GENETIC VARIATION IN BEECH POPULATIONS (*FAGUS SYLVATICA* L.) ALONG THE ALPINE CHAIN AND IN THE HUNGARIAN BASIN

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ABSTRACT

Seventy-eight European beech populations from the Alpine Chain and the Hungarian Basin were analyzed by means of 11 isozyme loci. By principal component analysis (PCA) the populations could be assigned to four groups: 1. French Alps, 2. Swiss Alps, 3. Northeastern Alps (Germany, western Austria), and 4. eastern Austria and Hungarian Basin. The first three canonical variates accounted approximately for 58% of the variation. The univariate approach showed that allele frequencies and genetic diversity did not vary similarly at all loci. Mean gene diversity was high and amounted roughly to 0.29. While for certain loci such as *Px–1* and *Px–2* gene diversity decreased from east to west, at other loci (e.g., *Sod-1*) diversity increased in populations along the transect from the Hungarian Basin to the French Alps. The longitudinal gradients of allele frequencies were in agreement with the postglacial history based on fossil pollen data. Correlations between allele frequencies and elevation were weak.

Key words: *Fagus sylvatica*, allozymes, genetic differentiation, Alps, Hungarian Basin

INTRODUCTION

In recent years many isozyme studies in European beech (*Fagus sylvatica* L.) have been carried out (*e.g.*, COMPS *et al.* 1987; 1990, 1991a and b; CUGUEN *et al.* 1988; MÜLLER-STARCK & ZIEHE 1991; GÖMÖRY *et al.* 1992a and b; MERZEAU *et al.* 1994a and b; VYŠNÝ *et al.* 1995; LEONARDI & MENOZZI 1995; see review in PAULE 1995; LARSEN 1996; HAZLER *et al.* 1997). Compared with many other forest tree species genetic diversity in beech at isozyme loci is high and genetic differentiation among regions occupied by different glacial source populations is well pronounced. The post-glacial history of beech has doubtless played an important role in shaping genetic structures. Additionally the range of natural ecological conditions and sometimes the scattered occurrence on the fringe of its range, as well as historical factors may have favoured genetic differentiation within and among populations.

It is often reported that highly variable populations are generally located in southern countries, where beech outlasted during the last glaciation. This hypothesis which is also stated for other species (GULLBERG *et al.* 1985), is supported by many generations within refuges whereas the time that has elapsed since the last glaciation has probably been too short to allow a higher degree of genetic differentiation in northern populations. However, gene diversity often expressed by the expected heterozygosity (H_e) (NEI 1973) is not always higher in refuge areas. High genetic differentiation of these refuge populations may be due to the presence of some fixed alleles not found in other regions (COULAUD 1994; HAZLER *et al.* 1997).

Other authors have studied the selection regimes over generations (GREGORIUS *et al.* 1986) or triggered by air pollution (MÜLLER-STARCK 1985, 1989; MÜLLER-STARCK & ZIEHE 1991). Certain isozyme loci, for instance genes coding aminopeptidases, have proven to be selectively adaptive or at least hitchhike with cryptic adaptive genes. Other studies have shown that genetic structures at isozyme loci are linked to climatic conditions caused by longitudinal and latitudinal (COMPS *et al.* 1990) or altitudinal differences (FELBER & THIEBAUT 1984; COMPS *et al.* 1987, 1990; GÖMÖRY *et al.* 1992b; PORTAL 1993; VYŠNÝ *et al.* 1995).

According to HUNTLEY & BIRKS (1983) the main glacial refuge was the Balkan region. From here beech has spread out through most of Europe toward the north and west. Unlike in oaks postglacial migration of beech was delayed. At the end of the Pleistocene 6% of the range was colonized and 5000 years later (end of the mid-Holocene) only 50% of the range was covered by *Fagus sylvatica* (LANG 1994, *loc. cit.* p. 162). At 7500

BP beech reached the Alps and it islikely that this mountain chain was an insurmountable obstacle under the climatic conditions of that period. Beech spread out toward the northwest approximately 6000 BP. It probably reached Hungary and the eastern part of the Alps first. Thus, it is interesting to test whether there are significant differences in genetic population structures along a transect from Hungary toward the west along the Alps. If differences can be verified, the question arises whether they are caused by phytogeographic history or by ecological conditions. Therefore, beech populations, along the Alpine chain (France, Switzerland, southern Germany, Austria) and in the Hungarian Basin were analyzed by means of allozymes.

