How to use GeneClass2?



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Please cite this reference if you use GeneClass2:

Piry S, Alapetite A, Cornuet, J.-M., Paetkau D, Baudouin, L., Estoup, A. (2004) GeneClass2: A Software for Genetic Assignment and First-Generation Migrant Detection. *Journal of Heredity* **95**:536-539.

WARNING: For reasons of international compatibility, the decimal separator used in GeneClass2 is a dot ".".

In order to use GeneClass2, your dataset(s) must be in a recognizable format. The file formats that are

recognized are: GenePop (alleles coded with 2 or 3 digits, or haploid) (Raymond & Rousset, 1995; see also "Genepop on the web"), FStat (Goudet, 1995), Genetix (Belkhir et al.), plus a XML (eXtended Markup Language) based format and an internally-used format (CIRAD-PRN). Please consult these references for detailed information on the file formats.

Note that it is preferable that the datafile contains as few missing data as possible (see Piry et al., 2004 for missing data management).

The language (French or English) can be choosen with the "Language" menu of GeneClass2.

It is worth noting that for most options chosen in the program a reference is indicated in a window: it is recomended to consult such references when detailed information are needed.

The following links provide more detailed instructions on the use of GeneClass2.

- 1. Detection of migrants
- 2. Assignment of individuals (or groups of individuals)
- 3. Data file conversion and description of population diversity

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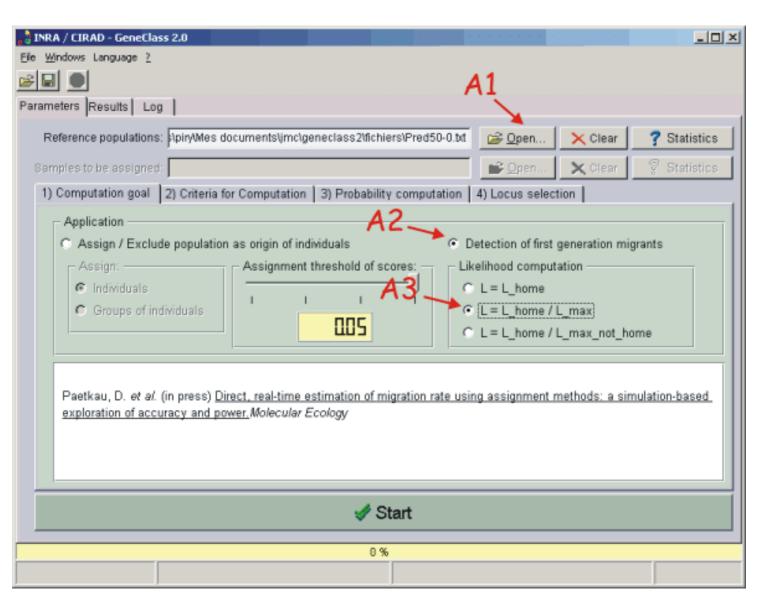
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- CBGP Website
- CBGP software Website

1) Detection of first generation migrants

Migrant detection needs a single data file that includes both the populations for which migrants will be seeked and the potential source populations for migrants.

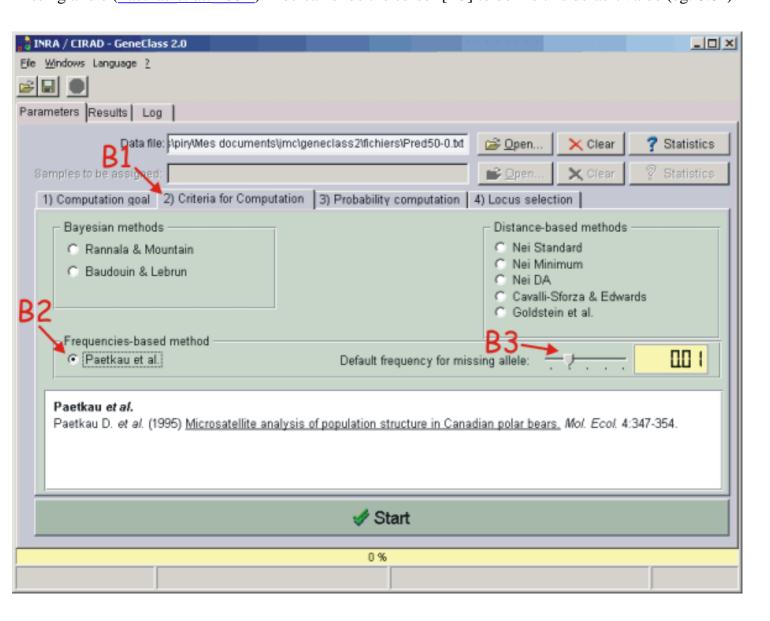
Start GeneClass2 from your "Start" menu, folder "CBGP". The splash-screen appears, followed by the main window of GeneClass2.

