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Biogeographic and Phylogenetic Investigations of the Pantropical Genus *Sargassum* (Fucales, Phaeophyceae) with Respect to Gulf of Mexico Species

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The genus *Sargassum*, despite considerable research efforts, is still one of the most systematically complex and problematic genera of the brown algae. In order to evaluate the adequacy of sectional to series level designations in the tropical subgenus, an ambitious effort was begun that will eventually include all of the east Isthmian species in a global phylogenetic analysis for the genus. This research effort is based on extensive seasonal field survey work and molecular-based parsimony analysis (chloroplast encoded ribulose-1, 5-bisphosphate carboxylase spacer region) of collected voucher specimens. The present analysis indicates that the Gulf/Caribbean species do not cluster as they are traditionally classified but form a strongly supported clade representing a single biogeographic unit in the *Malococarpicae* section of the subgenus *Sargassum*. Four of these species share an insertion/deletion event with some of the Pacific species, suggesting a shared evolutionary history between the two ocean basins.

Sargassum C. Agardh (1820) is one of the most species-rich, ecologically important, and widespread brown algal genera of subtropical and tropical regions around the world. The bulk of the species for the genus are distributed in the northern and southern regions of the Pacific basin, with the center of diversity in the Indo-Malay area (Nizamuddin, 1961; Phillips, 1995). Only 19 species have been reported east of the Isthmus of Panama and nine for the Gulf of Mexico, all classified in the tropical subgenus *Sargassum* (Taylor, 1960; Schneider and Searles, 1991; Wynne, 1998).

Sargassum is part of a relatively small (eight genera) Pacific-based family, the Sargassaceae (Kützing, 1843; Nizamuddin, 1961) in the order Fucales. The genus initially included 62 species organized into seven groups (C. Agardh, 1820). J. Agardh (1848, 1889) modified this original system to reflect phyletic concepts contending that species in the genus evolved from simple morphological forms to yield more complex forms. Subgeneric designations were based on relationships between the central axis and lamina, whereas subgeneric section and subsection designations were based on receptacle morphologies. For example, species within the *Malacocarpicae* section are typified by having smooth receptacles, whereas those in the *Acanthocarpicae* section have spiny receptacles. Series to subseries designations were based on subtle changes in reproductive, central axial, and lamina features. J. Agardh's (1889) system remains the systematic and phylogenetic framework for the genus today, with minor modifications added since (Fig. 1) (e.g., Grunow, 1915–1916; Setchell, 1931, 1933, 1936; Womersley, 1954; Abbott et al., 1988).

Today, over 400 hundred species are described in this system. Since its inception over 150 years ago, *Sargassum* has received considerable systematic effort, yet this genus is still by far one of the most systematically complex and problematic genera of the brown algae (e.g., Chiang et al., 1992; Kilar et al., 1992a). Taxonomic problems arise from the fact that many early species descriptions were based on fragmentary specimens that did not reflect high rates of phenotypic variability in key features, a worldwide distribution, and a substantial number of described species. Recent research has attempted to better account for inherent phenotypic variation (Ajisaka, 1992; Ajisaka et al., 1994, 1997; Kilar, 1992; Kilar et al., 1992b), yet species boundaries between many closely related species are still poorly defined (Ajisaka, 1992; Phillips, 1998; Phillips et al., 2000a). For example, insightful research on Atlantic and Gulf of Mexico species has documented high rates of phenotypic variability in several key characters used to define species (Kilar and Hanisak, 1988; Kilar, 1992; Kilar et al., 1992a). This work unequivocally demonstrates that boundaries between closely related species are masked by phenotypic variability and that some key features were of questionable value (Kilar and Hanisak, 1988; Kilar, 1992; Kilar et al., 1992a). Molecular markers have proved useful in such cases as *Sargassum* because they provide an independent means by which to evaluate species concepts (Phillips, 1998; Phil-

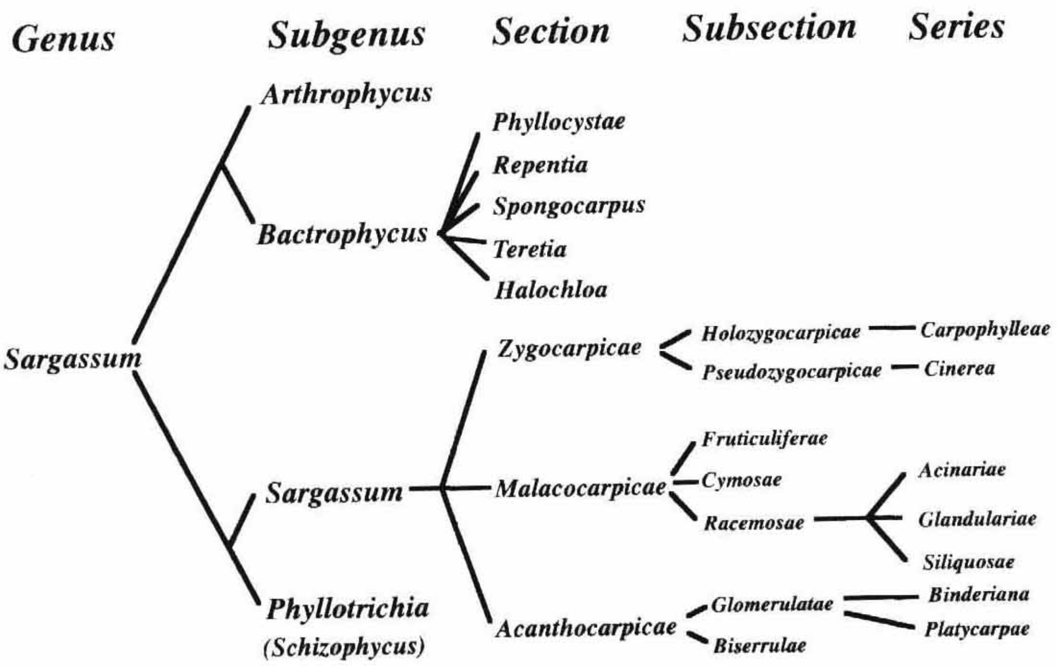

Fig. 1. Current systematic framework for *Sargassum* C. Agardh as compiled from J. Agardh (1843, 1889), Grunow (1915–1916), Setchell (1931, 1933, 1936), Womersley (1954), and Abbott et al. (1988).

lips et al., 2000). Research applying molecular-based characters to evaluate systematic concepts and species boundaries in Pacific basin *Sargassum* species made several fundamental discoveries (Phillips, 1998; Phillips et al., 2000). Most importantly, these analyses supported much (three) of J. Agardh's (1889) traditional subgeneric designations and all of the sectional designations in the subgenus *Sargassum*. Yet molecular analyses showed fundamental inadequacies in subsectional to series designations in the subgenus *Sargassum* (Phillips, 1998; Phillips et al., 2000).

