

# Genética Populacional e Filogeografia

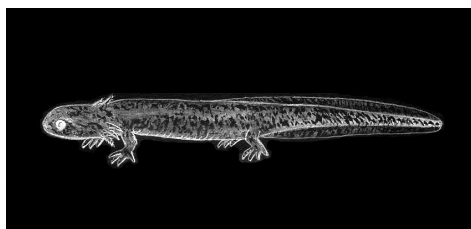
# Salamandra-lusitânica

## *Chioglossa lusitanica*

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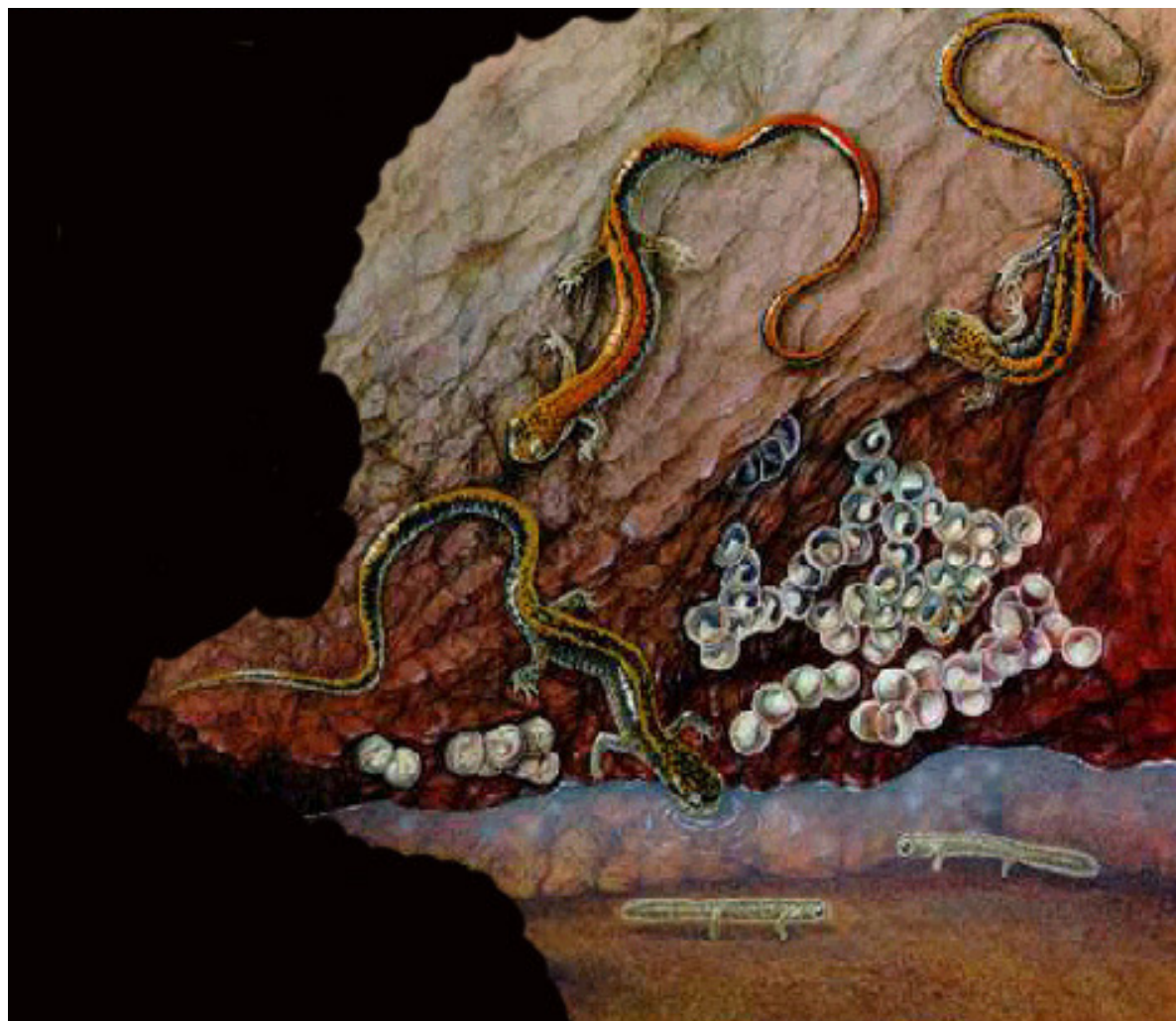
Adulto



Larva

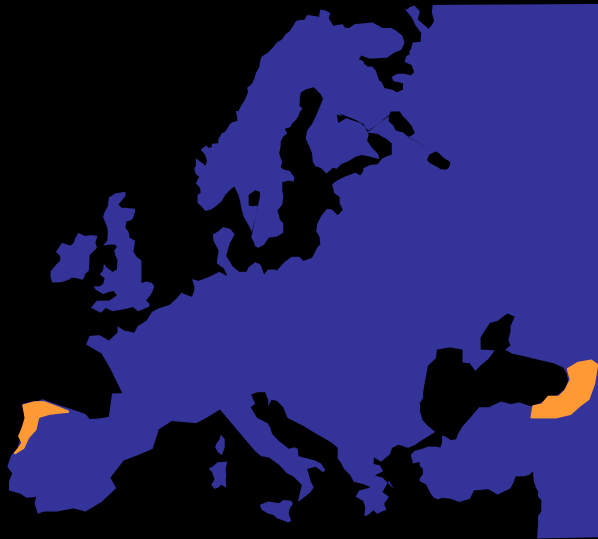
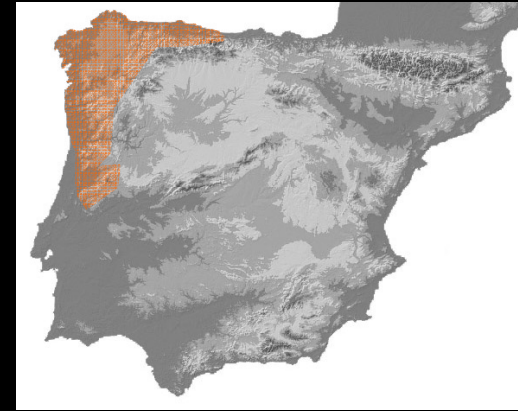


