

Supplementary Material

Integrative taxonomy in *Syllis prolifera* (Annelida, Syllidae): from a unique cosmopolitan species to a complex of pseudocryptic species

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Table S1. Sampling localities, substrates, collecting date, coordinates, catalogue numbers and GenBank accession numbers for all specimens used in the analysis.

Species	Code	Locality	Substrate	Collection date	Coordinates (decimal degrees)	Museum numbers	16S	COI
<i>Branchiosyllis cirropunctata</i> (Michel, 1909)	<i>Branchiosyllis cirropunctata</i>	Green Head, Australia	–	–	–	AMW.37728	JF903690	–
<i>Branchiosyllis exilis</i> (Gravier, 1900)	<i>Branchiosyllis exilis</i>	Darwin NT, Australia	–	–	–	AMW.37714	JF903692	–
	<i>Branchiosyllis exilis</i> 1	Port de la Selva, Girona, Spain	–	–	–	MNCN/ADN9571	–	EF123746
	<i>Branchiosyllis exilis</i> 2	Shark Bay, Australia	–	–	–	AMW.33389	JF903691	JF903783
	<i>Branchiosyllis exilis</i> 3	Coconut Island, Hawaii, USA	–	–	–	MNCN16.01/13275	JF903693	–
<i>Branchiosyllis thylacine</i> San Martín, Hutchings & Aguado, 2008	<i>Branchiosyllis thylacine</i>	Port Jackson New South Wales, Australia	–	–	–	AMW.37700	JF913951	–
<i>Haplosyllis spongicola</i> (Grube, 1855)	<i>Haplosyllis spongicola</i>	Banyuls-sur-mer, France	–	–	–	MNCN/ADN9578	EF123791	EF123751
	<i>Haplosyllis spongicola</i> 1	Shark Bay, Australia	–	–	–	AMW.33394	JF903698	–
<i>Syllis albae</i> Álvarez-Campos & Verdes, 2017	<i>Syllis albae</i>	ECIM beach, Las Cruces, Chile	Holdfasts of <i>Lessonia spicata</i> , intertidal	18 Jan 2013	33.501666, –71.631944	MNCN16.01/17500	KX792205	KX792209
<i>Syllis armillaris</i> (O.F. Müller, 1776)	<i>Syllis armillaris</i>	Cala Ratjada, Mallorca, Spain	–	–	–	MNCN16.01/13284	JF903727	–
<i>Syllis bella</i> (Chamberlin, 1919)	<i>Syllis bella</i> BA	'Sepok Point', between Balayan Bay and Batangas Bay, Luzon Island, Philippines	Coral rubble, 6-m depth	10 Dec 2010	13.683889, 120.895833	MNCN/ADN 85712	KX084859	KX084926
<i>Syllis crassicirrata</i> (Treadwell, 1925)	<i>Syllis crassicirrata</i> NZ	Cavalli Islands, Rainbow Warrior wreck, New Zealand	<i>Aglaophenia</i> sp. hydrozoans, 25-m depth	–	–36.842405, 174.771577	MNCN16.01/16869	KU182889	KU182896
<i>Syllis ehlersioides</i> (Marenzeller, 1890)	<i>Syllis ehlersioides</i>	Manazuru Peninsula, Japan	–	–	–	MNCN/ADN9606	EF123808	EF123773
<i>Syllis ferrari</i> Alós & Campoy, 1981	<i>Syllis ferrari</i>	Port de la Selva, Girona, Spain	–	–	–	MNCN/ADN 9608	EF123809	EF123775
<i>Syllis garciai</i> (Campoy, 1982)	<i>Syllis garciai</i>	O Grove, Galicia, Spain	–	–	–	MNCN/ADN9609	–	EF123776
<i>Syllis gracilis</i> (Grube, 1840)	<i>Syllis gracilis</i>	O Grove, Galicia, Spain	–	–	–	MNCN/ADN9611	EF123811	EF123778
	<i>Syllis gracilis</i> 2	Prince of Wales Island, Australia	–	–	–	–	JF903733	–
	<i>Syllis gracilis</i> AU5	MacGillivray Reef, Lizard Island, Australia	Coral rubble, 10-m depth	31 Aug 2010	–	AMW.41619	KU182891	KU182893
<i>Syllis hyalina</i> Grube, 1863	<i>Syllis hyalina</i>	Port de la Selva, Girona, Spain	–	–	–	MNCN/ADN9612	EF123818	EF123779
<i>Syllis magdalena</i> Wesenberg-Lund, 1962	<i>Syllis magdalena</i> CHI	ECIM beach, Las Cruces, Chile	Holdfasts of <i>Lessonia spicata</i> , intertidal	13 Jan 2014	33.501666, –71.631944	MNCN16.01/17475	KX792206	KX792211
<i>Syllis marugani</i> Aguado, San Martín & Nishi, 2006	<i>Syllis marugani</i>	Manazuru Peninsula, Japan	–	–	–	MNCN/ADN9614	EF123812	EF123780
<i>Syllis monilata</i> (Imajima, 1966)	<i>Syllis monilata</i>	Manazuru Peninsula, Japan	–	–	–	MNCN/ADN9615	EF123819	EF123781
<i>Syllis okadae</i> Fauvel, 1934	<i>Syllis okadae</i>	Manazuru Peninsula, Japan	–	–	–	MNCN/ADN9617	EF123814	EF123783
<i>Syllis patriciae</i> (Hartmann-Schröder, 1981)	<i>Syllis patriciae</i>	Shark Bay, Australia	–	–	–	–	JF903738	JF903781
<i>Syllis pectinans</i> Haswell, 1920	<i>Syllis pectinans</i> CHI	ECIM beach, Las Cruces, Chile	Holdfasts of <i>Lessonia spicata</i> , intertidal	13 Jan 2014	33.501666, –71.631944	MNCN16.01/17482	–	KX792210
<i>Syllis picta</i> (Kinberg, 1866)	<i>Syllis picta</i> AU134	High Rock, Lizard Island, Australia	Coral rubble, 20.1 m depth	11 Sep 2010	–	AMW41634	KU182890	–
<i>Syllis pigmentata</i> (Chamberlin, 1919)	<i>Syllis pigmentata</i>	Manazuru Peninsula, Japan	–	–	–	MNCN/ADN9607	–	EF123774
<i>Syllis prolifera</i> Krohn, 1852	SproGB01	Port Jackson, New South Wales, Australia	–	–	–	AMW.37076	JF903746	JF903780
	Spro0601	Casaraccio Pond, Sácer, Italy	<i>Mytilaster</i> sp., brackish	10 May 2013	40.912, 8.221	MNCN16.01/19318	–	PP494895
	Spro0602	–	–	–	–	MNCN16.01/19319	–	PP494893
	Spro0603	–	–	–	–	MNCN16.01/19320	–	PP494888
	Spro0604	–	–	–	–	MNCN16.01/19321	–	PP494894
	Spro0605	–	–	–	–	MNCN16.01/19322	–	PP494897
	Spro0606	–	–	–	–	MNCN16.01/19323	–	PP494890
	Spro0607	–	–	–	–	MNCN16.01/19324	–	PP494891
	Spro0608	–	–	–	–	MNCN16.01/19325	–	PP494896
	Spro0609	–	–	–	–	MNCN16.01/19326	–	PP494892
	Spro0610	–	–	–	–	MNCN16.01/19327	–	PP494889
	SproGB02	Cala Ratjada, Mallorca, Spain	–	–	–	MNCN16.01/13286	JF903740	–
	Spro0401	Port of Livorno, Livorno, Italy	<i>Corallina</i> sp., infralittoral	6 May 2013	43.534, 10.29	MNCN16.01/19328	–	PP494880
	Spro0402	–	–	–	–	MNCN16.01/19329	–	PP494886
	Spro0403	–	–	–	–	MNCN16.01/19330	–	PP494884
	Spro0404	–	–	–	–	MNCN16.01/19331	–	PP494881
	Spro0405	–	–	–	–	MNCN16.01/19332	–	PP494885
	Spro0406	–	–	–	–	MNCN16.01/19333	–	PP502860
	Spro0407	–	–	–	–	MNCN16.01/19334	–	PP494883
	Spro0504	Manarola, La Spezia, Italy	–	31 Mar 2013	44.107, 9.067	MNCN16.01/19335	–	PP502861
	Spro0505	–	–	–	–	MNCN16.01/19336	–	PP494882
	Spro0506	–	–	–	–	MNCN16.01/19337	–	PP494887
	Spro0507	–	–	–	–	MNCN16.01/19338	–	PP494885
	Spro0509	–	–	–	–	MNCN16.01/19339	–	PP502859
	SproGB03	Cabo de Gata, Almería, Spain	–	–	–	MNCN16.01/13285	JF903739	–
	Spro0108	La Caleta, Cádiz, Spain	<i>Corallina</i> sp., <i>Codium bursa</i> , unidentified sponges, and <i>Mesophyllum lichenoides</i> , intertidal	31 Aug 2015	36.527222, –6.313333	MNCN16.01/19340	–	PP495223
	Spro0117	–	<i>Corallina</i> sp., <i>Halopteris scoparia</i> , intertidal	29 Nov 2015	36.532222, –6.310833	MNCN16.01/19341	–	PP494868
	Spro0118	–	<i>Corallina</i> sp., unidentified sponges, and <i>Mesophyllum lichenoides</i> , intertidal	–	36.5325, –6.3125	MNCN16.01/19342	–	PP494866
	Spro0119	–	<i>Halopteris scoparia</i> , intertidal	–	36.5325, –6.3125	MNCN16.01/19343	–	PP494865
	Spro0120	–	<i>Corallina</i> sp., intertidal	12 Dec 2015	36.527222, –6.313333	MNCN16.01/19344	–	PP494860
	Spro0121	–	<i>Halopteris scoparia</i> , <i>Dyctiota dicotoma</i> , and unidentified brown algae, intertidal	12 Jul 2018	36.525833, –6.321666	MNCN16.01/19345	–	PP494861
	Spro0122	–	–	–	–	MNCN16.01/19346	–	PP502855

