"NIRA UPGRADING"

DEVELOPING THE USE OF NIRA FACILITIES FOR ROUTINE ANALYSIS OF EUCALYPTUS WOOD QUALITY TRAITS

Mission report

2007 May 25 – 31

Gilles Chaix
Philippe Vigneron

ASIA PULP & PAPER CO. - SINAR MAS GROUP - PT. ARARA ABADI & PT. WIRAKARYA SAKTI

CENTRE DE COOPERATION INTERNATIONALE EN RECHERCHE AGRONOMIQUE POUR LE DEVELOPPEMENT – BIOS DEPARTMENT

JUNE 2007

"NIRA UPGRADING"

DEVELOPING THE USE OF NIRA FACILITIES FOR ROUTINE ANALYSIS OF EUCALYPTUS WOOD QUALITY TRAITS

Draft Mission report

Gilles Chaix, Philippe Vigneron

The objective of this project is to develop the use of NIRA facilities for routine analysis of eucalyptus wood quality traits in the framework of general tree breeding research program and to validate calibration by studying the trait variability within the *Eucalyptus pellita* base population.

Problems and issues

Inclusion of wood quality traits in *Eucalyptus* selection and breeding strategies is becoming an important goal whatever the end uses are. Chemical, physical or anatomical characteristics affect both paper, sawn wood and charcoal yield and quality but the complex trait-trait relationships and the effect of growth are still poorly known. Choice of relevant characters for breeding, selection efficiency and genetic gain heavily rely to heritability values and magnitude of phenotypic variation within the breeding population as well as to genetic correlation between traits. Recent development of cost effective tools such as near infrared spectroscopy allow multi-trait assessment of large populations needed to provide genetic parameter estimations.

The general objectives of the current project entitled "NIRA UPGRADING" DEVELOPING THE USE OF NIRA FACILITIES FOR ROUTINE ANALYSIS OF EUCALYPTUS WOOD QUALITY TRAITS, jointly implemented by Arara Abadi Research Development Centre (Perawang) and CIRAD are to strengthen the capacity of AA RD in routinely using NIRS for effective inclusion of wood quality traits in Eucalyptus breeding and selection programme. The project focuses on capacity building and developing specific calibration equations for *Eucalyptus pellita*.

Objectives

1) to develop the use of NIRA facilities
2) to propose a routine analysis of eucalyptus wood quality traits in the framework of general tree breeding research program
3) to validate calibration by studying the trait variability within the *E. pellita* base population

Deliverables

1) Wood sampling strategy, collection and preservation in order to establish a reference collection for calibration
2) Advices on reference analysis methods (wet chemistry) in order to reduce experimental error
3) Training for one (possibly 2) AA scientist(s) on the field of hardware maintenance, spectral acquisition, spectral data treatment, calibration and validation procedures
4) Calibration equations for several wood and pulp properties: wood density, lignin and cellulose content, Kappa number, pulp yield, total extractive content

Work description

Relevant wood quality traits: basic density, lignin and cellulose content, extractives, pulp yield, Kappa number
Species: the project will focus on *Eucalyptus pellita*.
Sampling strategies:
Phenotypic variability - sampling the entire range of concentration of the constituents of interest anticipated in future samples to be predicted by the calibration
Intra tree variability - sampling the tree or the variety in order to represent or predict the actual average value of this tree or this variety
Accuracy of reference analysis: precision of wood trait measurement of references. Standard error and repeatability of measurement will be analysed prior to equation building.
Calibrations: 150-300 samples should represent the full range of values. A first sample will be defined according to the existing knowledge.
A portion of unknown samples must routinely be analysed by the reference methods to ensure that calibrations remain reliable. It may be necessary to update calibrations several times during the initial phases of use to incorporate "outlying" samples, until the calibrations become highly robust.
Complementary study implemented to apply and validate calibration equations:
Natural variability of the major traits within a representative sample of the *Eucalyptus pellita* base population will be studied in order to:
i) Validate calibration equations,
ii) Describe the phenotypic variability of wood quality traits within the base population, which will be used for further breeding.

Time schedule

First CIRAD mission: review of reference methods accuracy, technical training on wood sample collection and processing
AA: first wood sample collection for calibration, chemical analysis
France: 45-days AA scientist training (spectral acquisition, data treatment, calibration and validation procedures), first calibrations
AA: Extension of wood sample collection and calibrations
- Second CIRAD mission: validation of calibrations and data acquisition procedures.

	month											
	1	2	3	4	5	6	7	8	9	10	11	12
first cirad mission	X											
training in France				X	X							
Lab data acquisition	X	X	X	X	X	X	X	X	X	X	X	
Final mission												X

Key Performance Indicators

1/ Month 1: Cirad mission report on reference methods, sampling strategies for wood collection

2/ Month 5-6: a first set of infra red spectra will be collected and analyzed in relation with wood properties data provided by APP laboratory.
3/ Month 6-7: Training completed with delivery of training notes to support subsequent internal SMF staff training in NIRA operation.
4/ Month 12: final calibration equations are provided and ready to use for routine assessment of traits from which data were provided by APP
5/ Month 14: First appraisal of the range of wood properties variation is available. Guidelines for including wood properties in selection/breeding program of Eucalypts are provided.
6/ Month 14: Handbook for improvement of calibration equation quality is provided as basis for staff development and training.

Contents

Tables and figures lists

Acknowledgements

The mission benefited from deep and rich discussions with the different staffs involved in wood technology laboratory and tree breeding programme. Arara Abadi Research & Development Centre provided all the requested data and information for the implementation of the mission.

The consultants acknowledge all the people for their warm welcome and their large involvement in the success of the mission. In particular, deep, open and informative discussions were conducted with:

The wood laboratory staff and especially:

Mr Ruspandi, Mr Sabki and Ms Rosmithayani Wood technology section

The genetic and breeding program staff:

Dr Shen Tse An, Deputy Director Forestry, R&D, who organized and facilitated the mission, and with whom we had continuous exchanges of ideas and points of view all along the week.

MMr. A. BamBang, Wahyu Edi and their collaborators, Genetic Improvement programme

The mission was greatly facilitated by:

Miss Evi Eriana

Prof. Zhu Guang-Lian, head of R&D

This report was prepared by the forestry department of CIRAD for the account of Asia Pulp & Paper Co. (APP), the Sinar Mas Group (SMG), PT. Arara Abadi. It reflects CIRAD judgment in light of the information available to it at the time of preparation of this report.

Table 1. Other people involved during the mission

Name	Team
Heri Suryanto	Breeding
Arief Dariyanto	Acacia Breeding
Aqunq Estu W.	Acacia Breeding
Nurul Elmi Faid	Eucalyptus Breeding
Yago A. Nugralio	Nursery
Sari Yunam	Nursery
Anita Dewi M.	Nursery

1 EXECUTIVE SUMARY

The objective of this project is to develop the use of NIRA facilities for routine analysis of eucalyptus wood quality traits in the framework of general tree breeding research program and to validate calibration by studying the trait variability within the *E. pellita* base population.

The general objectives are to strengthen the capacity of AA RD in routinely using NIRS for effective inclusion of wood quality traits in Eucalyptus breeding and selection programme. The project focuses on capacity building and developing specific calibration equations for *Eucalyptus pellita*.

The main objectives of this mission are to:

- discuss the major traits to be studied and define a priority
- survey the accuracy of reference chemical analysis done in AA laboratory
- establish a detailed work plan for the project implementation
- define a detailed sampling strategy for calibration and validation set of tree
- plan the training period in Cirad laboratory.

The proposals are based on current data checking, commented laboratory and facilities visits, and intensive discussions between the experts and wood technology and breeding team.

Proposals contained in this report encompass five major points:

- the relevant wood quality traits for *Eucalyptus pellita,*
- the accuracy of reference analysis,
- the calibration plan,
- the sampling strategies for calibration set,
- the training in France.

The proposed work plan roughly includes **2400** laboratory analyses and **4000** spectral acquisitions and analysis in AA as well as in CIRAD laboratories. This unprecedented effort will lead to robust and powerful tools for implementation of tree breeding strategy.

Relevant wood properties

The first set of traits to be predicted is as follow: basic density, extractives, lignin and cellulose contents, pulp yield. A second priority set will be: kappa number, fiber length and coarseness.

Accuracy of reference analysis and Nir equipment

Chain processes from wood sample to spectral acquisition, through chemical and physical assessments were surveyed. Wood storage condition was discussed. An experimental protocol is proposed in order to measure the standard error of laboratory and determine the confidence of reference values.

Calibration plan

Existing AA calibration equations will be completed by adding available data which were not included in the previous equations. The same set of wood sample will be used to build new calibration in Cirad laboratory in order to cross-validate actual equations and establish cross-reference calibrations.

Considering the expected wide range of trait variation within the population to be predicted in the future, a novel sample collection is planned. Final sampling will include a pre-sampling step (300 trees) on which indirect survey of wood properties will be done with AA and CIRAD actual calibrations. Principal component analysis will allow selecting a final sample (150 trees) on which reference analysis will be done.

Sampling strategies for calibration set

We proposed a wood sampling collection for calibration/prediction set corresponding to 150 trees from the felling to the collection of different types of sample (solid wood, chips, sticks, drill, core, powder). All properties of these 150 trees will be measured **before** the training period in France. The sample fluxes (encompassing 1050 individual wood lots) are precisely described.

Training program in CIRAD laboratory

The wood technologist Nirs specialist will be trained in France during 6 weeks. The training program is based on chemometric methods, Nira management, spectra acquisition with APP wood samples sent to Cirad, and spectra analysis for calibration and prediction.

Time table of operation

Summary of time table is presented table 2 (see details in the text).