Many of the type species belonging to the subsections and series of the tropical subgenus *Sargassum* were described east of the Isthmus of Panama (see Taylor, 1960; Schneider and Searles, 1991; Wynne, 1998). To expand the Pacific-based phylogenetic analysis (Phillips, 1998; Phillips et al., 2000) and further test the adequacy of sectional to series level designations, an ambitious effort was begun that will eventually include all 19 of the east Isthmian species in a global phylogenetic analysis for the genus. This research effort includes extensive seasonal field survey work to evaluate distributional limits of Gulf of Mexico species and phylogenetic analysis based on ribulose-1, 5-bisphosphate carboxylase spacer region [or *rbc*LS spacer region including flanking regions of the genes encoding for the large (*rbc*L) and small (*rbc*S) subunits] of collected voucher specimens. We herein report our progress in addressing global issues within the genus, in evaluating traditional species boundaries, and in addressing biogeographic and phylogenetic hypotheses within coastal and pelagic species east of the Isthmus of Panama.

METHODS

Field survey work and species collection.—Several coastal sites were surveyed on a seasonal basis from Port Aransas, TX, to Sarasota, FL, to determine distribution limits of *Sargassum* species in the northwestern Gulf of Mexico (Fig. 2). Voucher specimens were collected from both saxicolous populations and drift material at these coastal sites and deposited at the LAF herbarium (University of Louisiana at Lafayette, Lafayette, LA). Offshore pelagic beds were surveyed during two separate oceanic cruises to Stetson Bank, TX, and Sonnier Bank, LA. Species were acquired from two localities in the Caribbean (Guadeloupe F.W.I. and the Panama Canal region) and from one site in the Florida Keys, FL, for inclusion in the phylogenetic analysis. Voucher material was preserved both in the desiccant silica gel for molecular analysis and in 5% formalin/seawa-

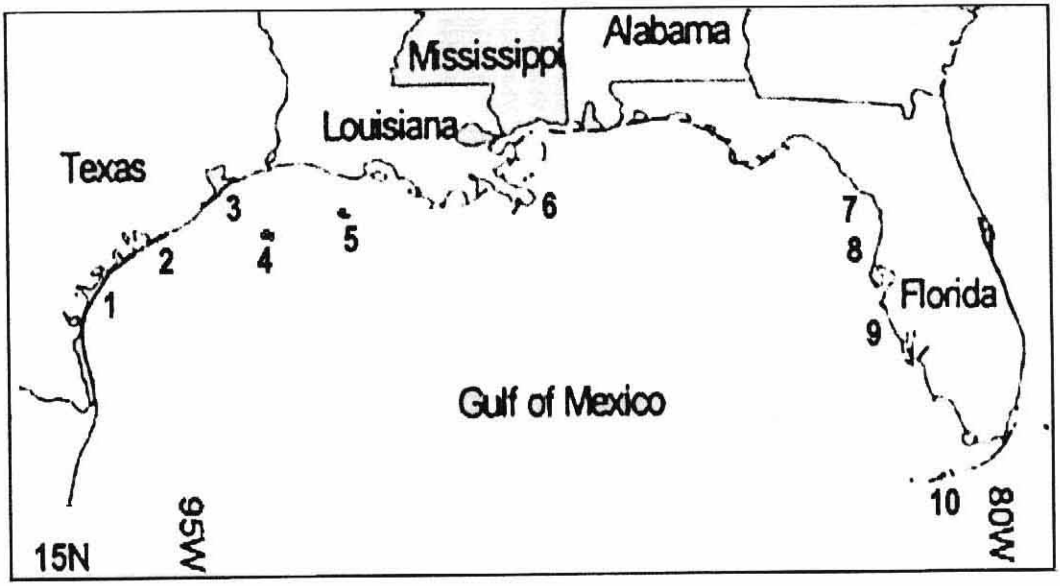

Fig. 2. Map of the Gulf of Mexico depicting coastal and oceanic collection localities. Collection sites are numbered as follows: 1 = Port Aransas, TX; 2 = Red Fish Bay, TX; 3 = Galveston, TX; 4 = Stetson Bank, TX; 5 = Sonnier Bank, LA; 6 = Grand Terre, LA; 7 = Homossassa, FL; 8 = Tarpon Springs, FL; 9 = Sarasota, FL; 10 = Key Largo, FL.

ter for morphological characterization. Traditional morphological features and regional keys (Taylor, 1960; Schneider and Searles, 1991; Wynne, 1998) were used to make initial species determinations.

DNA extraction.—Total DNA was extracted from 19 specimens representing eight species by a modified CTAB method and included in a comprehensive DNA collection for the genus (Phillips et al., 2001).

Polymerase chain reaction (PCR) and automatic sequencing.—The chloroplast-encoded *rbc*LS spacer region was amplified for all representative taxa by methods and with primers (CF, CF-NEW, CR, CR-NEW) as reported in Phillips (1998). PCR products were purified for sequencing reactions by PEG precipitation (Hillis et al., 1996). Sequencing reactions were accomplished with the Big Dye sequencing kit and protocol and run on an ABI Prism 310 genetic analyzer (PE Applied Biosystems, Foster City, CA). All PCR products (*rbc*LS spacer region) were sequenced in both directions.

Sequence and parsimony analysis.—Sequence data were compiled, viewed, and corrected within the software program Sequencher (Gene Codes Corp., Ann Arbor, MI). Sequences were aligned by eye and placed in a global *rbc*LS spacer region alignment for the genus. To address broader biogeographic and phylogenetic issues within the genus, *rbc*LS spacer region sequences from 22 *Sargassum* isolates were included from several localities in the Pacific basin (see Phillips, 1998; Phillips et al., 2000) along with those from 12 east Isthmian isolates (Table 1). A detailed analysis of Pacific species is already published, so results will focus on east Isthmian species (Phillips, 1998; Phillips et al. 2000). On the basis of the Pacific-based analyses, *Turbinaria ornata* J. Agardh (Sargassaceae) was used as the outgroup (Phillips, 1998).