OVOS



# *Chioglossa lusitanica*, uma salamandra peculiar

- Endémica da Península Ibérica
- Único representante do género *Chioglossa*



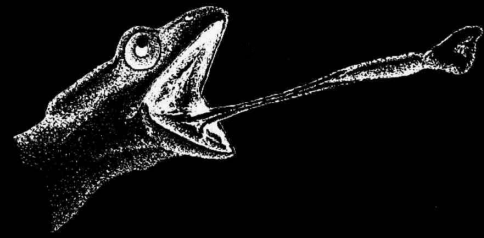
*Mertensiella caucasica*

# *Chioglossa lusitanica*



- **Particularidades únicas**

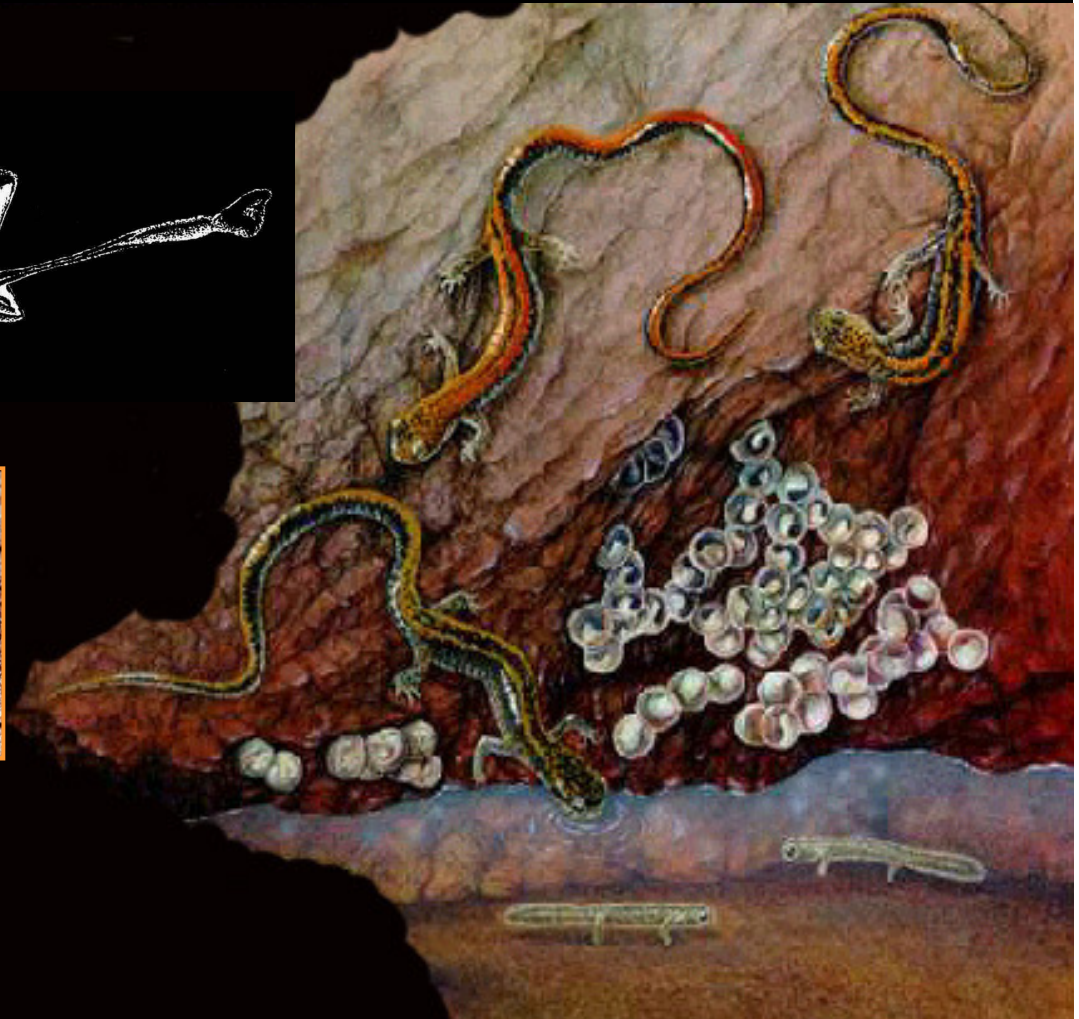
Língua preênsil com pedículo protractil



Autotomia da cauda



Respiração essencialmente cutânea



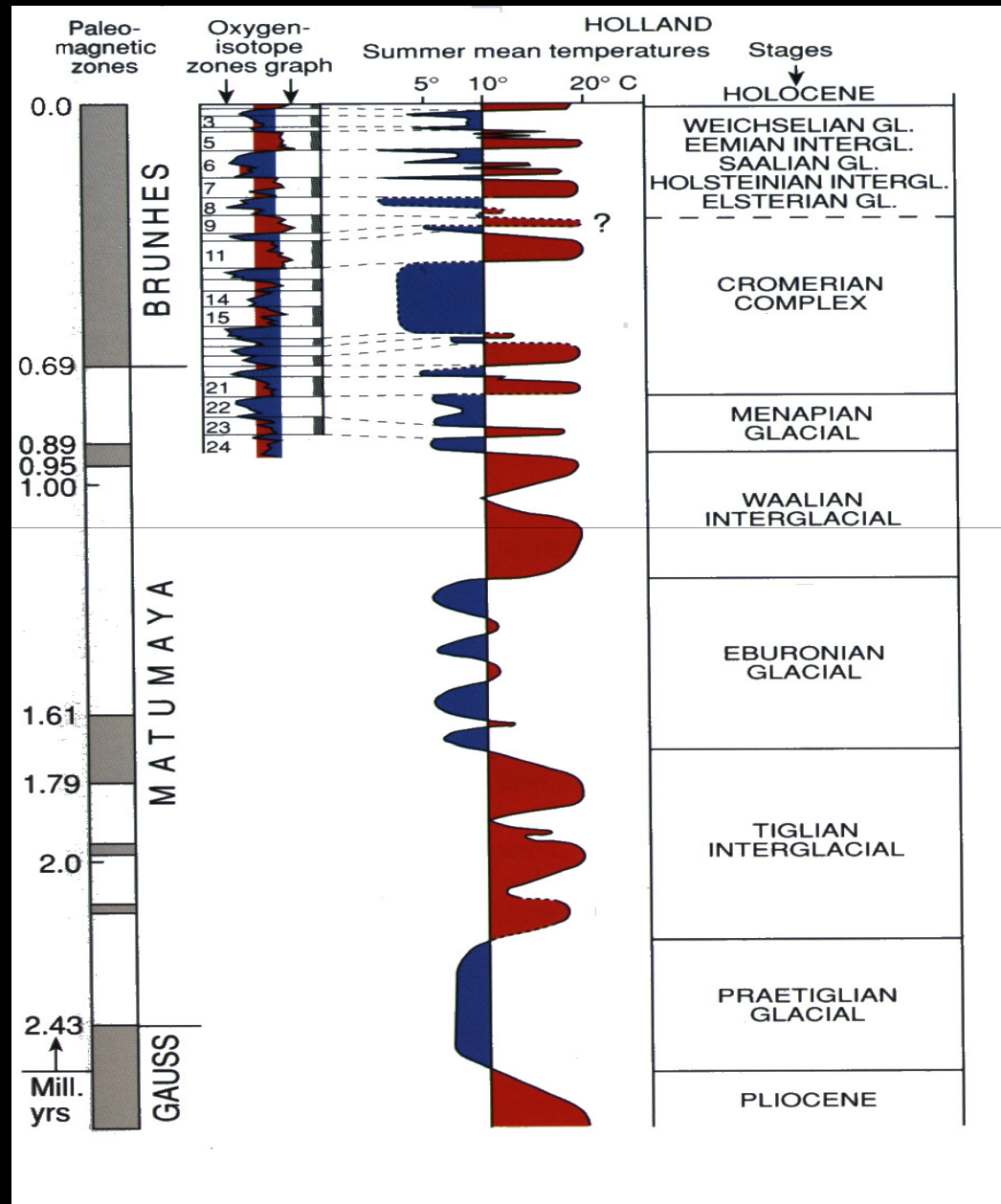
# Habitat

- Dependente de habitats saturados em humidade
- Baixa capacidade de dispersão



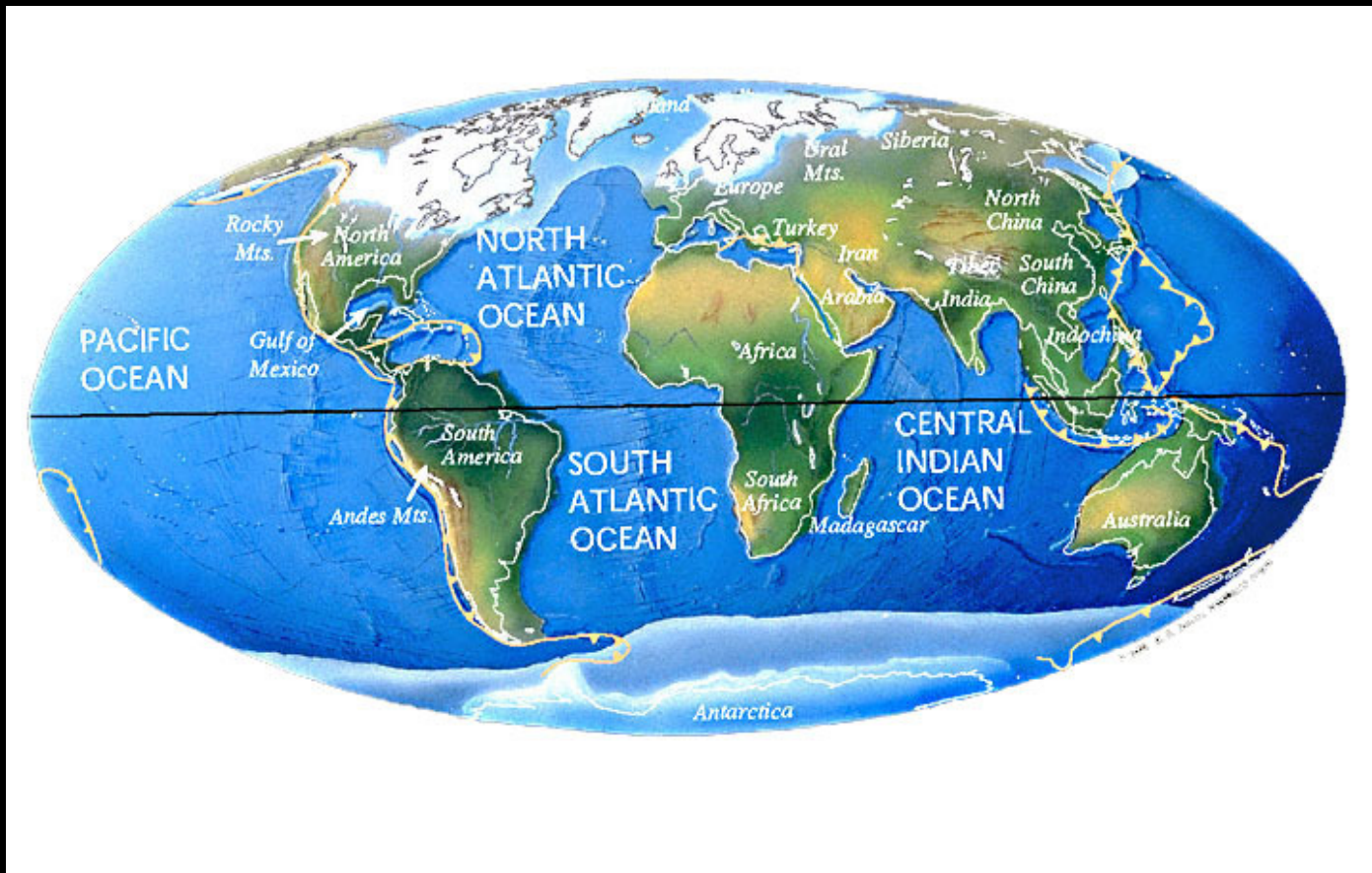


# Condições climáticas durante o Quaternário



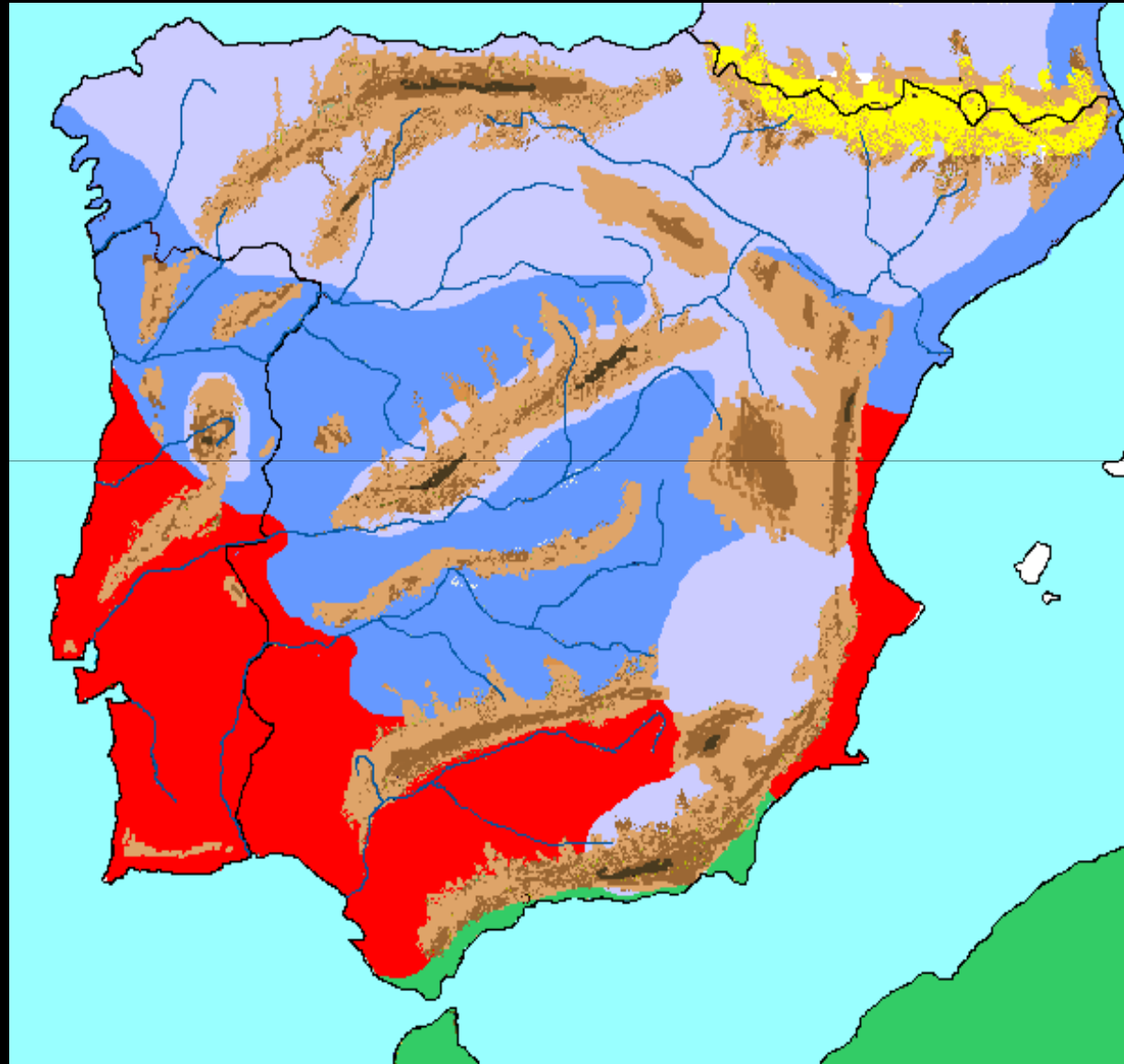
(Último Máximo Glaciar)

Durante os dois últimos milhões de anos o clima da Terra variou drasticamente, entre períodos glaciares e interglaciares





# Península Ibérica há 18-20 milhares de anos



 **Mountains**

 **Rivers**

 **Glacier ice**

 **Tundra**

 **Boreal forest**

 **Temperated mixed forest**

 **Mediterranean vegetation**

# Amostragem



## ❖ Variabilidade genética

1. polimorfismos proteicos; 2. microssatélites; e, 3. DNA mitocondrial (Citocromo b)

## ❖ Distribuição da variabilidade genética nas populações amostradas

ex. N<sup>o</sup> de alelos/haplótipos, diversidade haplotípica e nucleotídica, heterozigotia etc...

## ❖ Estrutura populacional

ex. Fst/Rst, árvores filogenéticas, métodos de agrupamentos Bayesianos, rede de haplótipos, mismatch, AMOVA

## ❖ Diferenciação/divergência e demografia

ex. distâncias genéticas, métodos de coalescência, efectivo populacional, expansão

❖ Interpretação com base na ecologia, barreiras geográficas (ex. rios, zonas de habitat) e factores climáticos

Salas	511	147147	170170	116116	172176	190194	207207	122122
Salas	512	147147	170170	116116	176176	186190	207207	122126
Salas	513	147147	170170	116116	176180	186190	207207	122122
Salas	514	147147	170170	116116	172180	186186	207207	122122
Salas	515	147147	170170	116116	172176	186190	207207	118122
Salas	516	147147	000000	116116	000000	186186	207211	118122
