Supplemental tables

Supplementary Table 1: Genome assembly statistics

step	# scaffolds	N50, Kb	% gap	BUSCO
10xGenomics, Supernova2	174165	33.877	3.073	C:62.9%[S:56.0%,D:6.9%],F:21.0%,M:16.1%,n:3950
Agouti with RNAseq	171284	35.23	8.804	C:64.7%[S:58.2%,D:6.5%],F:19.2%,M:16.1%,n:3950
Jelly on Nanopore	163793	40.024	7.147	C:67.6%[S:61.4%,D:6.2%],F:17.9%,M:14.5%,n:3950
Dovetail	39223	227.932		C:78.6%[S:73.7%,D:4.9%],F:6.4%,M:15.0%,n:5310

Supplementary Table 2: Transcriptome assembly statistics

sample	organ	# raw reads	# transcripts	# transcripts_1tpm_cutoff	after cdhit
AK393	stomach	101,419,756	211105	61363	53092
AK4334	lung	99,048,832	246972	86823	73408
AK2340	skin	155,144,638	290666	84145	71437
AK3438	eye	151,860,400	364970	88779	76814
AK626	brain	142,942,536	621519	169862	152663
merged	-	-	-	490972	228525

Supplementary Table 3: Gene family expansion/contraction
https://figshare.com/articles/dataset/Changes_in_cell_cycle_machinery_as_genetic_basis_for_polyploidy_stabilization_in_Australian_burrowing_frogs_Neobatrachus_/30576644?file=59724149

Supplementary Table 4: Sample overview
https://figshare.com/articles/dataset/Changes_in_cell_cycle_machinery_as_genetic_basis_for_polyploidy_stabilization_in_Australian_burrowing_frogs_Neobatrachus_/30576644?file=59724158

Supplementary Table 5: ABBA-BABA statistics

D	JK.D	V.JK.D.	Z	pvalue	nABBA	nBABA	nBlocks	H1	H2	H3	H4
-0.226253	-0.226253	8.00E-06	-78.910464	0	46711.21526	74029.13753	386	Nfulvus	Npictus	Nalbipes	Ldumerilli
0.109512	0.109512	1.00E-05	34.945628	0	69308.81975	55626.91236	386	Nsutor	Nwilsmorei	Nalbipes	Ldumerilli
0.085891	0.085891	1.90E-05	19.680214	0	74707.59391	62889.27748	386	Nalbipes	Nsutor	Nfulvus	Ldumerilli
0.034171	0.034171	1.20E-05	9.873014	0	63918.33891	59694.3127	386	Nalbipes	Nwilsmorei	Nfulvus	Ldumerilli
-0.065735	-0.065735	1.30E-05	-18.474778	0	50271.91652	57346.16222	386	Nsutor	Nwilsmorei	Nfulvus	Ldumerilli
-0.258839	-0.258839	8.00E-06	-89.660025	0	51184.83882	86935.81157	386	Nalbipes	Nfulvus	Npelobatoides	Ldumerilli

-0.393355	-0.393355	6.00E-06	-161.085107	0	46429.20843	106639.465	386	Nalbipes	Npictus	Npelobatoides	Ldumerilli
-0.112212	-0.112212	9.00E-06	-36.556505	0	58208.06246	72922.46474	386	Nalbipes	Nsutor	Npelobatoides	Ldumerilli
-0.068212	-0.068212	1.00E-05	-21.302818	0	56950.80332	65289.03872	386	Nalbipes	Nwilsmorei	Npelobatoides	Ldumerilli
-0.210298	-0.210298	8.00E-06	-75.783093	0	43114.9258	66077.92321	386	Nfulvus	Npictus	Npelobatoides	Ldumerilli
0.155947	0.155947	1.30E-05	43.539373	0	77534.6372	56614.53315	386	Nfulvus	Nsutor	Npelobatoides	Ldumerilli
0.195389	0.195389	1.20E-05	55.585044	0	80246.52704	54013.6124	386	Nfulvus	Nwilsmorei	Npelobatoides	Ldumerilli
0.295672	0.295672	8.00E-06	102.027653	0	97668.57858	53092.66628	386	Npictus	Nsutor	Npelobatoides	Ldumerilli
0.325074	0.325074	8.00E-06	113.067445	0	98769.93356	50308.44571	386	Npictus	Nwilsmorei	Npelobatoides	Ldumerilli
0.056898	0.056898	8.00E-06	20.766556	0	54382.79289	48527.41263	386	Nsutor	Nwilsmorei	Npelobatoides	Ldumerilli
0.023321	0.023321	1.70E-05	5.697195	0	60889.18677	58113.97111	386	Nalbipes	Nsutor	Npictus	Ldumerilli
4.00E-04	4.00E-04	1.20E-05	0.116023	0.907	54703.23753	54659.51581	386	Nalbipes	Nwilsmorei	Npictus	Ldumerilli
-0.030258	-0.030258	1.20E-05	-8.561145	0	44947.39298	47752.28171	386	Nsutor	Nwilsmorei	Npictus	Ldumerilli
-0.281701	-0.281701	8.00E-06	-101.132443	0	46440.85217	82866.92962	386	Nfulvus	Npictus	Nsutor	Ldumerilli
-0.246299	-0.246299	8.00E-06	-89.452657	0	45067.07887	74521.65282	386	Nfulvus	Npictus	Nwilsmorei	Ldumerilli

Supplementary Table 6: Pseudohaplotype assignments of tetraploids to the diploid species of origin

Nkunapalari

pseudohaplotype 1	pseudohaplotype 2	occurence	occurrence / total	occurrence / known
Npictus	Npictus	14	0,00475	0,010550113
Nalbipes	Nfulvus	721	0,24457	0,543330821
Nalbipes	Nalbipes	211	0,07157	0,159005275
Nalbipes	Nwilsmorei	26	0,00882	0,019593067
Nalbipes	Nsutor	122	0,04138	0,091936699
Nfulvus	Npictus	6	0,00204	0,004521477
Nalbipes	Npictus	136	0,04613	0,102486812
Nfulvus	Nfulvus	13	0,00441	0,009796534
Npelobatoides	Npictus	2	0,00068	0,001507159
Npelobatoides	Npelobatoides	7	0,00237	0,005275057
Nsutor	Nsutor	21	0,00712	0,01582517
Npictus	Nsutor	11	0,00373	0,008289375
Nsutor	Nwilsmorei	13	0,00441	0,009796534
Npictus	Nwilsmorei	3	0,00102	0,002260739
Nalbipes	Npelobatoides	2	0,00068	0,001507159
Nwilsmorei	Nwilsmorei	1	0,00034	0,00075358
Nfulvus	Nsutor	14	0,00475	0,010550113

Nfulvus	Nwilsmorei	4	0,00136	0,003014318
unknown		1621	0,54986	
known		1327	0,45014	
total		2948	1,00000	

Nsudellae

pseudohaplotype 1	pseudohaplotype 2	occurence	occurrence / total	occurrence / known
Nfulvus	Nsutor	21	0,00560	0,009446694
Nfulvus	Nfulvus	1691	0,45069	0,760683761
Nfulvus	Npictus	76	0,02026	0,034188034
Npictus	Npictus	331	0,08822	0,148897886
Nsutor	Nsutor	65	0,01732	0,029239766
Npictus	Nsutor	8	0,00213	0,00359874
Nalbipes	Nalbipes	17	0,00453	0,007647323
Npelobatoides	Npictus	5	0,00133	0,002249213
Npelobatoides	Npelobatoides	3	0,00080	0,001349528
Nalbipes	Npictus	2	0,00053	0,000899685
Nwilsmorei	Nwilsmorei	2	0,00053	0,000899685
Nalbipes	Nfulvus	1	0,00027	0,000449843
Nfulvus	Npelobatoides	1	0,00027	0,000449843
	unknown	1529	0,40752	
	known	2223	0,59248	
	total	3752	1,00000	

Naqulonius

pseudohaplotype 1	pseudohaplotype 2	occurence	occurrence / total	occurrence / known
Nfulvus	Npictus	42	0,02238806	0,032735776

Nfulvus	Nfulvus	1069	0,569829424	0,833203429
Nalbipes	Nfulvus	1	0,000533049	0,000779423
Npictus	Nsutor	10	0,00533049	0,007794232
Nfulvus	Nsutor	113	0,060234542	0,088074825
Nfulvus	Nwilsmorei	4	0,002132196	0,003117693
Npictus	Npictus	19	0,010127932	0,014809041
Nsutor	Nsutor	17	0,009061834	0,013250195
Nalbipes	Nwilsmorei	1	0,000533049	0,000779423
Nalbipes	Nsutor	1	0,000533049	0,000779423
Nsutor	Nwilsmorei	4	0,002132196	0,003117693
Nalbipes	Nalbipes	2	0,001066098	0,001558846
	unknown	593	0,316098081	
	known	1283	0,683901919	
	total	1876	1	

Supplementary Table 7:
https://figshare.com/articles/dataset/Changes_in_cell_cycle_machinery_as_genetic_basis_for_polyploidy_stabilization_in_Australian_burrowing_frogs_Neobatrachus_/30576644?file=60781930

Supplementary Table 8: Annotated genes that were under selection in all three tetraploid species.

