandl et al. 2021). However, in this research, FFCS in broadleaves showed a low correlation with site elevation, MAT and MAP (Table 3.5), indicating that these environmental variables are likely not the main drivers for the difference between FFCS in Continental and Alpine BGRs. These findings corroborate with the research by De Vos et al. (2015), in which elevation, MAT and MAP exhibited a low relative importance for predicting FFCS. FF sampling in the National scientific soil survey and OKFŠ project was conducted throughout the year. This is important to note because in the Continental BGR, FFCSs decline from the period after the leaf fall to the period before the leaf fall (Pernar et al. 2012). Considering that in the Continental BGR only 23% of FF samples were collected during the autumn and winter, while in the Alpine BGR this share was 39%, we may argue whether this contributed to the lower FFCS in the Continental BGR compared to the Alpine BGR. This emphasizes the importance of timing of FF sampling and implies that national sampling campaigns should strive at parallel samplings in different strata (the task which is frequently not practical for other reasons and usually increases the costs). In conifers, FFCS were significantly higher in the Mediterranean compared to the Alpine BGR (Table 3.4). This could be attributed to the observed trends of FFCS in conifers with environmental variables, i.e. an increasing trend with increasing MAT and a decreasing trend with increasing MAP (Table 3.5), which are associated with the Alpine and Mediterranean BGR. It is known that in the Mediterranean forests FF decomposition is reduced due to drier conditions which obstruct microbial activity (Santonja et al. 2017).

Furthermore, FFCS in conifers in the Mediterranean BGR was 7.92 t C ha<sup>-1</sup> (in this study), which is considerably lower compared to the reported national FFCS values ranging from 12.593 to 20.071 t C ha<sup>-1</sup> in young (40 years old) and mature (80 years old) black pine forest stands, respectively (Bakšić and Bakšić 2020). The reported national values correspond to carbon stocks of 13.7 t C ha<sup>-1</sup> in the forest floor in 30–60 years old Mediterranean pine tree species plantations in Spain (Herrero et al. 2016). Likewise, conifers in the Alpine BGR (i.e. spruce and fir) had on average 4.93 t C ha<sup>-1</sup> stored in the forest floor which is again considerably

lower compared to the national literature. Namely, Bakšić et al. (2023) summarized national studies on FFCS in fir forest communities and for Fir and Hard Fern (*Blechno-Abietetum* Horvat /1938/1950), FFCS was estimated to be 23.9 t C ha<sup>-1</sup>, which is substantially higher than the results from this study. These national studies were mainly focused on the development of regression models for the purpose of estimating fire fuel load mass and carbon stocks from forest floor depth. Field sampling was often local and designed to cover the maximum range of FF depths which lead to the prevailing selection of old stands. On the contrary, this study comprises data on a national scale, including managed and protected forests of different age, resulting in a wider range of environmental and management conditions, therefore a lower overall mean of FFCS is reasonable.

The importance of the accurate estimation of national FFCS has already been recognized (López-Senespleda et al. 2021) since this carbon pool is far from negligible (Pan et al. 2011). This study is the first attempt of estimating FFCS at the national level while taking into consideration the representation of the forest area in each BGR. Net carbon stock change in FF carbon pool in Croatia in the year 2021 was 14.53 kt C year<sup>-1</sup> (i.e. 53.3 kt CO<sub>2</sub> year<sup>-1</sup>, when using carbon to CO<sub>2</sub> conversion factor of 3.667) for the area of 63.58 kha that was reported under LC-FL subcategory (HR NIR 2023). The use of new carbon stock change (CSC) factor of 0.2405 t C ha<sup>-1</sup> would increase the reported net CO<sub>2</sub> removal from this carbon pool for 2.8 kt CO<sub>2</sub> year<sup>-1</sup>.

Currently, the conversion of other LU to Forest land is not separated by Forest land stratum, i.e. deciduous forest stratum, coniferous forest stratum and forests out of yield stratum in the Croatian NIR. Nevertheless, it is expected that in the near future, a spatially-explicit land use-change matrix will be available. This will allow the use of new CSC factors by Forest land stratum (Table 3.6) in the calculation of CO<sub>2</sub> emissions/removals. Assuming that all land conversion, whose area of 63.58 kha is reported in the last Croatian NIR (HR NIR 2023), would occur under single Forest land stratum, hypothetically net CO<sub>2</sub> removal from FFC pool would range from 47.1 kt CO<sub>2</sub> year<sup>-1</sup> (in case all land conversion would occur in the forests out of yield stratum) to 70.3 kt CO<sub>2</sub> year<sup>-1</sup> (in case all land conversion would occur in the coniferous forests).

# 4.3. The use of BBGCMuSo v.4.0 for estimating forest SOC stocks at the country level

The results from this study indicate that Biome-BGCMuSo (v4.0) could be considered useful for the estimation of the average national soil organic carbon stocks down to 30 cm (SOC<sub>30</sub>) for two investigated Forest land strata reported in the Croatian NIR, namely deciduous forests and coniferous forests strata (Table 3.7). Carbon stocks in both modelled and measured SOC<sub>30</sub> were found to be higher in coniferous forests in comparison to the deciduous forests (Table 3.7), which is in line with the results from the literature (Jandl et al. 2021). According to Osei et al. (2021), the differences in SOC between coniferous and deciduous forests are more pronounced in the top of the mineral soil, which can indicate that FFCS govern SOC. Higher FFCS in conifers compared to broadleaves, as confirmed in this study (Table 3.4), is in line with the observed differences in SOC<sub>30</sub> stocks.

Disaggregation of the results by Forest land stratum × BGR resulted with decreased accuracy of modelled SOC<sub>30</sub> (Table 3.9) and revealed model limitations regarding soil spatial variability. Namely, spatial variability of soil in Croatia is high (Bogunović et al. 1997) and this is also evident from the high coefficient of variance in measured SOC<sub>30</sub> in both deciduous and coniferous forests (Table 3.7, Table 3.8). However, the modelled SOC<sub>30</sub> could not replicate this variance in the same range (Table 3.8). One of possible reasons could be that in the modelling, the average management activities for each forest type were used (Table A.7), resulting with lower variability in the modelled SOC<sub>30</sub> in comparison to the measured SOC<sub>30</sub> that are under the effect of various management activities. Moreover, at this level, there are indications that the model showed underestimation of SOC<sub>30</sub> in the deciduous forests of the Mediterranean BGR and overestimation of SOC<sub>30</sub> in the coniferous forest of the Alpine BGR (Figure 3.4). The reason behind this observation could be that the same ecophysiological file (EPC) for a specific forest type was used across all BGRs without modifications of some parameters that may vary across sites (Merganičová et al. 2024). Carbon allocation parameters are known to be dependent on the environmental conditions (Merganičová et al. 2019) and are found to be influential on the Biome-BGC model results (Tatarinov and Cienciala 2006). From a group of allocation parameters used in the Biome-BGCMuSo model (Table A.3), the ratio of fine root to leaf carbon is considered to have the greatest impact on SOC (Van Noordwijk and Van de Geijn 1996, Mokany et al. 2006). In the regions that are under water stress, as it is a case with the Mediterranean BGR, root production is promoted to meet the plant water demands (Friedlingstein et al. 1999). In this study, deciduous forests were modelled using two EPC files, one for oak and one for beech forests. A defined ratio of fine root to leaf carbon in the EPC files of these forest types is more likely to meet the conditions in the BGRs where these forest types are common, Continental BGR (oak) and Alpine BGR (beech), and fails to meet the conditions in the Mediterranean BGR. Therefore, using a unique ratio of fine root to leaf carbon in modelling specific forest types in different environmental conditions is likely to be too low at dryer sites, resulting in the underestimation of SOC<sub>30</sub> in the Mediterranean BGR. Therefore, it is highly recommended to modify specific model parameters that are known to substantially vary with environment.

At the plot level, the poor correlation of modelled and measured carbon stocks in SOC<sub>30</sub> was observed (Figure 3.5), which is also evident in the negative Nash-Sutcliffe model efficiency coefficient (Table 3.9), meaning that the simulation results using the Biome-BGCMuSo v.4.0 model were not capable of representing the variability of the measured SOC<sub>30</sub> that reflects sitespecific conditions. More accurate modelling results at the plot level could potentially be obtained by using long-term data on management activities and land-use changes at the plot (or stand) level. However, it is rarely the case that a comprehensive site history for each modelled plot exists (this issue will be addressed in more detail in subchapter 4.4), raising the issue of accurate reconstruction of SOC within the model's spin-up phase (Thornton and Rosenbloom 2005). Moreover, out of various factors that determine SOC, not all of them are incorporated in the model logic. Aside from soil properties like soil texture and bulk density (Wiesmeier et al. 2019), SOC can be influenced by soil fauna, microbial biomass (Wiesmeier et al. 2019) and mycorrhiza (Soudzilovskaia et al. 2019), and these factors are not taken into account in Biome-BGCMuSo v.4.0. Studies that evaluate model performance at different scales and for different data frequencies, such as this study, are highly valuable for model developers as they can indicate possible gaps in the model logic and be useful for further model development. Considering the poor performance of the Biome-BGCMuSo v.4.0 model at the plot level, for testing the fourth hypothesis the improved model version, Biome-BGCMuSo v.6.2 was chosen, which is significantly enhanced regarding carbon and nitrogen soil budget as well as the soil hydrology.

# 4.4. The use of BBGCMuSo v.6.2 for estimating forest SOC stock changes at the forest level

Measured and modelled SOC<sub>30</sub> stocks showed no significant trend in a ten-year period (2012-2022) (Figure 3.13). Stable stocks of modelled SOC<sub>30</sub> over time can mainly be attributed to the intrinsic stability of modelled carbon stocks in SOM, which is a precondition for reaching a steady state at the end of the spinup phase (Golinkoff 2013). After a successful spinup, modelled SOC will remain relatively stable under undisturbed environmental conditions, i.e. no significant changes in meteorological conditions and nitrogen deposition and no stand-replacing management activities. On the other hand, the lack of observed trend in measured SOC<sub>30</sub> stocks can be explained by high spatial variability of the soil (CV  $\geq$  20% in this study), which poses a great challenge in detecting short-term temporal changes in soil carbon stocks (Goidts et al. 2009, Ortiz et al. 2013, Jandl 2022). This can be even more pronounced in the case of a small number of data points (N = 18 in this study), suggesting that longer time series and higher sampling density are required. Here it should be noted that the trends in the modelled and measured SOC<sub>30</sub>, although not statistically significant, were divergent (positive for modelled and negative for measured) (Figure 3.13).

The lack of statistically significant trend in the measured SOC<sub>30</sub> restricts statistical inference regarding the modelled SOC<sub>30</sub>. Therefore, the comparison of the trends in the measured and modelled SOC<sub>30</sub> should be considered inconclusive. Only the case when a statistically significant trend (both positive and negative) would be available from measured SOC<sub>30</sub> data would allow us to reliably determine if the model can adequately reproduce temporal changes in SOC<sub>30</sub>.

Aside from testing short-term SOC<sub>30</sub> changes at the forest level, the use of the chronosequence experiment allowed testing for long-term changes in SOC<sub>30</sub> throughout the rotation period of the stand. When looking at a specific sampling year (single-year chronosequence approach), no significant trend with the stand age was observed for the modelled and measured SOC<sub>30</sub>, which is in line with other similar studies (Peltoniemi et al. 2004, De Simon et al. 2012, Ostrogović Sever 2019). The non-existing age-trend in measured SOC<sub>30</sub> can again be explained by the high spatial variability of SOC<sub>30</sub> between the forest stands and a small number of stands (N = 6). The modelled SOC<sub>30</sub> showed high positive correlation with the clay content in the soil (Figure 3.15). Namely, chronosequence stands were grouped into the ones with soil clay content above 25% and higher SOC<sub>30</sub> (stands 101, 301, 701), and the ones with soil clay content below 25% and

lower SOC<sub>30</sub> (stands 102, 401, 601) (Figure 3.15). It is known that the clay content in the soil is a strong predictor of soil's carbon stabilisation capacities (Jackson et al. 2017, Hartley et al. 2021) since it is related to the formation of soil aggregates that protect soil organic matter from microbial decomposition (Oades 1988). Hence, clay content is known to have a positive effect on SOC (Jobbágy and Jackson 2000, Wiesmeier et al. 2019). The result from this study indicates that the soil clay content is stronger driver of modelled SOC<sub>30</sub> than the stand age.

