



**TWC/28/28**

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**DATE:** June 22, 2010




**INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS**  
GENEVA

**TECHNICAL WORKING PARTY ON AUTOMATION AND  
COMPUTER PROGRAMS**

**Twenty-Eighth Session**  
**Angers, France, June 29 to July 2, 2010**

COMBINATION OF MORPHOLOGICAL DISTANCE (GAIA) WITH GENOTYPIC  
DISTANCE IN THE FRAMEWORK OF "MANAGEMENT OF THE REFERENCE  
COLLECTION"


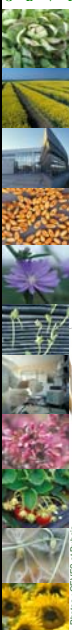

*Document prepared by experts from France*



Combination of morphological distance (GAIA)  
with genotypic distance in the framework of  
«management of the reference collection »

**UPOV –TWC/28**  
Angers, France, June 29 to July 2, 2010

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**Context**  
Framework of the inscription to the catalogue or legal protection

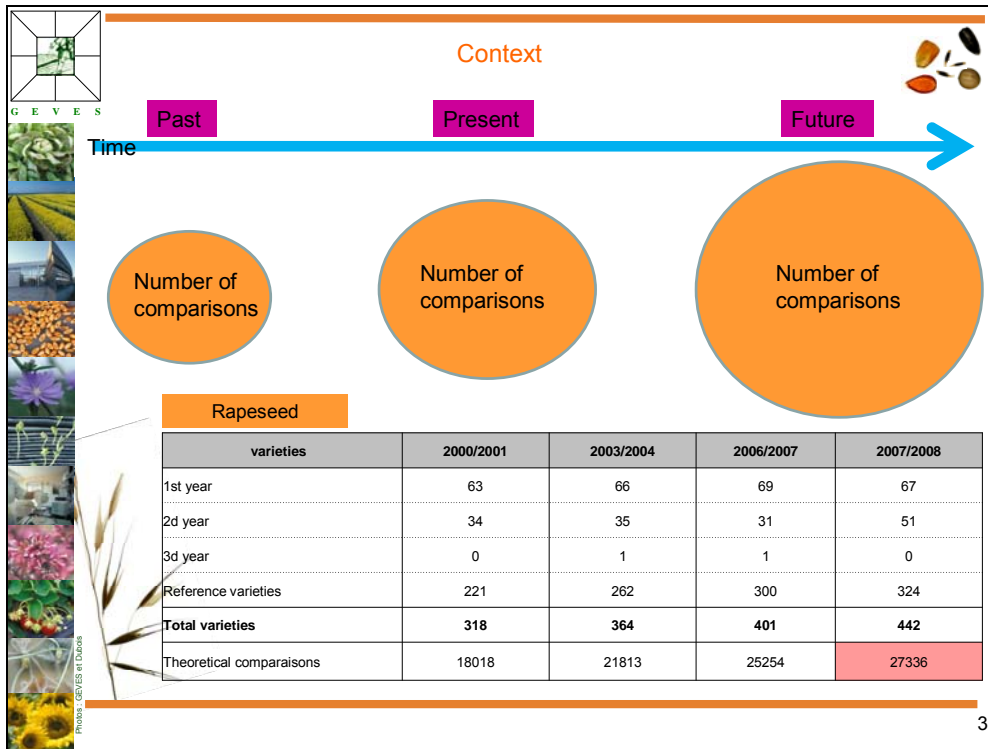
Quality of distinctness for new varieties

Comparison of all candidate varieties with each other

Comparison with other relevant varieties of common knowledge = Reference collection

Number of comparisons

2




**Context**


To maintain the high level of quality of the distinctness assessment, we need to find tools and procedures :


- To handle a great number of varieties = Huge number of comparisons
- Avoid prohibitive costs and lack of land for trials
- Avoid lengthening the duration of the tests

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## Context





Photos: GEVES et DUBOIS

**Main changes over the past:**

- development of the GAIA software to select the varieties which need to be grown in the field trials
- integration of characteristics derived from electrophoresis in combination with field characteristics (For maize only)


**Next step:**

- integration of genetic distances in combination with phenotypic characteristics to assess distinctness


**This combined approach** supposes prerequisites :


- Set of appropriate markers
- Choice of an appropriate distance
- Materials and protocols (Repetability and Reproducibility)
- Free acces of markers (No royalties)