Salas	517	147147	170170	116116	176176	194194	207207	122122
Salas	518	147147	000000	116116	000000	186190	000000	000000
Povedra	107	147147	170170	116116	176176	178182	207207	118122
Povedra	201	147147	170170	116116	176176	178182	207211	122130
Povedra	202	147147	170170	116116	176184	190190	207207	122122
Povedra	203	147147	170170	116116	172176	190190	207207	118122
Povedra	204	147147	170170	116116	172176	182186	207207	122122
Povedra	205	147147	170170	116116	176176	186202	207207	122122
Povedra	206	147147	170170	116116	176176	182186	207207	122122
Povedra	208	147147	170170	116116	176180	182190	207215	122122
Povedra	789	147147	170170	116116	176176	186194	207207	122122
Povedra	790	147147	170172	116116	172176	186186	207207	122122
Povedra	791	147147	170170	116116	172176	186190	207207	122130
Povedra	792	147147	170170	116116	176176	178186	203207	122126
Povedra	793	147147	170170	116116	164172	186190	207207	122126
Povedra	794	147147	170170	116116	176176	166182	207211	122126
Povedra	795	147147	170170	116116	176176	182190	207207	122122
Povedra	796	147147	170170	116116	168176	166202	207207	122122
Geres	463	147147	170170	116116	176180	182186	203207	118126
Geres	464	147147	170170	116116	176176	186186	207207	118122

## PROGRAMAS - DOWNLOAD

<http://www.discat.unipi.it/BioIMar/utilities/progs.htm>

<http://linkage.rockefeller.edu/soft/>

## ANÁLISE ON-LINE

<http://cbsuapps.tc.cornell.edu/index.aspx>

(Computational Biology Service Unit Web Computing Resources)

<http://genepop.curtin.edu.au> \*\*\*\*\*genepop

**The primary purpose of the original DOS based software is to compute exact tests for the following :**

- Hardy-Weinberg equilibrium
- Population differentiation, and
- Genotypic disequilibrium among pairs of loci

**The software computes estimates of classical population parameters, including:**

- Allele Frequencies and other basic information,
- Fst and other correlations,
- Maximum likelihood estimates in the presence of a NULL allele,
- Nm estimates by the private allele method.

