

Fern Phylogeny Based on *rbcL* Nucleotide Sequences

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ABSTRACT.—We analyzed nucleotide variation in *rbcL* (the gene encoding the large subunit of ribulose 1,5-bisphosphate carboxylase/oxygenase) from 99 genera of leptosporangiate ferns representing 31 of the 33 extant families. Phylogenetic relationships were inferred using three methods: neighbor joining, maximum parsimony, and maximum likelihood. All three methods resulted in optimal trees that were similar. Within the context of those taxa examined, these trees suggest that: 1) Polypodiaceae, Grammitidaceae and *Pleurosoriopsis* form a monophyletic group that is most derived among indusiate ferns; 2) *Davallia* is closely related to the Polypodiaceae; 3) *Tectaria* is related to Oleandraceae rather than to other members of Dryopteridaceae; 4) *Rumohra* and

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Elaphoglossum are closely related; 5) Dryopteridaceae are polyphyletic; 6) a monophyletic group consists of Polypodiaceae, Grammitidaceae, Davalliaceae, Oleandraceae, Nephrolepidaceae, Lomariopsidaceae, Dryopteridaceae, Thelypteridaceae, Blechnaceae, Aspleniaceae, and *Pleurosoriopsis*; 7) Dennstaedtiaceae, Monachosoraceae, Pteridaceae, Vittariaceae, and the previous clade form a monophyletic group; 8) Dennstaedtiaceae are polyphyletic; 9) tree ferns in the Cyatheaceae, Metaxyaceae, and Dicksoniaceae form a monophyletic group that emerged early in the diversification of leptosporangiate ferns; 10) Plagiogyriaceae and Loxomataceae emerge with the tree ferns; 11) heterosporous water ferns form a monophyletic group that diverged prior to the tree ferns; 12) Schizaeaceae, Cheiroleuriaceae, Dipteridaceae, Gleicheniaceae, Matoniaceae, and Hymenophyllaceae are basal to the heterosporous aquatic ferns; and 13) Osmundaceae are the most basal lineage of the leptosporangiate ferns.

Nucleotide variation in the gene encoding the large subunit of ribulose 1,5-bisphosphate carboxylase/oxygenase (*rbcL*) has provided the most extensive molecular data set for plant systematists (e.g., Chase et al., 1993). In the last five years almost every major lineage of land plants has been surveyed for *rbcL* sequence data (e.g., Chase et al., 1993; Manhart, 1994; Mishler et al., 1994), including three studies of ferns. Hasebe et al. (1993) analyzed *rbcL* from two leptosporangiate ferns (*Adiantum* and *Osmunda*) and two eusporangiate ferns (*Angiopteris* and *Botrypus*), and inferred their relationships with other land plants. Translated amino acid sequences, rather than DNA sequences, were used in that study, because variation in GC content across widely divergent taxa can bias phylogenetic analyses (Hasegawa et al., 1993). Maximum likelihood analyses supported monophyly of the ferns but relationships among basal taxa within the ferns were not fully resolved. Polymerase Chain Reaction (PCR)-mediated direct sequencing has recently become feasible and accumulation of fern *rbcL* data has accelerated. Hasebe et al. (1994) analyzed *rbcL* from 64 species (62 genera, 29 families sensu Kramer and Green, 1990) using neighbor joining and maximum parsimony methods. Wolf et al. (1994) analyzed *rbcL* from 45 species of dennstaedtioid ferns using maximum parsimony. The two studies produced almost concordant results, although both pointed out the need for more sequence data to increase taxon sampling, especially in large and diverse families such as Dryopteridaceae and Pteridaceae.

In this study, we compiled information from 180 fern *rbcL* sequences available to us (Appendix). We selected 107 taxa representing all major extant lineages of ferns and analyzed the data using three different tree construction methods: neighbor joining (NJ), maximum parsimony (MP), and maximum likelihood (ML). We also estimated the number of synonymous (Ks) and non-synonymous (Ka) nucleotide substitutions in fern *rbcL* sequences. The goals of our study were to 1) infer relationships among the major lineages of leptosporangiate ferns, 2) compare the results of three different tree making methods, 3) compare average interfamilial and intergeneric (intrafamilial) Ks and Ka in ferns with those in angiosperms, and 4) infer the phylogenetic positions of the historically problematic taxa *Loxoma*, *Orthiopteris*, and *Pleurosoriopsis*.

MATERIALS AND METHODS

We randomly selected one species from each genus for which *rbcL* data were available at the time of this analysis (Appendix), incorporating representatives from all extant families and subfamilies (see Fig. 1) except the Lophosoriaceae and Hymenophyllopsidaceae. We followed Kramer and Green's (1990) system for the delimitation of families. However, generic delimitation was not always concordant with Kramer and Green's (1990) system, and several genera were treated as in the original references from which sequence data were cited (see Appendix). The 107 selected sequences were used in the NJ and MP analyses. Due to limitations in computer capabilities and time, ML analyses were performed on a reduced data set of 72 species.

The *rbcL* sequences themselves were generated in ten different laboratories, and therefore specific protocols for DNA extraction, PCR amplification, cloning, and sequencing vary. Details are provided in the original citations given in the Appendix (see also Ranker, 1995).

The sequences could be aligned without any insertions or deletions. The 1206 bp region between base pair positions 73 and 1278 (from the initial methionine codon of *Marchantia polymorpha*; Ohyama et al., 1986) was used for phylogenetic analyses.

For the NJ analyses, we used PHYLIP version 3.5c (Felsenstein, 1993). In calculating the distance matrix, Kimura's 2-parameter model of nucleotide substitution (Kimura, 1980) was implemented, in which the transition (ts)/transversion (tv) ratio was fixed to 3 (Hasebe et al., 1994). Support for each internode was estimated using bootstrap resampling of nucleotide positions (Felsenstein, 1985; Felsenstein and Kishino, 1993). We also generated a NJ tree using the MOLPHY ver. 2.2 program (Adachi and Hasegawa, 1994). We calculated a distance matrix based on a maximum likelihood model of nucleotide substitution (HKY85 model; Hasegawa et al., 1985) using the "distance option" of NucML (Adachi and Hasegawa, 1994) with the assumption of $ts/tv=3$.

For the MP analyses, we used PAUP version 3.1 (Swofford, 1993). We searched for multiple islands of equally most parsimonious trees (Maddison, 1991) using the heuristic search method. We assigned equal weight to each codon position and conducted 500 searches using random-order-entry starting trees and nearest neighbor interchanges (NNI) branch swapping with MULPARS and STEEPEST DESCENT selected. The equally most parsimonious trees from these 500 searches were used as starting trees for tree bisection-reconnection (TBR) branch swapping with MULPARS and STEEPEST DESCENT selected (see Olmstead and Palmer [1994] for other search strategies for multiple islands with large data sets). We also implemented the differential character-state weighting model of Albert et al. (1993) and conducted 1000 searches using random-order-entry starting trees and NNI branch swapping with MULPARS and STEEPEST DESCENT selected. The equally most parsimonious trees from these 1000 searches were then used as starting trees for subtree pruning-grafting (SPR) and TBR branch swapping with MULPARS and STEEPEST DESCENT selected. Bootstrap (Felsenstein, 1985; Sanderson, 1989; Hillis and

Bull, 1993; Felsenstein and Kishino, 1993) and decay analyses (Bremer, 1988) were used to obtain a measure of confidence for each branch. Five hundred bootstrap replications were carried out with equal weighting, simple sequence addition, and NNI branch swapping with MULPARS and STEEPEST DESCENT selected. Bootstrap values were recorded for nodes supported in more than half the replicates. For the decay analysis, the equally most parsimonious trees from the heuristic searches above were used as starting trees for a further search using TBR branch swapping with MULPARS and STEEPEST DESCENT selected. All trees up to two steps longer than the equally most parsimonious trees were saved. The strict consensus trees were calculated to determine decay values for each branch, i.e., the number of steps that could be added to the tree before causing the branch to collapse.

For the ML data set, we eliminated 35 species that always clustered in monophyletic groups with other taxa with more than 85% bootstrap probability during preliminary analyses with NJ and MP methods. The data set containing 72 genera was analyzed on a Silicon Graphics Indigo II with IRIX 5.2 and the MIPS R4400 coprocessor using fastDNAmI version 1.0.6 (Felsenstein, 1981; Olsen et al., 1992). Thirty replications of the ML analysis were executed, each with different randomly determined orders of sequence addition and local branch swapping in effect. The "Categories" option was invoked to specify the different rates of substitution by codon position. Codons were categorized (1.0:0.39:8.0 for first:second:third codon positions, respectively) based on empirical estimates of base substitution rates at each codon position for fern *rbcL* sequences (Hasebe et al., 1994). As with the NJ and MP analyses, a ts/tv ratio of 3.0 was specified. One tree resulted from each of the 30 random sequence addition searches. Using fastDNAmI version 1.1 (Felsenstein, 1981; Olsen et al., 1994), we performed a statistical test (Kishino and Hasegawa, 1989) of each of these trees against the one with the best log-likelihood. This test uses the standard error of the difference of log-likelihood from the best ML tree to determine whether the log-likelihoods of any of the trees are significantly worse (lower) than that of the best ML tree. In addition, the "Global and User Tree" option was invoked to identify the best of these 30 trees and to carry out global branch swapping to attempt to find a more likely tree.

In the MP and ML analyses, the eusporangiate ferns (Marattiaceae and Ophioglossaceae) and Psilotaceae were designated as outgroup taxa based on previous molecular phylogenetic results (Hasebe et al., 1993; Hiesel et al., 1994; Manhart, 1994). Only a single taxon could be designated as the outgroup in the NJ analyses; we chose *Angiopteris* (Marattiaceae).

The number of synonymous and nonsynonymous nucleotide substitutions per site were counted by the NG method (Nei and Gojobori, 1986) for the taxa indicated in the Appendix, using the program kindly provided by N. Saitou (National Institute of Genetics, Mishima, Japan). The expected numbers of synonymous and nonsynonymous nucleotide substitutions per site (K_s and K_a , respectively) estimated by the Jukes and Cantor equation (Jukes and Cantor, 1969) were averaged among species-pairs in different families (interfamilial distances) and among pairs of species in different genera that formed a mono-

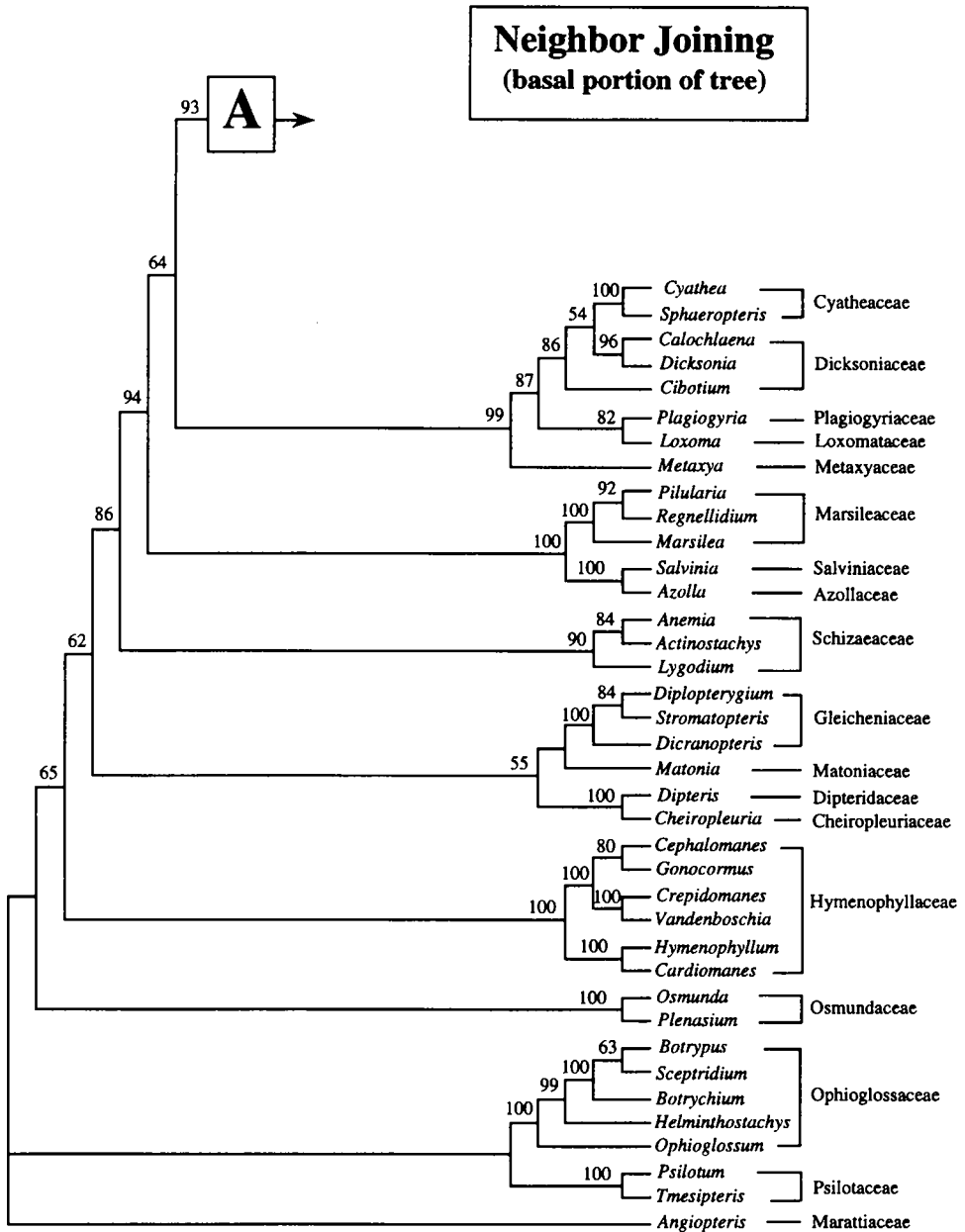


FIG. 1. Tree obtained using the neighbor joining (NJ) method. The branch lengths are arbitrary. Bootstrap values are indicated for nodes supported in $\geq 50\%$ of 1000 bootstrap replicates. Taxa shown in the tree are indicated in the Appendix. The tree was rooted by *Angiopteris* (Marattiaceae). The basal and upper portions of the tree are connected along the branch labeled "A".

Neighbor Joining
(upper portion of tree)

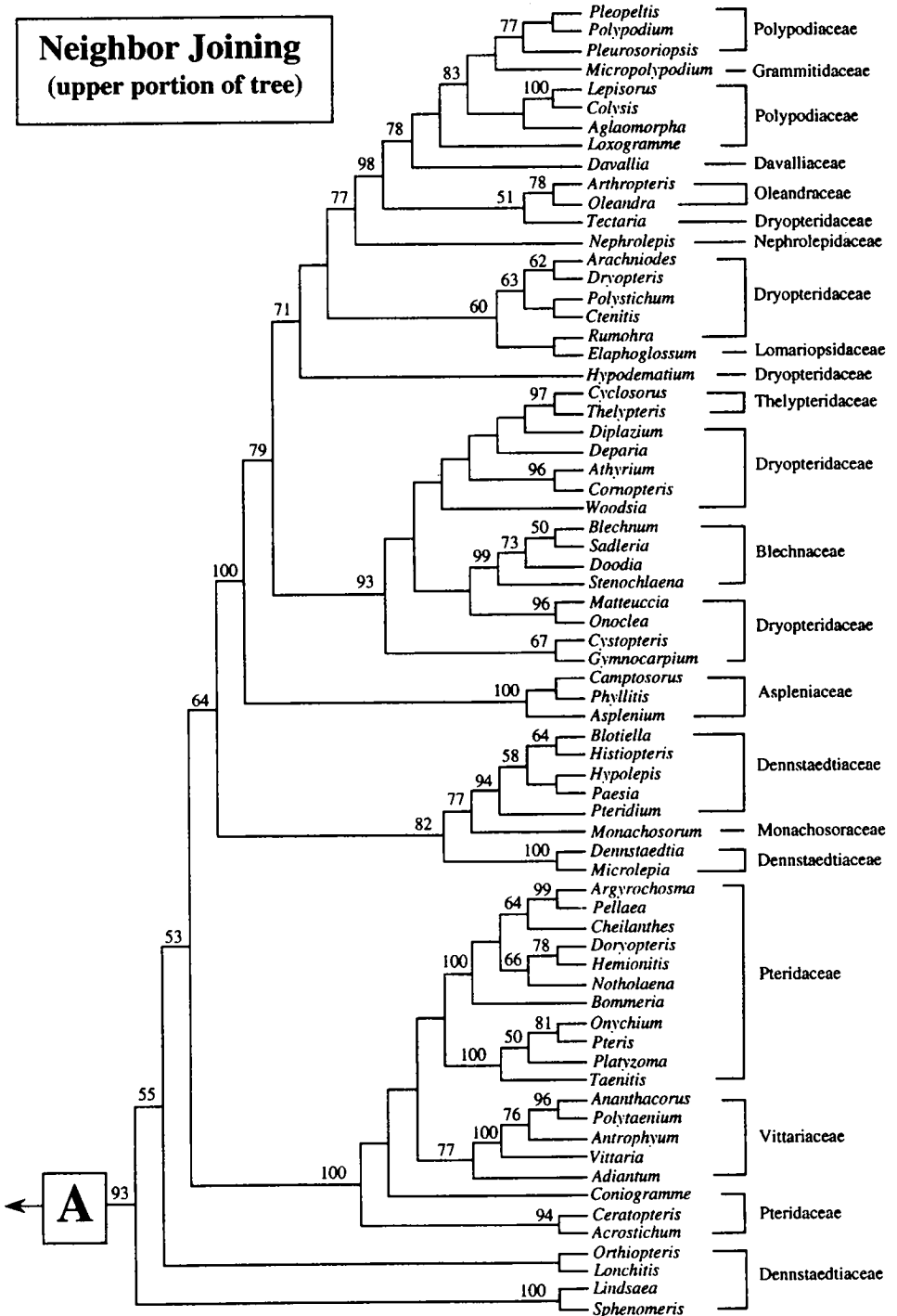


FIG. 1. Continued.

