

1 Matti J. Salmela^{1,2}, Stephen Cavers², Witold Wachowiak^{2,3}, Joan E. Cottrell⁴, Glenn
2 Iason⁵ & Richard A. Ennos¹
3
4 ¹ Institute of Evolutionary Biology, School of Biological Sciences, Ashworth
5 Laboratories, University of Edinburgh, Edinburgh EH9 3JT, UK
6 ² NERC Centre for Ecology and Hydrology Edinburgh, Bush Estate, Penicuik, Midlothian
7 EH26 0QB, UK
8 ³ Institute of Dendrology, Polish Academy of Sciences, Parkowa 5, 62-033 Kórnik,
9 Poland
10 ⁴ Forest Research, Northern Research Station, Roslin, Midlothian EH25 9SY, UK
11 ⁵ Macaulay Land Use Research Institute, Craigiebuckler, Aberdeen AB15 8QH, UK
12

13 **Understanding the evolution of native pinewoods in** 14 **Scotland will benefit their future management and** 15 **conservation**

17 **Summary**

18
19 Scots pine (*Pinus sylvestris* L.) is a foundation species in Scottish highland forests and a
20 national icon. Due to heavy exploitation, the current native pinewood coverage
21 represents a small fraction of the postglacial maximum. To reverse this decline, various
22 schemes have been initiated to promote planting of new and expansion of old pinewoods.
23 This includes the designation of seed zones for control of the remaining genetic resources.
24 The zoning was based mainly on biochemical similarity among pinewoods but, by

25 definition, neutral molecular markers do not reflect local phenotypic adaptation.
26 Environmental variation within Scotland is substantial and it is not yet clear to what
27 extent this has shaped patterns of adaptive differentiation among Scottish populations.
28 Systematic, rangewide common-environment trials can provide insights into the evolution
29 of the native pinewoods, indicating how environment has influenced phenotypic variation
30 and how variation is maintained. Careful design of such experiments can also provide
31 data on the history and connectivity among populations, by molecular marker analysis.
32 Together, phenotypic and molecular datasets from such trials can provide a robust basis
33 for refining seed transfer guidelines for Scots pine in Scotland and should form the
34 scientific basis for conservation action on this nationally important habitat.

35

36 **Introduction**

37

38 Scots pine (*Pinus sylvestris* L.) is a national icon in Scotland and a foundation species in
39 the Caledonian forest. Currently, 84 woodlands of different sizes of Scots pine woodland
40 in Scotland are recognized as native (Anonymous, 1998). In total these constitute less
41 than 1% of the maximum postglacial range and represent the only recognised UK
42 resource for this habitat, Caledonian pinewood, which receives protection under the EC
43 Habitats directive. These pinewoods represent the north-western extreme of the species'
44 distribution, which is one of the widest of all conifers, extending from northern Finland to
45 Turkey and from western Spain to eastern Siberia (Critchfield and Little, 1965), covering
46 a huge range of environments. In many places Scots pine is also a commercially important
47 timber species, its wood being used for construction, furniture and other products. In
48 Scotland, native trees of commercially desirable form persist in the relatively large
49 populations of e.g. Abernethy, Rothiemurchus and Glen Tanar (Mason et al., 2004).

50 Recognition of the biological, cultural and recreational value of the species to Scotland
51 has resulted in initiation of replanting programmes, and the commercial prospects for
52 native pine are currently being re-evaluated, e.g. in ‘Developing the Scots Pine Resource’
53 project in collaboration with institutes from the Nordic Countries (Macdonald et al.,
54 2008).

55
56 Adaptation to local climatic conditions through genetic differentiation is a widespread
57 feature of forest tree species (Howe et al., 2003; Savolainen et al., 2007). This
58 characteristic is of high importance for strategies focused on conservation of genetic
59 resources, especially under changing environments. The use of maladapted planting stock
60 or the uncontrolled translocation of non-local provenances may lead to losses in
61 productivity or dilution of local gene pools, impacts to which highly reduced or
62 fragmented populations may be especially vulnerable (McKay *et al.*, 2005). Despite the
63 many unique characteristics of Scottish pinewoods, the extent of possible local adaptation
64 has not been studied in detail.. The aims of this paper are to review current knowledge
65 about the evolution of pinewoods in Scotland and to demonstrate how understanding the
66 species’ history and evolution could provide valuable information with which to refine
67 current guidance on the conservation and expansion of the existing pinewood resources.
68 This is particularly timely given the Scottish government’s aim of increasing forest land
69 cover from 17.1 to 25% (*Scottish Forestry Policy 2006*, 2006).

70

71 **Re-colonisation and history of pinewoods in Scotland**

72

73 The last glaciation has strongly influenced the distributions of numerous species in
74 Europe as, during the last glacial maximum 23 000 – 18 000 years ago, ice covered the

75 majority of northern Europe (Svendsen et al., 1999). Pine survived through the ice age in
76 the Iberian, Italian and Balkan peninsulas (Bennett et al., 1991), but macrofossil evidence
77 for refugia have also been found in central parts of Europe (Birks and Willis, 2008; Willis
78 *et al.*, 2000; Willis and van Andel, 2004). Climate modelling suggests that these areas
79 would have been suitable for pine at that time (Cheddadi et al., 2006). Populations from
80 the Iberian and Apennine Peninsulas harbour unique seed-transmitted mitochondrial DNA
81 (mtDNA) variation that is not found elsewhere in Europe (Cheddadi *et al.*, 2006;
82 Pyhäjärvi *et al.*, 2008; Sinclair *et al.*, 1999; Soranzo *et al.*, 2000), and the Iberian
83 pinewoods have also been found to differ from other continental populations for
84 monoterpene and allozyme variation (Prus-Glowacki and Stephan, 1994; Tobolski and
85 Hanover, 1971). These patterns support the view that more northern pine populations
86 originate from refugia located north of the southern peninsulas and south of permafrost.
87
88 According to pollen studies, pine reached Scotland about 8,000 years ago and, appeared
89 first in the Wester Ross area in the northwest, and then shortly afterwards in the
90 Cairngorms (Birks, 1989), the latter presumably having spread northwards through
91 England (Bennett, 1995). Interpreting pollen data in species like pine can be challenging
92 due to its abundance and long dispersal distances, and therefore macrofossil data are
93 needed to verify presence of local populations (Birks, 2003). In fact, fossil stomata from
94 two sites in the Highlands indicate that pine was locally present 1,600-600 years earlier
95 than suggested by pollen data (Froyd, 2005). Contemporary populations from Wester
96 Ross differ from those in the rest of Scotland in their allozyme and monoterpene
97 frequencies, suggesting that the contemporary Scottish population derives from multiple
98 refugia (Forrest, 1980; Forrest, 1982; Kinloch et al., 1986). For example, in contrast to the
99 rest of the populations, the frequency of 3-carene in the northwest is very low (Forrest,

100 1980); biochemically, populations from this area seem more closely related to southern
101 European populations than those from north-central Europe, which are similar to the rest
102 of the Scottish pinewoods (Forrest, 1982). It is possible that the north-western trees
103 originate from refugia near southwest Ireland or western France (Ballantyne and Harris,
104 1994; Bennett, 1995), but this has not yet been verified by analysis of Irish macrofossils
105 or potentially native pinewood remnants. Alternatively natural selection or genetic drift
106 (random loss of genetic diversity due to e.g. sudden decrease in population size) may
107 account for the differences, as these populations are on the edge of the species' range and
108 under strong oceanic influence. The wet, mild climate is markedly different from that in
109 other parts of the range and provides potentially divergent selective pressures involving,
110 for example, pathogen attack, which may have driven biochemical differentiation.

