

THE LANCET Microbe

Supplementary appendix 2

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APPENDIX 2

Contents	Page
1. Supplementary methods	
1.1. Single-nucleotide polymorphism (SNP) and phylogenetic analysis	2
1.2. Neighbor-joining tree and genetic distance calculation	2
1.3. Multi-dimensional scaling analysis	2
1.4. References	4
2. Supplementary tables	
2.1. Supplementary Table 1. Characteristics of the 48 human sporotrichosis cases in Brazil	5
2.2. Supplementary Table 2. Characteristics of whole-genome sequences downloaded from NCBI	6
2.3. Supplementary Table 3. Quality control (QC) parameters for genomic analysis of <i>Sporothrix</i> sequences	7
2.4. Supplementary Table 4. Quality of <i>Sporothrix</i> sequences data generated by MycoSNP v1.4 pipeline	7
3. Supplementary figure	
3.1. Supplementary Figure 1. Mating types of <i>S. schenckii</i> and <i>S. brasiliensis</i> clades	10

Supplementary methods

Single-nucleotide polymorphism (SNP) and phylogenetic analysis. Single-nucleotide polymorphism (SNP) and phylogenetic analysis. SNPs were identified using MycoSNP v1.4 (<https://github.com/CDCgov/mycosnp-nf/>) as described by Bagal et. al. (1). This method prepares the reference, performs quality control, aligns reads to the reference genome and calls variants. *S. brasiliensis* strain 5110 was used as the reference genome (NCBI: txid1398154) (2). The pipeline runs in two steps post reference preparation. In the first step, raw WGS sequence files were processed for quality using FaQCs 2.10, aligned using the Burrows–Wheeler Aligner (BWA) v0.7.17 using the mem algorithm. Based on the quality control (QC) report and FaQCs output, samples were assessed for quality based on the metrics described in the supplementary table 3. For samples that passed the QC (supplementary table 4), GATK v 4.1.4.1 was used to call variants. Custom filters: 'QD < 2.0 || FS > 60.0 || MQ < 40.0 || DP < 10', were applied and a multi-FASTA file was generated and used for phylogenetic tree building. A maximum likelihood (ML) tree was built by the MycoSNP v 1.4 using FastTree v2.1.11, default parametrs (3). The consensus phylogenetic tree of 1,000 ultrafast bootstraps coupled with 1,000 Shimodaira–Hasegawa-like approximate likelihood ratio tests (SH-aLRT) was used to assess support for each clade (4, 5).

Neighbor-joining tree and genetic distance calculation. Neighbor-joining tree genetic distance calculations was built using MEGA 11 (6), using the multi-FASTA file from mycosnp v1.4. as input. The parameters used for the neighbor-joining tree were as follow: Test of Phylogeny 'none'; Substitution Type 'Nucleotide', Model/Method 'No. of differences'; Substitutions to Include 'd: Transitions + Transversions'; Rates among Sites 'Uniform Rates'; Gaps/Missing Data Treatment 'Complete deletion' by MEGA 11. The neighbor-joining tree was exported as 'Newick', 'Branch Lengths' option was selected, and maps were visualized together with metadata containing additional epidemiological data for each samples using Microreact (<http://microreact.org>).

Multi-dimensional scaling analysis. Multidimensional Scaling (MDS) plot was created using a patristic distance matrix, which is a measure of dissimilarity or difference between two taxa, taking account their shared evolutionary history (7). The resulting matrix is non-negative and symmetrical, conforming to the requirements of a dissimilarity matrix. The patristic distance matrix was constructed using the 'dendrophy' function in Python which utilizes a maximum likelihood tree, using the following script:

```
#!/usr/bin/Python

# usage: python CalculatePatristic_Distance.py ML_tree.tre >ML_PD.sqm

import dendropy
import sys,re
#from dendropy.calculate import treemeasure
from dendropy import treecalc

if len(sys.argv) !=2:
    print (" Usage: Calc_patristic_Dist.py tree(newick/nexus)")
    exit (1)

printC=sys.stdout.write
tree=dendropy.Tree.get_from_path(sys.argv[1],"newick")
pdm=treecalc.PatristicDistanceMatrix(tree)
```

```

for i in tree.taxon_set:
    printC (i.label+"\t"+i.label)
    for j in tree.taxon_set:
        printC ("\t"+str(pdm(i,j)))
    printC ("\n")
sys.stdout.flush()
'''

