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# MITOCHONDRIAL GENE GENEALOGY AND GENE FLOW AMONG ISLAND AND MAINLAND POPULATIONS OF A SEDENTARY SONGBIRD, THE GREY-CROWNED BABBLER (*POMATOSTOMUS TEMPORALIS*)

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*Abstract.*—Distinguishing between ongoing gene flow and purely historical association of populations can be difficult without data on times of population separation and effective population sizes. To help discriminate between these two scenarios, I examined mitochondrial DNA sequence diversity in three geographically close populations of the grey-crowned babbler (*Pomatostomus temporalis*) separated by water barriers of known age in the Northern Territory, Australia, using the polymerase chain reaction (PCR), direct sequencing, and genealogical methods of inference. PCR primers were designed to obtain sequences from region I, a highly variable segment of the control region. Sequence diversity in all populations was consistent with neutrality. In the population on Melville Island, a Pleistocene land-bridge island, sequence variability is as high as on the mainland and consists of two mitochondrial lineages differing by 2%. Phylogenetic analyses of the sequence variation observed among 44 individuals suggest that the number of times lineages in one population trace back to ancestors of a different population (between-population coalescent events) was too high to be compatible with a model of population divergence solely by drift since rising of the water barriers, implying instead recent or ongoing gene flow across water barriers. Similar estimates of $F_{st}$, the fraction of genetic diversity apportioned among populations, were obtained when calculated using the divergence times of alleles and when estimated from *Nm* values derived from trees and ranging from 0.29–0.55. Both the phylogenies and patterns of allelic divergence suggest that the population on Melville Island exchanges migrants with both continental populations, although statistical tests indicated that some alternative phylogenies implying restricted gene flow among the populations could not be discounted.

*Key words.*—Coalescent process, control-region sequences, intraspecific phylogeny, island populations, mitochondrial DNA.

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Population genetics is undergoing a minor revolution. The polymerase chain reaction (PCR) and direct-sequencing methods are for the first time making feasible large-scale surveys of population variation at the nucleotide level (Vigilant et al. 1989, 1991; Thomas et al. 1990; Di Rienzo and Wilson 1991; Schaeffer and Miller 1991; Edwards 1993; Prager et al. 1993). Sequence data provide a closer view of the molecular evolutionary process than does restriction analysis and have the potential to resolve better the branching pattern and depths of intraspecific phylogenetic trees (Kocher and Wilson 1991; Di Rienzo and Wilson 1991). Coincident with this technical advance is the development of theory ("genealogical" or "coalescent" theory) specifically designed to predict expected patterns in genealogical trees under a wide variety of population genetic models (Tajima 1983; Tavaré 1984; Takahata and Nei 1985; Slatkin 1989; Slatkin and Maddison 1989, 1990; Takahata 1989). Appropriately, the common ground of these two avenues is the phylogenetic tree, which unlike the frequencies of electrophoretic variants, can summarize both the history and extent of divergence of alleles (reviewed in Hudson 1991; Kreitman 1991).

Because of its lack of recombination, rapid evolutionary rate, and clonal (i.e., usually maternal) inheritance in animals, mitochondrial DNA (mtDNA) is especially suited for the study of closely related populations using genealogical trees (Avise et al. 1987; Avise 1989). The mtDNA control region, the major noncoding region containing the origin of heavy strand replication and the displacement loop ("D loop"), is well known

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as the most variable part of the animal mitochondrial genome (Brown 1985). In particular, a highly variable segment of the control region, region I, contains the highest proportion of variable sites of any mitochondrial segment and has been useful in defining closely related mitochondrial lineages within species (Vigilant et al. 1989, 1991; Thomas et al. 1990; Di Rienzo and Wilson 1991; Edwards 1993).

### *Importance of Genealogical Analysis*

Initial surveys of sequence variation in region I and other parts of the control region revealed high levels of sequence diversity; Vigilant et al.'s (1989) survey of 83 human control regions yielded 71 distinct mtNDA types. Such high rates of base substitution not only facilitate phylogenetic analysis of closely related genomes but necessitate the use of the newer genealogical models. This necessity arises in part from the fact that the levels of polymorphism now detectable using PCR can obscure the record of other population processes, such as migration when the mutation rate is high. For example, as pointed out by Slatkin and Maddison (1989), traditional calculation of Wright's (1951) $F_{st}$ (the fraction of genetic diversity apportioned among populations) from the frequencies of haplotypes in different populations would yield a statistic that incorporated the effects of *both* mutation and migration. The recent studies of human mtDNA show that the rate of mutation to new sequences keeps the population frequency of any one sequence and the chance of detecting identical sequences in separate localities quite low. Such data are dealt with poorly by traditional measures of $F_{st}$, which were designed under the assumption that migration, not mutation, was the primary factor producing the distribution of allelic types among populations. Additionally, like other phylogenetic methods of comparison (e.g., Ridley 1983; Harvey and Pagel 1991), genealogical analyses of alleles permit the explicit counting of historical events (e.g., migration events; Slatkin 1989; Slatkin and Maddison 1989, 1990). Of course, as with any method relying on a phylogeny, uncertainty in the phylogeny itself may prove a hindrance in the inference of population processes.

### *Avian mtDNA and Grey-Crowned Babblers*

The conclusion that most bird species show little population subdivision is based primarily on protein electrophoretic surveys of migratory species (Barrowclough 1983) and may need to be expanded to incorporate the results of recent mtDNA studies (e.g., Zink 1991; Taberlet et al. 1992) and electrophoretic surveys of sedentary species (Baker and Moeed 1987; Peterson 1992); nonetheless, some avian mtDNAs exhibit patterns with little phylogeographic structure (e.g., Ball et al. 1988). Initial results of the PCR approach, although directed toward more slowly evolving mtDNA segments than those available now (Quinn 1992), suggested that analyses of sequence diversity will provide additional details on the evolutionary history of bird populations (Edwards and Wilson 1990; Birt-Friesen et al. 1992).

Grey-crowned babblers (*Pomatostomus temporalis*) are a sedentary, social species with an extensive range throughout Australia and southern New Guinea. Although grey-crowned babblers have been traditionally classified into several subspecies, including a distinct form on Melville Island (*P. t. bamba*; Deignan 1964), more recent analyses showed that much of the morphological variation was clinal (Hall 1974; R. Schodde pers. comm. 1990) and suggested grouping the Australian populations into two major taxa (eastern "grey-crowned" *P. t. temporalis* and western "red-breasted" *P. t. rubeculus*). This division was supported by phylogenetic analysis of sequences from a portion of the cytochrome *b* gene (Edwards and Wilson 1990). However, this mtDNA segment proved useful for defining only major branches in the lineage of grey-crowned babblers and offered little resolution of more recent branching events.

Here I report on sequence variation in a 400-bp segment of region I among three geographically close populations of babblers in the Northern Territory, one of which is found on a land-bridge island, Melville Island. Unusual biogeographic settings such as water barriers are of particular interest in the study of sedentary species, because frequently the barriers can be dated and expectations of their effect on gene flow can be tested (e.g., Busack 1986; Capparella 1988). I use the gene genealogies derived from the sequences and the times of separation of the populations inferred from palynological data (see below) to help distinguish between two plausible explanations for the observation of shared mitochondrial lineages among populations (Avise et al. 1984; Neigel and Avise 1986; Takahata 1989; Slatkin and Maddison 1989): (1) ongoing gene flow and (2) retention (incomplete "sort-

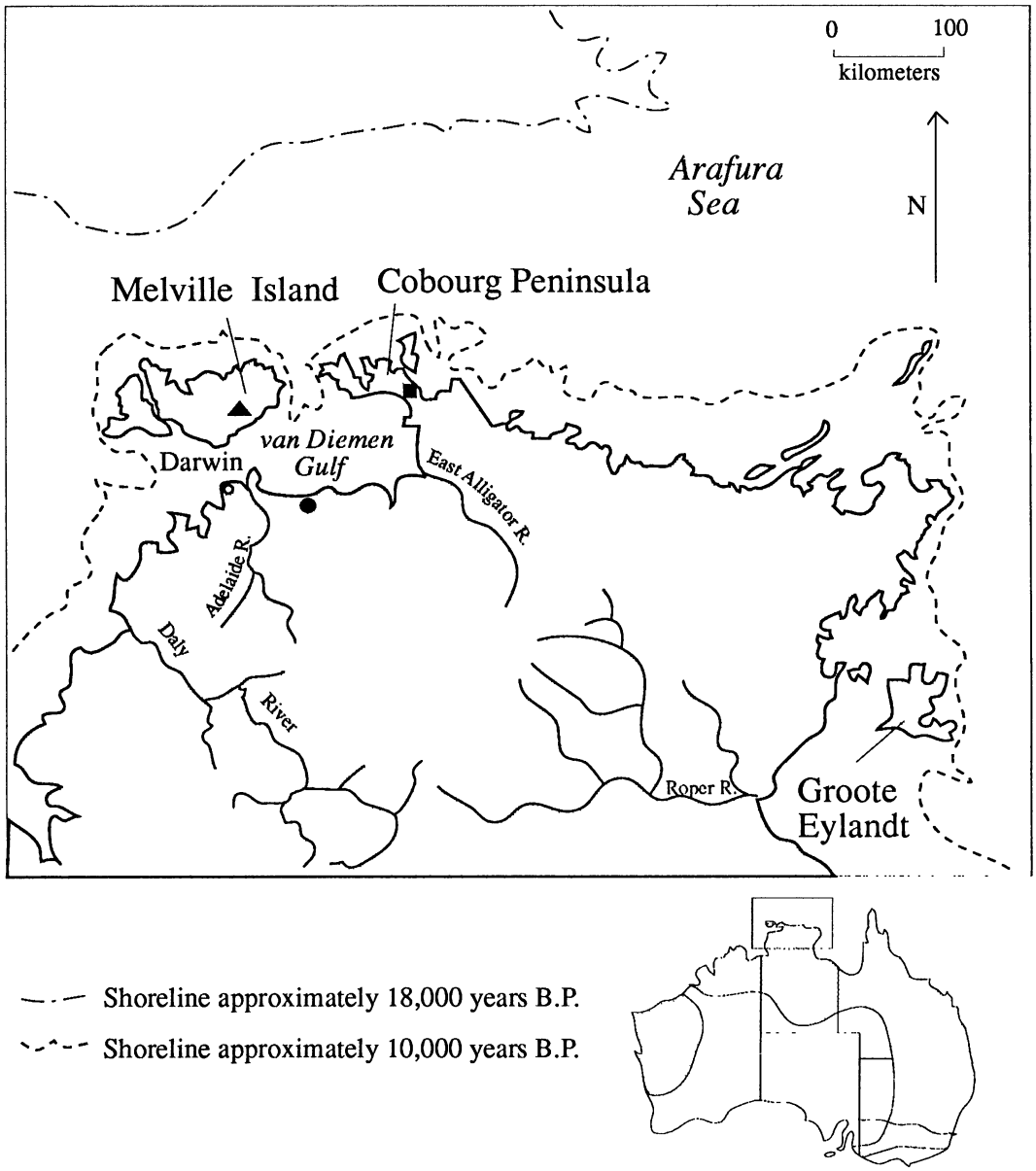



FIG. 1. Map showing the three sampling localities of babblers in the Northern Territory, Australia, in relation to Pleistocene water lines (modified from Woodroffe and Mulrennan [1991] with permission from the authors). The black circle, triangle, and square mark the locations of the Darwin, Melville Island, and Cobourg Peninsula populations, respectively. Inset: Distribution of the grey-crowned babbler in Australia (indicated by stippling).

ing") of ancestral mtDNA lineages in the absence of gene flow caused by large effective population size ($N_e$).

