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BMT/3/6 Add.

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INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS

GENEVA

**WORKING GROUP ON BIOCHEMICAL AND MOLECULAR TECHNIQUES
AND DNA-PROFILING IN PARTICULAR**

Third Session

Wageningen, Netherlands, September 19 to 21, 1995

**THE ESTIMATION OF MOLECULAR GENETIC DISTANCES
IN MAIZE FOR DUS AND ED PROTOCOLS: OPTIMIZATION
OF THE INFORMATION AND NEW APPROACHES OF KINSHIP**

Addendum prepared by experts from France

Two corrected pages of BMT/3/6

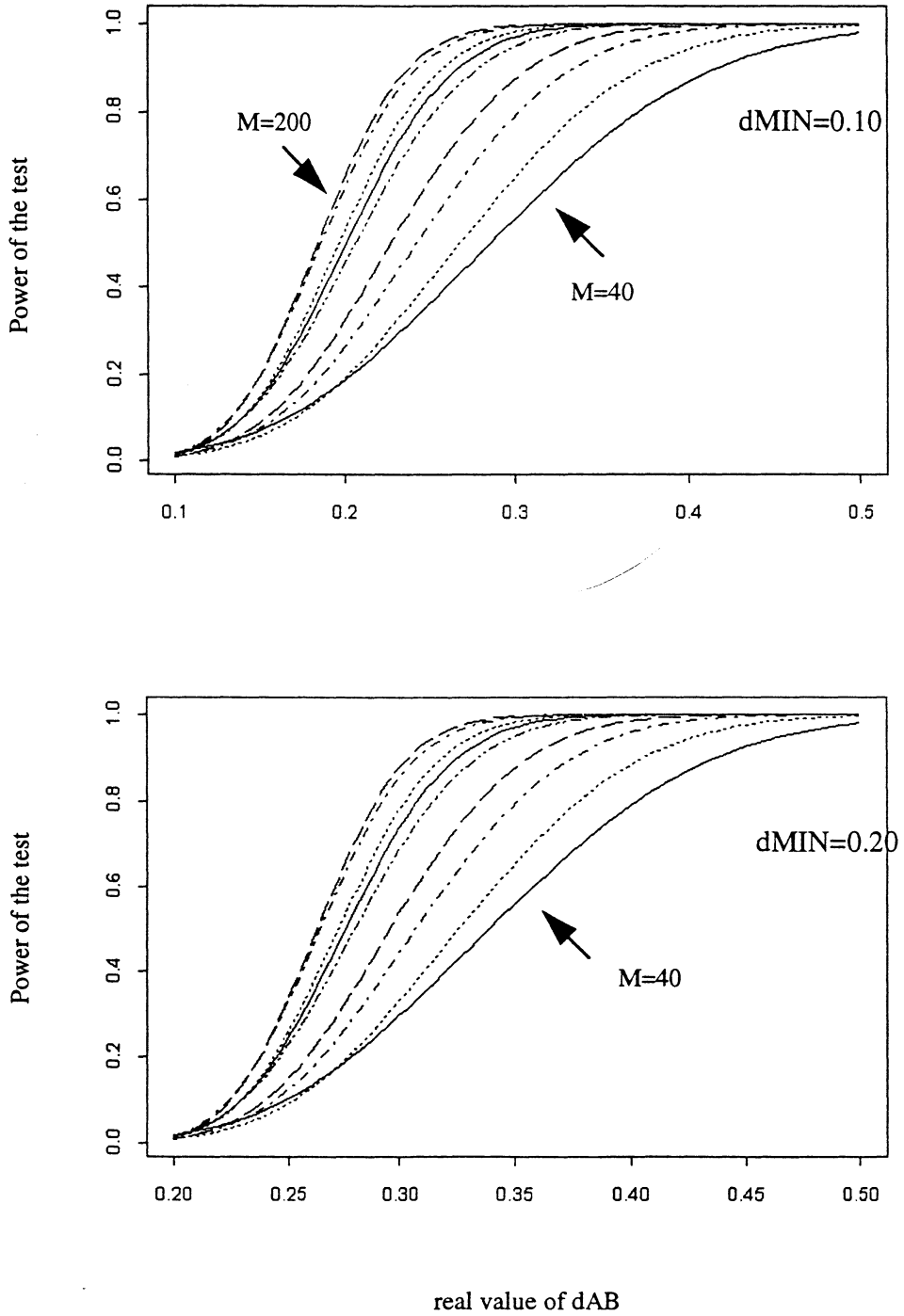


Figure 4 : Power of the test $d_{AB} < d_{MIN}$ at the 2% level for sample sizes ranging from 40 to 200 and for different values of d_{MIN} .

d_{MIN}	α	M	d_T
0.05	0.02	80	0.100
		160	0.087
0.10	0.02	80	0.175
		160	0.150
0.15	0.02	80	0.237
		160	0.212
0.20	0.02	80	0.300
		160	0.269
0.25	0.02	80	0.350
		160	0.319

Table 2 : Value of the effective minimum distance for the test of $H_0: d_{AB} \leq d_{MIN}$ with the Rogers distance.

Collection of Transparencies

**The estimation of molecular
genetic distances in Maize for
DUS and ED protocols**

**Optimisation of the information
and new approaches of kinship**

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B. Goffinet⁴, J. S. Smith⁵, Y. Dattée¹,
J. Guiard¹**

**Research program granted by the French
Ministry of Agriculture in the frame of a
C.T.P.S action.**

DUS = Distinction, Uniformity, Stability

ED = Essential Derivation

DUS = Distinction, Uniformity, Stability

ED = Essential Derivation

During this talk, I will speak about DUS for Distinction, Uniformity, Stability and about ED for Essential Derivation.