```

The values in the patristic distance matrix were used directly without scaling or normalization for performing multidimensional scaling using cmdscale R function. A plot was created to visualize the differences between samples using the qplot function in R, and the percentage of dissimilarity was done using the cmdscale R function using the following script:

```

library(ggplot2)
library(ggfortify)

# Reading the patristic distance matrix
D=read.table("ML_PD.sqm",header=FALSE, sep="\t")

# Calculate MDS of the data matrix D
mds.stuff<-cmdscale(data.matrix(D[3:ncol(D)]),eig=TRUE,x.ret=TRUE)
mds.var.per<-round(mds.stuff$eig/sum(mds.stuff$eig)*100,1)
mds.value<-mds.stuff$points

# Calculate the percentage of dissimilarity explained by the two dimensions

eigenvalues<-mds.stuff$eig
percentage_explained <- eigenvalues / sum(eigenvalues) * 100

# Print the results
cat("Percentage of Dissimilarity Explained by Dimension 1:", percentage_explained[1], "%\n")
cat("Percentage of Dissimilarity Explained by Dimension 2:", percentage_explained[2], "%\n")

# Adding the metadata info to the mds value table
D2<-D[,-1]
rownames(D2)<-D[,1]
mds.data<-data.frame(Sample=rownames(D2),X=mds.value[,1],Y=mds.value[,2])
head(mds.data)

#Sorted metadata info added to the mds values table
E=read.table("Corrects-list.txt",header=FALSE, sep="\t")
#head(E[1:2])
mds2<-cbind(E[1:2],mds.data)
#head(mds2)
colnames(mds2)<-c("id", "groups", "Sample", "X", "Y")
#head(mds2)

```

```
# plot the MDS table and show Metadata using different colors
qplot(X,Y,data=as.data.frame(mds2),color=groups)+
  theme_bw()+
  xlab("Coordinate 1")+
  ggtitle("MDS Plot:S. brasiliensis clades")+
  ylab("Coordinate 2")+
  geom_point( size=4)+
  #scale_shape_manual(values=c(15,2,3))+

scale_color_manual(values=c('#ffffff','#FF33F0','#35FF33','#FF3333','#33C9FF','#0000CD'))+
  guides(color = guide_legend(override.aes = list(size = 5)))
``
```

References

1. Ujwal R Bagal JP, Rory M Welsh, Elizabeth Misas, et al. MycoSNP: A Portable Workflow for Performing Whole-Genome Sequencing Analysis of *Candida auris*. *Methods Mol Biol*. 2022.
2. Teixeira MM, De Almeida LG, Kubitschek-Barreira P, Alves FL, Kioshima ÉS, Abadio AK, et al. Comparative genomics of the major fungal agents of human and animal Sporotrichosis: *Sporothrix schenckii* and *Sporothrix brasiliensis*. *BMC Genomics*. 2014;15(1):943.
3. Price MN, Dehal PS, Arkin AP. FastTree 2 – Approximately Maximum-Likelihood Trees for Large Alignments. *PLoS ONE*. 2010;5(3):e9490.
4. H Shimodaira MH. Multiple Comparisons of Log-Likelihoods with Applications to Phylogenetic Inference. *Mol Biol Evol*. 1999;16:1114-6.
5. Guindon S, Dufayard, J. F., Lefort, V., Anisimova, M., Hordijk, W., and Gascuel, O. New algorithms and methods to estimate maximum-likelihood phylogenies: assessing the performance of PhyML 3.0. *Syst Biol* 2010;59:07-321.
6. Koichiro Tamura GS, Sudhir Kumar. MEGA11: Molecular Evolutionary Genetics Analysis Version 11. *Mol Biol Evol*. 2021;38 (7):3022-7.
7. Gower JC. Some Distance Properties of Latent Root and Vector Methods Used in Multivariate Analysis. *Biometrika*. 1966;53:325-38.

