Genetic variation within and among populations of an arctic/alpine sweat bee (Hymenoptera: Halictidae)

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Abstract—We present the results of electrophoretic analyses of allozymes for eight population samples of the arctic/alpine sweat bee, *Lasioglossum* (*Evylaeus*) *boreale* Svensson, Sakagami and Ebmer. This holarctic species is found at high latitudes and at increasingly high elevations in the mountains of western North America as far south as southern Arizona. Our samples encompass a large proportion of the species' range; three samples are from Arizona, one from Utah, two from northern Canada, one from Mount Washington (the highest point in the eastern United States), and one from Sweden. Most samples had high levels of genetic variation compared with other bees, but the one from Sweden had low heterozygosity, suggesting that this location may have been comparatively recently colonized. The three northern North American samples were genetically similar despite the large geographic distances separating the localities (average >3000 km). In contrast, the southern United States samples were (with the exception of one pairwise estimate) genetically divergent despite the small geographic distances separating them (average <500 km). These results are consistent with earlier divergence among the southern populations, which are currently separated by regions of low elevation and inhospitable desert, than among the more northern ones. Although the data are not conclusive, they are suggestive of northward dispersal from refugia south of the ice sheets since the last glaciation.

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Résumé—Nous présentons ici les résulats d'analyses par électrophorèse des allozymes de huit populations de l'halicte arctique/alpin, *Lasioglossum* (*Evylaeus*) *boreale* Svensson, Sakagami et Ebmer. Cette espèce holarctique habite les latitudes élevées et on la retrouve de plus en plus en altitude dans les montagnes de l'ouest nord-américain jusque dans le sud de l'Arizona. Nos échantillons, trois d'Arizona, un d'Utah, deux du nord du Canada, un du Mont Washington (le point le plus élevé de l'est des États-Unis) et un de Suède, recouvrent une importante proportion de la répartition de l'espèce. Dans la plupart des échantillons, il y a une grande variation génétique par comparaison à d'autres abeilles, mais, dans l'échantillon de Suède, l'hétérozygotie est faible, ce qui semble indiquer que ce site n'a été colonisé que relativement récemment. Les trois échantillons provenant du nord de l'Amérique du Nord sont génétiquement semblables malgré les distances géographiques importantes entre les localités (en moyenne, plus de 3000 km). En revanche, il y a une

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divergence génétique entre les échantillons du sud des États-Unis (à l'exception d'un appariement) malgré les faibles distances géographiques entre les localités (en moyenne moins de 500 km). Ces résultats s'accordent avec l'hypothèse d'une divergence plus précoce entre les populations du sud qui sont séparées par des zones de faible altitude et par des déserts inhospitaliers qu'entre les populations du nord. Bien que ces résultats ne soient pas totalement concluants, ils indiquent que la dispersion s'est probablement faite vers le nord à partir de refuges situés au sud des glaces depuis la dernière glaciation.

[Traduit par la Rédaction]

Introduction

The growth and decay of large ice sheets in the northern hemisphere took place on numerous occasions since the origin of an arctic ice cap 2.4 million years ago (Hewitt 1996, 2000). The most recent ice-sheet expansion, referred to as the Wisconsin glaciation in North America, began approximately 25 000 years ago and lasted until about 10 000 years ago. When these ice sheets were at their largest, approximately 22 000 – 19 000 years ago (Yokoyama *et al.* 2000), almost all of Canada was covered by as much as 3500 m of ice. The extent of ice was much less in Europe, but in North America it extended as far south as 40°N (Hewitt 2000). The current distribution of the Canadian biota, and much of that throughout North America, must reflect dispersal from glacial refugia since the last glacial maximum.

Genetic studies of many temperate North American taxa have demonstrated patterns consistent with glacially induced vicariance events and post-glacial dispersal patterns. Examples include giant salamanders (Routman *et al.* 1994), gopher tortoises (Lamb *et al.* 1989), chickadees (Gill *et al.* 1993), song sparrows (Zink and Dittmann 1993), grasshopper mice (Riddle and Honeycutt 1990), woodrats (Hayes and Harrison 1992), and squirrels (Lamb *et al.* 1997). Species adapted to very short summer conditions and with arctic/alpine or subarctic/alpine distributions are of particular interest. This is because their current geographic ranges include northern populations in lowland areas that were previously ice covered and relict montane populations to the south, some of which would have been close to the southern margin of the ice sheets during glacial maxima. Alpine populations of arctic/alpine species may not have had as far to migrate following the end of the ice age if they passed this period in lowland localities near the mountains of western or eastern North America. These populations could simply move uphill to increasingly higher altitudes as the climate warmed. Conversely, the arctic populations of these species must have migrated northwards if they passed the most recent ice age south of the ice sheets (for diagrammatic representations of these scenarios see Hewitt 1996). Alternatively, they may have spread outwards from more northerly refugia of which the Beringian refugium is the best known. Beringia was an unglaciated region that extended from northeastern Siberia across Alaska to the Richardson Mountains in northern Yukon (Matthews 1979; Péwé 1983). Beringian refugia are not the only possible sources for cold-climate-adapted species to spread following retreat of the ice sheets. Other possibilities include Haida Gwai and Labrador coastal refugia, in which case dispersal would have been west to east or east to west, respectively (Matthews 1979; Clarke *et al.* 2001).

