

Eukaryotes, read depth

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Accumulation Curves within Samples.

We want to establish whether the actual read depth was sufficient. Are we likely to have found more phyla if we had had more reads? In order to answer this question, I have done a set of simulations, 1000 for each sample. Within each simulation, I have subsampled one sample until the total number of observed phyla in the sample had been found. I then plotted one accumulation curve as an example, and I collected statistics of all 1000 accumulation curves: The number of OTUs that needed to be read before we observed all phyla in the sample. There is one such number for each of 1000 simulations for each sample. I then report the maximum of these 1000 numbers for each sample, and the 95% quantile. For example, if the 95% quantile for one sample was 5000, then we can be 95% sure that we would have found all observed phyla by reading 5000 OTUs. If this number is substantially below the actual number of sequences read, we can be confident that we have read a sufficient number of sequences.

Would we have seen a rare phylum with the given read depth?

I have also used a second approach: Suppose we add an extra phylum to each sample, as one we have not observed, and suppose it is half as abundant as the least abundant phylum in this sample. How likely are we to have observed this extra phylum within the actual number of OTUs read? For example, sample 18S.1.1.C.PCM.MM contains Arthropoda (abundance 1315) and Streptophyta (abundance 143). Suppose there had been a third unobserved phylum in proportion of half the Streptophyta, how many reads would be required so that we may expect to see it? If this is less than the actual read depth of the sample, then this would be an indication that the read depth was sufficient. We can find the expected number of reads required by viewing the problem as an absorbing Markov Chain. For example, for a situation with 5 phyla, the initial state would be “{0,0,0,0,0}” (no phyla seen), and the final state would be “{1,1,1,1,1}” (all phyla seen). We can write down the transition matrix from the observed proportions of the phyla, and then we have the expected number of transitions before reaching the absorbing state in closed form.

The following table shows the results.

Table 1: Longtable

Sample	Observed		Simulation			Markov Chain	
	n phyla	Sample Abundance	p95	p100	frac	mn	low
18S.1.1.C.PCM.MM	2	1458	27	66	0.02	25	
18S.1.1.D.PCM.ME	10	18812	4005	7563	0.21	3434	
18S.1.1.F.PCM.ME	3	9020	784	1617	0.09	684	
18S.1.1.G.PCM.ME	6	5340	4158	5020	0.78	6231	*
18S.1.1.H.PCM.ME	4	35026	6796	19288	0.19	5840	
18S.1.10.B.PCM.MM	2	1363	6	14	0.00	7	
18S.1.10.C.PCM.MM	2	1689	13	32	0.01	11	
18S.1.10.D.PCM.ME	2	166	14	31	0.08	13	

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Table 1: Longtable (*continued*)

Sample	Observed		Simulation			Markov Chain	
	n phyla	Sample Abundance	p95	p100	frac	mn	low
18S.1.10.E.PCM.ME	3	1448	429	832	0.30	377	
18S.1.10.F.PCM.ME	5	20748	13630	19211	0.66	16139	
18S.1.10.G.PCM.ME	6	14319	5664	9538	0.40	5570	
18S.1.11.A.PCM.MM	2	4697	11	20	0.00	10	
18S.1.11.C.PCM.MM	6	19591	4123	8768	0.21	3267	
18S.1.11.D.PCM.ME	7	46082	725	1509	0.02	568	
18S.1.11.E.PCM.ME	8	7457	149	366	0.02	117	
18S.1.11.G.PCM.ME	8	15771	3655	7239	0.23	3383	
18S.1.2.B.PCM.MM	2	4369	2044	3546	0.47	2040	
18S.1.2.C.PCM.MM	6	21167	692	1642	0.03	563	
18S.1.2.D.PCM.ME	2	5487	7	19	0.00	8	
18S.1.2.E.PCM.ME	4	12105	1173	2623	0.10	975	
18S.1.2.F.PCM.ME	2	432	47	112	0.11	41	
18S.1.2.G.PCM.ME	7	11566	155	328	0.01	126	
18S.1.2.H.PCM.ME	7	3696	906	1583	0.25	801	
18S.1.3.B.PCM.MM	2	16	5	9	0.31	7	
18S.1.3.D.PCM.ME	5	6208	801	2009	0.13	727	
18S.1.3.E.PCM.ME	5	10298	1152	2407	0.11	1047	
18S.1.3.H.PCM.ME	3	503	95	182	0.19	85	
18S.1.4.B.PCM.MM	6	6076	50	119	0.01	41	
18S.1.4.D.PCM.ME	4	2362	71	146	0.03	56	
18S.1.4.E.PCM.ME	3	1163	433	831	0.37	453	
18S.1.4.F.PCM.ME	5	20475	15550	20372	0.76	23931	*
18S.1.4.H.PCM.ME	4	5947	4740	5827	0.80	7237	*
18S.1.5.B.PCM.MM	3	20482	4736	8964	0.23	3987	
18S.1.5.D.PCM.ME	2	568	442	555	0.78	664	*
18S.1.5.E.PCM.ME	2	9029	593	1529	0.07	449	
18S.1.5.F.PCM.ME	8	7140	3966	5612	0.56	4437	
18S.1.5.H.PCM.ME	4	15322	9946	13981	0.65	12186	
18S.1.6.B.PCM.MM	4	5890	20	67	0.00	17	
18S.1.6.D.PCM.ME	12	40399	11381	19871	0.28	10602	
18S.1.6.E.PCM.ME	5	18220	7307	12197	0.40	7098	
18S.1.6.F.PCM.ME	3	869	115	317	0.13	98	
18S.1.6.G.PCM.ME	2	1575	51	138	0.03	41	
18S.1.6.H.PCM.ME	6	5954	293	557	0.05	227	
18S.1.7.B.PCM.MM	3	86804	424	858	0.00	319	
18S.1.7.C.PCM.MM	5	953	98	233	0.10	85	
18S.1.7.E.PCM.ME	2	2037	76	140	0.04	65	
18S.1.7.F.PCM.ME	3	2433	229	444	0.09	189	
18S.1.7.G.PCM.ME	4	4294	114	276	0.03	90	
18S.1.7.H.PCM.ME	7	4688	837	1836	0.18	747	
18S.1.8.A.PCM.MM	2	16744	7	15	0.00	8	
18S.1.8.B.PCM.MM	3	2379	23	61	0.01	20	
18S.1.8.D.PCM.ME	5	4203	29	59	0.01	25	

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Table 1: Longtable (*continued*)

Sample	Observed		Simulation			Markov Chain	
	n phyla	Sample Abundance	p95	p100	frac	mn	low
18S.1.8.E.PCM.ME	5	7962	99	227	0.01	79	
18S.1.8.F.PCM.ME	7	20267	566	1514	0.03	430	
18S.1.8.H.PCM.ME	9	10536	1477	2955	0.14	1297	
18S.1.9.B.PCM.MM	2	2402	6	13	0.00	7	
18S.1.9.D.PCM.ME	4	11771	5380	8837	0.46	5619	
18S.1.9.E.PCM.ME	4	6820	5139	6628	0.75	7958	*
18S.1.9.G.PCM.ME	7	19521	11712	17241	0.60	15184	
18S.1.9.H.PCM.ME	5	28268	517	1148	0.02	406	
18S.2.1.A.PCM.ME	2	4934	333	613	0.07	275	
18S.2.1.B.PCM.ME	6	2600	189	375	0.07	165	
18S.2.1.C.PCM.LT	8	31078	2343	5840	0.08	1799	
18S.2.1.D.PCM.LT	8	9690	81	197	0.01	68	
18S.2.1.E.PCM.LT	6	9182	211	567	0.02	169	
18S.2.1.F.PCM.LT	5	30442	141	306	0.00	111	
18S.2.1.G.PCM.LT	5	8660	82	183	0.01	60	
18S.2.1.H.PCM.LT	10	14341	2376	4844	0.17	1980	
18S.2.10.A.PCM.ME	11	16069	1889	3325	0.12	1540	
18S.2.10.B.PCM.LT	10	12684	6698	10374	0.53	8375	
18S.2.10.D.PCM.LT	11	179903	10044	24426	0.06	8310	
18S.2.10.F.PCM.LT	6	13568	1949	5492	0.14	1765	
18S.2.10.G.PCM.LT	8	10778	5531	9015	0.51	6291	
18S.2.11.B.PCM.LT	10	13970	2090	3786	0.15	1602	
18S.2.11.C.PCM.LT	12	14834	933	1928	0.06	699	
18S.2.11.D.PCM.LT	8	5289	340	848	0.06	297	
18S.2.11.E.PCM.LT	4	20014	81	208	0.00	63	
18S.2.11.F.PCM.LT	7	6663	2216	4131	0.33	2227	
18S.2.2.A.PCM.ME	3	2354	68	177	0.03	58	
18S.2.2.B.PCM.ME	3	1691	37	80	0.02	33	
18S.2.2.C.PCM.LT	4	4167	45	137	0.01	38	
18S.2.2.D.PCM.LT	6	7093	5526	6970	0.78	8276	*
18S.2.2.E.PCM.LT	10	20629	3500	8821	0.17	3037	
18S.2.2.F.PCM.LT	8	14258	280	590	0.02	217	
18S.2.2.G.PCM.LT	8	19146	1488	3559	0.08	1168	
18S.2.3.A.PCM.ME	2	110403	1288	2638	0.01	1023	
18S.2.3.D.PCM.LT	11	5319	4213	4981	0.79	6461	*
18S.2.3.E.PCM.LT	4	4400	47	128	0.01	41	
18S.2.3.F.PCM.LT	12	14402	427	877	0.03	324	
18S.2.3.G.PCM.LT	3	5265	41	103	0.01	35	
18S.2.4.A.PCM.ME	6	26469	540	1070	0.02	412	
18S.2.4.B.PCM.ME	4	2813	103	253	0.04	85	
18S.2.4.C.PCM.LT	5	5776	79	162	0.01	60	
18S.2.4.D.PCM.LT	10	86257	30831	53971	0.36	28955	
18S.2.4.E.PCM.LT	5	14329	362	844	0.03	266	
18S.2.4.F.PCM.LT	11	71747	31562	47239	0.44	30744	

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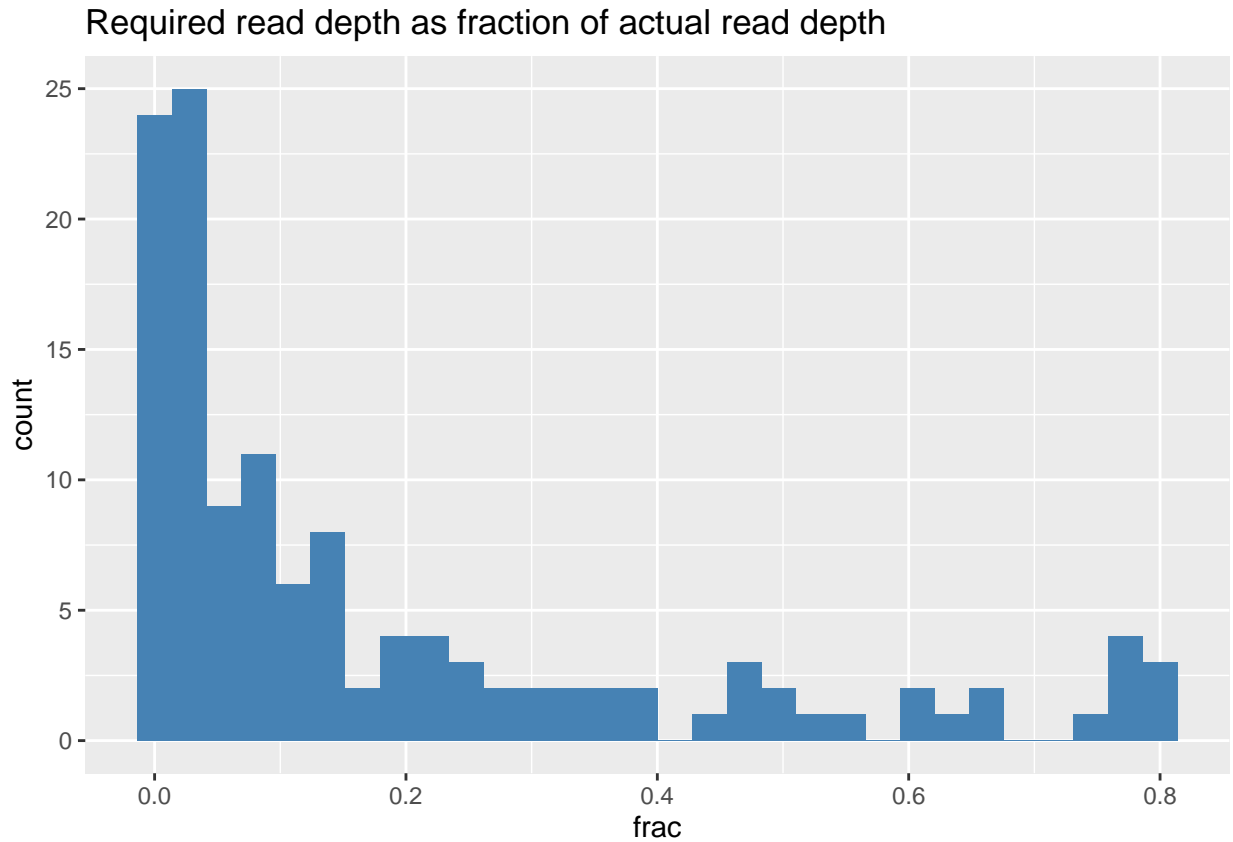
Table 1: Longtable (*continued*)

Sample	Observed		Simulation			Markov Chain	
	n phyla	Sample Abundance	p95	p100	frac	mn	low
18S.2.4.G.PCM.LT	4	4290	28	69	0.01	26	
18S.2.5.A.PCM.ME	6	6611	178	344	0.03	144	
18S.2.5.B.PCM.ME	8	2582	278	664	0.11	215	
18S.2.5.C.PCM.LT	8	12004	909	1812	0.08	649	
18S.2.5.D.PCM.LT	10	42924	5705	14945	0.13	4775	
18S.2.5.F.PCM.LT	6	2101	1341	1890	0.64	1635	
18S.2.5.G.PCM.LT	7	27119	12446	23387	0.46	12661	
18S.2.6.A.PCM.ME	12	1882	525	1184	0.28	494	
18S.2.6.B.PCM.ME	10	20668	708	1285	0.03	519	
18S.2.6.C.PCM.LT	7	27162	6455	12804	0.24	5766	
18S.2.6.D.PCM.LT	9	31444	2002	4264	0.06	1614	
18S.2.6.E.PCM.LT	6	30281	7782	18801	0.26	7071	
18S.2.6.F.PCM.LT	5	9004	91	188	0.01	78	
18S.2.7.A.PCM.ME	11	52727	42147	50990	0.80	61587	*
18S.2.7.C.PCM.LT	11	23378	14107	21586	0.60	18228	
18S.2.7.D.PCM.LT	8	88274	17101	32693	0.19	14268	
18S.2.7.E.PCM.LT	6	10969	261	500	0.02	201	
18S.2.7.F.PCM.LT	5	13291	205	410	0.02	161	
18S.2.8.A.PCM.ME	7	7936	4032	6662	0.51	4650	
18S.2.8.B.PCM.LT	7	13802	870	2306	0.06	649	
18S.2.8.C.PCM.LT	13	44312	14025	28044	0.32	13528	
18S.2.8.D.PCM.LT	11	19798	1696	5223	0.09	1259	
18S.2.8.E.PCM.LT	9	20792	937	2213	0.05	810	
18S.2.8.F.PCM.LT	6	2549	69	153	0.03	55	
18S.2.8.G.PCM.LT	7	23341	923	1943	0.04	781	
18S.2.9.A.PCM.ME	3	41336	993	2248	0.02	779	
18S.2.9.C.PCM.LT	7	26464	1427	2957	0.05	1078	
18S.2.9.D.PCM.LT	6	7431	406	821	0.05	285	
18S.2.9.E.PCM.LT	7	12351	991	1731	0.08	752	
18S.2.9.F.PCM.LT	7	12783	1782	3994	0.14	1458	
18S.2.9.G.PCM.LT	5	63942	8070	16670	0.13	6783	

- ‘n phyla’: The number of phyla observed in the sample.
- Sample abundance: The total abundance for the sample.
- p95: The total abundance required so that we found all phyla in the sample 95% of the time.
- p100: The highest abundance required to find all phyla in the sample over all 1000 simulations.
- frac: p95 divided by the sample abundance. For example, if this is 0.7, we can be 95% sure that we would have found all the phyla we did observe, with 70% of the actual read depth.
- mn: The expected of reads needed to observe an extra phylum, half as rare as the rarest phylum in the sample.
- low: Shows an asterisk if mn is greater than the actual read depth. This shows that the read depth was not sufficient in this case to detect to hypothetical, rare, extra phylum.

If p95 is well below the Sample Abundance, then this would give us confidence that the total read depth was sufficient. Leaving out those samples where there is only one phylum, where clearly there is no problem in making an accumulation curve, we see that this number is below 0.8 for all samples. In other words, we did not discover any new phyla beyond 80% of the actual read depth in any of our samples, in 95% of the

simulations. It seems to be reasonable to conclude that our read depth was sufficient.



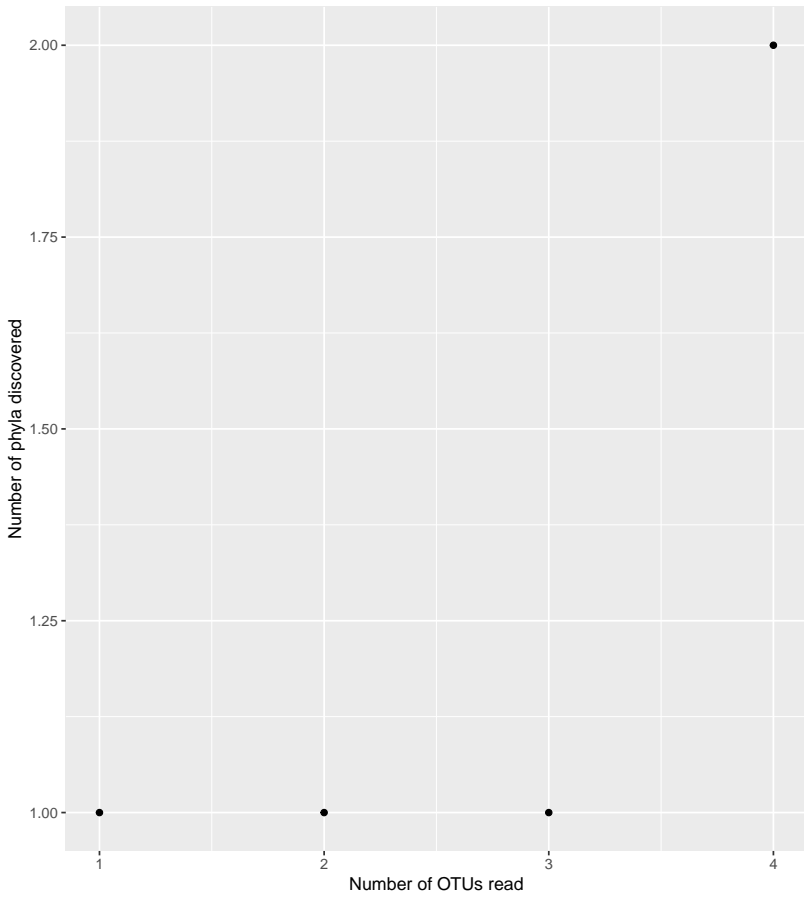
Required read depth here is the number of OTUs which was needed in 95% of the simulations to observe all the phyla actually observed in the sample.

For the second approach, we see that there are 8 samples in which we would miss the hypothetical rare phylum. In each of these samples, there is an observed phylum with only 2 OTUs observed, so that the extra phylum would have only 1 OTU, and we are missing something very rare indeed. This can never be entirely avoided.

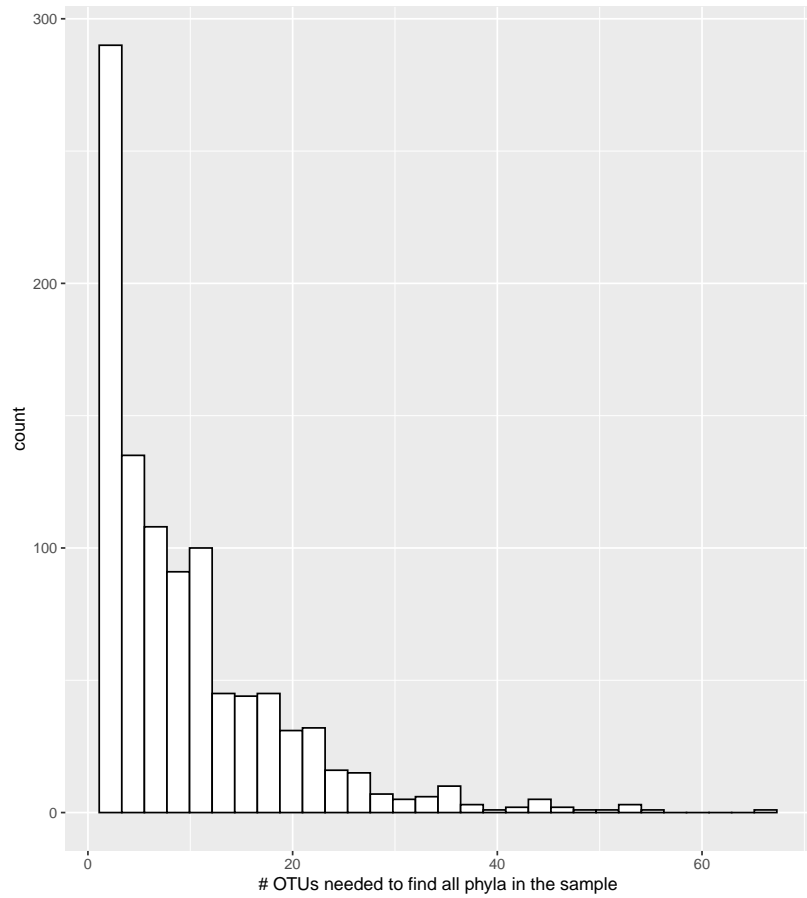
All in all, I think it is reasonable to assume that the actual read depth was sufficient. A limitation is that we have assumed sampling is random. In reality, OTUs of the same kind probably cluster together, but we have no way of modeling this, and this is the best we can do.

Appendix: Accumulation Curves: One example and 1000 simulations for each Sample in the Dataset.

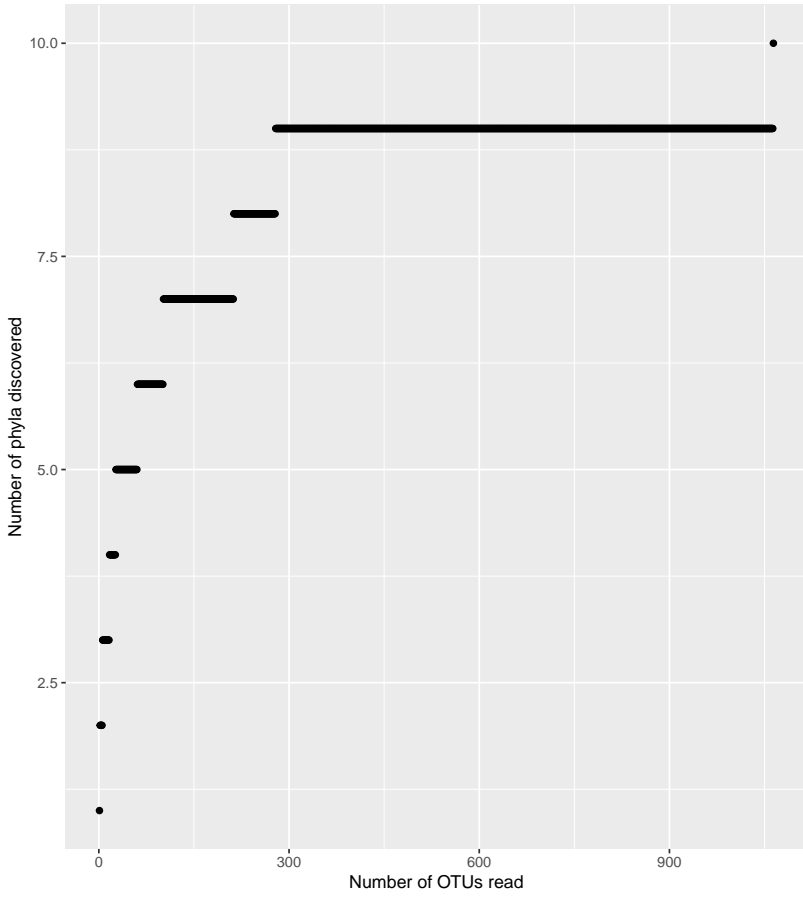
1. 18S.1.1.C.PCM.MM



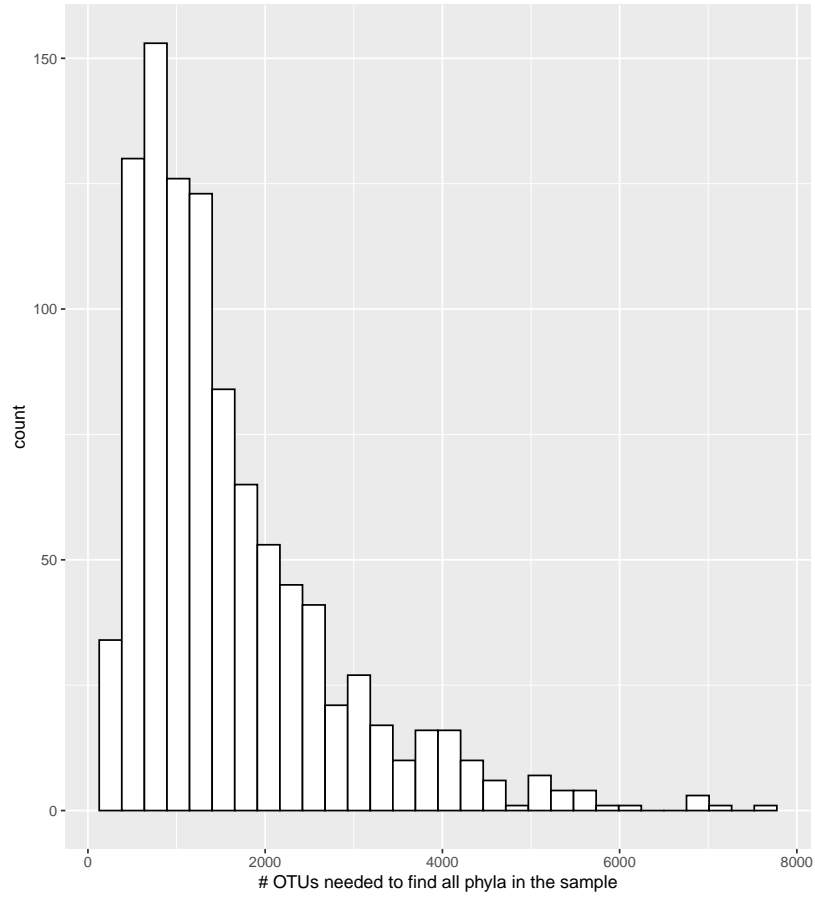
18S.1.1.C.PCM.MM



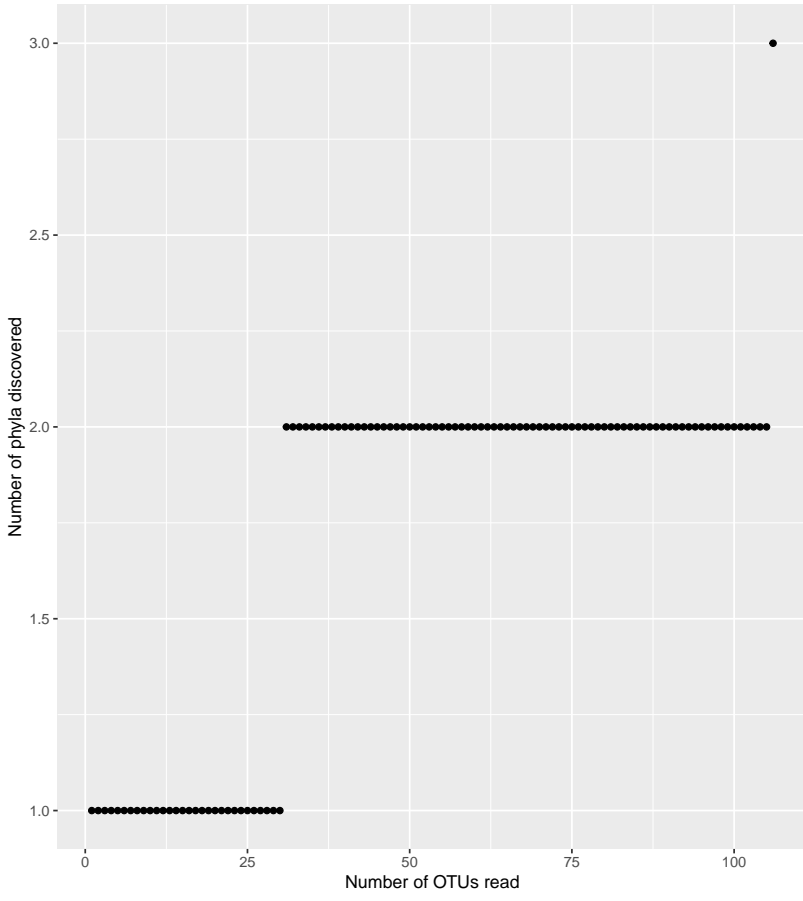
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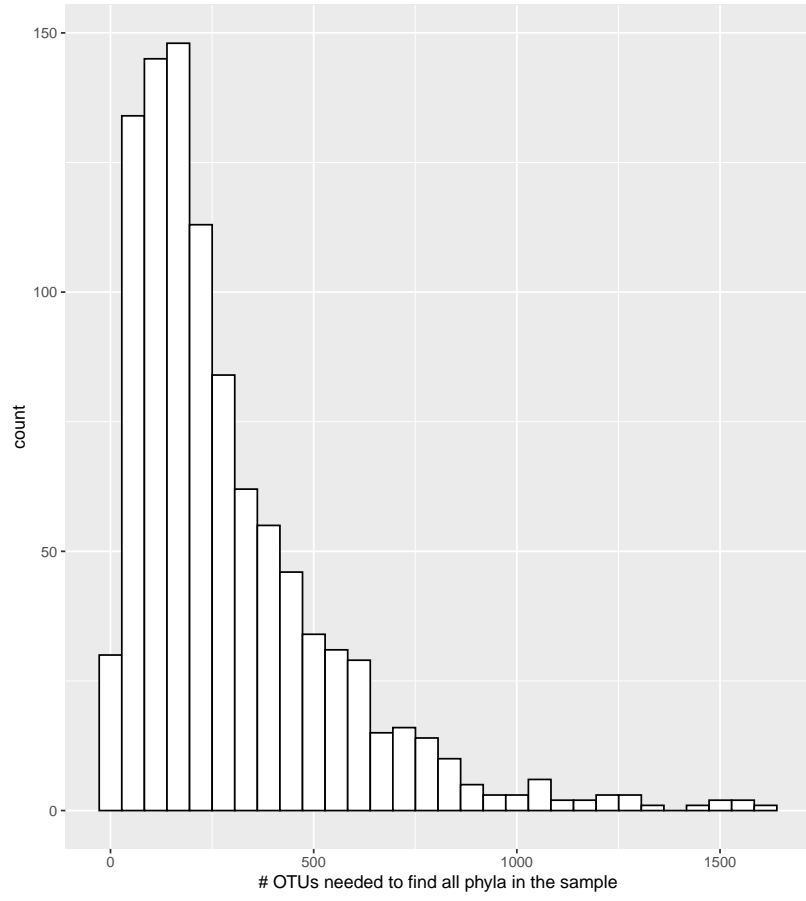
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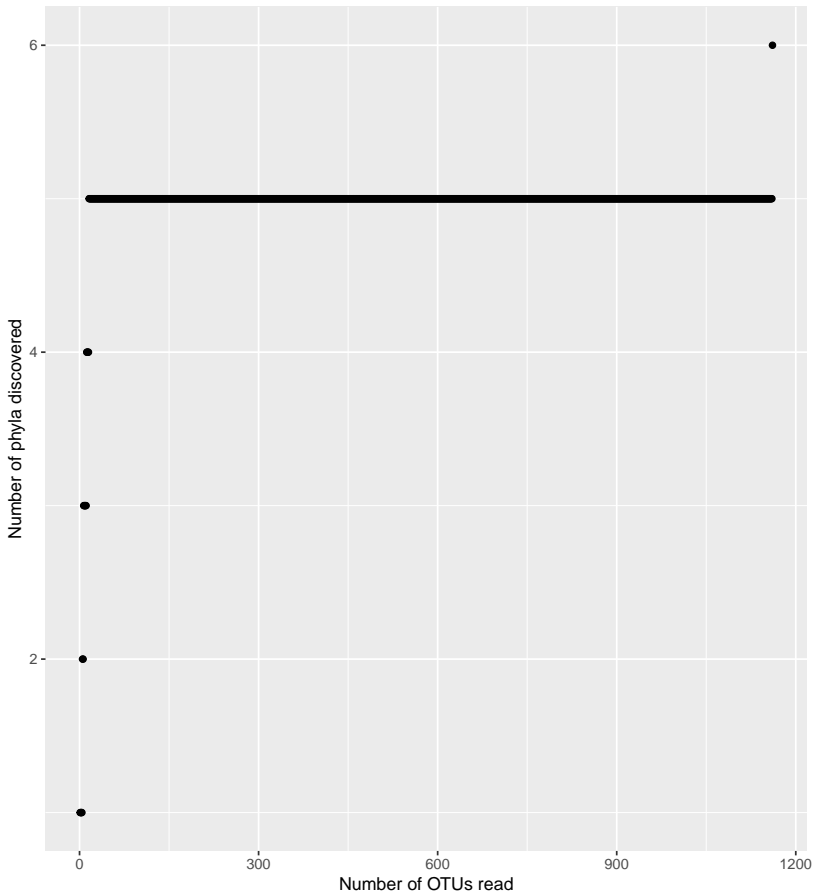
3. 18S.1.1.F.PCM.ME



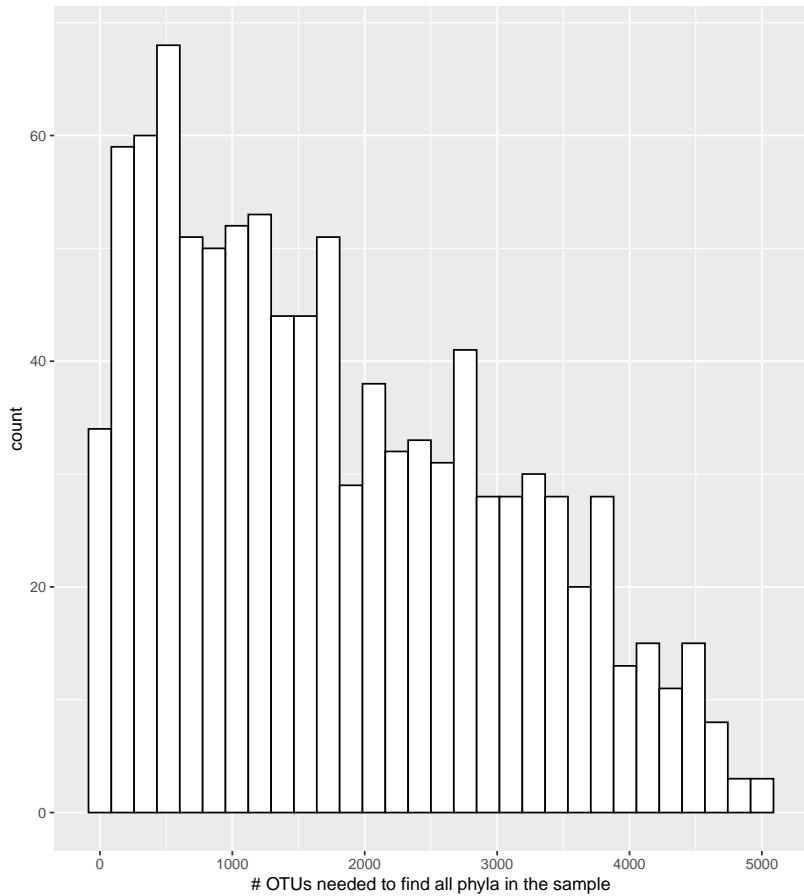
18S.1.1.F.PCM.ME



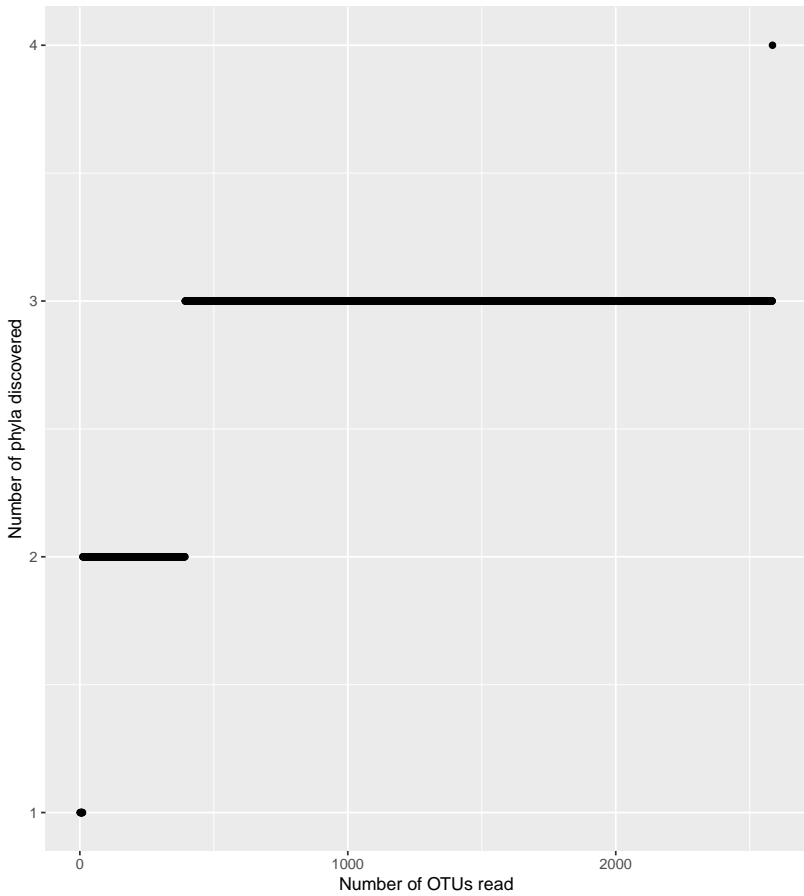
4. 18S.1.1.G.PCM.ME



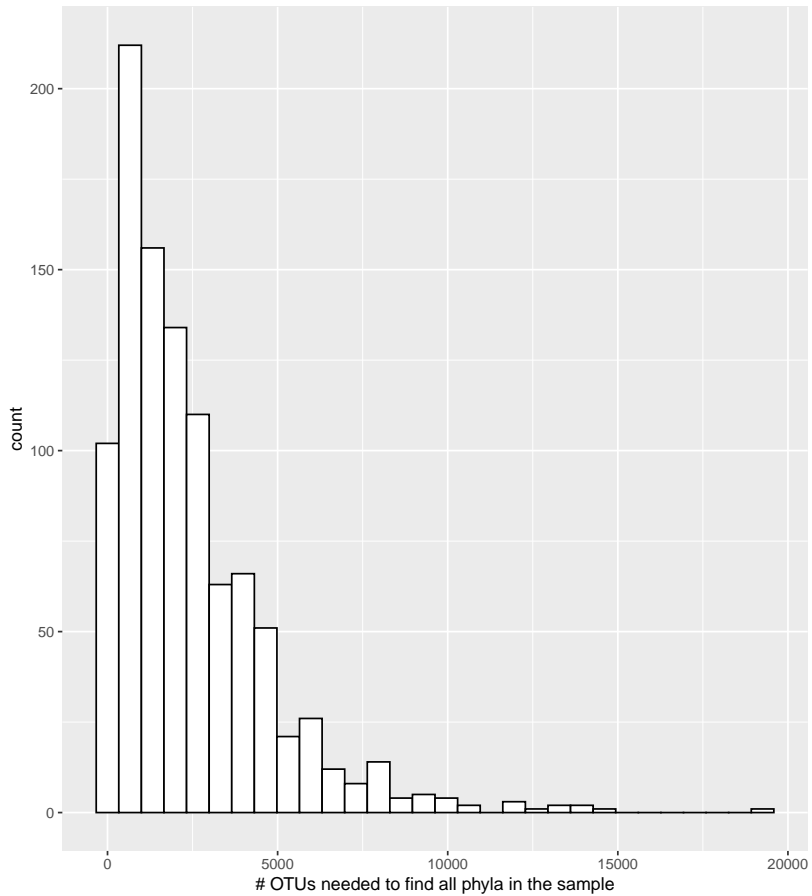
18S.1.1.G.PCM.ME



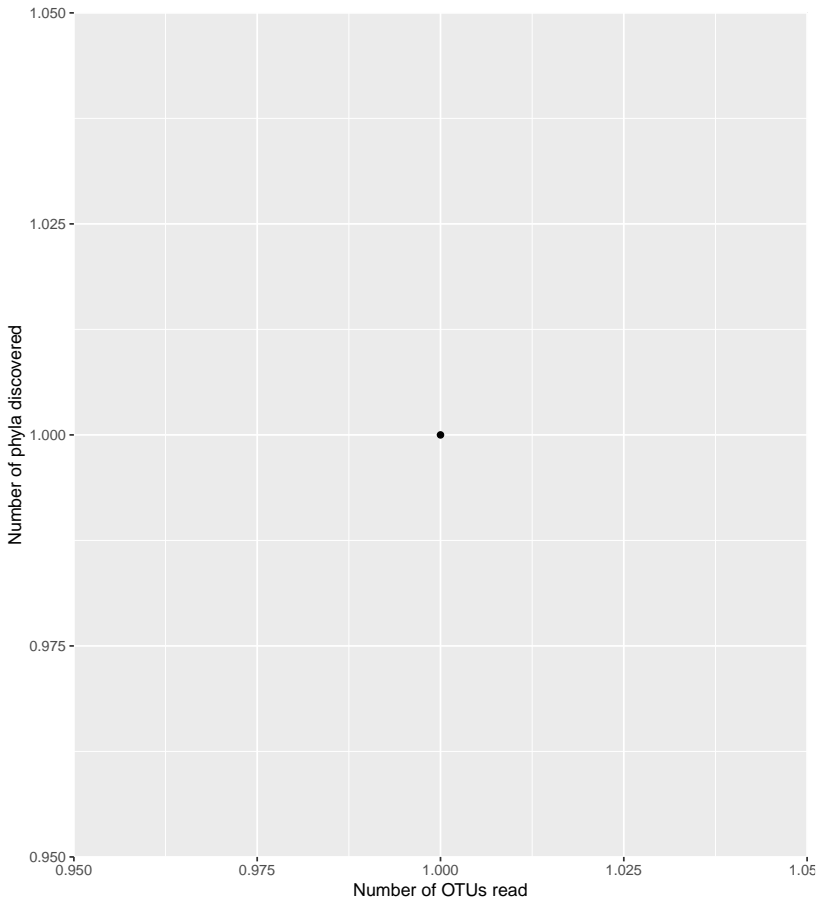
5. 18S.1.1.H.PCM.ME



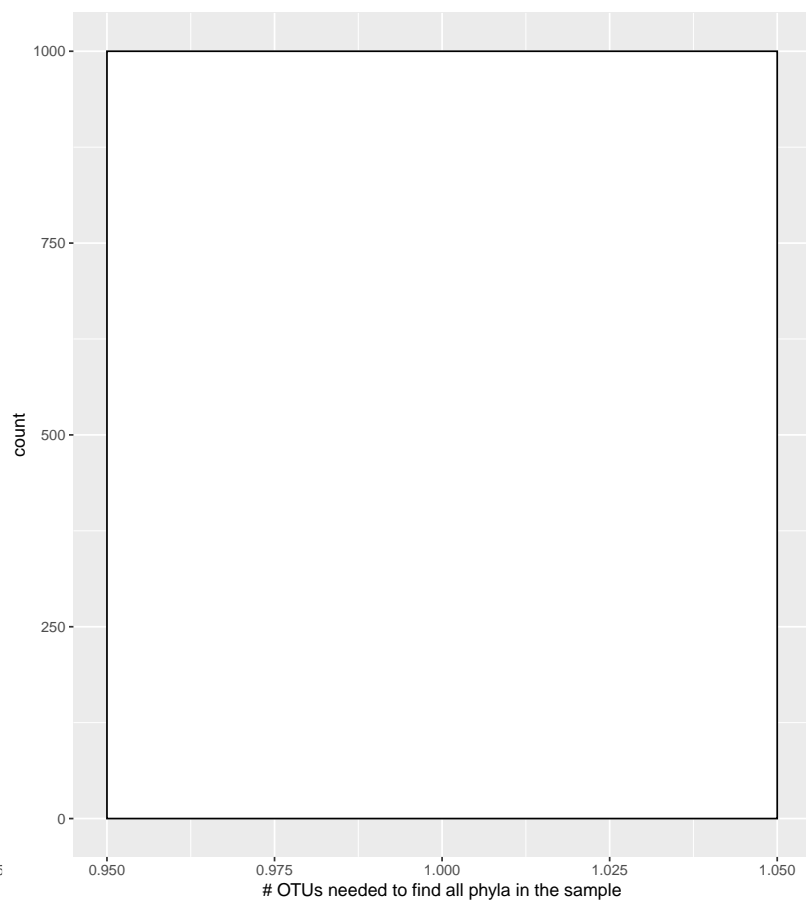
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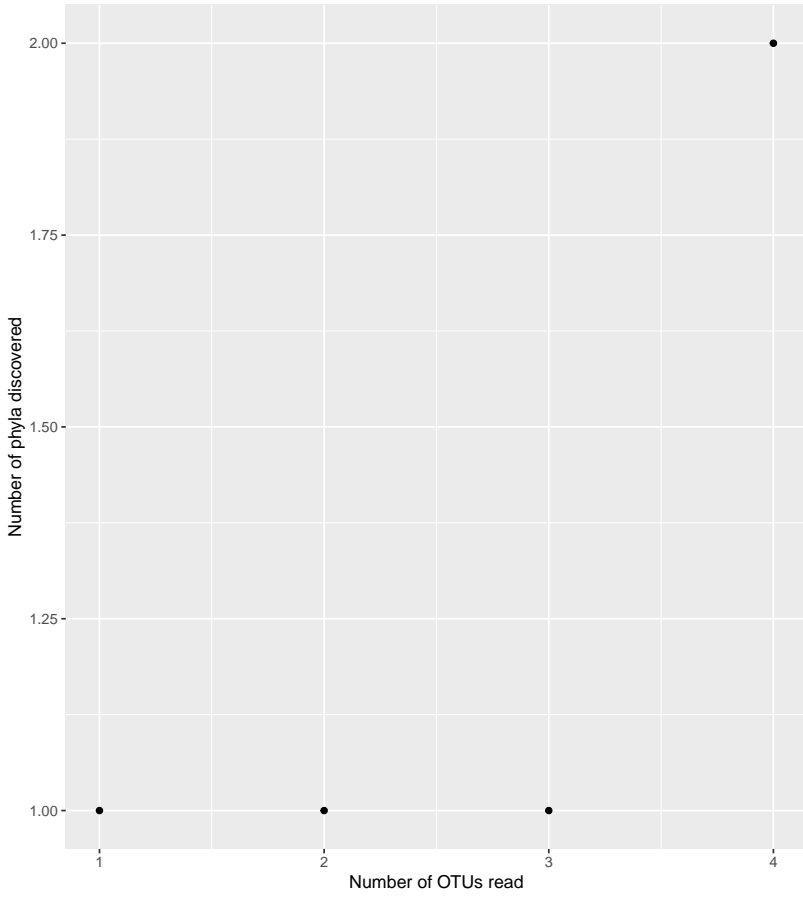
6. 18S.1.10.A.PCM.MM



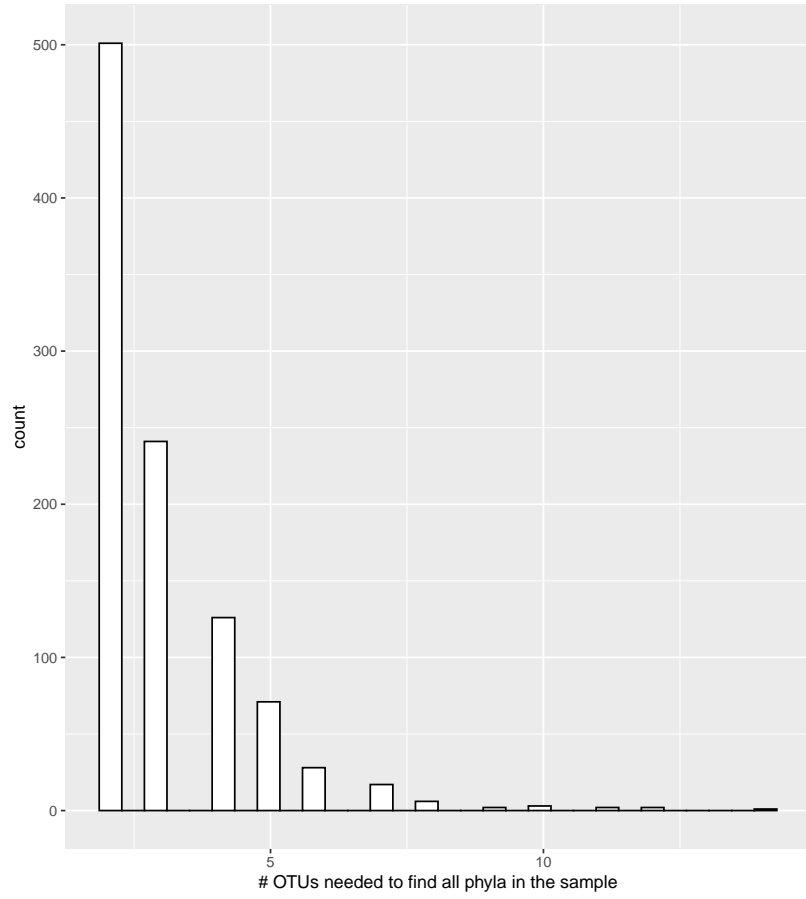
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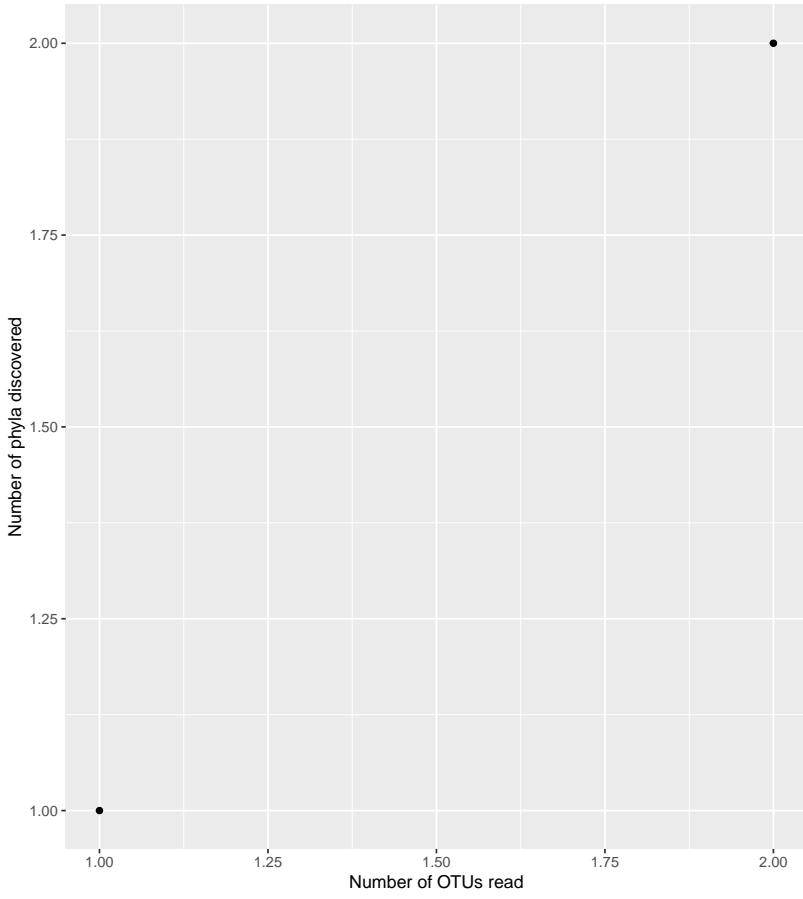
7. 18S.1.10.B.PCM.MM



