

**ESPA p4ges PROJECT**  
**Work Package Carbon (WP4-Carbon)**

**Manual for carbon stock quantification within the  
Ankeniheny-Zahamena Forest Corridor within the  
Below-Ground Biomass Survey framework  
(Manual n°2)**

**Period April 2014 - June 2015**

**Authors:** *RIANA Hary Andrisoa, RAZAFINDRAKOTO Mieja, JOURDAN Christophe, SAINT-ANDRE Laurent, RAZAFIMBELO Tantely, ANDRIAMANANJARA Andry, SANEHO Haritiana Julio Gabriel, RAMIFEHIARIVO Nandrianina, RANAIVOSON Ntsoa, RAZAFISOAZARA Mihajasoa, RAZAFIMANANTSOA Marie Paule, RAZAKAMANARIVO Herintsitohaina*

**Update:** *February, 2017*

## CONTEXT

The p4ges « *Can paying 4 global Ecosystem Services reduce poverty ?* » project in Madagascar aims to influence the development and the implementation of payment for ecosystemic services at international scale in order to alleviate poverty. Among ecosystemic services of interest, the carbon survey in the Ankeniheny Zahamena corridor (CAZ) in eastern Madagascar was carried out by the carbon workpackage (WP4; NE/K008692/1). Carbon stock assessment was performed in different land uses for five (5) pools defined by the IPCC (2006), were assessed 1) Aboveground biomass (AGB), 2) Belowground biomass (BGB), 3) Litter layer, 4) Dead wood (DW), 5) Soil Organic Carbon (SOC).

The present manual is established under the p4ges project “*Can paying 4global ecosystem services reduce poverty ?*” for the carbon survey conducted by WP4. It develops the second step of survey, focusing on belowground / Root that was based on data-results and information obtained through the use of Manual n°1 on classical survey. The content of this manual N°2 is mainly: i) an introduction part corresponding to the rationale of the root survey within p4ges project and ii) details on methodology process including: field and lab works, calculations ... allowing accurate belowground /root biomass and carbon quantification.

All datasets which were generated by methodology process described within this manual have been also archived at Environmental Information Data Centre - EIDC (<http://eidc.ceh.ac.uk>).

## COMPOSITION OF THE WP4-CARBON TEAM

### Central team

Dr. HDR RAZAKAMANARIVO Herintsitohaina  
Pr. RAZAFIMBELO Tantely  
Dr. ANDRIAMANANJARA Andry

### Collaborators

Dr. HDR SAINT ANDRE Laurent  
Dr. HDR JOURDAN Christophe  
Pr. RAJOELISON Gabrielle  
Dr RAMANANANTOANDRO Tahiana

### Assistants of research

RIANA Hary Andrisoa  
RAZAFINDRAKOTO Mieja Ambina  
SANEHO Haritiana Julio Gabriel  
RANAIVOSON Ntsoa  
RAMIFEHIARIVO Nandrianina  
HARINIRINA Ando

### Responsible of laboratory

RAZAFIMANANTSOA Marie Paule

### Field team

RAKOTONINDRINA Hobimiarantsoa  
RAZAFISOAZARA Mihajasoah Georgine  
RANDRIARILALA Tanteliniaina  
RANDRIANASOLO Heritiana Michel  
RAOELIJAONA Lanja Andriamialitadiavina  
RANAIVO HARIMALALA Tovohery Angelo

ANDRIANOTAHINA Harivony Honoré  
RABEKOTO Maharavo  
RANDRIANIRINAMIFIDIMANANTSOA T. V.  
RANAIVOARISON Adelin Modeste  
RANDRIANARIJAONA Jean Marcel  
RAZAFIARISON Justin Théodore

**GLOSSARY, ABBREVIATIONS and UNITS**

<b>Terms</b>	<b>Abbreviations and units</b>	<b>Description</b>
<b>Aboveground biomass</b>	<b>AGB (<i>kg</i> at tree level and <i>MgC.ha<sup>-1</sup></i> at site level)</b>	All biomass of living vegetation, both woody and herbaceous, above the soil including stems, branches, bark, seeds, and foliage. (IPCC, 2006)
<b>Ankeniheny Zahamena Forest Corridor</b>	<b>CAZ</b>	Remains of the evergreen forest of eastern Madagascar
<b>Belowground biomass</b>	<b>BGB (<i>kg</i> at tree level and <i>MgC.ha<sup>-1</sup></i> at site level)</b>	All biomass of live roots.
<b>Carbon</b>	<b>C</b>	Carbon in a defined pool
<b>Closed canopy</b>	<b>CC</b>	Primary forest formation
<b>Coarse root</b>	<b>CR</b>	Roots with diameter $\geq 10$ mm
<b>Coarse root above soil</b>	<b>CR0</b>	Coarse roots found above soil
<b>Diameter at breast height</b>	<b>DBH (<i>cm</i>)</b>	Diameter of the measured tree at 1.3m height
<b>Fine Root</b>	<b>FR</b>	Fine roots with diameter $< 2$ mm
<b>Height</b>	<b>H (<i>cm</i>)</b>	Height of the measured tree
<b>Intergouvernemental Panel on Climat Change</b>	<b>IPCC</b>	
<b>Laboratoire des RadioIsotopes</b>	<b>LRI</b>	Laboratory of soil carbon and biomass analysis- Antananarivo University
<b>Land use</b>	<b>LU</b>	The type of activity being carried out on a unit of land (IPCC, 2003)
<b>Medium root</b>	<b>MR</b>	Medium roots with $2 \text{ mm} \leq \text{diameter} < 10 \text{ mm}$
<b>Can paying for global ecosystem services reduce poverty</b>	<b>p4ges</b>	“Can Paying for ecosystem services reduce poverty?” project
<b>Plot</b>		Individual site (corresponding to a land use types) which was common for biophysical workpackages in which the survey was done
<b>Shrub fallow</b>	<b>SF</b>	A land use type corresponding to subsequent fallow cycle after deforestation (Styger <i>et al.</i> , 2007)
<b>Site</b>		Unit of sampling (corresponding to a plot) that can either an individual site or included within a transect site
<b>Tany maty or Degraded land</b>	<b>TM (or DL)</b>	A land use type corresponding to final stage of deforestation, or land that is abandoned (Styger <i>et al.</i> , 2007)
<b>Tree fallow</b>	<b>TF</b>	A land use type corresponding to first fallows following deforestation
<b>WorkPackage</b>	<b>WP</b>	Specialized group of researchers and assistants working on the different work

		packages of the p4ges project
<b>WorkPackage 1</b>	<b>WP1</b>	P4ges workpackage in charge of remote sensing and design sampling
<b>WorkPackage 2</b>	<b>WP2</b>	P4ges workpackage in charge of hydrology survey
<b>WorkPackage 4</b>	<b>WP4</b>	P4ges workpackage in charge of carbon assessment
<b>WorkPackage 5</b>	<b>WP5</b>	P4ges workpackage in charge of biodiversity survey
<b>Zone of Interest</b>	<b>ZOI</b>	Area of distribution of sampling effort throughout the spatial extent in order to capture the variation in CAZ and limit pseudo replication