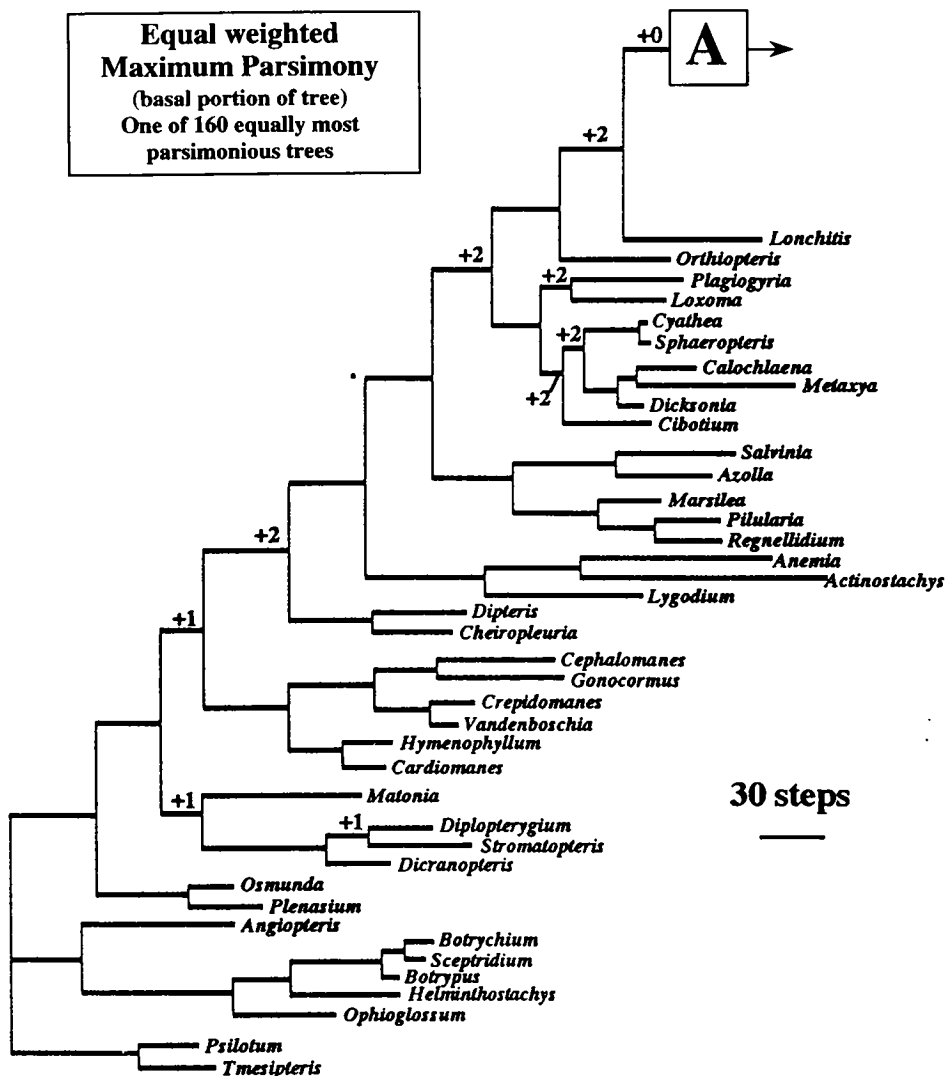


FIG. 2. An arbitrarily selected tree from the 160 equally most parsimonious trees in the maximum parsimony (MP) analyses with equal weighting. Branch lengths correspond to the number of nucleotide substitutions (ACCTRAN optimization). Decay indices are indicated on those nodes where the number of additional steps needed for a branch to collapse is ≤ 2 . A decay index of "+0" means that the node collapses in the strict consensus of the 160 equally most parsimonious trees. CI=0.196; RI=0.596; RCI=0.117. Taxa shown in the tree are indicated in the Appendix. The tree was rooted by the eusporangiate ferns and Psilotaceae. The basal and upper portions of the tree are connected along the branch labeled "A".

**Equal weighted
Maximum Parsimony**
(upper portion of tree)
One of 160 equally most
parsimonious trees

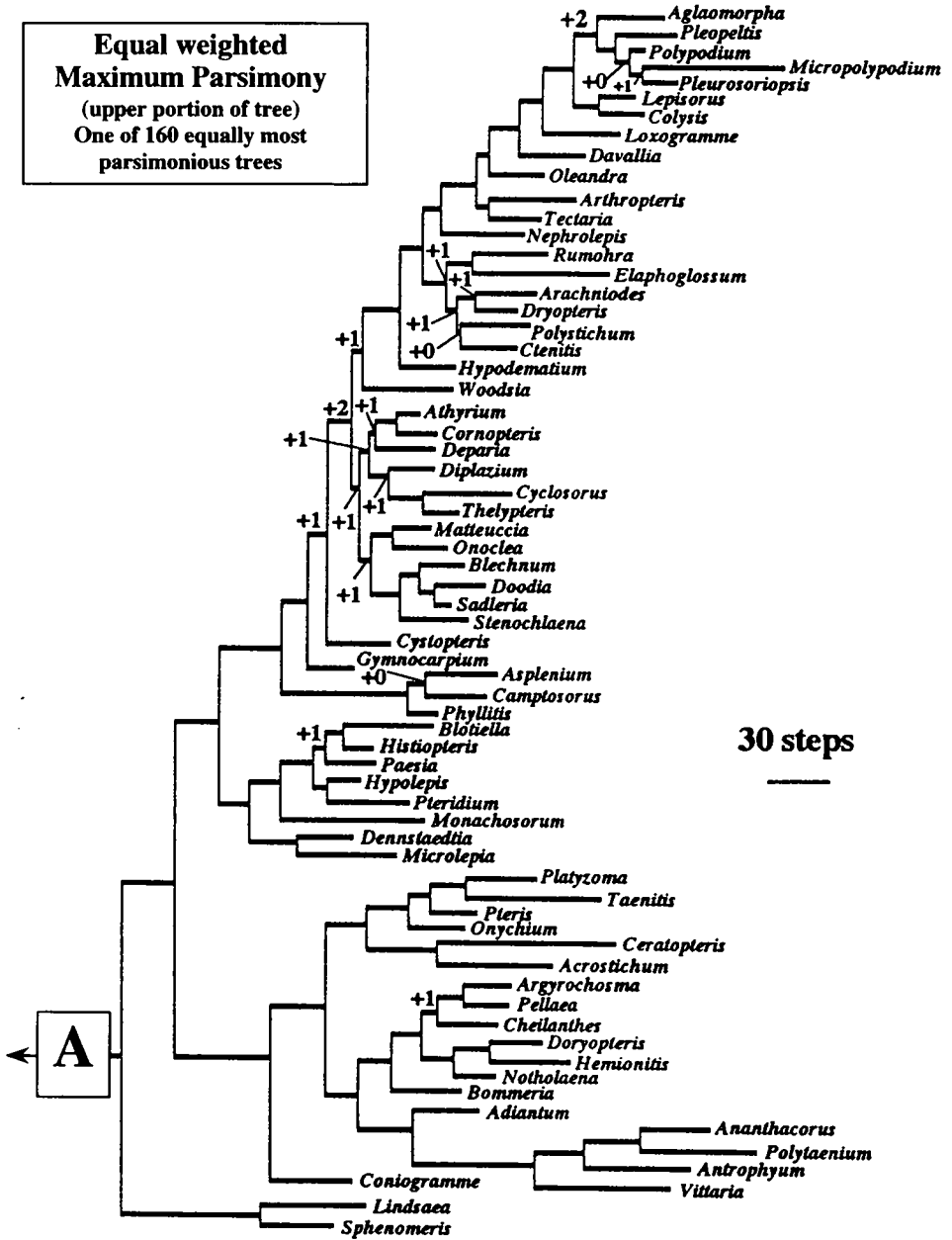


FIG. 2. Continued.

phyletic group within families (intergeneric distances). We also calculated the Ks and Ka values for angiosperm *rbcl* sequences available in the DNA databases DDBJ, EMBL, and NCBI. We selected 20 families for which more than 5

**Equal weighted
Maximum Parsimony
(basal portion of tree)
Strict consensus of 160 equally
most parsimonious trees**

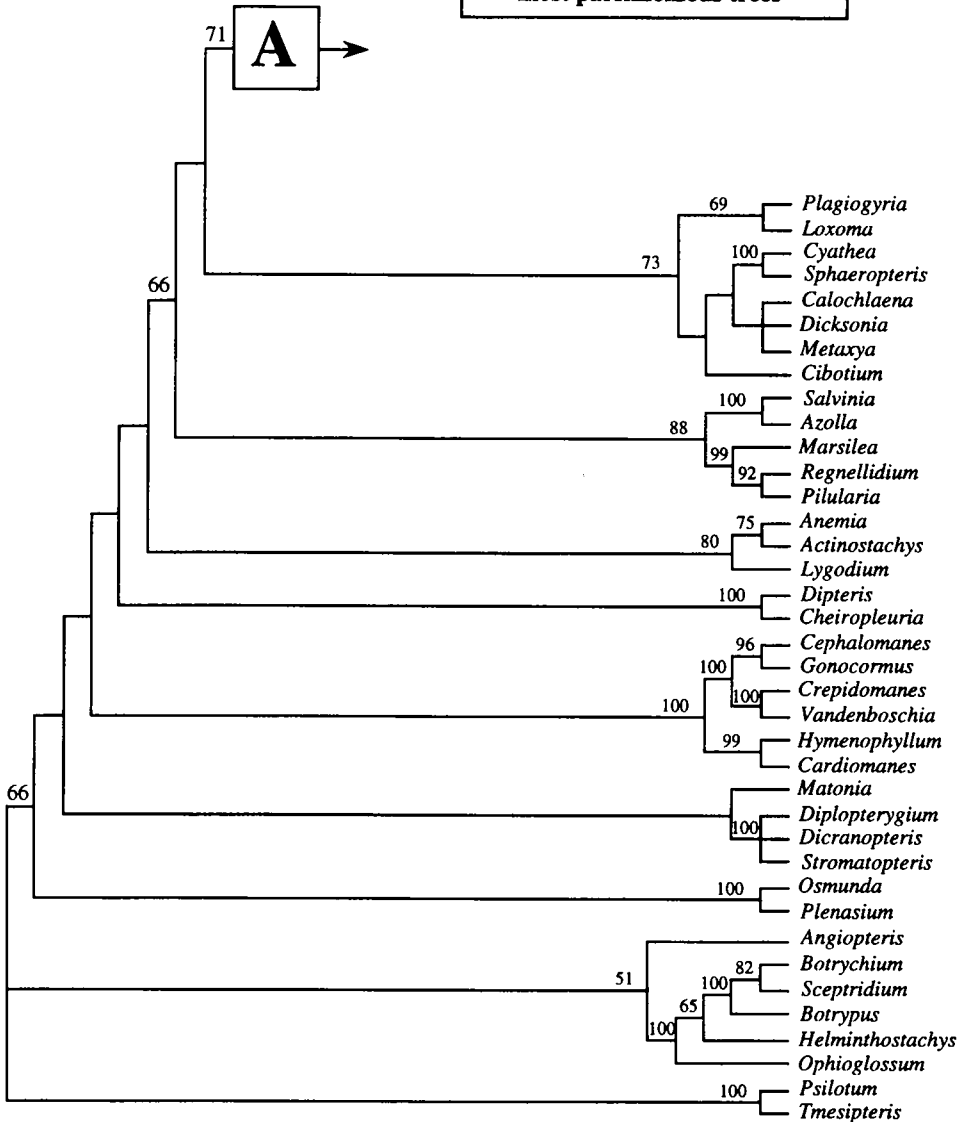


FIG. 3. Strict consensus of 160 equally most parsimonious trees obtained using maximum parsimony (MP) with equal weighting, simple sequence addition, and NNI branch swapping with MULPARS and STEEPEST DESCENT selected. Bootstrap values are indicated for nodes supported in $\geq 50\%$ of 500 bootstrap replicates. Taxa shown in the tree are indicated in the Appendix. The tree was rooted by the eusporangiate ferns and Psilotaceae. The basal and upper portions of the tree are connected along the branch labeled "A".

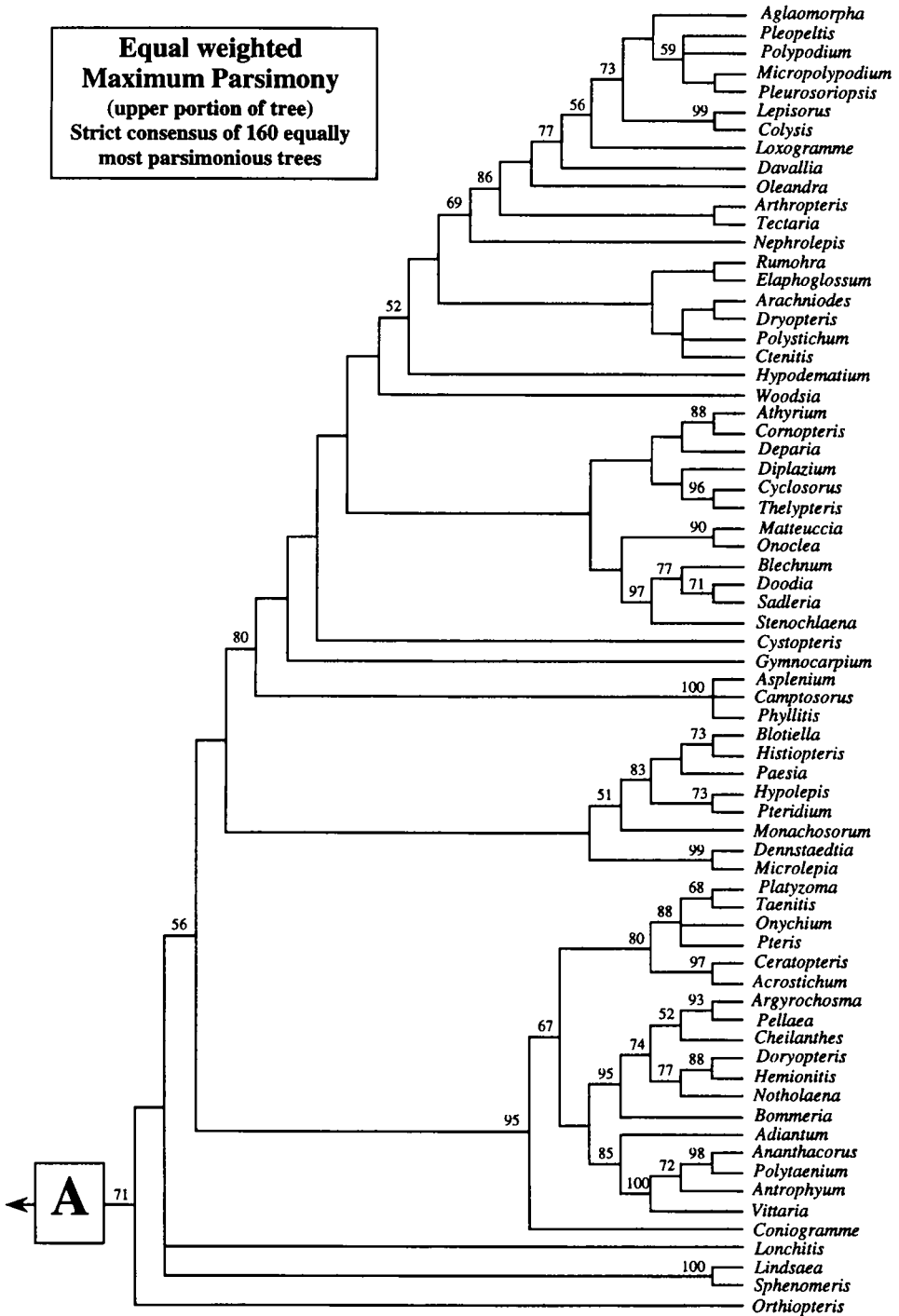


FIG. 3. Continued.