111 Biochemical similarity between northwest Scotland and southern Europe may reflect the
112 effects of adaptation in a similar direction. However, if variation was due to drift, this
113 would imply lack of gene flow between populations in western Scotland and elsewhere. In
114 their mtDNA study, Sinclair et al. (1999) found two molecular variants in Scotland, the
115 less common type being found in the western part of Scotland. Such differentiation
116 further supports the view of colonization from two directions. Similarly, multiple origins
117 might be suggested by the presence of a unique, paternally-inherited chloroplast DNA
118 (cpDNA) microsatellite allele that was found only in the Wester Ross area (Provan et al.,
119 1998). However, this variant could also represent a recent mutation. Had it been an
120 ancestral polymorphism it would have been surprising that the allele was restricted to the
121 area, considering efficient pollen-mediated transmission of cpDNA. Currently, the low
122 number of mtDNA haplotypes detected prevents precise definition of the colonisation
123 routes of pine in Europe (Naydenov *et al.*, 2007; Pyhäjärvi *et al.*, 2008; Sinclair *et al.*,
124 1999), but further evidence for separate evolutionary origins of eastern and western

125 pinewoods in Scotland has recently been found in candidate gene variation (Wachowiak
126 *et al.*, 2010).

127

128 **Current status of native Scottish pinewoods**

129

130 During its history in Scotland, pine has fluctuated in abundance, sometimes very rapidly,
131 due to various factors such as competition from deciduous tree species, decrease of
132 deciduous forests, climate change and human activity (Bennett, 1995). Nowadays, the
133 only natural pinewoods on the British Isles are patchily distributed in Scotland from
134 latitude 55 °N to 57 °N and from longitude 3 °W to 1 °W at altitudes up to 600 metres
135 (Mason *et al.*, 2004). According to the most recent available estimate, the native
136 pinewood area in Scotland covers 18,000 hectares in 84 separate pinewoods varying in
137 size from less than one to over 2,000 ha (Anonymous, 1998); some populations are small
138 and sparse consisting of little more than 100 trees at a density of less than one tree per
139 hectare (e.g. Martin, 1995). A substantial number of the native populations were already
140 identified and described in the influential book ‘The Native Pinewoods of Scotland’ by
141 Steven and Carlisle (1959). Natural pinewood regeneration is often prevented by grazing
142 of domestic livestock or wild deer, muir burning and planting of non-native trees
143 (Anonymous, 1998), and many of the populations have been reduced to very small
144 numbers due to human interference. Also, in the past, trees of poor growth form have
145 often been left in the forests while those considered to be superior from the silvicultural
146 perspective have been felled and extracted for timber (Mason *et al.*, 2004). In such cases,
147 the surviving trees could negatively affect the quality of later generations if they
148 contribute to mating (Ennos *et al.*, 1998; Mason *et al.*, 2004). However, the extent of such
149 practices is not known. In addition, undocumented quantities of trees of continental origin

150 have been introduced to Scotland since the 19th century (Forrest and Fletcher, 1995;
151 Taylor, 1993) which potentially could cause genetic contamination of local populations
152 via pollen flow. The coverage of Scots pine plantations, which are mainly used for timber
153 production, totals 100,000 ha (Mason *et al.*, 2004). However, the extent to which they
154 contribute to the pollen pool in Scotland is not known.

155

156 **Management of pinewoods in Scotland**

157

158 Since the late 1980s protection and expansion of pinewoods has been included in various
159 policies and grant schemes (Mason *et al.*, 2004). For example, the ‘Native Pinewood
160 Grant Scheme’ between 1989 and 2004 aided the regeneration of existing pinewoods and
161 created 48,000 ha of new pinewoods (16% natural regeneration, 84% plantations) while
162 the ‘Native Pinewood Habitat Action Plan’ aimed at increasing the remnant pinewood
163 area by 5,600 ha by 2005 and assisting natural regeneration (McIntosh, 2006). However,
164 there are no recent estimates available on the overall success of these projects or current
165 coverage of (semi-)natural pinewoods. To guide seed transfers, the Scottish pinewoods
166 have been divided into seven seed zones (figure 1) such that when (semi)-natural
167 pinewoods are being expanded, in order to qualify for grant support, planting stock must
168 come from within the same seed zone in an attempt to protect the local “genetic integrity”
169 (Anonymous, 1998). For other planting objectives, such as timber production, the rules
170 are somewhat less restrictive. The seed zones are based largely on monoterpene studies
171 (Forrest, 1980) so that biochemically similar pinewoods are clustered within one zone.

172 [Figure 1]

173

174 The general purpose of seed transfer guidelines in forestry is to prevent planting of
175 maladapted trees and to maximise survival and growth. Ideally, management of any
176 economically important tree species would include transfer restrictions, zoning species on
177 the basis of climate, soil and topography and the results of provenance trials replicated in
178 multiple environments (White et al., 2007). In other words, guidelines would take into
179 account broadly-assessed patterns of local adaptation, with quantified evaluation of the
180 phenotypic effects of seed transfers along environmental gradients. As adaptation is
181 driven by the environment, and the spatial scale of environmental heterogeneity can differ
182 widely among regions, transfer rules are not easily transferable between different
183 countries. Although apparently practical where field data are in short supply, applying
184 single-source data (such as monoterpenes and allozymes which can be considered
185 selectively neutral molecular markers) to devise seed zones is likely, at best, to poorly
186 reflect adaptive patterns (McKay and Latta, 2002; Merilä and Crnokrak, 2001) or, at
187 worst, result in detrimental effects on survival and growth if environmental conditions
188 vary greatly among the origin of seed and the plantation site. Hence, whilst variation at
189 molecular markers can accurately reflect other evolutionary features, such as population
190 structure, demography and mating system (i.e. relative levels of inbreeding and
191 outbreeding), they should be applied in combination with data on environments and
192 adaptively significant traits if seed zoning is to be meaningful.

193

194 **Local adaptation is common in trees**

195

196 Adaptations to local climate conditions have been described in many tree species using
197 provenance trials (see Howe et al., 2003; Savolainen et al., 2007 for reviews) and in Scots
198 pine in other parts of its range. Despite significant phenotypic differentiation, molecular