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## 1. Introduction

The carbon workpackage (WP4-carbon) team of the p4ges project “Can paying 4global ecosystem services reduce poverty” works on carbon quantification in the Ankeniheny Zahamena corridor (CAZ) in eastern Madagascar. C stock assessment was performed in different land uses for all five (5) pools (cf. Manual n°1). Among these 5 pools, special attention was given to root/below-ground biomass which is an important pool but little studied. Indeed, below ground biomass (BGB) constitutes a significant part of total forest biomass and could represents up to 40% of the total biomass (Cairns *et al.*, 1997). However, the root biomass is often omitted because study of all BGB and its distribution in the soil profile by direct methods are complicate, hard-working and time consuming (Vogt *et al.* 1998). Particularly for humid tropical forest in Madagascar, no study has yet examined this pool. In that sense, BGB was generally assessed indirectly by using the Root: Shoot ratio ( $R:S$ ) which corresponds to the relative biomass allocation between roots and other tree parameters such as above-ground (AGB).

Hence, the aim of the WP4 team, with the collaboration with other researchers from the International Center for promoting Agriculture and Development (CIRAD) and the High School of Agronomy-Department of Forest (ESSA-Forests), was to carry out an in-depth study of root component. For that, BGB part of each tree was subdivided into four categories: stump, coarse roots (CR with diameter  $\geq 10$  mm), medium roots (MR,  $2 \text{ mm} \leq \text{diameter} < 10 \text{ mm}$ ) and fine roots (FR, diameter  $< 2 \text{ mm}$ ) (Levillain *et al.*, 2011). Stump corresponds to the part of the tree joining the trunk of above-ground with root part (up to the first root ramification where the roots could be clearly individualized) (Wildy and Pate 2002; Saint-André *et al.*, 2005). The main objectives of this BGB survey were: i) to carry out direct measurement in some numbers of target trees for accurate BGB estimation and; ii) to compare defined methods of BGB quantifications on these same trees/same plots/same areas; iii) to calculate BGB at tree level ( $\text{kg.tree}^{-1}$ ) in order to develop later allometry equations which will allow to quantify BGB at stand level ( $\text{Mg.ha}^{-1}$ ).

## 2. Materials and Methods

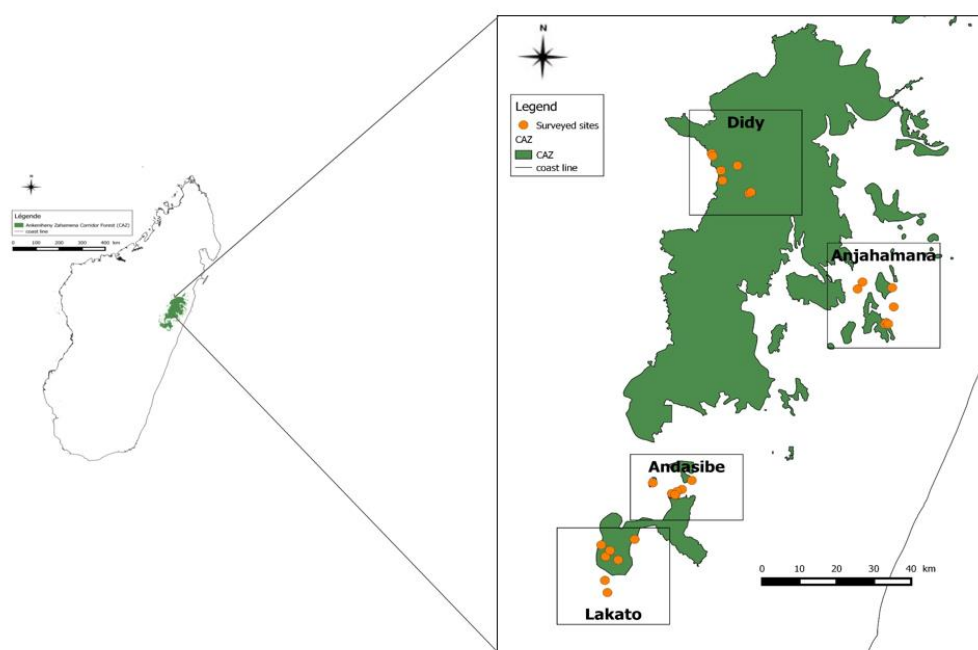
### 2.1. Study area

WP4 survey was conducted in humid tropical forest in Ankeniheny-Zahamena forest corridor (CAZ), located on the eastern Madagascar. This area, extends over 370,032 hectares (ha), is located between  $48^\circ 49' - 49^\circ 03'$  East and  $17^\circ 30' - 17^\circ 43'$  south. Four zones of interest (ZOIs) were selected for the purposes of the whole p4ges project: Lakato (ZOI1), Andasibe (ZOI2), Anjahamana (ZOI3) and Didy (ZOI4). The selection of four zones was based on a range of biophysical criteria (cf details in Manual

n°1): altitude, slope and other biophysical criteria as bioclimate zone, length of dry season, soil type. Moreover, others criteria were considered such as the deforestation history and access.

## 2.2. Process for selection of target trees

In-depth study of BGB only concerns Closed Canopy forest land use type in the four (4) ZOIs (cf Map1). Since direct measurement of root biomass is complicate, destructive and costly, selection of target trees is then mandatory (cf Figure 1). The process included selections of relevant ZOIs, sites and individual target trees showing specific species/diameter.



**Map 1: Location of all closed canopy sites per zone of interest (ZOI) in CAZ**

### 2.3.1. Identification of zones of interest for direct measurements

A previous step of forest inventory was carried out within all sites of closed canopy in the four ZOIs (Manual n°1) during which: name of species, DBH and its classes (Large tree/more than 20 cm, Medium tree between 20 and 10 cm, Small tree/less than 10 cm), and height (H) were recorded. In order to have an idea of AGB density, AGB at tree level ( $\text{kg.tree}^{-1}$ ) was estimated by applying on the inventoried data the allometry equation developed by Chave *et al.* in 2014.

$$\text{CHAVE 2014: } \text{AGB} = 0.0673 * (\text{WSG} * \text{DBH} * \text{DBH} * \text{H})^{0.976} \quad (\text{Equation 1})$$

This model developed by Chave *et al.* (2014) was considered because it was the most recent and already included any existing national allometry equations. Subsequently, total AGB at stand level



(Mg.ha<sup>-1</sup>) for each site was calculated by summing the AGB values of all trees within this considered site and upscaling this value to one hectare.

