LIFE 15/CCA/ES/125

Additional report Name: Above ground, below ground and carbon stock monitoring protocol

Action D1. Monitoring and project performance indicators

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INTRODUCTION

This document specifies all the monitoring parameters that must be evaluated during spring 2019 on the field trials. Methodologies are specified for above ground parameters (vegetation, fauna and Cocoon degradation), below ground parameters (soil and root system) and carbon storage on planted seedlings.

1. ABOVE GROUND MEASURES

Above ground vegetation monitoring intends to achieve three objectives at different scales:

- Test Cocoon effectiveness: determine survival rates, assess vigor, assess growth of planted seedlings, assess the effect on adjacent vegetation
- Test the adaptation of the selected species (adaptive restoration)
- Evaluate the restoration of the demonstration areas

In order to achieve the objectives of the above ground plant monitoring we propose to measure five groups of parameters:

- Vigor (survival and health)
- Growth rate
- Vegetation structure and composition

Additionally, fauna recruitment will be evaluated qualitatively. In order to evaluate Cocoon degradation it is necessary to assess the Cocoon device estate jointly with vigor and growth measures.

1.1 Seedling vigor

Tree/shrub vigor is a health indicator, also giving insights on survival rates. Vigor is assessed by the following semi-quantitative scores during their normal growing period:

3: Healthy seedling, with more than 75% of green, not wilted leaves. Also active growing points (apices) may be visible

2: Affected seedling, with 25-75% of the leaves being wilted, yellow or brown

1: Severely affected seedling with less than 25% of the leaves being green (i.e. the majority wilted, yellow of brown)

0: Presumably dead seedling with no or only wilted leaves. Seedlings, however, may still recover by resprouting after a rain event

R: Resprouted seedling

Cocoon vigor will be assessed at the seedlings monitored during 2017 and 2018 campaigns.

Figure 1. Seedlings of olive tree in different health stages, according to the proposed scores: healthy tree (left), affected tree (center) and resprouted tree (right).

Rodents grazing on saplings may cause negative growth. Moreover, plague infestation can affect seedlings development. Fast growth, wind or deficient root system development can cause seedling fall. All these situations must be recorded.

Figure 2. Other observations related to seedling health/growth, from left to right: grazing, broken stem, plague infestation, tree fall.

1.2 Seedling growth

Seedling growth will be assessed by measuring maximum plant height, from root crown to the shoot apex, using a measuring tape/stick. Be careful not to measure from Cocoon's lid. In order to also account for tree volume (multiple branching), stem diameter will be measured at the tree base (at the level of the Cocoon's lid, at 10 cm of the soil), using a caliper. Plant selection for growth measurements will be the same as vigor.

Figure 3. Taking height and diameter measures.

1.3 Vegetation structure

The growth of spontaneous vegetation is an indicator of the restoration of the site. The evaluation of the structure and composition of this vegetation will allow determining the degree of restoration of the site. Moreover, this measure will allow estimating aboveground biomass carbon stock in forests or rangelands. Vegetation structure and composition will not be evaluated in crops.

Structure will be evaluated quantifying cover types in each site or sub-site (differencing site specific variables: flat vs slopes, soil types, etc.). It is necessary to distinguish between herbaceous cover, woody species cover (shrubs and trees) and other soil cover types (litter, dry branches, plant remains), using the point transect method: a transect will be established extending a measuring tape of 25 m, and the evaluator will note the main cover type (herbaceous, woody, other) and the height of the dominant species every 20 cm. Woody species must be identified and height measured.

A minimum of 3 transects per hectare must be done in order to have representative measures. In slopes, transects must be done parallel and perpendicular to the slopes. Transects will be the same than 2017 campaign.

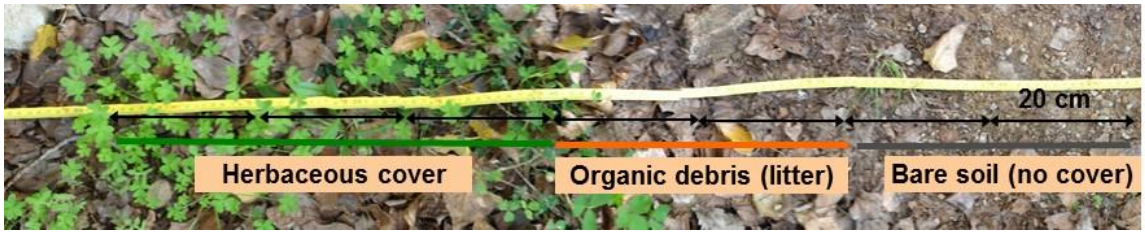


Figure 4. Different soil covers (herbaceous, litter, bare soil) in an evaluation transect.

1.4 Vegetation composition (biodiversity)

In order to determine vegetation composition the evaluator must identify all plant species (herbaceous, shrubs and trees, including planted ones) in the demonstration area (vegetation inventory) differencing each site or sub-site (according site specific variability: flat vs slopes, soil types, etc.). Monitor could take advantage of transect for doing this but probably he will need additional work in order to detect all the plants. For each plant, abundance estimation must be done, using this pattern ranks:

1: testimonial

2: low frequency (<25% soil cover)

3: high frequency (25%-75% soil cover)

4: dominant (>75% soil cover)

Vegetation composition should be evaluated at the same sub-sites than 2017 campaign.