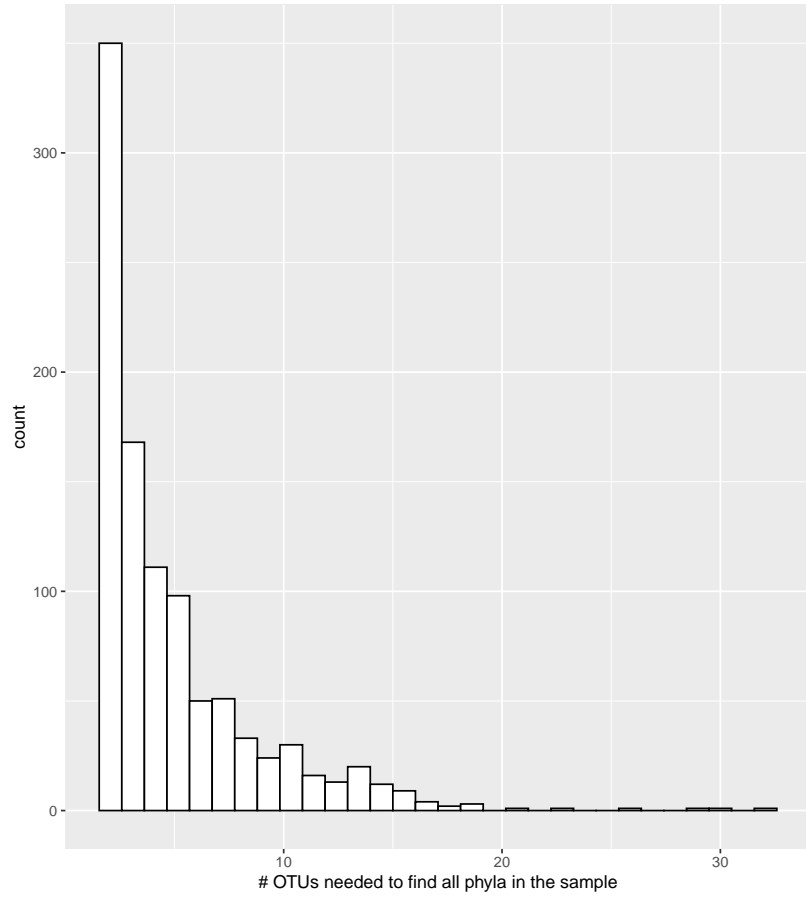
18S.1.10.B.PCM.MM



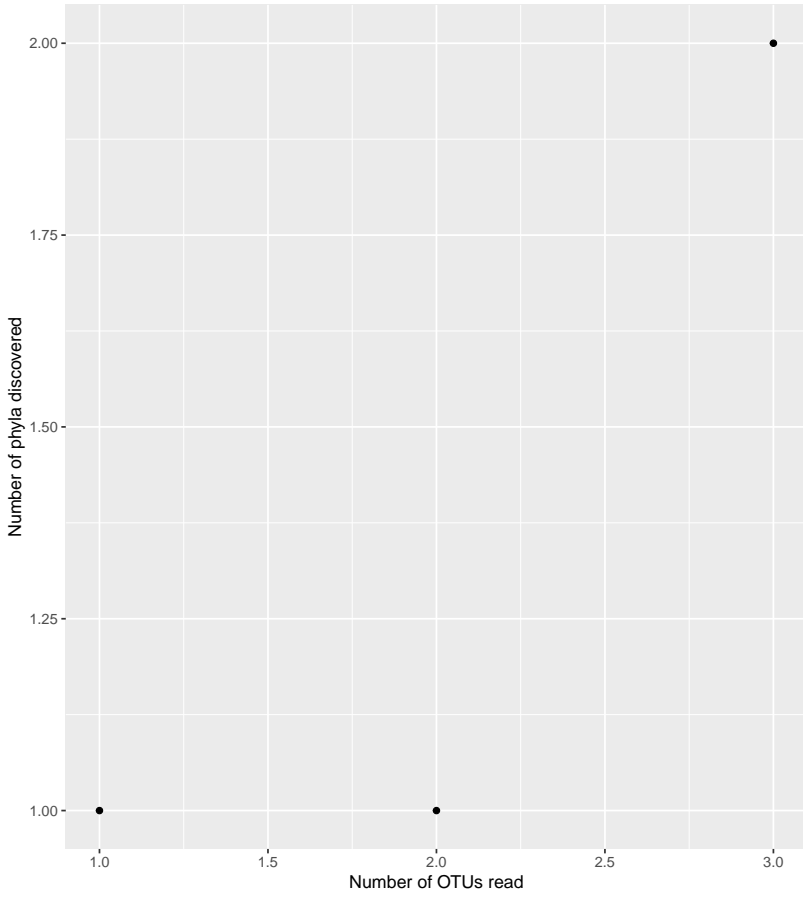
8. 18S.1.10.C.PCM.MM



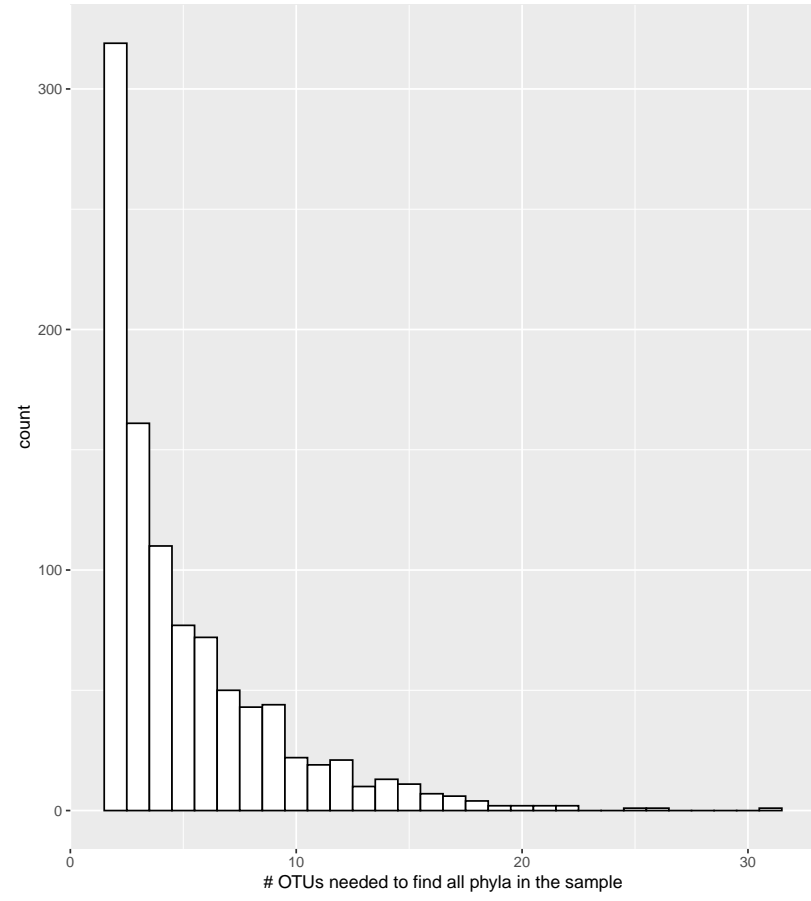
18S.1.10.C.PCM.MM



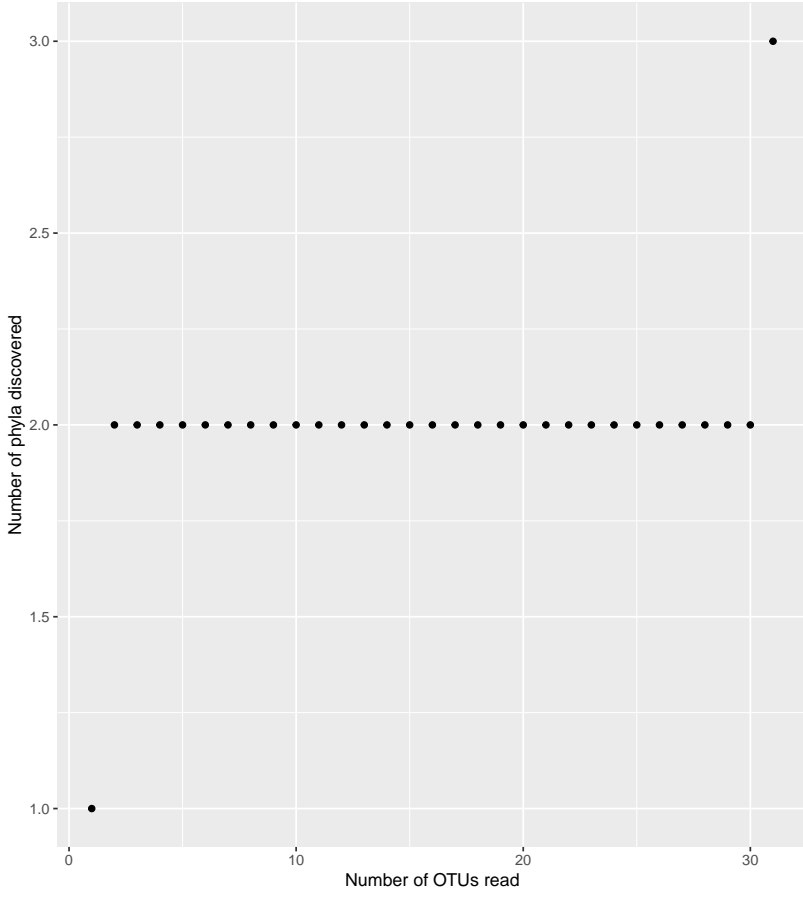
9. 18S.1.10.D.PCM.ME



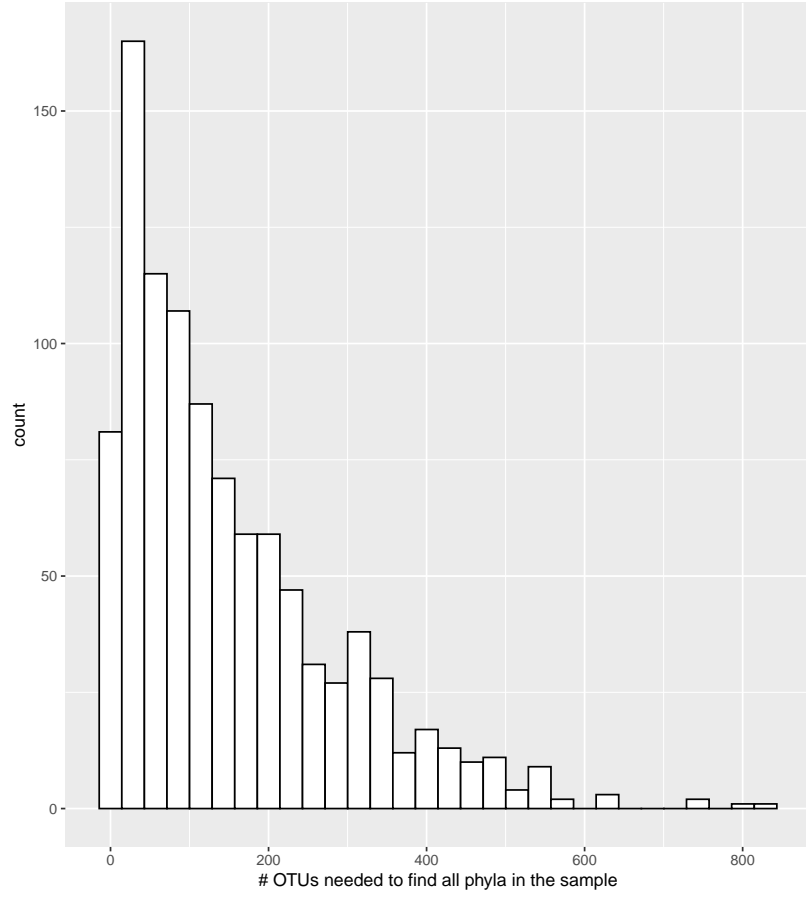
18S.1.10.D.PCM.ME



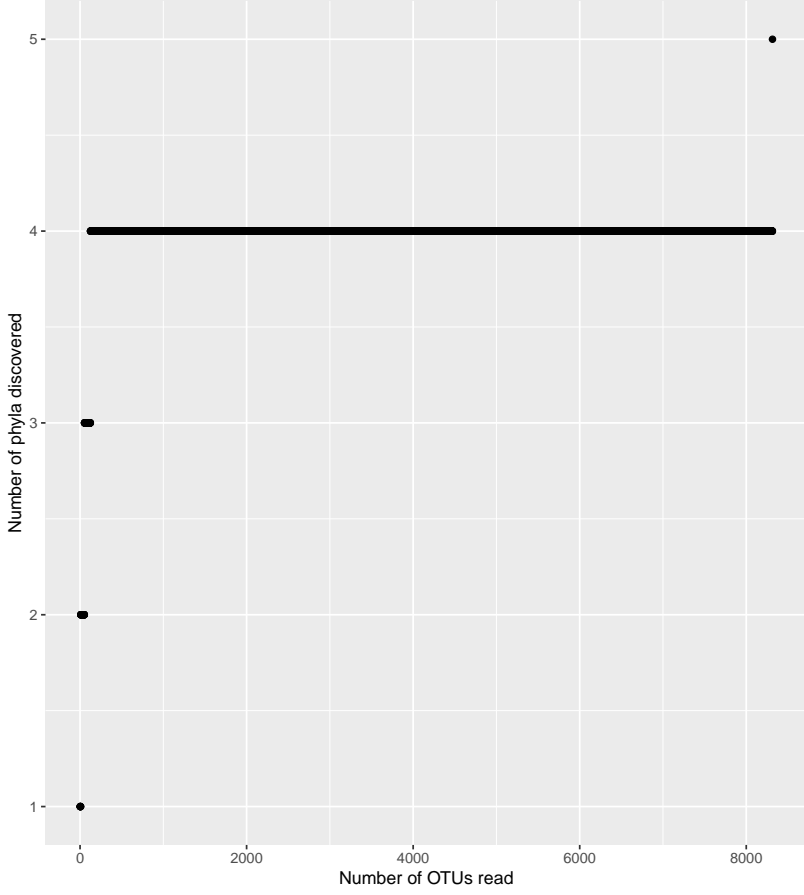
10. 18S.1.10.E.PCM.ME



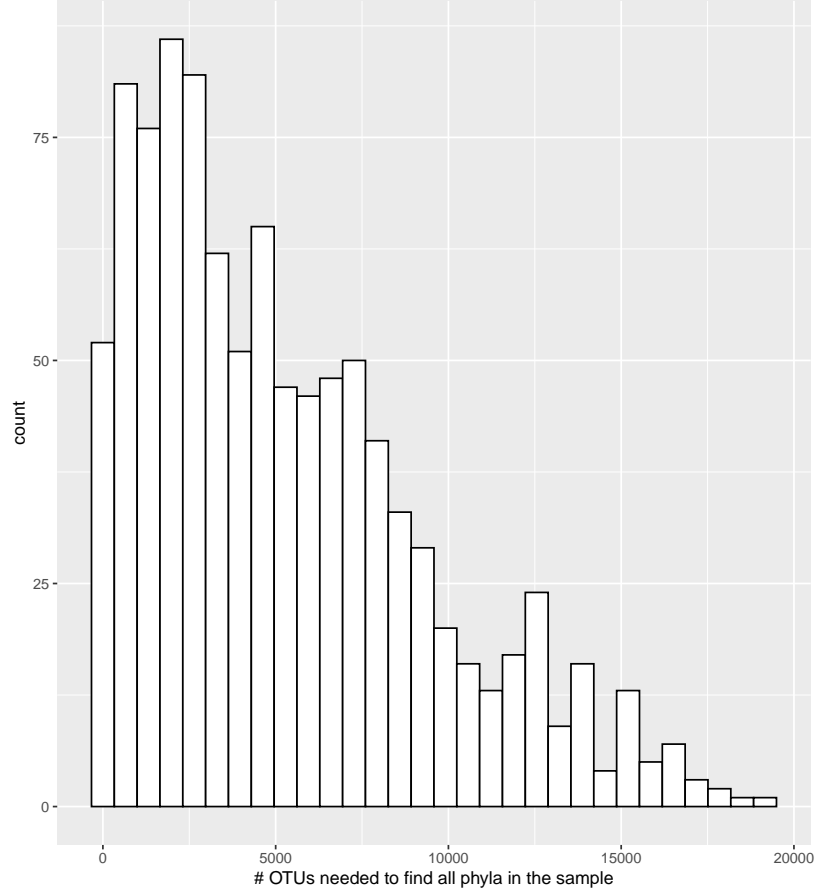
18S.1.10.E.PCM.ME



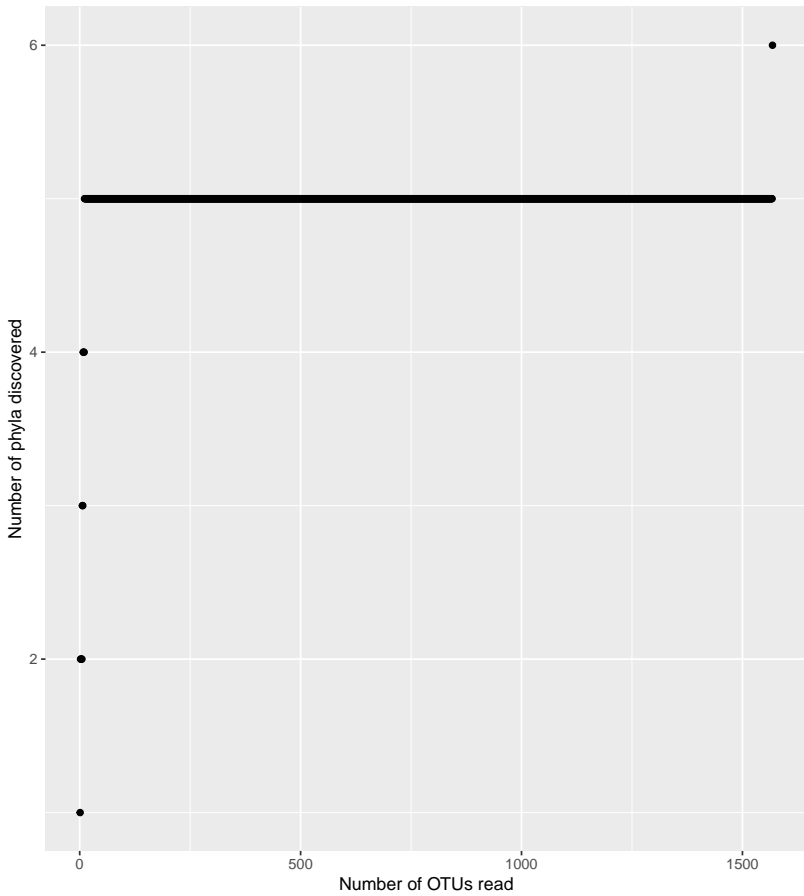
11. 18S.1.10.F.PCM.ME



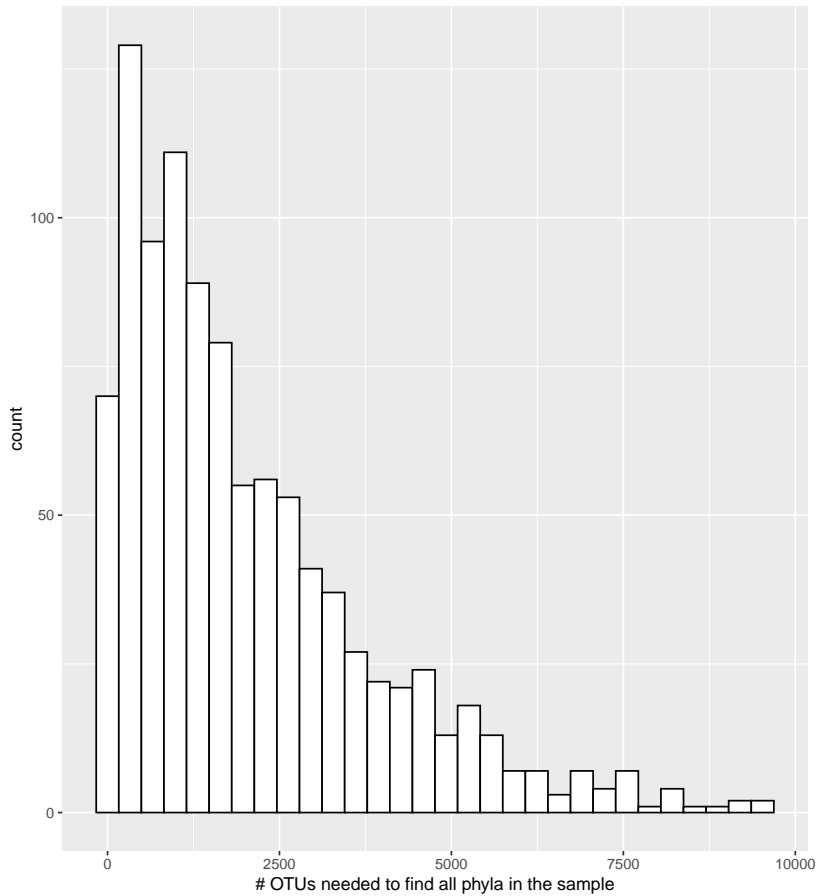
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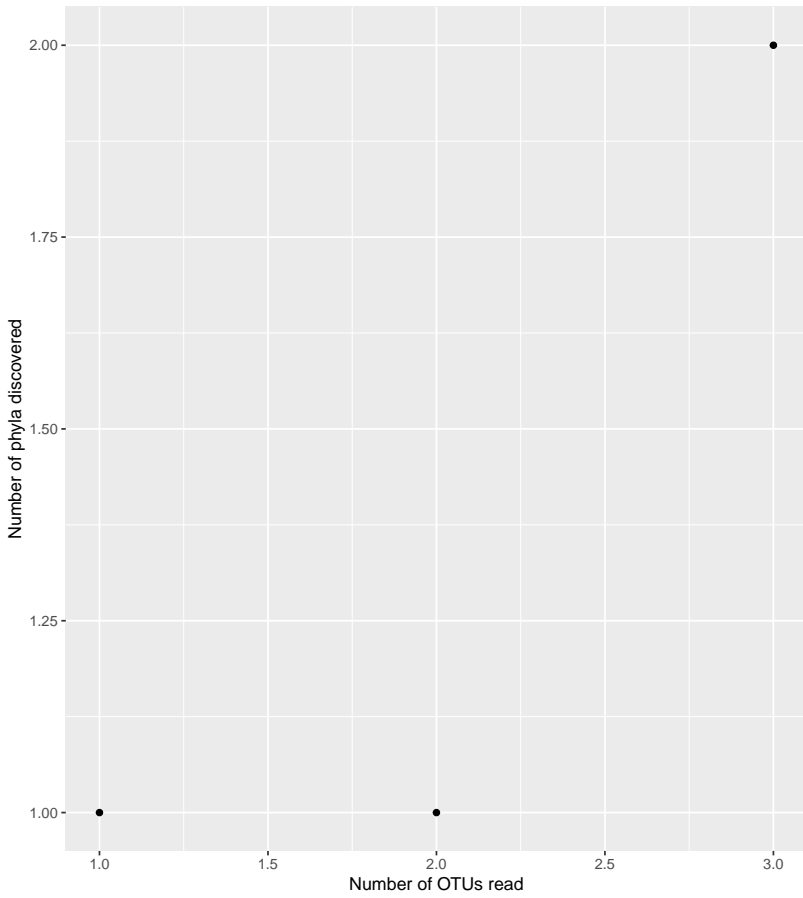
12. 18S.1.10.G.PCM.ME



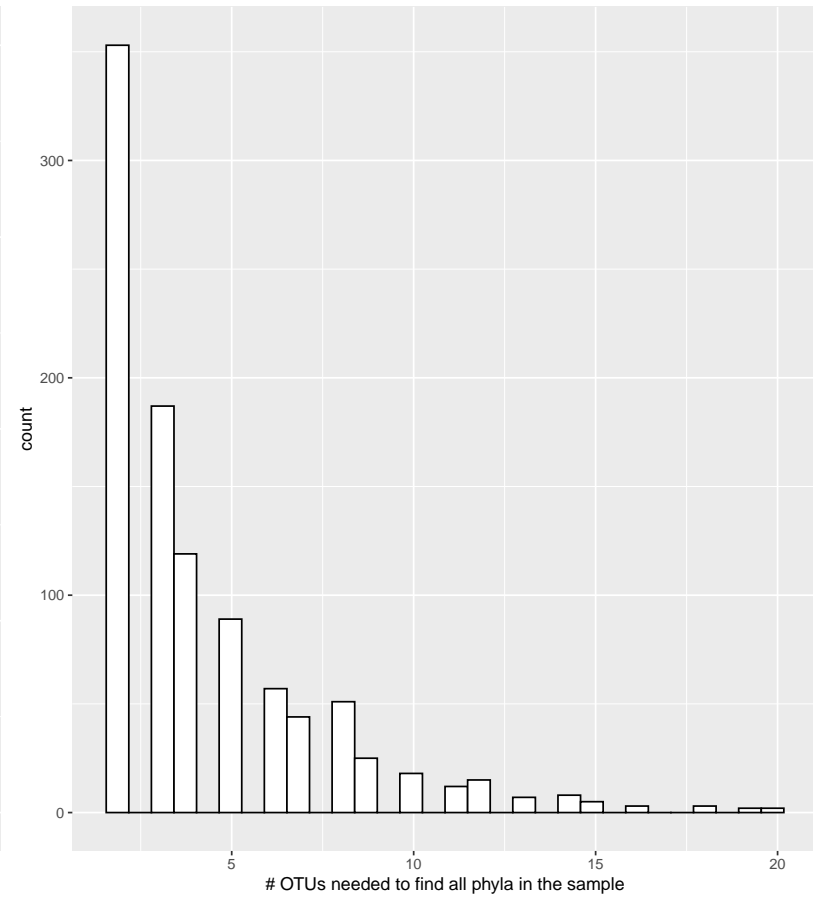
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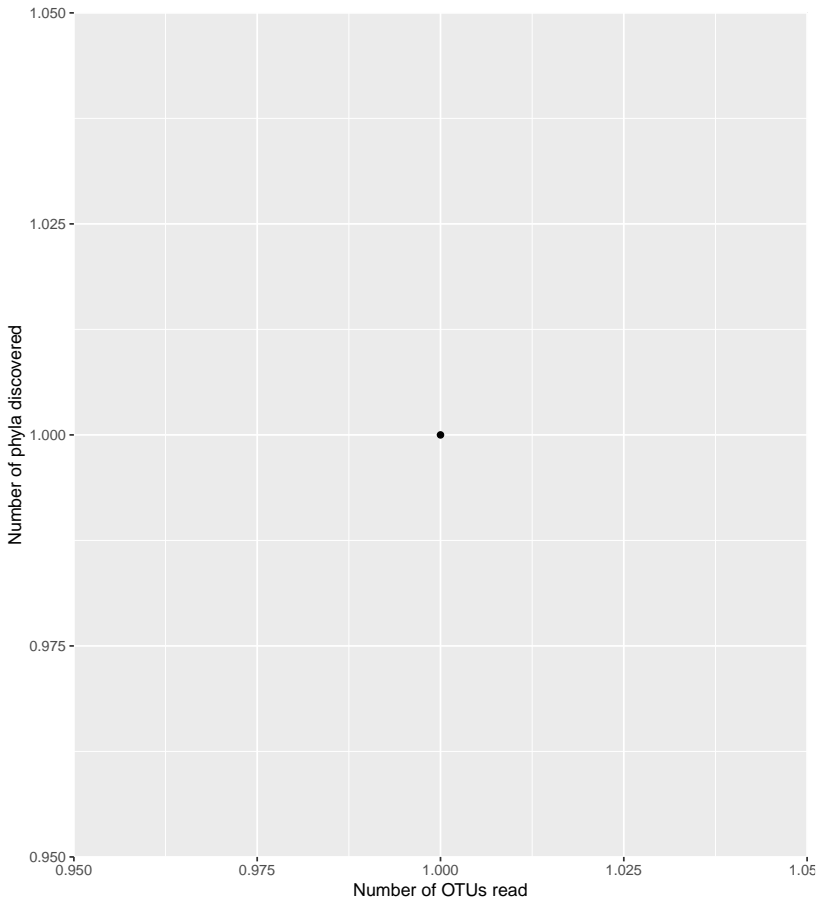
13. 18S.1.11.A.PCM.MM