Load a datafile by clicking on the "Open" button and choose a datafile in the selector [A1]. Choose the "Detection of first generation migrants" option in the first tab of the main window [A2]. Select the type of likelihood computation to use for migrant detection. For instance select " $L = L_home / L_max$ " which is the ratio of the likelihood computed from the population where the individual was sampled (L_home) over the highest likelihood value among all population samples including the population where the individual was sampled (L_max) [A3] (see Paetkau *et al.* 2004).



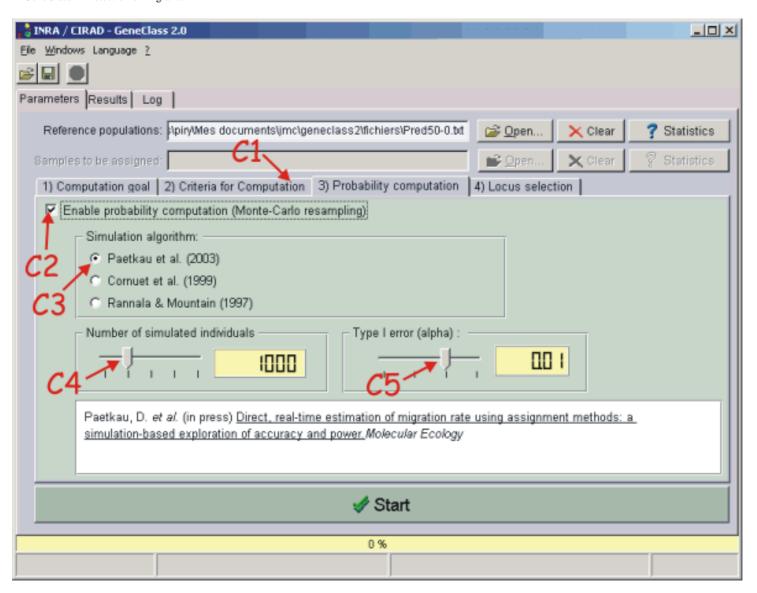
By clicking on the "2) Criteria for Computation" tab [B1] you can now select the criterion that will be used for likelihood computations. Bayesian and Frequencies-based methods appears to be better than distance-

based methods (see <u>Cornuet et al. 1999</u> for a comparative study). Let's for instance select <u>Paetkau et al.</u> (1995)'s criterion [**B2**]. This criterion needs a specific parameter which is the default frequency in the case of missing allele (<u>Paetkau et al. 2004</u>). You can slide the cursor [**B3**] to define this default value (eg. 0.01).