199 marker variation may show an opposite trend: due to efficient pollen-mediated mixing of
200 pollen pools even distant populations can seem very similar at molecular markers that are
201 not under selection (e.g. Karhu *et al.*, 1996). Phenotypic divergence is mostly driven by
202 environmental variation among sites. In the Northern hemisphere, due to differences in
203 length of the growing season and in the severity of seasonal periods of stress, trees
204 alternate between periods of active growth and dormancy in order to avoid frost damage
205 in the spring or the autumn (Howe *et al.*, 2003), and traits chosen for studies are usually
206 expected to be linked to these environmental factors. Growth patterns of Scots pine have
207 been extensively studied and, for example, timing of growth cessation is thought to be
208 influenced by both photoperiod and accumulated temperature (Koski and Sievänen,
209 1985). In common environment conditions, first-year pine seedlings from colder areas
210 generally set their terminal buds and become frost hardy earlier than the ones from
211 warmer conditions (e.g. Hurme *et al.*, 1997). Also, when seedlings from different parts of
212 Europe were grown under photoperiods typical of 50° latitude, seedlings from northern
213 regions set buds earlier compared to seedlings from more southern locations (Oleksyn *et*
214 *al.*, 1992). The same pattern has also been found in height growth cessation of older trees
215 (Oleksyn *et al.*, 2001; Repo *et al.*, 2000). In Sweden, provenance transfers from north to
216 south resulted in increased survival, but transferred provenances grew less than local ones
217 due to phenological differences (Eriksson *et al.*, 1980; Persson and Ståhl, 1990). On the
218 other hand, northward transfers increased mortality. Commonly, trees from sites
219 experiencing harsher – e.g. drier or colder - conditions grow more slowly than those
220 originating from milder environments, but they are also more tolerant of stress (Howe *et*
221 *al.*, 2003). Phenotypic divergence among populations is generally thought to be due to
222 differentiation at multiple underlying genes driven by diversifying selection (for reviews
223 on the genetic basis of complex trait variation in trees, see González-Martínez *et al.*,

224 2006; Howe *et al.*, 2003; Neale and Ingvarsson, 2008; Savolainen *et al.*, 2007), but so far
225 candidate gene studies in trees have revealed more about past demographic processes than
226 about effects of selection (see Lascoux *et al.*, 2008). However, additional factors can also
227 contribute: in Norway spruce (*Picea abies* (L.) Karst.) it appears that maternal effects, e.g.
228 differences due to environmental conditions during seed development, can greatly
229 influence trait variation (Johnsen *et al.*, 2005; Skrøppa, 1994; Skrøppa *et al.*, 1994), but in
230 Scots pine such effects seem much smaller (Ruotsalainen *et al.*, 1995).

231

232 For maintenance of natural patterns of adaptive variation, the safest option is usually to
233 use local seed material or seeds from an environment that matches conditions at the
234 planting site (Aitken *et al.*, 2008; McKay *et al.*, 2005). Using genotypes from other
235 locations might negatively affect the local population due to outbreeding depression
236 (hybridization among excessively diverged populations) leading to decreased fitness
237 (Frankham *et al.*, 2002). The definition of “local” depends on the species: in Douglas-fir
238 (*Pseudotsuga menziesii* (Mirb.) Franco), genetic differentiation can occur at 100-200
239 metres (Campbell, 1979), while populations of western white pine (*Pinus monticola*
240 Dougl.) seem genetically similar across a wide range covering 10° in both latitude and
241 longitude (Rehfeldt *et al.*, 1984). Relatively short transfers can actually be beneficial for
242 growth and survival: some conifer populations on the northern edge of the species
243 distributions have been found to perform better if transferred southward of their origin
244 (e.g. Savolainen *et al.*, 2007), while in other species a similar response can be seen in
245 transfers further north (Carter, 1996).

246

247 **Is Scots pine locally adapted in Scotland?**

248

249 The current abundance of pinewood in Scotland is only a small fraction of what it used to
250 be, and potentially the exploitation of the resources could have interfered with local
251 adaptation by randomly removing best-adapted trees. . However, the previous molecular
252 marker studies based on monoterpenes (Forrest, 1980; Forrest, 1982) and allozymes
253 (Kinloch *et al.*, 1986) and recent work on nucleotide variation in candidate genes
254 (Wachowiak *et al.*, 2010) show that even in relict populations, levels of molecular
255 variation are similar to those observed in the continuous part of the species' range and, as
256 is usual in the case of long-lived, randomly mating forest trees with effective gene flow by
257 pollen (Hamrick *et al.*, 1992), almost all of the variation was found within populations. In
258 theory, colonization events (such as postglacial migration) are expected to decrease
259 genetic variation through bottlenecks, but the life history characteristics of trees
260 (longevity, multiple age and size classes, overlapping generations and late reproduction)
261 seem to buffer against these effects (Austerlitz *et al.*, 2000). For example, due to their
262 postglacial colonisation history northern Fennoscandian Scots pine populations are much
263 more recently established than those from Central Europe (Willis *et al.*, 1998), but despite
264 their different histories the two parts of the range have very similar levels of nucleotide
265 variation at candidate genes (Pyhäjärvi *et al.*, 2007). Some quantitative traits have been
266 found to have less adaptive variation towards the northern range edges, but this pattern is
267 not seen in all traits, and differences could be caused by varying selection pressure
268 (Notivol *et al.*, 2007). In Scottish populations, low marker divergence among populations
269 suggests that gene flow among sites has, at least historically, been sufficient to
270 homogenise genetic variation across populations (Kinloch *et al.*, 1986). Also, when
271 comparing differentiation at cpDNA markers between Scotland and eight European
272 mainland populations, only around 1.5 % of the variation was found between populations,
273 indicating high levels of gene flow (Provan *et al.*, 1998). Within Scotland, 3.2% of the

274 variation was among populations. Glen Falloch, a relict population consisting of less than
275 100 trees, had the lowest diversity. Despite drastic changes in the abundance of Scots
276 pine in Scotland, it seems that the level of neutral molecular variation remains high, with
277 the majority of this variation being found mainly within populations.

278

279 Despite the relatively small area covered by pinewoods in Scotland, the environmental
280 conditions among them vary tremendously, providing potential for different selection
281 pressures to lead to local adaptation. Furthermore, Scotland's populations form a unique
282 part of the species' range due to its oceanic climate; only in western parts of Norway do
283 Scots pine woodlands occur in similar environments (Øyen et al., 2006). To summarise
284 climatic variation among Scottish native pinewood sites, we extracted data for all 84
285 pinewoods from the gridded (5 × 5 km) long-term average (1961-1990) UK Met Office
286 data. Details on the climate data generation can be found elsewhere (Perry and Hollis,
287 2005). The climate data indicate that some western populations in Scotland experience an
288 annual rainfall of close to 3,000 mm compared to only about 700 mm in the eastern parts.
289 The length of the growing season (the number of days with average temperature above
290 +5°C) varies from about 100 in some eastern pinewoods to 300 days near the west coast.
291 To study whether climatically similar pinewoods were found within each seed zone, we
292 performed a principal component analysis (PCA) to transform the seven variables into
293 two components (figure 2, table 1). The data suggest that different pinewood sites within
294 seed zones do not form climatically uniform clusters, which indicates that climatic
295 variation within one zone can be large. For example, the North West seed zone covers
296 areas with growing season lengths varying from about 130 to almost 300 days (table 2).
297 Because of this extensive within-zone variation and considering for instance the effects of
298 provenance transfers along latitudinal gradients in Sweden (Persson and Ståhl, 1990), it is

299 possible that current guidance results in seedlings being planted at non-optimal sites.
300 However, it should be kept in mind that this data was generated by interpolation from data
301 from weather stations which are not equally distributed across the country, and the
302 precision of models for different variables varies (Perry and Hollis, 2005). In addition to
303 climate, there is also variation in soil types; generally pine prefers freely-draining podzol
304 and ironpan soils with relatively low nutrient levels, but it is also found in brown earths,
305 gleys and peats (Mason et al., 2004). In wet conditions, poor drainage can lead to poor
306 growth and water-logging.