Species	Code	Locality	Substrate	Collection date	Coordinates (decimal degrees)	Museum numbers	16S	COI
	Spro0123					MNCN16.01/19347	PP495227	PP494862
	Spro0124					MNCN16.01/19348	PP495228	PP494863
	Spro0125					MNCN16.01/19349	–	PP494867
	Spro0126					MNCN16.01/19350	–	PP494864
	Spro0127					MNCN16.01/19351	–	PP502854
Lineage 4	Spro0116		<i>Corallina</i> sp., <i>Halopteris scoparia</i> , intertidal	28 Nov 2015		MNCN16.01/19352	–	PP502862
	Spro0107		<i>Corallina</i> sp., <i>Codium bursa</i> , unidentified sponges, and <i>Mesophyllum lichenoides</i> , intertidal	31 Aug 2015	36.527222, –6.313333	MNCN16.01/19353	–	PP494869
Lineage 5	Spro0301	Isla Capraia, Livorno, Italy	<i>Corallina</i> sp., infralittoral	18 Apr 2013	43.015, 9.822	MNCN16.01/19354	–	PP502856
	Spro0302					MNCN16.01/19355	PP502914	PP495222
	Spro0303					MNCN16.01/19356	–	PP494874
	Spro0304					MNCN16.01/19357	–	PP494877
	Spro0305					MNCN16.01/19358	–	PP494879
	Spro0306					MNCN16.01/19359	–	PP494872
	Spro0307					MNCN16.01/19360	–	PP494873
	Spro0308					MNCN16.01/19361	–	PP494870
	Spro0309					MNCN16.01/19362	–	PP494871
	Spro0310					MNCN16.01/19363	–	PP502857
	Spro0501	Manarola, La Spezia, Italy		31 Mar 2013	44.107, 9.067	MNCN16.01/19364	–	PP494878
	Spro0502				44.107, 9.067	MNCN16.01/19365	–	PP494876
	Spro0510				44.107, 9.067	MNCN16.01/19366	–	PP494875
<i>Syllis setoensis</i> (Imajima, 1966)	<i>Syllis setoensis</i> 1 BA	"Koala Point", Balayan Bay, Luzon Island, Philippines	Coral rubble, 5–16-m depth	5 Dec 2010	13.6975, 120.829167	MCZ25418	–	KU182894
	<i>Syllis setoensis</i> 2 BA	'Sepok Wall', between Balayan Bay and Batangas Bay, Luzon Island, Philippines	Coral rubble, 6–13-m depth	10 Dec 2010	13.683889, 120.895833	MNCN16.01/16870	–	KU182895
<i>Syllis tamarae</i> Álvarez-Campos & Verdes, 2017	<i>Syllis tamarae</i>	ECIM beach, Las Cruces, Chile	Holdfasts of <i>Lessonia spicata</i> , intertidal	12 Jan 2014	33.501666, –71.631944	MNCN16.01/17471	KX792208	KX792213
<i>Syllis tripantu</i> Álvarez-Campos & Verdes, 2017	<i>Syllis tripantu</i>	ECIM beach, Las Cruces, Chile	Holdfasts of <i>Lessonia spicata</i> , intertidal	18 Jan 2013	33.501666, –71.631944	MCZ25200	KX792207	KX792212
<i>Syllis vittata</i> Grube, 1840	<i>Syllis vittata</i>	Taipingjiao, China	<i>Sargassum thunbergii</i>	20 Mar 2008	36.033333, 120.35	OUC.POLYJ4.0808	GU362678	GU362691
<i>Syllis ypsiloides</i> Aguado, San Martín & ten Hove, 2008	<i>Syllis ypsiloides</i>	Coconut Island, Hawaii, USA	–	–	–	MNCN16.01/13291	JF903745	–

New generated sequences in bold.

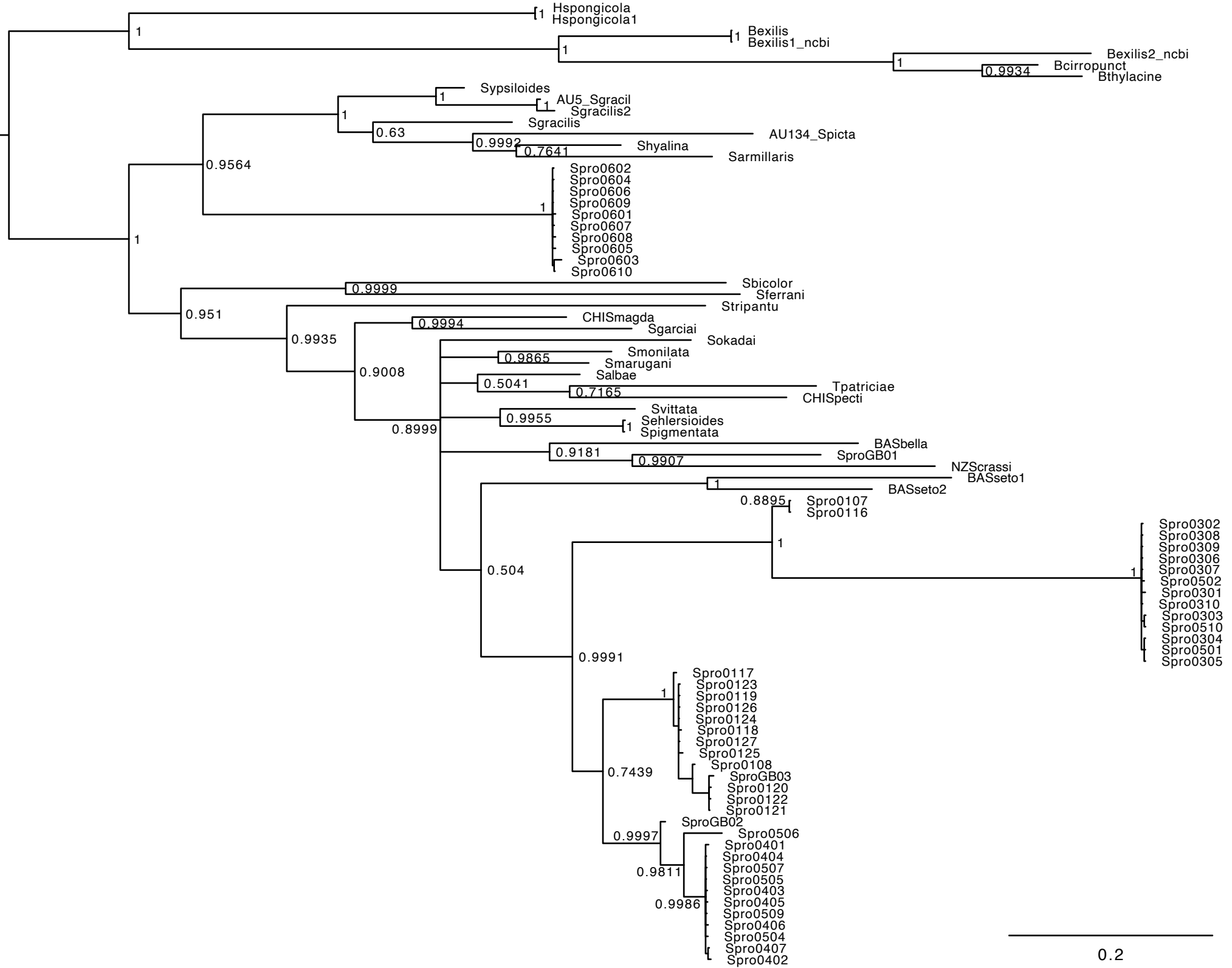


Fig. S1. Phylogenetic reconstruction of *Syllis prolifera* populations and other Syllinae species built by Bayesian Inference (BI) based on the concatenated mitochondrial dataset (16S rRNA and COI). Posterior probability scores are indicated in the nodes.

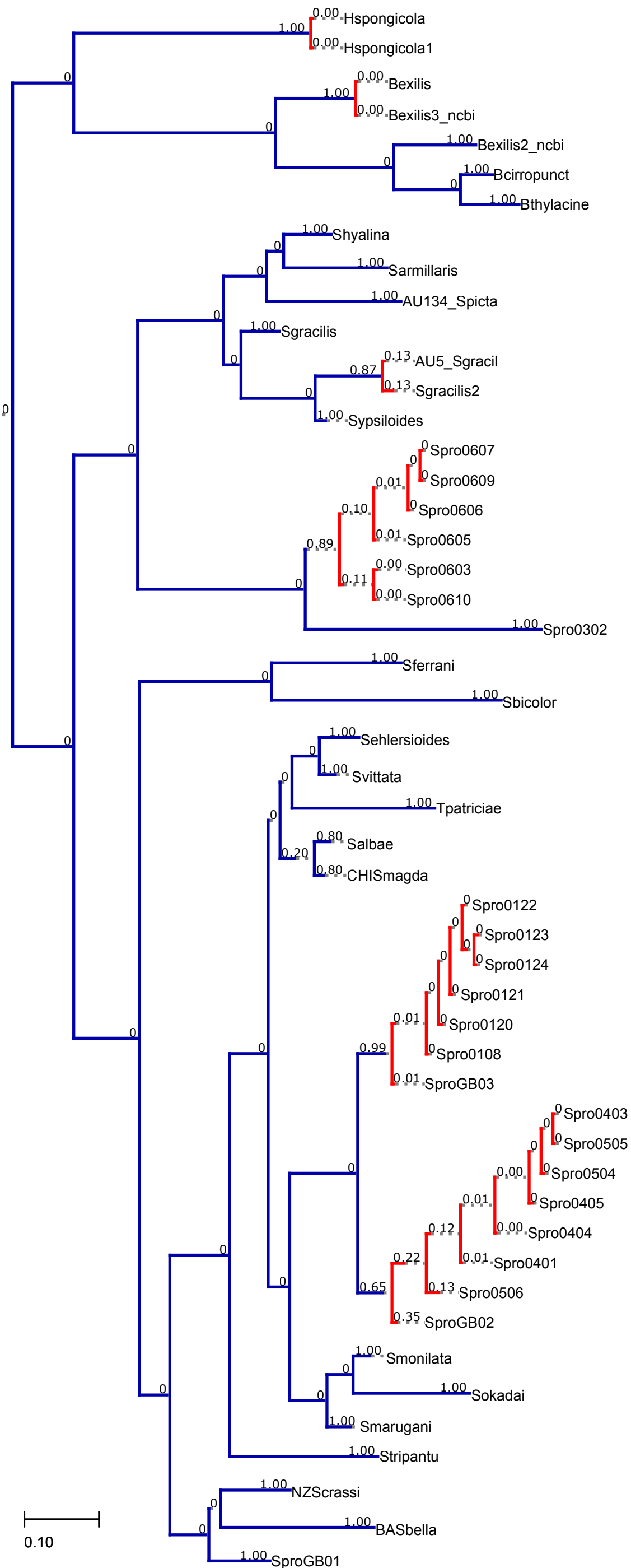


Fig. S2. bPTP results for *Syllis prolifera* species complex using 16S rRNA.

subsets/asap scores [2/5.0] [3/6.5] [1/2.0] [9/11.5] [4/9.0] [7/11.0] [8/11.5] [10/13.5] [6/11.0] [5/9.5]