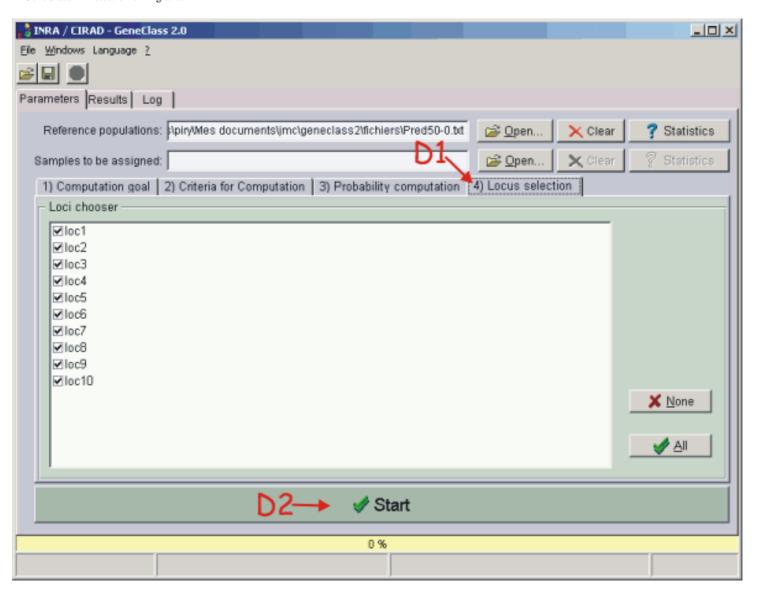
If you want to compute the probability that an individual is a resident (i.e. not a first generation migrant), click on the "3) Probability computation" tab [C1], and then check the "Enable probability computation (Monte-Carlo resampling)" box [C2].

You can now choose a resampling algorithm [C3] (eg. Paetkau *et al.* 2004; recommended for first generation migrants detection, but see Rannala & Mountain (1997) and Cornuet *et al.* (1999)). Slide the cursors to define the minimum number of simulated individuals [C4] (eg. 1000, default value or 10000 leading to a ten times longer but more precise computation), and the "Type I error (alpha)" cursor [C5] (eg. 0.01, default value, see Cornuet *et al.* (1999); Paetkau *et al.* (2004)):



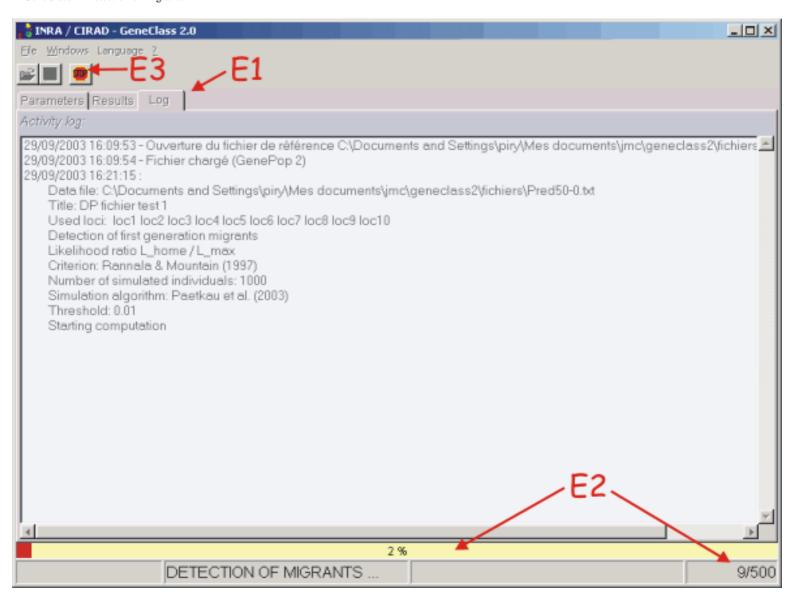
If needed, you can deselect some loci in the "Loci selection" tab [D1]. Deselected loci will be ignored during calculations.

Finally, click on the "Start" button [D2] to run the computation.