Considering these available information, two (2) ZOIs among the four (4) were chosen according to the obtained AGB at stand level (Mg.ha<sup>-1</sup>), based on existing extreme values. ZOI3 Anjahamana showed the highest average value and ZOI2 Andasibe showed the lowest average value, ZOI1 Lakato presented similar value as ZOI2 and ZOI4 Didy had medium value. While considering the access on each ZOI and site, ZOI3 Anjahamana and ZOI2 Andasibe were retained for all BGB direct measurements.

### **2.3.2. Selection of sites**

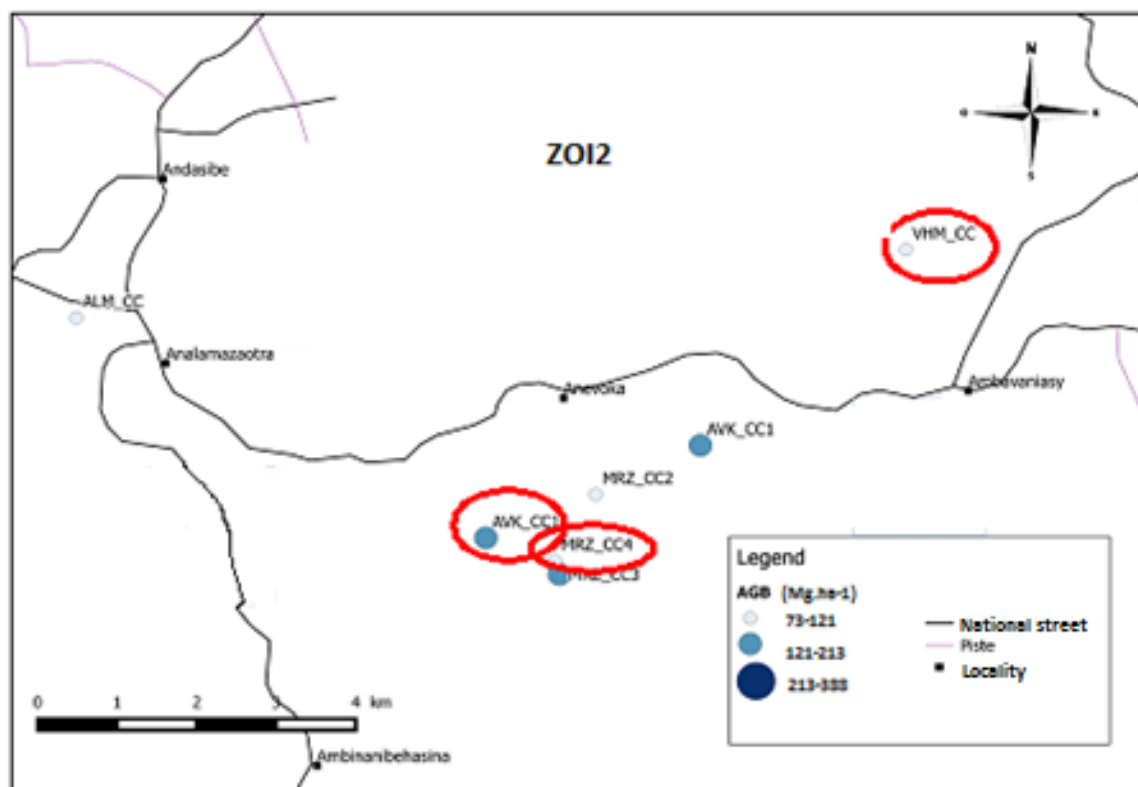
The selection of sites was based on the AGB values per site on the retained ZOIS and their relation with the representativeness of each tree size (proportion of trees per DBH class). In order to establish this relation, AGB per site was divided also to three classes (high, medium and low ranges of value) so that each class has the same number of values (a percentage of around 33% of the total number of sites for each class). Subsequently, maps were made showing the relationship of AGB per site (A for Map 2 and Map 4) and the contribution of each DBH class (B for Map3 and Map5). The aim was to search in each site, which category of tree (in terms of DBH class) contributed the most to its biomass (ABG). Afterwards, considering the contribution of this tree size into the AGB density per site (in Mg.ha<sup>-1</sup>) for each ZOI, at least 3 sites for each ZOI were selected for BGB direct measurements. This choice also took account of the access and any restrictions in field process in relation with the status of Protected Area (direct measurement not allowed in some part and some species within the CAZ).

### **2.3.3. Determination of target trees**

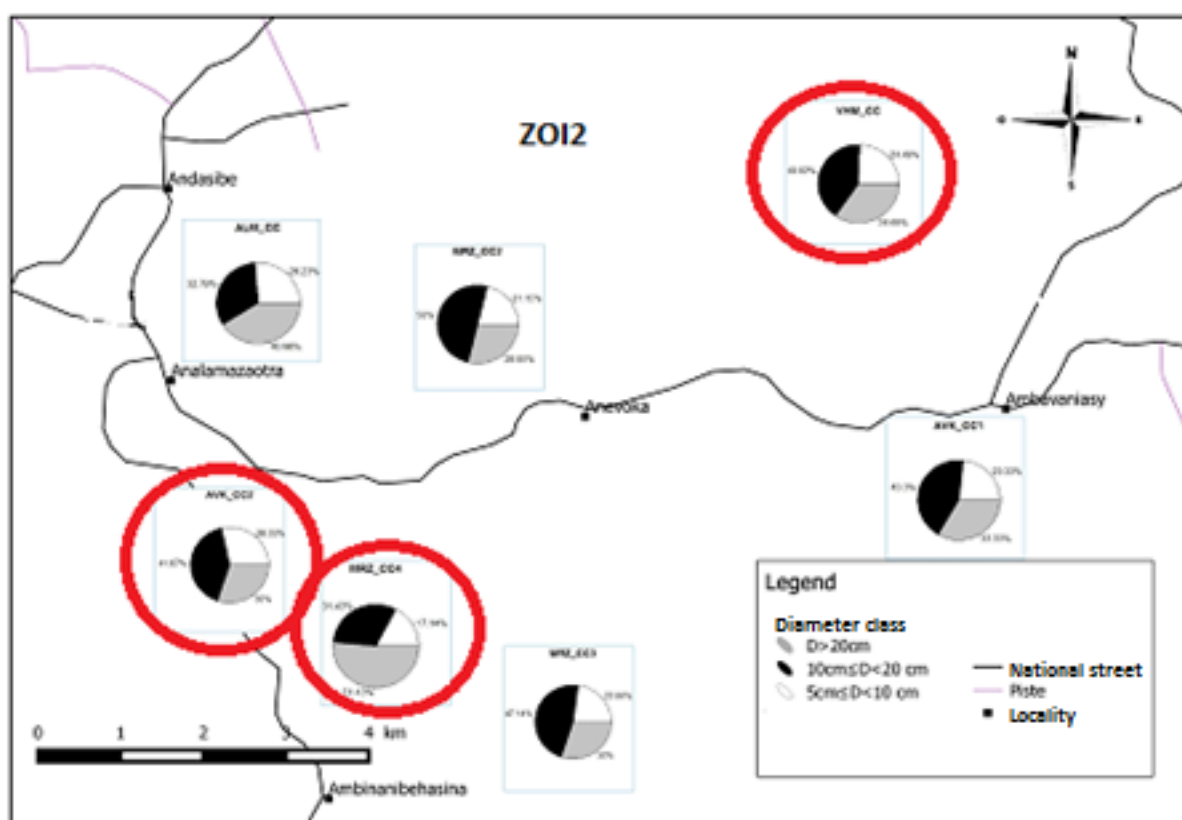
Considering the time allowed to the field work implementation, the related costs and the restriction in field process concerning the restricted species, the total number of target trees to be cut has been limited. Target trees were chosen from four (4) dominant species sorted out from the forest inventory with which WP4 team kept the three (3) most represented species per site and ZOI. Therefore, target trees were selected by considering the selection of zone, sites, diameter classes and species. To be noted that; three categories of trees were considered, large trees with  $DBH \geq 20\text{cm}$ , medium trees with  $10 \leq DBH < 20\text{cm}$  and small trees with  $5 \leq DBH < 10\text{cm}$ .