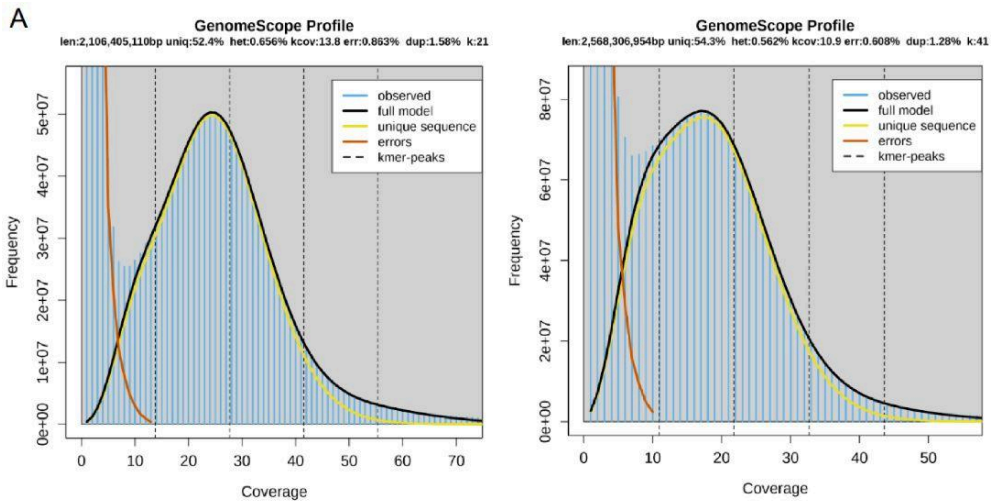
Functional group	Gene	Annotation	GO category
Crossover interference	g5917	SYCE2	synaptonemal complex assembly, structural unit of the synaptonemal complex
	g5068	Proline-rich protein 19	–
Chromosome packaging	g23417	Condensin-2 complex subunit D3	nuclear condensin complex, mitotic chromosome condensation
	g16961	Histone H2A	nucleosome, protein heterodimerization activity, DNA binding
	g5964	Histone H2B	nucleosome, protein heterodimerization activity, DNA binding
	g5970	Histone H3/CENP-A	nucleosome, protein

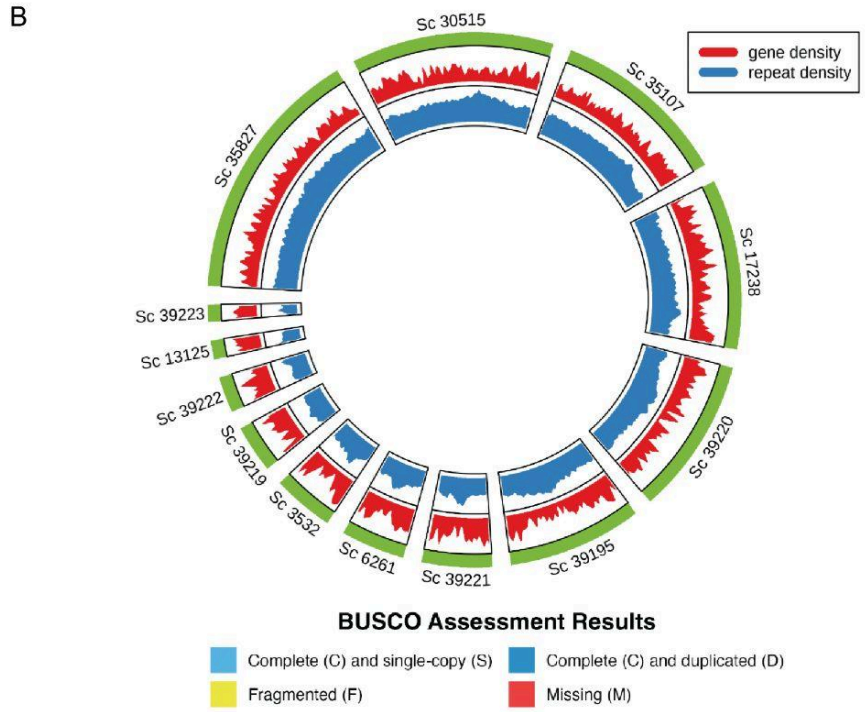
			heterodimerization activity, DNA binding
Spindle assembly	g2400	Kinesin-14 motor protein KifC1	DNA binding
	g11479	Tetratricopeptide repeat protein 34	
Other	g1430	Pancreatic trypsin inhibitor Kunitz domain	serine-type endopeptidase inhibitor activity
	g1431	Lipocalin/cytosolic fatty-acid binding domain	small molecule binding
	g1506	Allograft inflammatory factor 1	
	g22785	TAF1D	RNA polymerase transcription factor SL1 complex
	g23303	EMI domain profile	
	g23322	BolA protein	
	g23338	Iodothyronine deiodinase	thyroxine 5'-deiodinase activity
	g23627	1-phosphatidylinositol 4,5-bisphosphate phosphodiesterase eta-1	phosphatidylinositol-mediated signaling
	g9063	Glycosyl transferase, family 3	galactosyltransferase activity

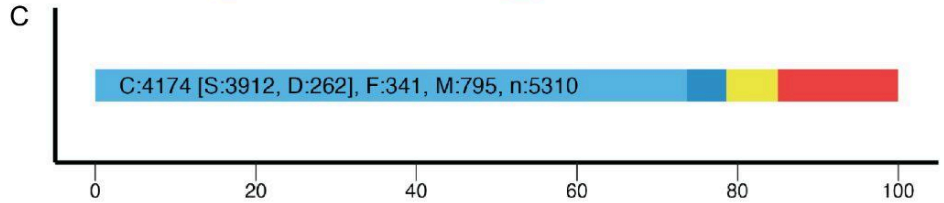
Supplementary Table 9: Introgression genes among tetraploid species.

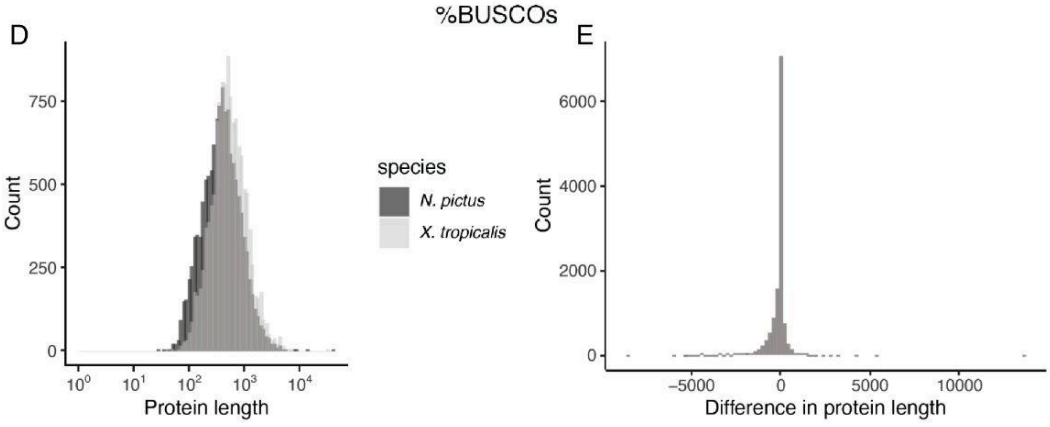
https://figshare.com/articles/dataset/Changes_in_cell_cycle_machinery_as_genetic_basis_for_polyploidy_stabilization_in_Australian_burrowing_frogs_Neobatrachus_/30576644?file=63103585

