Contrasting patterns of historical colonization in white oaks (*Quercus* spp.) in California and Europe

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Abstract

Phylogeography allows the inference of evolutionary processes that have shaped the current distribution of genealogical lineages across a landscape. In this perspective, comparative phylogeographical analyses are useful in detecting common historical patterns by either comparing different species within the same area within a continent or by comparing similar species in different areas. Here, we analyse one taxon (the white oak, genus *Quercus*, subgenus *Quercus*, section *Quercus*) that is widespread worldwide, and we evaluate its phylogeographical pattern on two different continents: western North America and Western Europe. The goals of the present study are: (i) to compare the chloroplast genetic diversity found in one California oak species vs. that found in the extensively studied European oak species (in France and the Iberian Peninsula); (ii) to contrast the geographical structure of haplotypes between these two taxa and test for a phylogeographical structure for the California species. For this purpose, we used the same six maternally inherited chloroplast microsatellite markers and a similar sampling strategy. The haplotype diversity within site as well as the differentiation among sites was alike in both taxa, but the Californian species has higher allelic richness with a greater number of haplotypes (39 vs. 11 in the European white oak complex). Furthermore, in California these 39 haplotypes are distributed locally in patches while in the European oaks haplotypes are distributed into lineages partitioned longitudinally. These contrasted patterns could indicate that gene movement in California oak populations have been more stable in response to past climatic and geological events, in contrast to their European counterparts.

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Introduction

The distribution of genealogical lineages on a landscape provides valuable information about the evolutionary history of species (Avise *et al*. 1987; Avise 2000) and regions of high conservation interest (Avise & Hamrick 1996; Smith & Wayne 1996; Moritz & Faith 1998). When we have such phylogeographical information for sets of species in a given region, we can gain additional insight about common historical and geographical patterns of this area (Arbogast & Kenagy 2001; Lapointe & Rissler 2005). The multispecies approach to comparative phylogeography (Bermingham & Moritz 1998) is particularly informative for a newly studied species when a set of species shows a congruent phylogeographical pattern in the examined area. An alternative multispecies approach to comparative phylogeography contrasts the phylogeographical structures of taxa from disjunct areas characterized by distinct climatic and geological histories (e.g. Magni *et al*. 2005). This alternative is beneficial when the local region of the species of interest does not have a congruent phylogeographical pattern, and when the region of comparison is well understood phylogeographically.

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In this study, we will illustrate the usefulness of this second approach with the case study of the genus *Quercus* (oak), a taxon that occurs throughout the northern hemisphere on continents that have experienced various climatic and geological changes. We will compare two oak taxa that belong to the same white oak section (subgenus *Quercus*, section *Quercus*), with overlapping latitudes at 36–41°N in regions of similar Mediterranean climate, but occurring in two different continents: western North America and Western Europe. We will compare the genetic structure of a Californian oak (*Quercus lobata*), for which no range-wide genetic data are available, with that of the European white oak complex, for which a well-founded documentation of the Pleistocene impact is available and which shows a clear geographical structure (Brewer *et al.* 2002; Petit *et al.* 2002a; Petit *et al.* 2002b). The motivation for this comparison is that different studies in California suggest a general phylogeographical pattern for fauna but no congruence among flora (Calsbeek *et al.* 2003; Thompson & Calsbeek 2005; but see Lapointe & Rissler 2005), making it difficult to interpret the phylogeographical pattern of the California oak relative to that of other codistributed taxa. Indeed, some plant and animal studies in California show a north–south split of spatial genetic structure near to the Transverse Ranges, as well as an east–west split between the Sierra Nevada and the Coastal Ranges (Liston *et al.* 1992; Wake 1997; Ledig 2000; Dawson *et al.* 2001; Calsbeek *et al.* 2003; Jacobs *et al.* 2004), suggesting that geographical barriers, possibly combined with climatological changes, may have affected the Californian fauna and flora (see Calsbeek *et al.* 2003; Lapointe & Rissler 2005). At a wider geographical scale, chloroplast (cp) genetic diversity for plants from Pacific North America has shown a trend for a north–south partitioning of genotypes that suggests two scenarios of recolonization processes (Soltis *et al.* 1997), both pointing to California as a large regional refugium for some species.

