OrgBalt LIFE project

EU LIFE Programme project
"Demonstration of climate change mitigation potential of nutrients rich organic soils in Baltic States and Finland"

REPORT

ON IMPLEMENTATION OF THE PROJECT

WORK PACKAGE

MONITORING OF THE IMPLEMENTATION OF PROJECT ACTIVITIES

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"LIFE OrgBalt compiled the first regional Baltic/ Finnish GHG emission factors for managed nutrient-rich organic soils (current and former peatlands), which have been made available for the customary scientific review and further verification for national GHG inventories in the hemiboreal region in Finland and the Baltic countries. While the project analysed selected CCM measures for drained organic soils in agriculture and forestry and developed spatial models and tools, it also identified remaining knowledge gaps. To bridge the remaining limitations and fill the gaps, it is essential to continue GHG measurements and model development, as well to broaden and complete the scope of the evaluated CCM measures in the after-LIFE-project period, notably by including rewetting and restoration of peatlands that are currently considered to be among the most recommended CCM measures on drained peatlands in the EU. In addition, the developed Simulation and PPC models still include limited macroeconomic considerations and lack assessment of all environmental impacts. For all these reasons, these models should be used carefully in CCM strategy development for identification of gaps in climate neutrality transition policy and funding frameworks and need further optimization for broader applicability as decision-making tools."

2022

CONTENTS

Abbreviations

ABBREVIATION	DEFINITION
AFOLU	agriculture, forestry and other land use
C	carbon
Ca	calcium
CCM	climate change mitigation measures
CO2	carbon dioxide
CH4	methane
DNA	deoxyribonucleic acid
EF	emission factor
GHG	greenhouse gas or greenhouse gases
GLOSOLAN	Global Soil Laboratory Network
IPCC	Intergovernmental Panel on Climate Change
IPCC Guidelines 2006	2006 IPCC Guidelines for National Greenhouse Gas Inventories
IPCC KP Supplement	2013 Revised Supplementary Methods and Good Practice Guidance Arising from the Kyoto Protocol
IPCC Wetlands Supplement	2013 Supplement to the 2006 Guidelines for National Greenhouse Gas Inventories: Wetlands
IR	infrared
IRS	infrared spectroscopy
ITS	internal transcribed spacer
IRGA	infra-red gas analyser
K	potassium
LSFRI Silava	Latvian State Forest Research institute "Silava"
Luke	Natural Resources Institute Finland "Luke"
LULUCF	land use, land use change and forestry
MEPRD	Ministry of Environmental Protection and Regional Development
Mg	magnesium
N	nitrogen
N2O	nitrous oxide
NEE	net ecosystem exchange
NO3	nitrate
OTU	operational taxonomic unit
P	phosphorus
PCR	polymerase chain reaction
pH	potential of hydrogen
rRNA	ribosomal ribonucleic acid
UT	University of Tartu
WOM	without measures
WAM	with additional measures

Introduction

The aim of monitoring the implementation of activities is to evaluate the impact of the implemented measures on greenhouse gas (GHG) emissions in the 17 demonstration sites and 36 reference sites established under action C3 and compare the identified impacts against the target indicators set out in the monitoring guidelines. In total, 53 sites are investigated.

The implementation of activities is monitored through three reports: initial, mid-term, and final.

The midterm monitoring report will include the description of the different field measurements used within the project to quantify greenhouse gas emissions from nutrient-rich organic soils and progress information on the first year of measurement activities.

One of the main tasks of the LIFE OrgBalt is, in fact, the improvement of methodologies for the calculation (Action C1) and projections (Actions C2 and C5) of GHG emissions from drained nutrient-rich organic soils (grassland, cropland, forest land and managed wetlands), thus contributing to the development of national GHG inventory systems and to the implementation of national and global CCM targets. The main indicators of the success of Actions C1, C2 and C5 will be that key sources of GHG emissions or CO_2 removals on organic soils are reported according to tier 3 methodology as preferred to tier 1 or tier 2 level reporting by the Intergovernmental Panel on Climate Change (IPCC) guidelines, as well as the impact of the climate change mitigation (CCM) measures implemented in managed cropland, grassland and forest land on organic soil.

GHG emissions in demo sites are monitored using GHG measurement methodologies applied in Action C1, including supplementary data on biomass production, weather conditions, soil and water properties. The long-term impact will be modelled using the scenario analysis tool elaborated within the scope of Action C2 and C5. Monitoring data will be used to update the scenario analysis tool for short-term actions like changes in crop rotation and the application of wood ash. However, the continuation of the measurements after completing the project is of special importance to elaborate accurate impact assessment curves of climate change mitigation (CCM) measures.

The gas measurements in all sampling sites (reference sites established within the scope of C1 and demo sites established within the scope of C3) will be used to improve GHG emission factors (EFs) elaborated in Action C1 and will be utilised in the final revision of the catalogue of CCM measures calculation and projections for WOM (Without measures) and WAM (With additional measures) projections, including the recommendation for application of CCM measures for the management of organic soils depending on land use, soil properties and climate projections.

Furthermore, considering high research value of the established demo sites, they will be used for monitoring GHG emissions from lands under transition period within the scope of the national CCM related research projects, as well as in training and education activities. Scientific outputs of the project will be monitored by the success of the implementation of the proposed methodologies and the publishing of Project results.

The benefits, results, and effectiveness of the LIFE OrgBalt project actions are measurable and will be evaluated and documented under the monitoring actions, compared with initial data, and checked if they are online with the project objectives and expected results. Specific indicators (measurements of CO_2, CH_4 and N_2O fluxes or emissions, Tier 3 level methodology for emission from relevant sources calculation under national GHG reporting, content of national reports related to international environmental policy agreements) to detect the impact of the project activities at local (demonstration site level) and national level, are selected and regular monitoring is foreseen.

The methodologies which will be applied to evaluate the project results are described in further chapters. Due to the rapid developments in this field, the methodologies may be updated according to up-to-date best practices. The impact of the project climate change mitigation targeted activities implemented within demonstration sites will be assessed by collecting and analysing the values of the reduction of the GHG emissions in the demonstration sites.

1 Monitoring methodology of the impact of activities

1.1 Field measurements

Organic soils contribute to the atmospheric greenhouse gas (GHG) concentrations, as they can either remove or emit GHG and perform as globally extensive carbon (C) and nitrogen (N) stores. Currently, both the IPCC (2006) agriculture, forestry and other land use (AFOLU) guidelines and the IPCC (2014) Wetlands Supplement may be used for reporting the annual GHG emissions or removals for soils under anthropogenic land uses. Area-based emission factors (EFs), describing the net annual soil GHG emissions/removals, have been developed to reflect the impacts of ecosystem type, land management, and environmental conditions. Countries may opt for different methodological levels in their GHG reporting, so-called Tier 1 to 3, where Tier 1 is the most straightforward approach with default EFs of the IPCC. The accuracy of EFs can be improved as more peer-reviewed data become available and quantify a wider set of specific management options and ecological conditions for a given country or region. In Life OrgBalt, we are working to form Tier-2 level EFs for soil CO_2, CH_4, and N_2O balances in monitoring included site types.

Quantifying the soil GHG balance, especially for carbon dioxide (CO_2), in forests and other ecosystems on organic soils is technically challenging. Monitoring needs to take into account that:

- C-sequestration into plant biomass takes place in a potentially voluminous and diverse vegetation community with uneven spatial distribution,
- C transfer from biomass into dead organic matter takes place both in the aboveground and belowground parts,
- physical and biochemical characteristics in organic soils change over time,
- CO_2 release through heterotrophic processes takes place both in recently deposited litter and in a soil composed of previously accumulated dead organic matter,
- CO_2 formed in the heterotrophic processes in the soil must be separated from similarly large CO_2 emissions formed in autotrophic root respiration in flux measurements,
- rates of biological processes change over the year and differ between years depending on weather conditions, stand development and management.

In this document, "soil CO_2 balance" includes C transfer fluxes to the soil as above- and belowground litter and losses by decomposition of litter and soil organic matter (Figure 1).

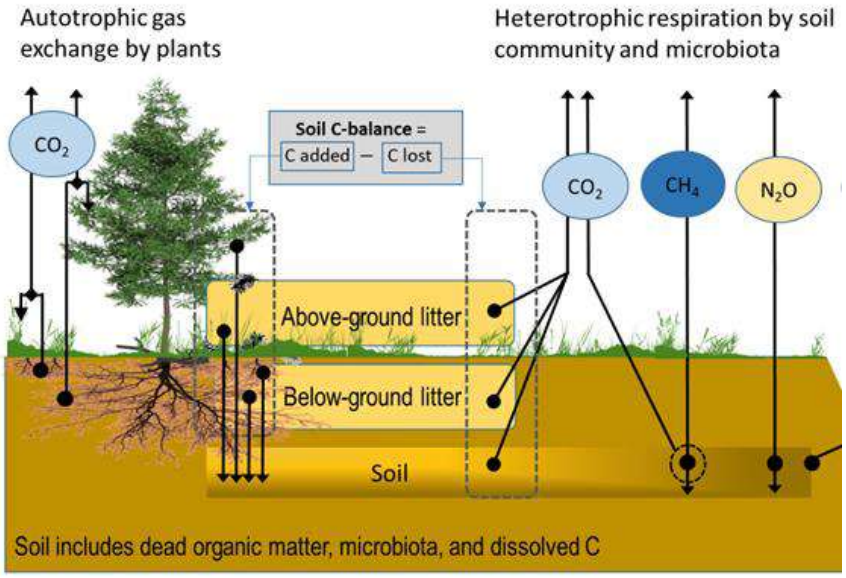

Figure 1. CO_2, CH_4, and N_2O fluxes and mass transfer components (arrows indicate flux/transfer direction) contribute to soil C-stock changes in a forest ecosystem on drained organic soil (as in IPCC, 2014), modified from Jauhiainen et al. (2019).