Supplemental figures

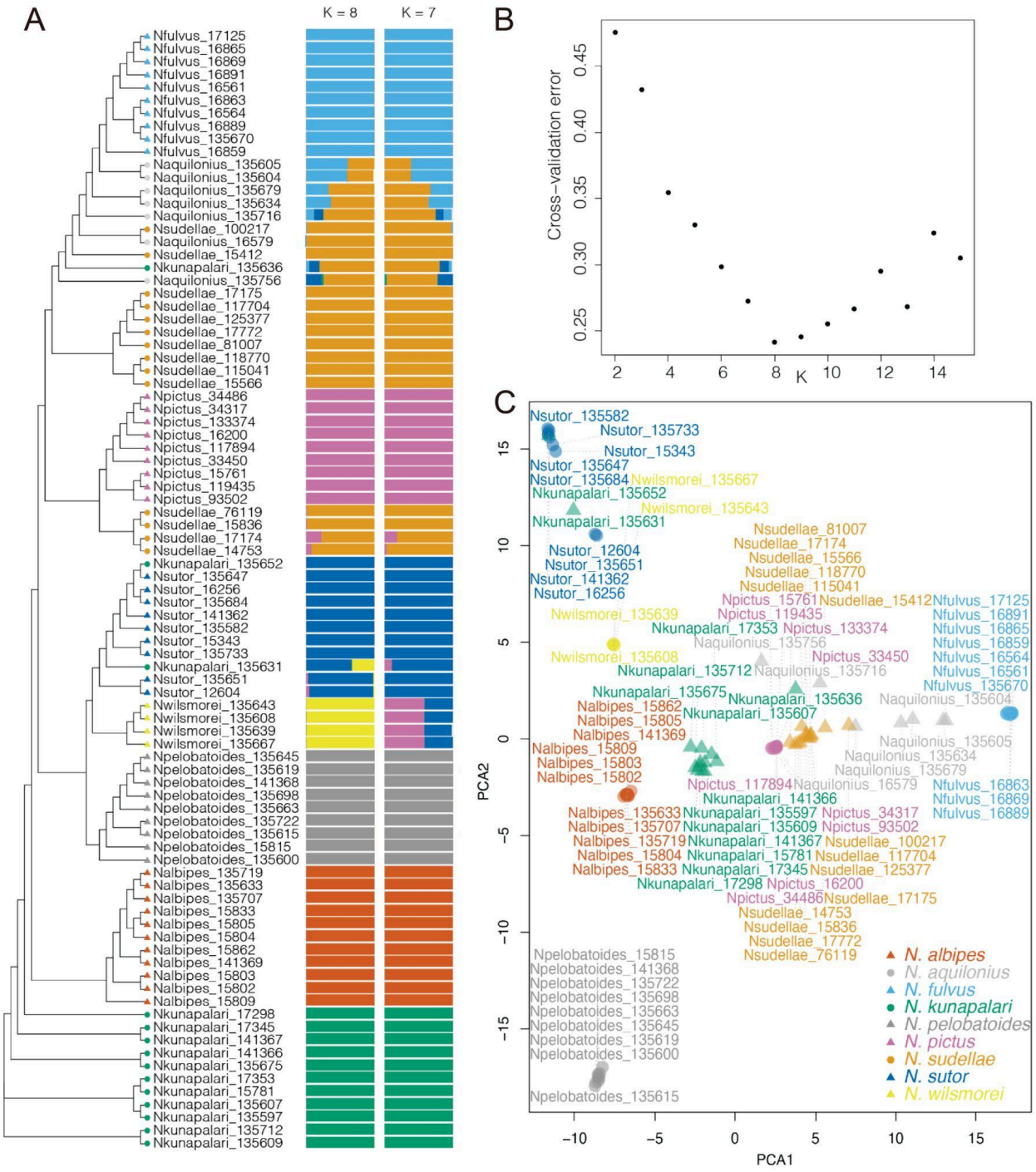

A
GenomeScope Profile
len:2,106,405,110bp uniq:52.4% het:0.656% kcov:13.8 err:0.863% dup:1.58% k:21
observed
full model
unique sequence
errors
kmer-peaks
Frequency
Coverage
GenomeScope Profile
len:2,568,306,954bp uniq:54.3% het:0.562% kcov:10.9 err:0.608% dup:1.28% k:41
observed
full model
unique sequence
errors
kmer-peaks
Frequency
Coverage


B
gene density
repeat density
Sc 30515
Sc 35107
Sc 17238
Sc 39220
Sc 39195
Sc 39221
Sc 6261
Sc 3532
Sc 39219
Sc 39222
Sc 13125
Sc 39223
Sc 35827
BUSCO Assessment Results
Complete (C) and single-copy (S)
Complete (C) and duplicated (D)
Fragmented (F)
Missing (M)

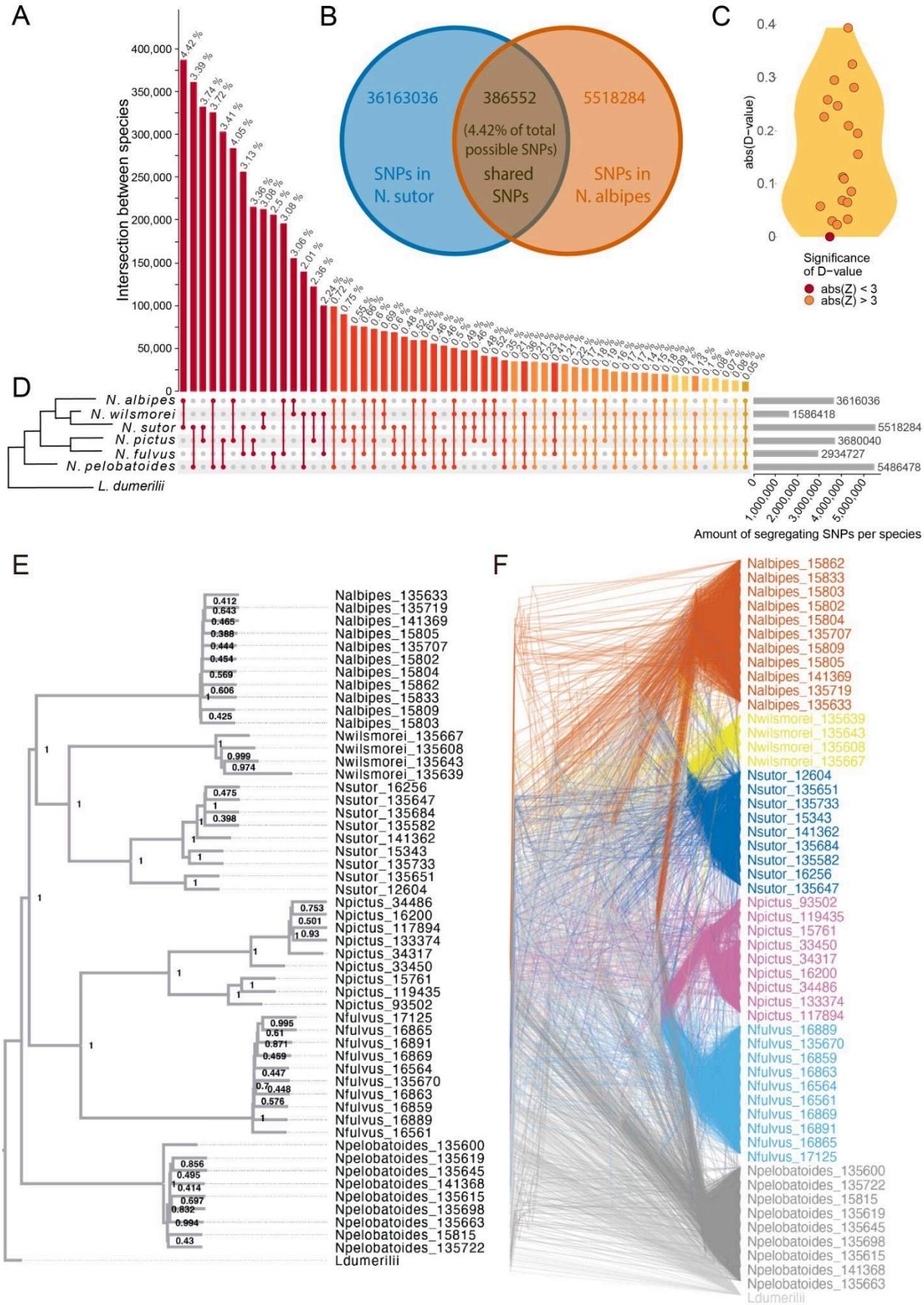

C
C:4174 [S:3912, D:262], F:341, M:795, n:5310
%BUSCOs


D
Count
Protein length
species
N. pictus
X. tropicalis
E
Count
Difference in protein length

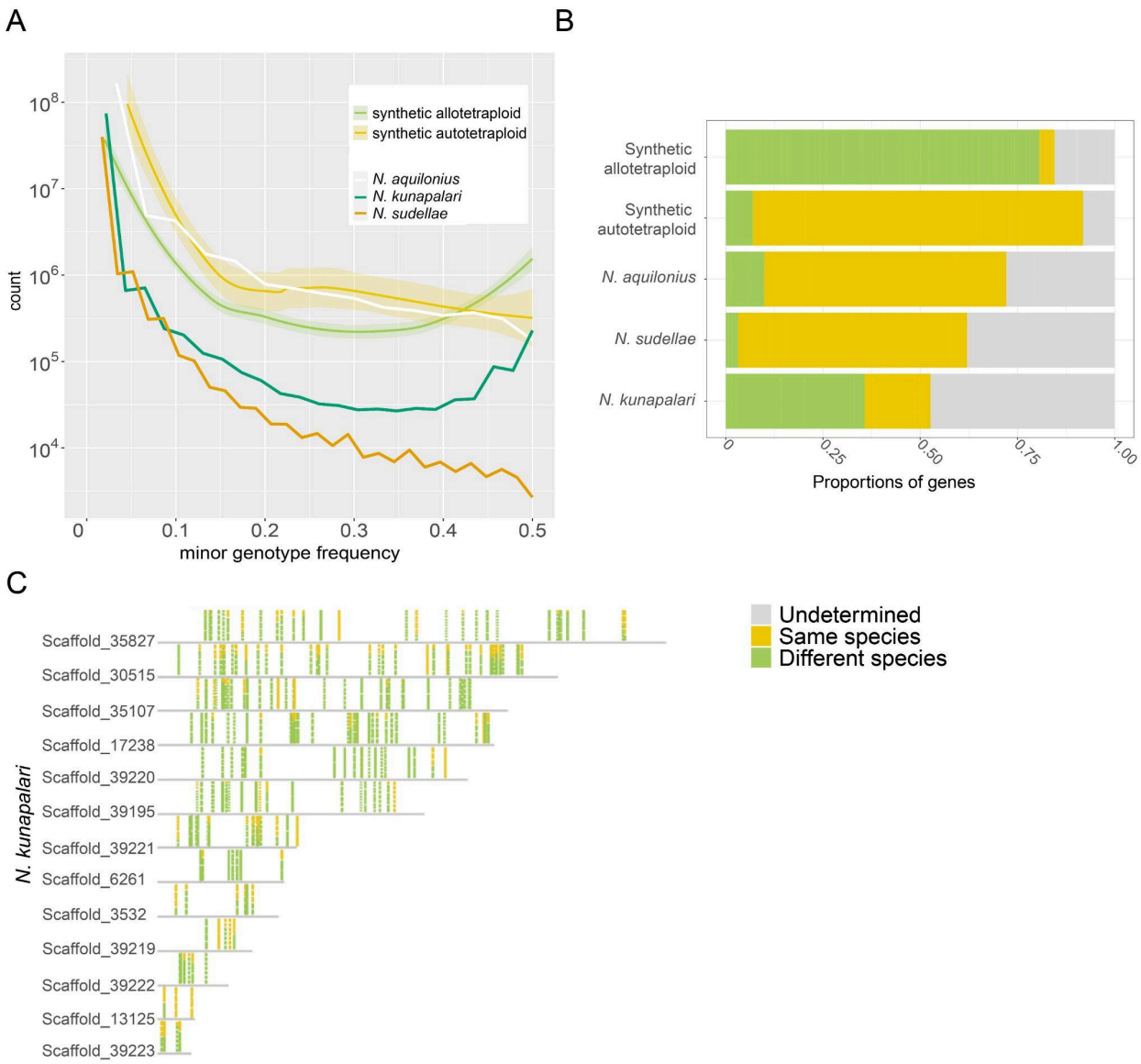
Supplementary Figure 1. A Genome size estimation of *N. pictus* based on k-mer histograms generated by JellyFish. Estimated genome size ranges from 2,106,405,110 to 2,568,306,954 (k-mer = 21-41). Estimation by Genomescope with k-mer = 21 (left) and k-mer = 41 (right). **B.** Gene and repeat density along the 13 biggest scaffolds of *N. pictus*. **C.** BUSCO Assessment results of the assembly of *N. pictus*. **D.** Protein length distribution of *N. pictus* and *X. tropicalis.* **E.** Comparison of protein length of *N. pictus* and *X. tropicalis*. Performed by blastp analysis of *N. pictus* against *X. tropicalis*. Proteins were filtered for evalue<0.0001 and pident>0.7, retaining 12451 of the original 22811 hits.