In contrast to the complex western North American pattern, syntheses of comparative phylogeographies of plants and animals in Europe show some common historical patterns among species, although individual species responded in diverse ways to those past historical events. In particular, the routes and speed of recolonization of many species mainly resulted from the impact of the Last Glacial Maximum (LGM), as well as from some climatic and topographic factors (Taberlet *et al.* 1998; Hewitt 2000). These common historical patterns are particularly evident in the white oak species complex, for which genetic structure and recolonization routes have been extremely well documented using chloroplast markers (Brewer *et al.* 2002; Petit *et al.* 2002a; Petit *et al.* 2002b). Overall, the results show a reduced diversity and high levels of fixation. Moreover, the geographical structure indicates a loss of haplotype richness northward and a longitudinal partitioning of the chloroplast lineages with discrete western, central and eastern lineages. The combination of these genetic data with palynological information has led to the conclusion that oaks migrated mainly northward, starting from three major southern refugia (Iberia, Italy and the Balkans), although largely cryptic refugia might have existed further north, perhaps up to 50°N (Brewer *et al.* 2002; Petit *et al.* 2002a). The main factors that controlled the spread of oaks were the climatic conditions (temperature and moisture) of the Late Glacial and Early Holocene (Holocene: 10 000 years to present), physical barriers such as the Alps, and competition between species (Brewer *et al.* 2002).

To conduct our comparative study, we used identical chloroplast microsatellite markers [simple strand repeat (SSR)] on trees of Californian valley oak (*Q. lobata*, distributed across the whole range) and European white oaks (five different species characterized by extensive cytoplasm sharing and sampled in France and the Iberian Peninsula: *Quercus robur*, *Quercus petraea*, *Quercus canariensis*, *Quercus faginea* and *Quercus pyrenaica*). Chloroplast markers are maternally inherited in oaks (Dumolin *et al.* 1995), allowing us to track recolonization processes through seeds. A previous study showed that chloroplast simple sequence repeats (cpSSRs) yield similar genetic patterns to chloroplast polymerase chain reaction-restriction fragment length polymorphism (cpPCR-RFLP) at the French scale for *Q. petraea* and *Q. robur* (Deguilloux *et al.* 2004) and, thus, constitute appropriate markers to infer historical recolonization processes in oaks. In California, we selected valley oak as our focal species because it is the most threatened oak species. However, we collected the other species of white oak whenever they were present in the same site. Even though *Q. lobata* has the potential to hybridize with other species (Miller & Lamb 1985), it does not do so in high frequency (e.g. Craft *et al.* 2002; Grivet and Sork, unpublished data). Thus, this study will focus on valley oak and we will discuss how the inclusion of other taxa will not change our results. Our two specific objectives were: (i) to compare the chloroplast genetic diversity found in California oak vs. that found in Europe; (ii) to contrast the geographical structure of haplotypes between these two taxa and test for a phylogeographical structure.

Material and method

The species

Quercus lobata (Née). The Valley oak is a deciduous tree endemic to California and native to Western USA. This tree is a major component of the oak savanna, oak woodland, and riparian forest habitats, especially before extensive agricultural clearing and flood control activities (groundwater pumping) began in the late 1880s (Griffin 1988; Pavlik *et al.* 1991). Valley oak is distributed in the Central

Valley (in scattered remnant patches or isolated trees) and surrounding foothills and valleys of the Sierra Nevada Mountains, Coast Ranges and Transverse Ranges, preferring deep, rich bottomland soils at elevations generally below than 600 m, some populations occurring above 1500 m (Griffin & Critchfield 1972). Acorns mature in one season and are dispersed by gravity, acorn woodpeckers, scrub jays, and possibly by small rodents. This species also hybridizes with other oak species (Nixon 2002) such as *Q. douglasii*, *Q. dumosa* and *Q. garryana* (Craft *et al.* 2002; V. Sork and D. Grivet, personal observation), but we do not address the issue of hybridization in this study because of our limited sampling and number of sites where these taxa are sympatric. Our preliminary analyses indicate that inclusion of other oak species will not change the findings reported here.

