

# How useful is DNA barcoding for large-scale inventories of tropical trees?

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# What is DNA barcoding?

« Use of a short gene sequence from standardized region of the genome as a tool for species identification »

Hebert & al. 2003

Barcode must be:

- Short <700pb
- Highly conserved yet variable enough



→ ATCTTGCCGA ACTAT



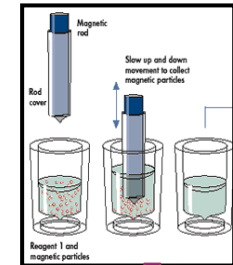
→ ATCTAGCCGCACTAT



→ GTATAGCTGCAAGAT



1. DNA  
Extraction

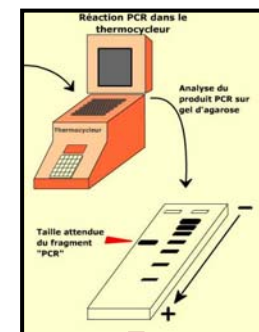
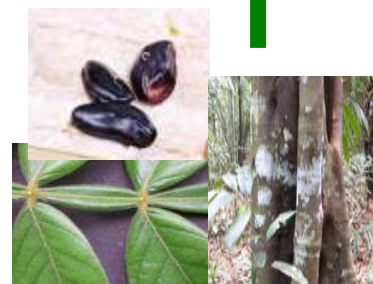


2. PCR

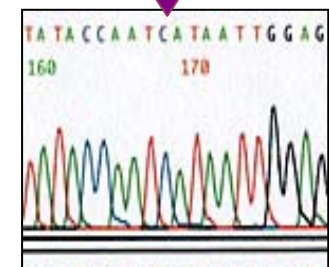
Reference DNA matrix

A	C	T	G	A	A	T	G
A	C	T	G	T	A	T	G
A	C	C	G	T	A	T	G
A	C	T	G	G	A	T	G

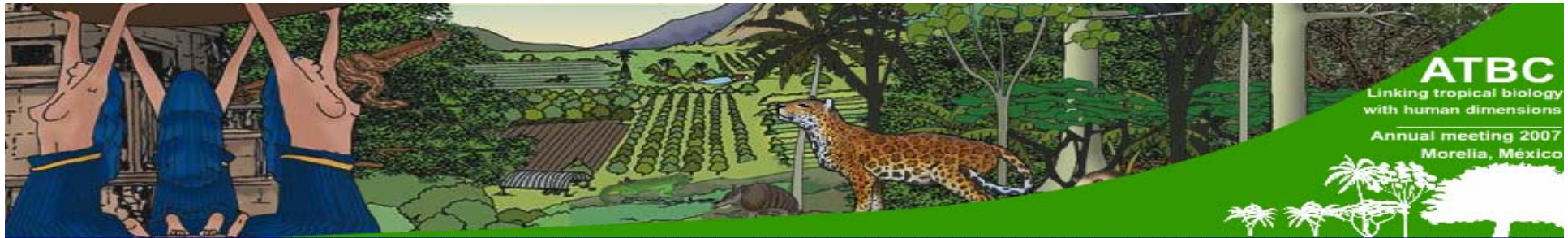
1,2,3



3. Sequencing







# ATBC'07 → test the utility of DNA barcoding for inventorying the tree community

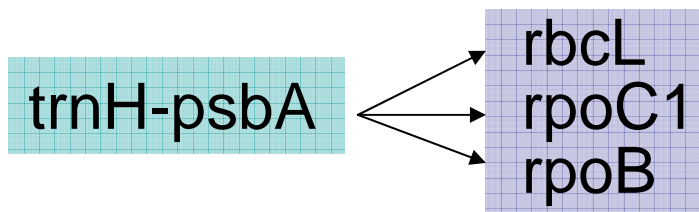
**rpoC1**

468 individuals  
38 families  
~ 170 species

Assignment  
→

100% family  
80% genus  
40% species

**Multilocus approach needed**



*Kress & Erickson 2007*



*Lahaye et al. 2008*

# Outline

## **1. Further tests of the DNA barcoding approach**

- 1.1 Which DNA region?
- 1.2 When molecules do not predict species identity

## **2. Applications to real field sampling**

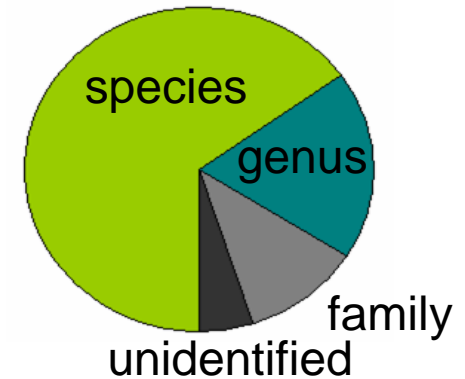
- 2.1 To identify unknown groups in a field inventory
- 2.2 To identify seedlings