Table 2. Simplified schedule proposal (the complete form is presented in the end of this document Table 7)

	2007								2008				
	May	June	July	August	September	October	November	December	January	February	March	April	May
First mission for Nira Upgrading													
Misson in Indonesia (Vigneron and Chaix)													
Performing previous data for EP													
Check the 300 sample set (Powder and chips) and the spectra data													
Nir acquisition on powder for 120 samples remaining and send samples to Cirad													
Calibration reviewed by Cirad and APP (BD, PY, Extratives, Lignin, Cellulose)													
Calibration set selection													
Collecting 40 gr drill samples (4 holes minimum 4 direction) on 300 trees													
Nir acquisition on 300 drills drying samples 20 gr													
Send to Cirad 300 drill samples, 5 gr per samples (1)													
Grounding 300 drills 15 gr													
Nir acquisition on 300 grounded samples and send samples to Cirad													
Cirad will send selected tree list of 150 trees + 20 trees to replace if necessary													
Wood sampling collection for calibration set													
Felling the 150 trees and collecting samples													
Preparing chips, sticks, grounded wood for wet chemistery													
Nir acquisition on chips, sticks, ground wood													
Separating powder for chemical analysis (35 > x > 180 mesh)													
Nir aquisition powder for chemical analysis (35 > x > 180 mesh)													
Extractives contents and Nir aquisition on fre extractive powder													
Lignin, cellulose, pulp yield, Basic density on disk and on chips													
Send to Cirad chips, drill, powder, powder for chemical analysis													
Fiber lentgh and Coarseness mearsurments and send to Cirad													
Repeatability of reference values													
Preparing sample (for basic density, pulp yield need 2 trees to fell)													
Nir acquisition on chips, sticks, ground wood													
Separating powder for chemical analysis (35 > x > 180 mesh)													
Extractive contents													
Nir aquisition on free extrative powder													
Lignin, cellulose, pulp yield, Basic density on disk and on chips													
Training in France													
Preparing sample in Indonesia													
Nir acquisition on solid wood, drill, chip samples													
Nir aquisition on powder for chemical analysis (35 > x > 180 mesh)													
Calibration equations (BD, PY, Extratives, Lignin, Cellulose, FL and Coars.)													
Calibration In routine													
First test of prediction in routine													
Outliers analysis (chemical and spectrum) and include in calibrations													
Second mission for Nira Upgrading													
Control of test in routine, perform the calibration equations in Cirad and APP													
First including wood properties in selection/breeding program of Eucalypts													
Handbook for improvement of calibration equation quality													

2 INTRODUCTION

Near-infrared spectroscopy has been recognized as a powerful analytical technique for rapid determination of various constituents in many agricultural and raw materials. The approach involves the acquisition of a reflectance spectrum after near-infrared radiation on ground or solid wood sample in a specific material plane (RT). The resulting NIR spectral information is then calibrated against standard analyses obtained using conventional analytical techniques (standard method) using linear regression.

Near infrared spectroscopy (NIRS) is used more and more in several fields where chemical analyses (usually costly and time consuming) are needed:

- food (quality control, chemical analysis, ...)
- agriculture (protein, sugar, ...)
- pharmaceutical (Control quality raw material and tablet products, ...)
- chemical industries (petroleum, polymer, ...)
- pulp and paper industries (first publication in 1988)

NIRS is a fast technique for qualitative and quantitative analyses of organic materials compare to chemical analysis. NIRS is a non-destructive spectral acquisition and requires a minimal sample preparation. NIRS is an ecologically-friendly methodology (no chemicals products) and less costly, compare to chemical analysis after investment of apparatus.

As described in the Figure 1, the Nirs tool should be used at different steps from the raw material characterization (chemical and/or physical wood properties) to the end product (pulp yield, kappa number).

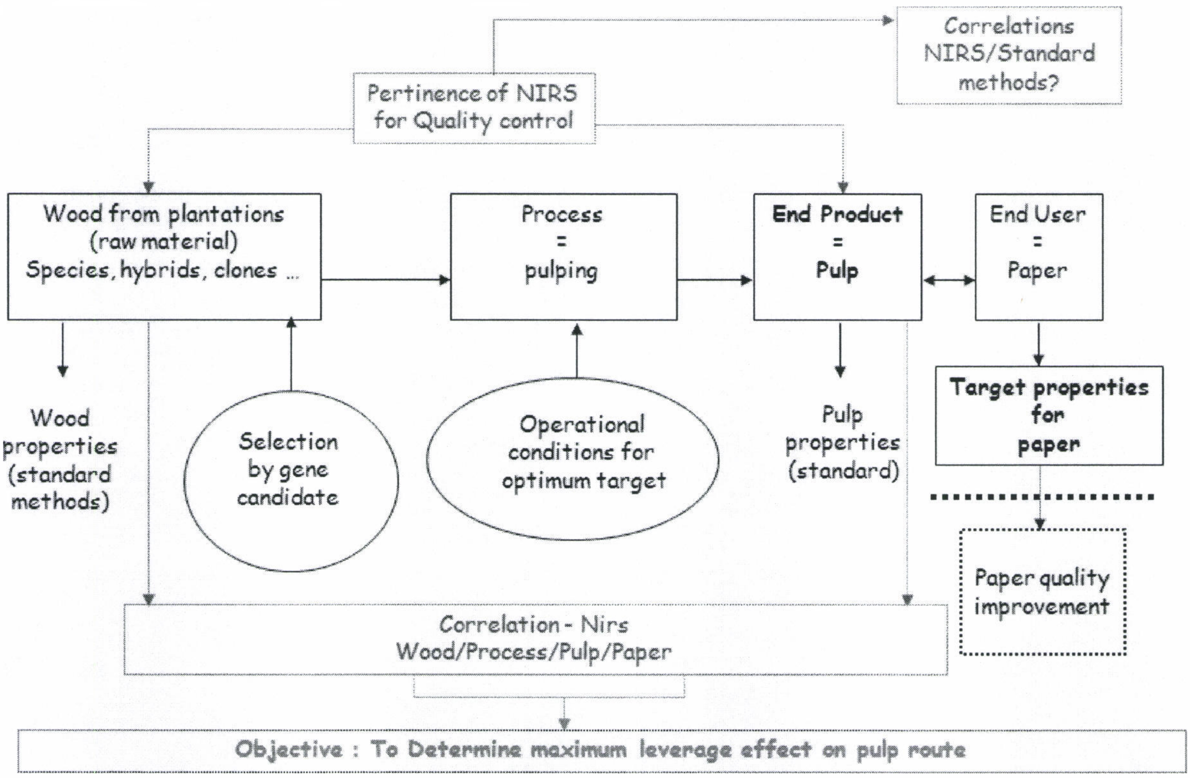


Figure 1. Nirs inputs during the paper production process

The calibration/prediction processes are based on a calibration set which is analysed both for wood properties and spectral data in term of absorbance. Theses two types of data were analysed by multiple regressions to build a prediction equation. Then this equation is used for the prediction of new samples. The calibration/prediction processes are described by the Figure 2.

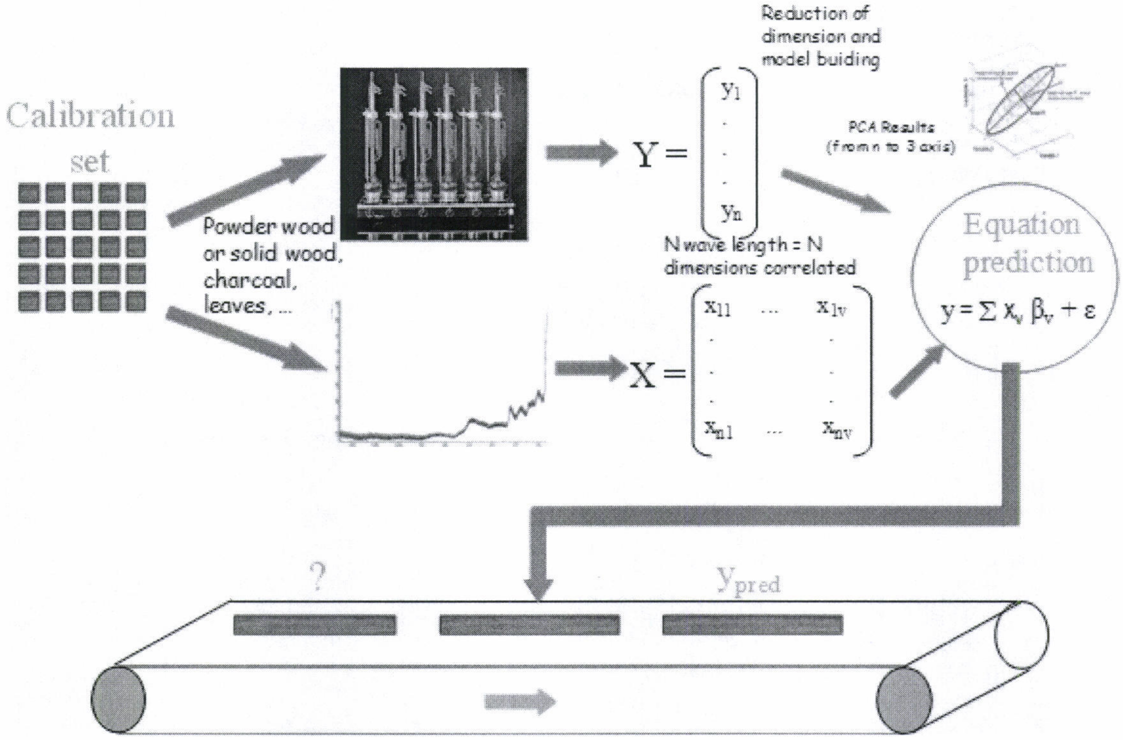


Figure 2. Calibration/prediction processes

NIRS approach is based on reference values and reduces the quantity of analysis and requires mathematical process and a rigorous step by step control (from samples, reference value control to spectra analysis) and of course requires a training of wood technologist. The samples have been analyzed as accurately and precisely as conventional techniques allow. Then standard methods and NIRS method will be compared in order to evaluate the potential of NIRS.

3 WORK PLAN

The general work plan presented in Figure 3 describes the work from the breeding population to the equation prediction building. The proposed method is based on a first set of 300 trees selected according to age, location, growth rate and soils type. Among them 150 trees are selected based on drill Nir information by Principal Components Analysis treatment or based on their predicted values obtained from the existing prediction equations. Wood properties are measured and one part (100) is used for the prediction equation building and the other part (50) for the validation of prediction equation. After that and during the prediction process, some samples could appear as outliers. These trees will be chemically assessed and included in the prediction set.