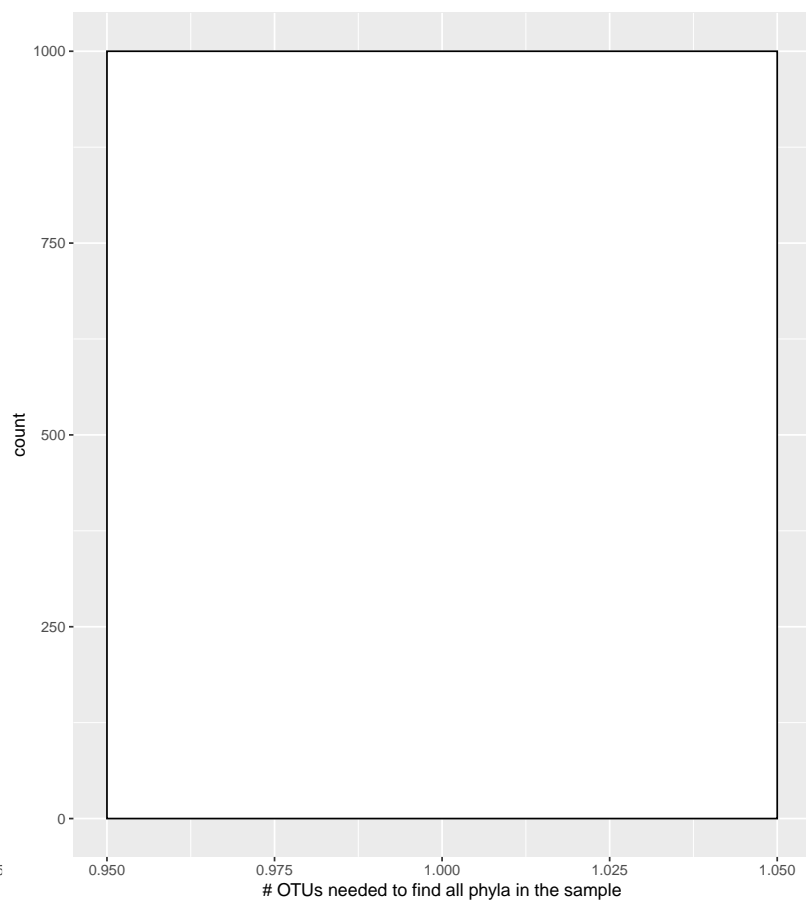
18S.1.11.A.PCM.MM



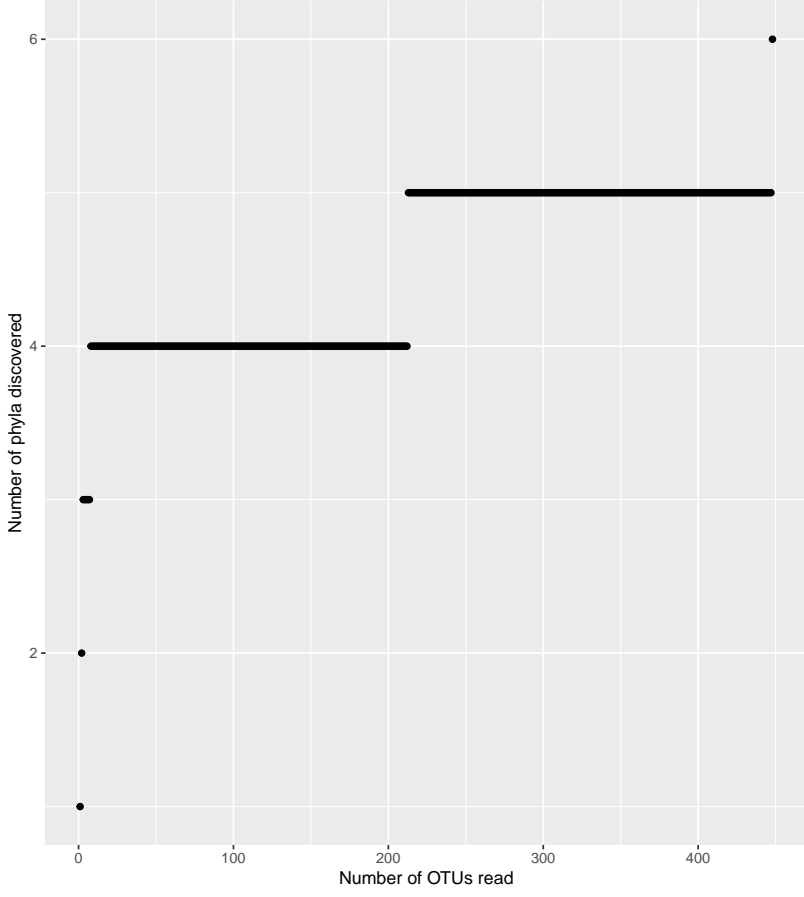
14. 18S.1.11.B.PCM.MM



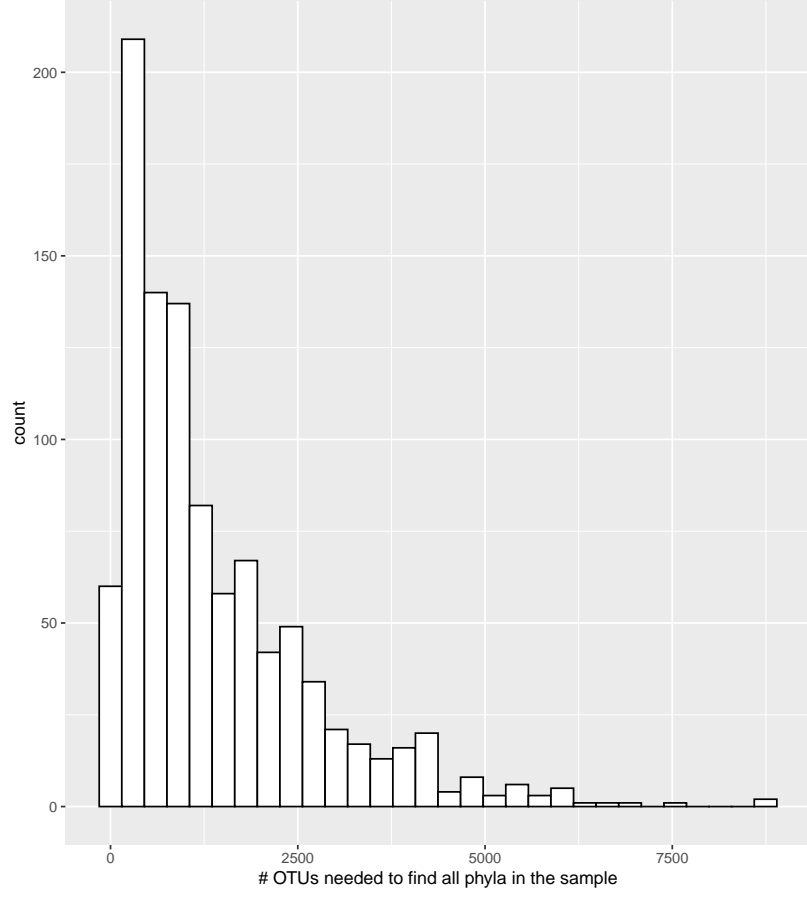
18S.1.11.B.PCM.MM



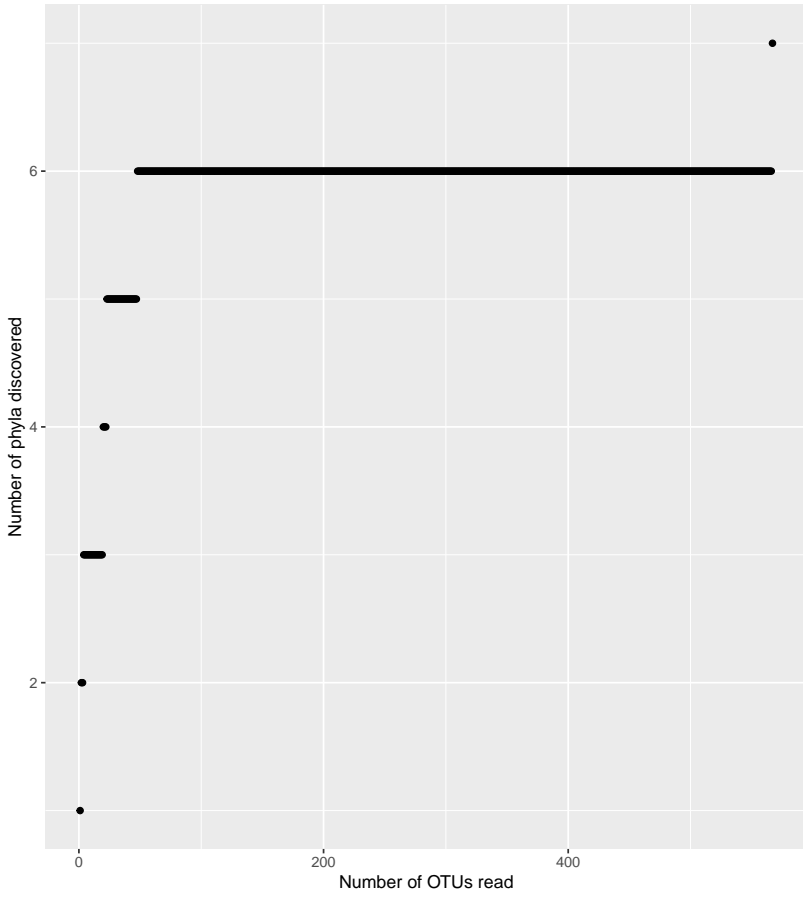
15. 18S.1.11.C.PCM.MM



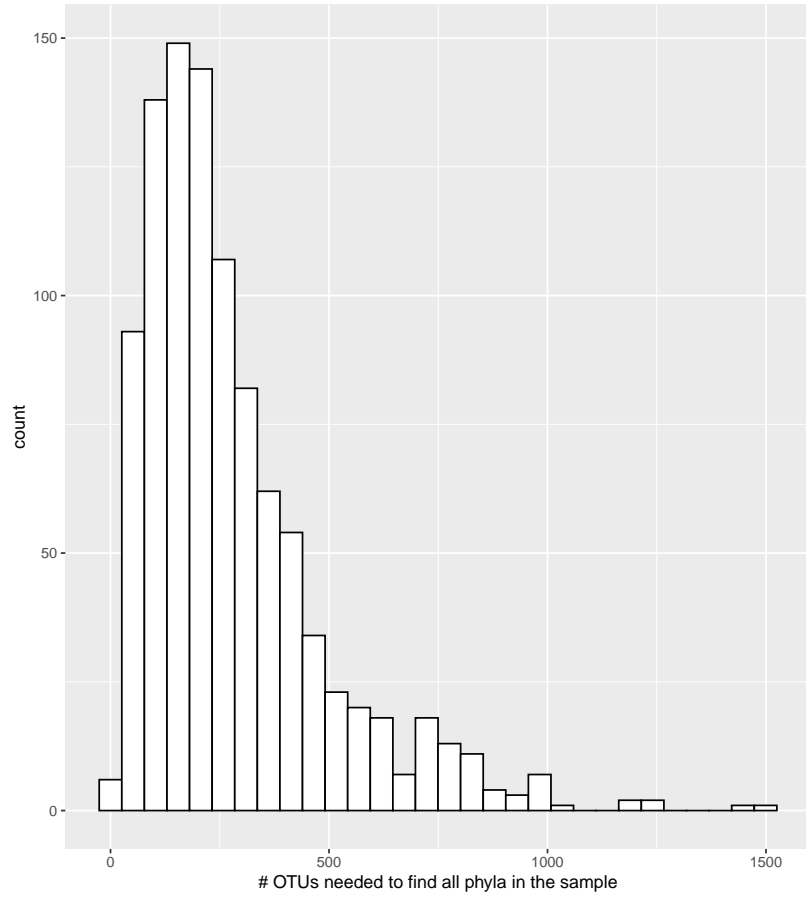
18S.1.11.C.PCM.MM



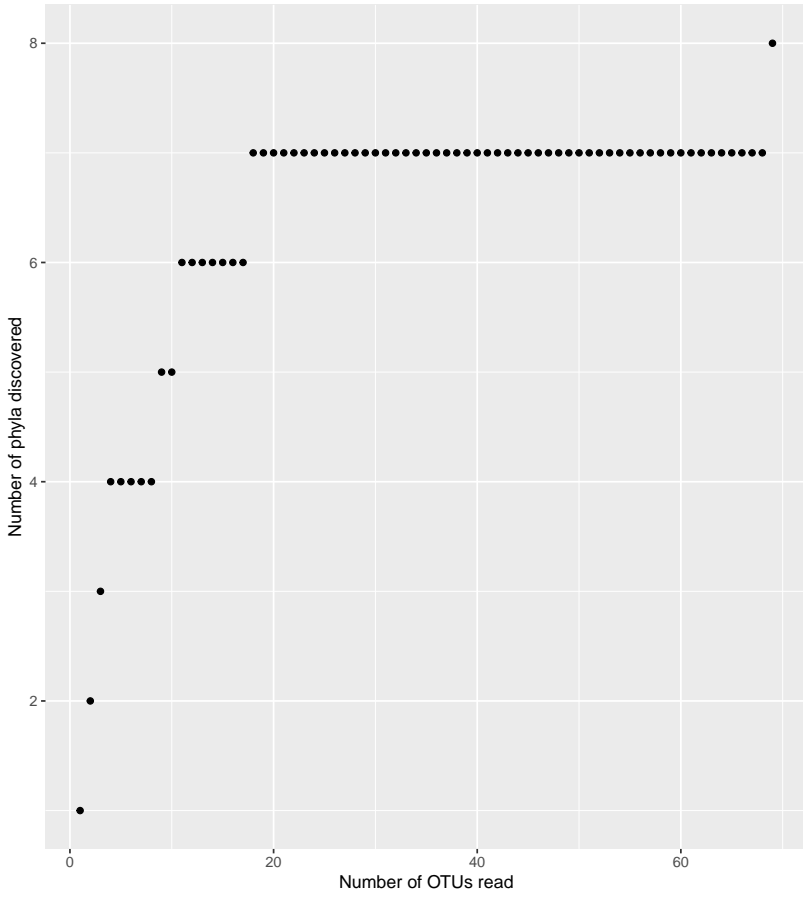
16. 18S.1.11.D.PCM.ME



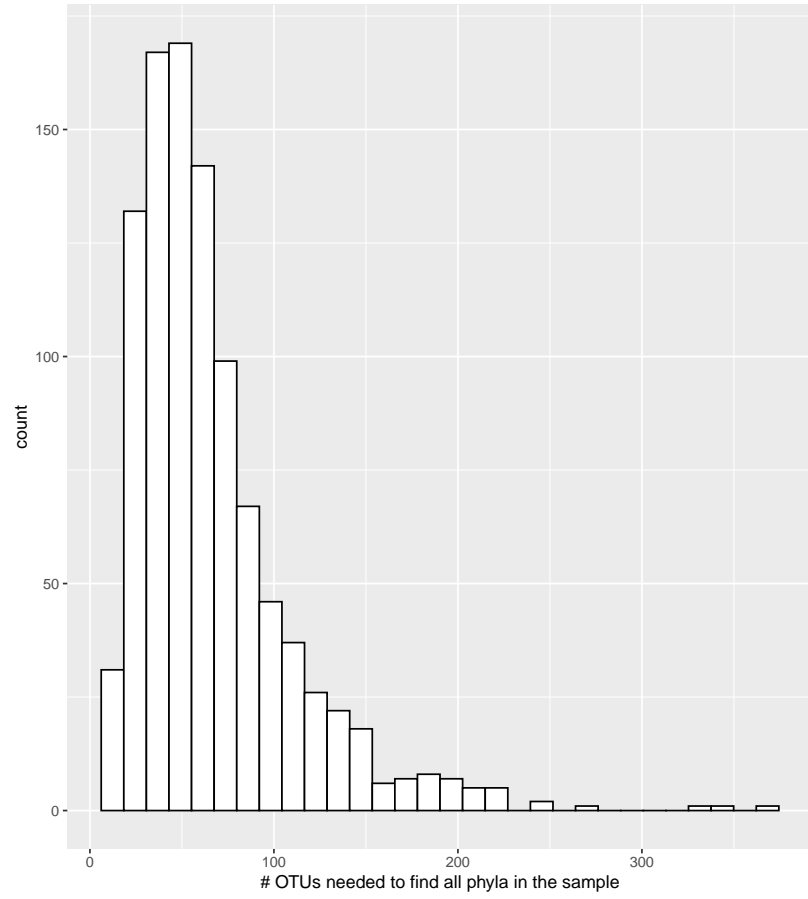
18S.1.11.D.PCM.ME



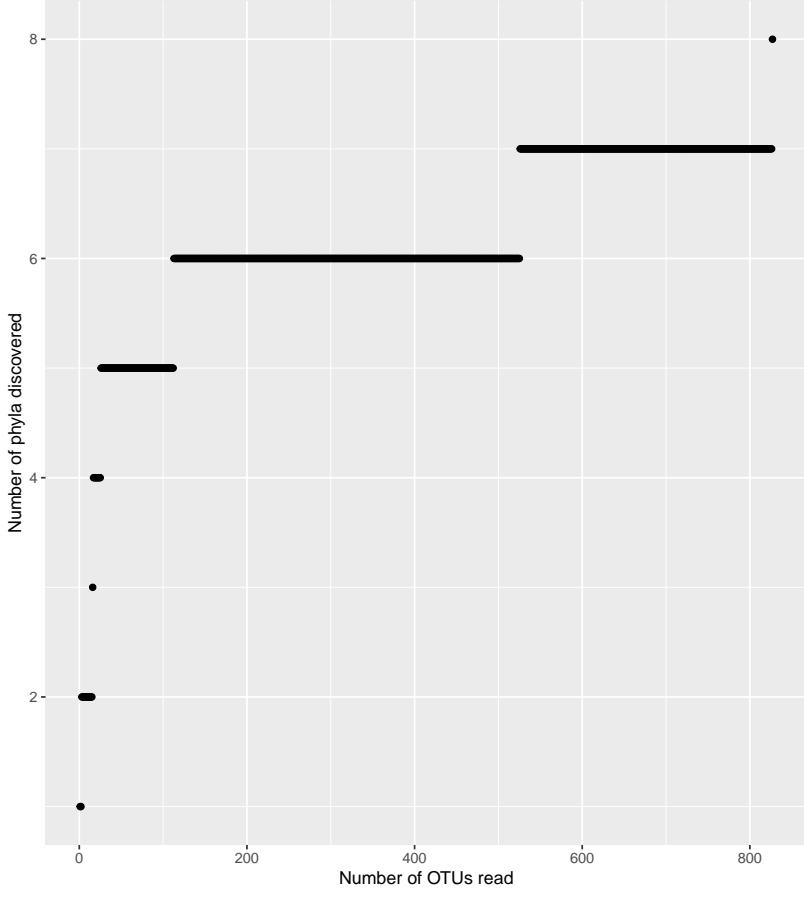
17. 18S.1.11.E.PCM.ME



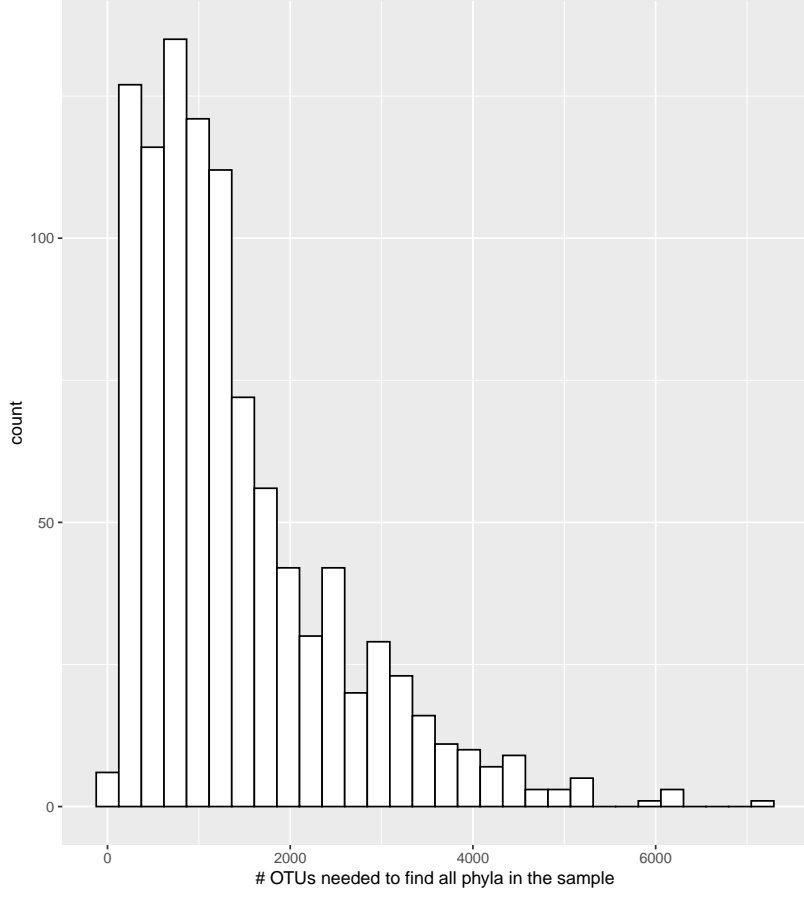
18S.1.11.E.PCM.ME



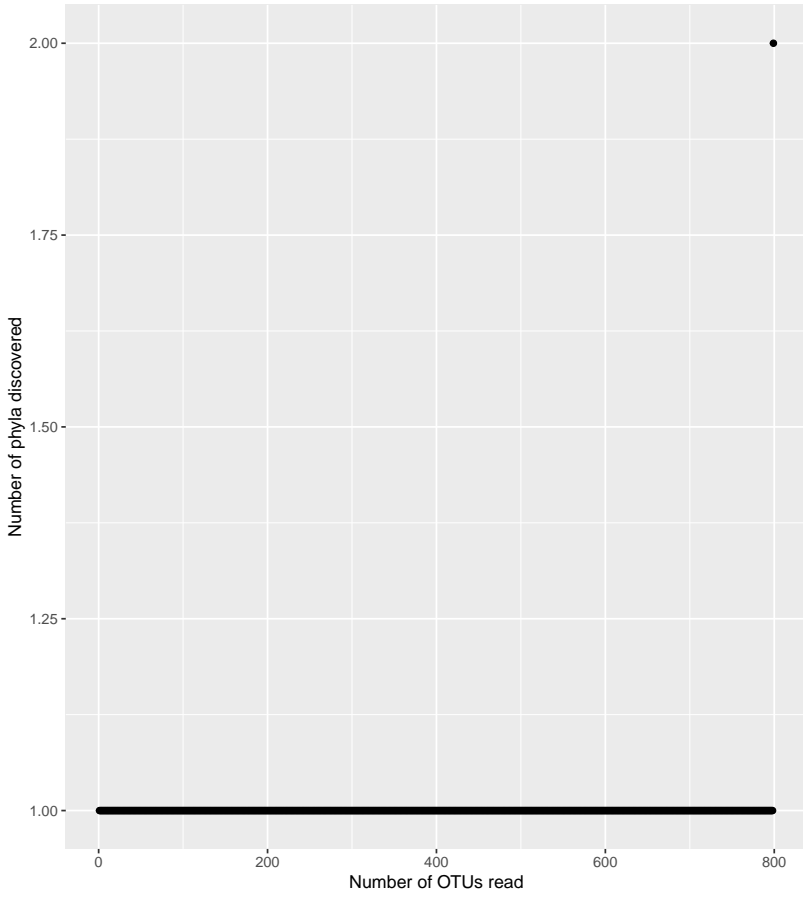
18. 18S.1.11.G.PCM.ME



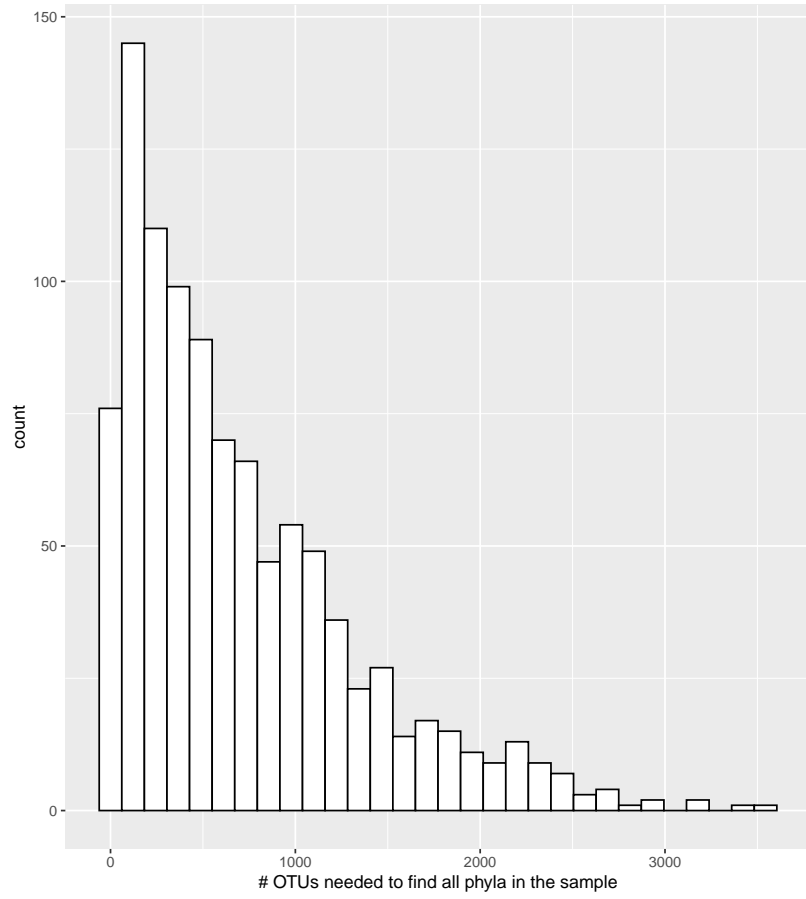
18S.1.11.G.PCM.ME



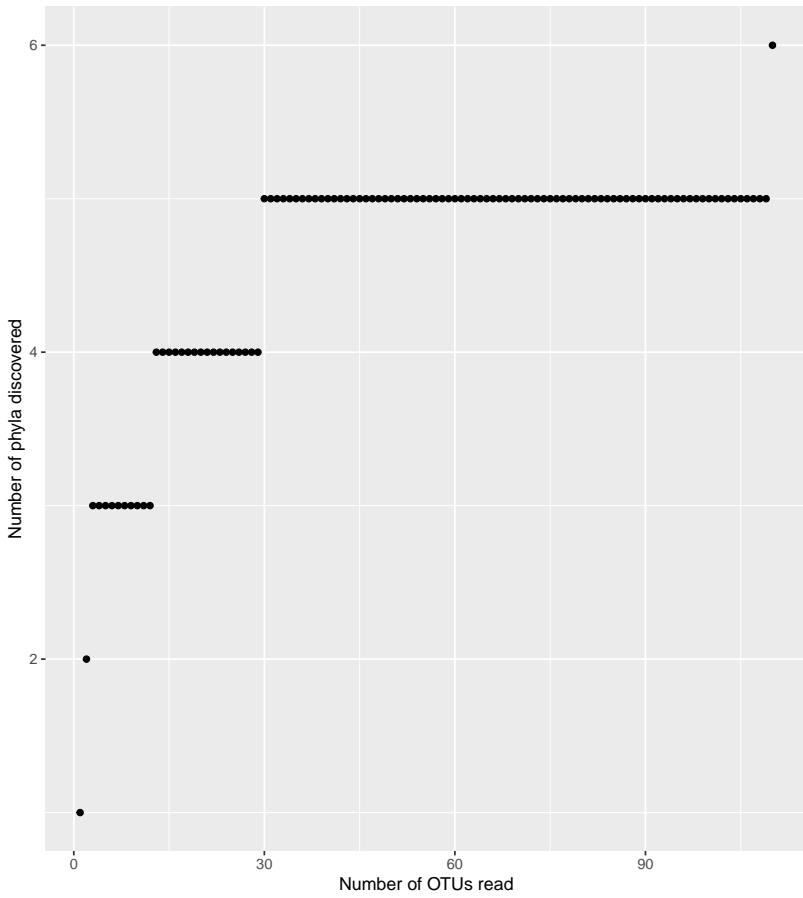
19. 18S.1.2.B.PCM.MM



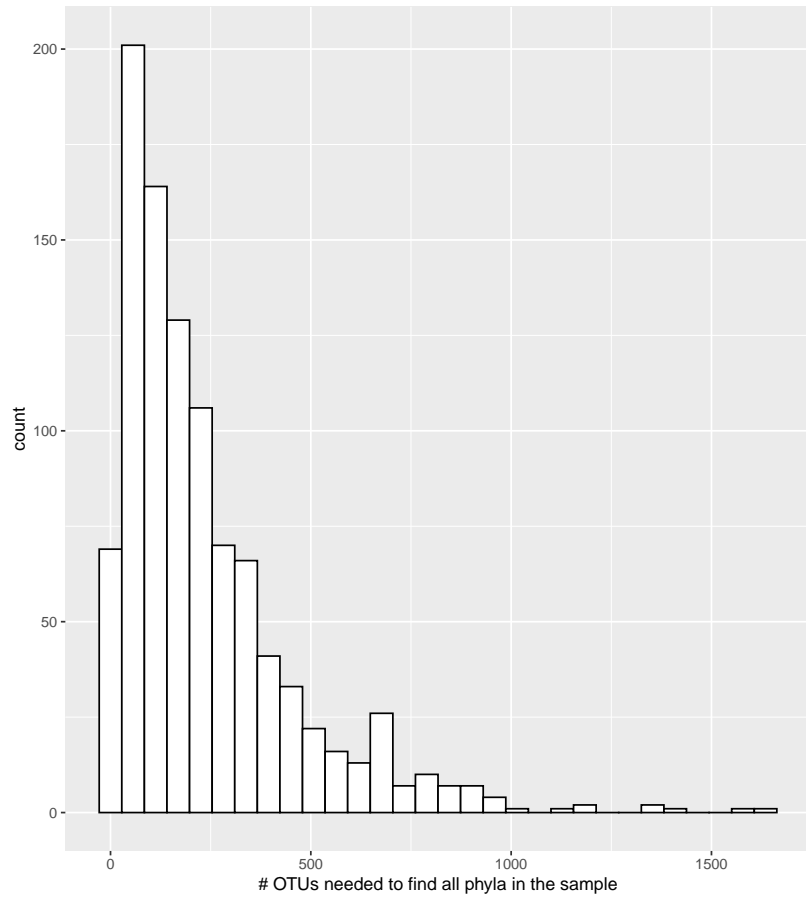
18S.1.2.B.PCM.MM



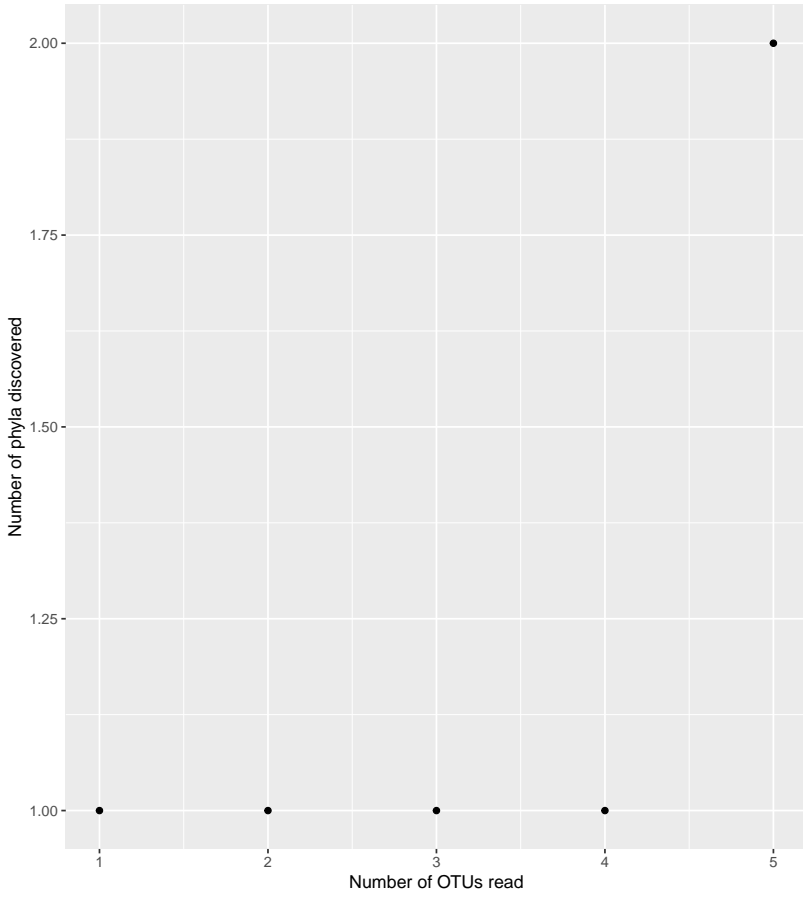
20. 18S.1.2.C.PCM.MM



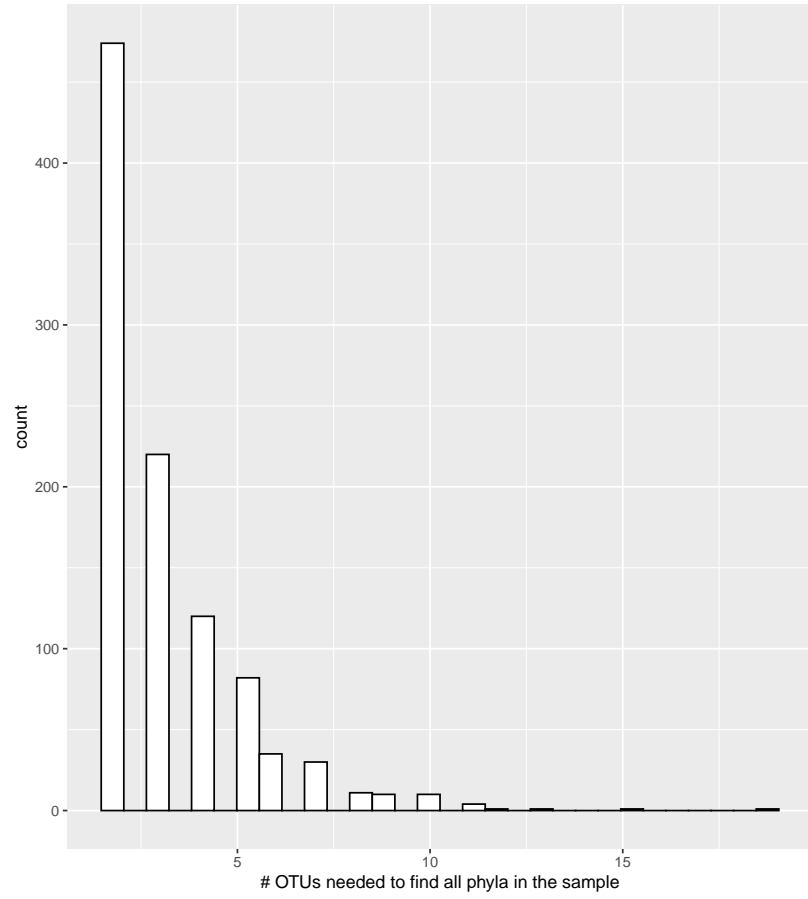
18S.1.2.C.PCM.MM



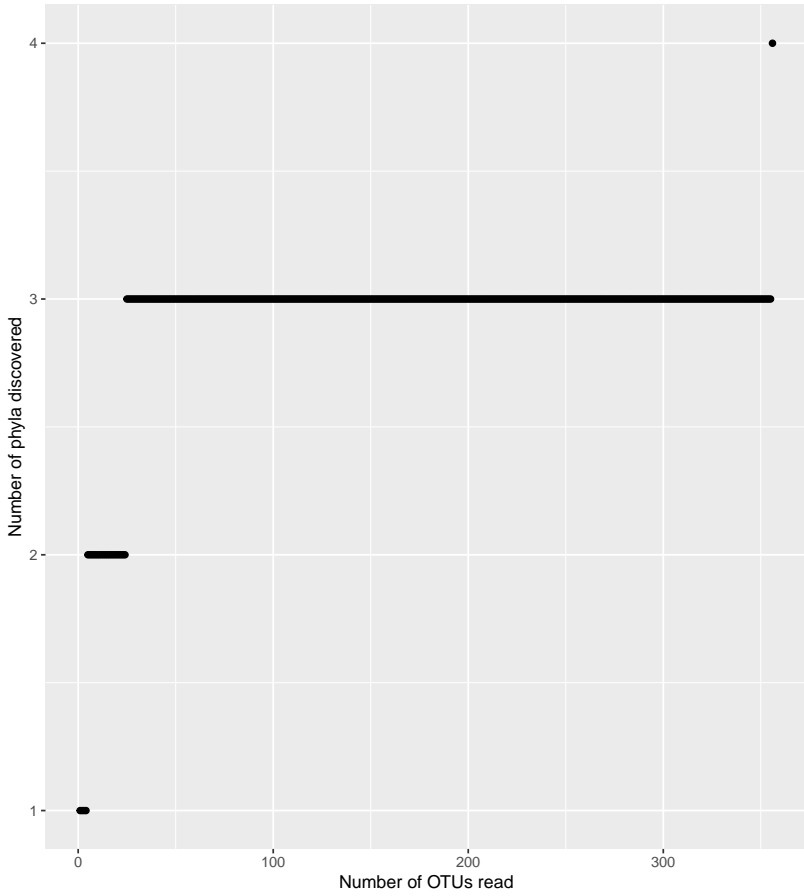
21. 18S.1.2.D.PCM.ME



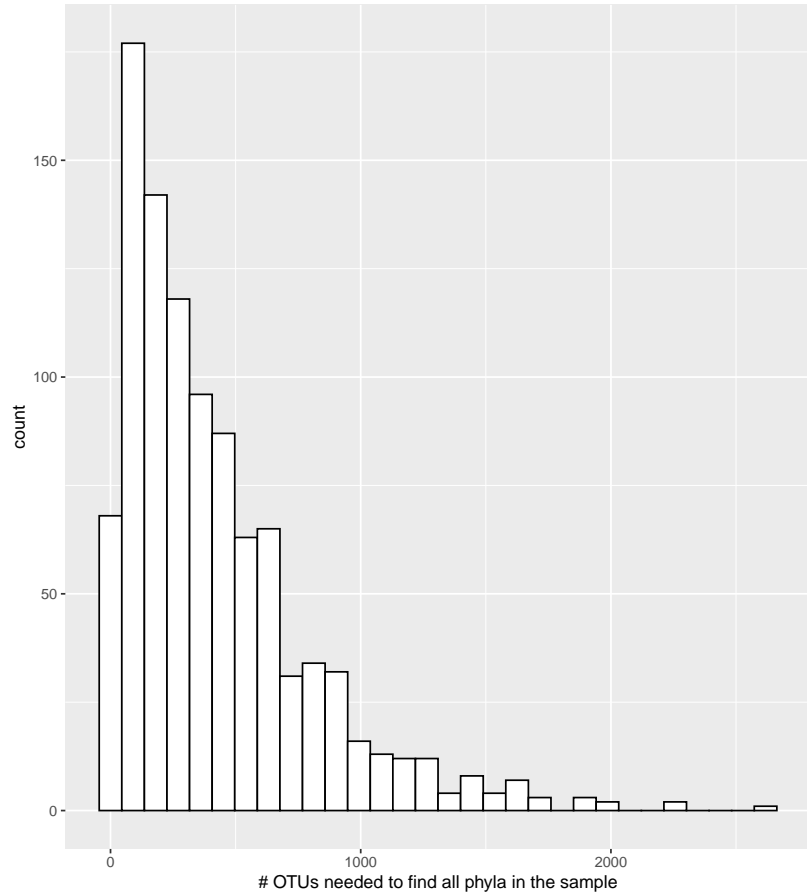
18S.1.2.D.PCM.ME



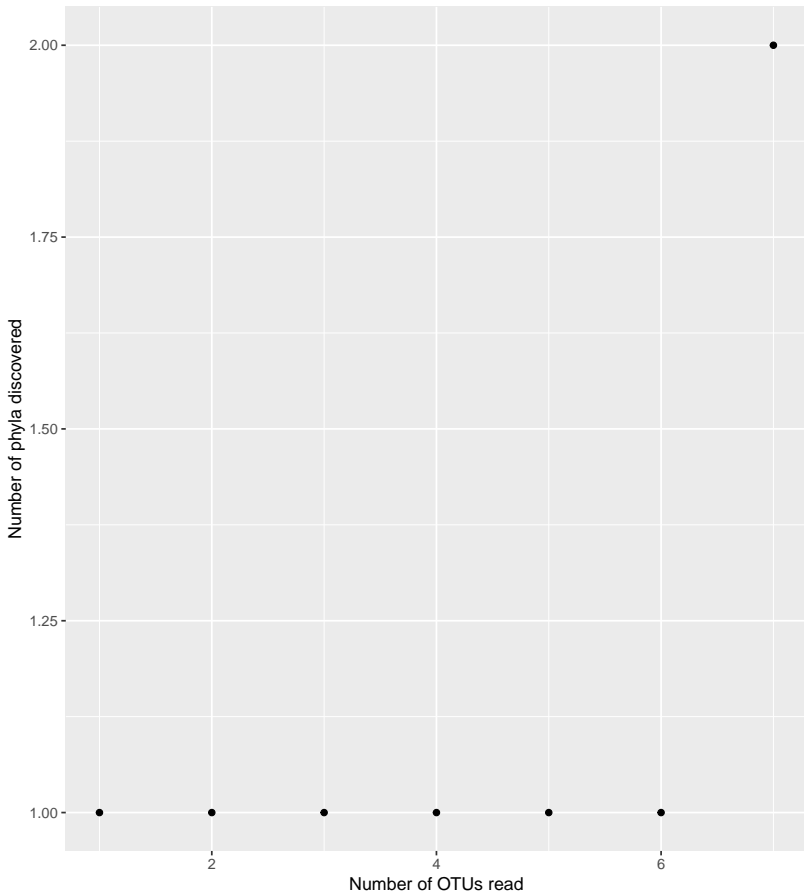
22. 18S.1.2.E.PCM.ME



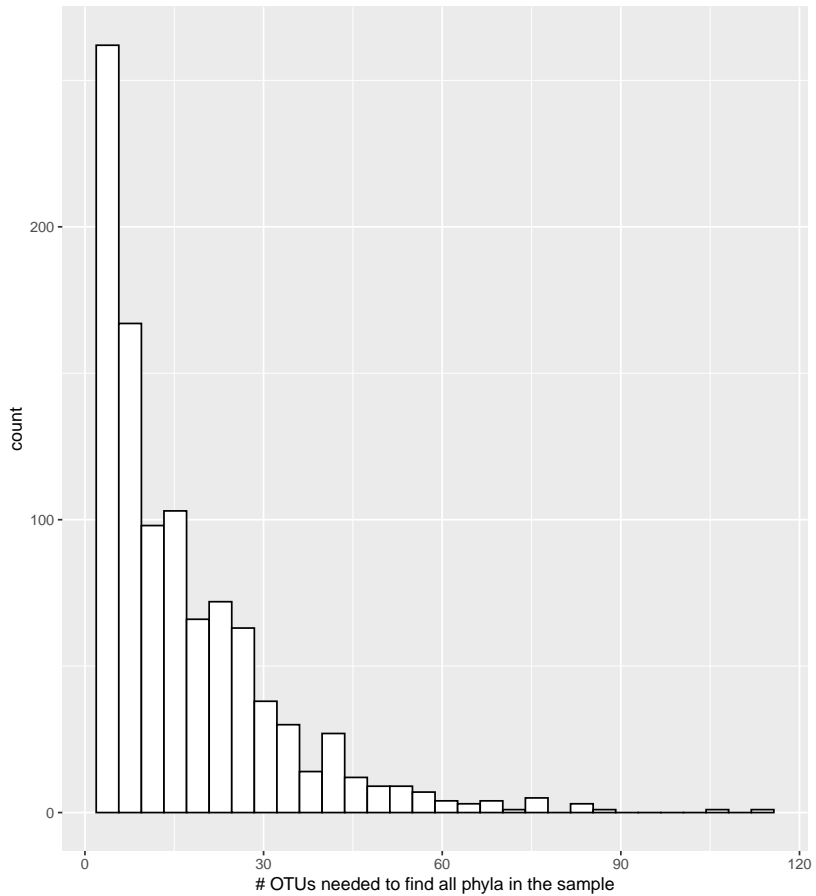
18S.1.2.E.PCM.ME



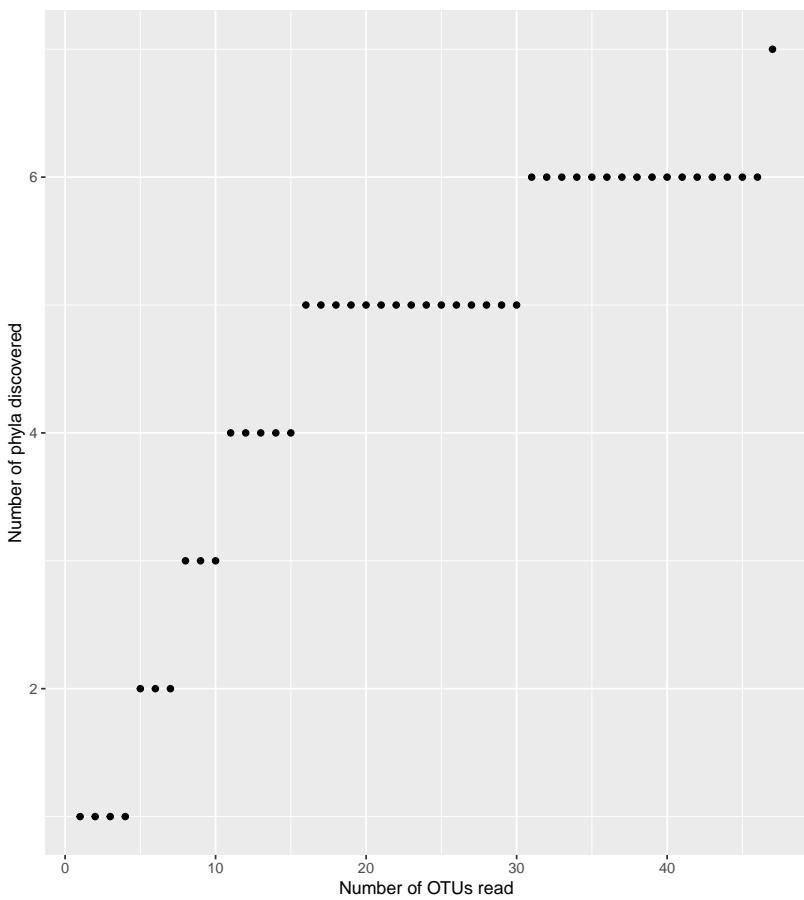
23. 18S.1.2.F.PCM.ME



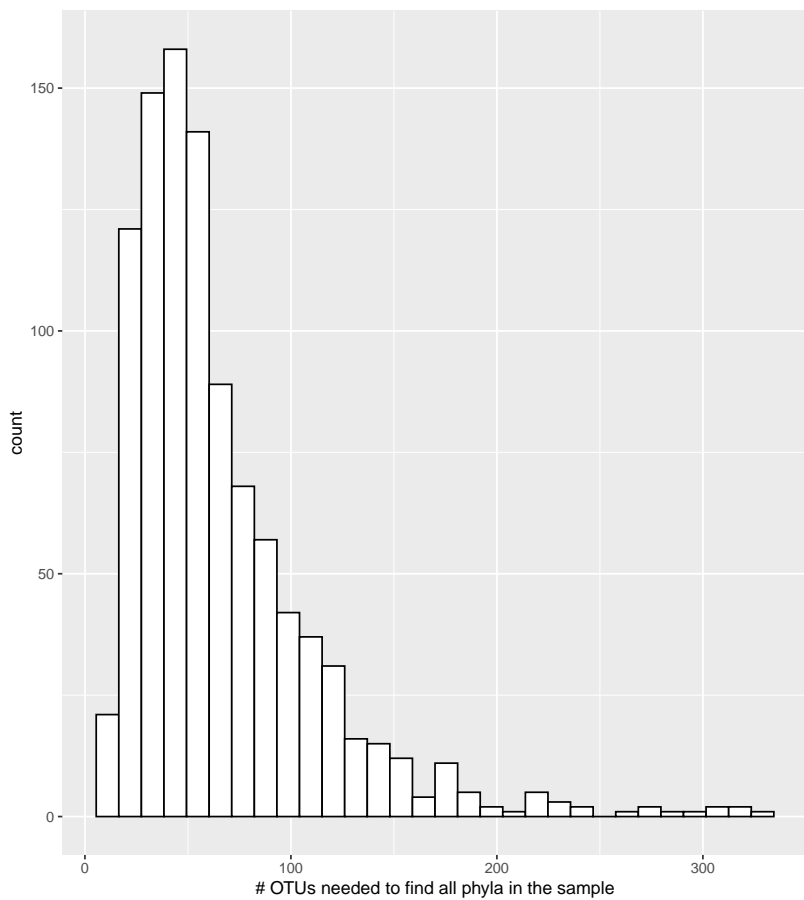
18S.1.2.F.PCM.ME



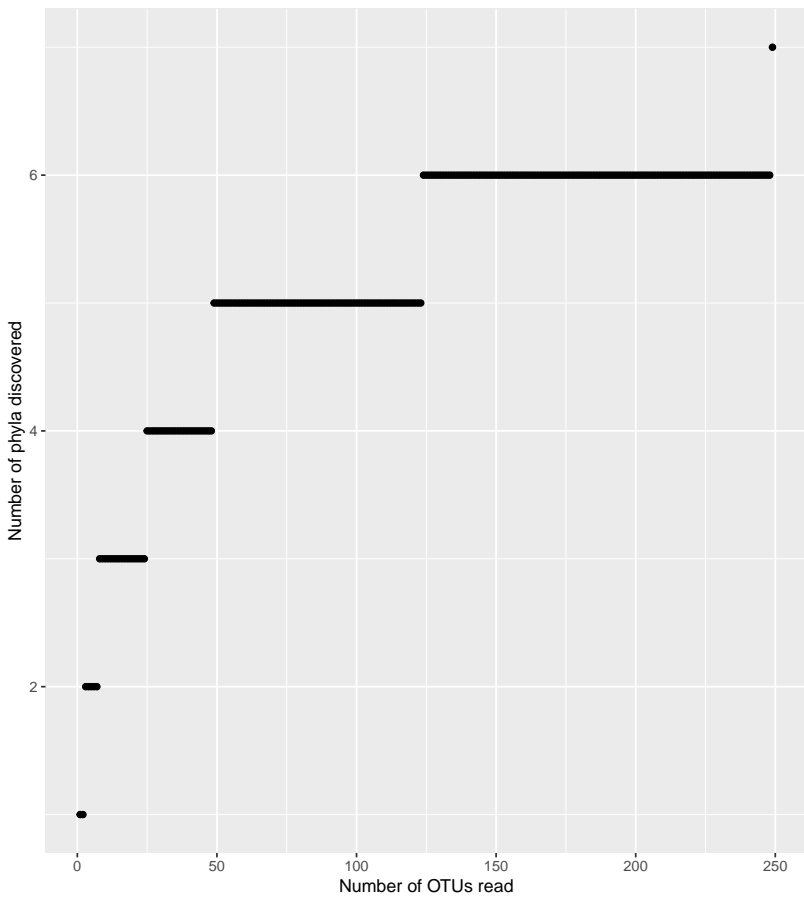
24. 18S.1.2.G.PCM.ME



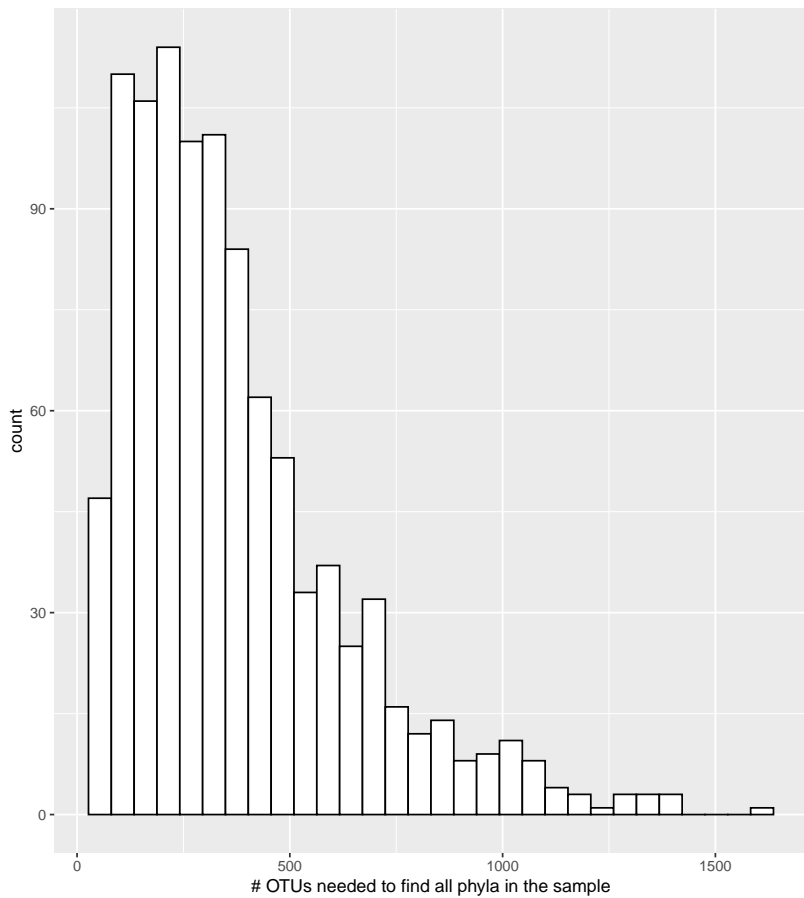
18S.1.2.G.PCM.ME



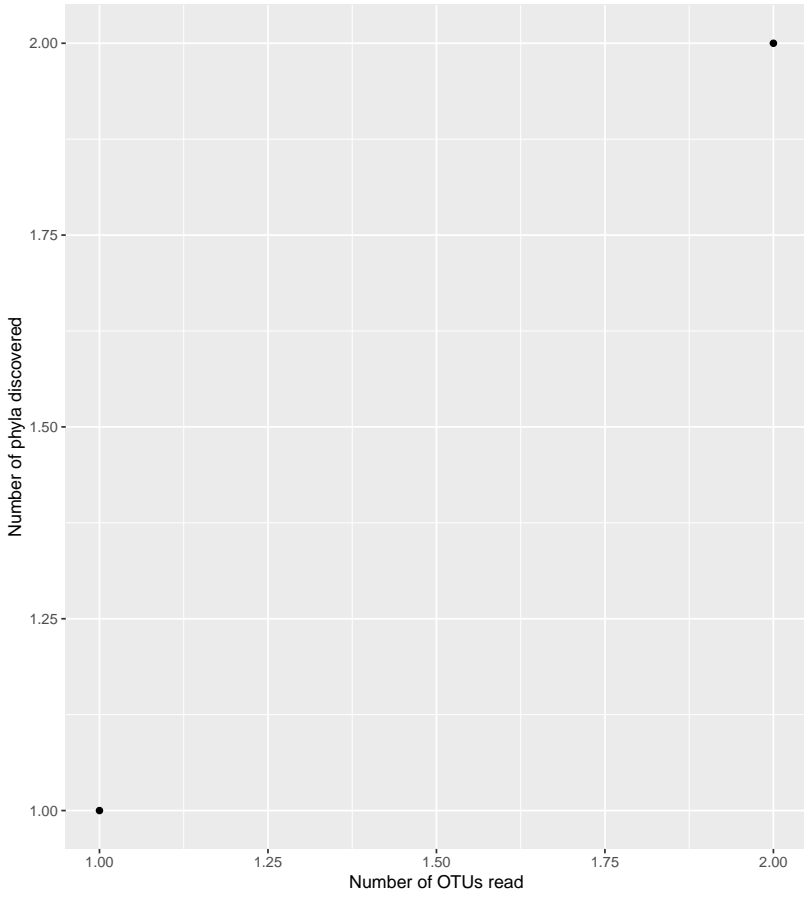
25. 18S.1.2.H.PCM.ME



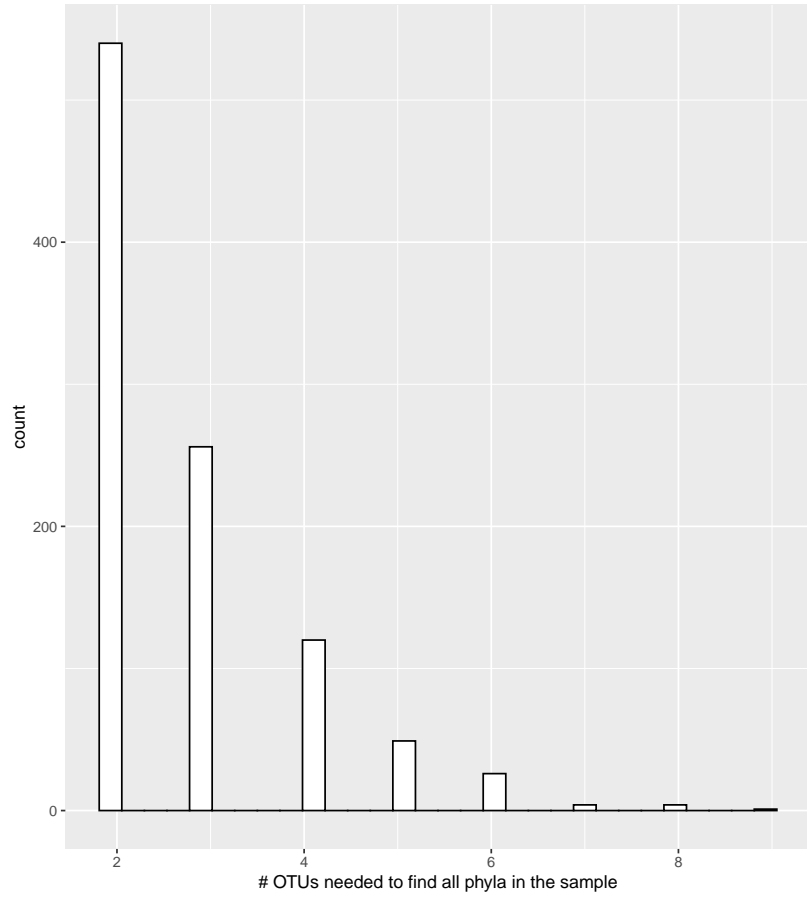
18S.1.2.H.PCM.ME

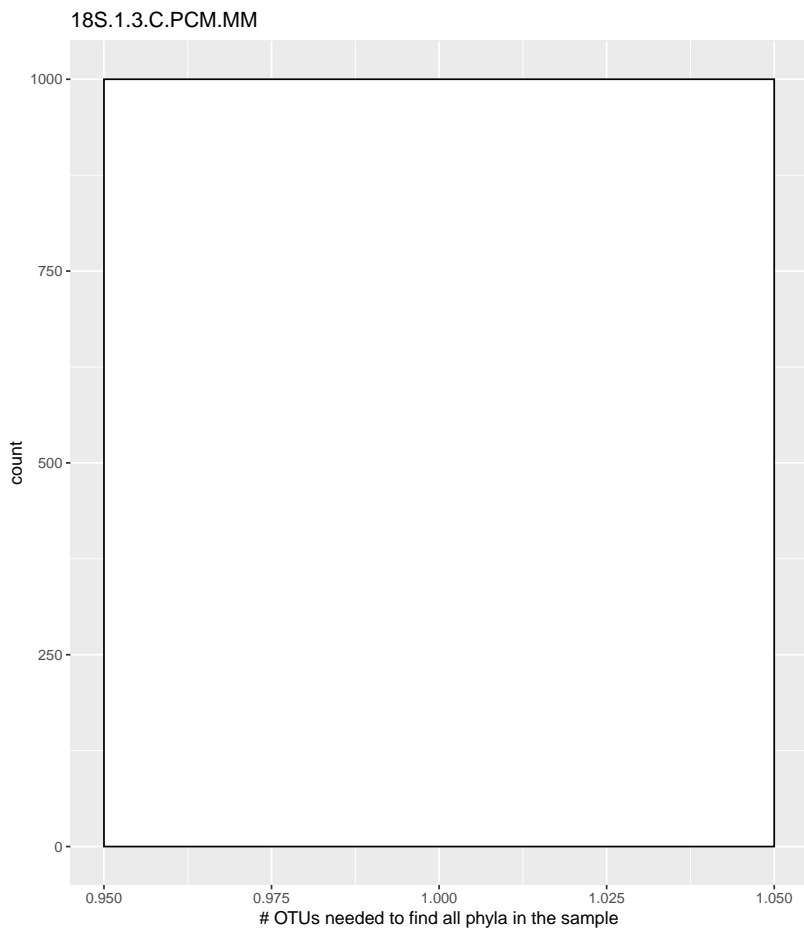
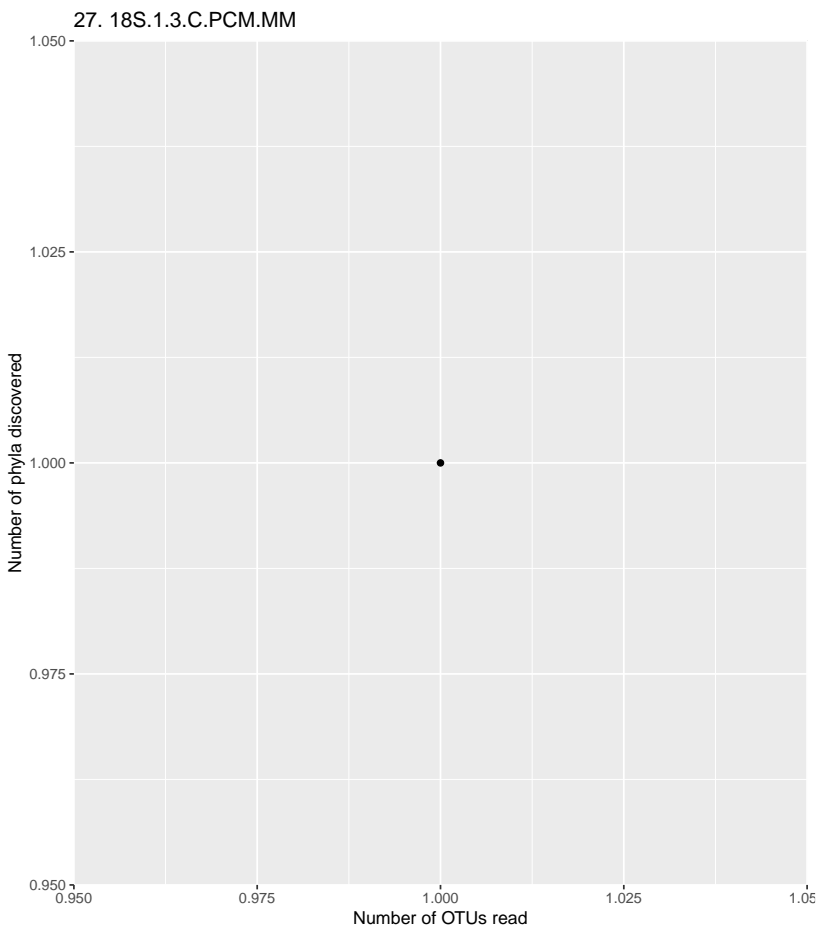