MATERIAL AND METHODS

Dormant buds were sampled from 78 *Fagus sylvatica* stands distributed from Hungary to the French Alps (Fig. 1), representing various climatic conditions and different altitudes from 200 to 1500 m a.s.l. (Tab. 1). In each stand 50 individuals were chosen at random. Methods used for enzyme extraction, electrophoresis, and genetic control are described in KIM (1979), THIEBAUT *et al.* (1982), MERZEAU *et al.* (1989) and in MÜLLER-STARCK & STARCKE (1993). A total of 10 isozyme systems was analyzed and 11 polymorphic loci used: *Acp–1* (acid phosphatase E.C. 3.1.3.2), *Got–1* (glutamate oxalo-acetate transaminase, E.C. 2.6.1.1), *Idh–1* (isocitrate dehydrogenase, E.C. 1.1.1.42), *Mdh–1* (malate dehydrogenase, E.C. 1.1.1.37), *Mnr–1* (menadione reductase, E.C. 1.6.99.2), 6-Pgd–1 (6-phosphogluconate dehydrogenase, E.C. 1.1.1.44), *Pgi–1* (phosphogluco-isomerase, E.C. 5.3.1.9), *Pgm–1* (phosphogluco mutase, E.C. 2.7.5.1), *Px–1* and *Px–2* (peroxidases, E.C. 1.11.1.7), and *Sod–1* (superoxide dismutase, E.C. 1.15. 1.1)

Mean allelic frequencies of each population were analyzed using the principal component analysis (PCA). In total frequencies of 15 alleles were analyzed

Table 1. Number of sampled populations according to altitudinal ranges and groups. 1 – French Alps; 2 – Swiss Alps; 3 – Northeastern Alps and environs; 4 – East Austria and Hungarian Basin

Altitude (m)	Groups			
	1	2	3	4
< 500	1	3	0	14
500–600	1	3	4	4
600–700	1	1	3	3
700–800	1	4	6	0
800–900	2	0	5	0
900–1000	3	2	2	0
1000–1100	4	0	3	0
> 1100	6	0	2	0
Total	19	13	25	21