1.5 Microsite evaluation

In order to detect the Cocoon effect on surrounding vegetation development we propose to perform a vegetation evaluation at microsite scale in a selection of seedlings. Cocoon effect on herbaceous vegetation cover and biomass will be evaluated tracing a 1m diameter circle around the seedling (using a plastic or metal circle, see Figure 5). For cover evaluation, it is recommendable to take an orthogonal picture covering all the area inside the circle and calculate the cover in the office (using a specific program or a grid panel). Another option is to measure herbaceous cover directly on the field using two perpendiculars 1m transects inside the circle. However this measure will be surely less accurate.

Figure 5. Example of an orthogonal picture of a sample circle of 1m diameter.

For determining plant biomass all the vegetation inside the circle (except the planted seedling, of course!!) will be harvested and weight (wet weight: weight at field; dry weight: weight after drying at 60°C for 4 days).

Microsite measures will be done for the two main species in each site. Measures will be taken in the same Cocoons selected for soils sampling and their associated controls.

1.6 Fauna evaluation

This parameter assesses the entry of animals to the restored area by counting footprints evidences and direct observations. The entry and establishment of macrofauna in restored areas, if do not causes damage to vegetation or soil, is an important step in ecosystem restoration. Fauna recruitment and entrance (use) will be measured through monitoring qualitative indicators during the field samplings: tracks (footprints), excrements, lairs, nests, direct observations. If possible, identify the species.

Figure 6. The presence of tracks, excrements or lairs are good indicators of fauna recruitment in restored areas.

1.7 Cocoon degradation

In order to qualitatively evaluate the Cocoon degradation/incorporation at soil, a scoring of the Cocoon devise will be done according to those categories:

1: Cocoon ok: with or without shelter, but with lid

2: Lid collapsed but bowl apparently in good state (without cracks, holes)

3: Bowl with signs of degradation (cracks, holes)

4: Highly degraded bowl (almost incorporated at soil)

Additionally, other observations as the presence of water in the bowl (WP, see Figure 8a), the presence of a soil pillar in the seedling hole (SP, see Figure 7d) and the colmatation with soil of the bowl (half silted bowl, HSB; fully silted bowl, FSB, see Figure 8b), will be noted.

Figure 7. Different degradation stages of the Cocoon, after 2-3 installation: (a) Cocoon in good state; (b) lid collapsed; (c) degraded bowl; (d) bowl almost incorporated at soil.

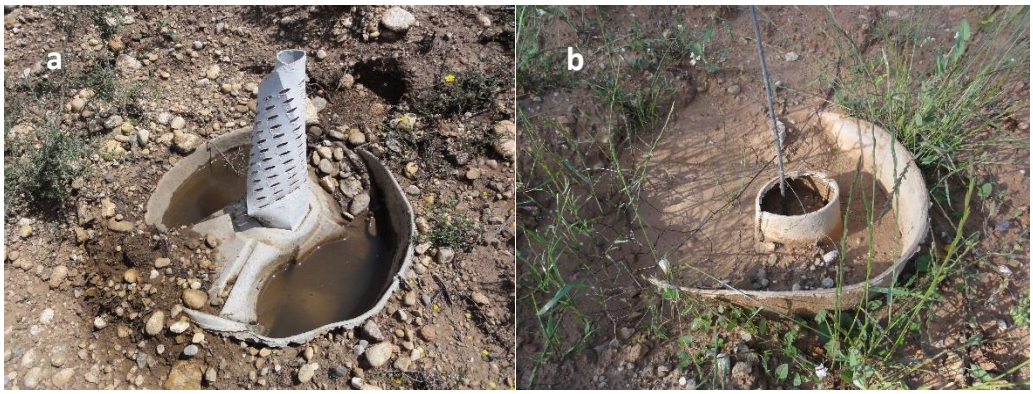


Figure 8. Other observations about the bowl: (a) presence of water in the bowl; (b) half silted bowl.

2. BELOW GROUND PROTOCOL (Soil and root final sampling)

As it was said in the previous version of the Below Ground Protocol: the reliability of a soil analysis depends on a good sampling. Soil tests and analysis will be done on a sample that is only a tiny, but representative, fraction of the field trial where Cocoons were installed and are causing "some effect". We assume that data from the analyses of the soil samples will show the effect of the Cocoon and plant growth in the soil of the trial area. Therefore, care must be taken to ensure that soil samples collected represent the soil influenced by the Cocoon and the seedling planted in it.

2.1 Soil sampling

In 2019, the last year of The Green Link Project, two representative soil samples at two different soil depths, topsoil layer (0-10 cm depth, **Ts**) and subsoil layer (20-30cm depth, **Ss**) will be taken in **spring** at each experimental site. THE NUMBER OF SAMPLES OF EACH SITE MUST BE THE SAME THAN IN THE FIRST SAMPLING (see table 1) AND THE LOCATION OF THOSE SAMPLES MUST BE THE CLOSEST POSSIBLE TO THE PLACE OF THE FIRST MONITORING. In the following paragraphs, how and where to take those samples is explained.

In each location (place of the first monitoring), the most representative species will be selected for soil and root sampling. It is very important that selected seedlings be the closest possible to the point where the first soil sampling was done (Fig. 1). In this point, a seedling planted with Cocoon is supposed to be located. Unearth that seedling that will be used also for root evaluation and carbon stock measurement. In the case that the soil sampling was not done in the place of this Cocoon, unearth the three selected seedlings closest to that point.

For the soil sampling, composite samples will be done by mixing the superficial soil (to do the Ts) collected from each of the three seedlings unearthed. The same will be done for the three sub-samples collected from the subsoil (to do the Ss). Every sample must be at least of 1.5 kg, trying to avoid rock fragments larger than 2 cm Ø, and visible rests of biomass (particularly those remaining after the microsite evaluation, section 1.5). Thus:

Ts. The top soil sample refers to the superficial layer of soil (depth has to be the first 10 cm). The sample will be taken precisely from the mass of soil introduced in the cylinder of the Cocoon box, where the plant is located, and in direct contact with the plant's root system.