Maximum parsimony analysis was accomplished with PAUP version 4.0b.1 (Swofford, 1998). Insertion/deletion events or indels, present in the intergenic spacer (IGS) portion of the *rbc*LS spacer region were coded as present or absent and placed at the end of the data set (Phillips, 1998; Phillips et al., 2000). All searches were done under the Fitch criterion of equal weights for all substitutions. Heuristic searches were accomplished with 100 random sequence additions, MULPARS, and nearest neighbor reconnection swapping algorithm. Trees found in the random searches were then used as a starting point for further analysis with MULPARS and tree bisection (TBR) until swapping was complete. Tree lengths were calculated on all characters. Support for nodes

TABLE 1. List of taxa collected during field surveys and isolates from the Pacific basin and elsewhere in the Caribbean.†

Taxon[b]	Location	Collector	Tissue	GenBank no.[c]
Sargassaceae				
†*Turbinaria ornata* J. Ag.	Sharks Cove, O'ahu, HI, U.S.A.	Naomi Phillips	Frozen	AF076688
Sargassum				
Arthrophycus				
†*Sargassum fallax* Sonder	Victoria, Australia	Gary Saunders	Silica gel	AF244333
Bactrophycus				
Teretia				
†*S. muticum* (Yendo) Fensh	San Francisco, CA, U.S.A.	Steve Obrebski	Fresh	AF244331
†*S. muticum* (Yendo) Fensh	Qingdao, China	Naomi Phillips	Silica gel	AF292068
†*S. thunbergii* (Mert.) Kunt.	Qingdao, China	Naomi Phillips	Frozen	AF244343
†*S. thunbergii* (Mert.) Kunt.	Shirahama, Japan	Tetsuro Ajisaka	Silica gel	AF244332
Phyllotrichia				
†*S. palmeri* Grunow	San Clemente Island, CA, U.S.A.	Steve Murray	Silica gel	AF244339
†*S. piluliferum* (Turner) C. Ag.	Shirahama, Japan	Naomi Phillips	Fresh	AF244340
†*S. patens* C. Ag.	Shirahama, Japan	Tetsuro Ajisaka	Silica gel	AF244338
Sargassum				
Zygocarpicae				
Holozygocarpicae				
†*S. polycystum* C. Ag.	Guam, Micronesia	Roy Tsuda	Silica gel	AF244346
†*S. polycystum* C. Ag.	Plau Besau, Malaysia	Tetsura Ajisaka	Silica gel	AF244344
Pseudozygocarpicae				
†*S. myriocystum* J. Ag.	Ryukyu, Japan	Tetsuro Ajisaka	Silica gel	AF244326
Malacocarpicae				
Fruticuliferae				
†*S. obtusifolium* J. Ag.	Kahala Beach Park O'ahu, HI, U.S.A.	Naomi Phillips	Fresh	AF244328
†*S. obtusifolium* J. Ag.	Ka ala wai, O'ahu, HI, U.S.A.	Naomi Phillips	Fresh	AF195516
†*S. polyporum* Montagne	Iriomote Islands, Japan	Tetsuro Ajisaka	Silica gel	AF244327
Cymosae				
†*S. polyphyllum* J. Ag.	Kahala Beach Park, O'ahu, HI, U.S.A.	Naomi Phillips	Fresh	AF076690
Racemosae				
Siliquosae				
†*S. siliquosum* J. Ag.	Port Dickson, Malaysia	Ching Lee Wong	Silica gel	AF244345
†*S. filipendula* C. Ag.	Sarasota, FL, U.S.A.	Naomi Phillips	Silica gel	AF301229
†*S. filipendula* C. Ag.	Galveston, TX, U.S.A.	Naomi Phillips	Silica gel	
†*S. mathiesonii* Kilar	Hommossassa, FL, U.S.A.	Naomi Phillips	Fresh	
Acinariae				

TABLE 1. Continued.

Taxon[b]	Location	Collector	Tissue	GenBank no.[c]
S. acinarium (L.) Setch.	Galveston, TX, U.S.A.	Naomi Phillips	Silica gel	
†*S. acinarium* (L.) Setch.	Port Aransas, TX, U.S.A.	Naomi Phillips	Silica gel	AF301227
S. acinarium (L.) Setch.	Grand Terre, LA, U.S.A.	Naomi Phillips	Silica gel	
Acanthocarpicae				
Glomerulatae				
Binderiana				
†*S. binderi* Sonder	Miyako Islands, Japan	Tetsuro Ajisaka	Silica gel	AF244337
†*S. echinocarpum* J. Ag.	Kahala Beach Park, O'ahu, HI, U.S.A.	Naomi Phillips	Fresh	AF292069
†*S. echinocarpum* J. Ag.	Ka ala wai, O'ahu, HI, U.S.A.	Naomi Phillips	Fresh	AF292070
†*S. echinocarpum* J. Ag.	Ka ala wai, O'ahu, HI, U.S.A.	Naomi Phillips	Fresh	AF076689
Glomerulatae				
Platycarpae				
†*S. cristaefolium* C. Ag.	Guam, Micronesia	Roy Tsuda	Silica gel	AF244335
†*S. duplicatum* J. Ag.	Ryukyu, Japan	Tetsuro Ajisaka	Silica gel	AF244324
†*S. platycarpum* Montagne	Guadeloupe, F. W. I.	Aline Renoux	Silica gel	AF301233
†*S. platycarpum* Montagne	San Blas, IS., Colon, Panama	Brian Wysor	Silica gel	AF301230
†*S. polyceratium* Montagne	Fort Randolf, Colon, Panama	Brian Wysor	Silica gel	AF301235
†*S. polyceratium* Montagne	Fort Randolf, Colon, Panama	Brian Wysor	Silica gel	AF301225
†*S. polyceratium* Montagne	Guadeloupe, F. W. I.	Aline Renoux	Silica gel	AF301232
†*S. hystrix* J. Ag.	Galeta, Panama	Brian Wysor	Silica gel	AF301228
Species in the subgenus *Sargassum* with unknown sectional to series placement				
†*S. natans* (L.) Gaillon	Stetson Bank, TX, U.S.A.	Naomi Phillips	Silica gel	AF301234
†*S. fluitans* Børgesen	Stetson Bank, TX, U.S.A.	Naomi Phillips	Silica gel	AF301231
†*S.* sp.	Plau Besau, Malaysia	Tetsuro Ajisaka	Silica gel	AF244341
†*Sargassum* sp.	Tonga	Tim Motley	Fresh	AF24434
Sargassum sp.	Grand Terre, LA, U.S.A.	Suzanne Fredericq	Silica gel	
Sargassum sp.	Grand Terre, LA, U.S.A.	Suzanne Fredericq	Silica gel	
Sargassum sp.	Grand Terre, LA, U.S.A.	Suzanne Fredericq	Silica gel	
†*Sargassum* sp.	Key Largo, FL, U.S.A.	Peter Vroom	Silica gel	
†*Sargassum* sp.	Plau Besau, Malaysia	Tetsuro Ajisaka	Silica gel	