Supplementary tables

Supplementary Table 1. Characteristics of the 48 human sporotrichosis cases in Brazil

Sample ID	Species	Brazil state	Year of specimen collection	Age (years) of associated case	Sex of associated case
B22092	<i>S. brasiliensis</i>	Bahia	2021	35	male
B22142	<i>S. brasiliensis</i>	Mato Grosso do Sul	2020		
B22061	<i>S. brasiliensis</i>	Mato Grosso do Sul	2019		
B22070	<i>S. brasiliensis</i>	Mato Grosso do Sul	2020	44	male
B22071	<i>S. brasiliensis</i>	Mato Grosso do Sul	2019	45	male
B22072	<i>S. brasiliensis</i>	Mato Grosso do Sul	2020	73	female
B22077	<i>S. brasiliensis</i>	Mato Grosso do Sul	2020	41	male
B22079	<i>S. brasiliensis</i>	Mato Grosso do Sul			
B22080	<i>S. brasiliensis</i>	Mato Grosso do Sul			
B22081	<i>S. brasiliensis</i>	Mato Grosso do Sul	2021	53	female
B22087	<i>S. brasiliensis</i>	Pernambuco	2021	18	female
B22089	<i>S. brasiliensis</i>	Rio de Janeiro	2021	24	female
B22099	<i>S. brasiliensis</i>	Rio de Janeiro	2019	57	female
B22086	<i>S. brasiliensis</i>	Rio Grande do Norte	2021	41	female
B22088	<i>S. brasiliensis</i>	Rio Grande do Norte	2021	59	male
B22091	<i>S. brasiliensis</i>	Rio Grande do Norte	2021	58	male
B22062	<i>S. brasiliensis</i>	São Paulo	2017	42	female
B22063	<i>S. brasiliensis</i>	São Paulo	2021		male
B22064	<i>S. schenckii</i>	São Paulo	2021		male
B22073	<i>S. brasiliensis</i>	São Paulo	2020	47	female
B22083	<i>S. brasiliensis</i>	São Paulo	2021	44	female
B22084	<i>S. brasiliensis</i>	São Paulo	2021		male
B22085	<i>S. brasiliensis</i>	São Paulo	2021	36	male
B22090	<i>S. brasiliensis</i>	São Paulo	2021	42	female
B22093	<i>S. brasiliensis</i>	São Paulo	2019	43	male
B22094	<i>S. brasiliensis</i>	São Paulo	2015	64	female
B22095	<i>S. brasiliensis</i>	São Paulo	2017	49	female
B22096	<i>S. brasiliensis</i>	São Paulo	2015	20	male
B22097	<i>S. brasiliensis</i>	São Paulo	2019	11	male
B22098	<i>S. brasiliensis</i>	São Paulo	2019	48	female
B22065	<i>S. schenckii</i>				
B22066	<i>S. brasiliensis</i>				
B22067	<i>S. brasiliensis</i>		2017		
B22068	<i>S. brasiliensis</i>		2013		
B22069	<i>S. brasiliensis</i>				female
B22074	<i>S. brasiliensis</i>		2020		male
B22137	<i>S. brasiliensis</i>		2016		female
B22138	<i>S. brasiliensis</i>				
B22139	<i>S. brasiliensis</i>		2020		
B22140	<i>S. brasiliensis</i>		2020		male
B22141	<i>S. brasiliensis</i>		2014		female
B22143	<i>S. globosa</i>		2013		
B22144	<i>S. brasiliensis</i>		2017		
B22145	<i>S. brasiliensis</i>		2016		
B22146	<i>S. brasiliensis</i>		2021		
B22147	<i>S. schenckii</i>		2022		
B22148	<i>S. brasiliensis</i>		2021		
B22149	<i>S. brasiliensis</i>		2021	23	female

Supplementary Table 2. Characteristics of whole-genome sequences downloaded from NCBI
<https://www.ncbi.nlm.nih.gov/sra/?term=Sporothrix>