The estimation of genetic distances in Maize for DUS and ED protocols

1. Introduction
2. Taking the genetic map into account
 - 2.1 Position of the problem
 - 2.2 Estimation of the covariances between distances at marker loci
 - 2.3 Experimental results
3. The ED approach
 - 3.1 Distance index
 - 3.2 ED protocol
4. The DUS approach
 - 4.1 Distance index
 - 4.2 DUS protocol
5. Discussion

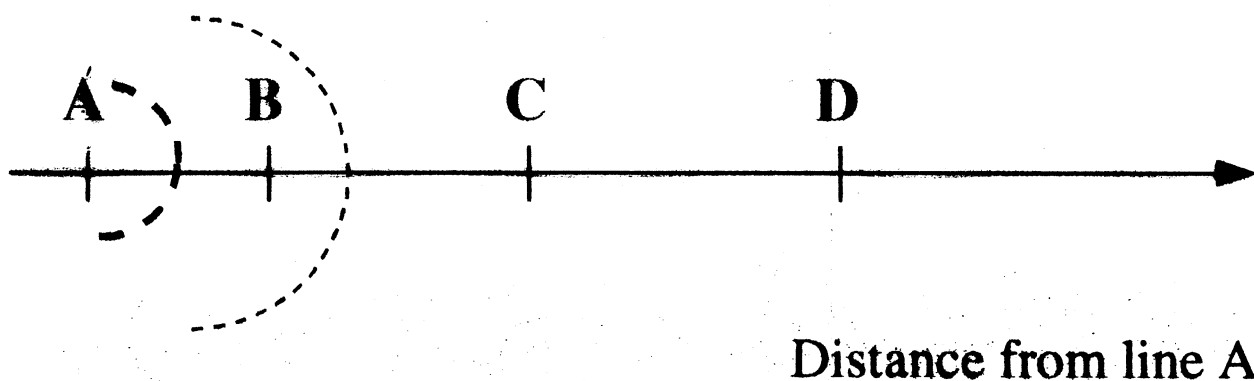
In Maize, a large number of molecular markers, located on the genetic map, are available.

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After a brief introduction, I would like to discuss the interest and the consequences of taking the genetic map into account, and apply it to the elaboration of new genetic distances. One specific for the ED approach, and one specific for the DUS approach.

Introduction



Distinction



Essential derivation

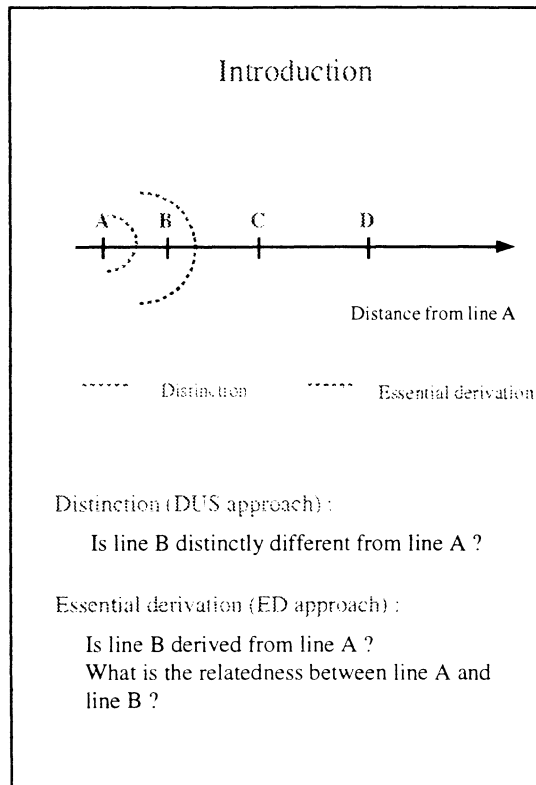
Distinction (DUS approach) :

Is line B distinctly different from line A ?

Essential derivation (ED approach) :

Is line B derived from line A ?

What is the relatedness between line A and line B ?



In Naize, both the DUS and ED approaches imply the comparison of two inbred lines, the line A and line B on the diagram. In the DUS approach, the question to answer is "Is line B distinctly different from line A?" while in the ED approach, the question to answer is "Is line B derived from line A?" or "What is the relatedness between lines A and B?". Both questions may be answered by computing a genetic distance between the two lines.

Introduction

The Rogers distance

L loci over the whole genome

M markers randomly sampled among the L loci

A and B = the two inbred lines to be compared

For each marker locus :

$d_{AB}^m = 0$ A and B identical at locus m

$d_{AB}^m = 1$ A and B are different at locus m

The Rogers distance

=

% of loci which differ between lines A
and B

among the M markers

Introduction

Statistical properties of the Rogers distance

They are a consequence of the following hypothesis :

**markers are randomly sampled
throughout the genome**

=

**no correlation between distances at
marker loci**

Introduction
Statistical properties of the Rogers
distance

They are a consequence of the
following hypothesis :

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=

no correlation between distances at
marker loci

I would just like to recall here that those statistical properties are a consequence of the hypothesis that the markers are sampled at random throughout the genome - In this case, there is no correlation between distances at marker loci or, in other words, the distance between two lines at one locus is independent on the distance between the two lines at another locus, whatever the distance between the loci -

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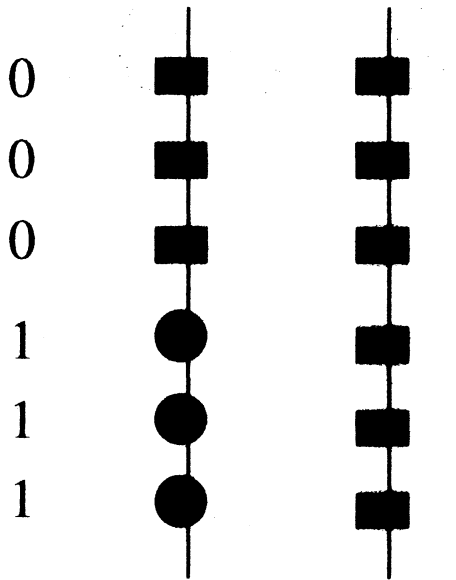
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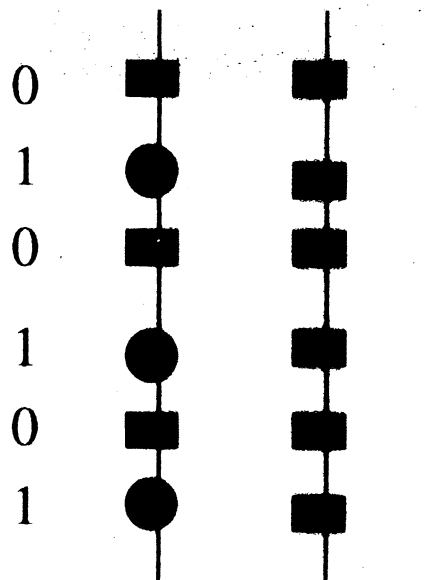
However, we've seen yesterday that a genetic map is available in more and more species like maize, tomato or maritime pine. In such species, once a set of markers has been chosen, it is often used repeatedly in different comparisons. Moreover, the genome coverage is not always perfect. For those reasons, and for other ones that I will detail now, it may be interesting to consider the genetic map as fixed and to reason conditionally to that given genetic map.