307

308 [Table 1, Table 2, Figure 2]

309

310 Some evidence of local adaptation in the native pinewoods exists, but the data currently
311 available is not extensive. Old provenance experiments set up by the Forestry
312 Commission in Scotland starting in the 1920s show that populations from the mainland of
313 Europe generally perform worse than Scottish material (Lines and Mitchell, 1965). Within
314 Scotland, trees transferred from continental to strongly oceanic areas usually perform
315 worse than local populations, possibly due to pathogen stress (Mason et al., 2004). Perks
316 and McKay (1997) found significant differences in root frost hardiness and growth in
317 seedlings from four provenances; for instance, seedlings from Loch Maree, located in the
318 west close to the Atlantic, had poorer height growth and slower development of frost
319 hardiness than other provenances. The only study where genetic parameters of adaptive
320 variation were estimated was by Perks and Ennos (1999) who also sampled four
321 provenances, each represented by 100 open-pollinated progeny (ten from each of ten
322 mother trees). Seedlings were grown at one site and measured at seven years of age.
323 Significant differentiation among populations was found in diameter, height and bud

324 burst. Adaptive variation was found in all of the measured characters, demonstrating the
325 presence of genetic variation for adaptively important traits, but due to the sample size,
326 estimates on the amount of adaptive variation are not precise. Also, while it was possible
327 to show clear differentiation among populations in the traits considered, geographic
328 coverage was too limited to offer a full picture of patterns of adaptive variation and the
329 study did not attempt to link observed trends to variation in climatic variables.

330

331 Ideally, in Scotland, the seed transfer guidelines for Scots pine should be based on
332 climatic and environmental characterisation of the remnant pinewoods and provenance
333 trials, ideally replicated in different environments. Considering the environmental
334 variation that exists within Scotland, differentiation in traits of adaptive importance such
335 as phenology, stress tolerance, and growth seems likely. For the maintenance of healthy
336 pinewoods in Scotland and to update existing seed transfer guidelines, it is essential to
337 study adaptive variation in a number of traits across the whole Scottish range.

338

339 **Maintenance of variation in pinewoods**

340

341 The current seed zones of Scots pine are meant to protect the “genetic integrity” of local
342 populations. However, the definition of genetic integrity remains unclear. While
343 maternally-inherited mtDNA variation might show differentiation among some of the
344 pinewoods (Sinclair et al., 1998), this does not mean that populations containing the
345 diverged mtDNA lineages are unconnected. In forest trees, the fact that most of the
346 variation measured with neutral molecular markers occurs within populations can be
347 attributed to efficient pollen-mediated gene flow (Hamrick et al., 1992). Pollen flow can
348 effectively mix the gene pools of populations even if they are under diversifying selection.

349 However, although a significant proportion of pollen can originate from sites located even
350 a few dozen kilometres away, the great majority of fertilizing pollen usually comes from
351 trees located within the same stand as the mother tree (e.g. Smouse and Sork, 2004).
352 Nonetheless, such mixing can contribute to the maintenance of variation in adaptive traits
353 (Barton, 1999; Slatkin, 1978) which in turn can facilitate adaptation to changing
354 conditions, as potentially beneficial alleles are introduced to the gene pool of the
355 population. Yeaman and Jarvis (2006) studied effects of environmental heterogeneity on
356 variation in height growth in 142 populations of lodgepole pine (*Pinus contorta* Douglas
357 ex. Loudon) and found that variability among the populations in drought occurrence,
358 annual precipitation and temperature explained 7-20% of the variation in height growth.
359 Due to influx of genetic variation from other populations gene flow can also counteract
360 adaptation, especially in peripheral populations (Garcia-Ramos and Kirkpatrick, 1997). It
361 is unlikely that the native pinewoods represent independently evolving units. Previous
362 studies on Scottish pinewoods show low levels of population differentiation (Kinloch *et*
363 *al.*, 1986; Provan *et al.*, 1998), although the gene flow estimates are indirect and may not
364 reflect current landscape-level processes (Smouse and Sork, 2004; Sork *et al.*, 1999).
365
366 Ongoing climate change is affecting forests all over the world, and changes in
367 temperature, rainfall and frequency of extreme weather events are expected (e.g. IPCC,
368 2007). In Scotland, models predict warmer summers and milder winters, with changes in
369 the distribution of rainfall (Ray, 2008). In the east, summers are predicted to become
370 drier, possibly leading to drought, while winters may become wetter, also a problem if it
371 leads to water-logging and anaerobic conditions in soils. Warmer conditions may help
372 pests and pathogens spread to new areas. For example, the northward spread of the pine
373 processionary moth (*Thaumetopoea pityocampa* Dennis and Schiff) in Italy has been

374 attributed to increasing winter temperatures (Battisti et al., 2005), and since the late
375 1990s, the occurrence of red needle blight, a fungal disease infecting a wide range of
376 *Pinus* species, has increased in the UK with first outbreaks occurring in Scotland in 2002
377 (Brown *et al.*, 2003). Changes in climate can lead to situations where environments are no
378 longer optimal for the populations growing in them. Trees have experienced warming
379 conditions before, following the retreat of continental ice at the end of the ice age (e.g.
380 Davis and Shaw, 2001). In current conditions the problem for trees is likely to be the rate
381 of change which is projected to be faster than that following the latest ice age. After the
382 last glaciation, European trees migrated at average speeds of around 100-700 metres per
383 year, depending on the species (Brewer et al., 2002; Magri et al., 2006). According to
384 Malcolm and Markham (2002), trees will have to be able to migrate at a rate of over 1,000
385 m per year to be able to keep pace with human-induced change. This time, however, trees
386 face environments already occupied by other species.

387

388 For a change in fitness of the population, selection must work on the variation present in
389 the population (Falconer and Mackay, 1996). Genetic variation in phenotypic traits can be
390 assessed by growing seedlings in a common environment in which environmental
391 variation is kept to a minimum (e.g. White *et al.*, 2007). Only variation that can be passed
392 on to the next generation has evolutionary significance; therefore, estimating levels of
393 such variation requires observations based on samples of a known family structure (e.g.
394 open-pollinated half-sib families). In the majority of the forest trees studied, populations
395 generally maintain high levels of adaptive variation (Aitken et al., 2008; Howe et al.,
396 2003; Savolainen et al., 2007), even in range-edge populations under extreme conditions
397 (Notivol *et al.*, 2007; Savolainen *et al.*, 2004). In traits with high levels of such variation,
398 the change in the phenotypic mean in response to new selection pressures can be rapid

399 (Falconer and Mackay, 1996). However, the life history characteristics of trees can slow
400 down rates of adaptation: they are long-lived, have long generation times and, due to
401 phenotypic plasticity, can continue to grow and reproduce even in changed environments
402 (Hamrick, 2004; Mátyás, 1996; Savolainen *et al.*, 2004; Savolainen *et al.*, 2007). For
403 example, Swedish provenance trials suggest that climate-related mortality in Scots pine
404 occurs mainly in the early stages (first 20 years) of a tree's life cycle (Persson and Ståhl,
405 1990). In addition, if grazing pressure prevents natural regeneration, the adaptive variation
406 present in seedlings is lost.