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## Context





Photos: GEVES et DUBOIS


**The methodology:**

- Characterize the reference collection using SSR markers
- Define 2 new threshold values for the morphological and biomolecular (BM) distance based on the DUS expert appreciation
  - a BM distance treshold, below which BM data are not considered
  - a GAIA threshold, below which BM data are not considered
- Combined these distances to improve the management of DUS trials

Species on which experience is available in France : Maize, Spring barley, Peach, lettuce and rapeseed

For this presentation I will take the example of SPRING BARLEY

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### Morphological distance estimated by GAIA


Software GAIA was developed by GEVES to optimize the distinctness tests while using knowledge of the crop experts in distinctness.

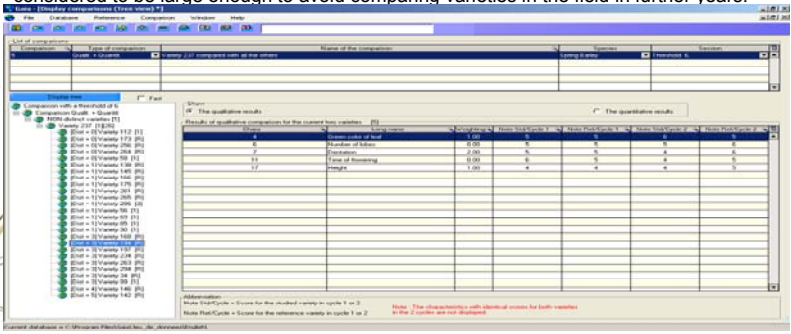
The principle of the software is to add the differences observed characteristic by characteristic, differences weighted by intensity and reliability of each characteristic.  
 Nb: Observed differences have a zero weight if not considered no reliable by the crop experts.


Varieties are compared pair-by-pair, and two varieties are declared "GAIA-distinct" if the addition of weightings (phenotypic distance) according to differences is greater than a threshold value (distinction threshold= Sdist) defined by the crop expert.

If two varieties are declared "GAIA-distinct", it means the phenotypic distance is considered to be large enough to avoid comparing varieties in the field in further years.