Soil CO_2 balance is estimated using the chambers-based measurement technique, which typically includes CO_2 exchange monitoring at the soil surface without ground vegetation and roots. Trenching (explained in subsequent paragraphs) prevents live root presence and regular sprout cutting prevents vegetation growth on the soil surface. Annual soil CO_2 balance is formed by using (1) summarised CO_2 flux data over the year in monitoring and (2) data on mass-based C stock changes, such as C inputs and decomposition as litter aboveground and belowground. Removal/inclusion of above ground litter in CO_2 flux monitoring needs to be considered in soil CO_2 balance equation, i.e., if the litter is removed from the measurement plots, the rates of both the input and decomposition of above ground litter need to be estimated.

For forming the EFs for methane (CH_4) and nitrous oxide (N_2O), there is no guidance on how living vegetation presence or litter dynamics should be taken into account in flux measurements, except that vegetation presence can be reported for CH_4 monitoring locations (IPCC, 2014). Wetland plants that have roots with aerenchymatous tissue are known to pipe out CH_4 from waterlogged peat layers (Askaer et al., 2011; Kokkonen et al., 2019) or attenuate the emissions in drained sites (Strack et al., 2006). Furthermore, belowground biomass disturbance, e.g., rhizosphere and mycorrhizal mycelia removal by trenching, has resulted in increased N_2O flux in drained organic forest soils (Ernfors et al., 2011). However, it seems clear that vegetation should be kept intact in studies of CH_4 and N_2O fluxes if possible. Annual soil CH_4 and N_2O balance are based on modelled fluxes over the year in monitoring.

The LIFE OrgBalt project aims to implement a wide range of innovative organic soil management measures to demonstrate how these areas can be managed sustainably, considering economic, social and climate aspects. 17 project demonstration sites have been established in Latvia and Finland. In

the project, GHG fluxes are monitored in 53 sites, including all project demonstration sites and reference sites. Table 1 shows the list of all implemented demonstration sites with a short description of the potential benefits of the applied climate change mitigation measures.

Table 1. LIFE OrgBalt demonstration sites

#	Country	Code	CCM measure	Potential CCM benefits
1	Latvia	LVC303	Paludiculture - afforestation of grassland with black alder and birch	Potential benefits of the establishment of forest paludiculture in rewetted grassland: • Reduced GHG emissions from the soil due to the improvement of the water regime by mounding and establishment of the network of shallow furrows to drain exceeding surface water • Reduction of risks associated with natural disturbances in forests with wet organic soils • Accumulation of CO2 in living and dead biomass, soil and litter and replacement effect of forest biofuel and harvested wood products
2	Latvia	LVC302	Conventional afforestation considering shorter rotation	Potential benefits of afforestation: • Reduced GHG emissions from soil • Accumulation of CO2 in living and dead biomass, soil and litter and replacement effect of forest biofuel and harvested wood products • Shorter rotation and more intensified management ensure higher yield and replacement effect, as well as reduces carbon losses due to root rot and other disturbances
3	Latvia	LVC308	Continuous forest cover as a forest regeneration method in spruce stands	Potential benefits of continuous forest cover: • Reduced CH_4 and N_2O emissions from soil due to avoiding of increase in the groundwater level after harvesting
4	Latvia	LVC307	Application of wood ash after commercial thinning in spruce stands	Potential benefits of wood ash application in the forest on organic soils: • Increased CO2 removals in living biomass, deadwood, soil, litter and harvested wood products due to improved growing conditions and additional increment in living biomass
5	Latvia	LVC311	Riparian buffer zone in forest land planted with black alder	Potential benefits of improved planting of black alder in riparian buffer zone: • Reduced GHG emissions from soil due to the improvement of the water regime by mounding and establishment of network of shallow furrows to drain exceeding surface water

#	Country	Code	CCM measure	Potential CCM benefits
				• Reduction of risks associated with natural disturbances in forests with wet organic soils • Accumulation of CO2 in living and dead biomass, soil and litter and replacement effect of forest biofuel and harvested wood products
6	Latvia	LVC309	Semi-natural regeneration of clear-felling sites with grey alder without reconstruction of drainage systems	Potential benefits of forest stand regeneration without reconstruction of drainage systems (from naturally wet or rewetted organic soils): • Reduced GHG emissions from the soil due to the improvement of the water regime by mounding and establishment of the network of shallow furrows to drain exceeding surface water • Reduction of risks associated with natural disturbances in forests with wet organic soils • Accumulation of CO2 in living and dead biomass, soil and litter and replacement effect of forest biofuel and harvested wood products
7	Latvia	LVC306	Agroforestry - fast growing trees and grass	Potential benefits of agroforestry: • Increased CO2 removals in living biomass and soil • Reduced GHG emissions from soil and replacement effect of woody and herbaceous biofuel and harvested wood products
8	Latvia	LVC310	Fast growing species in riparian buffer zones	Potential benefits of fast-growing species in riparian buffer zones: • Increased CO2 removals in living biomass and soil • Replacement effect of woody and herbaceous biofuel and harvested wood products • Avoided nutrients leakage from farmlands
9	Latvia	LVC301	Conversion of cropland used for cereal production into grassland considering periodic ploughing	Potential benefits of cropland conversion to grassland: • Reduced GHG emissions from soil • Increased carbon stock in soil and belowground biomass • Reduced risks of nutrient leaching and soil erosion
10	Latvia	LVC305	Controlled drainage of grassland considering even groundwater level during the whole vegetation period	Potential benefits of controlled drainage: • Reduced GHG emissions from organic soils due to reduced fluctuations of groundwater level

#	Country	Code	CCM measure	Potential CCM benefits
				• Reduced leaching of nutrients to surface water bodies • In summer drought additional water is available to meet crop demand ensuring higher carbon inputs into soil
11	Latvia	LVC304 a	Introduction of legumes in conventional farm crop rotation	Potential benefits of legumes in conventional crop rotation: • Reduced N2O emissions from soil reported in agriculture sector because of avoided mineral fertiliser application and gradual nitrogen input by symbiotic organisms • Increased carbon input with plants ensuring increased soil carbon stock
12	Latvia	LVC304 b	Introduction of legumes in conventional farm crop rotation	Potential benefits of legumes in conventional crop rotation: • Reduced N2O emissions from soil reported in agriculture sector because of avoided mineral fertiliser application and gradual nitrogen input by symbiotic organisms Increased carbon input with plants ensuring increased soil carbon stock
13	Latvia	LVC313	Strip harvesting in pine stands	Potential benefits of strip harvesting: • Reduced CH4 and N2O emissions from soil due to avoiding of increase of the groundwater level after harvesting in comparison to clear-felling
14	Latvia	LVC312	Forest regeneration (coniferous trees) without reconstruction of drainage systems	Potential benefits of forest regeneration with coniferous trees without reconstruction of drainage systems: • Reduced GHG emissions from soil due to improvement of water regime by mounding and establishment of network of shallow furrows to drain exceeding surface water • Reduction of risks associated with natural disturbances in forests with wet organic soils • Accumulation of CO2 in living and dead biomass, soil and litter and replacement effect of forest biofuel and harvested wood products
15	Finland	FIC301	**Continuous cover forestry on peatland.** Selective felling without full ditch network maintenance. Conventional clear cut	Potential benefits of continuous forest cover forestry practices: • Lower impact to environment conditions in forest stand • Remaining tree stand evapotranspiration controls soil water-table

#	Country	Code	CCM measure	Potential CCM benefits
			and uncut plots are used as comparison. Three sites in monitoring in South Finland.	• Reduced/no need for ditch network maintenance • Reduced change in soil CO2 emission after harvesting • Reduced inputs of water and plant nutrients to surface water bodies
16	Finland	FIC302	**Shifting to continuous cover forestry on peatland.** Forest regeneration following harvesting of overstorey. Conventional clearcut + ditch mounding + planting and uncut forest are used for comparison. Three sites in monitoring in South Finland.	Potential benefits of continuous forest cover forestry practices: • Lower impact on environmental conditions in the forest stand • Remaining tree stand evapotranspiration controls soil water-table • Reduced/no need for ditch network maintenance • Reduced change in soil CO2 emission after harvesting • Reduced inputs of water and plant nutrients to surface water bodies
17	Finland	FIC303	**Shifting to continuous cover forestry on peatland.** Forest regeneration following small gap harvesting and natural regeneration. A spruce shelter tree stand with natural regeneration is used as a comparison. Two sites in monitoring in North Finland.	Potential benefits of continuous forest cover forestry practices: • Lower impact on environmental conditions in the forest stand • Remaining tree stand evapotranspiration controls soil water-table • Reduced/no need for ditch network maintenance • Reduced change in soil CO2 emission after harvesting • Reduced inputs of water and plant nutrients to surface water bodies