407

408 Due to within-species genetic differentiation adaptive responses may vary among
409 populations from different parts of the range. According to Rehfeldt *et al.* (2002), the
410 immediate response to a warming climate will be positive in Scots pine populations
411 growing in harsh (suboptimal) conditions, e.g. northern parts of Europe, while populations
412 in mild (optimal) environments, e.g. southern Europe, will suffer. Using simulations,
413 Savolainen *et al.* (2004) concluded that while Finnish Scots pine populations have
414 potential to adapt in timing of bud set and frost hardiness, their response will be delayed
415 and will lag behind the moving optimum, partly because of the already established trees
416 growing at the site. Increased mortality could facilitate adaptation by creating open spaces
417 for regeneration (Kuparinen *et al.*, 2010). Specific forest management practices have also
418 been suggested as methods for enhancing adaptation; for instance, seedlings could be
419 transferred according to the predicted climate (St. Clair and Howe, 2007), or the interval
420 between recruitment events could be shortened (Kramer *et al.*, 2008).

421

422 Before specific provenances can be chosen for future climate, data on the effects of tree
423 transfers between variable sites and on variation of adaptively important traits is needed.

424 With such data in hand, models may be developed to test responses to specified variables,
425 although making predictions will remain challenging. Not only is adaptation a complex
426 process involving a number of traits simultaneously, but environmental change may also
427 involve changes in the structure of stands, stress frequency, growth rates and competition
428 (Richardson et al., 2007), and it is impossible to include all possible variables at the same
429 time. Current models have yet to combine genetics and ecology effectively, for example
430 models based on niche concepts often fail to take into account the possibility of
431 adaptation, while genetic models deal inadequately with ecology. There is a pressing
432 need, for climate change mitigation, for the development of new, landscape-scale models
433 that integrate these fields.

434

435 Studies on adaptive variation would also benefit from an understanding of current patterns
436 of genetic connectivity among forest fragments. For example, if only local material is
437 used for planting and gene flow is limited, local genetic “integrity” of small populations
438 will be maintained, but the population might become vulnerable to changing conditions
439 due to insufficient adaptive variation for natural selection to operate on. In the case of
440 isolated populations, variation could be introduced by bringing seedlings from other
441 locations; however, if gene flow occurs naturally and if natural regeneration occurs, such
442 practices might be unnecessary. Due to differences in the sizes of the native pinewoods
443 (from less than one to over 2,000 ha), there might also be variation in the patterns of
444 mating system. In small populations, random drift becomes a powerful force shaping
445 allele frequencies, and along with inbreeding, this can lead to lower fitness as detrimental
446 alleles increase in frequency (Frankham *et al.*, 2002). Like other pines, Scots pine is
447 mainly outcrossing (Muona and Harju, 1989), i.e. matings usually occur between
448 unrelated trees, but self-pollination, the most severe form of inbreeding, is also possible

449 due to the lack of a genetic system preventing self-fertilization (Sarvas, 1962). Normally,
450 selfed embryos are aborted early in their development due to early inbreeding depression.
451 However, in stands with limited numbers of trees, bi-parental inbreeding (mating between
452 relatives) is a potential risk. Despite efficient gene flow, inbreeding might become a
453 significant factor when isolation is extreme. In Scots pine, gene flow and mating system
454 have been studied in e.g. Spanish populations occurring in isolated stands in mountainous
455 regions. Although the proportions of self-pollination were eight times larger (25% vs. 3%)
456 in a population of 36 trees spread across a 15-ha area compared to that of larger
457 populations covering thousands of hectares (Robledo-Arnuncio et al., 2004), the rates
458 were nevertheless low when the degree of isolation of the trees is taken into account. In
459 the small population, 4.3 % of the pollen originated from other populations, the closest
460 one being located 30 km away (Robledo-Arnuncio and Gil, 2005). Kärkkäinen et al.
461 (1996) documented variation in levels of inbreeding depression within larger populations
462 in Finland: outcrossing rates in northern populations were somewhat lower than in the
463 south, but inbreeding depression was weaker in the north, possibly due to selection having
464 already removed detrimental recessive alleles exposed by inbreeding. Understanding the
465 mating system is also beneficial for studies on adaptive variation in phenotype, as
466 departures from the assumed family structure can lead to biased estimates of adaptively
467 significant genetic variation (Namkoong, 1966; Squillace, 1974).

468

469 **Conclusions**

470

471 Due to its economic and biological significance across two continents, Scots pine is one
472 of the most thoroughly studied forest tree species in the world, and its biology has been
473 studied from DNA to the whole ecosystem level. Native remnant pinewoods of Scotland

474 represent a distinct part of the distribution because of their proximity to the Atlantic
475 Ocean, highly variable climate conditions and the 500 km distance to the closest
476 continental populations. Scots pine's current coverage in Scotland is only a fraction of
477 what it used to be, but there are plans to expand old and plant new native woodlands. . If
478 local adaptation has occurred and provided that other management practices (e.g.
479 cultivation and deer management) support successful regeneration, modifications to
480 existing seed transfer guidelines could improve the effectiveness of re-plantation efforts
481 by minimising seedling mortality due to maladapted stock and the consequent ecological,
482 economic and strategic effects. Current transfer rules are based primarily on molecular
483 variation that does not reflect the likely pattern of environmental adaptation across
484 Scotland. To update the existing management guidelines, further research is
485 recommended, with a particular focus on the following:

486

- 487 1. Rangewide progeny trials are needed to characterise general trends of adaptive
488 variation in traits such as phenology, growth, and stress tolerance in relation to the
489 environment. Such data can also reveal whether plantations have diluted local
490 adaptation in native pinewoods.
- 491 2. Effects of provenance transfers along climatic gradients can be obtained by
492 replicating trials at multiple environmentally diverse sites across Scotland. In such
493 a design, the performance of local trees can also be compared to that of trees from
494 more distant sources.
- 495 3. Neutral genetic markers should be used to assess other types of natural processes
496 occurring in populations, such as effects of population fragmentation, mating
497 system variation and current gene flow dynamics.

498 4. Potential role of pollen contamination can be assessed by e.g. observing
499 synchronisation of reproductive events between plantations and nearby native
500 woodlands.

501

502

503 **Funding**

504

505 This work was supported by Scottish Forestry Trust (MJS' PhD studentship), NERC and
506 the EU-funded Network of Excellence EVOLTREE (FP6, contract #016322; WW's
507 mobility grant under IA4 - Human resource exchange).

508

509 **Acknowledgments**

510

511 The authors wish to thank UK Met Office for allowing the use of their climate data, Chris
512 Quine, Editor Gary Kerr and an anonymous reviewer for comments that improved the
513 manuscript.

514

515

516 **References**

517

518 Aitken SN, Yeaman S, Holliday JA, Wang T, Curtis-McLane S. Adaptation, migration or
519 extirpation: climate change outcomes for tree populations. *Evolutionary*
520 *Applications* (2008) 1:95-111.

521 Anonymous. *Caledonian Pinewood Inventory (CD-ROM)* (1998): Forestry Authority
522 Scotland.

523 Austerlitz F, Mariette S, Machon N, Gouyon PH, Godelle B. Effects of colonization
524 processes on genetic diversity: Differences between annual plants and tree species.
525 *Genetics* (2000) 154:1309-1321.

526 Ballantyne CK, Harris C. *The Periglaciation of Great Britain*. (1994) Cambridge:
527 Cambridge University Press.

528 Barton NH. Clines in polygenic traits. *Genetical Research* (1999) 74:223-236.

529 Battisti A, et al. Expansion of geographic range in the pine processionary moth caused by
530 increased winter temperatures. *Ecological Applications* (2005) 15:2084-2096.

531 Bennett KD. *Post-glacial dynamics of pine (Pinus sylvestris) and pinewoods in Scotland*.
532 In: Scottish Natural Heritage--Aldhous JR, ed. (1995) Edinburgh: Forestry
533 Commission, The Royal Society for the Protection of Birds. 23-39.