**The package also has some ecumenical utilities which convert the input GENEPOP format files to formats used by other statistical packages such as LinkDos, FSTAT, MONTY, BIOSYS, and ANOVA file for heterozygosity.**

# Análise da diversidade genética (parâmetros standard)

- ❖ N° de alelos/locus
- ❖ N° médio de alelos/população
- ❖ Frequência alélica/população
- ❖ Riqueza alélica/população
- ❖ Heterozigotia observada
- ❖ Heterozigotia esperada
- ❖ HWE
- ❖ LD

# Análise da diversidade genética

❖ GENETIX

❖ GENEPOP

❖ FSTAT

❖ ARLEQUIN

❖ GENALEX



**GENETIX**

❖ Construção do Infile – phylogenetix.xls –  
**transformar** - phylogenetix.txt  
(texto separado por tabulações, formato txt)

❖ Abrir o genetix

1. fichier > importer (escolher o nosso ficheiro) > tabulation > 3 chiffres
2. Seleccionar variabilité

Nº médio de alelos em cada população

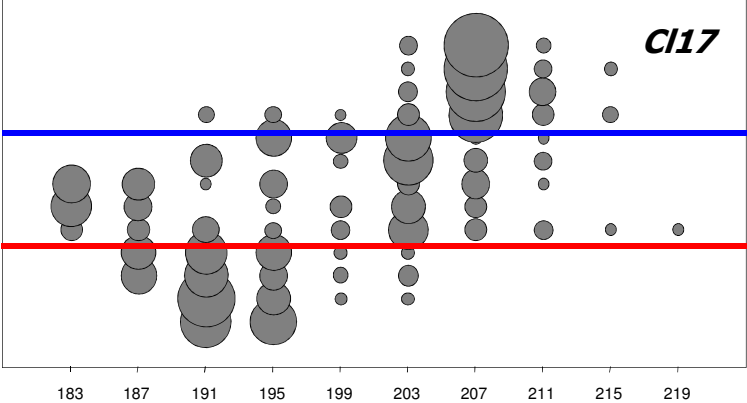
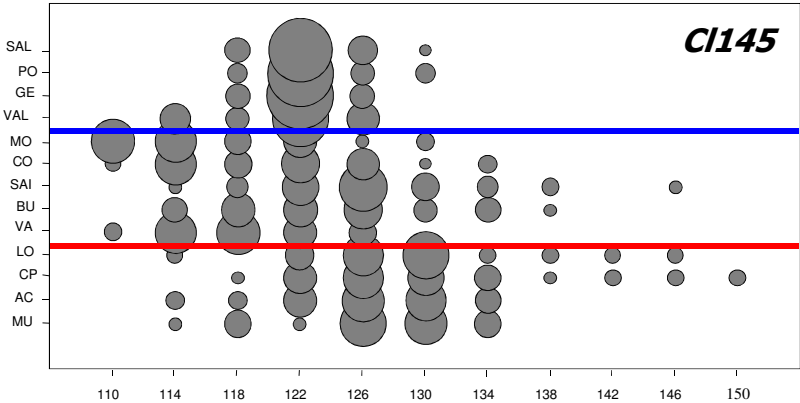
Heterozigotia esperada (He) em cada população

Variação da He com a latitude (construir gráfico – ver ficheiro  
DistHe.xls)

# Phylogeography

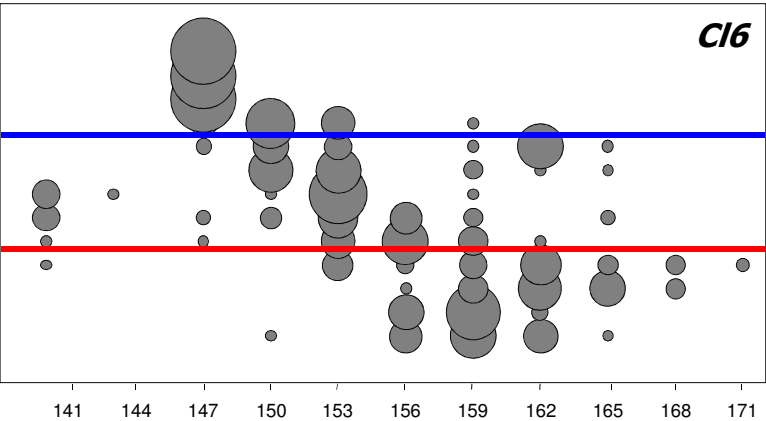
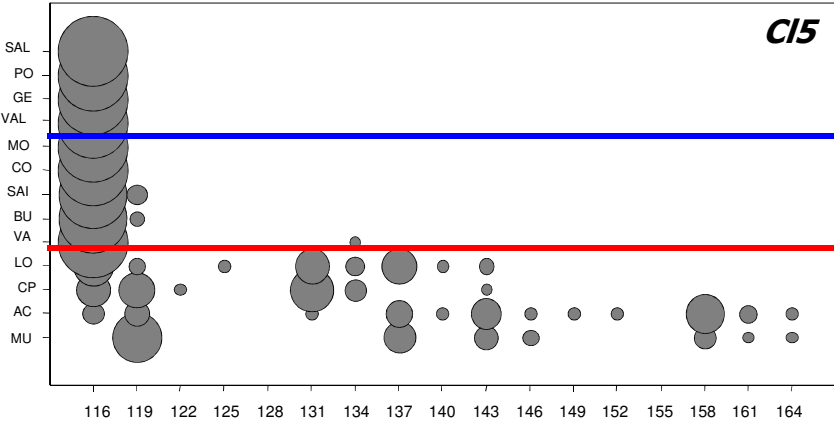


## Microsatellites



Douro river

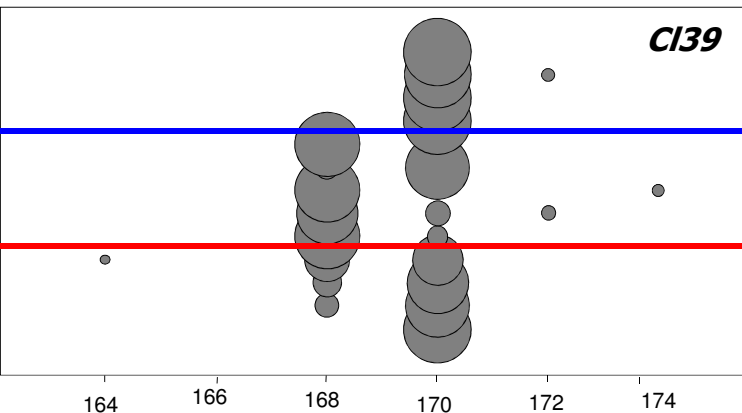
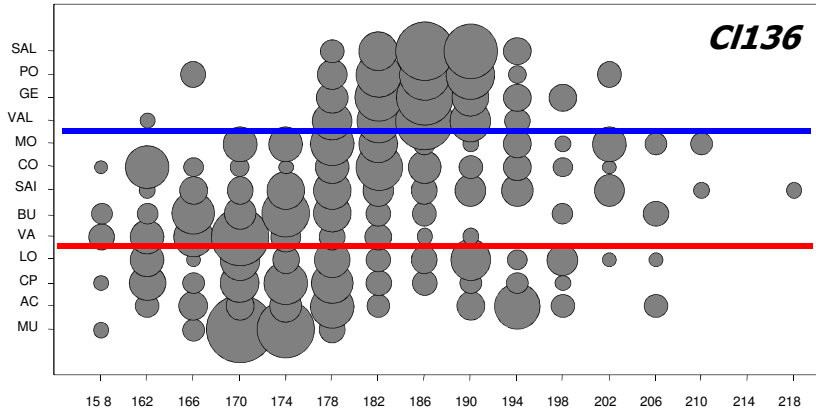
Mondego river