1.1.1 Greenhouse gas flux monitoring

Two dark closed chamber methods are used to monitor GHG fluxes between soil and the atmosphere in field conditions. In both chamber methods, a known area and volume of airspace on top of the monitored soil surface are closed by a chamber headspace. GHG concentration increases inside the chamber over the time of the deployment period, and the GHG flux rate is determined by combining information on the closed soil surface area, the volume of the closed airspace, and the GHG concentrations over the deployment period. The practical difference between the methods is the timing between the air sampling event at the field and the GHG gas concentration analysis that provides the final GHG flux reading. The first method involves a series of individual air samples collected during deployment time from the closed chamber at the field, storing the samples for

transportation, subsequent GHG concentration analysis in the laboratory and calculation of the GHG fluxes (hereafter referred also as method-1). The second method involves closing the monitored airspace by closed chamber and circulation of air between the closed chamber and GHG analyser, and instant GHG concentration analysis and flux readout provided at the field (hereafter referred also as method-2). The first method is often referred to as the 'static chamber method' and the latter as the 'dynamic chamber method'.

Traditionally the static chamber method has been more practical because (1) the GHG concentration analysis is based on common laboratory equipment and the analytical method by gas chromatography, and (2) several important GHG species, including CO_2, CH_4 and N_2O, can be analysed from the same gas sample, which usually makes the cost per sample affordable. The downside of the method is general slowness and labour intensiveness (e.g., long deployment time at air sample collection, especially for CH_4 and N_2O, potentially long time in sample transport/storage before the analysis by gas chromatography) before the actual GHG fluxes can be calculated.

The first portable gas analysers suitable for use in field conditions during vegetation season and using the dynamic chambers were for CO_2 data collection (trademarks such as ADC, EGM, Licor, etc.). Monitoring multiple GHG species (CO_2 and/or CH_4 and/or N_2O) has become possible in field conditions only recently due to technical development in instrumentation, and the price of analysers (e.g., Licor, Picarro, Gasmet, etc.) have gradually become more affordable. The key benefit of this method (in comparison to static chambers) is speed due to short deployment time and instantly available flux readout(s) for GHG(s). Instantly available GHG flux readout at the monitoring location allows renewed flux monitoring if a technical failure occurs (e.g., chamber leakage). Short deployment time also makes it possible to collect GHG data from a higher number of monitoring points/conditions than the static chamber method. The downside of the approach includes the high price of the analyser, still somewhat developing techniques for use in demanding weather/climate conditions and sites, and analyser-specific limitations in GHG species included.

'Method-1' on-site gas sampling using dark closed static chambers (e.g., Hutchinson and Livingston, 1993; Ojanen et al., 2010) is used to measure total ecosystem respiration (R_{total} CO_2) of the soil, CH_4 and N_2O. Collars (Ø 50 cm) in 5 replicates are pre-installed in the soil to form permanent bases for chambers. Vegetation within the collar enclosed soil surfaces is not disturbed. During field management operations, collars in cropland and grassland sites are temporarily removed. During a 30-60 minute (depending on the volume of the chambers) long deployment period, four air samples are drawn from the cylindrical chamber headspace into pre-evacuated glass bottles. CH_4 and N_2O concentrations are analysed in the lab using gas chromatography to analyse soil net gas exchange determination for these gases. Method-1 is used in every site during winter as this method is not so demanding for weather conditions.

In grasslands, the transparent closed dynamic chamber is also used to assess the net ecosystem exchange of CO_2 during the growing period. 'Method-2' is used for in-situ CO_2 flux monitoring by using a closed dynamic chamber (Järveoja et al., 2016; Ojanen et al., 2012). Concentration change and flux are determined using a portable gas analyser (e.g. EGM-4, EGM-5, Licor). On each site, 3 permanent flux monitoring point groups (i.e. sub-plots) are established for heterotrophic soil CO_2

emissions monitoring. Each flux monitoring point group includes 3 monitoring points (Ø 30 cm), i.e. total of 9 monitoring points at each site. To prevent autotrophic root respiration contributions into CO_2 fluxes, flux monitoring enclosed surfaces are trenched and root-ingrowth preventing cloth is installed beforehand (belowground litter deposition and carbon loss as CO_2 will be determined separately). All monitoring surfaces will be kept free from litter during monitoring (litter deposition and emissions from litter decomposition will be determined separately). The soil respiration chamber is set gas-tightly on the soil surface. During each flux measurement, CO_2 concentration and temperature inside the chamber are recorded over a deployment period of up to 3 min. A higher number of monitoring points is reserved for CO_2 monitoring based on the high importance of this specific greenhouse gas from drained organic soils (IPCC, 2014). This approach yields a sufficient amount of observed data of CO_2 emissions, keeping in mind that several different processes, both spatially and temporarily, contribute to the emission (Hiraishi et al., 2013), and monitoring by IRGA allows relatively fast CO_2 flux data collection. After each monitoring round at the field, GHG flux data is uploaded to the server maintained in Luke. Data quality is automatically pre-screened based on agreed criteria and stored on the server. Fluxes stored on the server can be accessed at any time. Still, annual flux calculation can be performed after a complete one-year-long dataset becomes available.

Fluxes of CO_2, CH_4 and N_2O will be calculated from the change in gas concentration in the chamber headspace over time, adjusted by the ground area enclosed by the collar, volume of chamber headspace, air density and molar mass of gas at the measured chamber. Flux monitoring at each site will be continued at least monthly for 24 months. The same sampling and flux calculation methods are applied both for reference and demo sites but also the same time period is used for sampling to guarantee comparability of data between the sites and countries.

As the final outcome, gaseous flux monitoring data will provide the soil net balance for CH_4 and N_2O fluxes over the monitoring period ('method-1'). For estimating soil net CO_2 flux at all monitoring sites, heterotrophic CO_2 fluxes estimated by the 'method-2' will be combined with relevant mass-based C-flux flows in above- and belowground litter for providing complete soil net CO_2 flux. In addition, soil net CO_2 balance in non-forested sites will be estimated from modelled net ecosystem CO_2 exchange based on in-situ collected data.

1.1.2 Tree stand biomass measurements

Carbon fluxes mediated by vegetation are estimated by measurements of plant biomass and production (Ojanen et al., 2013; Uri et al., 2017). Tree stand aboveground and belowground biomass (coarse root) estimation are based on measuring the tree stand diameter distribution (breast height diameter) of all trees on the sample plot, and further parameters (e.g., tree height and length of the live crown) for sample trees. Sample tree data forms a complementary set of variables for all trees. Biomass of different stand components (stems, branches, foliage, stump and coarse root systems) are estimated with allometric functions that use breast height diameter, either alone or together with the complementary variables, as explanatory variables (see Figure 2, Figure 3). Such functions are available for all our common forest tree species (e.g., Zianis et al., 2005; Liepiņš et al., 2017). Biomass production estimations are based on the annual diameter growth of measured sample trees.

The growth data will be used to construct diameter distributions and the complementary set of variables for the stand in consecutive years. The allometric functions will be fitted into these data sets, and the annual biomass production will be estimated as the difference between biomass values of consecutive years. Values will be transformed per square meter using a sample plot area.

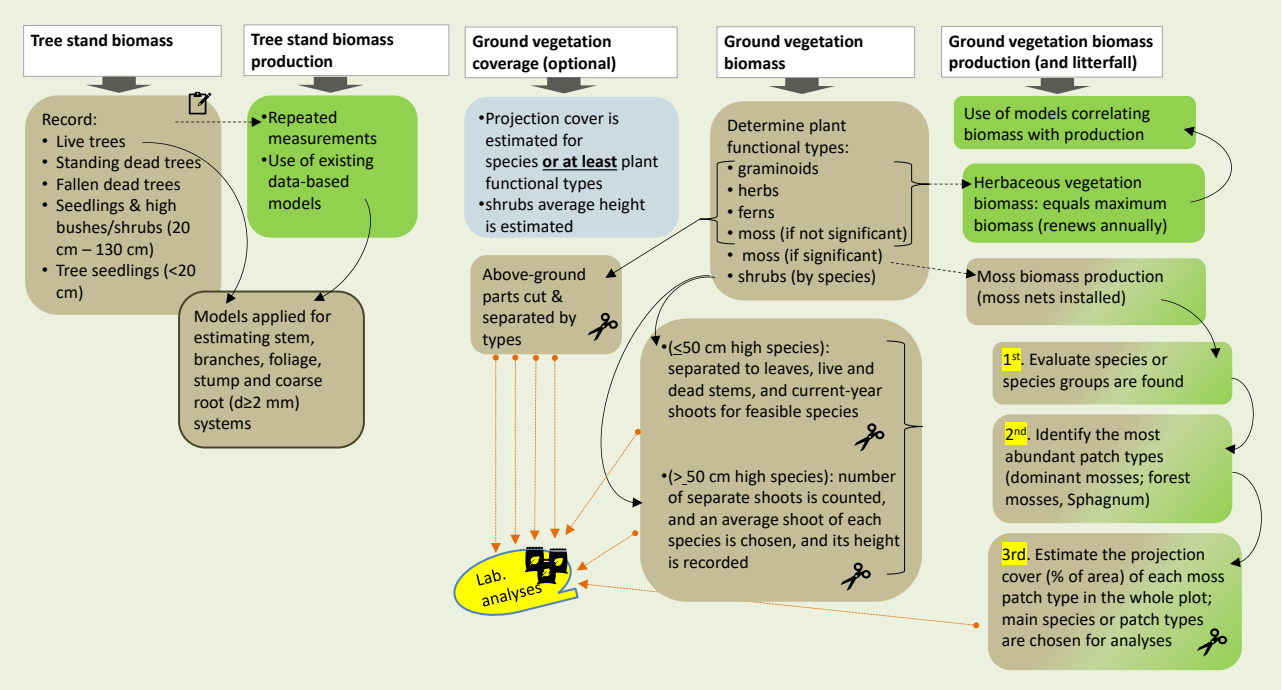

Figure 2. Outline of planned aboveground biomass and biomass production determination (uin tree, understory and ground layers in Life OrgBalt.