534 Bennett KD, Tzedakis PC, Willis KJ. Quaternary refugia of North European trees.
535 *Journal of Biogeography* (1991) 18:103-115.

536 Birks HH. The importance of plant macrofossils in the reconstruction of Lateglacial
537 vegetation and climate: examples from Scotland, western Norway, and Minnesota,
538 USA. *Quaternary Science Reviews* (2003) 22:453-473.

539 Birks HJB. Holocene isochrone maps and patterns of tree-spreading in the British Isles.
540 *Journal of Biogeography* (1989) 16:503-540.

541 Birks HJB, Willis KJ. Alpines, trees, and refugia in Europe. *Plant Ecology & Diversity*
542 (2008) 1:147 - 160.

543 Brewer S, Cheddadi R, de Beaulieu JL, Reille M. The spread of deciduous *Quercus*
544 throughout Europe since the last glacial period. *Forest Ecology and Management*
545 (2002) 156:27-48.

546 Brown A, Rose D, Webber J. *Red band needle blight of pine* (2003) Edinburgh, UK:
547 Forest Research.

548 Campbell RK. Geneecology of Douglas-fir in a watershed in the Oregon Cascades.
549 *Ecology* (1979) 60:1036-1050.

550 Carter KK. Provenance tests as indicators of growth response to climate change in 10
551 north temperate tree species. *Canadian Journal of Forest Research-Revue*
552 *Canadienne De Recherche Forestiere* (1996) 26:1089-1095.

553 Cheddadi R, et al. Imprints of glacial refugia in the modern genetic diversity of *Pinus*
554 *sylvestris*. *Global Ecology and Biogeography* (2006) 15:271-282.

555 Critchfield WB, Little E. *Geographic distribution of the pines of the world* (1965): U.S.
556 Department of Agriculture. 97.

557 Davis MB, Shaw RG. Range shifts and adaptive responses to Quaternary climate change.
558 *Science* (2001) 292:673-679.

559 Ennos RA, Worrell R, Malcolm DC. The genetic management of native species in
560 Scotland. *Forestry* (1998) 71:1-23.

561 Eriksson G, Andersson S, Eiche V, Ifver J, Persson A. *Severity index and transfer effects*
562 *on survival and volume production of Pinus sylvestris in Northern Sweden*. In:
563 *Studia Forestalia Suecica* (1980) Uppsala: The Swedish University of Agricultural
564 Sciences, College of Forestry.

565 Falconer DS, Mackay TFC. *Introduction to Quantitative Genetics*. (1996) Third edn.:
566 Pearson Education Limited, Essex.

567 Forrest GI. Genotypic variation among native Scots pine populations in Scotland based on
568 monoterpene analysis. *Forestry* (1980) 53:101-128.

569 Forrest GI. Relationship of some European Scots pine populations to native Scottish
570 woodlands based on monoterpene analyses. *Forestry* (1982) 55:19-37.

571 Forrest GI, Fletcher AM. *Implications of genetic research for native pinewood
572 conservation*. In: Our Pinewood Heritage--Aldhous JR, ed. (1995) Edinburgh:
573 Forestry Commission, The Royal Society for the Protection of Birds.

574 Frankham R, Ballou JD, Briscoe DA. *Introduction to Conservation Genetics*. (2002)
575 Cambridge: Cambridge University Press.

576 Froyd CA. Fossil stomata reveal early pine presence in Scotland: implications for
577 postglacial colonization analyses. *Ecology* (2005) 86:579-586.

578 Garcia-Ramos G, Kirkpatrick M. Genetic models of adaptation and gene flow in
579 peripheral populations. *Evolution* (1997) 51:21-28.

580 González-Martínez SC, Krutovsky KV, Neale DB. Forest-tree population genomics and
581 adaptive evolution. *New Phytologist* (2006) 170:227-238.

582 Hamrick JL. Response of forest trees to global environmental changes. *Forest Ecology
583 and Management* (2004) 197:323-335.

584 Hamrick JL, Godt MJW, Sherman-Broyles SL. Factors influencing levels of genetic
585 diversity in woody plants species. *New Forests* (1992) 6:95-124.

586 Howe GT, Aitken SN, Neale DB, Jermstad KD, Wheeler NC, Chen THH. From genotype
587 to phenotype: unraveling the complexities of cold adaptation in forest trees.
588 *Canadian Journal of Botany-Revue Canadienne De Botanique* (2003) 81:1247-
589 1266.

590 Hurme P, Repo T, Savolainen O, Pääkkonen T. Climatic adaptation of bud set and frost
591 hardiness in Scots pine (*Pinus sylvestris*). *Canadian Journal of Forest Research-*
592 *Revue Canadienne De Recherche Forestiere* (1997) 27:716-723.

593 IPCC. *Climate Change 2007: Impacts, Adaptation and Vulnerability. Contribution of*
594 *Working Group II to the Fourth Assessment Report of the Intergovernmental*
595 *Panel on Climate Change--*Parry ML, Canziani OF, Palutikof JP, van der Linden
596 PJ, Hanson CE, eds. (2007) Cambridge, UK: Cambridge University Press.

597 Johnsen Ø, Dæhlen OG, Østreng G, Skrøppa T. Daylength and temperature during seed
598 production interactively affect adaptive performance of *Picea abies* progenies.
599 *New Phytologist* (2005) 168:589-596.

600 Karhu A, et al. Do molecular markers reflect patterns of differentiation in adaptive traits
601 of conifers? *Theoretical and Applied Genetics* (1996) 93:215-221.

602 Kärkkäinen K, Koski V, Savolainen O. Geographical variation in the inbreeding
603 depression of Scots pine. *Evolution* (1996) 50:111-119.

604 Kinloch BB, Westfall RD, Forrest GI. Caledonian Scots pine - origins and genetic
605 structure. *New Phytologist* (1986) 104:703-729.

606 Koski V, Sievänen R. *Timing of growth cessation in relation to the variations in the*
607 *growing season*. In: *Crop physiology of forest trees--*Tigerstedt P, Puttonen MA,
608 Koski V, eds. (1985) Helsinki, Finland: Helsinki University Press. 167–193.

609 Kramer K, et al. Bridging the gap between ecophysiological and genetic knowledge to
610 assess the adaptive potential of European beech. *Ecological Modelling* (2008)
611 216:333-353.

612 Kuparinen A, Savolainen O, Schurr FM. Increased mortality can promote evolutionary
613 adaptation of forest trees to climate change. *Forest Ecology and Management*
614 (2010) 259:1003-1008.

615 Lascoux M, Pyhäjärvi T, Källman T, Savolainen O. Past demography in forest trees: what
616 can we learn from nuclear DNA sequences that we do not already know? *Plant*
617 *Ecology & Diversity* (2008) 1:209 - 215.

618 Lines R, Mitchell AF. *Results of some older Scots pine provenance experiments*. In:
619 Forestry Commission Report on Forest Research 1964-1965 (1965) London:
620 HMSO 172-194.

621 Macdonald E, Cooper G, Davies I, Freke B. Scots pine timber: current utilisation and
622 future market prospects in Scotland. *Scottish Forestry* (2008) 62:12-21.

623 Magri D, et al. A new scenario for the Quaternary history of European beech populations:
624 palaeobotanical evidence and genetic consequences. *New Phytologist* (2006)
625 171:199-221.