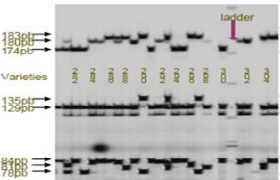




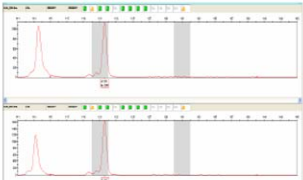



### Genotypic distance computed by NEPER (GEVES)

**Licor**



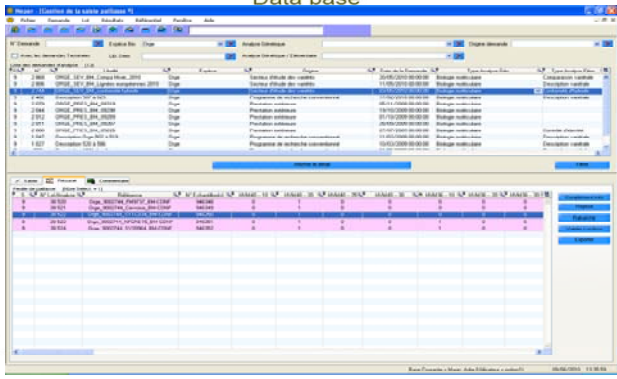
**ABI capillary system**




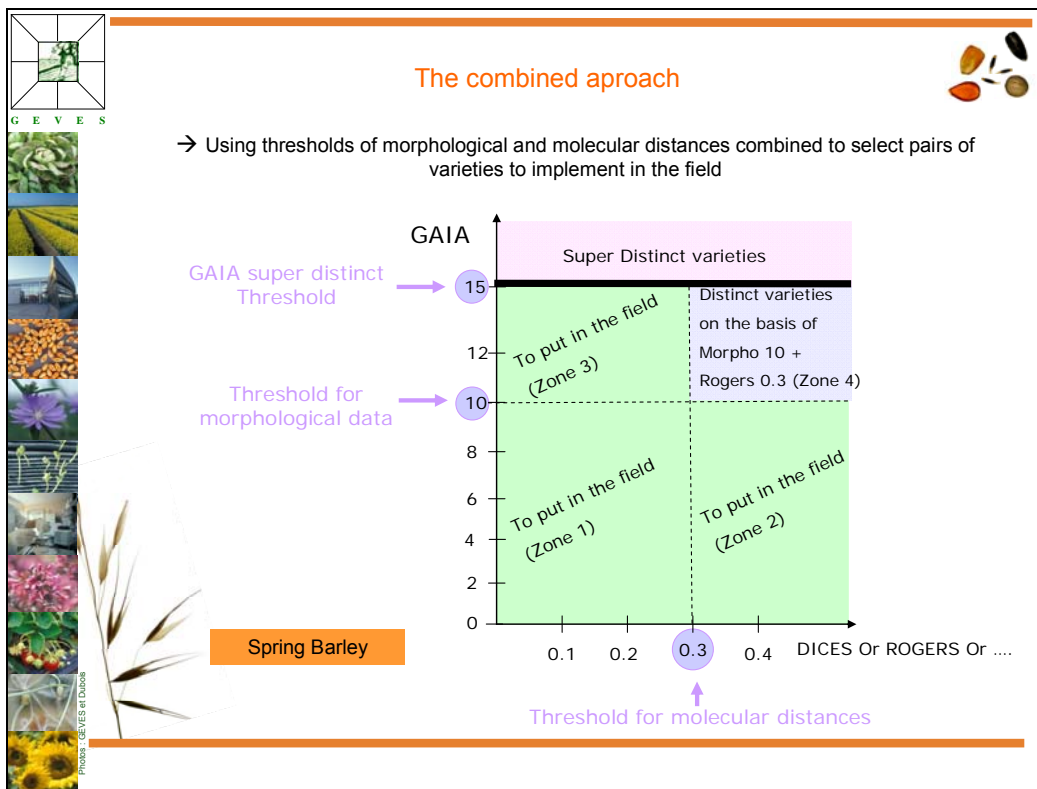
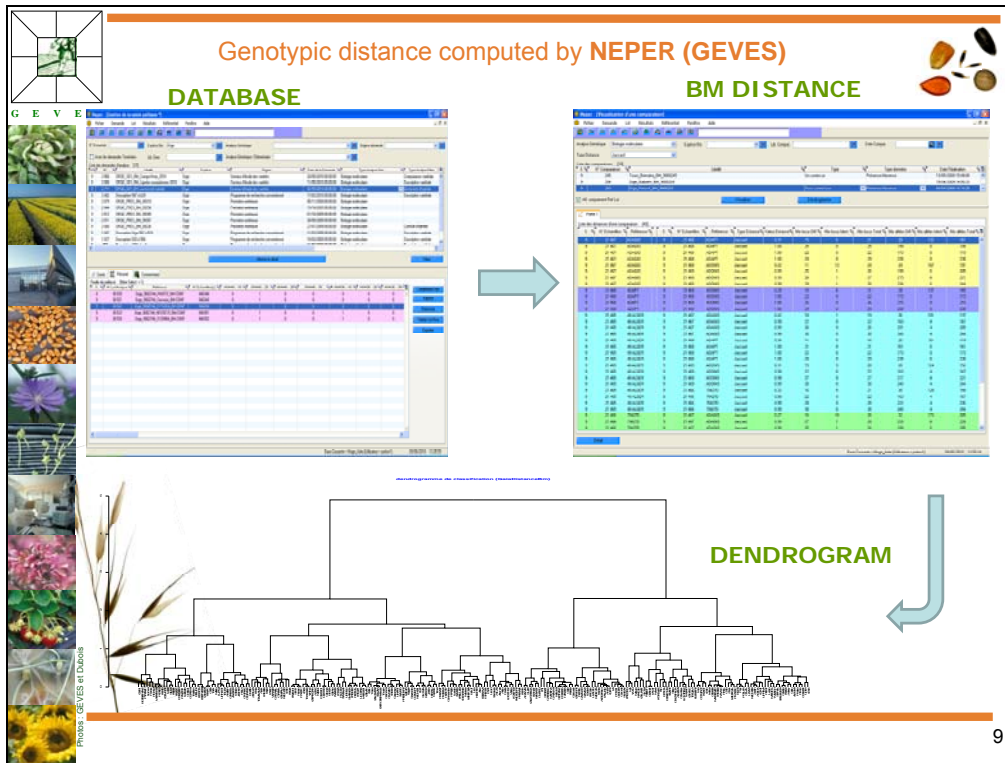