On the other hand, analysing the long-term SOC<sub>30</sub> dynamics using repeated chronosequence approach (all sampling years combined), revealed a significant increasing trend of measured SOC<sub>30</sub> with the stand age, while modelled SOC<sub>30</sub> still showed no age-trend (Figure 3.14). The resampling of the chronosequence experiment has already been recognised as important in monitoring forest development (Yanai et al. 2003). Nevertheless, when using the chronosequence approach for observing SOC dynamics there are certain limits one needs to be aware of. The nature of management activities changes over time, e.g. using horses vs tractors (Yanai et al. 2003), and different mechanization systems affect the forest soil differently (Clayton 1990). Namely, we may argue whether the highest measured SOC<sub>30</sub> observed in the oldest stand (701), compared to other stands, is due to the different harvesting method in the past, due to the stand age, or both.

When observing the age-trend in the measured SOC<sub>30</sub>, it was possible to note that the stand 601 did not fit within the 95% confidence interval of the observed trend (Figure 3.14). Chronosequence stands are similar regarding the soil characteristics with no substantial differences in the soil type, soil texture, bulk density and soil CF (Ostrogović 2013), indicating that the observed lower SOC<sub>30</sub> in this stand could be a consequence of some local anomaly. By investigating historical maps (web page <a href="https://www.arcanum.com/en/maps/">https://www.arcanum.com/en/maps/</a>), the indications of the land-use change were noted at the location of the stand 601 (Figure 4.1). Namely, in the period 1783–1784 (Molnár et al. 2014) it appears that the area of the stand 601 was under nonforest land, possibly cropland, while in the period 1865–1869 (Timár et al. 2006) this area was recorded as a forest land (Figure 4.1). This indicates that the land in the stand 601 was afforested between the years 1784 and 1865, while other forest stands from the chronosequence were recorded as forests already in the period 1783-1784 (maps not shown). In other words, forest stand 601 can maximally be the second forest generation, while other chronosequence stands have surely more than two forest generations. This could potentially explain lower measured SOC<sub>30</sub> in this stand, resulting in it not fitting well into the observed long-term age trend. The observation of site historical development suggests that the time-period longer than a default of 20 years (IPCC 2006) could be needed to reach forest SOC after the land conversion to forest land.



**Figure 4.1.** The approximate location of the stand 601 (in yellow rectangle) in the period 1783–1784 (Molnár et al. 2014) (left panel), period 1865–1869 (Timár et al. 2006) (middle panel) and in the year 2024 (right panel).

#### 4.5. Calibration of BBGCMuSo v.6.2 model

In addition to the discussion on the results directly related to the research hypothesis four, several issues observed during the model calibration call for further commenting. The calibration of models within a Biome-BGC family is usually performed using high-frequency data available from flux measurement stations, mostly eddy covariance sites (Chiesi et al. 2007, Sándor et al. 2016, Hidy et al. 2016a). It was observed that Biome-BGC can simulate carbon fluxes effectively, but fails in representing the woody biomass stocks (Maselli et al. 2009). This study demonstrated that the model calibration using solely high-frequency data (carbon fluxes) could result in a reduced model ability for predicting long-term data (carbon stocks) (Table 3.11, Figure 3.9). Likewise, model calibration for solely carbon stocks resulted with increased discrepancy between the modelled and measured carbon fluxes (Table 3.11, Figure 3.10, Figure 3.11). These findings highlight the importance of taking into account datasets with a different temporal resolution when performing model calibration.

Obtaining observation data of different temporal resolution from a single site poses a challenge. Therefore, the EC site in Jastrebarski lugovi is highly valuable for its long-term measurements of both carbon fluxes and carbon stocks (Anić et al. 2018). Still, a single-site calibration (this study) is considered to potentially "overfit" the parameters for the calibration site, and reduce model robustness when applied to other sites (Blyth et al. 2011). Therefore, a multi-site calibration (Merganičová et al. 2024) by combining observation data from multiple sites that

have available data on carbon fluxes and carbon stocks in the pedunculate oak forest would be desired.

It is widely accepted that the sensitivity analysis should be performed for any new model version, since with model development new parameters are introduced to the model and relations between already included parameters and output variables may change. Some studies also emphasize the importance of performing forest type-specific SA as they observed different levels of impact of specific parameters on carbon stocks and carbon fluxes between broadleaves and conifers (Tatarinov and Cienciala 2006). In this study, in the All-At-a-Time SA output variable daily NEE was found to be most sensitive to the changes in parameters' nitrogen fixation (Nfix), maintenance respiration per kg of tissue N (MRperN) and specific leaf area (SLA) (Figure 3.8). Generally, parameters related to photosynthesis and respiration have the highest influence on carbon fluxes in comparison to other parameters (Pillai et al. 2019, Liu et al. 2022). Likewise, parameter fraction of leaf N in Rubisco (FLNR) is considered as a key influential parameter on carbon fluxes in the deciduous and coniferous forests in the temperate zone (Raj et al. 2014, Ren et al. 2022). However, in this study FLNR did not show high influence on carbon flux (less than 10%) (Figure 3.6 and Figure 3.8), corroborating the findings from Tatarinov and Cienciala (2006). Here it should be noted that in this study, parameter ranges were adjusted to avoid model collapse as it could cause unrealistically high sensitivity of variables. Uncertainty of the model results can originate from the defined parameter range used in the SA (Liu et al. 2022), which makes finding the appropriate parameter range a critical, yet challenging choice (Hasan et al. 2017). Finally, besides ecophysiological parameters, the uncertainty of model outputs can also originate from model input data (such as meteorology) or site parameters (such as soil parameters) (Post et al. 2008).

#### 5. CONCLUSIONS

The carbon stock in dead wood pool estimated in this study is lower by 11–27% (depending on the Forest land stratum) compared to the corresponding value currently used in the Croatian National GHG Inventory Report (NIR), which is calculated using fresh wood basic densities and carbon fraction of 50%, suggesting that the national dead wood volume-to-carbon conversion factors by decay classes obtained in this study should be used to improve the accuracy of the Croatian NIR.

Newly compiled national database on forest floor carbon stocks can facilitate the increase of net CO<sub>2</sub> removals for 5% under this carbon pool within *Land converted to Forest land* subcategory of Forest land in the Croatian NIR. Significantly higher forest floor carbon stocks in the coniferous forests stratum compared to other Forest land strata emphasizes the need for stratification of land conversion activity data (area) within the Croatian NIR, with respect to the Forest land stratum in order to improve the accuracy of net CO<sub>2</sub> removal estimate from forest floor carbon pool.

Process-based Biome-BGCMuSo v.4.0 model showed to be a suitable tool for the estimation of the overall mean of soil organic carbon down to 30 cm depth (SOC<sub>30</sub>) for deciduous and coniferous forests strata reported in the Croatian NIR. Although after the disaggregation of the results with respect to biogeographical regions the model still represented the measured SOC<sub>30</sub> well, indications of underestimating SOC<sub>30</sub> stocks in the Mediterranean BGR and overestimating SOC<sub>30</sub> stocks in the Alpine BGR were observed. This highlights the need for further development of the model inner logic and routines, as well as additional calibration of model parameters to account for particularities regarding species and biogeographical regions.

The parameter values obtained in calibration of the Biome-BGCMuSo model v.6.2 depend on the type of data used in calibration. Namely, optimized parameter values obtained when using carbon stock data were different from optimized parameter values obtained when daily carbon fluxes data, or both, were used in calibration. This highlights the importance of using different temporal resolution datasets in the calibration of process-based models.

No significant trend was observed either in measured or in modelled SOC<sub>30</sub> using the Biome-BGCMuSo v.6.2 model in the pedunculate oak forest, represented by the chronosequence experiment, during the investigated period from 2012 to 2022. The obtained results could be attributed to high spatial variability observed in the measured SOC<sub>30</sub> and

intrinsic stability of model carbon stocks in soil organic matter under similar conditions. Although there is no disagreement in the trends between the measured and modelled SOC<sub>30</sub>, the trends were divergent (negative for measured and positive for modelled SOC<sub>30</sub>), suggesting that more thorough research, including longer time series and higher sampling density, is required.

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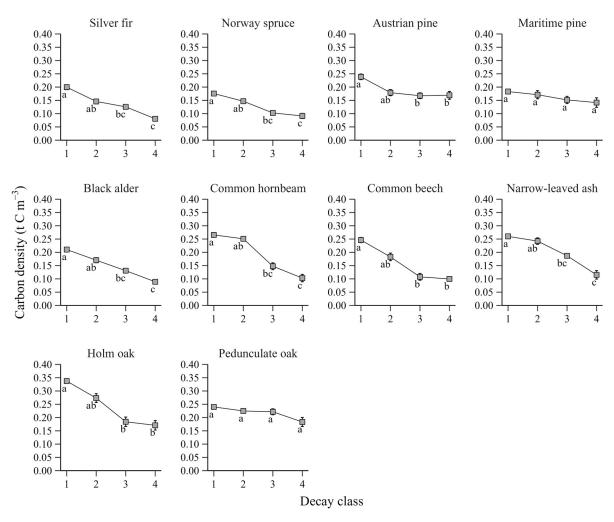
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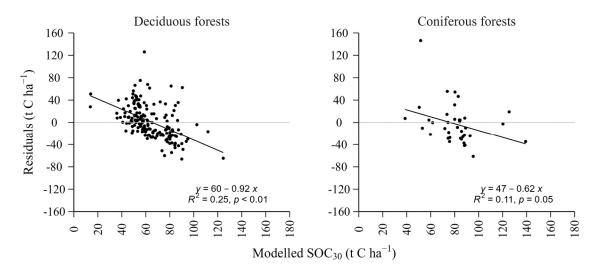
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## **APPENDIX**



**Figure A.1.** Dead wood carbon density (mean  $\pm$  s.e.) by decay classes for silver fir (*Abies alba* Mill.), Norway spruce (*Picea abies* (L.) Karst.), Austrian pine (*Pinus nigra* Arnold), maritime pine (*Pinus pinaster* Aiton), black alder (*Alnus glutinosa* (L.) Gaertn.), common hornbeam (*Carpinus betulus* L.), common beech (*Fagus sylvatica* L.), narrow-leaved ash (*Fraxinus angustifolia* Vahl), holm oak (*Quercus ilex* L.) and pedunculate oak (*Quercus robur* L.). Different lowercase letters next to data points indicate a statistically significant difference (p < 0.05) in dead wood carbon density between different decay classes for a given tree species. The number of samples per tree species and decay class is nine.



**Figure A.2.** Residual analysis of soil organic carbon in the mineral soil layer down to 30 cm depth  $(SOC_{30})$  at the plot level for different Forest land strata.

**Table A.1.** The number of dead wood samples collected within dead wood study, grouped according to different tree species groups, biogeographical regions and tree species.

Tree species group	Biogeographical region	Tree species	Number of analysed samples
		Black alder (Alnus glutinosa (L.) Gaertn.)	36
		Common hornbeam (Carpinus betulus L.)	36
D 11	Continental	Narrow-leaved ash (Fraxinus angustifolia Vahl)	36
Broadleaves		Pedunculate oak (Quercus robur L.)	36
	Alpine	Common beech (Fagus sylvatica L.)	36
	Mediterranean	Holm oak (Quercus ilex L.)	36
<b>Broadleaves Total</b>			216
	Alata	Silver fir (Abies alba Mill.)	36
Const.	Alpine	Norway spruce (Picea abies (L.) Karst.)	36
Conifers	M . 1'4	Austrian pine (Pinus nigra Arnold)	36
	Mediterranean	Maritime pine (Pinus pinaster Aiton)	36
<b>Conifers Total</b>			144
GRAND TOTAL			360

**Table A.2.** Distribution of plots within biogeographical regions and according to main forest ecosystems and tree species in Forest ecosystem database (ForecoDB).

