



**GCIRC Rapeseed/Mustard Technical Meeting**  
**Feb 2-4, 2009**  
**New Delhi, India**



Structure of **genetic diversity** and elaboration of **core collections** for the three related *Brassica* species: *B. napus*, *B. oleracea*, *B. rapa*

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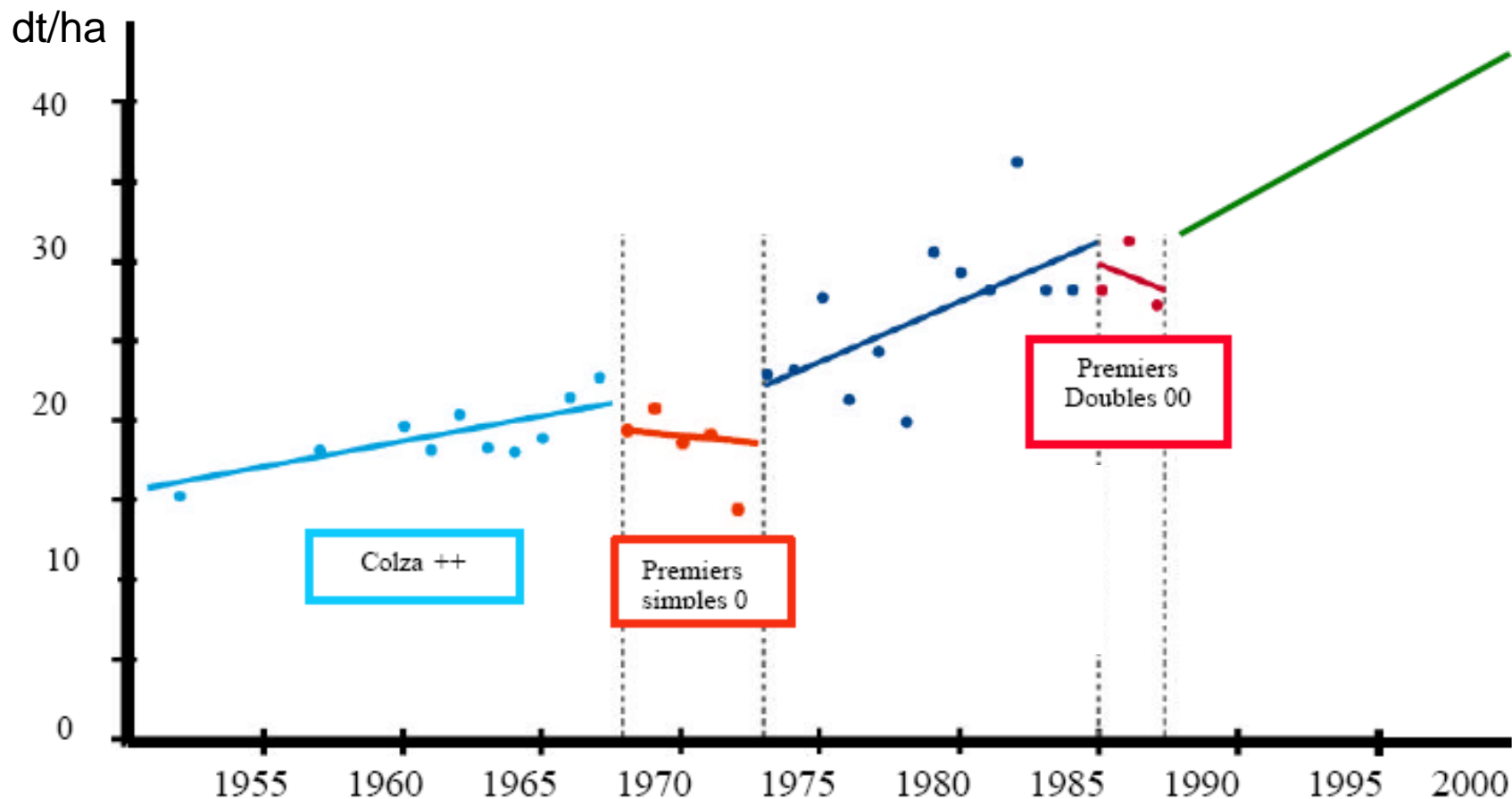


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# Towards a reduction of exploited *B. napus* diversity



=> Need to broaden *B. napus* genetic diversity by exploiting its progenitors

Source CETIOM

# Rationales

- How is Brassica genetic diversity organized?
- How to answer to new breeding challenges ?
  - Exploiting *Brassica* genetic diversity