[a] Collaborative use of DNA in the collection may be arranged by contacting the authors.

[b] Taxa are listed in taxonomic order, including subgenera, sections, subsections, and series for the genus *Sargassum*. Those taxa successfully sequenced for the *rbc*LS spacer region and used in phylogenetic analysis are indicated by †.

[c] Accession numbers represent *rbc*LS spacer region GenBank entries for each sequenced taxon.

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                    1                                                                                                100
T.ornataHI          ...GACCTTTAAAAGCA.GCTTTAGATCATTGGAAAGATATT.ACTTTTGAATATACTTCAACAGACACACCTGATTTTGTTGAAGTAGCAACTGAAAGT
S.duplicatumJP      ...GACCTTTAAAAGCA.GCTTTAGATTTATGGAAAGATATT.ACCTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTAGCAACTGAAAAT
S.myriocystumMA     ...GACCTTTAAAAGCA.GCTTTAGATTTATGGAAAGATATT.ACCTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTAGCAACTGAAAAT
S.polyporumJP       ...GACCCTTAAAAGCA.GCTTTAGATCTATGGAAAGATATT.ACCTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT
S.obtusifoliumHI    ..GGACCTTTAAAAGCA.GCTTTAGATCTATGGAAAGATATT.ACCTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT
S.polyphyllumHI     ...GACCTTTAAAAGCA.GCTTTAGATCTATGGAAAGATATT.ACCTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT
S.echinocarpumHI    GCGGACCTTTAAAAGCA.GCTTTAGATCTATGGAAAGATATT.ACTTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTAGCAACTGAAAGT
S.muticumCH         ....................TTTAGATCTATGGAAAGATATT.ACTTTTGAATATACTTCAACAGACACACCTGATTTTGTTGAAGTAGCAACTGAAAAT
S.muticumCA         ..............................TGGAAAGATATT.ACTTTTGAATCTACTTCAACAGACACACCTGATTTTGTTGAAGTAGCAACTGAAAAT
S.thunbergiiJP      ..............................TGGAAAGATATT.ACTTTTGAATATACTTCAACAGACACACCTGATTTTGTTGAAGTAGCGACTGAAAGT
S.fallaxAU          ..............................TGGAAAGATATT.ACTTTTGAATATACTTCAACAGACACACCTGATTTTGTTGAAGTAGCAACTGAAAGT
S.echinocarpumHI    ..............................TGGACAGATATT.ACTTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTACCAACTGAAAGT
S.cristaefoliumGM   ..............................TGGAAAGATATT.ACTTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTAGCAACTGAAAGT
S.echinocarpumHI    ..............................TGGAAAGATATT.ACTTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTAGCAACTGAAAGT
S.binderiJP         ............................................CTTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTACCAACTGAAAGT
S.patensJP          ....................TTTAGATCTATGGAAAGATATT.ACCTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTAGCAACTGAAAGT
S.palmeriCA         ......................................TATA.ACTTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTTCCAACTGAAAGT
S.piluliferumJP     ..............................TGGAAAGATATT.ACCTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT
S.sp.MA             ..............................TGGAAAGATATT.ACCTTTGAATATACTTCAACAGATACACC.GATTTTGTTGA.GTAGCAACTGAAAAT
S.sp.TA             ..............................TGGAAAGATATT.ACTTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTAGCAACTGAAAGT
S.thunbergiiCH      ............................................CTTTTGAATATACTTCAACAGACACACCTGATTTTGTTGAAGTAGCGACTGAAAGT
S.polycystumGM      ....................TTTAGATCTATGGAAAGATATT.ACTTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTAGCAACTGAAAGT
S.polycystumMA      ............................................CCTTTGAATGTCCTCCAACAGATACACCTGATTTTGTTGAAGTAGCAACTGAAAAT
S.sp.KL             ........................................TT.ACTTTCGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT
S.acinariumTX       TGCGGACCTTTAAAAGCAGCTTTAGATCTATGGAAAGATATT.ACCTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT
S.siliquosumMA      TGCGGACCTTATTAAGCAGCTTTAGAACTATGGAAAGATATT.ACCTTTGAATATACTTCAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT
S.polyceratiumPA    TGCGGACCTTTAAAAGCAGCTTTTGATCTATGGAAAGATATTACCTTTGAATATACTT.CAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT
S.sp.PA             TGCGGACCTTTAAAAGCAGCTTTAGATCTATGGAAAGATATTACCTTTGAATATACTT.CAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT
S.polyceratiumPA    TGCGGACCTTTAAAAGCAGCTTTAGATCTATGGAAAGATATTACCTTTGAATATACTT.CAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT
S.hystrixPA         TGCGGACCTTTAAAAGCAGCTTTAGATCTATGGAAAGATATTACCTTTGAATATACTTACAACAGATACACCTGATTTTGTTGAAGTTGCTACTGAAAGT
S.filipendulaFL     TGCGGACCTTTAAAAGCAGCTTTAGATCTATGGAAAGATATTACCTTTGAATATACTT.CAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT
S.platycarpumGP     TGCGGACCTTTAAAAGCAGCTTTAGATCTATGGAAAGATATTACCTTTGAATATACTT.CAACAGATACACCTGATTTTGTTGAAGTAGCAACTGAAAGT
S.fuitansPEL        TGCGGACCTTTAAAAGCAGCTTTAGATCTATGGAAAGATATTACCTTTGAATATACTT.CAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT
S.polyceratiumGP    TGCGGACCTTTAAAAGCAGCTTTAGATCTATGGAAAGATATTACCTTTGAATATACTT.CAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT
S.platycarpumPA     TGCGGACCTTTAAAACCCGCTTTAGATCTATGGAAAGATATTACCTTTGAATATACTT.CGACAGATACACCTGATTTTGTTGAAGTAGCAACTGAAAGT
S.natansPEL         TGTGGACCTTTAAAAGCAGCTTTAGATCTATGGAAAGATATTACCTTTGAATATACTT.CAACAGATACACCTGATTTTGTTGAAGTTGCAACTGAAAGT