Following all these criteria, in total, 54 target trees were selected for direct measurements of BGB (cf Figure 1):

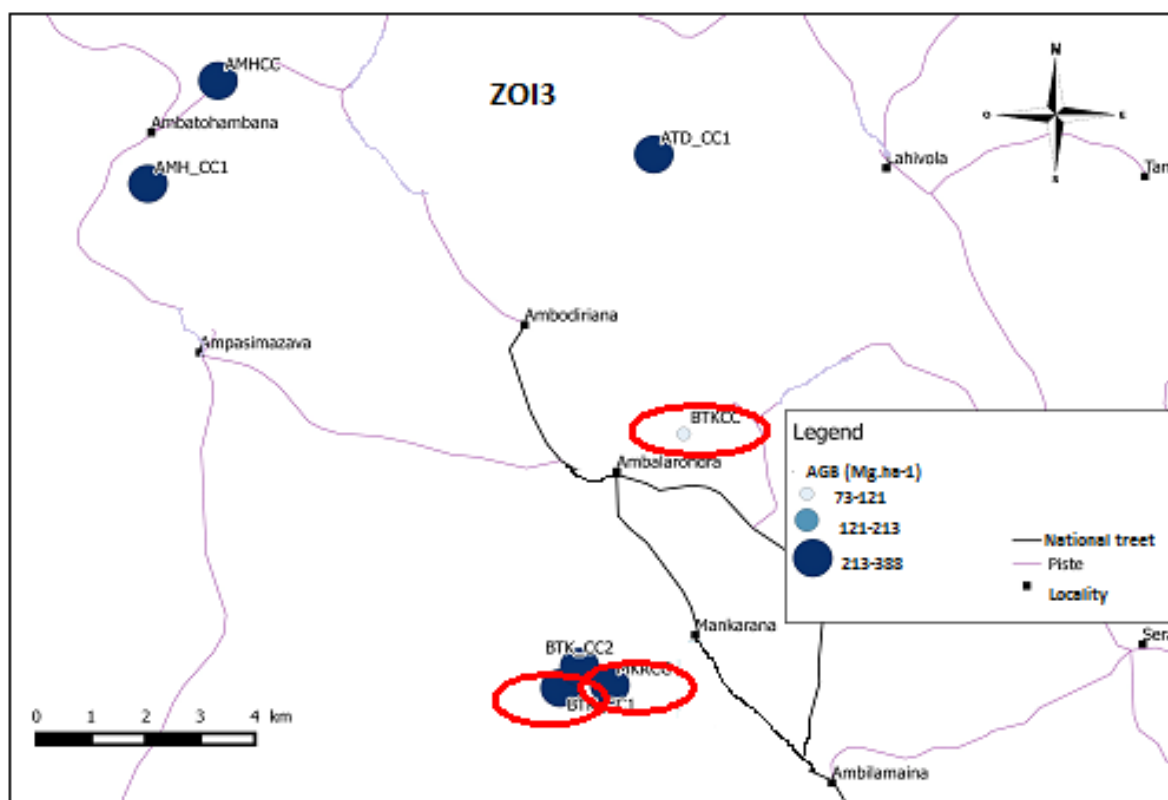
*2 zones \* 3 sites/zone \* 3 species per site \* 3 diameter class / species*



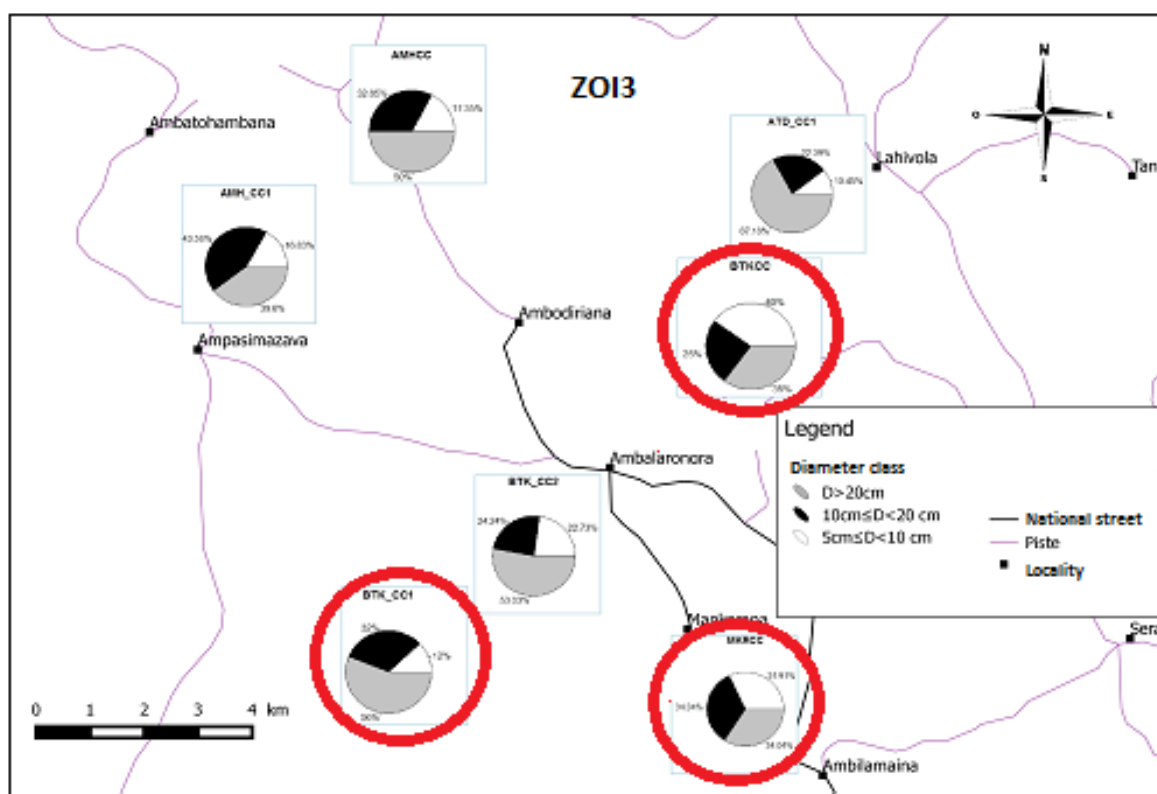
Map 2: A- AGB density ( $\text{Mg.ha}^{-1}$ ) for ZOI2 with the selected sites contoured in red