## MATERIALS AND METHODS

*Field Methods and Sampling of Babbler Populations.*—In the present study, I used a total of 44 individuals from three red-breasted populations (Darwin, Melville Island, Cobourg Peninsula; fig. 1); four individuals from the grey-crowned subspecies were used as outgroups (table 1). In 1987, I collected tissues from individuals in the Darwin population for mtDNA isolation ($N = 15$; see Edwards and Wilson 1990 and below). Populations on Melville Island ($N = 15$) and the Cobourg Peninsula ($N = 14$) were

sampled nondestructively. We lured individuals into triangulated mist nets using tape recordings of "yahoo" and distress calls (King 1980). We pierced the brachial vein with a sterile syringe, collected 200–400 μL of blood in 500 μL of an EDTA (ethylenediamine tetraacetate)-based storage buffer (Arctander 1988), and kept samples at room temperature until refrigeration or permanent storage at −70°C was available (from 2 h to 14 d). The iris color of each individual indicated age; we used categories described in Counsilman and King (1977) and J. L. Brown et al. (1982) and lumped their "medium brown" and "dark brown" irises as "brown" (see legend, table 1). Because babblers are highly social, with social units generally consisting of a mated pair and 1–15 auxiliary "helpers at the nest" (Brown et al. 1983; Brown 1987; Ford 1989; pers. obs.), we usually caught multiple individuals from single, easily delimited social units (table 1). I consider the potential influence of this social structure on the interpretation of results presented (see below). The 44 individuals studied came from 19 social units and included five brown-eyed juveniles; one of these juveniles was the sole representative of its social unit (table 1). Some individuals from the Darwin population were previously screened for variation in a short piece of the cytochrome *b* gene (see table 1 and Edwards and Wilson 1990; see also Edwards 1993).

Melville Island is a low-lying Pleistocene landbridge island situated approximately 20 km from the closest mainland point. Submergence of the van Diemen Gulf is thought to have last separated Melville Island from the mainland and adjacent Cobourg Peninsula about 8000 and 10,000 B.P., respectively, before which the gulf underwent repeated fluctuations like much of the Arafura Sea to the north (Veevers 1969; Jongsma 1970; reviewed in Woodroffe and Mulrennan 1991; R. Fensham pers. comm. 1992). Thus, in the model of population divergence based solely on genetic drift, I use these times as operational, maximum times of separation of populations of babblers, which were presumably distributed continuously throughout suitable habitats in the region during periods of land emergence. Such distributions have been suggested for other animal and plant species separated elsewhere in the Australian region by contemporary water barriers (Schodde and Calaby 1972; Moran et al. 1989).

*DNA Isolation, Polymerase Chain Reaction, and Direct Sequencing.*—mtDNAs from the Darwin population and one of the outgroup individuals (table 1) were isolated by CsCl gradient centrifugation as described (Brown 1980; Shields and Wilson 1987; see Edwards and Wilson 1990). For the remaining samples, total genomic DNA was isolated by digesting 50–100 μL of the combination of blood plus storage buffer for 3–12 h with 7 μL of 25% SDS (sodium dodecyl sulfate) and 15 μL of proteinase K (15 μg/mL) in 450 μL

TABLE 1. *Pomatostomus temporalis rubeculus* individuals and social units sampled and mtDNAs observed in three localities in the Northern Territory. All individuals are adults (>3½ yr old, cream irises) unless marked with a letter in parentheses indicating eye color as follows: B, brown, juvenile; L, light brown, ca. 3 yr; F, flecked, ca. 3½ yr. After the colon following each individual or set of individuals is the social unit designation of those individuals. Asterisks by individuals in the Darwin population denote those also characterized for sequence variation in the cytochrome *b* gene (Edwards and Wilson 1990). See Materials and Methods for details.

| Locality | mtDNA type | Individual number: social unit |
|---|---|---|
| 100 km E Darwin | 1 | 152, 153(B), 155(L)*: 13 |
| | 2 | 156*: 13 |
| | 3 | 157, 158: 14; 160: 15; 161, 162, 164, 165(L), 167 (B): 16 |
| | 4 | 159: 15 |
| | 5 | 168*: 17 |
| | 6 | 170*: 18 |
| Totals | | 15: 6 |
| Pikertaramoor, Melville Island | 7 | 483: 43; 489(B): 46; 490, 491, 493: 47 |
| | 8 | 484: 44; 495(F): 49; 497(L): 50 |
| | 9 | 485: 44 |
| | 10 | 487: 45 |
| | 11 | 486, 488: 45 |
| | 12 | 494: 48 |
| | 13 | 496(L): 49 |
| | 14 | 498: 50 |
| Totals | | 15:8 |
| Cobourg Peninsula | 15 | 520: 57 |
| | 16 | 521: 57 |
| | 17 | 522: 57; 523(F): 58 |
| | 18 | 524(B): 59 |
| | 19 | 525: 59 |
| | 20 | 527(F): 59 |
| | 21 | 528: 59; 529: 60 |
| | 22 | 530(B), 531(F): 60; 532, 533, 534: 61 |
| Totals | | 14: 5 |
| All places and social units | | 44: 19 |
| *P. t. temporalis* (outgroups) | O1–O4 | 4 |

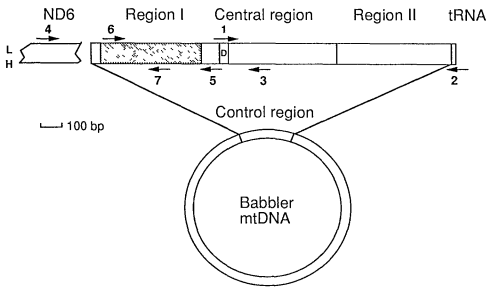



FIG. 2. Strategy for primer design, amplification, and sequencing of the 5′ end of the babbler control region (region I). Arrows indicate primers on either the light (L) or heavy (H) strand. Initial sequences of the control region were obtained with primer 1, directed toward the conserved "D" box as shown, and primer 2, complementary to the phenylalanine tRNA (Quinn and Wilson 1993). From knowledge of this sequence and partial alignment with the homologous region in chicken mtDNA (Desjardins and Morais 1990), I designed primer 3, which was used in conjunction with a primer nested in the center of the gene for NADH dehydrogenase subunit 6 (primer 4, designed by Alan Cooper, University of Wellington, New Zealand) to amplify a 1.3-kb segment spanning the entire 5′ half of the control region, $tRNA^{glu}$ (not shown) and nearly all of the ND6 gene. Partial sequences of this segment were utilized to design primers 5–7, which are used to define region I in this study (see Materials and Methods). The primer sequences, in the 5′ → 3′ direction and with laboratory designations in parentheses, are: 3 (HD4), CCCGACCAGCTGCATCTGTG; 4 (ND6C), CCGAGACAACCCACGCACAAG; 5 (HD14), GGGGAATGTAGGCTCCT; 6 (LD11), GGGTATGTATATCTTGCATACAATTAT; 7 (HD6), TCGTTTACGTGAGGTCGT. Primers 3 and 4 as well as primer 3 in conjunction with NDGE (CCATAACCAACAACCTGTCAAT; not shown, located to the right of primer 4), amplify a homologous segment of mtDNA in a wide range of passerine and nonpasserine bird species, whereas the sequences of primers 5–7 were designed specifically to amplify *Pomatostomus temporalis* mtDNA.

of SET buffer (0.15 M NaCl, 0.05 M Tris, 1 mM EDTA, pH 8.0; Wetton et al. 1987). Proteins and other cellular debris were precipitated with 350 μL of 5 M NaCl, after which the tubes were spun at 14,000 rpm for 30 min (Medrano et al. 1990). DNA in 450 μL of the supernatant was precipitated, dried, and stored in 10 mM Tris (pH 8.0) and 1 mM EDTA for enzymatic amplification.

Double-stranded PCR amplification, generation of single-stranded templates by the unbalanced primer method (Gyllensten and Erlich 1988), and direct sequencing were performed as described (Kocher et al. 1989). Amplifications were performed in a Perkin Elmer Cetus Thermocycler, generally for 30 cycles, with each cycle consisting of 40 s at 94°C, 1 min at 50°C, and 1 min at 72°C and with the limiting primer diluted 100- to 200-fold for production of single-stranded templates for sequencing. Three primers (5, 6, and 7 in fig. 2) were used to amplify and sequence region I. These primers span a 429-bp segment of babbler region I, from which 400 bp of sequence were routinely obtained. Sequences were aligned by eye using the sequence editor ESEE (Cabot and Beckenbach 1989).

### *Phylogenetic Analyses*

*Estimates of Sequence Divergence, Phylogenetic Trees, and Population Parameters.*—The computer package PHYLIP (version 3.4, Felsenstein 1991) on a SUN SPARC station was used for all calculations of sequence divergence and tree building (except generation of user trees, see below). Biased base composition of the sequences and the potential for rate heterogeneity among nucleotide sites influenced the choice of methods for calculating sequence divergence (Edwards 1992). I used the maximum-likelihood model of Hasegawa et al. (1985) as implemented in PHYLIP's DNADIST program, with a transition/transversion ratio of 20:1 and the assumption of equal rates of change among all sites. Because there is evidence (Wakeley 1993) that rates of change among sites in these sequences are heterogeneous, I also estimated sequence divergence with an assumption of a gamma distribution of rates among sites (Jin and Nei 1990). With the latter approach, I assumed a rate parameter ($\alpha$) of 0.5, which specifies a high level of skew in rates; this value was suggested by the mtDNA analysis of Wilson et al. (1989). For phylogenetic reconstructions, I employed maximum-likelihood (DNAML; Felsenstein 1981) and distance algorithms. The distance methods included the Fitch-Margoliash (1967) algorithm; KITSCH (assumes equal evolutionary rates among lineages; Felsenstein 1984); and the neighbor-joining method (Saitou and Nei 1985).

I used cladistic methods to estimate the extent and pattern of gene flow among populations and to test for population structure. The minimum number of inferred migration events, *s*, was counted using a parsimony reconstruction of geographic states (MacClade version 3.0β; Maddison and Maddison 1992) as described in Slatkin and Maddison (1989). Situations in which lineages stemming from common ancestors are currently found in different contemporary populations could also be caused by historical association of populations in the absence of gene flow (e.g., fig. 8 of Slatkin and Maddison 1989).

When the values of $s$ are assumed to reflect gene flow, they are referred to as migration events and are converted to estimates of $Nm$ (the average number of migrants per generation between populations, where $N$ is the effective population size in a Wright-Fisher model and $m$ is the fraction of each population undergoing migration) using table 1 in Slatkin and Maddison (1989). In such cases, although the value of $s$ is usually unambiguous, there can be ambiguity as to which branches of the tree to assign migration events most parsimoniously. I reconstructed migration events on the trees using DELTRAN (in which events are delayed as much as possible), as opposed to ACCTRAN (in which events are forced to occur as soon as possible moving from the root to the tips of the tree). An important assumption of the cladistic method is that the sequences being analyzed are evolving in a selectively neutral fashion. The method proposed by Tajima (1989) was used to test this hypothesis.

Assuming equilibrium, I calculated $F_{st}$ in two ways to compare the results of different methods: (1) by equating $F_{st}$ with $1/(1 + 2Nm)$, where $Nm$ was estimated from $s$ (Hudson et al. 1992); and (2) using an approach relying on average coalescence times of alleles within and between populations (modification of eq. 8 in Slatkin [1991] and M. Slatkin [pers. comm. 1992]; cf. Lynch and Crease [1990]):

$$F_{st} = \frac{\bar{t}_b - \bar{t}_w}{\bar{t}_b}, \quad (1)$$

where $\bar{t}_b$ and $\bar{t}_w$ are the mean times to common ancestry of all alleles between and within populations, respectively. If we assume a strict molecular clock (which follows from neutrality assumptions of the equation), then

$$\frac{\bar{t}_b - \bar{t}_w}{\bar{t}_b} = \frac{\bar{d}_b - \bar{d}_w}{\bar{d}_b}, \quad (2)$$

where $\bar{d}_b$ and $\bar{d}_w$ are the mean sequence divergence of all alleles between and within populations, respectively, calculated from the distance matrix. Lynch and Crease's (1990) method, which uses a Jukes and Cantor (1969) correction for restriction site data but is otherwise similar to equation (2), was also used by treating each nucleotide as a restriction site (M. Lynch pers. comm. 1992). Whereas the first method of calculating $F_{st}$ incorporates information on the phylogenetic relationships of the sampled alleles, the second method (eq. 2) incorporates information on the extent of divergence among alleles and their frequency in each population.