26. 18S.1.3.B.PCM.MM

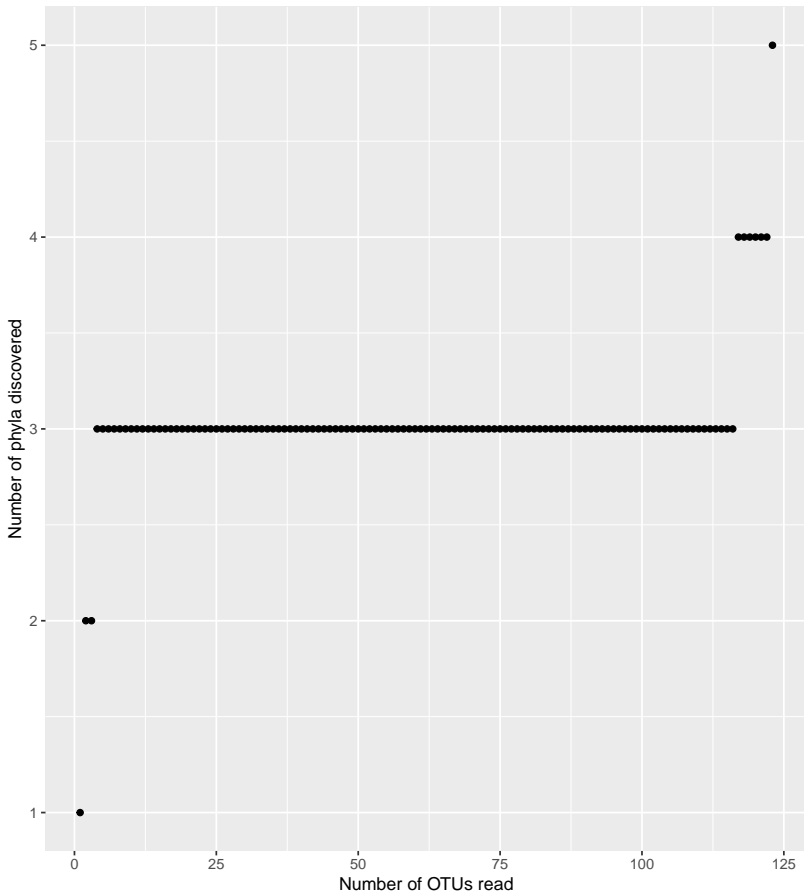


18S.1.3.B.PCM.MM

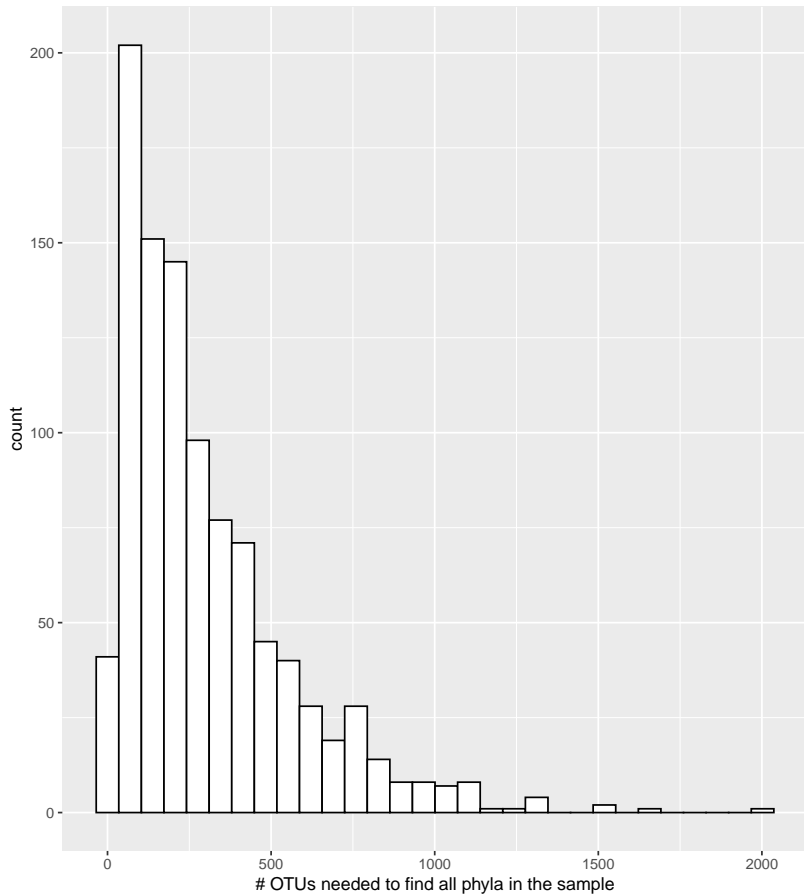




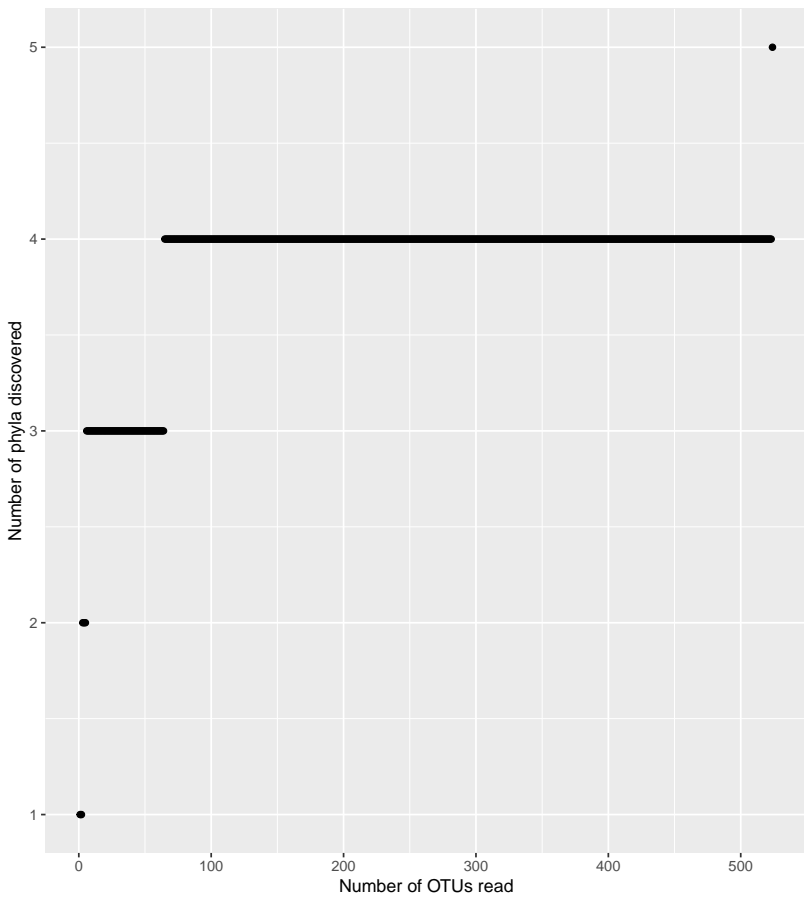
28. 18S.1.3.D.PCM.ME



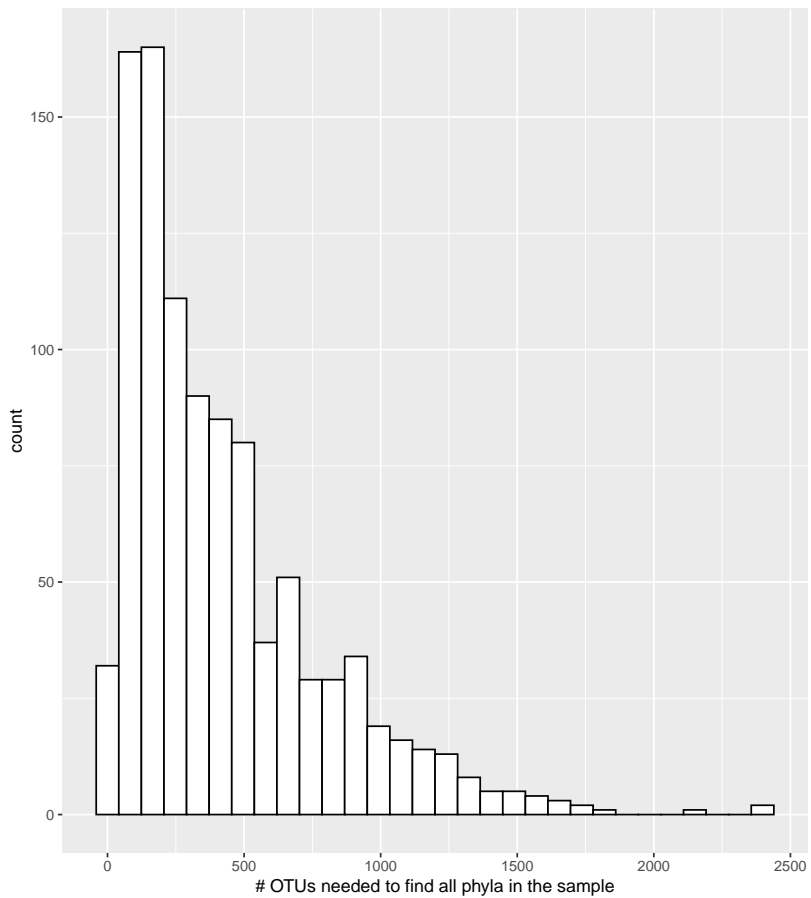
18S.1.3.D.PCM.ME



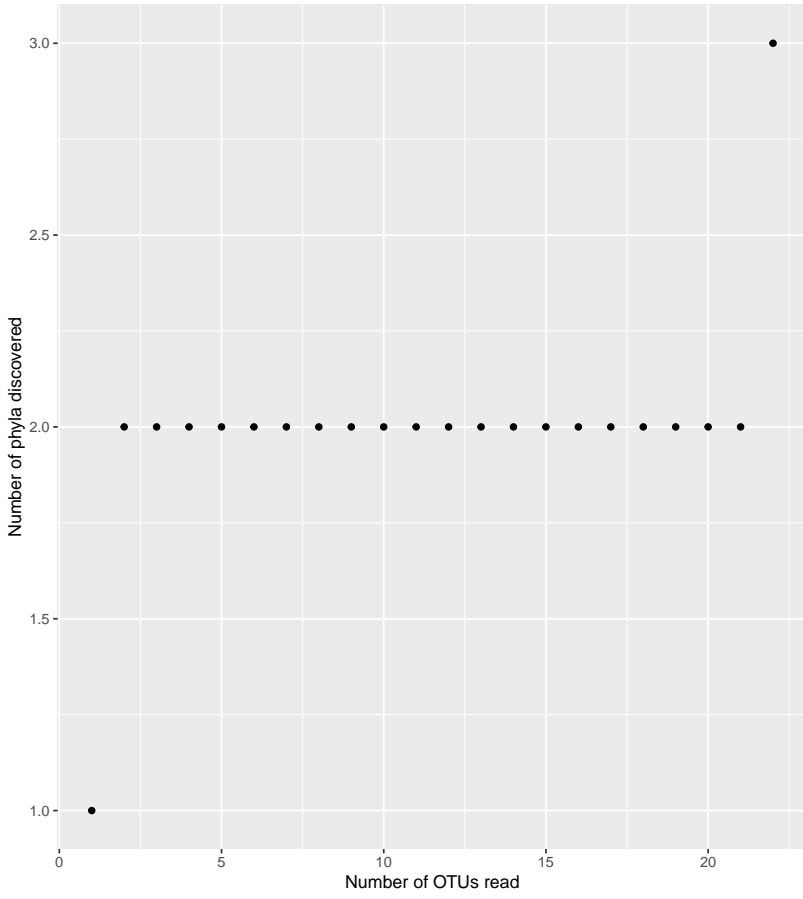
29. 18S.1.3.E.PCM.ME



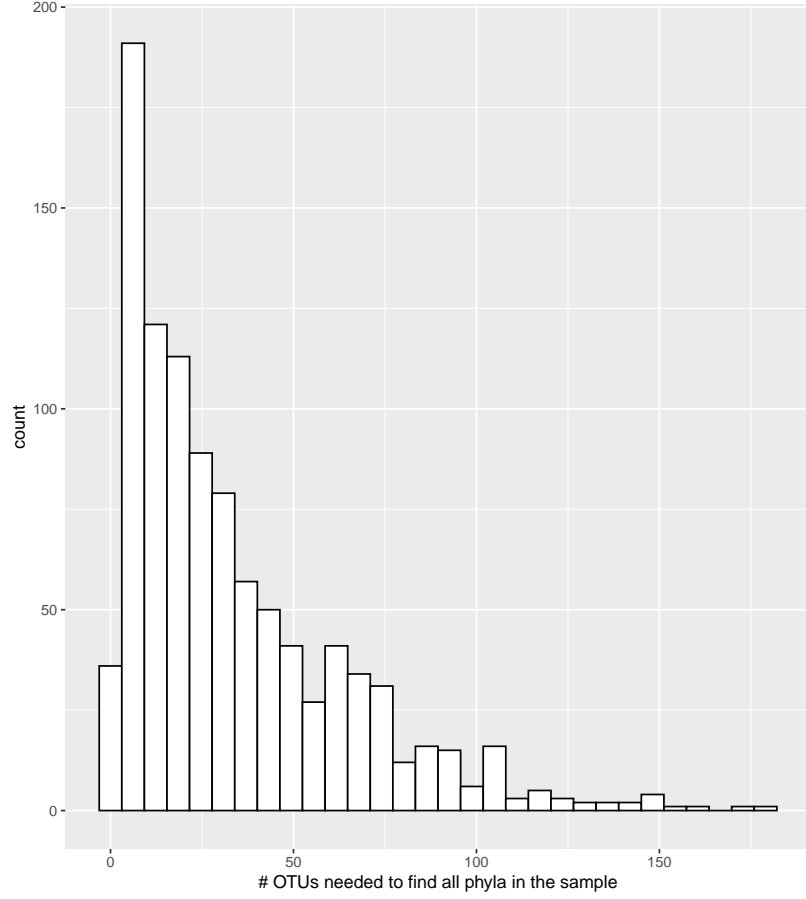
18S.1.3.E.PCM.ME



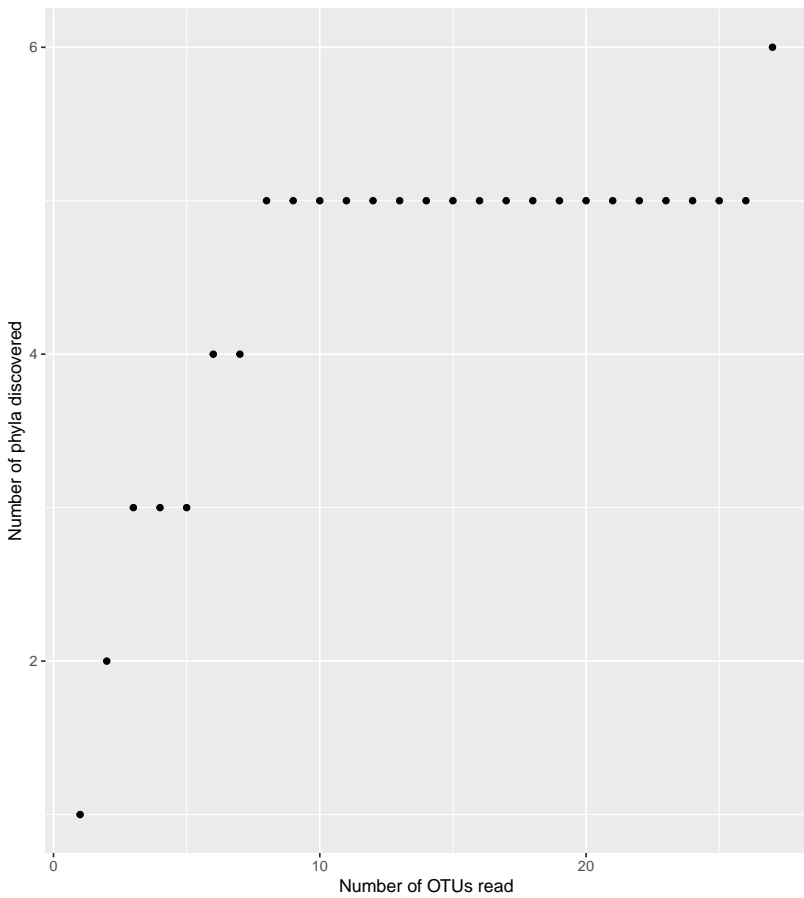
30. 18S.1.3.H.PCM.ME



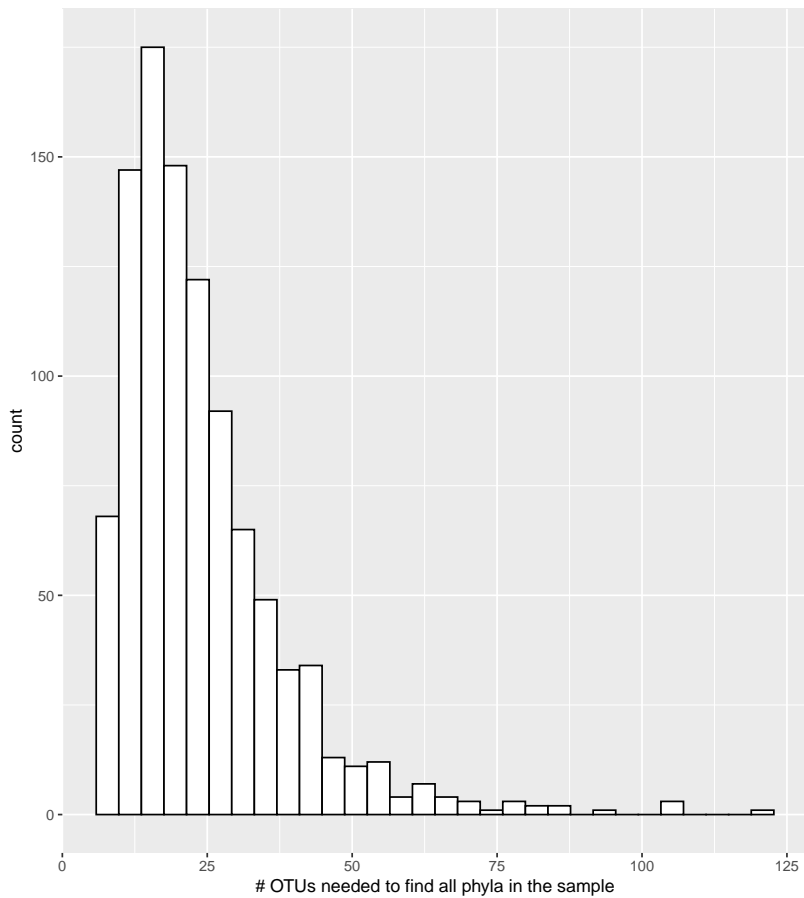
18S.1.3.H.PCM.ME



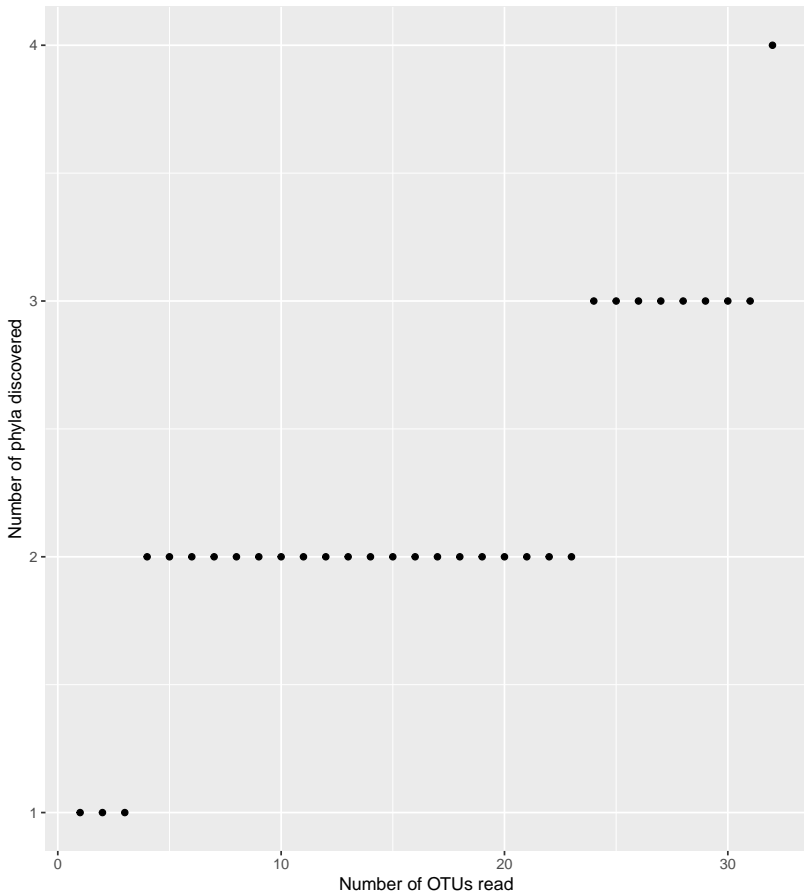
31. 18S.1.4.B.PCM.MM



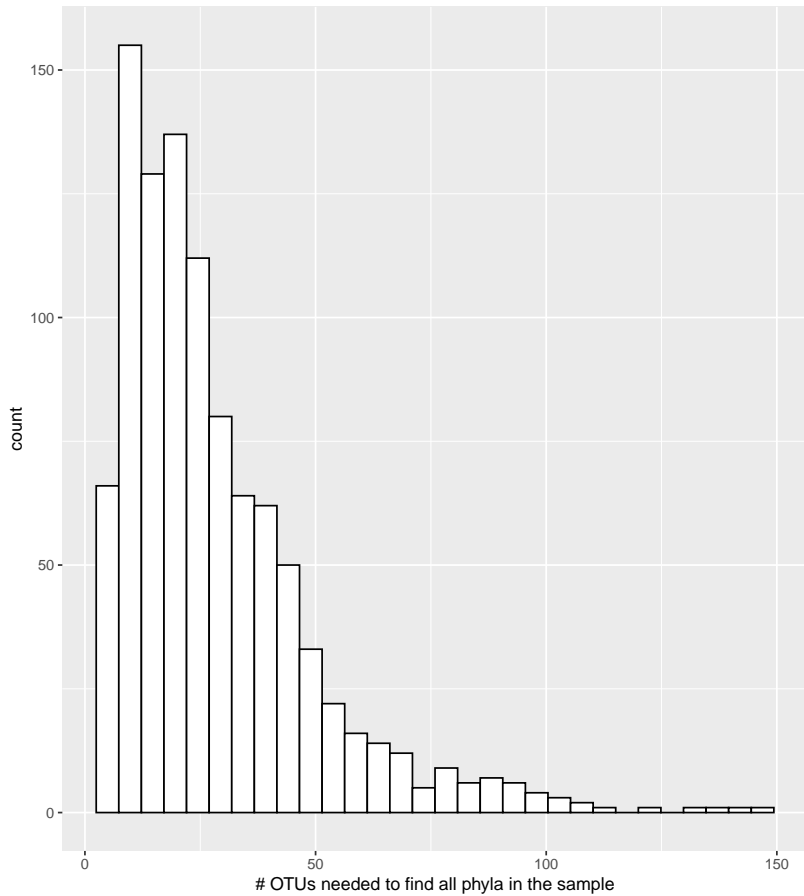
18S.1.4.B.PCM.MM



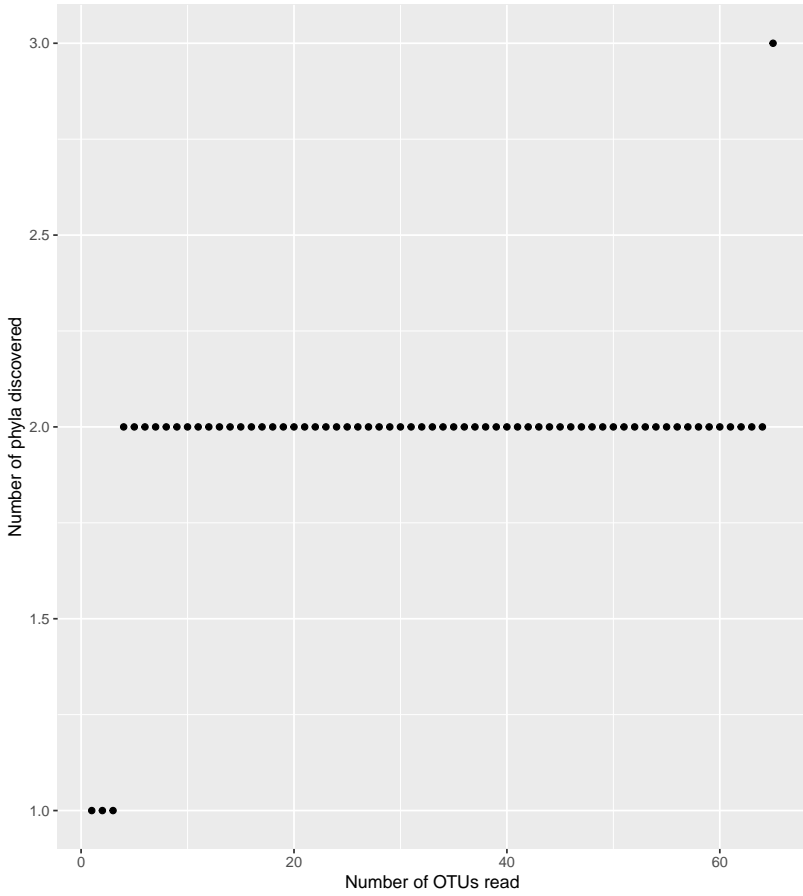
32. 18S.1.4.D.PCM.ME



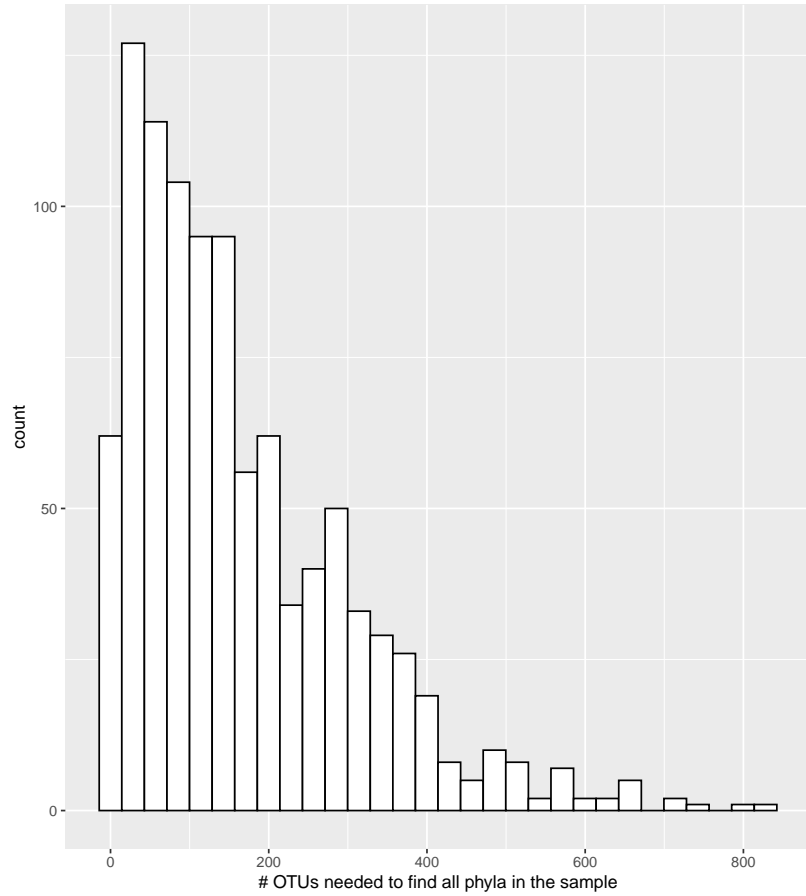
18S.1.4.D.PCM.ME



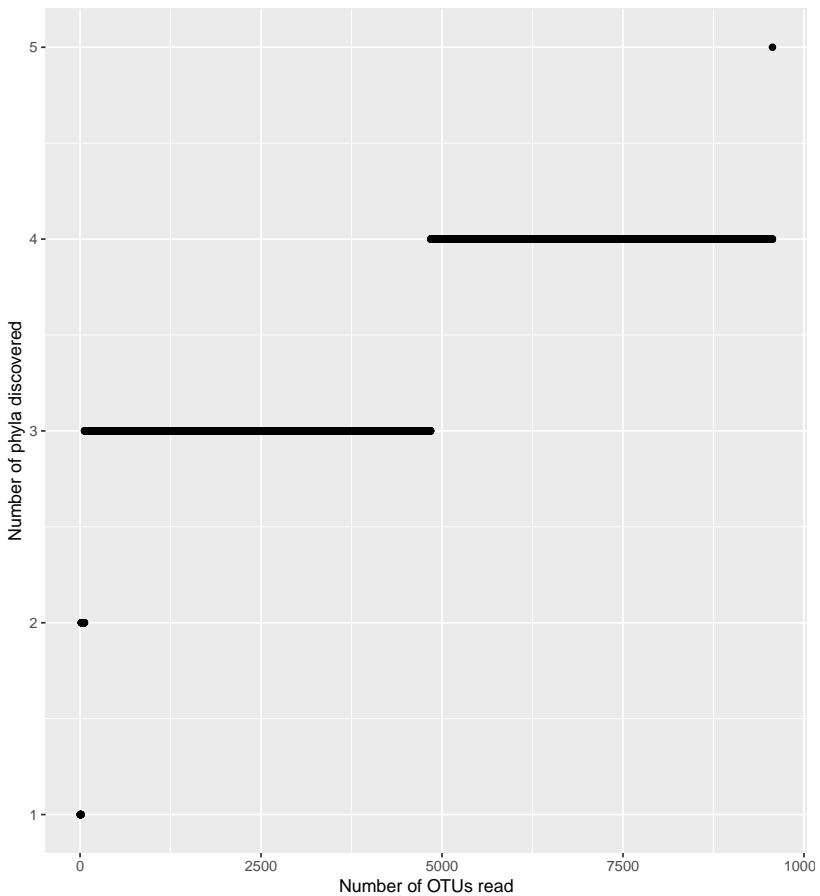
33. 18S.1.4.E.PCM.ME



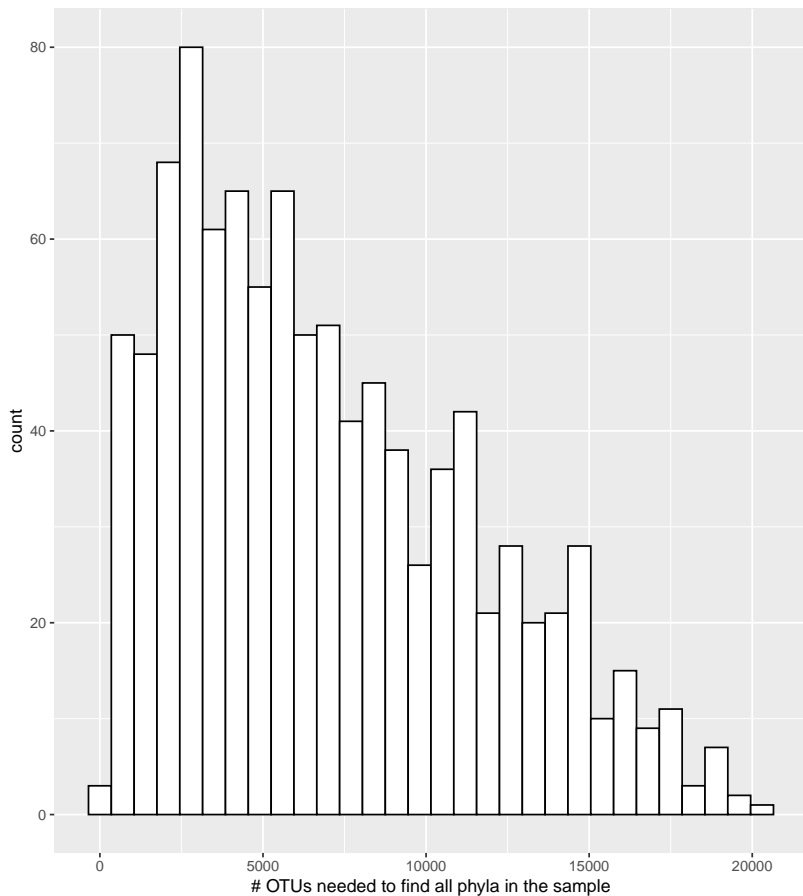
18S.1.4.E.PCM.ME



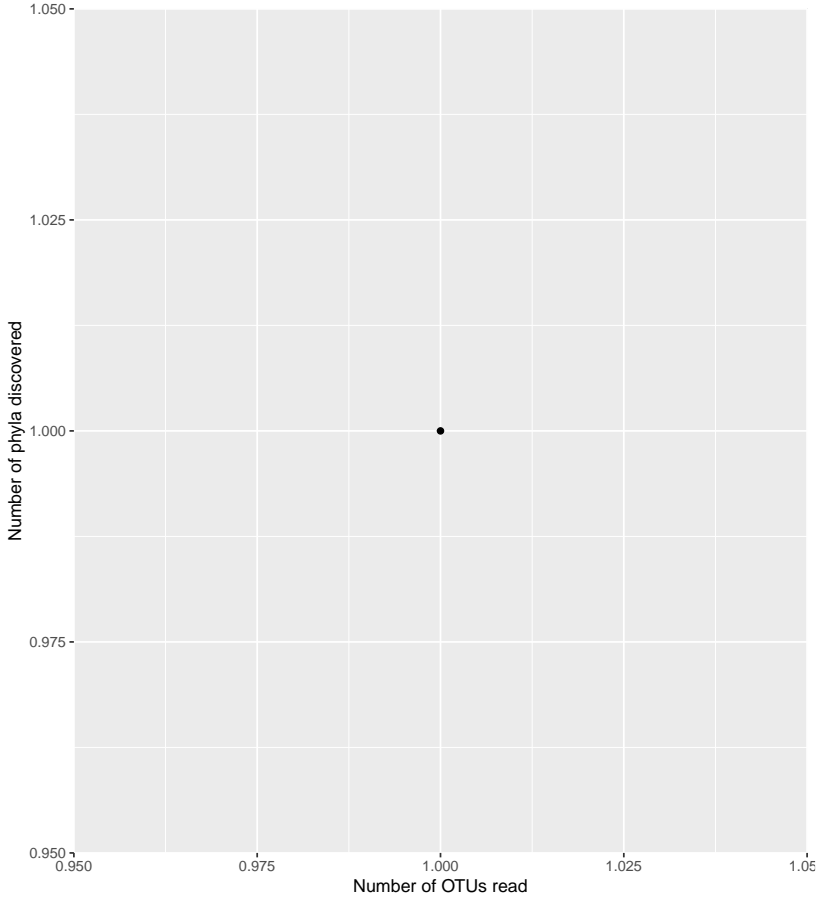
34. 18S.1.4.F.PCM.ME



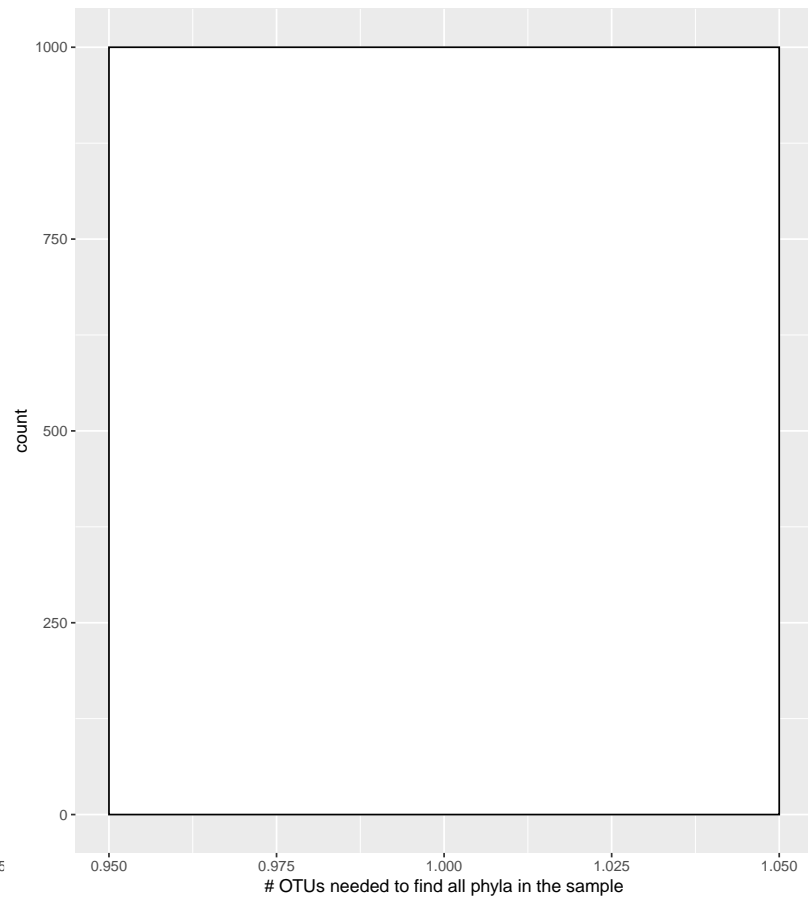
18S.1.4.F.PCM.ME



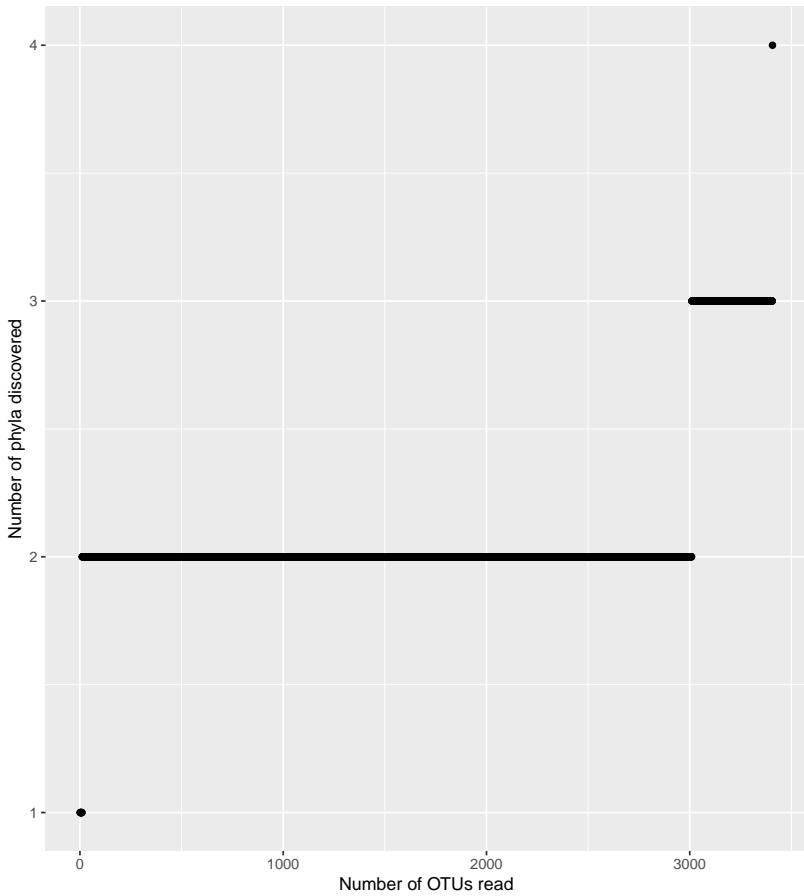
35. 18S.1.4.G.PCM.ME



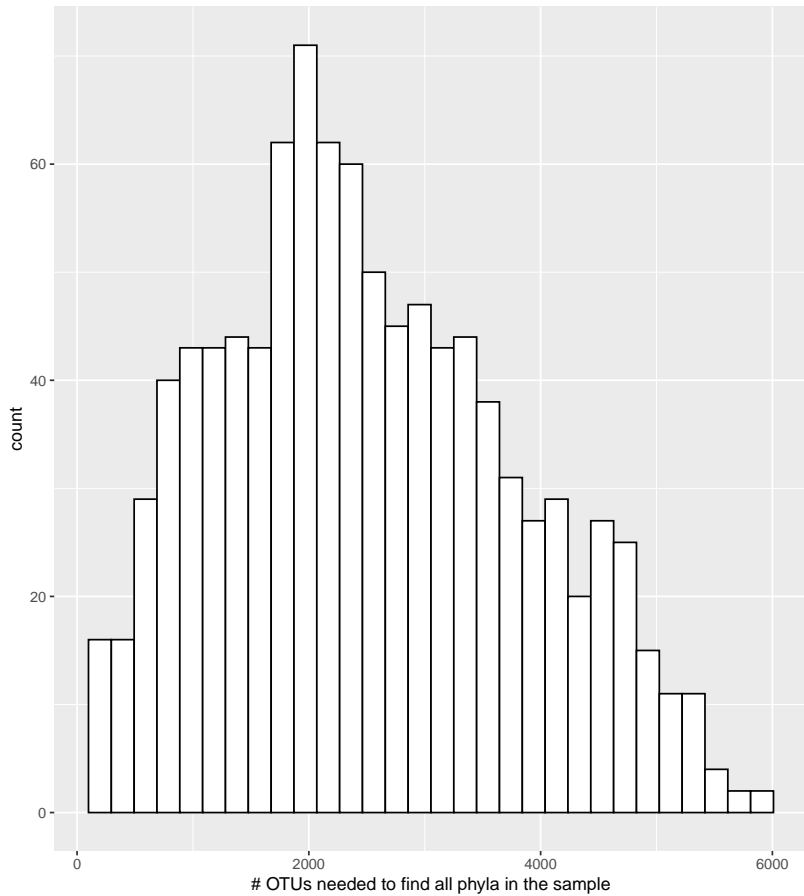
18S.1.4.G.PCM.ME



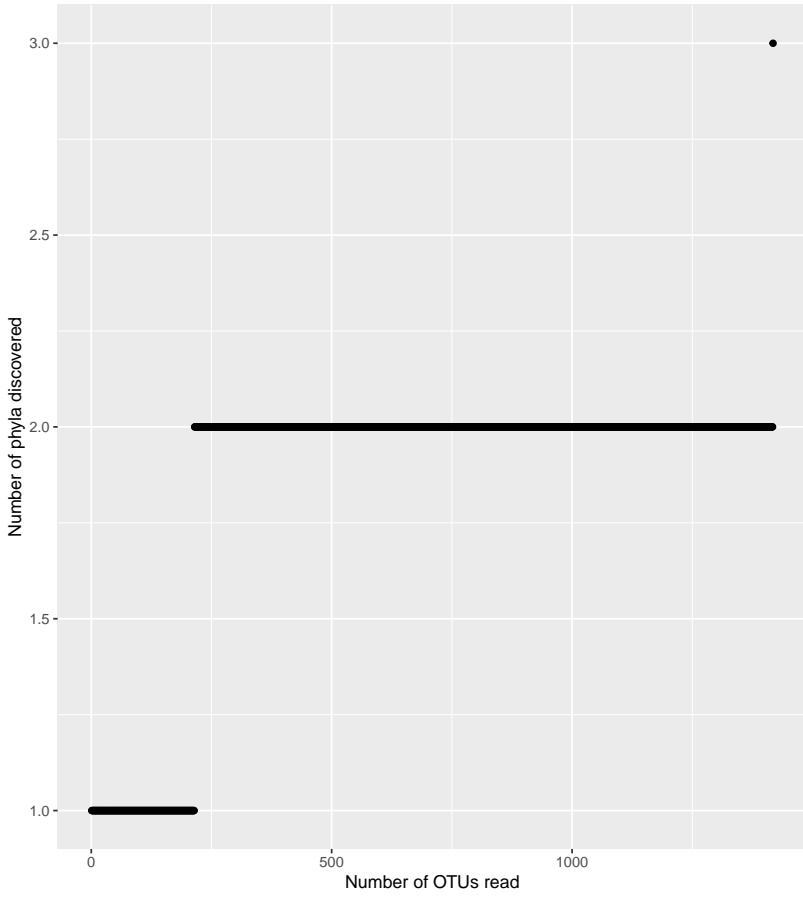
36. 18S.1.4.H.PCM.ME



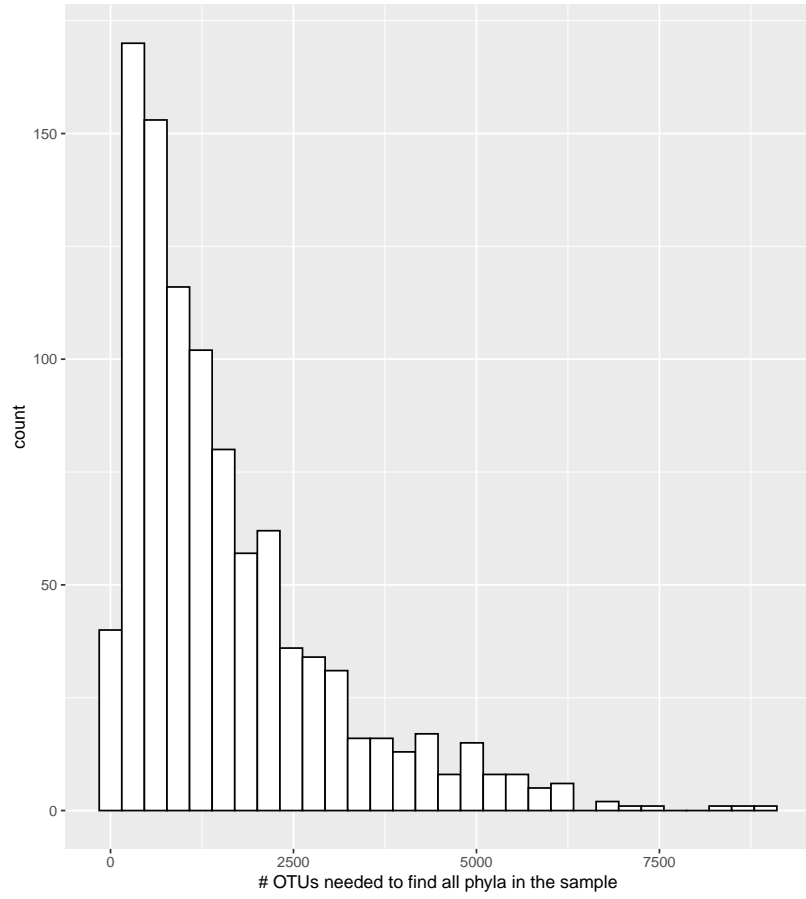
18S.1.4.H.PCM.ME

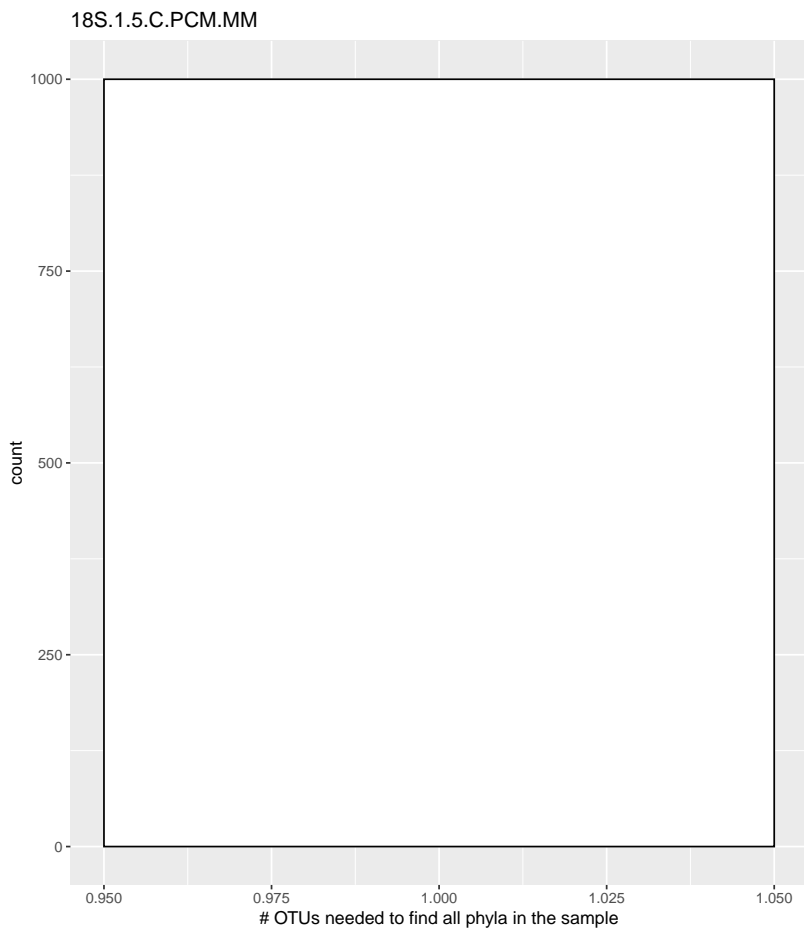
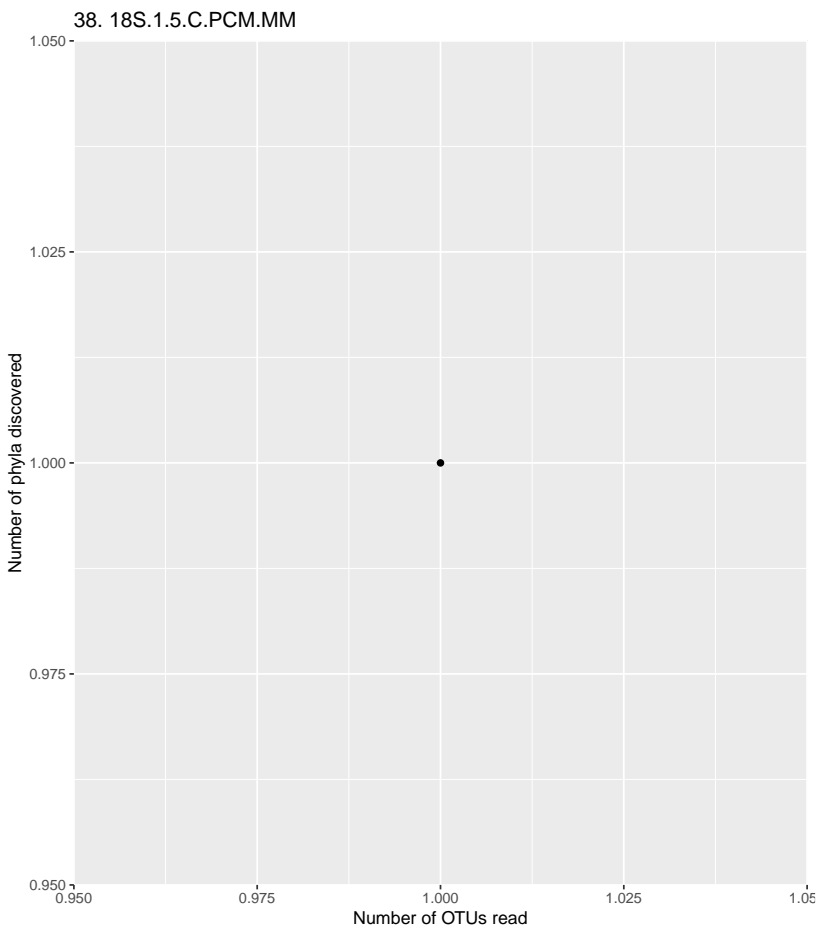


37. 18S.1.5.B.PCM.MM

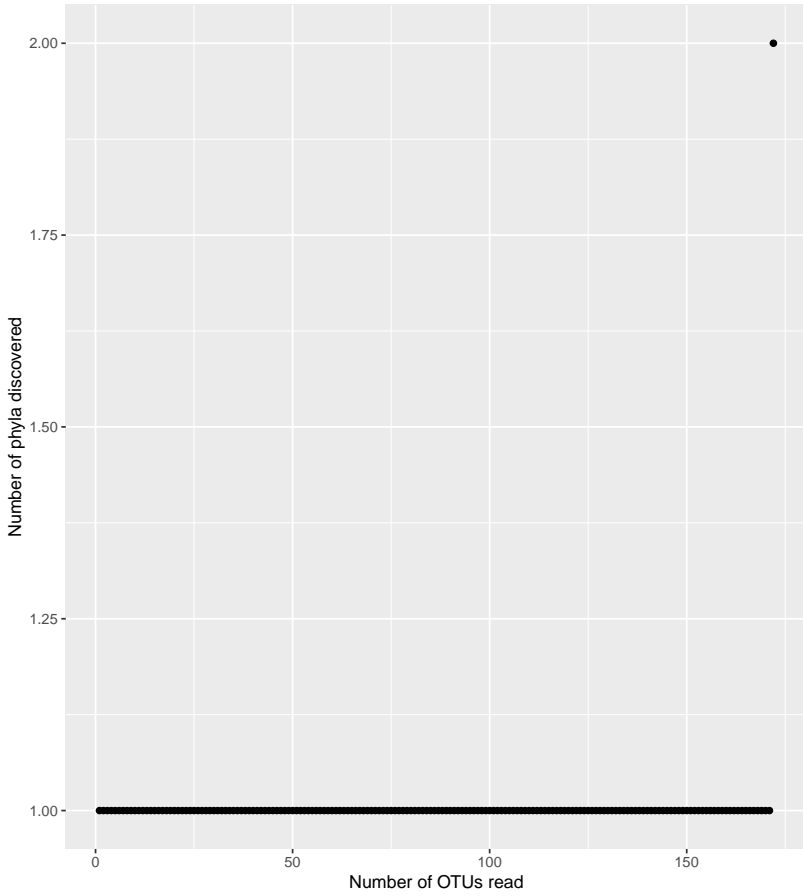


18S.1.5.B.PCM.MM

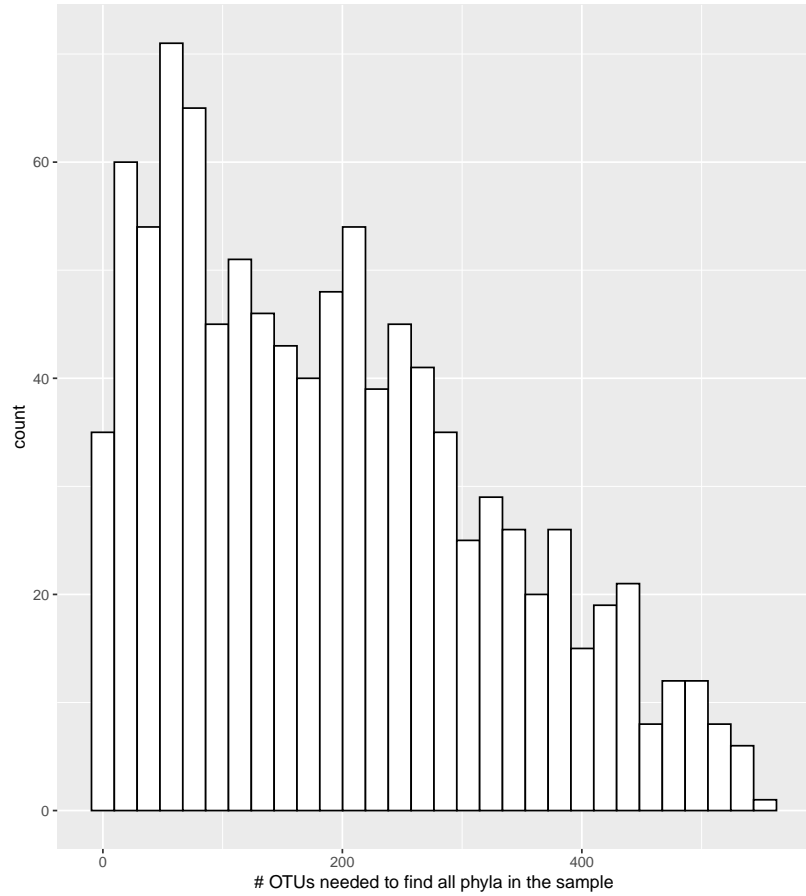




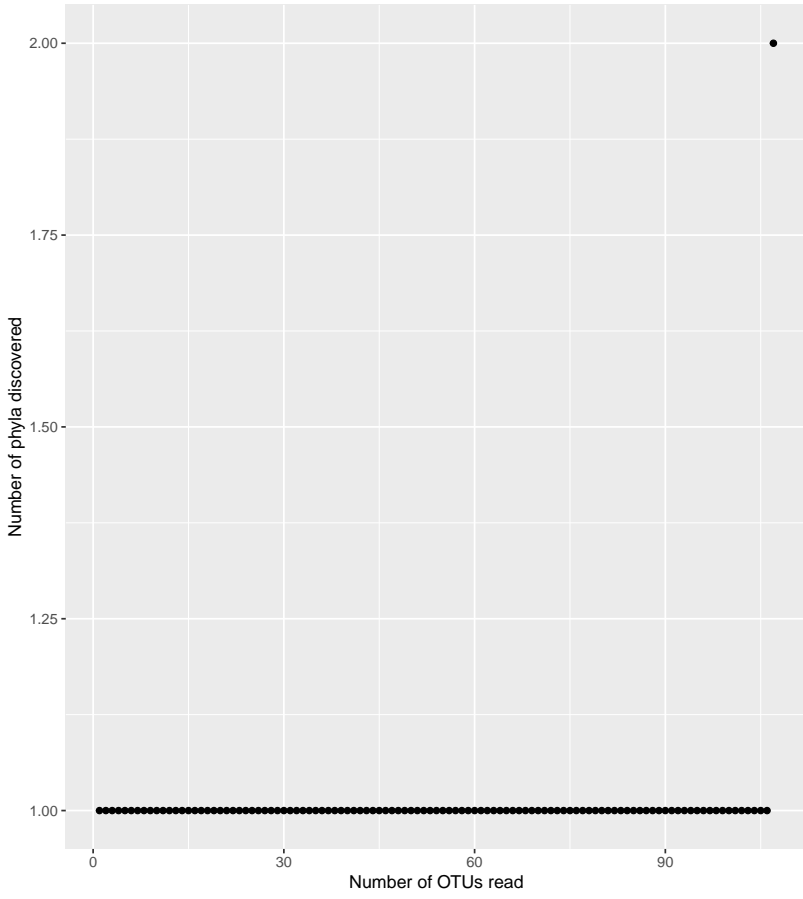
39. 18S.1.5.D.PCM.ME



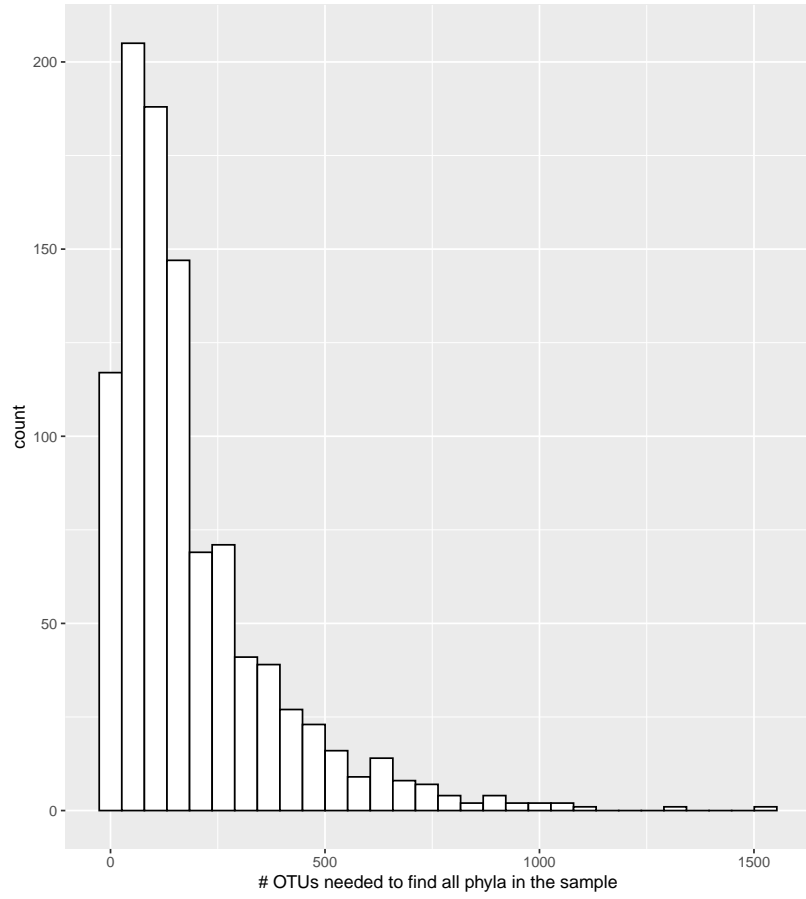
18S.1.5.D.PCM.ME



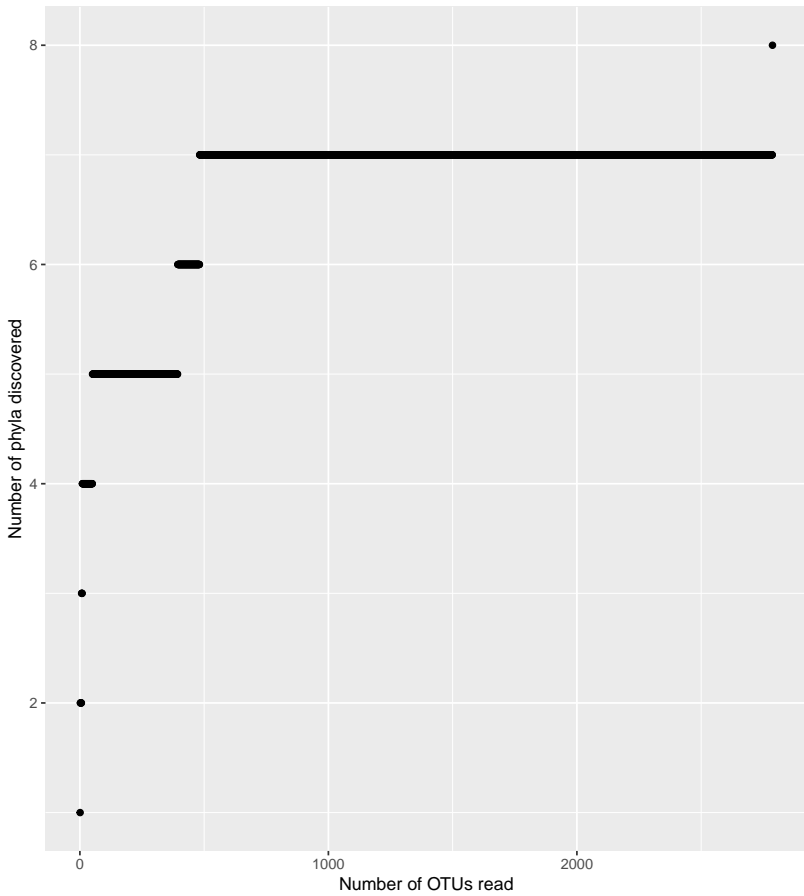
40. 18S.1.5.E.PCM.ME



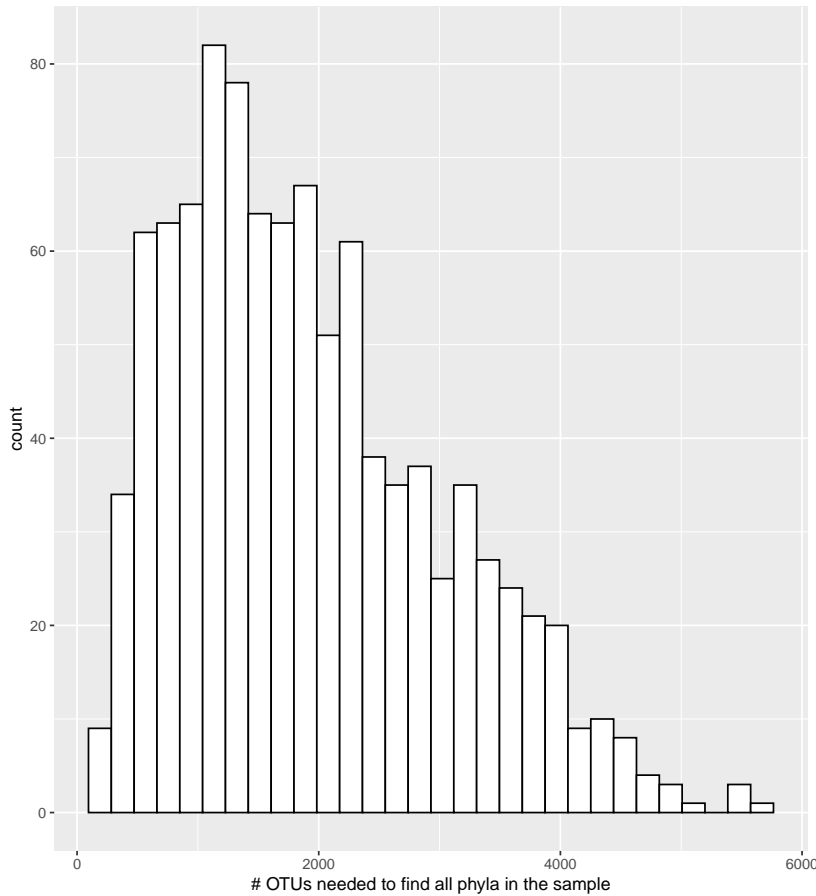
18S.1.5.E.PCM.ME



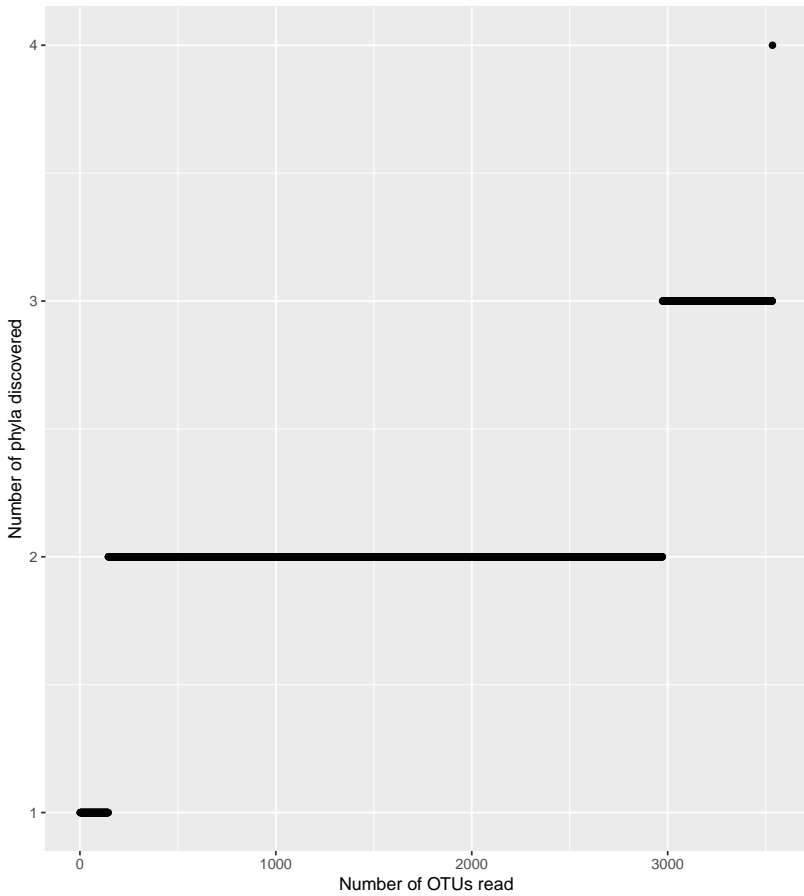
41. 18S.1.5.F.PCM.ME



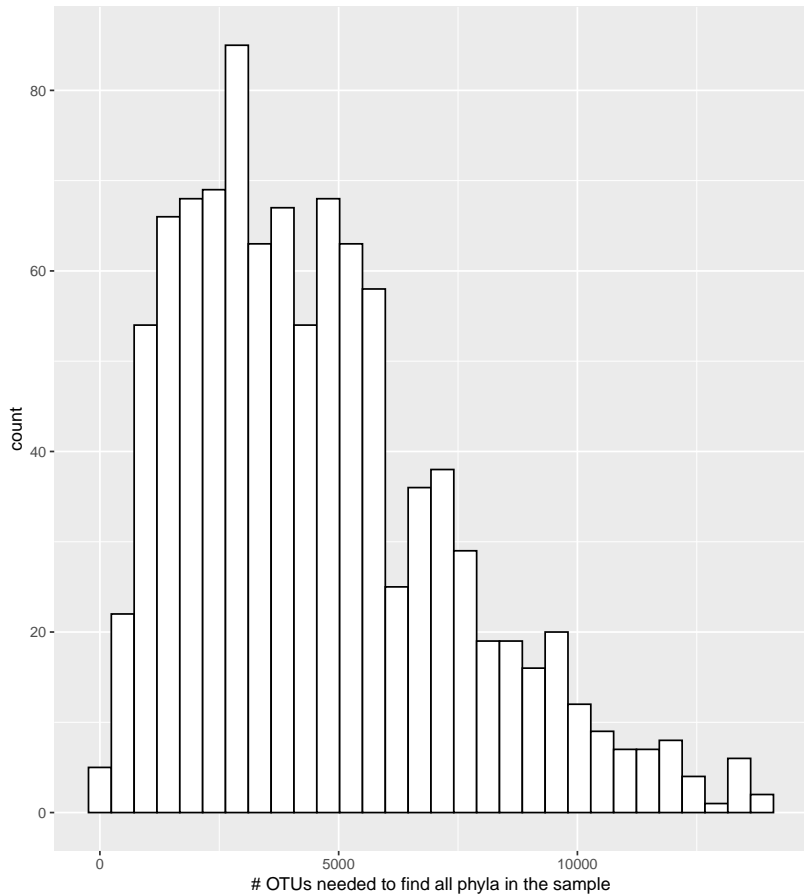
18S.1.5.F.PCM.ME



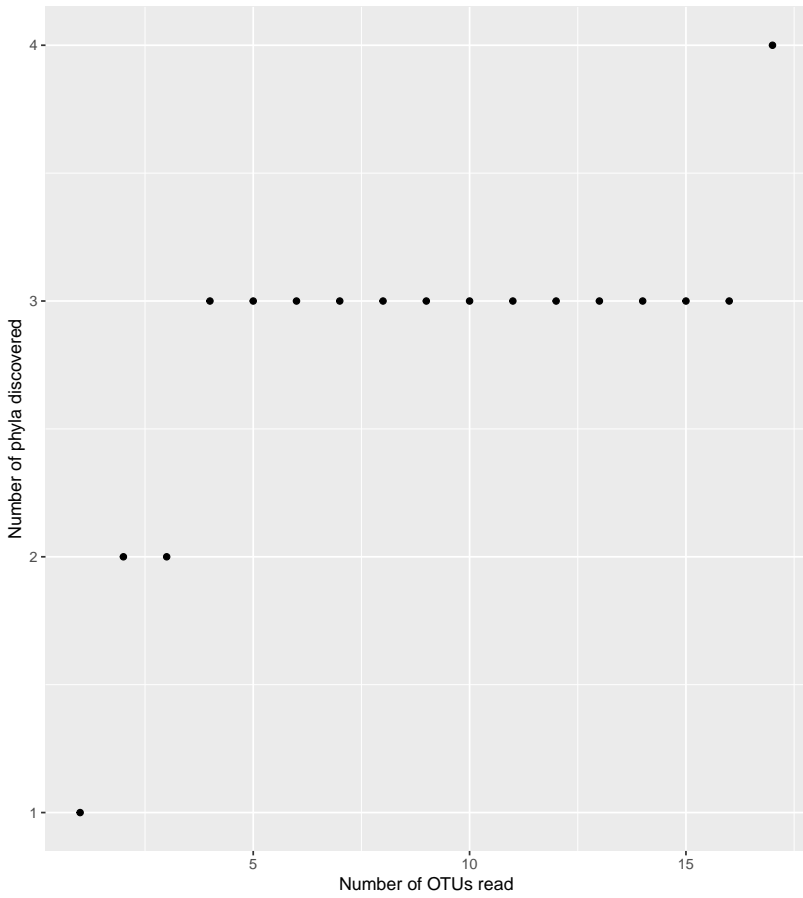
42. 18S.1.5.H.PCM.ME



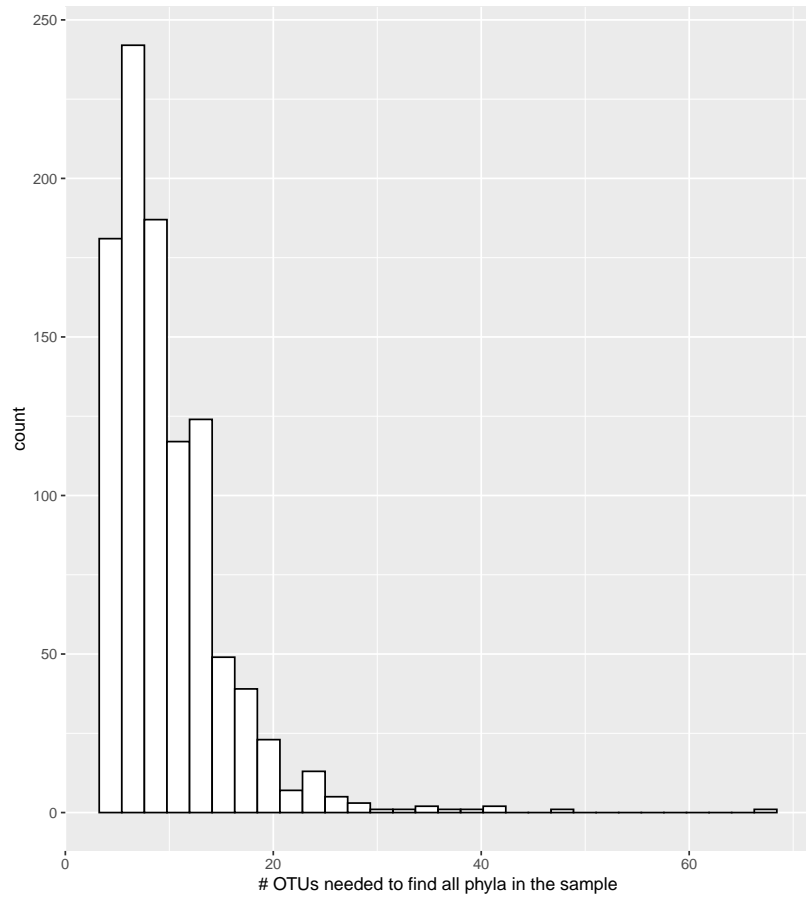
18S.1.5.H.PCM.ME

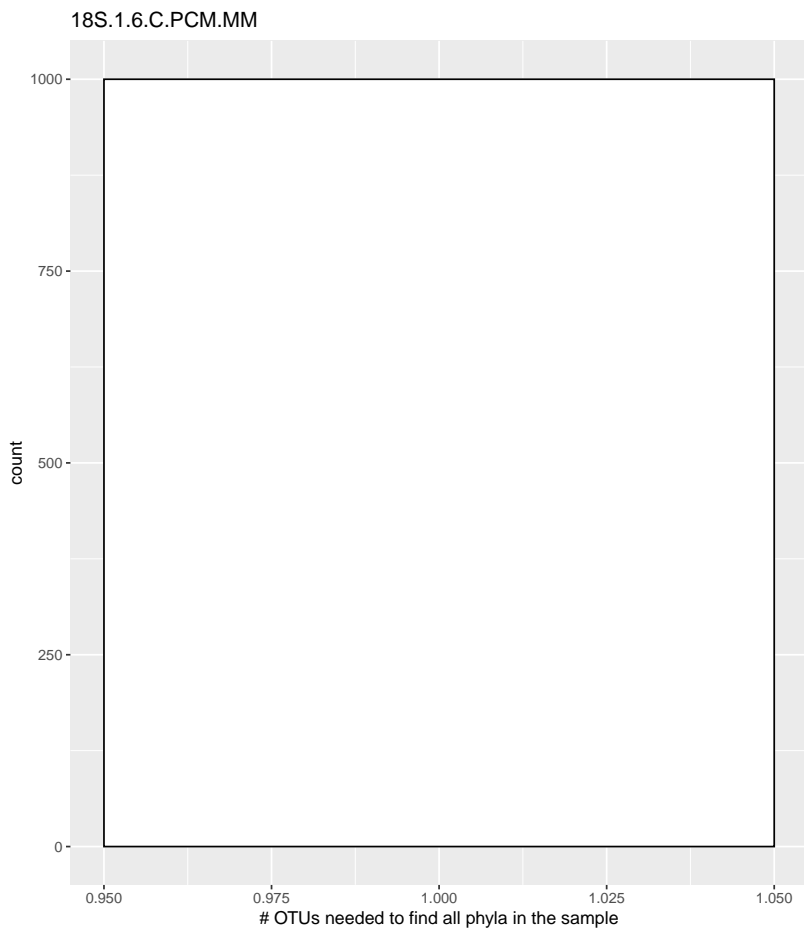
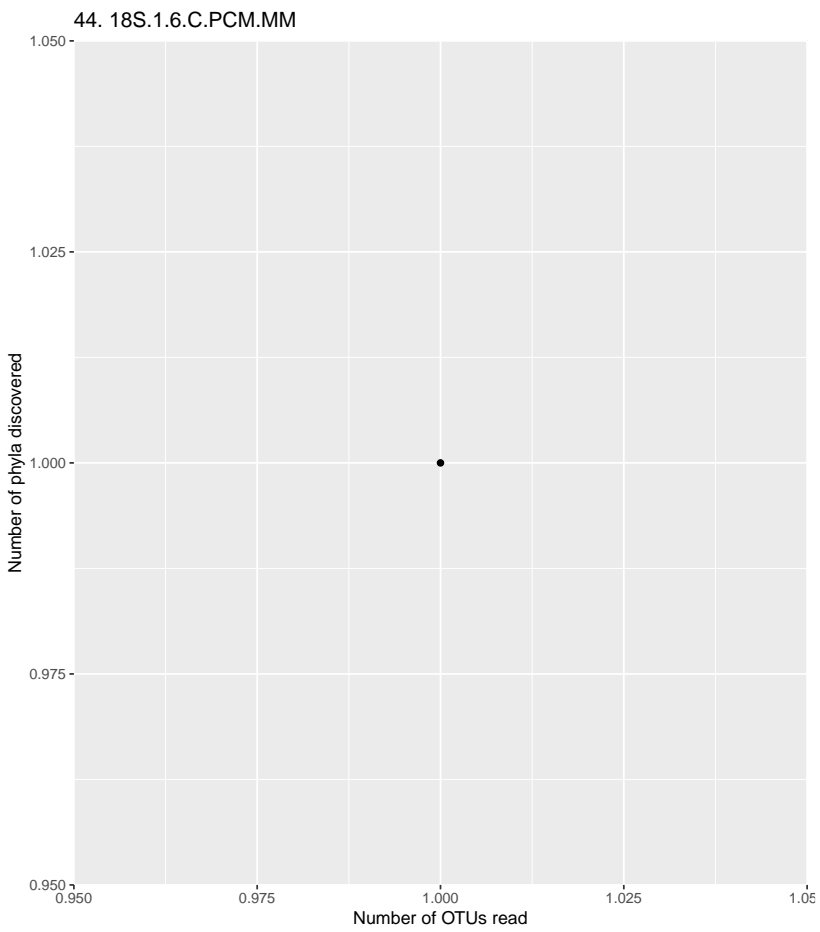