European white oak complex. We analysed five oak species from France and the Iberian Peninsula. In France, we analysed the two sympatric species *Quercus robur* and *Quercus petraea*. In Spain, we analysed three additional oak species: *Quercus canariensis*, *Quercus faginea*, and *Quercus pyrenaica*. Extensive cytoplasm sharing among European white oak species has been reported (Dumolin-Lapegue *et al.* 1998, 1999 for France; Olalde *et al.* 2002 for Spain), and the cpDNA pattern reflects thus both historical gene flow and introgression. However, the systematic local sharing of haplotypes for the different oak species, combined with their different ecological requirements and ability to hybridize, allowed the interpretation of their pattern of historical colonization without interference with the introgression process (Petit *et al.* 2002c). Acorns are dispersed by gravity, birds (particularly the Eurasian jay) and mammals.

Brief history of climatic and geological events

Unlike Europe, where the Quaternary period had a major impact on species distribution (e.g. Hewitt 2000), most major climatic and continental changes in California occurred during the Tertiary and this period probably played the pivotal role in Californian species distribution (Millar 1996). Here, we briefly summarize the geological and climatic major events that characterized California from the Tertiary and later periods, and attempt to connect these events to the distribution of oak populations. About 20 species of oaks appeared in California at the Early Miocene [24 million years ago (Ma)] (Keator 1998), a period characterized by the establishment of the Mediterranean climate (Raven & Axelrod 1978). These favourable climatic conditions can be connected to the establishment of oak woodland that became dominant in west-central California before the Middle Pliocene (Pliocene: 5.3–1.8 Ma) (Griffin 1988). During the Late Pliocene and continuing throughout the Pleistocene (Pleistocene: 1.8 Ma to 10 000 years), uplifts

of the Sierra Nevada and the Coastal Ranges occurred due to the movement of the Pacific Ocean floor under the North American plate (for a recent brief review on uplifts on the West Coast, see Jacobs *et al.* 2004). These mountain ranges may have influenced oak distribution between high and low elevations on the west side of the Sierra Nevada and between coastal and inland areas in the Coastal Ranges. In western North America, climatic conditions during the Pleistocene were different from those in Europe. While neither the ice sheet nor the permafrost reached California, presumably cooler temperature and increased rainfall during this period (Thompson *et al.* 1993) influenced the distribution of the California flora. Scarce pollen data available in California suggest that oak populations followed climate variations between glacial and interglacial periods (Heusser 2000; Pisias *et al.* 2001), and this response to climate changes must have left similar genetic fingerprint on several oak taxa despite their different ecological specificities.

The recent history of Europe related to oak history is described in detail elsewhere (Brewer *et al.* 2002; Petit *et al.* 2002a). In summary, oak populations in Europe have been greatly affected by the climatic changes between glacial and interglacial periods during the Quaternary. In particular, the low levels of temperature and moisture characterizing the LGM limited the distribution of oaks to parts of Europe where favourable topography and microclimates existed (mostly along the Mediterranean borderlands and around the Black Sea; Brewer *et al.* 2002). These climatic conditions resulted in vegetation principally composed of herbaceous plants (Pons 1984; cited in Brewer *et al.* 2002), leaving vast open space for the early colonization of oaks. The spread of oaks took place in two steps (Brewer *et al.* 2002): (i) in the Late Glacial (13 000–11 000 years BP) the distribution was correlated with shifts in climate (increased temperature and precipitation); (ii) and in the beginning of the Holocene (10 000 years BP to present) the changes were controlled by competition between species, landscape topography (e.g. the Alps), and other edaphic factors.

Sampling

In California, we sampled leaves from two to eight individuals per population (mean: 3.2) of *Q. lobata* from 35 sites, yielding a total of 111 individuals. After all collections, leaf tissue was stored in –80 °C pending DNA extraction.

For the Europe oak species, we used DNA from one to five individuals per population (mean: 4.0) from 92 sites, yielding a total of 367 individuals. These DNA samples have been obtained from previous studies where they had been analysed for cpPCR-RFLP: 48 populations in France (116 individuals, cf. Petit *et al.* 2002c) and 44 populations in the Iberian Peninsula (251 individuals, cf. Olalde *et al.* 2002).

Laboratory methodology

DNA isolation for the California samples. Frozen leaves were ground with the Mixer Mill MM 301 (Retsch), and Doyle & Doyle (1990) protocol has been used for DNA isolation.