↓ Scoring ↓

↓ Data base ↓








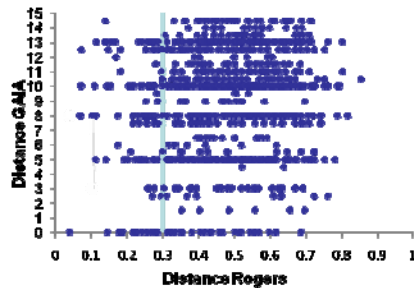


GEVES




Photos: GEVES et Dabois

### Correlation between molecular and morphological data ?

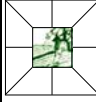


Spring Barley




= There is no relationship between the two distances used  $\Rightarrow$  how to define an appropriate way of integrating molecular data into the decision ?

We set special experiments to observe the degree of similarities/differences between varieties, and compare it with the molecular distances




GEVES



Photos: GEVES et Dabois

### Protocol of the expert experiment

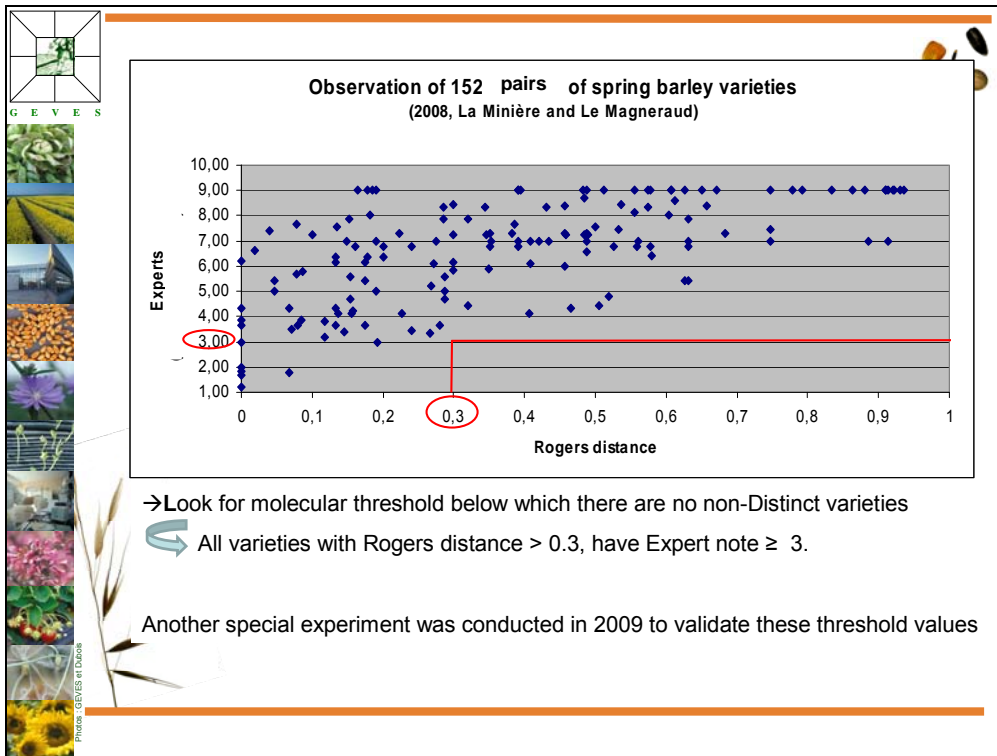


- **Material** : 500 spring barley varieties
- **Markers** : 30 SSR markers, 234 alleles, High polymorphism, Good level of uniformity
- **Field design** : couple of varieties side-by-side on 2 sites (Le Magneraud / La Miniere)
- **Visual assessment realized by 11 experts (Geves + Breeders)**

**Scale of similarity:**

1. the two varieties have similarities
3. the two varieties are distinct but close
5. the comparison was useful, but the varieties are clearly distinct
7. the comparison should have been avoided because the varieties are very different
9. the comparison should have been avoided because the varieties are totally different