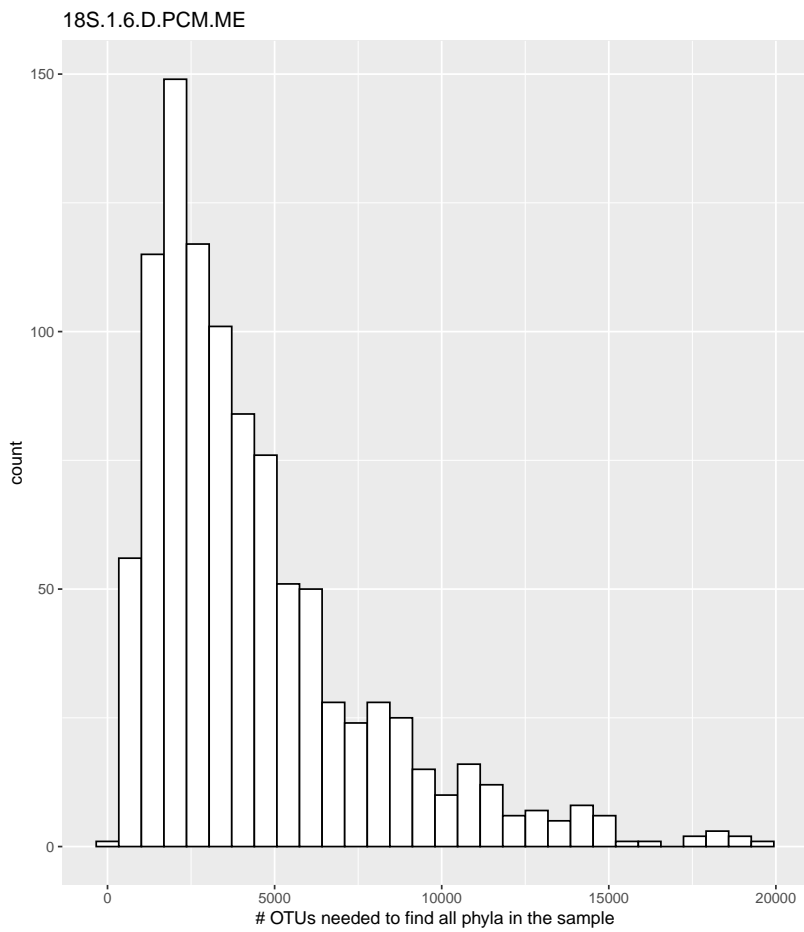
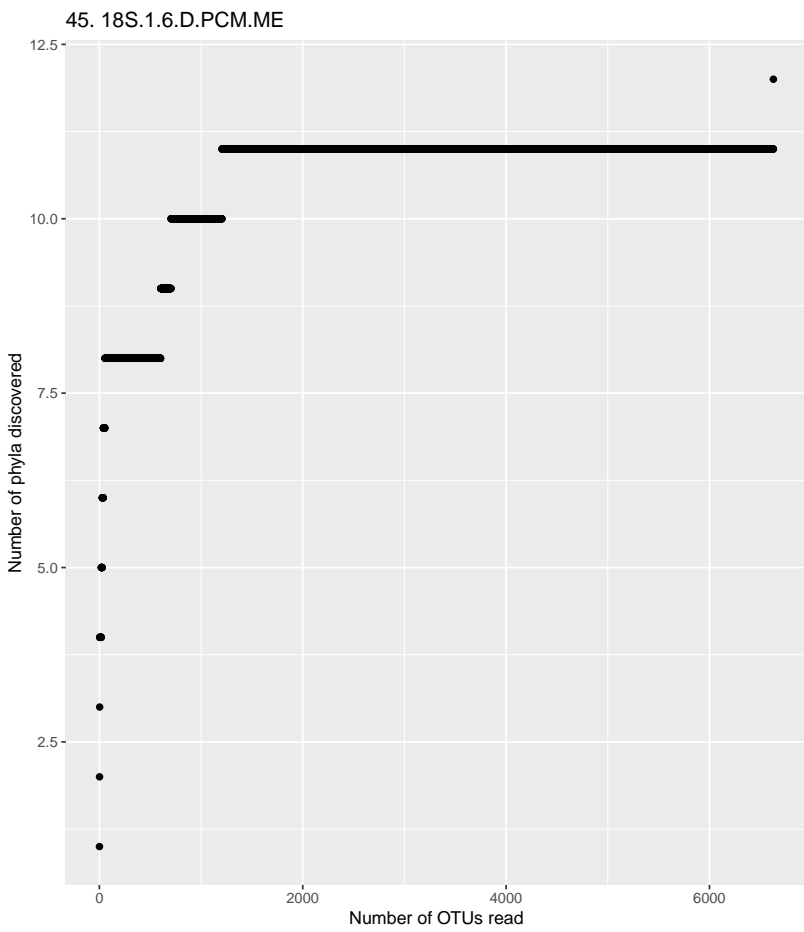
43. 18S.1.6.B.PCM.MM



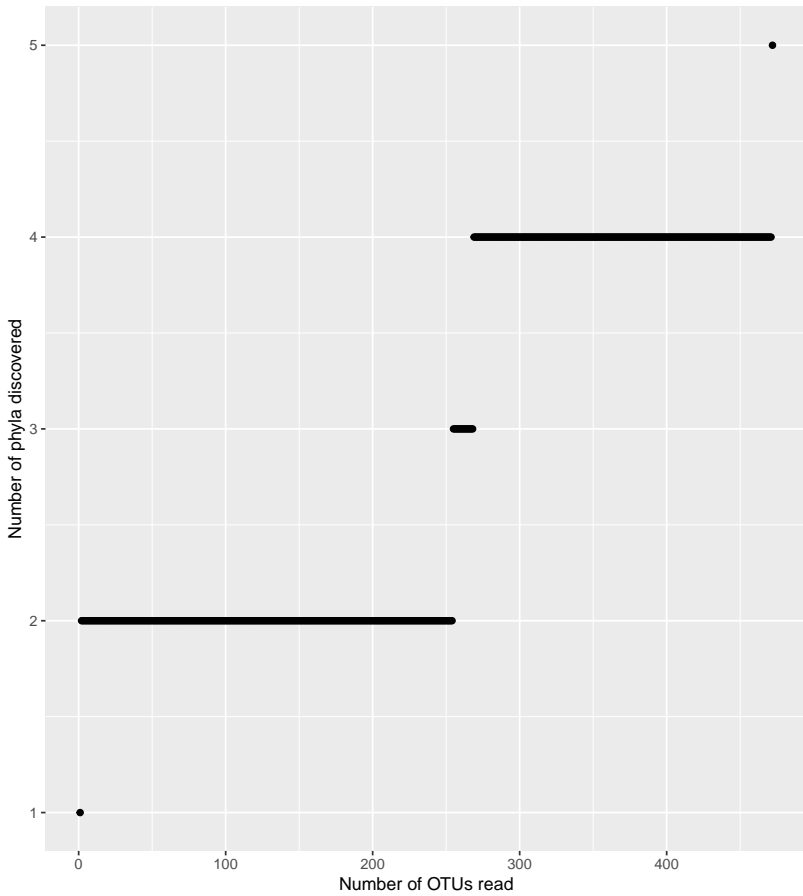
18S.1.6.B.PCM.MM



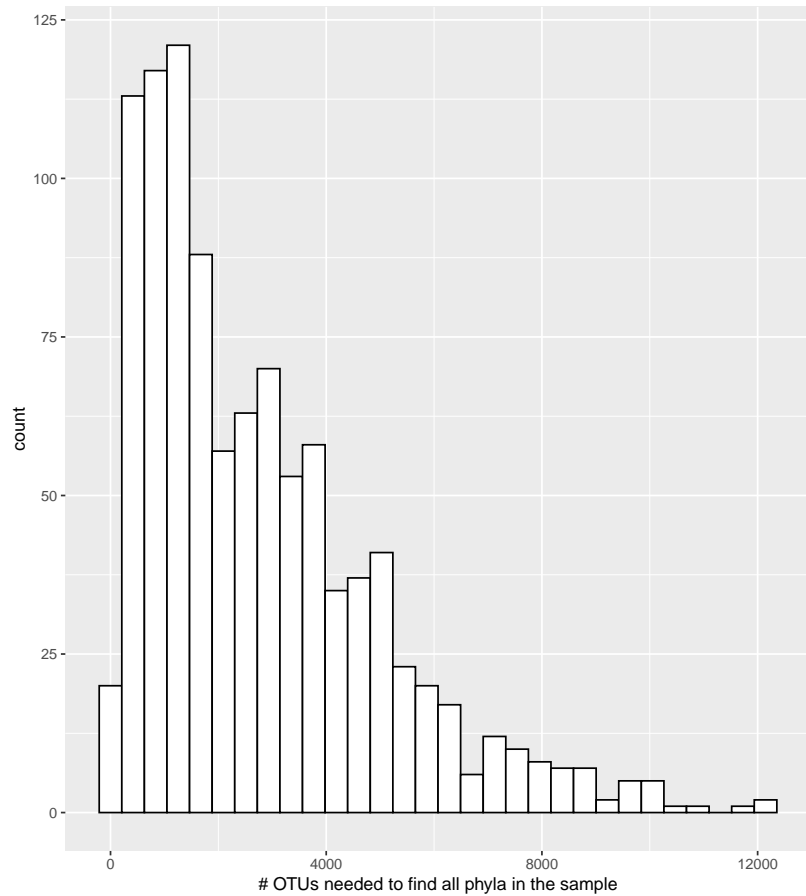




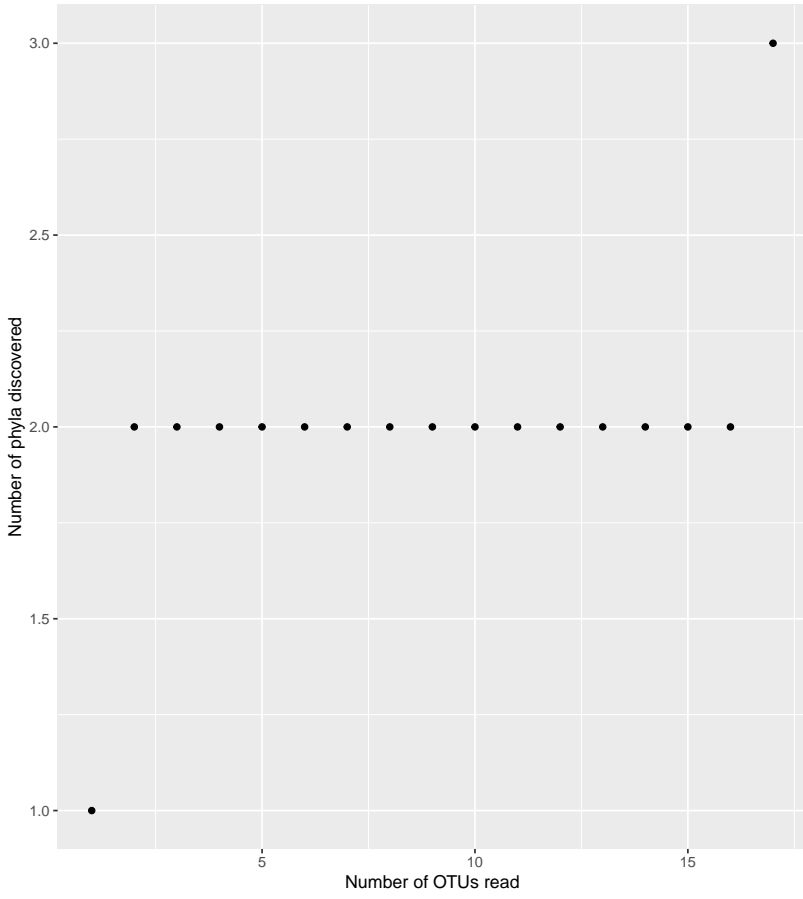
46. 18S.1.6.E.PCM.ME



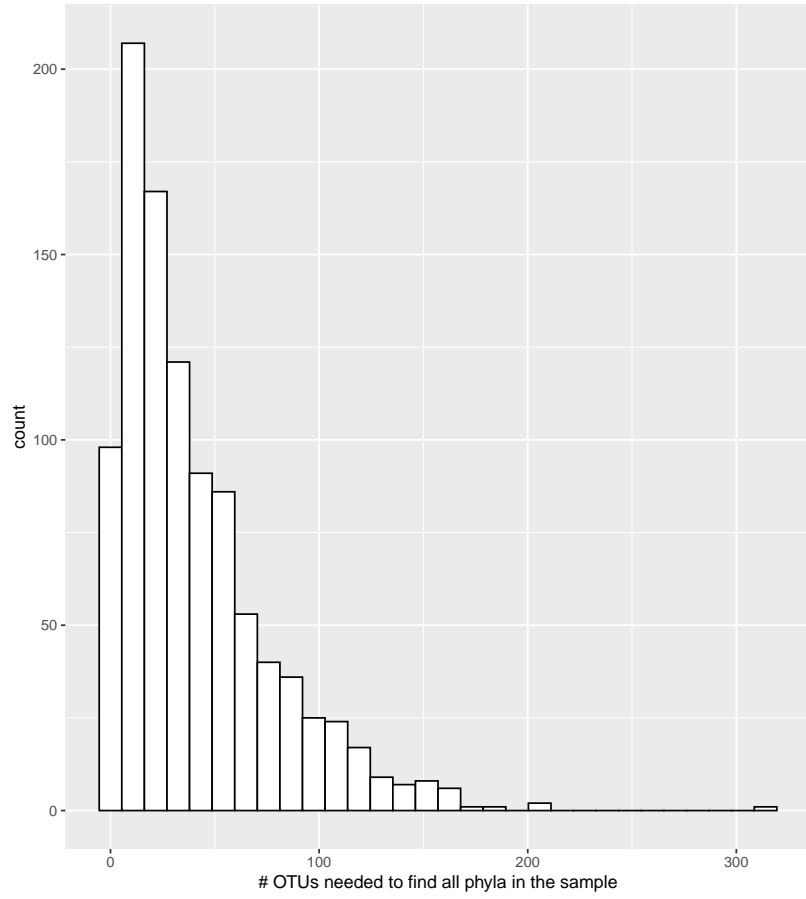
18S.1.6.E.PCM.ME



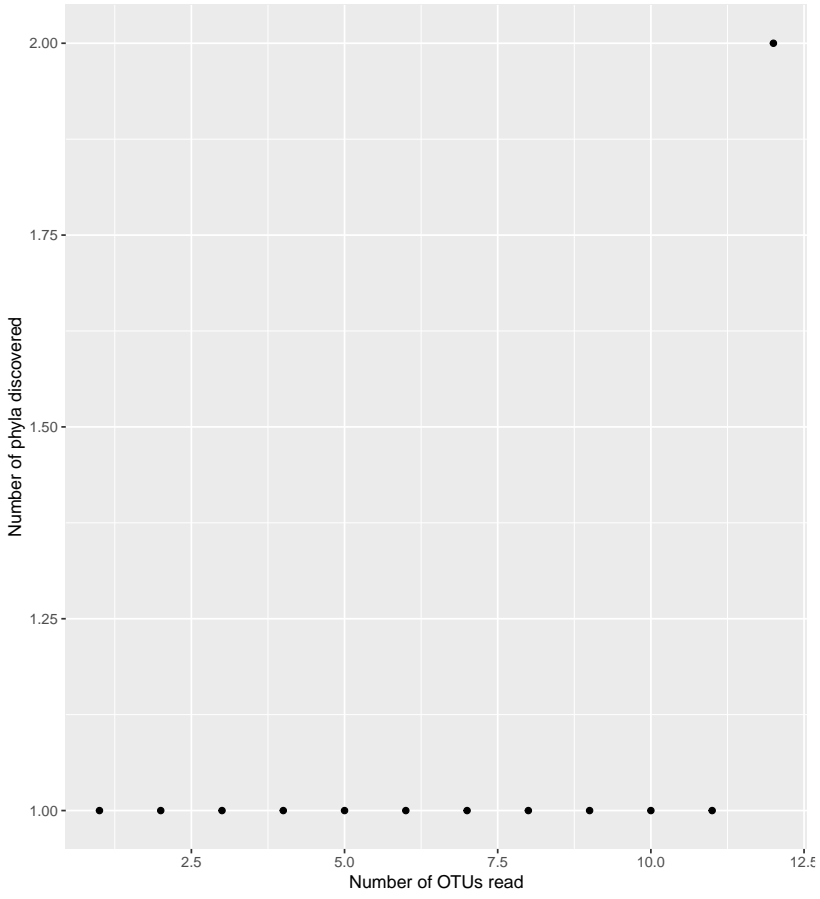
47. 18S.1.6.F.PCM.ME



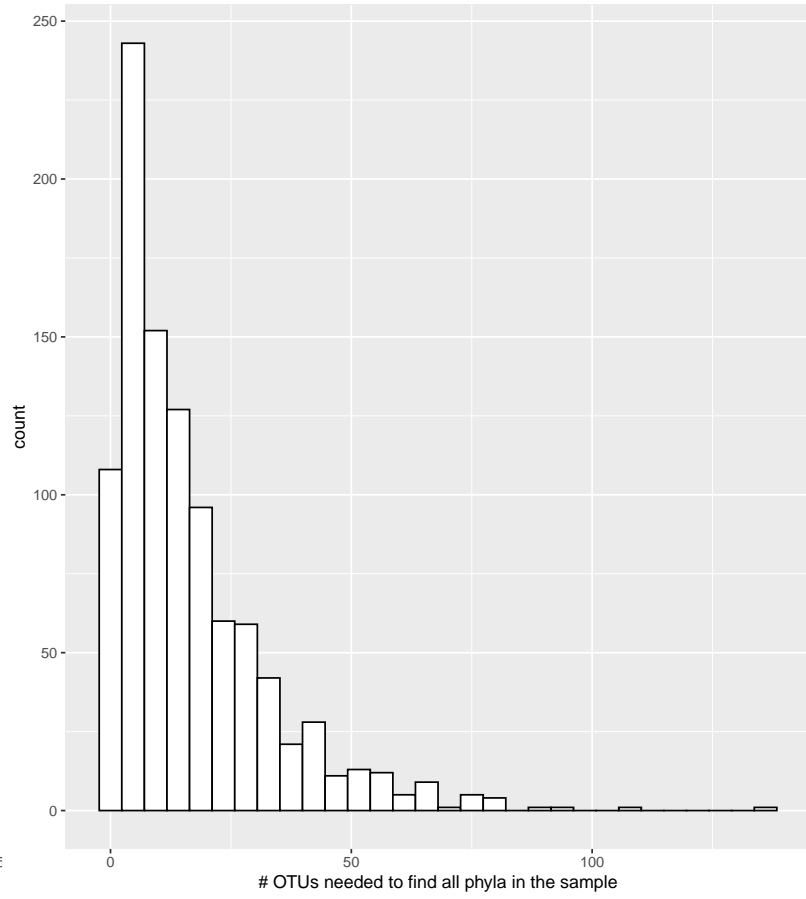
18S.1.6.F.PCM.ME



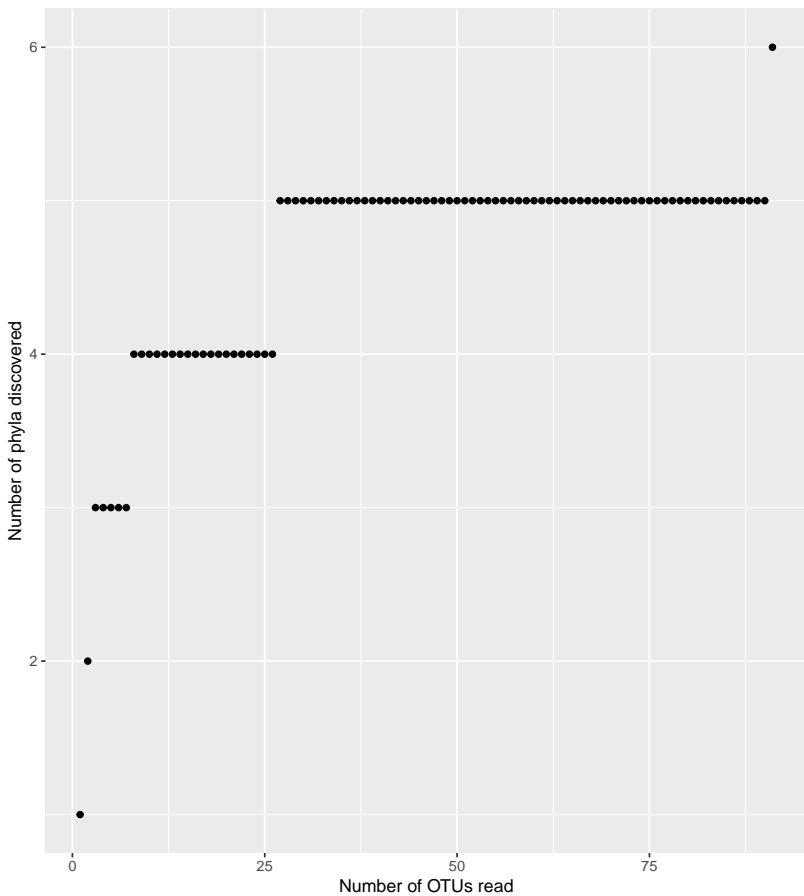
48. 18S.1.6.G.PCM.ME



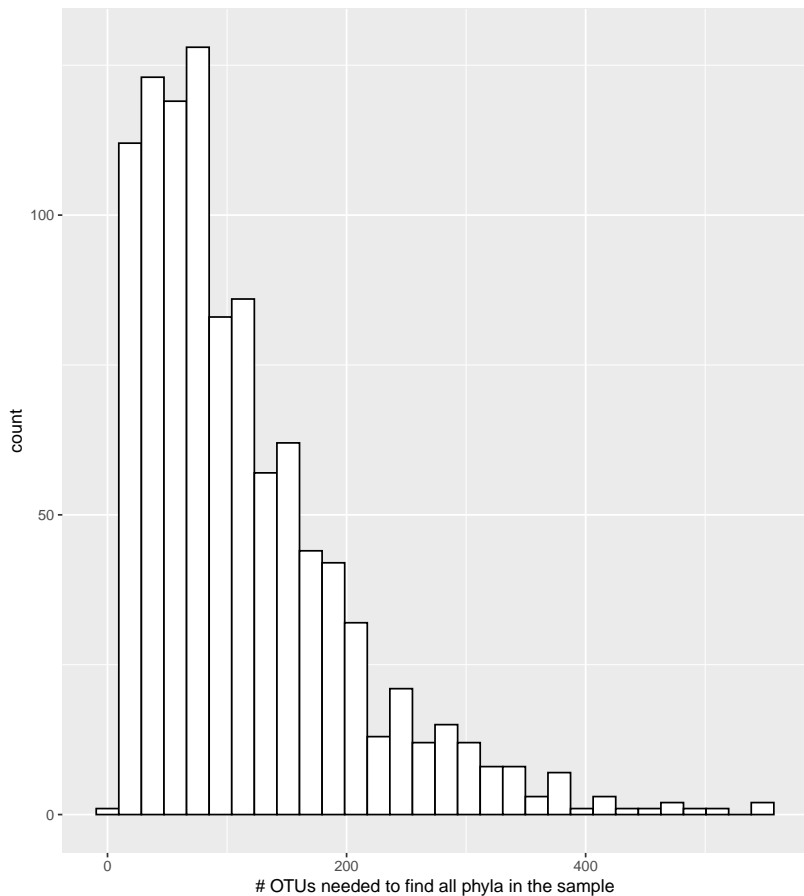
18S.1.6.G.PCM.ME

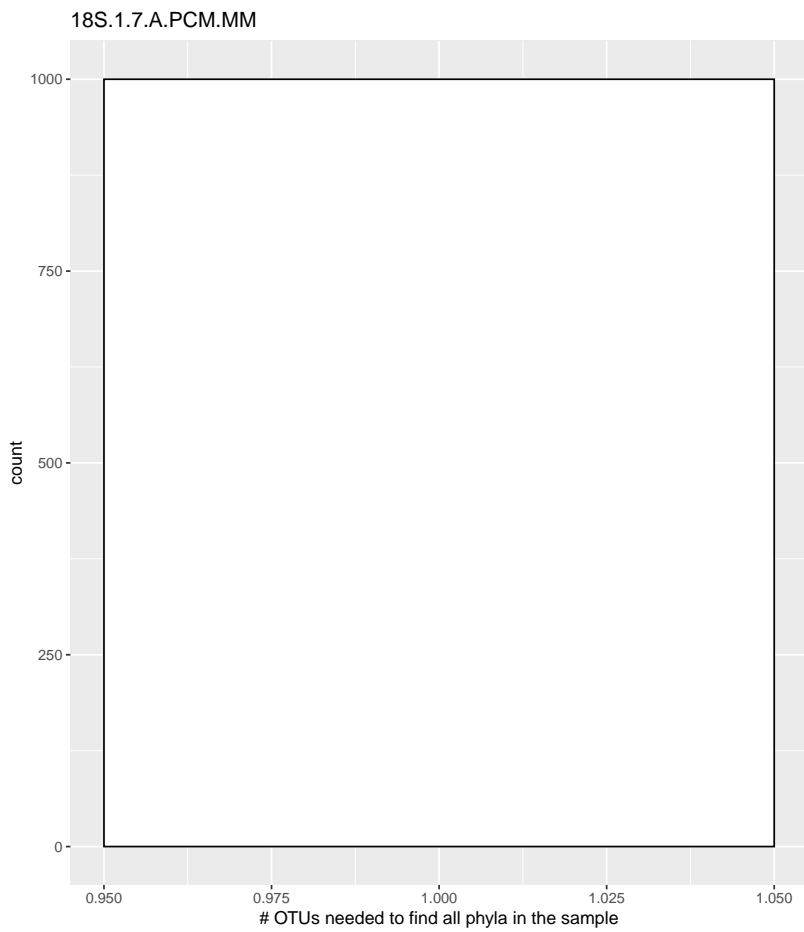
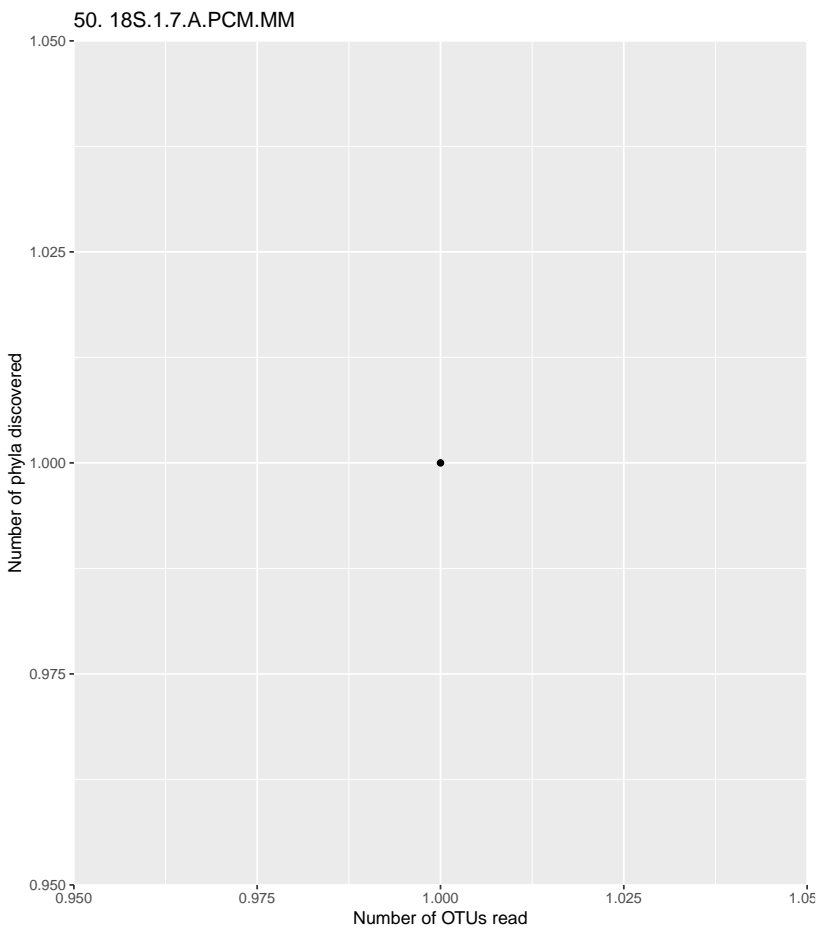