626 Malcolm JR, Markham A, Neilson RP, Garaci M. Estimated migration rates under
627 scenarios of global climate change. *Journal of Biogeography* (2002) 29:835-849.

628 Martin PJ. *First steps in the regeneration of Glen Falloch pinewood*. In: Our Pinewood
629 Heritage--Aldhous JR, ed. (1995) Edinburgh: Scottish Natural Heritage, Forestry
630 Commission, The Royal Society for the Protection of Birds.

631 Mason WL, Hampson A, Edwards C. *Managing the Pinewoods of Scotland*. (2004)
632 Edinburgh: Forestry Commission.

633 Mátyás C. Climatic adaptation of trees: rediscovering provenance tests. *Euphytica* (1996)
634 92:45-54.

635 McIntosh B. Native pinewoods in Scotland: perspectives on policy and management.
636 *Forestry* (2006) 79:303-307.

637 McKay JK, Christian CE, Harrison S, Rice KJ. "How local is local?" - A review of
638 practical and conceptual issues in the genetics of restoration. *Restoration Ecology*
639 (2005) 13:432-440.

640 McKay JK, Latta RG. Adaptive population divergence: markers, QTL and traits. *Trends*
641 *in Ecology & Evolution* (2002) 17:285-291.

642 Merilä J, Crnokrak P. Comparison of genetic differentiation at marker loci and
643 quantitative traits. *Journal of Evolutionary Biology* (2001) 14:892-903.

644 Muona O, Harju A. Effective population sizes, genetic variability, and mating system in
645 natural stands and seed orchards of *Pinus sylvestris*. *Silvae Genetica* (1989)
646 38:221-228.

647 Namkoong G. Inbreeding effects on estimation of genetic additive variance. *Forest*
648 *Science* (1966) 12:8-13.

649 Naydenov K, Senneville S, Beaulieu J, Tremblay F, Bousquet J. Glacial vicariance in
650 Eurasia: mitochondrial DNA evidence from Scots pine for a complex heritage
651 involving genetically distinct refugia at mid-northern latitudes and in Asia Minor.
652 *BMC Evolutionary Biology* (2007) 7.

653 Neale DB, Ingvarsson PK. Population, quantitative and comparative genomics of
654 adaptation in forest trees. *Current Opinion in Plant Biology* (2008) 11:149-155.

655 Notivol E, García-Gil MR, Alía R, Savolainen O. Genetic variation of growth rhythm
656 traits in the limits of a latitudinal cline in Scots pine. *Canadian Journal of Forest*
657 *Research-Revue Canadienne De Recherche Forestiere* (2007) 37:540-551.

658 Oleksyn J, Reich PB, Tjoelker MG, Chalupka W. Biogeographic differences in shoot
659 elongation pattern among European Scots pine populations. *Forest Ecology and*
660 *Management* (2001) 148:207-220.

661 Oleksyn J, Tjoelker MG, Reich PB. Growth and biomass partitioning of populations of
662 European *Pinus sylvestris* L. under simulated 50° and 60° N daylengths: evidence
663 for photoperiodic ecotypes. *New Phytologist* (1992) 120:561-574.

664 Øyen B-H, Blom HH, Gjerde I, Myking T, Saetersdal M, Thunes KH. Ecology, history
665 and silviculture of Scots pine (*Pinus sylvestris* L.) in western Norway - a literature
666 review. *Forestry* (2006) 79:319-329.

667 Perks MP, Ennos RA. Analysis of genetic variation for quantitative characters between
668 and within four native populations of Scots pine (*Pinus sylvestris*). *Botanical*
669 *Journal of Scotland* (1999) 51:103-110.

670 Perks MP, McKay HM. Morphological and physiological differences in Scots pine
671 seedlings of six seed origins. *Forestry* (1997) 70:223-232.

672 Perry M, Hollis D. The development of a new set of long-term climate averages for the
673 UK. *International Journal of Climatology* (2005) 25:1023-1039.

674 Persson B, Ståhl EG. Survival and yield of *Pinus sylvestris* L. as related to provenance
675 transfer and spacing at high altitudes in northern Sweden. *Scandinavian Journal of*
676 *Forest Research* (1990) 5:381-395.

677 Provan J, et al. Gene-pool variation in Caledonian and European Scots pine (*Pinus*
678 *sylvestris* L.) revealed by chloroplast simple-sequence repeats. *Proceedings of the*
679 *Royal Society of London Series B-Biological Sciences* (1998) 265:1697-1705.

680 Prus-Glowacki W, Stephan BR. Genetic variation of *Pinus sylvestris* from Spain in
681 relation to other European populations. *Silvae Genetica* (1994) 43:7-14.

682 Pyhäjärvi T, Garcia-Gil MR, Knürr T, Mikkonen M, Wachowiak W, Savolainen O.
683 Demographic History Has Influenced Nucleotide Diversity in European *Pinus*
684 *sylvestris* Populations. *Genetics* (2007) 177:1713-1724.

685 Pyhäjärvi T, Salmela MJ, Savolainen O. Colonization routes of *Pinus sylvestris* inferred
686 from distribution of mitochondrial DNA variation. *Tree Genetics & Genomes*
687 (2008) 4:247-254.

688 Ray D. *Impacts of climate change on forestry in Scotland – a synopsis of spatial*
689 *modelling research* (2008): Forestry Commission Scotland.

690 Rehfeldt GE, Hoff RJ, Steinhoff RJ. Geographic patterns of genetic variation in *Pinus*
691 *monticola*. *Botanical Gazette* (1984) 145:229-239.

692 Rehfeldt GE, Tchebakova NM, Parfenova YI, Wykoff WR, Kuzmina NA, Milyutin LI.
693 Intraspecific responses to climate in *Pinus sylvestris*. *Global Change Biology*
694 (2002) 8:912-929.

695 Repo T, Zhang G, Ryyppö A, Rikala R, Vuorinen M. The relation between growth
696 cessation and frost hardening in Scots pines of different origins. *Trees - Structure*
697 *and Function* (2000) 14:456-464.

698 Richardson DM, et al. Human impacts in pine forests: past, present, and future. *Annual*
699 *Review of Ecology, Evolution, and Systematics* (2007) 38:275-297.

700 Robledo-Arnuncio JJ, Alía R, Gil L. Increased selfing and correlated paternity in a small
701 population of a predominantly outcrossing conifer, *Pinus sylvestris*. *Molecular*
702 *Ecology* (2004) 13:2567-2577.

703 Robledo-Arnuncio JJ, Gil L. Patterns of pollen dispersal in a small population of *Pinus*
704 *sylvestris* L. revealed by total-exclusion paternity analysis. *Heredity* (2005) 94:13-
705 22.

706 Ruotsalainen S, Nikkanen T, Haapanen M. Effect of seed-maturing conditions on the
707 growth and hardiness of one-year old *Pinus sylvestris* seedlings. *Forest Genetics*
708 (1995) 2:189-198.

709 Sarvas R. Investigations on the flowering and seed crop of *Pinus sylvestris*.
710 *Communicationes Instituti Forestalis Fenniae* (1962) 53:1-198.

711 Savolainen O, Bokma F, García-Gil R, Komulainen P, Repo T. Genetic variation in
712 cessation of growth and frost hardiness and consequences for adaptation of *Pinus*
713 *sylvestris* to climatic changes. *Forest Ecology and Management* (2004) 197:79-89.