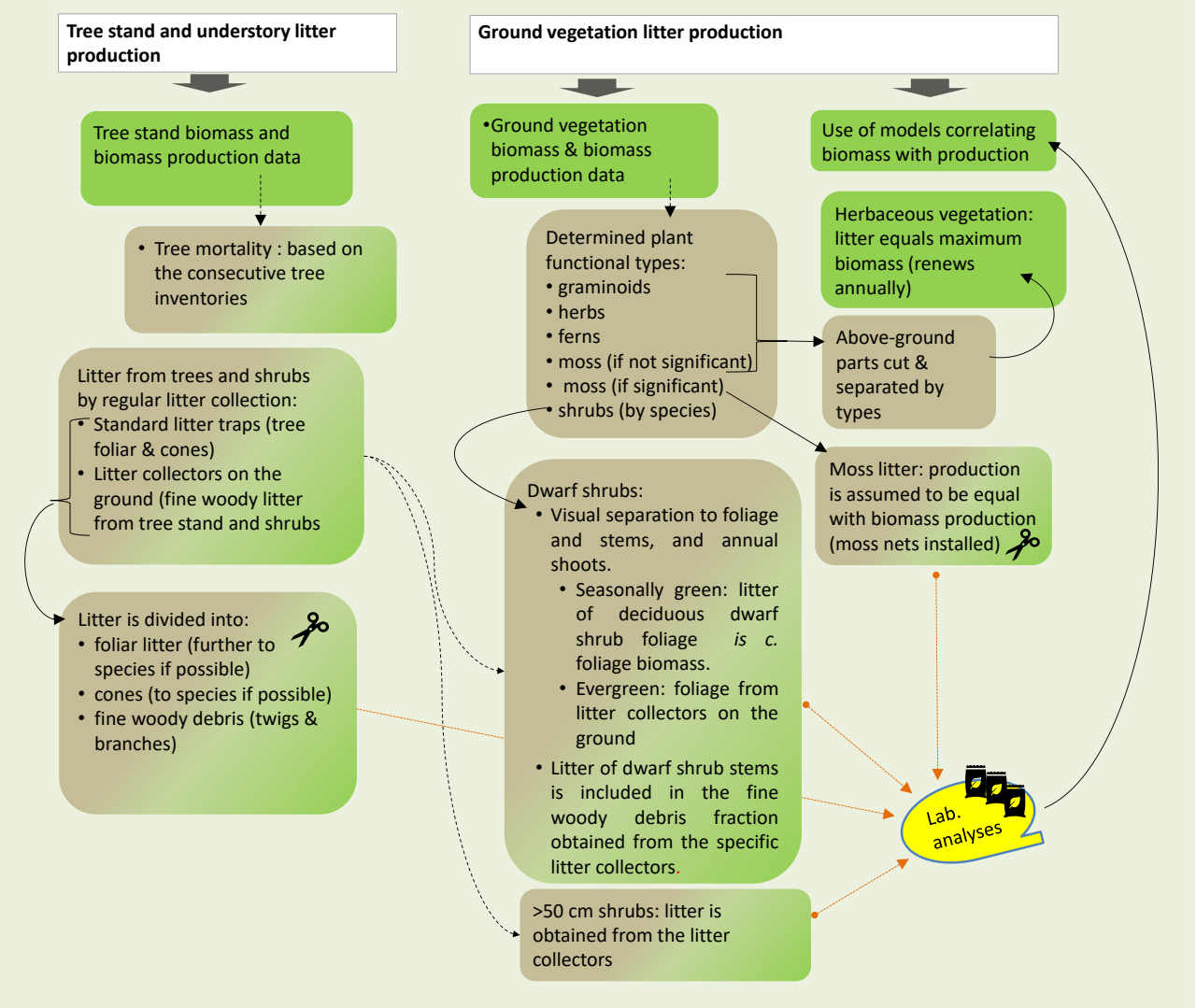

Figure 3. Outline of planned aboveground litter production in tree, understory and ground layers in Life OrgBalt.

1.1.3 Ground vegetation measurements

The aboveground biomass of the ground vegetation is measured by harvesting, drying and weighing the aboveground vegetation of small plots at the time of peak biomass in summer 2021 in Finland and 2022 in the Baltic states (see Figure 2). The samples are separated into plant functional types (shrubs, graminoids, forbs, and mosses, as applicable). For deciduous shrubs, the biomass is separated into leaves and stems. For all shrubs, current-year shoots are separated. Shrub stem radial growth will be estimated using literature data for plots with substantial shrub layer. Otherwise, deciduous leaves and current-year shoots will be considered as annual biomass production. For herbaceous plants, total biomass is regarded as annual aboveground production. Values are transformed per square meter using a sample plot area. Existing data on correlations between biomass and annual production rates in different species are applied where possible and further developed in forest sites to ease laborious harvesting, separation, and drying work.

Fine root biomass (<2mm) is estimated from volume-exact soil cores, analysed down to the rooting zone lower limit in 10-cm sections (see Figure 4). The end of live-root occurrence is confirmed from the samples. Roots are separated from soil by hand, washed free of soil, dried and weighted, and soil bulk density will be used to generalise root mass per sample volume to values per square meter.

Fine-root production is estimated by the ingrowth-core method modified for peat soils (Laiho et al., 2014; Bhuiyan et al., 2017), or the root mesh method (Uri et al., 2017) for annual plants. The amount of ingrown roots represents fine-root production over the 1-2 years-long incubation period, which will be generalised into annual production per square meter. Pilot studies suggest that two years of incubation time is needed for sites with perennial vegetation (Bhuiyan et al., 2017 and unpublished data). In the root mesh method, roots grown through the strips during the incubation period and thereafter measured for a known volume on both sides of the strip represent production. This simpler method is enough where branching and radial growth of existing root systems need not be considered. Fine-root turnover (litter input) is estimated as production per biomass. Roots in biomass and ingrowth core samples are separated into tree and ground vegetation roots to the extent possible; this task is labour intensive and requires expertise.

Ground vegetation coverage measurements and ground vegetation biomass sampling (biomass and biomass production samples) were made in Finland 2020, and biomass samples are in lab analyses. Ground vegetation coverage measurements and ground vegetation biomass sampling in the Baltic states are started in 2021 and 2022 by utilising on-site harvested samples, which are possibly supplemented modelling-based approaches to ease the large workload involved. As a part of ground vegetation biomass monitoring, moss nets were installed on forest sites with abundant moss coverage during autumn 2020. Sampling for fine root biomass determination will be made in 2022. Root ingrowth cores were set in forest sites in 2020.