*Cladistic Hypothesis Testing and Statistical Analysis.*—Trees generated under alternate assumptions of gene flow provided a statistical framework for testing both the robustness of the patterns of gene flow observed in the "best" trees and the ability of the sequence data to discriminate between alternate hypotheses of gene flow. To test for the significance of the levels of population subdivision detected, I compared the observed $s$ with a null distribution of $s$ observed in 500 random trees generated using the "random joining" procedure (Maddison and Slatkin 1991) in MacClade (Maddison and Maddison 1992). Because individuals within social units may not be a random sample from each population as a whole, I repeated this procedure using (1) the subset of sequences that were all distinct from one another and (2) a subset consisting of one randomly chosen individual from each of the 19 social units. This action was taken so as not to inflate $s$ by including essentially "redundant" individuals, that is, individuals that may be genetically correlated primarily because of group structure. In testing the significance of the number of geographic transitions on a tree, the cladistic method assumes that the tree used to calculate the observed $s$ is the true tree. To test the power of the sequence data to discriminate between the best topologies and those specifying extended isolation of populations (Neigel and Avise 1986; Slatkin 1989), I used the constraints option in PAUP (version 3.0r, Swofford 1991) to build the most parsimonious trees constrained to assume a topology minimizing $s$ over the tree, that is, in which the mtDNAs of each population were monophyletic. I applied the User Trees option in DNAML in PHYLIP to evaluate the likelihood of such trees relative to that of the best tree using the Kishino and Hasegawa (1989) test. Computational limitations make it difficult to use likelihood tests such as the Kishino-Hasegawa test to compare large samples of trees with the "best" observed tree, although methods of identifying complete sets of trees that are compatible with a given data set are being developed in the context of parsimony (Templeton et al. 1992).

## Results

*The Sequences Compared.*—Using primers 5 and 7 in figure 2, a total of 400 bp were read from each of the 48 babbler mtDNAs (figs. 2, 3). Alignment of the sequences was achieved by assuming 1-bp gaps at position 203 in the 44 *rubeculus* sequences and at positions 198 and 235

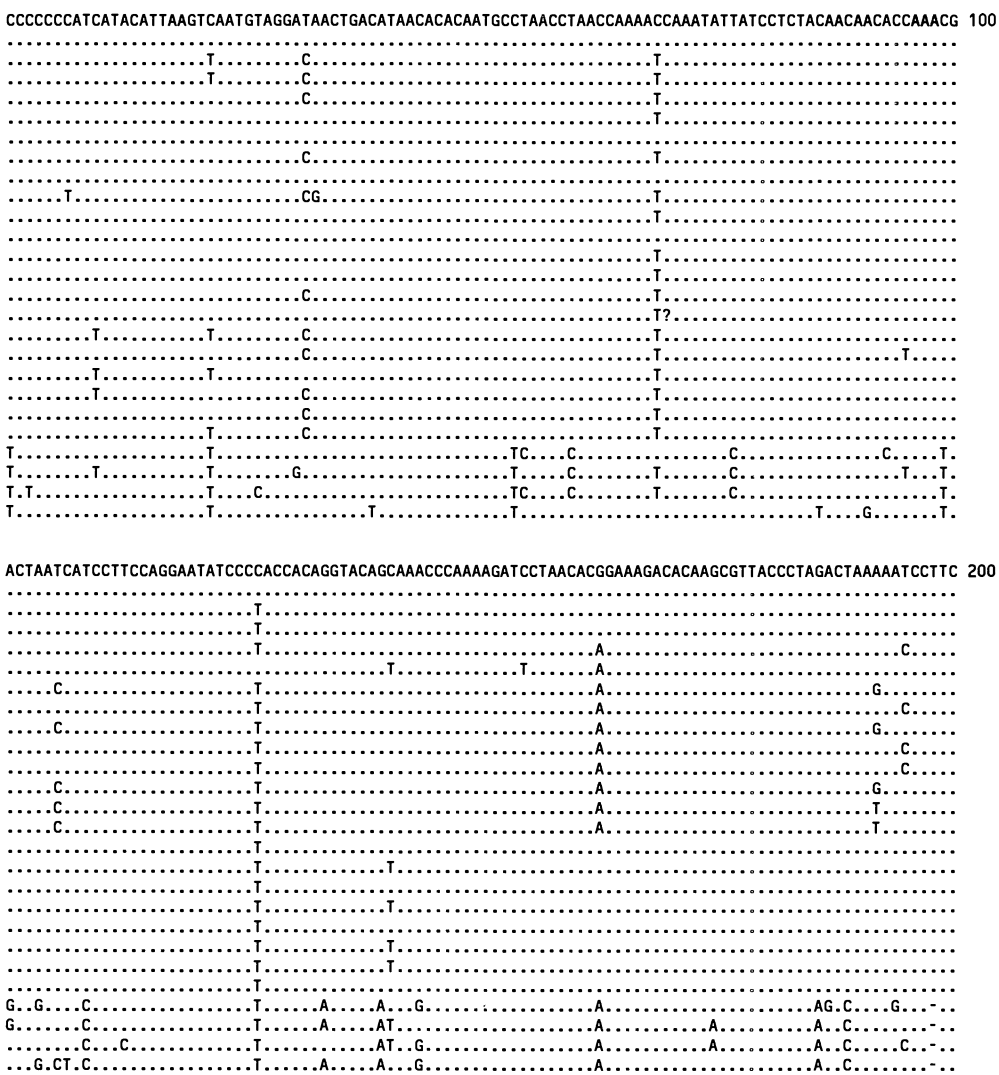
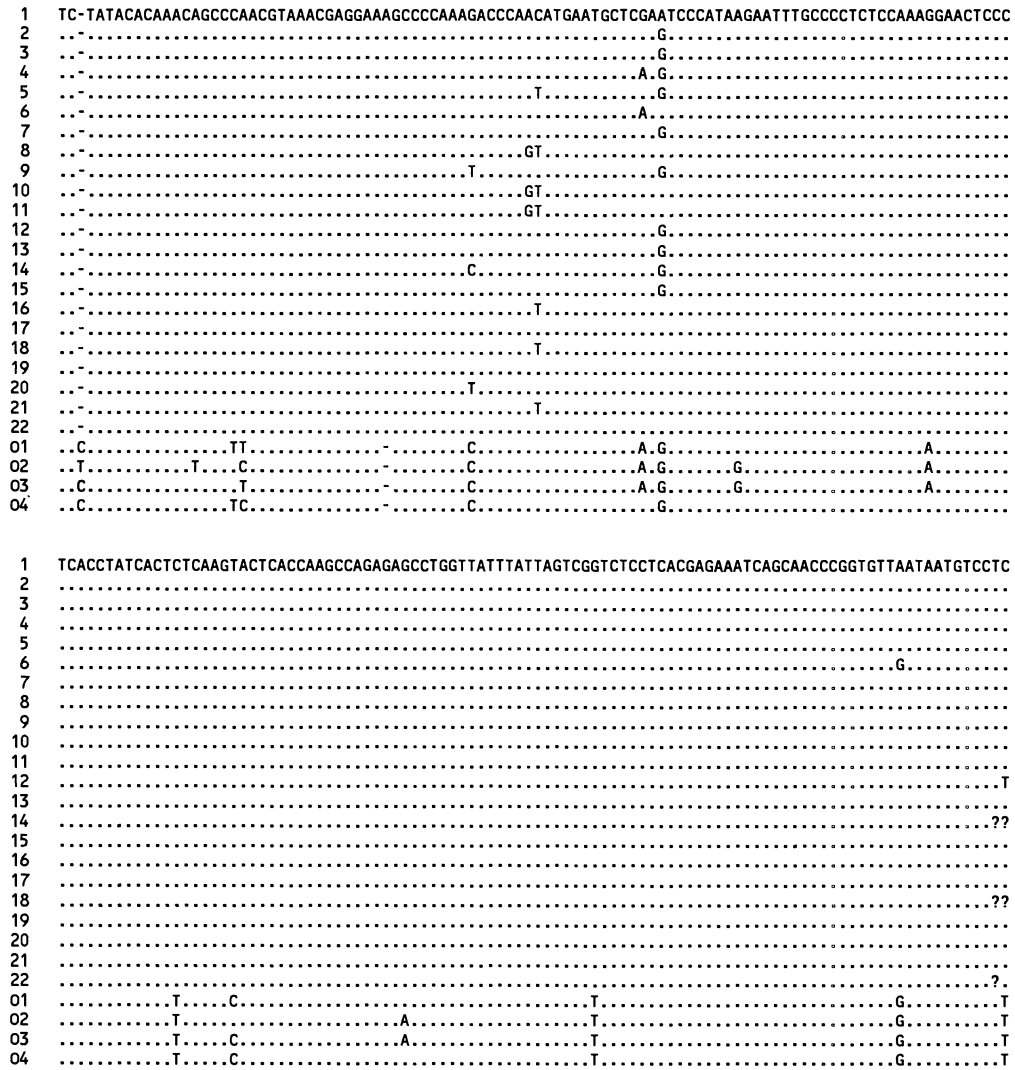


FIG. 3. Twenty-two region I sequences from 44 grey-crowned babblers in the Northern Territory aligned with four outgroup sequences. Individual sequences are designated by a number indicating the mtDNA type (see table 1: 1–6, Darwin; 7–14, Melville Island; 15–22, Cobourg Peninsula). The four *Pomatostomus temporalis*

in the four outgroup *temporalis* sequences (fig. 3). A total of 21 sites were variable among the *rubeculus* sequences, 15 of which were "phylogenetically informative" from a strict parsimony perspective. There were 22 distinct sequences ("types" or "haplotypes") in the three study populations; all four *temporalis* outgroups were distinct from *rubeculus* and from one another. As expected for bird mtDNA (Kocher et al. 1989; Edwards et al. 1991), the base composition of the sequences is significantly biased, with a deficiency of guanines (G, mean = 12.6%) and thymines (T, 19.8%), and an excess of adenines (A, 36.2%) and cytosines (C, 31.5%). Only 2 of the 21 variable sites displayed polymorphisms involving transversions (all types of substitutions other than C ↔ T or A ↔ G), a pattern also consistent with patterns of base substitution found in other closely related vertebrate mtDNAs (W. M. Brown et al. 1982; Aquadro and Greenberg 1983; Wilson et al. 1985; Kocher et al. 1989).

*Neutrality, Population Variability, and Inferences from Phylogenetic Trees.*—Table 2 summarizes the sequence variation and presents tests




*temporalis* individuals used as outgroups are indicated by O1 (Morehead, Western Province, Papua New Guinea), O2 (Hughenden, Queensland), O3 (Parkes, New South Wales), and O4 (Coen, Queensland). Gaps were inserted at bases 198, 203, and 235 to improve alignment.

of neutrality within and among each of the three populations. Tajima (1989) suggested that the difference in the estimate of $\Theta = 4N\mu$ derived from the number of segregating (polymorphic) sites ($S$) and the average number of nucleotide differences ($\hat{k}$) could be used to detect deviations from neutral evolution. (Here, as below, $N$ is the effective population size and $\mu$ the substitution rate per genome per generation.) His $D$ statistic evaluates differences in these two estimates, with a large absolute value indicating a deviation from neutrality. The sequence variability within each of the three populations and in the pooled sample was entirely consistent with neutrality (table 2).