714 Savolainen O, Pyhäjärvi T, Knürr T. Gene flow and local adaptation in trees. *Annual*
715 *Review of Ecology, Evolution, and Systematics* (2007) 38:595-619.

716 *Scottish Forestry Policy 2006*. (2006): Forestry Commission Scotland, Edinburgh.

717 Sinclair WT, Morman JD, Ennos RA. Multiple origins for Scots pine (*Pinus sylvestris* L.)
718 in Scotland: evidence from mitochondrial DNA variation. *Heredity* (1998) 80:233-
719 240.

720 Sinclair WT, Morman JD, Ennos RA. The postglacial history of Scots pine (*Pinus*
721 *sylvestris* L.) in western Europe: evidence from mitochondrial DNA variation.
722 *Molecular Ecology* (1999) 8:83-88.

723 Skrøppa T. Growth rhythm and hardiness of *Picea abies* progenies of high-altitude
724 parents from seed produced at low elevations. *Silvae Genetica* (1994) 43:95-100.

725 Skrøppa T, Nikkanen T, Ruotsalainen S, Johnsen Ø. Effects of sexual reproduction at
726 different latitudes on performance of the progeny of *Picea abies*. *Silvae Genetica*
727 (1994) 43:298-304.

728 Slatkin M. Spatial patterns in distributions of polygenic characters. *Journal of Theoretical*
729 *Biology* (1978) 70:213-228.

730 Smouse PE, Sork VL. Measuring pollen flow in forest trees: an exposition of alternative
731 approaches. *Forest Ecology and Management* (2004) 197:21-38.

732 Soranzo N, Alía R, Provan J, Powell W. Patterns of variation at a mitochondrial
733 sequence-tagged-site locus provides new insights into the postglacial history of
734 European *Pinus sylvestris* populations. *Molecular Ecology* (2000) 9:1205-1211.

735 Sork VL, Nason J, Campbell DR, Fernandez JF. Landscape approaches to historical and
736 contemporary gene flow in plants. *Trends in Ecology & Evolution* (1999) 14:219-
737 224.

738 Squillace AE. Average genetic correlations among offspring from open-pollinated forest
739 trees. *Silvae Genetica* (1974) 23:149-156.

740 St. Clair JB, Howe GT. Genetic maladaptation of coastal Douglas-fir seedlings to future
741 climates. *Global Change Biology* (2007) 13:1441-1454.

742 Steven HM, Carlisle A. *The Native Pinewoods of Scotland*. (1959) Edinburgh: Oliver and
743 Boyd.

744 Svendsen JJ, et al. Maximum extent of the Eurasian ice sheets in the Barents and Kara Sea
745 region during the Weichselian. *Boreas* (1999) 28:234-242.

746 Taylor C. Report on the activities of the native pinewood managers. *Scottish Forestry*
747 (1993) 48:102-109.

748 Tobolski JJ, Hanover JW. Genetic variation in the monoterpenes of Scotch pine. *Forest*
749 *Science* (1971) 17:293-299.

750 Wachowiak W, Salmela MJ, Ennos RA, Iason G, Cavers S. High genetic diversity at the
751 extreme range edge: nucleotide variation at nuclear loci in Scots pine (*Pinus*
752 *sylvestris* L.) in Scotland *Heredity* (DOI: 10.1038/hdy.2010.118) (2010).

753 White TL, Adams WT, Neale DB. *Forest Genetics*. (2007) Wallingford, UK: CABI
754 Publishing.

755 Willis KJ, Bennett KD, Birks HJB. *The late Quaternary dynamics of pines in Europe*. In:
756 Ecology and Biogeography of *Pinus*--Richardson DM, ed. (1998) Cambridge, UK:
757 Cambridge University Press. 107-121.

758 Willis KJ, Rudner E, Sumegi P. The full-glacial forests of central and southeastern
759 Europe. *Quaternary Research* (2000) 53:203-213.

760 Willis KJ, van Andel TH. Trees or no trees? The environments of central and eastern
761 Europe during the Last Glaciation. *Quaternary Science Reviews* (2004) 23:2369-
762 2387.

763 Yeaman S, Jarvis A. Regional heterogeneity and gene flow maintain variance in a
764 quantitative trait within populations of lodgepole pine. *Proceedings of the Royal*
765 *Society B-Biological Sciences* (2006) 273:1587-1593.

766
767
768

769 Table 1 List of climatic variables used in the principal component (PC) analysis. Values
 770 in the table are correlation coefficients that vary between -1 (strong negative correlation)
 771 and 1 (strong positive correlation); the further the coefficient is from zero, the stronger the
 772 association between the variable and the PC. PC1 is the main component, explaining 69%
 773 of the variation.

Variable	PC1	PC2
Length of the growing season	-0.45	-0.12
February mean temperature	-0.45	-0.12
July mean temperature	-0.35	-0.47
Annual extreme temperature range	0.10	-0.72
Air frost days per year	0.44	-0.03
Ground frost days per year	0.43	-0.13
Annual precipitation	-0.30	0.46
Percentage of variation	69.20	23.99

774

775

776 Table 2 Range of climatic variation in four variables within each seed zone according to
777 the UK Met Office long-term average data (Perry and Hollis, 2005). Seed zones: EC=East
778 Central, N=North, NC=North Central, NE=North East, NW=North West, SC=South
779 Central, SW=South West. Climatic variables: LGS=length of the growing season,
780 FMT=February mean temperature, JMT=July mean temperature, AP=annual
781 precipitation.

782

Seed zone	LGS (days)		FMT (°C)		JMT (°C)		AP (mm)	
	min	max	min	max	min	max	min	max
EC	154	216	-0.8	1.0	10.6	13	743	1223
N	162	251	-0.5	2.4	10	13.9	1215	1778
NC	208	299	0.6	4.0	11.1	14.4	1346	2900
NE	108	234	-2.0	1.9	9.4	13.7	785	1343
NW	134	295	-0.9	4.0	8.5	14	1912	2790
SC	219	238	0.8	1.8	11.9	13.4	1159	2904
SW	179	297	0.0	3.9	9.7	14.1	1563	2934

783

784

785

786

787 Figure 1 Map of the current Scots pine seed zones in Scotland.

788

789 Figure 2 Plot of the first two principal components, which account for 69 and 24% of total
790 variation, respectively, of climatic variation among 84 native pinewood sites. The seven
791 variables used are shown in table 1. Current seed zones are represented by different
792 symbols, and the closer the populations are in the graph, the more similar they are
793 climatically. PC1 represents a gradient in annual rainfall and temperature: populations
794 with more negative values are generally located in the west (high rainfall, mild climate);
795 positive values represent more eastern pinewoods with less rainfall and colder winters.

796

797 *This is a pre-copy-editing, author-produced PDF of an article accepted for publication in*
798 *Forestry following peer-review. The definitive publisher-authenticated version Salmela,*
799 *Matti J.; Cavers, Stephen; Wachowiak, Witold; Cottrell, Joan E.; Iason, Glenn R.; Ennos,*
800 *Richard A. 2010 Understanding the evolution of native pinewoods in Scotland will*
801 *benefit their future management and conservation. Forestry, 83. 535-545 is available*
802 *online at <http://forestry.oxfordjournals.org/content/83/5/535.abstract>*

803

804

805

806

807

808

809

810

811

812

813

814

815

816