⇒ A real need for defining core collections

# Objectives

Study the **genetic diversity** of

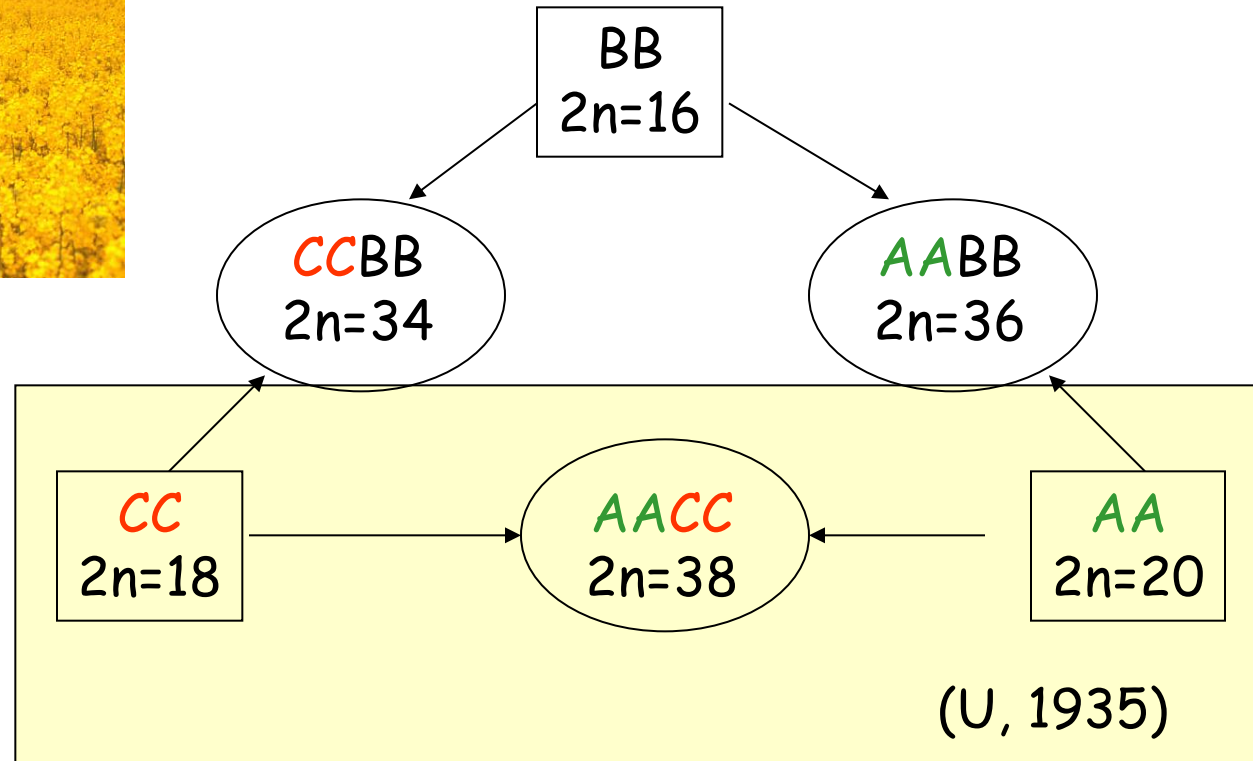
- a semi-autogamous species *Brassica napus*
- two allogamous species : *Brassica oleracea*,  
*Brassica rapa*

**Connect the diversity** of *Brassica napus*, *rapa* and *oleracea*

**Deliver public core collections** to the scientific community

# Which available genetic diversity for *Brassica napus* ?

# Which diversity ? Relationships between Brassicae



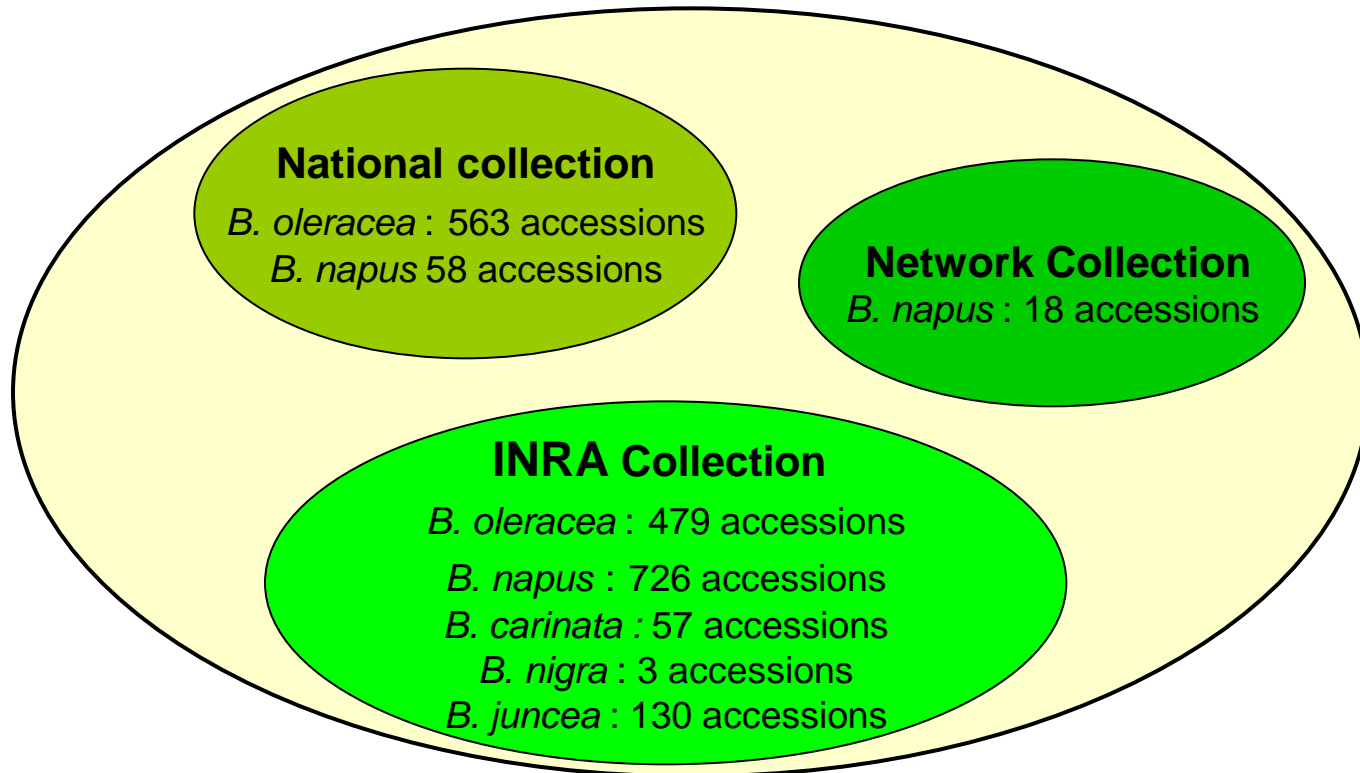
# Which available genetic diversity for the scientific community ?