Maximum Parsimony

weighted (Albert et al., 1993)

(basal portion of tree)

Strict consensus of 8 equally
most parsimonious trees

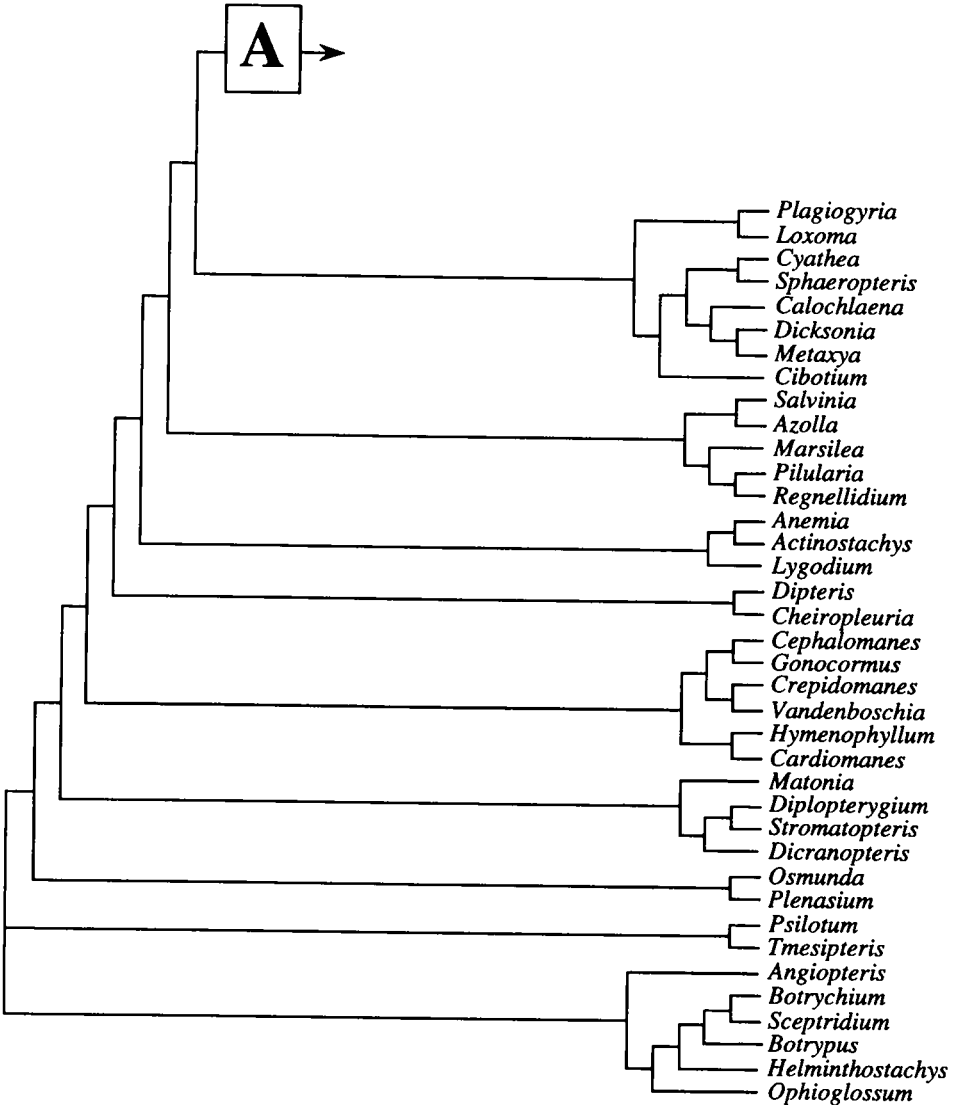


FIG. 4. Strict consensus tree of the eight equally most parsimonious trees obtained using maximum parsimony (MP) with the weighting criterion of Albert et al. (1993). Branch lengths are arbitrary. Taxa shown in the tree are indicated in the Appendix. The tree was rooted by the eusporangiate ferns and Psilotaceae. The basal and upper portions of the tree are connected along the branch labeled "A".

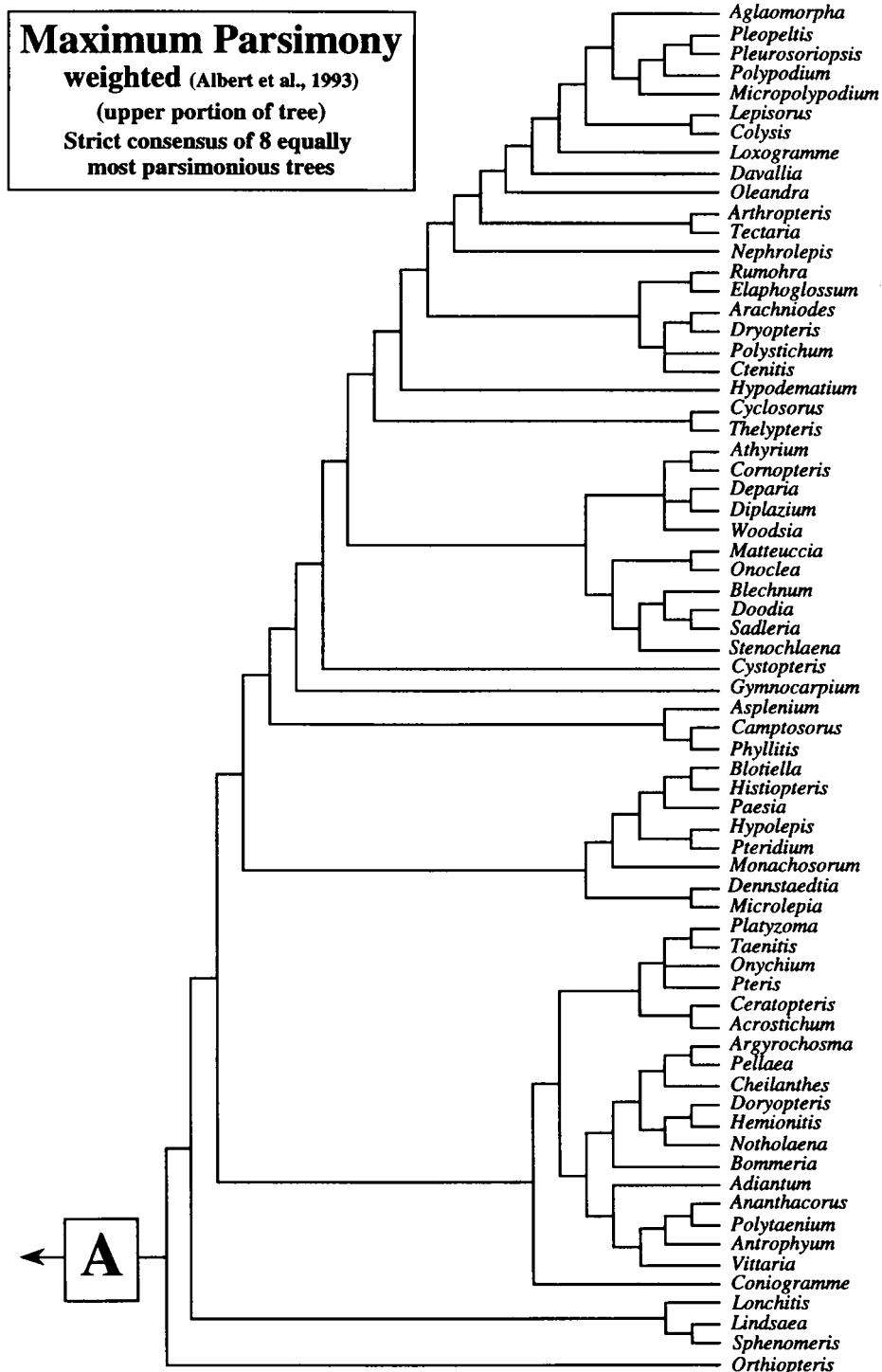


FIG. 4. Continued.

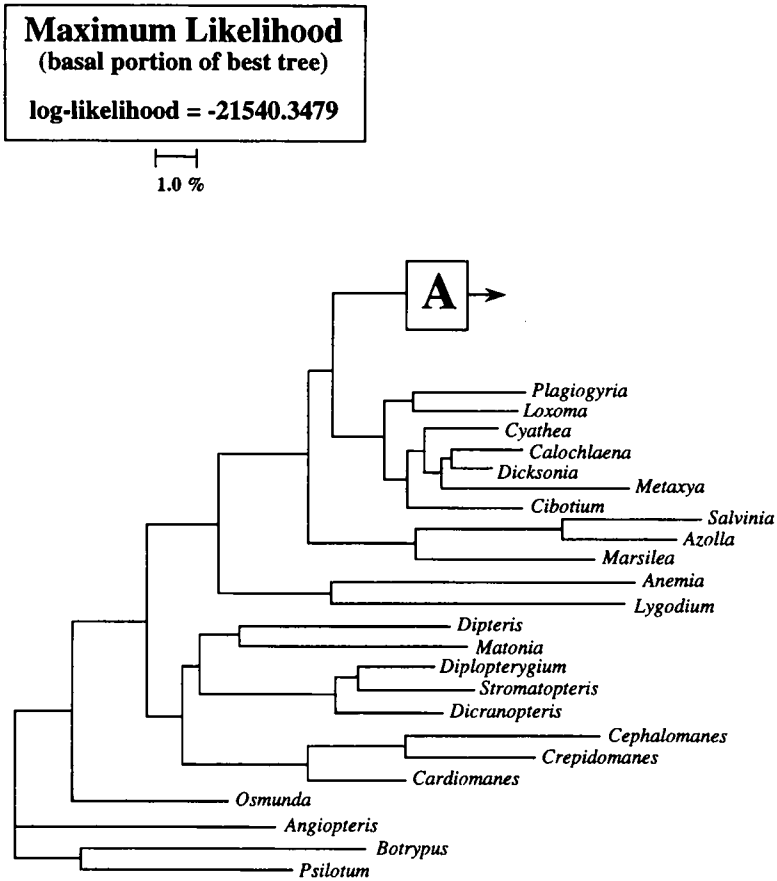


FIG. 5. The best maximum likelihood tree with a log-likelihood of -21540.3479 obtained after 30 random sequence addition searches with local branch swapping. Codons were categorized (1.0: 0.39:8.0 for first:second:third codon positions, respectively), and ts/tv was assumed as 3.0. Taxa shown in the tree are indicated in the Appendix. The tree was rooted by the eusporangiate ferns and Psilotaceae. The basal and upper portions of the tree are connected along the branch labeled "A".

genera were reported in the databases. The dataset and the complete table of K_s and K_a values are available from K. Ueda upon request.

RESULTS

The NJ tree obtained using Kimura's 2-parameter model is shown in Fig. 1. The NJ analysis with a maximum likelihood model of nucleotide substitution resulted in the same tree.

Using the MP method with equal weighting, 160 equally most parsimonious trees of 5978 steps were found in two islands (40 and 120 trees, respectively). Using random-order-entry starting trees, the bigger island was found at the first

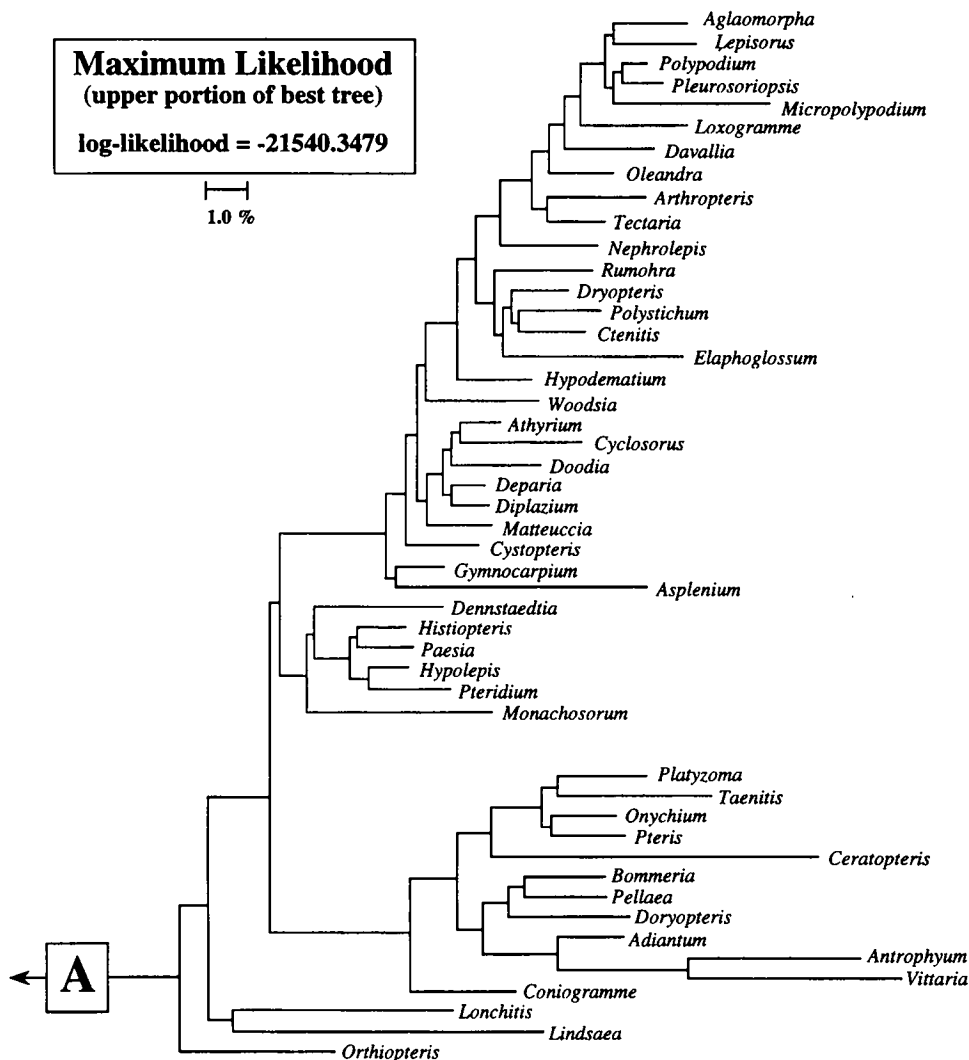


FIG. 5. Continued.

search and the smaller island at the 48th search. The same strict consensus tree was calculated for both islands. These trees had a consistency index of 0.196, a retention index of 0.596, and a rescaled consistency index of 0.117. One arbitrarily selected tree from the 160 equally most parsimonious trees is shown (with decay values) in Fig. 2. The strict consensus of the 160 equally most parsimonious trees is shown in Fig. 3. The weighted MP analysis resulted in eight equally most parsimonious trees from one island found at the first search using random-order-entry starting trees. These eight trees were then used as starting trees for further searches with both SPR and TBR swapping. Additional trees of shorter or equal length were not found. The strict consen-

Maximum Likelihood

(basal portion of tree)

Strict consensus of 22 trees

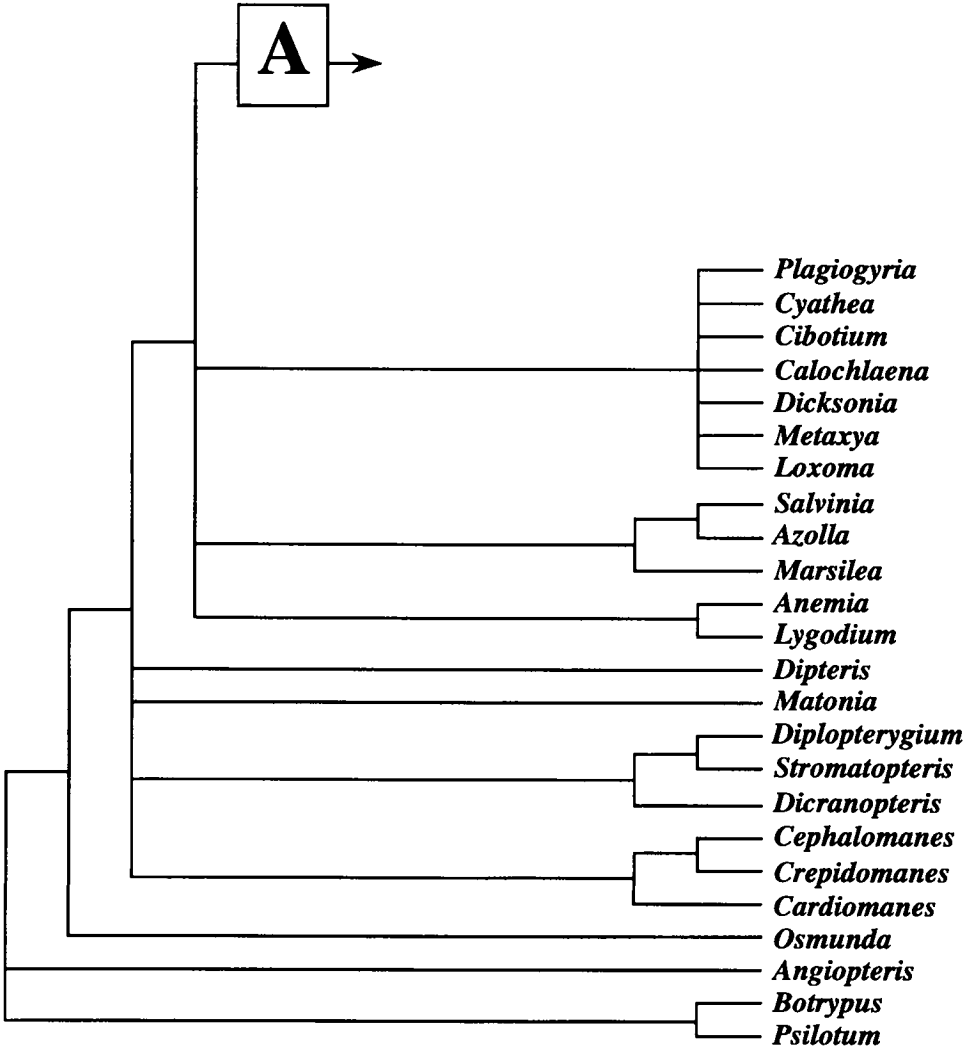


FIG. 6. Strict consensus tree of the best ML tree and the 21 trees whose log-likelihoods were not significantly worse than the best ML tree. Taxa shown in the tree are indicated in the Appendix. The tree was rooted by the eusporangiate ferns and Psilotaceae. The basal and upper portions of the tree are connected along the branch labeled "A".