Microsatellite screening. Six chloroplast microsatellites have been used to screen for chloroplast polymorphism for both French-Iberian and Californian populations. Ccmp10 was designed on *Nicotiana tabacum* (Weising & Gardner 1999), while μdt1, μdt3, μdt4, μcd4, and μcd5 were designed on *Quercus robur* (Deguilloux *et al.* 2003). The PCR conditions were performed with the following profile: 5 min at 94 °C followed by 25 cycles of 1 min denaturing at 94 °C, 1 min annealing at the appropriate annealing temperature (following author's indications: Weising & Gardner 1999; Deguilloux *et al.* 2003), and 1 min of elongation time at 72 °C, with a final extension step of 8 min at 72 °C. The PCR solution contained 1 × reaction buffer (Promega), the four dNTPs (each 0.2 mM), 2.5 mM of MgCl$_2$, 0.2 μM of each primer, 0.5 mg/mL of BSA, and 0.5 U of *Taq* polymerase (Promega). After amplification, PCR products were ran at least twice on acrylamide gel. The colour codes used to represent the European SSR haplotype-lineages have been chosen in accordance to the cpPCR-RFLP haplotype-lineages found in France and Iberian Peninsula (Petit *et al.* 2002c; Olalde *et al.* 2002).

Microsatellite sequencing. For each length variant identified in Californian or French populations, one individual was sequenced to check that the observed polymorphism was due to a difference in the number of repeats rather than a modification of the flanking regions. Repeat number from Iberian length variants was deduced from the size of the PCR product run on acrylamide gel with controls of known number of repeats.

Statistical analyses

Genetic diversity. We estimated genetic diversity and differentiation using the program called CPSSR (www. pierroton.inra.fr/genetics/labo/Software/), as described in Pons & Petit (1995, 1996): the within-population diversity (H_S), the overall diversity (H_T), the differentiation among-populations (G_{ST}), as well as the equivalent coefficient of differentiation (R_{ST}), which takes into account the similarities among haplotypes (by accounting for variance in allele size).

To control for the difference in sampling size and microsatellite lengths when comparing genetic diversity of the two oak taxa, we used a double standardization method: first, by using rarefaction, and, then by accounting for different mean number of repeats (Petit *et al.* 2005). Rarefaction size was set to 100 individuals and computed using the program, RAREFAC (www.pierroton.inra.fr/ genetics/labo/Software/). The expected number of different alleles in equal-sized samples of 100 is noted $\hat{A}_{[100]}$. We then used equation 1 of Petit *et al.* (2005) to compute $\hat{A}_{[100,10]}$, the standardized value of allelic richness for a mean number of repeats = 10. We chose this value of 10 as it is close to the overall mean value in the two species (10 and 11 for the European and American populations, respectively, Table 1).

Spatial genetic structure. The program CPSSR was further used to test for the existence of a phylogeogeographical structure (Burban *et al.* 1999): the directly measured R_{ST} values were compared with those obtained after random permutation of haplotype identities, and we tested whether R_{ST} and G_{ST} were significantly different. When $R_{ST} > G_{ST}$, it means that similar alleles are geographically close to each other, suggesting that the populations have a phylogeographical structure.

Primers	European oaks		California valley oak	
	# alleles	# repeats	# alleles	# repeats
μdt1	3	11, 12, 13	4	11, 12, 13, 14
μdt3	4	8, 9, 10, 11	4	10, 11, 13, 14
μdt4	3	9, 10, 11	2	11, 12
μcd4	3	10, 11, 12	4	10, 11, 12/13*, 14
μcd5	2	8, 9	3	9, 10, 11
ccmp10	2	9, 10	4	9, 10, 11, 12
mean number	3.33	10	3.67	11
$\hat{A}_{[100]}$	9.53		36.35	
$\hat{A}_{[100,10]}$	9.47		34.17	
# haplotypes	11		39	

Table 1 Allelic richness and number of repeats (poly A/T) for the six microsatellites, and number of haplotypes (standardized for the difference in sampling size and microsatellite lengths) for California and European oak populations

*, ambiguity of the exact number of repeats for this allele (two sequences of the same individual indicate 12 poly A/T in one case, and 13 poly A/T in the second case).