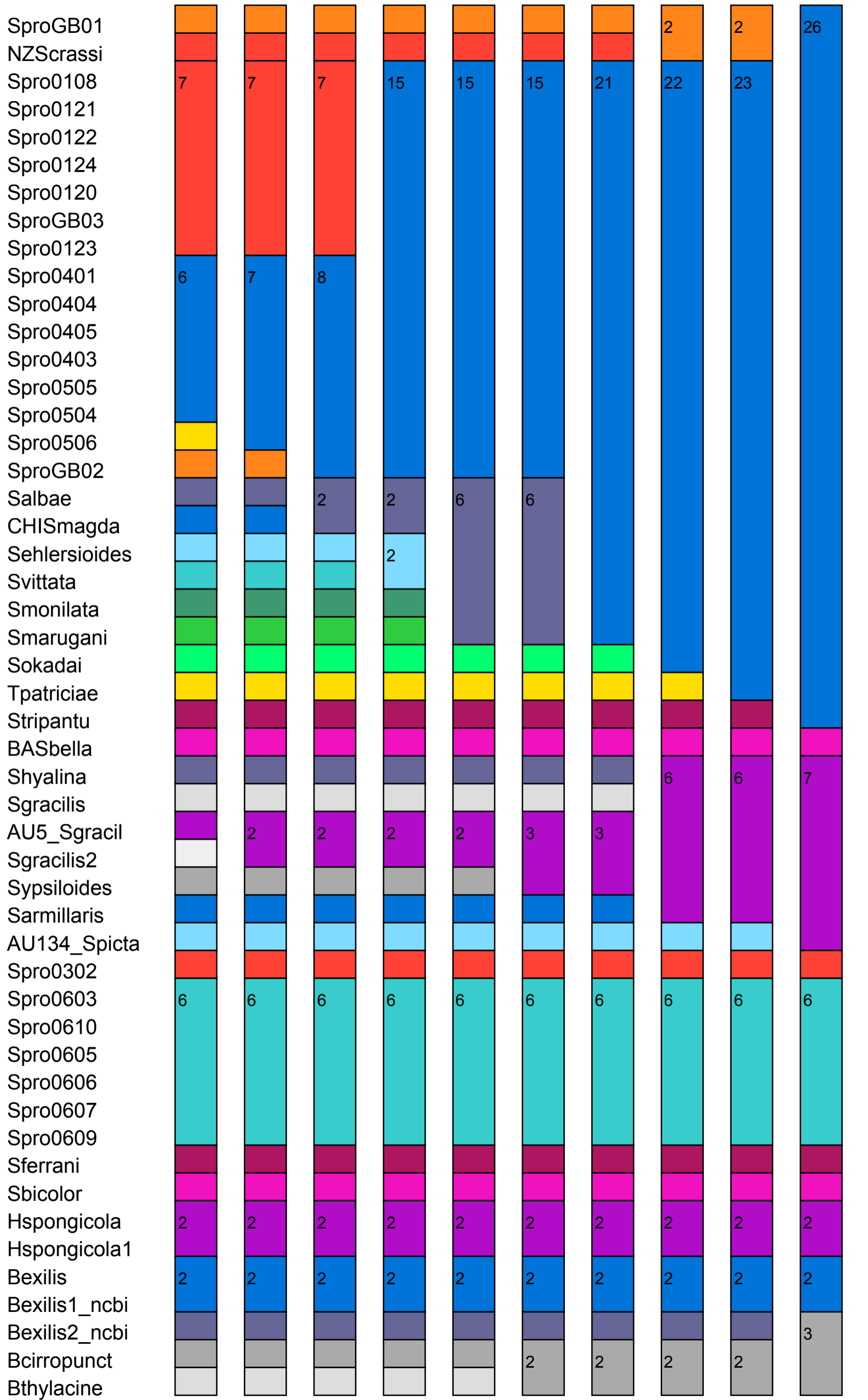


Fig. S3. ASAP results for *Syllis prolifera* species complex using 16S rRNA.

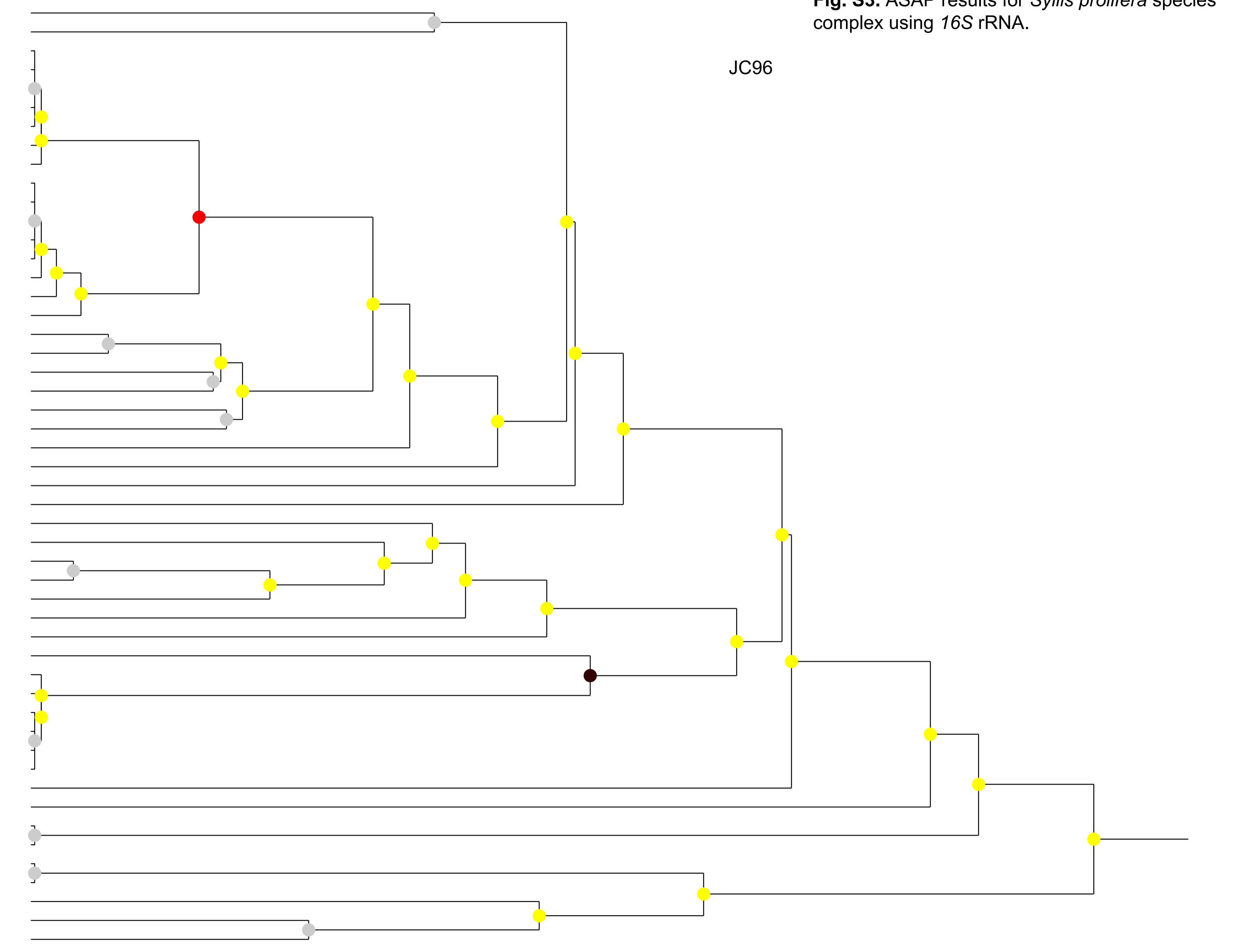


Fig. S3. (Cont.)