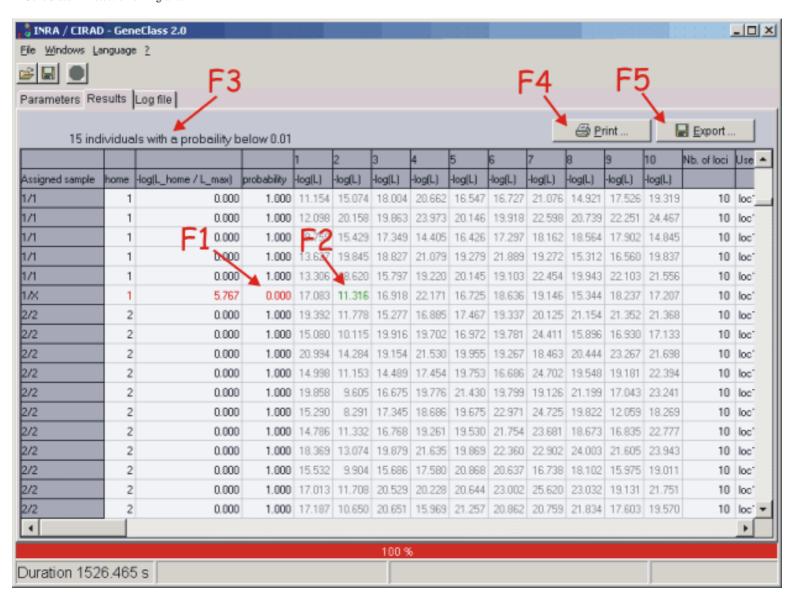


The program displays the "Log file" [E1] that displays the running parameters. The progress bar and a counter shows the state of the computation [E2].

The "Stop" button [E3] allows the computation to be aborted.



Once computations are finished, the results are displayed in a grid where potential F0 migrants (<u>Paetkau et al.</u>, 2004) are labelled in red (p < threshold) [F1] and the most likely population in green [F2]. The number of individuals with a probability below the threshold value is also indicated [F3]. Results can be printed ("Print" button [F4]) or exported in csv format ("Export" button [F5]).



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2) Assignment or exclusion of individuals (or groups of individuals)

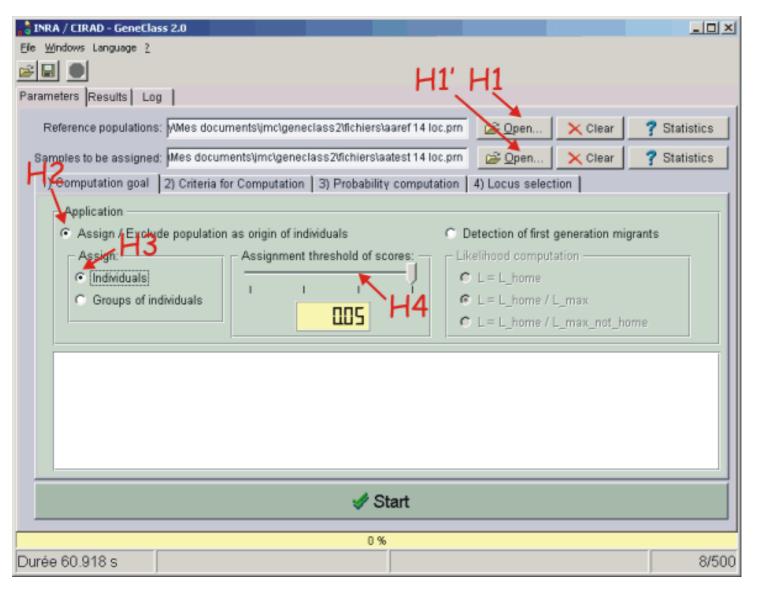
Start GeneClass2 from your "Start" menu, folder "CBGP". The splash-screen appears, then the main window of GeneClass2.

Load a reference datafile by clicking on the upper "Open" button [H1] and choose the datafile in the selector. Load a to-be-assigned individuals datafile by clicking on the lower "Open" button [H1'] and choose the datafile in the selector.