Sample accession	strain	Year of isolation	Geographic origin	Host
SRR12483721	<i>Sporothrix schenckii</i> SsEM7	2012	Colombia, Medellin	<i>Homo sapiens</i>
SRR12483722	<i>Sporothrix schenckii</i> SsMS1	2012	Colombia, Medellin	<i>Homo sapiens</i>
SRR12483723	<i>Sporothrix brasiliensis</i> A5	2018	Brazil, Distrito Eederal	<i>Canis lupus familiaris</i>
SRR12483724	<i>Sporothrix schenckii</i> A0003	2015	Brazil, Distrito Distrito Federal	<i>Felis catus</i>
SRR12483725	<i>Sporothrix brasiliensis</i> A0001	2016	Brazil, Distrito Federal	<i>Felis catus</i>
SRR12483726	<i>Sporothrix brasiliensis</i> s48605	2016	Brazil, Rio de Janeiro	<i>Homo sapiens</i>
SRR12483728	<i>Sporothrix brasiliensis</i> s28606	2005	Brazil, Rio de Janeiro	<i>Homo sapiens</i>
SRR12483729	<i>Sporothrix brasiliensis</i> s15677	1999	Brazil, Rio de Janeiro	<i>Homo sapiens</i>

Supplementary Table 3. Quality control (QC) parameters for genomic analysis of *Sporothrix* sequences

QC Metric	Thresholds
Average Q Score (Phred Score) After Trimming	≥ 28
GC After Trimming	51% - 59%
Reference Length Coverage After Trimming	≥ 20X
Mean Coverage Depth	≥ 20X

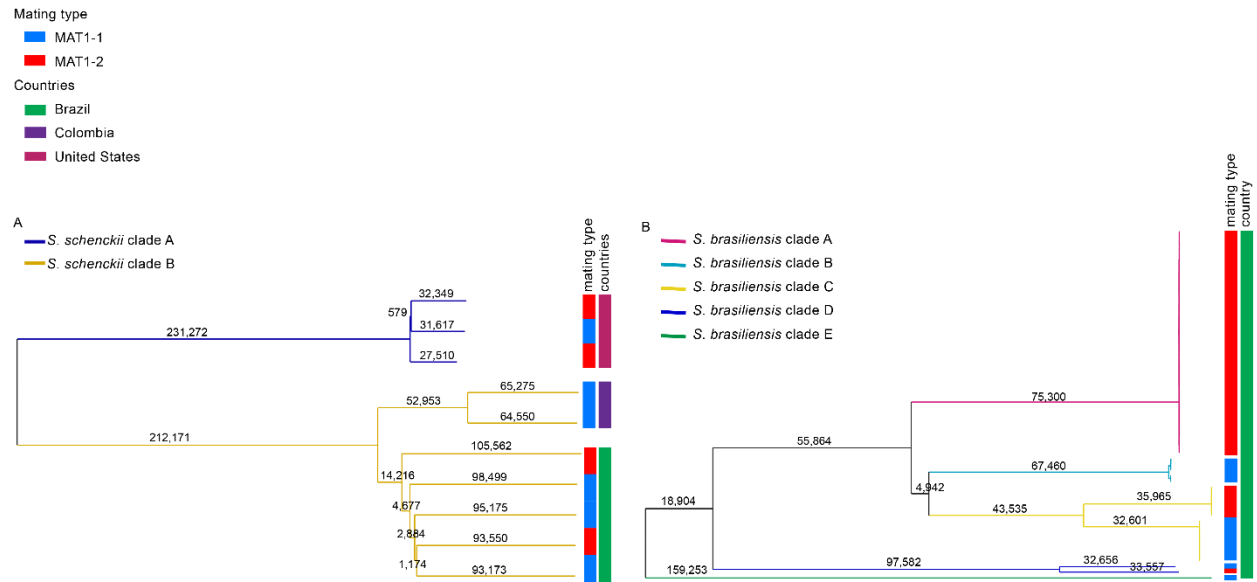
Supplementary Table 4. Quality of *Sporothrix* sequences data generated by MycoSNP v1.4 pipeline
(<https://github.com/CDCgov/mycosnp-nf/>)