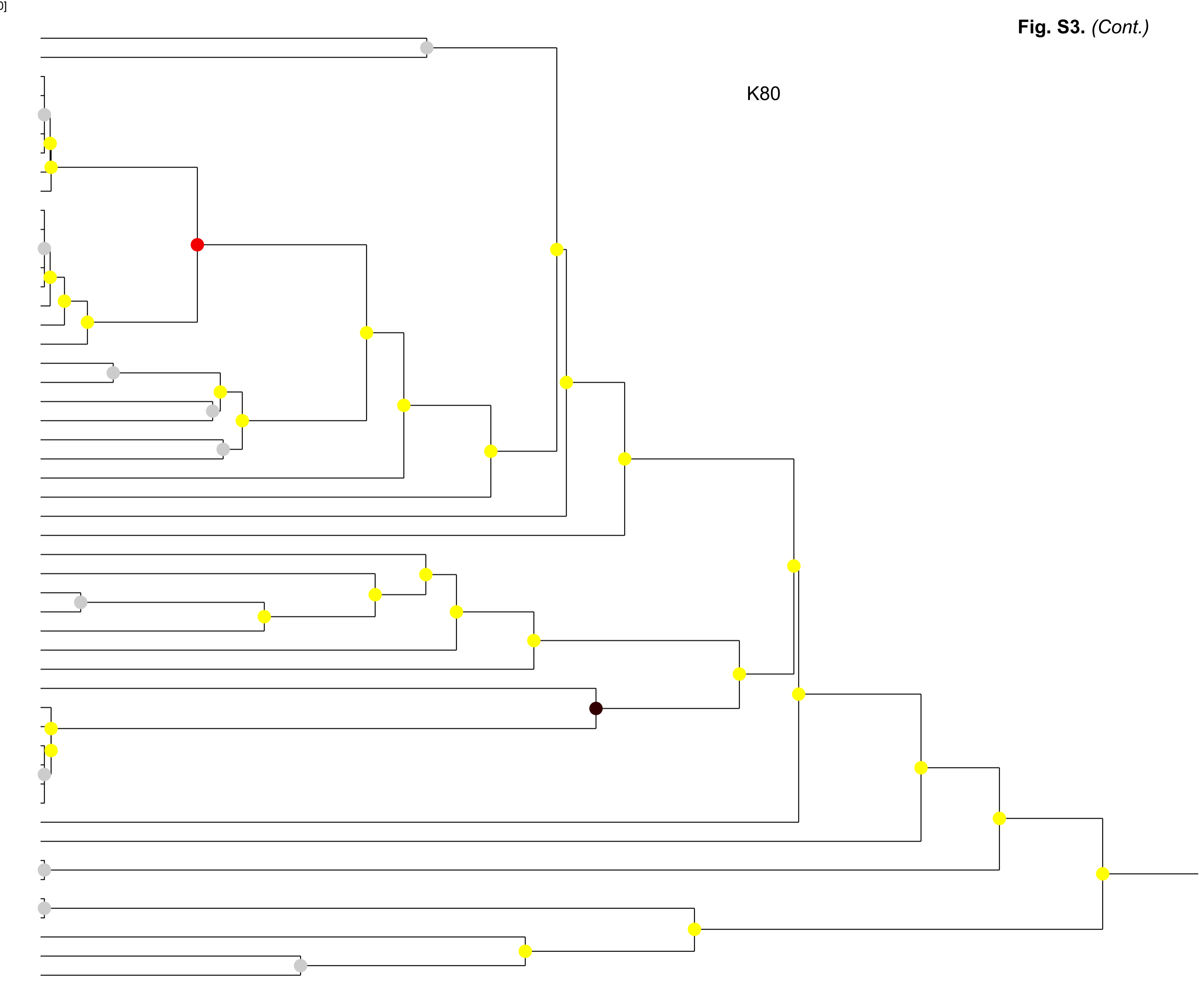
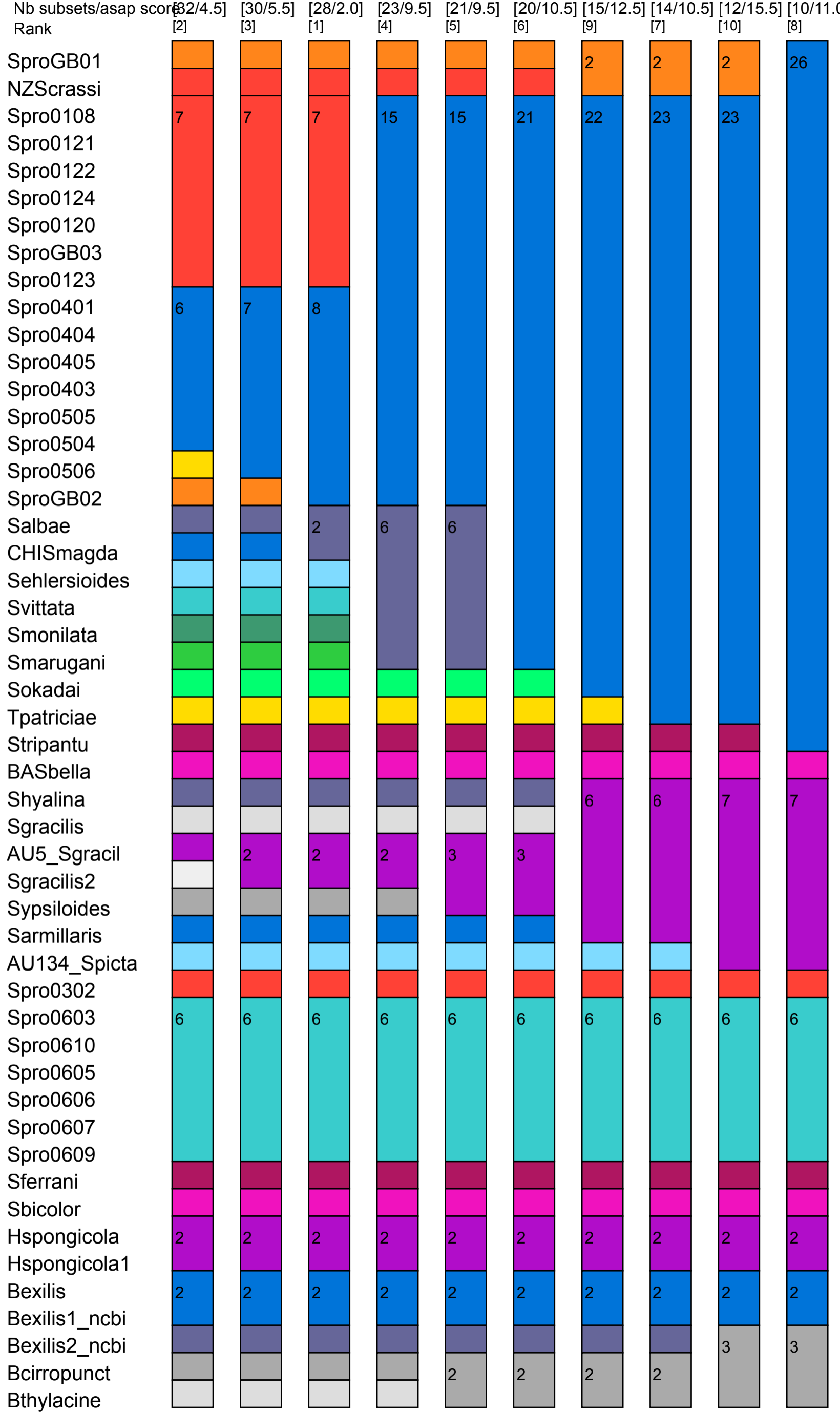
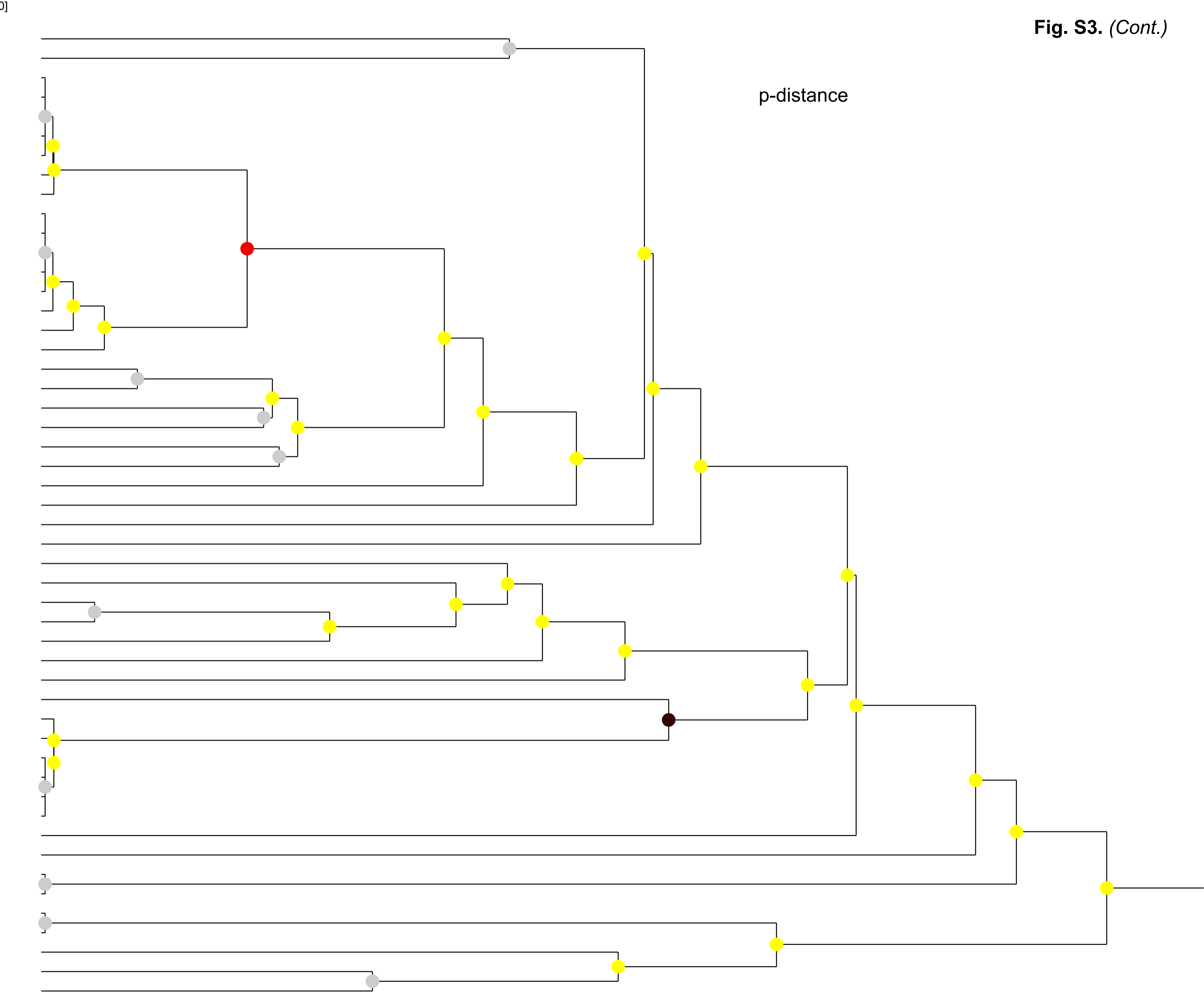
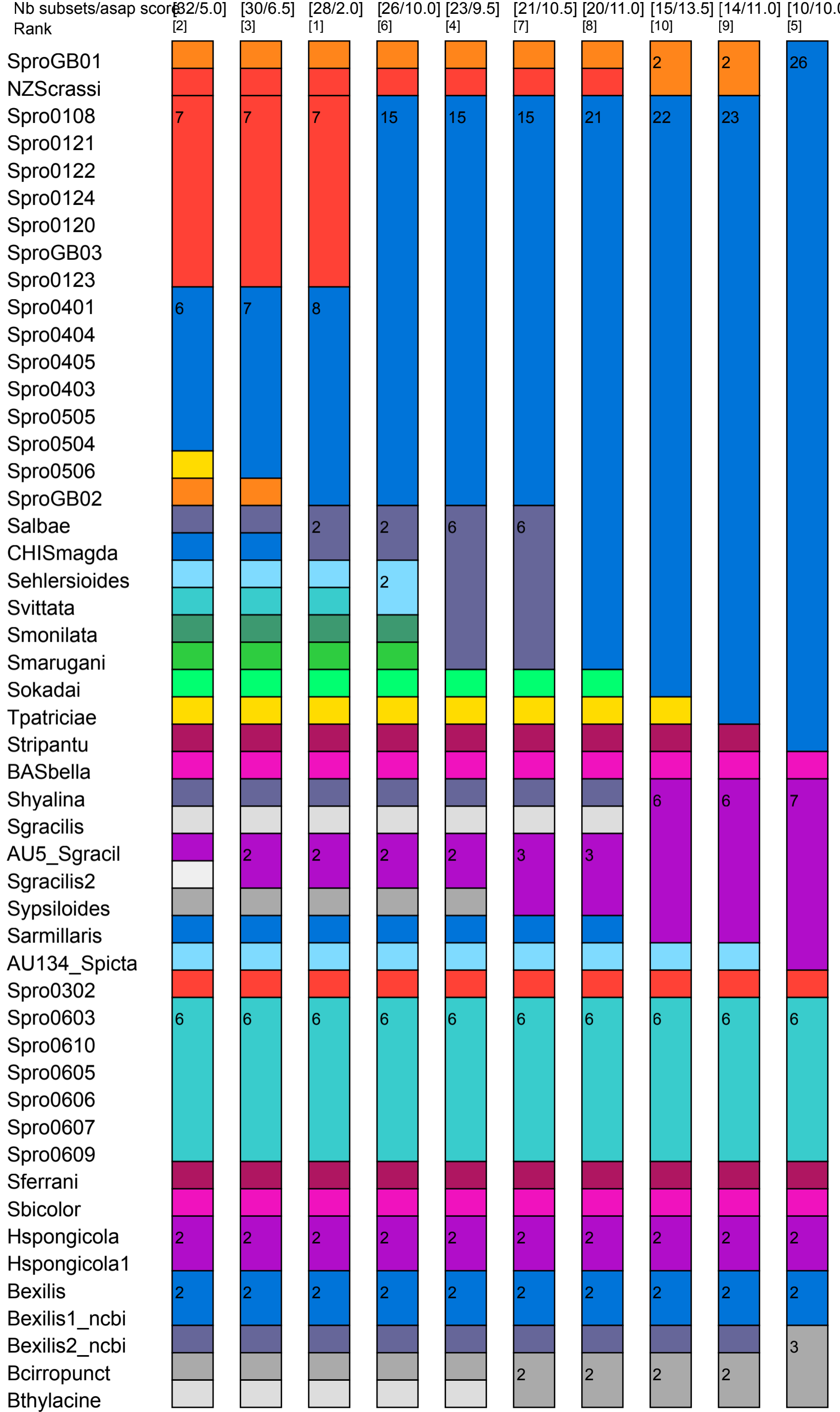


Fig. S3. (Cont.)