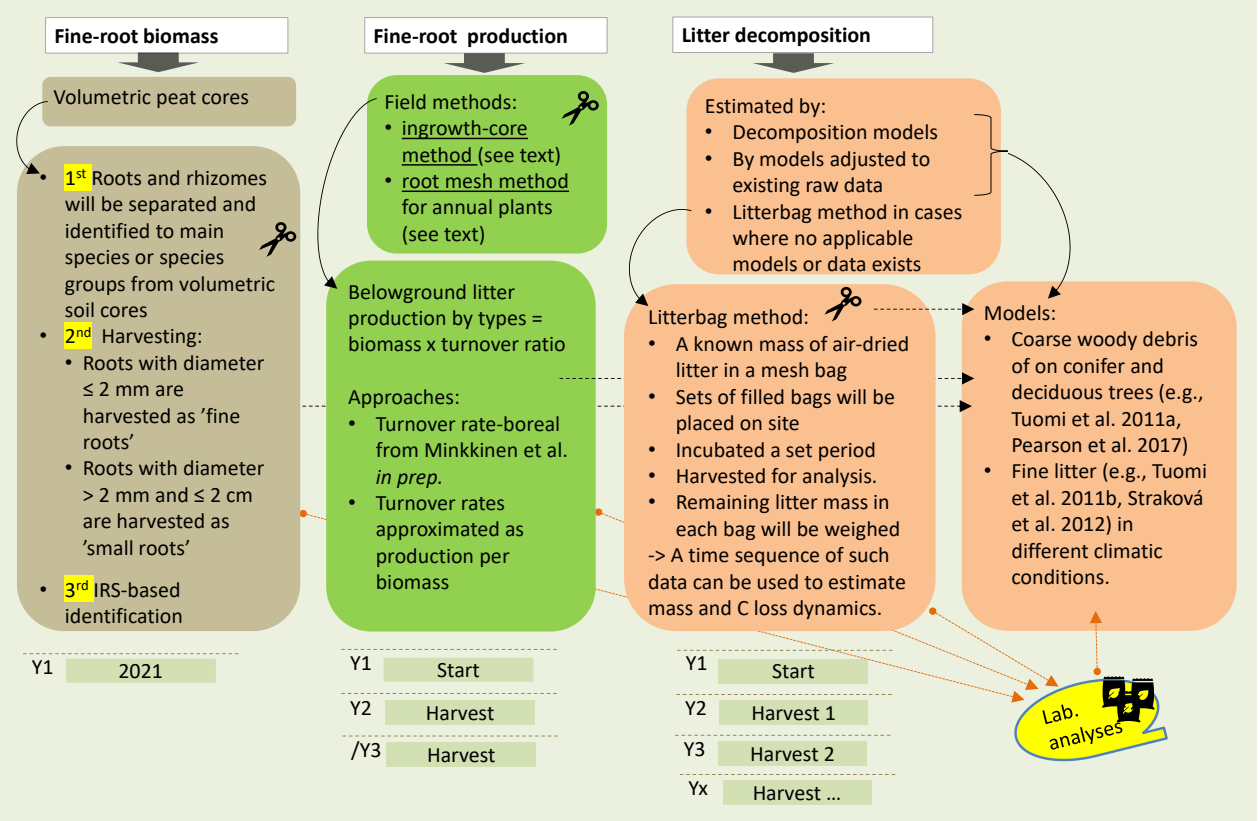

Figure 4. Outline of belowground fine-root biomass determination, biomass production determination, and belowground decomposition determination in LifeOrgBalt.

1.1.4 Carbon inputs with dead biomass and carbon loss rates

Estimates of current carbon stock in litter and deadwood are obtained by the area-based sampling in each site. For forested sites, annual tree mortality estimates will be based on monitoring data from other projects or tree mortality models (e.g., Jutras et al., 2003), where applicable. Estimates on the *amount of deadwood* will be made as the data from tree stand biomass becomes available.

Carbon input with the annual aboveground litter from perennial plants are based on a recurrent collection of litter from litter traps of known area (e.g., Ojanen et al., 2013; Uri et al., 2017), following the litter classification and analysis by methodology defined for ICP forest monitoring (see Figure 3). Litter traps will be set at the sites at the time of site establishment and the start of gaseous flux monitoring. For annual plants, the annual biomass production equals also the amount of litter input (i.e. annual plant litter estimates are based on ground vegetation biomass monitoring). Annual fine-root litter input rates are based on the production/biomass ratio as described in previous chapters.

Decomposition of *aboveground litter* C pools is estimated using decomposition models, separately for the coarse woody debris of conifer and deciduous trees (e.g., Pearson et al., 2017; Tuomi, Rasinmaki, et al., 2011; Tuomi et al. 2011a; Pearson et al., 2017), and fine litter (e.g., Strakova et al., 2012; Tuomi et al., 2011a) in different climatic conditions. The litterbag method (Strakova et al., 2012) is used for estimating litter decomposition rates in cases where no applicable models exist (see Figure 5).

Figure 5. Examples of decomposition experiment litterbags containing a known amount of tree twig litter with two diameter classes (left) and different litter types harvested from litter collector (right).

Typical litter types on the chosen experiment sites should be used to collect new litter decomposition data. The listed materials include deciduous leaves (alder or birch), needles (spruce or pine), dead shrubs (*Filipendula* sp. or *Rubus chamaemorus*), small twigs (diam. <5 mm), thicker twigs (diam. 10 mm < x < 20 mm), *Sphagnum* moss (if abundant on-site), forest mosses (if abundant on-site), unsorted (twig- free) litter from litter collectors. The suggested litter types for the study in the Baltic states are based on conditions at the suggested/selected sites (Table 2) for this experiment.

Table 2. Suggested forest sites for the decomposition study in the Baltic states based durin the planning process in September 2021

Country	Black alder	Birch	Pine	Spruce
Lithuania	LTC109	LTC108	-	LTC104
Estonia	EEC108	EEC106	EEC105	EEC104
Latvia	LVC109	LVC108	LVC110 (LVC107)[(1)]	LVC106
[(1)] Optional addition to include (old stand) for adding number of sites to 3 sites studied				

Litter traps collecting litterfall from trees and ground level were set at forest sites during autumn 2020 in all partner countries. The traps are emptied for deposited litter materials monthly during the warm season and after snow melts. Existing litter collections in Finland were upgraded according to LIFE OrgBalt standards. Previous year litter materials are currently in analysis or data is ready (drying and fractioning of litter types).

Litter decomposition study materials have been collected in selected sites starting from 2020. Litterbags were prepared and set to the chosen sites in spring 2021 in Finland, and in the Baltic states, similar litterbag sets were set at field sites in spring 2022. Harvesting of the litter bags will be after 1, 2, 3 (and 4) years after the experiment start. Pre-existing materials and data (from former applicable studies) are currently surveyed for possibilities to use in decomposition modelling.

1.1.5 Characterising soil microbial communities

We concentrate on the whole microbiota: fungi, archaea and bacteria in this work. This is because the main GHG in drained organic sites is CO_2. The microbiome is analysed by amplicon sequencing using ITS and 16S primers. We will concentrate on the forested organic sites LifeOrgBalt has to offer. 30 sites are included in the analysis, totalling 180 separate soil samples. Selected forest sites can be grouped to include differences in tree composition (deciduous, conifer, and mixed tree stands), tree stand age, and typical water table levels in soil (high and low water table sites). In each chosen site, soil sampling (performed in August /September 2021) was made at each of the three subplots with two treatments; trenched and un-trenched. Sampling made in un-trenched conditions (points established for N_2O, CH_4 and total CO_2 monitoring) includes soil environment with semi-decomposed organic soil, recent belowground litter, living roots, mycorrhizae, soil animals etc.. In contrast, living roots are excluded from the trenched conditions (points established for heterotrophic CO_2 monitoring). This chosen main strategy is adjusted according to the data on CO_2 emissions on these drained soils.

Soil samples were collected in August – September 2021, starting from the Northmost sites. Each participating country is responsible for the national sampling and sample shipment in frozen condition to laboratories in Finland. Soil samples (10 x 10 x 10 cm sample) are taken from only one depth below the litter layer at c. 15 cm depth in the soil profile.

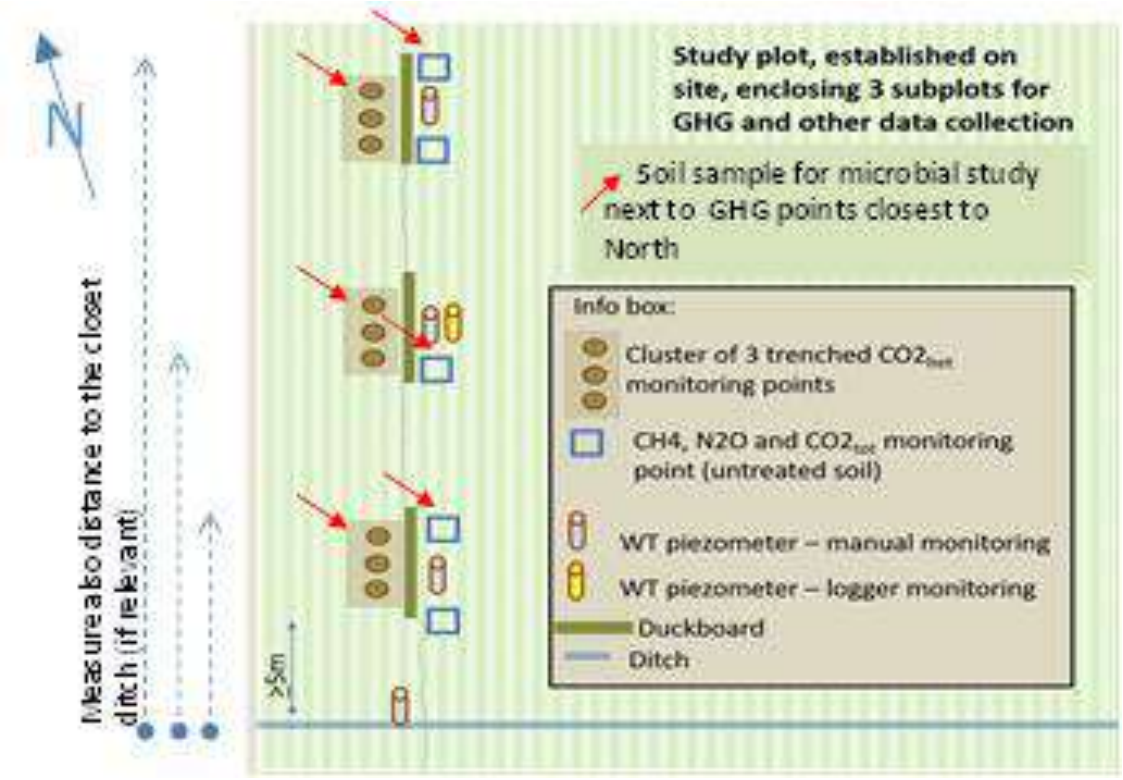

Figure 6. Soil sampling strategy in each included forest site.