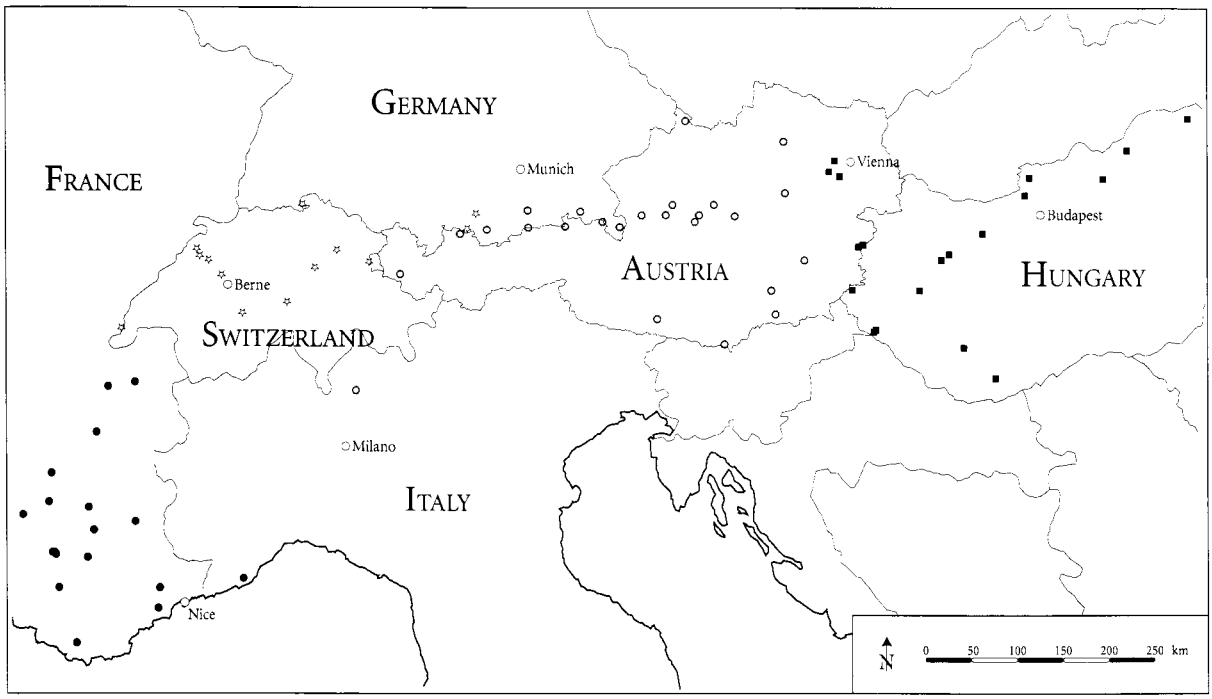

Figure 1. Location of the beech populations sampled along the east-west transect. The broken lines divide the four main groups displayed by PCA. ●● French Alps; ☆ – Swiss Alps; ✡ – Mortheastern Alps and environs; ■ East Austria and Hungarian Basin

Table 2. Mean allele frequencies at each locus within groups

Locus	Alleles	French Alps	Swiss Alps	Northeastern Alps (Germany and western Austria)	Eastern Austria and Hungarian Basin
Acp–1	108	0.076	0.006	0.032	0.148
	100*	0.705	0.710	0.785	0.631
	84*	0.219	0.284	0.183	0.221
Got–1	105	0.209	0.154	0.057	0.074
	100*	0.791	0.846	0.943	0.926
Idh–1	116*	0.172	0.216	0.301	0.288
	100*	0.810	0.782	0.693	0.709
	84	0.018	0.002	0.006	0.003
Mdh–1	22	0.218	0.283	0.323	0.229
	18*	0.782	0.717	0.677	0.771
Mnr–1	126	0.000	0.002	0.002	0.006
	100*	0.950	0.936	0.916	0.910
	74	0.001	0.002	0.018	0.003
	63*	0.049	0.060	0.064	0.081
Pgi–1	113	#	0.000	#	#
	100	0.985	0.991	0.989	0.988
	87	0.015	0.009	0.010	0.011
Pgm–1	106	0.182	0.328	0.374	0.236
	100*	0.816	0.672	0.626	0.764
	93	0.002	0.000	#	0.000
6–Pdg–1	112	0.003	0.002	0.018	0.012
	100*	0.942	0.933	0.867	0.840
	84*	0.055	0.065	0.115	0.148
Px–1	105	0.214	0.225	0.364	0.371
	100*	0.786	0.775	0.636	0.629
Px–2	39	0.124	0.079	0.157	0.180
	26*	0.779	0.797	0.740	0.755
	13*	0.097	0.124	0.103	0.065
Sod–1	112	0.212	0.222	0.085	0.050
	100*	0.788	0.778	0.915	0.950
Number of sampled populations		19	13	25	21

as variables: one allele at each diallelic locus and two alleles at each triallelic locus (Table 2). At locus *Mnr–1*, where 4 alleles were found only two alleles were subjected to PCA, because $Mnr\text{–}1^{74}$ and $Mnr\text{–}1^{126}$ were rare variants. Similarly for *Pgm–1* only the most frequent allele $Pgm\text{–}1^{100}$ was considered. *Pgi–1* was not included in multivariate analysis due to its minor polymorphism. PCA is a method employed to describe patterns among the populations in a multidimensional space by which principal axes in this space are aligned sequentially in the direction of greatest variances. No *a priori* hypothesis is imposed, instead PCA is used to generate a hypothesis on the geographical pattern. Different population groups based on the PCA were compared using the Mann-Whitney test based on gene diversity calculated according to NEI's formula (1973) as $H_e = 1-\sum p_i^2$, where p_i is the frequency of ith allele. Gene diversities were compared using a non-parametric

test because their distribution is unknown. Mean allele frequencies were correlated to longitude and altitude, respectively. Mahalonobis' genetic distance was calculated as described in DE VIENNE & DAMERVAL (1985). Furthermore, a linear multiple regression analysis between allele frequencies, longitude and altitude was performed.