Project	Forest ecosystem/	Biog	eographic	al region	Tota
Troject	Tree species	Continental	Alpine	Mediterranean	1018
	Oak	101	4	7	11
	Pedunculate oak (Quercus robur L.)	48		1	4
	Sessile oak (Quercus petraea (Matt.) Liebl)	26	1		2
	Turkey oak (Quercus cerris L.)	4	2	1	
	Pubescent oak (Quercus pubescens Willd.)	1	1	3	
	Other	22		2	2
	Beech	52	27	3	8
	Common beech (Fagus sylvatica L.)	41	24	3	6
	Other	11	3		1
	Pine	2		4	
National	Austrian pine (Pinus nigra Arnold)	2		4	
scientific	Fir/Spruce	1	24		2
soil survey	Silver fir (Abies alba Mill.)	1	18		1
	Norway spruce (Picea abies (L.) Karst.)		6		
	Forests out of yield	4	1	17	2
	Shrub/maquis	4		8	1
	Pubescent oak (Quercus pubescens Willd.)			5	
	Holm oak (Quercus ilex L.)			2	
	Turkey oak (Quercus cerris L.)			1	
	Other (oak)			1	
	Other (beech)		1		
	BROADLEAVES	157	32	27	2
	CONIFERS	3	24	4	3
Total		160	56	31	2
	Oak			8	
	Pubescent oak (Quercus pubescens Willd.)			4	
	Turkey oak (Quercus cerris L.)			1	
	Other			3	
	Pine			10	]
	Aleppo pine ( <i>Pinus halepensis</i> Mill.)			6	
OKFŠ project	Austrian pine ( <i>Pinus nigra</i> Arnold)			4	
	FOOY			9	
	Pubescent oak (Quercus pubescens Willd.)			4	
	Holm oak (Quercus ilex L.)			4	
	Turkey oak (Quercus cerris L.)			1	
	BROADLEAVES			17	
	CONIFERS			10	
Total				27	2
Grand Total		160	56	58	27

NOTE: "Other" indicate tree species that are common in associated forest ecosystem, e.g. alder, ash, or lime in oak forests and sycamore maple or hornbeam in beech forests. Data source: National scientific soil survey and OKFŠ project (see subchapter 2.4.1).

**Table A.3.** Ecophysiological constants (EPC) file for the Biome-BGCMuSo v4.0 model for Oak, Beech, Pine and Fir/Spruce forest ecosystems. Values for Oak are from Hidy et al. (2016a) for Oak forest and values for Beech, Pine and Fir/Spruce are from Cienciala and Tatarinov (2006) and Hidy et al. (2016a) for Oak forest. Some of the parameters were adjusted according to specified references or field measurement. Lowercase letters in the Reference/remarks column refer to the forest type: a) Oak, b) Beech, c) Pine, d) Fir/Spruce.

2 3

4

Parameter name	Unit -	a) <b>OAK</b>	b) <b>BEECH</b>	c) PINE	d) FIR/ SPRUCE	References/remarks	
FLAGS							
biome type flag (1 = WOODY 0 = NON-WOODY)	(flag)	1	1	1	1		
woody type flag (1 = EVERGREEN 0 = DECIDUOUS)	(flag)	0	0	1	1		
photosyn. type flag (1 = C3 PSN 0 = C4 PSN)	(flag)	1	1	1	1		
phenology flag (1 = MODEL PHENOLOGY 0 = USER-SPECIFIED PHENOLOGY)	(flag)	0	1	1	1		
Q10 flag (1 = temperature dependent q10 value; 0= constans q10 value)	(flag)	1	1	1	1		
acclimation flag (1 =acclimation 0 = no acclimation)	(flag)	1	1	1	1		
CO2 conductance reduction flag (0: no effect, 1: multiplier)	(flag)	1	1	1	1		
soil hydrological calculation method (0: Richards, 1: DSSAT)	(flag)	1	1	1	1		
discretization level of SWC calculation (0: low, 1: medium, 2: high)	(int)	0	0	0	0		
soil temperature calculation method (0: Zheng, 1: DSSAT)	(flag)	0	0	0	0		
ECOPHYSIOLOGICAL PARAMETERS							
yearday to start new growth (when phenology flag = 0)	(yday)	80	0	0	0	a) personal assessment	
yearday to end litterfall (when phenology flag = 0)	(yday)	310	0	0	0	a) personal assessment	
transfer growth period as fraction of growing season	(prop.)	0.3	0.2	0.3	0.3		
litterfall as fraction of growing season	(prop.)	0.3	0.2	0.3	0.3		
base temperature	(Celsius)	5	5	5	5		
growing degree day for start of fruit allocation	(Celsius)	1000	1000	1000	1000		
growing degree day for start of leaf senescence (0: no 1:yes)	(Celsius)	2350	2350	2350	2350		
annual leaf and fine root turnover fraction	(1/yr)	1	1	0.4	0.26	c-d) White et al. 2000	
annual live wood turnover fraction	(1/yr)	0.7	0.7	0.7	0.7		
annual whole-plant mortality fraction	(1/vegper)	0.02	0.005	0.01	0.005	b, d) White et al. 2000	

			Paramete			
Parameter name	Unit -	a) <b>OAK</b>	b) BEECH	c) PINE	d) FIR/ SPRUCE	References/remarks
annual fire mortality fraction	(1/yr)	0	0	0	0	
(ALLOCATION) new fine root C : new leaf C	(ratio)	0.8	0.9	1.5	0.66	a-d) adjusted
(ALLOCATION) new fruit c : leaf c (>0: yes, 0: no)	(ratio)	0.14	0	0	0	
(ALLOCATION) soft stem c : leaf c (>0: yes, 0: no)	(ratio)	N/A	N/A	N/A	N/A	
(ALLOCATION) new woody stem C : new leaf C	(ratio)	2	2	2	2.2	a, b, c) adjusted
(ALLOCATION) new live wood C : new total wood C	(ratio)	0.16	0.154	0.076	0.1	b) Pietsch et al. 2005
(ALLOCATION) new root C : new stem C	(ratio)	0.26	0.115	0.39	0.21	b) Pietsch et al. 2005; c) adjusted
(ALLOCATION) current growth proportion	(ratio)	0.5	0.5	0.5	0.5	
C:N of leaves	(kgC/kgN)	29.43	21.73	46.1	40.18	a-d) genus-specific field measurements
C:N of leaf litter, after retranslocation	(kgC/kgN)	44.05	45.14	69.62	51.19	a-d) genus -specific field measurements
C:N of fine roots	(kgC/kgN)	43	47.6	57.6	58	b) Pietsch et al. 2005; c-d) White et al. 2000
C:N of fruit	(kgC/kgN)	78.14	NO	NO	NO	a) genus-specific field measurements
C:N of soft stem	(kgC/kgN)	N/A	N/A	N/A	N/A	
C:N of live wood	(kgC/kgN)	73.5	50	58	37.1	b) Pietsch et al. 2005
C:N of dead wood	(kgC/kgN)	451	550	826	730	b) Pietsch et al. 2005; c) White et al. 2000
leaf litter labile proportion	(DIM)	0.2	0.124	0.26	0.28	b) Pietsch et al. 2005
leaf litter cellulose proportion	(DIM)	0.56	0.561	0.49	0.38	b) Pietsch et al. 2005
fine root labile proportion	(DIM)	0.34	0.34	0.23	0.23	b) Pietsch et al. 2005
fine root cellulose proportion	(DIM)	0.44	0.44	0.41	0.41	b) Pietsch et al. 2005
fruit litter labile proportion	(DIM)	0.3	NO	NO	NO	
fruit litter cellulose proportion	(DIM)	0.29	NO	NO	NO	
soft stem litter labile proportion	(DIM)	N/A	N/A	N/A	N/A	
soft stem litter cellulose proportion	(DIM)	N/A	N/A	N/A	N/A	
dead wood cellulose proportion	(DIM)	0.75	0.77	0.7	0.7	b) Pietsch et al. 2005
canopy water interception coefficient	(1/LAI/d)	0.038	0.034	0.044	0.045	b) Pietsch et al. 2005; c-d) White et al. 2000
canopy light extinction coefficient	(DIM)	0.54	0.6	0.508	0.5	b) Pietsch et al. 2005; c-d) White et al. 2000

			Paramete	r's value		
Parameter name	Unit -	a) OAK	b) BEECH	c) PINE	d) FIR/ SPRUCE	References/remarks
all-sided to projected leaf area ratio	(DIM)	2	2	2.6	2.6	
canopy average specific leaf area (projected area basis)	(m²/kgC)	34.5	34.5	6.8	7.8	c) White et al. 2000
ratio of shaded SLA:sunlit SLA	(DIM)	2	2	2	2	
fraction of leaf N in Rubisco	(DIM)	0.088	0.162	0.055	0.053	b) Pietsch et al. 2005
fraction of leaf N in PEP Carboxylase	(DIM)	N/A	N/A	N/A	N/A	
maximum stomatal conductance (projected area basis)	(m/s)	0.0024	0.005	0.0025	0.002	
cuticular conductance (projected area basis)	(m/s)	0.00006	0.00006	0.00006	0.00006	
boundary layer conductance (projected area basis)	(m/s)	0.005	0.01	0.01	0.009	
relative SWC (prop. to FC) to calc. soil moisture limit 1 (-9999: not used)	(prop)	1	1	1	1	a-d) adjusted
relative SWC (prop. to SAT) to calc. soil moisture limit 2 (-9999: not used)	(prop)	0.99	0.99	1	1	a-d) adjusted
relative PSI (prop. to FC) to calc. soil moisture limit 1 (-9999: not used)	(prop)	-9999	-9999	-9999	-9999	
relative PSI (prop. to SAT) to calc. soil moisture limit 2 (-9999: not used)	(prop)	-9999	-9999	-9999	-9999	
vapour pressure deficit: start of conductance reduction	(Pa)	200	600	600	610	
vapour pressure deficit: complete conductance reduction	(Pa)	2550	3000	2500	3100	b) Pietsch et al. 2005
senescence mortality coefficient of aboveground plant material	(prop.)	0.01	0.01	0.01	0.01	
senescence mortality coefficient of belowground plant material	(prop.)	0.01	0.01	0.01	0.01	
senescence mortality coefficient of leaf (after maturity)	(prop.)	0.025	0.025	0.025	0.025	
turnover rate of wilted standing biomass to litter	(prop.)	0.01	0.01	0.01	0.01	
turnover rate of cut-down non-woody biomass to litter	(prop.)	0.01	0.01	0.01	0.01	a-d) adjusted (KM)
N denitrification proportion	(prop.)	0.01	0.01	0.01	0.01	
bulk N denitrification proportion (WET)	(prop.)	0.02	0.02	0.02	0.02	
bulk N denitrification proportion (DRY)	(prop.)	0.01	0.01	0.01	0.01	
N mobilen proportion	(prop.)	0.1	0.1	0.1	0.1	
symbiotic+asymbiotic fixation of N	(kgN/m²/yr)	0.005	0.0005	0.002	0.0005	a-d) adjusted
ratio of the storage and the actual pool mortality due to management	(prop.)	0.9	0.9	0.9	0.9	
critical value of soil stress coefficient	(prop.)	0.5	0.5	0.5	0.5	
critical number of stress days after which senescence mortality is complete	(days)	90	90	90	90	
maximum depth of rooting zone	(m)	0.1-0.63	0.1-0.63	0.1-0.63	0.1-0.63	a-d) site-specific

Parameter name	Unit	a) <b>OAK</b>	b) <b>BEECH</b>	c) <b>PINE</b>	d) FIR/ SPRUCE	References/remarks
root distribution parameter	(DIM)	3.67	1.5	3.67	3.67	b) adjusted (KM)
maturity coefficient	(prop.)	0.5	0.5	0.5	0.5	
growth resp per unit of C grown	(prop.)	0.3	0.3	0.3	0.3	
maintenance respiration in kgC/day per kg of tissue N	(kgC/kgN/d)	0.218	0.4	0.218	0.4	a, c) adjusted

NOTE: NO – not occurring, i.e. parameters that are not relevant due to model conditions set by flags; N/A – not available, i.e. parameters that are not present for woody vegetation. An additional 28 parameters at the end of the epc file were kept fixed and are not presented in the Table.

6

**Table A.4.** Ecophysiological constants (EPC) file for the Biome-BGCMuSo v6.2 model used in model calibration and validation for pedunculate oak forest. *A priori* are the initial values of the parameters used in the model calibration, Min and Max are minimum and maximum parameters' values, respectively, that defined the parameter range used in the sensitivity analysis and calibration, and Optimised are parameter's optimised values used in the model validation. *A priori* values are from Hidy et al. (2016a) for oak forest, a generic EPC file for deciduous broadleaf forests and from proposed values from Hidy et al. (2021). Some of the parameters were adjusted according to personal assessment or field measurement. Abbreviations are given for the parameters used in the model calibration. Optimised parameters are indicated with grey shading.