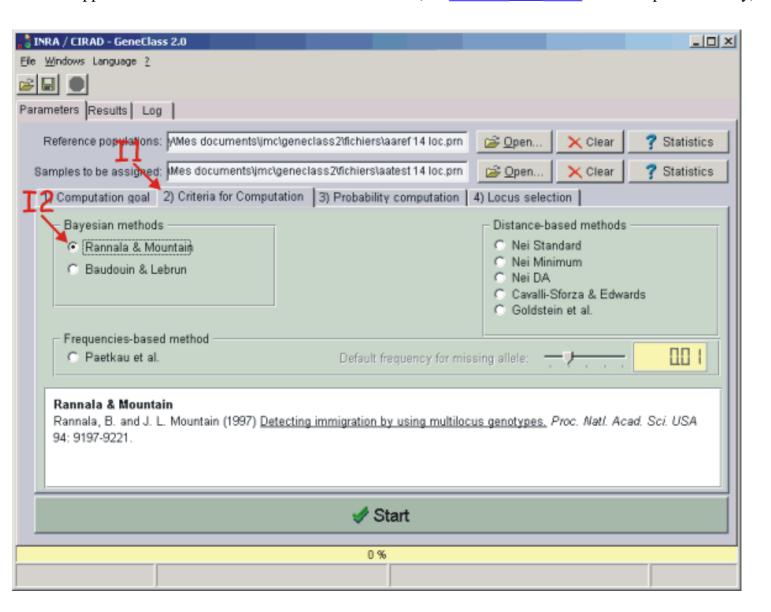
Note that for self-assignment purposes only a single file is needed (i.e. the reference file [H1]).

Choose "Assignment" [H2] in the first tab of the main window and "Individuals" [H3] in the "Assign:" box. Note that assignment statistics can be also computed for groups of individuals rather than individual genotypes (Baudouin & Lebrun, 2000). Groups of individuals must be coded as populations in the samples file.

The assignment threshold (without probability computation) can be set by sliding the "Assignment threshold of scores" cursor [H4] (see computations without associated probabilities section for a definition of scores).

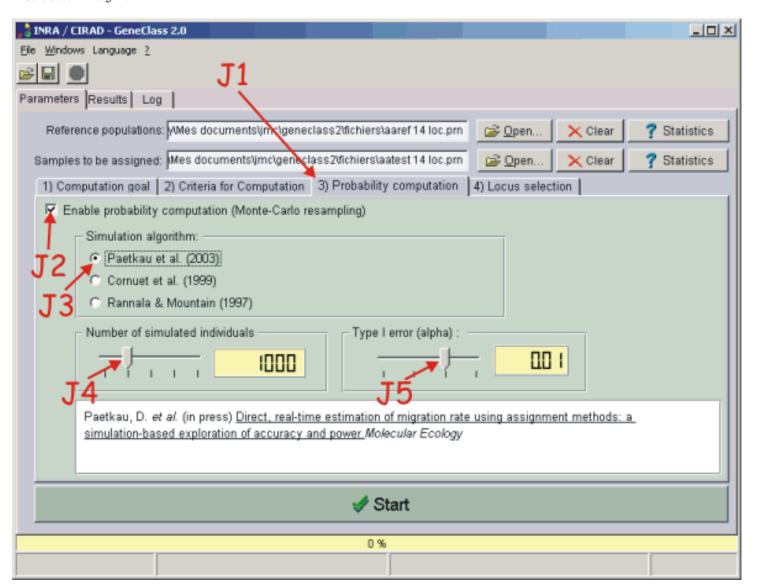


Click on the "2) Criteria for Computation" [I1] tab and choose the desired criterion (e.g. "Rannala & Mountain" [I2], see Rannala & Mountain, 1997 and Cornuet et al., 1999). Bayesian and Frequencies-based methods appears to be better than distance-based methods (see Cornuet et al. 1999 for a comparative study).