Map 3: B- Contribution of diameter class for ZOI2 with selected sites contoured in red



Map 4: A- AGB density ( $\text{Mg.ha}^{-1}$ ) for ZOI3 with the selected sites contoured in red



Map 5: B- Contribution of diameter class for ZOI3 with selected sites contoured in red

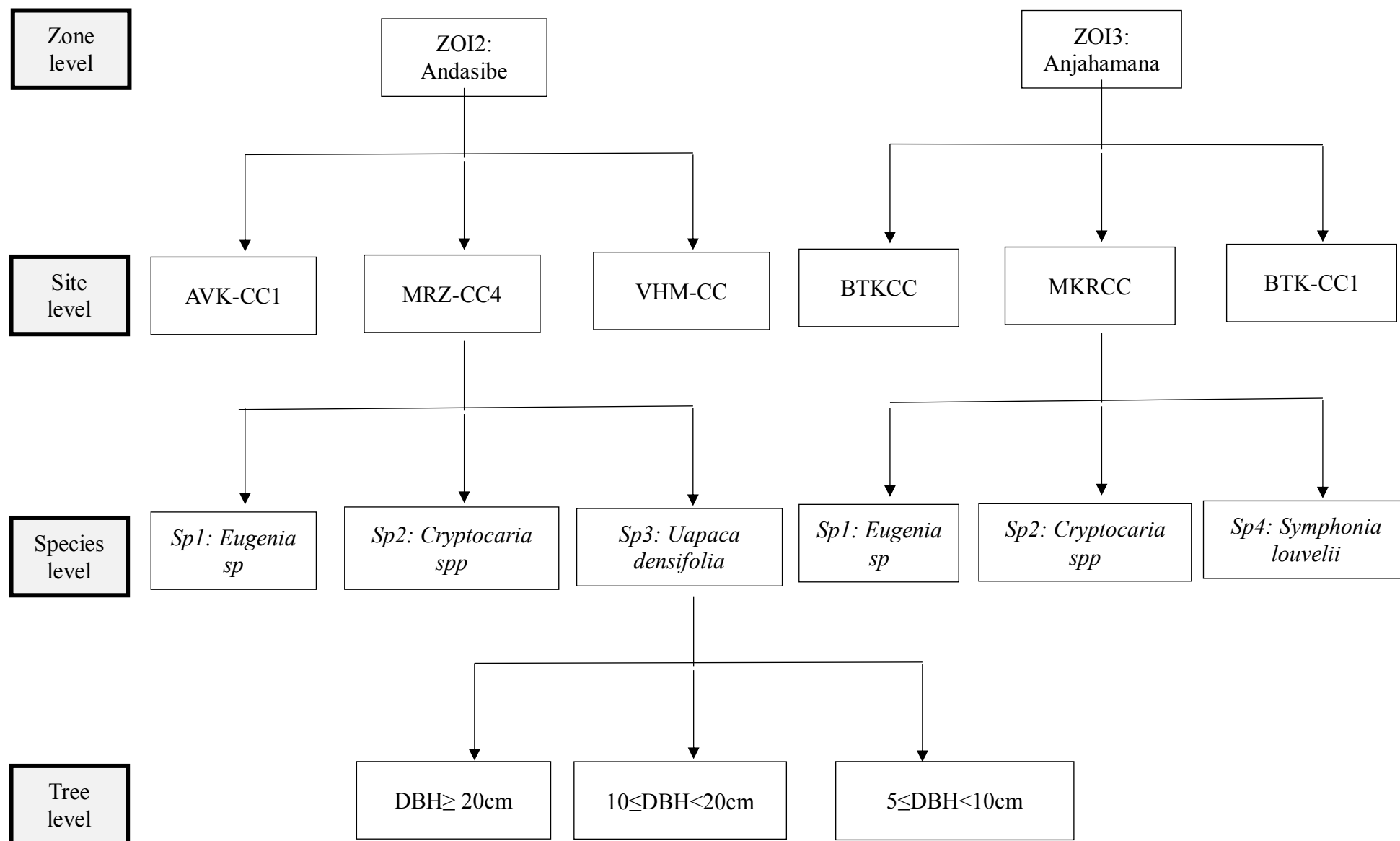


Figure 1: Process for selection of target trees for direct measurement

## 2.3. Description of methods developed in the field

For the BGB measurement in field, two main methods were adopted:

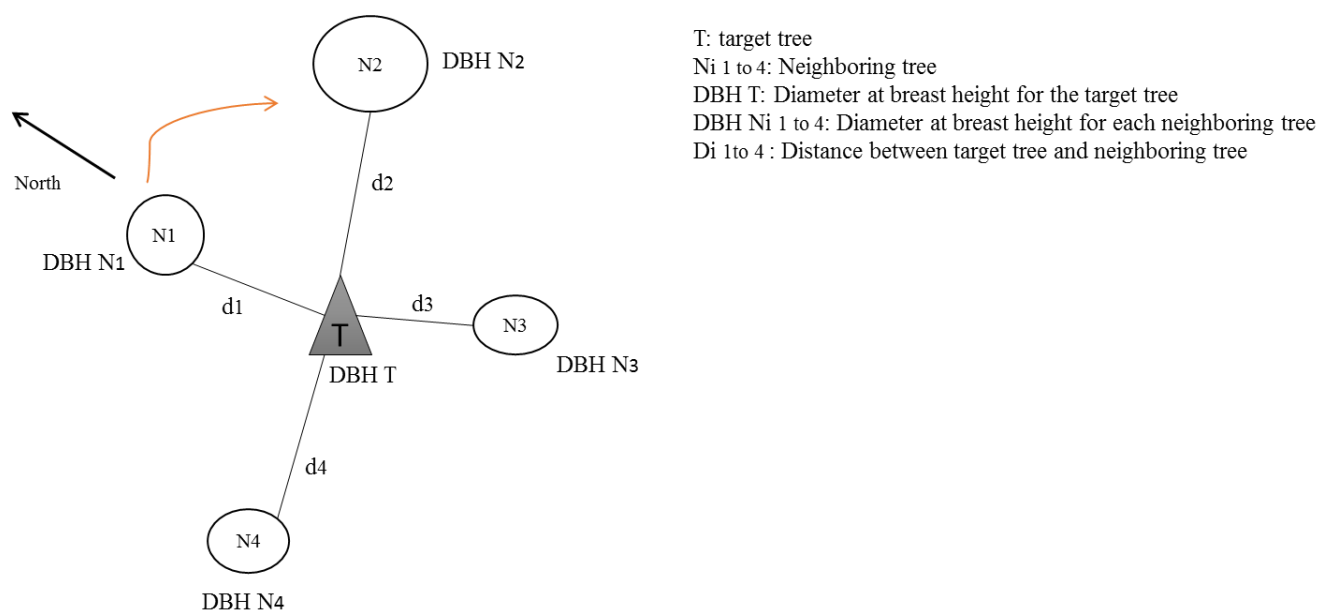
- Method of *Voronoi polygon*
- Method of *Pits*