Douro river

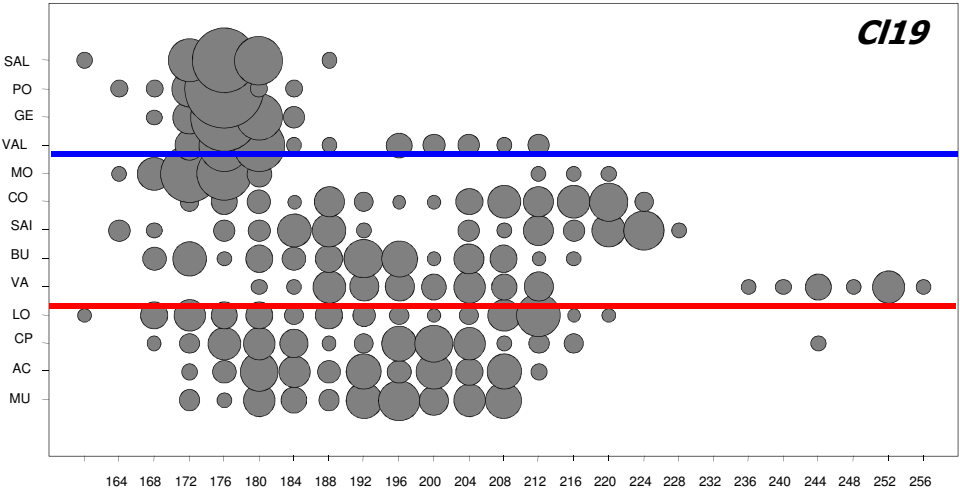
Mondego river

# Phylogeography



Douro river

Mondego river



Douro river

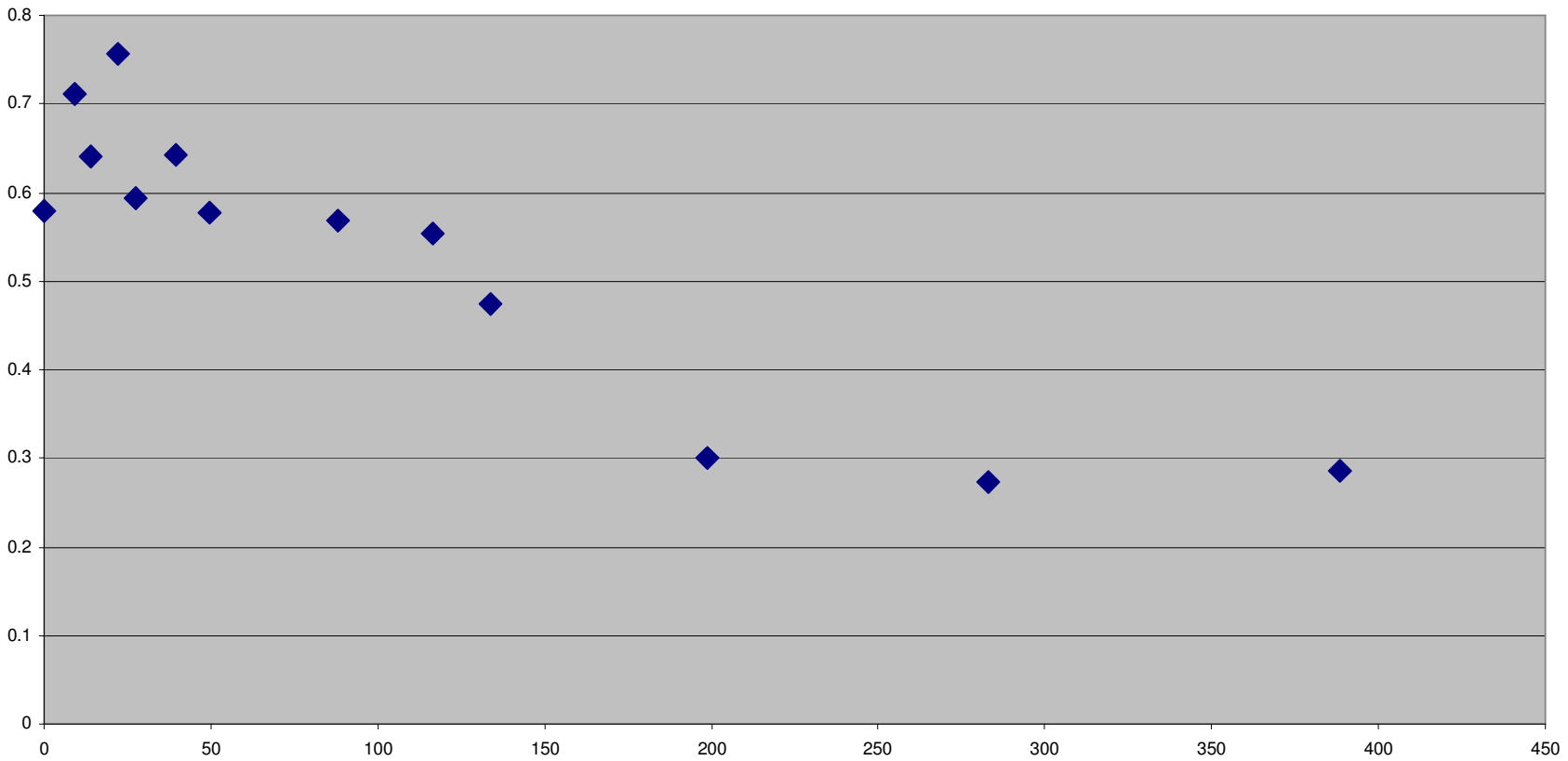
Mondego river

# Proteínas

## Microssatélites

	Hexp.	H n.b.	Hobs.	P(0.95)	P(0.99)	Nbre	Moyen d'allèles/locus
Salas	0.2861	0.2924	0.3063	0.5714	0.5714	2.8571	
Ecart-type	0.3235	0.3298					
Povedra	0.2729	0.2817	0.2946	0.5714	0.7143	3.5714	
Ecart-type	0.3009	0.3056					
Geres	0.2998	0.3075	0.2857	0.5714	0.5714	2.8571	
Ecart-type	0.324	0.3159					
Valongo	0.474	0.4854	0.4612	0.7143	0.7143	4.7143	
Ecart-type	0.3435	0.3317					
Montem	0.553	0.5672	0.5857	0.8571	0.8571	5.5714	
Ecart-type	0.3367	0.3473					
Covelo	0.5686	0.5802	0.5714	0.8571	0.8571	6.5714	
Ecart-type	0.3478	0.3239					
Saide	0.578	0.5927	0.5313	1	1	7.2857	
Ecart-type	0.3451	0.3195					
Bucaco	0.6423	0.6559	0.6222	0.8571	1	6.8571	
Ecart-type	0.3234	0.3067					
Varzea	0.5939	0.6083	0.5866	0.8571	1	7	
Ecart-type	0.3529	0.348					
Lfiscal	0.7564	0.7716	0.7529	1	1	8.4286	
Ecart-type	0.1318	0.1242					
LCPera	0.7119	0.7284	0.7241	1	1	7.4286	
Ecart-type	0.2114	0.1835					
Acor	0.6398	0.6577	0.6915	1	1	6.8571	
Ecart-type	0.2727	0.2899					
Muradal	0.5794	0.5935	0.6327	0.8571	0.8571	5.1429	