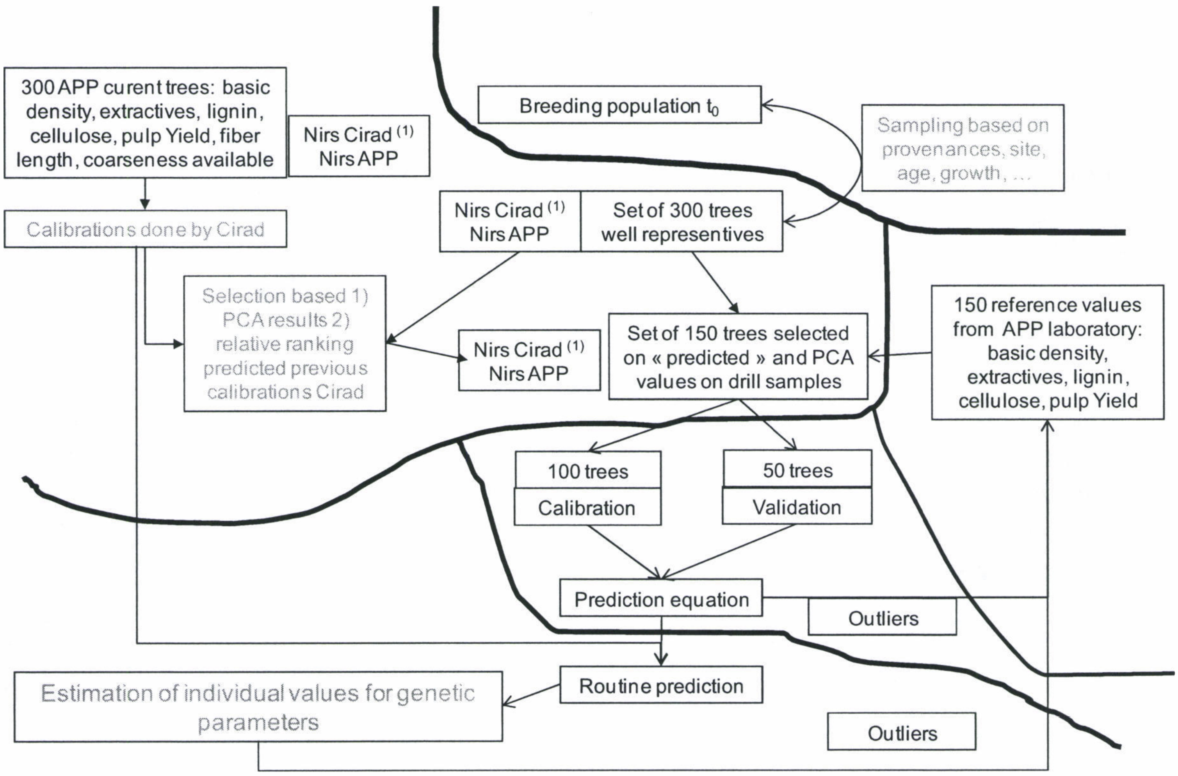


Figure 3. Work plan activities

4 RELEVANT WOOD QUALITY TRAITS FOR *EUCALYPTUS PELLITA*

According to the discussion held during the mission, the first set of traits to be predicted is as follow: basic density, extractives, lignin and cellulose contents, pulp yield. A second priority set will be: kappa number, fiber length and coarseness.

5 ACCURACY OF REFERENCE ANALYSIS AND NIR EQUIPMENT

5.1 CHECKING CHEMICAL ANALYSIS, NIR EQUIPMENT, AND TYPE OF SAMPLES

The determination of wood extractives content is carried out using Soxhlet extractors, in accordance with standard Tappi T204 cm-97 (ethanol-benzene 1:2 and acetone extraction) and Tappi T280 pm-99 (acetone extraction).

Residual lignin was determined as insoluble Klason lignin content following the standard procedure (TAPPI method T222 om-98). In this method, the polysaccharides (cellulose and polyoses) are hydrolyzed by sulfuric acid solution, the acid-insoluble lignin is filtered off, dried and weighed.

Celluloses is determined according to TAPPI Test method T203. An air-dried sample was weighed to an equal of 1.50 g of oven-dried sample to the nearest 0.10 mg. The pulp sample was extracted consecutively with 17.5 % and 9.45 % NaOH solutions at 25 °C. The soluble fraction, consisting of beta- and gamma-celluloses, is determined volumetrically by oxidation with potassium dichromate, and the alphacellulose, is the insoluble fraction, derived by difference.

Basic density measured by difference between anhydrous weight and saturated weight and volume displacement is based on two types of sample: chips and solid disk.

5.2 NIRA TESTS

Spectral acquisition: several type of sample showed the perfect functioning of the Nir Apparatus. But, the check cell, which is used as permanent control, is lost and APP R&D Department should buy one. Considering the high cost of such check cell, a discount price could probably be obtained through Cirad contact with FOSS Company.

An existing calibration based on 180 samples is currently used in AA laboratory (Table 3). Nevertheless, the calibration sample is suspected to display a quite narrow variability, the trees being CPTs, and then inappropriate in order to predict out of range values.

This existing calibration seems to provide acceptable results in term of efficiency (R^2 close to 0.8) except for cellulose content, but was never validate with an appropriate validation set.

Table 3. Description of trees concerning the existing calibration

Type of tree	Location	Age (year)	Number of tree
CPTS	Jambi	3.3-5	59
CPTS	Parawang	2.3-6	44
Clone EP05	-	2-5	18
Seedling	Malako	13.5	10
CPTS	Jambi	5	15
Clone EP05	Tapung	5	35

5.3 SAMPLE STORAGE CONDITIONS

Both for the spectrophotometer efficiency stability, sample conservation, repeatability, calibration and prediction quality, it's compulsory to maintain the apparatus and the wood samples at constant temperature and humidity conditions. The goal is to maintain the humidity of the wood sample at the same humidity during NIRS and prediction process because water absorbance bands affect the wood specific absorbance. APP could dedicate the Nirs room for NIRS activity and long term storage of calibration sets collection which will be used as permanent control. The samples to be predicted will be stored in the same room at least one week before spectral acquisition in order to stabilize temperature and water content.

5.4 Identifying the chemical and physical laboratory capacities

Each sample is analyzed two times so the capabilities are 3 samples per day during 5 days per week, 15 samples per week for extractives, lignin and cellulose contents, basic density and pulp yield. The R&D APP laboratory capacities should be sufficient for the routine analysis. For the genetic analysis which need data on several hundreds or several thousands of samples these capacities are far from sufficient to provide the data to breeders in due time.

5.5 Standard error of laboratory

The precision of prediction equation heavily relies on quality of wood trait assessment.

Quality of assessment depends on the precision of the apparatus used and on the repeatability. In order to estimate the repeatability (i.e. the consistency of the result over several assessments of the same trait with the same sample) we propose to implement the following:

- two different samples
- 10 analyses for each of them (for all the traits) using routinely used methods.

The methodology is presented in the Figure 4.

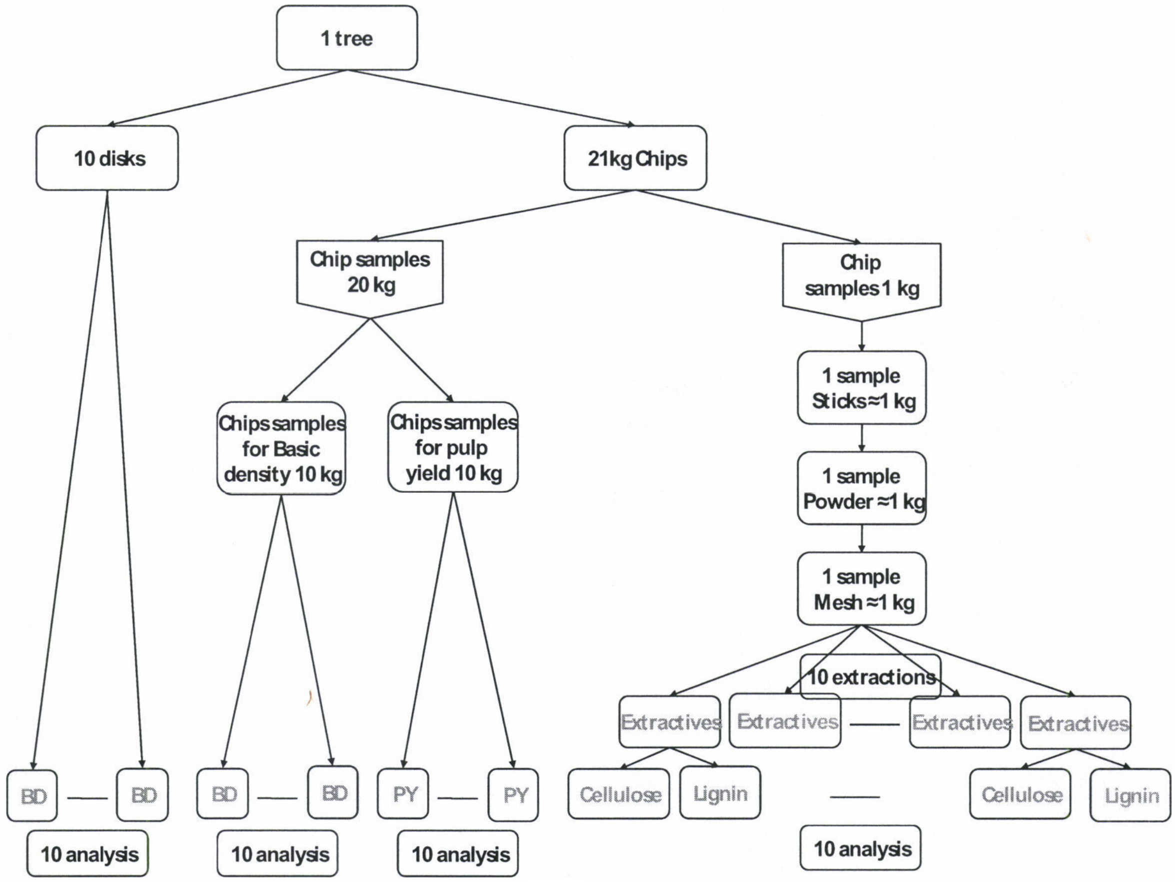


Figure 4. Methodology proposed for standard error of laboratory estimation

5.6 SAMPLING WOOD STRATEGY (SEE § 7.1)

The intra-tree variability conduct to collect three logs along the bole. For each we propose to prepare two type of sample regrouped by destructive samples and non destructives samples (Figure 5). The first group concerns the solid wood part for chips (chemical properties, pulp yield, and basic density), solid wood for radial variability studies and a disk for collection. The second group concerns the drill and core samples.