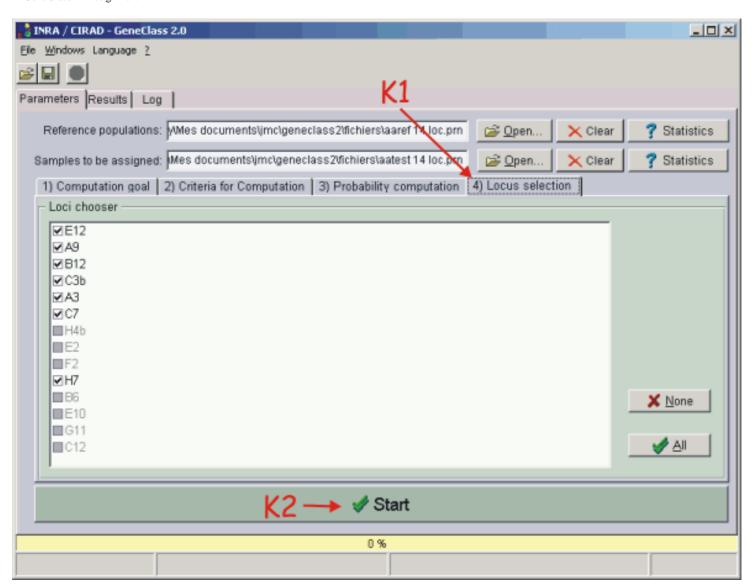
If you want to compute the probability that an individual belongs to each reference population, click on the "3) Probability computation" tab [J1], and then check the "Enable probability computation (Monte-Carlo resampling)" box [J2].

You can now choose a resampling algorithm [J3], eg. Paetkau *et al.* (2004) (recommended, but see Rannala & Mountain 1997 and Cornuet *et al.* 1999). Slide the cursor [J4] to define the minimum number of simulated individuals (eg. 1000, default value or 10000, leading to ten times longer but more precise computation), and the cursor [J5] setting the type one error eg. 0.01, default value (see Cornuet *et al.* 1999; Paetkau *et al.* 2004).

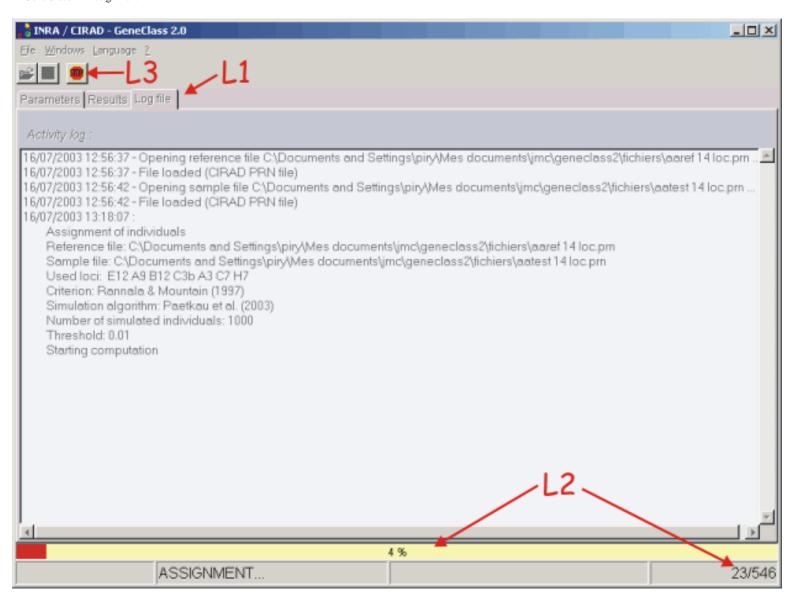


If needed, you can deselect some loci in the "Loci selection" tab [K1]. Deselected loci will be ignored during calculations.

Finally, click on the "Start" button [K2] to run the computation.