Gene banks

Panels, diversity sets and core collections



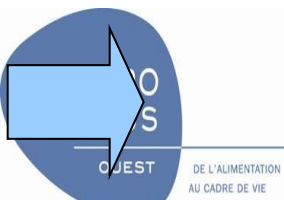
# French Brassica Genetic Resources Center (BGRC)



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# International organisation of *Brassica* diversity study

- RESGEN Project
  - *Brassica* core collections



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## EU project RESGEN CT99 109-112

**"*Brassica* Collections for Broadening Agricultural Use" including  
"Characterising and utilising genetic variation in *Brassica carinata* for its  
exploitation as an oilseed crop"**

The RESGEN CT99 109-112 project on the genetic resources of *Brassica* was carried out in the framework of the European Community Programme on the Conservation, Characterisation, Collection and Utilisation of Genetic Resources in Agriculture. The project was funded by The European Commission and started January 1<sup>st</sup> 2000.

### Core collections of the four *Brassica* species

Each of the four subgroups of the project developed their own core collection. The size of the four core collections is as follows:

- *B. oleracea*: 395 accessions
- *B. rapa*: 100 accessions
- *B. napus*: 150 accessions
- *B. carinata*: 80 accessions

<http://documents.plant.wur.nl/cgn/pgr/brasedb/brasresgen.htm>

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# International organisation of *Brassica* diversity study

- RESGEN Project
  - *Brassica* core collections (Institute of Crop Science and Plant Breeding Giessen, Germany)
- BnDFFs «Diversity Fixed Foundation Sets» (OREGIN ([www.oregin.info](http://www.oregin.info)))



**A Diversity Fixed Foundation Set (DFFS): is defined as**  
*"an informative set of genetically fixed lines representing a structured sampling of diversity across a gene pool"*

Species	Set	Number of lines	Founder line seed available	Fixed line seed available	Founder DNA available	Fixed line DNA available	detail	MTA	Project	Contact
<i>B. napus</i>	BnaDFFS	188	152	56	152	53	 Oct 08	-		Graham Teakle (WHRI)
<i>B. oleracea</i>	BolDFFS	376	-	-	-	-	 Jan 09	-		Graham Teakle (WHRI)
<i>B. rapa</i>	BraDFFS	-	-	-	-	-	-	-	-	Guusje Bonnema (MUR)
<i>B. C genome</i>	BCgDFFS	88 (tbc)	-	-	-	-	-	-		Paul Hand (WHRI)



A web based User Interface has been developed to query the population Area of CropStore, to view data summaries and download Excel workbooks containing detailed data - [Available Here \(CropStore :: Populations\)](#)

[www.oregin.info](http://www.oregin.info)

# International organisation of *Brassica* diversity study

- RESGEN Project
  - *Brassica* core collections
- BnDFFs «Diversity Fixed Foundation Sets» (OREGIN ([www.oregin.info](http://www.oregin.info)))
- *rapa* collection: China – Wageningen (G. Bonnema)
- Collections of Vavilov Research Institute of Plant Industry (*napus*, *rapa* and *oleracea*)