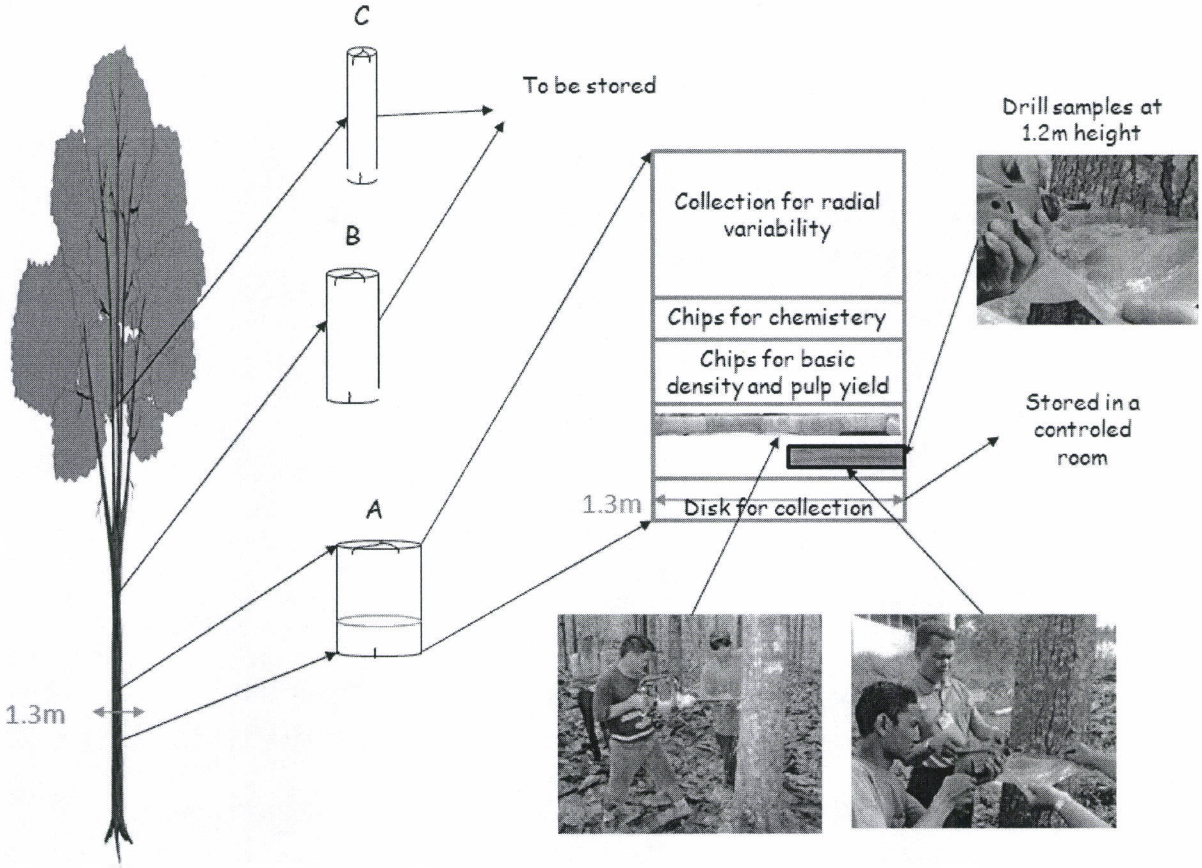


Figure 5. Sampling wood proposed for the calibration set

6 CALIBRATION PLAN

6.1 USE OF EXISTING CALIBRATION EQUATION

The wood technology laboratory has already developed different calibration equations based on previous sample sets (181 wood samples, see 5.2). 119 additional samples with chemical analysis are available. New spectral acquisition will be done for these 300 wood samples using the newly reappeared FOSS apparatus. A new calibration equation will be established (Table 3).

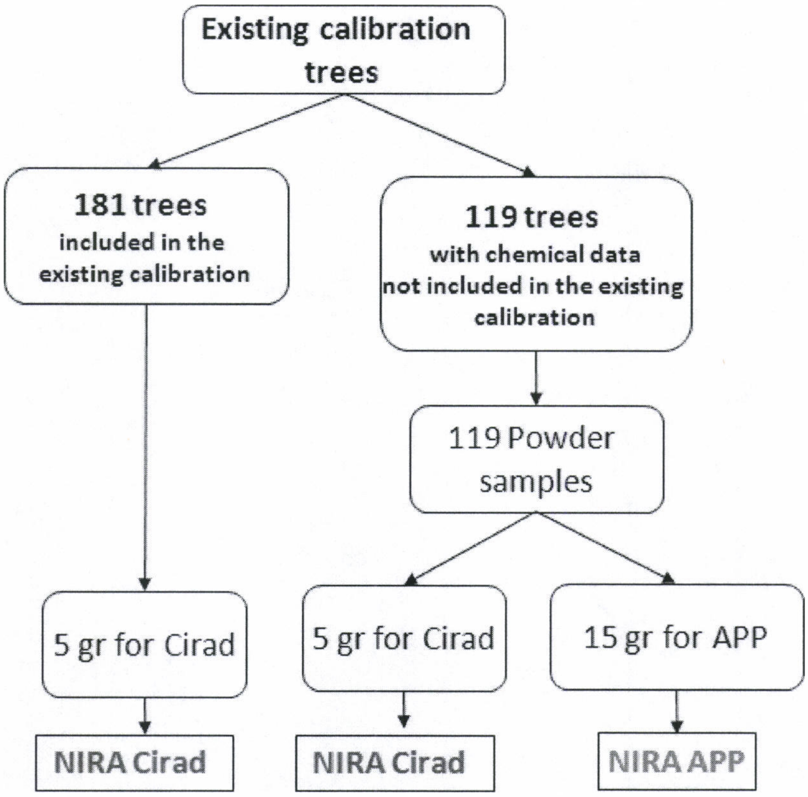


Figure 6. Description of work concerning existing calibration set

At the same time, the wood powder (5 gr per sample) will be send to CIRAD in order to develop calibration with another apparatus and to cross checking the two calibrations. These calibration equations will be used in order to obtain a first prediction of the new sample set (see 6.3).

6.2 SELECTION OF A NEW CALIBRATION SET

As for any prediction based on regression methods, the highest precision is obtained within the range of variation covered by the calibration set, in other words, the set of data used to establish the regression model has to fully cover the expected range of concentration of the constituents of interest anticipated in future samples to be predicted. Genotypic, physiologic and environmental effects on wood properties, even if poorly known, have long been recognized. Age to age variations and the transition from juvenile to more mature wood is a major source of both within and between tree wood variability. It implies that sampling strategy needs to consider several factors such as age, location (i.e. mineral and peat land), genotype (provenances or progenies).

As described in figure 1, the final sample of 150 trees will be selected as follow:

- Initial sample of 300 trees a priori selected upon age, location and genotype
- Initial prediction of tree performances (for wood quality traits of interest) using previous available NIRA calibrations (from CIRAD as well as from AA)
- Choose of 150 trees based on a principal components analysis (PCA) of the 300 (time the number of traits) predicted values.

Chemical analysis will be performed on these 150 wood samples and used for calibration (100 trees) and validation (50 trees). The resulting calibration will be used for final prediction of the 300 initial trees.

6.2.1 PRE SAMPLING OF 300 TREES.

The objective is to build calibration equation for several wood quality traits (lignin, cellulose, pulp yield, basic density for a first run) which are usable for selection, i.e. with special focus on 3-5 years old. Nevertheless, in order to build robust equations usable for a larger range of ages, some younger and older trees will be sampled.

The 300 individuals will be sampled from:

<u>Before 3 or 3.5 years old: 50 to 60 trees</u>
- ± 35 *Eucalyptus pellita* seedlings in a commercial plot planted in 2004 on mineral site
- ± 20 *Eucalyptus pellita* seedlings planted on peat land, 2.5 to 3 years old

- ± 5 trees from EP05 at age 3 if possible from mineral and peat.

<u>From 3 to 5 years old: ± 200 trees</u>
- Full sib progeny trial EP03L, planted July 2003, 24 pure *E. pellita* FS families + EP05 clone,

The ortet selection will be done this year (2007). Sampling will be done in 1 replicate in order to keep the trial usable. According to the survival rates, 5 to 10 trees per FS family will be sampled, in order to reach a total of 170 to 180 individuals (even more if possible). In addition, 5 individual trees from EP05 clone will be sampled in the same block. This sample will be used, after calibration, for a first analyse of wood quality traits variance components.

- Seedling Seed Orchard EP06S which will be thinned soon will provide 20 to 30 'big' trees to match the requirement of size variability. (1 tree per family).

<u>Over 5 years old: ± 50 to 60 trees</u>

- Full sib progeny trial EP03K: 5.5 years old, 13 pure *E. pellita* FS families, 2 trees/family, 5 trees for EP05 clone: 31 trees.
- EP02E, 6.5 years old: 10 trees
- EP04F, 7.5 years old: 10 trees
- 5 trees from EP05 at age 7 if possible

6.2.2 HOW TO CHOOSE THE TREES:

For each trial, each family, each age, we need to sample the full range of size variation, excepted very poor or abnormal trees (avoid unhealthy, broken, low branching, low forked trees which could display unusual wood properties with reaction wood, decays….).