49. 18S.1.6.H.PCM.ME

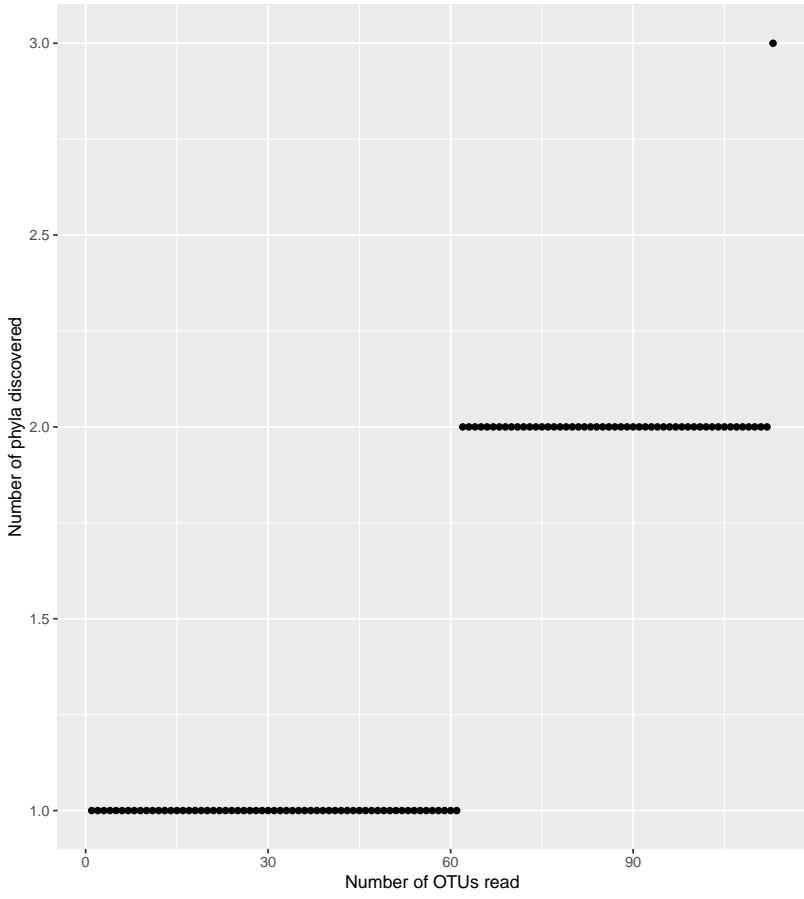


18S.1.6.H.PCM.ME

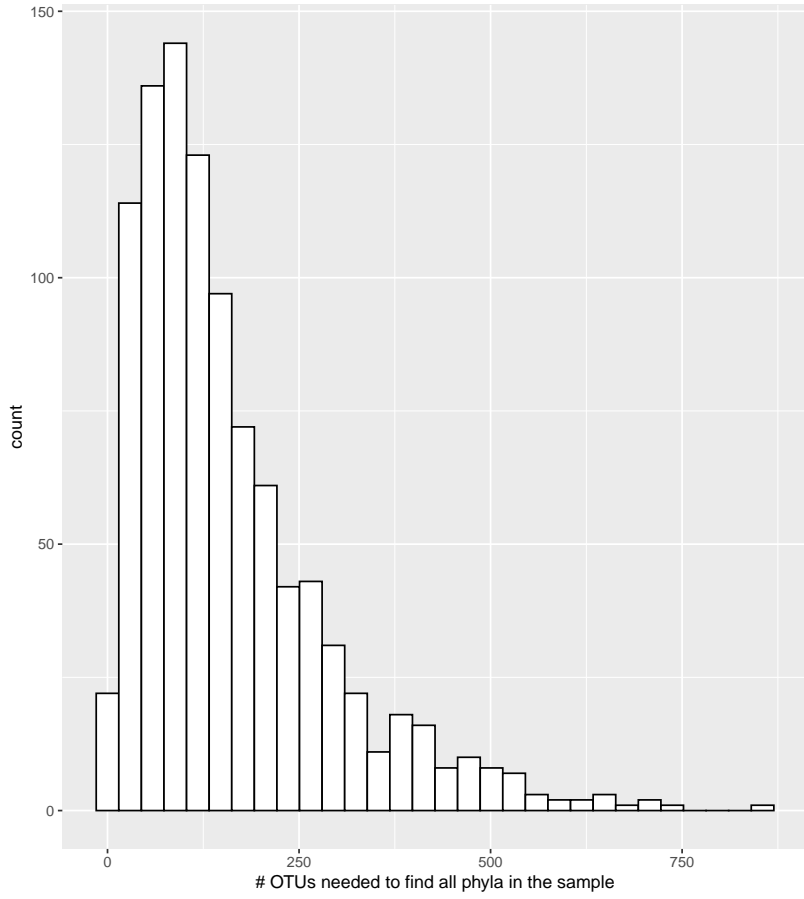




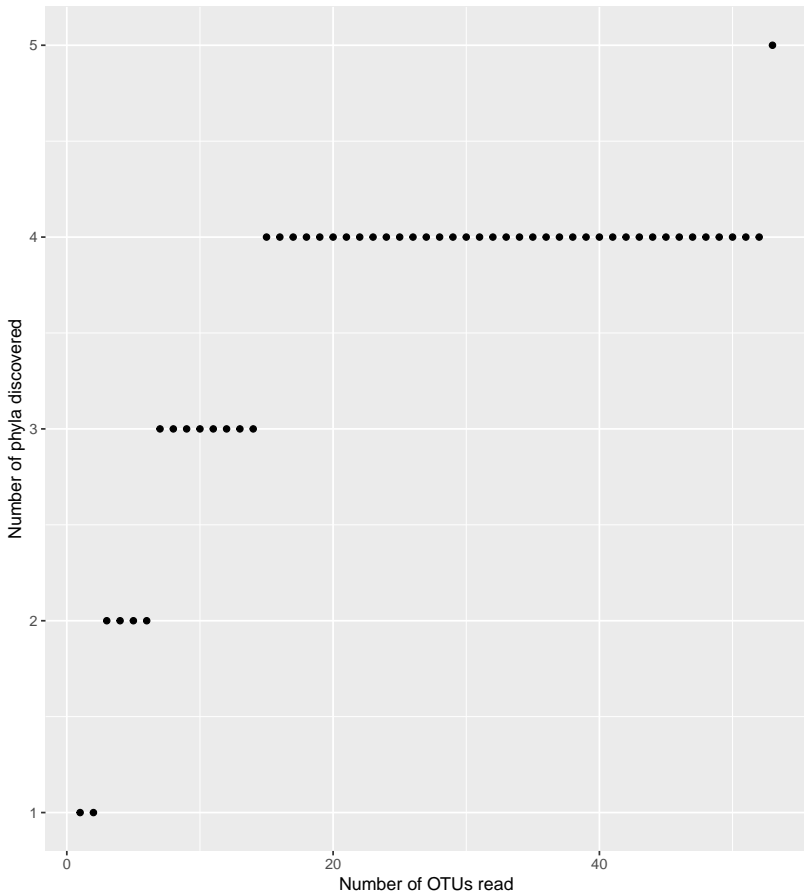
51. 18S.1.7.B.PCM.MM



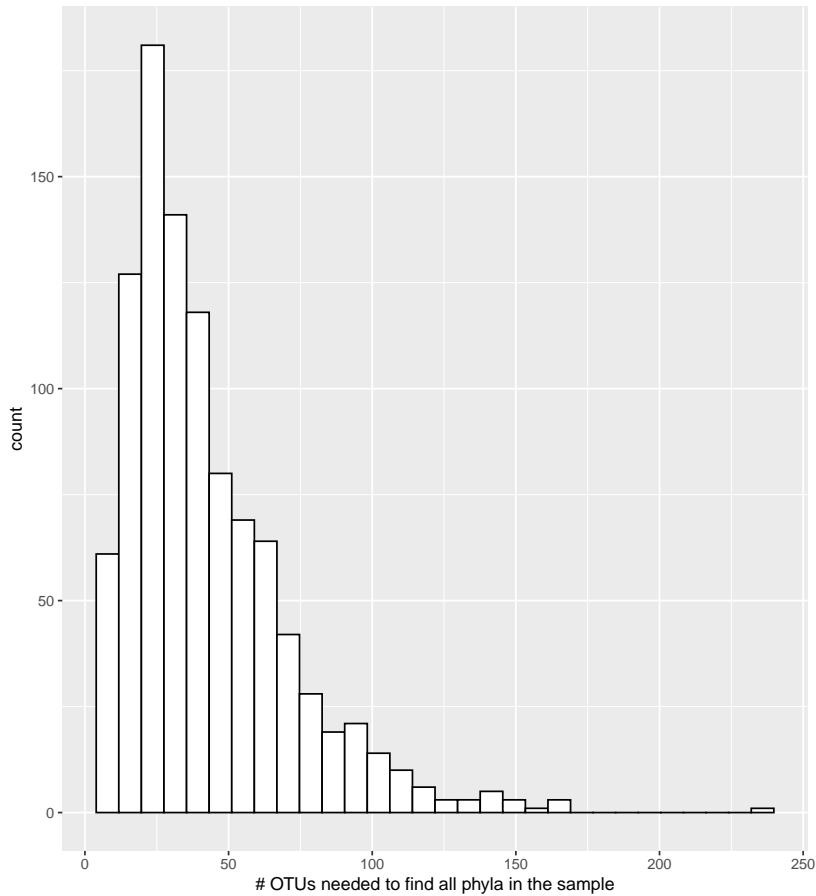
18S.1.7.B.PCM.MM



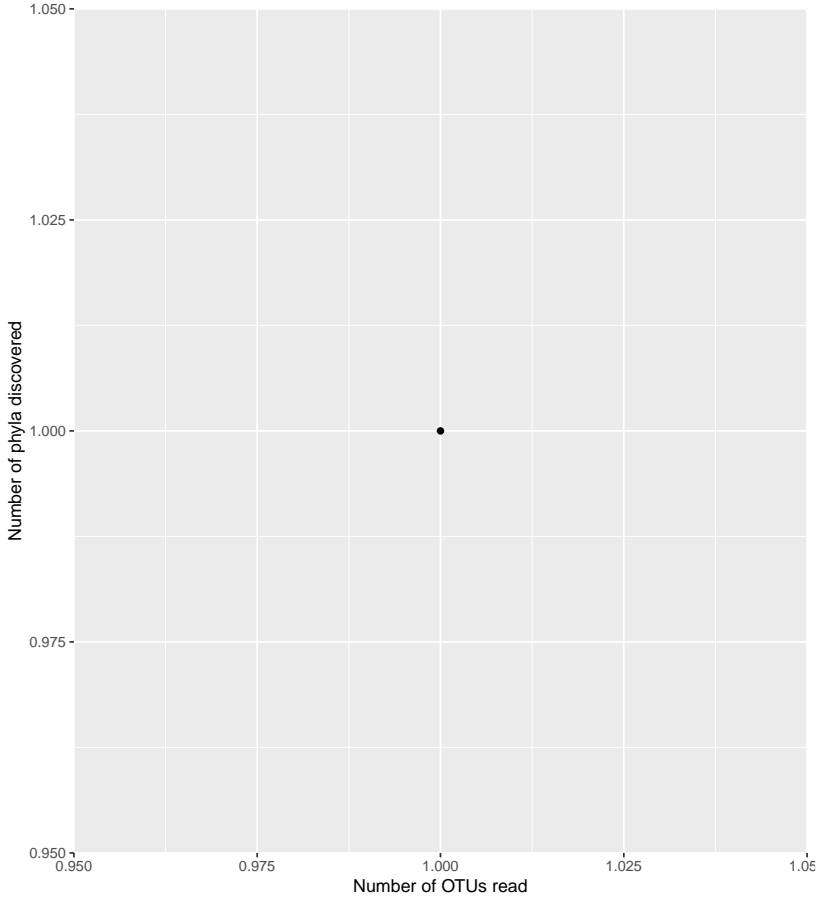
52. 18S.1.7.C.PCM.MM



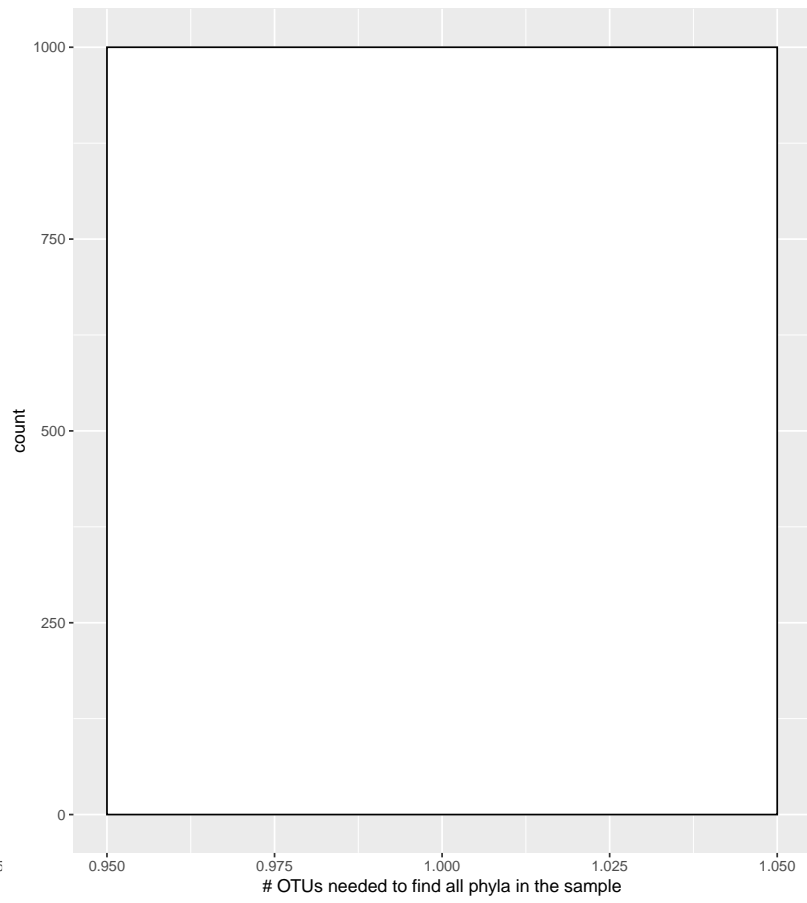
18S.1.7.C.PCM.MM



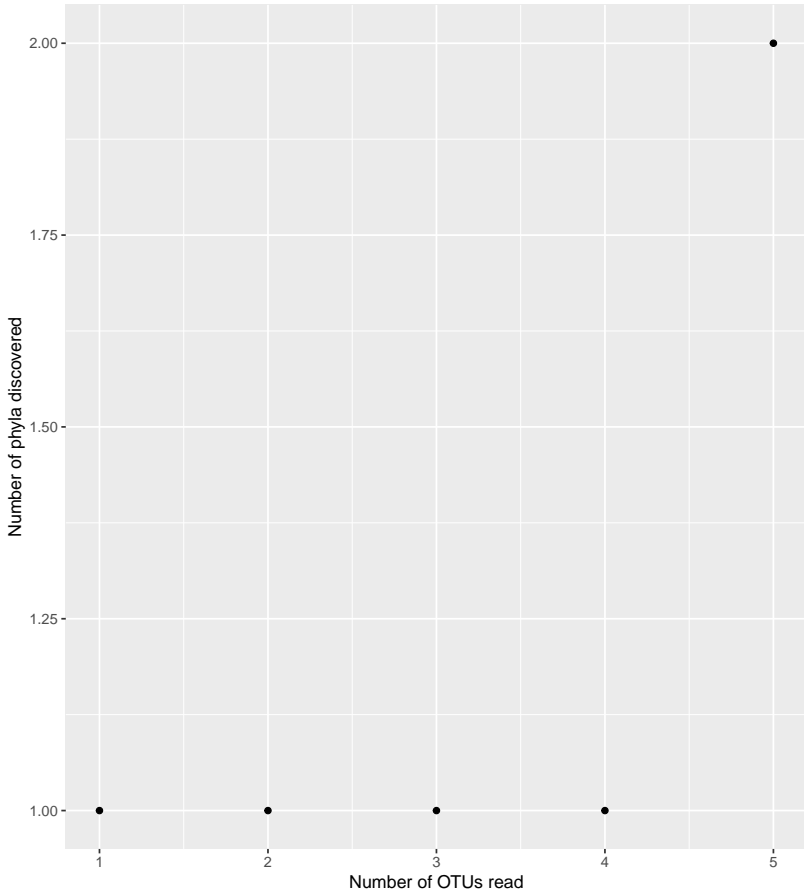
53. 18S.1.7.D.PCM.ME



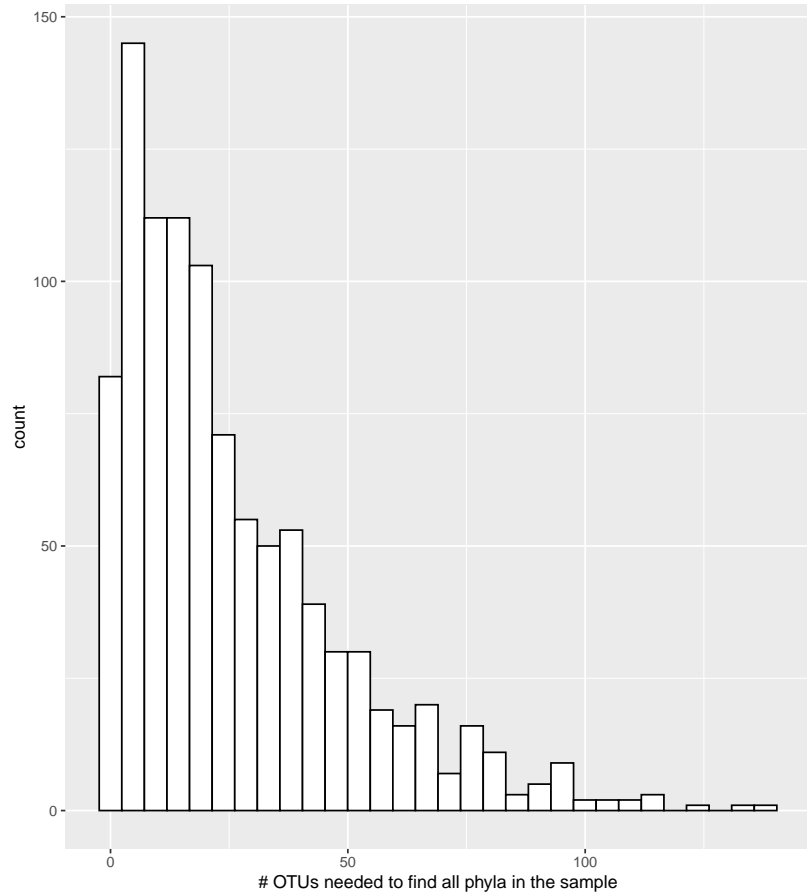
18S.1.7.D.PCM.ME



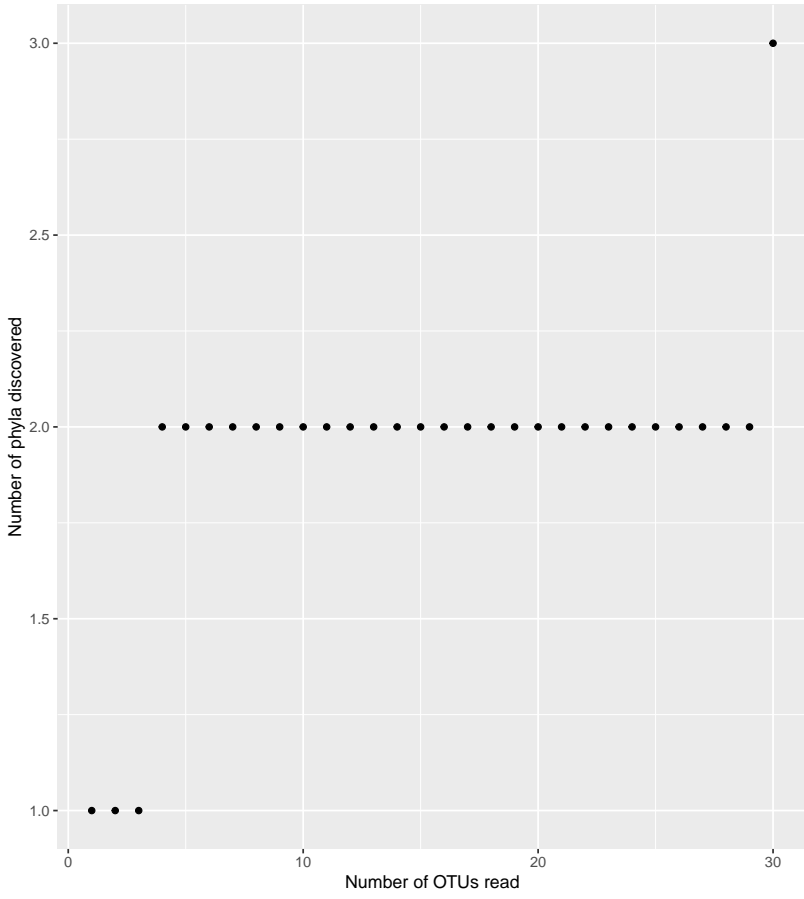
54. 18S.1.7.E.PCM.ME



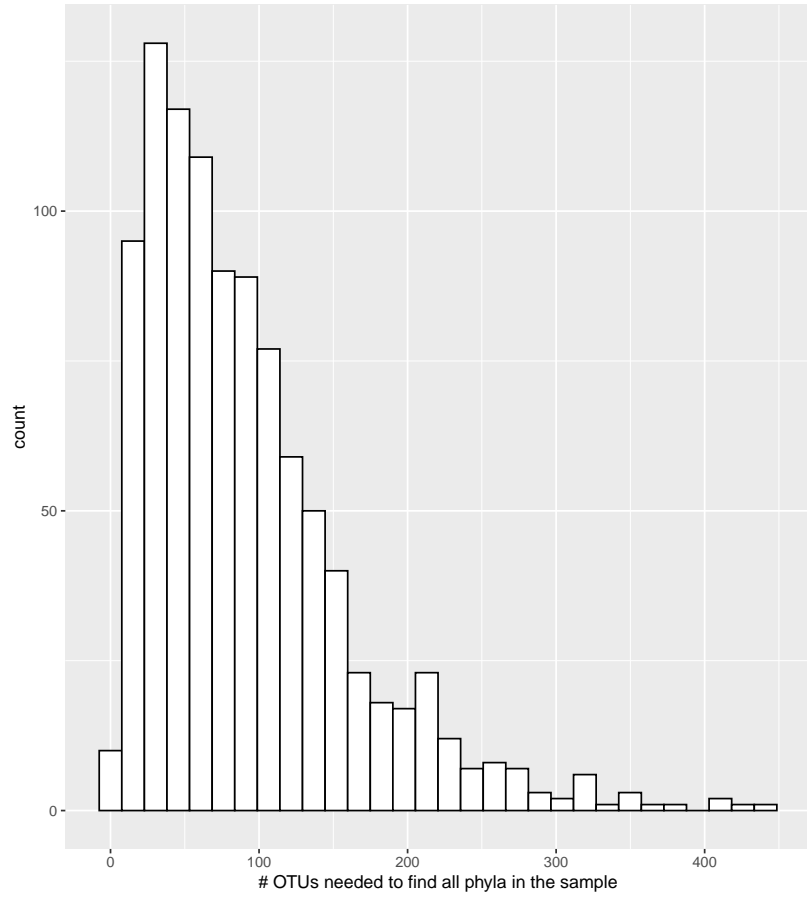
18S.1.7.E.PCM.ME



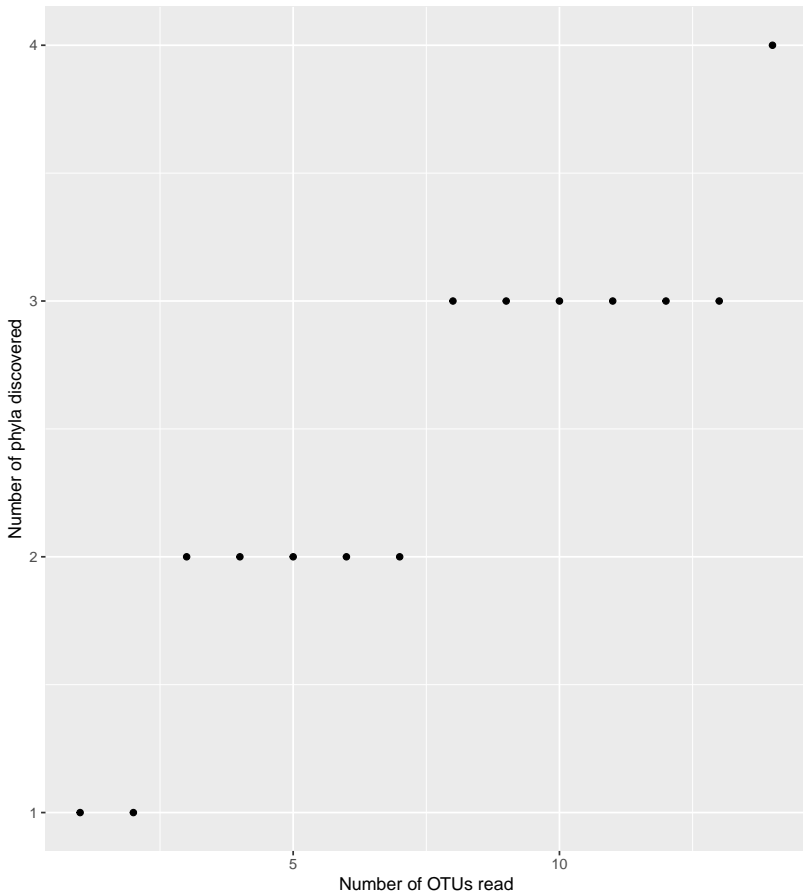
55. 18S.1.7.F.PCM.ME



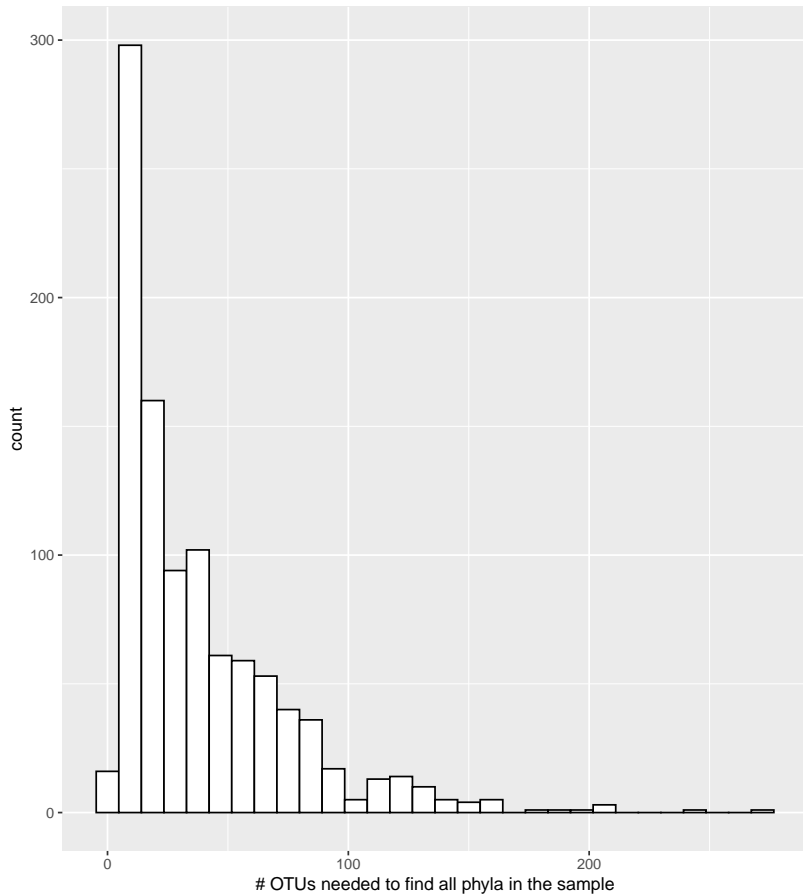
18S.1.7.F.PCM.ME



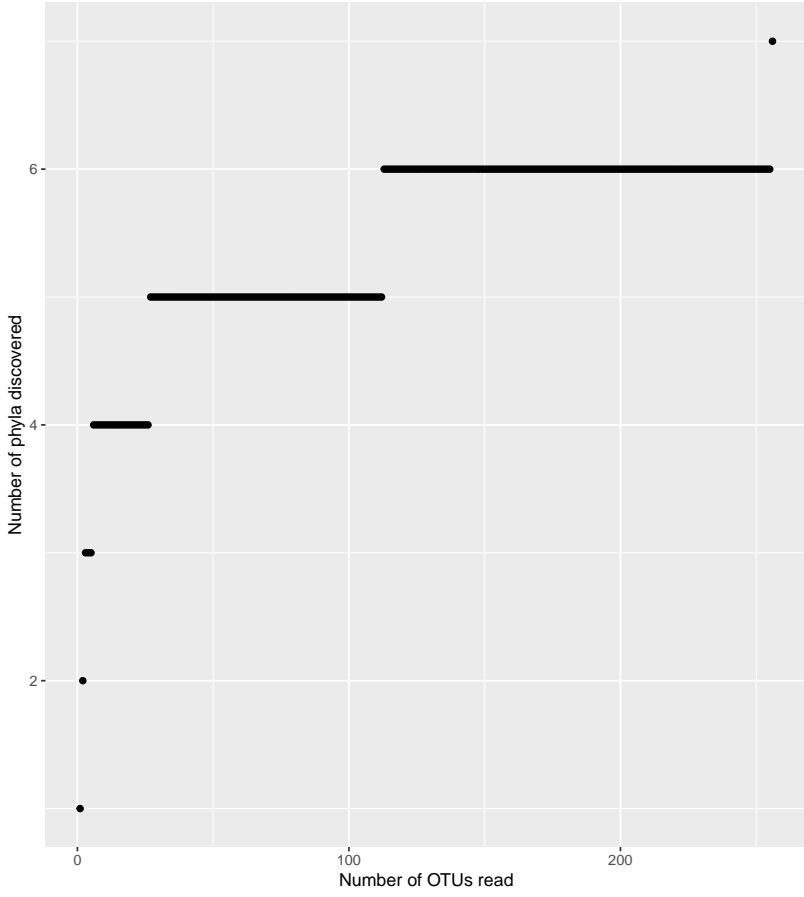
56. 18S.1.7.G.PCM.ME



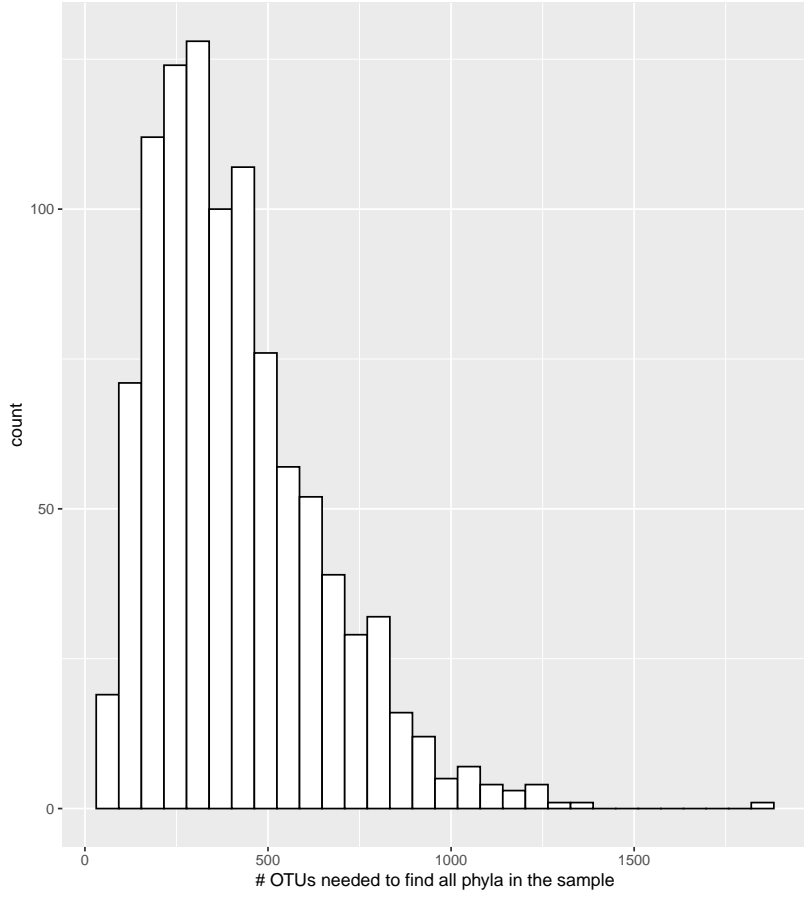
18S.1.7.G.PCM.ME



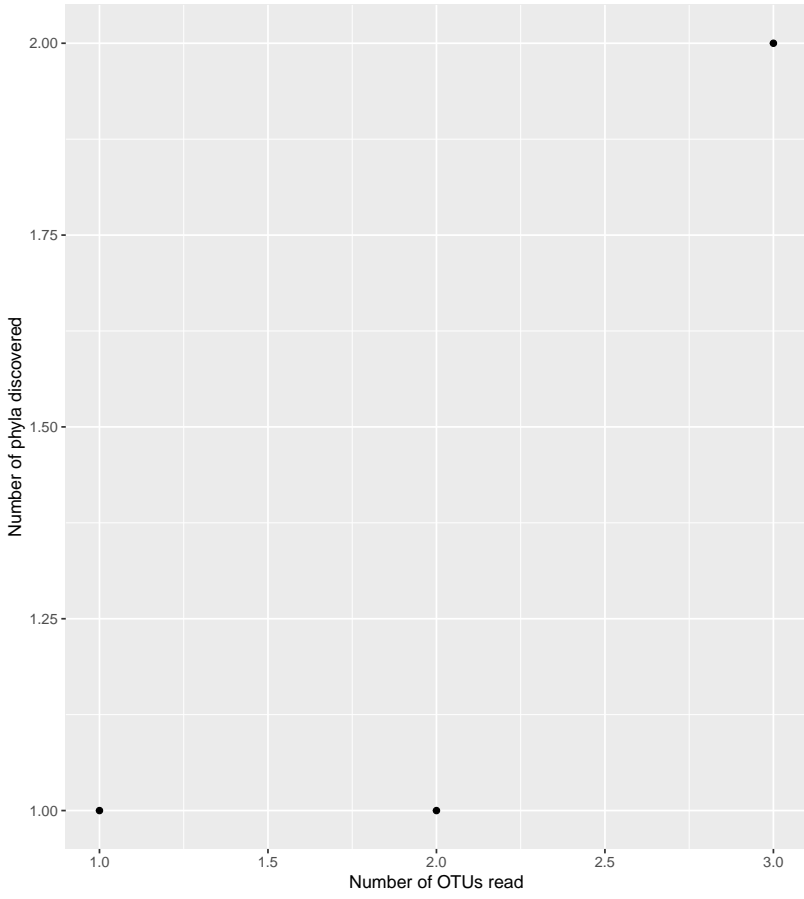
57. 18S.1.7.H.PCM.ME



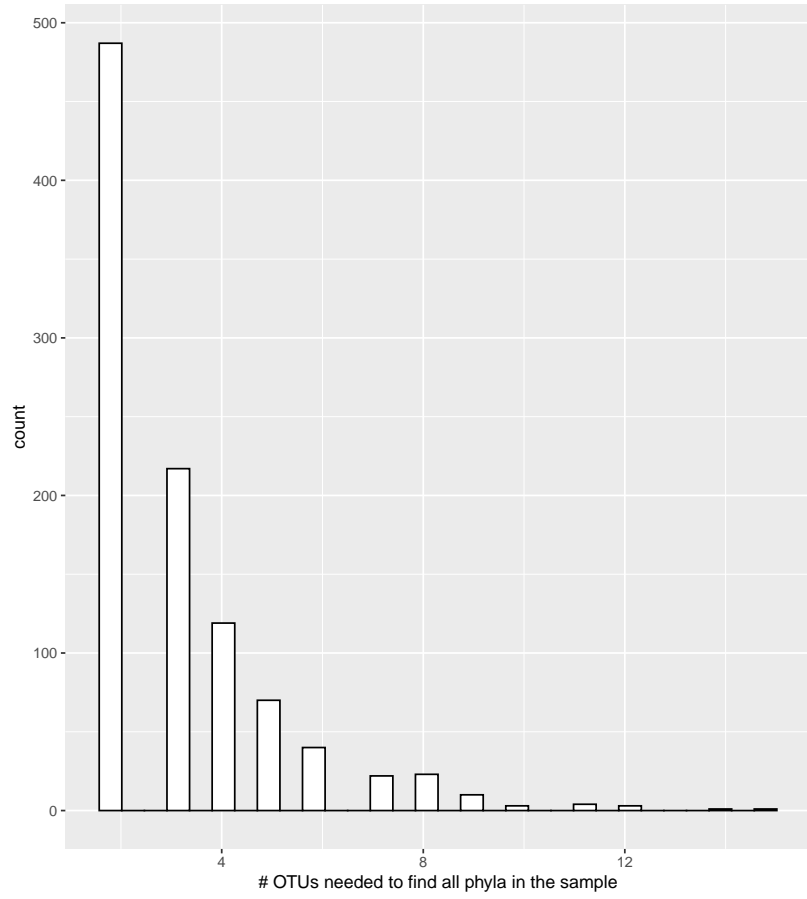
18S.1.7.H.PCM.ME



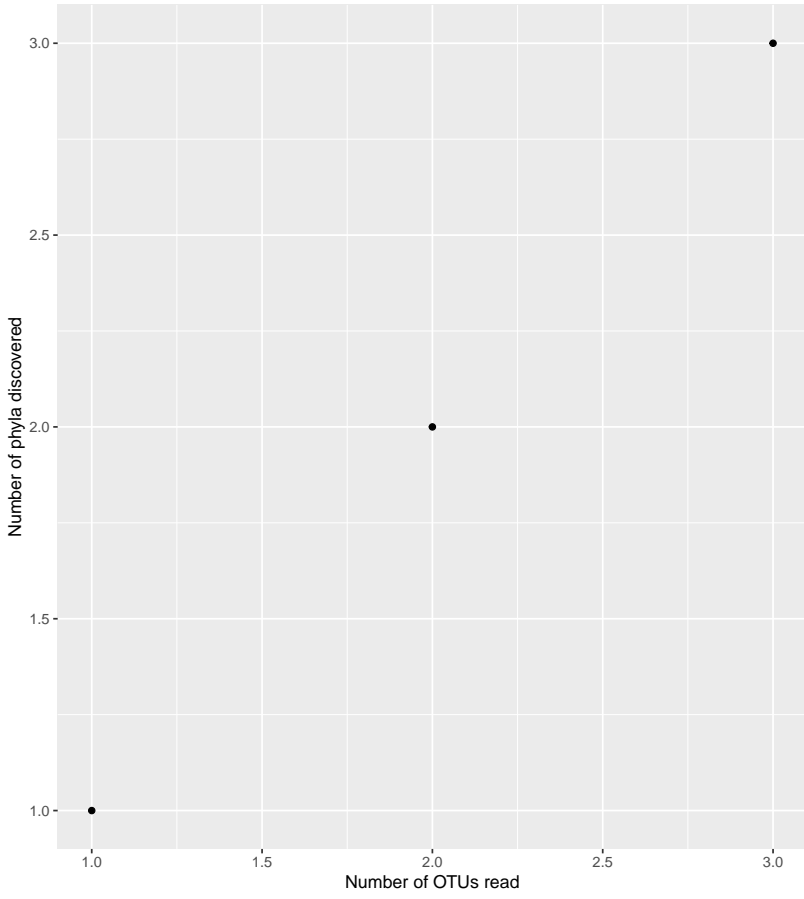
58. 18S.1.8.A.PCM.MM



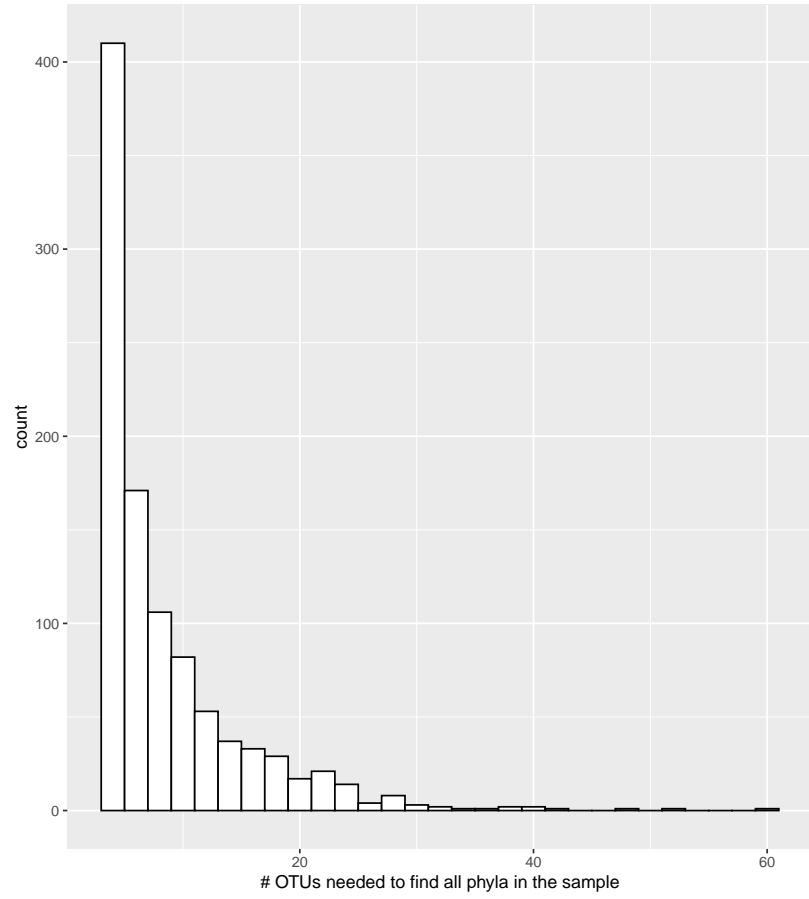
18S.1.8.A.PCM.MM



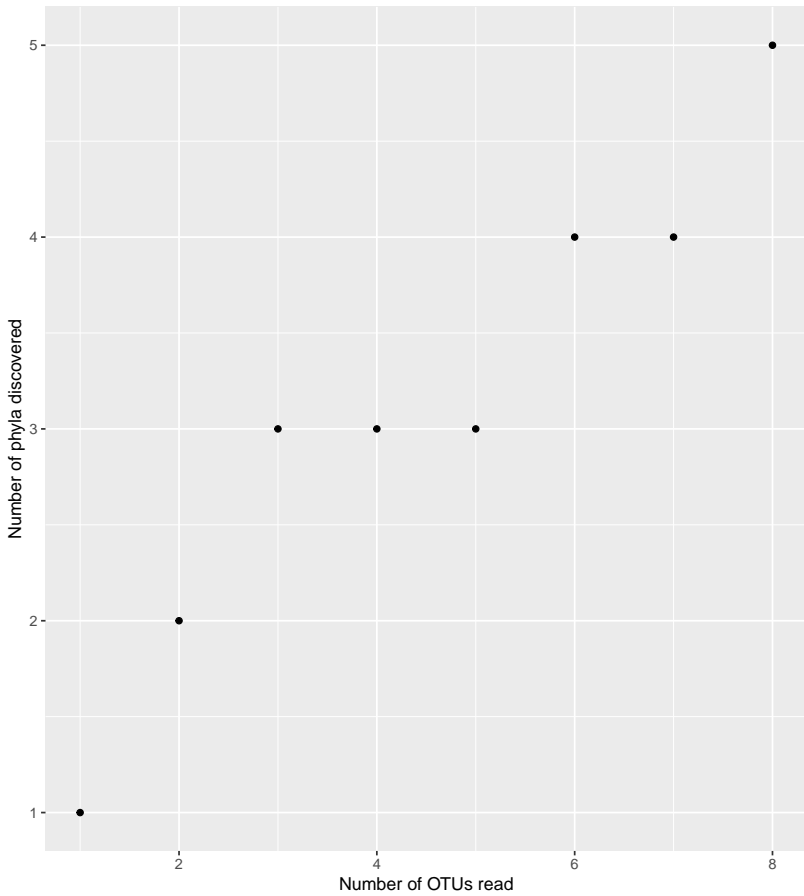
59. 18S.1.8.B.PCM.MM



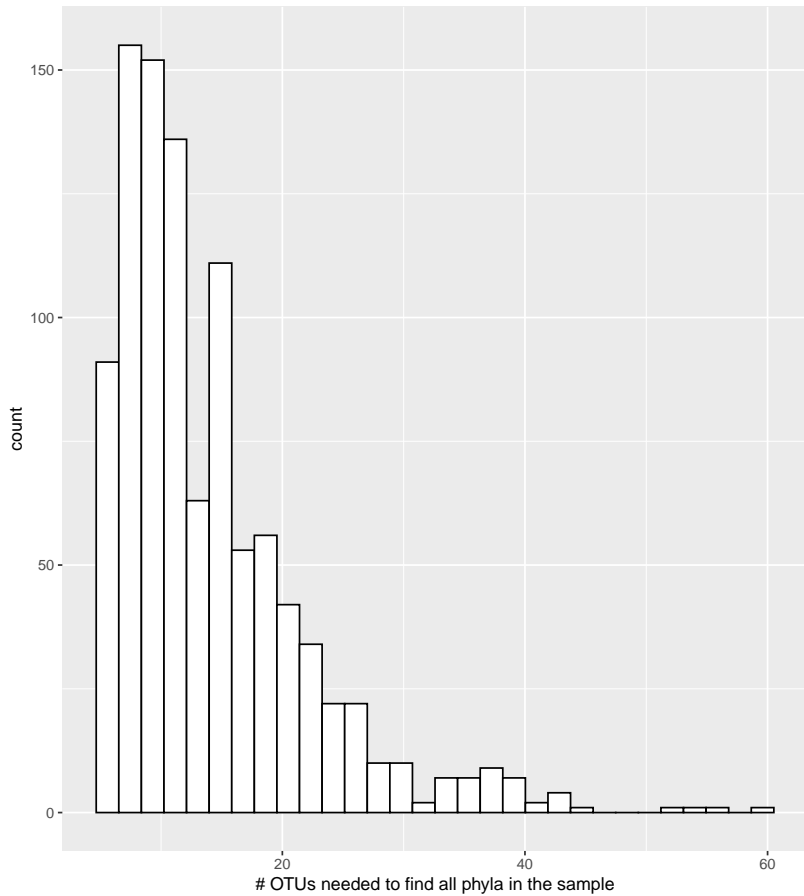
18S.1.8.B.PCM.MM



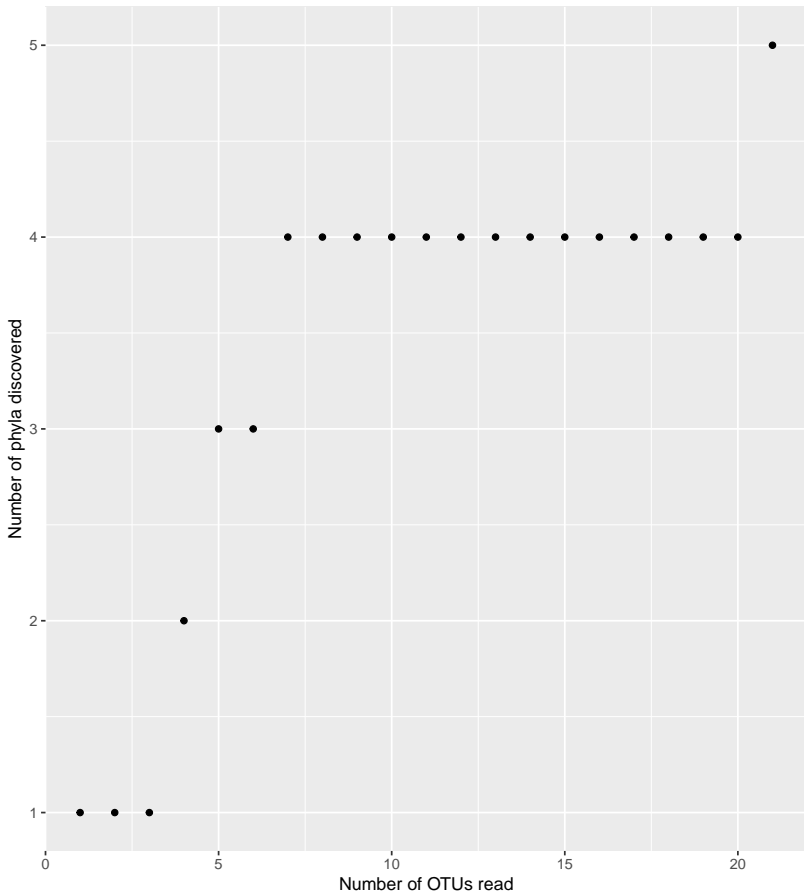
60. 18S.1.8.D.PCM.ME



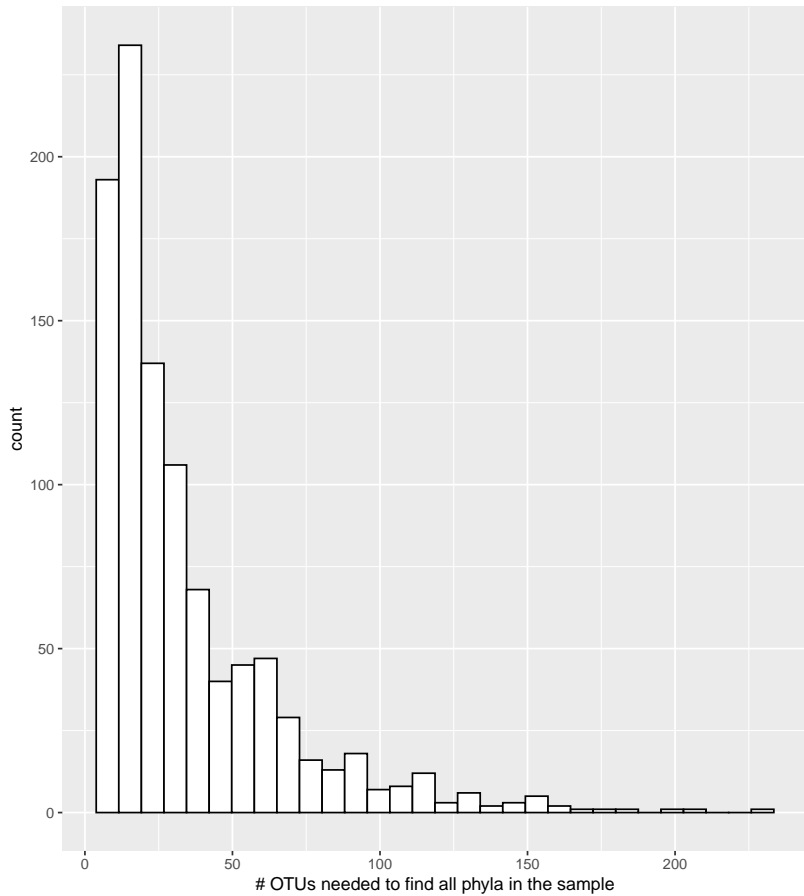
18S.1.8.D.PCM.ME



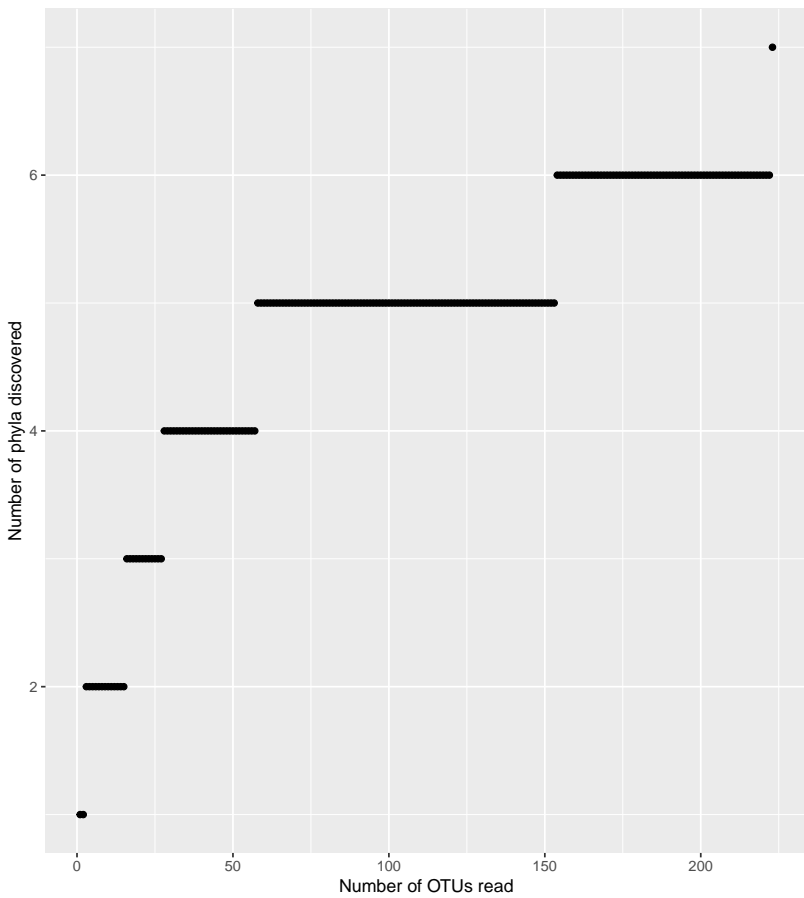
61. 18S.1.8.E.PCM.ME



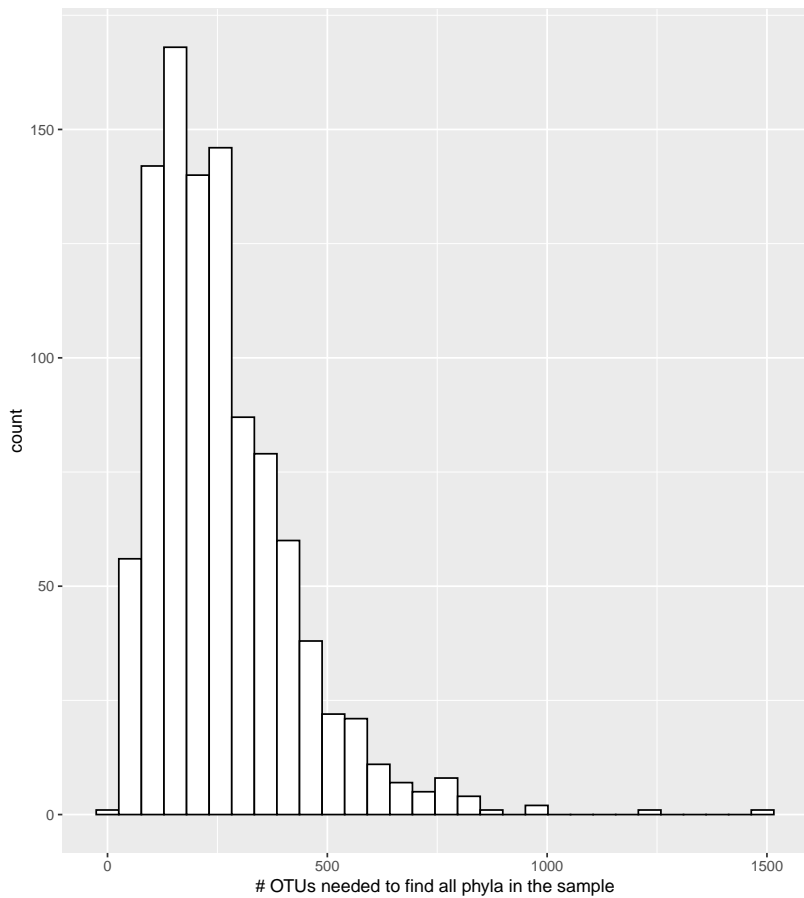
18S.1.8.E.PCM.ME



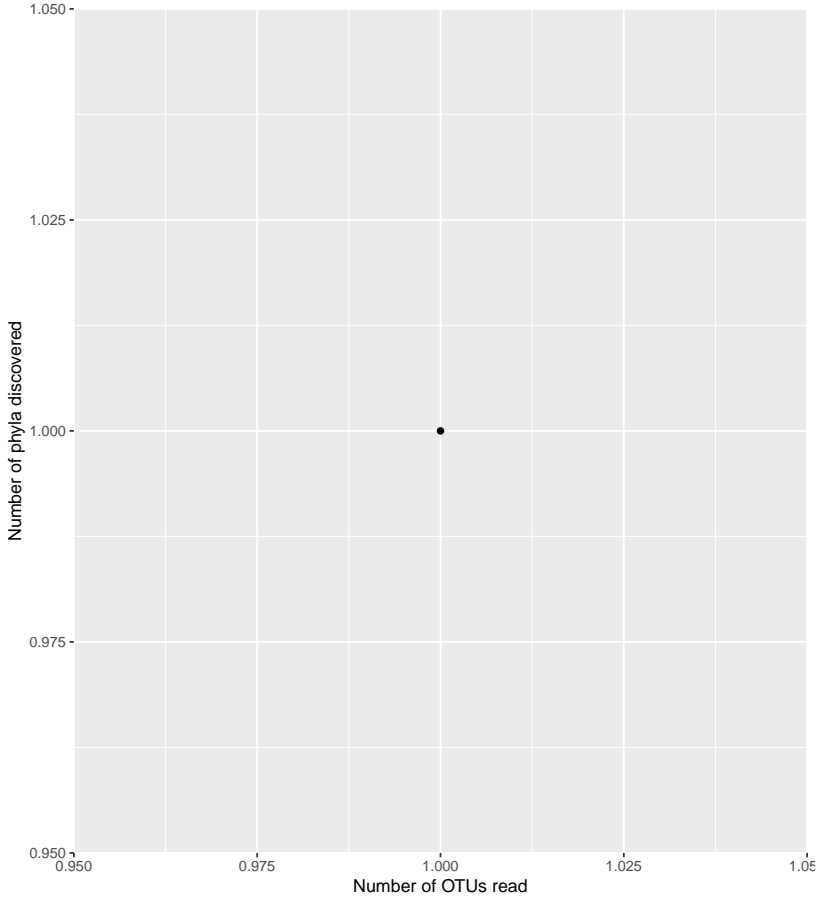
62. 18S.1.8.F.PCM.ME



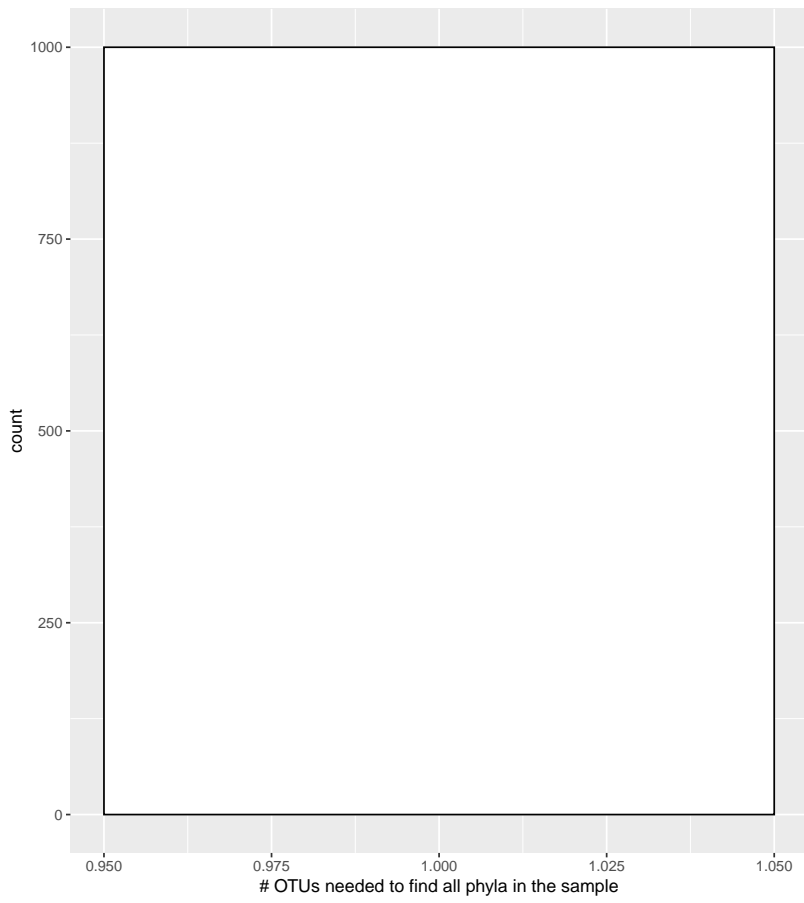
18S.1.8.F.PCM.ME