Lasioglossum (*Evylaeus*) *boreale* Svensson, Sakagami and Ebmer (Hymenoptera: Halictidae) is a ground-nesting bee with an arctic–subarctic/alpine distribution. It was originally described from specimens from high altitude on the Japanese Island of Hokkaido and from lowland elevations at high latitude in northern Sweden (Svensson *et al.* 1977). It has since been reported from arctic Canada (Sakagami and Toda 1986). We have discovered that this species is also found across Canada from the Northwest

Territories to Newfoundland. More interestingly, it also occurs at high altitude at least as far south as the Graham Mountains in southeastern Arizona in the west and on Mount Washington, New Hampshire, in the east. It is possible that this species was described from North America prior to the work of Svensson *et al.* (1977). Indeed, superficial comparison suggests that the type specimen of *Lasioglossum* (*Evylaeus*) *dasiphorae* (Cockerell), collected from high altitude in Colorado and described in 1904, may be conspecific with the species we are referring to here; however, pending detailed morphological revisions of this taxonomically difficult group and further genetic sampling to assess species-level status of various populations (Packer and Taylor 1997), we will refer to the taxon studied herein as *L. boreale*.

With a distribution that encompasses almost the entire area affected directly by ice-sheet expansion, extralimital areas to the south, and the Beringian refugium, this species is of biogeographic interest. In this paper we present the results of allozyme analyses of eight populations of *L. boreale* from most of its North American range. We used these data to compare levels of genetic variability among populations and to investigate whether there are any clear patterns that permit discrimination among alternative scenarios concerning the post-glacial dispersal of this species. In particular, we investigated whether the patterns of allozyme variation are consistent with a southern distribution during glacial times with a resulting northward movement of some populations or with it having had a Beringian glacial refugium with subsequent expansion to the south and east.

We used two methods to assess these hypotheses. First, gene diversity may be expected to be higher in populations where a species has persisted for a considerable length of time than in areas to which it has recently migrated (Hewitt 1996). This would be true if the migration involved considerable distances over many generations, as would be the case for northern populations if they had followed the retreating ice sheets. This pattern is expected because gradual dispersal over large distances by small organisms with comparatively low dispersal distances will result in small effective population sizes, especially along the advancing front. Thus, more recently colonized areas may be expected to have lower levels of genetic diversity (Hewitt 1996).

Secondly, we looked at interpopulation levels of genetic divergence. If all North American populations are derived from a single source since the last ice age, they should all be genetically similar because of the short period of time that has elapsed since they colonized the continent. Conversely, if recolonization was from two or more refugia we may expect to see a more disjunct population genetic structure. It is also possible that montane populations in the southwest of the United States could be genetically more distinctive both from one another and from the arctic/subarctic populations. This could be the case for two reasons. First, the southern populations may have dispersed uphill as soon as the ice started retreating, and thus have become separated earlier than the more northern ones. Second, they may have persisted south of the ice over one or more ice ages perhaps with different populations isolated in different mountain ranges during different interglacials. In either scenario (or in a combination of the two) the southernmost populations would have become differentiated the earliest, whereas gene flow among subarctic populations would have continued until more recently and may still be in operation.

Lastly, we assessed patterns of isolation by distance among the samples. Isolation by distance is detected by plotting the mean number of migrants (N_m) between populations per generation against the geographic distance between them on log–log plots (Slatkin 1993). The N_m values obtained from F statistics are an indication of historical levels of gene flow (Packer and Owen 2001). Residual homogeneity in allele frequencies among samples may lead to high estimates of gene flow even when contemporary population processes and geographic distribution render gene flow impossible. Here, we used this method more as a means of graphical representation of resulting genetic differentiation rather than as suggesting that gene flow is occurring among populations now (for detailed criticisms of the use of N_m to represent actual gene flow see Whitlock and McCauley 1999). Because of the predicted complexity in the biogeographic history of this species, we did not expect the negative relationship that may be found when populations are at equilibrium (Slatkin 1993; Peterson and Denno 1998). Nonetheless, inspection of the resulting plot can reveal whether or not geographically different subsets of the data exhibit different patterns of genetic differentiation with distance (Slatkin 1993).

Methods

Samples

Lasioglossum boreale was collected from a range of localities in North America and one site in Sweden (Table 1). The three sites in Arizona are from two mountain ranges. The Graham Mountains (also known as the Pinaleno Mountains) are one of a series of isolated ranges in southeastern Arizona (for a map of these ranges see Masta 2000). They are separated by desert lowlands of an elevation of approximately 500 m. Greens Peak and Bill Williams mountains are part of a more continuous high-altitude (>1800 m) region in northern Arizona and represent the southernmost extent of the Rocky Mountains. This region continues northwards and encompasses the Uinta Mountain site in Utah. With the exception of Mount Washington, the remaining sites are all of low elevation (Table 1).