To determine the scale of historical gene flow among populations, we performed a spatial autocorrelation analysis based on genetic distance methods for multiallelic and multilocus loci (Smouse & Peakall 1999) with the GENALEX version 5 software (www.anu.edu.au/BoZo/GenAlEx/).

Results

Comparison of the cpSSR variation in the French-Iberian and Californian populations

For five of the six microsatellite markers, the California valley oak samples displayed an equal or higher number of alleles compared to the European oak complex (Table 1). Furthermore, all markers showed a higher number of repeats in the Californian populations compared to the French-Iberian populations (Table 1). The combination of alleles from the six loci resulted in a total of 39 haplotypes for the California valley oak compared to 11 in Western Europe oaks (Table 1).

The California oak populations displayed similar genetic diversity to that of the European oak populations, i.e. low within population diversity ($H_S = 0.285$ vs. 0.114) and substantial overall population diversity ($H_T = 0.979$ vs. 0.755). This diversity was similarly partitioned among populations in California ($G_{ST} = 0.709$, $R_{ST} = 0.824$) and in Western Europe ($G_{ST} = 0.849$, $R_{ST} = 0.838$).

Geographic distribution of the cpSSR haplotypes. The 11 haplotypes detected in the French-Iberian populations belong to different lineages (Fig. 1b) originating from distinct refugia and they followed different migration routes. By contrast, the 39 haplotypes of *Quercus lobata* are locally clustered across California — nearby populations sharing the same haplotypes — and show an east–west partition: 28 haplotypes (including all 16 found only once) are located within 16 western sites ($n = 54$) and 10 haplotypes are located within 16 eastern sites ($n = 47$) (Fig. 1a).

For the European populations included in this study, R_{ST} and G_{ST} were not significantly different from each other
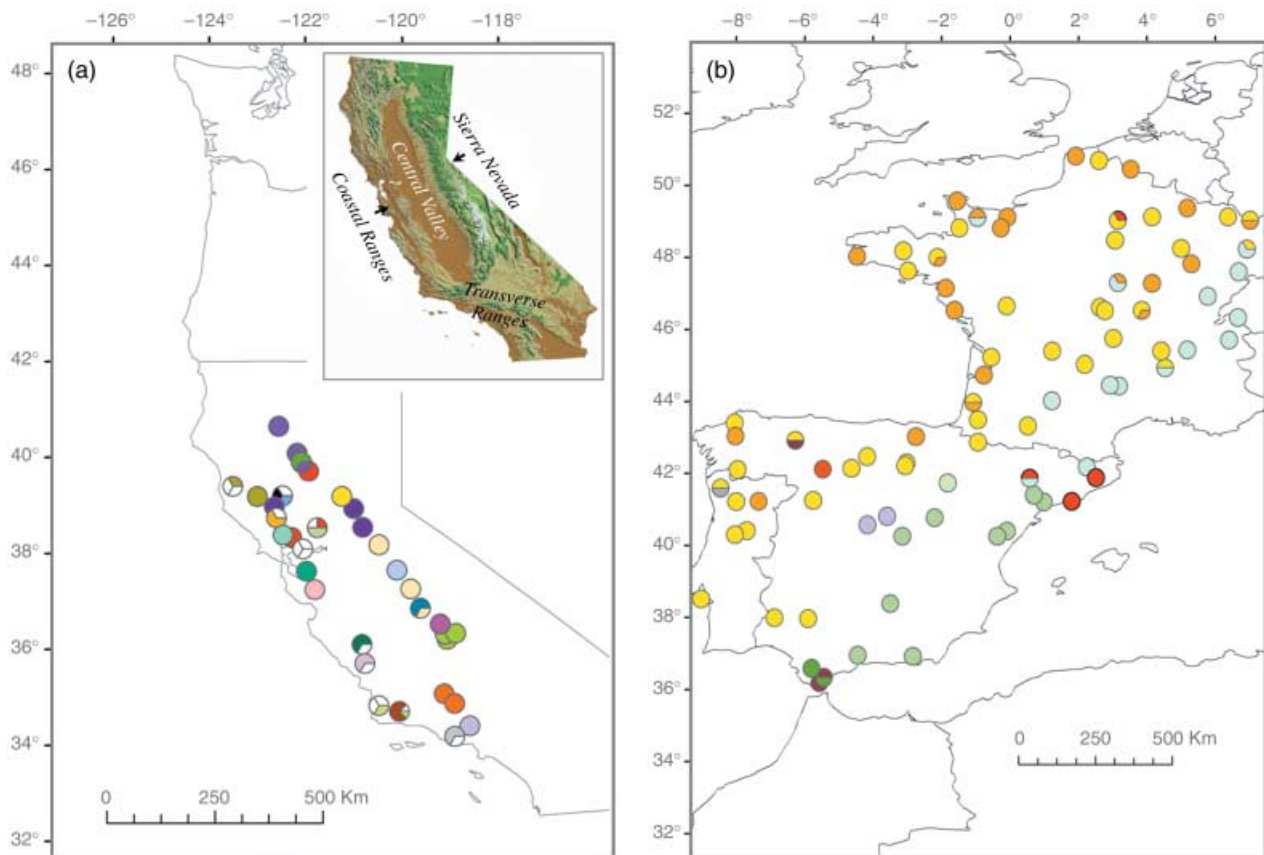
Fig. 1 Geographic distribution of (a) the 39 chloroplast microsatellite haplotypes across California (the white colour corresponds to haplotypes found only once); (b) the 11 chloroplast microsatellite haplotypes across France and the Iberian Peninsula. (Circle diameters are not proportional to sample size). The California shaded relief map was produced by the USGS (http://geography.sierra.cc.ca.us/booth/California/other/cal_links.htm).