Demonstra warms	Abbreviation	TT24		D-f			
Parameter name	Abbreviation	Unit	a) A priori	b) Min	c) Max	d) Optimised	- References/remark
Flags							
biome type flag (1 = woody 0 = non-woody)		(flag)	1			1	
woody type flag (1 = evergreen 0 =deciduous)		(flag)	0			0	
photosynthesis type flag (1 = C3 PSN $0$ = C4 PSN)		(flag)	1			1	
Parameter name							
yearday to start new growth		(yday)	80			80	a) personal assessment
yearday to end litterfall		(yday)	310			310	a) personal assessment
transfer growth period as fraction of growing season	GP	(prop.)	0.3	0.05	0.3	0.3	
litterfall as fraction of growing season	LP	(prop.)	0.3	0.2	0.6	0.3	
base temperature	T_base	(Celsius)	5	0	7	5	
minimum temperature for growth displayed on current day (-9999: no T-depend.)		(Celsius)	-9999			-9999	
optimal1 temperature for growth displayed on current day (-9999: no T-depend.)		(Celsius)	-9999			-9999	
optimal2 temperature for growth displayed on current day (-9999: no T-depend.)		(Celsius)	-9999			-9999	
maximum temperature for growth displayed on current day (-9999: no T-depend.)		(Celsius)	-9999			-9999	
minimum temperature for carbon assimilation on current day (-9999: no limit)		(Celsius)	-9999			-9999	
optimal1 temperature for carbon assimilation on current day (-9999: no limit)		(Celsius)	-9999			-9999	
optimal2 temperature for carbon assimilation on current day (-9999: no limit)		(Celsius)	-9999			-9999	
maximum temperature for carbon assimilation on current day (-9999: no limit)		(Celsius)	-9999			-9999	
annual leaf and fine root turnover fraction		(1/yr)	1			1	
annual live wood turnover fraction	WTF	(1/yr)	0.7	0.5	1	0.7	
annual fire mortality fraction	FM	(1/yr)	0	0	0.05	0	
whole-plant mortality fraction in vegetation period	WPM	(1/year)	0.02	0.01	0.1	0.02	

Davameter name	Abbussiati	II:-:4		Paramet	- References/remark			
Parameter name	Abbreviation	Unit	a) A priori	b) Min	c) Max	d) Optimised	- Keierences/remark	
C:N of leaves	CN_lv	(kgC/kgN)	22.76	17	27	22.76	<ul> <li>a) species-specific field measurements</li> </ul>	
C:N of leaf litter, after retranslocation	CN_li	(kgC/kgN)	33.96	23	65	33.96	a) species-specific field measurements	
C:N of fine roots	CN_ro	(kgC/kgN)	43	24	74	43		
C:N of fruit	CN_fr	(kgC/kgN)	66.42	33	80	66.42	a) species-specific field measurements	
C:N of soft stem		(kgC/kgN)	N/A			N/A		
C:N of live wood	CN_lw	(kgC/kgN)	73.5	60	100	73.5		
C:N of dead wood	CN_dw	(kgC/kgN)	451	400	550	451		
dry matter carbon content of leaves		(kgC/kgDM)	0.5			0.5		
dry matter carbon content of leaf litter		(kgC/kgDM)	0.5			0.5		
dry matter carbon content of fine roots		(kgC/kgDM)	0.5			0.5		
dry matter carbon content of fruit		(kgC/kgDM)	0.5			0.5		
dry matter carbon content of soft stem		(kgC/kgDM)	0.5			0.5		
dry matter carbon content of live wood		(kgC/kgDM)	0.5			0.5		
dry matter carbon content of dead wood		(kgC/kgDM)	0.5			0.5		
leaf litter labile proportion	LLap	(DIM)	0.2	0.1	0.4	0.2		
leaf litter cellulose proportion	LCeP	(DIM)	0.56	0.1	0.69	0.56		
fine root labile proportion	RLaP	(DIM)	0.34	0.1	0.5	0.34		
fine root cellulose proportion	RCeP	(DIM)	0.44	0.1	0.6	0.44		
fruit litter labile proportion	FLaP	(DIM)	0.3	0.1	0.6	0.3		
fruit litter cellulose proportion	FCeP	(DIM)	0.29	0.1	0.6	0.29		
soft stem litter labile proportion		(DIM)	N/A			N/A		
soft stem litter cellulose proportion		(DIM)	N/A			N/A		
dead wood cellulose proportion	WCeP	(DIM)	0.75	0.5	0.9	0.75		
canopy water interception coefficient	CWIC	(1/LAI/d)	0.038	0.01	0.06	0.038		
canopy light extinction coefficient	CLEC	(DIM)	0.54	0.3	0.7	0.54		
potential radiation use efficiency		(g/MJ)	NO			NO		
radiation parameter1 (Jiang et al.2015)		(DIM)	NO			NO		
radiation parameter2 (Jiang et al.2015)		(DIM)	NO			NO		

		** **		Paramete	Deferences/nemark		
Parameter name	Abbreviation	Unit -	a) A priori	b) Min	c) Max	d) Optimised	- References/remark
ll-sided to projected leaf area ratio	SLA_PA	(DIM)	2	1.5	2.5	2	
atio of shaded SLA:sunlit SLA	ShSLA_suSLA	(DIM)	2	0.6	5	2	
raction of leaf N in Rubisco	FLNR	(DIM)	0.088	0.08	0.3	0.088	
raction of leaf N in PEP Carboxylase		(DIM)	NO			NO	
naximum stomatal conductance (projected area basis)	MSC	(m/s)	0.0024	0.002	0.01	0.003	
outicular conductance (projected area basis)	CC	(m/s)	0.00006	0.0000 1	0.0001 8	0.00006	
oundary layer conductance (projected area basis)	BLC	(m/s)	0.005	0.004	0.09	0.005	
naximum height of plant		(m)	40			40	a) personal assessment
tem weight corresponding to maximum height		(kgC/m²)	700			700	a) personal assessment
plant height function shape parameter (slope)		(DIM)	1.1			1.1	a) personal assessment
naximum depth of rooting zone	MRD	(m)	1.0	0.8	4.1	1.0	
oot distribution parameter	rootDistr	(DIM)	3.67	0.5	4	3.67	
oot weight corresponding to max root depth		$(kgC/m^2)$	0.4			0.4	
oot depth function shape parameter (slope)		(DIM)	0.5			0.5	
oot weight to root length conversion factor		(m/kg)	NO			NO	
growth resp per unit of C grown	GRC	(DIM)	0.3	0.1	0.4	0.345	
naintenance respiration in kgC/day per kg of tissue N	MRperN	(kgC/kgN/d)	0.218	0.1	0.4	0.138	a) adjusted
heoretical maximum prop. of non-structural and structural carbohydrates	NSC:SCmax	(DIM)	0.1	0.07	0.3	0.152	
prop. of non-structural carbohydrates available for maintenance respiration	NSC_2MR	(DIM)	0.3	0.25	0.5	0.3	
ymbiotic+asymbiotic fixation of N	Nfix	$(kgN/m^2/yr)$	0.0036	0.0001	0.005	0.0017	
ime delay for temperature in photosynthesis acclimation	Tau	(day)	10	1	50	10	
SWC ratio to calc. soil moisture limit 1 (prop. to FC-WP)	VWCratio_lim1	(prop)	0.9	0.1	0.9	0.9	
SWC ratio to calc. soil moisture limit 2 (prop. to SAT-FC)	VWCratio_lim2	(DIM)	0.985	0.5	1	0.985	
ninimum of soil moisture limit2 multiplicator (full anoxic stress value)	Min_soilstress2	(prop)	0.4	0	1	0.4	
apor pressure deficit: start of conductance reduction	VPDS	(Pa)	200	100	1500	200	
rapor pressure deficit: complete conductance reduction	VPDC	(Pa)	2550	2200	3500	2550	
naximum senescence mortality coefficient of aboveground plant material	SMCA	(prop.)	0.01	0	0.014	0.01	
naximum senescence mortality coefficient of belowground plant material	SMCB	(prop.)	0.01	0	0.014	0.01	

Dougnaton name	Abbussistian	TI:4		Paramet	er's value		- References/remark
Parameter name	Abbreviation	Unit	a) A priori	b) Min	c) Max	d) Optimised	- References/remark
maximum senescence mortality coefficient of non-structured plant material	SMCL	(prop.)	0	0	0.003	0	
lower limit extreme high temperature effect on senescence mortality	SNSC_ext1	(Celsius)	30	30	39	30	
upper limit extreme high temperature effect on senescence mortality	SNSC_ext2	(Celsius)	40	31	50	40	
turnover rate of wilted standing biomass to litter	TRWB	(prop.)	0.01	0.01	0.1	0.01	
turnover rate of non-woody cut-down biomass to litter	TRCN	(prop.)	0.05	0.01	0.1	0.05	
turnover rate of woody cut-down biomass to litter	TRCW	(prop.)	0.0009	0.0001	0.1	0.0009	
drought tolerance parameter (critical value of DSWS)	DSWScrit	(prop.)	90	0	100	90	
effect of soil stress factor on photosynthesis (1: full effect, 0: no effect)	Ssef	(dimless)	0	0	0.2	0	
length of phenophase		(Celsius)	10000			10000	
leaf ALLOCATION		(ratio)	0.258			0.258	a) converted (from Hidy et al. 2016a)
fine root ALLOCATION		(ratio)	0.245			0.245	<ul><li>a) converted (from Hidy et al. 2016a)</li></ul>
fruit ALLOCATION		(ratio)	0.036			0.036	<ul><li>a) converted (from Hidy et al. 2016a)</li></ul>
soft stem ALLOCATION		(ratio)	N/A			N/A	
live woody stem ALLOCATION		(ratio)	0.059			0.059	a) converted (from Hidy et al. 2016a)
dead woody stem ALLOCATION		(ratio)	0.307			0.307	<ul><li>a) converted (from Hidy et al. 2016a)</li></ul>
live coarse root ALLOCATION		(ratio)	0.015			0.015	a) converted (from Hidy et al. 2016a)
dead coarse root ALLOCATION		(ratio)	0.08			0.08	a) converted (from Hidy et al. 2016a)
canopy average specific leaf area (projected area basis)	SLA	(m <sup>2</sup> /kgC)	34.5	16	66.7	36.95	
current growth proportion	CGP	(prop.)	0.5	0.1	0.7	0.5	
maximum lifetime of plant tissue		(°Cd)	10000			10000	

NOTE: NO – not occurring, i.e. parameters that are not relevant due to model conditions set by flags; N/A – not available, i.e. parameters that are not present for woody vegetation. Crop-specific and growing season blocks of parameters are not presented in the Table as they had no effect in model simulations in this study.

**Table A.5.** Soil properties input file for the Biome-BGCMuSo v6.2 model used in the model calibration and validation. Values on the left side are groups of parameters (in bold) and parameter values with units (in parenthesis) and on the right side are parameter descriptions. Values that changed in the model validation are indicated with grey shading.