Taking the genetic map into account position of the problem

 d_{AB} 

line A

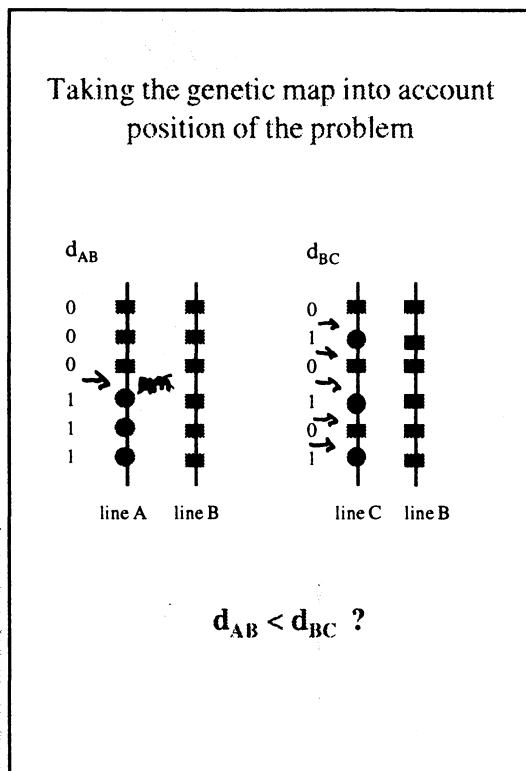
line B

 d_{BC} 

line C

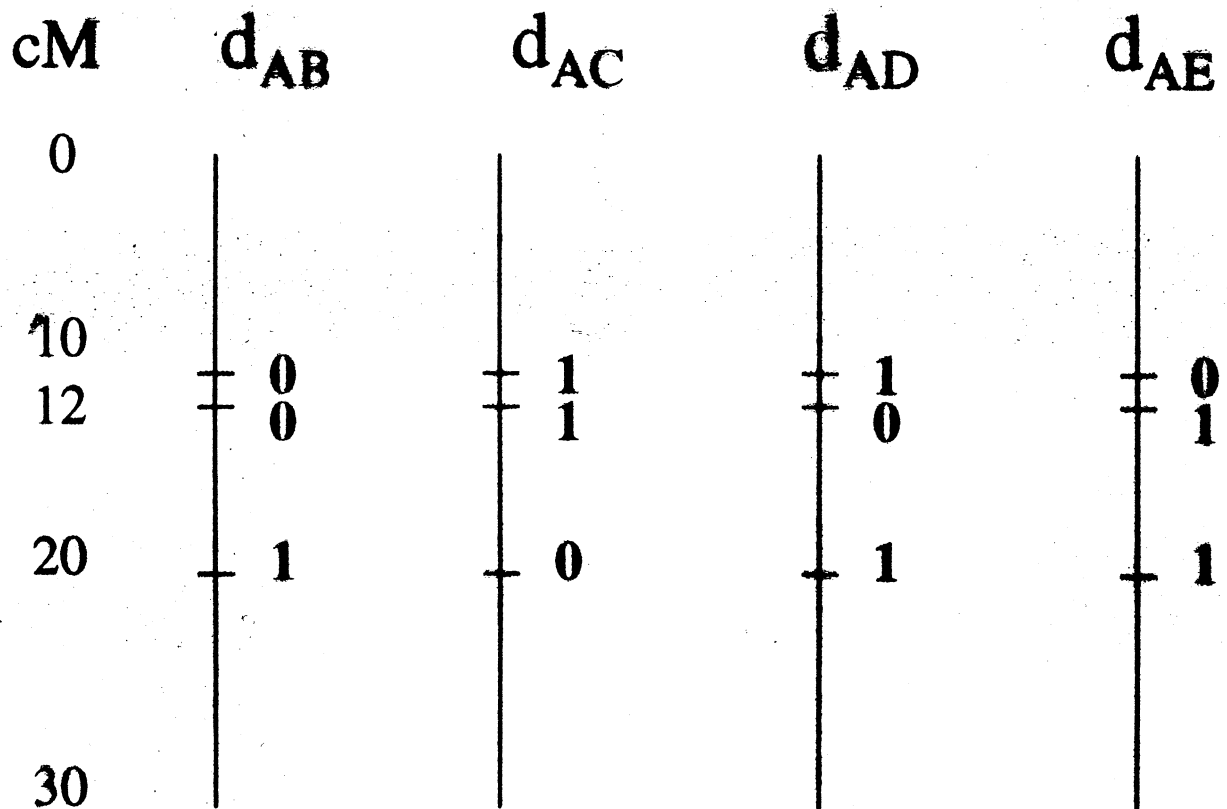
line B

$$d_{AB} < d_{BC} ?$$



Suppose, for example, that we have six markers, all situated on the same chromosome and that we want to compare line A and line B on this diagram. At each marker locus, there are two alleles, the green boxes and the red circles. Line A and B differ here by three markers out of six, and so do lines B and C. However, if we consider the differences in terms of recombination events, one recombination event is sufficient to give line A from an hybrid between line B and another line,

Taking the genetic map into account position of the problem



Rogers 0.33

0.66

0.66

0.66

Wished 0.5

0.5

>0.66

<0.66

Taking the genetic map into account

Position of the problem

Conditionally to a given genetic map,

If lines A and B are related, distances at marker loci are no more independent,

Therefore, finding the “best” estimator of the genetic distance
=
estimating the covariances between distances at marker loci.