($P = 0.694$). In contrast, for the California populations, R_{ST} was significantly higher than G_{ST} ($P = 0.006$) indicating the presence of a phylogeographical structure for *Q. lobata* at the studied scale.

Spatial genetic structure. Spatial autocorrelation analysis identified a significant clustering for European oak populations distant from 250 km or less ($0 \leq r \leq 0.2$; Fig. 2a). The California oak populations showed significant correlation within the first 200 km and the range of those values were greater than those found in the European data ($0 \leq r \leq 0.3$; Fig. 2b).

Discussion

Our comparison of two white oak taxa living in continents with different histories reveals contrasting patterns of genetic structure, suggesting that California valley oak has a very different evolutionary history than that of the European oak complex. The higher haplotype richness and the patchy genetic structure observed in California valley oak populations suggest that these populations were not affected as strongly by past climatic changes as those from Western Europe. By benchmarking the California populations against the European ones, it appears that California populations were more persistent locally, maintaining more ancient genetic structure, and without evidence of recent contraction to a few refugia.

Allelic richness provides one source of evidence indicating that environmental changes during the Pleistocene and earlier epochs did not seriously affect California oak populations. We found higher allelic richness in *Quercus lobata* for all six microsatellite markers but one, which translates into a more than threefold higher number of haplotypes in California valley oak (39) compare to the European oak complex (11). The most likely explanation for this difference would be that the Californian populations had not experienced a bottleneck like the European populations, allowing them to accumulate and conserve more diversity through time. Ascertainment bias (Ellegren *et al.* 1995; cited in Crawford *et al.* 1998) can be ruled out, since five of the microsatellites have been designed for European oaks and the sixth was based on *Nicotiana* cpDNA sequence. In any case, we used a recent method developed to control for both sample size and number of repeats while comparing the diversity of the two oak species (Petit *et al.* 2005). The standardized estimates confirm that the higher allelic richness for *Q. lobata* is not an artefact of the use of longer alleles in one species compared to the other, even though the difference between the California oak species and the European oak complex is reduced a bit once the data are corrected.

The disparity between the two sets of populations in the geographical structure of their haplotypes provides additional evidence that the two oak taxa have different histories of gene movement. First, in the California species, there is evidence of a phylogeographical structure ($R_{ST} > G_{ST}$), implying a greater local stability of the cpDNA lineages relative to their constitutive haplotypes in these populations. Second, the abundant haplotypes in California are organized in geographically localized patches, whereas haplotypes in Europe are restricted to lineages partitioned longitudinally (see Petit *et al.* 2002c). These distinct distributions are reflected by the autocorrelation patterns: both oak species show a positive correlation between genetic and geographical distances for relatively short distances (approximately 200 km), but the Californian populations show a stronger correlation that vanishes faster compared to the European ones. This pattern indicates that California populations did not colonize vast areas as did their European counterparts.