The microbial community study is currently in the laboratory work phase. DNA from the soil samples was extracted in December 2021. As described for the whole procedure in Kosunen et al. (2020) the DNA was extracted from the samples using a NucleoSpin soil kit (Macherey Nagel, Germany). Nanodrop One (Thermo Scientific) is used to measure DNA concentrations. Currently DNA samples are on trays ready for shipment to sequencing. The bacterial and fungal community structure will be assessed with amplicon sequencing targeting the 16S and ITS regions, respectively. As a deviation from the proposal plan, the main focus will be on the microbial decomposer community involved in in the CO_2 emissions because (1) it is likely to be the main GHG gas species emitted from these primarily drained nutrient-rich organic soils, and (2) it would be challenging to sample soil profile depths to below the ground water level necessary for studying methanogens and methanotrophs.

ITS2 region for fungi and V4 region of 16S SSU rRNA for bacteria are amplified by polymerase chain reaction (PCR). The fragments are then sequenced with the MiSeq platform (Illumina) by utilising the MiSeq v3 kit. PipeCraft 1.0 pipeline software is used for quality filtering as well as the removal of artefacts, primer-dimers and primers from the raw 16S rRNA and ITS sequence reads. After assembling paired-end reads and a two-step quality filtering, an OTU table is created from the sequence reads. OTUs are then annotated taxonomically using BLAST and a reference ITS2 database (sh_genral_release_dynamic_01.12.2018.fasta) from UNITE and a 16S rRNA database (SILVA_123_SSURef_Nr99_tax_silva.fasta) from SILVA to find representative fungal and bacterial sequences, respectively. After quality filtering, functional information of fungal guilds of OTUs is derived from FUNGuild. Sequence annotation is planned to take place by Luke in 2022.

1.1.6 Soil screening with infrared spectroscopy (IRS, FTIR)

Information on soil nutrient concentrations and other soil properties, e.g., soil organic matter characteristics, are needed for many purposes. The rates of many soil processes and, consequently, soil greenhouse gas emissions depend at least to some extent on the nutrient regime of the site (IPCC 2014). Infrared spectroscopy (IRS) is a rapid, cost-effective and relatively easy-to-use technique that has long been used for the characterisation of different sample materials, including the determination of several chemical and biological characteristics of soils (e.g., Holmgren and Nordén, 1988; Confalonieri et al. 2001; Terhoeven-Urselmans et al., 2008; Cécillon et al. 2009; Bellon-Maurel and McBratney 2011; Krumins et al., 2012; Hayes et al., 2015; Straková and Laiho, 2016). IRS has long been applied in characterising samples with complex chemical compositions, including peat. Infrared radiation is the region of electromagnetic radiation where wavelengths range from ca. 780 nm to ca. 1 mm. Infrared waves are thus longer than those of visible light. Infrared spectroscopy is based on each chemical bond absorbing infrared radiation in a specific manner that depends on the nature of the bond. Thus, an infrared absorbance spectrum, showing for each wavelength or wavenumber the proportion of radiation absorbed by the sample, shows the relative abundance of different chemical bonds in the sample, that is, a summary of the chemical composition of the sample (e.g., Coates, 2000). The power of IRS is based on each chemical bond present in a sample absorbing IR radiation in a specific manner that depends on the nature of the bond. Thus, an IR absorbance spectrum, showing for each wavelength or wavenumber the proportion of radiation absorbed by the sample, shows the relative abundance of different chemical bonds in the sample. IR spectra thus summarise the whole chemical composition of the sample. The spectra can either be used for direct interpretation of the absorbance intensities at different wavelengths or be reduced into a smaller number of variables that contain summarised information on the systematic variation in the spectra by, e.g., Principal Component Analysis (PCA) or other multivariate methods (Adamczyk et al., 2016). Such summary variables may then be used as predictive variables (e.g., Vávrová et al., 2008), in our case, for GHG emissions. These approaches can be combined by first seeking the characteristics of the spectra that have the best predictive power and then interpreting them (Adamczyk et al., 2016).

The LIFE OrgBalt project tested IRS as such solution for peat and soil samples collected in cool temperate moist climate zone in forest land, cropland and grassland. In parallel, peat samples collected previously in the LIFE REstore project (from 42 GHG measurement and demo sites) were used to cover the full spectrum of peat properties – from nutrient-poor Sphagnum peat to fertile peat of mesotrophic bogs. Soil samples from peatlands with various land uses and samples from naturally wet and drained forest stands with different forest site type classification were selected. Sample set dominated by organic soils with some exceptions of mineral soil from deeper soil layers. The project ensures comparability with the GLOSOLAN network by utilising the GLOSOLAN specifications-based equipment and procedures. The spectroscopic analyses on the LIFE REstore project samples were run in Silava laboratories in 2021. This study aimed to start building a spectral library for organic soils (including peat) and create initial calibration models to evaluate the method's potential to predict pH value and C, N, P, K, Ca, Mg and humic acid concentration in peat samples.

This study aimed to start building a spectral library for organic soils (including peat) and to create initial calibration models to evaluate the method's potential to predict pH value and C, N, P, K, Ca and Mg concentration in peat samples. The results are reported in LIFE OrgBalt Mid Term reporting (Annex C2_03 Harmonized methodology for characterising peat properties using infrared screening method). In the scope of this study, the residual prediction deviation value (RPD) was considerably lower than the 2 signals - a possible difficulty in applying the current methodological approach for quantitative analyte prediction in unknown samples. The highest potential of prediction performance was observed for pH, Ca, and Mg, but the lowest perspective for P and K. C, N and humic acid as well as other parameter prediction performance, may be improved by primary increasing count and variety of calibration samples (spectra) and secondary by increasing count of measurement replicates for the same sample to discard replicates that increases relative standard deviation of prediction replicates above the threshold, e.g. 10 %. It was observed that mostly the highest performance of analyte prediction in peat samples was for prediction models elaborated by the peat soil calibration data set only; the addition of forest soil sample spectra to the calibration data set did not improve model performance.

Nevertheless, also for such calibration data sets with peat soils only, PCA often indicated significant spectral differences that could have added uncertainty to values predicted by the model. In the scope of the study separation of spectra by PCA did not improve model quality as model robustness may have decreased to the insufficient number of spectra. The higher number of spectra would allow for making separate calibration models by focusing more on PCA results. Afterwards, these models could be applied to unknown samples by the guidance of values of spectral residues and Mahalanobis distance to match appropriate models and unknown spectra. Another potential solution for improving model prediction capabilities may be improving sample preparation procedures, e.g., ensuring more homogenous samples.

The first part of the activity (building the FTIR library) was implemented in 2021 and is reported in LIFE OrgBalt Midterm reporting. The second part of FTIR analyses, based on LIFE OrgBalt soil samples, will be conducted in 2022. Analytical comparisons are planned to include IRS data comparison with GHG fluxes, as well as with soil properties - pH, N, P, K, Ca, Mg, C and ash content in parallel to the implementation of conventional methods. Results will be published in a peer-reviewed scientific article.

1.1.7 Soil and water analyses

A comprehensive evaluation of soil properties down to 100 cm depth will be done in all gas fluxes measurement plots while establishing the reference and demonstration sites. Soil properties are implemented once during the project implementation, in 2021-2022. Soil sampling and analyses will be performed according to ICP Forest guidelines (Cools and de Vos, 2010; Konig et al., 2010), methodology providing comparable results. Sampling will be done in 3 repetitions in every reference and demo site or using a method providing comparable results. A good procedure is sampling at north and south from gas measurement sites, as close as possible to gas sampling & measurement sites. Sampling sites will be located in a flat area representing average conditions in a reference or demo site. 100 cm3 undisturbed soil samples will be collected at 0-10, 10-20, 20-30, 30-40, 40-50 cm depth

and disturbed samples at 50-75 and 75-100 cm depth. After collection, samples are transferred to plastic bags with labels containing information on the project, sampling plot, repetition, depth and date.

Additionally, litter samples (10 x 10 cm to the whole depth) are collected nearby soil sampling sites in forest land. Small pits can be dug to collect samples if sampling with an auger is impossible. Litter samples in the field or in the laboratory should be cleaned from plants' green (living) parts.

Soil and litter samples will be collected in the spring and summer of 2021 or 2022. However, the sampling period is not critical as far as the total content of elements is determined.