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**Taking the genetic map into
account :**

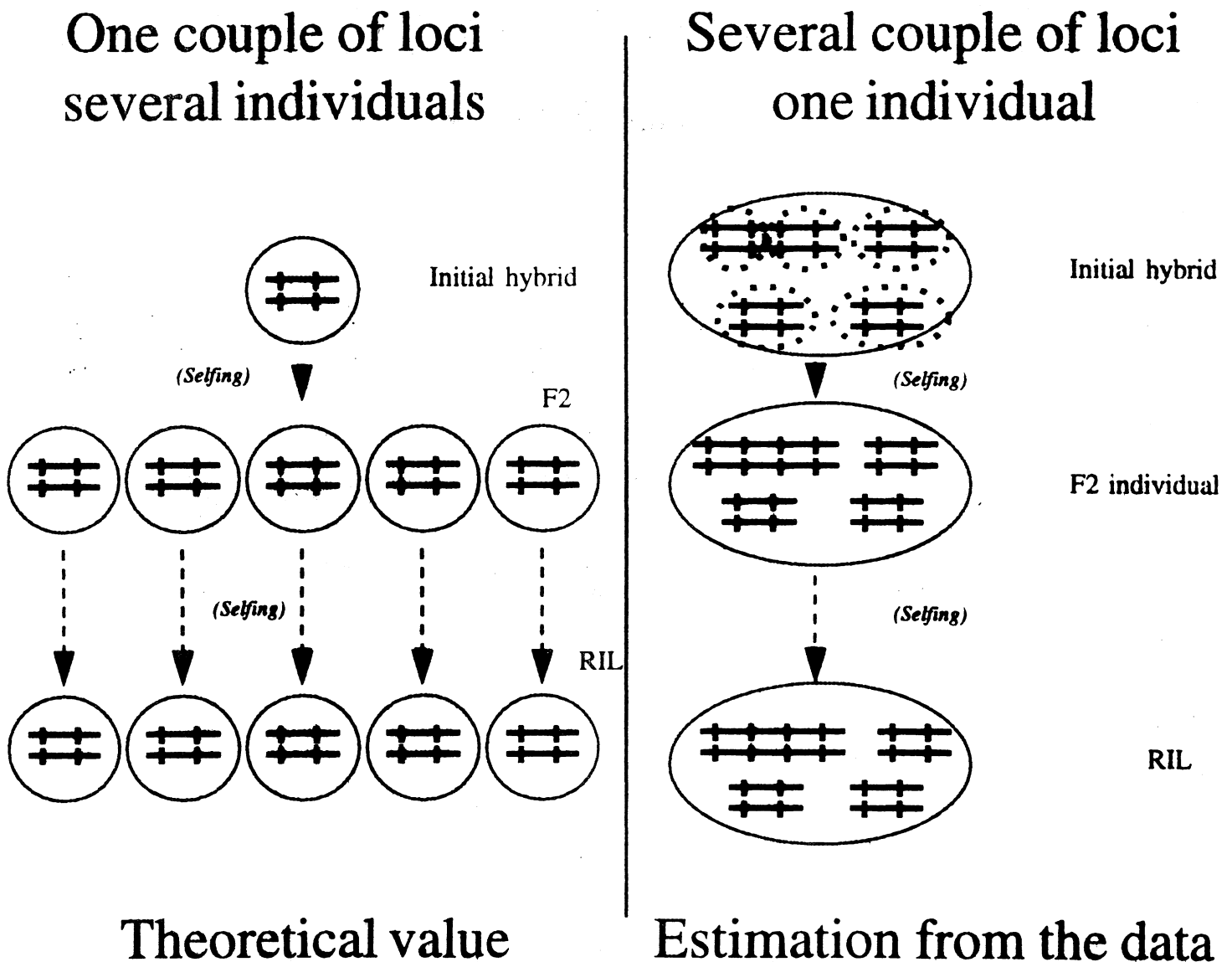
**Covariances between distances at
marker loci**

**How can two inbred lines be
derived each from another ?**

Example :

Selfing from an hybrid

The measurement of covariances between distances at marker loci



The observation of several couples of loci
within one individual brings valuable
informations about the genetical history of the