Ss. The subsoil sample will be taken at a depth between 20 and 30 cm, below the Cocoon box (the box has a height of around 22 cm). Particularly important is to sample the soil around or within the deeper root system developed by the seedling.

2.1.1 Equipment

Use clean sampling tools and avoid contaminating the soil sample during their mixing or packaging. For instance, a small amount of fertilizer residue on your hands or tools, can distort the analysis results. Please, pay careful attention to not contaminate your tools with the humus (organic soil generally black) of the original seedling.

The necessary equipment for soil sampling will be:

- Spade or shovel preferably made of steel.
- Plastic bucket or large plastic bag for collecting and mixing subsamples.
- Plastic bag to contain about 1.5 kg of the final composite sample to be sent to CIDE.
- Waterproof marker to label the plastic bag to identify the sample.

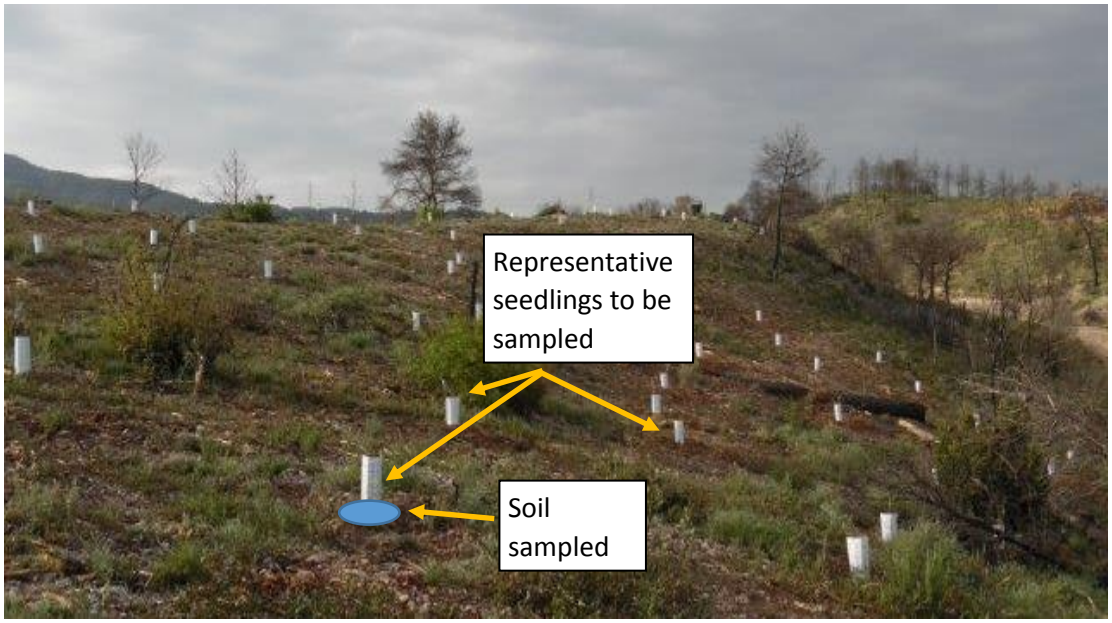


Figure 1. Selecting the seedlings to be removed (the closest to the point where the first soil sampling was done)

2.1.2 Methods

In summary, every partner has to:

1. Locate the point where the first soil sampling was done.
2. Identify the selected most representative species with Cocoon close to this point.
3. Choose at least three (3) seedlings with Cocoon of each selected species.
4. Remove the shelter (if still exists) and the Cocoon box. Please be very careful when removing the box in order to disturb the less possible the root system and the soil within and around it (Fig. 2).
5. Unearth the seedling avoiding to break the root system and trying to not disturb the first 10 cm corresponding to the Ts (Fig. 3).

6. Dig a hole around the system with the help of a spade or a shovel until you can take out the whole seedling (above and below ground biomass).
7. For the Ts, do a sub-sample with the soil of the first 10 cm of the selected seedling. This part should be done very carefully to not break the small roots and avoiding to include the humus (organic soil generally black) of the original seedling.
8. Repeat step 7 with at least two seedlings more. Put all sub-samples together in a plastic bag a mark it with: Name of the sample, depth and date.

Fig. 2. Seedling after removing the shelter (if still exists) and the Cocoon box.

9. If you haven't dug the hole around the root system, do so and take the Ss. For this, do a sub-sample with the dug soil, which should be located below each removed box and between 20 and 30 cm depth. This part should also be done very carefully to not break the root system and, eventually, to not sample the humus (organic soil generally black) of the original seedling.
10. Repeat step 9 with at least two seedlings more. Put all sub-samples together in a plastic bag a mark it with: Name of the sample, depth and date.

Fig. 3. Unearthing the seedling and the whole root system (not disturbed).

Very Important:

- DO NOT sieve or dry the samples. We will do it at CIDE

- Soil samples must be sent to CIDE immediately after being taken. Biological analyses should be done as soon as possible.

Table 1. Samples of the first sampling campaign.