Supplementary Figure 2. A. Unrooted Neighbour-joining tree of neutral (4fds) sites, accompanied by admixture results for K=8 and K=7 . **B.** Cross Validation error for ADMIXTURE results amount of clusters (K) between 2 and 14. **C.** Principal component analysis of variation on 4fds. Tetraploid individuals are circles, diploid individuals are triangles.

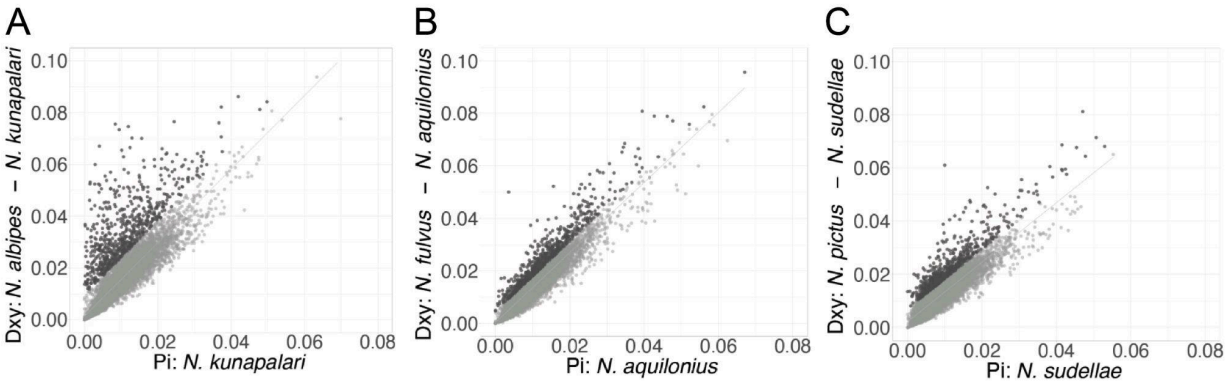

A
B
C
D
E
F
Intersection between species
36163036
386552
(4.42% of total possible SNPs)
5518284
SNPs in N. sutor
shared SNPs
SNPs in N. albipes
abs(D−value)
Significance of D−value
abs(Z) < 3
abs(Z) > 3
N. albipes
N. wilsmorei
N. sutor
N. pictus
N. fulvus
N. pelobatoides
L. dumerilii
3616036
1586418
5518284
3680040
2934727
5486478
Amount of segregating SNPs per species
Nalbipes_135633
Nalbipes_135719
Nalbipes_141369
Nalbipes_15805
Nalbipes_135707
Nalbipes_15802
Nalbipes_15804
Nalbipes_15862
Nalbipes_15833
Nalbipes_15809
Nalbipes_15803
Nwilsmorei_135667
Nwilsmorei_135608
Nwilsmorei_135643
Nwilsmorei_135639
Nsutor_16256
Nsutor_135647
Nsutor_135684
Nsutor_135582
Nsutor_141362
Nsutor_15343
Nsutor_135733
Nsutor_135651
Nsutor_12604
Npictus_34486
Npictus_16200
Npictus_117894
Npictus_133374
Npictus_34317
Npictus_33450
Npictus_15761
Npictus_119435
Npictus_93502
Nfulvus_17125
Nfulvus_16865
Nfulvus_16891
Nfulvus_16869
Nfulvus_16564
Nfulvus_135670
Nfulvus_16863
Nfulvus_16859
Nfulvus_16889
Nfulvus_16561
Npelobatoides_135600
Npelobatoides_135619
Npelobatoides_135645
Npelobatoides_141368
Npelobatoides_135615
Npelobatoides_135698
Npelobatoides_135663
Npelobatoides_15815
Npelobatoides_135722
Ldumerilii

Supplementary Figure 3. A. UpSet plot showing the number of shared biallelic single nucleotide polymorphisms (SNPs) between diploid *Neobatrachus* species. The color from dark red to light orange indicates the number of species sharing SNPs from two (dark red) to three, four, five, or all six (light orange) diploid species. Bars are ordered in descending order based on the number of shared SNPs (i.e., intersection region shown in B), and labeled with the percentage of shared SNPs out of the total SNPs (intersection/union) found in the compared species. The horizontal bar plot on the right shows the total number of biallelic SNPs present in each species. **B.** Venn diagram showing how shared SNPs (intersection) are identified in genic regions between a pair of species. The numbers shown here correspond to the first bar on the vertical barplot. **C.** D-statistics from ABBA-BABA analysis for the six diploid *Neobatrachus* species. Each possible combination in concordance with the species tree in Fig. 3F is reported. D-statistics are considered significant if the Z-score is greater than 3 or lower than −3. **D.** Tree topology of the diploid *Neobatrachus* species is shown on the left. The eastern banjo frog, *Limnodynastes dumerilii dumerilii*, was used as an outgroup. **E.** Consensus tree of 385 gene trees of all diploid *Neobatrachus* individuals and *Limnodynastes dumerilii*, based on the maximum number of quartet topologies in the gene trees, weighted by gene tree discordance, constructed using ASTER[158] (v1.15.2.3) and visualized using FigTree (v.1.4.4). Node labels indicate the percentage of quartets among the gene trees that agree with the branch. **F.** Densitree of 385 gene trees of all diploid *Neobatrachus* individuals and outgroup *Limnodynastes dumerilii.*