After collection, samples are transported to LSFRI Silava laboratory of Forest environment and air-dried. Then all samples will be dried at 105ºC degrees, weighted to determine bulk density, milled and screened through a 1 mm sieve, and samples for elemental analyses will be milled and sieved through a 0.25 mm sieve. After the preparation of samples following parameters will be determined: bulk density, pH, N, P, K, Ca, Mg, C and ash content. Parameters which will be determined in soil and reference methods are provided in Table 2.

Table 3. Parameters and reference methods of soil analyses

No.	Parameter	Reference method	Application1
1.	Sample pre-treatment	ISO 11464	IR
2.	Soil Moisture Content	ISO 11465	IR
3.	Bulk Density	ISO 11272 (adopted to organic material)	I
4.	pH	ISO 10390	IR
5.	Organic Carbon (C)	ISO 10694	I
6.	Total nitrogen (N)	ISO 13878	IR
7.	Aqua regia extractable phosphorus (P), potassium (K), calcium (Ca) and magnesium (Mg)	ISO 11466	IR2
8.	Ash content	ISO 1171	I

Water samples (0.5 L per piezometer per time) will be collected from piezometers during every site visit (monthly base on average), simultaneously with gas sampling. Sampling is done from one of the piezometers. The other should be used for continuous water level measurement, and additional 2 piezometers should be used for manual water level measurement during site visits if the sample plot is split into subplots. Water samples after collection are transported in a cold camera and stored in a freezer at a low temperature (4ºC). Once per month, all samples are transported to Latvia for analysis. This can be done simultaneously with the transportation of gas bottles for gas analyses. At LSFRI Silava following parameters will be determined in water N total, NO_3^-, P, K, Ca, Mg, DOC). Additional parameters, e.g., Hg may be considered in case of additional funding to determine the linkage between environmental conditions and Hg outputs into water. Parameters which will be determined in water samples and reference methods are provided in Table 3.

Table 4. Parameters and reference methods of water analyses

No.	Parameter	Reference method
1.	Sample pre-treatment	ISO 10523, ISO 7888
2.	pH	ISO 10523
3.	Electrical conductivity	BS EN 27888
5.	Total N, NO_3^-, TOC	ISO 10304-1, ISO 12260, BS EN 1484
6.	Dissolved K, Ca and Mg	ISO 7980, ISO 9964-3
7.	Total P	ISO 6878

The results of the analyses will be used to determine possible correlations and covariations with GHG fluxes, particularly, after the proposed actions are implemented in the project demo sites. Water properties will be used as additional parameters to increase the elaborate GHG emission models' accuracy and improve the ability to predict GHG fluxes under different management scenarios and land uses.

1.2 Modelling

The SUSI peatland simulator is aimed for application in boreal and tropical climate zone to calculate growth response on the drainage of organic soils, including estimation of soil carbon losses. SUSI peatland simulator is based on the assumption that forest growth is limited by the accessibility of nutrients, which are released during the decomposition of organic matter. The increased groundwater level is slowing down the decomposition of organic matter and the availability of nutrients, reducing the growth of trees and carbon losses. Susi peatland simulator is aimed at the parametrisation of these variables. The main modelling aim is to upgrade the SUSI peatland simulation for use in projecting CC scenarios and make the software useable within the LIFE OrgBalt region. Furthermore, SUSI will be delivered as open-source software to be readily and widely adaptable for drained organic soil research and land use studies.

The SUSI peatland simulator is developed, but its improvement is an ongoing process. The effort placed on this task has been increased by the addition of postdoctoral researcher Jani Anttila to the project. Considerable effort is now made to improve the accessibility of the simulator. This includes writing documentation, and user instructions, improving the readability of model output, creating well-explained example use cases, as well as improving the actual user interface to the simulation code via Jupyter notebooks and from the command line. The model has also been made publicly available on Github at https://github.com/annamarilauren/susi so that researchers and developers can access the source code and suggest improvements directly to the maintainers.

The current challenge in applying the SUSI model in Baltic countries is generating the appropriate input data. These data need to contain specifics, such as tree biomass partitioning into branches, leaves, roots, etc., which need to be estimated with statistical models appropriate to the site and tree

species. More effort and cooperation are currently directed toward achieving this task of creating suitable inputs for the model.

1.3 Post-2023 impact assessment

A replicability and transferability strategy has been published under action A2 to multiply the impact of the Project results during its implementation and to replicate and transfer its findings after its end, in order to reach a wider audience and implement its results in further sites and regions, other than the Project demo sites.

A key role in this respect is represented by the elaboration of a Simulation model (SM) under action C5. The simulation model will serve as a policy planning/decision support tool for the development of GHG emissions projections at a national level and the analysis of the socio-economic impact for 2 scenarios – with and without implementation of CCM measures - with dynamic background information on changes of technical conditions of drainage systems. The elaboration of these models will be possible on the bases of the results of Activities C1 and C2, namely the elaboration of a catalogue of climate change mitigation measures, including a socio-economic impact assessment, the improvement of GHG emission factors and of the methodologies for GHG inventory reporting together with the related national reports, and finally the elaboration of mathematical equations and tools for GHG projections from organic soils. The simulation model will be proposed as an evaluation tool to determine the extent to which measures should be implemented in each evaluated country. This will support the development and the evaluation of climate change mitigation measures related projects in the context of the Common Agricultural Policy. The simulation model's main targets are policy and decision-makers, consultants, non-governmental organisations of farmers and foresters, and individual stakeholders (major foresters and farmers). The model will include data on organic soils at a national level and the potential for land-use change according to the 17 climate reduction scenarios identified in the project. Data on organic soils and their use in each evaluated country will be integrated. Feedback from the involved stakeholders will be collected during the dissemination, training and networking activities planned under actions E.2 and E.3, i.e., National workshops, Thematic Workgroup meetings, Networking workshops on the national level and Experience exchange visits. Feedback will be gathered to improve the developed models as well as to evaluate the results obtained through them in terms of GHG emissions reductions and the socio-economic impacts under different management scenarios. In addition, the project envisages a total of 10 training seminars -2 for each country - which are planned to be organised at two levels - one for consultants and the other for individual stakeholders, i.e., landowners and managers. During training workshops, the simulation tool will be presented to give a national perspective of the implemented climate change mitigation measures.

2 Overview of the implementation of field activities

In the LIFE OrgBalt project, the fieldwork has been carried out in all demonstration and reference sites, 53 sites in Latvia, Lithuania, Estonia, and Finland.

2.1 Greenhouse gas flux monitoring

The first regular sampling round was performed in Estonia and Latvia at the end of the year 2020. In Lithuania, regular sampling was started in spring 2021. Sampling was started in Finland in spring 2020.

GHG fluxes measurements and sampling have been done by using three different methods:

1) Static dark chamber method (see sub-chapter 1.1.1. 'Method -1') to measure N_2O, CH_4 and heterotrophic respiration (CO_2) during the wintertime.
2) Heterotrophic respiration – soil CO_2 emissions monitoring during the warmer period (see sub-chapter 1.1.1., 'Method-2'),
3) NEE - transparent chamber (in grasslands) during the vegetation period.

On regular bases, data quality check and flux calculations have been done:

- Raw data quality check of the measurement data with static dark chamber and the flux calculations of N_2O and CH_4
- Raw data quality check and heterotrophic respiration flux calculations (by using flux calculation platform created by Luke)
- Raw data quality check of NEE.

GHG measurements overview in the project partner countries:

- **Finland:** measurements are carried out every third week during the warm season. In wintertime, monitoring is approximately every five/six weeks because of typically low temperatures (challenges to have analyser in function) and snow conditions (challenges to have site access). Monitoring was started in May 2020, the first-year flux data set has been checked and annual flux estimates, including temperature adjustments to CO_2 data, are planned to be formed in April 2022. Flux data collection at the demo sites continues until the end of April 2022.
- **Estonia**: all the measurements are carried out twice per month (see Table 56). During each measurement campaign, the gas samples were collected in 6 replicates (6 chambers) and from each chamber during the 1 h-long deployment time, 4 samples were collected. In total, 5760 gas samples were collected and analysed.

Table 56. Review of measurements in Estonia

2021	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Total
EEC101	X	X	X	X	X	X	X	X	X	X	X	X	24
EEC102	X	X	X	X	X	X	X	X	X	X	X	X	24

2021	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Total
EEC103	X	X	X	X	X	X	X	X	X	X	X	X	24
EEC104	X	X	X	X	X	X	X	X	X	X	X	X	24
EEC105	X	X	X	X	X	X	X	X	X	X	X	X	24
EEC106	X	X	X	X	X	X	X	X	X	X	X	X	24
EEC107	X	X	X	X	X	X	X	X	X	X	X	X	24
EEC108	X	X	X	X	X	X	X	X	X	X	X	X	24
EEC109	X	X	X	X	X	X	X	X	X	X	X	X	24
EEC110	X	X	X	X	X	X	X	X	X	X	X	X	24

- **Latvia**: measurements are carried out monthly (see Table 7). During each measurement campaign, the gas samples were collected in 5 replicates (5 chambers) and from each chamber during the deployment time, 4 samples were collected. In total, 6740 gas samples were collected and analysed.