This sampling will allow:
- to survey a large range of age and size: 2.5 to 7.5 years old
- to have a first survey of evolution with age for clone EP05
- to implement a first analyse of genetic and environmental variance components of wood quality traits (EP03L)

6.3 WORKPLAN FOR THE SELECTION OF CALIBRATION SET

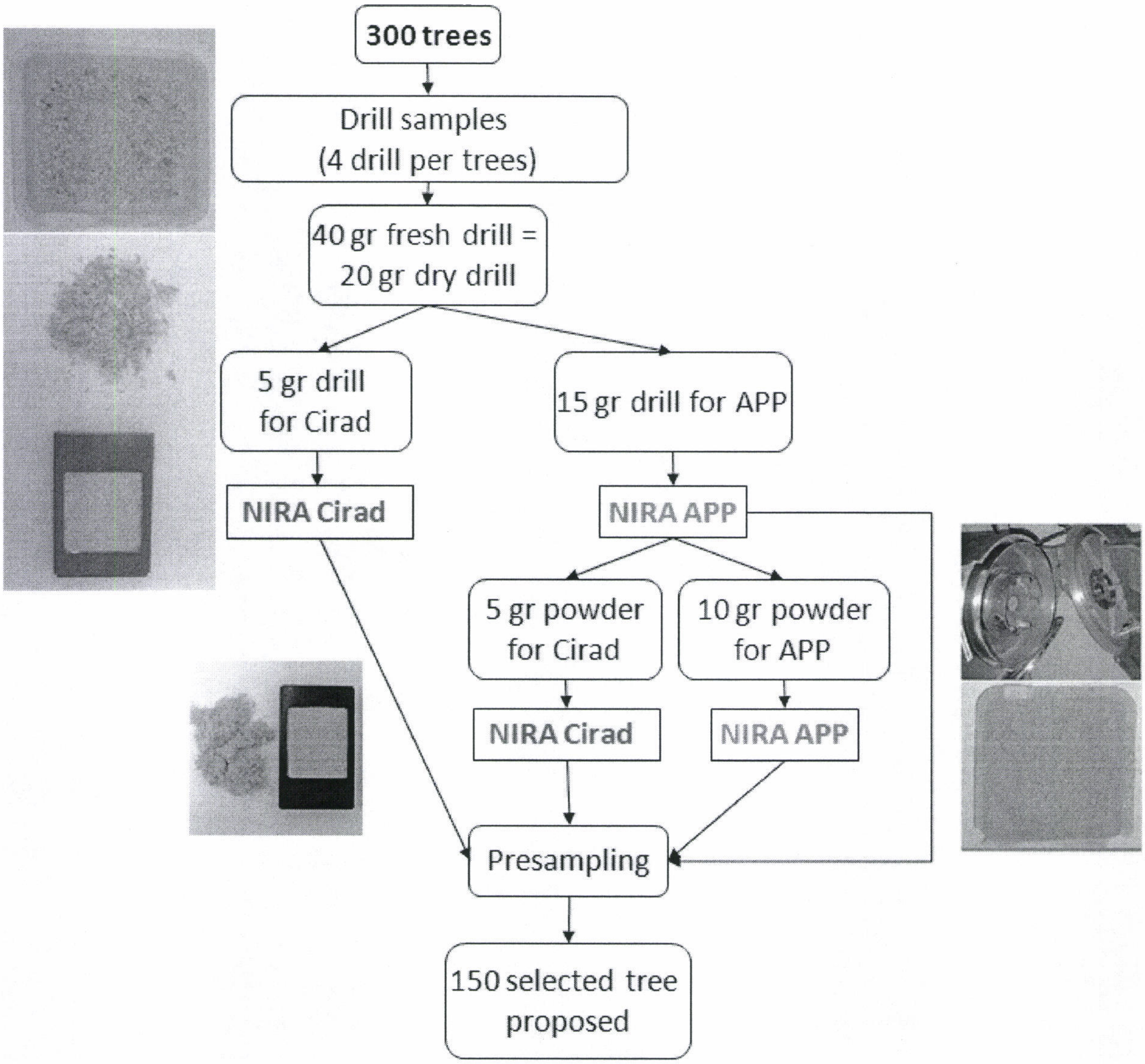


Figure 7. Work plan for calibration set selection

Collecting 40 gr drill samples (4 holes minimum 4 direction) on 300 trees

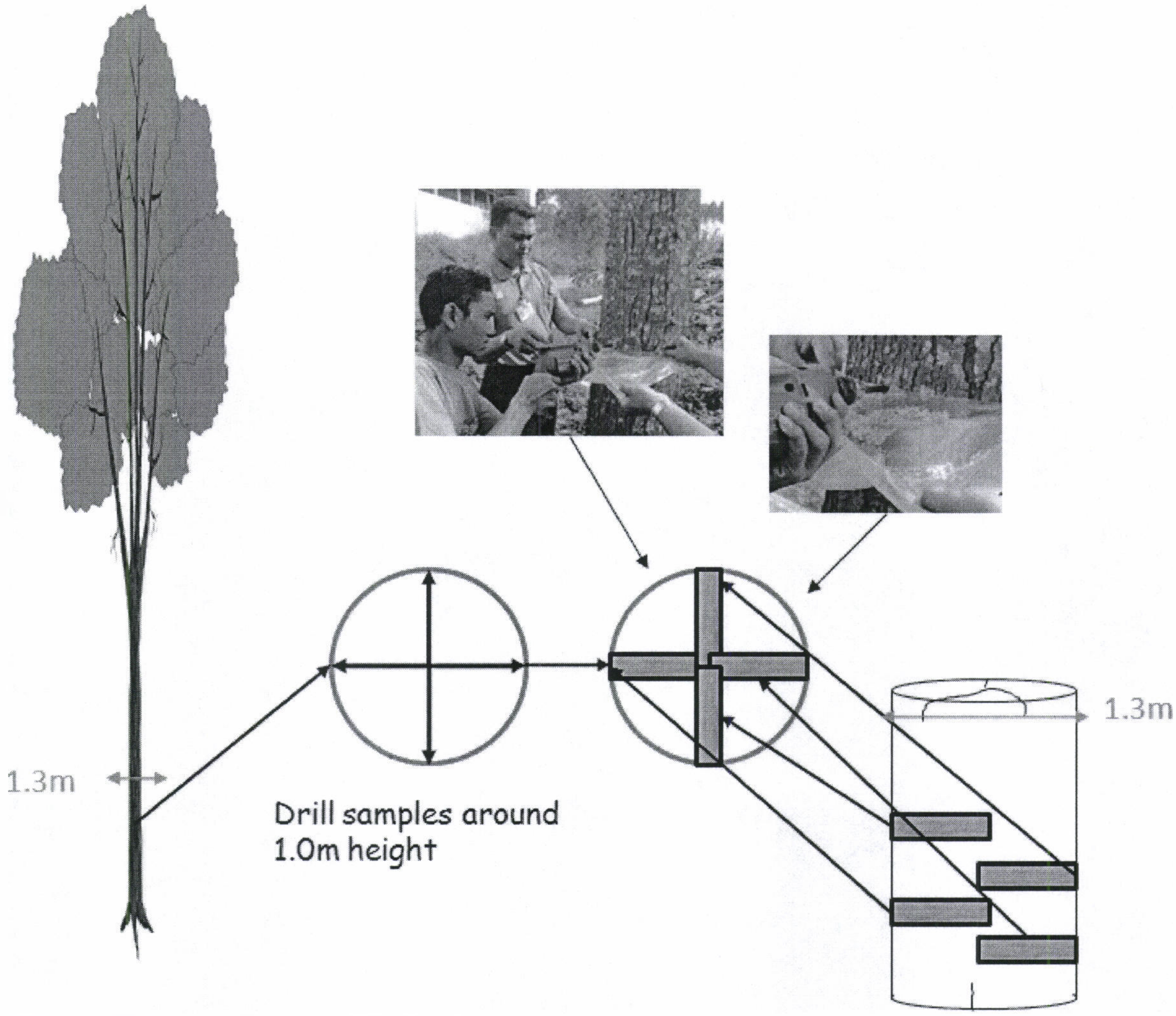


Figure 8. Drill collection proposed

As described in the Figure 8. Drill collection proposed we propose to collect only drill samples. According to the total required quality, 40 grams (20 g of dried wood) corresponding to 4 drill will be collected per tree in different direction and at different height around 1 meter height above ground.

The 300 drill samples corresponding to the 33 trees will be dried in conditioned (24h/24h) room during 1 week before the spectral acquisition to obtain 20 gr for each. Five gram per sample will be sent to Cirad, 15 gr will be grounded before spectral acquisition on APP Nira.

A set of 150 trees will be selected among the 300 first ones based on the Principal Component Analysis of the drill spectrum NIR. All types of combinations of variables should be represented from spectral point of view. The variation in all directions should be as largest as possible, but limited to the spectra region of interest. The Cirad and APP previous calibrations should be used, not for prediction but to classify the samples. This selection should be controlled in according to the previous selection criteria (age, soils, height, and diameter). This operation could be done in APP lab and Cirad lab and allows a gain of time before the complete control of APP apparatus.

Cirad will propose one list of 150 trees and one 20 trees supplementary list which will be use for the prediction equation building based on wood properties and spectral data.