Bees were collected from flowers, and for some northern localities, from roads and other structures where they were basking. Bees were stored in Eppendorf tubes® on ice in the field and identified while cold using the morphological characteristics outlined by Svensson *et al.* (1977): the weak propodeal carina in females and the unique retrorse lobe of the gonostylus in males readily separate *L. boreale* from sympatric relatives. Bees were killed by storage in liquid nitrogen and were transported to the laboratory where they were stored at –80°C.

Genetic analysis

Each individual was divided into two pieces: metasoma and head plus mesosoma, so as to maximize the number of allozyme loci that could be stained for these small (approximately 7 mm long) bees. Horizontal starch gel electrophoresis was used to resolve 25 loci using methods that have been described elsewhere (Packer and Owen 1989, 1990).

The computer program BIOSYS (Swofford and Selander 1989) was used to estimate Nei's (1978) unbiased expected heterozygosity. Observed heterozygosity is not presented here because many of our samples were hemizygous males. Mean number of alleles per locus and the proportion of loci that were polymorphic are not reported because they are heavily influenced by sample size, which varied substantially.

Two methods were used to investigate levels of genetic differentiation among samples. First, genetic distance and identity values were calculated using Nei's (1978) unbiased estimators. Second, N_m values were calculated among pairs of populations using the program MONTY (Slatkin 1993). This program uses an estimate of F_{ST}, which is not influenced by population size (Weir and Cockerham 1984), and calculates N_m from this using the equation $F_{ST} = 1/(4N_m + 1)$.

Table 1. Localities where samples of *Lasioglossum* (*Evylaeus*) *boreale* were obtained.

Locality	Abbreviation	Latitude (N)	Longitude (W)	Altitude (m)	Date
Shannon Campground, Mt. Graham, Graham Mountains, Arizona	GM	32°	109°	2740	August 1992
Lookout tower, Green's Peak, White Mountains, Arizona	GP	34°	109°	3047	August 1992
Lookout tower, Bill Williams Mountain, Arizona	BW	35°	112°	2786	August 1992
Lily Lake Campground, Uinta Mountains, Utah	UM	40°	111°	3047	August 1992
Alpine garden, Mt. Washington, New Hampshire	MW	44°	71°	1917	June and August 1992
McGill Subarctic Research Station, Schefferville, Quebec	SC	54°	66°	500	June 1993
Inuvik Research Station, Inuvik, North West Territories	IN	68°	133°	30	August 1991
Lake Torneträsk, Abisko, Sweden	AB	68°	18°	385	August 1993

The Mann Whitney U test (Sokal and Rohlf 1995) was used to investigate differences in heterozygosity among some groups of samples and we present an unweighted pair group mathematical average (UPGMA) phenogram (Sokal and Sneath 1973) for a graphical representation of the genetic differences among populations. The latter should not be taken as indicating the evolutionary relationships among the samples.

Results

Twenty-five loci were scored for all populations (Table 2). Twelve of these were variable within or among populations (Table 3).

The Bill Williams Mountain sample from Arizona was monomorphic at all loci, perhaps because of its small sample size ($n \leq 10$ haploid genomes). The larger sample from Abisko, Sweden, was also monomorphic at all loci except for Pep_{gl} and had a low heterozygosity (H_{exp}) estimate of 0.006 (Table 3). The remaining samples all had H_{exp} values between 0.035 and 0.057, all somewhat higher than the average for 10 samples of related species, which had a mean of 0.029 and ranged from 0.008 to 0.06 (Packer and Owen 1992). For the North American samples, there appears to be no clear pattern in the genetic variability data in terms of the biogeographic hypotheses we were testing. The H_{exp} values between the three northern North American samples (Inuvik, Schefferville, and Mount Washington) and the three from Arizona and Utah (Bill Williams Mountain sample omitted from the analysis because of small sample size) did not differ ($U = 5$, $P > 0.2$). The marked lack of variation in the Swedish population, despite a reasonable sample size, strongly suggests recent invasion of this area.

Some loci exhibited fixed differences among the samples as follows: the locus *Gda* showed no within population variation, but there were three electromorphs, one fixed for the Graham Mountains sample, a second fixed in the other two Arizona samples, and the third fixed in all other samples. The Swedish sample was fixed for a fast electromorph at the locus *Acy*, which was not found in any of the other samples. Variation at Pep_{gl} was complex in that the Swedish sample was almost fixed for a unique allele and the Graham Mountains sample was fixed for an allele that was found elsewhere only rarely in two northern samples (Table 3).

Table 2. List of allozyme loci scored.