Location	ID	Samples
Valencia	1	TOUS 1 (0-10cm)
	2	TOUS 1 (20-30cm)
	3	TOUS 2 (0-10cm)
	4	TOUS 2 (20-30cm)
	5	TOUS 3 (0-10cm)
	6	TOUS 3 (20-30cm)
Alicante	7	JIJONA 1 (0-10cm)
	8	JIJONA 1 (20-30cm)
	9	JIJONA 2 (0-10cm)
	10	JIJONA 2 (20-30cm)
	11	JIJONA 3 (0-10cm)
	12	JIJONA 3 (20-30cm)
Italy	13	BIOPOPLAR parte alta 10cm
	14	BIOPOPLAR parte alta 30cm
	15	BIOPOPLAR parte bassa 10cm
	16	BIOPOPLAR parte bassa 30cm
Cataluña	17	EL BRUC camp Ts (0-10)
	18	EL BRUC camp Ss (20-30)
	19	EL BRUC bosc Ts (0-10)
	20	EL BRUC bosc Ss (20-30)
Almería	21	Almería Ts (0-10)
	22	Almería Ss (20-30)
Greece	23	Upper plot (observatory) 0-10
	24	Upper plot (observatory) 20-30
	25	Black land (upper) 0-10
	26	Black land (upper) 20-30
	27	Black land (lower) 0-10
	28	Black land (lower) 20-30
	29	Sensoterra 0-10
	30	Sensoterra 20-30
	31	Next to Sensoterra 0-10
	32	Next to Sensoterra 20-30
	33	Sloped area 0-10
	34	Sloped area 20-30
Canarias	35	Pendiente ladera alta Ts 0-10
	36	Pendiente ladera alta Ss 20-30
	37	Terraza ladera alta Ts 0-10
	38	Terraza ladera alta Ss 20-30
	39	Terraza ladera baja Ts 0-10
	40	Terraza ladera baja Ss 20-30
Valencia	41	TOUS 4 0-10

Location	ID	Samples
	42	TOUS 4 20-30
	43	TOUS 5 0-10
	44	TOUS 5 20-30
Alicante	45	JIJONA 4 0-10
	46	JIJONA 4 20-30
	47	JIJONA 5 0-10
	48	JIJONA 5 20-30
	49	JIJONA 6 0-10
	50	JIJONA 6 20-30
Almería	51	Almería rambla Ts 0-10
	52	Almería rambla Ss 20-30
Cataluña	53	EL BRUC CT Ts 0-10
	54	EL BRUC CT Ss 20-30
	55	EL BRUC CT Fons Ts 0-10
	56	EL BRUC CT Fons Ss 20-30
Greece	57	Next to Grey Ts 0-10
	58	Next to Grey Ss 20-30
	59	Above black Ts 0-10
	60	Above black Ss 20-30
	61	Grey Plot Ts 0-10
	62	Grey Plot Ss 20-30
	63	Unit 5 Ts 0-10
	64	Unit 5 Ss 20-30

2.2 Root evaluation

Root evaluation will be done at the Cocoons selected for soil sampling, and the associated controls. Additionally, all seedlings unearthed for carbon stock measures (complete harvest, see section 3.1) will be also measured.

2.2.1 Methods

Every partner has to:

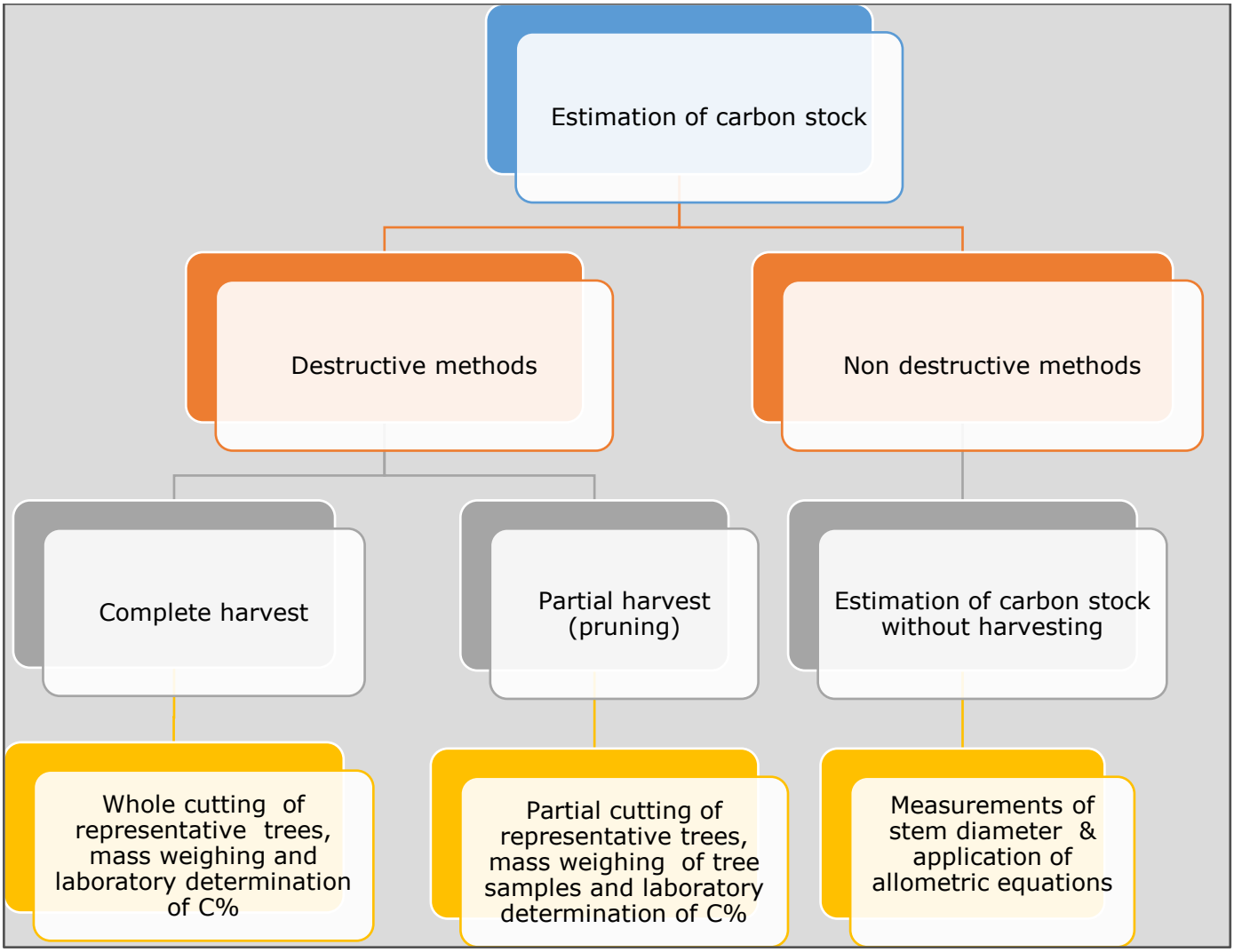
1. Below and above ground biomass should be separated (by using one of the tools suggested in the Plant biomass stock protocol).
2. Take pictures of the unearthed seedling (roots and stem), using a measuring tape/stick as a reference.
3. Length of the main root should be measured at the field (maximum length of the root system).
4. The weight of the root system should be determined in the lab after cleaning the roots (wet weight) and drying the whole plant (60°C for 4 days), dry weight. Be sure that soil is not adhered to the roots.
5. Put below (root system) and above ground biomass in separate plastic bags properly identified. Label in every bag must explain: seedling code (including seedling species), collecting and delivery date.
6. Send all the samples to CERTH's lab as soon as possible for carbon stock determination.