7 SAMPLING STRATEGIES FOR CALIBRATION SET

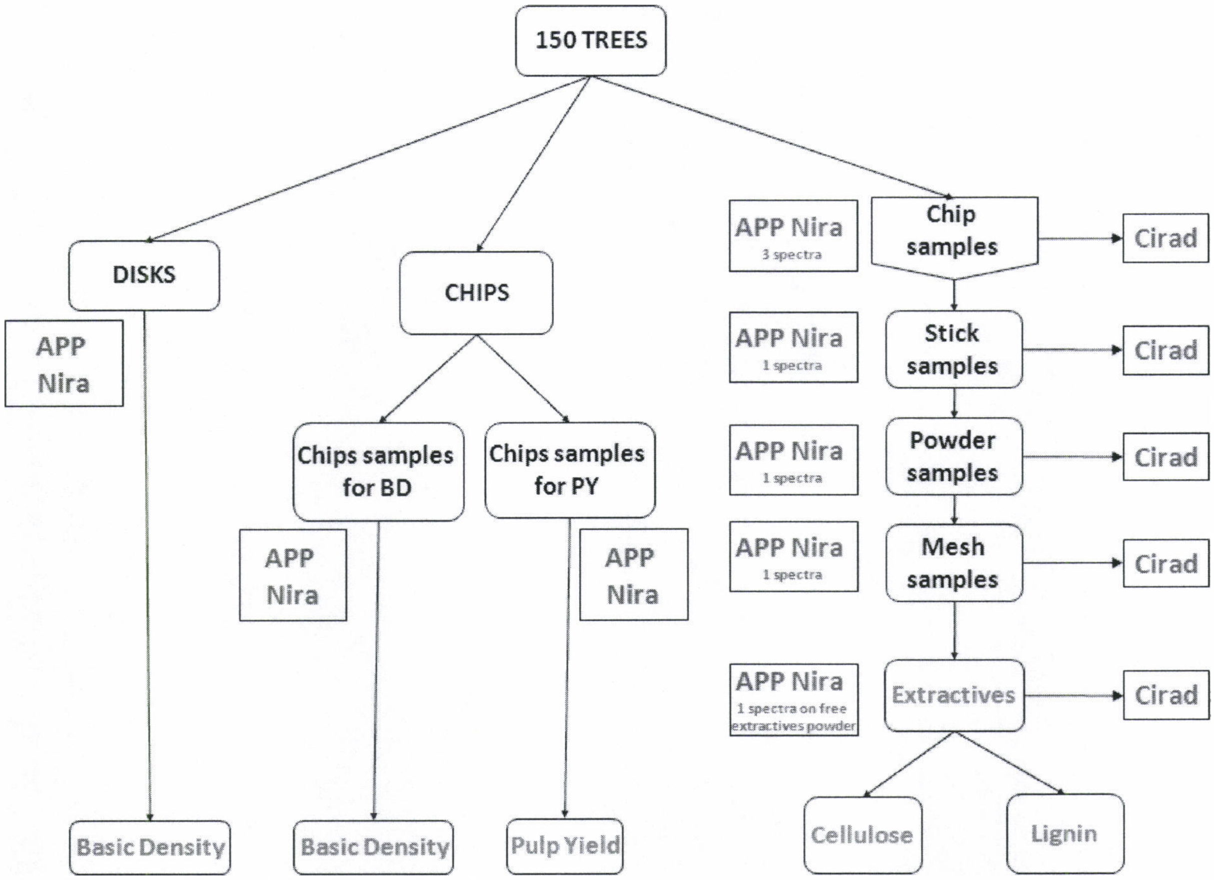


Figure 9. Global sampling strategy for calibration/prediction process

7.1 WOOD SAMPLING COLLECTION FOR CALIBRATION SET

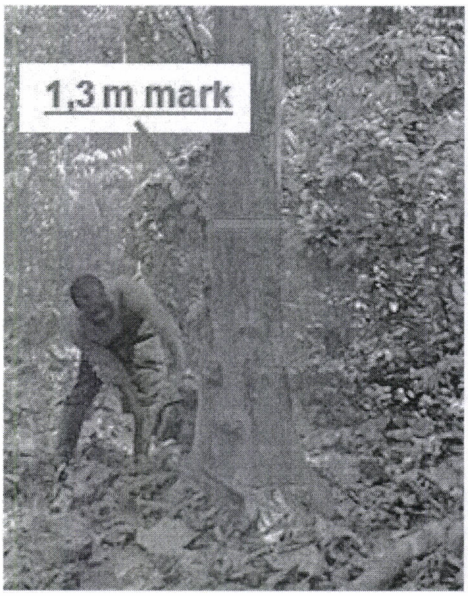
7.1.1 FELLING 150 SELECTED TREES

During felling down the 150 trees, diameter will be assessed every meter for each tree. Data will be used later to develop a general taper equation and volume table. Field operations before feeling are presented by the following point illustrated by pictures (pictures concern teak and not eucalyptus).

1- Determine north direction on standing tree

2- Mark or scratch the North direction to the highest level on standing tree

3- Measure girth at BH (1,30 m)
4- Mark the 1,30 m level on standing tree
5- Take photographs and morphology of the tree
6- Fell the tree at ground level and continue marking the north direction by means of the chainsaw (the mark must be slightly deeper than the bark depth, i.e. it must mark the wood). If the crown and the branches are cross cutted it is possible to return the stem with the help of a turning device this procedure will facilitated the North marking.


1,3 m mark

7.1.2 Collecting wood samples

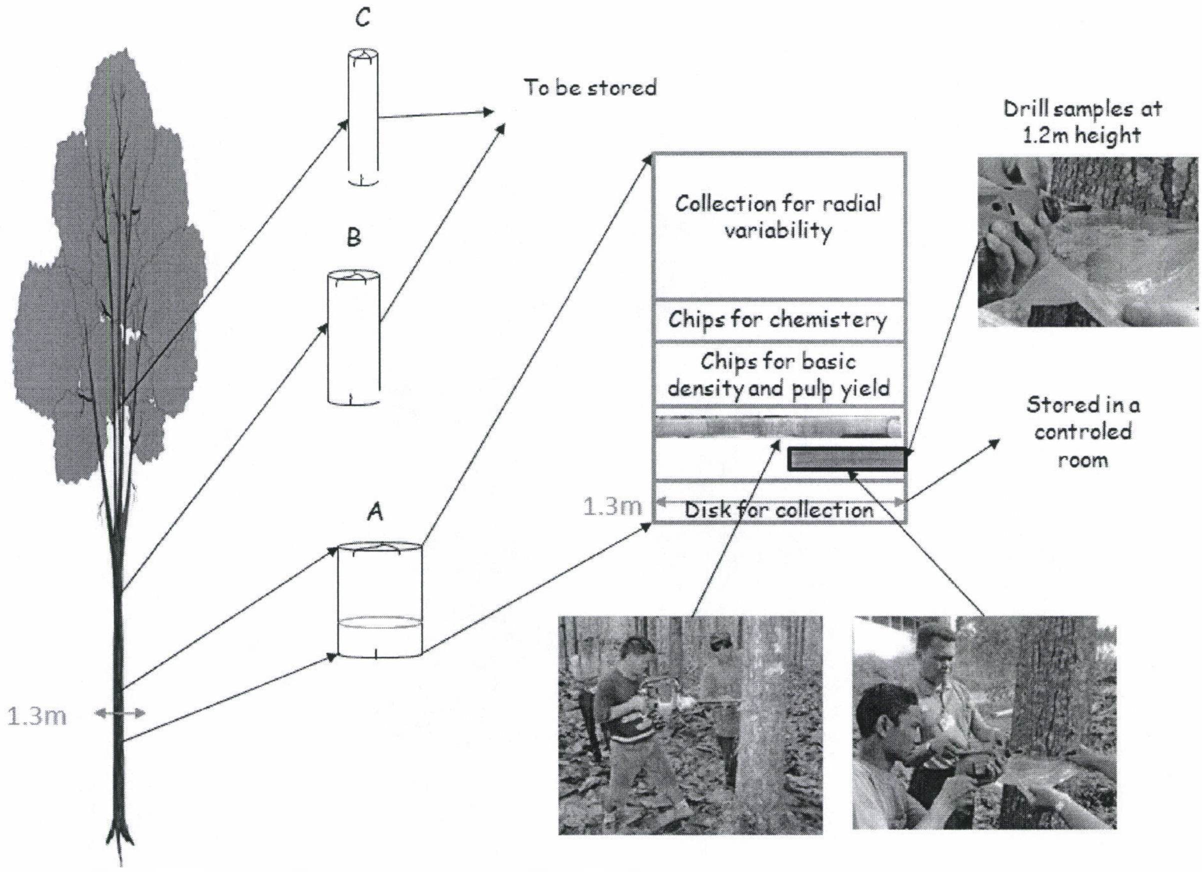


Figure 10. Wood collection process and type of samples

Trees will be cut and three logs by trees will collect (A: 1.2-2.2 m height, B: +/- 50cm around the 25% of commercial volume height, C: +/- 50cm around the 75% of commercial volume height). The length of each log depends on the diameter (Figure 10).

- The north direction will be marked on each sample
- The part A will be designed for the wood properties analysis and several samples of wood will collect:

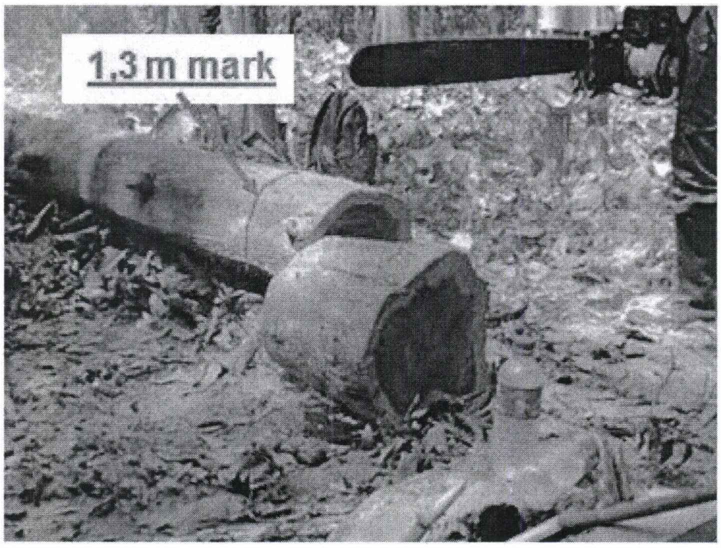
* One disk 50 mm of thickness stored in a controlled room.
    * Drill samples (4 holes) and core samples (2 cores)
    * One disk for chips (basic density and pulp yield)
* One disk for chips and sticks for wet chemistry
* One disk for further analysis included radial variability

- The part B and C will be designed for further analysis (within tree variability)

Field operations after feeling are presented by the following point illustrated by pictures.

1- Determine 1.2 m level using 1.3 m level as reference

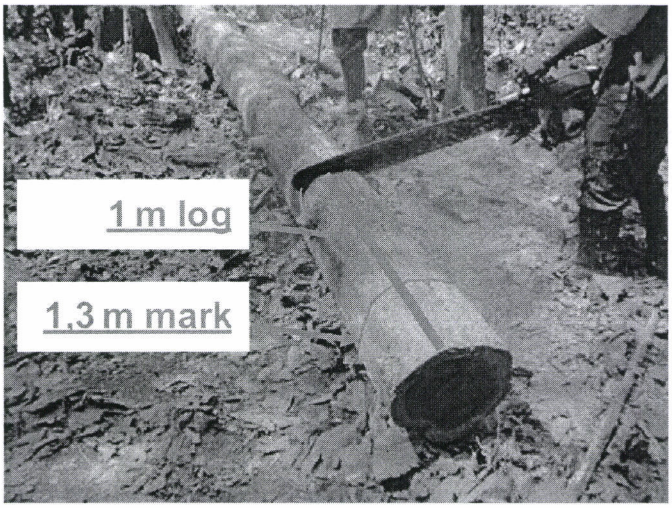
2- Cross cut at 1.2 m level



3- Mark the bottom part (with permanent colour) writing the tree number and the height of the section (in this case 1.2 m)

4- Measure 1 m from this section and mark the level on the bark

5- Cross cut at this level in order to obtain the 1 m small log for disks and destructive sampling



6- Mark the bottom part of the remaining log, writing the tree number and the height of the section (in this case should be 1.2 + 1 = 2,2 m)

7- Mark the X m level on the remaining log corresponding to 25% of commercial volume

8- Cross cut at that level (the length of each log depends on the diameter)

9- Mark the bottom part of the remaining log, writing the tree number and the height of the section (in this case should be 1.2 + 1 + X = Y.y m)

10- Repeat for the log corresponding to 75% of commercial volume

Summary:

The other logs are stored for further analysis.