Sample Name	Reads After Trimming	Paired Reads After Trimming	Unpaired Reads After Trimming	GC After Trimming	Average Q Score After Trimming	Reference Length Coverage After Trimming	Mean Coverage Depth	Reads Mapped	status
B10282	9256752 (100.00 %)	9256752 (100.00 %)	0 (0.00 %)	54.14%	34	68.21	61.78	8891995 (96.83%)	Passed
B11252	9311272 (100.00 %)	9311272 (100.00 %)	0 (0.00 %)	53.96%	34.03	68.2	61.41	8910037 (96.59%)	Passed
B24667	2737178 (99.99 %)	2736984 (99.99 %)	194 (0.01 %)	54.74%	32.38	20.69	18.07	2611135 (96.08%)	Passed
B22061	35560897 (99.15 %)	35551108 (99.97 %)	9789 (0.03 %)	55.44%	34.52	219.54	202.39	33137191 (98.94%)	Passed
B22062	25069745 (99.30 %)	25063114 (99.97 %)	6631 (0.03 %)	55.08%	34.55	161.84	149.45	23375138 (98.94%)	Passed
B22063	22423393 (99.73 %)	22417750 (99.97 %)	5643 (0.03 %)	55.04%	34.5	150.23	138.16	20843321 (98.93%)	Passed
B22064	20111519 (99.71 %)	20105168 (99.97 %)	6351 (0.03 %)	55.16%	34.33	139.94	122.17	18377791 (96.96%)	Passed
B22065	20597423 (99.80 %)	20593688 (99.98 %)	3735 (0.02 %)	55.24%	34.42	140.56	122.76	18789473 (96.73%)	Passed
B22066	25208943 (99.77 %)	25202354 (99.97 %)	6589 (0.03 %)	55.95%	34.45	173.42	159.3	23492306 (98.36%)	Passed
B22067	26443265 (99.68 %)	26435870 (99.97 %)	7395 (0.03 %)	55.25%	34.49	179.42	163.15	24417911 (98.23%)	Passed
B22068	15212618 (99.77 %)	15207872 (99.97 %)	4746 (0.03 %)	53.56%	34.34	103.72	91.44	13690980 (97.05%)	Passed
B22069	32242501 (99.68 %)	32235342 (99.98 %)	7159 (0.02 %)	54.96%	34.5	210.54	193.55	29957404 (98.94%)	Passed
B22070	29737187 (99.75 %)	29730940 (99.98 %)	6247 (0.02 %)	55.67%	34.52	197.58	182.58	27760670 (99%)	Passed
B22071	25956134 (99.82 %)	25949736 (99.98 %)	6398 (0.02 %)	55.49%	34.56	174.14	161.75	24359364 (99.05%)	Passed
B22072	26560235 (99.68 %)	26554026 (99.98 %)	6209 (0.02 %)	56.09%	34.46	176.88	164.96	25038831 (99.11%)	Passed
B22073	23077128 (98.91 %)	23069640 (99.97 %)	7488 (0.03 %)	54.96%	34.54	151.01	138.61	21399665 (98.87%)	Passed
B22074	24054055 (99.68 %)	24048628 (99.98 %)	5427 (0.02 %)	55.65%	34.48	155.53	142.52	22396447 (98.29%)	Passed
B22077	21156227 (98.60 %)	21149442 (99.97 %)	6785 (0.03 %)	55.47%	34.5	139.41	129	19785204 (98.95%)	Passed
B22079	19402395 (99.32 %)	19396922 (99.97 %)	5473 (0.03 %)	55.41%	34.56	129.06	119.37	18106135 (99.01%)	Passed
B22080	29604425 (98.68 %)	29596862 (99.97 %)	7563 (0.03 %)	55.10%	34.48	198.97	181.95	27367181 (98.87%)	Passed
B22081	18105340 (99.59 %)	18100944 (99.98 %)	4396 (0.02 %)	54.71%	34.45	123.32	114.09	16936863 (98.84%)	Passed
B22083	18326089 (99.62 %)	18320650 (99.97 %)	5439 (0.03 %)	55.08%	34.43	126.37	117.49	17229296 (98.92%)	Passed
B22084	20026673 (99.43 %)	20021078 (99.97 %)	5595 (0.03 %)	55.56%	34.44	136.46	126.45	18753553 (98.97%)	Passed
B22085	21457659 (99.34 %)	21452156 (99.97 %)	5503 (0.03 %)	55.41%	34.41	145.71	132.43	19808726 (98.24%)	Passed