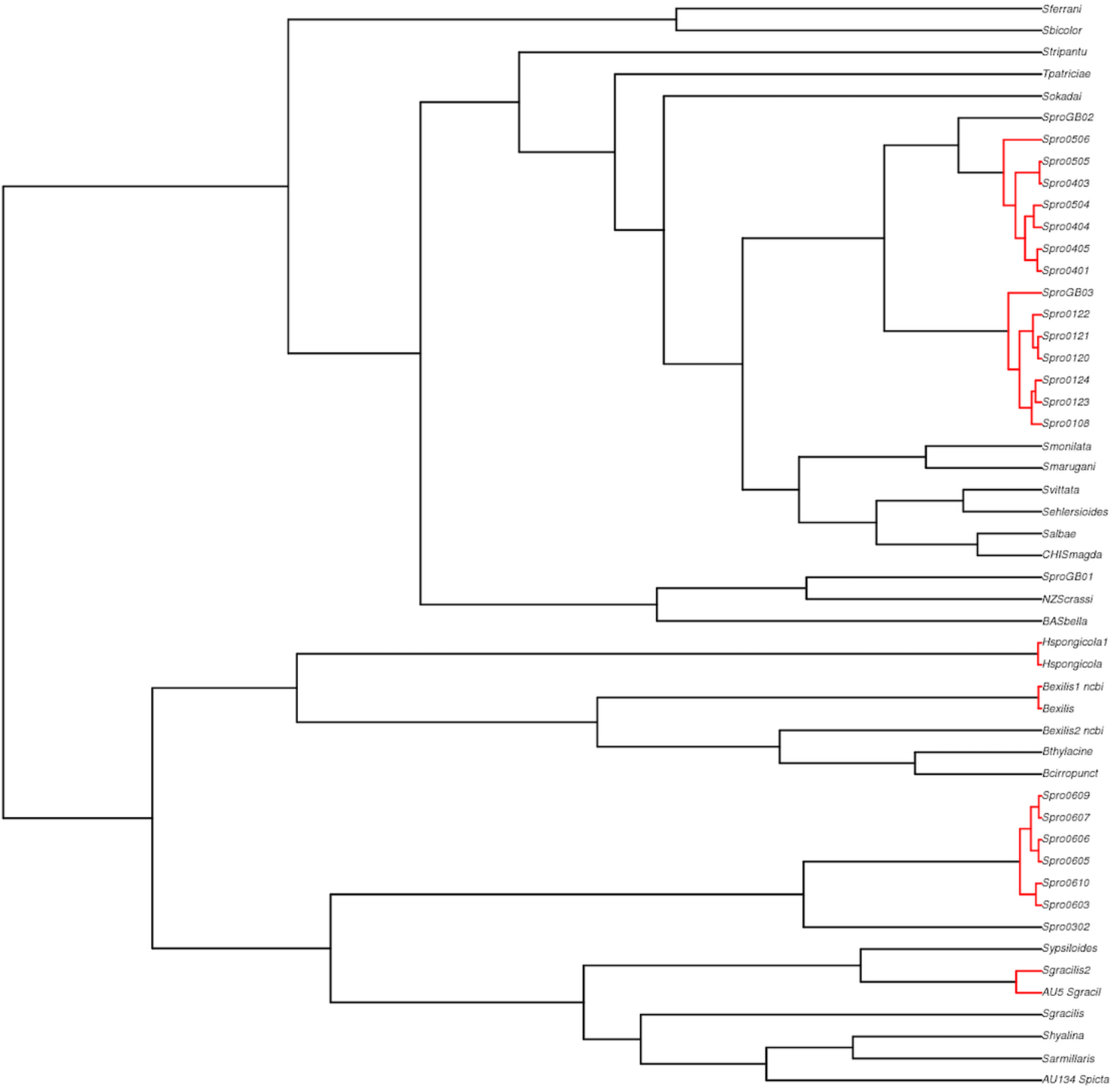
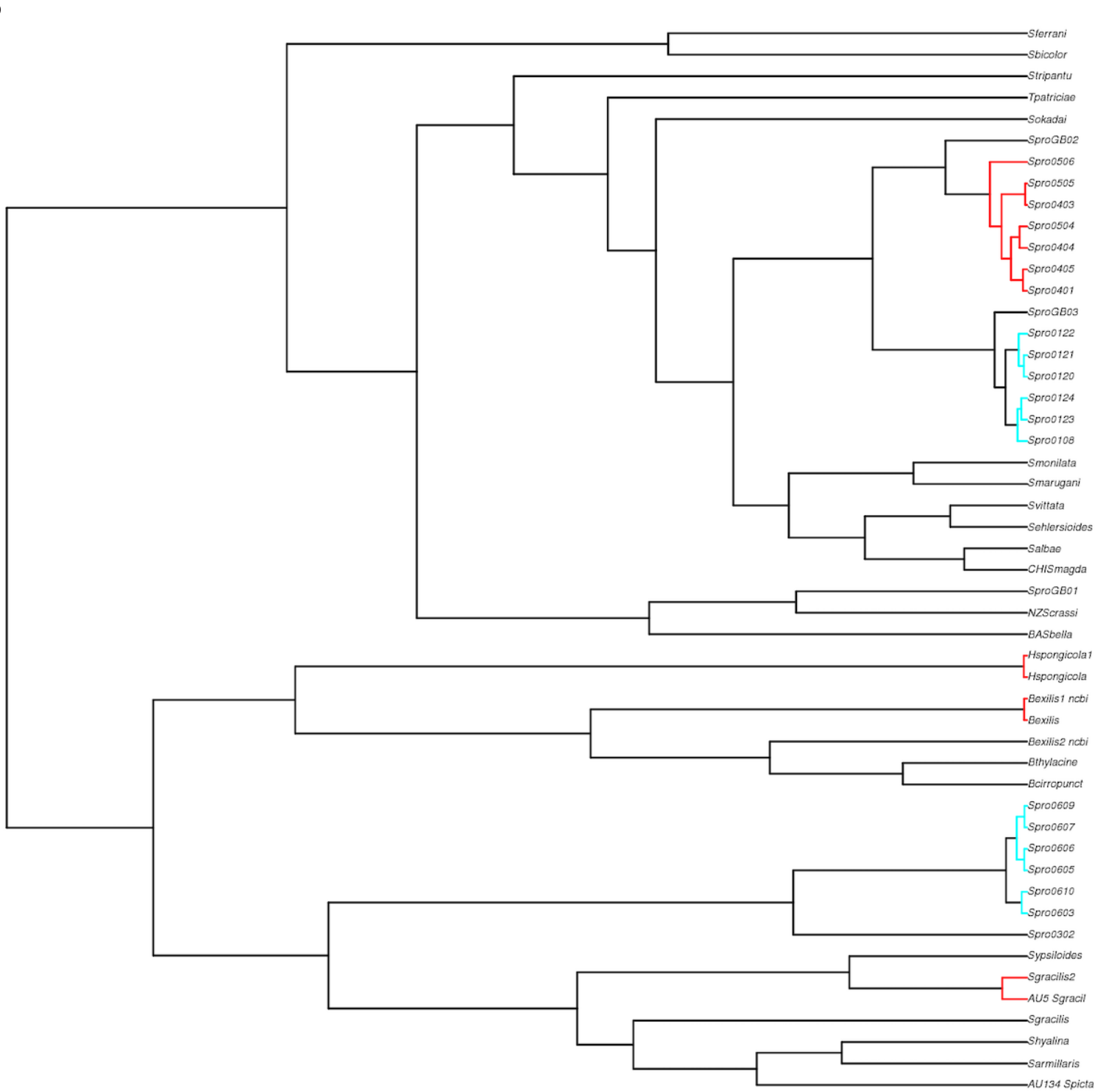
A**B**

Fig. S4. GMYC results for *Syllis prolifera* species complex using 16S rRNA. A, single-threshold variant. B, multi-threshold variant.

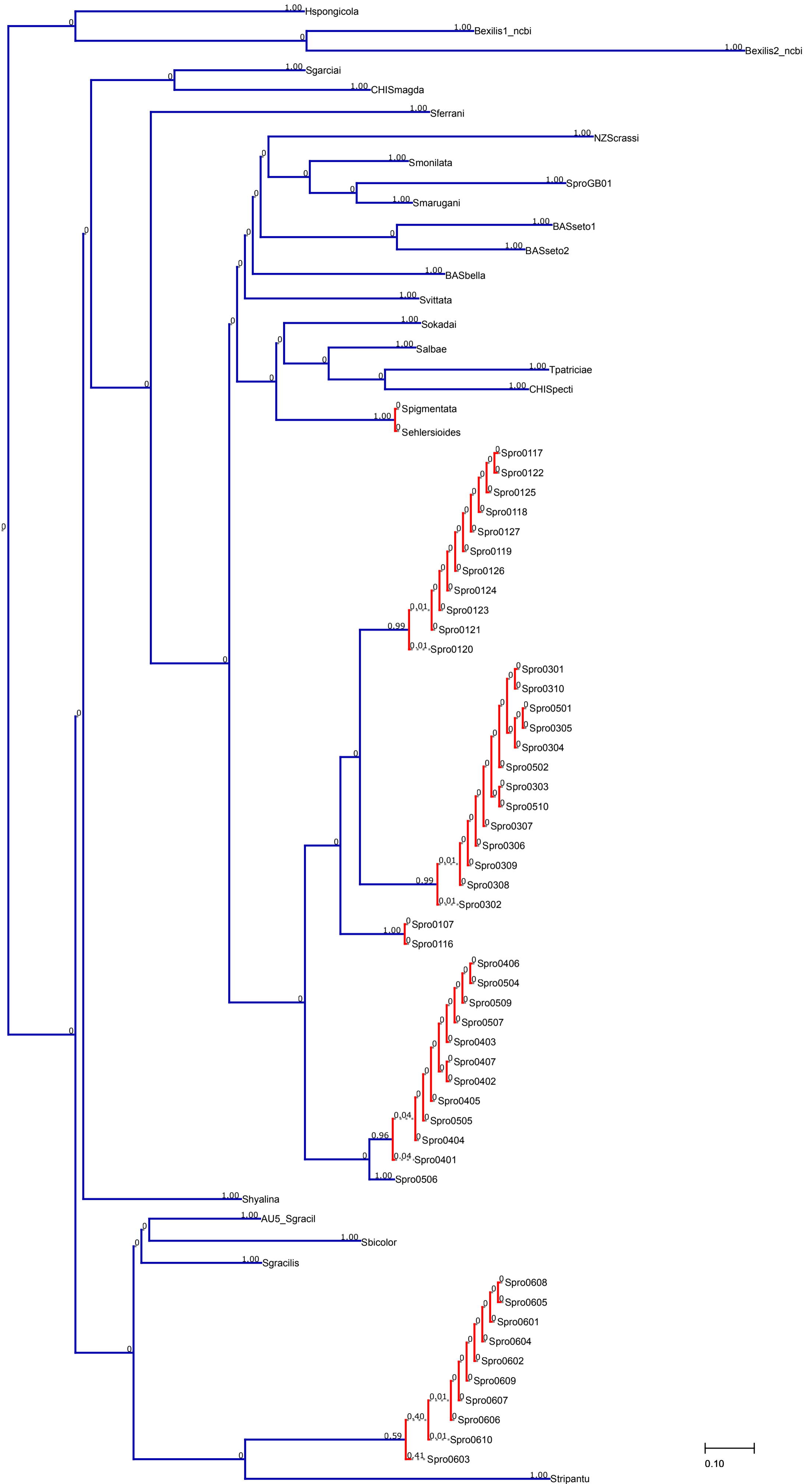


Fig. S5. bPTP results for *Syllis prolifera* species complex using COI.