Sequences in the Melville Island population exhibited the highest mean divergence. No mtDNA types were found in more than one population (table 1). The 22 distinct sequences ("types") were analyzed using the DNAML program and the matrix of nucleotide divergence estimates (table 3) was analyzed with three methods (fig. 4). Estimates of sequence divergence using the gamma approach were generally higher than those using the maximum-likelihood ap-

TABLE 2. Summary of sequence variation and tests of neutrality within and among three populations of *Pomatostomus temporalis rubeculus*.

| Population | $N$ | Number of mtDNA types | Maximum (percent) divergence* | Diversity† | Tests of neutrality‡ | | |
|---|---|---|---|---|---|---|---|
| | | | | | Number of variable sites ($S$) | Mean percent divergence (mean number of differences, $\hat{k}$) | $D$ statistic |
| Darwin | 15 | 6 | 2.32 | 0.71 | 12 | 0.88 (3.52) | −0.18 (NS) |
| Melville Island | 15 | 8 | 2.85 | 0.87 | 12 | 1.24 (4.96) | 1.34 (NS) |
| Cobourg Peninsula | 14 | 8 | 1.53 | 0.87 | 8 | 0.66 (2.64) | 0.09 (NS) |
| All places | 44 | 22 | 2.85 | 0.94 | 21 | 0.94 (3.76) | −0.72 (NS) |

* Calculated using maximum-likelihood divergences and assuming a 20:1 transition/transversion ratio.
† Calculated according to the equation $h = (1 - \Sigma x_i^2)n/(n - 1)$, where $x_i$ is the frequency of the $i$th mtDNA type (Nei 1987).
‡ Calculated according to Tajima (1989). None of the values for $D$ can reject an hypothesis of neutrality (NS). See Results.

proach (table 3). Distance trees based on the gamma distances were highly concordant with analyses based on maximum-likelihood distances (not shown). All trees show that mtDNAs from Melville Island fall into two distinct clusters, one of which (types 7, 9, 12–14) consists of a lineage or lineages near the base of the entire clade, and the other of which (types 8, 10, 11) is more closely related to the lineages from other populations. In none of the trees do the mtDNAs from any population form a monophyletic group.

*Gene Flow or Retention of Ancestral Mitochondrial Lineages?*—In principle, the pattern of population para- and polyphyly observed in figure 4 could be caused by either ongoing gene flow (Slatkin and Maddison 1989, 1990) or retention of ancestral alleles in contemporary populations (Takahata 1989). To help distinguish between these two possibilities, I counted the total inferred number of between-population coalescent events ($s$) for each tree in figure 4, which was either 4, 5, or 6, for the Kitsch, Fitch-Margoliash and neighbor-joining, and maximum-likelihood trees, respectively (mean = 5; for trees based on gamma distances, $s$ increased to 5 for the Kitsch tree and decreased to 4 for the neighbor-joining tree). Figure 8 of Slatkin and Maddison (1989) shows a simulation of the relationship between the expectation of $s$ ($\bar{s}$) and $t/N_e$, the ratio of the time since divergence (in generations) of two populations and the effective population size for mitochondrial genes, assuming divergence caused solely by random drift (lineage extinction) with no migration. As expected under such a model, $s$ will be high immediately after separation of the populations (low $t/N_e$), reflecting retention of mitochondrial lineages, but is expected to decrease as the time because divergence increases relative to $N_e$, reflecting the approach to fixation (monophyly of mtDNAs in each population).

Because Slatkin and Maddison (1989) performed simulations of $s$ for two populations, I needed to scale the $s$ value observed among the three study populations (i.e., 5) by a factor $2/r$ (where $r$ is the number of populations, or 3) to compute the $s$ value expected under a similar amount of gene flow or drift for two populations ($s^*$; Slatkin and Maddison 1989); this yields $s^* \approx 3.3$. For $s^* \approx 3.3$, $t/N_e \approx 0.25$ (Fig. 8 in Slatkin and Maddison 1989). First-year breeders among babblers are on average about 3 yr old (Brown 1987), which implies approximately 2700–3300 generations ($t$) in the 8000–10,000 yr since the assumed separation of the populations by water barriers. Thus, an $N_e$ of $t/0.25 = 10{,}800$–$13{,}200$ would be required to observe 3.3 between-population coalescent events in contemporary populations after a divergence time of this length. Because this seems large for a long-term $N_e$ in a passerine bird (Barrowclough and Shields 1984),

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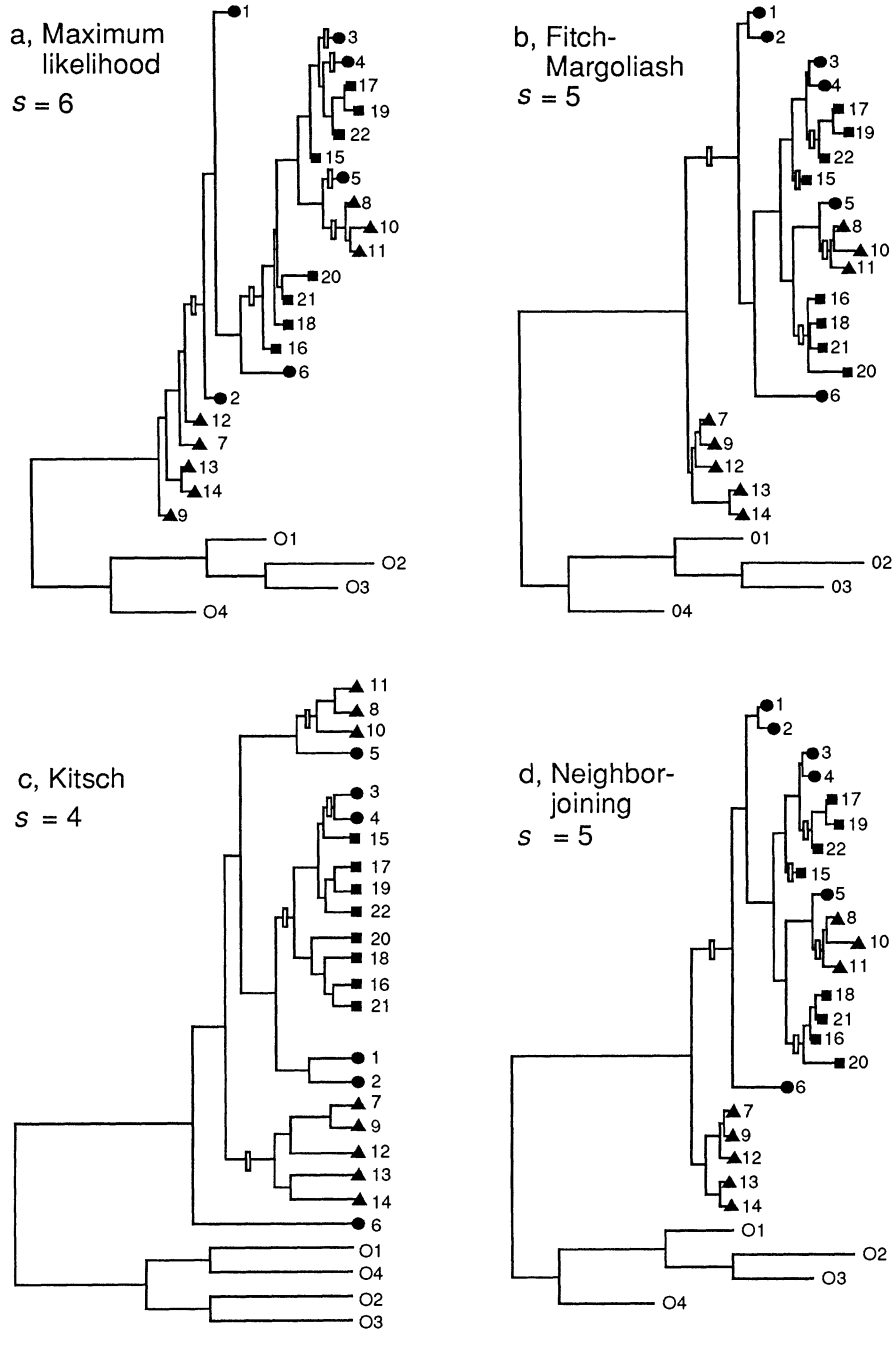
FIG. 4. Four phylogenetic trees built from the sequence data in figure 3. (a) Maximum-likelihood, using a 20:1 transition/transversion ratio; (b) Fitch-Margoliash; (c) Kitsch; (d) Neighbor-joining. The distance trees (b–d) are based on a matrix of maximum-likelihood divergence estimates between each of the 26 distinct sequences (table 3). In each tree, sequences are designated by the type numbers (1–22) indicated in table 1 and figure 3. Black circles, triangles, and squares represent individuals from the Darwin, Melville Island, and Cobourg Peninsula




populations, respectively (fig. 1). The number of inferred between-population coalescent events, *s*, is indicated for each tree, and inferred migration events are denoted by hollow bars on each tree. The particular reconstructions of migration events *delay* character changes as one moves up the tree (DELTRAN option; Maddison and Maddison 1992), and other equally parsimonious reconstructions are possible for each tree. The trees were rooted using the four sequences from the *temporalis* subspecies as outgroups (O1–4; see legend, fig. 3). Branch lengths are approximate.

TABLE 3. Estimates of nucleotide sequence divergence among 26 region I sequences. Above the diagonal are estimated numbers of substitutions per site assuming a gamma distribution of rates with $\alpha = 0.5$ (Jin and Nei 1990). Below the diagonal are estimated numbers of substitutions per site using the maximum-likelihood method and a transition/transversion ratio of 20:1 (Felsenstein 1991). mtDNA type designations are those listed in table 1.