# The first target:

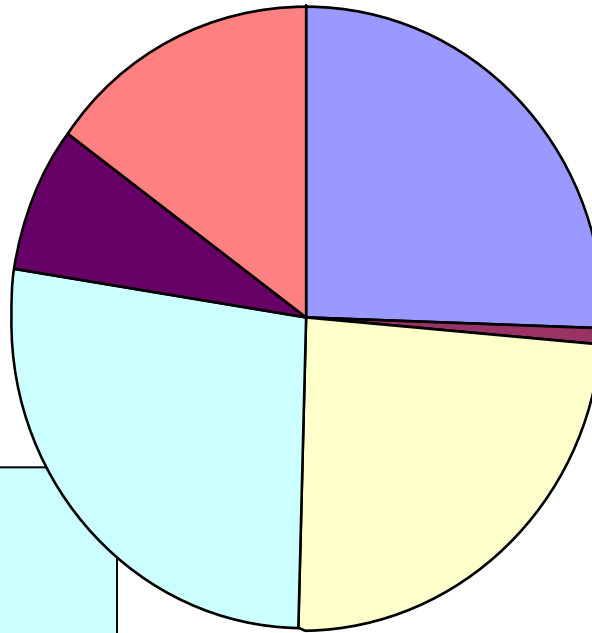
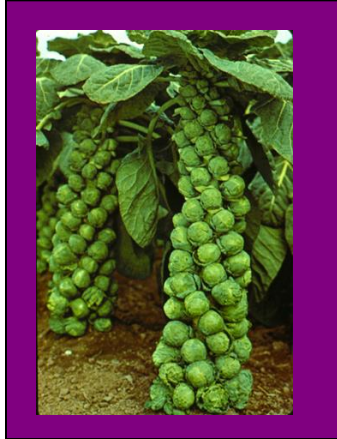
## *Brassica oleracea*

# Plant material



# Brassica oleracea collection

384 accessions



- acephala
- alboglabra
- capitata
- botrytis
- oleracea
- non défini



# Definition of SSR marker sets

## SSR pool

Public markers

582 SSR derived from EST Génoplante

40 SSR Australia (bo)

113 SSR Japan (br)

103 SSR USA (bo)

80 SSR Agriculture Canada

BBSRC England

Celera

Private markers

130 SSR Trait Genetics

Quality indice

Position on genetic map

DnxY

### 1<sup>st</sup> SSR choice

- Quality

- distribution on maps

Position on  
Traits Genetics  
map

**2<sup>nd</sup> SSR Choice** depending on pilote  
project

*12 napus*

*6 oleracea*

*6 rapa*

Set 76 SSR rapa



Set 76 SSR napus



Set 76 SSR oleracea

# Data mining

Definition of an analysis strategy based on *Brassica oleracea* data

# Current results

## Study of 384 *B. oleracea* accessions using 22 SSR

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# General strategy

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## The ECPGR *Brassica* Database

[Centre for Genetic Resources, The Netherlands \(CGN\)](#)

Database managers:  
[F. Menting](#) and [drs. N. Bas](#)



<http://documents.plant.wur.nl/cgn/pgr/brasedb/default.htm>



A large core collection of 384 accessions (RESGEN cc) defined according to cultigroups and origins of accessions



1. Acquisition of molecular data based on public SSR markers

2. Study of the genetic diversity structure

3. Sample and validate a core collection

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# Methods: Definition of core collection