3. CARBON STOCK

Forest areas are among the most important carbon sinks of the terrestrial ecosystem. Forest vegetation takes up the carbon dioxide in the process of photosynthesis. In this natural process, the carbon dioxide is absorbed from the atmosphere, and then, the carbon is stored in the following carbon stocks (1):

- above ground biomass
- below ground biomass or root biomass
- forest litter
- woody debris or dead wood
- soil organic matter or soil carbon

The methods used for the estimation of carbon stock are summarized in Graph 1.



Graph 1. Methods for the estimation of carbon stock

The methods that are proposed for the estimation of carbon stock into the framework of Green Link project are:

1. Complete harvesting of species (destructive method)
2. Estimation of carbon stock using partial harvest (destructive method)
3. Estimation of carbon stock using allometric equations based on field measurements (non-destructive method)

3.1 Destructive method

Partial harvest method involves indirect measurement and/or sub-sampling of the tree (pruning) to estimate the tree biomass. This method does not give as accurate measure of tree biomass as the complete harvesting method. The estimation of carbon stock through partial harvest method involves three distinct parts:

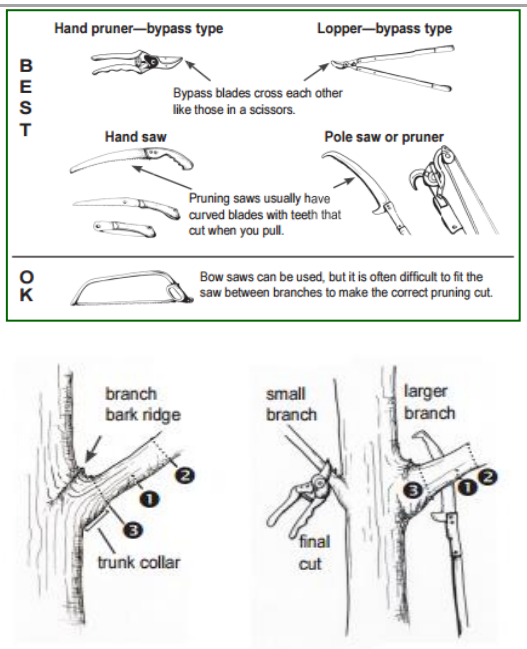
1. Tree volume estimation through field measurements of basal diameter and tree height
2. Tree partial harvesting (pruning), weighting of prunings of each tree, and sample collection for laboratory analysis for the determination of carbon concentration
3. Conversion of carbon content laboratory results to carbon stock per species considering the volume and the number of each species planted respectively.

Complete harvest method of the species can give accurate measurement of carbon stock. All parts of species can be sent to the laboratory for the carbon stock analysis.

The destructive method is divided in two different parts. The first one refers to the field activities for taking all the measurements needed by all partners, and the second one refers to the lab activities for estimating the amount of carbon stock, which concerns only CERTH.

The proposed method for the field activities is described below:

Step 1	Selection of representative species from each of the planted species (5-12 trees)	
Step 2	Measurement of stem diameter (at 10 cm from the bottom) and tree height of the aforementioned trees for volume estimation using a caliper and tape	**$V = (\pi D^2)/4 * h$** where: **V:** tree volume (m^3) (considering the stem of the tree has cylindrical shape) **D:** stem diameter (m) **H:** tree height (m)

Step 3	Pruning of the selected trees in order to collect representative samples without affecting tree growth. Note: Weight the mass of each sample separately which will be used to calculate tree volume	
Step 4	Place the collected samples in plastic bags and send them to CERTH laboratory for the determination of carbon content	**The total mass of sample should be 1-2kg. For that reason, the pruning procedure will be applied in more than one tree of each type**.