$\rightarrow$  These expert scores are compared to molecular distances



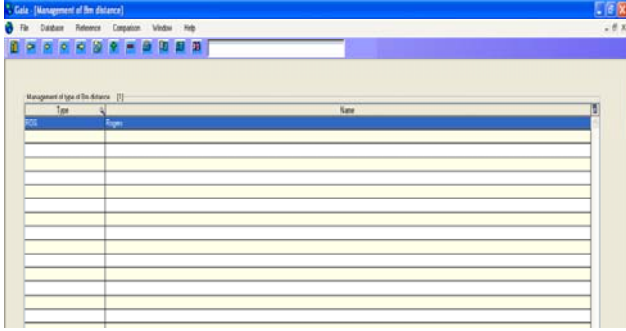
- ### Conclusion of these combined approach on spring barley
- ❑ The threshold values have been fixed :
    - Morphological treshold = GAIA 10
    - Molecular treshold = ROGERS 0.3
  - ❑ The reference collection is fully characterized
  - ❑ UPOV has validated in principle the combined approach
  - ❑ A number of TWC documents are available This is not a full list  
BMT/8/14, BMT/8/14 Add., TWC/20/2, BMT/4/7 Rev. 2, TWC/14/18, TWC/14/8, TWC/14/15



Implementation of the combined approach through Gaia software :

**How to integrate biomolecular data?**

Before integration, you have to define types of distance that you want to manage through the window «Management of Bm distance »



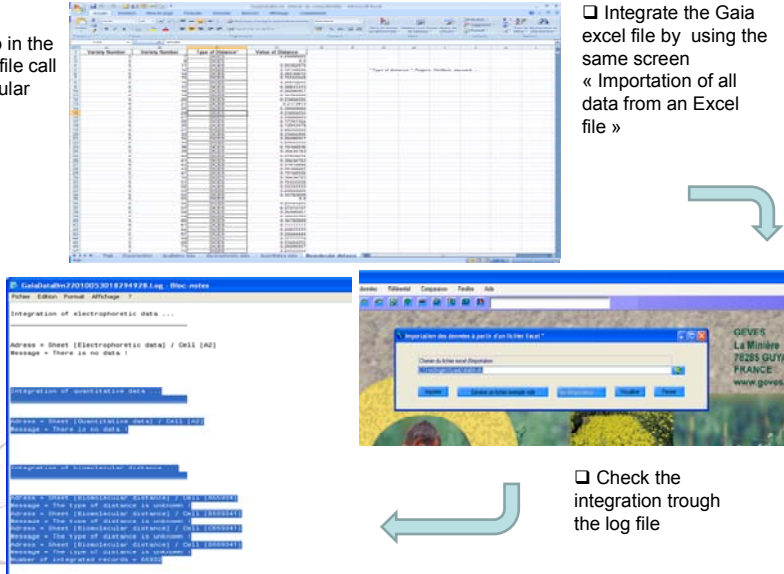
Photos: GEVES et Dubois

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Implementation of the combined approach through Gaia software :

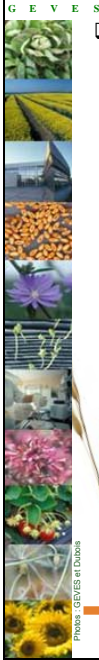
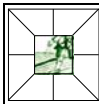
**How to integrate biomolecular data?**

- ❑ New Tab in the Gaia excel file call « Biomolecular Distance »
- ❑ Integrate the Gaia excel file by using the same screen « Importation of all data from an Excel file »
- ❑ Check the integration through the log file



Photos: GEVES et Dubois

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### Implementation of the combined approach through Gaia software :

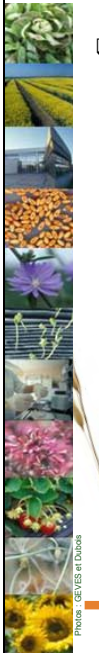
#### How to display biomolecular data?



❑ You can display Bm distance integrated into Gaia database by using the window « Management of data ».