## 2. Study of genetic diversity structure

Bayesian approach  
Software structure 2.2

*Group definition and affiliation of accessions to groups*

## 3. Sampling core collection

**Maximisation (M)**  
(MSTRAT software)

**Random (R)**

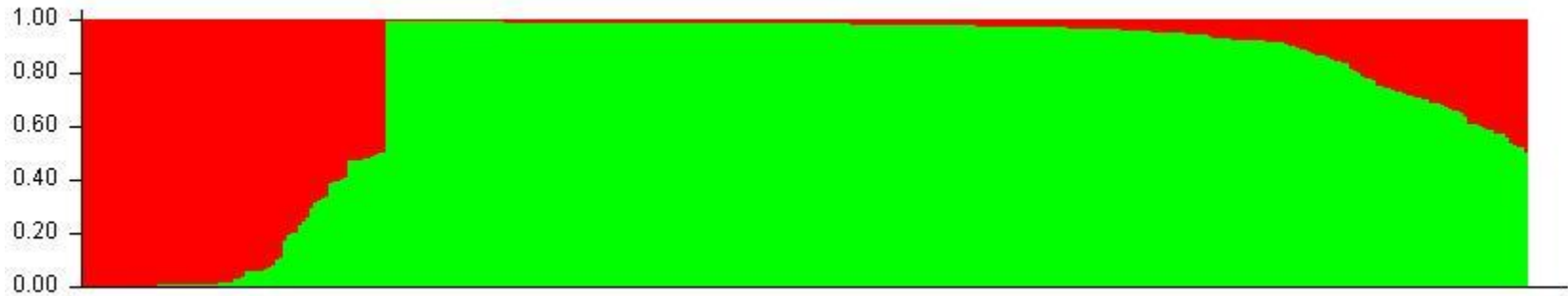
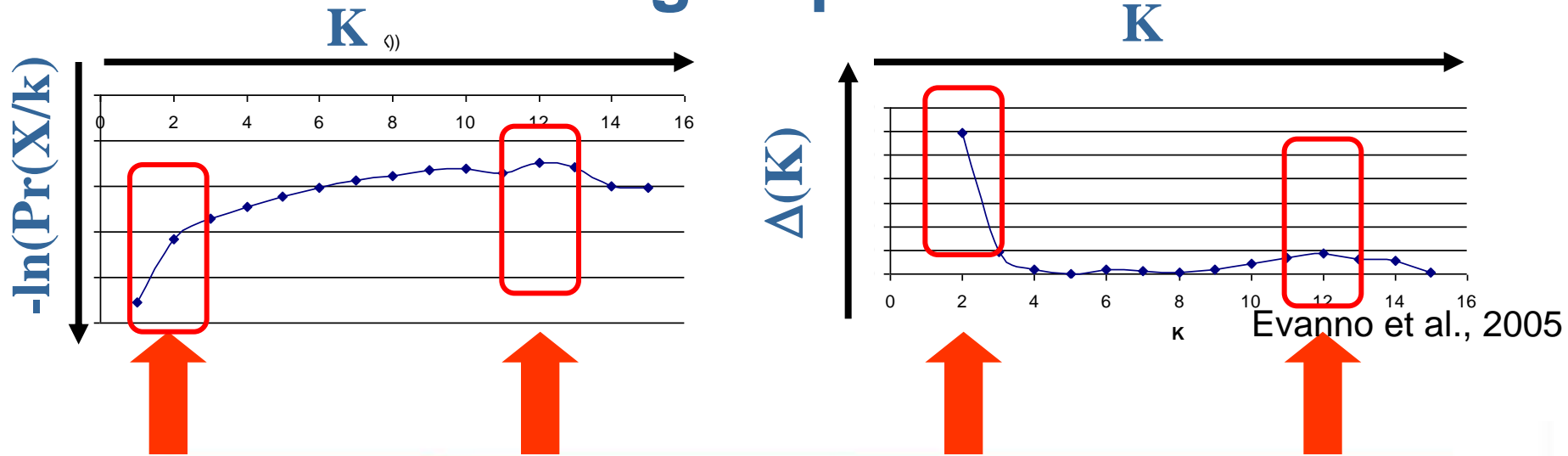
Methods based on  
diversity structure results