3.1.1 Additional instruction for pruning

In order to coordinate with the variety of the species' growth, a general template for the species' pruning was developed. The following three cases presented the harvesting categories:

Case 1: *Species with satisfying growth (many branches for harvesting)*

Partial harvesting: In this category will be included all the seedlings have a great survival rate and branches enough for harvesting 50-100gr of each tree. From each species, 1kg must be collected for the laboratory analysis (further instructions in the next section). The samples fragmented will be sent to CERTH's lab for completing the carbon stock measurement.

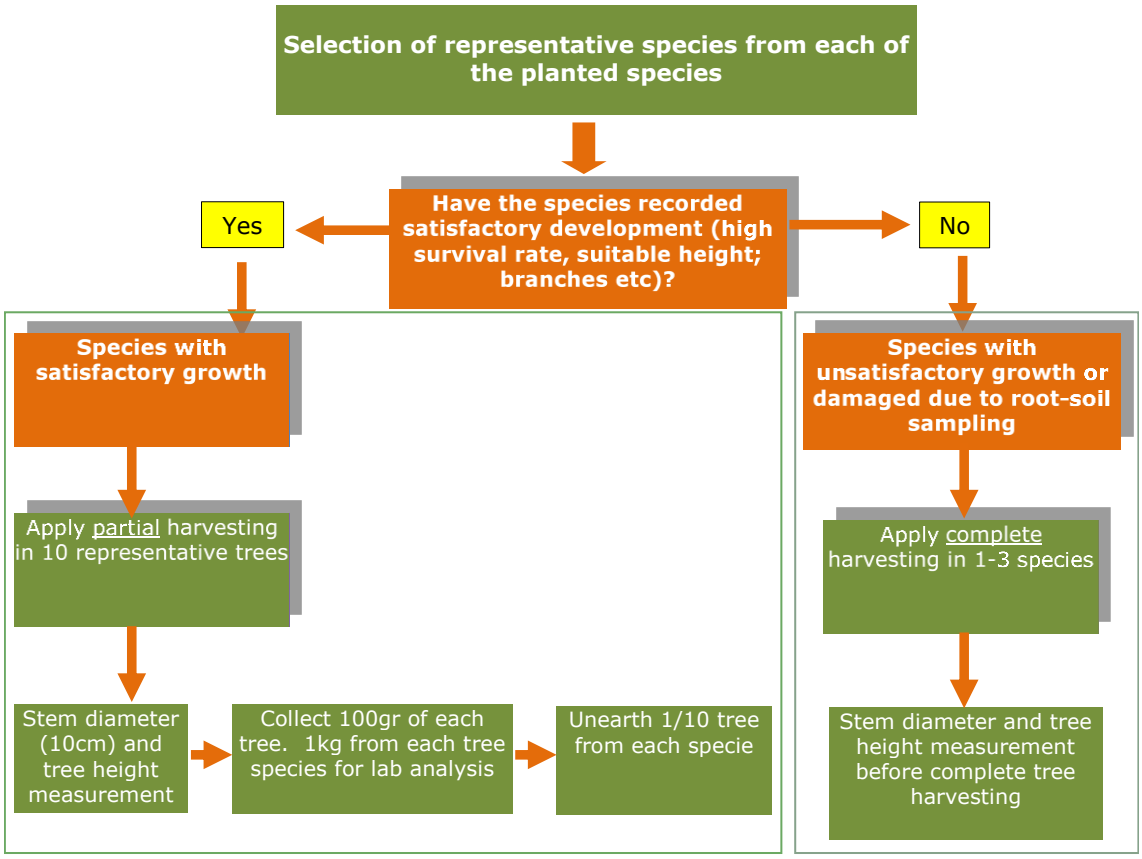
Case 2: *Species with inadequate growth (no or small branches, small height, tree's age < 3 years old)*

Complete harvesting: Three individuals per species (in order to ensure representativity) with not satisfying growth, with small or no branches, must totally be unearthed in order to estimate the carbon stock.

Case 3: *Species affected from the soil sampling*

Complete harvesting: In the project's framework, soil sampling from some species will take place. From this procedure, specific species will be affected and destroyed. Taking profit of this soil sampling, the affected species can be harvested completely and sent to CERTH's lab for the carbon stock analysis.

These three harvesting categories are summarized in Graph 2.



Graph 2: Field procedure for carbon stock estimation

3.1.2 How to harvest the species of trees and shrubs

a. Partial tree harvesting

Each partner has to collect a representative sample of each tree species in the trial area. The representative sample must be from at least **10 trees (subsamples)** of the **same tree species** in order to conduct an accurate measurement of carbon stock. It would be better if the sampled trees have a medium distance among each other. The pruning of each specie suggested weighting **1 kg. Each subsample (sample of one tree) should weight 100gr. Therefore, the total weight of the sample of each tree species would be 1 kg (10 trees of the same specie * 100gr/tree = 1000gr).**

The samples must be fragmented before sent to CERTH's lab for better logistic and management. The fragmented samples should be in plastic labelled bags. The label must have the following information: **tree species, number of trees collected, kilograms, collected date, site, delivery date.**

b. Complete tree harvesting

In order to have a complete measurement of carbon stock, it is preferable to harvest completely one seedling of each species. By this way, CERTH will have the chance to measure not only the above ground carbon stock, but also the below ground. Furthermore, the below ground estimation of carbon stock will be also estimated with the equation mentioned in the following table.

By cutting the whole seedling, each partner has to weight the fragmented parts (stem, branches, and root) after removing (cleaning) the soil adhered to roots and drying all the parts at 60°C for 4 days (this will be an important data for the lab work). The fragmented parts (stem, branches, and root) of the seedlings collected would be sent in different plastic bags to CERTH.