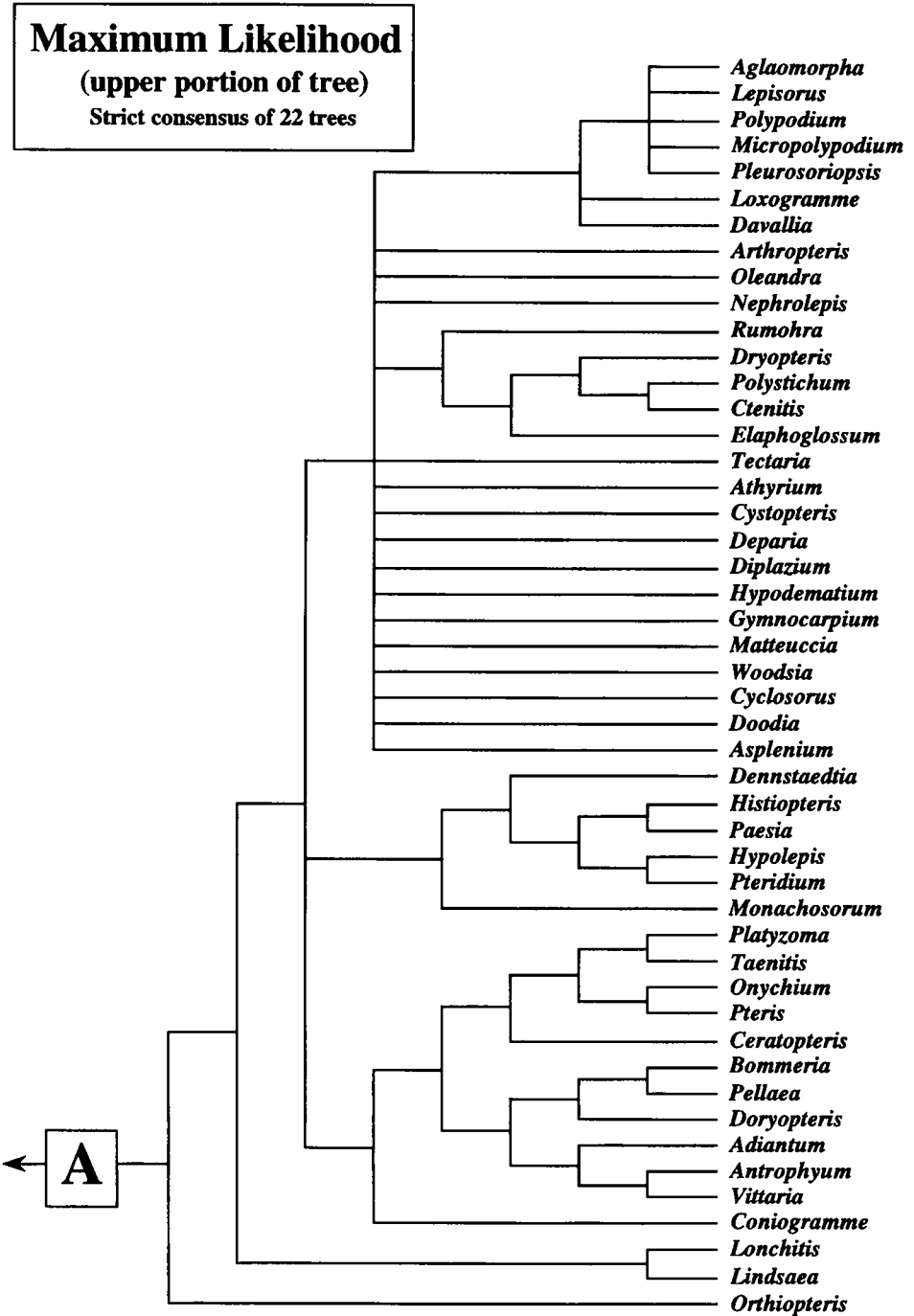


FIG. 6. Continued.

TABLE 1. Mean and standard error (SE) of synonymous (Ks) and nonsynonymous (Ka) nucleotide substitutions per nucleotide site in ferns and angiosperms. The t-test is based on the difference between the mean values of fern and angiosperm families.

| | | Ferns | | Angiosperms | | t-test | |
|---------------|----|-------|-------|-------------|-------|--------|----------|
| | | Mean | SE | Mean | SE | t | P |
| Interfamilial | Ks | 1.170 | 0.461 | 0.372 | 0.111 | 1789 | < 0.0001 |
| | Ka | 0.028 | 0.008 | 0.025 | 0.010 | 351 | < 0.0001 |
| Intergeneric | Ks | 0.411 | 0.258 | 0.104 | 0.074 | 29 | < 0.0001 |
| | Ka | 0.018 | 0.011 | 0.013 | 0.006 | 8 | < 0.0001 |

sus tree of these eight equally most parsimonious trees is shown in Fig. 4. All of these equally most parsimonious trees obtained by the weighted MP analysis corresponded in topology to the trees 2 steps longer (5980 steps) than the equally most parsimonious trees obtained using the equal weighting criterion.

The ML analysis produced 25 different trees, because some of the 30 random addition sequence searches resulted in the same trees. The ML tree with the best log-likelihood (-21540.3479) is shown in Fig. 5. Three of the 30 random addition sequence searches resulted in finding the same best tree. A statistical test of the variance of log-likelihood differences (Kishino and Hasegawa, 1989) between each of the 24 less likely trees and the best ML tree was carried out to determine whether any of the trees were significantly worse than the best ML tree. Three trees had a significantly lower log-likelihood than that of the best ML tree, whereas the log-likelihoods of the other 21 trees were not significantly worse than that of the best ML tree. The strict consensus tree of these 21 trees and the best ML tree is shown in Fig. 6. Although global branch swapping was executed on all of the 30 trees, no trees with a higher log-likelihood than the best ML tree (Fig. 5) were obtained.

The average interfamilial and intergeneric Ks and Ka values for ferns and angiosperms are shown in Table 1. A t-test shows that these values are significantly higher in ferns than in angiosperms at the 1% level. The intrafamilial Ks and Ka values among fern and angiosperm genera are shown in Table 2.

We also attempted to calculate Ks and Ka values using the Li et al. (1985) method, however, the "LWL91" program (Li, 1993) kindly provided by Li (The University of Texas, Houston) could do the calculations for only up to 30 species-pairs. We were unable to expand the taxon number limitation of the program. Preliminary indications were that Ks and Ka values were very similar using both methods.

DISCUSSION

PHYLOGENETIC ANALYSES.—The results of computer simulation studies have shown that ML tends to be the most robust method to infer phylogenetic relationships (Kobayashi-Fukami and Tateno, 1991; Hasegawa et al., 1991), although the method takes relatively more time than other methods. MP may give erroneous results under some conditions (Hendy and Penny, 1989; DeBry,

TABLE 2. Average estimated number and standard error of nucleotide substitutions per nucleotide site between genera in a family. Data for angiosperms are drawn from DNA databases, and are available from K. Ueda upon request. For families, the number in parentheses indicates the number of genera used for this calculation. Ks = the estimated number of synonymous nucleotide substitutions per nucleotide site and standard error. Ka = the estimated number of nonsynonymous nucleotide substitutions per nucleotide site and standard error.

| Fern family | Ks | Ka | Angiosperm family | Ks | Ka |
|-----------------------|--------------|----------------|---------------------|----------------|-----------------|
| Aspleniaceae (3) | 0.18 ± 0.012 | 0.014 ± 0.0046 | Acanthaceae (10) | 0.14 ± 0.037 | 0.011 ± 0.0033 |
| Blechnaceae (4) | 0.16 ± 0.035 | 0.010 ± 0.0069 | Araceae (8) | 0.17 ± 0.088 | 0.015 ± 0.0052 |
| Cyatheaceae (2) | 0.027 | 0.0051 | Araceae (6) | 0.034 ± 0.0074 | 0.0098 ± 0.0030 |
| Dennstaedtiaceae (4) | 0.24 ± 0.10 | 0.011 ± 0.0040 | Asteraceae (26) | 0.088 ± 0.034 | 0.016 ± 0.0054 |
| Dicksoniaceae (3) | 0.23 ± 0.068 | 0.016 ± 0.0071 | Betulaceae (5) | 0.043 ± 0.019 | 0.0035 ± 0.0014 |
| Gleicheniaceae (2) | 0.29 | 0.0098 | Bromeliaceae (7) | 0.035 ± 0.010 | 0.0044 ± 0.0021 |
| Hymenophyllaceae (4) | 0.42 ± 0.086 | 0.038 ± 0.0079 | Campanulaceae (12) | 0.23 ± 0.086 | 0.017 ± 0.0070 |
| Marsileaceae (3) | 0.27 ± 0.048 | 0.012 ± 0.0033 | Caryophyllaceae (5) | 0.15 ± 0.040 | 0.0076 ± 0.0028 |
| Oleandraceae (2) | 0.21 | 0.013 | Epacridaceae (5) | 0.15 ± 0.048 | 0.016 ± 0.0040 |
| Ophioglossaceae (3) | 0.35 ± 0.24 | 0.017 ± 0.0057 | Ericaceae (12) | 0.14 ± 0.042 | 0.016 ± 0.0053 |
| Osmundaceae (2) | 0.14 | 0.02 | Fagaceae (5) | 0.11 ± 0.10 | 0.012 ± 0.0086 |
| Polypodiaceae (6) | 0.22 ± 0.058 | 0.013 ± 0.0032 | Geraniaceae (5) | 0.15 ± 0.057 | 0.0040 ± 0.0017 |
| Pteridiaceae (11) | 0.51 ± 0.15 | 0.020 ± 0.012 | Lamiaceae (7) | 0.12 ± 0.029 | 0.017 ± 0.0049 |
| Schizaeaceae (3) | 1.40 ± 0.36 | 0.029 ± 0.0078 | Loganiaceae (7) | 0.16 ± 0.052 | 0.017 ± 0.0048 |
| Thelypteridiaceae (2) | 0.21 | 0.0087 | Magnoliaceae (5) | 0.014 ± 0.011 | 0.0085 ± 0.0070 |
| Vittariaceae (2) | 0.37 | 0.03 | Nymphaeaceae (7) | 0.061 ± 0.037 | 0.0054 ± 0.0021 |
| | | | Onagraceae (7) | 0.12 ± 0.047 | 0.011 ± 0.0037 |
| | | | Poaceae (10) | 0.13 ± 0.049 | 0.017 ± 0.0074 |
| | | | Saxifragaceae (22) | 0.093 ± 0.096 | 0.0078 ± 0.0039 |
| | | | Solanaceae (16) | 0.045 ± 0.024 | 0.010 ± 0.0056 |

1992; Zharkikh and Li, 1993; Takezuki and Nei, 1993) especially when the rate of nucleotide substitution is quite high or not constant (Felsenstein and Sober, 1986). However, those studies are based on the simulation of a small number of data sets using simple models, and more studies are necessary to reveal the applicability of each method for real data sets. In this study we therefore used the three different methods (NJ, MP, and ML) to infer phylogenetic relationships, and incorporated all of the results in the following discussion.

Optimal trees recovered from all three methods of tree construction had a similar overall topology. The trees we recovered were not completely congruent with any formerly proposed phylogenetic scheme (e. g., Bower, 1928; Holtum, 1949; Wagner, 1969; Nayar, 1970; Bierhorst, 1971; Mickel, 1974; Lovis, 1977), although each scheme is at least in part consistent with our results. The phylogenetic patterns observed in our analyses are discussed here in order, from the most basal lineages to the uppermost branches.

Osmundaceae (*Osmunda* and *Plenasium*) tended to be a sister taxon to all other leptosporangiate ferns on the *rbcL* trees. This phylogenetic position was supported with less than 50% bootstrap probability in both the NJ and equally weighted MP analyses. The distinctness of the Osmundaceae from other leptosporangiate ferns has been documented using both morphological and molecular data (e.g., Gifford and Foster, 1988; Stein et al., 1992). The basal branching of the Osmundaceae is concordant with the fossil record (Stewart and Rothwell, 1993). Although a phylogenetic relationship of Osmundaceae to *Plagiogyria* has been suggested (e. g., Mickel, 1974), *rbcL* trees do not support this hypothesis. The placement of Plagiogyriaceae as a sister group to Osmundaceae in the MP tree (Fig. 2) would require 78 additional steps.

The gleichenioid ferns (Cheiropleuriaceae, Dipteridaceae, Gleicheniaceae, and Matoniaceae) and Hymenophyllaceae were basally situated in all trees (Figs. 1–6). The relationships were nearly identical to those inferred by Hasebe et al. (1994). Our NJ tree (Fig. 1) weakly supported monophyly of the gleichenioids, and intergeneric relationships were not well resolved. A relationship between the gleichenioid and polypodioid (Grammitidaceae and Polypodiaceae) ferns has been suggested (e.g., Bierhorst, 1971; Holtum, 1973; Gifford and Foster, 1988), but detailed morphological analyses by Jarrett (1980) did not support this hypothesis, suggesting instead that Polypodiaceae are among the most derived, mainly indusiate fern families. The *rbcL* analyses here (Figs. 1–6) and in Haufler and Ranker (1995) clearly support the latter hypothesis.

Stromatopteris is usually treated as a subfamily of the Gleicheniaceae (e.g., Kramer, 1990a) or as a separate family (Bierhorst, 1971). The *rbcL* analyses strongly support the monophyly of *Dicranopteris*, *Diplopterygium*, and *Stromatopteris*. Although generic relationships within the family were not inferred with high statistical confidence (Figs. 1 and 3), it appears from the *rbcL* results (Figs. 1–6) that the segregation of *Stromatopteris* as a separate family would result in a paraphyletic Gleicheniaceae *sensu stricto*. Furthermore, as shown in Tables 1 and 2, the *Ks* value between *Diplopterygium* and *Stromatopteris* (0.29) was much smaller than the average interfamilial *Ks* value (1.17 ± 0.46)

TABLE 3. Differences in the estimated numbers of synonymous nucleotide substitutions per nucleotide site (Ks) between two fern species, labeled A and B, where species C is a reference species for each Ks value. This difference for each species pair is in the column labeled Ks(A,C)–Ks(B,C), and those values indicated with an asterisk (*) are significantly different from the null hypothesis: Ks(A,C)–Ks(B,C) = 0 at the 5% level. The column labeled SpA/SpB contains the ratio of the differences in the estimated number of nucleotide substitutions between species A and species B.

| Species A | Species B | Species C | Ks(A,C)–Ks(B,C) | SpA/SpB |
|----------------------|--------------------|--------------------|-----------------|---------|
| <i>Anemia</i> | <i>Salvinia</i> | <i>Dipteris</i> | 0.011 ± 0.28 | 1.0 |
| <i>Actinostachys</i> | <i>Salvinia</i> | <i>Dipteris</i> | –0.084 ± 0.28 | 0.94 |
| <i>Lygodium</i> | <i>Salvinia</i> | <i>Dipteris</i> | –0.023 ± 0.29 | 0.98 |
| <i>Ceratopteris</i> | <i>Acrostichum</i> | <i>Doryopteris</i> | 0.048 ± 0.071 | 1.2 |
| <i>Ceratopteris</i> | <i>Pteris</i> | <i>Doryopteris</i> | 0.15 ± 0.073* | 1.6 |
| <i>Ceratopteris</i> | <i>Taenitis</i> | <i>Doryopteris</i> | 0.12 ± 0.071 | 1.5 |
| <i>Ceratopteris</i> | <i>Onychium</i> | <i>Doryopteris</i> | 0.17 ± 0.071* | 1.7 |
| <i>Ceratopteris</i> | <i>Platzzoma</i> | <i>Doryopteris</i> | 0.10 ± 0.074 | 1.5 |

for ferns and closer to the mean intergeneric Ks value (0.41 ± 0.26). Our results are therefore consistent with Kramer's (1990a) treatment of these genera in one family.

Cardiomanes has been treated in a separate subfamily from other genera in Hymenophyllaceae because it has a thicker lamina than other filmy ferns (Iwatsuki, 1990). The *rbcL* trees (Figs. 1–6) suggest that this segregation of *Cardiomanes* results in the paraphyly of the remaining genera (subfamily Hymenophylloideae). Thus, revision of subfamilial classification in Hymenophyllaceae may be necessary after more sequence data for this family are accumulated.

Schizaeaceae were the next clade to diverge in all three methods of analysis (Figs. 1, 2, and 5). As shown in Tables 1 and 2, the average Ks value among the three genera in Schizaeaceae (*Actinostachys*, *Anemia*, and *Lygodium*) was 1.4 ± 0.36 , which is higher than the average intergeneric Ks value (0.41 ± 0.26) and higher than the average interfamilial Ks value (1.17 ± 0.46) for ferns. This suggests that the genera in Schizaeaceae diverged from each other at an earlier time than did most families and genera of ferns. An alternative hypothesis is that *rbcL* has an accelerated rate of base substitution in the Schizaeaceae. However, the latter hypothesis is not supported, because the results of the relative rate tests (Wu and Li, 1985) using *Dipteris* as a reference species did not show any significant rate variation between each of the schizaeaceous lineages and *Salvinia* (Table 3). This result was also supported when we used *Calochlaena* or *Lindsaea* instead of *Salvinia* (data not shown). Thus, we interpret the high sequence divergence among schizaeaceous genera as evidence of a long evolutionary history. This is consistent with the substantial morphological divergence among these genera (Kramer, 1990b) and a long fossil history (Stewart and Rothwell, 1993). This result supports the classification in which the four genera are treated as separate families (e.g., Pichi Sermolli, 1977). Pteridaceae (Wagner, 1969; Mickel, 1974; Holttum, 1973; Lovis, 1977; Kramer, 1990c) and Marsileaceae (Bierhorst, 1971; Pichi Sermolli, 1977; Lovis, 1977) have been

postulated to have a close relationship to Schizaeaceae, but our analyses do not support these hypotheses.

Monophyly of the heterosporous water ferns was supported by morphological and fossil evidence (Rothwell and Stockey, 1994), by a previous molecular study that included *Marsilea* and *Salvinia* (Hasebe et al., 1994), and by this study, where we used all extant water fern genera (*Azolla*, *Marsilea*, *Pilularia*, *Regnellidium*, and *Salvinia*). The trees in this study (Figs. 1–4) show the same intergeneric relationships among the water ferns as those inferred by Rothwell and Stockey (1994).

The tree ferns (Cyatheaceae and Dicksoniaceae) diverged after the water ferns in most of the *rbcL* analyses (Figs. 1, 2, 4, and 5), although this order was not well supported by the bootstrap analyses for either NJ or MP analyses (Figs. 1 and 3). Monophyly of the tree ferns was also inferred by Hasebe et al. (1994) and Wolf et al. (1994). The incorporation of *rbcL* data for additional tree fern taxa in the present study provided some fresh insights into relationships within this clade. For example, *Loxoma* (Loxomataceae) and *Plagiogyria* (Plagiogyriaceae) are depicted as sister taxa within the tree fern clade in the NJ, MP and ML analyses, although bootstrap support for the monophyletic relationship between *Loxoma* and *Plagiogyria* is not strong, especially with MP. Morphological evidence that supports *Loxoma* and *Plagiogyria* as sister taxa is lacking; therefore this hypothesis requires further testing. Nevertheless, *rbcL* data provide strong support for inclusion of both *Loxoma* and *Plagiogyria* within the tree fern clade. Some morphological traits that support this relationship are sporangial annuli, spore wall, and gametophyte morphology (Atkinson, 1973; Tryon and Tryon, 1982; Tryon and Lugardon, 1990). Overall generic relationships among tree ferns, resolved by the different algorithms, were not concordant. Monophyly of *Dicksonia* and *Calochlaena* was supported by the NJ and ML methods, but not by the MP method.