| mt-DNA type | Darwin | | | | | | Melville Island | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 |
| 1 | — | 0.0030 | 0.0130 | 0.0160 | 0.0190 | 0.0160 | 0.0130 | 0.0190 | 0.0160 | 0.0240 | 0.0160 | 0.0160 | 0.0160 | 0.0180 |
| 2 | 0.0025 | — | 0.0100 | 0.0130 | 0.0160 | 0.0190 | 0.0100 | 0.0210 | 0.0130 | 0.0270 | 0.0190 | 0.0130 | 0.0130 | 0.0160 |
| 3 | 0.0127 | 0.0101 | — | 0.0030 | 0.0100 | 0.0240 | 0.0160 | 0.0160 | 0.0180 | 0.0240 | 0.0160 | 0.0160 | 0.0160 | 0.0180 |
| 4 | 0.0153 | 0.0127 | 0.0025 | — | 0.0130 | 0.0210 | 0.0190 | 0.0190 | 0.0210 | 0.0270 | 0.0190 | 0.0130 | 0.0130 | 0.0160 |
| 5 | 0.0179 | 0.0153 | 0.0101 | 0.0127 | — | 0.0240 | 0.0160 | 0.0050 | 0.0180 | 0.0100 | 0.0080 | 0.0190 | 0.0130 | 0.0160 |
| 6 | 0.0153 | 0.0179 | 0.0231 | 0.0205 | 0.0232 | — | 0.0240 | 0.0240 | 0.0270 | 0.0300 | 0.0210 | 0.0270 | 0.0210 | 0.0240 |
| 7 | 0.0127 | 0.0101 | 0.0153 | 0.0179 | 0.0153 | 0.0231 | — | 0.0210 | 0.0030 | 0.0270 | 0.0190 | 0.0030 | 0.0050 | 0.0080 |
| 8 | 0.0179 | 0.0205 | 0.0153 | 0.0179 | 0.0051 | 0.0232 | 0.0205 | — | 0.0240 | 0.0050 | 0.0030 | 0.0240 | 0.0180 | 0.0210 |
| 9 | 0.0154 | 0.0128 | 0.0179 | 0.0206 | 0.0180 | 0.0259 | 0.0025 | 0.0232 | — | 0.0300 | 0.0210 | 0.0050 | 0.0080 | 0.0080 |
| 10 | 0.0231 | 0.0257 | 0.0204 | 0.0231 | 0.0101 | 0.0284 | 0.0257 | 0.0050 | 0.0285 | — | 0.0080 | 0.0300 | 0.0240 | 0.0270 |
| 11 | 0.0153 | 0.0179 | 0.0179 | 0.0205 | 0.0076 | 0.0205 | 0.0179 | 0.0025 | 0.0206 | 0.0076 | — | 0.0210 | 0.0160 | 0.0180 |
| 12 | 0.0153 | 0.0127 | 0.0178 | 0.0205 | 0.0179 | 0.0258 | 0.0025 | 0.0231 | 0.0051 | 0.0284 | 0.0205 | — | 0.0080 | 0.0080 |
| 13 | 0.0153 | 0.0127 | 0.0127 | 0.0153 | 0.0127 | 0.0206 | 0.0051 | 0.0179 | 0.0076 | 0.0231 | 0.0153 | 0.0076 | — | 0.0030 |
| 14 | 0.0181 | 0.0155 | 0.0155 | 0.0181 | 0.0155 | 0.0234 | 0.0077 | 0.0208 | 0.0077 | 0.0260 | 0.0181 | 0.0077 | 0.0025 | — |
| 15 | 0.0101 | 0.0076 | 0.0025 | 0.0050 | 0.0076 | 0.0205 | 0.0127 | 0.0127 | 0.0153 | 0.0178 | 0.0153 | 0.0153 | 0.0102 | 0.0129 |
| 16 | 0.0102 | 0.0127 | 0.0127 | 0.0153 | 0.0127 | 0.0153 | 0.0179 | 0.0127 | 0.0206 | 0.0179 | 0.0102 | 0.0205 | 0.0153 | 0.0181 |
| 17 | 0.0127 | 0.0153 | 0.0050 | 0.0076 | 0.0153 | 0.0231 | 0.0205 | 0.0153 | 0.0232 | 0.0205 | 0.0179 | 0.0231 | 0.0179 | 0.0207 |
| 18 | 0.0154 | 0.0180 | 0.0128 | 0.0154 | 0.0128 | 0.0206 | 0.0232 | 0.0128 | 0.0260 | 0.0179 | 0.0154 | 0.0232 | 0.0206 | 0.0234 |
| 19 | 0.0101 | 0.0127 | 0.0076 | 0.0101 | 0.0179 | 0.0205 | 0.0179 | 0.0179 | 0.0206 | 0.0231 | 0.0153 | 0.0205 | 0.0153 | 0.0181 |
| 20 | 0.0154 | 0.0179 | 0.0127 | 0.0153 | 0.0180 | 0.0206 | 0.0232 | 0.0180 | 0.0205 | 0.0232 | 0.0206 | 0.0258 | 0.0206 | 0.0207 |
| 21 | 0.0127 | 0.0153 | 0.0101 | 0.0127 | 0.0101 | 0.0179 | 0.0205 | 0.0101 | 0.0232 | 0.0153 | 0.0127 | 0.0231 | 0.0179 | 0.0207 |
| 22 | 0.0102 | 0.0127 | 0.0025 | 0.0050 | 0.0127 | 0.0205 | 0.0179 | 0.0127 | 0.0206 | 0.0179 | 0.0153 | 0.0205 | 0.0153 | 0.0181 |
| 01 | 0.0864 | 0.0832 | 0.0831 | 0.0802 | 0.0894 | 0.0864 | 0.0832 | 0.0956 | 0.0830 | 0.1016 | 0.0925 | 0.0802 | 0.0865 | 0.0808 |
| 02 | 0.0740 | 0.0709 | 0.0708 | 0.0739 | 0.0769 | 0.0799 | 0.0651 | 0.0829 | 0.0648 | 0.0887 | 0.0798 | 0.0621 | 0.0681 | 0.0626 |
| 03 | 0.0898 | 0.0865 | 0.0803 | 0.0775 | 0.0866 | 0.0775 | 0.0865 | 0.0928 | 0.0863 | 0.0988 | 0.0896 | 0.0834 | 0.0837 | 0.0780 |
| 04 | 0.0931 | 0.0898 | 0.0836 | 0.0807 | 0.0838 | 0.0807 | 0.0898 | 0.0899 | 0.0895 | 0.0959 | 0.0868 | 0.0867 | 0.0870 | 0.0812 |

TABLE 3. Continued.

| Cobourg Peninsula | | | | | | | | *P. t. temporalis* | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 15 | 16 | 17 | 18 | 19 | 20 | 21 | 22 | 01 | 02 | 03 | 04 |
| 0.0100 | 0.0100 | 0.0130 | 0.0160 | 0.0100 | 0.0160 | 0.0130 | 0.0100 | 0.0980 | 0.0830 | 0.1020 | 0.1070 |
| 0.0080 | 0.0130 | 0.0160 | 0.0190 | 0.0100 | 0.0160 | 0.0130 | 0.0100 | 0.0980 | 0.0830 | 0.1020 | 0.1070 |
| 0.0100 | 0.0100 | 0.0130 | 0.0160 | 0.0100 | 0.0160 | 0.0130 | 0.0100 | 0.0980 | 0.0830 | 0.1020 | 0.1070 |
| 0.0080 | 0.0130 | 0.0160 | 0.0190 | 0.0100 | 0.0160 | 0.0130 | 0.0050 | 0.0900 | 0.0830 | 0.0860 | 0.0900 |
| 0.0080 | 0.0130 | 0.0160 | 0.0130 | 0.0190 | 0.0180 | 0.0100 | 0.0130 | 0.1020 | 0.0870 | 0.0980 | 0.0940 |
| 0.0210 | 0.0160 | 0.0240 | 0.0210 | 0.0210 | 0.0210 | 0.0190 | 0.0210 | 0.0980 | 0.0900 | 0.0860 | 0.0900 |
| 0.0130 | 0.0190 | 0.0210 | 0.0240 | 0.0100 | 0.0160 | 0.0130 | 0.0100 | 0.0980 | 0.0830 | 0.1020 | 0.1070 |
| 0.0130 | 0.0130 | 0.0160 | 0.0130 | 0.0190 | 0.0180 | 0.0100 | 0.0130 | 0.1100 | 0.0940 | 0.1060 | 0.1030 |
| 0.0160 | 0.0210 | 0.0240 | 0.0270 | 0.0210 | 0.0210 | 0.0240 | 0.0210 | 0.0940 | 0.0720 | 0.0980 | 0.1030 |
| 0.0190 | 0.0190 | 0.0210 | 0.0190 | 0.0240 | 0.0240 | 0.0160 | 0.0190 | 0.1190 | 0.1030 | 0.1140 | 0.1110 |
| 0.0160 | 0.0100 | 0.0190 | 0.0160 | 0.0160 | 0.0210 | 0.0130 | 0.0160 | 0.1060 | 0.0900 | 0.1020 | 0.0980 |
| 0.0160 | 0.0210 | 0.0240 | 0.0240 | 0.0210 | 0.0270 | 0.0240 | 0.0210 | 0.0900 | 0.0680 | 0.0940 | 0.0980 |
| 0.0100 | 0.0160 | 0.0180 | 0.0210 | 0.0160 | 0.0210 | 0.0180 | 0.0160 | 0.0970 | 0.0750 | 0.0930 | 0.0980 |
| 0.0130 | 0.0180 | 0.0210 | 0.0240 | 0.0180 | 0.0210 | 0.0210 | 0.0180 | 0.0900 | 0.0680 | 0.0870 | 0.0910 |
| — | 0.0100 | 0.0080 | 0.0100 | 0.0100 | 0.0100 | 0.0080 | 0.0050 | 0.0980 | 0.0830 | 0.0940 | 0.0980 |
| 0.0101 | — | 0.0130 | 0.0050 | 0.0100 | 0.0100 | 0.0030 | 0.0100 | 0.1060 | 0.0910 | 0.0940 | 0.0990 |
| 0.0076 | 0.0127 | — | 0.0130 | 0.0030 | 0.0080 | 0.0100 | 0.0030 | 0.1020 | 0.0870 | 0.0900 | 0.1030 |
| 0.0102 | 0.0051 | 0.0128 | — | 0.0160 | 0.0100 | 0.0030 | 0.0100 | 0.1110 | 0.0950 | 0.0900 | 0.1030 |
| 0.0101 | 0.0101 | 0.0025 | 0.0153 | — | 0.0100 | 0.0130 | 0.0050 | 0.0980 | 0.0830 | 0.0860 | 0.0980 |
| 0.0102 | 0.0102 | 0.0076 | 0.0102 | 0.0102 | — | 0.0080 | 0.0101 | 0.1110 | 0.0950 | 0.0900 | 0.1030 |
| 0.0076 | 0.0025 | 0.0101 | 0.0025 | 0.0127 | 0.0076 | — | 0.0080 | 0.1100 | 0.0940 | 0.0980 | 0.1030 |
| 0.0050 | 0.0102 | 0.0025 | 0.0102 | 0.0050 | 0.0102 | 0.0076 | — | 0.0980 | 0.0830 | 0.0940 | 0.0980 |
| 0.0862 | 0.0926 | 0.0893 | 0.0961 | 0.0862 | 0.0952 | 0.0955 | 0.0863 | — | 0.0420 | 0.0550 | 0.0450 |
| 0.0738 | 0.0799 | 0.0768 | 0.0833 | 0.0738 | 0.0825 | 0.0828 | 0.0739 | 0.0392 | — | 0.0720 | 0.0690 |
| 0.0834 | 0.0836 | 0.0804 | 0.0809 | 0.0774 | 0.0802 | 0.0865 | 0.0835 | 0.0505 | 0.0645 | — | 0.0420 |
| 0.0867 | 0.0869 | 0.0898 | 0.0904 | 0.0867 | 0.0895 | 0.0898 | 0.0868 | 0.0423 | 0.0618 | 0.0396 | — |

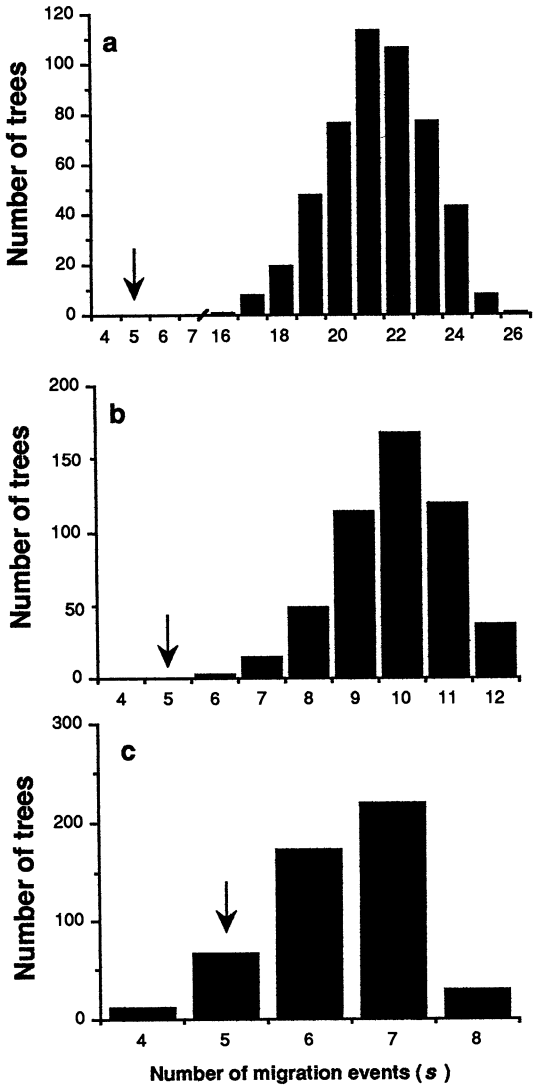



FIG. 5. Null expectations of the number of inferred between-population coalescent events, *s*, along trees containing various subsets of the 44 individuals. Distributions expected considering (a) all 44 individuals from the three populations, (b) the 22 distinct sequences among the three populations, (c) one individual from each of the 19 social groups in the sample. Each distribution was generated using 500 random trees built on the random-joining procedure, in which pairs of taxa join (coalesce) with equal probability (Maddison and Slatkin 1991). The arrows in each panel indicate the observed value of *s* (= 5) averaged over the four trees in figure 4.

particularly a social one, I conclude that most or all of the observed between-population coalescent events are in fact caused by recent migration.