Enzyme	Symbol	Enzyme nomenclature number	No. of loci
β-*N*-Acetylhexosaminidase	*Aha*	3.2.1.52	1
Adenylate kinase	*Ak*	2.7.4.3	1
Aldehyde dehydrogenase (NAD$^+$)	*Alddh*	1.2.1.3	1
Aminoacylase	*Acy*	3.5.1.14	1
Arginine kinase	*Ark*	2.7.3.3	1
Diaphorase (NADPH)	*Diap*	1.6.99.*	1
Diaphorase (NAD)	*Dia*	1.8.1.*	1
Fumarate hydratase	*Fum*	4.2.1.2	1
Glyceraldehyde-3-phosphate dehydrogenase	*Gapd*	1.2.1.12	1
Glycerol-3-phosphate dehydrogenase (NAD$^+$)	*G3pd*	1.1.1.8	2
Glucose-6-phosphate isomerase	*Gpi*	5.3.1.9	1
Guanine deaminase	*Gda*	3.5.4.3	1
Hydroxyacylglutathione hydrolase	*Hagh*	3.1.2.6	1
D-2-Hydroxy-acid dehydrogenase	*Had*	1.1.99.6	1
Hexokinase	*Hk*	2.7.1.1	1
Isocitrate dehydrogenase (NADP$^+$)	*Idh*	1.1.1.42	1
Malate dehydrogenase	*Mdh*	1.1.1.37	2
Malate dehydrogenase (NADP$^+$)	*Me*	1.1.1.40	1
Peptidase (glycine–leucine)	Pep_{gl}	3.4.11–13	1
Phosphogluconate dehydrogenase	*6Pgd*	1.1.1.43	1
Phosphoglucomutase	*Pgm*	5.4.2.2	1
Pyruvate kinase	*Pk*	2.7.1.40	1
Triose-phosphate isomerase	*Tpi*	5.3.1.1	1

Genetic-distance data (Table 4) showed little differentiation among the three northern populations but showed considerable differentiation of the Graham Mountains bees from the other two samples from Arizona and all other samples (Fig. 1). The Uinta Mountain sample showed intermediate levels of differentiation from both the Arizona and more northern bees but was somewhat more similar to the latter and clustered with them in the UPGMA analysis (Fig. 1). The Swedish sample was also distinctive (Table 4), clustered with the Bill Williams and Green's Peak samples from Arizona in the distance tree (Fig. 1) but was individually most similar to the Uinta Mountains sample (Table 4).

The isolation by distance plot (Fig. 2) showed that the populations across the range of this species did not demonstrate isolation by distance. There were several clusters of points. To the top right are three points where both gene flow and geographic distances are high. These were the three pairwise comparisons among the northern North American populations: Mount Washington, Schefferville, and Inuvik. The only other high gene flow estimate was between Bill Williams Mountain and Green's Peak, the two localities with the second smallest interlocality distance (Table 4) included in our study. The intervening land is also of moderately high elevation (>1800 m). Low levels of gene flow are found for the other pairwise estimates from Arizona: Graham Mountains and both Bill Williams Mountain and Green's Peak, these two points are closest to the lower left-hand corner of the plot. Similarly low levels of gene flow are found between the three samples from Arizona and the one from Utah despite the comparatively small geographic distances separating these localities. These are just to the

TABLE 3. Allele frequencies, sample sizes and expected heterozygosities for samples of *Lasioglossum* (*Evylaeus*) *boreale*.

	Sampling locality*							
Locus	GM	GP	BW	UM	MW	SC	IN	AB
Acy								
A	0.000	0.118	0.000	0.000	0.000	0.000	0.000	0.000
B	1.000	0.882	1.000	1.000	1.000	1.000	1.000	0.000
C	0.000	0.000	0.000	0.000	0.000	0.000	0.000	1.000
n	26	17	10	22	23	26	34	15
Aha								
A	0.091	0.167	0.000	0.000	0.000	0.000	0.087	0.000
B	0.818	0.833	1.000	1.000	1.000	1.000	0.913	1.000
C	0.091	0.000	0.000	0.000	0.000	0.000	0.000	0.000
n	11	6	4	14	19	28	23	26
Alddh								
A	0.913	1.000	1.000	0.889	1.000	1.000	1.000	1.000
B	0.087	0.000	0.000	0.111	0.000	0.000	0.000	0.000
n	23	10	4	18	23	14	30	26
Gda								
A	0.000	1.000	1.000	0.000	0.000	0.000	0.000	0.000
B	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
C	0.000	0.000	0.000	1.000	1.000	1.000	1.000	1.000
n	37	8	1	34	36	12	46	27
Gpd-2								
A	0.975	1.000	1.000	1.000	0.442	0.292	0.346	1.000
B	0.025	0.000	0.000	0.000	0.558	0.708	0.654	0.000
n	40	11	3	33	43	24	52	22
Gpi								
A	1.000	1.000	1.000	1.000	1.000	0.939	1.000	1.000
B	0.000	0.000	0.000	0.000	0.000	0.061	0.000	0.000
n	23	12	7	28	41	33	50	26
Hagh								
A	0.000	0.000	0.000	0.045	0.000	0.000	0.000	0.000
B	1.000	1.000	1.000	0.909	1.000	1.000	1.000	1.000
C	0.000	0.000	0.000	0.045	0.000	0.000	0.000	0.000
n	27	9	1	22	19	14	31	20
Hk-2								
A	1.000	1.000	1.000	1.000	1.000	0.905	1.000	1.000
B	0.000	0.000	0.000	0.000	0.000	0.095	0.000	0.000
n	27	13	9	31	42	42	54	34
Mdh-2								
A	0.350	0.182	0.000	0.000	0.029	0.094	0.018	0.000
B	0.650	0.818	1.000	1.000	0.971	0.906	0.982	1.000
n	40	11	1	29	35	32	55	34
Me								
A	0.000	0.000	0.000	0.000	0.048	0.000	0.000	0.000
B	1.000	1.000	1.000	1.000	0.952	1.000	1.000	1.000
n	28	12	8	28	42	28	50	33

Table 3 (***concluded***).