B22086	19416209 (99.74 %)	19410236 (99.97 %)	5973 (0.03 %)	56.30%	34.43	133.87	122.72	18034663 (98.41%)	Passed
B22087	23927292 (99.34 %)	23921008 (99.97 %)	6284 (0.03 %)	55.28%	34.52	162.03	148.59	22151615 (98.93%)	Passed
B22088	27165987 (99.81 %)	27159610 (99.98 %)	6377 (0.02 %)	58.77%	34.62	187.7	88.07	14186897 (54.05%)	Passed
B22089	22074248 (99.21 %)	22068092 (99.97 %)	6156 (0.03 %)	55.14%	34.44	147.65	135.79	20528632 (98.81%)	Passed
B22090	26424353 (99.67 %)	26419268 (99.98 %)	5085 (0.02 %)	55.51%	34.55	180.23	164.44	24454289 (98.28%)	Passed
B22091	23373938 (99.81 %)	23367756 (99.97 %)	6182 (0.03 %)	55.36%	34.48	162.15	148.71	21747559 (98.12%)	Passed
B22092	19172421 (99.52 %)	19161648 (99.94 %)	10773 (0.06 %)	51.05%	34.34	126.82	100.94	15552030 (97%)	Passed
B22093	25255978 (99.29 %)	25248474 (99.97 %)	7504 (0.03 %)	55.92%	34.47	170.9	155.65	23342082 (98.28%)	Passed
B22094	21438086 (99.24 %)	21431240 (99.97 %)	6846 (0.03 %)	54.66%	34.47	143.78	132.28	19938304 (98.76%)	Passed
B22095	32516974 (98.16 %)	32506970 (99.97 %)	10004 (0.03 %)	55.25%	34.49	209.07	192.03	30195916 (98.84%)	Passed
B22096	28217662 (99.70 %)	28211416 (99.98 %)	6246 (0.02 %)	55.75%	34.48	194.17	177.73	26200699 (98.35%)	Passed
B22097	19243656 (99.20 %)	19238490 (99.97 %)	5166 (0.03 %)	53.44%	34.43	126.33	112.96	17547195 (97.5%)	Passed
B22098	32579576 (98.36 %)	32568120 (99.96 %)	11456 (0.04 %)	55.32%	34.52	210.15	189.43	29824650 (98.06%)	Passed
B22099	27082265 (99.04 %)	27075510 (99.98 %)	6755 (0.02 %)	54.52%	34.53	179.24	164.69	25157052 (98.83%)	Passed
B22101	27957762 (98.69 %)	27948784 (99.97 %)	8978 (0.03 %)	54.91%	34.54	181.95	166.37	25856537 (98.79%)	Passed
B22102	22694855 (99.69 %)	22688716 (99.97 %)	6139 (0.03 %)	55.83%	34.44	156.45	145.13	21281287 (99%)	Passed
B22103	26392988 (99.75 %)	26387042 (99.98 %)	5946 (0.02 %)	56.04%	34.56	178.36	157.21	24222123 (97%)	Passed
B22104	25197390 (98.06 %)	25187808 (99.96 %)	9582 (0.04 %)	55.17%	34.5	162.02	149.06	23430578 (98.9%)	Passed
B22137	79578755 (99.35 %)	79546156 (99.96 %)	32599 (0.04 %)	52.19%	34.46	513.14	369.06	58400552 (97.27%)	Passed
B22138	23489444 (99.20 %)	23483162 (99.97 %)	6282 (0.03 %)	52.55%	34.54	151.42	129	20402124 (97.24%)	Passed
B22139	19166564 (99.39 %)	19157088 (99.95 %)	9476 (0.05 %)	53.63%	34.44	123.29	109.47	17270127 (98.45%)	Passed
B22140	18755163 (99.54 %)	18747550 (99.96 %)	7613 (0.04 %)	53.14%	34.49	121.94	106.66	16715481 (97.32%)	Passed
B22141	27603790 (99.37 %)	27596050 (99.97 %)	7740 (0.03 %)	55.91%	34.61	177.93	162.18	25501450 (98.35%)	Passed
B22142	23601514 (99.56 %)	23595802 (99.98 %)	5712 (0.02 %)	55.55%	34.61	154.25	141.56	21865875 (99.1%)	Passed
B22143	20820536 (99.02 %)	20816032 (99.98 %)	4504 (0.02 %)	56.34%	34.99	112.17	81	16825138 (88.09%)	Passed
B22144	28548338 (98.87 %)	28540722 (99.97 %)	7616 (0.03 %)	54.84%	34.6	177.83	157.74	25567515 (98.9%)	Passed
B22145	27771980 (98.12 %)	27763832 (99.97 %)	8148 (0.03 %)	56.22%	34.86	158.8	144.41	25433586 (99.22%)	Passed
B22146	24340053 (99.31 %)	24334726 (99.98 %)	5327 (0.02 %)	57.00%	34.66	159.18	146.18	22618219 (98.67%)	Passed
B22147	23697517 (99.04 %)	23692512 (99.98 %)	5005 (0.02 %)	56.48%	34.75	146.04	127.9	21539859 (96.95%)	Passed
B22148	20612654 (99.08 %)	20612654 (100.00 %)	0 (0.00 %)	56.52%	34.72	121.8	100.3	16835446 (97.97%)	Passed
B22149	23433543 (98.92 %)	23427566 (99.97 %)	5977 (0.03 %)	56.52%	34.64	147.26	134.6	21676227 (98.69%)	Passed
B22150	21227338 (98.80 %)	21222504 (99.98 %)	4834 (0.02 %)	55.95%	34.7	134.94	124.18	19689869 (99.21%)	Passed
B22151	25947045 (98.31 %)	25938246 (99.97 %)	8799 (0.03 %)	55.26%	34.79	147.59	132.18	23413551 (99.07%)	Passed
B22152	48643175 (99.38 %)	48616922 (99.95 %)	26253 (0.05 %)	52.48%	34.65	295.41	200.41	33781095 (96.81%)	Passed