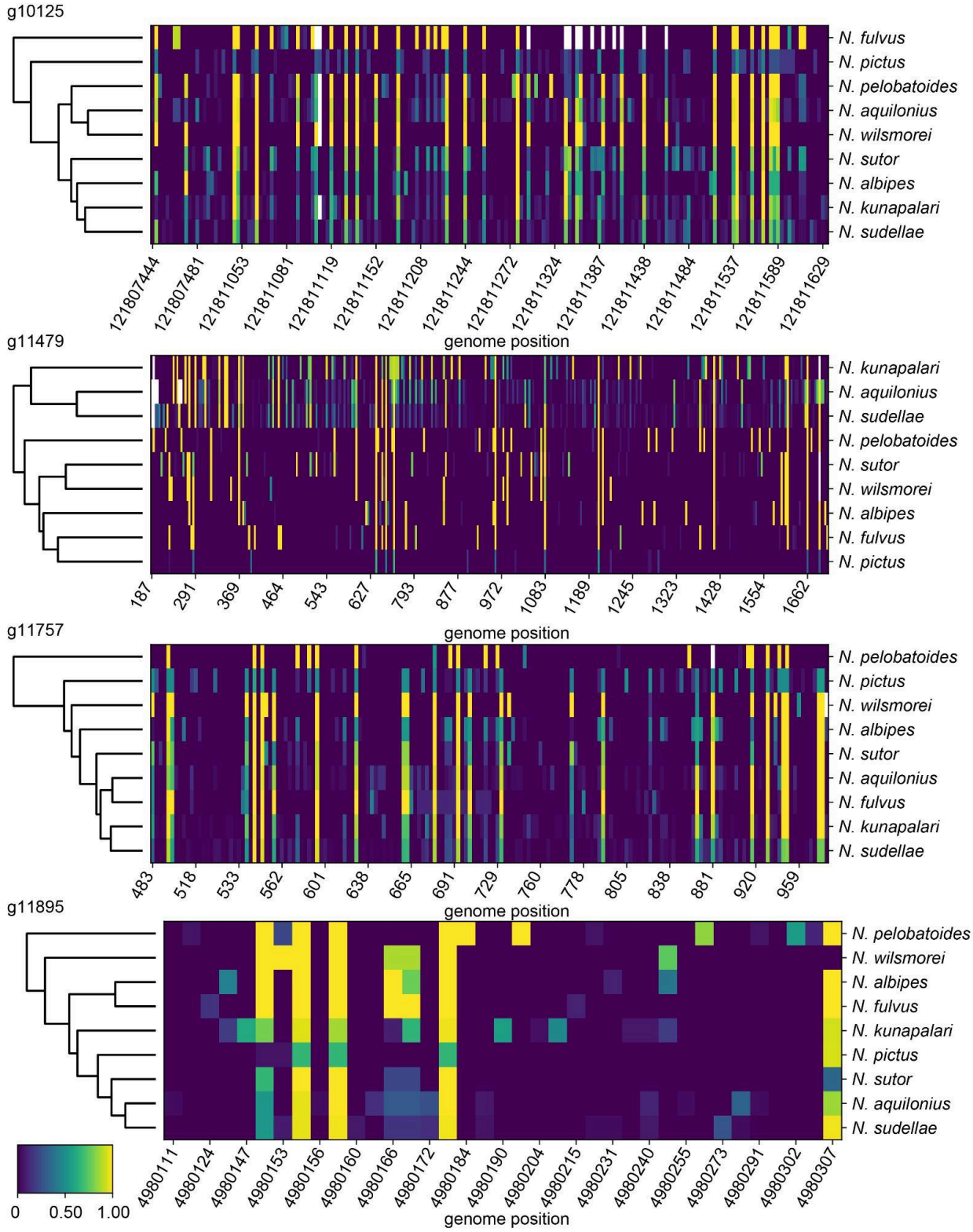


Supplementary Figure 4. A. Site frequency spectra of natural tetraploid *N. sudellae*, *N. aquilonius* and *N. kunapalari* in comparison with synthetic auto- and allotetraploids constructed *in silico* from the diploid species combinations. Synthetic allotetraploid populations exhibit a pronounced peak at intermediate frequency when mapped to a diploid reference due to fixed divergence between the diploid species, while autotetraploid populations do not. The shape of the site frequency spectra of the natural tetraploid *N. sudellae* and *N. aquilonius* resembles synthetic autotetraploids, while *N. kunapalari* shows an increase of minor genotype counts at 0.5 frequencies similar to synthetic allotetraploids. **B**. Overall phylogenetic assignment of natural and synthetic tetraploid pseudohaplotypes to the matching or discordant diploid species. **C**. Species-wide phylogenetic assignment of pseudohaplotypes of N. kunapalari to the matching (yellow) or discordant (green) diploid species along the genome. Each individual sample from N. kunapalari was assigned matching or discordant parental haplotypes, together forming a dotted bar at each locus. The dots correspond to an assignment of one individual at this locus. None of the longest scaffolds (chromosomes) are consistently assigned to matching (yellow) or discordants (green) parents,

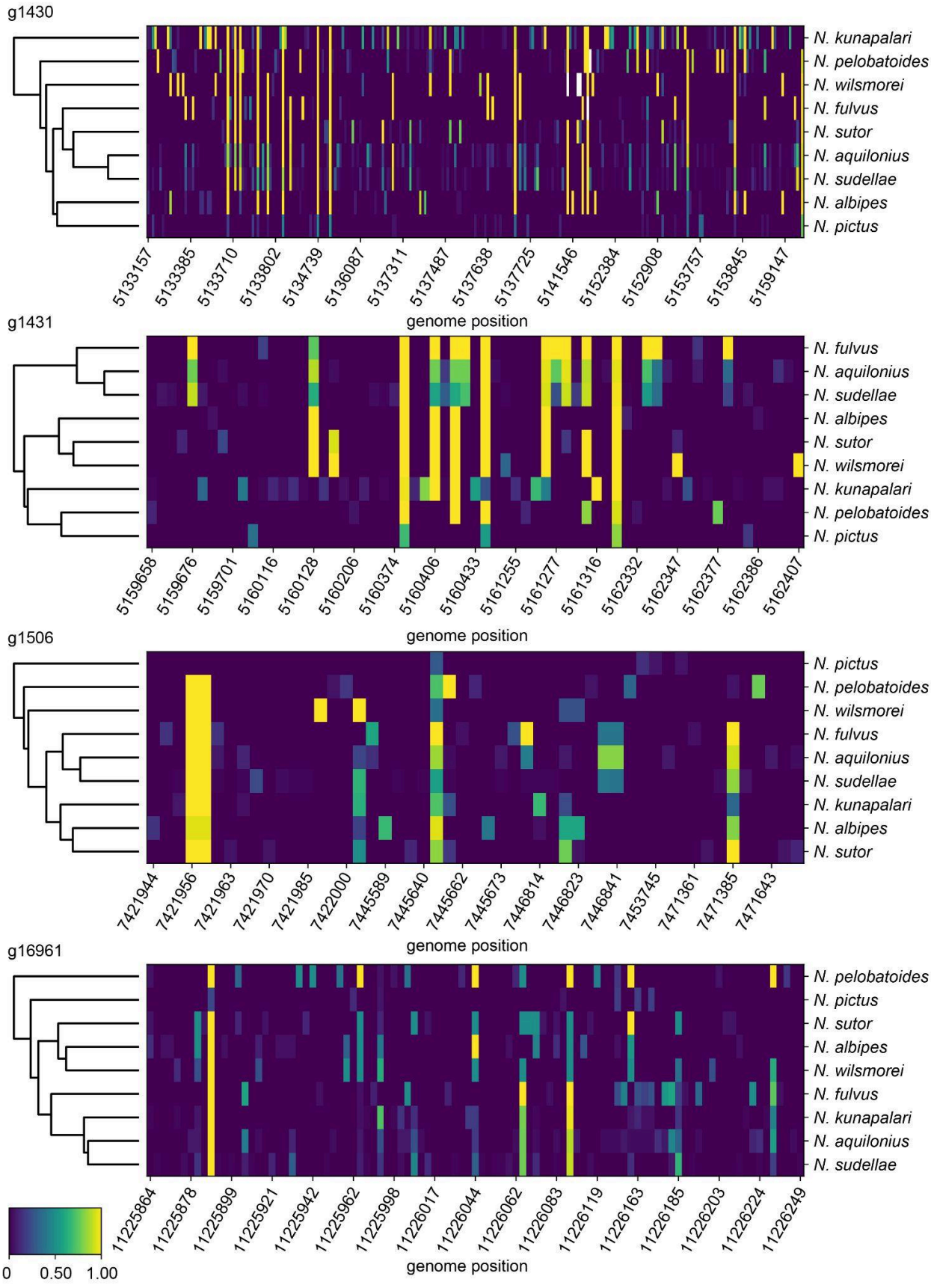
suggesting that *N. kunapalari* has mixed inheritance mode and homeologs can recombine throughout the genome.



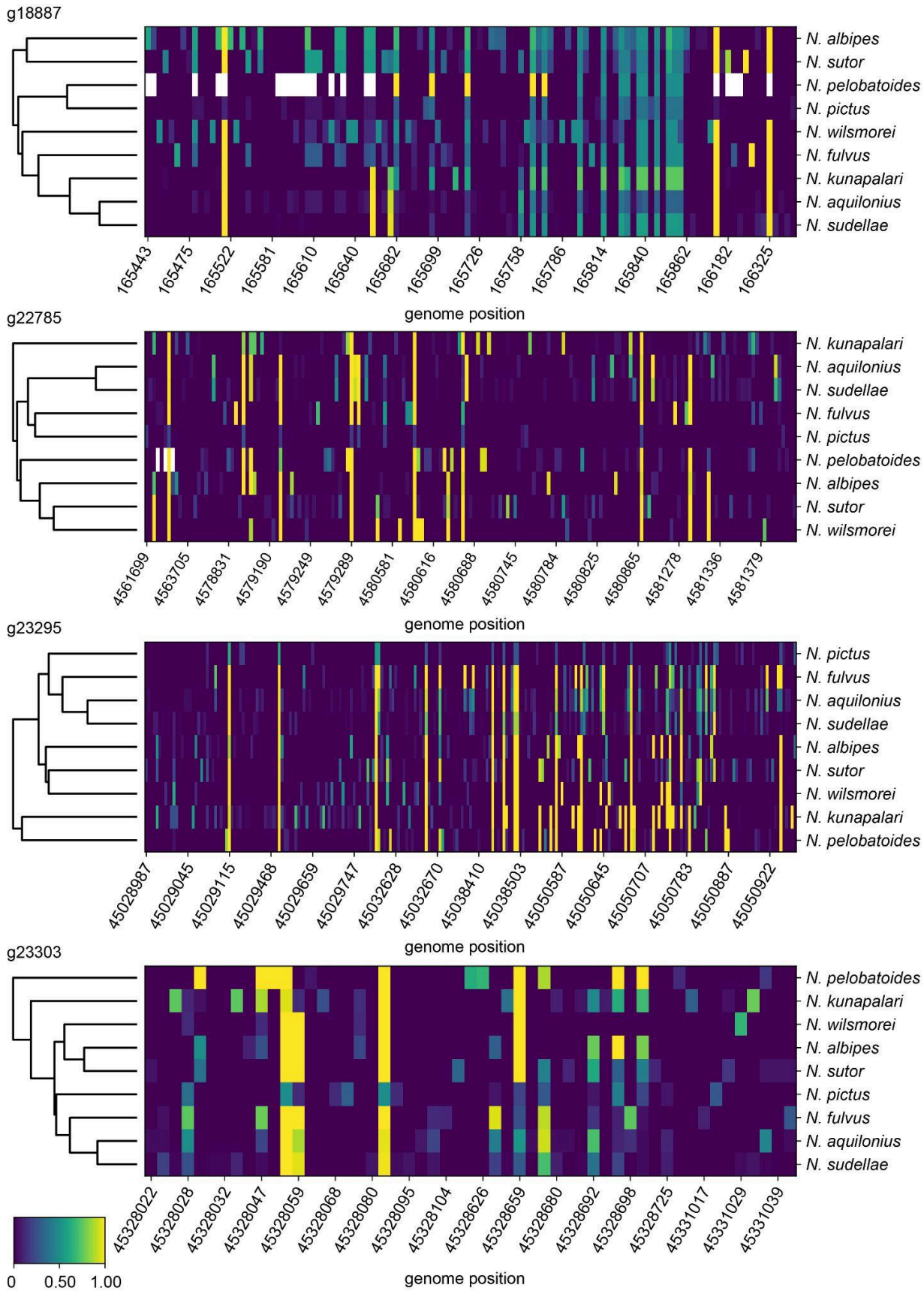
Supplementary Figure 5. Scatterplots showing the relationship between nucleotide diversity within tetraploid species and nucleotide divergence per diploid–tetraploid species pair per gene: (A) 2n *N. albipes* - 4n *N. kunapalari*; (B) 2n *N. fulvus* - 4n *N. aquilonius*; (C) 2n *N. pictus* - *N. sudellae* . The top 5% residual outliers from linear regression (gray line) are denoted in dark gray.