**The measurement of covariances
between distances at marker loci
estimation from the data**

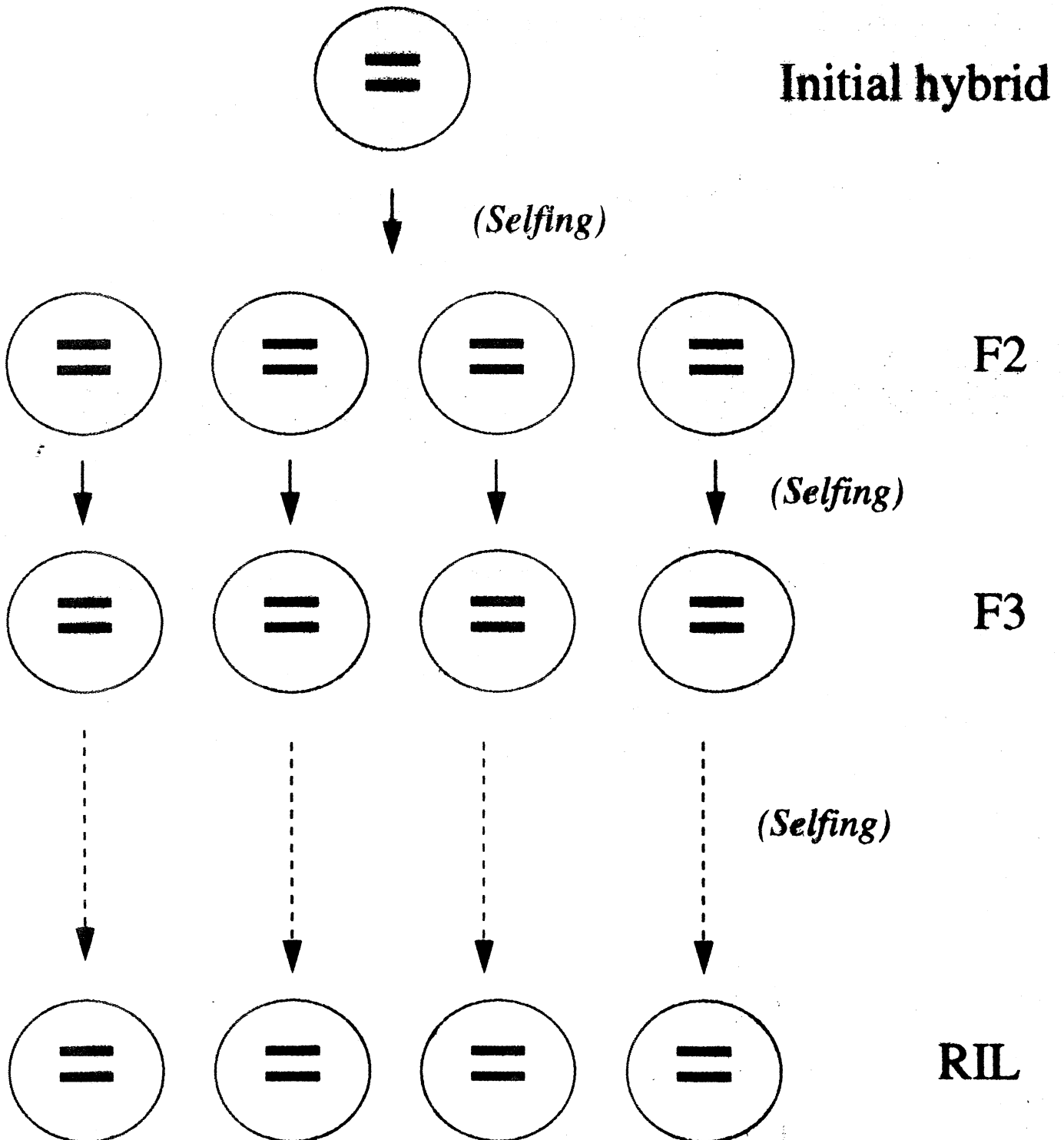
Data set

locus	1	2	3	4	5
cM	5	15	20	35	40
d_{AB}^k	0	0	1	0	1

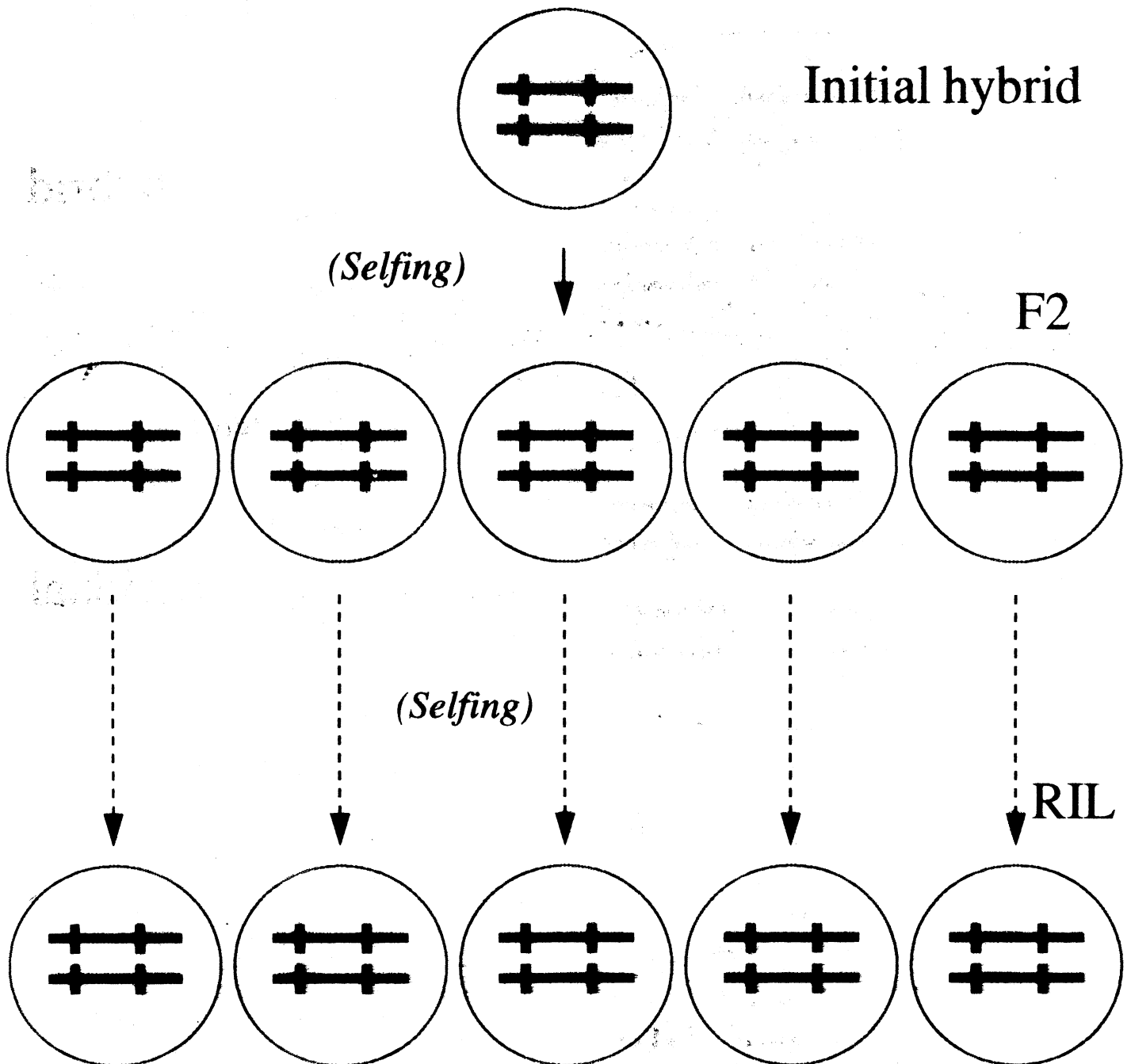
After rearranging

locus	locus	rec. rate	d_{AB}^k	d_{AB}^m
1	1	0	0	0
1	2	0.10	0	0
1	3	0.15	0	1
1	4	0.30	0	0
1	5	0.35	0	1
2	2	0	0	0
2	3	0.05	0	1

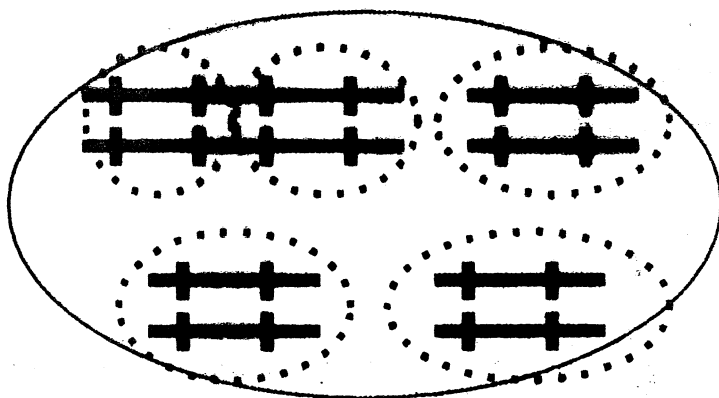
Segregation of one marker locus under selfing