                    101                                                                                              200
                       rbcL stop codon
                       ___
T.ornataHI          CCATAAAGTTATTCTGTTCTACAGTTTAAT******TTTTATTATAAATC**********TAAAAGAATATCTT****************AGCGGGCTTT
S.duplicatumJP      CCGTAAATCTATTCTGTTC*****TTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.myriocystumMA     CCGTAAATCTATTCTGTTC*****TTTAAT******TTTTACGA****************TAAAAAAACAGATT****************GATGAATTTT
S.polyporumJP       CCTTAAACCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.obtusifoliumHI    CCTTAAACCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAGAACAGATT****************GATGAATTTT
S.polyphyllumHI     CCTTAAACCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.echinocarpumHI    AACTAAATCTATTCTGTTCTACAGTTTATT******CTTTACTA****************TAAAAGAACGGATT****************AATGAATTTT
S.muticumCH         CCTTAGATCTATTCTGTTCTGTAGTTTAAT******TTTTACTA****************TAAAAGAACAGACT****************AATGAATTTT
S.muticumCA         CCTTAGATCTATTCTGTTCTGTAGTTTAAT******CTTTACTA****************TAAAAGAACGGATT****************AATGAATTTT
S.thunbergiiJP      CCTTAAATTTATTCTATTCTATAGTGTAAT******TTTTACTA****************TAAAAGAACAGACT****************AATGAATTTT
S.fallaxAU          ACCTAAATCTATTCTGCTCTATAGTTTAAT******TTTTACTA****************TAAAAGAGCAGACT****************AATGAATTTT
S.echinocarpumHI    AACTAAATCTATTCTGTTCTACAGTTTAAT******CTTTACTA****************TAAAAGAACGGATT****************AATGAATTTT
S.cristaefoliumGM   AACTAAATCTATTCTGTTCTACAGTTTAAT******CTTTACTA****************TAAAAGAACGGATT****************AATGAATTTT
S.echinocarpumHI    AACTAAATCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAGAACGGATT****************AATGAATTTT
S.binderiJP         AACTAAATCTATTCTGTCCTACAGTTTAAT******CTTTACTA****************TAAAAGAATGGATT****************AATGAATTTT
S.patensJP          AACTAAATCTATTCTGTTCTACAGTTTAAT******CTTTACTA****************TAAAAGAACGGATT****************AATGAATTTT
S.palmeriCA         CCCTAAACCTATTCTGTTCTA.AGTTTAAT******CTTTACTA****************TAAAAGAACAGATT****************GATGAATTTT
S.piluliferumJP     CCTTAAACCTATTCTGTCCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.sp.MA             CCGTAAATCTATTCTGTTC*****TTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.sp.TA             AACTAAATCTATTCTGTTCTACAGTTTAAT******CTTTACTA****************TAAAAGAACGGATT****************AATGAATTTT
S.thunbergiiCH      CCTTAGATTTATTCTATTCTGTAGTGTAAT******TTTTACTA****************TAAAAGAACAGACT****************AATGAATTTT
S.polycystumGM      CACTAAATCTATTCTGTTCTACAGTTTAAT******CTTTACTA****************TAAAAGAACGGATT****************AATGAATTTT
S.polycystumMA      CCCTAAATCTATCCTGTCC*****TTTAAT******CTTTACGA****************TAAAAAAACAGATT****************GATGAATTTT
S.sp.KL             CCTTAAACCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.acinariumTX       CCTTAAACCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.siliquosumMA      CCTTAAACCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.polyceratiumPA    CCTTAAACCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.sp.PA             CCTTAAACCTATTCTGTTCTATAGTTTTAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.polyceratiumPA    CCTTAAACCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.hystrixPA         CCTTAAACCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.filipendulaFL     CCTTAAACCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.platycarpumGP     AAATAAATCTATTCTGTTCTCTAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.fuitansPEL        CCTTAAACCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.polyceratiumGP    CCTTAAACCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.platycarpumPA     AACTAAATCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT
S.natansPEL         CCTTAAACCTATTCTGTTCTATAGTTTAAT******CTTTACTA****************TAAAAAAACAGATT****************GATGAATTTT

                    201                                                                                              300
T.ornataHI          **AAT*CTTTT*TAATA*CTTTACATTA*AAATAAAAAACATAAAAAGTTTGGTAGTTAACTAAAAATAAAACTAAAATATTT*ACATTAGATAAAAGAA
S.duplicatumJP      **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.myriocystumMA     **AAC*CTTTT*TAATA*TTTTACATTA*AAAAAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAAATAAATAA
S.polyporumJP       **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.obtusifoliumHI    **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.polyphyllumHI     **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAGAAAGCTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.echinocarpumHI    **AAT*ATTTT*TAATA*TTTTACATTA*AAATAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.muticumCH         **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACAGAAAAAGTTTAGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.muticumCA         **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACAGAAAAAGTTTAGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.thunbergiiJP      **AAT*CTTTT*TAATA*TTTTATATTA*AGACAAAAGATAGAAAAAGTTTAGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.fallaxAU          **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAAAAAATTTAGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.echinocarpumHI    **AAT***********A*TTTTACATTA*AAATAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.cristaefoliumGM   **AAT*** *******A*TTTTACATTA*AAATAAAAGACATAAAAAGTT.GGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.echinocarpumHI    **AAT*TTTTT*TAATA*TTTTACATTA*AAATAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.binderiJP         **AAT*ATTTT*TAATA*TTTTACATTA*AAATAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
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Fig. 3. Global *rbc*L-S spacer region alignment exhibiting seven indels with the Hawaiian–Guam–Gulf of Mexico specific indel highlighted in gray. Geographic codes with the first and last letter of the site follow taxon names with PEL = pelagic, * = gaps; and ● = missing characters.