### 2.3.1. Method of Voronoi polygon

#### a. Description of the method

*Voronoi polygon* is the polygon of occupancy and the elementary space which is formed by the intersection of the perpendicular lines that pass through the midpoints of the lines connecting the center of the sampled tree to the center of the nearest neighboring trees (Santantonio *et al.*, 1977; Saint-André *et al.*, 2005). Here a simplified voronoi were used because of tropical forest complexity, where trees have different ages and different dimensions (large trees are neighbors of small trees). We assumed that the influence of their respective root system is linked to their global dimension. And all the roots of the target tree that grow outside the Voronoi polygon are balanced by those of neighboring trees growing inside the polygon (Levillain *et al.* 2011).

#### b. Experimental design



**Figure 2: Experimental design for method of voronoï at the first step**

Based on this experimental design of the voronoi polygon method (figure 2), different steps were followed (figure3 and figure 4), like:

- Research of the target tree following the criteria of the selection explained in previous sections
- Numbering neighbouring trees according to the direction of a hand of a watch

- Record of the dendrometric parameters such as name of species, DBH, H of the target tree and each neighbouring tree
- Measurement of the distance between target tree and each neighbouring trees ( $d_i$ )
- Determination of the importance of the size of the target tree considering the size ( $x_i$ ) of each neighbouring tree

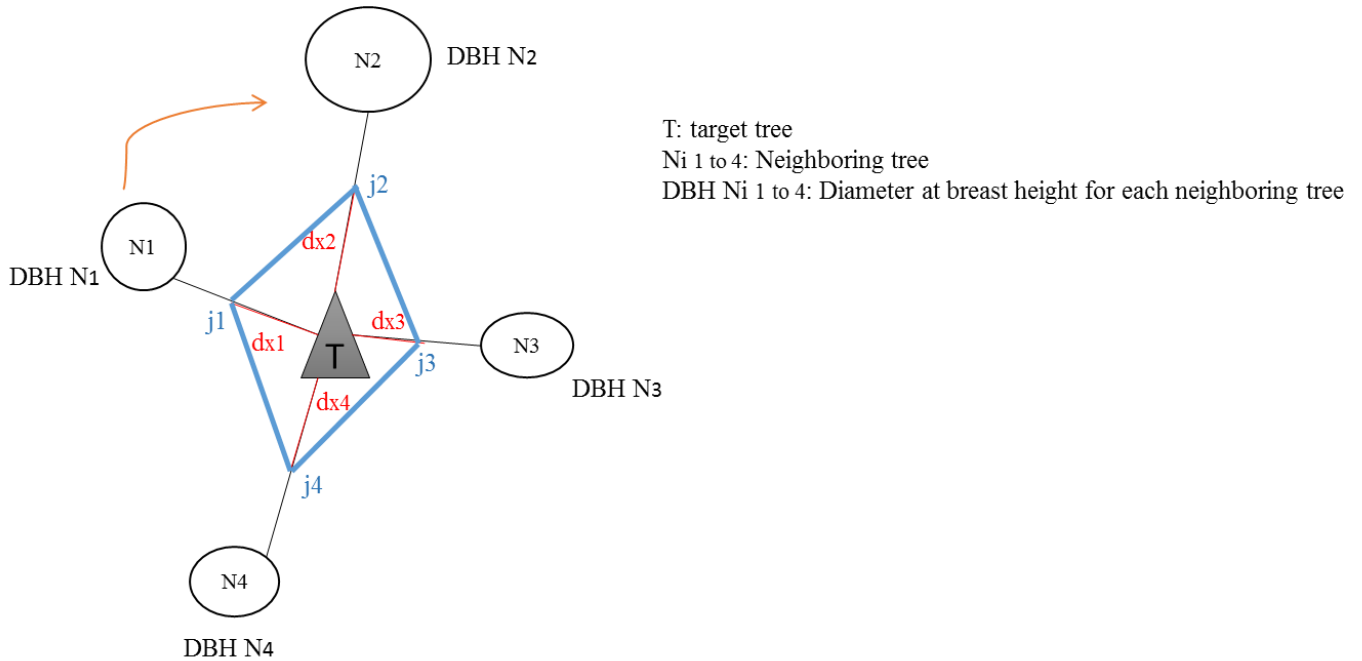
$$x_i \% = \frac{DBH\ T + DBH\ N_i}{DBH\ T} * 100$$

(In our example, I varied from 1 to 4, assuming that there are 4 neighbours' trees)

- Calculation of the distance  $dx_i$  corresponding to the space  $x_i\%$  from the target tree. Then,  $dx_i$  was materialized in the field and was used for real delimitation of the voronoi polygon

$$dx_i = x_i \% * d_i \text{ with } 1:1 \text{ to } 4$$

The junction  $j_i$  ( $i: 1$  to  $4$ ) of each distance  $dx_i$  allowed the final delimitation of the polygon.