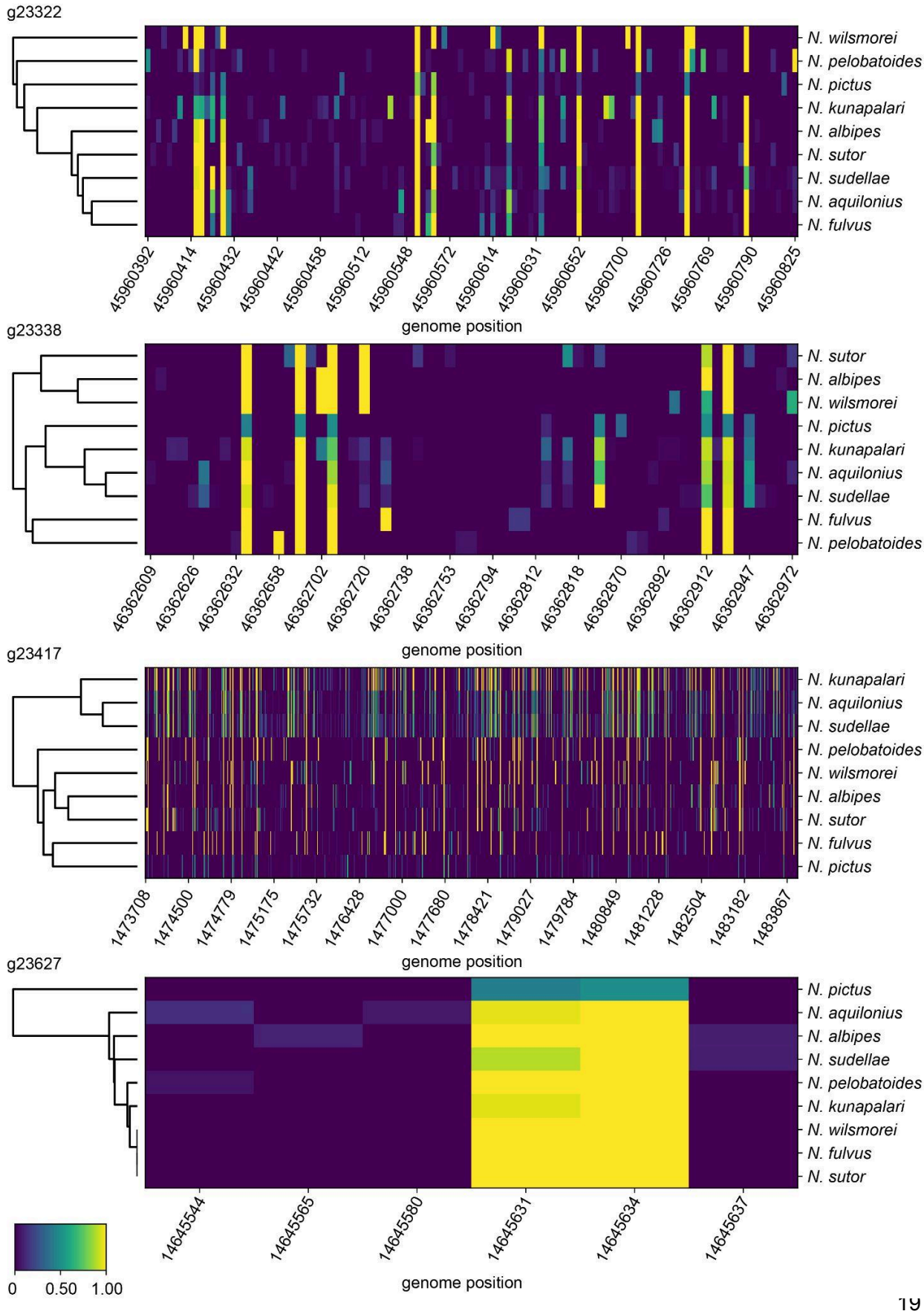
Supplementary Figure 6: Biallelic SNPs in CDS of genes under selection, colored and clustered by proportion of alternative allele per site per species (yellow: alternative allele is fixed in species; white: missing data in that species). [1-4 of 27]


g1430
N. kunapalari
N. pelobatoides
N. wilsmorei
N. fulvus
N. sutor
N. aquilonius
N. sudellae
N. albipes
N. pictus
5133157
5133385
5133710
5133802
5134739
5136087
5137311
5137487
5137638
5137725
5141546
5152384
5152908
5153757
5153845
5159147
genome position
g1431
N. fulvus
N. aquilonius
N. sudellae
N. albipes
N. sutor
N. wilsmorei
N. kunapalari
N. pelobatoides
N. pictus
5159658
5159676
5159701
5160116
5160128
5160206
5160374
5160406
5160433
5161255
5161277
5161316
5162332
5162347
5162377
5162386
5162407
genome position
g1506
N. pictus
N. pelobatoides
N. wilsmorei
N. fulvus
N. aquilonius
N. sudellae
N. kunapalari
N. albipes
N. sutor
7421944
7421956
7421963
7421970
7421985
7422000
7445589
7445640
7445662
7445673
7446814
7446823
7446841
7453745
7471361
7471385
7471643
genome position
g16961
N. pelobatoides
N. pictus
N. sutor
N. albipes
N. wilsmorei
N. fulvus
N. kunapalari
N. aquilonius
N. sudellae
11225864
11225878
11225899
11225921
11225942
11225962
11225998
11226017
11226044
11226062
11226083
11226119
11226163
11226185
11226203
11226224
11226249
genome position
0
0.50
1.00

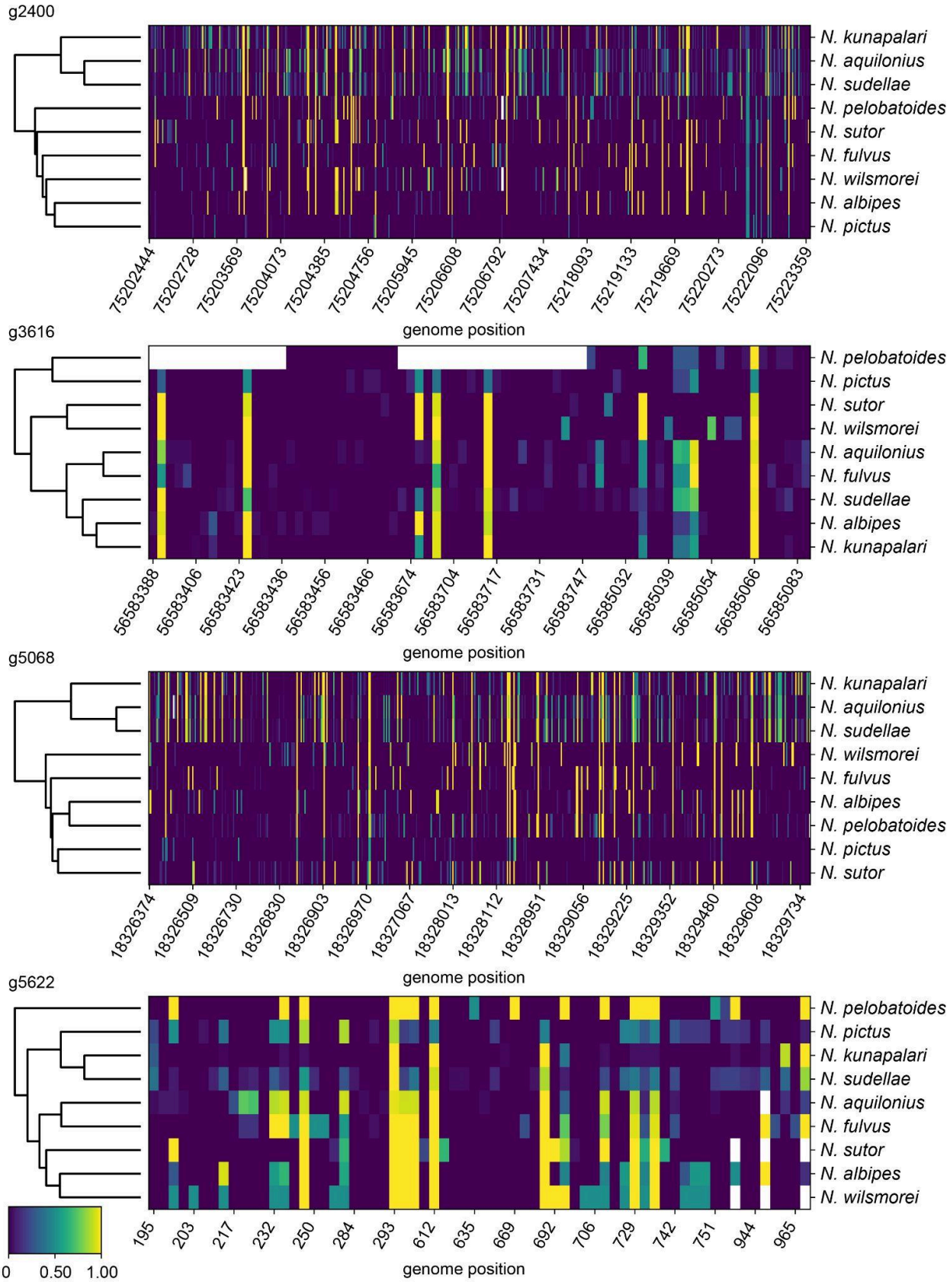
Supplementary Figure 6 cont.: Biallelic SNPs in CDS of genes under selection, colored and clustered by proportion of alternative allele per site per species (yellow: alternative allele is fixed; white: missing data in that species). [5-8 of 27]