	Sampling locality*							
Locus	GM	GP	BW	UM	MW	SC	IN	AB
Pep_{gl}								
A	0.000	1.000	1.000	0.227	0.000	0.000	0.054	0.933
B	0.000	0.000	0.000	0.591	0.000	0.000	0.000	0.000
C	0.000	0.000	0.000	0.000	0.000	0.000	0.135	0.000
D	0.000	0.000	0.000	0.182	0.933	0.923	0.811	0.067
E	1.000	0.000	0.000	0.000	0.067	0.077	0.000	0.000
n	32	12	9	22	30	26	37	30
Pgm-2								
A	0.000	0.000	0.000	0.000	0.000	0.125	0.000	0.000
B	1.000	1.000	1.000	1.000	1.000	0.875	1.000	1.000
n	23	7	3	22	31	24	43	24
H_{exp}	0.046	0.038	0.000	0.043	0.035	0.057	0.045	0.006
Mean (n)	28.1	10.7	5.0	25.3	32.0	25.3	42.1	24.9

* Abbreviations as in Table 1.

Table 4. Genetic (above) and geographic (below) distances among the eight samples of *Lasioglossum* (*Evylaeus*) *boreale*.

	Sampling locality*							
	GM	GP	BW	UM	MW	SC	IN	AB
GM	—	0.101	0.105	0.092	0.114	0.123	0.118	0.155
GP	173.2	—	0.002	0.076	0.114	0.126	0.112	0.094
BW	367.7	288.9	—	0.072	0.110	0.122	0.108	0.096
UM	890.7	728.2	610.1	—	0.039	0.050	0.040	0.069
MW	3598.1	3471.8	3630.1	3243.1	—	0.002	0.001	0.104
SC	4176.9	4021.1	4098.8	3578.5	1211.8	—	0.002	0.116
IN	4242.9	4092.6	3901.1	3372.5	4421.0	3648.0	—	0.103
AB	8038.5	7865.5	7815.0	7205.5	5534.1	4371.3	4679.9	—

* Abbreviations as in Table 1.

right of the aforementioned pair of points (Fig. 2). The tight cluster to the right of the plot with intermediate N_{m} values represents gene flow between the Utah sample and the three northern ones. The large number of points just beneath these represents low levels of gene flow and large geographic distances. All of the data involving the sole European sample fall here, as do comparisons between Arizona and northern North American samples. The lowest gene flow estimate, at the bottom right-hand corner of the plot is between Abisko, Sweden, and the Graham Mountains, Arizona.

To summarize the genetic differentiation data: there was high genetic differentiation among some of the geographically closest populations (*e.g.*, the Graham Mountains and Green's Peak, which are separated by only 173 km, D = 0.101). Conversely, some of the geographically most distant samples are genetically similar (*e.g.*, Mount Washington and Inuvik, which are separated by over 4400 km, D = 0.001).

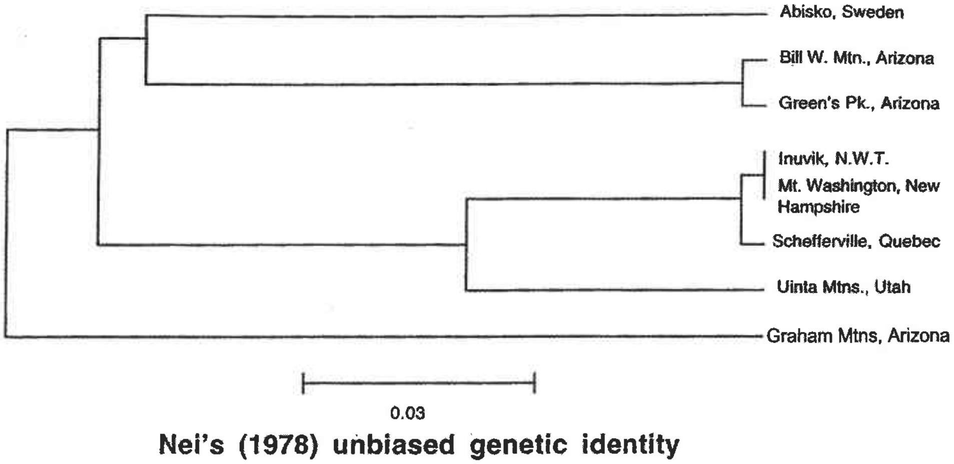

Figure 1. Unweighted pair group mathematical average (UPGMA) phenogram based upon pairwise genetic distances among samples of *Lasioglossum* (*Evylaeus*) *boreale*. Note that this pattern should not be taken as representing the possible evolutionary relationships among the samples, rather it is used solely to show the relative levels of genetic divergence among populations.