Table 7. Review of measurements in Latvia

2021	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Total
LVC101	X	X	X	X	X	X	X		X	X	X	X	11
LVC102	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC103	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC104	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC105	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC106	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC107	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC108	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC109	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC110	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC111	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC112	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC113	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC114	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC115	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC116	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC301	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC302	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC303	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC304	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC305	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC306						X	X	X	X	X	X	X	7
LVC307	X	X	X	X	X	X	X	X	X	X	X	X	12

2021	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Total
LVC308	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC309	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC310						X	X	X	X	X	X	X	7
LVC311	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC312	X	X	X	X	X	X	X	X	X	X	X	X	12
LVC313	X	X	X	X	X	X	X	X	X	X	X	X	12

- **Lithuania**: measurements are carried out monthly from October 2021(see Table 8). During each measurement campaign, the gas samples were collected in 5 replicates (5 chambers) and from each chamber during the 1 hour long deployment time, 4 samples were collected. In total, 1080 gas samples were collected and analysed.

Table 8. Review of measurements in Lithuania

2021	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Total
LTC101			X			X	X				X	X	5
LTC102			X								X	X	3
LTC103			X	X		X	X		X		X	X	7
LTC104			X		X		X		X		X	X	6
LTC105			X		X			X		X	X	X	6
LTC106			X	X	X		X			X	X	X	7
LTC107			X			X		X		X	X	X	6
LTC108			X			X	X				X	X	5
LTC109						X	X				X	X	4
LTC110			X			X				X	X	X	5

Soil heterotrophic respiration measurements ('Method-2') were done at the same frequency as the static dark chamber measurements ('Method-1'): in Estonia twice per month, in Latvia and Lithuania once per month using the portable gas analysers (EGM-4 and EGM-5). Collected data were uploaded to the flux calculation platform created by Luke, and the first initial data quality check and the flux calculations were done, data analyses are in progress. The additional plan for the year 2022 is to finalise the heterotrophic respiration flux calculation platform.

NEE measurements with transparent chambers are carried out on the grasslands at the same frequency as other GHG flux measurements in Estonia and Latvia from April to November 2021. The flux calculations are not done yet, and are planned to do in spring 2022. There is also a plan to create a platform/system for the NEE flux calculations (transparent chamber method), examine an option the integration with heterotrophic respiration data calculation platform, or study the possibilities to automate annual flux calculation. However, these novel automation developments are considered LIFE OrgBalt inspired spin-offs that need additional funding support as they were not part of the original project plan.

2.2 Biomass-related measurements quantifying annual production

Tree biomass measurements in Finland were made in 2016 at all sites, repeated during LIFE OrgBalt 2020 at FIC301 and FIC302 sites, and are planned to be made in spring 2022 at FIC303. In Latvia, the measurements were done in 2021 on all forest sites, and data calculation and analyses are in progress. In Estonia and Lithuania, the tree stands aboveground and belowground (coarse root) biomass estimations are not done yet and are planned for July 2022.

Aboveground biomass sampling on the grassland and cropland has been carried out in all Baltic states at the time of maximum vegetation growth – at the end of July or the beginning of August 2021. Samples have been dried, weighted and chemical parameters analyses (C and N content) are in progress. Belowground samples from the grassland were collected in April 2021 and the beginning of August 2021. All the root samples have been washed out, sorted, dried, weighted, and data in files. Chemical analyses are planned for spring 2022.

Moss biomass and moss biomass production in forest sites. In Finland, in 2020, installed moss nets were harvested in 2021, materials are in analysis for site FIC303, and the data is ready for sites FIC301 and FIC302.

In Finland, ground vegetation cover and biomass collection were made in 2020 and 2021. Materials for sites FIC301 and FIC302 are analysed, and data in files; site FIC303 material analyses continue in 2022. Collected biomass data has been used in the testing possibility to model shrubs biomass based on simplified shrubs data – the preliminary results from this method development are promising and will be further studied in 2022. In Latvia, the measurements were done in the summer of 2021 on all sites, and data calculation and analyses are in progress. In Estonia and Lithuania, estimations of annual biomass production and litter inputs from ground vegetation have been planned for July/August 2022.

Fine root biomass samples (soil cores) were collected from the Finland sites in 2020, the materials were analysed in 2021, and the data is now in files. In Latvia, the samples were collected, washed out, sorted, weighted and analysed in 2021. Fine root biomass sampling has not yet started in Lithuania and Estonia and is planned for autumn 2022.

In Finland, fine root production measurements started by root sock incubation at 2 sites in 2020. samples from previously stated incubation at one site were harvested in autumn 2020, analysed in 2021 and are now in data files. Fine root production measurements are to be started in the Baltic states in 2022

2.3 Carbon inputs with dead biomass and carbon loss rates

Litter traps were set at the forest sites at the start of gaseous flux monitoring and litter material collection was followed simultaneously with the flux monitoring. All litter materials collected so far are separated infractions, dried, weighted and data is in files.

Decomposition studies. Litter decomposition study materials in Finland were collected in selected sites in 2020. Litterbags were prepared and set to the chosen sites in spring 2021, and the first bags will be harvested in spring 2022. Pre-existing materials and data (from former applicable studies) are currently surveyed for possibilities to use in decomposition modelling.

The decomposition experiment is also planned in Estonia, Latvia and Lithuania but will be performed in the spring and autumn of 2022.

2.4 Characterising soil microbial communities

The microbial community study is currently in the laboratory work phase. Soil samples were collected in August & September 2021, and DNA from the soil samples was extracted in December/January 2021. Currently DNA samples are on trays waiting for shipment to sequencing. Sequence annotation is planned to take place by Luke in 2022.

2.5 Soil screening with infrared spectroscopy (IRS, FTIR)

The first part of the activity (building the FTIR library) was implemented in 2021 and is reported in LIFE OrgBalt Midterm reporting 2021. The second part of FTIR analyses, based on LIFE OrgBalt soil samples, will be conducted in 2022. Analytical comparisons are planned to include IRS data comparison with GHG fluxes, as well as with soil properties - pH, N, P, K, Ca, Mg, C and ash content in parallel to the implementation of conventional methods.

2.6 Soil and water analyses

Physio-chemical analyses of soil and water samples were done in ISO 17025 accredited Laboratory of Forest Environment of Latvian State Forest Research Institute "Silava".

Soil samples collected in 2020 and 2021 were shipped to the laboratory of "Silava" for further processing. 640 soil samples from 38 sapling plots and various soil depths (0-10; 10-20; 20-30; 30-40; 40-50; 50-75, and 75-100) have been collected in Latvia, while 116 soil samples have been collected in Finland. Sample processing started in 2021 and the parameters like bulk density, total carbon, total nitrogen pH and ash content have been determined; analysis of HNO_3 extractable parameters K, Ca, Mg, P will continue in 2022. FTIR analyses on the samples are planned for 2022. According to the LVS ISO 11464 (2005) standard, the soil samples were prepared for analysis. Chemical parameters were determined to organic soil milled till fine powder and fine earth fraction ($D < 2$ mm) of mineral soil (prepared according to LVS ISO 11277) were analysed according to standard methods (Table 9). Organic carbon concentration (g kg^{-1}) in soil was calculated as the difference between total carbon concentration and inorganic carbon (carbonate) concentration. Analysis of ash content was used to calculate the content of organic matter.

Table 9. Soil sample analysis methods

Parameter	Unit	Method principle	Standard method
Bulk density	kg m^{-3}	Gravimetry	LVS ISO 11272:2017
pH_{CaCl2}	log unit	Potentiometry	LVS ISO 10390:2021
Total carbon	g kg^{-1}	Elementary analysis (dry combustion)	LVS ISO 10694:2006
Total nitrogen	g kg^{-1}	Elementary analysis (dry combustion)	LVS ISO 13878:1998
$CaCO_3$	g kg^{-1}	Volumetry	ISO 10693
HNO_3 extractable K, Ca, Mg and P	g kg^{-1}	ICP-OES	LVS EN ISO 11885:2009
Ash content	g kg^{-1}	Combustion	LVS CEN/TS 14775:2004

Water samples have been collected once per month simultaneously with gas flux measurements. Samples from Estonia and Lithuania have been shipped to "Silava" for the analyses. All the samples are analysed by standard methods (Table 10). In total 332; 43 and 102 water samples have been collected in Latvia, Lithuania and Estonia accordingly, and all parameters have been analysed.

Table 10. Water sample analysis methods

Water samples			
pH	log unit	Potentiometry	LVS ISO 10523:2012
Conductivity (EC)	µS cm^{-1}	Conductometry	LVS EN 27888:1993
Total nitrogen (N)	mg L^{-1}	Catalytic oxidation	LVS EN 12260:2004
NO_3 and PO_4	mg L^{-1}	Ion chromatography	ISO 10304-1:2007
NH_4	mg L^{-1}	Photometry	LVS ISO 7150-1:1984
DOC	mg L^{-1}	Catalytical combustion	LVS EN 1484:2000

Water analyses for the sites in Finland are made in Luke because there was no budgeting available for water analytical tools at the field (YSI meter), and regular transport of water samples to Silava was considered a risk for the water quality due to long distance. Samples from the Finnish sites have been collected regularly at times of gaseous flux measurements since spring 2021, and water analyses have been made locally in Luke Laboratories. Data on the collected water samples are ready.

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