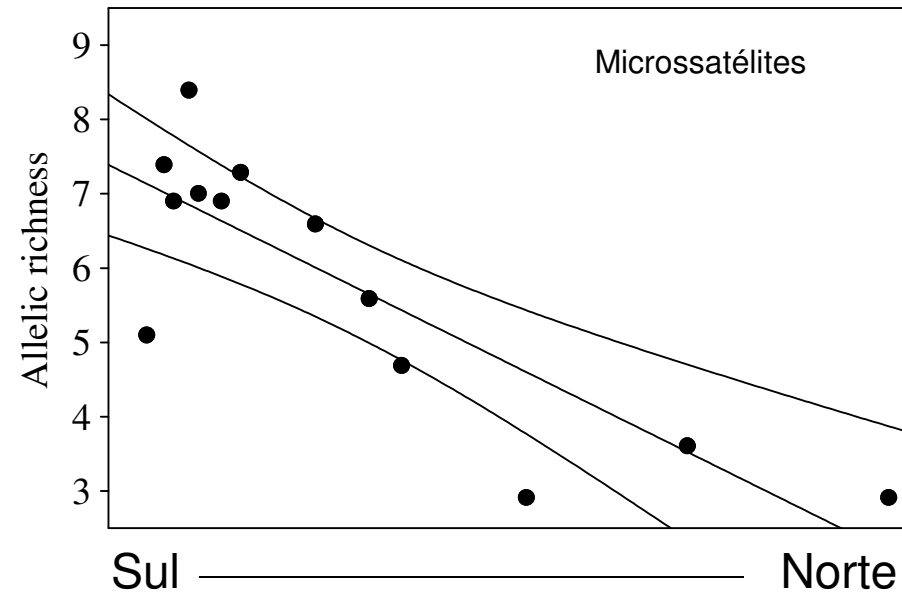
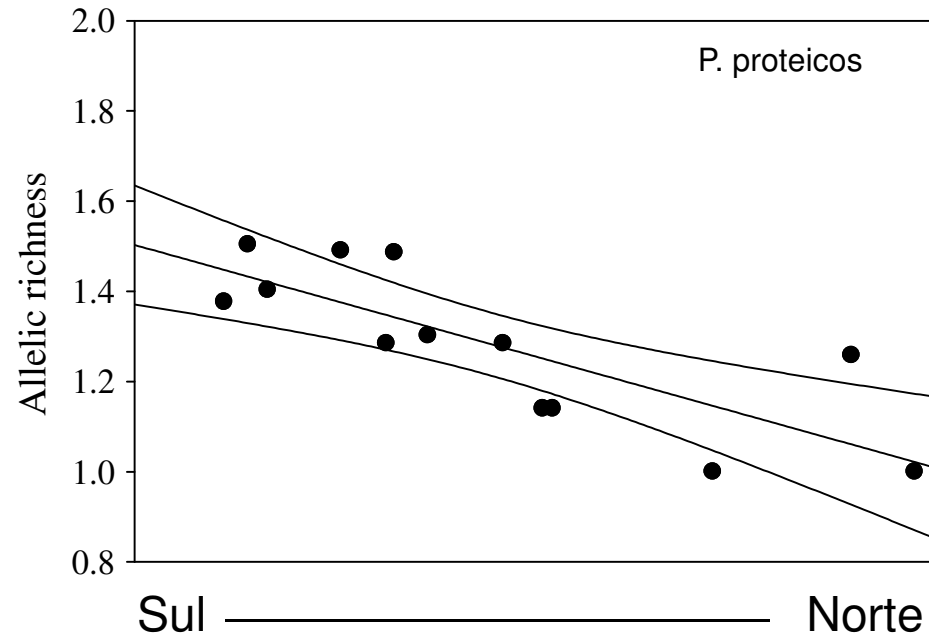
	Hexp.	H n.b.	Hobs.	P(0.95)	P(0.99)	Nbre	Moyen d'allèles/locus
SAL	0	0	0	0	0	1	
Ecart-type	0	0					
FS	0.0223	0.0233	0.0119	0.1429	0.1429	1.2857	
Ecart-type	0.0616	0.0315					
GE	0.0666	0.068	0.0559	0.1429	0.1429	1.1429	
Ecart-type	0.18	0.1479					
BA	0.0645	0.0665	0.0982	0.1429	0.1429	1.1429	
Ecart-type	0.176	0.2599					
BJ	0.1223	0.1274	0.1132	0.2857	0.2857	1.2857	
Ecart-type	0.2183	0.1942					
VAL	0.0758	0.0767	0.0765	0.1429	0.5714	1.5714	
Ecart-type	0.1548	0.1538					
MO	0.1195	0.1224	0.1429	0.2857	0.4286	1.5714	
Ecart-type	0.2003	0.2349					
TA	0.112	0.1154	0.1092	0.2857	0.2857	1.2857	
Ecart-type	0.206	0.1873					
CO	0.1591	0.1612	0.1507	0.2857	0.4286	1.7143	
Ecart-type	0.2503	0.2346					
SAI	0.0972	0.0991	0.1099	0.2857	0.2857	1.5714	
Ecart-type	0.1698	0.1877					
BU	0.1361	0.1382	0.1494	0.4286	0.4286	1.7143	
Ecart-type	0.1978	0.2214					
VA	0.1191	0.122	0.1489	0.2857	0.4286	1.4286	
Ecart-type	0.2006	0.2523					
LVI	0.271	0.2766	0.278	0.7143	0.8571	2	
Ecart-type	0.214	0.225					
CP	0.1351	0.1374	0.0977	0.2857	0.7143	1.7143	
Ecart-type	0.1737	0.1343					
AC	0.0436	0.0446	0.034	0.2857	0.2857	1.2857	
Ecart-type	0.0771	0.0597					
MUR	0.0363	0.0374	0.0408	0.1429	0.2857	1.2857	
Ecart-type	0.0842	0.0932					



Sul



Norte (km )



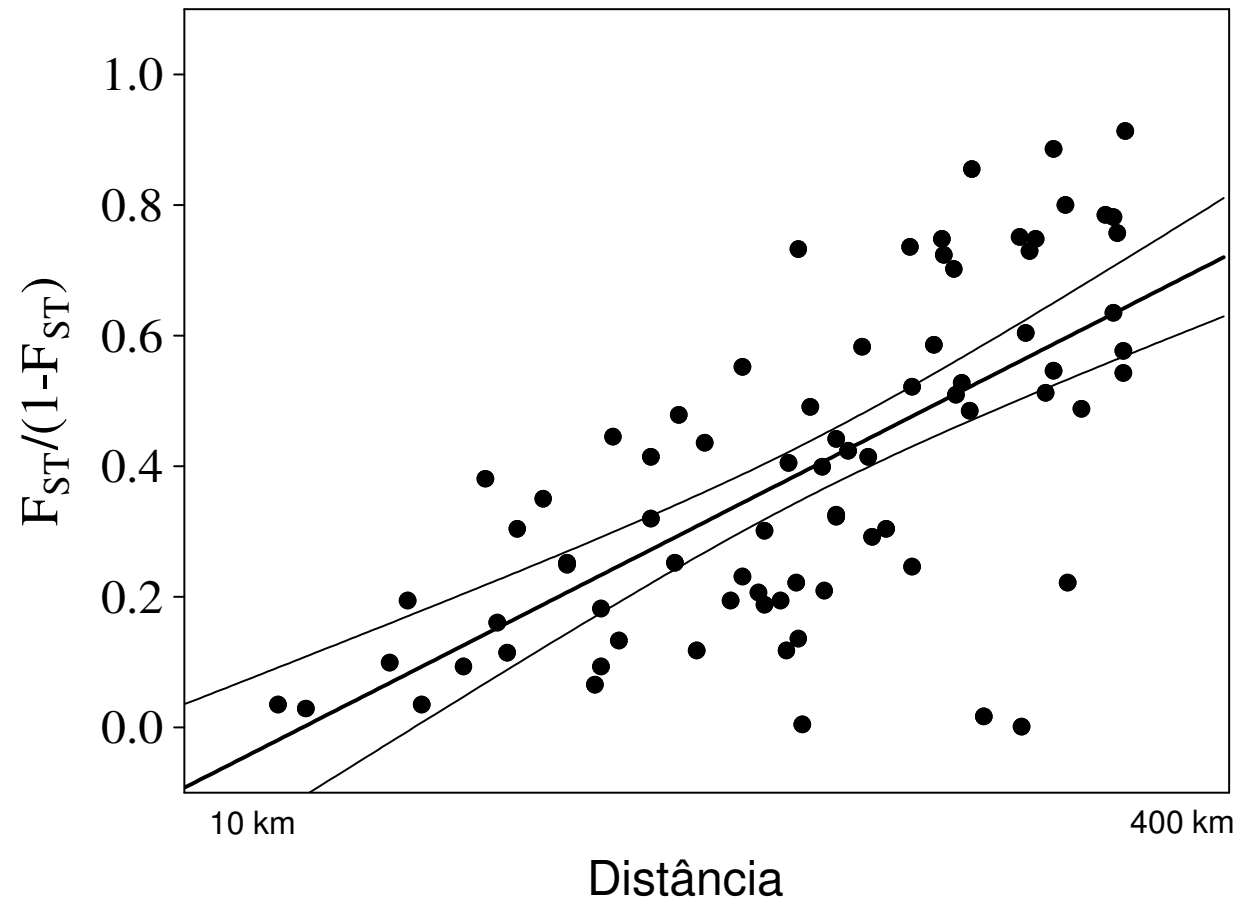
# Subestruturação

$F_{st}$  Global?