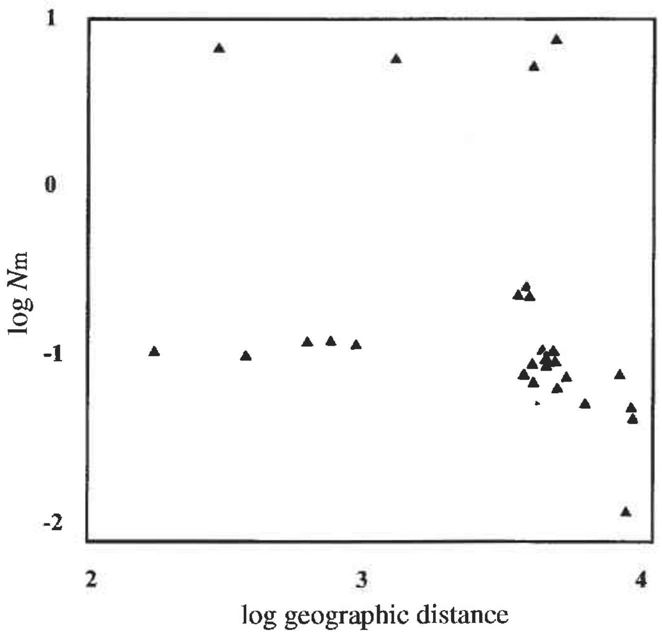

Figure 2. Isolation by distance plot for pairwise estimates of the logarithm of the number of migrants per generation (N_m) against the logarithm of the geographic distance separating the localities where *Lasioglossum* (*Evylaeus*) *boreale* was sampled.

Discussion

The migration of organisms as a result of large-scale climate change is a subject of considerable interest given the reality of global warming. It has been well established that the geographic ranges of many northern hemisphere insects have shifted northwards over the past 100 years (Parmesan 1996). Rapid climate change also occurred at the end of the most recent ice age with ice-sheet area decreasing by 10% over only a few hundred years but with glacial retreat declining in rapidity thereafter (Yokoyama *et al.* 2000). Thus, between the initiation of glacial retreat and the hypsithermal (Flandrian) of about 8000 years ago, much of southern Canada and the northern United States changed from being covered in ice to supporting complex ecosystems under climatic conditions that were slightly warmer than they are now. This dramatic change happened over only a few thousand years.

Assuming it persisted in a refugium to the south of the ice sheets, as a species adapted to cold climates, *L. boreale* would have had to migrate northwards at the same rate as the glaciers retreated or adapt to increasing temperatures (Pielou 1991). An additional route to survival would have been available for populations in mountainous regions, as these could move uphill to track short summer habitats at higher elevations. Populations of short summer-adapted species on the "sky islands" of the southwestern United States would have become separated from one another early in the process of glacial retreat. Those in the contiguous Rockies may have been in genetic contact with one another comparatively more recently, as mountains farther north would have become increasingly suitable for *L. boreale* as the climate warmed. Furthermore, more northerly mountains would become suitable at increasingly lower elevations because of the influence of high latitudes added to the cooling effect of altitude. As temperatures became similar to those of today, *L. boreale* could persist barely above sea level at the northern edge of its range (*e.g.*, at Inuvik, 50 m above sea level). Meanwhile, those populations on the lowest altitude mountains would probably have died out as the climate warmed beyond their ecological tolerance at the peak of their particular mountain range. This effect would have been most acute during the glacial hypsithermal of 8000 years ago. An altitude of at least 2500 m would seem to be required for the species to have persisted to the present in the southernmost portions of its range in Arizona (L Packer, unpublished observation).

Reduced levels of genetic variation are expected in populations that have dispersed over large distances over relatively short periods of time (Hewitt 1996). For example, species with current ranges that include regions north of the glacial limit but which passed the last glacial maximum to the south of the ice sheets would be expected to have a pattern of decreasing genetic variation from south to north. Armbruster *et al.* (1998) found a significant reduction in H_{exp} with increasing latitude for the pitcherplant mosquito, *Wyeomyia smithii* (Coq) (Diptera: Culicidae). Similarly, Ashworth *et al.* (1996) found reduced mtDNA haplotype diversity in eastern arctic and subarctic populations of the ground beetle *Amara alpina* Paykull (Coleoptera: Carabidae), a species for which their molecular data strongly supported a Beringian refugium origin for all present day northern populations.

We did not find reduced genetic variation in our northern samples compared with those from the southwestern United States; however, all of the southern samples were collected from isolated mountain peaks. We expect *L. boreale* to persist in small populations on these mountaintops with a concomitant reduction in expected levels of genetic variability compared with larger populations in the more extensive areas of suitable habitat found in the northern portions of the species' range. These isolated mountaintop localities would, presumably, have harboured even smaller populations during the glacial hypsithermal of 8000 years ago. Consequently, we do not take the lack of pattern in heterozygosity among North American samples as arguing against northward migration of the ancestors of the more northern populations following the retreat of the glaciers; however, the sample from Sweden has the lowest recorded heterozygosity for any of the 26 bee species we have studied (Packer and Owen 1992;

Rosenmeier and Packer 1993; Packer *et al.* 1999 and references therein; JS Taylor and L Packer, unpublished data). This is suggestive of recent colonization of this area, presumably from farther east. It would be interesting to obtain additional samples of this species from across arctic and subarctic regions of the Old World to investigate this further.