g18887
N. albipes
N. sutor
N. pelobatoides
N. pictus
N. wilsmorei
N. fulvus
N. kunapalari
N. aquilonius
N. sudellae
165443
165475
165522
165581
165610
165640
165682
165699
165726
165758
165786
165814
165840
165862
166182
166325
genome position
g22785
N. kunapalari
N. aquilonius
N. sudellae
N. fulvus
N. pictus
N. pelobatoides
N. albipes
N. sutor
N. wilsmorei
4561699
4563705
4578831
4579190
4579249
4579289
4580581
4580616
4580688
4580745
4580784
4580825
4580865
4581278
4581336
4581379
genome position
g23295
N. pictus
N. fulvus
N. aquilonius
N. sudellae
N. albipes
N. sutor
N. wilsmorei
N. kunapalari
N. pelobatoides
45028987
45029045
45029115
45029468
45029659
45029747
45032628
45032670
45038410
45038503
45050587
45050645
45050707
45050783
45050887
45050922
genome position
g23303
N. pelobatoides
N. kunapalari
N. wilsmorei
N. albipes
N. sutor
N. pictus
N. fulvus
N. aquilonius
N. sudellae
45328022
45328028
45328032
45328047
45328059
45328068
45328080
45328095
45328104
45328626
45328659
45328680
45328692
45328698
45328725
45331017
45331029
45331039
genome position
0
0.50
1.00

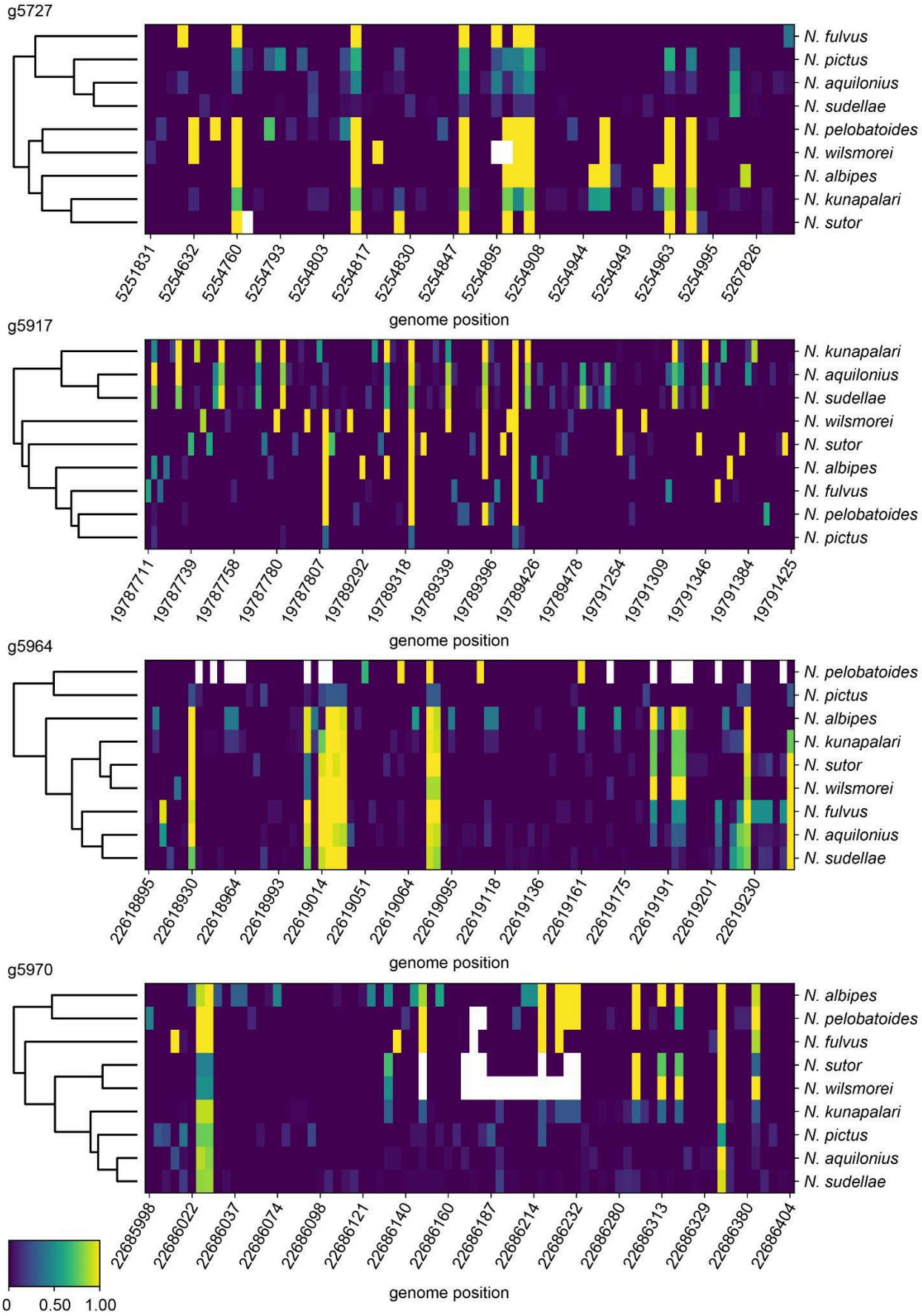
Supplementary Figure 6 cont.: Biallelic SNPs in CDS of genes under selection, colored and clustered by proportion of alternative allele per site per species (yellow: alternative allele is fixed; white: missing data in that species). [9-12 of 27]


g23322
N. wilsmorei
N. pelobatoides
N. pictus
N. kunapalari
N. albipes
N. sutor
N. sudellae
N. aquilonius
N. fulvus
45960392
45960414
45960432
45960442
45960458
45960512
45960548
45960572
45960614
45960631
45960652
45960700
45960726
45960769
45960790
45960825
genome position
g23338
N. sutor
N. albipes
N. wilsmorei
N. pictus
N. kunapalari
N. aquilonius
N. sudellae
N. fulvus
N. pelobatoides
46362609
46362626
46362632
46362658
46362702
46362720
46362738
46362753
46362794
46362812
46362818
46362870
46362892
46362912
46362947
46362972
genome position
g23417
N. kunapalari
N. aquilonius
N. sudellae
N. pelobatoides
N. wilsmorei
N. albipes
N. sutor
N. fulvus
N. pictus
1473708
1474500
1474779
1475175
1475732
1476428
1477000
1477680
1478421
1479027
1479784
1480849
1481228
1482504
1483182
1483867
genome position
g23627
N. pictus
N. aquilonius
N. albipes
N. sudellae
N. pelobatoides
N. kunapalari
N. wilsmorei
N. fulvus
N. sutor
14645544
14645565
14645580
14645631
14645634
14645637
genome position
0
0.50
1.00

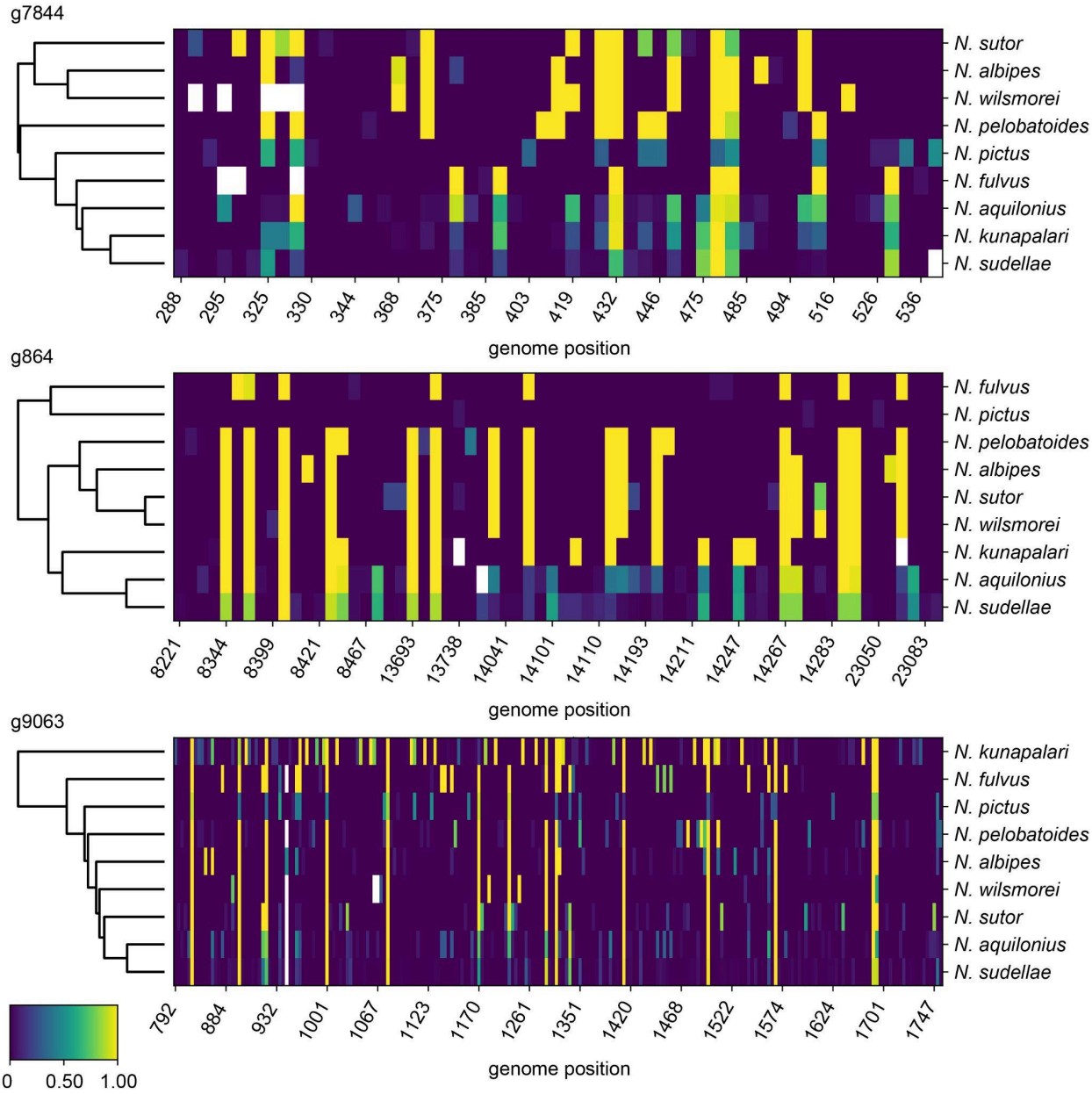
Supplementary Figure 6 cont.: Biallelic SNPs in CDS of genes under selection, colored and clustered by proportion of alternative allele per site per species (yellow: alternative allele is fixed; white: missing data in that species). [13-16 of 27]