Segregation of one couple of marker loci under selfing

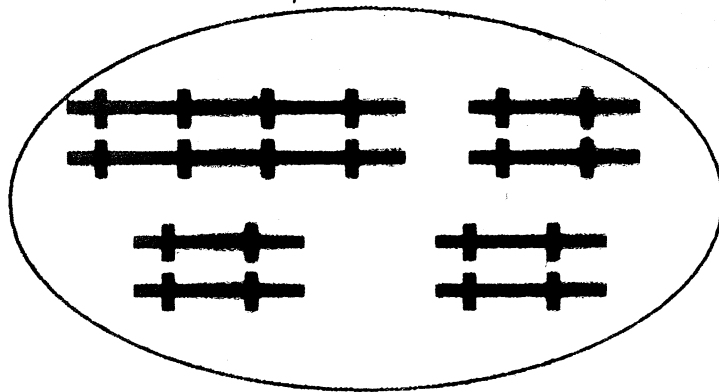


Segregation of five couples of marker loci within one individual under selfing



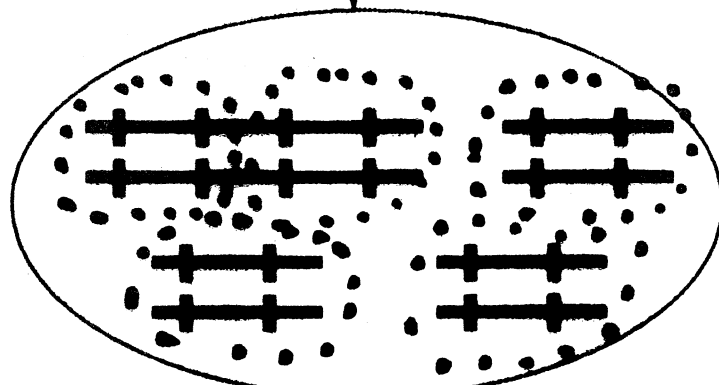
Initial hybrid

(Selfing)



F2 individual

(Selfing)



RIL

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The measurement of covariances between distances at marker loci

Experimental results

Data set 1 :

50 Maize Recombinant Inbred lines

derived by selfing from an hybrid
without selection.

Molecular data on 170 public RFLP
probes. (A. Charcosset)

Data set 2 :

**37 highly selected, elite inbred lines of
Maize from the central US Corn Belt.**

Molecular data on 110 public RFLP
probes. (S. Smith)