For example, the plastic bags for an olive tree sample will be one bag for the fragmented stem, one bag for the fragmented branches, and one bag for the root.

Each plastic bag must have a label with the appropriate information such as **tree species, kilograms, collected date, site, delivery date, and the part of the tree (stem, branches and root).**

c. Shrubs harvesting

Shrubs' root system usually is well grown and fibrous. For the complete harvesting of the shrubs species is needed to pay attention on how to unearth the seedling because of the complexity the root system may have. A shovel for digging around the shrub will be a necessary tool in order not to hurt the root system.

After the uprooting of the shrubs, it must be divided in the two main parts. In one plastic bag put the above ground material, and in another one, put the root system harvested.

Each plastic bag must have a label with the appropriate information such as **shrub specie, kilograms, collected date, site, delivery date, and the part of the shrub (stem & branches, root).**

The next steps concern only **CERTH** for the lab estimation of carbon stock and are given below:

Step 1	Estimation of above ground biomass carbon stock (CS_{AGB}) per species taking into account volume and wood density	**CS_{AGB}/tree = (C%)*V*ρ** **where:** **C%: carbon content (w/w)** **V: tree volume (m^3)** **P: wood density (kg/m^3)**
Step 2	Estimation of above ground carbon stock (CS_{AGB}) based on the laboratory results and the number of planted species	**CS_{AGB} = N *(CS/tree)** where: **CS_{AGB} :** carbon stock derived from above ground biomass **N:** number of trees
Step 3	Calculation of below ground biomass carbon stock (CS_{BGB}) using the a shoot to root ration	**CS_{BGB}= 0.27*CS_{AGB}**
Step 4	Estimation of total carbon stock (CS_t)	**CS_t= CS_{BGB} + CS_{AGB}**

Table 1. Species in all demonstration areas

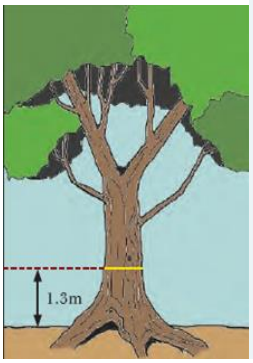
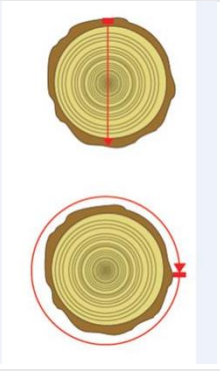
Tree species	Demonstration sites						
	El Bruc	Alicante	Valencia	Almeria	Gran Canaria	Calabria	Ptolemais
Olea europaea	X	X	X		X	X	
Quercus ilex. subsp. ilex (truffle)	X	X	X				
Quercus ilex. subsp. Ballota	X	X	X				
Quercus faginea	X						
Ceratonia siliqua	X	X	X				
Ficus carica	X	X				X	
Prunus dulcis	X	X	X	X			
Juglans regia	X		X				
Prunus avium	X						
Prunus spinosa	X						
Teraclinis articulata		X	X				
Pinus halepensis		X	X				
Arbutus unedo		X	X				
Cistus albidus		X					
Punica Granatum		X	X			X	
Celtis australis			X				
Tamarix				X			
Rosmarinus				X			
Juniperus turbinata						X	
Pistacia atlantica						X	
Robinia Pseudoacacia							X
Cupressus sempervirens							X
Macedonian Oak							X

3.2 Estimation of carbon stock using allometric equation (non - destructive method)

The allometric equations convert tree diameter at 1.3 m height (dbh) to biomass, or biomass expansion factors that convert standing volume to biomass. Then, tree biomass is converted to carbon using a standard conversion factor.

In the framework of **LIFE Green Link** project, and considering that the trees of the project have not the appropriate height to measure the stem diameter (at the 1.3 m height), we can measure the stem diameter at 10 cm height. In order to evaluate the allometric method results for carbon stock, we can compare the results of this non-destructive method with those of the destructive one.

The non-destructive method for the carbon stock estimation includes the following steps:

Step 1	Selection of representative trees of each tree species (4-12 trees)	
Step 2	Measurement of tree stem diameter at 1.3m *(10 cm-LIFE Green Link project)* above the soil surface using a caliper of girth tape	 
Step 3	Calculation of above ground biomass (AGB) per tree using an allometric equation based on stem diameter	**AGB = 0.139D$^{2.32}$** (kg/ tree; for regions with a rainfall height lower than 1500mm per year) **AGB = 0.118D$^{2.53}$** (kg/ tree; for regions with a rainfall height between 1500 and 4000mm per year)
Step 4	Calculation of below ground biomass (BGB) per tree using a shoot to root ration	According to European Guidelines for the calculation of BGB, following ratio shall be used: **BGB = 0.27xAGB (kg/ tree)**

Step 5	Conversion of AGB and BGB to carbon stock (CS) by multiplying by 0.47	**CS = 0.47x(AGB+BGB) (kg/tree)**
Step 6	Estimation of total carbon stock based on land area or number of species	**TCS = 0.47x(AGB+BGB)xN,** where **N**: number of trees per specie

All partners have to complete only **Step 1 and Step 2** for this non-destructive method. From **Step 3 to Step 6** only concerns CERTH's actions for the carbon stock calculation.