subsets/asap score [33/17.5] [32/9.5] [31/4.0] [30/1.5] [30/1.5] [30/4.0] [28/14.0] [25/11.5] [23/13.0] [10/8.5]

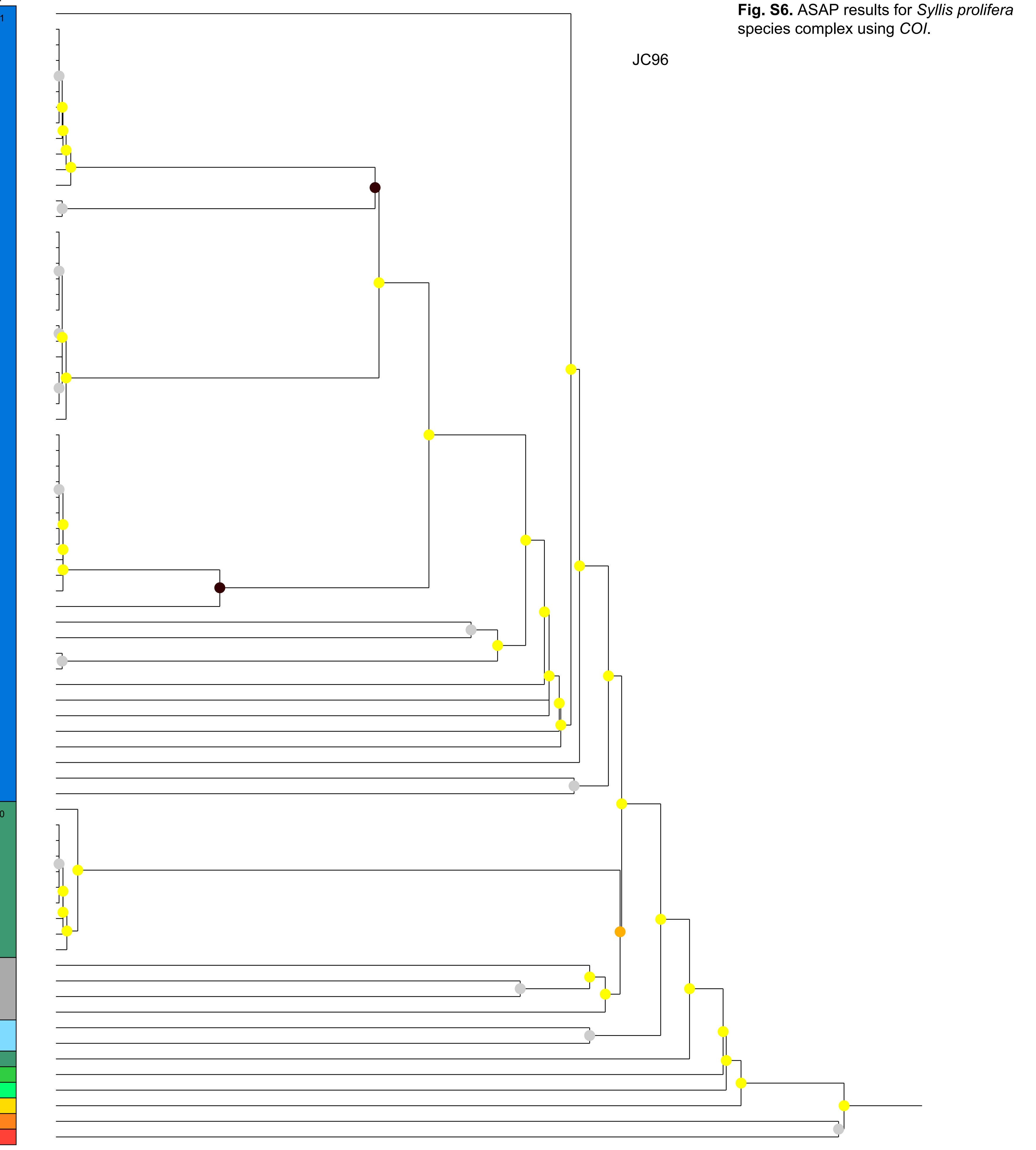
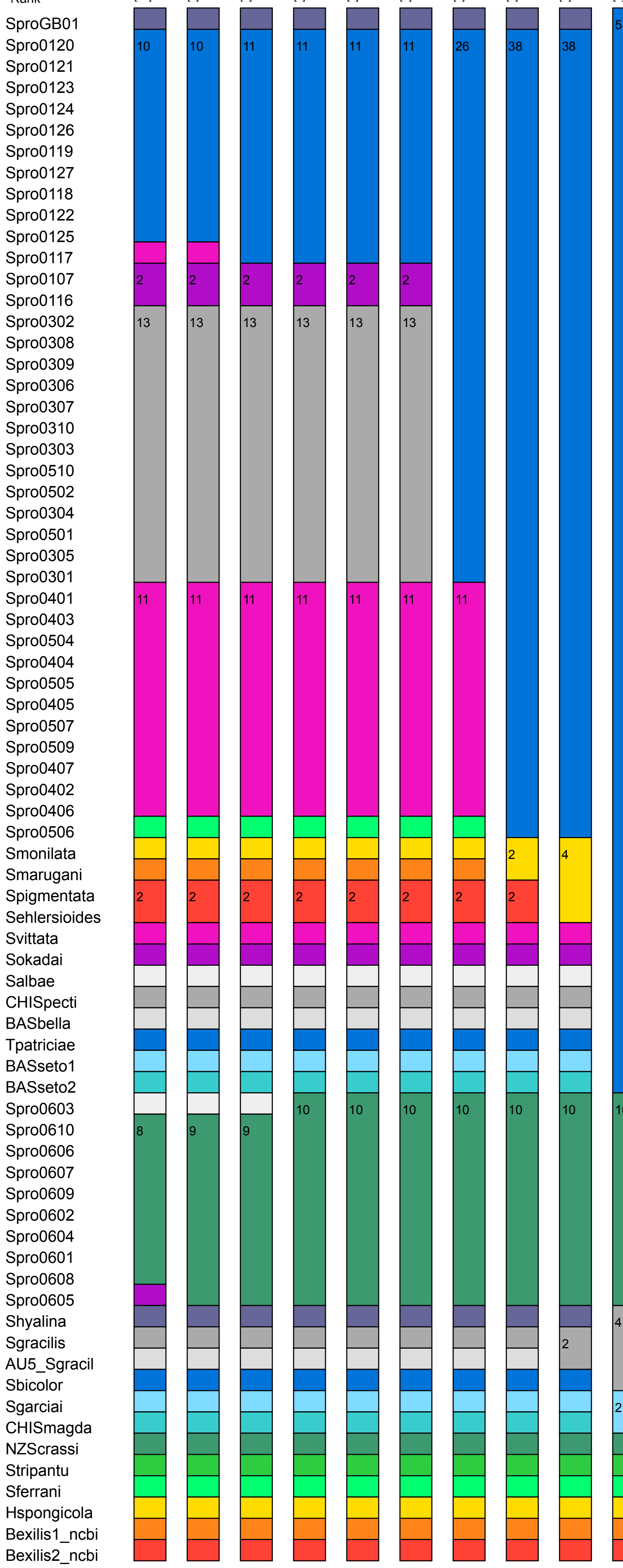


Fig. S6. ASAP results for *Syllis prolifera* species complex using *COI*.

nb subsets/asap score [52/10.5] [31/4.5] [30/1.5] [30/1.5] [30/4.0] [28/13.5] [25/11.5] [13/16.0] [10/13.0] [2/15.5]

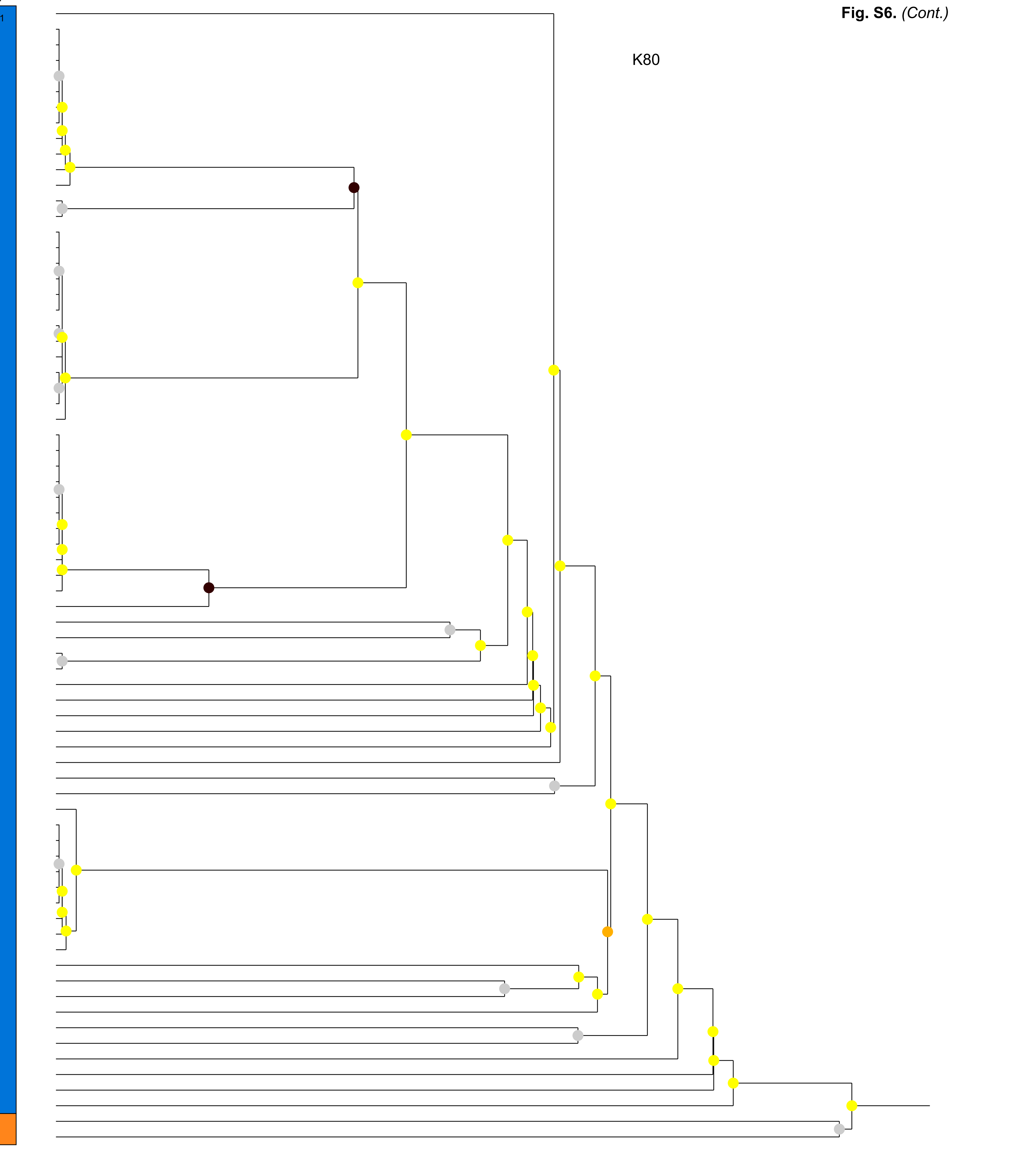
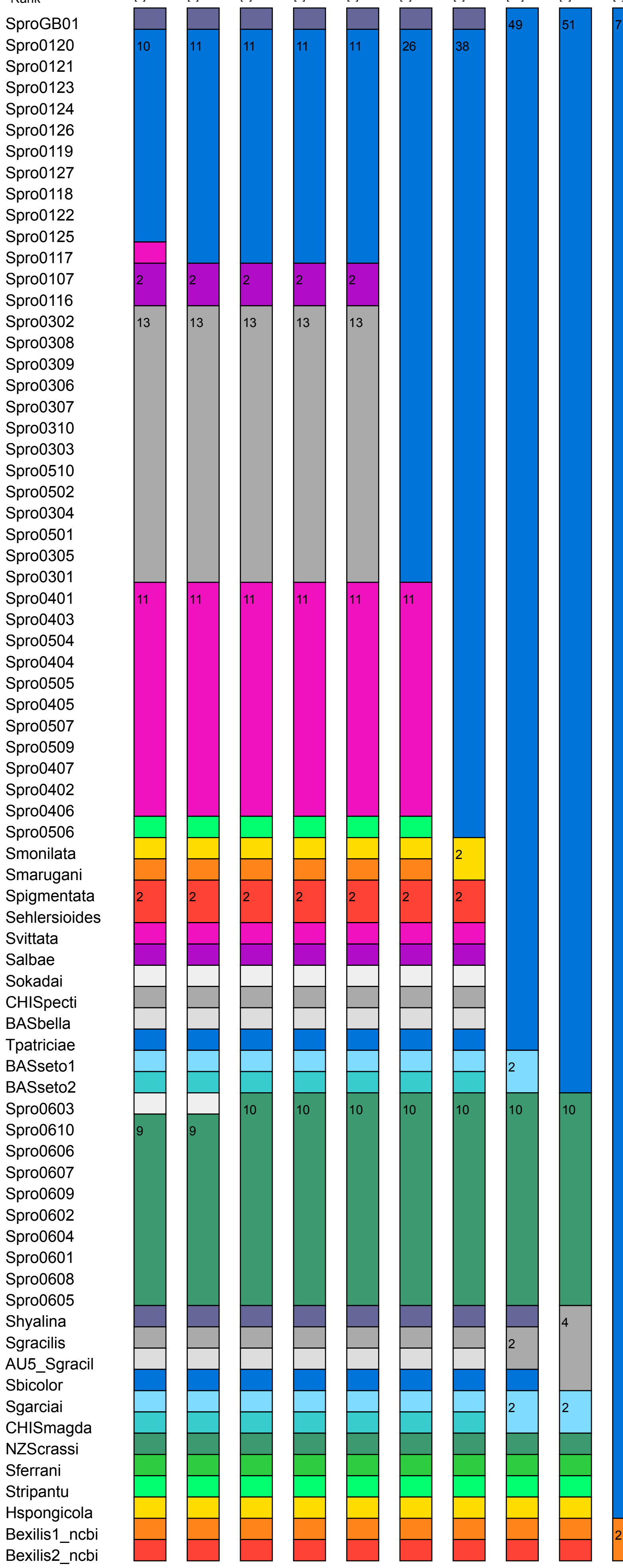