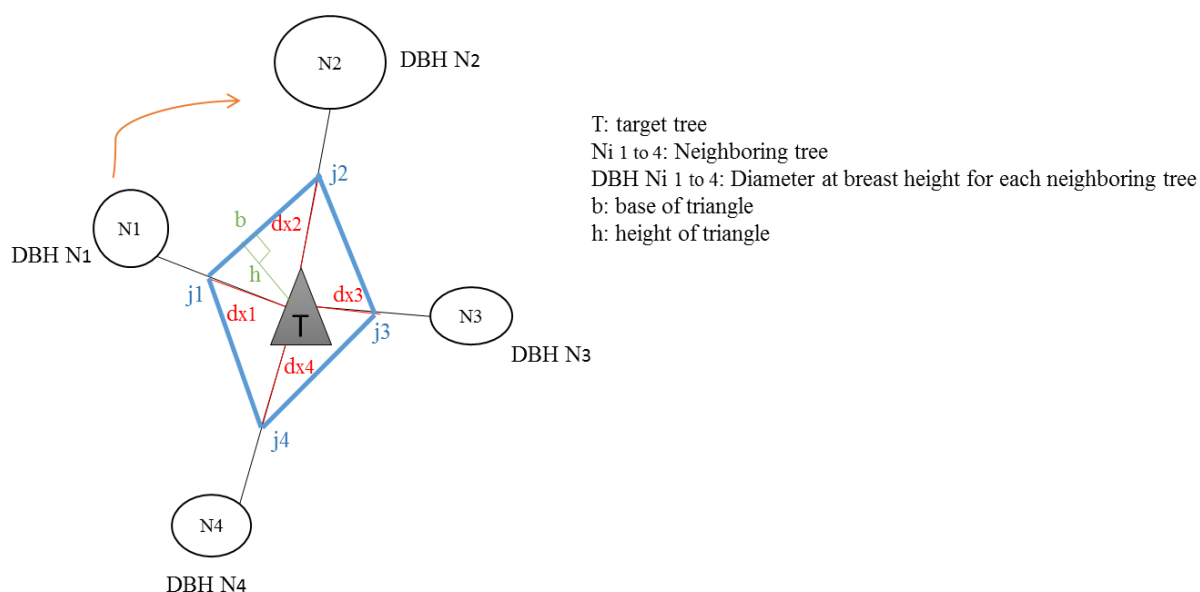


**Figure 3: Step for the materialization of the final delimitation of voronoi polygon**

- For the calculation of the eventual area of the voronoi polygon (cf figure 4), height and the base of each triangle which composed the polygon was plotted and the area of each triangle was calculated by the formula:

$$\text{Area of triangle: } (b * h)/2$$

With  $h$ : height (m),  $b$ : base (m)



**Figure 4: Step of calculation of the total area of voronoi polygon**

The area of the whole polygon was then calculated by summing areas of all existing triangles.

#### c. Collection of root data

In order to better ensure representativeness of all direct measurements, for each Voronoi polygon, one triangle was chosen for small tree with  $5 \leq \text{DBH} < 10\text{cm}$  and two triangles for medium and large trees with  $10 \leq \text{DBH} < 20\text{cm}$  and  $\text{DBH} \geq 20\text{cm}$  respectively. When two triangles were retained, one of them was located at the down of slope/tree and the one at the top of slope/tree.

In each triangle, excavation of soil and extraction of each category of roots (FR, MR, and CR) was done. Three soil horizons were considered considering general observation in the field: top soil from 0 to 10 cm, subsoil from 10 to 30cm and deep soil 30-50 cm. For CR, the roots above soil (CR0) were also considered.

Carefully, categories of roots of the target tree were distinguished from the neighboring trees. The stump and pivot were weighed separately and samples were collected. Then, roots were washed, weighed and samples were collected, weighed and put inside bag for laboratory analysis. The total fresh weight of each component was obtained in the field using electronic balances.

Furthermore, each ramification of CR of the target tree was measured to have a more accurate estimation of roots. The process of ramification measurement was showed in figure 5. The ramification is defined here by each section of CR from the base of the stump. The following measurements are performed:

- Length of each ramification until 1cm diameter

- vertical and horizontal diameters of each root at the beginning and at the ending of each ramification when the root is cone-shaped
- The mean of diameter at the ending of each ramification between horizontal and vertical diameter:

$$DfM = (DfH + DfV)/2$$

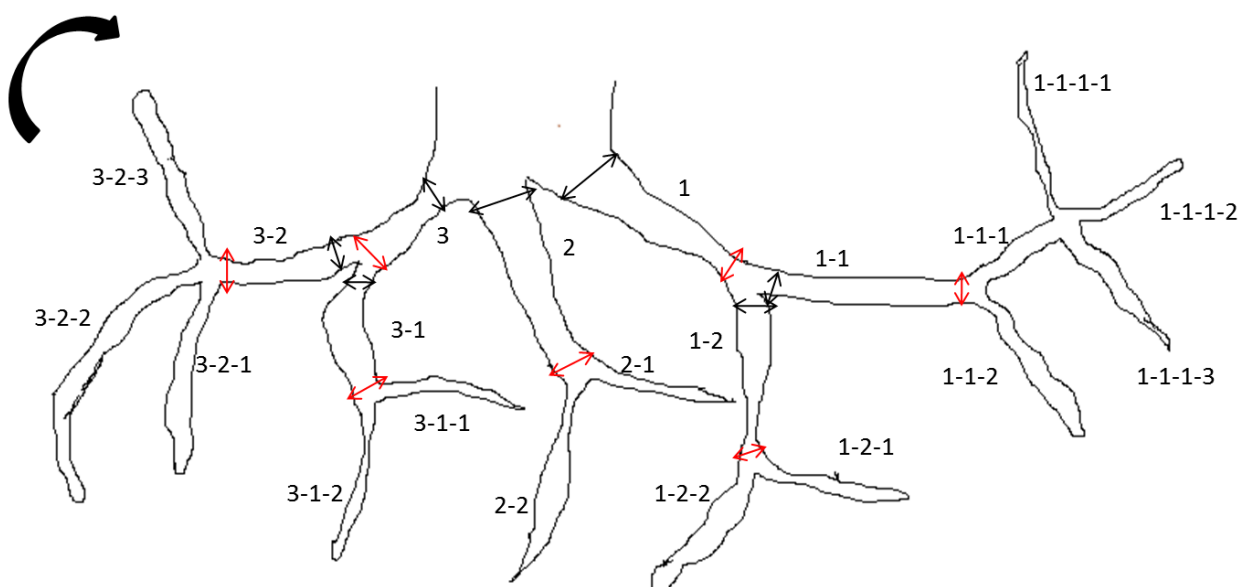
with DfM: the mean of diameter at the end of ramification; DfH: Diameter at the end of ramification horizontal and DfV: Diameter at the end of ramification vertical.

- Weight of each ramification

After measuring and weighing each ramification, some samples are collected, weighed and put inside bag for laboratory analysis.