Pteridaceae are a large family, sometimes subdivided into six groups (Tryon and Tryon, 1982; Tryon et al., 1990; Table 4). As in a previous study (Hasebe et al., 1994), *rbcL* data suggest that members of the Pteridaceae share a recent common ancestor. However, the Pteridaceae are not a monophyletic group because the Vittariaceae are placed in the Pteridaceae as a sister group to *Adiantum* (Figs. 1–6). This relationship did not change, even when the number of taxa in Vittariaceae was increased (Crane et al., 1995). The following five monophyletic groups were recognized with a high degree of confidence in all three tree construction methods: Group 1 = *Onychium*, *Platyzoma*, *Pteris*, and *Taenitis*; Group 2 = *Acrostichum* and *Ceratopteris*; Group 3 = *Argyrochosma*, *Bommeria*, *Cheilanthes*, *Doryopteris*, *Hemionitis*, *Notholaena*, and *Pellaea*; Group 4 = *Coniogramme*; and Group 5 = *Adiantum* and the Vittariaceae. These five monophyletic groups were partly congruent with traditional classifications (Table 4). A more detailed analysis of relationships in Group 3 is provided by Gastony and Rollo (1995).

A relationship between evolutionary rate and life history factors was demonstrated in seed plants by Bousquet et al. (1992), with *rbcL* evolving more rapidly in annual plants than in perennials. We used the relative rate test (Wu

TABLE 4. Comparison of classification systems of the Pteridaceae.

| | Tryon and Tryon (1982) | Tryon et al. (1990) | This <i>rbcl</i> study (Figs. 1-4) |
|----------------------|------------------------|---------------------|---------------------------------------|
| <i>Platyzoma</i> | Platyzomateae | Platyzomatoideae | Group 1 |
| <i>Taenitis</i> | Taenitideae | Taenitidoideae | Group 1 |
| <i>Onychium</i> | Cheilantheae | Taenitidoideae | Group 1 |
| <i>Pteris</i> | Pterideae | Pteridoideae | Group 1 |
| <i>Ceratopteris</i> | Ceratopterideae | Ceratopteridoideae | Group 2 |
| <i>Acrostichum</i> | Pterideae | Pteridoideae | Group 2 |
| <i>Argyroschisma</i> | Cheilantheae | Cheilanthoideae | Group 3 |
| <i>Pellaea</i> | Cheilantheae | Cheilanthoideae | Group 3 |
| <i>Cheilanthes</i> | Cheilantheae | Cheilanthoideae | Group 3 |
| <i>Doryopteris</i> | Cheilanthese | Cheilanthoideae | Group 3 |
| <i>Hemionitis</i> | Cheilantheae | Cheilanthoideae | Group 3 |
| <i>Notholaena</i> | Cheilantheae | Cheilanthoideae | Group 3 |
| <i>Bommeria</i> | Cheilantheae | Cheilanthoideae | Group 3 |
| <i>Contiogramme</i> | Taenitideae | Cheilanthoideae | Group 4 |
| <i>Adiantum</i> | Adiantaeae | Adiantoideae | Group 5 |

and Li, 1985) to compare *Ceratopteris* (an annual fern) to some perennial species in the Pteridaceae, using *Doryopteris* as a reference species (Table 3). When we compared the evolutionary rate of *Ceratopteris* to *Onychium* or *Pteris*, the null hypothesis of constancy in evolutionary rate in both lineages was rejected at the 5% significance level. However, the null hypothesis was not rejected for other perennial pteroid ferns (Table 3). More data from other gene sequences are necessary to determine whether the evolutionary rate of *rbcl* is accelerated in annual ferns as in annual angiosperms.

A detailed analysis of phylogenetic relationships in Dennstaedtiaceae sensu lato was provided by Wolf et al. (1994) and Wolf (1995). The taxa of Dennstaedtiaceae sampled here formed four distinct groups, but the relationships among the groups were weakly resolved. The four groups are: Group 1 = Dennstaedtiaceae sensu stricto (*Blotiella*, *Dennstaedtia*, *Histiopteris*, *Hypolepis*, *Microlepia*, *Paesia*, and *Pteridium*) and Monachosoraceae; Group 2 = *Lonchitis*; Group 3 = *Lindsaea* and *Sphenomeris*; and Group 4 = *Orthiopteris*.

The most derived indusiate ferns (Aspleniaceae, Blechnaceae, Davalliaceae, Dryopteridaceae, Grammitidaceae, Lomariopsidaceae, Nephrolepidaceae, Oleandraceae, Polypodiaceae, Thelypteridaceae) diverged after the dennstaedtioid (Dennstaedtiaceae and Monachosoraceae) and pteroid (Pteridaceae and Vittariaceae) ferns. All of our analyses are consistent with the branching of the Aspleniaceae at the base of this group. The relationships among the Blechnaceae, Thelypteridaceae, and Dryopteridaceae subfamily Athyrioideae sensu Kramer and Kato (1990) were different in the three tree construction methods.

The *rbcl* results indicate that Dryopteridaceae sensu Kramer et al. (1990) are polyphyletic, as inferred by Hasebe et al. (1994) and Wolf et al. (1994). For example, *Tectaria* does not cluster with other members of the Dryopteridaceae (also reported by Wolf et al., 1994), but shows a close relationship to the Oleandraceae. Although *Tectaria* and *Ctenitis* are usually included in the same sub-

group of Dryopteridaceae, they appear on divergent branches in all the *rbcl* trees (Figs. 1–5). This result was confirmed even when we included other species of *Tectaria* and *Ctenitis*: *T. gaudichaudii* (Wolf et al., 1994), *T. fauriei*, and *C. sinii* (Hasebe et al., unpublished data) in our analyses.

RbcL data show that the Polypodiaceae, Davalliaceae, Grammitidaceae, Oleandraceae, and Nephrolepidaceae share a recent common ancestor with some members of the Dryopteridaceae. Within the polypodioids (Grammitidaceae and Polypodiaceae), *Loxogramme* is the most basal. Additional details of relationships within the Polypodiaceae are provided in Haufler and Ranker (1995). *Loxogramme* shares several morphological characters with Grammitidaceae and Polypodiaceae, and Tryon and Tryon (1982) suggested that *Loxogramme* was derived from the common ancestor of both groups, which is supported by the *rbcl* trees (Figs. 1–5).

Pleurosoriopsis is a small epilithic fern distributed only in eastern Asia. Phylogenetic relationships of this genus are controversial and *Pleurosoriopsis* has been placed in Pteridaceae (Copeland, 1947), Aspleniaceae (Löve et al., 1977), Grammitidaceae (Tryon and Tryon, 1982) and Pleurosoriopsidaceae (Kurita and Ikebe, 1977). Our results suggest a close affinity of *Pleurosoriopsis* with the Polypodiaceae and Grammitidaceae, and support the hypothesis of Tryon and Tryon (1982) based on spore germination and morphological characters of the petiolar trichomes, spores, and gametophytes.

REPLY TO SMITH (1995).—Sixteen questions concerning fern relationships from the standpoint of non-molecular data were posed by Smith (1995, this volume). The global analyses of *rbcl* nucleotide sequence data provide responses for all of them except one. Below we list each question and the answer suggested by *rbcl* data. 1) Where goes *Saccoloma*? We analyzed *Orthiopteris*, which is often segregated from *Saccoloma* in the paleotropics. The MP and ML *rbcl* trees (Figs. 2–6) placed *Orthiopteris* basal to Dennstaedtiaceae sensu lato. Wolf (1995) added two species of *Saccoloma* to his analysis and found them to emerge within the *Hypolepis* clade (*Blotiella* to *Pteridium* in Fig. 2), but with *Orthiopteris* in the same position as we found it in the global analysis. 2) What are the relationships of *Monachosorum*? This genus is often included in the Dennstaedtiaceae and the *rbcl* analyses support this treatment. Wolf et al. (1994) and Wolf (1995) further resolved the basal position of *Monachosorum* to the *Hypolepis* clade. 3) What are the relationships of *Ceratopteris*? *Ceratopteris* is in the Pteridaceae and is most closely related to *Acrostichum*. 4) What are the affinities of *Pleurosoriopsis*? *Pleurosoriopsis* is closely allied to Grammitidaceae and Polypodiaceae. 5) What are the relationships of Hymenophyllopsidaceae? We have been unable to get living material of this family, which is endemic to the highlands of the Roraima formation in South America. 6) What are the relationships of the Hymenophyllaceae? Hymenophyllaceae are basally situated in *rbcl* trees. 7) What is the relationship, or lack thereof, between Polypodiaceae and Grammitidaceae? The members of these families used in the present study form a monophyletic group. 8) What are the relationships of Plagiogyriaceae? Plagiogyriaceae are closely related to

the tree ferns (Cyatheaceae, Dicksoniaceae, and Metaxyaceae) and appear to have an affinity with *Loxoma*. 9) What are the relationships and origin of the heterosporous fern families? The heterosporous ferns are more closely related to the most derived leptosporangiate ferns (including the tree fern group) than they are to the Schizaeaceae. Azollaceae, Salviniaceae, and Marsileaceae form a strongly supported, monophyletic group. 10) What are the relationships of Psilotaceae? *Psilotum* and *Tmesipteris* form a monophyletic group in the NJ tree (Fig. 1; tree rooted by *Angiopteris* only) and do not have a close relationship with any leptosporangiate ferns including *Stromatopteris* (contrary to Bierhorst, 1968, p.266), which is a sister taxon to *Diplopterygium*. This result also was supported when we assigned only *Angiopteris* as an outgroup in the MP and ML analyses (analysis not shown). *Psilotum* and *Tmesipteris* grouped with Ophioglossaceae in trees produced from plastid 16S (Manhart, 1995) and nuclear 18S ribosomal RNA sequences (Wolf et al., unpublished). 11) What are the relationships of Thelypteridaceae? Thelypteridaceae do not form a sister group with Cyatheaceae (Holtum and Sen, 1961) or Aspleniaceae (Nayar, 1976; Pichi Sermolli, 1977), but are closely related to the athyrioids (*Athyrium* to *Woodsia* in Fig. 1) in our analyses. 12) What are the relationships among the Schizaeaceae? Schizaeaceae form a monophyletic group. In Schizaeaceae, *Lygodium* is sister to *Actinostachys* and *Anemia*. 13) Are the Pteridaceae (and Vittariaceae) most closely related to or derived from the Schizaeaceae, or are they more closely allied to the higher leptosporangiate ferns? Pteridaceae diverged from Dennstaedtiaceae *sensu lato* and are not among the most derived clades of indusiate, leptosporangiate ferns; nor are they closely related to Schizaeaceae. 14) Are the Dennstaedtiaceae allied with the Dicksoniaceae or are they part of a higher leptosporangiate clade, ancestral to, or an early offshoot from the line leading to the dryopteroids, asplenioids, and blechnoids? Dennstaedtiaceae are polyphyletic with some taxa diverging after the tree ferns, and the rest forming a monophyletic clade closely related to the most derived indusiate ferns. 15) What is a reasonable circumscription of Dryopteridaceae? Dryopteridaceae are polyphyletic and share a common ancestor with the Aspleniaceae and Oleandraceae (Fig. 1). Dryopteridaceae *sensu lato* perhaps should be divided after more detailed analyses. 16) Do the higher leptosporangiate families, including Aspleniaceae, Blechnaceae, Davalliaceae, Dennstaedtiaceae *sensu lato*, Dryopteridaceae, Grammitidaceae, Pteridaceae, and Polypodiaceae form a clade? All together, these taxa form a well-supported, monophyletic group.

KS AND KA VALUES.—Ks and Ka values are useful for comparing the variation in nucleotide substitutions per nucleotide site among lineages. Average Ks and Ka values between families and between genera within families were significantly higher in ferns than in angiosperms (Table 1). There are at least two explanations for this: 1) fern genera and families are older than those of angiosperms, on average, and have therefore accumulated more nucleotide substitutions, or 2) nucleotide substitution rates are accelerated in ferns (Hasebe et al., 1993). To choose between these will require inferring the correct phy-

logenetetic relationships among land plants, identifying an appropriate outgroup for the ferns, and then applying the relative rate test (Wu and Li, 1985; Muse and Weir, 1992).

FUTURE PROSPECTS.—Global analyses of *rbcL* sequences in leptosporangiate ferns have provided possible answers to many questions in fern systematics. So far, we have extensive data for only one plastid gene. Inferences from this study must be tested with non-molecular data and also with more conserved or more rapidly evolving regions of the fern genome. A comparison of trees obtained from both *rbcL* and non-molecular data is reported elsewhere in this volume (Pryer et al., 1995). Some fern plastid genomes have been shown to be maternally inherited (Gastony and Yatskievych, 1992). This may lead to conflicts between *rbcL* phylogenies and those based on biparentally inherited, nuclear-encoded, gene sequence data, although the possibility of conflicts at the interfamilial level, where hybridization has not been reported in ferns, is much lower than at the infrageneric or intergeneric levels.

One of the most limiting factors in this type of study is data analysis. Computational speed is unlikely to be improved by many orders of magnitude in the near future and thus a more efficient approach may be to examine smaller data sets, although this can lead to the problem of long branch attraction, especially when using maximum parsimony analysis (Felsenstein, 1978; Hendy and Penny, 1989; Huelsenbeck and Hillis, 1993). Selection of tree construction methods is another problem that has not been fully studied. In our analyses, the results of three different methods gave almost concordant results for relationships among ferns, giving us confidence in our results.

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APPENDIX. Pteridophyte *rbcL* sequence data compiled for studies in this issue of the American Fern Journal. These are arranged alphabetically by family, subfamily, and genus. Taxonomy of families follows Kramer and Green (1990). Taxa used for the calculation of the expected number of synonymous and nonsynonymous nucleotide substitutions per nucleotide site (Ks and Ka, respectively) are indicated in a column labeled Ks/Ka.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/ Ka | Figures in this study |
|--------------------|---|--|-----------------------------|---------------------|--------------------------|--------|-----------------------|
| Aspleniaceae | <i>Asplenium antiquum</i> Makino | Japan, Tokyo | <i>Murakami 596901</i> (TI) | U30596 | This study (Murakami) | * | 1,2,3,4,5,6 |
| Aspleniaceae | <i>Asplenium filipes</i> Copel. | Japan, Kagoshima | <i>Murakami 596902</i> (TI) | U30605 | Hasebe et al., 1994 | | |
| Aspleniaceae | <i>Asplenium griffithianum</i> Hook. | Japan, Kagoshima | <i>Murakami 596903</i> (TI) | | Murakami et al., unpubl. | | |
| Aspleniaceae | <i>Asplenium incisum</i> Thunb. | Japan, Mie | <i>Murakami 596904</i> (TI) | | Murakami et al., unpubl. | | |
| Aspleniaceae | <i>Asplenium nidus</i> L. | Laos, Vientiane | <i>Murakami 596905</i> (TI) | | Murakami et al., unpubl. | | |
| Aspleniaceae | <i>Asplenium nidus</i> L. | U.S.A., Guam | MBG 896449 (UTC) | U05907 | Wolf et al., 1994 | | |
| Aspleniaceae | <i>Asplenium normale</i> D. Don | Japan, Mie | <i>Murakami 596906</i> (TI) | | Murakami et al., unpubl. | | |
| Aspleniaceae | <i>Asplenium normale</i> D. Don var. <i>boreale</i> Ohwi ex Kurata | Japan, Wakayama | <i>Murakami 596907</i> (TI) | | Murakami et al., unpubl. | | |
| Aspleniaceae | <i>Asplenium normale</i> D. Don var. <i>shimurae</i> H. Ito | Japan, Mie | <i>Murakami 596908</i> (TI) | | Murakami et al., unpubl. | | |
| Aspleniaceae | <i>Asplenium oligophlebium</i> Baker | Japan, Mie | <i>Murakami 596909</i> (TI) | | Murakami et al., unpubl. | | |
| Aspleniaceae | <i>Asplenium pekinense</i> Hance | Japan, Wakayama | <i>Murakami 596910</i> (TI) | | Murakami et al., unpubl. | | |
| Aspleniaceae | <i>Asplenium prolongatum</i> Hook. | Japan, Mie | <i>Murakami 596911</i> (TI) | | Murakami et al., unpubl. | | |
| Aspleniaceae | <i>Asplenium ritoense</i> Hayata | Japan, Wakayama | <i>Murakami 596912</i> (TI) | | Murakami et al., unpubl. | | |
| Aspleniaceae | <i>Asplenium ruprechtii</i> Kurata (= <i>Camptosorus sibiricus</i> Rupr.) | Japan, Toyama | <i>Murakami 596918</i> (TI) | U30606 | This study (Murakami) | * | 1,2,3,4 |