# BRIDGE project

2 ha → 1028 trees dbh > 10cm

Botanical  
identification



**48 families  
129 genera  
255 species**

Marker

rpoc1

rbcL

rpoB

ycf5

matK

trnH-psbA

## 1.1 Which DNA region?

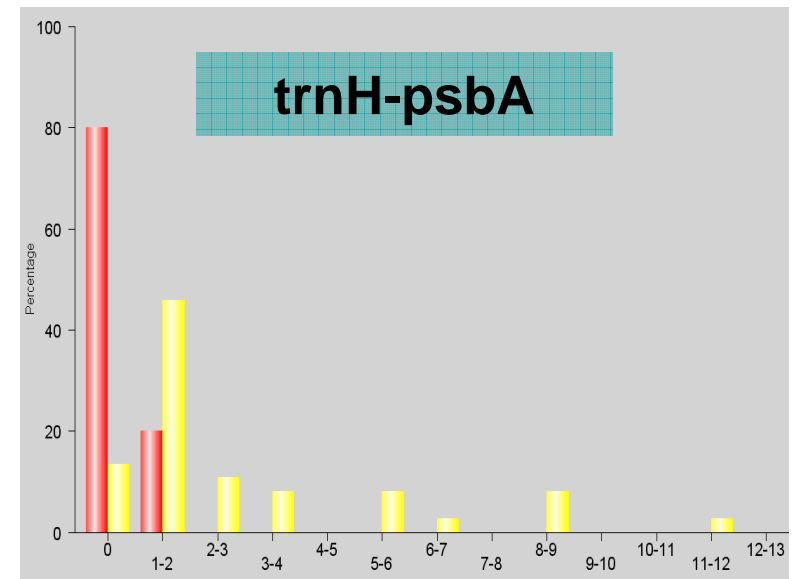
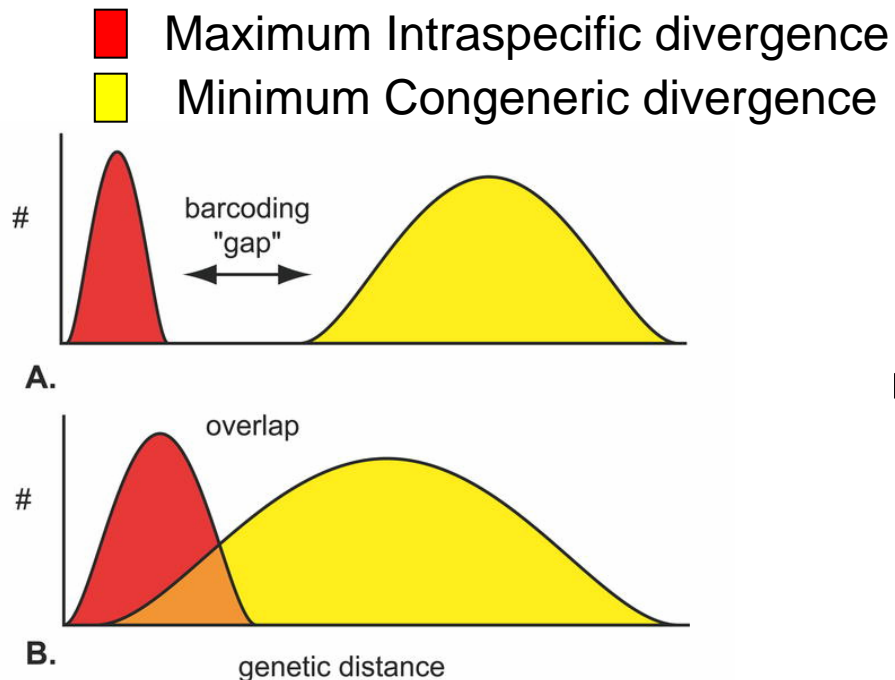
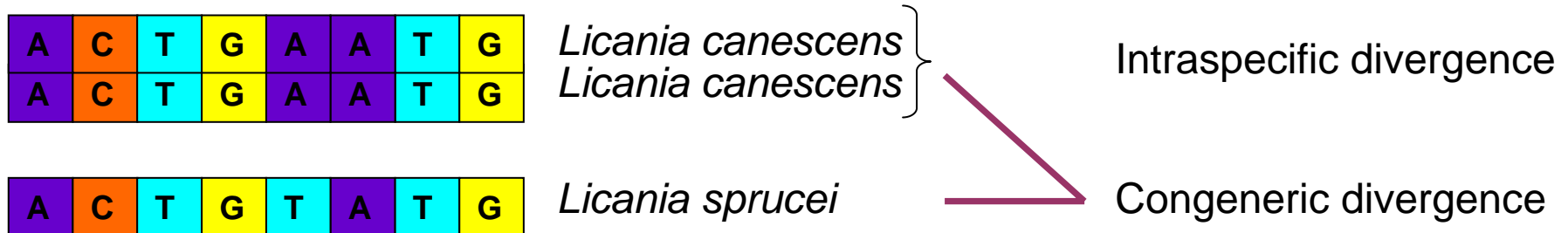
# Universality of DNA markers