The *s* values derived from the trees suggest

TABLE 4. Estimates of average levels of gene flow (*Nm*) and $F_{st}$ for three populations of babblers in the Northern Territory. In (a) the minimum number of inferred migration events (*s*) was calculated for each tree in figure 4 and converted to estimates of *Nm* as described (Slatkin and Maddison 1989); $F_{st}$ here is calculated as $1/(1 + 2Nm)$. In (b), values of $F_{st}$ were calculated from the three types of sequence divergence estimates in the table according to equation 2 (Materials and Methods) and Lynch and Crease (1990); values of *Nm* values were extracted from $F_{st}$ assuming the above relationship.

| Method | *Nm* | Confidence interval for *Nm*† | $F_{st}$ |
|---|---|---|---|
| a) Migration events‡ | | | |
| DNAML | 1.2 | 0.8–2.1 | 0.29 |
| Fitch-Margoliash | 0.9 | 0.4–1.6 | 0.36 |
| Kitsch | 0.6 | 0.3–1.1 | 0.45 |
| Neighbor-joining | 0.9 | 0.4–1.6 | 0.36 |
| b) Coalescence times | | | |
| ML distances | 0.4 | NA§ | 0.55 |
| Gamma distances | 0.8 | NA | 0.39 |
| Jukes-Cantor‖ | 0.6 | NA | 0.46* |

† Confidence intervals are based on the observed *s* for each tree ± one standard deviation about *s* (Slatkin and Maddison 1989).

‡ Using values for $N = 16$ in table 1 of Slatkin and Maddison (1989).

§ Not applicable.

‖ $F_{st}$ calculated by methods for restriction site data in Lynch and Crease (1990; see Materials and Methods).

* Significant geographic subdivision at the $P < 0.01$ level (Lynch and Crease 1990).

*Nm* values of 0.6–1.2 (mean 0.9) among the three populations (table 4). The conclusion of significant population subdivision is reinforced by comparison of the observed *s* with its null distribution given the number of populations and individuals sampled (fig. 5) and random mating; here complete panmixis can be rejected for all but the case in which I assume independence of only one individual per social group (fig. 5c). Inspection of *s* values between each pair of populations suggests that the Melville Island population exchanges migrants with the Cobourg Peninsula and mainland populations in roughly equal proportions, and that gene flow between the two continental populations may be slightly higher (table 5). If the number of randomly sampled individuals from each population is in fact lower than the actual sample sizes due to genetic similarities within social groups, these estimates of *Nm* would be biased downward from the true values of *Nm* for these populations, suggesting an even higher level of gene flow.

Table 4 shows that estimates of *Nm* using *s* ($\langle Nm_s \rangle$; Hudson et al. 1992) were comparable

TABLE 5. The number of inferred between-population coalescent events, $s$, for different phylogenetic trees (D, Darwin; M, Melville Island; C, Cobourg Peninsula). To calculate $s$ values between pairs of populations, one of the indicated trees (fig. 4) was assumed, all the mtDNA types found in one of the three populations were removed, and $s$ was counted for the remaining two populations. Thus, the sums of the $s$ values in these pairwise comparisons do not add up to the $s$ values observed over the entire tree for all three populations. Assuming sample sizes of 16 per population, $Nm$ values corresponding to an observed $s$ of 1, 2, 3, or 4 are less than 0.1, 0.3, 0.7, and 1.3, respectively (see Slatkin and Maddison 1989, 1990).

| Tree | $s$ for pairs of populations | | |
|---|---|---|---|
| | D and M | D and C | M and C |
| DNAML | 2 | 4 | 2 |
| Fitch-Margoliash | 2 | 3 | 2 |
| Kitsch | 2 | 2 | 1 |
| Neighbor-joining | 2 | 3 | 2 |

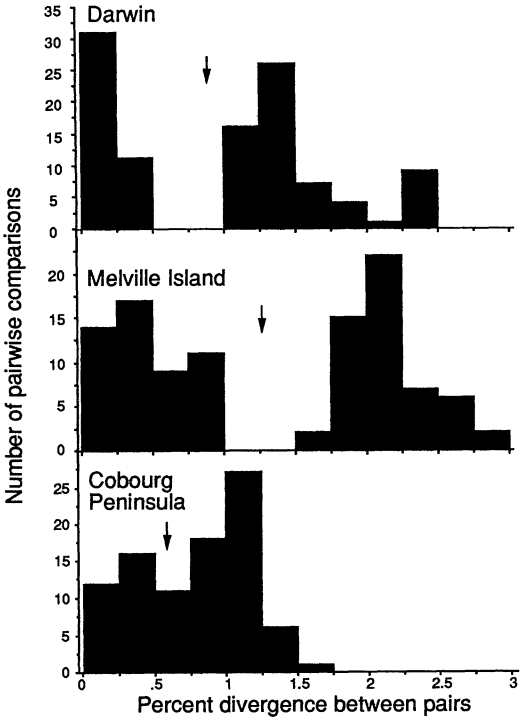



FIG. 6. Distributions of all pairwise divergence estimates (coalescence times) within each of the three populations. The histograms plot all pairwise comparisons of sequence divergences, regardless of whether some sequences in a population were identical or not. Arrows indicate mean divergences over all comparisons.

across tree-making methods; the average $Nm_s$ is only slightly higher than the average $Nm$ estimated using the divergences (coalescence times) of alleles ($\langle Nm_F \rangle$; Hudson et al. 1992). Because $Nm$ among these babbler populations is generally small, $Nm_F$ is likely to provide a better estimate of the true $Nm$ than $Nm_s$ (Hudson et al. 1992).

*Patterns of Allelic Divergence.*—The mean and distribution of pairwise divergences (coalescence times) of sampled alleles also provide information about the evolutionary history of populations (Tajima 1983; Slatkin 1987; Strobeck 1987; Avise et al. 1988), but the lack of statistical independence of pairwise comparisons and the large stochastic variance associated with the mean make the distributions difficult to interpret (Tajima 1983; Slatkin and Hudson 1991). Figure 6 shows the distribution of pairwise sequence divergences within each of the three populations. Both the Darwin and Melville populations show distinctly bimodal distributions. This pattern is reflected in the phylogenetic analyses (fig. 4), which show at least two divergent lineages for each of these populations; on Melville Island these lineages differ by about 2% (about 6–11 sites). The distribution in the Cobourg population is more normal (fig. 6).

Strobeck (1987) devised a test for detecting gene flow by comparing the average number of site differences in sequences from a single subpopulation, $\Theta$, which is equivalent to $4N\mu$ regardless of migration rates with other subpopulations, with the number of distinct sequences from the same subpopulation, $\Theta'$, which is increased by alleles immigrating from other subpopulations (see also Slatkin 1987). If the average number of nucleotide differences in a population is high relative to the number of distinct sequences, then migration from other subpopulations can be inferred. For the Melville Island population ($N = 15$), the number of distinct sequences is 8, whereas the average divergence among sequences is 1.2%, corresponding to an average difference of about 5 (table 2); these values suggest that the sequences are not divergent enough to invoke migration from other subpopulations (Strobeck 1987; table 1). However, the evidence for gene flow contained in analyses of mtDNA diversity in multiple populations probably exceeds that contained in a single population (Strobeck 1987).

*Statistical Significance of the Trees.*—The phylogenetic hypotheses produced by the various methods were tested against one another and against phylogenies expected under different hypotheses of gene flow. For example, if no gene flow were occurring among the populations, the mtDNAs from each population would form

monophyletic groups, assuming sufficient time for complete sorting of lineages; with extreme inbreeding and temporal stability of social units, the mtDNAs within all populations and social units would be monophyletic. The trees produced by the various methods were all statistically compatible with one another (table 6). However, the data were consistently able to reject evolutionary hypotheses in which both populations and social units were monophyletic (trees 11–16, table 6). Yet the data could not distinguish between the "best" trees and those specifying the most parsimonious arrangements assuming no gene flow (trees 5–9, table 6). Because the competing hypotheses assuming no gene flow were the most parsimonious hypotheses, they represent stringent competing hypotheses, and the likelihoods of these trees are nearly at the rejection level. For example, given a tree specifying no migration, movement of one individual (#485) next to another member of its social unit (#484) was enough to produce a significantly poorer fit to the data than the best trees (tree 10, table 6).

## Discussion

### *Genealogical Analysis and Inferences of Population History*

The analysis of population processes in the context of phylogenetic trees has important advantages over traditional, frequency-based population genetic models and will be particularly important for interpretation of variability in rapidly evolving mtDNA segments (Avise 1989; Slatkin and Maddison 1989; Takahata 1989; Hudson 1991). For example, I found no sharing of mtDNA sequences between localities, a situation implying that the mutation rate in this segment of the control region is high relative to the rates of migration. Taken alone, however, such a pattern is consistent both with no gene flow and extensive gene flow among populations; thus I could detect exchanges of individuals between populations only using inferences drawn from a phylogenetic tree. Recently Brown and Chapman (1991) also used the genealogical approach to infer gene flow in killifish, but the presence of several mtDNA types common to two or more populations provided at least some direct evidence of the movement (or persistence) of genes across populations. The mutation rate for the mtDNA restriction sites employed by these authors was low enough to ensure that estimates of $F_{st}$ based on traditional methods would not be greatly compromised. However, the rate of evolution in region I of the control region has been estimated to be over ten times the average rate detected across the entire mitochondrial genome using the restriction approach (Vigilant et al. 1989; Kocher and Wilson 1991), and over 100 times faster than that of a nuclear pseudogene (Hasegawa and Horai 1991). Such high mutation rates necessitate using models that take into account mutation as well as migration as a significant evolutionary force (Slatkin and Maddison 1989; Lynch and Crease 1990; Slatkin 1991).

However, a purely genealogical approach to inferences about gene flow has some practical drawbacks, the most important of which is the difficulty obtaining statistically significant phylogenies of closely related molecular lineages. Although the number of coalescent events inferred from the "best" trees was higher than that expected given no migration since the separation of Melville Island from the mainland, the sequence data themselves were insufficient to reject hypotheses of complete restriction of gene flow among populations ($Nm = 0$; table 6). The limited resolution achievable among such closely related populations is reflected in the extremely short branches in the trees (fig. 4), although higher numbers of variable sites and levels of sequence divergence have been detected in a continent-wide study of this species (Edwards 1993). Many gene genealogies among closely related lineages have not been tested statistically against alternative hypotheses, and it may be that many of the branches on which migration events are inferred will be short, particularly if there is high gene flow. In the maximum-likelihood tree, for example, the mean length of branches on which migration events are inferred is longer than all other branches in the tree, but the difference is marginal depending on the assumptions used (e.g., ACCTRAN or DELTRAN) to reconstruct the migration events (ACCTRAN: $t_{40} = 2.39$, $P < 0.05$; DELTRAN: $t_{40} = 1.47$, $P > 0.10$; excluding the branch to the outgroup). Ultimately, the ability to distinguish "ongoing" gene flow from recent cessation of gene flow will depend on the resolution of the genetic data. Fortunately, the situations in which genealogical methods have their greatest power (i.e., low to moderate levels of gene flow; Slatkin and Maddison 1989) are also those in which significant resolution of major branching events can best be achieved by sequence data.