N° Variety	Name of the variety	N° Variety	Name of the variety	Type	Distance
26_296	1980025.B	723	Provence	PROG	0.4300
26_296	1980025.B	722	Provence	PROG	0.5400
26_296	1980025.B	728	Wakigelt	PROG	0.5500
26_296	1980025.B	727	Levee	PROG	0.5700
26_296	1980025.B	726	Arctique	PROG	0.5750
26_296	1980025.B	741	Blanc	PROG	0.5800
26_296	1980025.B	742	Bonanza	PROG	0.5800
26_296	1980025.B	742	Estacion	PROG	0.4200
26_296	1980025.B	2.439	Medallona	PROG	0.5750
26_296	1980025.B	2.472	Bananae	PROG	0.4800
26_296	1980025.B	2.712	Fume	PROG	0.5750
26_296	1980025.B	2.713	Prunier	PROG	0.5700
26_296	1980025.B	2.806	Cyrien	PROG	0.7200
26_296	1980025.B	2.865	Irani	PROG	0.7300
26_296	1980025.B	2.869	Luzerne	PROG	0.7800
26_296	1980025.B	2.878	Bonaf	PROG	0.6800
26_296	1980025.B	2.879	Deja	PROG	0.4800
26_296	1980025.B	3.889	Chassane	PROG	0.6000
26_296	1980025.B	2.882	Luz	PROG	0.5100
26_296	1980025.B	2.883	Prunier	PROG	0.6200
26_296	1980025.B	2.929	Styrie	PROG	0.6200
26_296	1980025.B	2.936	Luzerne	PROG	0.5200
26_296	1980025.B	2.969	Provence	PROG	0.6900
26_296	1980025.B	2.969	Provence	PROG	0.6200
26_296	1980025.B	2.993	Provence	PROG	0.6000
26_296	1980025.B	2.994	Pila	PROG	0.4400
26_296	1980025.B	3.000	Provence	PROG	0.5100
26_296	1980025.B	3.001	Provence	PROG	0.5600
26_296	1980025.B	3.002	Provence	PROG	0.4900
26_296	1980025.B	3.006	Provence	PROG	0.4400
26_296	1980025.B	3.018	Provence	PROG	0.6100
26_296	1980025.B	3.023	Toussin	PROG	0.4800
26_296	1980025.B	3.216	Provence	PROG	0.4800
26_296	1980025.B	3.329	Styrie	PROG	0.5900
26_296	1980025.B	4.118	Provence	PROG	0.5800
26_296	1980025.B	4.480	Provence	PROG	0.4900
26_296	1980025.B	4.904	Provence	PROG	0.5200
26_296	1980025.B	4.926	Provence	PROG	0.5800
26_296	1980025.B	4.926	Styrie	PROG	0.5800

❑ From this window you can also display a dendrogram to see the variability of your distance




### Implementation of the combined approach through Gaia software :

#### How to declare a comparison which uses biomolecular data?




- ❑ You have to define through a session the 2 new threshold values :
- a BM distance threshold, below which BM data are not considered
  - a GAIA threshold, below which BM data are not considered

N°	Characteristic	Long name	Unit name	Type
1	1	1	1	1
2	2	2	2	2
3	3	3	3	3
4	4	4	4	4
5	5	5	5	5
6	6	6	6	6
7	7	7	7	7
8	8	8	8	8
9	9	9	9	9
10	10	10	10	10
11	11	11	11	11
12	12	12	12	12
13	13	13	13	13
14	14	14	14	14
15	15	15	15	15
16	16	16	16	16
17	17	17	17	17
18	18	18	18	18
19	19	19	19	19
20	20	20	20	20
21	21	21	21	21
22	22	22	22	22
23	23	23	23	23
24	24	24	24	24
25	25	25	25	25
26	26	26	26	26
27	27	27	27	27
28	28	28	28	28
29	29	29	29	29
30	30	30	30	30
31	31	31	31	31
32	32	32	32	32
33	33	33	33	33
34	34	34	34	34
35	35	35	35	35
36	36	36	36	36
37	37	37	37	37
38	38	38	38	38
39	39	39	39	39
40	40	40	40	40
41	41	41	41	41
42	42	42	42	42
43	43	43	43	43
44	44	44	44	44
45	45	45	45	45

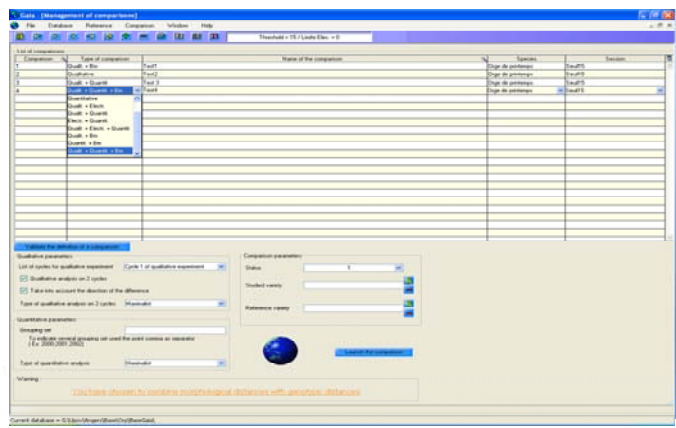


Implementation of the combined approach through Gaia software :