Fig. S6. (Cont.)

nb subsets/asap score [33/17.0] [32/8.5] [31/4.0] [30/1.5] [30/1.5] [30/4.0] [28/11.5] [25/12.5] [23/14.5] [10/10.5]

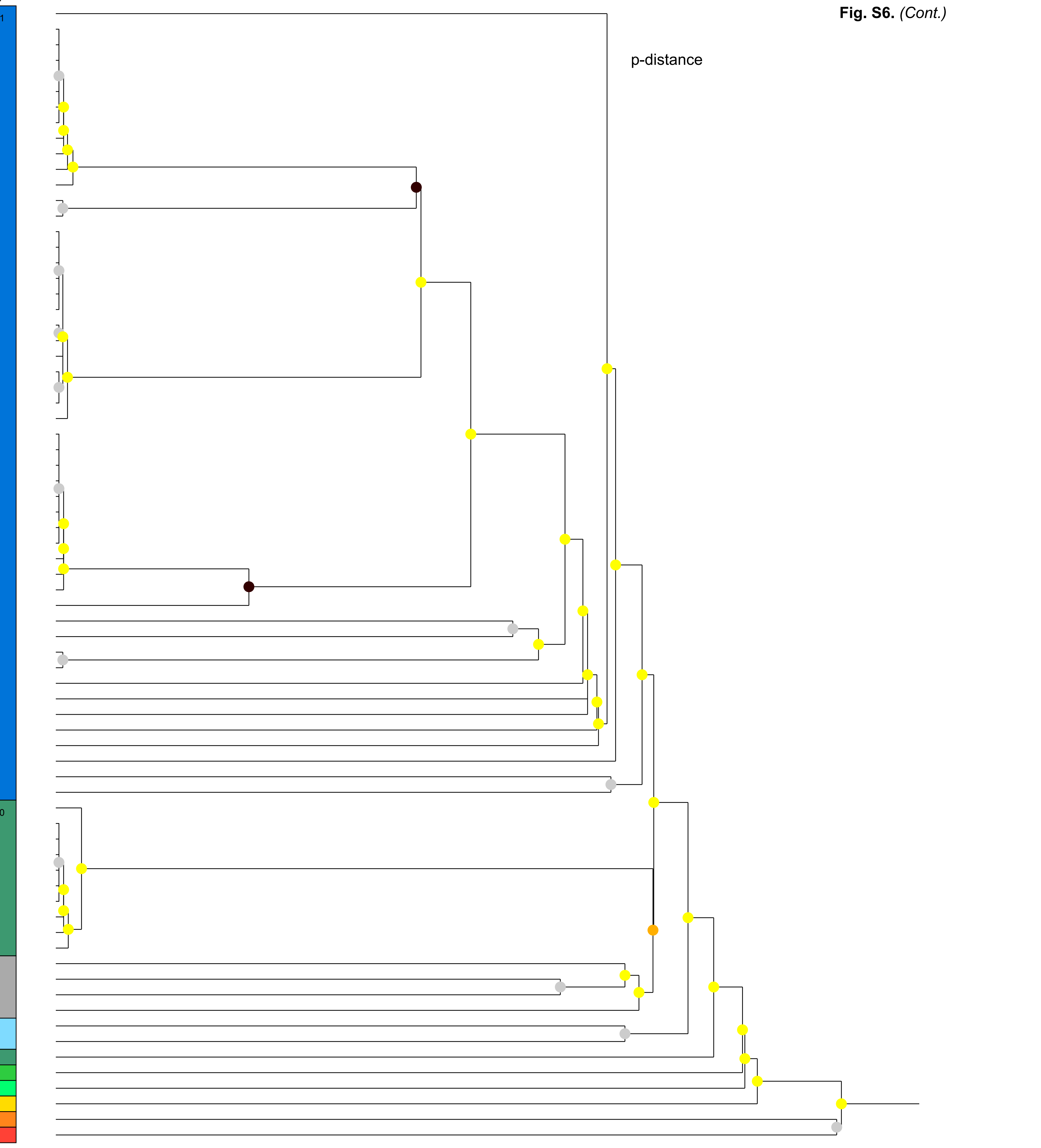
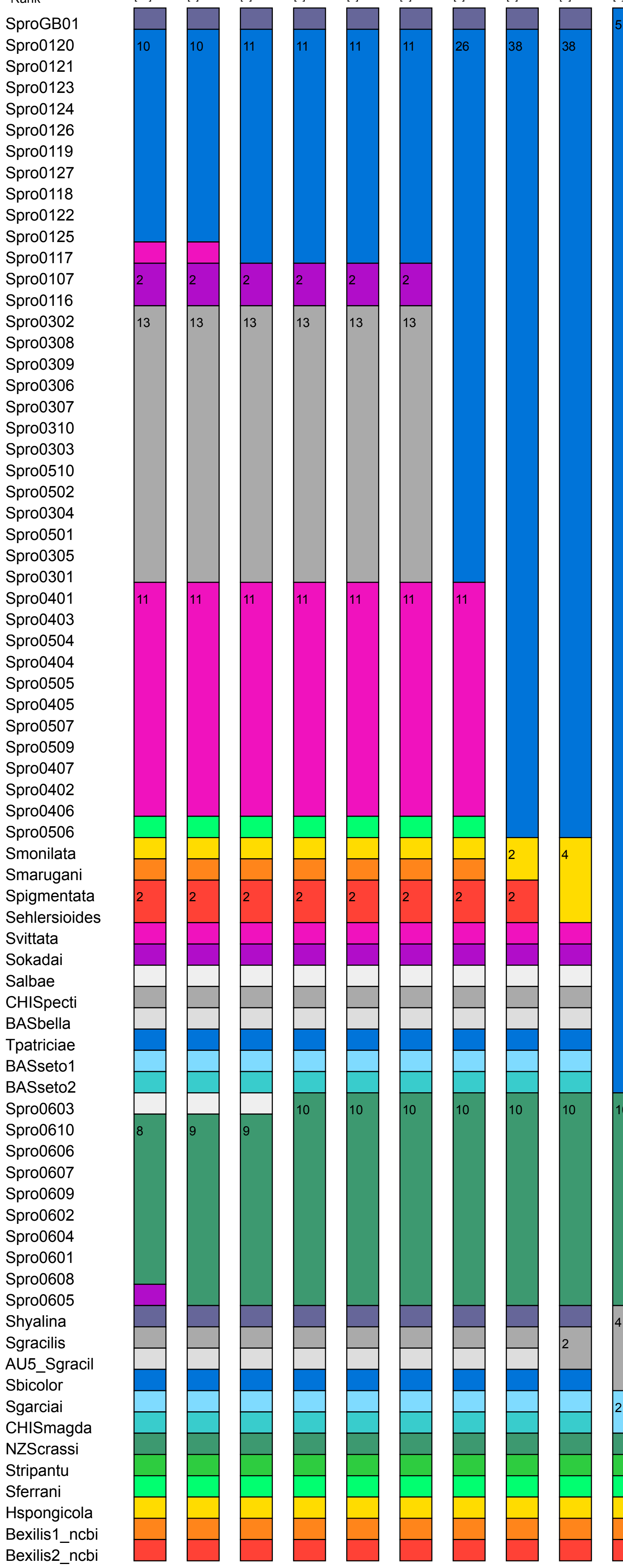


Fig. S6. (Cont.)