RESULTS

Principal component analysis of allele frequencies

The first three axes derived from PCA accounted for 58.8% of the variation explained. The first axis (31.8%, Fig. 2) clearly separated the French and Swiss populations (positive coordinates) from all the others (negative coordinates). This was due to higher frequencies of *Px–1*^{100}, *6-Pgd–1*^{100}, and *Idh–1*^{100} in France and Switzerland and to lower estimates for *Got–1*^{100}, *6-Pgd–1*^{84}, *Sod–1*^{100} and *Idh–1*^{116} in both regions (Table 1). The second axis (13.7%) corresponded to variations of allele frequencies at *Acp–1* and *Mnr–1* but, it did not separate geographical groups (Fig. 2). The third axis (13.3%) partially separated the French group (group 1 including one northwestern Italian population) from the Swiss group (group 2 including one French population and two German populations, close to Switzerland) (Fig. 3, Table 1). This separation is mainly due to the high frequency of *Pgm–1*^{100} in French populations as well as the different values for *Acp–1*^{84} and *Mdh–1*^{18}. It also discriminated well the western Austrian and south German populations (group 3 including 1 northern Italian population) from Hungarian and eastern Austrian populations (group 4) (Fig. 3, Table 2) due to high loadings of *Acp–1*^{100} and *Pgm–1*^{100}. Table 2 confirms these results. Mahalanobis' distance was lower between the western groups and the eastern groups than between any other pair of groups.

Table 3. Mahalanobis' genetic distances between groups. 1 – French Alps; 2 – Swiss Alps; 3 – Northeastern Alps and environs; 4 – East Austria and Hungarian Basin

Groups	1	2	3
2	2.183		
3	2.457	2.348	
4	2.445	2.585	2.227

Gene diversity (Nei's expected heterozygosity)

Comparison of gene diversity among groups (Tables 4 and 5) complements previous results. The Mann-

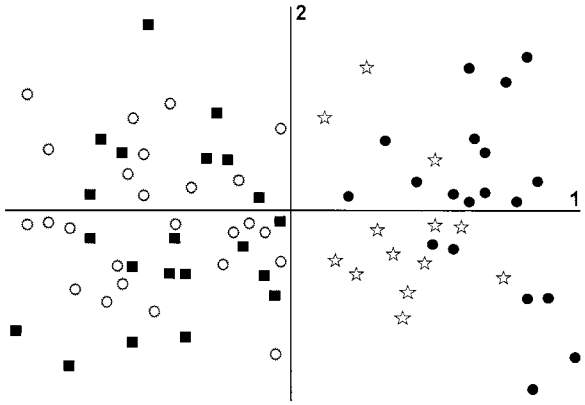

Figure 2. Principal component analysis including 10 loci and 15 variables (allelic frequencies)> Total variance explained by axis 1: 31.8 %, axis 2: 13.7 %. ●● – French Alps, ☆ – Swiss Alps, ○ – Northeastern Alps and environs; ■ – East Austria and Hungarian Basin.

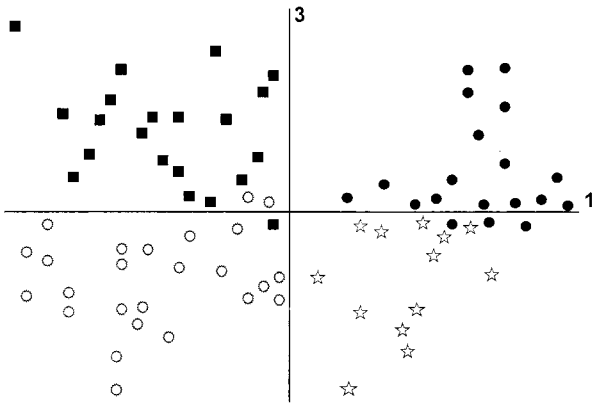

Figure 3. Principal component analysis including 10 loci and 15 variables (allelic frequencies)> Total variance explained by axis 1: 31.8 %, axis 3: 13.3 %. ● – French Alps, ☆ – Swiss Alps, ○ – Northeastern Alps and environs; ■ – East Austria and Hungarian Basin.

Whitney test compared the level of gene diversity between the 4 groups discriminated by the multivariate analysis. While gene diversity between French and Swiss populations (group 1 and 2) and between the northeastern Alps and eastern Austria including Hungary (group 3 and 4) did not differ significantly at most loci, other pair-wise comparisons showed distinct differences often at high probability levels.

Mean gene diversity (Table 4) increased slightly from west to east. Significantly higher estimates were found in the eastern groups 3 and 4 at *Idh–1, Mnr–1, 6-Pgd–1, Px–1,* and *Px–2* compared with the remaining populations while *Got–1* and *Sod–1* were very polymorphic in France and Switzerland and much lesser diversity was detected in eastern beech populations. Gene diversity increased at *Mdh–1* and *Pgm–1* from group 1 to groups 2 and 3 and decreased in group 4.