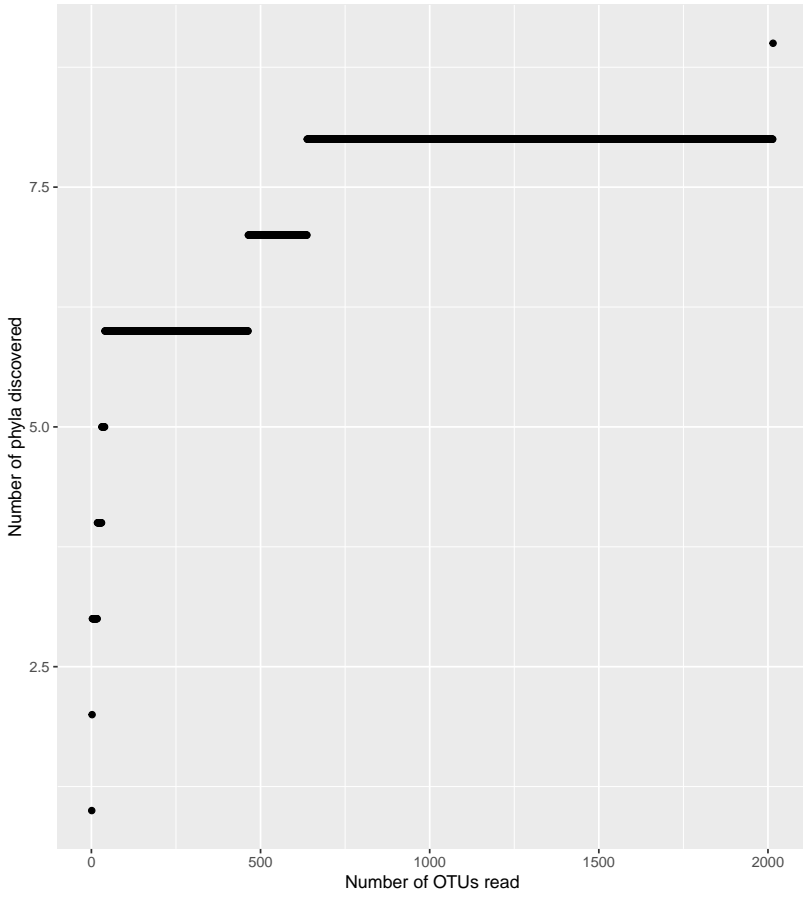
63. 18S.1.8.G.PCM.ME



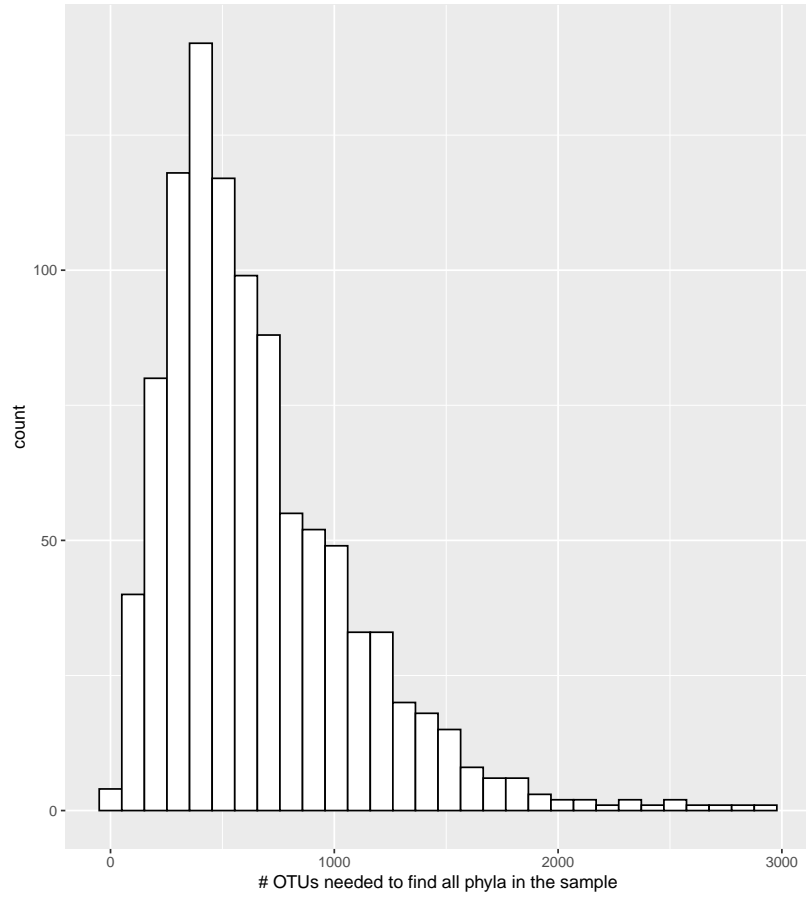
18S.1.8.G.PCM.ME



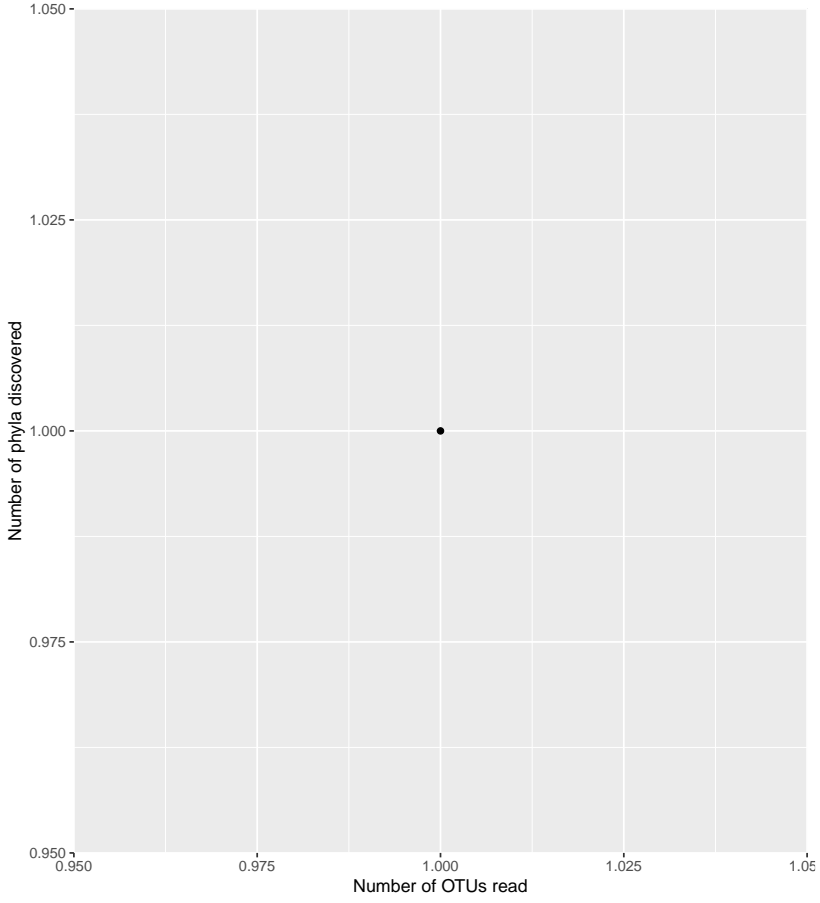
64. 18S.1.8.H.PCM.ME



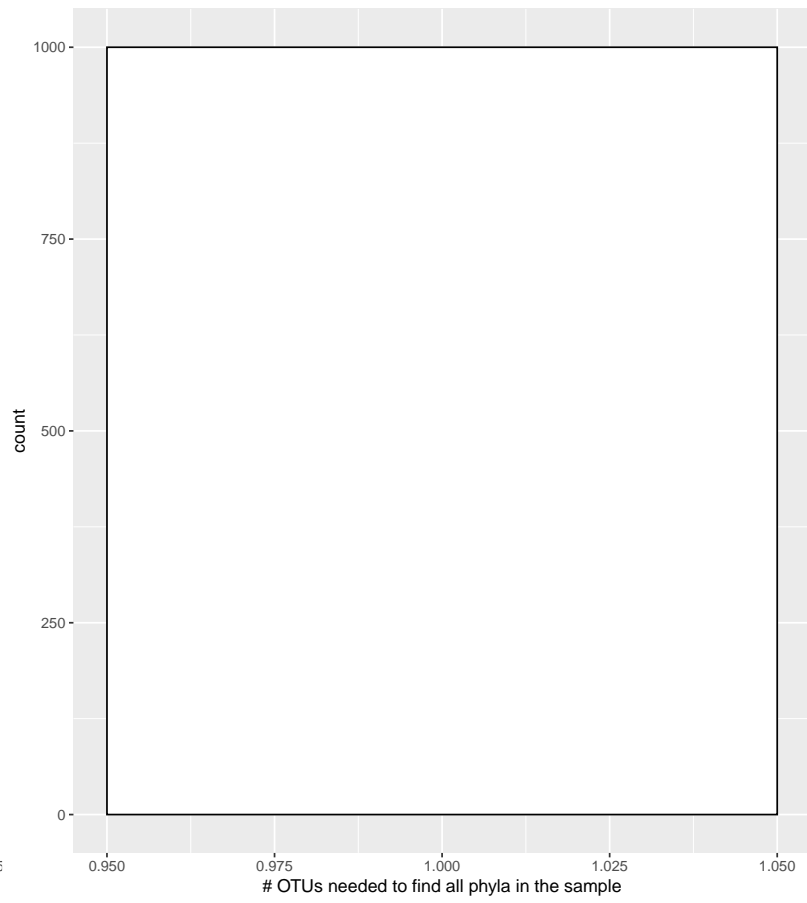
18S.1.8.H.PCM.ME



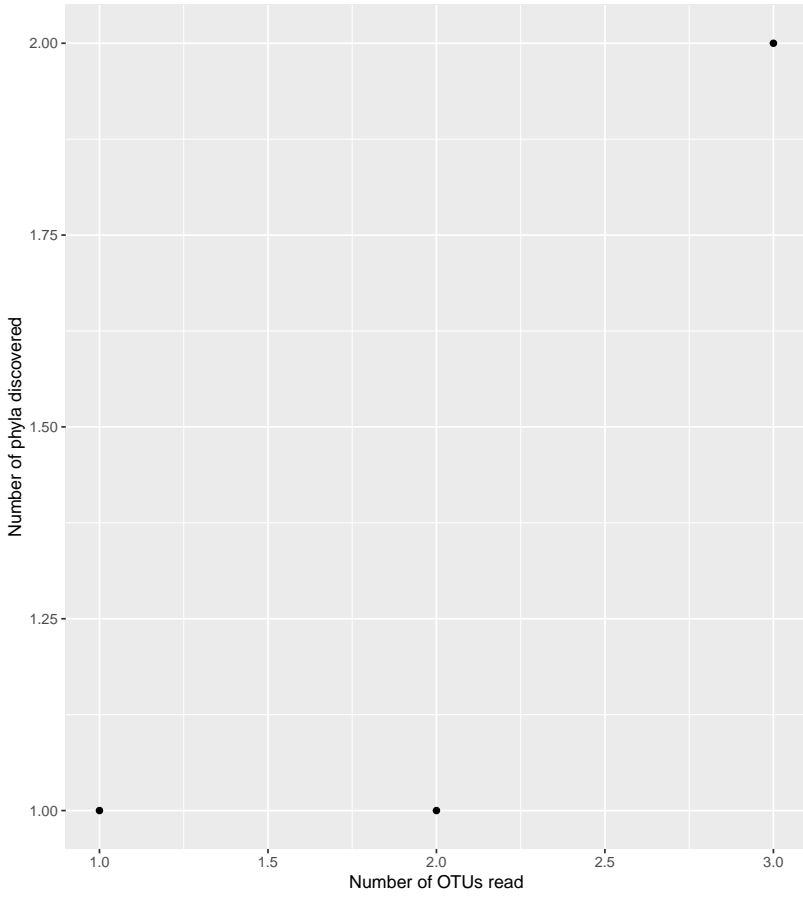
65. 18S.1.9.A.PCM.MM



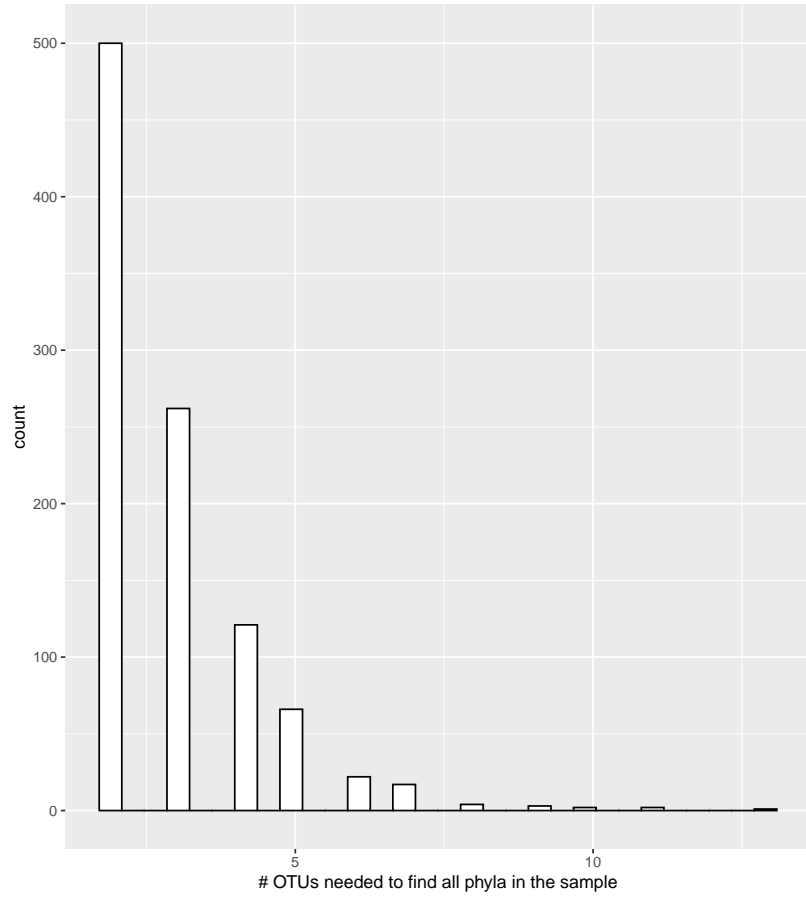
18S.1.9.A.PCM.MM



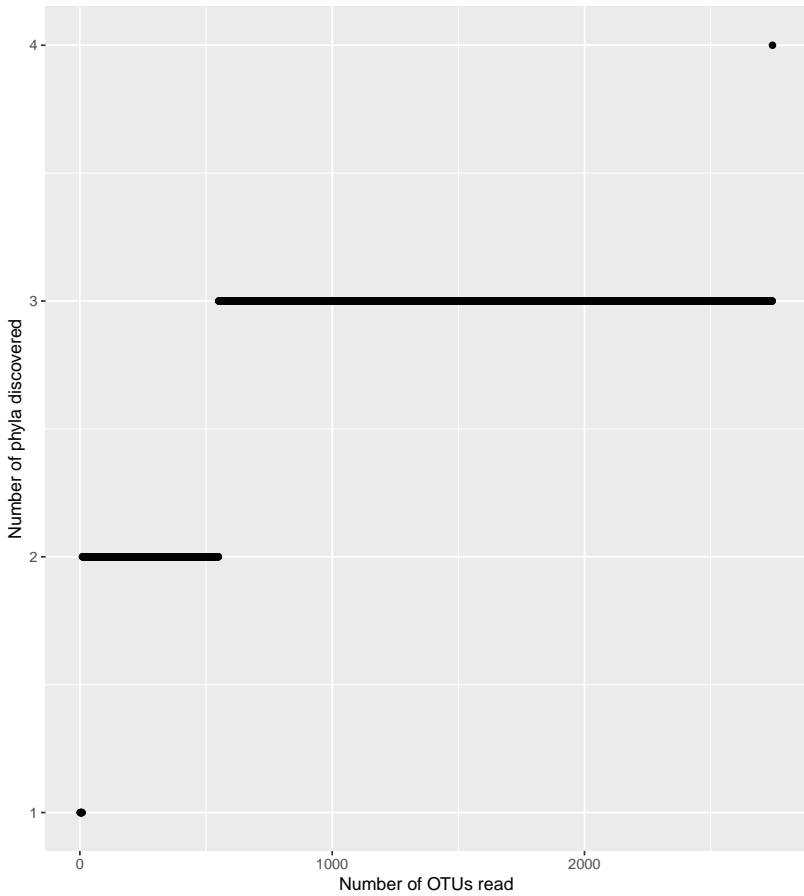
66. 18S.1.9.B.PCM.MM



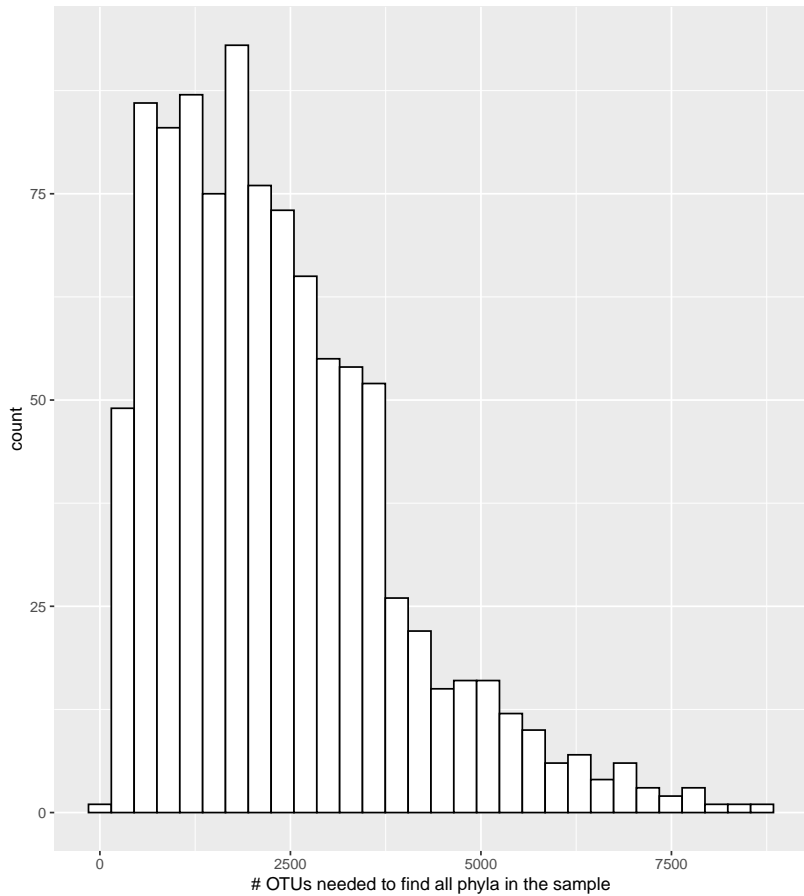
18S.1.9.B.PCM.MM



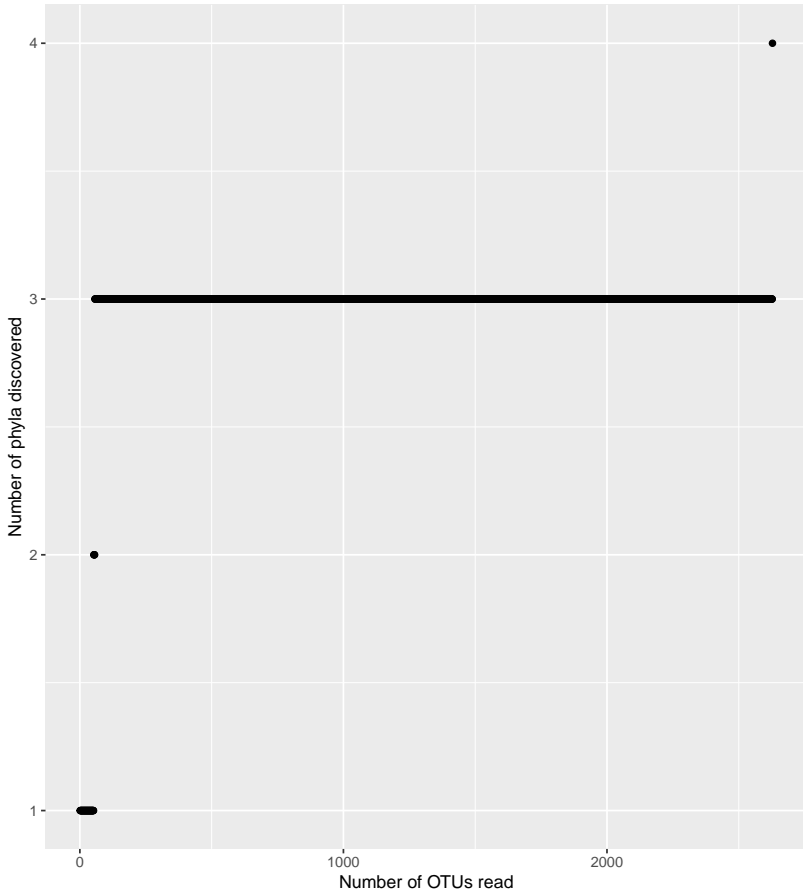
67. 18S.1.9.D.PCM.ME



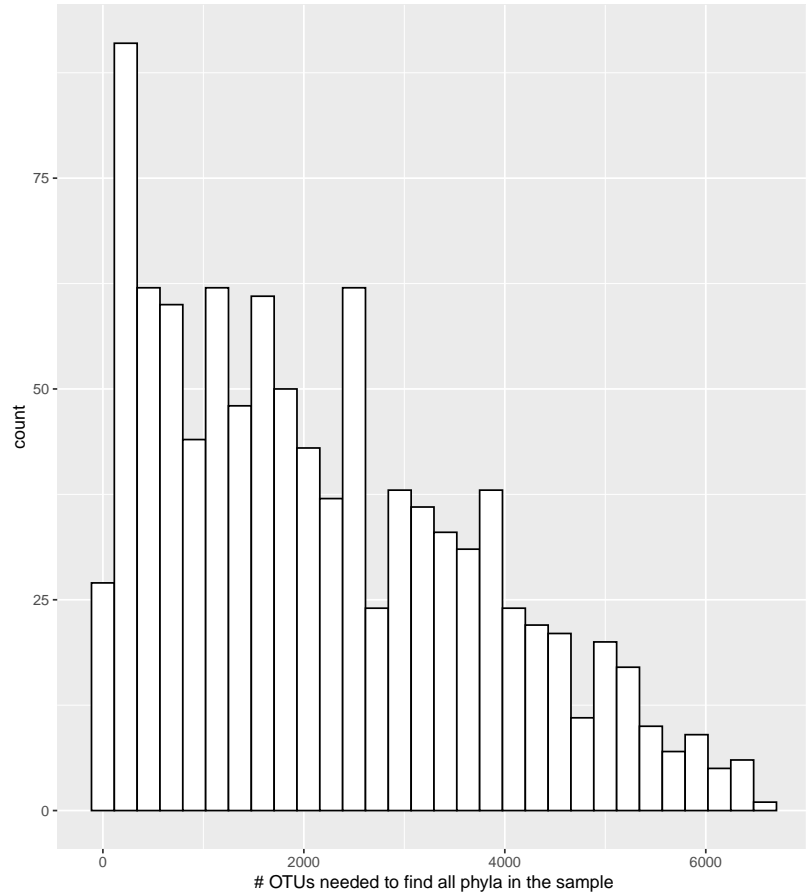
18S.1.9.D.PCM.ME



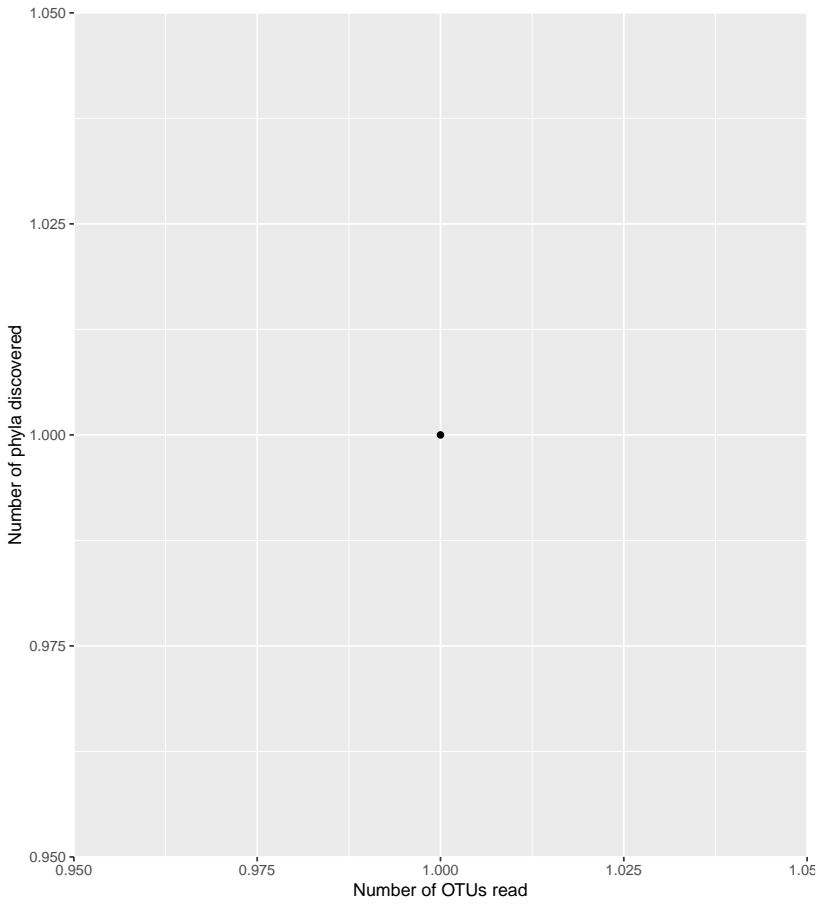
68. 18S.1.9.E.PCM.ME



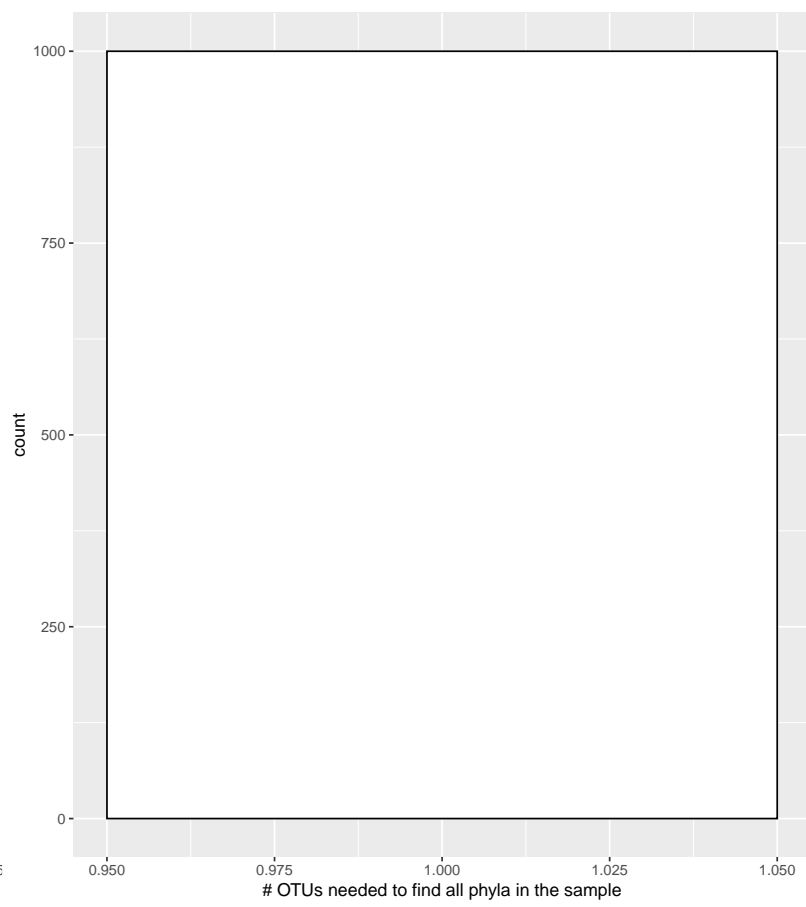
18S.1.9.E.PCM.ME



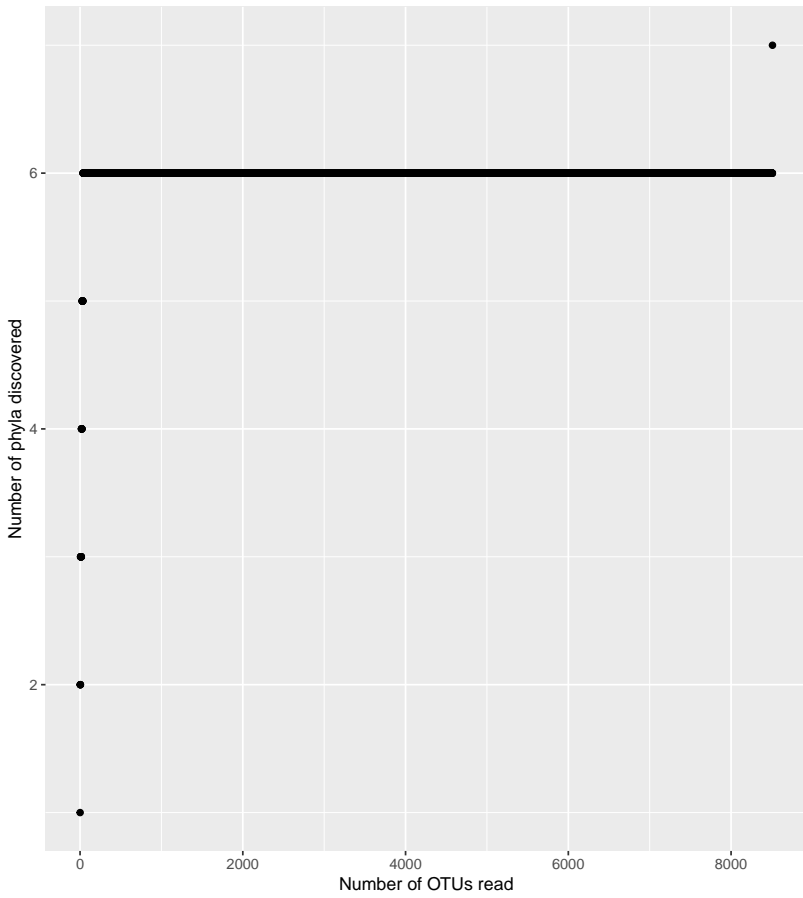
69. 18S.1.9.F.PCM.ME



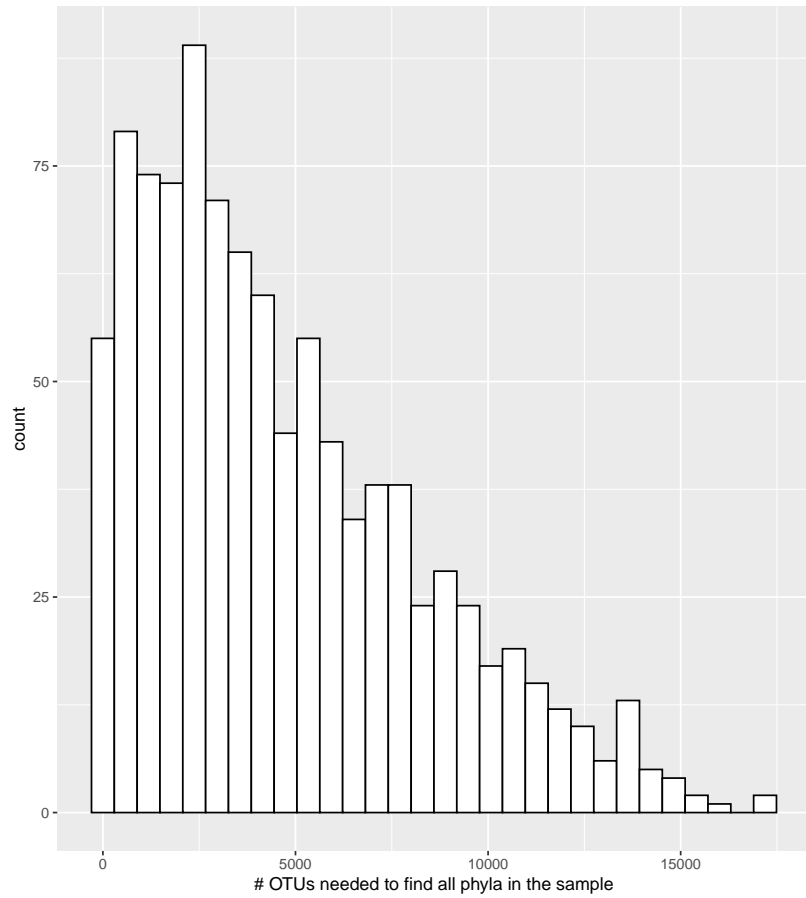
18S.1.9.F.PCM.ME



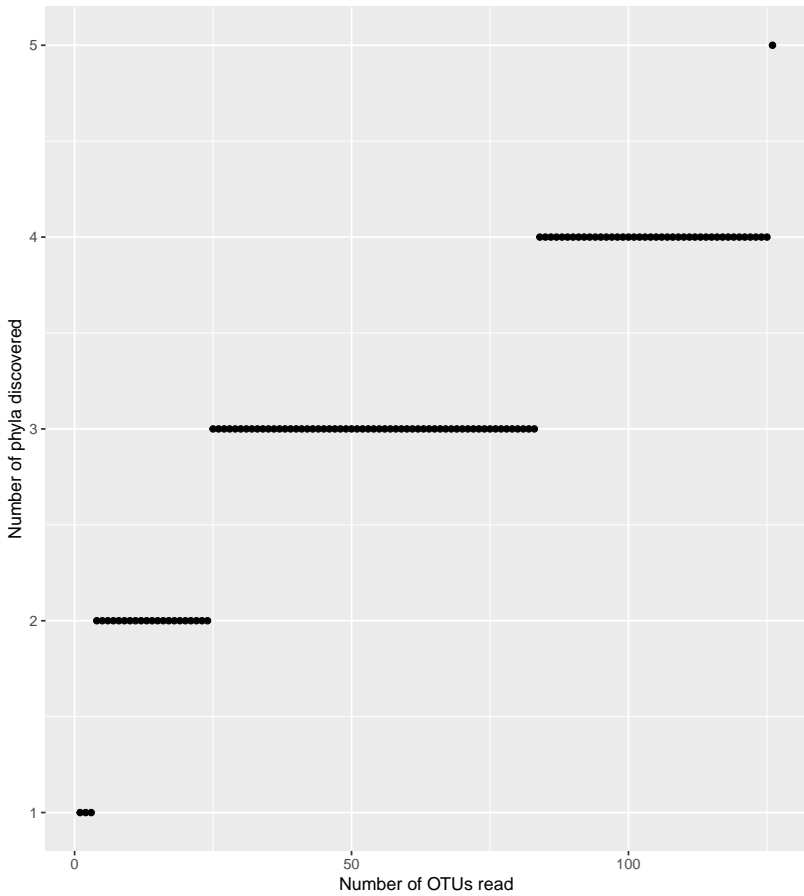
70. 18S.1.9.G.PCM.ME



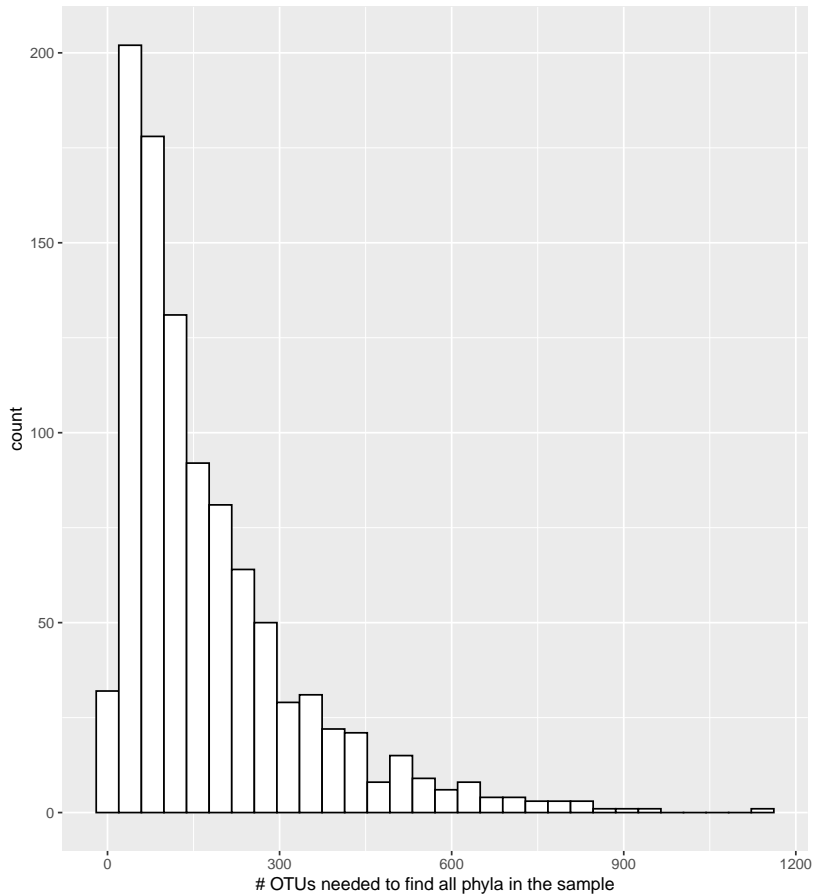
18S.1.9.G.PCM.ME



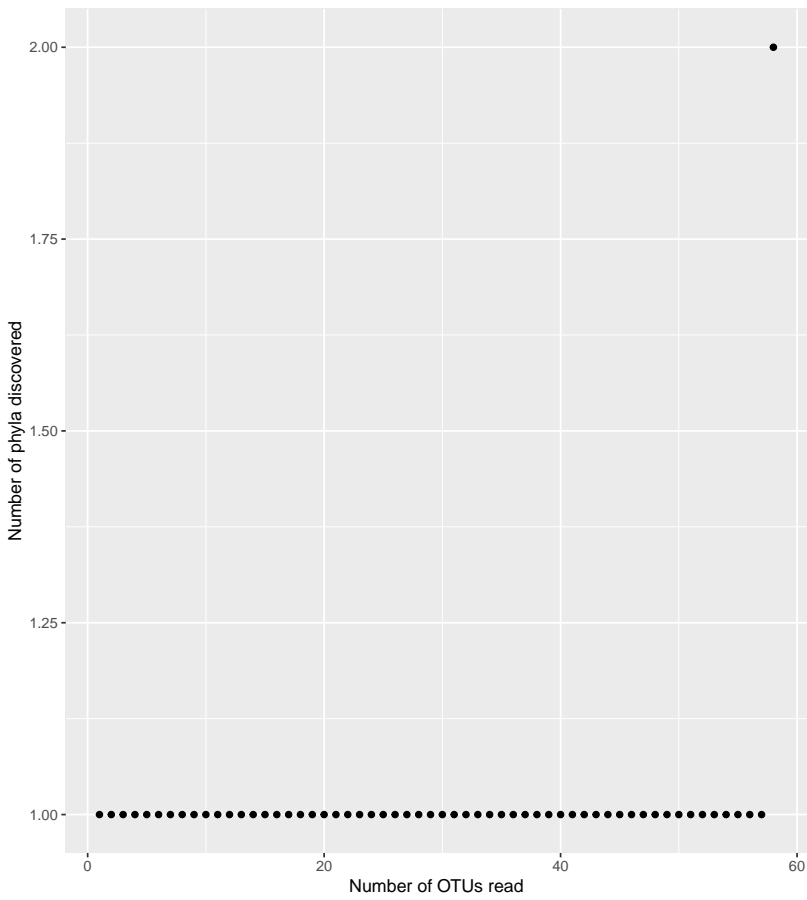
71. 18S.1.9.H.PCM.ME



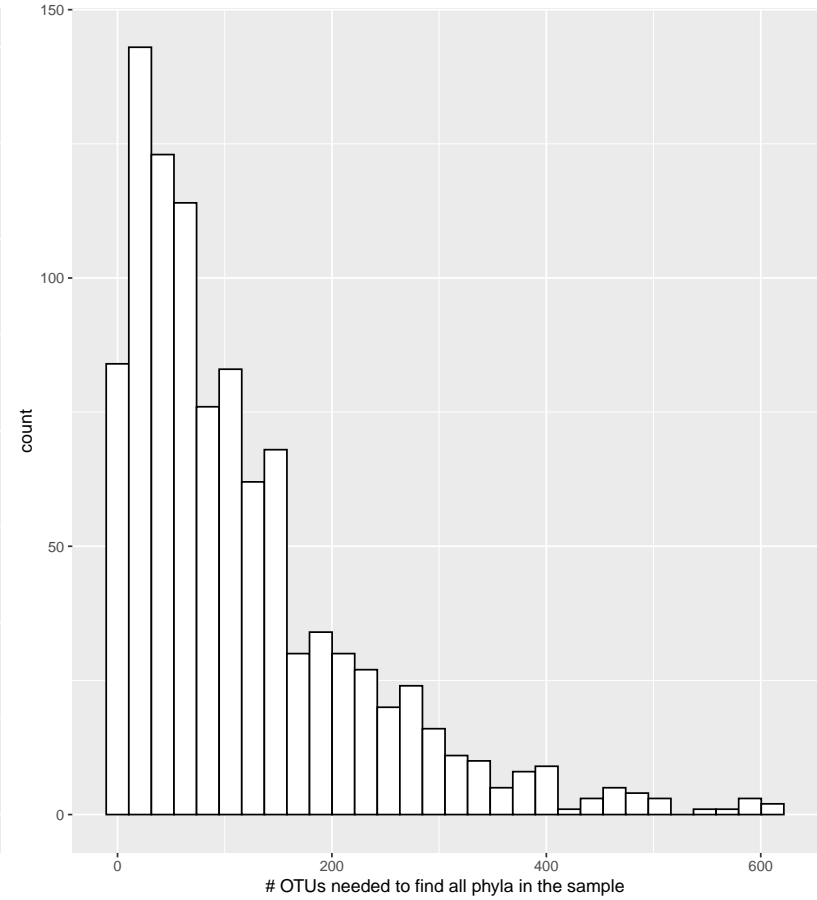
18S.1.9.H.PCM.ME



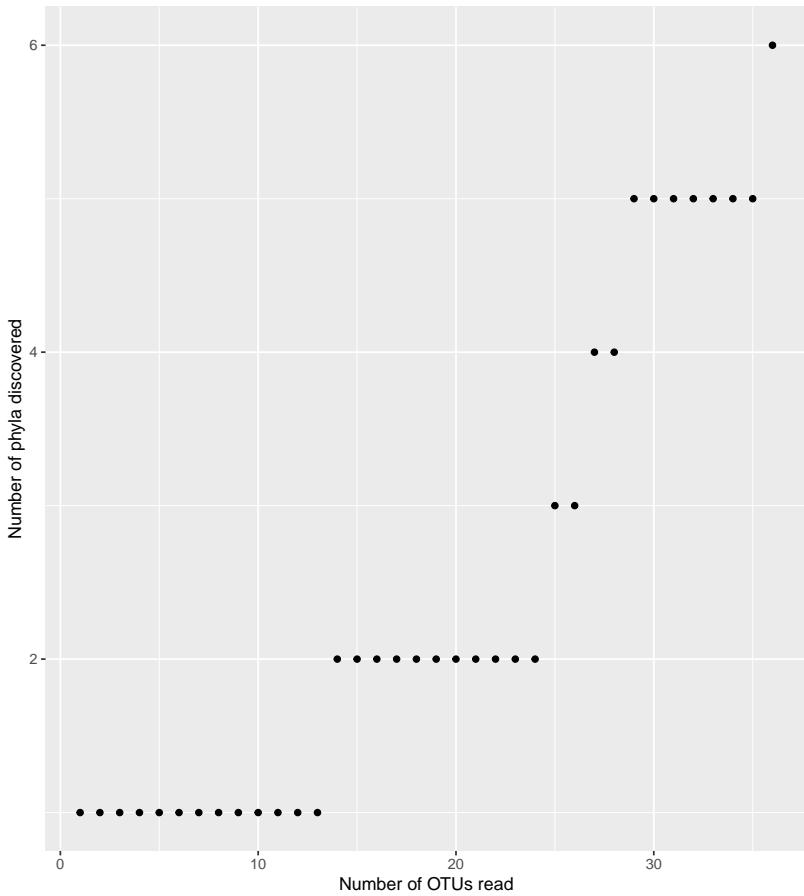
72. 18S.2.1.A.PCM.ME



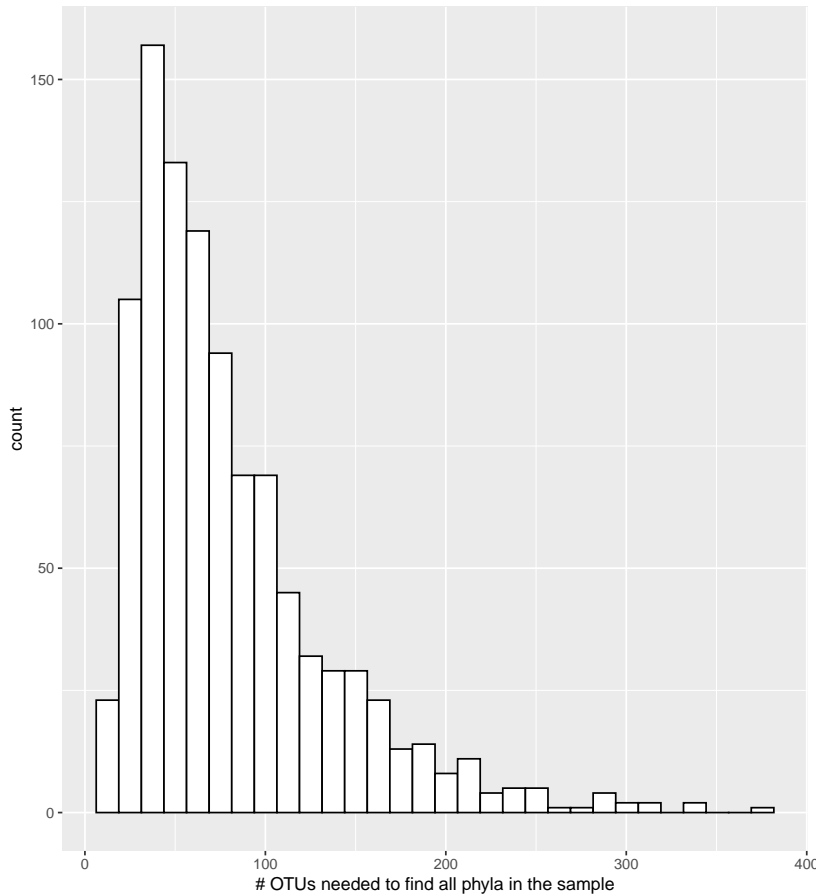
18S.2.1.A.PCM.ME



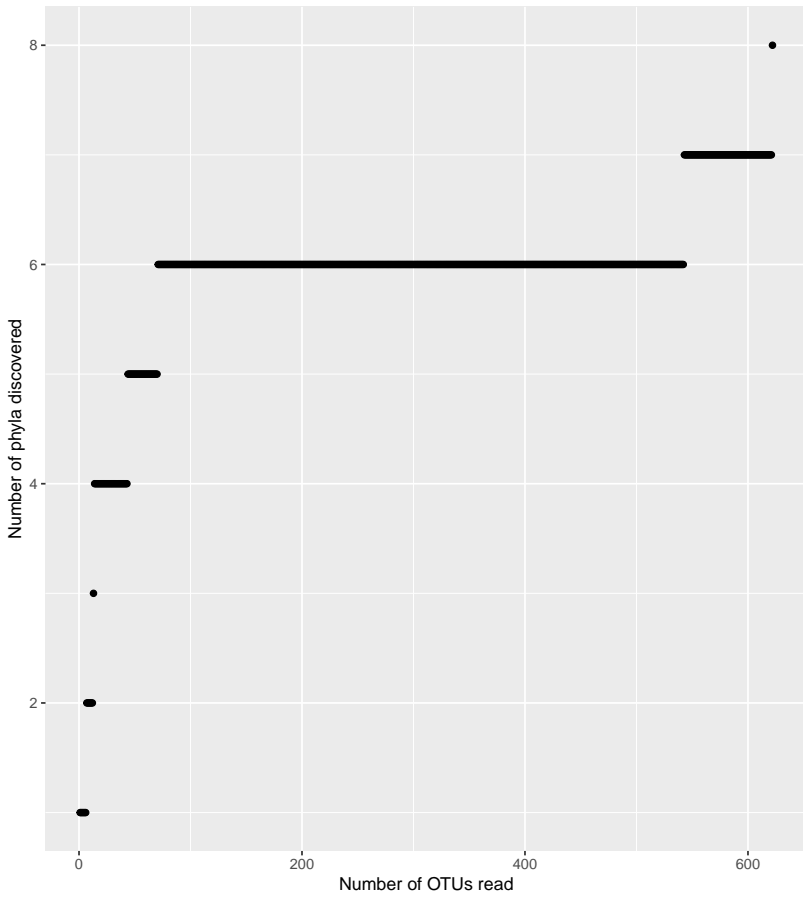
73. 18S.2.1.B.PCM.ME



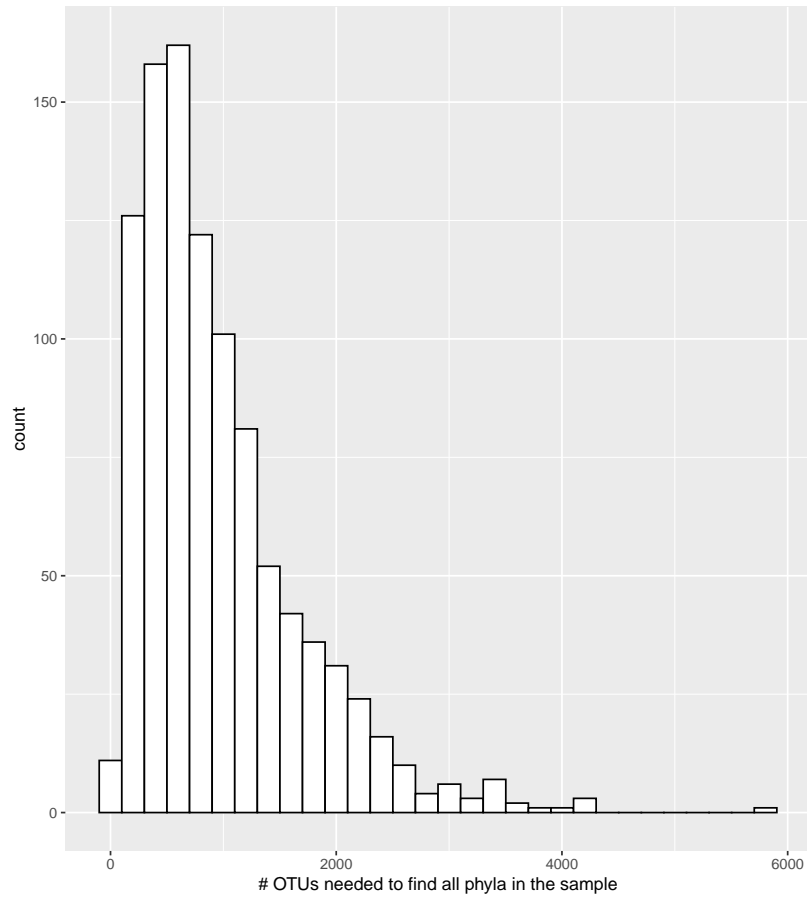
18S.2.1.B.PCM.ME



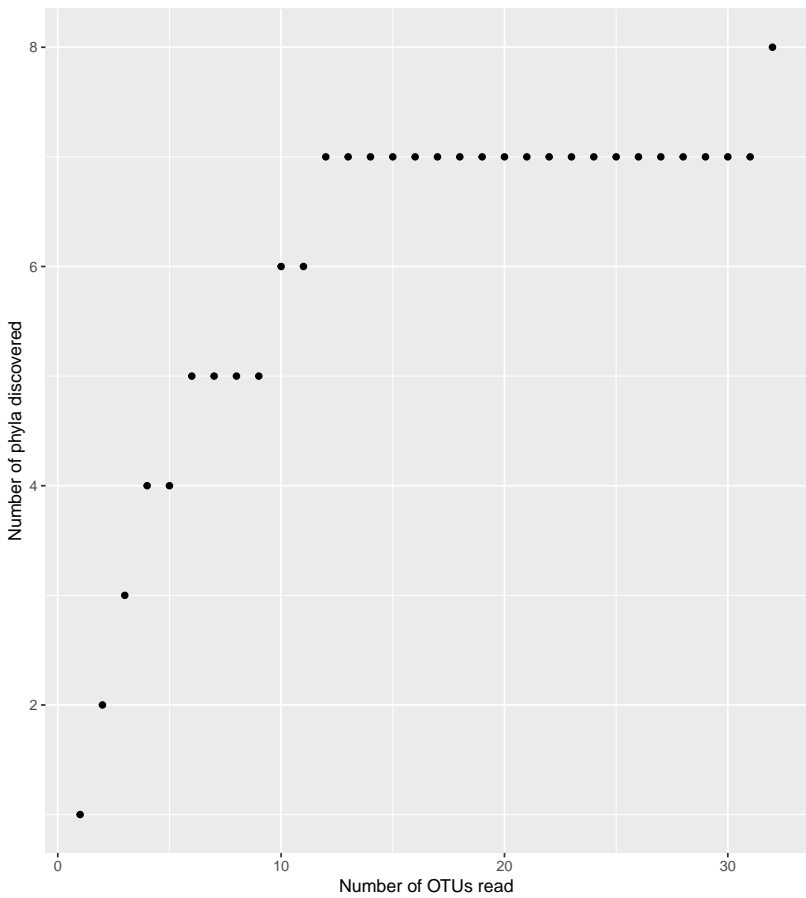
74. 18S.2.1.C.PCM.LT



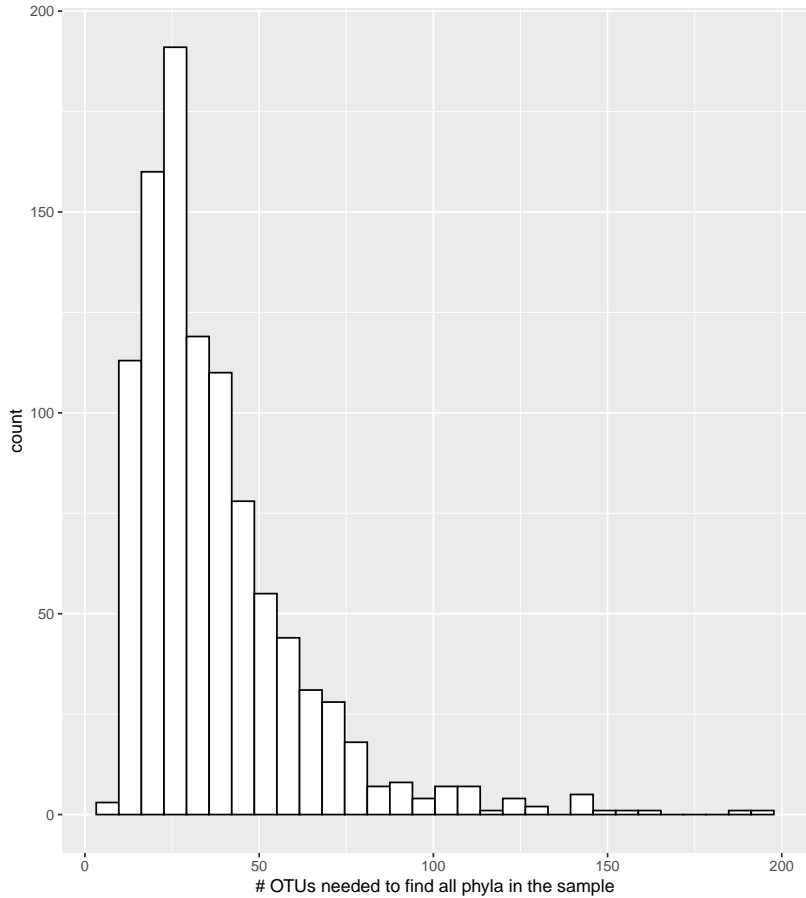
18S.2.1.C.PCM.LT



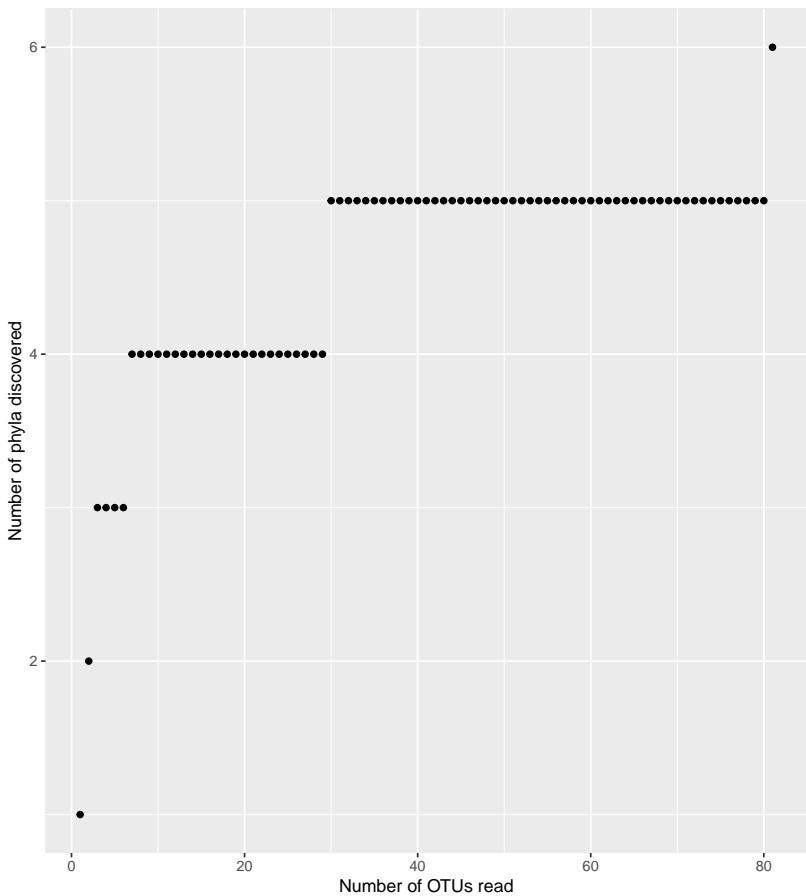
75. 18S.2.1.D.PCM.LT



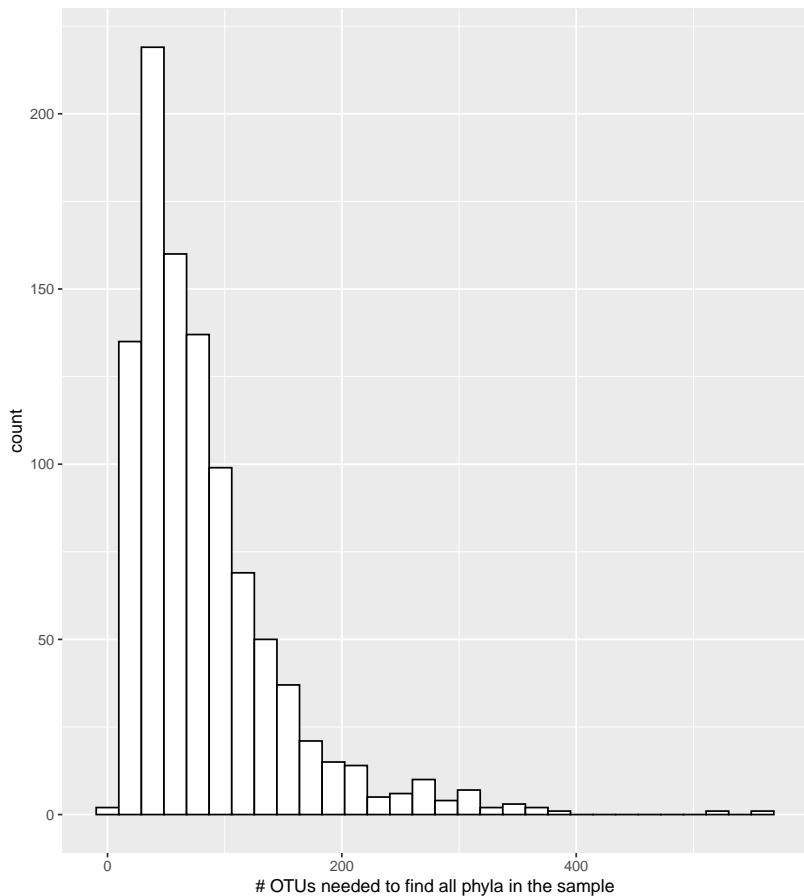
18S.2.1.D.PCM.LT