**Constant (C)**

**Logarithmic (L)**

**Proportional (P)**

# 2 - Structure of the collection revealed by a bayesian approach: from 2 to 15 groups



**Cauliflowers and Broccoli**

**Broccoli and others cultigroups**

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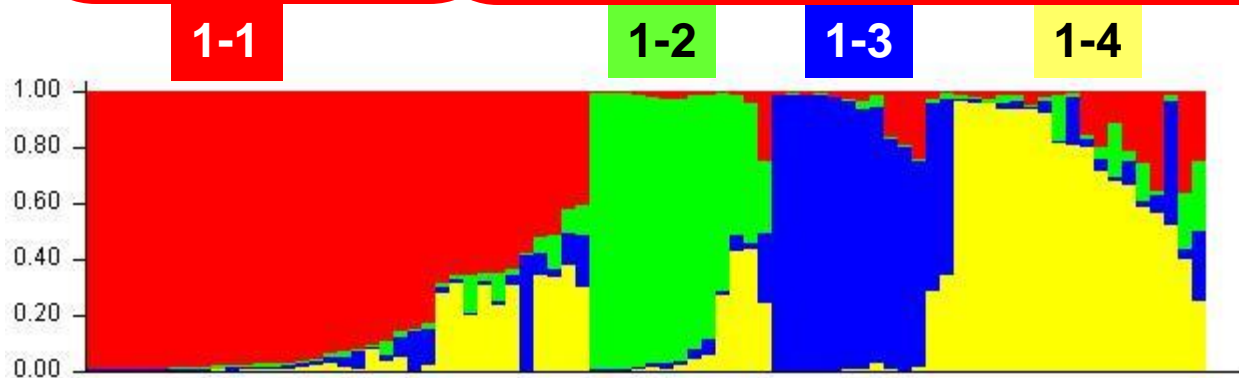
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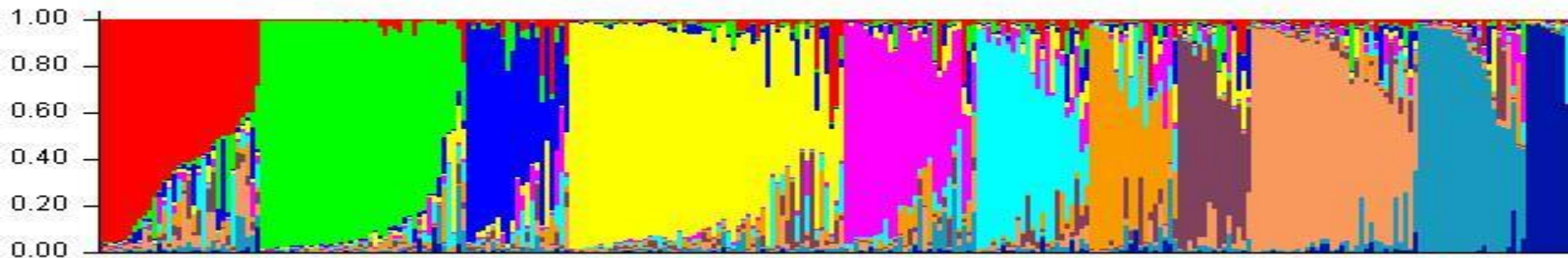
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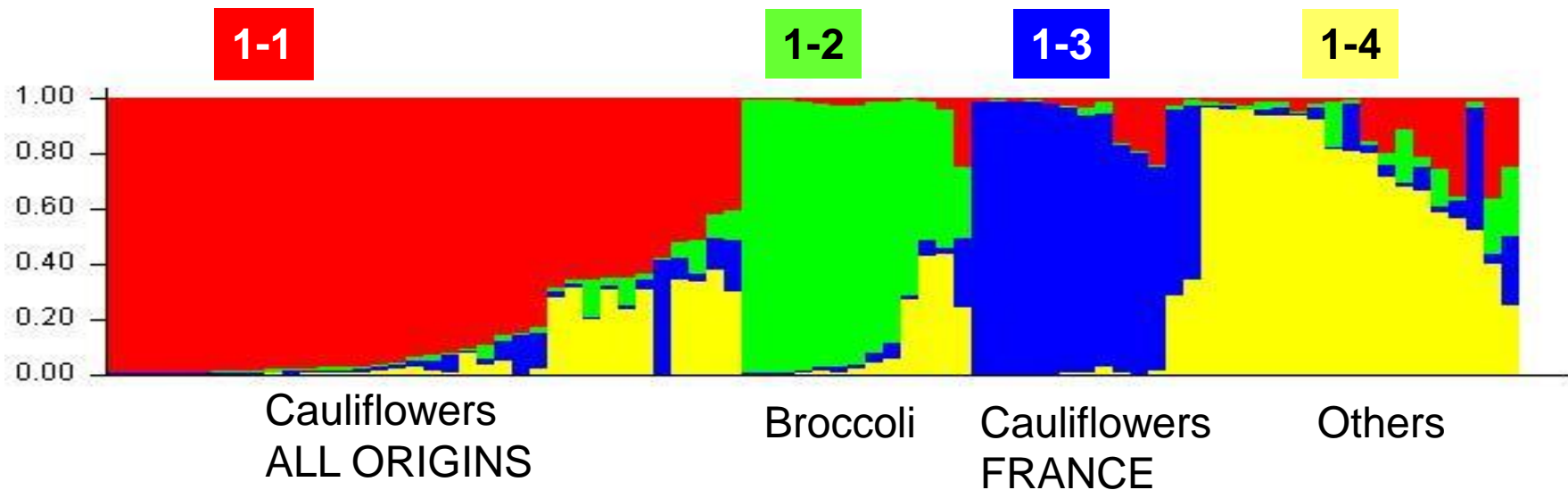




80 accessions







Cultigroups  
good indicators

*B. o. acephala*  
*acephala medullosa*  
marrow-stem kale

2-1

*B. Oleracea gemmifera*  
Brussels sprouts

*B.o. acpehala sabellica* curly kale

Geographic origin are  
good structure

*B.o. capitata*  
cabbage

2-4

*B.o. botrytis italica* (broccoli)  
*B.o. acephala gongylodes* - Kohlrabi  
(chou-rave) (ITALY)

2-5

*B.o. hilarionis, campestris,*

All cultigroups  
EGYPT, TURKEY SYRIA

2-6

*B.o. capitata costata, acephala*  
Cabbages, tronchuda cabbage, kale  
PORTUGAL SPAIN