First log for wood analysis and calibration/prediction process

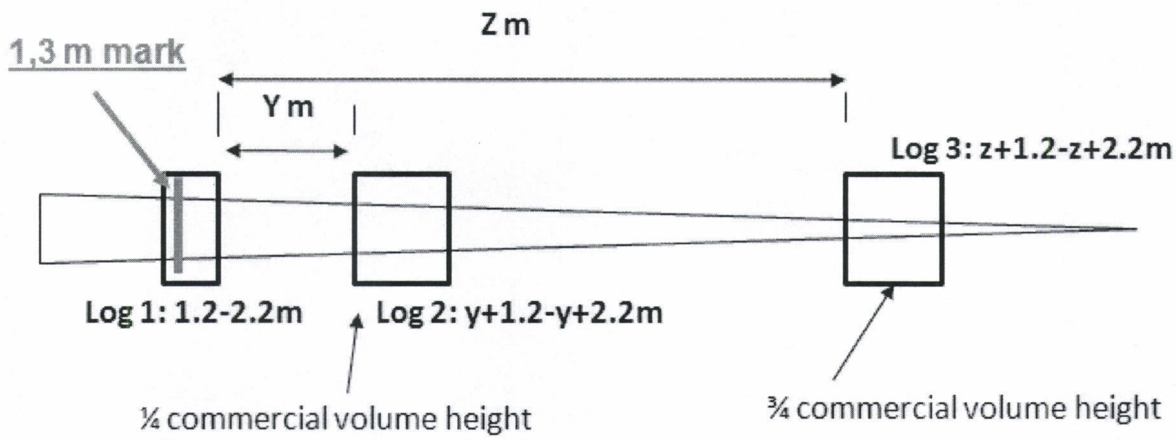


Figure 11. Summary of sampling along the bole

7.1.3 Chemical Measure on the wood sample

The wet chemistry analysis need 12 weeks corresponding to extractives, lignin and cellulose contents (150 samples x 2 times = 300 analysis). The chemical measures route and samples flow are presented in the Figure 12.

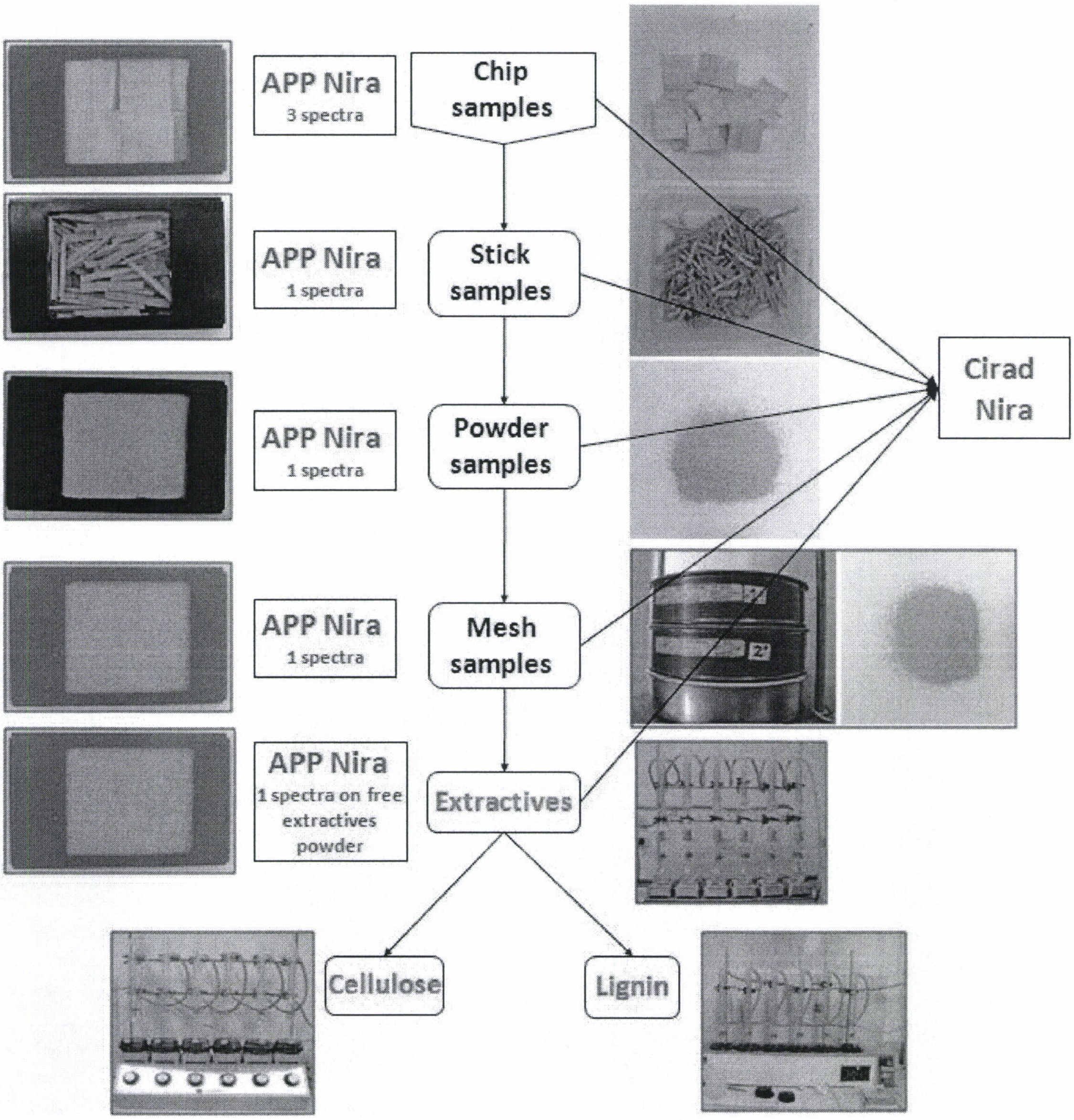


Figure 12. Chemical analysis process for calibration set (Cirad need 15 gram for chips and sticks and 5 gram for powder, mesh)

7.1.4 Basic density and Pulp Yield measures

The basic density and pulp yield need 6 weeks. The chemical measures are presented in the Figure 13.

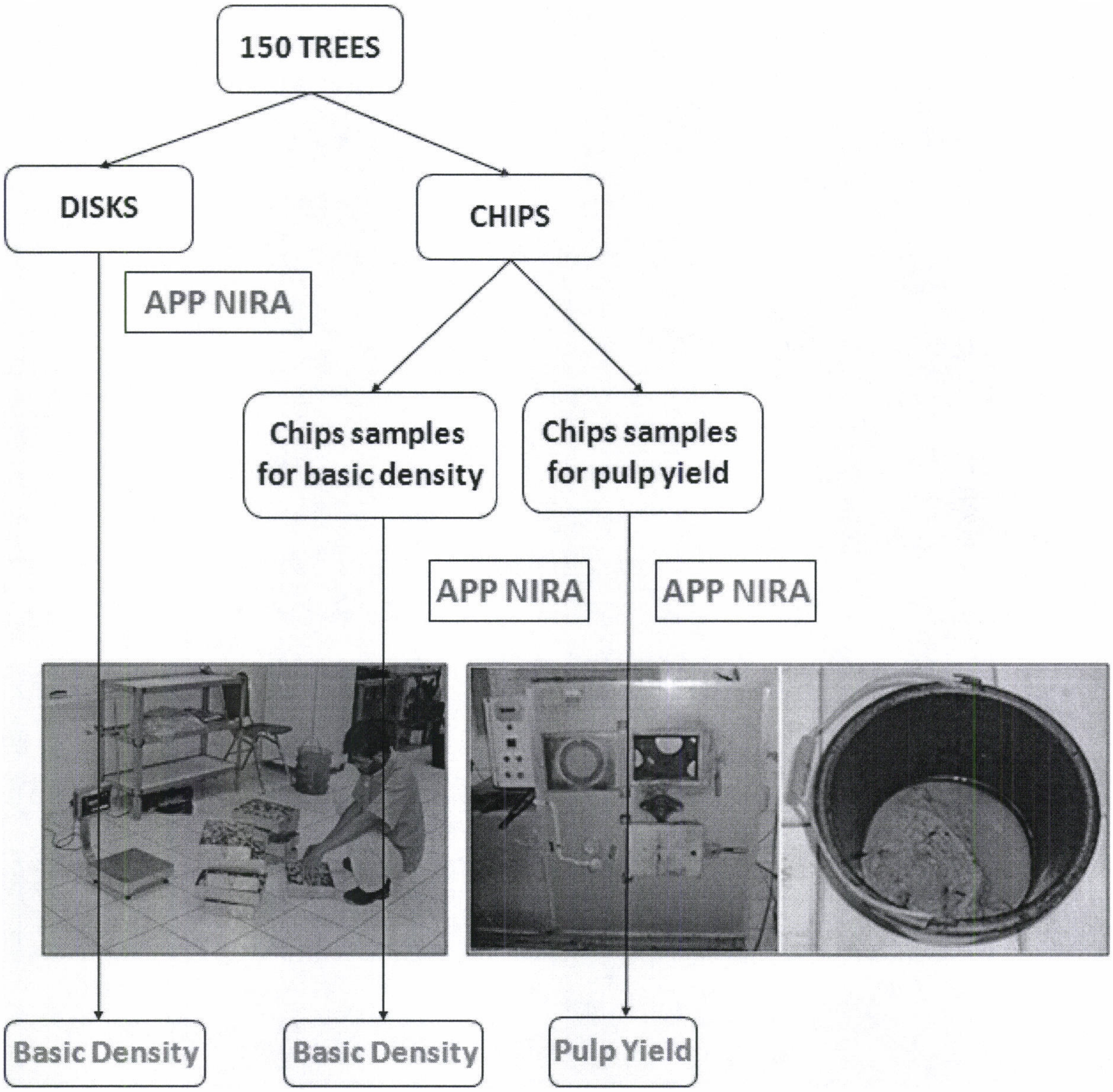


Figure 13. Basic density and pulp yield for calibration/prediction process

7.1.5 Fiber length and Coarseness measurement

Because fiber length and coarseness measurement are not priority for calibration training these measure will be done later. These need 6 weeks.

7.2 SUMMARY OF SAMPLE FLOW FROM APP TO CIRAD

In order to compare/verify APP and CIRAD NIR apparatus, Cirad needs both the entire samples and the data from the lab to build the calibration based on the spectra data from Nir Cirad apparatus. The Figure 14 shows the summary of wood sample flow according to the three steps (Existing calibration, presampling for calibration set selection and calibration prediction set trees).