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S.patensJP           **AAT*ATTTT*TAATA*TTTTACATTA*AAATAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.palmeriCA          **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.piluliferumJP      **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAGCTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.sp.MA              **AAC*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.sp.TA              **AAT*ATTTT*TAATA*TTTTACATTA*AAATAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.thunbergiiCH       **AAT*CTTTT*TAATA*TTTTATATTA*AGACAAAAGATAGA.AAAGTTTAGTAGTTAACTCAAAATTAAATTAAAATATTT*ACTTTAAATAAAATAA
S.polycystumGM       **AAT*ATTTT*TAATA*TTTTACATTA*AAATAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAGAATAA
S.polycystumMA       **AAC*CTTTT*TAATA*TTTTACATTAAAAAAAAAAGACTTAAAA.GTTGGGTGTTAACCTAAAAATAAATTTAAATTTTTA*CCTTTAAATTATATAA
S.sp.KL              **AAT*CTTTT*TAATA*TTTTACATTA*AAACAGAAGACATAAAAAGTTTGGAAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.acinariumTX        **AAT*********A*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.siliquosumMA       **AAT*TTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAGAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.polyceratiumPA     **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.sp.PA              **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.polyceratiumPA     **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.hystrixPA          **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.filipendulaFL      **AAT*********A*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.platycarpumGP      **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.fuitansPEL         **AAT*********A*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.polyceratiumGP     **AAT*********A*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATAAAATTATTT*ACATTAAATAAAATAA
S.platycarpumPA      **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAAACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA
S.natansPEL          **AAT*CTTTT*TAATA*TTTTACATTA*AAACAAAAGACATAAAAAGTTTGGTAGTTAACTAAAAATAAAATTAAAATATTT*ACATTAAATAAAATAA


                     301                                                                                     389
                               rbcS start codon

T.ornataHI           TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGACGAGCAAATTAAAAATCAAGTTG-+-+++-..
S.duplicatumJP       TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAGATTAAAAATCAAGTTG++++++-..
S.myriocystumMA      TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAGATTAAAAATCAAGTTT++++++-..
S.polyporumJP        TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACATCAAGTTG-+++++-..
S.obtusifoliumHI     TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACATCAAGTTG-+++++-..
S.polyphyllumHI      TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACATCAAGTTG-+++++-..
S.echinocarpumHI     TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGACGAGCAAATTAAAAGTCAAGTTG-+++++-..
S.muticumCH          TTGAAGAGTAATGAGACTTAC.CAAGGATGTTTTTCATTTTTACCAGATTTAAGTGACGAACAAATTAAACATCAAGTTG-+++++-..
S.muticumCA          TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGACGAACAAATTAAACATCAAGTTG-+++++-..
S.thunbergiiJP       TTGAAGAGTAATGAGACTTACACATGGATGTTTTTCATTTTTACCAGATTTAAGTGACGAACAAATTAAACATCAAGTTG-+++++-..
S.fallaxAU           TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGACGAGCAAATTAAAAATCAAGTTA-+++++-..
S.echinocarpumHI     TTGAAGAGTAATGAGACTTACCCAGGGATGTTTTCCATTTTTACCAAATTTAAGTGACGAGCAAATTAAAAGTCAAGTTG-++++++..
S.cristaefoliumGM    TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGACGAGCAAATTAAAAGTCAAGTTG-++++++..
S.echinocarpumHI     TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCCAATTTAAGTGACGAGCAAATTAAAAGTCAAGTTG-+++++-..
S.binderiJP          TTGAAGAGTAATGAGACTTACACAAGGATGTTTT.CATTTTTACCAGATTTAAGTGACGAGCAAATTAAAAGTCAAGTGG-+++++-..
S.patensJP           TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGACGAGCAAATTAAACATCAAGTTG-+++++-..
S.palmeriCA          TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGACGAGCAAATTAAACGTCAACTGG-+++++-..
S.piluliferumJP      TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACGTCAAGTGG-+++++-..
S.sp.MA              TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCGGATTTAAGTGATGAGCAAATTAAAA.TCAA.TGG++++++-..
S.sp.TA              TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGACGAGCAAATTAAAAGTCAA.TGG-+++++-..
S.thunbergiiCH       TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGACGAACAAATTAAACATCAAGTTG-+++++-..
S.polycystumGM       TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAACTGACGAACAATCTAATCGGCAAGTCG-+++++-..
S.polycystumMA       TGGAAAATTAATGAGACTTACCCCGGGATGTTTTCCATTTTTACC..ATT..............................++++++-..
S.sp.KL              TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACATCAAGTTG-+++++-..
S.acinariumTX        TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGTTTTAAGTGATGAGCAAATTAAACATCAAGTTG-++++++..
S.siliquosumMA       TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACATCAAGTTG-+++++-..
S.polyceratiumPA     TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACATCAAGTTG-+++++-..
S.sp.PA              TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACATCAAGTTG-+++++-..
S.polyceratiumPA     TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACATCAAGTTG-+++++-..
S.hystrixPA          TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACATCAAGTTG-+++++-..
S.filipendulaFL      TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACATCAAGTTG-++++++..
S.platycarpumGP      TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACATCAAGTTG-+++++-..
S.fuitansPEL         TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACATCAAGTTG-++++++..
S.polyceratiumGP     TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAACATCAAGTTG-++++++..
S.platycarpumPA      TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGACAAATTAAAAATCAAGTTGG-+++++-..
S.natansPEL          TTGAAGAGTAATGAGACTTACACAAGGATGTTTTTCATTTTTACCAGATTTAAGTGATGAGCAAATTAAAATCAAGTTGG-+++++-..
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Fig. 3. Continued.

was accomplished with bootstrap proportion values (Felsenstein, 1985) based on 1,000 resamplings of heuristic searches with MULPARS and the TBR swapping algorithm.