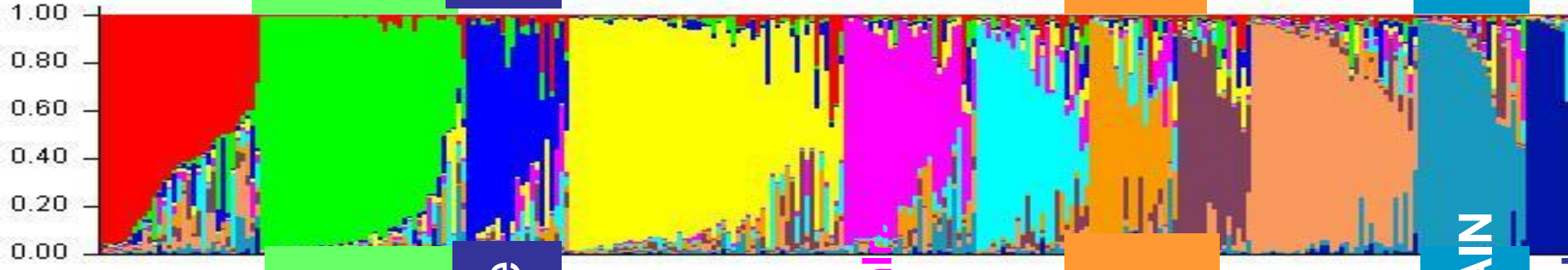
2-8

2-9

*B.o. capitata alba* white cabbage SPAIN

*B.o. acephala alboglabra* – chinese kale

2-11



# 3 - Estimation of core collection size

Constant (C)   Proportional (P)   Logarithmic (L)   Maximisation (M)   Random (R)

## Collection structure

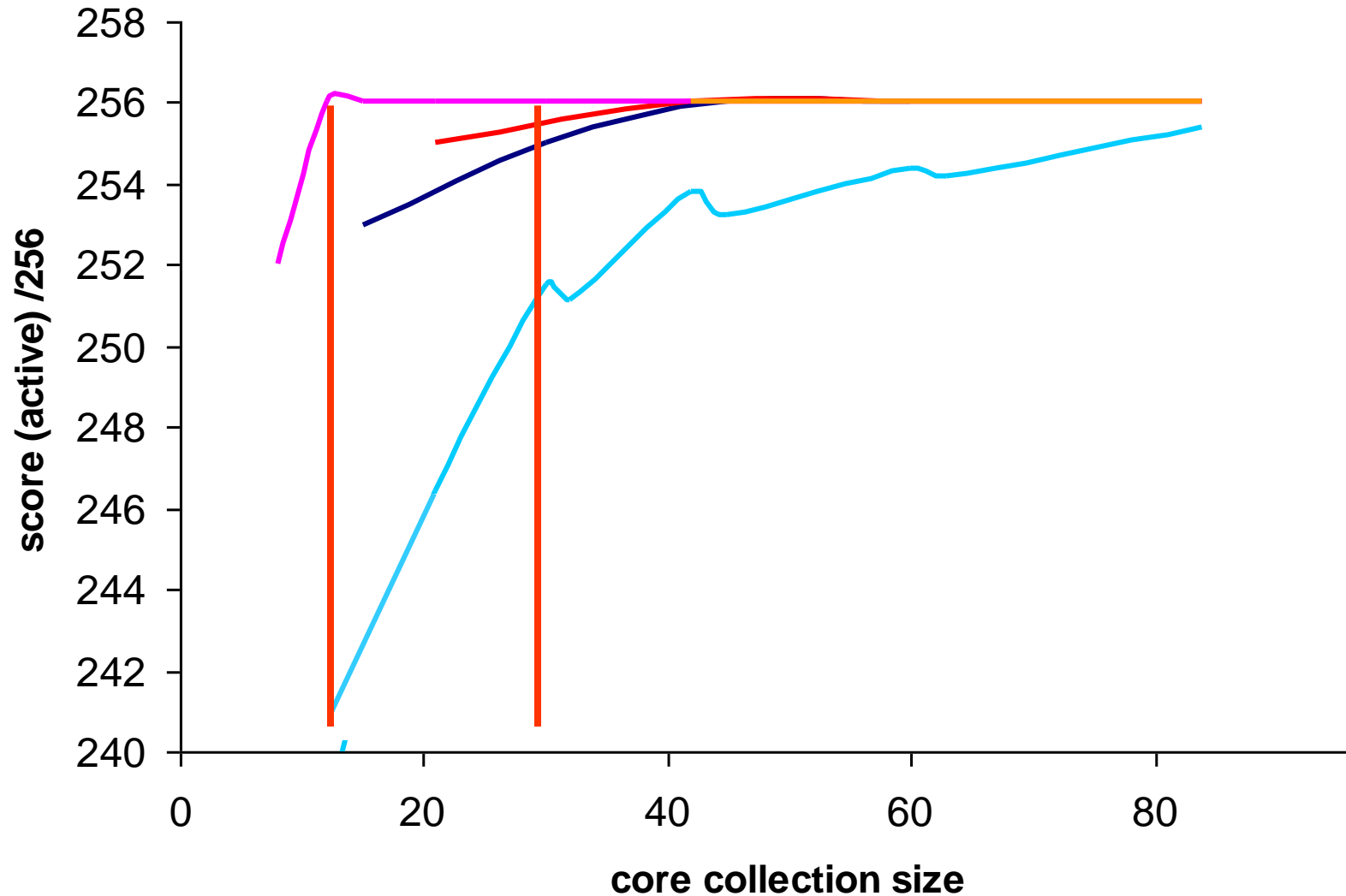
group	size
1-1	16
1-2	13
1-3	13
1-4	18
2-1	33
2-2	42
2-3	21
2-4	56
2-5	27
2-6	23
2-7	18
2-8	15
2-9	34
2-10	22
2-11	9