The genetic structure observed for valley oak in California most likely reflects the impacts of the Tertiary, a period characterized by most climatic and continental changes (Millar 1996). The palynological records from the Sierra Nevada (Woolfenden 2003), the Coastal Ranges (Adam *et al.* 1981; Heusser 1995), and the Central Valley (Davis 1999) favour the scenario that Californian populations have not gone totally extinct for a very long time (from the late Pleistocene and on). The pollen cores globally show that oak is rare in California, and that populations declined during glacials and increased during interglacials (Heusser 2000; Pisias *et al.* 2001). The patchy distribution of the Californian haplotypes may reflect the repeated range
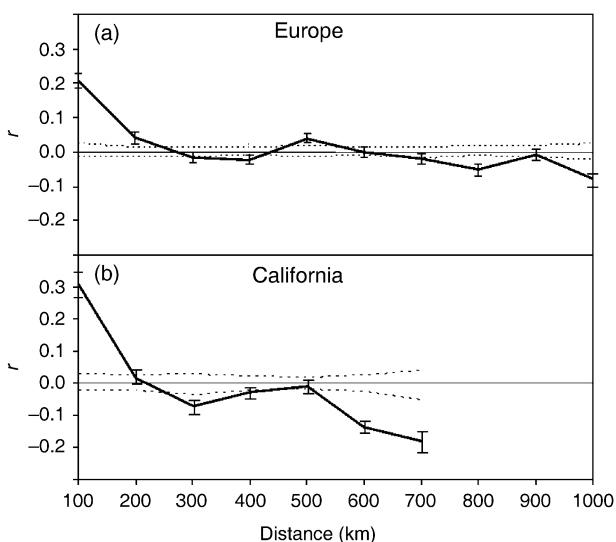
Fig. 2 Correlogram showing the genetic correlation (a) among pairs of European oak individuals and (b) among pairs of California oak individuals, as a function of distance (in kilometres). The dashed lines bound the 95% confidence interval about the null hypothesis of no spatial genetic structure.

contractions, expansions, and migrations of oak populations during successive glacial and interglacial periods. This scenario has also been proposed for California red oaks (*Quercus agrifolia*, *Quercus kelloggii*, *Quercus wislizeni* and *Quercus parvula*) by nuclear data that show a high population differentiation at the California scale (Dodd & Kashani 2003).

In North America, the only other oak species studied so far across its entire range is the northern red oak (*Quercus rubra*), which is found in central and eastern North America (Magni *et al*. 2005). This species shows a lower level of genetic structure than the European oaks and is characterized by one major haplotype geographically widespread. These results might be explained by the persistence of populations further north, compared to Europe, resulting in a more restricted colonization, and by increased seed movements in red oaks compared to white oaks, supported by recent ecological evidence (Steele *et al*. 2004). The climatic conditions during the LGM were different in the East coast than in Europe or in California: oak populations were close to the ice sheet leading to less opportunity of long distance events and founding events, and only the Appalachians, which run southwest and northeast, constituted a geographical barrier (Magni *et al*. 2005). These interspecific and intercontinental comparisons point out the importance of climatic, geological, and recolonization processes in shaping taxa genetic diversity.

An issue that often arises when discussing genetic patterns of oaks is the impact of hybridization. In Europe, several studies have shown that the species status in oaks does not need to be considered when looking at the haplotype distribution because of extensive cpDNA exchanges between species (references cited in Petit *et al*. 2002b). Haplotypes are shared among sympatric species and only the very rare ones are restricted to one species, resulting in similar genetic partitioning across the species range. The consequence is that each of the five European species analysed in our study show, when analysed individually at the European scale (Petit *et al*. 2002b), lower overall haplotype diversity, lower within population diversity, but higher genetic differentiation among populations (except for *Quercus pyrenaica*, this species representing only 21% of our sample size) compared to *Q. lobata*. However, the direction and speed of recolonization of the different species may have been influenced by their ecological behaviour (Petit *et al*. 2002b).