SOILPR	OP FILE - MuSo6	.2 - extended (standalone) version - extraSOIparameters.txt file is not needed
SOIL G	ENERIC PARAM	METERS
2	(m)	depth of soil
12	(ppm)	C:N ratio of stable soil pool (soil4)
0.1	(prop.)	NH <sub>4</sub> mobilen proportion
107	(s/m)	aerodynamic resistance (Wallace and Holwill, 1997)
PEGOL	ADOCUMENON NAME	
	(dimless)	CRIFICATION AND DENITRIFICATION PARAMETERS
1.75 17	(dimless)	parameter 1 for tscalar function of decomposition parameter 2 for tscalar function of decomposition
2.6	(dimless)	parameter 3 for tscalar function of decomposition
40	(dimless)	parameter 4 for tscalar function of decomposition
-5	(Celsius)	minimum soil temperature for decomposition
10	(m)	e-folding depth of decomposition rate depth scalar
0.2	* *	net mineralization proportion of nitrification
0.2	(prop.)	maximum nitrification rate
0.02	(1/day)	
0.02	(prop.)	coefficient of N <sub>2</sub> O emission of nitrification parameter 1 for pHscalar function of nitrification
1	(dimless)	• • •
5.2	(dimless) (dimless)	parameter 2 for pHscalar function of nitrification
0.55	, ,	parameter 3 for pHscalar function of nitrification parameter 4 for pHscalar function of nitrification
1	(dimless)	parameter 1 for tscalar function of nitrification
12	(dimless) (dimless)	parameter 2 for tscalar function of nitrification
2.6	(dimless)	parameter 3 for tscalar function of nitrification
30	(dimless)	parameter 4 for tscalar function of nitrification
0.1		minimum WFPS for scalar of nitrification calculation
0.1	(prop.)	
0.45	(prop.) (prop.)	lower optimum WFPS for scalar of nitrification calculation higher optimum WFPS for scalar of nitrification calculation
0.2		minimum value for saturated WFPS scalar of nitrification calculation
0.2	(prop.) $(1/gCO_2)$	soil respiration related denitrification rate
2	(dimless)	denitrification related $N_2/N_2O$ ratio multiplier
0.55		critical WFPS value for denitrification
0.55	(prop)	Chucai WFF3 value foi deniumeation
RATE S	SCALARS	
0.39	(DIM)	respiration fractions for fluxes between compartments (11s1)
0.55	(DIM)	respiration fractions for fluxes between compartments (12s2)
0.29	(DIM)	respiration fractions for fluxes between compartments (14s3)
0.28	(DIM)	respiration fractions for fluxes between compartments (s1s2)
0.46	(DIM)	respiration fractions for fluxes between compartments (s2s3)
0.55	(DIM)	respiration fractions for fluxes between compartments (s3s4)
0.7	(1/day)	rate constant scalar of labile litter pool
0.07	(1/day)	rate constant scalar of cellulose litter pool
0.014	(1/day)	rate constant scalar of lignin litter pool
0.07	(1/day)	rate constant scalar of fast microbial recycling pool

SOILP	ROP F	ILE - M	uSo6.2	- extend	led (star	dalone)	) versio	ı - extra	SOIpar	ameters.txt file i	s not needed		
0.014	(1/0	day)								rate consta	ant scalar of medium microbial recycling pool		
0.0014	(1/	dow)								roto ao	notant cooler of clay, migrabial recycling neel		
	`	day)									enstant scalar of slow microbial recycling pool		
0.0001		day)		rate constant scalar of recalcitrant SOM (humus) pool rate constant scalar of physical fragmentation of coarse woody debris									
0.0000	(1/0	day)							rate cor	istant scarar or p	mysical fragmentation of coarse woody deoris		
SOIL	MOIS	TURE I	PARAN	IETER	s								
6	(m	m)									limit of first stage evaporation		
20	(m	m)									maximum height of pond water		
1	(di	mless)									curvature of soil stress function		
-9999	(d	imless)						n	neasureo	l runoff curve nu	umber (0: no runoff, -9999: model estimation)		
0.002	(pı	rop.)								fract	ion of dissolved part of SOIL1 organic matter		
0.002	(pı	op.)								fract	ion of dissolved part of SOIL2 organic matter		
0.001	(pı	op.)								fract	ion of dissolved part of SOIL3 organic matter		
0.001	(pı	rop.)								fract	ion of dissolved part of SOIL4 organic matter		
2	_	mless)								surfa	ce residue (e.g. mulch) parameter: layer effect		
1	(kg	gC/m2)								surface re	esidue (e.g. mulch) parameter: critical amount		
100	(di	mless)									parameter 1 for surface residue function		
0.75	(di	mless)									parameter 2 for surface residue function		
0.75	(di	mless)									parameter 3 for surface residue function		
0.5	(di	mless)								sur	face residue parameter: evaporation reduction		
0.88	`	mless)									parameter 1 for diffusion calculation (tipping)		
35.4	`	mless)									parameter 2 for diffusion calculation (tipping)		
5	`	mless)								•	parameter 3 for diffusion calculation (tipping)		
0	`	ag)									flag for GW-method		
-9999	(m										capillary fringe [m]		
-		METER	S										
212.5		IM)									emission bulk density dependence parameter1		
1.81	(D	IM)									emission bulk density dependence parameter2		
-1.353		IM)								•	ion soil water content dependence parameter1		
0.2	(D	IM)								•	ion soil water content dependence parameter2		
1.781	(D	IM)								soil CH <sub>4</sub> emiss	ion soil water content dependence parameter3		
6.786	(D	IM)								soil CH <sub>4</sub> emiss	ion soil water content dependence parameter4		
0.010	(D	IM)								soil CH <sub>4</sub> emis	ssion soil temperature dependence parameter1		
COIL	COME	OCITI	ONI A NI	D CHA	DACTI	DICTI	CVAL	HEC (	0000	o measured dat			
13.2	13.4	10.4	8.2	8.2	8.2	8.2	8.2	8.2	8.2	o measured dat (%)	sand percentage by volume in rock-free soil		
40.8	38.8	35.1	32.4	32.4	32.4	32.4	32.4	32.4	32.4	(%)	silt percentage by volume in rock-free soil		
5.1	5.1	5.1	5.1	5.1	5.1	5.1	5.1	5.1	5.1	(dimless)	soil pH		
-9999	-9999	-9999	-9999	-9999	-9999	-9999	-9999	-9999		(dimless)	soil pri soilB		
-9999 -9999	-9999 -9999	-9999	-9999 -9999	-9999 -9999	-9999 -9999	-9999	-9999 -9999			(g/cm <sup>3</sup> )	bulk density		
-9999 -9999	-9999 -9999	-9999	-9999 -9999	-9999 -9999	-9999 -9999	-9999	-9999 -9999			_	•		
			-9999 -9999	-9999 -9999	-9999 -9999	-9999 -9999	-9999 -9999			$(m^3/m^3)$	SWC at saturation		
-9999	-9999									$(m^3/m^3)$	SWC at field capacity		
-9999	-9999	-9999	-9999	-9999	-9999	-9999	-9999	-9999	-9999	$(m^3/m^3)$	SWC at leasure as is matter a state		
-9999	-9999	-9999	-9999	-9999	-9999	-9999	-9999	-9999		$(m^3/m^3)$	SWC at hygroscopic water content		
-9999	-9999	-9999	-9999	-9999	-9999	-9999	-9999	-9999		(dimless)	drainage coefficient		
-9999	-9999	-9999	-9999	-9999	-9999	-9999	-9999	-9999	-9999	(cm/day)	hydraulic conductivity at saturation		

**Table A.6.** Soil texture, clay, sand and silt proportion (%), and pH by Observation site / stand and soil depths, used in SOI files in the Biome-BGCMuSo v6.2 model calibration and validation.

Observation site / Stand	Soil depth (cm)	Soil texture			
		Clay (%)	Silt (%)	Sand (%)	pН
Eddy covariance site	0-3	40.8	40.8	13.2	5.1**
•	3-10	38.8	38.8	13.4	
	10-30	35.1	35.1	10.4	
	30-60	32.4	32.4	8.2	
Chronosequence experiment					
102	0-3	22.3	48.3	29.4	4.9**
	3-10	21.0	51.3	27.7	
	10-30	23.0	47.3	29.8	
	30-60	22.9	46.0*	31.1*	
101	0-3	26.6	50.5	22.9	4.9**
	3-10	26.1	50.3	23.6	
	10-30	29.4	45.5	25.2	
	30-60	31.6	43.1*	25.3*	
301	0-3	33.3	46.8	19.9	4.6**
	3-10	30.6	46.0	23.4	
	10-30	34.1	42.6	23.4	
	30-60	35.5	41.1*	23.4*	
401	0-3	24.5	50.7	24.8	4.5**
	3-10	23.0	50.2	26.8	
	10-30	25.4	48.5	26.2	
	30-60	26.0	48.4*	25.6*	
601	0-3	24.3	51.7	24.0	4.6**
	3-10	22.8	48.6	28.6	
	10-30	25.5	48.6	26.0	
	30-60	26.7	50.4*	22.9*	
701	0-3	25.2	44.3	30.5	4.5**
	3-10	25.1	51.4	23.5	
	10-30	26.2	51.2	22.6	
	30-60	27.6	49.5*	22.9*	

NOTE: \*applies to all deeper soil layers; \*\*equal to all 10 soil layers. Soil texture: clay content (<0.002 mm), silt content (0.002–0.02 mm), and sand content (0.02–2 mm); pH, sampling year 2012 (Data source: Ostrogović 2013).

**Table A.7.** Description of management activities for different forest ecosystems used in the Biome-BGCMuSo model v4.0 simulations.

Management activities	Forest ecosystem			
Management activities	Oak	Beech	Pine	Fir/Spruce
Rotation (years)	140	100	60	Uneven-aged management
Thinning rate, 1/10 y (%)	15	15	15	30
Thinning rate, 1/1 y used in model $(\%)$	2.1	2.35	2.92	3

NOTE: The explanation of the method for e.g. Oak forests is as follows: Oak forests in Croatia are even-aged managed, with average thinning of 15% performed every 10 years with regeneration cuts (2-3) performed during the last 10 years of the rotation period. To perform spatial modelling, thinning and regeneration cuts needed to be distributed among different locations. In the absence of this information average annual thinning intensity was estimated to ensure evenly distributed thinning at the spatial scale. Under the assumption of a rotation period of 140 years (prescribed rotation for pedunculate oak), annual thinning intensity should account for a 1.5% thinning rate during a 130 year period and 100% regeneration cut during the whole rotation period, which sums to 2.1%. Fir/Spruce forests are unevenly aged and managed with thinning of 30% performed every 10 years, i.e. average annual thinning intensity is 3%.

**Table A.8.** Initialization file (INI) used for spinup model run in model Biome-BGCMuSo v.4.0. On the left side are sections (in bold) with input data and on the right side are descriptions with information on type of data or unit (in parenthesis).

BBGCMuSo4 simulation spinup run	
MET_INPUT	
metdata\430176_FORESEE_v4_1900-1999.mtc43	(filename) met file name
4	(int) number of header lines in met file
RESTART	
0	(flag) $1 = \text{read restart}$ ; $0 = \text{dont read restart}$
1	(flag) $1 = \text{write restart}$ ; $0 = \text{dont write restart}$
0	(flag) 1 = use restart metyear; 0 = reset metyear
restart\muso4.endpoint	(filename) name of the input restart file
restart\muso4.endpoint	(filename) name of the output restart file
TIME_DEFINE	
100	(int) number of meteorological data years
100	(int) number of simulation years
1900	(int) first simulation year
1	(flag) $1 = \text{spinup run}; 0 = \text{normal run}$
6000	(int) maximum number of spinup years
CLIM_CHANGE	
0.0	(degC) - offset for Tmax
0.0	(degC) - offset for Tmin
1.0	(degC) - multiplier for PRCP
1.0	(degC) - multiplier for VPD
1.0	(degC) - multiplier for RAD
CO2_CONTROL	
1	(flag) 0=constant; 1=vary with file
290.0	(ppm) constant atmospheric CO <sub>2</sub> concentration
CO2\CO2_CMIP5_1900-1999.txt	(filename) name of the CO <sub>2</sub> file
NINER CONTROL	
NDEP_CONTROL 1	(flag) 0=constant; 1=vary with file
0.000200	(kgN/m²/yr) wet+dry atmospheric deposition of N
nitrogen\Ndep_hhs_1900-1999.txt	(filename) name of the N-dep file
SITE	
13.9 13.9 13.9 13.9 13.9 13.9	(%) sand percentage by volume in rock-free soil
34.9 34.9 34.9 34.9 34.9 34.9	(%) silt percentage by volume in rock-free soil
131.8	(m) site elevation
43.00	(degrees) site latitude (- for S.Hem.)
0.20	(DIM) site shortwave albedo
15.64	(Celsius) mean annual air temperature
18.83	(Celsius) mean annual air temperature range
-9999	(dimless) measured runoff curve number
-9999 -9999 -9999 -9999 -9999 -9999	(g/cm <sup>3</sup> ) bulk density (no data: -9999)