cc sizes tested



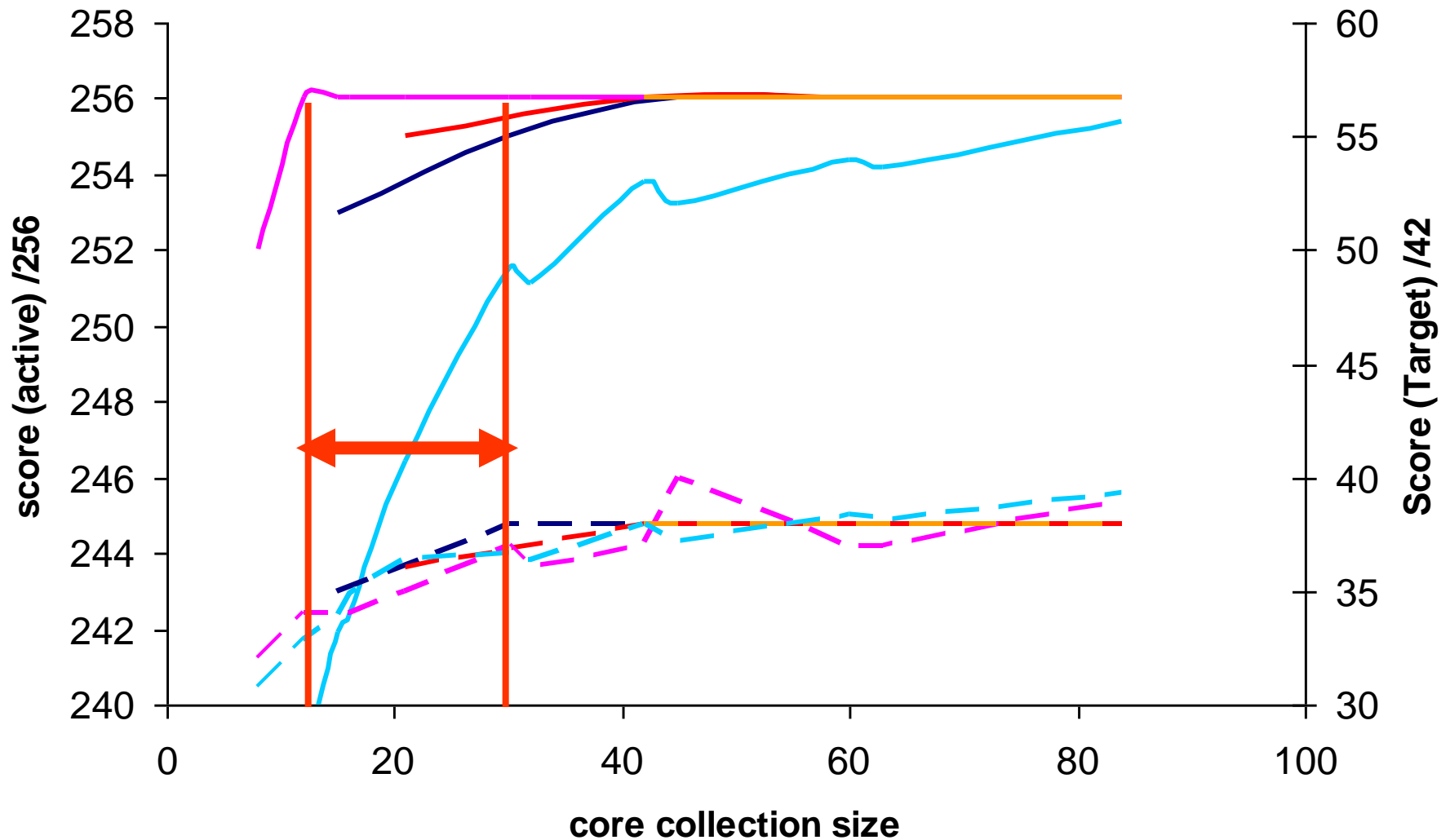
	C	P	L	M	R
8				X	X
15	X			X	X
16				X	X
21			X	X	X
30	X			X	X
32				X	X
42		X	X	X	X
45	X			X	X
60	X			X	X
63			X	X	X
84		X	X	X	X

# Allele capture with the different strategies



— C\_active — L\_active — M\_active — P\_active — R\_active

# Towards nested core collections



# Perspectives

- ▶ The presented results are preliminary
  - Completed with more SSR
  - Completed with targeted markers such as SNP defined in QTL regions.
  
- ▶ *B. napus* core collection in 2009

# collaborations

- Thanks to genbank curators who provided the seeds
- platform INRA Clermont-Ferrand / GAP Division
- WUR
- Vavilov Plant Institute
- Open to other collaborations



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# The team

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**Cyril Falentin**  
Gilles Boutet

Geoffrey Wagner  
Anthony Ollivier

**Anne Laperche**  
Michel Renard  
Maria Manzanares-Dauleux

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