There are fixed differences among some of our populations: the Swedish sample is fixed for a different electromorph at *Acy*, the Graham Mountains bees are fixed for a unique allele at *Gda*, and this locus also completely differentiates the other two Arizona samples from all other bees. Application of the phylogenetic species concept to these data (Davis and Nixon 1992; Packer and Taylor 1997) would suggest that we might be dealing with three species rather than just one; however, this approach to genetic data is somewhat controversial (Avise 2000; Wheeler and Meier 2000). We would not want to make any strong statements on the species-level status of the various populations we have sampled in the absence of larger sample sizes from the same locations in combination with intensive sampling of intervening sites. Even if the fixed genetic differences among populations did reflect species-level differences, this would not alter the biogeographic scenarios we discuss below.

Our genetic-distance and gene-flow data indicate relative homogeneity among the three northern samples despite the large geographic distances separating them. Genetic distances between the Mount Washington sample and the other two northern ones are very small and all three also have similar heterozygosity levels. Habitat suitable for *L. boreale* on Mount Washington is presently isolated from other suitable areas by at least 500 km. It is possible that the species occurs on the Gaspé Peninsula of Quebec, but it does not occur on the small region of tundra on Mount Katahdin in Maine (JS Taylor, unpublished observations). It is unlikely that the genetic similarity among the Mount Washington and the other two northern samples could have resulted from migration from Beringia eastwards and southwards to the northern edge of the Appalachian Mountains. If this were the case lower levels of genetic variation would be expected from west to east, as found for *A. alpina* (Ashworth *et al.* 1996). We suggest that the northern populations passed the recent ice age south of the ice sheet and dispersed northwards following the retreat of the glaciers. As the climate warmed further, a population was left behind on Mount Washington as the nearby lower elevation habitat became too warm for *L. boreale.*

Populations on isolated mountaintops in the southwestern United States exhibit high levels of genetic differentiation even over small geographic distances. This suggests that the southern populations may have become separated from one another a long time ago, perhaps preceding the most recent ice age. A similar conclusion was reached in a detailed study of a jumping spider from intermediate altitudes in the “sky islands”. Masta (2000) studied mtDNA differentiation of the jumping spider *Habronattus pugillis* Griswold (Aranea: Salticidae) from southeastern Arizona and found that for reasonable clock rates of mtDNA divergence, the predicted dates of separation among populations preceded the last ice age. Even for the two most recently separated groups of populations, Masta (2000) estimated the divergence time to be 30 000 years ago, with other populations diverging as early as 2 millions years ago. Although one must take molecular-clock estimates as being only a very rough indication of divergence times, these values are not consistent with divergence since the last ice age. Because of its interesting geographic distribution, similar molecular studies of *L. boreale* could prove particularly illuminating.

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References

Armbruster P, Bradshaw WE, Holzapfel CM. 1998. Effects of postglacial range expansion on allozyme and quantitative genetic variation of the pitcher-plant mosquito *Wyeomyia smithii*. *Evolution* **52**: 1697–704

Ashworth AC, Schwert DP, Reiss RA. 1996. Biogeography of arctic beetles: integrating the results of paleontological and molecular population genetic studies. *Arctic Insect News* **7**: 12–5

Avise, JA. 2000. Cladists in wonderland. *Evolution* **54**: 1828–32

Clarke TE, Levin DB, Kavanaugh DH, Reimchen TE. 2001. Rapid evolution in the *Nebria gregaria* Group (Coleoptera: Carabidae) and the Paleogeography of the Queen Charlotte Islands. *Evolution* **55**: 1408–18

Davis JI, Nixon KC. 1992. Populations, genetic variation, and the delimitation of phylogenetic species. *Systematic Biology* **41**: 421–35

Gill GB, Nostrom AM, Mack AL. 1993. Speciation in North American chickadees: I Patterns of mtDNA genetic divergence. *Evolution* **47**: 195–212

Hayes JD, Harrison RG. 1992. Variation in mitochondrial DNA and the biogeographic history of woodrats (*Neotoma*) of the eastern United States. *Systematic Biology* **41**: 331–44

Hewitt GM. 1996. Some genetic consequences of ice ages, and their role in divergence and speciation. *Biological Journal of the Linnean Society* **58**: 247–76

Hewitt GM. 2000. The genetic legacy of the Quarternary ice ages. *Nature (London)* **405**: 907–13

Lamb T, Avise JC, Gibbons JW. 1989. Phylogeographic patterns in mitochondrial DNA of the desert tortoise (*Xerobates agassizi*) and evolutionary relationships among the North American gopher tortoises. *Evolution* **43**: 76–87