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data- base acces- sion | Literature citation | Ks/ Ka | Figures in this study |
|-----------------------|--|---|--|---------------------------------|-------------------------------|-----------|--------------------------|
| Aspleniaceae | <i>Asplenium serellii</i> Hook. | Japan, Mie | <i>Murakami 596913</i> (TI) | | Murakami et al., un- publ. | | |
| Aspleniaceae | <i>Asplenium trichomanes</i> L. | Japan, Mie | <i>Murakami 596914</i> (TI) | | Murakami et al., un- publ. | | |
| Aspleniaceae | <i>Asplenium tripteropus</i> Nakai | Japan, Wakayama | <i>Murakami 596915</i> (TI) | | Murakami et al., un- publ. | | |
| Aspleniaceae | <i>Asplenium wilfordii</i> Mett. ex Kuhn | Japan, Mie | <i>Murakami 596916</i> (TI) | | Murakami et al., un- publ. | | |
| Aspleniaceae | <i>Asplenium wrightii</i> D.C. Ea- ton ex Hook. | Japan, Mie | <i>Murakami 596917</i> (TI) | | Murakami et al., un- publ. | | |
| Aspleniaceae | <i>Phyllitis scolopendrium</i> (L.) Newn. | Japan, Fukushima | <i>Hasebe 26544</i> (TI) | U30607 | Hasebe et al., 1994 | * | 1,2,3,4 |
| Azollaceae | <i>Azolla caroliniana</i> Willd. | U.S.A., cultivated | <i>Pryer 959a</i> (DUKE) | U24185 | This study (Pryer) | * | 1,2,3,4,5,6 |
| Blechnaceae | <i>Blechnum orientale</i> L. | Japan, Kagoshima | TI698 (TI) | U05606 | Hasebe et al., 1994 | * | 1,2,3,4 |
| Blechnaceae | <i>Blechnum occidentale</i> L. | U.S.A., Hawaii | <i>Wolf & Anderson-Wong</i> 242 (UTC) | U05909 | Wolf et al., 1994 | | |
| Blechnaceae | <i>Blechnum brasiliense</i> Desv. | Unknown | MBG 801368 (UTC) | U05910 | Wolf et al., 1994 | | |
| Blechnaceae | <i>Doodia maxima</i> J. Sm. | Unknown | MBG U508 (UTC) | U05921 | Wolf et al., 1994 | * | 1,2,3,4,5,6 |
| Blechnaceae | <i>Doodia media</i> R. Br. | Unknown | MBG 801282 (UTC) | U05922 | Wolf et al., 1994 | | |
| Blechnaceae | <i>Sadleria pallida</i> Hook. & Arm. | U.S.A., Hawaii | <i>Ranker 1287</i> (COLO) | U05943 | Wolf et al., 1994 | * | 1,2,3,4 |
| Blechnaceae | <i>Woodwardia fimbriata</i> Sm. | U.S.A., California | MBG 897131 (UTC) | U05950 | Wolf et al., 1994 | | |
| Blechnaceae | <i>Stenochlaena tenuifolia</i> (Desv.) Moore | Unknown | MBG 840392 (UTC) | U05945 | Wolf et al., 1994 | | |