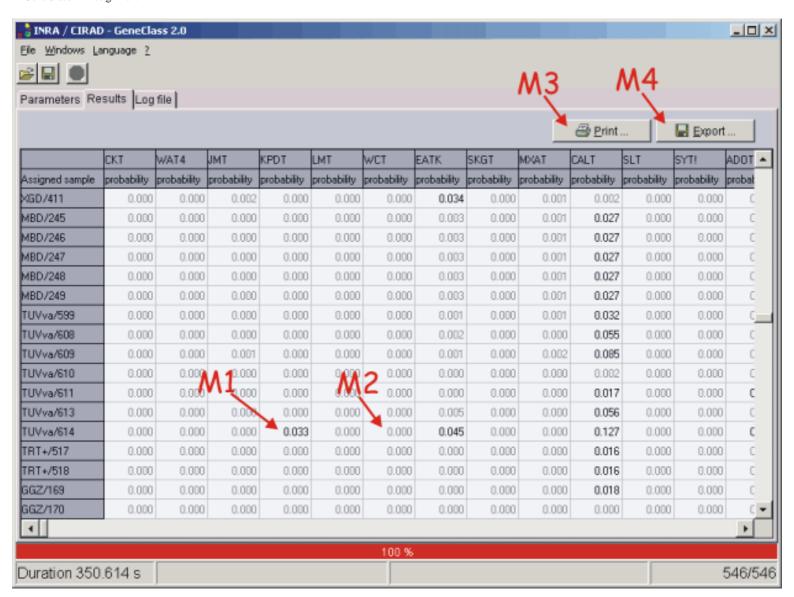


The program displays the "Log file" [L1] that displays the running parameters. The progress bar and a counter shows the state of the computation [L2]. The "Stop" button [L3] allows the computation to be aborted.



Once computations are finished, the results are displayed in a grid in which the probability for each individual belonging to each reference population is displayed [M1] (Cornuet et al., 1999). If a given individual's probability in a given reference population is lower than the previously defined threshold, the value is greyed [M2].

Results can be printed ("Print" button [M3]) or exported in csv format ("Export" button [M4]).



Computations without associated probabilities:

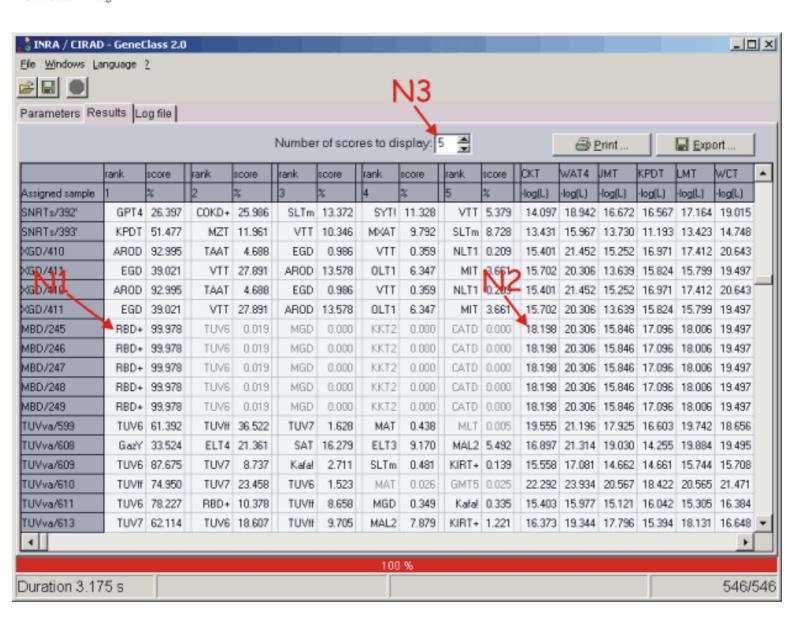
If you did not check the "Enable probability computation (Monte-Carlo resampling)" box [J2] the results are displayed as follows: The five first pairs of columns show, for each individual, the most likely populations and their relatives scores (*cf.* note below) in decreasing order [N1]. The right part of the grid displays -log values of the likelihoods [N2].

The number of displayed scores can be changed with the spin edit area [N3].

Note: In a reference file with k populations the score of an individual i in a population l is computed as follow:

$$score_{i,l} = \frac{L_{i,l}}{\sum_{j=1}^{k} L_{i,j}},$$

with Li,l the likelihood value of the individual i in the population l.