Table 4. Mean gene diversity within groups at the different loci

Locus	French Alps	Swiss Alps	Northeastern Alps (Germany and western Austria)	Eastern Austria and Hungarian Basin
Acp–1	0.449	0.415	0.349	0.531
Got–1	0.331	0.261	0.108	0.137
Idh–1	0.314	0.342	0.429	0.414
Mdh–1	0.341	0.406	0.437	0.353
Mnr–1	0.095	0.120	0.156	0.167
Pgi–1	0.030	0.017	0.020	0.022
Pgm–1	0.299	0.441	0.468	0.361
6–Pgd–1	0.110	0.125	0.235	0.272
Px–1	0.336	0.349	0.463	0.467
Px–2	0.368	0.343	0.417	0.393
Sod–1	0.334	0.345	0.156	0.095
Mean	**0.273**	**0.288**	**0.294**	**0.292**

Table 5. Comparison of gene diversity (NEI 1973) among groups. Non parametric Mann-Whitney's test. 1 – French Alps; 2 – Swiss Alps; 3 – Northeastern Alps and environs; 4 – East Austria and Hungarian Basin.

Locus	1–2	1–3	1–4	2–3	2–4	3–4
Acp–1	ns	ns	*	***	***	***
Got–1	**	***	***	***	***	ns
Idh–1	ns	***	***	***	***	ns
Mdh–1	**	***	ns	*	**	***
Mnr–1	ns	***	***	*	**	ns
Pgm–1	***	***	**	*	***	***
6–Pgd–1	ns	***	***	***	***	ns
Px–1	ns	***	***	***	***	ns
Px–2	ns	*	ns	**	*	ns
Sod–1	ns	***	***	***	***	ns

ns – non significant; * – $0.01 < p < 0.05$; ** – $0.001 < p < 0.01$; *** – $p < 0.001$

An exceptional high value at *Acp–1* was estimated for group 4.

Longitudinal and altitudinal gradients

Based on above-mentioned results it is tempting to speculate that significant correlations exist between certain allele frequencies and longitude. The multiple linear regression (data not shown) which included all populations showed that there is a strong negative correlation ($r = -0.585$) between longitude and elevation. This correlation is mainly due to the very different distribution of the altitudinal ranges of stands in the groups. Samples originating from eastern Austria and Hungary (group 4) were mostly taken from low elevations (< 700 m) and since the allele frequencies often differed from those of other groups collinearity is likely. Hence certain data may bias the multiple regression and were therefore excluded. The reduced data set was then subjected to an additional linear regression. This was done based on the 57 populations from groups 1, 2 and 3 (from France to western Austria) after grouping the populations into 8 altitudinal classes to reduce the differences in altitudinal range from one group to another. The negative correlation between longitude and elevation decreased noticeably ($r = -0.180$) and altitudinal and longitudinal effects were assumed to be separate.

Linear regressions showed that most allele frequencies vary along different longitudes (Table 6). Considering those allele frequencies for which correlations were established at a significant level of $p < 0.01$, frequencies of $Idh–1^{116}$, $Mnr–1^{63}$, $6–Pgd–1^{84}$, $Px–1^{105}$, and $Px–2^{39}$ increase from west to east while $Got–1^{105}$, $Idh–1^{100}$, $Mnr–1^{100}$, $6–Pgd–1^{100}$, $Sod–1^{112}$ decrease in the same direction. $Px–2^{13}$ is most frequent in Switzer-

Table 6. Significancy of correlations between allele frequencies and longitude or altitude for data of groups 1 to 3.