Pairwise  $F_{st}$  ? i.e  $F_{st}$  entre cada par de populações?



Fst = 0.26



FSTAT

❖ Construção do Infile - Genetix – **abrir** - phylogenetix.txt - Outils- converter em Phylogenetix. dat

❖ Colocar esse ficheiro dentro da pasta do Fstat

❖ Abrir o Fstat > open (nosso ficheiro .dat)

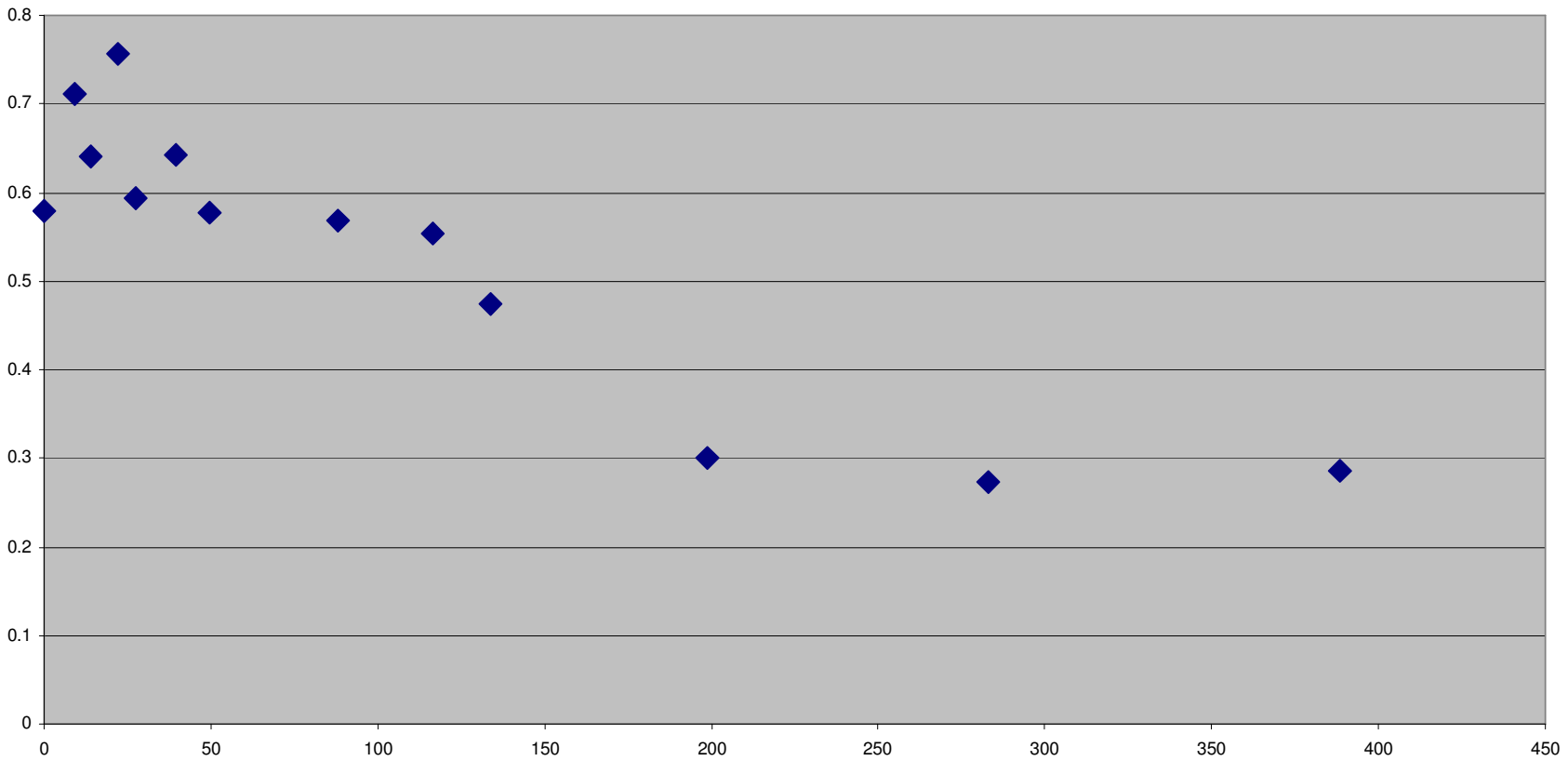
Riqueza alélica (*Allelic richness*)

Hardy-Weinberg

Genotypic-disequilibrium

Diferenciação entre cada par de populações (*Population differentiation*)

Comparação da riqueza alélica, heterozigotia observada e Fst entre as populações a norte e a sul do rio Douro



Sul



Norte (km )