RESULTS

Field survey work.—Only three saxicolous species (*Sargassam filipendula* C. Agardh, *Sargassum acinarium* (L.) Setchell, *Sargassum mathiesonii* Kilar) were found in coastal sites from Port Aransas, TX, to Sarasota, FL. In the offshore pelagic *Sargassum* beds surveyed at Stetson Bank, TX, and Sonnier Bank, LA, three morphotypes were found. Two of these morphotypes fit traditional species descriptions for *Sargassum natans* (L.) Gaillon and *Sargassum fluitans* Børgesen, with one exemplifying an intermediary morphology (see Taylor, 1960; Schneider and Searles, 1991). Our preliminary field survey data suggested that five species and a nondescribed morphotype in the pelagic beds occupied the upper northwestern Gulf of Mexico.

Molecular analyses.—Nineteen samples representing eight species and several biogeographic isolates for the Gulf of Mexico and Caribbean were successfully extracted for total cellular DNA and included in a data set comprising 24 Pacific basin isolates (Table 1). Twelve of the 19 extracted DNAs were successfully amplified for the *rbc*LS spacer region. Twelve PCR products were sequenced and included in the present phylogenetic analysis (Table 1). All sequences were unambiguously aligned and included 400 characters, with 269 characters constant characters, 64 variable uninformative characters, and 64 informative characters (Fig. 3). The east Isthmian isolates were found to share the order- and family-specific indels found in the Pacific taxa, as shown in Figure 3, along with sharing an indel with four of the Hawaiian/Micronesian isolates (Fig. 3) (Phillips, 1998). The IGS region within *Sargasssum* species ranges from approximately 140 to 161 depending on indel events, whereas for *T. ornata,* the IGS region was 166 bases. Twenty equally most parsimonious trees (length = 202, Consistency Index = 0.728, Retention Index = 0.272, Rescaled Consistency Index = 0.617) were obtained with bootstrap values mapped on the phylogram with the most common topology (Fig. 4).

All east Isthmian species are classified in the subgenus *Sargassum,* with *S. acinarium* (L.) Setchell in the *Malacocarpicae* section, *Racemosae* subsection, *Acinariae* series and *S. filipendula* C. Agardh in the same section and subsection but in the *Siliquosae* series (Table 1). The other species are classified in the *Acanthocarpicae* section, *Glomerulatae* subsection in the *Binderiana* or *Platycarpae* series (Table 1) (J. Agardh, 1889). All east Isthmian isolates included in the molecular analysis branch within the subgenus *Sargassum,* affirming their traditional subgeneric classification (J. Agardh, 1889). Consistent with its traditional classification, the isolate representing *S. acinarium* clustered with *Malacocarpicae* species, but the subsection (*Racemosae*) and series (*Acinariae*) level ranks were not resolved (Fig. 4). The other east Isthmian isolates clustered in a biogeographic unit separate from the Pacific basin *Malacocarpicae* species in a topology incongruous with their traditional placement. Ten of these 11 isolates formed a highly diverged (10 base pairs), well-supported internal clade. Aside from the *Sargassum platycarpum* clade, little internal structure was present to support subsectional or series level ranks. Lack of clustering of the *Sargassum polyceratium* Montagne and *Sargassum echinocarpum* J. Agardh isolates suggested polyphyly for these species (Fig. 4).

In terms of overall systematic inferences within the genus, three of the four subgeneric designations, but not that of *Phyllotrichia,* were supported in the phylogeny (Fig. 4). The three sections (*Acanthocarpicae, Malacocarpicae, Zygocarpicae*) of the *Sargassum* subgenus were well supported, but the *Malacocarpicae* section appeared polyphyletic (Fig. 4). Further support for the basal branching of the tropical subgenus was obtained beyond a Pacific-based phylogenetic analysis (Phillips, 1998; Phillips et al., 2000a). The *Acanthocarpicae* species formed another well-supported clade but with little internal resolution of series designations or species boundaries. Confirming the traditional classification, the ***Bactrophycus*** species clustered together in a well-supported group sister to *Arthrophycus* subgenus.

DISCUSSION

Inferences about distributional limits of *Sargassum* within coastal regions of the Gulf of Mexico are limited at this time to the northwestern and northeastern regions between Port Aransas, TX, and Sarasota, FL. In these regions, preliminary field survey work suggests a significant reduction in species diversity over reported records (see Taylor, 1960; Schneider and Searles, 1991; Wynne, 1998), but the data must be considered cautiously until more comprehensive fieldwork is accomplished. Future field research efforts will include more seasonal data and southern regions of both Texas and Florida, providing a comprehensive distribution map for *Sargassum* species within the Gulf of Mexico.

Extensive pelagic beds of *Sargassum* occur in both the Gulf of Mexico and the Atlantic basin (Taylor, 1960; Schneider and Searles, 1991; Wynne, 1998). These beds comprise diverse and complex ecosystems that are of considerable ecological significance (e.g., Stoner and Greening, 1984; Philips and Zeman, 1990; Calder, 1995; Stachowicz and Lindquist, 1997; Schofield et al., 1998). Yet, this is the first study to evaluate the specific composition of the pelagic beds and the relationships of these to other coastal *Sargassum* species with molecular tools. Three morphotypes (one wide, one narrow, and one intermediary) were found to compose offshore pelagic beds. On the basis of morphological features, the wide morphotype fits the *S. fluitans* Børgesen concept, the narrow morphotype fits the *S. natans* (L.) Gaillon concept, with the intermediary morphotype of uncertain specific standing. Preliminary molecular analysis including isolates for both the wide and narrow morphotypes supports these morphotypes as two separate species and provides some exciting insights into possible relationships between the wide morphotype (*S. fluitans*) and the coastal species. Although weakly supported, the *S. fluitans* isolate clusters with the isolates of *S. polyceratium* and *S. filipendula* and shares a Pacific-specific indel. This evidence suggests a shared evolutionary history between these three species yet provides limited insight into species boundaries. Future expanded analyses will aid in testing boundaries