Locus	*Got–1*	*Idh–1*		*Mnr–1*		*6–Pgd–1*		*Px–1*	*Px–2*			*Sod–1*
Alleles	105	100	116	63	100	84	100	105	13	26	39	112
Longitude	***	***	***	**	**	***	***	***	**	**	***	***
Altitude	*		*						*			*

* – $0.01 < p < 0.05$; ** – $0.001 < p < 0.01$; *** – $p < 0.001$

Table 7. Survey of results obtained for beech with respect to correlation between allelic frequencies and altitude. + : positive correlation; – : negative correlation; (a) and (b), see comments in the text

Loci Alleles	*Acp–1*	*Got–1* *105*	*Idh–1*	*Mnr–1*	*Pgm–1* *100*	*Px–1* *105*	*Px–2* *13*	*Sod–1* *112*
Western Europe FELBER & THIÉBAUT 1984						+		
Atlantic range COMPS *et al.* 1987		+				+		
Central Europe COMPS *et al.* 1990						(a)	(b)	
France GÖMÖRY *et al.* 1992	92 +	+	100 – 116 +	63 – 100 +	+			
Spain and Pyrenees PORTAL 1993	100 + 108 –	+	116 –			+		–
Italy COULAUD 1994			84 –	100 – 126 +			–	–
Carpathians VYŠNÝ *et al.* 1995				100 +				
Present study		+	116 –	100 Group 1,2 + Group 3 –			+	+

land and decreases both in westwards and eastwards direction.

The analysis of genetic data in relation to altitudes is intricate because (1) strong correlations between most allele frequencies and longitude and (2) the confounding effect of altitude and longitude exist. Correlations between altitude and allele frequencies were tested as follows: (1) within each of the first 3 groups (groups exhibit considerable altitudinal variations) and (2) within pairs of groups only for allele frequencies which did not vary significantly between the two groups. Four allele frequencies are correlated with altitudes ($0.01 < p < 0.05$) (Table 6): $Got\text{–}1^{105}$ and $Idh\text{–}1^{116}$ both in groups 1 and 2 (positive and negative correlations, respectively), $Px\text{–}2^{13}$ and $Sod\text{–}1^{112}$ in group 3 (positive correlations).

Rare alleles

Rare alleles, not mentioned in Table 2, occurred in single populations only: $Got\text{–}1^{90}$ and $Pgi\text{–}1^{76}$ were found exclusively in group 3 (Germany and western Austria) while $Got\text{–}1^{95}$ and $Pgi\text{–}1^{113}$ were detected in group 3 as well as in group 4 (eastern Austria and Hungary).

DISCUSSION

A high degree of genetic polymorphism was found in the populations studied. In general estimates were very similar than those reported for other broadleaf species in Central Europe (see review by MÜLLER-STARCK & ZIEHE 1991).

At *Pgi–1* gene diversity was very low in all populations (H_T = 0.022). This finding was to be expected because gene diversity at this locus is generally very low in Central European beech populations, while diversity is very high in Southeastern Europe (GÖMÖRY *et al.* 1993). Overall estimates for the four groups as well as single locus estimates fit well to the H_T-values reported in beech of other Central European regions (*e.g.*, KONNERT 1995; BELLETTI & LANTERI 1996).

Multivariate analysis displayed 4 pools of beech populations and genetic distances increased with geographic distance. These results suggest an evident correlation between genetic structure of the beech populations studied and longitude. How can this correlation be explained biologically? Most authors who have studied postglacial beech migrations agree that the Balkan peninsula and southern Italy (Calabria) are important refuges and that the first one probably predominates (HUNTLEY & BIRKS 1983; HUNTLEY *et al.* 1989; VAQUIER 1995, for review see also LANG 1994). Genetic studies based on isozymes (e.g., GÖMÖRY *et al.* 1993) or chloroplast DNA (DEMESURE *et al.* 1996) support fossil pollen finding in this respect. Very likely beech immigrated from the south toward the north and the west. It is interesting in this context that gene diversity estimates of the populations studied in this paper are slightly lower than in the foothills of southern Alps (cf. present data with BELETTI & LANTERI 1996) but higher compared to beech populations in the northern Alpine Foreland (cf. present data with KONNERT 1995).

The longitudinal gradient of genetic structures observed in the beech populations studied seems to confirm the role of migration history in shaping these structure patterns. Beech stands from Hungary and eastern Austria are surely older in evolutionary terms than those located on the northern slope of the Alps in western Austria, Germany and Switzerland. This may explain the different degrees of genetic differentiation observed. Therefore, the geographic pattern in beech in the Alpine range is similar to that of silver fir, for which also longitudinal trends were detected (BREITENBACH-DORFER *et al.* 1997). The group from the French Alps may have evolved in a more complex way (COMPS *et al.* in preparation). It is probably the result of at least two migration paths occurring at different times, one along the northern slope of the Alps, and a second through northern Italy. Furthermore it cannot be surely excluded that small refuges in southern France existed (JALUT *et al.* 1975; TRIAT-LAVAL 1979) which may have played a role in shaping the genetic structure; these groups are genetically similar to Pyrenean populations as well as to Swiss ones (GÖMÖRY *et al* 1992b; COULAUD 1994). Allele frequencies and gene diversity did not vary in an identical fashion at all loci with longitude: 6 out of 10 loci present a greater gene diversity in the eastern part of the Alps, while only 2 out of 10 loci are more polymorphic in France and Switzerland. Therefore mean diversity is slightly higher in groups 3 and 4. This may confirm some previous results which show that beech gene diversity tends to be higher in evolutionary older populations, particularly in southern refuges (GULLBERG *et al.* 1985; COMPS *et al.* 1991a).