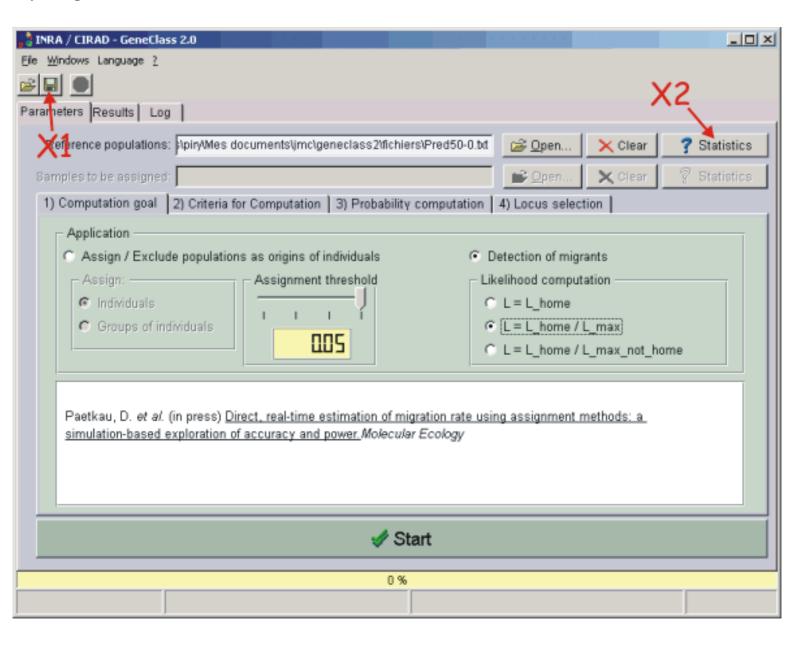


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3) Data file conversion and description of population diversity

Datafiles can be exported into another format using the "Floppy" button [X1] in the upper bar, or with the "Save reference file as..." item in the "File" menu. GeneClass2 can therefore be used as a file-format converter between applications.

A preview of the usual statistics describing the diversity of populations can be obtained before or after any computation from the "Statistics" button [X2] in the main window.



The result is a window with a grid containing allele frequencies, for each locus and for each population, heterozygotes proportion and Nei's gene diversity (Heterozygozity *sensu* Nei, 1987).

This grid can be printed or exported in csv format from the "File" menu [Y1].

LOCUS	Populations:	1	2	3	4	5	6	7	8	9	10
loc1	Populations:	1	2	3	4	5	6	7	8	9	10
Genes number		100	100	100	100	100	100	100	100	100	100
Alleles number		10	10	6	9	12	9	8	9	10	8
Frequencies	44	0	0	0.090	0.050	0.010	0	0	0	0.060	0
Frequencies	45	0.270	0.100	0	0.020	0.120	0.010	0.240	0.250	0.050	0.040
Frequencies	46	0	0	0	0	0	0.030	0.180	0.040	0.030	0.010
Frequencies	47	0.050	0.280	0	0.020	0.080	0	0.070	0.380	0.270	0.170
Frequencies	48	0.030	0.070	0.040	0.180	0	0.070	0	0	0.040	0
Frequencies	49	0.240	0.120	0.030	0.060	0.230	0.240	0.100	0.090	0.430	0.520
Frequencies	50	0.220	0.090	0.020	0	0.100	0.170	0.150	0.150	0.060	0.070
Frequencies	51	0	0.010	0	0.170	0.020	0.060	0.150	0	0	0
Frequencies	52	0.030	0.150	0	0.020	0.140	0.110	0	0.010	0	0
Frequencies	53	0.020	0.040	0	0	0.010	0	0	0.030	0.010	0.060
Frequencies	54	0	0	0	0	0.150	0	0.050	0	0	0
Frequencies	55	0	0	0	0	0.010	0	0	0	0	0
Frequencies	56	0.010	0	0	0	0	0	0	0	0	0
Frequencies	57	0.100	0.050	0.660	0.460	0.070	0.010	0	0.040	0.040	0.110
Frequencies	58	0.030	0.090	0.160	0.020	0.060	0.300	0.060	0.010	0.010	0.020
Heterozygotes proportion		0.880	0.800	0.600	0.780	0.880	0.760	0.840	0.760	0.740	0.700
Ne7s gene diversity		0.814	0.858	0.533	0.727	0.874	0.810	0.853	0.766	0.736	0.685
loc2	Populations:	1	2	3	4	5	6	7	8	9	10
Genes number		100	100	100	100	100	100	100	100	100	100
Allejes number	,	ß	6	7	6	7	6	6	7	6	7.

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