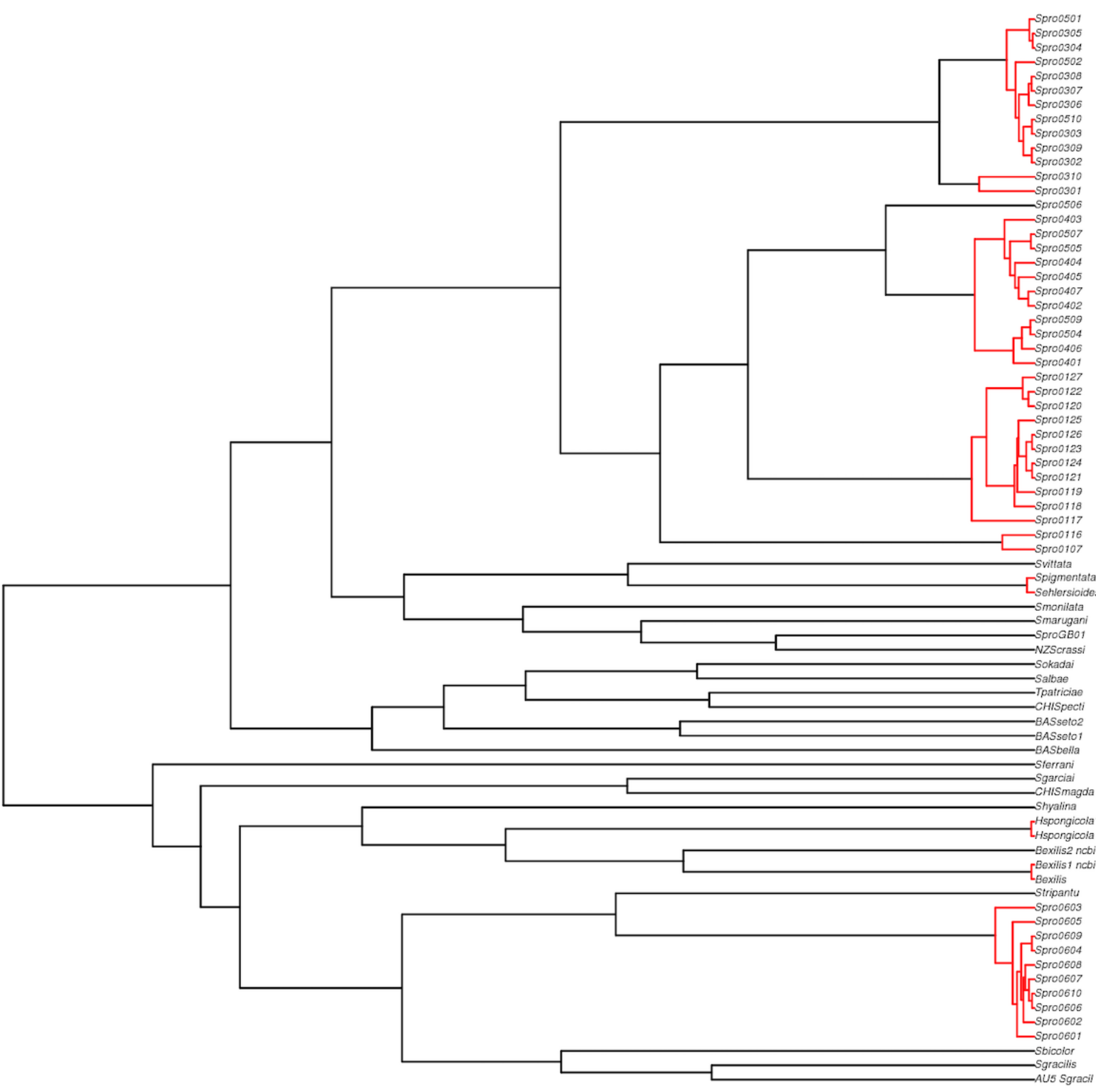
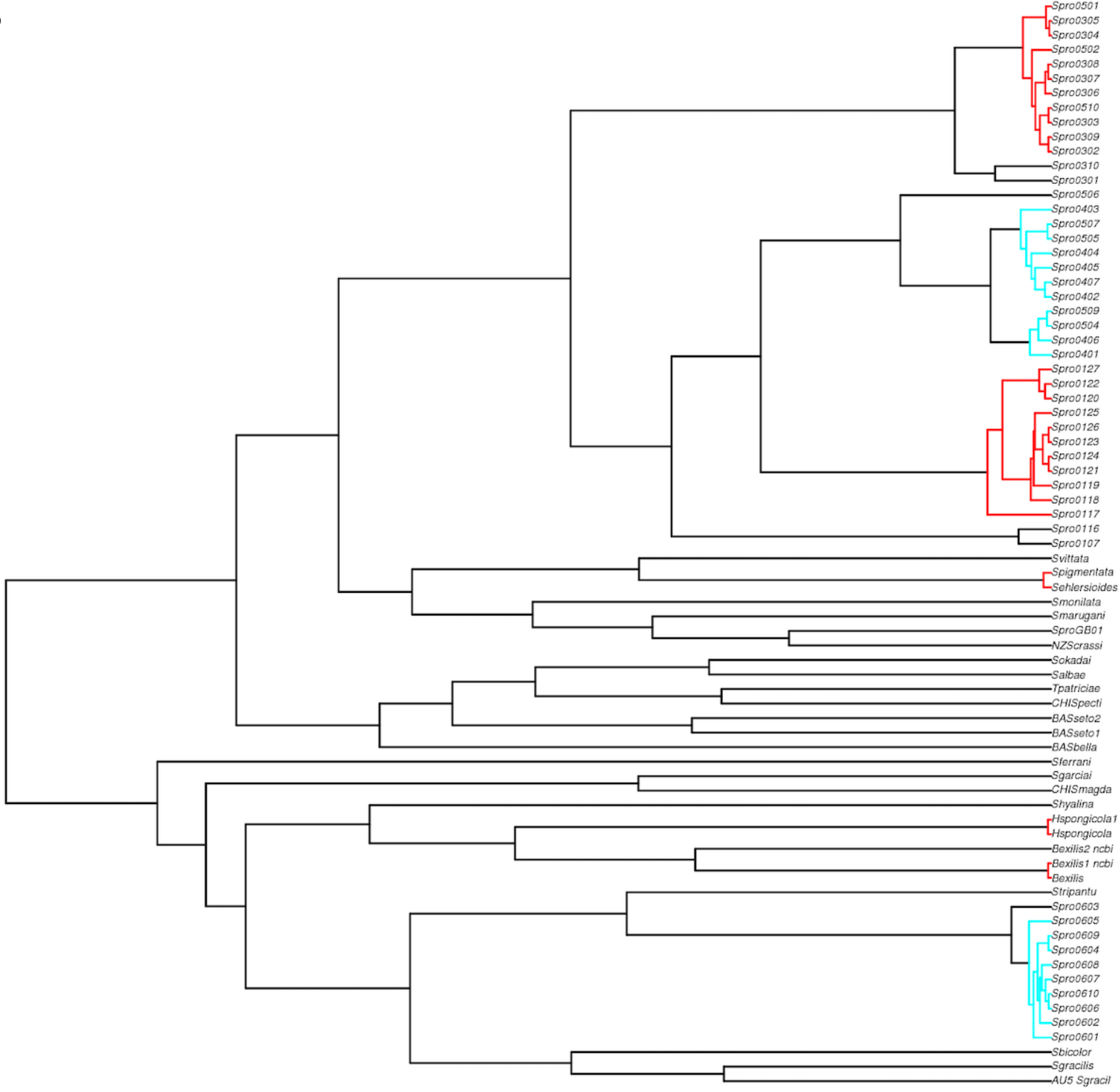
A**B**

Fig. S7. GMYC results for *Syllis prolifera* species complex using COI. A, single-threshold variant. B, multi-threshold variant.

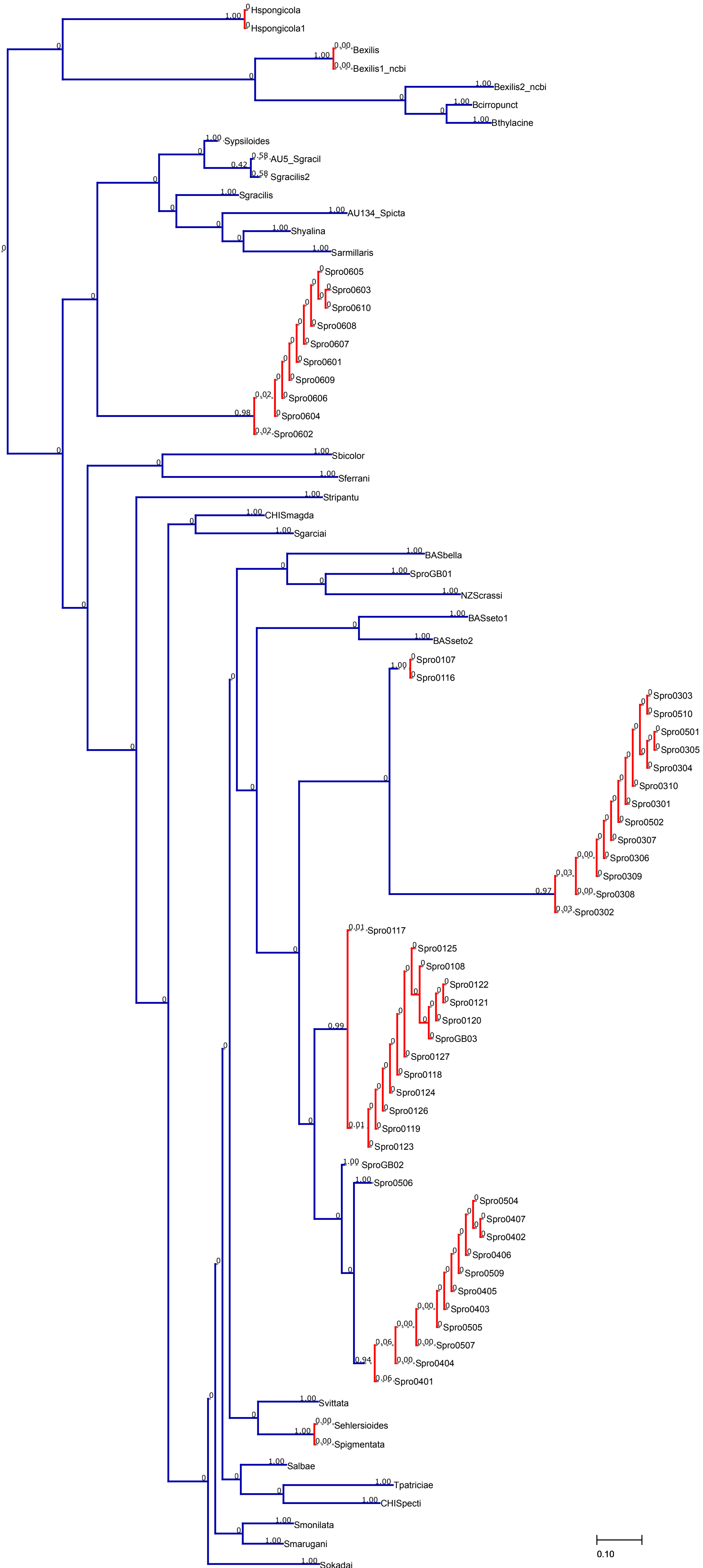


Fig. S8. bPTP results for *Syllis prolifera* using the concatenated mitochondrial dataset (16S rRNA and COI).