Lamb T, Jones TR, Wettstein PJ. 1997. Evolutionary genetics and phylogeography of tassel-eared squirrels (*Sciurus abierti*). *Journal of Mammalogy* **78**: 117–33

Masta, S. 2000. Phylogeography of the jumping spider *Habronattus pugillis* (Araneae: Salticidae): recent vicariance of sky island populations? *Evolution* **54**: 1699–711

Matthews, JV Jr. 1979. Tertiary and quarternary environments: historical background for an analysis of the Canadian insect fauna. pp. 7–30 *In* HV Danks (Ed), *Canada and its insect fauna. Memoirs of the Entomological Society of Canada* **108**

Nei M. 1978. Estimation of average heterozygosity and genetic distance from a small number of individuals. *Genetics* **89**: 583–90

Packer L, Owen RE. 1989. Isozyme variation in *Halictus rubicundus*: a primitively eusocial bee (Hymenoptera: Halictidae). *The Canadian Entomologist* **121**: 1049–58

——— 1990. Allozyme variation, linkage disequilibrium and diploid male production in a primitively social bee *Augochlorella striata* (Hymenoptera: Halictidae). *Heredity* **65**: 241–8

——— 1992. Variable enzyme systems in the Hymenoptera. *Biochemical Systematics and Ecology* **20**: 1–7.

——— 2001. Population genetic aspects of pollinator decline. *Conservation Ecology* **5**(1): 4. Available from http://www.consecol.org/vol5/iss1/art4 (accessed on 5 April 2001)

Packer L, Taylor JS. 1997. How many cryptic species are there? An application of the phylogenetic species concept to genetic data for some comparatively well known bee species. *The Canadian Entomologist* **129**: 587–94

Packer L, Porsa A, Plateaux-Quénu C, Plateaux L. 1999. A cryptic species allied to *Evylaeus villosulus* (Kirby) (Hymenoptera: Halictidae). *Annales de la Societe Entomologique de France* **35**: 165–71

Parmesan C. 1996. Climate and species' range. *Nature (London)* **382**: 765–6

Peterson MA, Denno RF. 1998. The influence of dispersal and diet breadth on patterns of genetic isolation by distance in phyophagous insects. *American Naturalist* **152**: 428–46

Péwé, TL. 1983. The periglacial environment in North America during Wisconsin time. pp 157–89 *In* HE Wright Jr. (Ed), *Late-quarternary environments of the United States*. Minneapolis: University of Minnesota Press

Pielou EC. 1991. *After the ice age: the return of life to glaciated North America*. Chicago: University of Chicago Press

Riddle BR, Honeycutt RL. 1990. Historical biogeography in North American arid regions: an approach using mitochondrial-DNA phylogeny in grasshopper mice (genus *Ochomys*). *Evolution* **44**: 1–14

Rosenmeier L, Packer L. 1993. A comparison of genetic variation in two sibling species pairs of haplodiploid insects. *Biochemical Genetics* **31**: 185–200

Routman E, Wu R, Templeton AR. 1994. Parsimony, molecular evolution and biogeography: the case of the North American giant salamander. *Evolution* **48**: 1799–809

Sakagami SF, Toda MJ. 1986. Some arctic and subarctic solitary bees collected at Inuvik and Tuktoyaktuk, NWT, Canada (Hymenoptera: Apoidea). *The Canadian Entomologist* **118**: 395–405

Slatkin M. 1993. Isolation by distance in equilibrium and non-equilibrium populations. *Evolution* **47**: 264–79

Sokal RR, Rohlf FJ. 1995. *Biometry*. 3rd edition. New York: WH Freeman and Co

Sokal RR, Sneath PHA. 1973. *Numerical taxonomy*. New York: WH Freeman and Co

Svensson BG, Ebmer PAW, Sakagami SF. 1977. *Lasioglossum* (*Evylaeus*) *boreale*, a new Halictinae (Hymenoptera: Apoidea) species found in northern Sweden and on Hokkaido, Japan, with notes on its biology. *Entomologica Scandinavica* **8**: 219–29

Swofford DL, Selander RB. 1989. *BIOSYS-1: a computer program for the analysis of allelic variation in population genetics and biochemical systematics*. Release 1.7. Champaign: Illinois Natural History Survey

Weir BS, Cockerham CC. 1984. Estimating *F*-statistics for the analysis of population structure. *Evolution* **38**: 1358–70

Wheeler QD, Meier R (*Editors*). 2000. *Species concepts and phylogenetic theory*. New York: Columbia University Press

Whitlock MC, McCauley DE. 1999. Indirect measures of gene flow and migration: $F_{ST} \neq 1/(4N_m + 1)$. *Heredity* **82**: 117–25

Yokoyama Y, Lambeck K, De Deckker P, Johnston P, Fields LK. 2000. Timing of the last glacial maxima from observed sea-level minima. *Nature (London)* **406**: 713–6

Zink RM, Dittmann DL. 1993. Gene flow, refugia, and evolution of geographic variation in the song sparrow (*Melospiza melodia*). *Evolution* **47**: 717–29

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