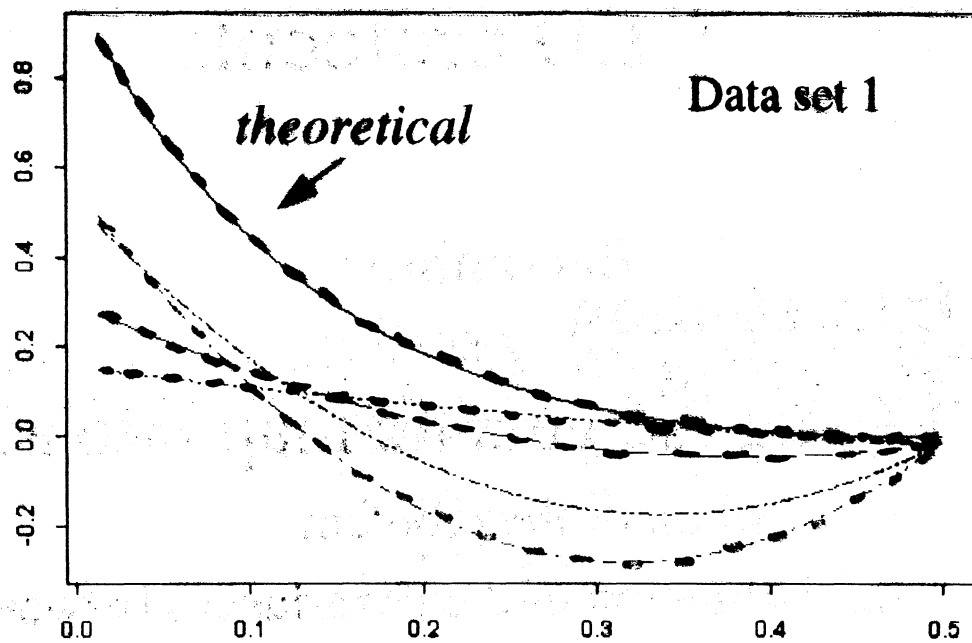
theoretical value = expected for

R II

The measurement of covariances between distances at marker loci

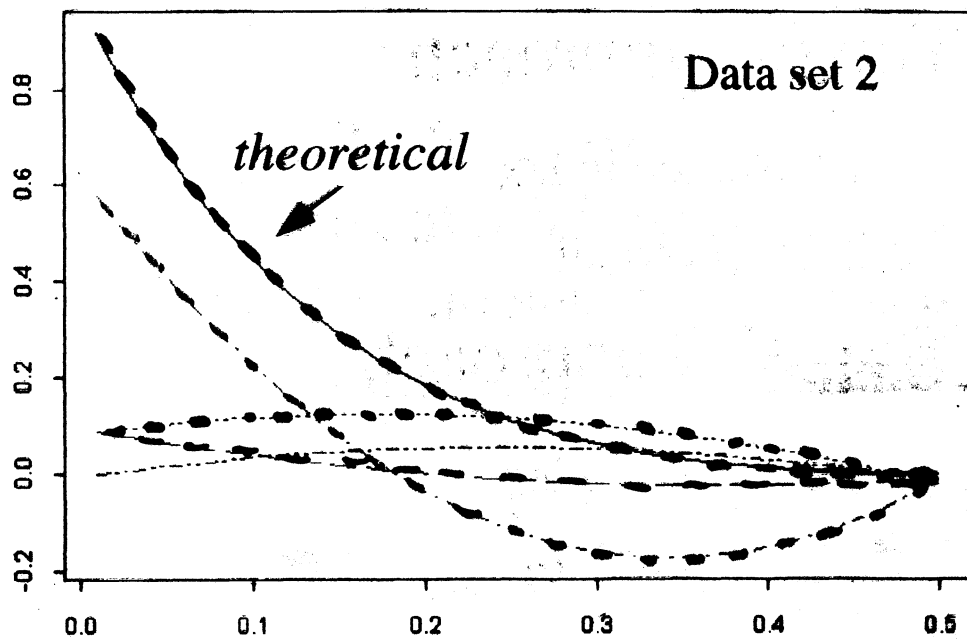
Experimental results

Correlation between distances at marker loci



Rogers

0.233
0.398
0.331
0.173



Rogers

0.318
0.518
0.482
0.145

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The ED approach

determination of the distance

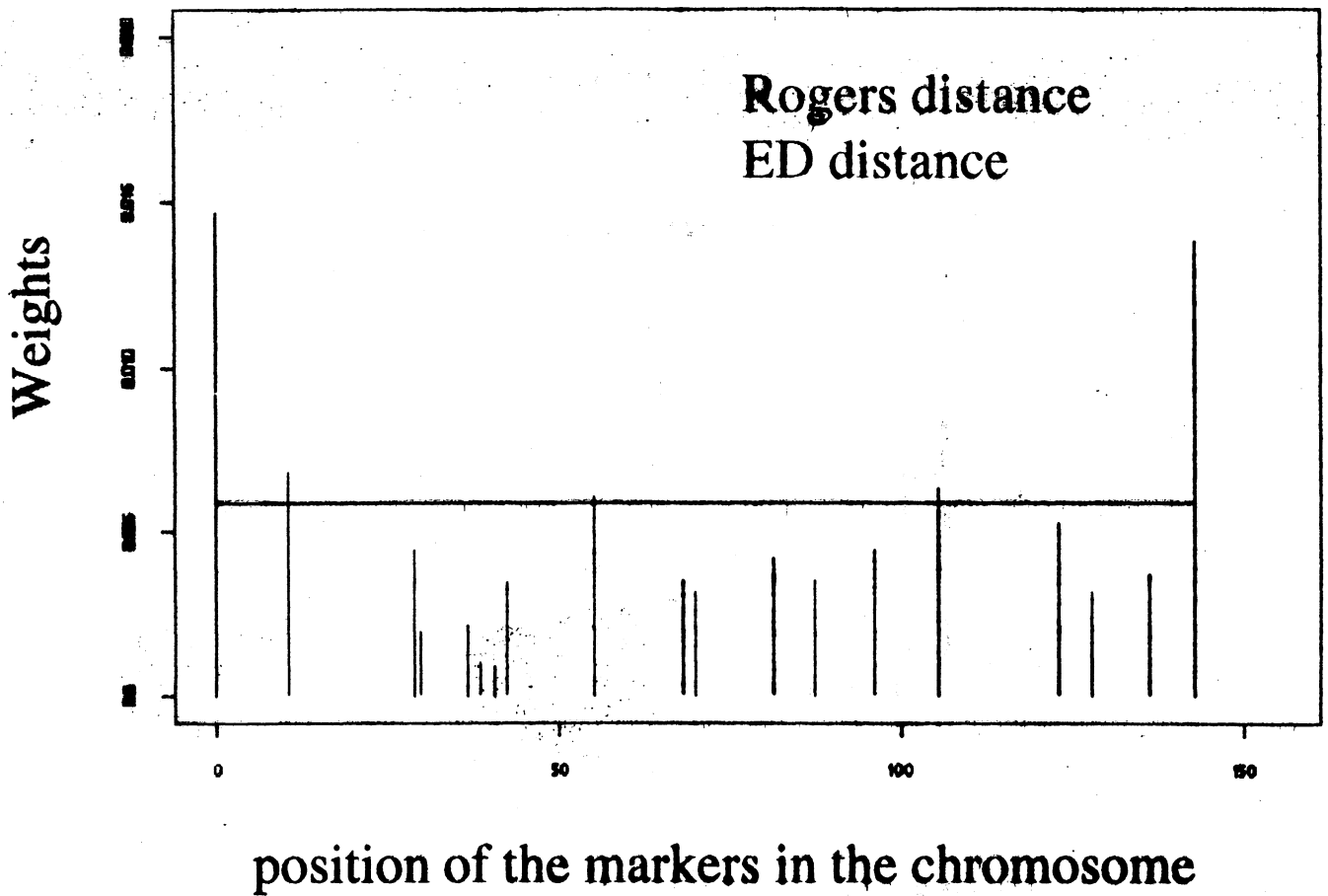
The ED distance

weights each marker locus in order to minimize the sampling variance, conditionally to the genetic map.

The weight of each marker locus is proportional to the covariances between this locus and all the other marker loci

The ED distance example

Weights of 17 Maize RFLP markers on chromosome 8



ED protocol

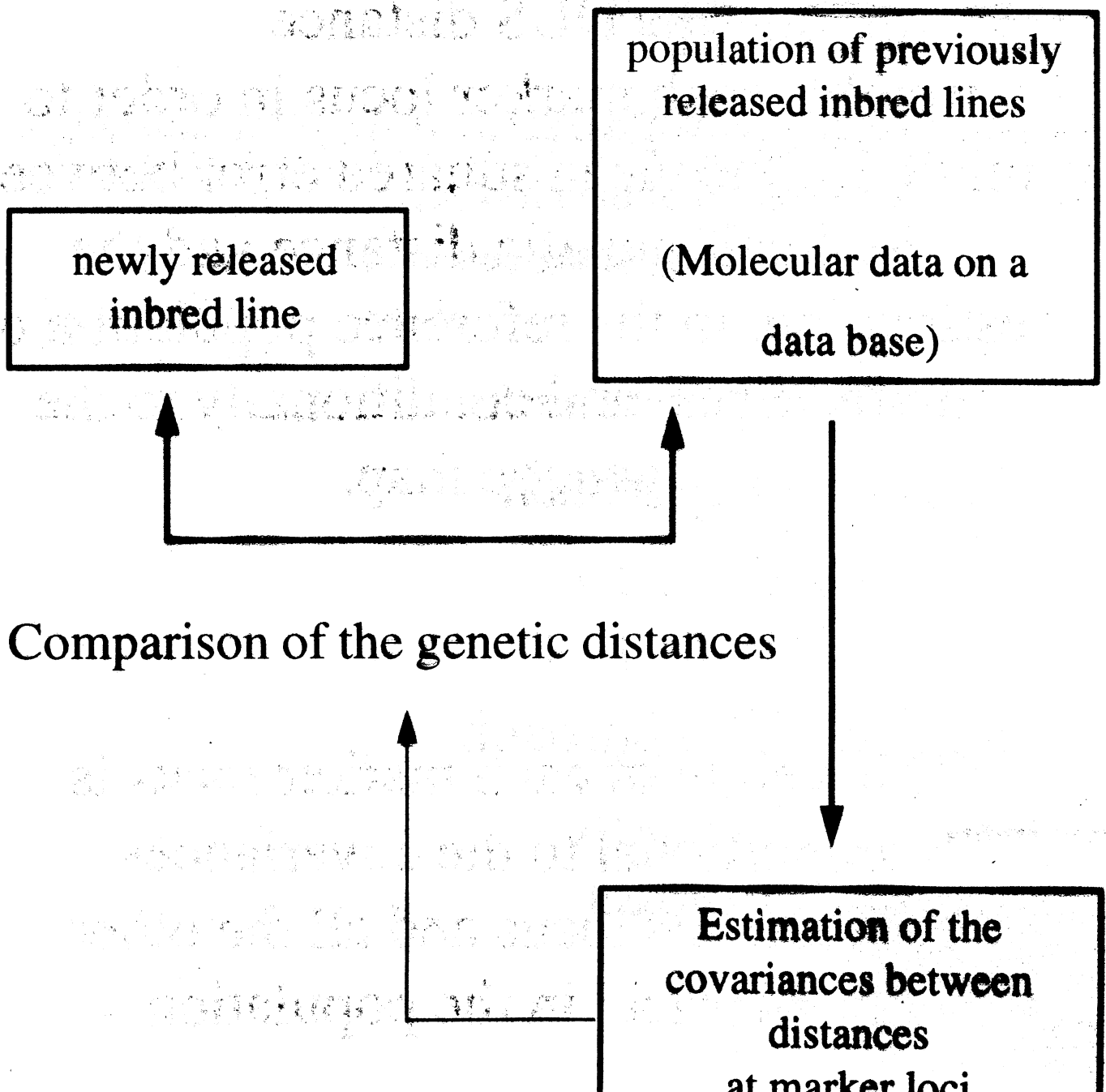
- (1) Choose an appropriate set of markers with known position on the genetic map
- (2) Compute the d_{AB}^k 's
- (3) Use the data to find the best suited value for the covariances between distances
- (4) Compute the ED genetic distance and its sampling variance by using (2) and (3).