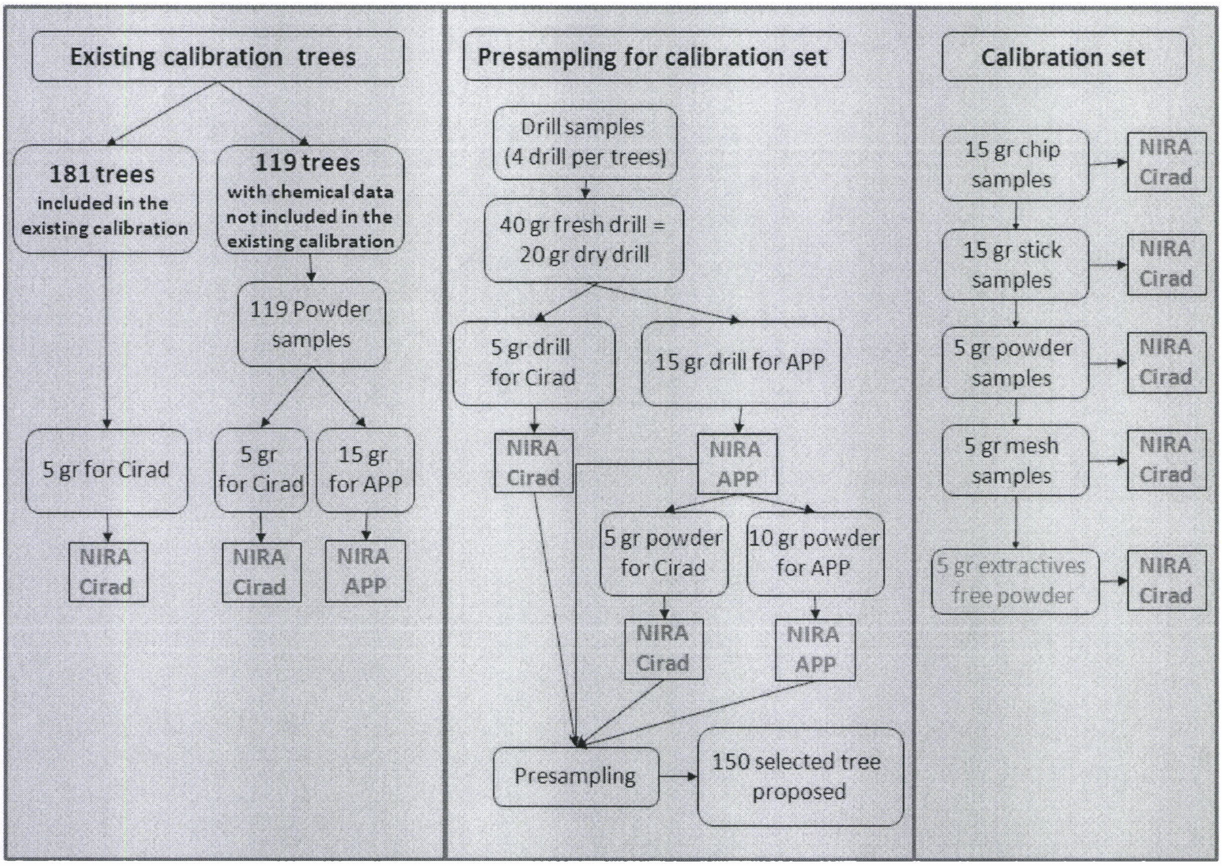


Figure 14. Summary of wood sample flow from APP to Cirad

7.3 NOTATION AND NUMBERING SYSTEM

The sample identification is a crucial point.

Thee files will be compiled from all the current data files concerning the NIRA activities:

- One sample identification file with Species, location, trials …
- One chemical and physical file where each laboratory analysis data will be referenced
- One spectral file with details on spectral acquisition.

We recommended the methodology and the forms as described in Table 4, Table 5, Table 6.

Table 4. Sample identification file proposed

Numerous sample	Species	Trials	Provenance	Descendance	Bloc	Repetition	Location	Soil	Age	Feeling date	Sampe collected	Date	Assessor
1													
2													
3													
4													
5													
6													
7													
8													
9													
10													
11													
12													
13													
14													
15													
16													
17													
18													
19													
20													
21													
22													
23													

Species
1: Eucalyptus pellita
2: Acacia crassicarpa
3: Acacia mangium
4:
5:
6:

Soils
1: mineral soil
2: pet land
3:
4:

Sample collected
1: Chips wood
2: Sticks wood
3: Powder wood
4: Mesh wood
5: Extractive free wood
6: Solid wood

Table 5. Example of chemical and physical file proposed

Numerous sample	Numerous analysis	Extractives	Date	Assessor	Cellulose	Date	Assessor	Lignin	Date	Assessor	BD chips	Date	Assessor	BD Disk	Date	Assessor
1	1															
1	2															
1	3															
2	4															
2	5															
2	6															
2	7															
3	8															
1	9															
1	10															
1	11															
1	12															
4	13															

Pulp yield	Date	Assessor	Coarseness	Date	Assessor	Fiber lenght	Date	Assessor

Table 6. Spectral file proposed

Numerous sample	Numerous analysis	Numerous spectra	Type sample	Face for solid wood	Assessor	Date spectra acquisition	Number scan	Accurancy (nm)
1	2	1						
1	2	2						
1	3	3						
2	3	4						
3	3	5						
4	5	6						
5	2	7						
6	5	8						
7	6	9						
		10						
		11						
		12						
		13						

Type sample
1:Chips wood
2: Sticks wood
3: Powder wood
4: Mesh wood
5: Extractive free wood
6: Solid wood
Face
1: radial
2: axial
3: tangential

8 TRAINING IN FRANCE

M. Ruspandi will be train in France (Montpellier) during 45 days on the wood samples of APP previously sent and CIRAD samples.

The training program is as follow:

- Chemometric methods training by CIRAD 2 weeks
- WINISI and FOSS Spectrophotometer training 1 week
- Wood spectral acquisition of APP samples 1 week on Cirad apparatus
- PCA on wood spectral and selection of the calibration and validation sets 2 weeks

The work program related to the previous points is the following:

1- Spectra acquisition on drill samples

2- Spectra acquisition on chips and sticks

3- Spectra acquisition on powders

3- Spectra acquisition on powders for chemical analysis (35 > x > 180 mesh)

5- Calibration equations with first data group (basic density, pulp yield, extractives, lignin, and cellulose)

6- Calibration equations with second data group (fiber length and coarseness)

7- Work plan to achieve until the next mission

Table 7. Planning of NIRA up grading for APP

	2007								2008				
	May	June	July	August	September	October	November	December	January	February	March	April	May
First mission for Nira Upgrading													
Misson in Indonesia (Vigneron and Chaix)	X												
Definition of workplan	X												
Check of chimical analysis, Nir equipment, type of samples	X												
Performing previous data for EP (180 nir+wp, 120 wp)													
Check spectra of current samples set on the Nira Computer (180 are used now)	X												
Check the 300 sample set (Powder and chips)		X											
Nir acquisition on powder for 120 samples remaining		X											
Send to Cirad 300 grounded samples, 5 gr per samples (1)		X											
Calibration reviewed by Cirad (BD, PY, Extratives, Lignin, Cellulose)		X											
Calibration reviewed by APP (BD, PY, Extratives, Lignin, Cellulose)		X											
Calibration reviewed by APP in France (BD, PY, Extratives, Lignin, Cellulose)										X			
Calibration set selection													
Collecting 40 gr drill samples (4 holes minimum 4 direction) on 300 trees		X											
Drying 300 drill samples at room conditions		X											
Nir acquisition on 300 drills dryning samples 20 gr		X											
Send to Cirad 300 drill samples, 5 gr per samples (1)		X											
Grounding 300 drills 15 gr		X	X										
Nir acquisition on 300 grounded samples			X										
Send to Cirad 300 grounded samples, 5 gr per samples (1)			X										
Data spectra analysis by PCA and prediction with previous calibration from Cirad and APP			X										
Selection of 150 trees			X										
Cirad will send selected tree list of 150 trees + 20 trees to replace if necessary			X										
Wood sampling collection for calibration set													
Felling the 150 trees				X									
Collecting samples (disk, core, drill, part for chips, part for solid wood sample)				X	X								
Preparing chips, sticks, grounded wood for wet chemistery					X								
Nir acquisition on chips (3 scans by sample)					X								
Nir acquisition on sticks (1 scan by sample)					X								
Nir aquisition on ground wood (1 scan by sample)					X								
Separating powder for chemical analysis (35 > x > 180 mesh)					X								
Nir aquisition powder for chemical analysis (35 > x > 180 mesh) (1 scan per sample)					X								
Extractives contents					X	X	X						
Nir aquisition on fre extractive powder					X	X	X						
Lignin contents						X	X	X					
Cellulose contents						X	X	X					
Pulp yield on chips						X	X						
Basic density on disk and on chips						X	X						
Send to Cirad chips, drill, powder, powder for chemical analysis (5 gr / samples)								X					
Fiber lentgh and Coarseness mearsurments									X	X			
Send the values to Cirad								X		X			

Table 8. Planning of NIRA up grading for APP

	May	June	July	August	September	October	November	December	January	February	March	April	May
Repeatability of reference values													
Preparing sample (for basic density, pulp yield need 2 trees to fell)			■										
Nir acquisition on chips (3 scans by sample)			■										
Nir acquisition on sticks (1 scan by sample)	■		■										
Nir aquisition on ground wood (1 scan by sample)			■										
Separating powder for chemical analysis (35 > x > 180 mesh)			■										
Extractive contents				■									
Nir aquisition on free extrative powder													
Lignin contents				■									
Cellulose contents				■									
Pulp yield on chips				■									
Basic density on disk and on chips				■									
Training in France													
Preparing sample in Indonesia								■					
Nir acquisition on solid wood samples									■	■			
Nir acquisition on drill samples									■	■			
Nir acquisition on chips 2									■	■			
Nir aquisition on powder for chemical analysis (35 > x > 180 mesh)										■			
Calibration equations (BD, PY, Extratives, Lignin, Cellulose, FL and Coars.)										■			
Calibration in routine													
First test of prediction in routine											■		
Outliers analysis (chemical and spectrum) and include in calibrations											■	■	
Second mission for Nira Upgrading													
Control of test in routine												■	■
Perform the calibration equations in Cirad and APP												■	■
First including wood properties in selection/breeding program of Eucalypts												■	■
Handbook for improvement of calibration equation quality													■