TABLE 6. Evaluation of competing phylogenetic hypotheses of gene flow using the Kishino and Hasegawa (1989) test (ln $L$ = natural log of the likelihood). Trees 5–16 represent a randomly chosen subset of equally parsimonious trees fulfilling the particular constraint described under Pattern; see Materials and Methods and Results.

| Tree | Method of construction | Pattern | Hypothesis | ln $L$ | Difference in ln $L$ | SD in ln $L$ | Rejected? ($P < 0.05$) |
|---|---|---|---|---|---|---|---|
| | | | a) Trees in figure 4 | | | | |
| 1 | Maximum-likelihood | Observed trees | Observed level of gene flow | −1060.4 | ← best | | |
| 2 | Fitch-Margoliash | | | −1065.5 | −5.1 | 9.2 | No |
| 3 | Kitsch | | | −1064.1 | −3.7 | 10.1 | No |
| 4 | Neighbor-joining | | | −1071.9 | −11.5 | 17.2 | No |
| | | | b) Alternative hypotheses of gene flow | | | | |
| 5 | Parsimony | Populations monophyletic | No gene flow between populations | −1081.4 | −21.0 | 14.9 | No |
| 6 | Parsimony | | | −1081.4 | −21.0 | 14.9 | No |
| 7 | Parsimony | | | −1083.1 | −22.7 | 16.8 | No |
| 8 | Parsimony | | | −1080.1 | −19.7 | 16.9 | No |
| 9 | Parsimony | | | −1087.1 | −26.7 | 17.4 | No |
| 10 | Parsimony | Populations monophyletic with type 8 next to type 9 | | −1118.0 | −57.6 | 20.4 | Yes |
| 11 | Parsimony | Populations monophyletic, social units monophyletic | No gene flow, no dispersal between social units | −1150.5 | −90.1 | 26.1 | Yes |
| 12 | Parsimony | | | −1152.2 | −91.8 | 26.6 | Yes |
| 13 | Parsimony | | | −1153.5 | −93.1 | 26.5 | Yes |
| 14 | Parsimony | | | −1155.6 | −95.2 | 26.4 | Yes |
| 15 | Parsimony | | | −1162.7 | −102.4 | 26.7 | Yes |
| 16 | Parsimony | | | −1153.7 | −93.3 | 27.4 | Yes |

### *Implications of Genetic Diversity in Melville Island Babblers*

The phylogenies inferred from the sequences and the times of population divergence suggested by geological data imply that under reasonable assumptions about effective population sizes the observed number of between-population coalescent events was too high to be explained solely by retention of mitochondrial lineages since separation of the populations; thus ongoing gene flow is probably occurring. The variation in region I detected within and among the three populations, separated from one another by only 150 km, suggests that this part of the control region will be useful in examining the evolutionary dynamics of closely related populations of birds.

Together the phylogenetic analyses, pairwise $s$ values, and the bimodal distribution of sequence divergences in the Melville Island population (fig. 6) are consistent with Melville exchanging migrants with both the Cobourg and the Darwin populations (table 5), although at a level low enough to permit the buildup of significant differentiation across the water barriers separating them. Although bimodal distributions such as those in the top two panels of figure 6 are not expected for randomly mating populations of constant size (Slatkin and Hudson 1991), the phylogenies suggest that migration could also be a factor explaining the presence of divergent sequences. In contrast to studies reporting reduced genetic diversity in island populations (e.g., Johnson and Martin 1988; Ashley and Wills 1987), the mtDNA diversity on Melville Island is comparable to that on the Cobourg Peninsula (table 2), implying that the water barrier has not significantly influenced the number of survivng mitochondrial lineages here.

In addition to the potential barrier to gene flow imposed by the van Diemen Gulf (fig. 1), grey-crowned babblers are thought to be "sedentary," with little exchange of individuals between populations. In the Neotropics, water barriers older and narrower than the one involved here are thought to limit gene flow and contribute to the differentiation of avian species (Capparella 1988), and recent surveys of birds (Johnson and Martin 1988; Degnan and Moritz 1992) and rodents (Ashley and Wills 1987) suggest reduced gene flow between island and mainland taxa (but see Zink et al. 1987). My results suggest that babblers are either blown over the Gulf by the frequent

seasonal storms of the area, or are actively migrating across, perhaps partly via the Vernon Islands between Darwin and Melville Island. Such movements have in some cases resulted in the presence of divergent mitochondrial lineages within single social units (e.g., lineages 8 and 9 in social unit 44; table 1, fig. 4), consistent with results from restriction mapping (Edwards and Wilson 1990). Such migration events have important consequences for levels of relatedness within social units, the dynamics of kin selection, and the divergence of babbler populations (Edwards 1993).

Grey-crowned babblers are also found on another land-bridge island in the Northern Territory, Groote Eylandt (fig. 1) and have been documented on islands off the tip of Cape York, Queensland (Draffan et al. 1983). The analyses suggest that passive or active migration to these islands, as opposed to population persistence after rising post-Pleistocene water barriers, is a plausible explanation for the occurrence of babblers on such islands. If true, this finding has implications for theories of species coexistence in island communities. Diamond (1975) suggested that the patterns of species distributions and community structure on land-bridge islands off New Guinea were the result of either interspecific competition or taxon-specific resistance to crossing water barriers. In New Guinea, rufous babblers (*P. isidori*) are found primarily in lowland rainforest, both on the main island and on two of the three land-bridge islands most remote from the main island (Beehler et al. 1986; Diamond 1987). If *P. isidori* possesses colonizing capabilities similar to those implicated for *P. temporalis,* it would be expected to occur on islands closer to New Guinea as well. Thus, the distributional pattern displayed by *P. isidori* is more consistent with the presence of true Pleistocene relicts than is that of *P. temporalis.* Molecular genealogical studies of conspecific populations on New Guinea and satellite islands should provide additional insight into the relative roles of competition, habitat availability, and colonizing ability in shaping contemporary avian communities.

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## Literature Cited

Aquadro, C. F., and B. D. Greenberg. 1983. Human mitochondrial DNA variation and evolution: analysis of nucleotide sequences from seven individuals. Genetics 103:287–312.

Arctander, P. 1988. Comparative studies of avian DNA by restriction fragment length polymorphism analysis: convenient procedures based on blood samples from live birds. Journal of Ornithology 129:205–216.

Ashley, M., and C. Wills. 1987. Analysis of mitochondrial DNA polymorphisms among Channel Island deer mice. Evolution 41:854–863.

Avise, J. C. 1989. Gene trees and organismal histories: a phylogenetic approach to population biology. Evolution 43:1192–1208.

Avise, J. C., J. Arnold, R. M. Ball, E. Bermingham, T. Lamb, J. E. Neigel, C. A. Reeb, and N. C. Saunders. 1987. Intraspecific phylogeography: the mitochondrial DNA bridge between population genetics and systematics. Annual Review of Ecology and Systematics 18:489–522.

Avise, J. C., R. M. Ball, and J. Arnold. 1988. Current versus historical population sizes in vertebrate species with high gene flow: a comparison based on mitochondrial DNA polymorphism and inbreeding theory for neutral mutations. Molecular Biology and Evolution 5:331–344.

Avise, J. C., J. E. Neigel, and J. Arnold. 1984. Demographic influences on mitochondrial DNA lineage survivorship in animal populations. Journal of Molecular Evolution 20:99–105.

Baker, R. J., and A. Moeed. 1987. Rapid genetic differentiation and founder effect in colonizing populations of common mynas. Evolution 41:525–538.

Ball, R. M., Jr., S. Freeman, F. C. James, E. Bermingham, and J. C. Avise. 1988. Phylogeographic population structure of red-winged blackbirds assessed by mitochondrial DNA. Proceedings of the National Academy of Sciences, USA 85:1558–1562.

Barrowclough, G. F. 1983. Biochemical studies of

microevolutionary processes. Pp. 223–261 *in* A. H. Brush and G. A. Clark, Jr., eds. Perspectives in ornithology. Cambridge University Press, Cambridge.

Barrowclough, G. F., and G. F. Shields. 1984. Karyotypic evolution and long-term effective population sizes of birds. Auk 101:99–102.

Beehler, B. M., T. K. Pratt, and D. A. Zimmerman. 1986. Birds of New Guinea. Princeton University Press, Princeton, N.J.

Birt-Friesen, V. L., W. A. Montevecchi, A. J. Gaston, and W. S. Davidson. 1992. Genetic structure of thick-billed murre (*Uria lomvia*) populations examined using direct sequence analysis of amplified DNA. Evolution 46:267–272.

Brown, B. L., and R. W. Chapman. 1991. Gene flow and mitochondrial DNA variation in the killifish, *Fundulus heteroclitus*. Evolution 45:1147–1161.

Brown, J. L. 1987. Helping and communal breeding in birds: ecology and evolution. Princeton University Press, Princeton, N.J.

Brown, J. L., E. R. Brown, and S. D. Brown. 1982. Morphological variation in a population of grey-crowned babblers: correlations with variables affecting social behavior. Behavioral Ecology and Sociobiology 10:281–287.

Brown, J. L., D. D. Dow, E. R. Brown, and S. D. Brown. 1983. Socioecology of the grey-crowned babbler (*Pomatostomus temporalis*): population structure, unit size, and vegetation correlates. Behavioral Ecology and Sociobiology 13:115–124.

Brown, W. M. 1980. Polymorphism in mitochondrial DNA of humans as revealed by restriction endonuclease analysis. Proceedings of the National Academy of Sciences, USA 77:3605–3609.

———. 1985. The mitochondrial genome of animals. Pp. 95–130 *in* R. J. MacIntyre, ed. Molecular evolutionary genetics. Plenum Press, New York.

Brown, W. M., E. M. Prager, A. Wang, and A. C. Wilson. 1982. Mitochondrial DNA sequences of primates: tempo and mode of evolution. Journal of Molecular Evolution 18:225–239.

Busack, S. 1986. Biogeographic analysis of the herpetofauna separated by the formation of the Strait of Gibraltar. National Geographic Research 2:17–36.

Cabot, E. L., and A. T. Beckenbach. 1989. Simultaneous editing of multiple nucleic acid and protein sequences with ESEE. Computer Applications in the Biosciences 5:233–234.

Capparella, A. P. 1988. Genetic variation in neotropical birds: implications for the speciation process. Proceedings of the International Ornithological Congress 19:1658–1664.

Counsilman, J. J., and B. King. 1977. Ageing and sexing the grey-crowned babbler (*Pomatostomus temporalis*). Babbler 1:23–41.

Degnan, S., and C. Moritz. 1992. Mitochondrial DNA variation in Australian silvereyes (*Zosterops lateralis*). Auk 109:800–811.

Deignan, H. G. 1964. Subfamily Timaliinae. Pp. 240–247 *in* E. Mayr and R. Paynter, Jr., eds. Checklist of the birds of the world. Museum of Comparative Zoology, Harvard University, Cambridge, Mass.

Desjardins, P., and R. Morais. 1990. Sequence and gene organization of the chicken mitochondrial genome. A novel gene order in higher vertebrates. Journal of Molecular Biology 212:599–634.

Diamond, J. 1975. Assembly of species communities. Pp. 342–444 *in* T. Case and J. Diamond, eds. Evolution of ecological communities. The Belknap Press, Harvard University, Cambridge, Mass.

———. 1987. Flocks of brown and black New Guinean birds: a bicoloured mixed-species foraging association. Emu 87:201–211.

Di Rienzo, A., and A. C. Wilson. 1991. Branching pattern in the evolutionary tree for human mitochondrial DNA. Proceedings of the National Academy of Sciences, USA 88:1597–1601.

Draffan, R.D.W., S. T. Garnett, and G. J. Malone. 1983. Birds of the Torres Strait: an annotated list and biogeographical analysis. Emu 83:207–234.

Edwards, S. V. 1992. Gene flow and mitochondrial DNA evolution in social babblers (*Pomatostomus*). Ph.D. dissertation. University of California, Berkeley, Calif.