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The DUS approach

determination of the distance



The DUS approach

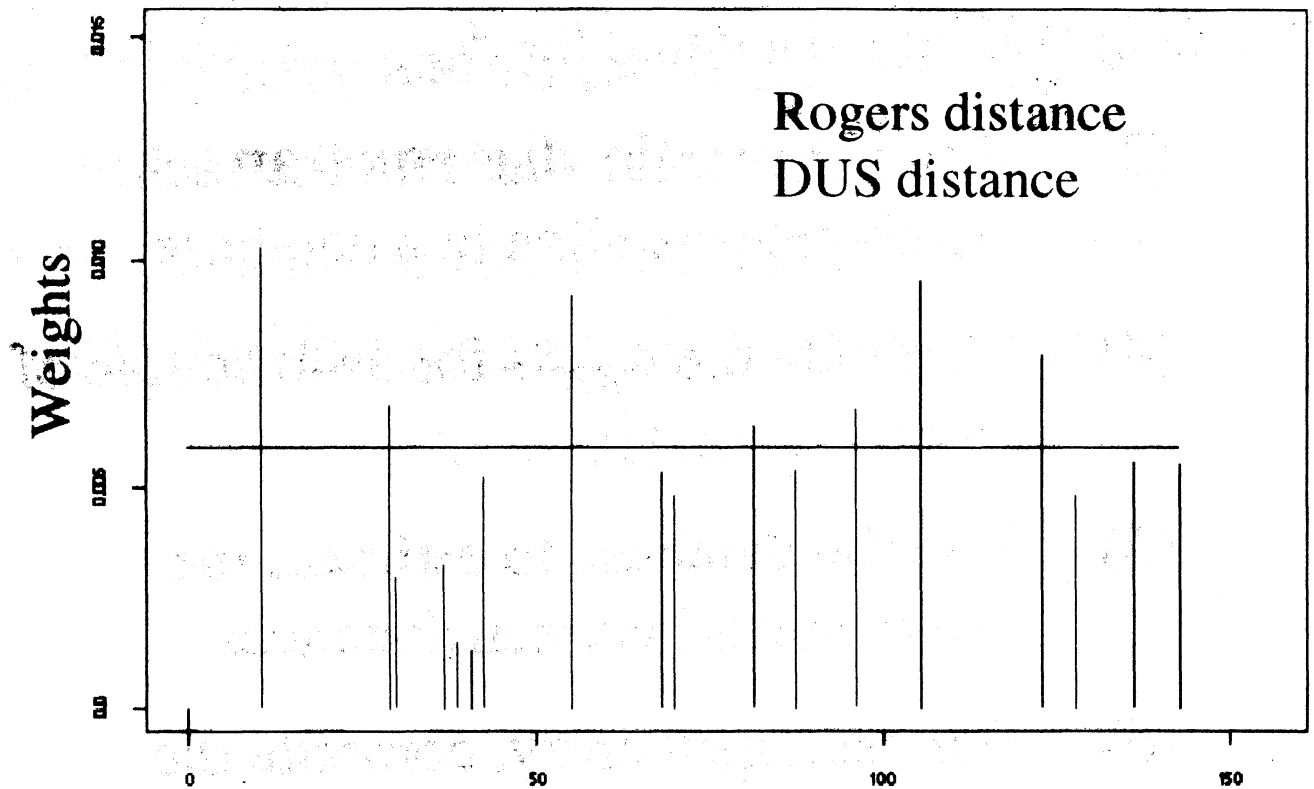
determination of the distance

The DUS distance
weights each marker locus in order to minimize the mean squared error between the "real" genetic distance and the estimation, in the reference population of inbred lines and conditionally to the genetic map.

The weight of each marker locus is proportional to the covariances between this locus and all the other marker loci in the population

The DUS distance example

Weights of 17 Maize RFLP markers on chromosome 8



position of the markers in the chromosome

population = RIL

DUS protocol

- (1) **Choose an appropriate set of markers** with known position on the genetic map
- (2) **Choose a value for d_{MIN}**
- (3) **Enter the molecular data for each newly released line in a database**
- (4) **Compute the d_{AB}^k 's for each couple of lines**
- (5) **Use the database to estimate the covariances between distances**
- (6) **For each new entry, compute the DUS genetic distance and its sampling variance by using (4) and (5)**
- (7) **Test $H_0 : d_{\text{AB}} < d_{\text{MIN}}$**

Discussion

Comparison of the three distances