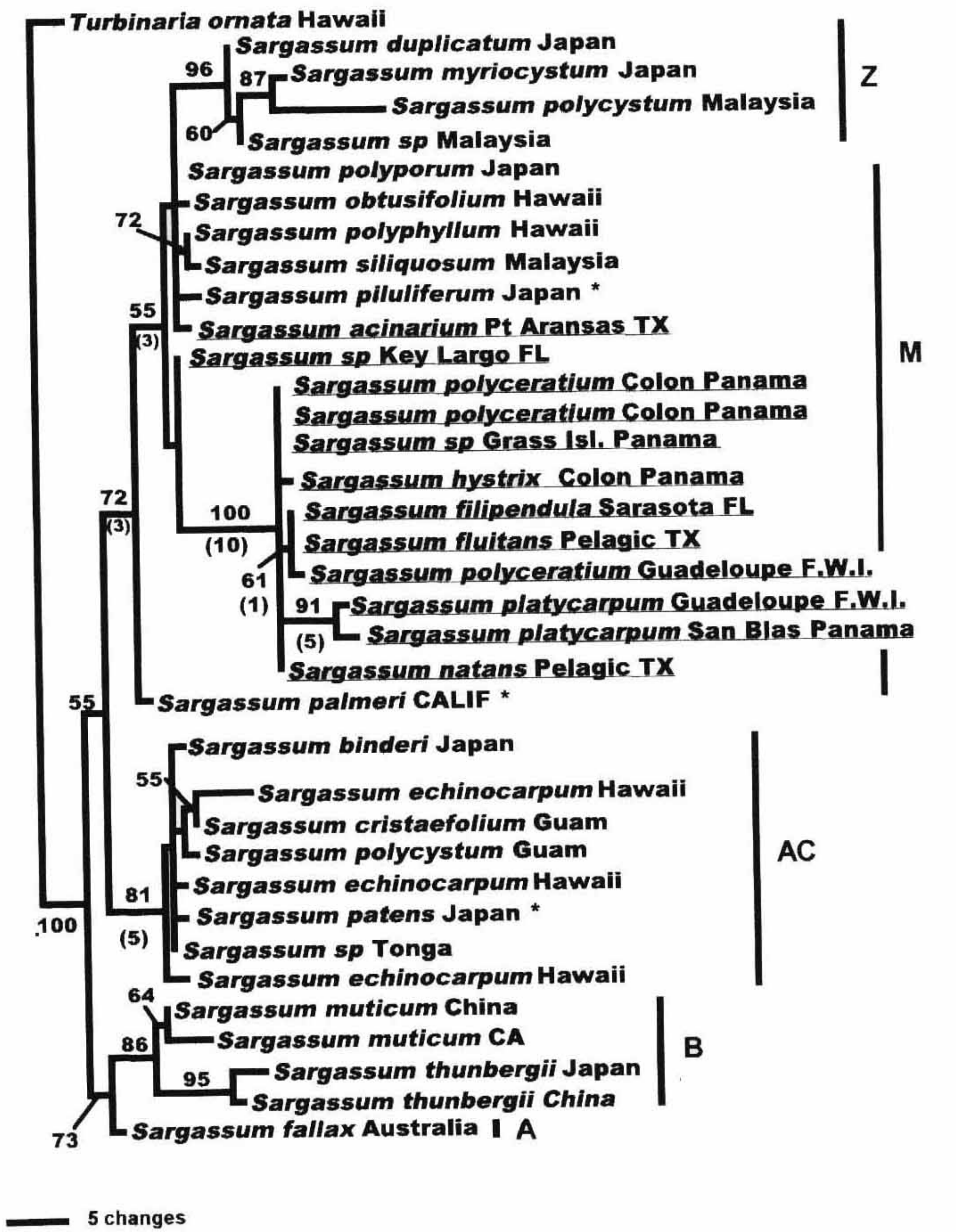

Fig. 4. Global *rbc*LS spacer region phylogram based on 36 aligned sequences including the outgroup *Turbinaria ornata* J. Agardh. Bootstrap values represent percentages based on 1,000 replications of the resampled data (Felsenstein, 1985). Numbers of base pair changes for specific nodes are in parentheses. Sequences representing the subgenus *Phyllotrichia* are asterisked, sequences representing the Gulf/Caribbean are underlined, M represents sequences for the *Malococarpicae* section, AC represents sequences for the *Acanthocarpicae* section, Z represents sequences for the *Zygocarpicae* section, B represents sequences for the subgenus *Bacrophycus*, and A represents sequences for the subgenus *Arthrophycus*.

between these three pelagic morphotypes and the coastal species.

This expanded global parsimony analysis including additional species in the sections, subsections, and series of the pantropical subgenus *Sargassum* has provided new insights into the systematics and biogeography of this genus. Preliminary analyses indicate that 10 of the 12 isolates for the Gulf of Mexico and Caribbean form a separate, well-supported species group, inconsistent with traditional sectional to series designations for the species included within this group. This result, along with a shared Pacific-specific indel, suggests a close but independent evolutionary history for these species as compared with Pacific basin *Malacocarpicae* species and reflects a history not dictated by systematics but by biogeography. It also brings into question the validity of these systematic designations and the morphological characters on which they are based. If this trend is substantiated in subsequent expanded analyses, sectional to series level designations will need to be seriously re-evaluated. But, more excitingly, if the Gulf/Caribbean species remain in a biogeographic unit, this may enable us to utilize the close of the Isthmus of Panama to time speciation events within *Sargassum*. The Gulf/Caribbean taxa also seem to share biogeographic affinities with the Californian endemic species *Sargassum palmeri*, as do the *Malacocarpicae* and *Zygocarpicae* Pacific species (Phillips, 1998; Phillips et al., 2000).

As to establishing clear species boundaries between closely related east Isthmian or Pacific *Sargassum* subgenus species, the sequence data unfortunately provide little additional resolution except for affirming the specific standing of *S. platycarpum* and the two pelagic morphotypes. These data also suggest that *S. polyceratium* of the Gulf of Mexico/Caribbean is polyphyletic as is *S. echinocarpum* of the Pacific as currently described. Hopefully, future expanded analysis based on both morphological and molecular characters will aid in establishing species boundaries within this polymorphic and difficult genus in both ocean basins.

By expanding the Pacific basin analysis to include the Gulf of Mexico/Caribbean species, we obtained confirmation of the systematic and phylogenetic trends that were observed in the Pacific basin analysis (Phillips, 1998; Phillips et al., 2000) while obtaining greater resolution of the basal relationships of the subgenus *Sargassum*. As we continue to expand these analyses, hopefully more and more exciting results will unfold and aid us in meeting our long-term systematic and phylogenetic research goals within this species-rich and difficult genus.

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