BBGCMuSo4 simulation spinup run	
-9999 -9999 -9999 -9999 -9999 -9999	(m <sup>3</sup> /m <sup>3</sup> ) SWC at SAT (no data: -9999)
-9999 -9999 -9999 -9999 -9999 -9999	(m³/m³) SWC at FC (no data: -9999)
-9999 -9999 -9999 -9999 -9999 -9999	(m <sup>3</sup> /m <sup>3</sup> ) SWC at WP (no data: -9999)
-9999 -9999 -9999 -9999 -9999	(m³/m³) SWC at HW (no data: -9999)
EPC_FILE	
epc\121_14.epc	(filename) EPC filename
W_STATE	
0.0	(kg/m²) water stored in snowpack
0.5	(DIM) initial soil water as a proportion of saturation
C_STATE	
0.03	(kgC/m²) first-year maximum leaf carbon
0.0	(kgC/m²) first-year maximum stem carbon
0.0	(kgC/m²) coarse woody debris carbon
0.0	(kgC/m²) litter carbon, labile pool
0.0	(kgC/m²) litter carbon, unshielded cellulose pool
0.0	(kgC/m²) litter carbon, shielded cellulose pool
0.0	(kgC/m²) litter carbon, lignin pool
0.0	(kgC/m²) soil carbon, fast microbial recycling pool
0.0	(kgC/m²) soil carbon, medium microbial recycling pool
0.0	(kgC/m²) soil carbon, slow microbial recycling pool
0.0	(kgC/m²) soil carbon, recalcitrant SOM (slowest)
	(kge/m ) son euroon, reculentant both (stowest)
N_STATE	
0.0	(kgN/m <sup>2</sup> ) litter nitrogen, labile pool
0.0	(kgN/m <sup>2</sup> ) soil nitrogen, labile pool
GROWING_SEASON	
5.00	(kg/m²) crit. amount of snow limiting photosyn.(if no data: 999.9)
0	(flag) use GSI index to calculate growing season
20.00	(Celsius) limit1 (under:full constrained) of HEATSUM index
60.00	(Celsius) limit2 (above:unconstrained) of HEATSUM index
0.00	(Celsius) limit1 (under:full constrained) of TMIN index
5.00	(Celsius) limit2 (above:unconstrained) of TMIN index
4000	(Pa) limit1 (above:full constrained) of VPD index
1000	(Pa) limit2 (under:unconstrained) of VPD index
0	(s) limit1 (under:full constrained) of DAYLENGTH index
0	(s) limit2 (above:unconstrained) of DAYLENGTH index
10	(day) moving average (to avoid the effects of extreme events)
0.1	GSI limit1 (greater that limit -> start of vegper)
0.01	GSI limit2 (less that limit -> end of vegper)
intvar\GSI_sp.txt	file of the estimated start and end of the VP
OUTPUT_CONTROL	
outputs\spinup	(txt) prefix for output files
control\ctrl_spinup.txt	(txt) file of the BBGC variables (control)
0	(flag) 1 = write daily output $0 = \text{no daily output}$
V	(mag) 1 – write daily output 0 = no daily output

BBGCMuSo4 simulation spinup run	
0	(flag) $1 = \text{monthly avg of daily variables } 0 = \text{no monthly avg}$
0	(flag) $1 = \text{annual avg of daily variables}$ $0 = \text{no annual avg}$
0	(flag) $1 = \text{write annual output } 0 = \text{no annual output}$
1	(flag) for on-screen progress indicator
DAILY_OUTPUT	
3	number of daily output variables
623	GPP
622	NEE
649	TR
ANNUAL_OUTPUT	
3	number of annual output variables
636	vegC
638	soilC
639	totalC
MANAGEMENT_SECTION	
PLANTING	
0	(flag) do PLANTING? 0=no; 1=yes; filepath=reading from file
30 -9999 -9999 -9999 -9999 -9999	(yday) PLANTING day
10.0 -9999 -9999 -9999 -9999 -9999	(double) quantity of seed (kg seed/ha)
40.0 -9999 -9999 -9999 -9999 -9999	(%) C content of seed
90.0 -9999 -9999 -9999 -9999 -9999	(%*) useful part of seed
THINNING	
MGM\121_thinning_sp.txt	(flag) do THINNING? 0=no; 1=yes; filepath=reading from file
30 -9999 -9999 -9999 -9999 -9999	(yday) THINNING day
0.0 -9999 -9999 -9999 -9999 -9999	(prop) thinning rate
90.0 -9999 -9999 -9999 -9999 -9999	(%) transported part of stem
0.0 -9999 -9999 -9999 -9999 -9999	(%) transported part of leaf
MOWING	(flag) do MOWING? 0=no; 1=yes; filepath=reading from file
0	(flag) mowing method? 0 - on fixday, 1 - if LAI greater than a fixed value
0	(int) fixed value of the LAI before MOWING (fixvalue method)
6.0	(int) fixed value of the LAI after MOWING (fixvalue method)
1.0	(yday*) MOWING day
150 234 -9999 -9999 -9999 -9999	(int*) value of the LAI after MOWING (fixday method)
1.0 1.0 -9999 -9999 -9999 -9999	(%*) transported part of plant materia
95.0 95.0 -9999 -9999 -9999 -9999	
GRAZING	
0	(flag) do GRAZING? 0=no, 1=yes; filepath=reading from file
156 262 -9999 -9999 -9999 -9999	(yday*) first day of GRAZING
202 311 -9999 -9999 -9999 -9999	(yday*) last day of GRAZING
650.0 650.0 -9999 -9999 -9999 -9999	(kg/LSU) weight equivalent of one unit
0.69 1.15 -9999 -9999 -9999 -9999	(LSU/ha*) animal stocking rate: Livestock Units per hectare
13.0 13.0 -9999 -9999 -9999 -9999	(kg dry matter/LSU*) daily ingested dry matter
1.50 1.50 -9999 -9999 -9999 -9999	(prop*) trampling effect (transform, standing dead biome to litter)

BBGCMuSo4 simulation spinup run	(6(♥) ,' CDM', 1 C 1
25.0 25.0 -9999 -9999 -9999 -9999 -9999	(%*) ratio of DM intake formed excremen
100.0 100.0 -9999 -9999 -9999 -9999	(%*) ratio of excrement returning to litte
40.0 40.0 -9999 -9999 -9999 -9999	(%*) carbon content of dry matte
2.0 2.0 -9999 -9999 -9999 -9999	(%*) N content of manur
40.0 40.0 -9999 -9999 -9999 -9999	(%*) C content of manur
0.02 0.02 -9999 -9999 -9999 -9999	(kgN <sub>2</sub> O-N:kgN*) manure emission factor for direct N <sub>2</sub> O emission
0.95 0.95 -9999 -9999 -9999 -9999	(dimless) fraction of nitrogen excretion managed in manure management system
11.0 11.0 -9999 -9999 -9999 -9999	(kgCH <sub>4</sub> /LSU/yr*) manure emission factor for CH <sub>4</sub> emission
58.0 58.0 -9999 -9999 -9999 -9999	(kgCH <sub>4</sub> /LSU/yr*) fermentation emission factor for CH <sub>4</sub> emission
HARVESTING	
0	(flag) do HARVESTING? 0=no, 1=yes; filepath=reading from file
200 -9999 -9999 -9999 -9999 -9999	(yday*) HARVESTING day
0.010 -9999 -9999 -9999 -9999 -9999	(m²/m²*) LAI after HARVESTING (snag
100.0 -9999 -9999 -9999 -9999 -9999	(%*) transported part of plant materia
PLOUGHING	
0	(flag) do PLOUGHING? 0=no, 1=yes; filepath=reading from fil
200 -9999 -9999 -9999 -9999 -9999	(yday*) PLOUGHING da
1 -9999 -9999 -9999 -9999 -9999	(type*) PLOUGHING depth (0:shallow, 1:medium, 2:deep
0.10 -9999 -9999 -9999 -9999 -9999	(prop*) dissolving coefficient of ploughed biome to litte
FERTILIZING	
0	(flag) do FERTILIZING? 0=no, 1=yes; filepath=reading from fil
60 120 -9999 -9999 -9999 -9999	(yday*) FERTILIZING da
30.0 30.0 -9999 -9999 -9999 -9999	(kgN/ha/day*) (nitrogen from fertilization per day
17.0 17.0 -9999 -9999 -9999 -9999	(%*) nitrate content of fertilize
17.0 17.0 -9999 -9999 -9999 -9999	(%*) ammonium content of fertilize
5.0 5.0 -9999 -9999 -9999 -9999	(%*) carbon content of fertilize
70.0 70.0 -9999 -9999 -9999 -9999	(%*) labile fraction of fertilize
20.0 20.0 -9999 -9999 -9999 -9999	(%*) unshielded cellulose fraction of fertilize
0.0 0.0 -9999 -9999 -9999 -9999	(%*) shielded cellulose fraction of fertilize
10. 109999 -9999 -9999 -9999	(%*) lignin fraction of fertilize
0.05 0.05 -9999 -9999 -9999 -9999	(prop*) dissolving coefficier
90.0 90.0 -9999 -9999 -9999 -9999	(%*) useful pa
0.01 0.01 -9999 -9999 -9999 -9999	$(kgN_2O\text{-}N:kgN^*)$ emission factor for N-addition
IRRIGATION	
0	(flag) do IRRIGATION? 0=no, 1=yes; filepath=reading from fil
150 250 -9999 -9999 -9999 -9999	(yday*) IRRIGATION da
30. 30.0 -9999 -9999 -9999 -9999	(kgH <sub>2</sub> O/m <sup>2</sup> /day*) amount of water
END_INIT	

**Table A.9.** Initialization file (INI) used for the normal model run in model Biome-BGCMuSo v.4.0. On the left side are sections (in bold) with input data and on the right side are descriptions with information on the type of data or unit (in parenthesis).

BBGCMuSo4 simulation normal run	
MET_INPUT	
metdata/430176_FORESEE_v4_2021-2100_NCC_HIRHAM5_rcp85.mtc43	(filename) met file name
4	(int) number of header lines in met file
RESTART	
1	(flag) $1 = \text{read restart}$ ; $0 = \text{dont read restart}$
0	(flag) $1 = \text{write restart}$ ; $0 = \text{dont write restart}$
0	(flag) $1 = use restart metyear; 0 = reset metyear$
restart\muso4.endpoint	(filename) name of the input restart file
restart\muso4.endpoint	(filename) name of the output restart file
TIME_DEFINE	
17	(int) number of meteorological data years
17	(int) number of simulation years
2000	(int) first simulation year
0	(flag) 1 = spinup run; 0 = normal run
6000	(int) maximum number of spinup years
CLIM_CHANGE	
0.0	(degC) - offset for Tmax
0.0	(degC) - offset for Tmin
1.0	(degC) - multiplier for PRCP
1.0	(degC) - multiplier for VPD
1.0	(degC) - multiplier for RAD
COL CONTROL	
CO2_CONTROL 1	(flag) 0=constant; 1=vary with file
390.0	(ppm) constant atmospheric CO <sub>2</sub> concentration
CO2\CO2_ML_2008-2012.txt	(filename) name of the CO <sub>2</sub> file
NDEP_CONTROL	(0.)0
1	(flag) 0=constant; 1=vary with file
0.001400	(kgN/m²/yr) wet+dry atmospheric deposition of N
nitrogen\Ndep_hhs_2021-2100.txt	(filename) name of the N-dep file
SITE	
13.9 13.9 13.9 13.9 13.9 13.9	(%) sand percentage by volume in rock-free soil
34.9 34.9 34.9 34.9 34.9 34.9 34.9	(%) silt percentage by volume in rock-free soil
131.8	(m) site elevation
43.00	(degrees) site latitude (- for S.Hem.)
0.20	(DIM) site shortwave albedo
15.64	(Celsius) mean annual air temperature
18.83	(Celsius) mean annual air temperature range
-9999	(dimless) measured runoff curve number
-9999 -9999 -9999 -9999 -9999 -9999	(g/cm³) bulk density (no data: -9999)

BBGCMuSo4 simulation normal run	
-9999 -9999 -9999 -9999 -9999 -9999	(m³/m³) SWC at SAT (no data: -9999
-9999 -9999 -9999 -9999 -9999 -9999	(m³/m³) SWC at FC (no data: -9999
-9999 -9999 -9999 -9999 -9999 -9999	(m <sup>3</sup> /m <sup>3</sup> ) SWC at WP (no data: -9999
-9999 -9999 -9999 -9999 -9999 -9999	(m³/m³) SWC at HW (no data: -9999
EPC_FILE	
epc\121_14.epc	(filename) EPC filename
W_STATE	
0.0	(kg/m²) water stored in snowpack
0.5	(DIM) initial soil water as a proportion of saturation
C_STATE	
0.001	(kgC/m²) first-year maximum leaf carbon
0.0	(kgC/m²) first-year maximum stem carbon
0.0	(kgC/m²) coarse woody debris carbon
0.0	(kgC/m²) litter carbon, labile poo
0.0	(kgC/m²) litter carbon, unshielded cellulose poo
0.0	(kgC/m²) litter carbon, shielded cellulose poo
0.0	(kgC/m²) litter carbon, lignin poo
0.0	(kgC/m²) soil carbon, fast microbial recycling poo
0.0	(kgC/m²) soil carbon, medium microbial recycling poo
0.0	(kgC/m²) soil carbon, slow microbial recycling poo
0.0	(kgC/m²) soil carbon, recalcitrant SOM (slowest
N_STATE	
0.0	(kgN/m²) litter nitrogen, labile poo
0.0	(kgN/m <sup>2</sup> ) soil nitrogen, labile poo
GROWING_SEASON	
5.00	(kg/m <sup>2</sup> ) crit. amount of snow limiting photosyn.(if no data: 999.9
0	(flag) use GSI index to calculate growing season
1.00	(Celsius) limit1 (under:full constrained) of HEATSUM inde
10.00	(Celsius) limit2 (above:unconstrained) of HEATSUM inde
-2.00	(Celsius) limit1 (under:full constrained) of TMIN inde
5.00	(Celsius) limit2 (above:unconstrained) of TMIN inde
4000	(Pa) limit1 (above:full constrained) of VPD inde
1000	(Pa) limit2 (under:unconstrained) of VPD inde
0	(s) limit1 (under:full constrained) of DAYLENGTH inde
0	(s) limit2 (above:unconstrained) of DAYLENGTH inde
10	(day) moving average (to avoid the effects of extreme events
0.1	GSI limit1 (greater that limit -> start of vegper
0.01	GSI limit2 (less that limit -> end of vegper
GSI.txt	file of the estimated start and end of the V
OUTPUT_CONTROL	
output/jlug	(txt) prefix for output file
control\ctrl_normal.txt	(txt) file of the BBGC variables (control
1	(flag) $1 = \text{write daily output } 0 = \text{no daily output}$