↔ Diameter at the beginning of the ramification

↔ Diameter at the end of the ramification



**Figure 5: Process of root ramification measurement with the voronoi method**

### 2.3.2. Method of pits

#### a. Description of the method

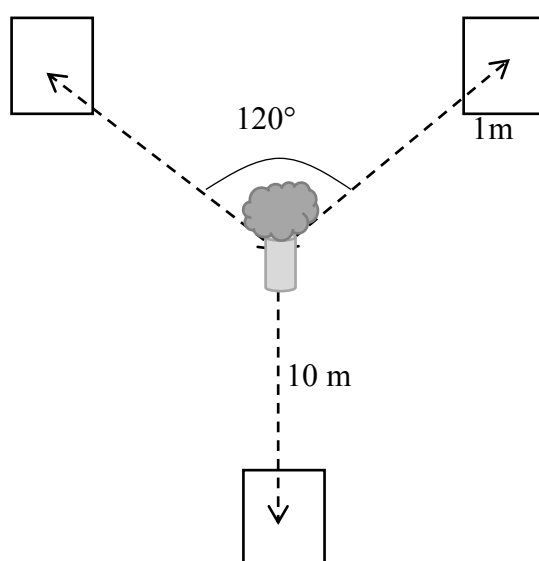
Another method of estimation the root biomass around the target trees was also tested. It consisted in excavating all roots in three pits around the target (and with no tree) and was used in some references



that we assumed not appropriate for humid tropical forest like herein (underestimate a lot BGB quantity).

#### b. Experimental design

After choosing the sampled tree, three pits of 1m<sup>2</sup> corresponding to the three vertices of an equilateral triangle (with the central point which is the sampled tree) were delimited. Distance between the center of the triangle and each apex was 10 m and the angle between the two vertices from the center of the triangle is 120 ° (Figure 6).



**Figure 6: Design sampling of root survey with the method of pits**

#### c. Collection of root data

The same general activities as collection data with the voronoï method were done in pits method. With consideration of the three depths, roots were carefully excavated from each pit, using hand tools and sorted by their diameter into the three categories of roots (FR, MR and CR). Then, roots were washed, weighed and samples were collected for lab works.

## 2.4. Laboratory works

At the laboratory, the samples were dried in an oven stove at 75°C and weighted frequently with precision scale until the stabilization of the weight. Generally, the process took around 48 hours for fine and medium roots, and could be more than a week to reach constant weight of coarse roots and stump.

## 2.5. Calculation of root biomass at tree level

The oven-dry process in the laboratory provided dry weights for all root biomass samples. For each method, these dry weights were reported on the total weight which were recorded in the field in order to get the total dry weight of all root system of each target tree.

For the voronoï method, the area of surveyed triangles and the whole polygon were considered in the calculation. These data of root biomass at tree level were used afterwad to develop allometry equations in order to have the root biomass at stand level (data processing on going).

## 2.6. Aboveground biomass measurement by direct method

Studies about AGB and BGB were conducted in parallel since trees were already slaughtered. For that, after choosing the target tree, fresh weights of AGB components (trunk, leaves and branches) were directly measured (Picard *et al.*, 2012). The collection of data concerning the AGB measurement was summarized in the figure 7.

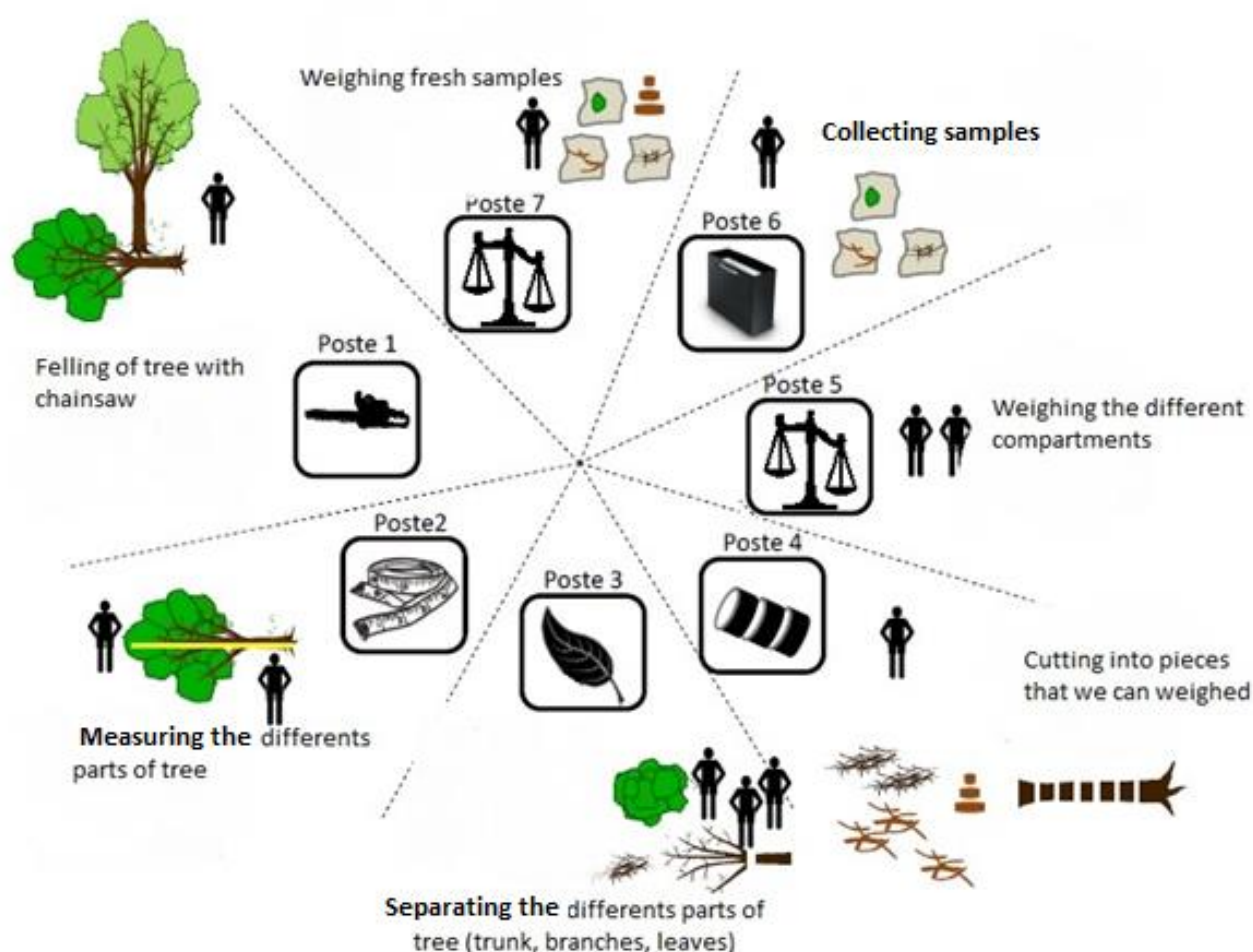


Figure 7: Process in the field for data collection of aboveground biomass components

### 3. Conclusion

All the obtained data were in process for publication of development of allometric models for root biomass and above ground biomass. Furthermore, the knowledge of carbon stock in this belowground pool is relevant for ensuring sustainability of ecosystem services provided by humid Malagasy tropical forest; this, a part from the continuity of Reducing Emissions from Deforestation and Degradation (REDD+) projects in rainforest of Madagascar that were launched in the Eastern region, like those of Ankeniheny-Zahamena Forest Corridor (CAZ).

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