**How to declare a comparison which uses biomolecular data?**



- ❑ Define one of the three type of comparison that use Bm data (Quali + Bm, Quanti + Bm or Quali + Quanti + Bm)
- ❑ Select the session you wish to compute




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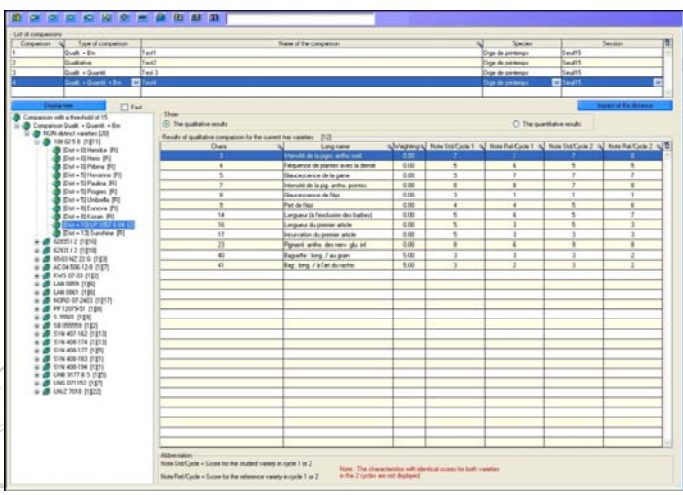


Implementation of the combined approach through Gaia software :


**How to display a comparison which uses biomolecular data?**



- ❑ You can display the comparison by using one of the two mode as usual : Tree view or Tabular view




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### Implementation of the combined approach through Gaia software :

#### How to display a comparison which uses biomolecular data?



☐ If you want to see graphically the impact of Bm distances, you have also the possibility to get data used by the following graph

Relationship between phenotypic distance and genotypic distance

Phenotypic Distance / Genotypic Distance

Genotypic distance


Phenotypic distance

- Varieties not distinguished (Below Morpho. Threshold & Below Genes. Threshold) (Zone 1)
- Varieties not distinguished (Below Morpho. Threshold) (Zone 2)
- Varieties not distinguished (Below Morpho. Threshold but Below Genes. Threshold) (Zone 3)
- Varieties distinguished (Zone 4)
- Morpho. Threshold
- Genes. Threshold
- GAIA upper defined threshold (Points above this line aren't showed on this graph)


Graph data


Zone	Index	Studied variety	Name of studied variety	Status	Reference variety	Name of side
4	1	26 382	NORD 07 2403	1	6 693	Gauche
4	2	26 382	NORD 07 2403	1	15 195	Gate
4	3	26 382	NORD 07 2403	1	17 286	Poet
4	4	26 382	NORD 07 2403	1	17 291	SW Mito
4	5	26 382	NORD 07 2403	1	743	Salon
4	6	26 382	NORD 07 2403	1	5 295	Sebastian
4	7	26 382	NORD 07 2403	1	8 630	SW Verde
4	8	26 382	NORD 07 2403	1	8 976	Marber
4	9	26 382	NORD 07 2403	1	12 472	SW Arto
4	10	26 382	NORD 07 2403	1	14 930	Chudron
4	11	26 382	NORD 07 2403	1	22 893	SYN 407143
4	12	26 382	NORD 07 2403	1	22 896	S 16048
4	13	26 382	NORD 07 2403	1	22 916	MAQW 79162
4	14	26 382	NORD 07 2403	1	152 306	Phinea
4	15	26 385	SW 07 33	1	2 803	Phlog
4	16	26 388	AC 04 506 12 0	1	153 296	Haves
4	17	26 388	AC 04 506 12 0	1	3 001	Flageul
4	18	26 388	AC 04 506 12 0	1	5 127	Druha
4	19	26 388	AC 04 506 12 0	1	7 468	Mangal
4	20	26 388	AC 04 506 12 0	1	7 483	Troada
4	21	26 388	AC 04 506 12 0	1	10 393	Beulin
4	22	26 388	AC 04 506 12 0	1	12 810	Marthe
4	23	26 388	AC 04 506 12 0	1	17 500	Bolina
4	24	26 390	SYN 407 162	1	5 900	Nausange
4	25	26 390	SYN 407 162	1	12 835	Macore
4	26	26 390	SYN 407 162	1	17 791	Charol

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## THANK YOU FOR YOUR ATTENTION !





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