BBGCMuSo4 simulation normal run	
0	(flag) $1 = \text{monthly avg of daily variables } 0 = \text{no monthly avg}$
0	(flag) $1 = \text{annual avg of daily variables}$ $0 = \text{no annual avg}$
0	(flag) $1 = \text{write annual output } 0 = \text{no annual output}$
1	(flag) for on-screen progress indicator
DAILY_OUTPUT	
3	number of daily output variables
623	GPP
622	NEE
649	TR
ANNUAL_OUTPUT	
3	number of annual output variables
636	vegC
638	soilC
639	totalC
MANAGEMENT_SECTION	
PLANTING	
0	(flag) do PLANTING? 0=no; 1=yes; filepath=reading from file'
100 -9999 -9999 -9999 -9999 -9999	(yday) PLANTING day
10.0 -9999 -9999 -9999 -9999 -9999	(double) quantity of seed (kg seed/ha)
40.0 -9999 -9999 -9999 -9999 -9999	(%) C content of seed
90.0 -9999 -9999 -9999 -9999 -9999	(%*) useful part of seed
THINNING	
0	(flag) do THINNING? 0=no; 1=yes; filepath=reading from file
200 -9999 -9999 -9999 -9999 -9999	(yday) THINNING day
0.5 -9999 -9999 -9999 -9999 -9999	(prop) thinning rate
100.0 -9999 -9999 -9999 -9999 -9999	(%) transported part of stem
100.0 -9999 -9999 -9999 -9999 -9999	(%) transported part of leaf
MOWING	(flag) do MOWING? 0=no; 1=yes; filepath=reading from file
0	(flag) mowing method? 0 - on fixday, 1 - if LAI greater than a fixed value
0	(int) fixed value of the LAI before MOWING (fixvalue method)
6.0	(int) fixed value of the LAI after MOWING (fixvalue method)
1.0	(yday*) MOWING day
150 234 -9999 -9999 -9999 -9999	(int*) value of the LAI after MOWING (fixday method)
1.0 1.0 -9999 -9999 -9999 -9999	(%*) transported part of plant material
95.0 95.0 -9999 -9999 -9999 -9999	
GRAZING	
0	(flag) do GRAZING? 0=no, 1=yes; filepath=reading from file
156 262 -9999 -9999 -9999 -9999	(yday*) first day of GRAZING
202 311 -9999 -9999 -9999 -9999	(yday*) last day of GRAZING
381. 3819999 -9999 -9999 -9999	(kg/LSU) weight equivalent of one unit
0.69 1.15 -9999 -9999 -9999 -9999	(LSU/ha*) animal stocking rate: Livestock Units per hectare
13.0 13.0 -9999 -9999 -9999 -9999	(kg dry matter/LSU*) daily ingested dry matter
1.50 1.50 -9999 -9999 -9999 -9999	(prop*) trampling effect (transform. standing dead biome to litter)

BBGCMuSo4 simulation normal run	
25.0 25.0 -9999 -9999 -9999 -9999	(%*) ratio of DM intake formed excremen
100.0 1009999 -9999 -9999 -9999	(%*) ratio of excrement returning to litte
40.0 409999 -9999 -9999 -9999	(%*) carbon content of dry matte
2.0 2.0 -9999 -9999 -9999 -9999	(%*) N content of manur
40.0 409999 -9999 -9999 -9999	(%*) C content of manur
0.01 0.01 -9999 -9999 -9999 -9999	$(kgN_2O-N:kgN^*)$ manure emission factor for direct $N_2O$ emission
0.93 0.93 -9999 -9999 -9999 -9999	(dimless) fraction of nitrogen excretion managed in manure management system
11.0 11.0 -9999 -9999 -9999 -9999	(kgCH <sub>4</sub> /LSU/yr*) manure emission factor for CH <sub>4</sub> emissio
58.0 58.0 -9999 -9999 -9999 -9999	(kgCH <sub>4</sub> /LSU/yr*) fermentation emission factor for CH <sub>4</sub> emission
HARVESTING	
0	(flag) do HARVESTING? 0=no, 1=yes; filepath=reading from file
200 -9999 -9999 -9999 -9999 -9999	(yday*) HARVESTING da
1.0 -9999 -9999 -9999 -9999 -9999	(m²/m²*) LAI after HARVESTING (snag
100.0 -9999 -9999 -9999 -9999 -9999	(%*) transported part of plant materia
PLOUGHING	
0	(flag) do PLOUGHING? 0=no, 1=yes; filepath=reading from fil
201 -9999 -9999 -9999 -9999 -9999	(yday*) PLOUGHING da
1 -9999 -9999 -9999 -9999 -9999	(type*) PLOUGHING depth (0:shallow, 1:medium, 2:deep
0.10 -9999 -9999 -9999 -9999 -9999	(prop*) dissolving coefficient of ploughed biome to litte
FERTILIZING	
0	(flag) do FERTILIZING? 0=no, 1=yes; filepath=reading from fil
60 200 -9999 -9999 -9999 -9999	(yday*) FERTILIZING da
30. 309999 -9999 -9999 -9999	(kgN/ha/day*) (nitrogen from fertilization per day
17. 179999 -9999 -9999 -9999	(%*) nitrate content of fertilize
17. 179999 -9999 -9999 -9999	(%*) ammonium content of fertilize
5.0 5.0 -9999 -9999 -9999 -9999	(%*) carbon content of fertilize
70. 709999 -9999 -9999 -9999	(%*) labile fraction of fertilize
20. 209999 -9999 -9999 -9999	(%*) unshielded cellulose fraction of fertilize
0.0 0.0 -9999 -9999 -9999 -9999	(%*) shielded cellulose fraction of fertilize
10. 109999 -9999 -9999 -9999	(%*) lignin fraction of fertilize
0.05 0.05 -9999 -9999 -9999 -9999	(prop*) dissolving coefficien
90.0 90.0 -9999 -9999 -9999 -9999	(%*) useful pa
0.1 0.1 -9999 -9999 -9999 -9999	(kgN <sub>2</sub> O-N:kgN*) emission factor for N-addition
IRRIGATION	
0	(flag) do IRRIGATION? 0=no, 1=yes; filepath=reading from fil
150 250 -9999 -9999 -9999 -9999	(yday*) IRRIGATION da
30. 30.0 -9999 -9999 -9999 -9999	(kgH <sub>2</sub> O/m2/day*) amount of water
END_INIT	

**Table A.10.** Initialization file (INI) used for normal model run in the Biome-BGCMuSo v.6.2 model. On the left side are sections (in bold) with input data and on the right side are descriptions with information on type of data or unit (in parenthesis).

BBGCMuSo 6.2 simulation normal run	
MET_INPUT	
MET/1900-2017.met	(filename) met file name
4	(int) number of header lines in met file
365	(int) number of simdays in last simyear (truncated year: <= 365)
RESTART	
1	(flag) $1 = \text{read restart}$ ; $0 = \text{dont read restart}$
0	(flag) $1 = \text{write restart}$ ; $0 = \text{dont write restart}$
RST/jst_apriori_muso6.rst	(filename) name of the input restart file
RST/jst_apriori_muso6.rst	(filename) name of the output restart file
TIME_DEFINE	
10	(int) number of simulation years
2008	(int) first simulation year
0	(flag) $1 = \text{spinup run}$ ; $0 = \text{normal run}$
6000	(int) maximum number of spinup years
COA CONTROL	
CO2_CONTROL	(flac) O constant 1 como mid. file
1	(flag) 0=constant; 1=vary with file
395.0	(ppm) constant atmospheric CO2 concentration
CO2/CO2.txt	(filename) name of the CO2 file
NDEP_CONTROL	
1	(flag) 0=constant; 1=vary with file
0.001400	(kgN/m²/yr) wet+dry atmospheric deposition of N
NDEP/Ndep.txt	(filename) name of the N-dep file
SITE	
110.0	(m) site elevation
45.62	(degrees) site latitude (- for S.Hem.)
0.20	(DIM) site shortwave albedo
10.60	(Celsius) mean annual air temperature
21.0	(Celsius) mean annual air temperature range
0.50	(prop.) proprortion of NH4 flux of N-deposition
SOIL_FILE	
SOI/jst_apriori.soi	(filename) SOIL filename
SON/31_aprion.son	(menanc) soil menanc
EPC_FILE	
jst_apriori.epc	(filename) EPC filename
MANAGEMENTE EN E	
MANAGEMENT_FILE	(6) (10)(6)
MGM/jst_muso6.mgm	(filename) MGM filename (or "none")
CIMULATION CONTROL	

 ${\bf SIMULATION\_CONTROL}$ 

BBGCMuSo 6.2 simulation normal	
0	(flag) phenology flag (1 = MODEL PHENOLOGY 0 = USER-SPECIFIED FENOLOGY)
0	(flag) vegper calculation method if MODEL PHENOLOGY is used (0: original, 1: GSI)
0	(flag) transferGDD flag (1= transfer calc. from GDD 0 = transfer calc. from EPC)
1	(flag) q10 flag (1 = temperature dependent q10 value; 0= constans q10 value)
1	(flag) acclimation flag of photosynthesis (1 = acclimation $0$ = no acclimation)
1	(flag) acclimation flag of respiration $(1 = acclimation 0 = no acclimation)$
1	(flag) CO <sub>2</sub> conductance reduction flag (0: no effect, 1: multiplier)
0	(flag) soil temperature calculation method (0: Zheng, 1: DSSAT)
0	(flag) soil hydrological calculation method (0: tipping DSSAT, 1: Richards)
0	(int) discretization level of soil hydr.calc.[Richards-method] (0: low, 1: medium, 2: high)
0	(flag) photosynthesis calculation method (0: Farquhar, 1: DSSAT)
0	(flag) evapotranspiration calculation method (0: Penman-Montieth, 1: Priestly-Taylor)
0	(flag) radiation calculation method (0: SWabs, 1: Rn)
0	(flag) soilstress calculation method (0: based on VWC, 1: based on transp. demand)
W_STATE	
0.0	(kg/m²) water stored in snowpack
1.0	(DIM) initial soil water as a proportion of field capacity
CN_STATE	
0.001	(kgC/m <sup>2</sup> ) first-year maximum leaf carbor
0.001	(kgC/m <sup>2</sup> ) first-year maximum fine root carbon
0.001	(kgC/m²) first-year maximum fruit carbon
0.001	(kgC/m <sup>2</sup> ) first-year maximum softstem carbon
0.001	(kgC/m <sup>2</sup> ) first-year maximum live woody stem carbor
0.001	(kgC/m <sup>2</sup> ) first-year maximum live coarse root carbor
0.0*	(kgC/m <sup>2</sup> ) coarse woody debris carbon
0.0*	(kgC/m²) litter carbon, labile poo
0.0*	(kgC/m²) litter carbon, unshielded cellulose poo
0.0*	(kgC/m²) litter carbon, shielded cellulose poo
0.0*	(kgC/m²) litter carbon, lignin poo
0.0*	(kgC/m <sup>2</sup> ) soil carbon, fast microbial recycling poo
0.0*	(kgC/m <sup>2</sup> ) soil carbon, medium microbial recycling poo
0.0*	(kgC/m²) soil carbon, slow microbial recycling poo
0.0*	(kgC/m²) soil carbon, recalcitrant SOM (slowest)
0.0*	(kgN/m²) litter nitrogen, labile poo
0.0*	(kgN/m²) soil mineralized nitrogen, NH <sub>4</sub> pool
0.0*	(kgN/m²) soil mineralized nitrogen, NO <sub>3</sub> pool
CLIM_CHANGE	
0.0	(degC) - offset for Tmax
0.0	(degC) - offset for Tmir
1.0	(degC) - multiplier for PRCF
1.0	(degC) - multiplier for VPD
1.0	(degC) - multiplier for RAD
CONDITIONAL_MANAGEMEN	T_STRATEGIES
0	(flag) conditional mowing ? 0 - no, 1 - yes
	(m <sup>2</sup> /m <sup>2</sup> ) fixed value of the LAI before MOWING