3.3 References

1. **Intergovernmental Panel on Climate Change.** Guidelines for National Greenhouse Gas Inventories. Institute of Global Environmental Strategies (IGES). *Agriculture, Forestry and other and-use.* Volume IV, 2006.
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4. SUMMARY TABLE, SPRING 2019 MEASURES (ABOVE GROUND, BELOW GROUND, CARBON STOCK)

ABOVE GROUND	
Task/Measure	**How**
Vigor	Visual evaluation (same seedlings than 2017 and 2018 campaign)
Growth	Height and diameter measures (same seedlings than 2017 and 2018 campaign)
Vegetation structure	Cover and height measures taken in 25 m transects (same transects than 2017 campaign)
Vegetation composition (plant diversity)	Plant identification and species abundance (same sub-sites than 2017 campaign)
Microsite evaluation	Cover measures and harvesting in 1 m circle (diameter) around seedlings (Cocoons where soil sampling has been done and respective controls)
Fauna evaluation	Visual indicators (footprints, tracks, nests, direct observation…), same sub-sites than 2017 campaign
Cocoon degradation	Visual evaluation (same seedlings than vigor and growth measures)
BELOW GROUND	
Task/Measure	**How**
Soil sampling	Topsoil layer (0-10 cm depth, Ts), subsoil layer (20-30cm depth, Ss); 1.5 kg per sample, avoiding rock fragments > 2 cm Ø (same sub-sites than 2017 campaign, same number of samples)
Root evaluation	Root length, weight and dry biomass measure (Cocoons where soil sampling has been done and respective controls) + all seedlings unearthed for carbon stock measures (1 seedling per species)
CARBON STOCK	
Task/Measure	**How**
Complete harvest (shrubs and trees)	Unearthing, cleaning (roots), drying (60°C for 4 days) and weighting the different parts of the seedling (stem, branches, and root) (Cocoons where soil sampling has been done + one seedling of the other species)
Partial tree harvesting	Pruning branches in species with high development (10 trees (subsamples) of each species, 100 gr per tree= 1kg)

ANNEX

ABOVE GROUND MEASURES FIELD FORMS

VIGOR, GROWTH AND COCOON ASSESSMENT FORM

Date of measurement:			**Site:**		**Sub-site:**	
Seedling identification (code)	**Adjacent control (code)**	**Vigor[1]**	**Height (cm)**	**Diameter (mm)**	**Cocoon degradation[2]**	**Other observations[3]**

[1]Vigor codes:

3: Healthy tree, with more than 75% of green, not wilted leaves. Also active growing points (apices) may be visible

2: Affected tree, with 25-75% of the leaves being wilted, yellow or brown

1: Severely affected tree with less than 25% of the leaves being green (i.e. the majority wilted, yellow of brown)

0: Presumably dead tree with no or only wilted leaves. Tree seedlings, however, may still recover by resprouting after a rain event

R: resprouted tree

[2]Cocoon degradation codes:

1: Cocoon ok: with or without shelter, but with lid

2: Lid collapsed but bowl apparently in good state (without craks, holes)

3: Bowl with signs of degradation (cracks, holes)

4: Highly degraded bowl (almost incorporated at soil)

[3]Other observations codes:

G: Grazing

B: Broken stem

P: Plague infestation

T: Tree fall

WP: presence of water in the bowl

SP: presence of a soil pillar

HSB: half silted bowl

FSB: fully silted bowl

ESTRUCTURE MEASURES FORM

Date of measurement: **Site:** **Sub-site:**

	0 m					1 m					2 m					3 m					4 m				
	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100
Herbaceous																									
Woody																									
Org. debris																									
	5 m					6 m					7 m					8 m					9 m				
	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100
Herbaceous																									
Woody																									
Org. debris																									
	10 m					11 m					12 m					13 m					14 m				
	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100
Herbaceous																									
Woody																									
Org. debris																									
	15 m					16 m					17 m					18 m					19 m				
	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100
Herbaceous																									
Woody																									
Org. debris																									
	20 m					21 m					22 m					23 m					24 m				
	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100	0 - 20	20 - 40	40 - 60	60 - 80	80 - 100
Herbaceous																									
Woody																									
Org. debris																									

BIODIVERSITY MEASURES FORM

Date of measurement: **Site:** **Sub-site:**

SPECIE	ABUNDANCE

Abundance codes:

1: testimonial

2: low frequency (<25% soil cover)

3: high frequency (25%-75% soil cover)

4: dominant (>75% soil cover)

FAUNA EVALUATION FORM

Date of measurement: **Site:** **Sub-site:**

OBSERVATION TYPE	PRESENCE	ABSENCE	SPECIE/S
FOOTPRINTS			
EXCREMENTS			
LAIRS			
NESTS			
DIRECT OBSERVATIONS			

MICROSITE EVALUATION

Seedling identification (code): CONTROL Harvested vegetation weight (wet weight, g):

TRANSECT 1

	0 m				0.5 m
	0 - 10	10-20	20-30	30-40	40-50
Herbaceous					
	0.5 m				1 m
	50-60	60-70	70-80	80-90	90 - 100
Herbaceous					

TRANSECT 2

	0 m				0.5 m
	0 - 10	10-20	20-30	30-40	40-50
Herbaceous					
	0.5 m				1 m
	50-60	60-70	70-80	80-90	90 - 100
Herbaceous					

Seedling identification (code): COCOON Harvested vegetation weight (wet weight, g):

TRANSECT 1

	0 m				0.5 m
	0 - 10	10-20	20-30	30-40	40-50
Herbaceous					
	0.5 m				1 m
	50-60	60-70	70-80	80-90	90 - 100
Herbaceous					

TRANSECT 2

	0 m				0.5 m
	0 - 10	10-20	20-30	30-40	40-50
Herbaceous					
	0.5 m				1 m
	50-60	60-70	70-80	80-90	90 - 100
Herbaceous					