Marker	Sequencing success %	
rpoc1	98	
rbcL	96	
rpoB	97	Moraceae
ycf5	88	Rubiaceae Myristicaceae Apocynaceae
trnH-psbA	89	
matK	30	



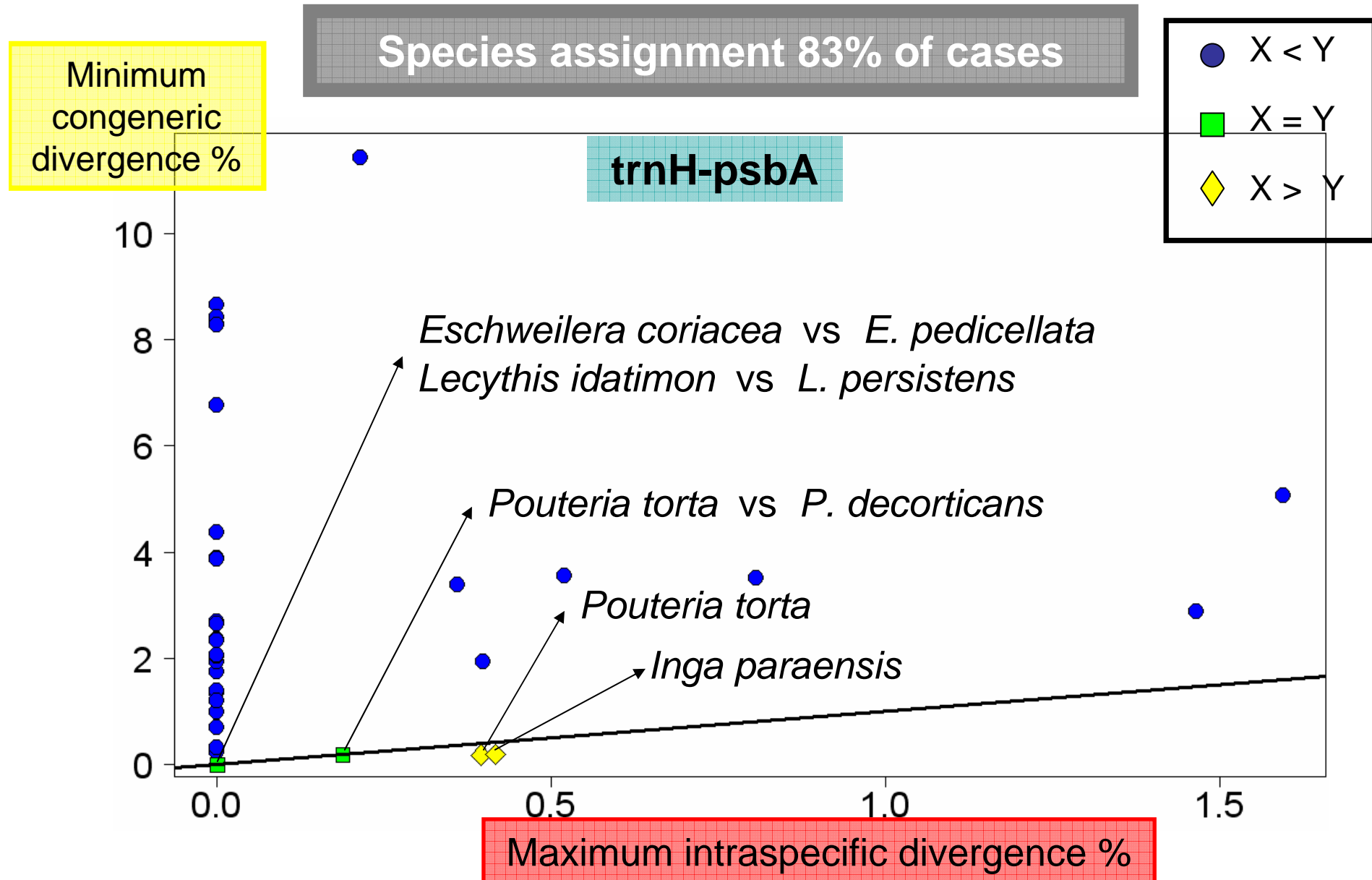
## 1.1 Which DNA region?