BBGCMuSo 6.2 simulation norma	al run
0.0	(m²/m²) fixed value of the LAI after MOWING
0.0	(%) transported part of plant material after MOWING
0	(flag) conditional irrigation? 0 - no, 1 - yes
0.0	(prop) SMSI before cond. IRRIGATION (-9999: SWCratio is used)
0.0	(prop) SWCratio of rootzone before cond. IRRIGATION (-9999: SMSI is used)
0.0	(prop) SWCratio of rootzone after cond. IRRIGATION
0.0	(kgH <sub>2</sub> O/m2) maximum amount of irrigated water
OUTPUT_CONTROL	
output/jst_n	(filename) output prefix
1	(flag) writing daily output (0 = no; 1 = binary; 2 = ascii; 3 = on-screen)
0	(flag) writing monthly average of daily output (0 = no; 1 = binary; 2 = ascii; 3 = on-screen)
0	(flag) writing annual average of daily output (0 = no; 1 = binary; 2 = ascii; 3 = on-screen)
2	(flag) writing annual output (0 = no; 1 = binary; 2 = ascii; 3 = on-screen)
1	(flag) for on-screen progress indicator
DAILY_OUTPUT	
3	number of daily output variables
3007	daily_nee
3009	daily_gpp
3014	daily_tr
ANNUAL_OUTPUT	
8	number of annual output variables
3024	cum_nee
3025	cum_gpp
3029	cum_tr
3158	LDaboveC_w
3060	litrC
458	soilC[0]
459	soilC[1]
460	soilC[2]
END_INIT	

**Table A.11.** Initialization file (INI) used for spinup model run in model calibration. On the left side are sections (in bold) with input data, and on the right side are descriptions with information on type of data or unit (in parenthesis).

BBGCMuSo 6.2 simulation spinup run	
MET_INPUT	
MET/1900-2017.met	(filename) met file name
4	(int) number of header lines in met file
365	(int) number of simdays in last simyear (truncated year: <= 365)
RESTART	
0	(flag) $1 = \text{read restart}$ ; $0 = \text{dont read restart}$
1	(flag) $1 = \text{write restart}$ ; $0 = \text{dont write restart}$
RST/jst_apriori_muso6.rst	(filename) name of the input restart file
RST/jst_apriori_muso6.rst	(filename) name of the output restart file
TIME_DEFINE	
107	(int) number of simulation years
1900	(int) first simulation year
1	(flag) $1 = \text{spinup run}; 0 = \text{normal run}$
6000	(int) maximum number of spinup years
CO2_CONTROL	
1	(flag) 0=constant; 1=vary with file
290.0	(ppm) constant atmospheric CO <sub>2</sub> concentration
CO2/CO2.txt	(filename) name of the CO <sub>2</sub> file
NDEP_CONTROL	
1	(flag) 0=constant; 1=vary with file
0.000200	(kgN/m²/yr) wet+dry atmospheric deposition of N
NDEP/Ndep.txt	(filename) name of the N-dep file
SITE	
110.0	(m) site elevation
45.62	(degrees) site latitude (- for S.Hem.)
0.20	(DIM) site shortwave albedo
10.60	(Celsius) mean annual air temperature
21.0	(Celsius) mean annual air temperature range
0.50	(prop.) proprortion of NH <sub>4</sub> flux of N-deposition
SOIL_FILE	
SOI/jst_apriori.soi	(filename) SOIL filename
EPC_FILE	
jst_apriori.epc	(filename) EPC filename
MANAGEMENT_FILE	
MGM/jst_muso6.mgm	(filename) MGM filename (or "none")
SIMULATION_ CONTROL	

BBGCMuSo6.2 simulation spinup run	
0 (flag) pheno	ology flag (1 = MODEL PHENOLOGY 0 = USER-SPECIFIED FENOLOGY)
0 (flag) veg	pper calculation method if MODEL PHENOLOGY is used (0: original, 1: GSI)
0 (fla	ng) transferGDD flag (1= transfer calc. from GDD 0 = transfer calc. from EPC)
1	$(flag) \ q10 \ flag \ (1 = temperature \ dependent \ q10 \ value; \ 0 = constans \ q10 \ value)$
1	(flag) acclimation flag of photosynthesis (1 = acclimation $0$ = no acclimation)
1	(flag) acclimation flag of respiration (1 = acclimation $0 = no$ acclimation)
1	(flag) $CO_2$ conductance reduction flag (0: no effect, 1: multiplier)
0	(flag) soil temperature calculation method (0: Zheng, 1: DSSAT)
0	(flag) soil hydrological calculation method (0: tipping DSSAT, 1: Richards)
0 (int) discr	etization level of soil hydr.calc.[Richards-method] (0: low, 1: medium, 2: high)
0	(flag) photosynthesis calculation method (0: Farquhar, 1: DSSAT)
0 (flag) e	vapotranspiration calculation method (0: Penman-Montieth, 1: Priestly-Taylor)
0	(flag) radiation calculation method (0: SWabs, 1: Rn)
0 (flag	) soilstress calculation method (0: based on VWC, 1: based on transp. demand)
W_STATE	
0.0	(kg/m²) water stored in snowpack
1.0	(DIM) initial soil water as a proportion of field capacity
CN_STATE	
0.001	(kgC/m²) first-year maximum leaf carbon
0.001	(kgC/m²) first-year maximum fine root carbon
0.001	(kgC/m²) first-year maximum fruit carbon
0.001	(kgC/m²) first-year maximum softstem carbon
0.001	(kgC/m²) first-year maximum live woody stem carbon
0.001	(kgC/m²) first-year maximum live coarse root carbon
0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	(kgC/m²) coarse woody debris carbon
0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	(kgC/m <sup>2</sup> ) litter carbon, labile pool
0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	(kgC/m²) litter carbon, unshielded cellulose pool
0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	(kgC/m²) litter carbon, shielded cellulose pool
0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	(kgC/m²) litter carbon, lignin pool
0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	(kgC/m²) soil carbon, fast microbial recycling pool
0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	(kgC/m²) soil carbon, medium microbial recycling pool
0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	(kgC/m <sup>2</sup> ) soil carbon, slow microbial recycling pool
0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	(kgC/m²) soil carbon, recalcitrant SOM (slowest)
0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	(kgN/m²) litter nitrogen, labile pool
0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	(kgN/m²) soil mineralized nitrogen, NH <sub>4</sub> pool
0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0 0.0	(kgN/m²) soil mineralized nitrogen, NO <sub>3</sub> pool
CLIM_CHANGE	
0.0	(degC) - offset for Tmax
0.0	(degC) - offset for Tmin
1.0	(degC) - multiplier for PRCP
1.0	(degC) - multiplier for VPD
1.0	(degC) - multiplier for RAD
CONDITIONAL_MANAGEMENT_STRATEGIES	
0	(flag) conditional mowing ? 0 - no, 1 - yes
0.0	$(m^2/m^2)$ fixed value of the LAI before MOWING

BBGCMuSo6.2 simulation spinup run	
0.0	(m <sup>2</sup> /m <sup>2</sup> ) fixed value of the LAI after MOWING
0.0	(%) transported part of plant material after MOWING
0	(flag) conditional irrigation? 0 - no, 1 - yes
0.0	(prop) SMSI before cond. IRRIGATION (-9999: SWCratio is used)
0.0	(prop) SWCratio of rootzone before cond. IRRIGATION (-9999: SMSI is used)
0.0	(prop) SWCratio of rootzone after cond. IRRIGATION
0.0	(kgH <sub>2</sub> O/m <sup>2</sup> ) maximum amount of irrigated water
OUTPUT_CONTROL	
output/jst_s	(filename) output prefix
1	(flag) writing daily output ( $0 = no$ ; $1 = binary$ ; $2 = ascii$ ; $3 = on-screen$ )
0	(flag) writing monthly average of daily output $(0 = no; 1 = binary; 2 = ascii; 3 = on-screen)$
0	(flag) writing annual average of daily output $(0 = no; 1 = binary; 2 = ascii; 3 = on-screen)$
2	(flag) writing annual output (0 = no; 1 = binary; 2 = ascii; 3 = on-screen)
1	(flag) for on-screen progress indicator
DAILY_OUTPUT	
3	number of daily output variables
3007	daily_nee
3009	daily_gpp
3014	daily_tr
ANNUAL_OUTPUT	
8	number of annual output variables
3024	cum_nee
3025	cum_gpp
3029	cum_tr
3158	LDaboveC_w
3060	litrC
458	soilC[0]
459	soilC[1]
460	soilC[2]
END_INIT	

## **CURRICULUM VITAE**

Doroteja Bitunjac (born Dimoski) was born on 29<sup>th</sup> of February 1992 in Zagreb, Croatia. She finished primary and high school in Velika Gorica. In 2015, she graduated from the Faculty of Forestry University of Zagreb, with a master's thesis under the title "Effect of establishment methods on structural properties of young Pedunculate oak (*Quercus robur* L.) stands in forest basin Kalje". On the occasion of the 117th anniversary of the Faculty of Forestry, University of Zagreb, she was awarded "Academic Milan Anić" for completing Graduate Studies with great success.

Her career at the Croatian Forest Research Institute, Division for Forest Management and Forestry Economics, started in 2016 when she was employed as a professional trainee and was involved in the project "Soil carbon stock changes and calculation of soil organic carbon, total soil nitrogen C:N trends" (Nr:10-14-1442/79) of Croatian Environment Agency, for a one-year period. From September 2020 onwards, she has been employed in the same Division, as a research assistant on a Croatian Science Foundation project "Modelling Forest Carbon Stocks, Fluxes and Forest Risks under Future Climate Scenarios – MODFLUX" (HRZZ-IP-2019-4-6325). In the same year, she enrolled in Postgraduate Doctoral Studies of Forestry and Wood Technology at the Faculty of Forestry and Wood Technology University of Zagreb.

By now, she has actively participated in numerous international and domestic scientific conferences and held two invited lectures on abroad scientific institutions. At the European Geoscience Union conference, in the years 2023 and 2024, she was a co-convener and co-chairperson of the sessions under the topics of estimation of carbon stocks and fluxes in the ecosystems, as a support for the National GHG Inventory Reports under the UNFCCC. Aside from the MODFLUX project she actively participates in the COST action PROCLIAS (CA19139). Since January 2022 she has held the position of Assistant Editor in the scientific journal SEEFOR-South-east European Forestry. Aside from scientific work, she is involved in the preparation of studies and reports needed for the enhancement of the Croatian National GHG Inventory Reports.

## Scientific publications

1. Merganičová K, Merganič J, Dobor L, Hollós R, Barcza Z, Hidy D, Sitková Z, Pavlenda P, Marjanović H, Kurjak D, Bošeľa M, **Bitunjac D**, Ostrogović Sever MZ., Novák J, Fleischer

- P, Hlásny T, 2024. Biogeochemical model Biome-BGCMuSo v6.2 provides plausible and accurate simulations of carbon cycle in Central European beech forests. Geoscientific Model Development. [preprint, in review].
- 2. **Bitunjac D**, Ostrogović Sever MZ, Sever K, Merganičová K, Marjanović H, 2023. Dead wood volume-to-carbon conversion factors by decay class for ten tree species in Croatia and eight tree genera globally. Forest Ecology and Management 549:121431.
- 3. Ostrogović Sever MZ, Barcza Z, Hidy D, Kern A, **Dimoski D**, Miko S, Hasan O, Grahovac B, Marjanović H, 2021. Evaluation of the Terrestrial Ecosystem Model Biome-BGCMuSo for Modelling Soil Organic Carbon under Different Land Uses. Land 10(9):968.