For California oaks, we have analysed the samples of the two other white oak species that have the potential to hybridize with *Q. lobata*. If we were to include those data, the differences in allelic richness with those of Europe would be even greater, increasing the total number of haplotypes to 45. We found five haplotypes unique to *Quercus douglasii* (eight populations sampled, five in sympatry with *Q. lobata*), and one haplotype unique to *Quercus garryana* (six populations sampled, none in sympatry with *Q. lobata*). Unlike the European oaks and unlike the work of Whittemore & Schaal (1991) on the white oaks of the eastern United States, these Californian white oak species do not always share the same haplotypes when sympatric. *Quercus douglasii* and *Q. garryanna* are capable of sharing the same haplotype with *Q. lobata* (*Q. lobata* share four haplotypes with *Q. douglasii*, and four with *Q. garryana*), but in only two out of the five localities with both *Q. lobata* and *Q. douglasii*, did they share the same haplotypes. We did not find all three species at the same locality, but we found one haplotype shared by all three in adjacent sites (north of San Francisco). Our tentative conclusion that white oak in California hybridize less frequently than those in Europe is substantiated by a recent study on nuclear genetic variation in *Q. lobata* and *Q. douglasii*, which found minimal hybridization (Craft *et al*. 2002). Further work is now underway in our laboratory to identify congeneric gene exchange in California white oaks.

The observation that haplotypes are so locally restricted in California is quite unexpected given that these populations seem to have been present from long past. Our tentative explanation is that populations contracted significantly at various times during periods of unfavourable climatic conditions (as supported by pollen data) and expanded through subsequent local diffusion and limited long distance dispersal. This limited long distance dispersal, which is quite different from the European pattern, might reflect that valley oak acorns were less frequently dispersed at long distances. Alternatively, colonization success may have been low following occasional long distance dispersal events, due to a lack of adaptation to new local conditions or to competition with established seedlings in the local populations. In Western Europe, the presence of a vast plain and lower competition with other taxa would have allowed oak populations to migrate over much larger distances, and across wider available space. Thus, we conclude that the patchy and localized distribution of haplotypes observed in valley oak reflects a range expansion at short distances from discontinuous populations. The preliminary analysis of *Q. douglasii* and *Q. garryana* indicates that these two species also show this patchy structure (Fig. 3), thus supporting our hypothesis that the geological history of California is a primary shaper of the current genetic structure of oak populations.

In conclusion, the high genetic diversity and patchy genetic structure found in California oak populations — which contrast with the pattern of their European counterparts — suggest that California valley oak populations were more stable in response to past events that affected them: (i) California oak populations have been less strongly affected by past climatic changes, and, in particular, these populations did not experience a great impact due to the LGM;
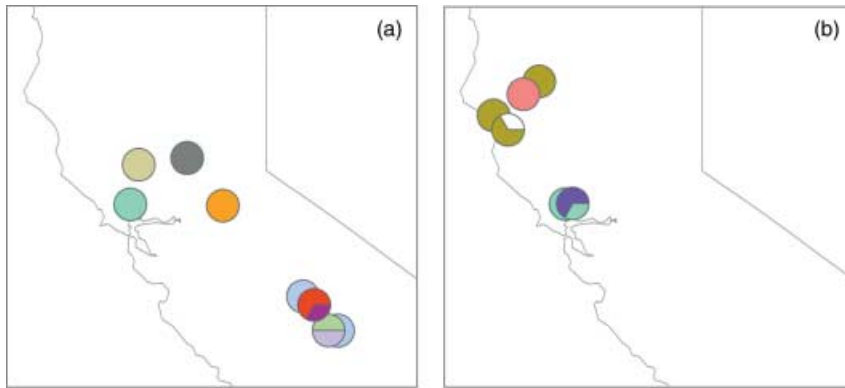
Fig. 3 Geographic distribution of the chloroplast microsatellite haplotypes for samples collected for (a) *Quercus douglasii* and (b) *Quercus garryana*.

(ii) the spread of California oak populations has been more restricted across the landscape, due either to less long distance dispersal events or to poor colonizing success. Our study illustrates the usefulness of interregional phylogeography in inferring the past-history of California oaks in an area of complex evolutionary history.

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