# Variability of DNA markers



## 1.1 Which DNA region?

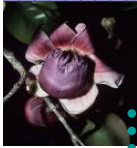
# Differentiating closely related species



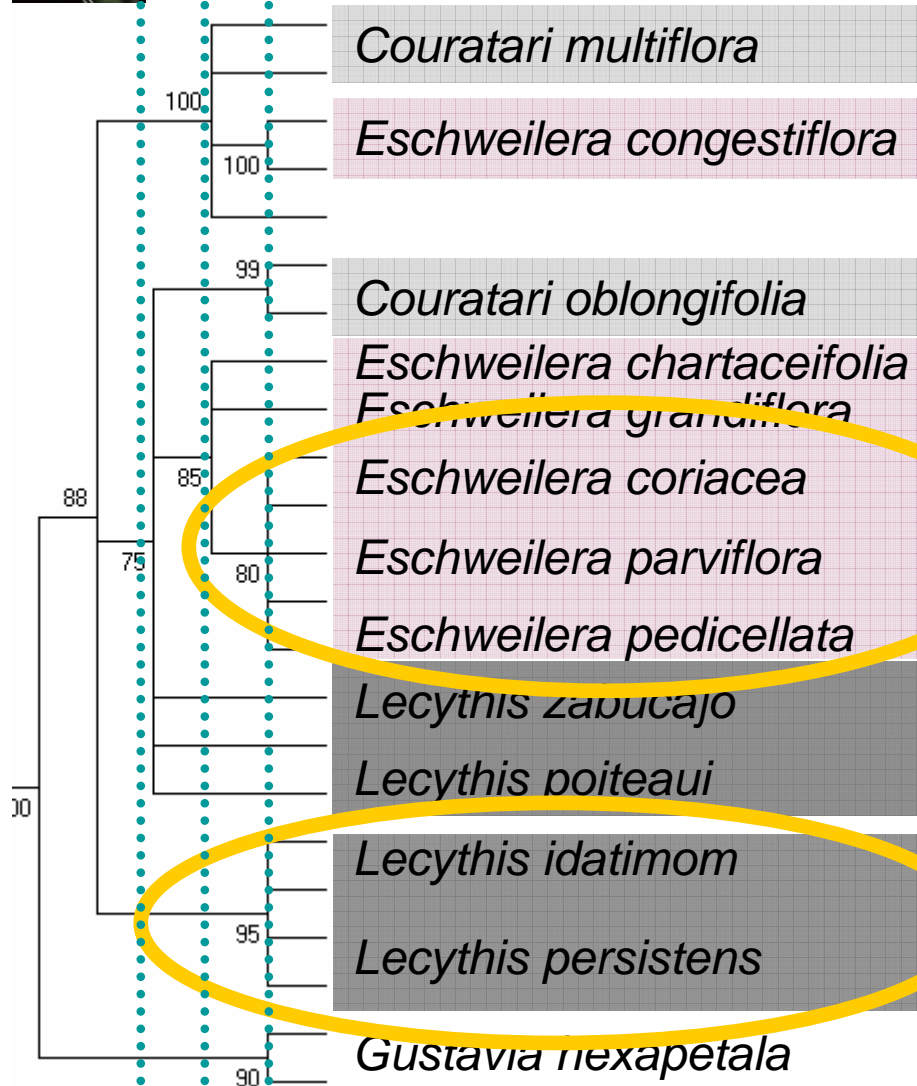


## 1.2 When molecules do not predict species identity

### Particular cases



#### Lecythidaceae



#### 1. Low genetic variation



ITS

#### 2. Polyphyletic genus



Identification  
of lineages

**rbcL**

Mori et al. 2005





## **2. APPLICATIONS to real field sampling**



## 2.1 Application to real sampling: Identifying “unknown” trees after an inventory

100 trees → 80 identified to the family  
20 unidentified

trnH-psbA + rbcL	% individuals
family	100 %
genus	80 %
species	20 %

DNA sequence absent from data repositories



## 2.1 Application to real sampling: Identifying seedlings



252 seedlings  
collected

### Botanical survey

17% species  
43% genus  
60% family  
40% unidentified

### Molecular approach

trnH-psbA + rpoc1

37% species  
79% genus  
97% family  
3% unidentified

# Conclusions and Perspectives

- Molecular approach → Reliable tool in tropical trees identification  
→ Standard & Fast

*trnH-psbA*  *rbcL*  
*rpoC1* 80%-90% → species

- DNA barcode reference needed

*rbcL*: 66% genus, 40% family **absent in Genbank**

*trnH-psbA* 80% genus, 45% family **absent in Genbank**

Cost per barcode region → \$5



78.800 angiosperms → \$400.000

# Conclusions and Perspectives

- Application in permanent plots and future inventories



Homogenize identification across  
permanent plots



Better insights about species distribution

- Dietary of animals

- Wood Forensics

Convention on International Trade in Endangered Species of Wild Fauna and Flora  
CITES



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