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/Ka | Figures in this study |
|-------------------------------------|---|--|---|---------------------|---------------------|-------|-----------------------|
| Blechnaceae (Stenochlaenoideae) | <i>Stenochlaena palustris</i> (Burm.) Bedd. | Singapore, Singapore | <i>Hasebe 27701</i> (TI) | U05652 | Hasebe et al., 1994 | * | 1,2,3,4 |
| Cheitroleuriaceae | <i>Cheitroleuria bicuspis</i> (Blume) C. Presl | Japan, Okinawa | <i>Yokoyama 27619</i> (TI) | U05607 | Hasebe et al., 1994 | * | 1,2,3,4 |
| Cyatheaceae | <i>Cyathea lepifera</i> (J. Sm. ex Hook.) Copel. (= <i>Sphaeropteris lepifera</i> (J. Sm. ex Hook.) R. Tryon) | Japan, Okinawa | <i>Yokoyama 5176</i> (TI) | U05616 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Cyatheaceae | <i>Cyathea</i> sp. | U.S.A., Hawaii, cultivated | <i>Wolf & Anderson-Wong 256</i> (UTC) | U05914 | Wolf et al., 1994 | * | 1,2,3,4 |
| Cyatheaceae | <i>Sphaeropteris cooperi</i> (F. Muell.) R. Tryon | Australia | MBG 894468 (UTC) | U05944 | Wolf et al., 1994 | * | 1,2,3,4 |
| Davalliaceae | <i>Davallia denticulata</i> (Burm. f.) Mett. | Unknown | MBG 896474 (UTC) | U05917 | Wolf et al., 1994 | * | 1,2,3,4,5,6 |
| Davalliaceae | <i>Davallia mariesii</i> T. Moore ex Baker | Japan, Jukushima | <i>Hasebe 26557</i> (TI) | U05617 | Hasebe et al., 1994 | | |
| Dennstaedtiaceae (Dennstaedtiodeae) | <i>Blotiella pubescens</i> (Kaulf.) R. Tryon | La Réunion | <i>Strasberg s.n.</i> (REU, UTC) | U05911 | Wolf et al., 1994 | | 1,2,3,4 |
| Dennstaedtiaceae (Dennstaedtiodeae) | <i>Coptodipteris wilfordii</i> (Moore) Nakai & Momose | Japan, Mt. Fuji | <i>Togashi 360</i> (UC) | U18635 | Wolf, 1995 | | |
| Dennstaedtiaceae (Dennstaedtiodeae) | <i>Dennstaedtia punctilobula</i> (Michx.) Moore | U.S.A., Vermont | <i>Paris s.n.</i> (UTC) | U05918 | Wolf et al., 1994 | * | 1,2,3,4,5,6 |
| Dennstaedtiaceae (Dennstaedtiodeae) | <i>Dennstaedtia samoensis</i> (Brack.) Moore | Vauatu | Kew 1971-5068 (UTC) | U18637 | Wolf, 1995 | | |
| Dennstaedtiaceae (Dennstaedtiodeae) | <i>Dennstaedtia auriculata</i> (Nalvarrete) | Ecuador | <i>Navarrete s.n.</i> (QCA) | U18636 | Wolf, 1995 | | |
| Dennstaedtiaceae (Dennstaedtiodeae) | <i>Histiopteris incisa</i> (Thumb.) J. Sm. | Australia, Queensland | <i>Kato et al. 237</i> (TI) | U05627 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/ Ka | Figures in this study |
|---------------------------------------|---|--|---|---------------------|---------------------|-----------|-----------------------|
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Histiopteris incisa</i> (Thunb.) J. Sm. | New Caledonia | UCBG 57.0776 (UTC) | U05926 | Wolf, 1995 | | |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Hypolepis aspidioides</i> Christ | Western Samoa | Kew #1965-58402 (UTC) | U18634 | Wolf, 1995 | | |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Hypolepis hostilis</i> (Kunze) C. Presl | Ecuador | <i>Navarrete s.n.</i> (QCA) | U18638 | Wolf, 1995 | | |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Hypolepis muelleri</i> Wakef. | Australia | <i>Thomson 189</i> (UTC) | U05927 | Wolf, 1995 | | |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Hypolepis punctata</i> (Thunb.) Mett. | Japan, Nara | <i>Hasebe 27605</i> (TI) | U05628 | Hasebe et al., 1994 | * | 1, 2, 3, 4, 5, 6 |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Leptolepia novae-zelandiae</i> (Col.) Mett. ex Diels | New Zealand | <i>Parris 11713</i> (UTC) | U18639 | Wolf, 1995 | | |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Lonchitis hirsuta</i> L. | Puerto Rico | <i>Axelrod 4221</i> (UPRRP, UTC) | U05929 | Wolf et al., 1994 | * | 1, 2, 3, 4, 5, 6 |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Lonchitis mannii</i> (Baker) Alston | Cameroon | <i>Thomas 2764</i> (UC) | U18641 | Wolf, 1995 | | |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Microlepia strigosa</i> (Thunb.) C. Presl | U.S.A., Hawaii | <i>Wolf & Anderson-Wong 249</i> (UTC) | U05931 | Wolf, 1995 | | 1, 2, 3, 4 |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Microlepia szechuanica</i> Chung | Unknown | From spores (UTC) | U18643 | Wolf, 1995 | | |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Microlepia platyphylla</i> (Don) J. Sm. | China | From spores (UTC) | U18642 | Wolf, 1995 | | |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Paesia scaberula</i> (Rich.) Kuhn | Unknown | From spores (UTC) | U05937 | Wolf et al., 1994 | | 1, 2, 3, 4, 5, 6 |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Pteridium aquilinum</i> (L.) Kuhn | U.S.A., Idaho | <i>Wolf 237</i> (UTC) | U05939 | Wolf, 1995 | | |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Pteridium aquilinum</i> (L.) Kuhn | Japan, Tochigi | <i>Hasebe 27606</i> (TI) | U05646 | Hasebe et al., 1994 | * | 1, 2, 3, 4, 5, 6 |
| Dennstaedtiaceae (Dennstaedtioidaeae) | <i>Pteridium esculentum</i> (G. Forst.) Cockayne | Australia | UCBG 56.0657 (UTC) | U05940 | Wolf, 1995 | | |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/Ka | Figures in this study |
|-------------------------------------|--|--|------------------------|---------------------|---------------------|-------|-----------------------|
| Dennstaedtiaceae (Lindsaeoideae) | <i>Lindsaea ensifolia</i> Sw. | U.S.A., Hawaii | Palmer s.n. (UTC) | U05928 | Wolf, 1995 | | |
| Dennstaedtiaceae (Lindsaeoideae) | <i>Lindsaea parasitica</i> (Roxb. ex Griffith) Hieron. | Brunei | Kew #1991-1934 (UTC) | U18640 | Wolf, 1995 | | |
| Dennstaedtiaceae (Lindsaeoideae) | <i>Lindsaea odorata</i> Roxb. | Japan, Okinawa | Hasebe & Lin C106 (TI) | U05630 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Dennstaedtiaceae (Lindsaeoideae) | <i>Odontosoria chinensis</i> (L.) J. Sm. (= <i>Sphenomeris chinensis</i> (L.) Maxon) | U.S.A., Hawaii | Ranker 1231 (COLO) | U05934 | Wolf, 1995 | | 1,2,3,4 |
| Dennstaedtiaceae (Lindsaeoideae) | <i>Odontosoria chinensis</i> (L.) J. Sm. (= <i>Sphenomeris chinensis</i> (L.) Maxon) | Japan, Okinawa | Hasebe 245B (TI) | U05651 | Hasebe et al., 1994 | * | |
| Dennstaedtiaceae (Lindsaeoideae) | <i>Odontosoria scandens</i> (Desv.) C. Chr. | Puerto Rico | Axelrod 5353 (UPRRP) | U05935 | Wolf, 1995 | | |
| Dennstaedtiaceae (Lindsaeoideae) | <i>Tapetidium melanesicum</i> Kramer | Fiji | Smith 9382 (UC) | U18647 | Wolf, 1995 | | |
| Dennstaedtiaceae (Lindsaeoideae) | <i>Tapetidium luzonicum</i> (Hook.) Kramer | Taiwan | Huang & Kao 7525 (UC) | U18646 | Wolf, 1995 | | |
| Dennstaedtiaceae (Saccolomatoideae) | <i>Orthopteris kingii</i> (Bedd.) Holttum | Indonesia, Bogor | Hasebe 27604 (TI) | U18644 | Wolf, 1995 | | 1,2,3,4,5,6 |
| Dennstaedtiaceae (Saccolomatoideae) | <i>Saccoloma moluccanum</i> (Bl.) Mett. | Philippines | Emer 9067 (VT) | U18649 | Wolf, 1995 | | |
| Dennstaedtiaceae (Saccolomatoideae) | <i>Saccoloma elegans</i> Kaulf. | Ecuador | Barrington 1064 (VT) | U18645 | Wolf, 1995 | | |
| Dicksoniaceae | <i>Calochlaena dubia</i> (R.Br.) M.D. Turner & R.A. White | Australia, Queensland | Kato et al. 201 (TI) | U05615 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/Ka | Figures in this study |
|---------------------------------|---|--|---------------------------|---------------------|------------------------|-------|-----------------------|
| Dicksoniaceae | <i>Calochlaena villosa</i> (C. Chr.) M.D. Turner & R.A. White | Papua N.G. | Kew #1974-94 (UTC) | U05912 | Wolf et al., 1994 | | |
| Dicksoniaceae | <i>Culcita conifolia</i> (Hook.) Maxon | Costa Rica | Barrington s.n. (UTC) | U18648 | Wolf, 1995 | | |
| Dicksoniaceae | <i>Cibotium barometz</i> (L.) J. Sm. | Japan, Kagoshima | TI1288 (TI) | U05610 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Dicksoniaceae | <i>Cibotium glaucum</i> (J. Sm.) Hook. & Arn. | U.S.A., Hawaii | SA 82-159 (UTC) | U05913 | Wolf et al., 1994 | | |
| Dicksoniaceae | <i>Dicksonia antarctica</i> Labill. | Australia, Queensland | Hasebe 27615 (TI) | U05618 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Dicksoniaceae | <i>Dicksonia antarctica</i> Labill. | Australia | MBG 830474 (-2) (UTC) | U05919 | Wolf et al., 1994 | | |
| Dipteridaceae | <i>Dipteris conjugata</i> Reinw. | Japan, Okinawa | Hasebe 27618 (TI) | U05620 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Dryopteridaceae (Dryopterideae) | <i>Arachniodes aristata</i> (G. Forst.) Tindale | Japan, Mie | Yokoyama 5170 (TI) | U30608 | This study (Yokoyama) | * | 1,2,3,4 |
| Dryopteridaceae (Dryopterideae) | <i>Cyrtium hookerianum</i> (Pr.) C. Chr. | Japan, Oh-ita | Yokoyama 5169 (TI) | | Hasebe et al., unpubl. | | |
| Dryopteridaceae (Dryopterideae) | <i>Dryopteris cristata</i> (L.) A. Gray | U.S.A. | MBG 791348 (UTC) | U05923 | Wolf et al., 1994 | * | 1,2,3,4,5,6 |
| Dryopteridaceae (Dryopterideae) | <i>Dryopteris dickinsii</i> (Fr. & Sav.) C. Chr. | Japan, Kumamoto | Hasebe 26555 (TI) | U05622 | Hasebe et al., 1994 | | |
| Dryopteridaceae (Dryopterideae) | <i>Polystichum munitum</i> (Kaulf.) C. Presl | U.S.A., Idaho | Soltis & Soltis s.n. (WS) | U05938 | Wolf et al., 1994 | | |
| Dryopteridaceae (Dryopterideae) | <i>Polystichum tripterum</i> (Kunze) C. Presl | Japan, Mie | Yokoyama 5171 (TI) | U30832 | This study (Yokoyama) | | 1,2,3,4,5,6 |
| Dryopteridaceae (Onocleaceae) | <i>Matteuccia struthiopteris</i> (L.) Todaro | U.S.A., cultivated | Wolf 464 (UTC) | U05930 | Wolf et al., 1994 | * | 1,2,3,4,5,6 |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/Ka | Figures in this study |
|----------------------------------|--|--|--------------------------|---------------------|----------------------|-------|-----------------------|
| Dryopteridaceae (Onocleaceae) | <i>Matteuccia struthiopteris</i> (L.) Todaro | Japan, Fukushima | <i>Sano 16</i> (CBM) | D43917 | Sano et al., unpubl. | | |
| Dryopteridaceae (Onocleaceae) | <i>Onoclea sensibilis</i> L. | Japan, Fukushima | <i>Hasebe 26565</i> (TI) | U05640 | Hasebe et al., 1994 | * | 1,2,3,4 |
| Dryopteridaceae (Onocleaceae) | <i>Onoclea sensibilis</i> L. | U.S.A. | MBG 791216 (UTC) | U05936 | Wolf et al., 1994 | | |
| Dryopteridaceae (Onocleaceae) | <i>Woodsia obtusa</i> (Spreng.) Torr. | U.S.A., Missouri | <i>Raven 27640</i> (UTC) | U05949 | Wolf et al., 1994 | | |
| Dryopteridaceae (Onocleaceae) | <i>Woodsia polystichoides</i> D.C. Eaton | Japan, Fukushima | <i>Hasebe 27602</i> (TI) | U05657 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Dryopteridaceae (Physenatiaceae) | <i>Athyrium filix-femina</i> (L.) Roth ex Mertens | unknown | MBG 871825 (UTC) | U05908 | Wolf et al., 1994 | * | 1,2,3,4,5,6 |
| Dryopteridaceae (Physenatiaceae) | <i>Athyrium niponicum</i> (Mett.) Hance | Japan, Tokyo | <i>Sano 38</i> (CBM) | D43891 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physenatiaceae) | <i>Athyrium shearerii</i> (Bak.) Ching | Japan, Aichi | <i>Sano 41</i> (CBM) | D43892 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physenatiaceae) | <i>Athyrium vidalii</i> (Miq.) Koidz. | Japan, Chiba | <i>Sano 25</i> (CBM) | D43893 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physenatiaceae) | <i>Athyrium yokoscense</i> (Fr. & Sav.) H. Christ | Chiba, Japan | <i>Sano 22</i> (CBM) | D43894 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physenatiaceae) | <i>Cornopteris crenulatoserrulata</i> (Makino) Nakai | Japan, Fukushima | <i>Sano 19</i> (CBM) | D43896 | This study (Sano) | * | 1,2,3,4 |
| Dryopteridaceae (Physenatiaceae) | <i>Cornopteris decurrens-alata</i> (Hook.) Nakai | Japan, Nagasaki | <i>Sano 18</i> (CBM) | D43897 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physenatiaceae) | <i>Cystopteris fragilis</i> (L.) Bernh. | U.S.A., Utah | <i>Taye 2755</i> (UTC) | U05916 | Wolf et al., 1994 | * | 1,2,3,4,5,6 |
| Dryopteridaceae (Physenatiaceae) | <i>Deparia bonincola</i> (Nakai) M. Kato | Japan, Tokyo | <i>Sano 12</i> (CBM) | D43899 | This study (Sano) | * | 1,2,3,4,5,6 |
| Dryopteridaceae (Physenatiaceae) | <i>Deparia conilii</i> (Fr. & Sav.) M. Kato | Japan, Kagoshima | <i>Sano 32</i> (CBM) | D43901 | Sano et al., unpubl. | | |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data- base access- sion | Literature citation | Ks/ Ka | Figures in this study |
|-------------------------------------|--|---|---------------------------|----------------------------------|----------------------|-----------|--------------------------|
| Dryopteridaceae (Physematiaceae) | <i>Deparia fenzliana</i> (Luerss.) M. Kato | U.S.A., Hawaii | <i>Sano 14</i> (CBM) | D43900 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Deparia kiusiana</i> (Koidz.) M. Kato | Japan, Oh-ita | <i>Sano 13</i> (CBM) | D43902 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Deparia okuboana</i> (Makino) M. Kato | China, Yun-nan | <i>Sano 21</i> (CBM) | D43903 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Deparia otomasui</i> (Kurata) Serizawa | Japan, Kumamoto | <i>Sano 28</i> (CBM) | D43904 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Deparia petersenii</i> (Kunze) M. Kato | New Zealand, Auckland | <i>Sano 10</i> (CBM) | D43905 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Deparia prolifera</i> (Kaulf.) Hook. & Grev. | U.S.A., Hawaii | <i>Sano 15</i> (CBM) | D43906 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Deparia pycnosora</i> (H. Christ) M. Kato | Japan, Tochigi | <i>Sano 26</i> (CBM) | D43907 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Deparia viridifrons</i> (Maki- no) M. Kato | Japan, Chiba | <i>Sano 24</i> (CBM) | D43908 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Diplazium cavalerianum</i> (Christ) M. Kato | Japan, Chiba | <i>Sano 11</i> (CBM) | D43909 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Diplazium donianum</i> (Mett.) Tard. | Japan, Kagoshima | <i>Sano 29</i> (CBM) | D43911 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Diplazium esculentum</i> (Retz.) Sw. | Japan, Kagoshima | <i>Hasebe 276001</i> (TI) | U05619 | Hasebe et al., 1994 | * | 1, 2, 3, 4, 5, 6 |
| Dryopteridaceae (Physematiaceae) | <i>Diplazium lonchophyllum</i> Kunze | Unknown | MBG 903385 (UTC) | U05920 | Wolf et al., 1994 | | |
| Dryopteridaceae (Physematiaceae) | <i>Diplazium mesosorum</i> (Ma- kino) Koidz. | Japan, Tochigi | <i>Sano 44</i> (CBM) | D43910 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Diplazium squamigerum</i> (Mett.) Matsum. | Japan, Saitama | <i>Sano 20</i> (CBM) | D43912 | Sano et al., unpubl. | | |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/Ka | Figures in this study |
|----------------------------------|---|--|---|---------------------|------------------------|-------|-----------------------|
| Dryopteridaceae (Physematiaceae) | <i>Diplazium subsinuatum</i> (Wall. ex Hook. & Grev.) Tagawa | Japan, Kagoshima | <i>Sano 30</i> (CBM) | D43913 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Diplazium tomitarocnum</i> Masam. | Japan, Kagoshima | <i>Sano 43</i> (CBM) | D43914 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Diplazium wichurae</i> (Mett.) Diels | Japan, Shizuoka | <i>Sano 13</i> (CBM) | D43915 | Sano et al., unpubl. | | |
| Dryopteridaceae (Physematiaceae) | <i>Gymnocarpium dryopteris</i> (L.) Newman | U.S.A., Idaho | <i>Wolf 238</i> (UTC) | U05925 | Wolf et al., 1994 | * | 1,2,3,4,5,6 |
| Dryopteridaceae (Physematiaceae) | <i>Gymnocarpium oyamense</i> (Baker) Ching | Japan, Saitama | <i>Hasebe 483</i> (TI) | U05626 | Hasebe et al., 1994 | | |
| Dryopteridaceae (Physematiaceae) | <i>Hypodematum crenatum</i> (Forssk.) Kuhn subsp. <i>fau-rieti</i> (Kodama) K. Iwats. | Japan, Saitama | <i>Sano 17</i> (CBM) | D43916 | This study (Sano) | * | 1,2,3,4,5,6 |
| Dryopteridaceae (Rumohreae) | <i>Rumohra adiantiformis</i> (G. Forst.) Ching | Australia, Victoria | <i>Hasebe & Nishida 5200</i> (TI) | U05648 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Dryopteridaceae (Rumohreae) | <i>Rumohra adiantiformis</i> (G. Forst.) Ching | La Réunion | <i>Strausberg s.n.</i> (REU) | U05942 | Wolf et al., 1994 | | |
| Dryopteridaceae (Tectarieae) | <i>Ataxipteris sinii</i> (Ching) Holttum | Japan, Kagoshima | <i>Sano 40</i> (CBM) | D43898 | Hasebe et al., unpubl. | | |
| Dryopteridaceae (Tectarieae) | <i>Ctenitis eatonii</i> (Baker) Ching | Taiwan, Nantoh | <i>Hasebe 27600</i> (TI) | U05614 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Dryopteridaceae (Tectarieae) | <i>Lastreopsis munita</i> (Mett.) Tindale | Australia, New South Wales | <i>Kato et al. 244</i> (TI) | | Hasebe et al., unpubl. | | |
| Dryopteridaceae (Tectarieae) | <i>Tectaria devexa</i> (Kunze) Copel. | Japan, Okinawa | <i>Sano 50</i> (CBM) | D43918 | This study (Yokoyama) | * | 1,2,3,4,5,6 |
| Dryopteridaceae (Tectarieae) | <i>Tectaria gaudichaudii</i> (Mett.) Maxon | U.S.A., Hawaii | <i>Wolf 268 & Anderson-Wong</i> (UTC) | U05946 | Wolf et al., 1994 | | |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/ Ka | Figures in this study |
|---------------------------------------|---|--|---|---------------------|-----------------------|-----------|-----------------------|
| Equisetaceae | <i>Equisetum arvense</i> L. | U.S.A., Michigan | <i>Manhart 05/29/87-1</i> (TAMU) | L11053 | Manhart, 1994 | | |
| Gleicheniaceae (Gleichenioideae) | <i>Dicranopteris linearis</i> (Burm. f.) Underw. | U.S.A., Hawaii | <i>Wolf 255 & Anderson-Wong</i> (UTC) | U18626 | Wolf, 1995 | | 1,2,3,4,5,6 |
| Gleicheniaceae (Gleichenioideae) | <i>Diplazium glaucum</i> (Houtt.) Nakai | China, Yun-nan | <i>Murakami 27621</i> (TI) | U05624 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Gleicheniaceae (Stromatopteridoideae) | <i>Stromatopteris moniliformis</i> Mett. | New Caledonia | <i>Hasebe 27621</i> (TI) | U05653 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Grammitidaceae | <i>Micropolypodium okuboi</i> (Yatabe) Hayata | Japan, Kagoshima | <i>Hasebe 26575</i> (TI) | U05658 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Hymenophyllaceae (Cardiomanioideae) | <i>Cardiomanes reniforme</i> (G. Forst.) C. Presl | New Zealand, Waitomo | <i>Yokoyama 5174</i> (TI) | U30833 | This study (Yokoyama) | * | 1,2,3,4,5,6 |
| Hymenophyllaceae (Hymenophylloideae) | <i>Cephalomanes thysanostomum</i> (Makino) K. Iwats. | Japan, Okinawa | <i>Hasebe 26549</i> (TI) | U05608 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Hymenophyllaceae (Hymenophylloideae) | <i>Crepidomanes birmanicum</i> (Bedd.) K. Iwats. | Japan, Tokyo | <i>Hasebe 27622</i> (TI) | U05613 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Hymenophyllaceae (Hymenophylloideae) | <i>Gonocormus minutus</i> van den Bosch (= <i>Crepidomanes minutus</i> (Blume) K. Iwats.) | Japan, Tokyo | <i>Hasebe 27623</i> (TI) | U05625 | Hasebe et al., 1994 | * | 1,2,3,4 |
| Hymenophyllaceae (Hymenophylloideae) | <i>Fymenophyllum fucoides</i> (Sw.) Sw. | Costa Rica | <i>Crane 930613-10</i> (ISC) | U20933 | Crane et al., 1995 | | 1,2,3,4 |
| Hymenophyllaceae (Hymenophylloideae) | <i>Vandenboschia davallioides</i> (Gaud.) Copel. | U.S.A., Hawaii | <i>Wolf 248 & Anderson-Wong</i> (UTC) | U05948 | Wolf, 1995 | | 1,2,3,4 |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/ Ka | Figures in this study |
|--------------------|---|--|---------------------------|---------------------|------------------------|-----------|-----------------------|
| Isoetaceae | <i>Isoetes melanopoda</i> Gay & Durieu | U.S.A., Texas | Manhart 03/10/88-1 (TAMU) | L11054 | Manhart, 1994 | | |
| Lomariopsidaceae | <i>Elaphoglossum hybridum</i> (Bory) Moore | La Réunion | T. Ranker 1375 (COLO) | U05924 | Wolf et al., 1994 | * | 1,2,3,4,5,6 |
| Lomariopsidaceae | <i>Elaphoglossum yoshinagae</i> (Yatabe) Makino | Japan, Kagoshima | Hasebe 26559 (TI) | U05623 | Hasebe et al., 1994 | | |
| Loxomataceae | <i>Loxoma cunninghamii</i> Cunn. | New Zealand, New Plymouth | Hasebe 939 (TI) | U30834 | This study (Yokoyama) | * | 1,2,3,4,5,6 |
| Lycopodiaceae | <i>Lycopodium digitatum</i> Dill. ex A. Braun | U.S.A., Michigan | Manhart 06/12/88 (TAMU) | L11055 | Manhart, 1994 | | |
| Marattiaceae | <i>Angiopteris evecta</i> (G. Forst.) Hoffm. | U.S.A., Hawaii | Nagata 12/20/88 (TAMU) | L11052 | Manhart, 1994 | | 1,2,3,4,5,6 |
| Marattiaceae | <i>Angiopteris lygodifolia</i> Ros. | Japan, Shizuoka | no information | X58429 | Yoshinaga et al., 1992 | | |
| Marsileaceae | <i>Marsilea macropoda</i> Engelm. ex A. Braun | U.S.A., Alabama | Pryer 959 (DUKE) | U24263 | This study (Pryer) | * | 1,2,3,4,5,6 |
| Marsileaceae | <i>Marsilea quadrifolia</i> L. | Japan, Yamaguchi | Hasebe 26546 (TI) | U05633 | Hasebe et al., 1994 | | |
| Marsileaceae | <i>Marsilea quadrifolia</i> L. | U.S.A., cultivated | Manhart 07/31/91-2 (TAMU) | L13480 | Manhart, 1994 | | |
| Marsileaceae | <i>Pilularia americana</i> A. Br. | U.S.A., Georgia | Pryer 978 (DUKE) | U24263 | This study (Pryer) | * | 1,2,3,4 |
| Marsileaceae | <i>Regnellidium diphyllum</i> Lindm. | U.S.A., cultivated | Pryer 977 (DUKE) | U24262 | This study (Pryer) | * | 1,2,3,4 |
| Matoniaceae | <i>Matonia pectinata</i> R. Br. | Malaysia, Selangor | Hasebe 27620 (TI) | U05634 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Metaxiaceae | <i>Metaxya rostrata</i> (Kunth) C. Presl | Costa Rica | Conant s.n. (LSC) | U05635 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Monachosoraceae | <i>Monachosorum arakii</i> Tagawa | Japan, Mie | Hasebe 590 (TI) | U05636 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Monachosoraceae | <i>Monachosorum henryi</i> H. Christ | Taiwan | Moran 5461 (HAST. MO. F) | U05932 | Wolf et al., 1994 | | |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/Ka | Figures in this study |
|--------------------|---|--|----------------------------------|---------------------|---------------------|-------|-----------------------|
| Nephrolepidaceae | <i>Nephrolepis cordifolia</i> (L.) C. Presl | Japan (TI), cultivated | <i>Hasebe 26556</i> (TI) | U05637 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Nephrolepidaceae | <i>Nephrolepis cordifolia</i> (L.) C. Presl | cultivated | MBG 834344 (UTC) | U05933 | Wolf et al., 1994 | | |
| Nephrolepidaceae | <i>Nephrolepis hirsutula</i> (G. Forst.) C. Presl | Japan, Tokyo | <i>Yokoyama 2000</i> (TI) | U05638 | Hasebe et al., 1994 | | |
| Oleandraceae | <i>Arthropteris beckeri</i> (Hook.) Mett. | Australia, Queensland | <i>Kato et al. 205</i> (TI) | U05605 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Oleandraceae | <i>Oleandra pisillaris</i> (Sw.) C. Chr. | Malay Peninsula | <i>Hasebe 27603</i> (TI) | U05639 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Ophioglossaceae | <i>Botrychium biternatum</i> (Sav.) Underw. | U.S.A., Tennessee | <i>Wilson 5979</i> (TAMU) | L13474 | Manhart, 1994 | * | 1,2,3,4 |
| Ophioglossaceae | <i>Botrychium strictum</i> Underw. (= <i>Botrypus strictus</i> (Underw.) Holub) | Japan, Kanagawa | <i>Hasebe 27626</i> (TI) | B14881 | Hasebe et al., 1993 | * | 1,2,3,4,5,6 |
| Ophioglossaceae | <i>Helminthostachys zeylanica</i> (L.) Hook. | Japan | <i>Sahashi s.n.</i> (UNC) | L40907 | This study (Hauk) | | 1,2,3,4 |
| Ophioglossaceae | <i>Ophioglossum engelmannii</i> Prantl | U.S.A., Texas | <i>Manhart 04/20/89-1</i> (TAMU) | L11058 | Manhart, 1994 | * | 1,2,3,4 |
| Ophioglossaceae | <i>Botrychium multifidum</i> (S. G. Gmelin) Rupr. (= <i>Sceptridium multifidum</i> (Gmel.) Nishida ex Tagawa) | U.S.A., Michigan | <i>Hauk 579</i> (UNC) | L40906 | This study (Hauk) | | 1,2,3,4 |
| Osmundaceae | <i>Osmunda cinnamomea</i> L. var. <i>fokiensis</i> Copel. | Japan, Tochigi | <i>Hasebe 27624</i> (TI) | D14882 | Hasebe et al., 1993 | * | 1,2,3,4,5,6 |
| Osmundaceae | <i>Plenasium bromelifolium</i> C. Presl (= <i>Osmunda banksifolia</i> (C. Presl) Kuhn) | Indonesia, East Kalimantan | <i>Kato et al. 23273</i> (TI) | U05645 | Hasebe et al., 1994 | * | 1,2,3,4 |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/Ka | Figures in this study |
|-------------------------------|---|--|--|---------------------|--------------------------|-------|-----------------------|
| Plagiogyriaceae | <i>Plagiogyria japonica</i> Nakai | Japan, Mie | <i>Hasebe 27614</i> (TI) | U05643 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Polypodiaceae (Drynariaceae) | <i>Aglaomorpha speciosa</i> (Blume) Roos | cultivated (TI) 90-690 | <i>Hasebe 26660</i> (TI) | U05642 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Polypodiaceae (Lepidoreae) | <i>Lepisorus thunbergianus</i> (Kaulf.) Ching | Japan, Tokyo | <i>Hasebe 512</i> (TI) | U05629 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Polypodiaceae (Loxogrammeae) | <i>Loxogramme graminifolia</i> (Baker) C. Chr. | Japan, Fukuoka | <i>Hasebe 26661</i> (TI) | U05631 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Polypodiaceae (Microsoreae) | <i>Colysis shintensis</i> (Hayata) H. Ito | Japan, Kagoshima | <i>Hasebe 26551</i> (TI) | U05612 | Hasebe et al., 1994 | * | 1,2,3,4 |
| Polypodiaceae | <i>Pleopeltis macrocarpa</i> (Bory de Willd.) Kaulf. | Costa Rica, Alajuela | <i>Andrews 456</i> (KANU) | U21152 | Haufler and Ranker, 1995 | * | 1,2,3,4 |
| Polypodiaceae | <i>Pleopeltis thysanolepis</i> (A. Braun ex Klotzsch) Andrews and Windham | Mexico, Oaxaca | <i>Andrews 1000</i> (KANU) | U21153 | Haufler and Ranker, 1995 | * | 1,2,3,4 |
| Polypodiaceae (Polypodiaceae) | <i>Polyopodium amorphum</i> Suksdorf | U.S.A., Washington | <i>Haufler s.n.</i> (KANU) | U21142 | Haufler and Ranker, 1995 | * | 1,2,3,4,5,6 |
| Polypodiaceae (Polypodiaceae) | <i>Polyopodium appalachianum</i> Haufler and Windham | U.S.A., New Hampshire | <i>Haufler s.n.</i> (KANU) | U21141 | Haufler and Ranker, 1995 | * | 1,2,3,4,5,6 |
| Polypodiaceae (Polypodiaceae) | <i>Polyopodium australe</i> Fée | England, Cornwall | <i>Haufler s.n.</i> (KANU) | U21140 | Haufler and Ranker, 1995 | * | 1,2,3,4,5,6 |
| Polypodiaceae (Polypodiaceae) | <i>Polyopodium fauriei</i> H. Christ | Japan, Tochigi | <i>Yahara s.n.</i> (TI) | U21148 | Haufler and Ranker, 1995 | * | 1,2,3,4,5,6 |
| Polypodiaceae (Polypodiaceae) | <i>Polyopodium glaberrimum</i> Mickel and Beitel | Mexico, Oaxaca | <i>Barrington 922</i> (KANU) | U21143 | Haufler and Ranker, 1995 | * | 1,2,3,4,5,6 |
| Polypodiaceae (Polypodiaceae) | <i>Polyopodium glycyrrhiza</i> D.C. Eaton | U.S.A., California | <i>Haufler s.n.</i> (KANU) | U21146 | Haufler and Ranker, 1995 | * | 1,2,3,4,5,6 |
| Polypodiaceae (Polypodiaceae) | <i>Polyopodium macaronesicum</i> A. Bobrov | Canary Islands, Tenerife | <i>Hennipman FS 082</i> (Hennipman's personal herbarium) | U21151 | Haufler and Ranker, 1995 | * | 1,2,3,4,5,6 |