———. 1993. Long-distance gene flow in a cooperative breeder detected in genealogies of mitochondrial DNA sequences. Proceedings of the Royal Society of London, Series B 252:177–185.

Edwards, S. V., and A. C. Wilson. 1990. Phylogenetically informative length polymorphism and sequence variability in mitochondrial DNA of Australian songbirds (*Pomatostomus*). Genetics 126: 695–711.

Edwards, S. V., P. Arctander, and A. C. Wilson. 1991. Mitochondrial resolution of a deep branch in the genealogical tree for perching birds. Proceedings of the Royal Society of London, Series B 243:99–107.

Felsenstein, J. 1981. Evolutionary trees from DNA sequences: a maximum likelihood approach. Journal of Molecular Evolution 17:368–376.

———. 1984. Distance methods for inferring phylogenies: a justification. Evolution 38:16–24.

———. 1991. PHYLIP 3.4 manual. University of California Herbarium, Berkeley, Calif.

Fitch, W. M., and E. Margoliash. 1967. Construction of phylogenetic trees. Science 155:279–284.

Ford, H. A. 1989. Ecology of birds: an Australian perspective. Surrey Beatty and Sons, Chipping Norton, New South Wales, Australia.

Gyllensten, U. B., and H. A. Erlich. 1988. Generation of single-stranded DNA by the polymerase chain reaction and its application to direct sequencing of the *HLA-DQA* locus. Proceedings of the National Academy of Sciences, USA 85:7652–7656.

Hall, B. P. (ed.) 1974. Birds of the Harold Hall Australian Expeditions: 1962–1970. Trustees of the British Museum of Natural History, London.

Harvey, P., and M. Pagel. 1991. The comparative method in evolutionary biology. Oxford University Press, Oxford.

Hasegawa, M., and S. Horai. 1991. Time of the deepest root for polymorphism in human mitochondrial DNA. Journal of Molecular Evolution 32:37–42.

Hasegawa, M., H. Kishino, and T. Yano. 1985. Dating of the human-ape splitting by a molecular clock of mitochondrial DNA. Journal of Molecular Evolution 22:160–174.

Hudson, R. R. 1991. Gene genealogies and the coalescent process. Oxford Survey of Evolutionary Biology 7:1–44.

Hudson, R. R., M. Slatkin, and W. P. Maddison. 1992.

Estimation of levels of gene flow from DNA sequence data. Genetics 132:583–589.

Jin, L., and M. Nei. 1990. Limitations of the evolutionary parsimony method of phylogenetic analysis. Molecular Biology and Evolution 7:82–102.

Johnson, N. K., and J. A. Martin. 1988. Evolutionary genetics of flycatchers. II. Differentiation in the *Empidonax difficilis* complex. Auk 105:177–191.

Jongsma, D. 1970. Eustatic sea level changes in the Arafura Sea. Nature 228:150–151.

Jukes, T. H., and C. R. Cantor. 1969. Evolution of protein molecules. Pp. 21–123 *in* H. N. Monroe, ed. Mammalian protein metabolism, vol. III. Academic Press, New York.

King, B. R. 1980. Social organization and behavior of the grey-crowned babbler, *Pomatostomus temporalis*. Emu 80:59–76.

Kishino, H., and M. Hasegawa. 1989. Evaluation of the maximum likelihood estimate of the evolutionary tree topologies from DNA sequence data, and the branching order in Hominoidea. Journal of Molecular Evolution 29:170–179.

Kocher, T. D., W. K. Thomas, A. Meyer, S. V. Edwards, S. Pääbo, F. X. Villablanca, and A. C. Wilson. 1989. Dynamics of mitochondrial DNA evolution in animals: amplification and sequencing with conserved primers. Proceedings of the National Academy of Sciences, USA 86:6196–6200.

Kocher, T. D., and A. C. Wilson. 1991. Sequence evolution of mitochondrial DNA in humans and chimpanzees: control region and a protein-coding region. Pp. 391–413 *in* S. Osawa and T. Honjo, eds. Evolution of life: fossils, molecules, and culture. Springer, Tokyo.

Kreitman, M. 1991. Detecting selection at the level of DNA. Pp. 204–221 *in* R. K. Selander et al., eds. Evolution at the molecular level. Sinauer, Sunderland, Mass.

Lynch, M., and T. J. Crease. 1990. The analysis of population survey data on DNA sequence variation. Molecular Biology and Evolution 7:377–394.

Maddison, W. P., and D. R. Maddison. 1992. MacClade: analysis of phylogeny and character evolution, version 3.0. Sinauer, Sunderland, Mass.

Maddison, W. P., and M. Slatkin. 1991. Null models for the number of evolutionary steps in a character on a phylogenetic tree. Evolution 45:1184–1197.

Medrano, J. F., E. Aasen, and L. Sharrow. 1990. DNA extraction from nucleated red blood cells. BioTechniques 8:43.

Moran, G. F., O. Muona, and J. C. Bell. 1989. *Acacia mangium*: a tropical forest tree of the coastal lowlands with low genetic diversity. Evolution 43:231–235.

Nei, M. 1987. Molecular evolutionary genetics. Columbia University Press, New York.

Neigel, J. E., and J. C. Avise. 1986. Phylogenetic relationships of mitochondrial DNA under various demographic models of speciation. Pp. 515–534 *in* S. Karlin and E. Nevo, eds. Evolutionary processes and theory. Academic Press, New York.

Peterson, A. T. 1992. Philopatry and genetic differentiation in the *Aphelocoma* jays. Biological Journal of the Linnean Society 47:249–260.

Prager, E. M., R. D. Sage, U. Gyllensten, W. K. Thomas, R. Hubner, C. S. Jones, L. Noble, J. B. Searle, and A. C. Wilson. 1993. Mitochondrial DNA sequence diversity and the colonization of Scandinavia by house mice from East Holstein. Biological Journal of the Linnean Society 50:85–122.

Quinn, T. W. 1992. The genetic legacy of Mother Goose—phylogeographic patterns of lesser snow goose (*Chen caerulescens caerulescens*) maternal lineages. Molecular Ecology 1:105–117.

Quinn, T. W., and A. C. Wilson. 1993. Sequence evolution in and around the mitochondrial control region of birds. Journal of Molecular Evolution 37: 417–425.

Ridley, M. 1983. The explanation of organic diversity: the comparative method and adaptations for mating. Clarendon Press, Oxford.

Saitou, N., and M. Nei. 1985. The neighbor-joining method: a new method for reconstructing phylogenetic trees. Molecular Biology and Evolution 4:406–425.

Schaeffer, S. W., and E. L. Miller. 1991. Nucleotide sequence analysis of *Adh* genes estimates the time of geographic isolation of the Bogota population of *Drosophila pseudoobscura*. Proceedings of the National Academy of Sciences, USA 88:6097–6101.

Schodde, R., and J. H. Calaby. 1972. The biogeography of the Australo-Papuan bird and mammal faunas in relation to the Torres Strait. Pp. 257–300 *in* D. Walker, ed. Bridge and barrier: the natural and cultural history of the Torres Strait. Research School Pacific Studies (Australian National University), Canberra, Australia.

Shields, G. F., and A. C. Wilson. 1987. Calibration of mitochondrial DNA evolution in geese. Journal of Molecular Evolution 24:212–217.

Slatkin, M. 1987. The average number of sites separating DNA sequences drawn from a subdivided population. Theoretical Population Biology 32:42–49.

———. 1989. Detecting small amounts of gene flow from phylogenies of alleles. Genetics 121:609–612.

———. 1991. Inbreeding coefficients and coalescence times. Genetical Research 58:167–175.

Slatkin, M., and R. R. Hudson. 1991. Pairwise comparisons of mitochondrial DNA sequences in stable and exponentially growing populations. Genetics 129:555–562.

Slatkin, M., and W. P. Maddison. 1989. A cladistic measure of gene flow inferred from the phylogenies of alleless. Genetics 123:603–613.

———. 1990. Detecting isolation by distance using phylogenies of genes. Genetics 126:249–260.

Strobeck, C. 1987. Average number of nucleotide differences in a sample from a single subpopulation: a test for population subdivision. Genetics 117:149–153.

Swofford, D. L. 1991. PAUP, phylogenetic analysis using parsimony, version 3.0r. Illinois Natural History Survey, Champaign, Ill.

Taberlet, P., A. Meyer, and J. Bouvet. 1992. Unusual mitochondrial DNA polymorphism in two local populations of the blue tit (*Parus caeruleus*). Molecular Ecology. *In press.*

Tajima, F. 1983. Evolutionary relationship of DNA sequences in finite populations. Genetics 105:437–460.

———. 1989. Statistical method for testing the neutral mutation hypothesis by DNA polymorphism. Genetics 123:585–595.

Takahata, N. 1989. Gene genealogy in three related populations: consistency probability between gene and population trees. Genetics 122:957–966.

Takahata, N., and M. Nei. 1985. Gene genealogy and variance of interpopulational nucleotide differences. Genetics 110:325–344.

Tavaré, S. 1984. Lines-of-descent and genealogical processes, and their applications in population genetics models. Theoretical Population Biology 26: 119–164.

Templeton, A., K. A. Crandall, and C. F. Sing. 1992. A cladistic analysis of phenotypic associations with haplotypes inferred from restriction endonuclease mapping and DNA sequence data. III. Cladogram estimation. Genetics 132:619–633.

Thomas, W. K., S. Pääbo, F. X. Villablanca, and A. C. Wilson. 1990. Spatial and temporal continuity of kangaroo rat populations shown by sequencing mitochondrial DNA from museum specimens. Journal of Molecular Evolution 31:101–112.

Veevers, J. J. 1969. Paleogeography of the Timor Sea region. Paleogeography, Paleoclimatology, Paleoecology 6:125–140.

Vigilant, L., R. Pennington, H. Harpending, T. D. Kocher, and A. C. Wilson. 1989. Mitochondrial DNA sequences in single hairs from a southern African population. Proceedings of the National Academy of Sciences, USA 86:9350–9354.

Vigilant, L., M. Stoneking, H. Harpending, K. Hawkes, and A. C. Wilson. 1991. African populations and the evolution of human mitochondrial DNA. Science 253:1503–1507.

Wakeley, J. 1993. Substitution rate variation among sites in hypervariable region 1 of human mitochondrial DNA. Journal of Molecular Evolution 37:613–623.

Wetton, J. H., R. E. Carter, D. T. Parkin, and D. Walters. 1987. Demographic study of a wild house sparrow population by DNA fingerprinting. Nature 327:147–149.

Wilson, A. C., R. L. Cann, S. M. Carr, M. George, U. B. Gyllensten, K. M. Helm-Bychowski, R. G. Higuchi, S. R. Palumbi, E. M. Prager, R. D. Sage, and M. Stoneking. 1985. Mitochondrial DNA and two perspectives on evolutionary genetics. Biological Journal of the Linnean Society 26:375–400.

Wilson, A. C., E. A. Zimmer, E. M. Prager, and T. D. Kocher. 1989. Restriction mapping in the molecular systematics of mammals: a retrospective salute. Pp. 407–419 *in* B. Fernholm et al., eds. The hierarchy of life. Molecules and morphology in phylogenetic analysis. Elsevier Science, Amsterdam, The Netherlands.

Woodroffe, C., and M. Mulrennan. 1991. The past, present, and future extent of tidal influence in Northern Territory coastal wetlands. Pp. 83–104 *in* I. Moffatt and A. Webb, eds. North Australian research: some past themes and new directions. North Australian Research Unit, Australian National University, Canberra, Australia.

Wright, S. 1951. The genetical structure of populations. Annals of Eugenics 15:323–354.

Zink, R. M. 1991. Geography of mitochondrial DNA variation in two sympatric sparrows. Evolution 45: 329–340.

Zink, R. M., D. F. Lott, and D. W. Anderson. 1987. Genetic variation, population structure, and evolution of California quail. Condor 89:395–405.

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