Studies of the occurrence of rare alleles in Europe (COULAUD 1994; HAZLER *et al.* 1997; COMPS *et al.* in preparation) show that they are most frequent in the eastern part of Europe, in the Balkan peninsula and in Italy, that is to say in areas which are known to have been refuges during the last glaciation. The presence of these rare alleles may be a more convincing argument than gene diversity to confirm existence and location of these refuges. Austria and southern Germany are the northwestern limits for the rare *Got-1*^{90}, *Got-1* ^{95}, and *Pgi–1*^{76}, which seems to confirm the northern slope of the Alps as an important migration path toward the west.

Correlations between allele frequencies and altitude have been observed in several previous studies on genetic variation in beech (FELBER & THIEBAUT 1984; COMPS *et al* 1987, 1990; GÖMÖRY *et al.*1992b; PORTAL 1993; VYŠNÝ *et al.*1995). Conversely LEONARDI & MENOZZI (1995) found no significant correlations. In their study, genetic differentiation was pronounced probably due to very diverse ecological conditions especially with respect to elevation. However, allele frequencies at isozmye loci did not vary with altitudes. Which conclusion can be drawn from these different findings? In different parts of the range allele frequencies at *Got–1* often vary with altitudes (see references given in Table 7). In all cases positive correlation appears for the most rare allele *Got–1*^{105}, indicating that the polymorphism of beech stands increases significantly with altitudes, sometimes with a probability of $p < 0.001$ (COMPS *et al.* 1987). Interpretation of the allelic structure at *Px–1* is more complex. Generally, polymorphic populations occur predominantly in areas outside the optimal range of beech where climatic conditions limit the distribution. Thus polymorphism at this locus is higher at high altitudes close to subalpine areas or at low altitudes in Mediterranean regions

where temperature and aridity become limiting (FELBER & THIEBAUT 1984; THIEBAUT 1984; COMPS *et al.* 1987; PORTAL 1993). Diversity at this locus is mainly low in regions where beech encounters optimal site conditions and frequency of $Px\text{–}1^{105}$ is not significantly correlated with altitudes (COMPS *et al.* 1990). Since only very few populations were sampled at high altitudes in the present study, this may explain the lack of significant correlation between $Px\text{–}1^{105}$ and elevation (Table 7, a). However, it is surprising that no correlatio-n was found at *Px–1* in Italy (COULAUD 1994).

Correlations between allele frequencies at *Px–2* and elevation do not generally exist. However, frequency of $Px\text{–}2^{13}$ was found to be negatively correlated in COULAUD (1994) and positively correlated in the present study (Table 7, b). Gene diversity at *Px-2* was found to be high in regions where the altitudinal range can vary erratically (COMPS *et al.* 1990). This study does not confirm these results, however, particularly in group 4 where a very low altitudinal range in the sampled stands (500 m) corresponds to a relatively high diversity at this locus.

Concerning other isozyme loci, correlations between allele frequencies and altitude are more ambiguous. In fact, some indications were found that certain loci may be selectively adaptive with respect to elevation (*Acp–1*, *Pgm–1;* GÖMÖRY *et al.* 1992) or data suggest contradictory trends in different regions at *Idh–1* and *Mnr–1* (see references given in Table 7). So, with exception of *Px–1* and *Got–1*, associations of allele frequencies with altitudes are very vague and this can be interpreted that loci under study are probably adaptively neutral. Moreover, results suggest that beech populations in a given region are not particularly differentiated and the effects of migration and gene exchange contribute to rather large effective population sizes. This may also be interpreted as a sign of limited adaptation to local conditions, as has been stated for some other species, primarily conifers (MÁTYÁS 1996).

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