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/Ka | Figures in this study |
|----------------------------------|---|--|------------------------------------|---------------------|--------------------------|-------|-----------------------|
| Polypodiaceae (Polypodiaceae) | <i>Polypodium pellucidum</i> Kaulfuss | U.S.A., Hawaii | Haufler s.n. (KANU) | U21149 | Haufler and Ranker, 1995 | | |
| Polypodiaceae (Polypodiaceae) | <i>Polypodium plectosorum</i> Kunze | Mexico, Chiapas | Barrington 925 (KANU) | U21144 | Haufler and Ranker, 1995 | | |
| Polypodiaceae (Polypodiaceae) | <i>Polypodium rhodopleuron</i> Kunze | Mexico, Vera Cruz | Barrington 926 (KANU) | U21145 | Haufler and Ranker, 1995 | | |
| Polypodiaceae (Polypodiaceae) | <i>Polypodium scouleri</i> Hook. & Grev. | U.S.A., California | Haufler s.n. (KANU) | U21150 | Haufler and Ranker, 1995 | | |
| Psilotaceae | <i>Psilotum nudum</i> L. | Japan (TI), cultivated | Hasebe 27627 (TI) | U30835 | This study (Hasebe) | | 1,2,3,4,5,6 |
| Psilotaceae | <i>Psilotum nudum</i> L. | U.S.A., cultivated | Manhart 04/06/88-1 (TAMU) | L11059 | Manhart, 1994 | | |
| Psilotaceae | <i>Tmesipteris oblancoolata</i> Copel. | Indonesia, Anbon | Hasebe 27628 (TI) | U30836 | This study (Hasebe) | | 1,2,3,4 |
| Pteridaceae (Adiantoideae) | <i>Adiantum capillus-veneris</i> L. | Japan (TI), cultivated | Hasebe 26558 (TI) | D14880 | Hasebe et al., 1993 | | |
| Pteridaceae (Adiantoideae) | <i>Adiantum pedatum</i> L. | Japan, Fukushima | Hasebe 26547 (TI) | U05602 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Pteridaceae (Adiantoideae) | <i>Adiantum raddianum</i> C. Presl | U.S.A., Hawaii | Wolf 244 & Anderson-Wong (UTC) | U05906 | Wolf et al., 1994 | | |
| Pteridaceae (Ceratopteridoideae) | <i>Ceratopteris thalictroides</i> (L.) Brongn. | Japan (TI), cultivated | Hasebe 27607 (TI) | U05609 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Pteridaceae (Cheilantheoideae) | <i>Argyrochosma delicatula</i> (Maxon & Weath.) Windham (= <i>Notholaena delicatula</i> Maxon & Weath.) | Mexico, Nuevo León | Yatskievych & Gastony 89-229 (IND) | U19500 | Gastony and Rollo, 1995 | | 1,2,3,4 |
| Pteridaceae (Cheilantheoideae) | <i>Argyrochosma fendleri</i> (Kunze) Windham (= <i>Notholaena fendleri</i> Kunze) | U.S.A., New Mexico | Sullivan et al. s.n. (IND) | U27727 | Gastony and Rollo, 1995 | | |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/Ka | Figures in this study |
|-----------------------------------|--|--|---|---------------------|-------------------------|-------|-----------------------|
| Polypodiaceae (Cheilanthesoideae) | <i>Bommeria ehrenbergiana</i> (Klotzsch) Underw. | Mexico, Hidalgo | <i>Yatskievych & Gastony</i> 89-203 (IND) | U19497 | Gastony and Rollo, 1995 | | 1,2,3,4,5,6 |
| Polypodiaceae (Cheilanthesoideae) | <i>Cheilanthes albofusca</i> Baker | China, Yunnan | <i>Li & Xiang S-4L</i> (IND) | U19498 | Gastony and Rollo, 1995 | | |
| Polypodiaceae (Cheilanthesoideae) | <i>Cheilanthes allosuroides</i> Mett. | Mexico, Jalisco | <i>Yatskievych & Gastony</i> 89-237 (IND) | U27239 | Gastony and Rollo, 1995 | | 1,2,3,4 |
| Polypodiaceae (Cheilanthesoideae) | <i>Cheilanthes aurea</i> Baker | Mexico, Oaxaca | <i>Yatskievych & Gastony</i> 89-256 (IND) | U28786 | Gastony and Rollo, 1995 | | |
| Polypodiaceae (Cheilanthesoideae) | <i>Cheilanthes bonariensis</i> (Willd.) Proctor | Mexico, Michoacan | <i>Yatskievych & Gastony</i> 89-246 (IND) | U19499 | Gastony and Rollo, 1995 | | |
| Polypodiaceae (Cheilanthesoideae) | <i>Cheilanthes decora</i> (Brack.) R. Tryon & A. Tryon | U.S.A., Hawaii | <i>Flynn</i> s.n. (IND) | U27446 | Gastony and Rollo, 1995 | | |
| Polypodiaceae (Cheilanthesoideae) | <i>Cheilanthes duclouxii</i> (H. Christ) Ching | China, Yunnan | <i>Li & Xiang S-18L</i> (IND) | U27447 | Gastony and Rollo, 1995 | | |
| Polypodiaceae (Cheilanthesoideae) | <i>Cheilanthes horridula</i> Maxon | Mexico, Nuevo León | <i>Gastony 90-10-1</i> (IND) | U27448 | Gastony and Rollo, 1995 | | |
| Polypodiaceae (Cheilanthesoideae) | <i>Cheilanthes intramarginalis</i> (Kauf. ex Link) Hook. (= <i>Mildella intramarginalis</i> (Kauf. ex Link) Trev.) | Mexico, Hidalgo | <i>Yatskievych & Gastony</i> 89-207 (IND) | U27449 | Gastony and Rollo, 1995 | | |
| Polypodiaceae (Cheilanthesoideae) | <i>Cheilanthes lanosa</i> (Michx.) D. C. Eaton | U.S.A., Indiana | <i>Hegeman</i> s.n. (IND) | U27205 | Gastony and Rollo, 1995 | | |
| Polypodiaceae (Cheilanthesoideae) | <i>Cheilanthes rigida</i> (Sw.) Mett. (= <i>Cheiloplecton rigidum</i> (Sw.) Fée) | Mexico, Puebla | <i>Yatskievych & Gastony</i> 89-284 (IND) | U29133 | Gastony and Rollo, 1995 | | |
| Polypodiaceae (Cheilanthesoideae) | <i>Coniogramme japonica</i> (Thunb.) Diels | Japan, Saga | <i>Hasebe</i> 26662 (TI) | U05611 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/Ka | Figures in this study |
|-------------------------------|--|--|--------------------------------------|---------------------|-------------------------|-------|-----------------------|
| Peridaceae (Cheilantheoideae) | <i>Doryopteris concolor</i> (Langsd. & Fisch.) Kuhn (= <i>Cheilanthes concolor</i> (Langsd. & Fisch.) R. Tryon & A. Tryon) | Taiwan, Nantoh | Hasebe 27609 (TI) | U05621 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Peridaceae (Cheilantheoideae) | <i>Doryopteris pedata</i> (L.) Fée var. <i>palmata</i> (Willd.) Hicken | Mexico, Hidalgo | Riba 1746 (IND) | U27206 | Gastony and Rollo, 1995 | | |
| Peridaceae (Cheilantheoideae) | <i>Hemionitis elegans</i> Davenp. | Mexico, Oaxaca | Yatskiyevych & Gastony 89-258 (IND) | U27729 | Gastony and Rollo, 1995 | | |
| Peridaceae (Cheilantheoideae) | <i>Hemionitis levyi</i> Fourn. | Mexico, Oaxaca | Yatskiyevych & Gastony 89-253 (IND) | U27725 | Gastony and Rollo, 1995 | | 1,2,3,4 |
| Peridaceae (Cheilantheoideae) | <i>Llavea cordifolia</i> Lag. | Mexico, Nuevo León | Yatskiyevych & Gastony 89-224A (IND) | U27726 | Gastony and Rollo, 1995 | | |
| Peridaceae (Cheilantheoideae) | <i>Notholaena rosei</i> Maxon | Mexico, Oaxaca | Yatskiyevych et al. 83-453 (IND) | U27728 | Gastony and Rollo, 1995 | * | 1,2,3,4 |
| Peridaceae (Cheilantheoideae) | <i>Notholaena sulphurea</i> (Cav.) J. Sm. | Mexico, Puebla | Yatskiyevych & Gastony 89-248 (IND) | U28254 | Gastony and Rollo, 1995 | | |
| Peridaceae (Cheilantheoideae) | <i>Pellaea andromedifolia</i> (Kaulf.) Fée | U.S.A., California | Gastony 86-8 (IND) | U19501 | Gastony and Rollo, 1995 | * | 1,2,3,4,5,6 |
| Peridaceae (Cheilantheoideae) | <i>Pellaea boivinii</i> Hook. | Madagascar, Fianarantsoa | Liede & Conrad 2626 (IND) | U29132 | Gastony and Rollo, 1995 | | |
| Peridaceae (Cheilantheoideae) | <i>Pellaea cordifolia</i> (Sessé & Moc.) A.R. Sm. | U.S.A., Texas | Gastony 87-3 (IND) | U28253 | Gastony and Rollo, 1995 | | |
| Peridaceae (Cheilantheoideae) | <i>Pellaea pringlei</i> Davenp. | Mexico, Morelos | Gastony 87-11-16 (IND) | U28787 | Gastony and Rollo, 1995 | | |
| Peridaceae (Cheilantheoideae) | <i>Pellaea rotundifolia</i> (G. Forst.) Hook. | cultivated, Indiana Univ. | Gastony s.n. (IND) | U28788 | Gastony and Rollo, 1995 | | |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data-base accession | Literature citation | Ks/Ka | Figures in this study |
|--------------------------------|--|--|--------------------------------|---------------------|-------------------------|-------|-----------------------|
| Peridaceae (Cheilanthesoideae) | <i>Trachypteris pinnata</i> (Hook. f.) C. Chr. | Bolivia, Santa Cruz | Windisch 6088 (SJRP) | U27450 | Gastony and Rollo, 1995 | | |
| Peridaceae (Platyzomatoideae) | <i>Platyzoma microphyllum</i> R. Br. | Australia, Queensland | Kato et al. 303 (TI) | U05644 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Peridaceae (Peridoideae) | <i>Acrostichum aureum</i> L. | Indonesia, Java | Hasebe 27613 (TI) | U05601 | Hasebe et al., 1994 | * | 1,2,3,4 |
| Peridaceae (Peridoideae) | <i>Pleurosoriopsis makinoi</i> (Maxim.) Fomin | Japan, Sugadaira | Yokoyama 5175 (TI) | U30837 | This study (Yokoyama) | | 1,2,3,4,5,6 |
| Peridaceae (Peridoideae) | <i>Pteris fauriei</i> Hieron | Japan, Tokyo | Hasebe 26553 (TI) | U05647 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Peridaceae (Peridoideae) | <i>Pteris vittata</i> L. | U.S.A., Hawaii | Wolf 257 & Anderson-Wong (UTC) | U05941 | Wolf et al., 1994 | | |
| Peridaceae (Taenitidoideae) | <i>Onychium japonicum</i> (Thunb.) Kunze | Japan, Tokyo | Hasebe 422 (TI) | U05641 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Peridaceae (Taenitidoideae) | <i>Taenitis blechnoides</i> (Willd.) Sw. | Singapore, Singapore | Hasebe 27608 (TI) | U05654 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Salvinaceae | <i>Salvinia cucullata</i> Roxb. ex Bory | cultivated (TI) | Hasebe 26550 (TI) | U05649 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Schizaeaceae | <i>Actinostachys digitata</i> (L.) Wall. | Japan, Tokyo | Yokoyama 27617 (TI) | U05650 | Hasebe et al., 1994 | * | 1,2,3,4 |
| Schizaeaceae | <i>Anemia mexicana</i> Klotzsch | Australia, cultivated | Hasebe 26543 (TI) | U05603 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Schizaeaceae | <i>Lygodium japonicum</i> (Thunb.) Sw. | Japan, Kanagawa | Hasebe 27616 (TI) | U05632 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Schizaeaceae | <i>Lygodium japonicum</i> (Thunb.) Sw. | U.S.A., cultivated | Manhart 07/13/91-1 (TAMU) | L13479 | Manhart, 1994 | | |
| Selaginellaceae | <i>Selaginella apoda</i> (L.) Spring | U.S.A., cultivated | Manhart 04/06/88-2 (TAMU) | L11280 | Manhart, 1994 | | |
| Thelypteridaceae | <i>Cyclosorus opulentus</i> (Kauf.) Nakaike | Ecuador | MBG 920036 (UTC) | U05915 | Wolf et al., 1994 | * | 1,2,3,4,5,6 |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data- base acces- sion | Literature citation | Ks/ Ka | Figures in this study |
|-----------------------|--|---|-------------------------------|---------------------------------|----------------------|-----------|--------------------------|
| Thelypteridaceae | <i>Thelypteris acuminata</i> (Houtt.) Morton | Japan, Chiba | Sano 39 (CBM) | D43919 | Sano et al., unpubl. | * | |
| Thelypteridaceae | <i>Thelypteris beddomei</i> (Baker) Ching | Japan, Saga | Hasebe 26564 (TI) | U05655 | Hasebe et al., 1994 | * | 1,2,3,4 |
| Thelypteridaceae | <i>Thelypteris palustris</i> (Salisbury) Schott var. <i>pubescens</i> (Laws.) Pernald | U.S.A. | MBG 810566 (UTC) | U05947 | Wolf et al., 1994 | | |
| Vittariaceae | <i>Ananthacorus angustifolius</i> Underw. & Maxon | Costa Rica | Farrar CR289 (ISC) | U20932 | Crane et al., 1995 | | 1,2,3,4 |
| Vittariaceae | <i>Anetium citrifolium</i> Splitg. | Costa Rica | Crane 930626-26 (ISC) | U21284 | Crane et al., 1995 | | |
| Vittariaceae | <i>Anurophyum boryanum</i> (Willd.) Spreng. | La Réunion | Ranker 1538 (ISC) | U20930 | Crane et al., 1995 | | |
| Vittariaceae | <i>Anurophyum ensiforme</i> Hook. | Mexico | Farrar 1994 (Mexico) (ISC) | U20931 | Crane et al., 1995 | | |
| Vittariaceae | <i>Anurophyum plantagineum</i> (Cav.) Kaulf. | Fiji | Drake s.n. (ISC) | U21285 | Crane et al., 1995 | | |
| Vittariaceae | <i>Anurophyum reticulatum</i> (G. Forst.) Kaulf. | Australia, Queensland | Kato et al. 431 (TI) | U05604 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Vittariaceae | <i>Hecistopteris pumila</i> (Spreng.) J. Sm. | Costa Rica | Crane 930626-25 (ISC) | U21286 | Crane et al., 1995 | | |
| Vittariaceae | <i>Polytaenium cajanense</i> (Desv.) Benedict | Costa Rica | Crane 930617-5 (ISC) | U20934 | Crane et al., 1995 | | |
| Vittariaceae | <i>Polytaenium lanceolatum</i> (L.) Benedict | Costa Rica | Crane 930626-37 (ISC) | U21287 | Crane et al., 1995 | | |
| Vittariaceae | <i>Polytaenium lineatum</i> (Sw.) J. Sm. | Costa Rica | Clark s.n. Not vouch- ered | U20935 | Crane et al., 1995 | | 1,2,3,4 |
| Vittariaceae | <i>Vittaria anguste-elongata</i> Hayata | Taiwan | Chiou 920722 (ISC) | U21291 | Crane et al., 1995 | | |
| Vittariaceae | <i>Vittaria dimorpha</i> K. Müll. | Costa Rica | McAlpin, NYBG (ISC) | U21292 | Crane et al., 1995 | | |

APPENDIX. Continued.

| Family (subfamily) | Species | Locality (country, state, or province) | Source/Voucher | Data- base access- sion | Literature citation | Ks/ Ka | Figures in this study |
|-----------------------|--|---|-----------------------------|----------------------------------|---------------------|-----------|--------------------------|
| Vittariaceae | <i>Vittaria ensiformis</i> Sw. | La Réunion | Ranker 1542 (ISC) | U21290 | Crane et al., 1995 | | |
| Vittariaceae | <i>Vittaria flexuosa</i> Fée | Japan, Kagoshima | Hasebe 27613 (TI) | U05656 | Hasebe et al., 1994 | * | 1,2,3,4,5,6 |
| Vittariaceae | <i>Vittaria gardneriana</i> Fée | Ecuador | Clark 1130 (ISC) | U21294 | Crane et al., 1995 | | |
| Vittariaceae | <i>Vittaria graminifolia</i> Kaulf. | Costa Rica | Farrar CR312 (ISC) | U21295 | Crane et al., 1995 | | |
| Vittariaceae | <i>Vittaria isoetifolia</i> Bory | La Réunion | Ranker 1533 (ISC) | U20936 | Crane et al., 1995 | | |
| Vittariaceae | <i>Vittaria lineata</i> (L.) J.E. Sm. | U.S.A. | Lassiter 84-09-15B (ISC) | U20937 | Crane et al., 1995 | | |
| Vittariaceae | <i>Vittaria minima</i> (Baker) Benedict | Costa Rica | Moran 3180 (ISC) | U21288 | Crane et al., 1995 | | |
| Vittariaceae | <i>Vittaria remota</i> Fée | Costa Rica | Moran 3180 (ISC) | U21289 | Crane et al., 1995 | | |
| Vittariaceae | <i>Vittaria stipitata</i> Fée | Costa Rica | Crane 930611-10 (ISC) | U21293 | Crane et al., 1995 | | |
| Vittariaceae | <i>Vittaria zosterifolia</i> Willd. | Taiwan | Chiou 920723 (ISC) | U21296 | Crane et al., 1995 | | |