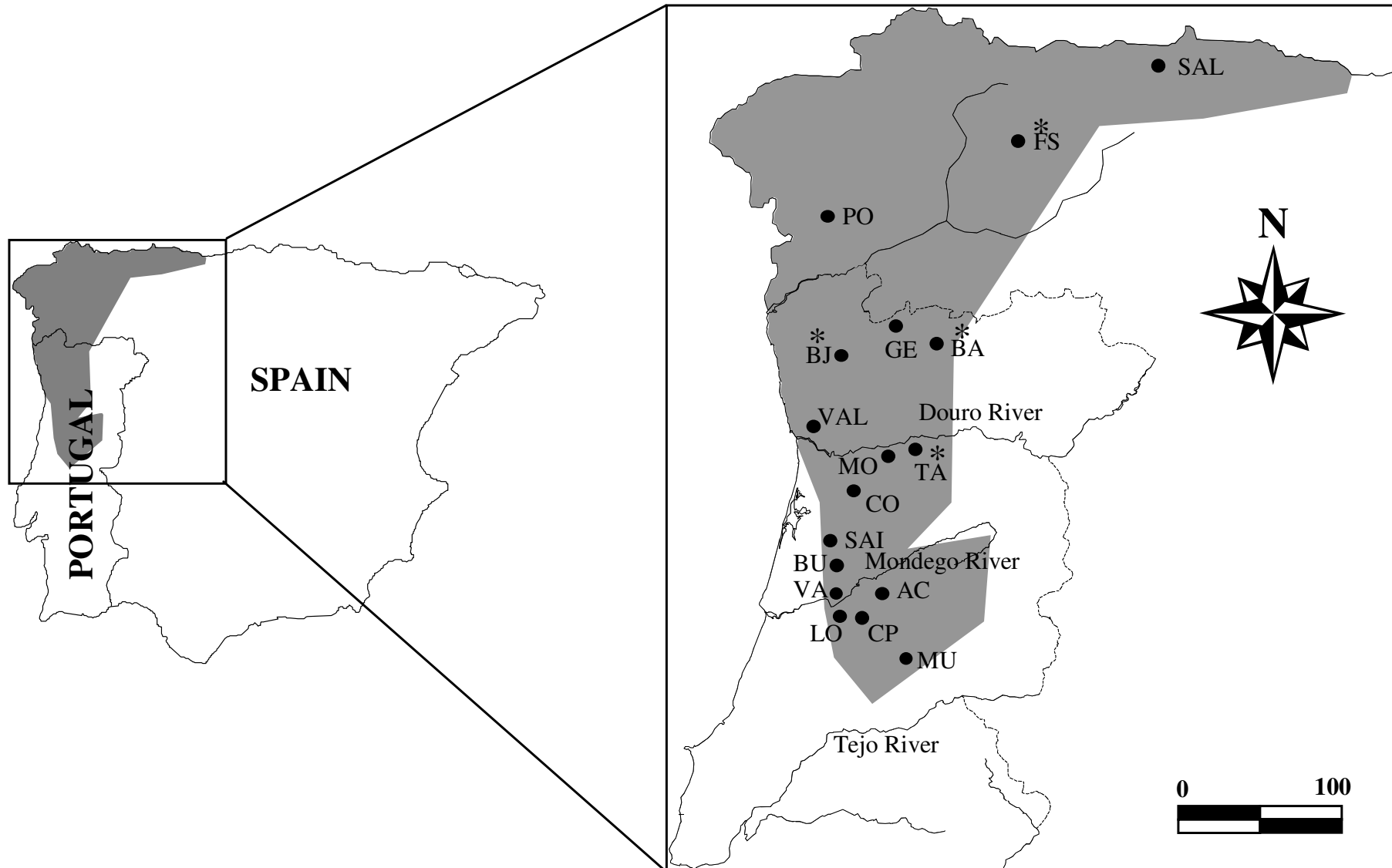


GENEPOP

GENEPOP ON THE WEB

<http://genepop.curtin.edu.au/>

# Phylogeography

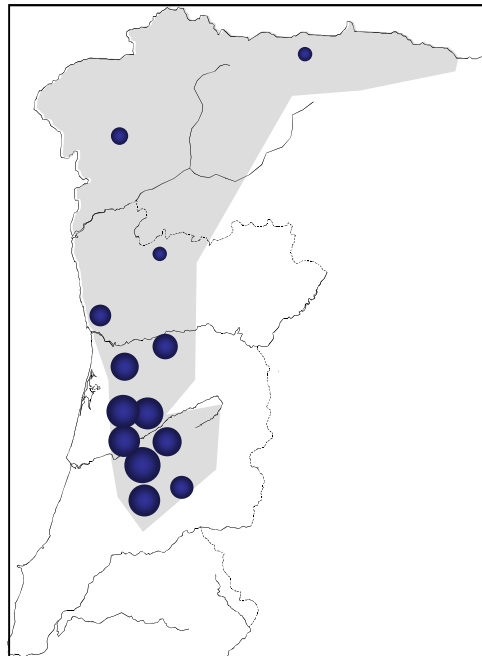




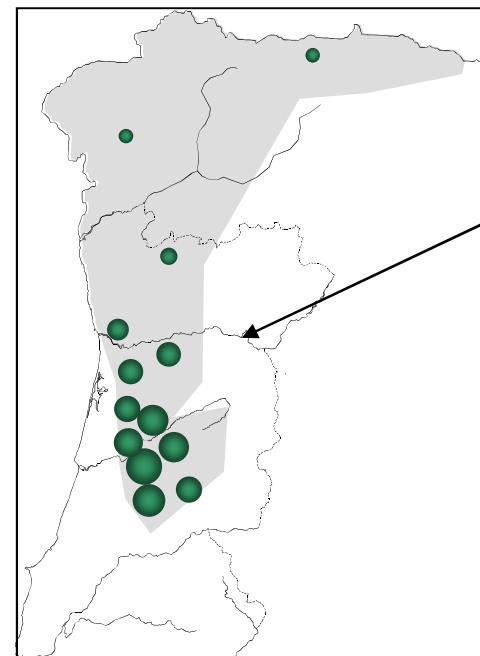
# Phylogeography



Allelic richness



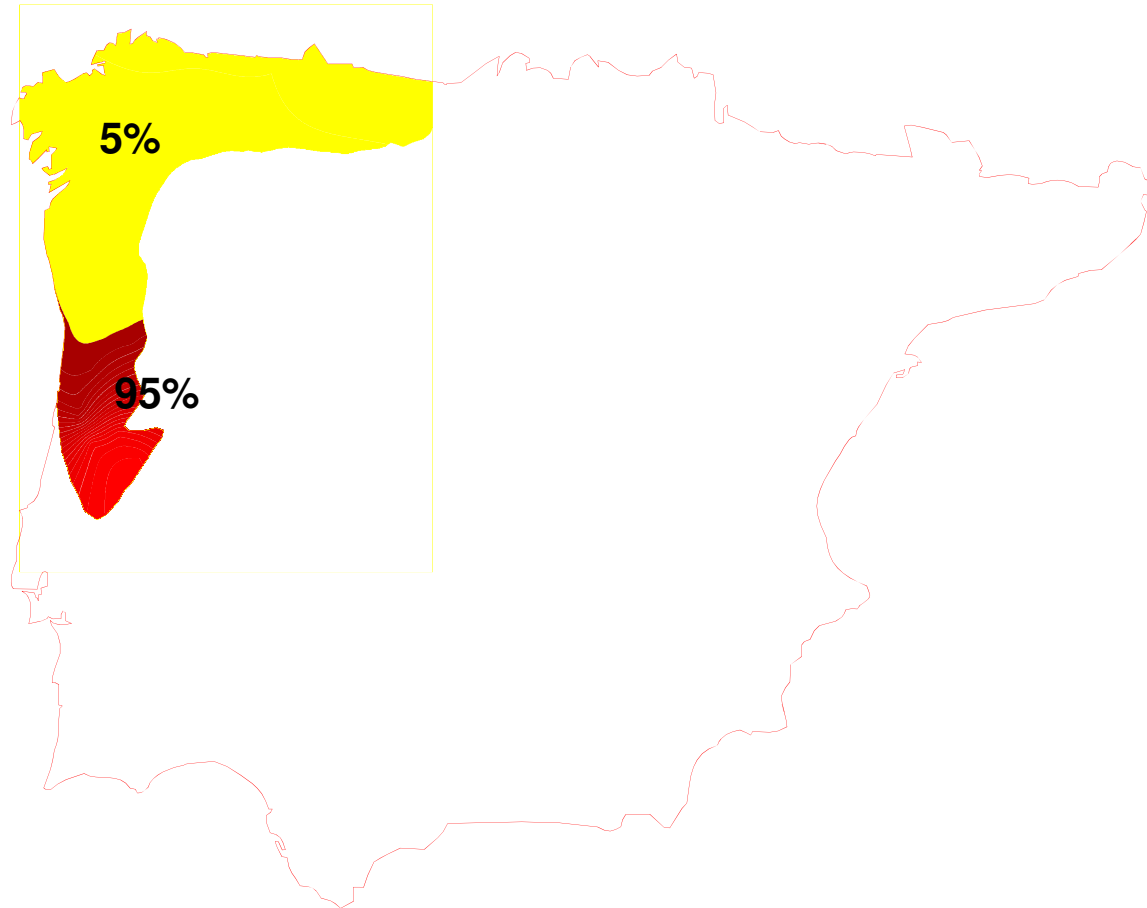
Expected heterozygosity



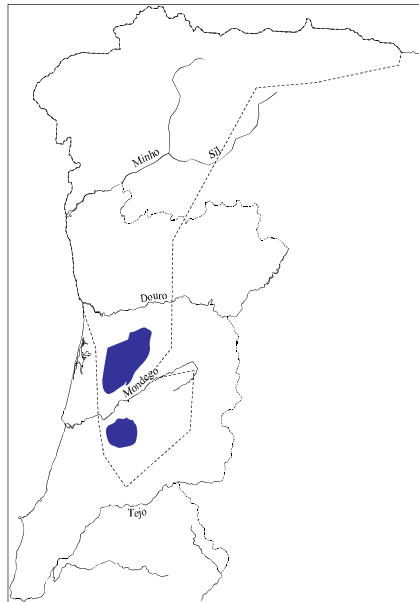
Douro River

# Previous genetic studies

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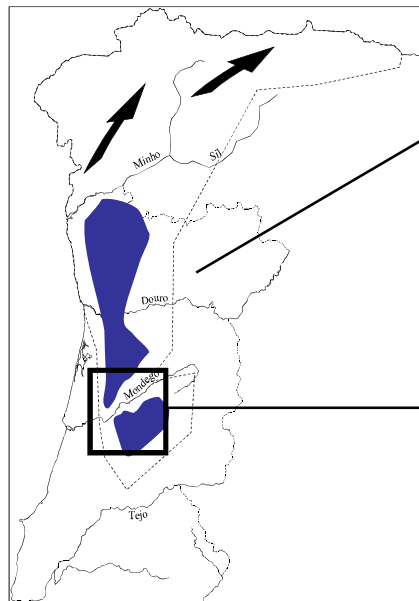
# Cenário Biogeográfico



Fragmentação da distribuição em refúgios glaciares durante o Plioceno /Plistoceno (ca. 2Ma)



Distribuição actual



Expansão pós-glacial



Contacto secundário