B22153	27025280 (99.52 %)	27018968 (99.98 %)	6312 (0.02 %)	56.07%	34.71	163.03	150.49	25138041 (99.24%)	Passed
B22154	23762922 (99.43 %)	23756534 (99.97 %)	6388 (0.03 %)	56.38%	34.62	150.58	138.43	22029841 (99.29%)	Passed
B22155	25009945 (99.15 %)	25003448 (99.97 %)	6497 (0.03 %)	56.34%	34.76	155.97	143.69	23220679 (99.26%)	Passed
B22156	23602703 (99.79 %)	23597318 (99.98 %)	5385 (0.02 %)	56.20%	34.72	152.57	140.58	21912932 (99.33%)	Passed
B22157	26740953 (99.06 %)	26735126 (99.98 %)	5827 (0.02 %)	55.84%	34.7	165.78	151.27	24585715 (99.23%)	Passed
B22159	29562570 (98.81 %)	29554686 (99.97 %)	7884 (0.03 %)	55.08%	34.68	180.99	166.09	27384154 (98.95%)	Passed
SRR12483721	26595558 (95.58 %)	25442664 (95.67 %)	1152894 (4.33 %)	53.35%	33.53	115.71	99.22	23803317 (94.17%)	Passed
SRR12483722	27899291 (94.97 %)	26502652 (94.99 %)	1396639 (5.01 %)	52.56%	33.5	121.22	103.21	24833213 (94.5%)	Passed
SRR12483723	18062910 (99.97 %)	18057970 (99.97 %)	4940 (0.03 %)	53.60%	35.37	81.6	64.62	14497522 (93.41%)	Passed
SRR12483724	13980446 (99.97 %)	13975668 (99.97 %)	4778 (0.03 %)	54.70%	35.29	63.15	49.26	11400561 (93.64%)	Passed
SRR12483725	22130161 (99.93 %)	22115384 (99.93 %)	14777 (0.07 %)	52.97%	35.45	99.97	69.32	15537852 (91.84%)	Passed
SRR12483726	19419587 (99.94 %)	19407498 (99.94 %)	12089 (0.06 %)	52.66%	35.46	87.72	59.87	13327377 (95.93%)	Passed
SRR12483728	16945194 (99.93 %)	16933226 (99.93 %)	11968 (0.07 %)	53.26%	35.45	76.55	52.75	11758937 (96.27%)	Passed
SRR12483729	21315936 (99.93 %)	21300952 (99.93 %)	14984 (0.07 %)	53.34%	35.43	96.29	65.74	14630182 (96.29%)	Passed

Supplementary figures



Supplementary Figure 1. Mating types of *S. schenckii* and *S. brasiliensis* clades. A) Neighbor-Joining phylogenetic tree of *S. schenckii* isolates from Brazil, Colombia, and USA by clade. **B)** Neighbor-Joining phylogenetic tree of *S. brasiliensis* isolates from Brazil by clade. Branch lengths represent SNPs.