g2400
N. kunapalari
N. aquilonius
N. sudellae
N. pelobatoides
N. sutor
N. fulvus
N. wilsmorei
N. albipes
N. pictus
75202444
75202728
75203569
75204073
75204385
75204756
75205945
75206608
75206792
75207434
75218093
75219133
75219669
75220273
75222096
75223359
genome position
g3616
N. pelobatoides
N. pictus
N. sutor
N. wilsmorei
N. aquilonius
N. fulvus
N. sudellae
N. albipes
N. kunapalari
56583388
56583406
56583423
56583436
56583456
56583466
56583674
56583704
56583717
56583731
56583747
56585032
56585039
56585054
56585066
56585083
genome position
g5068
N. kunapalari
N. aquilonius
N. sudellae
N. wilsmorei
N. fulvus
N. albipes
N. pelobatoides
N. pictus
N. sutor
18326374
18326509
18326730
18326830
18326903
18326970
18327067
18328013
18328112
18328951
18329056
18329225
18329352
18329480
18329608
18329734
genome position
g5622
N. pelobatoides
N. pictus
N. kunapalari
N. sudellae
N. aquilonius
N. fulvus
N. sutor
N. albipes
N. wilsmorei
195
203
217
232
250
284
293
612
635
669
692
706
729
742
751
944
965
genome position
0
0.50
1.00

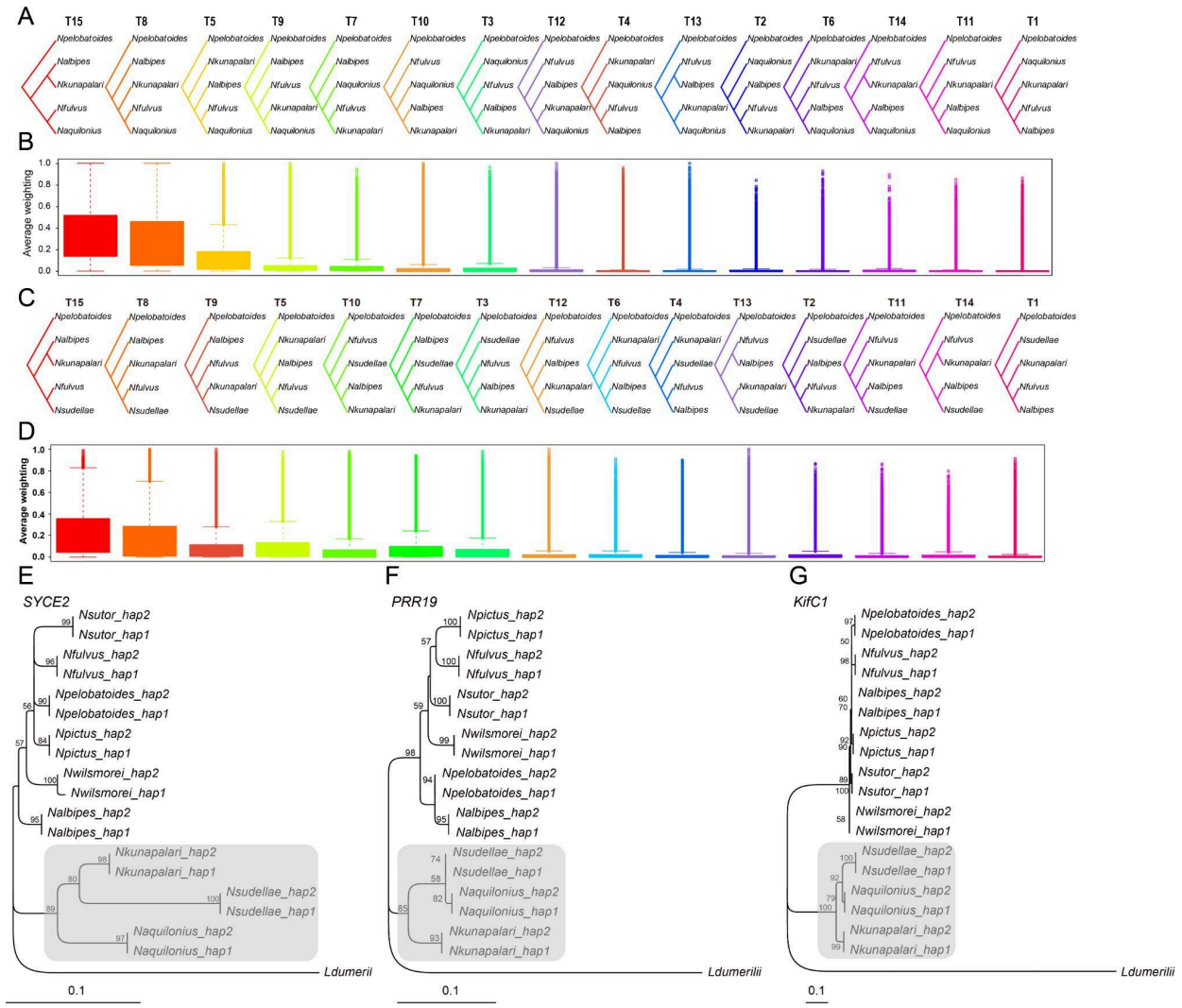
Supplementary Figure 6 cont.: Biallelic SNPs in CDS of genes under selection, colored and clustered by proportion of alternative allele per site per species (yellow: alternative allele is fixed; white: missing data in that species). [17-20 of 27]


g5727
N. fulvus
N. pictus
N. aquilonius
N. sudellae
N. pelobatoides
N. wilsmorei
N. albipes
N. kunapalari
N. sutor
5251831
5254632
5254760
5254793
5254803
5254817
5254830
5254847
5254895
5254908
5254944
5254949
5254963
5254995
5267826
genome position
g5917
N. kunapalari
N. aquilonius
N. sudellae
N. wilsmorei
N. sutor
N. albipes
N. fulvus
N. pelobatoides
N. pictus
19787711
19787739
19787758
19787780
19787807
19789292
19789318
19789339
19789396
19789426
19789478
19791254
19791309
19791346
19791384
19791425
genome position
g5964
N. pelobatoides
N. pictus
N. albipes
N. kunapalari
N. sutor
N. wilsmorei
N. fulvus
N. aquilonius
N. sudellae
22618895
22618930
22618964
22618993
22619014
22619051
22619064
22619095
22619118
22619136
22619161
22619175
22619191
22619201
22619230
genome position
g5970
N. albipes
N. pelobatoides
N. fulvus
N. sutor
N. wilsmorei
N. kunapalari
N. pictus
N. aquilonius
N. sudellae
22685998
22686022
22686037
22686074
22686098
22686121
22686140
22686160
22686187
22686214
22686232
22686280
22686313
22686329
22686380
22686404
genome position
0
0.50
1.00

Supplementary Figure 6 cont.: Biallelic SNPs in CDS of genes under selection, colored and clustered by proportion of alternative allele per site per species (yellow: alternative allele is fixed; white: missing data in that species). [21-24 of 27]



Supplementary Figure 6 cont.: Biallelic SNPs in CDS of genes under selection, colored and clustered by proportion of alternative allele per site per species (yellow: alternative allele is fixed; white: missing data in that species). [25-27 of 27]



Supplementary Figure 7.: Topologies scans among tetraploids and candidate-gene phylogenies based on true haplotypes. A, C, The 15 possible rooted topologies for the two five-taxon datasets used in topology weighting analyses, with *N. pelobatoides* as the outgroup. The first dataset examines tetraploid relationships involving *N. kunapalari* and *N. aquilonius* (A), and the second examines relationships involving *N. kunapalari* and *N. sudellae* (C). B, D, Boxplots showing the genome-wide distribution of topology weighting for each topology shown in A and C, respectively. Introgression topologies 9, 12, and 13 showed the strongest support and were therefore summed in downstream analyses (Fig. 3F, G) to represent introgression phylogenetic signals among tetraploids. E–G, True haplotype gene trees for *SYCE2, PRR19,* and *KifC1*, reconstructed from 18 linked-read genome assemblies representing six diploid and three tetraploid species. The grey shaded boxes highlight the clade containing the three tetraploid species.