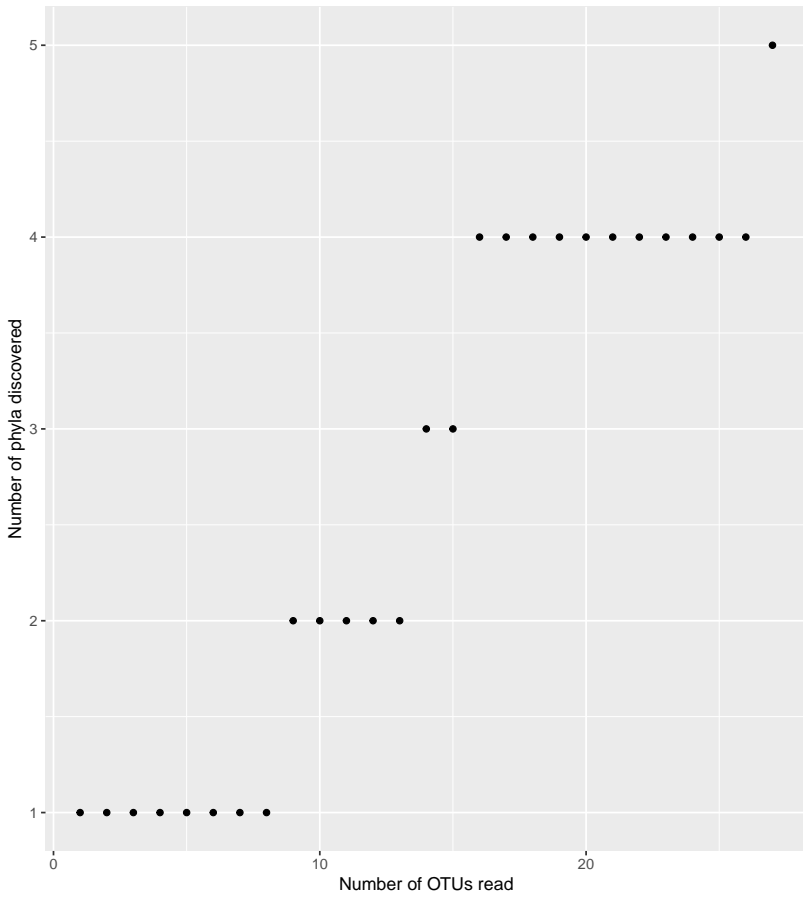
76. 18S.2.1.E.PCM.LT



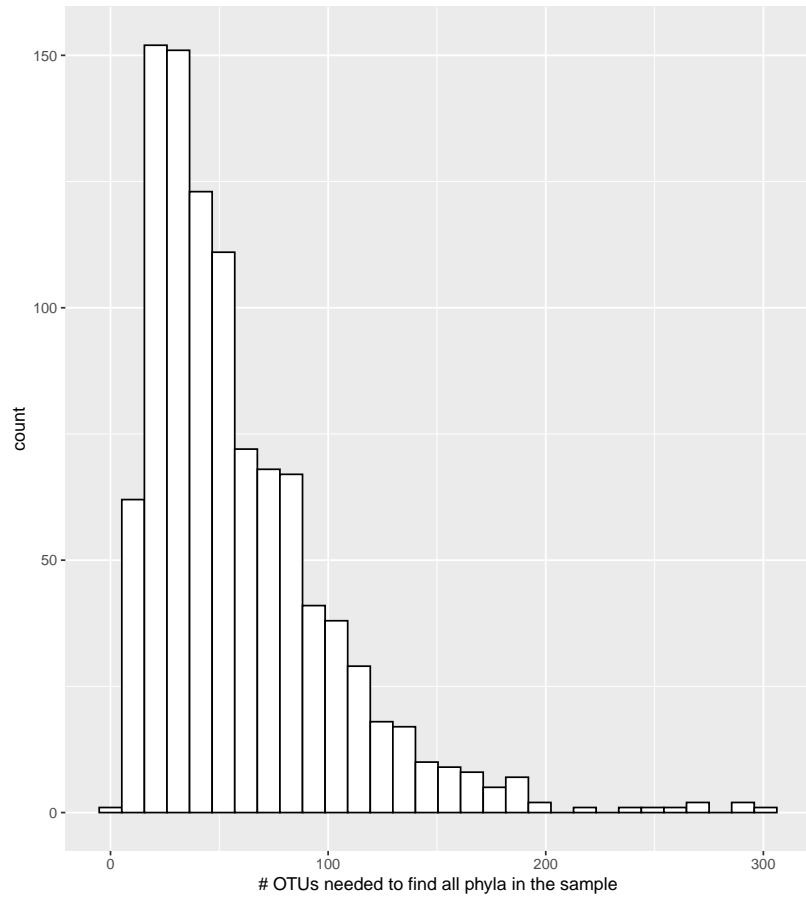
18S.2.1.E.PCM.LT



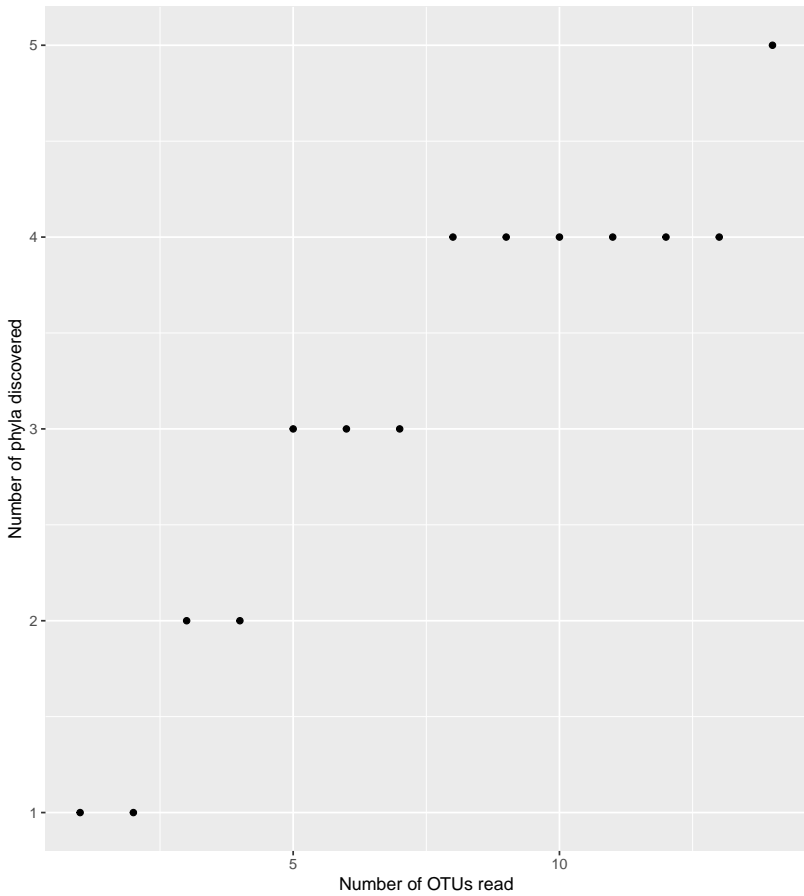
77. 18S.2.1.F.PCM.LT



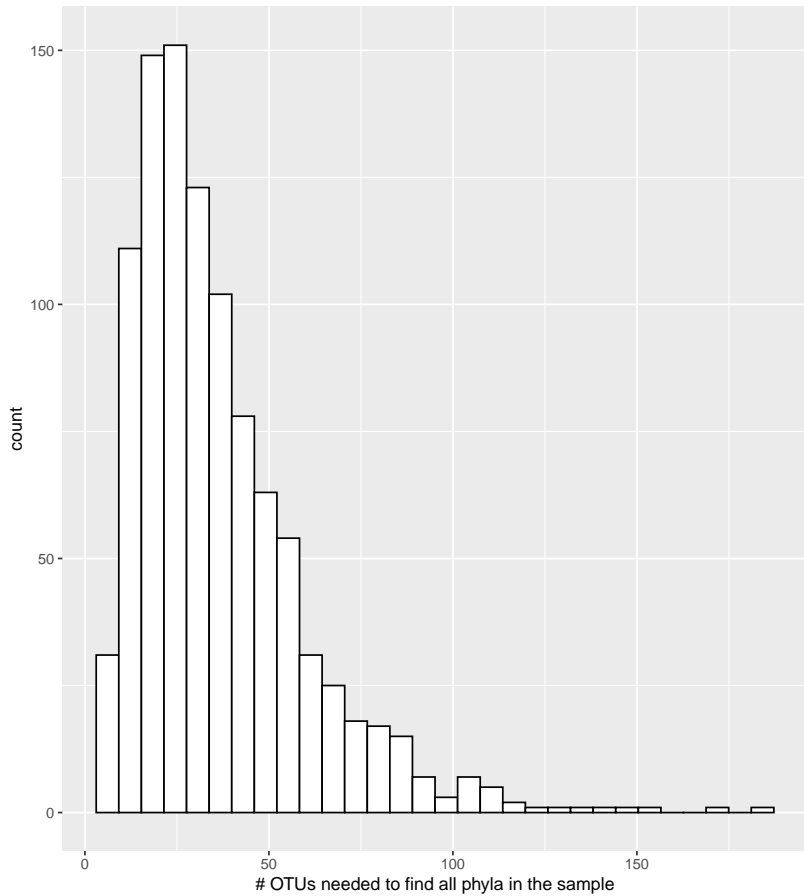
18S.2.1.F.PCM.LT



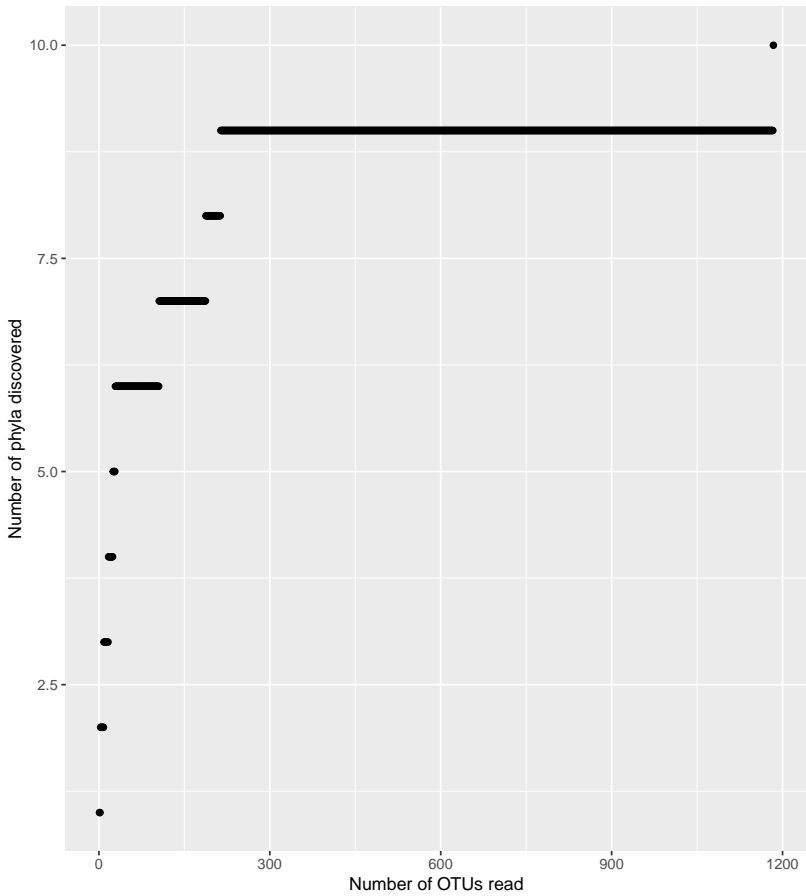
78. 18S.2.1.G.PCM.LT



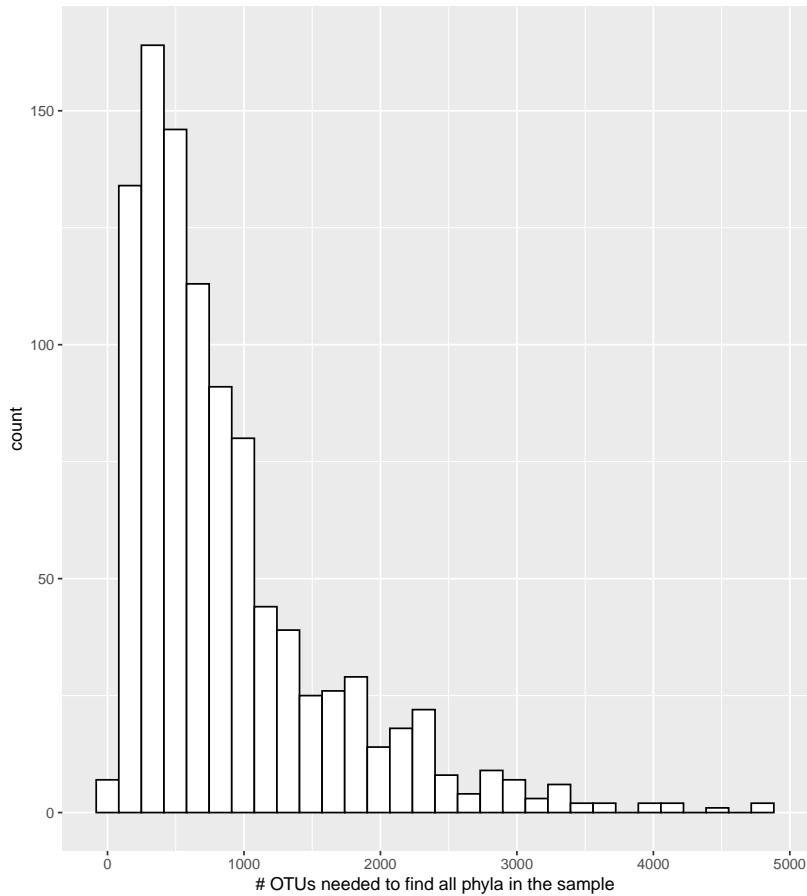
18S.2.1.G.PCM.LT



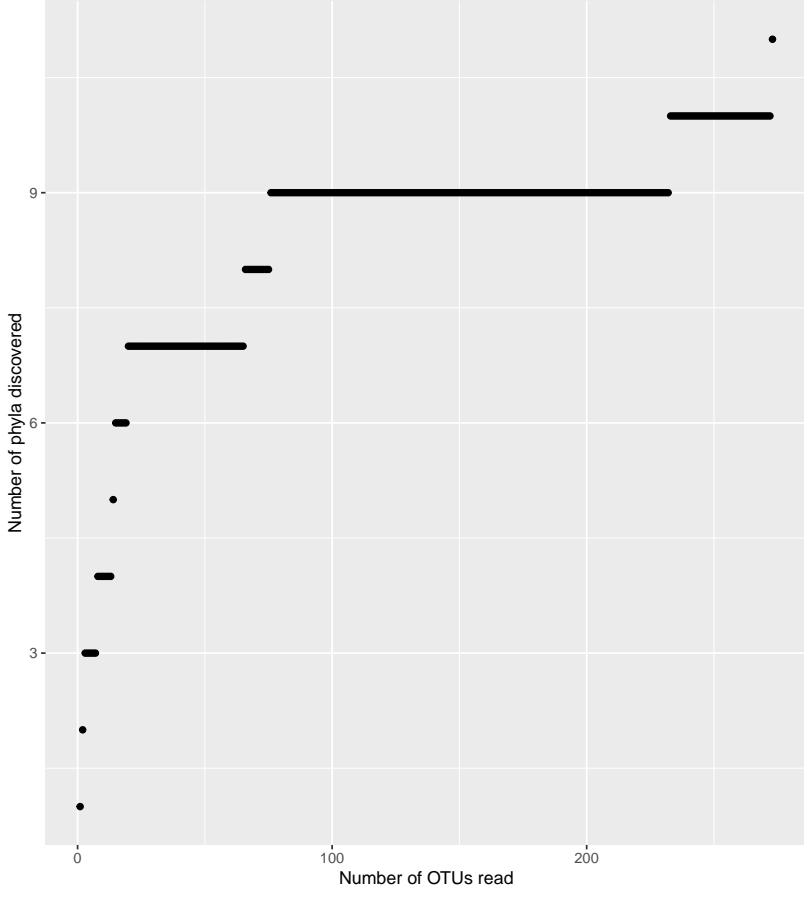
79. 18S.2.1.H.PCM.LT



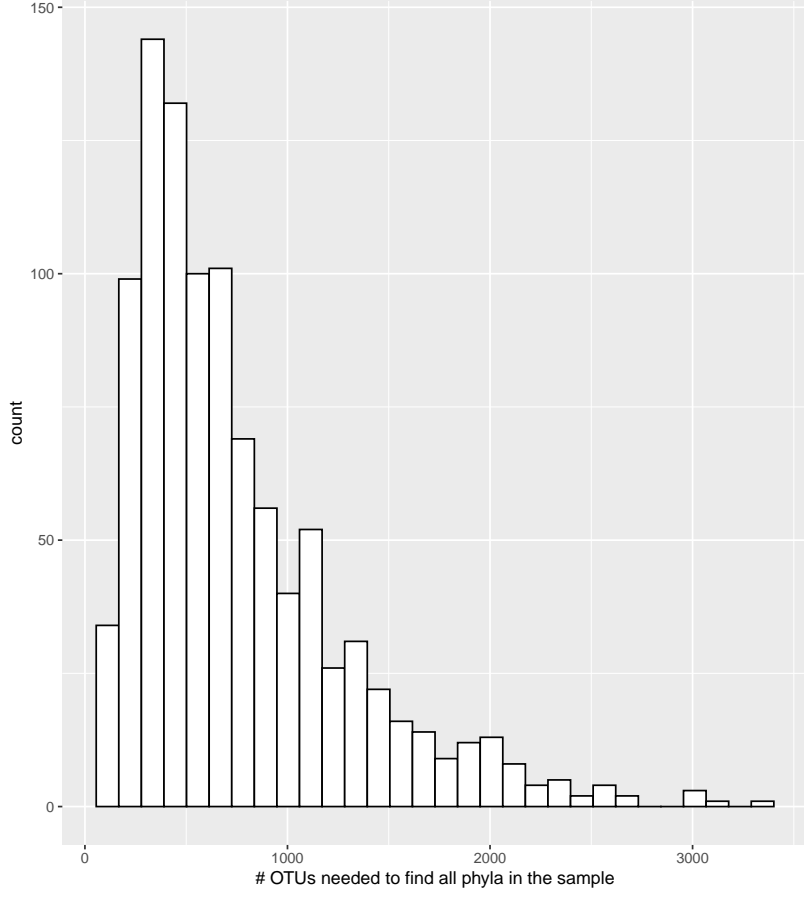
18S.2.1.H.PCM.LT



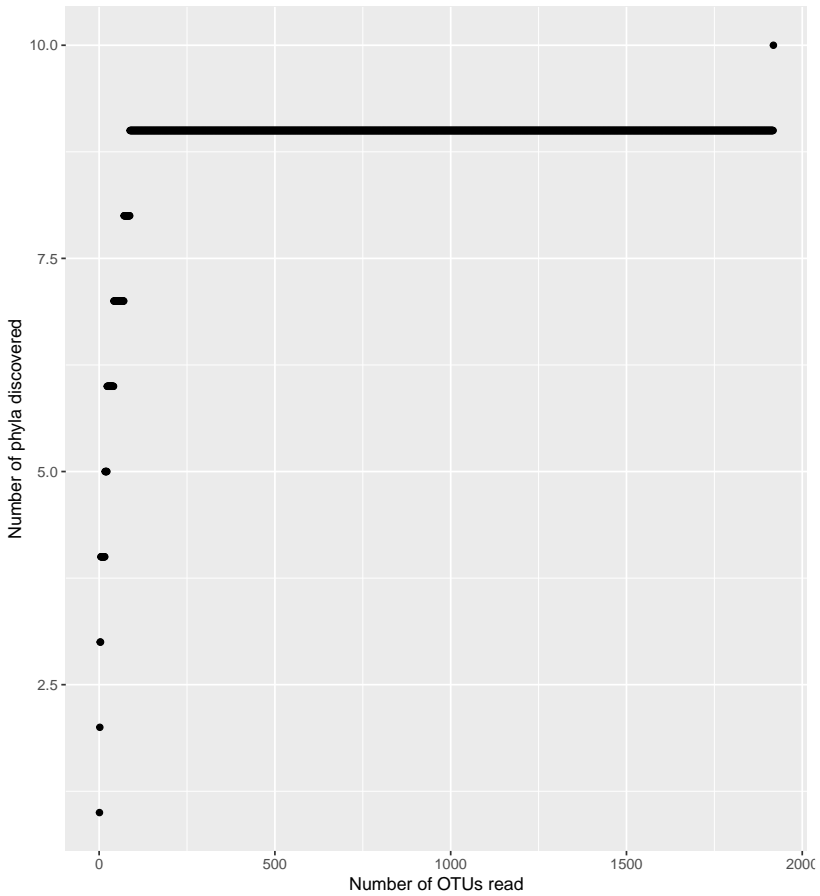
80. 18S.2.10.A.PCM.ME



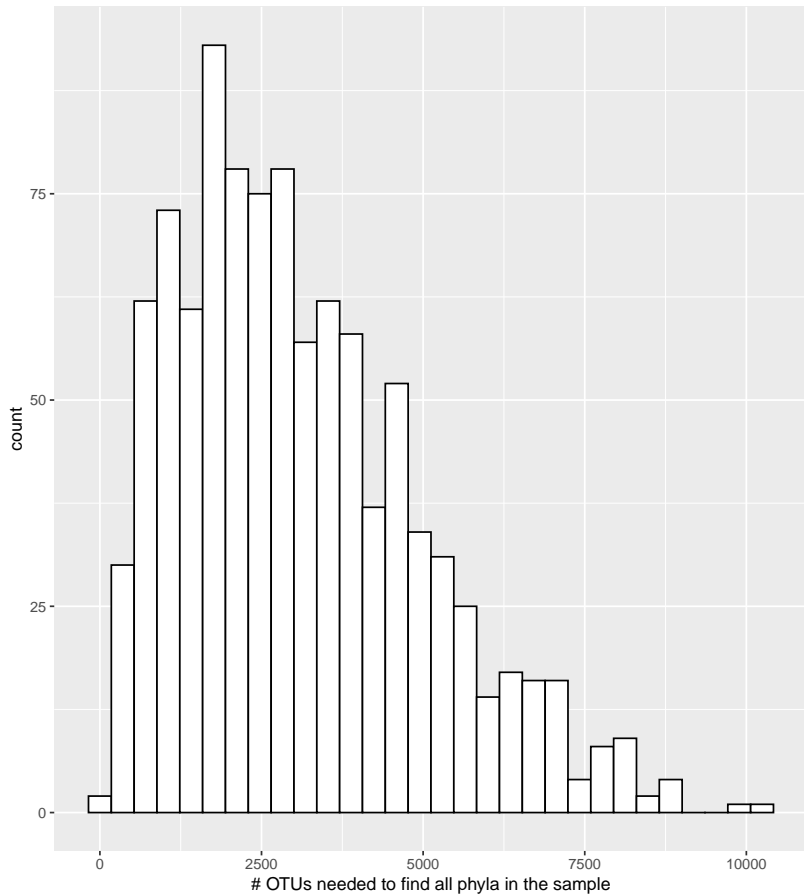
18S.2.10.A.PCM.ME



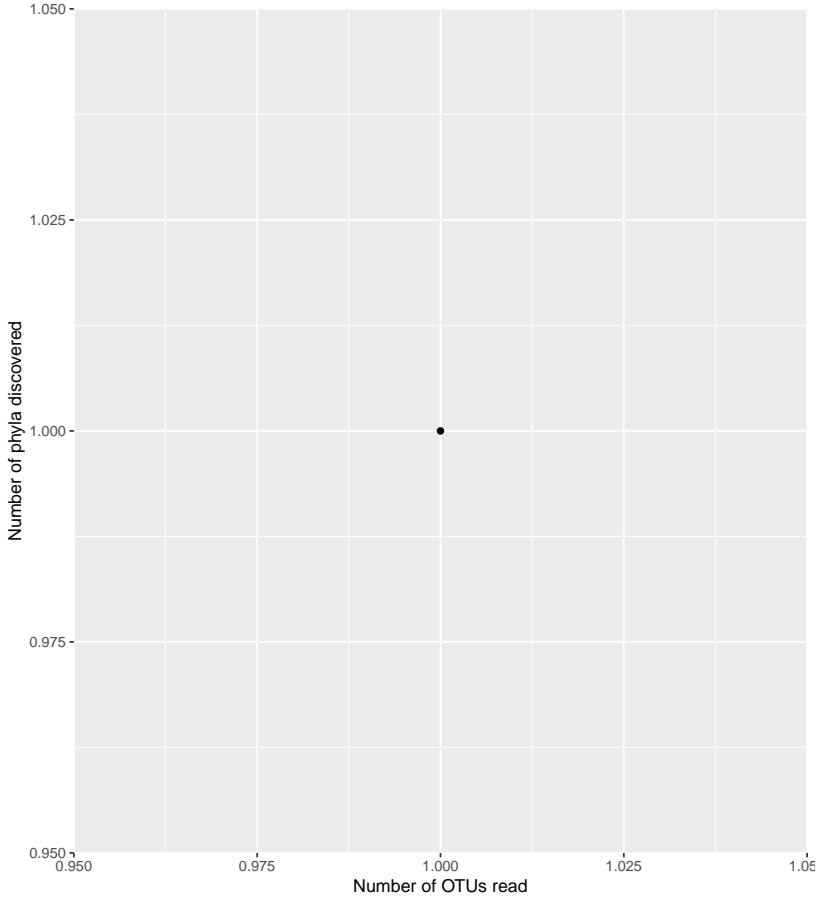
81. 18S.2.10.B.PCM.LT



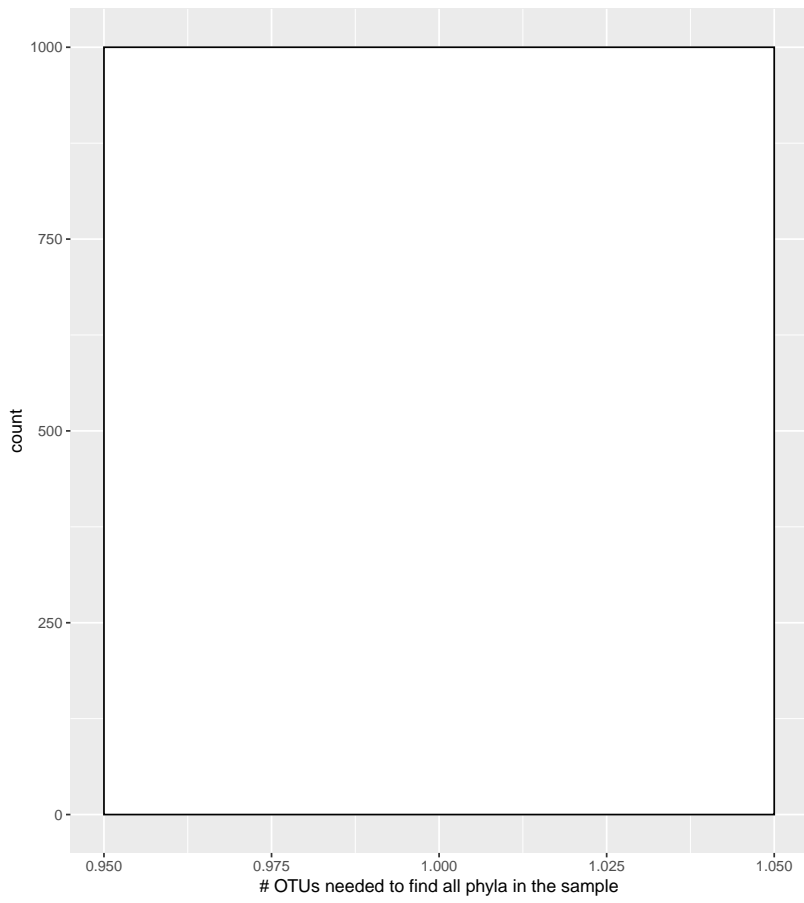
18S.2.10.B.PCM.LT



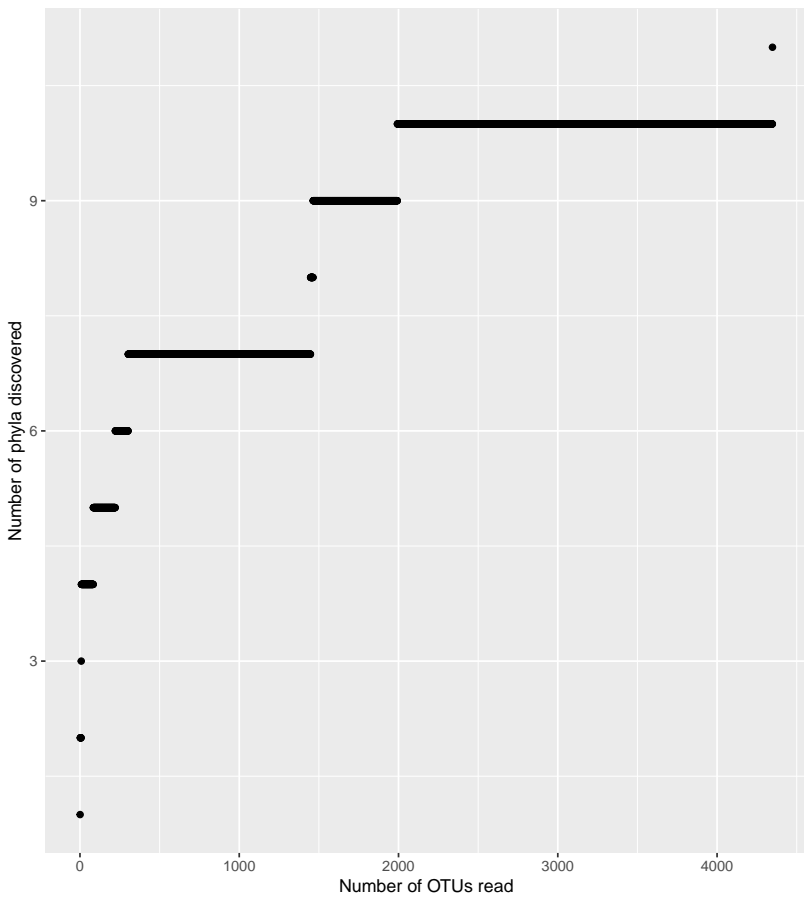
82. 18S.2.10.C.PCM.LT



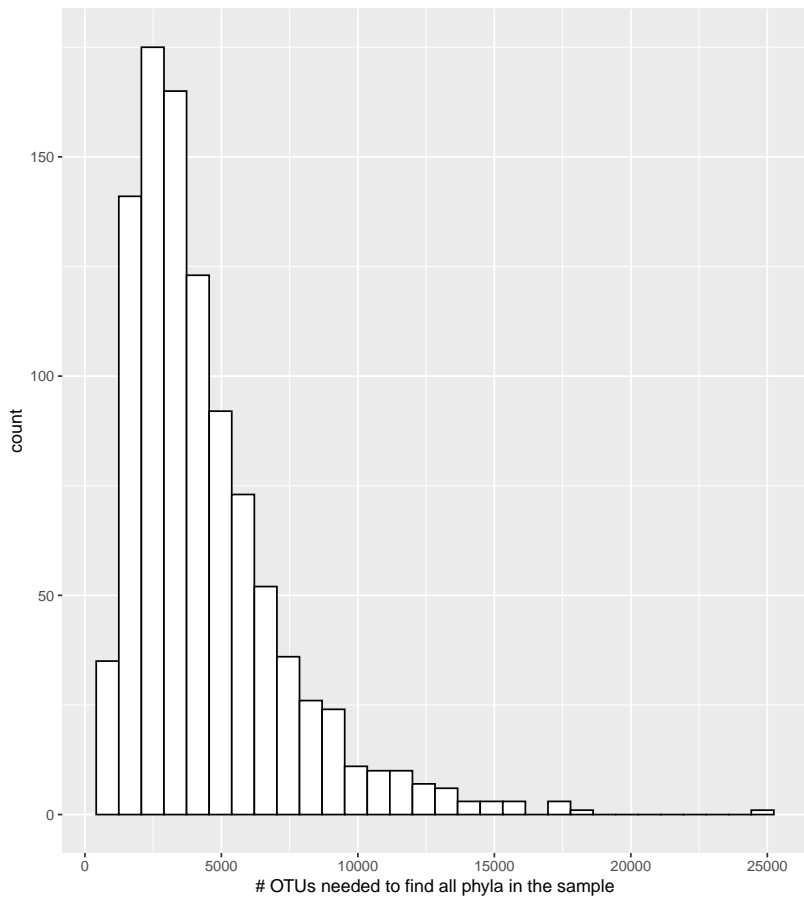
18S.2.10.C.PCM.LT



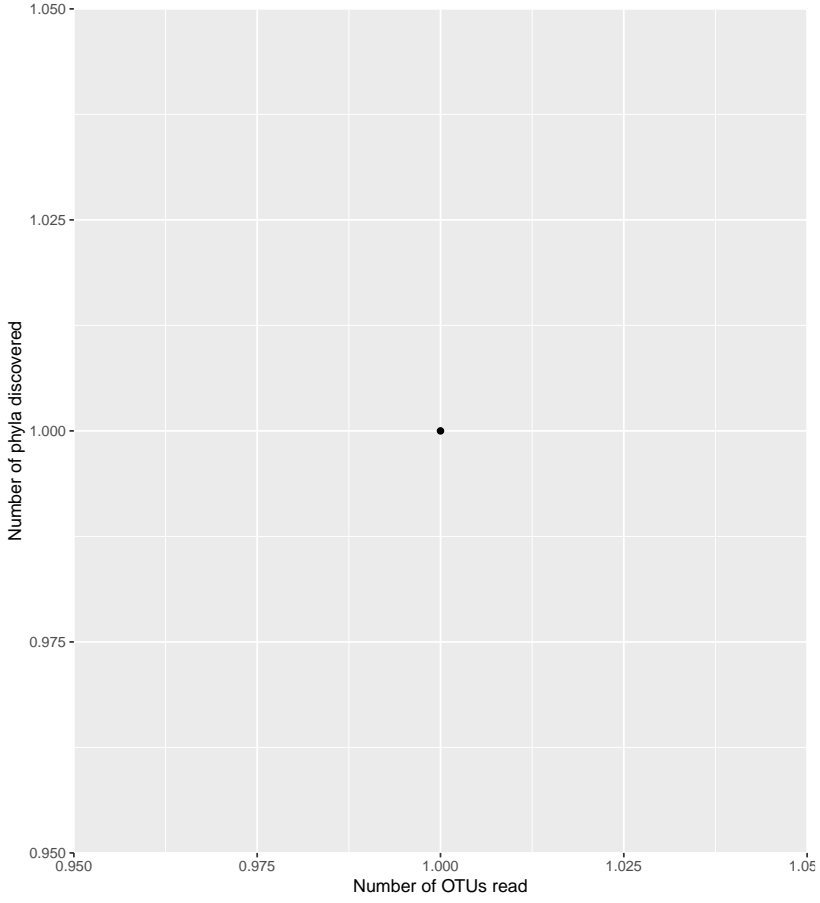
83. 18S.2.10.D.PCM.LT



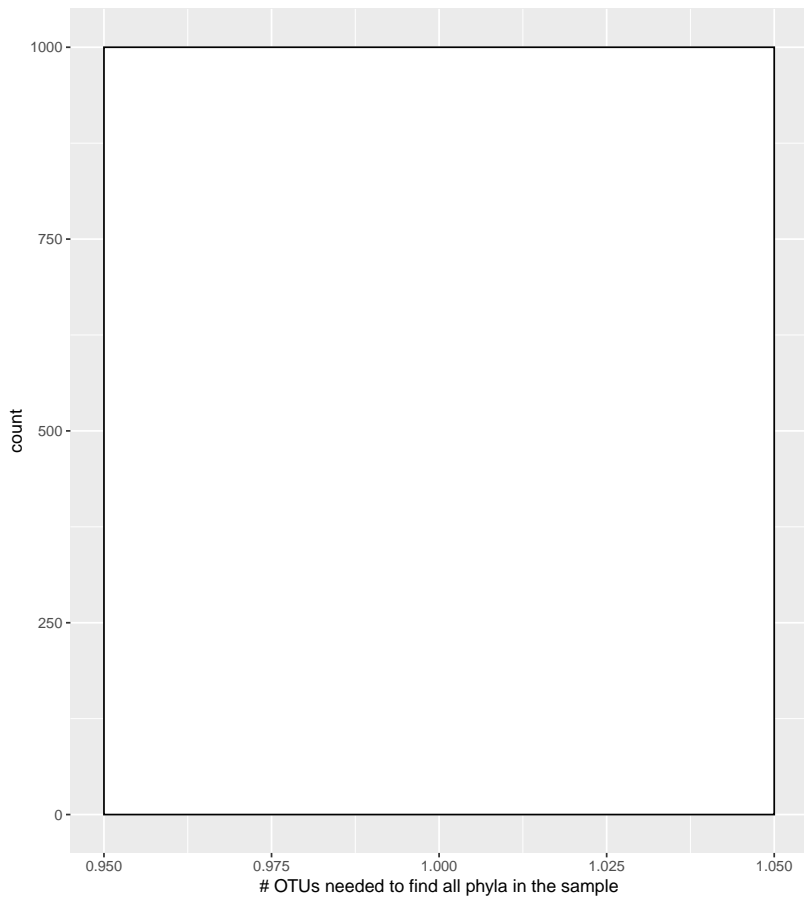
18S.2.10.D.PCM.LT



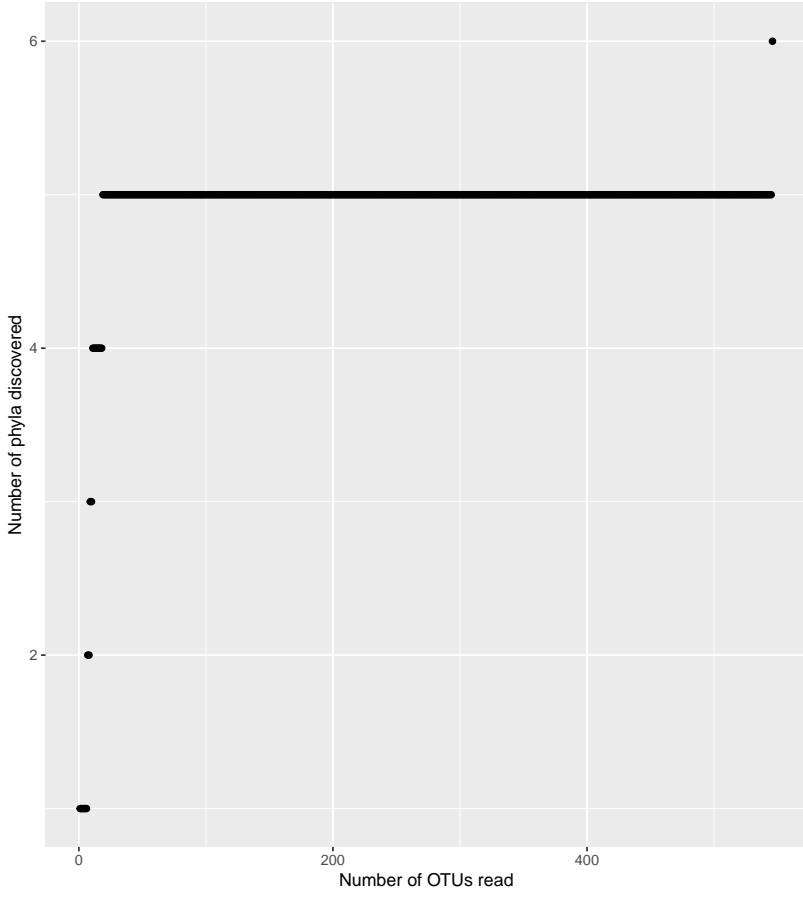
84. 18S.2.10.E.PCM.LT



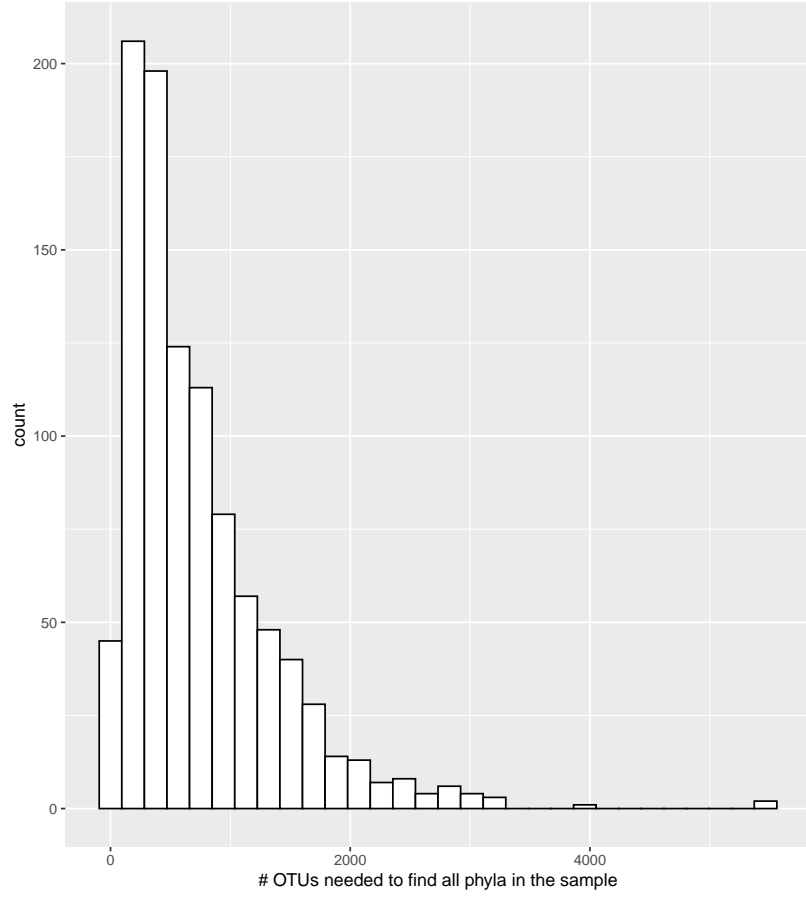
18S.2.10.E.PCM.LT



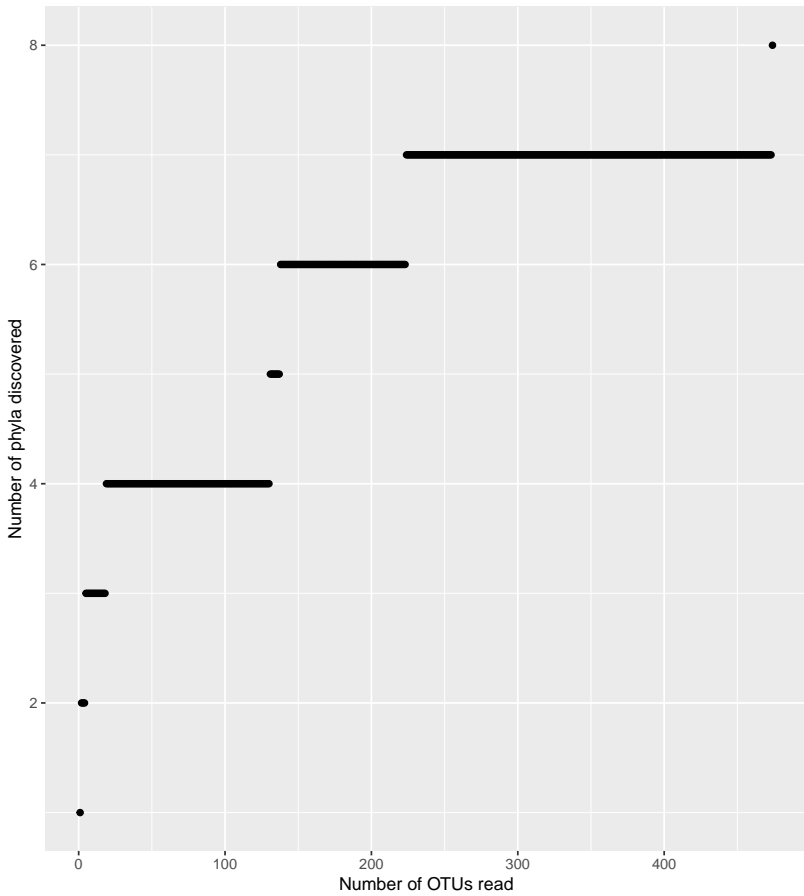
85. 18S.2.10.F.PCM.LT



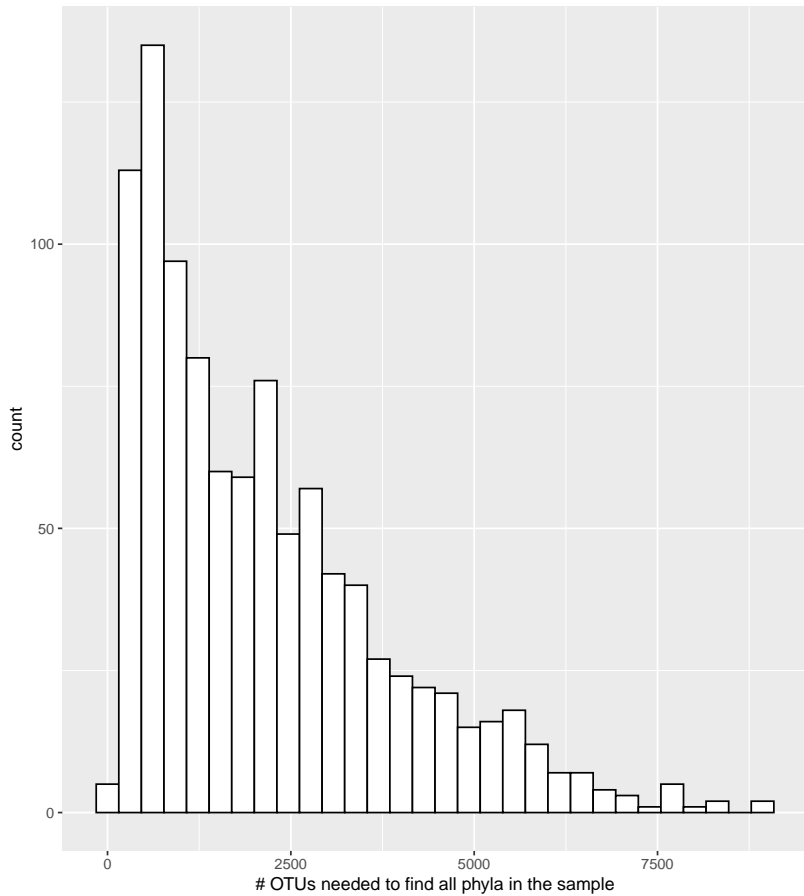
18S.2.10.F.PCM.LT



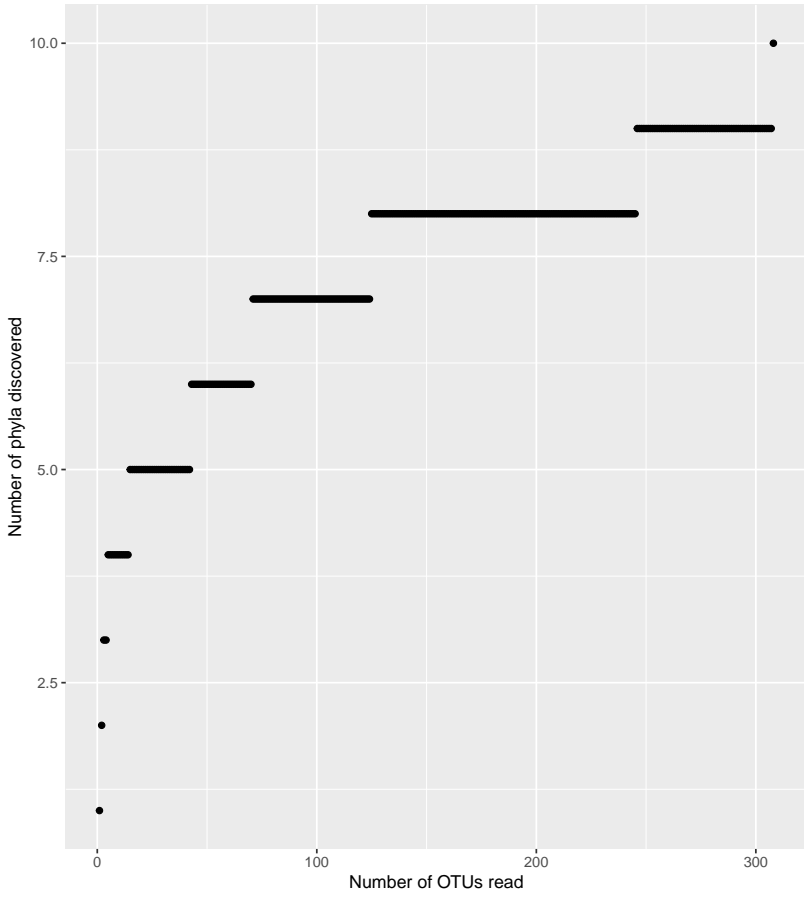
86. 18S.2.10.G.PCM.LT



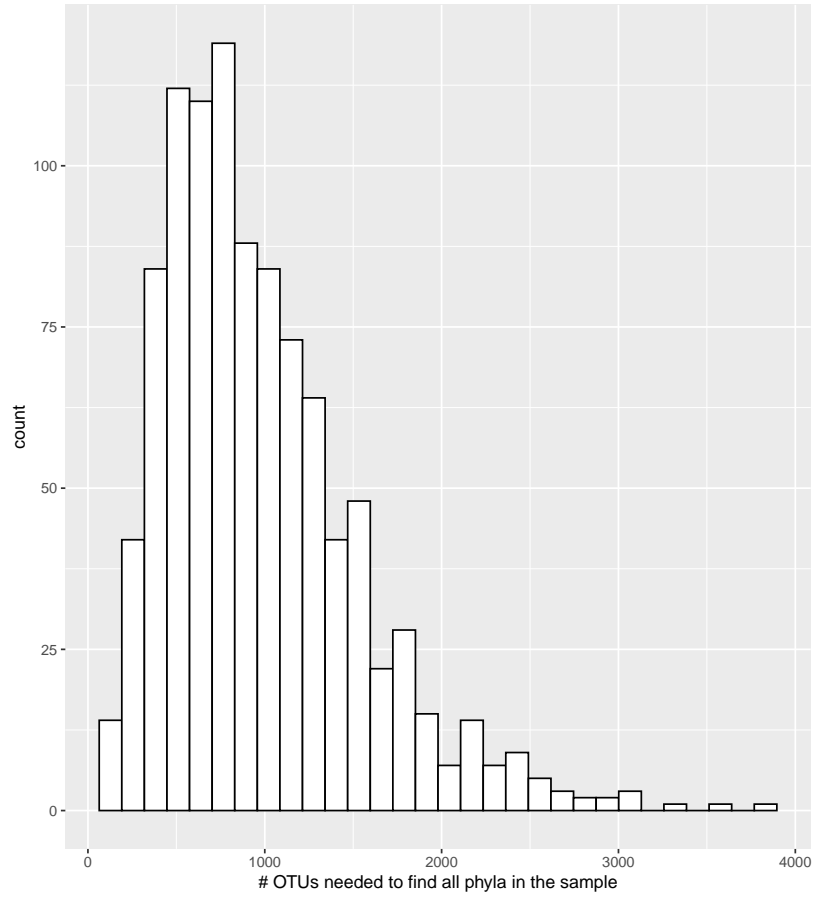
18S.2.10.G.PCM.LT



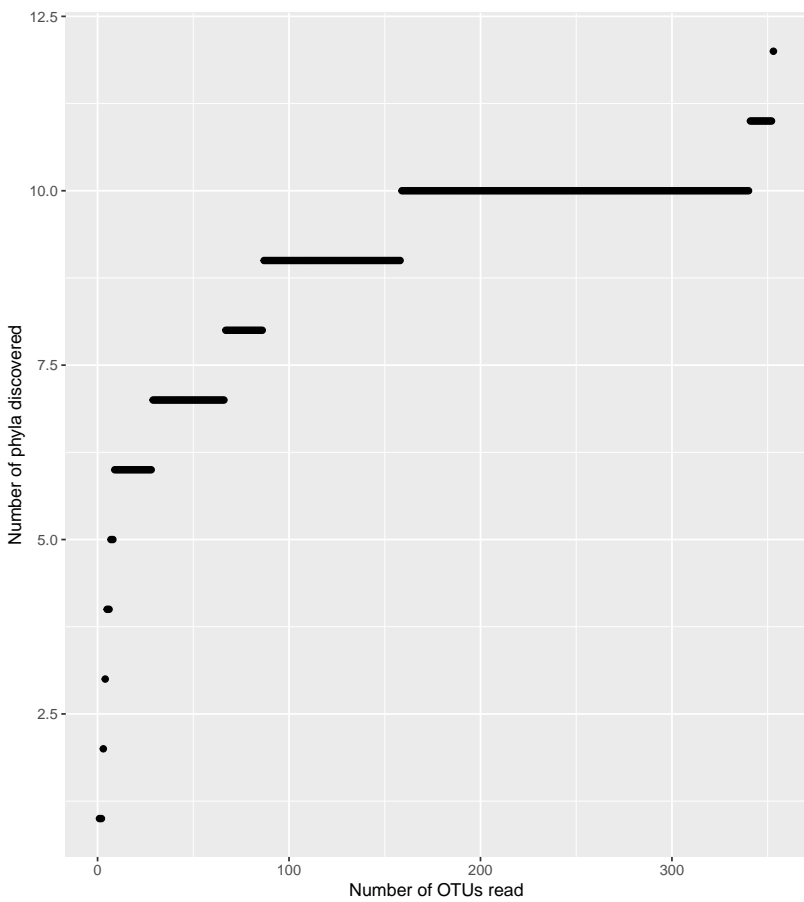
87. 18S.2.11.B.PCM.LT



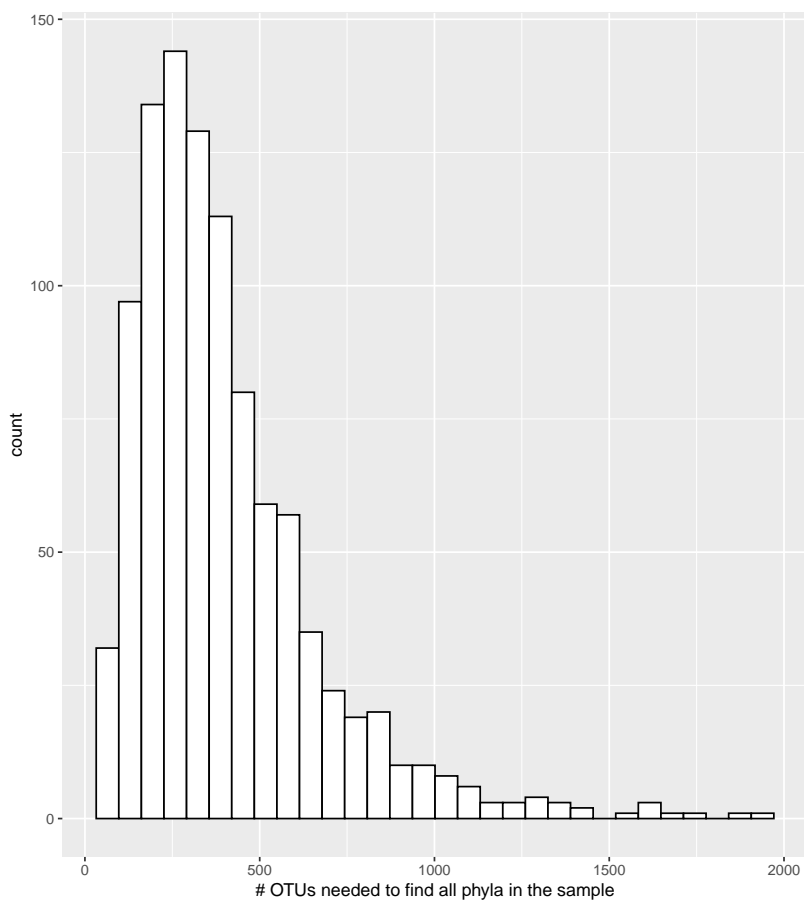
18S.2.11.B.PCM.LT



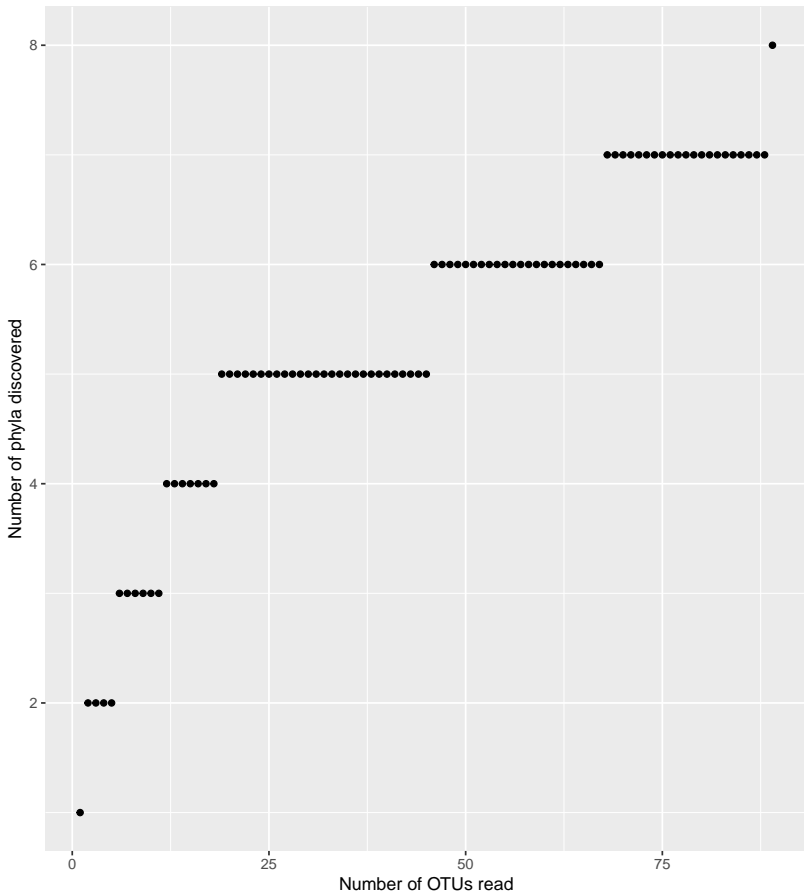
88. 18S.2.11.C.PCM.LT



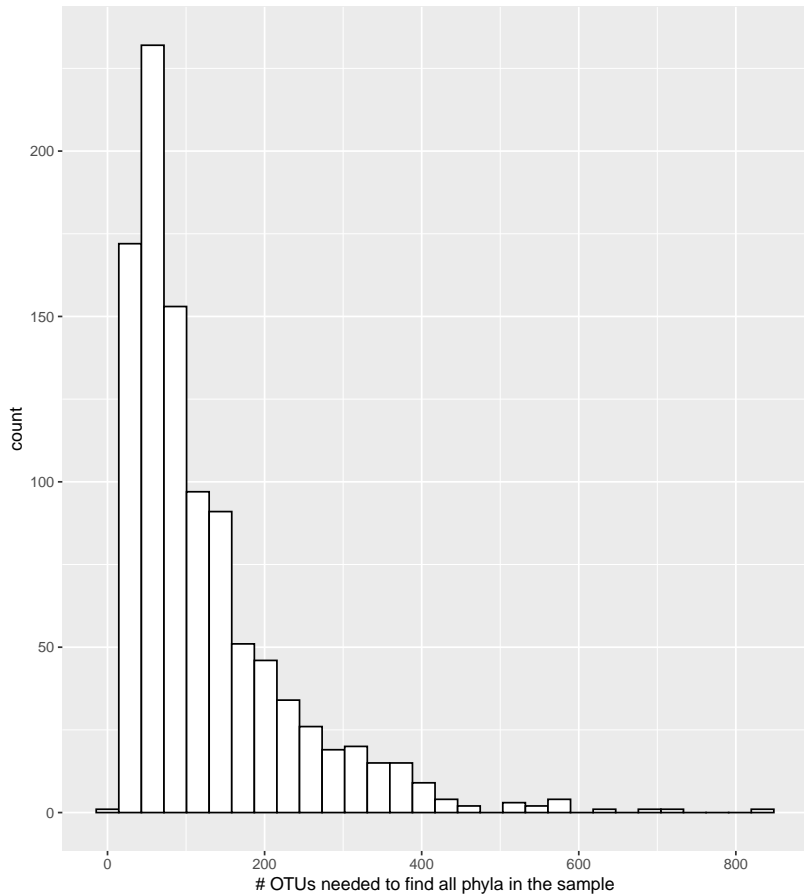
18S.2.11.C.PCM.LT



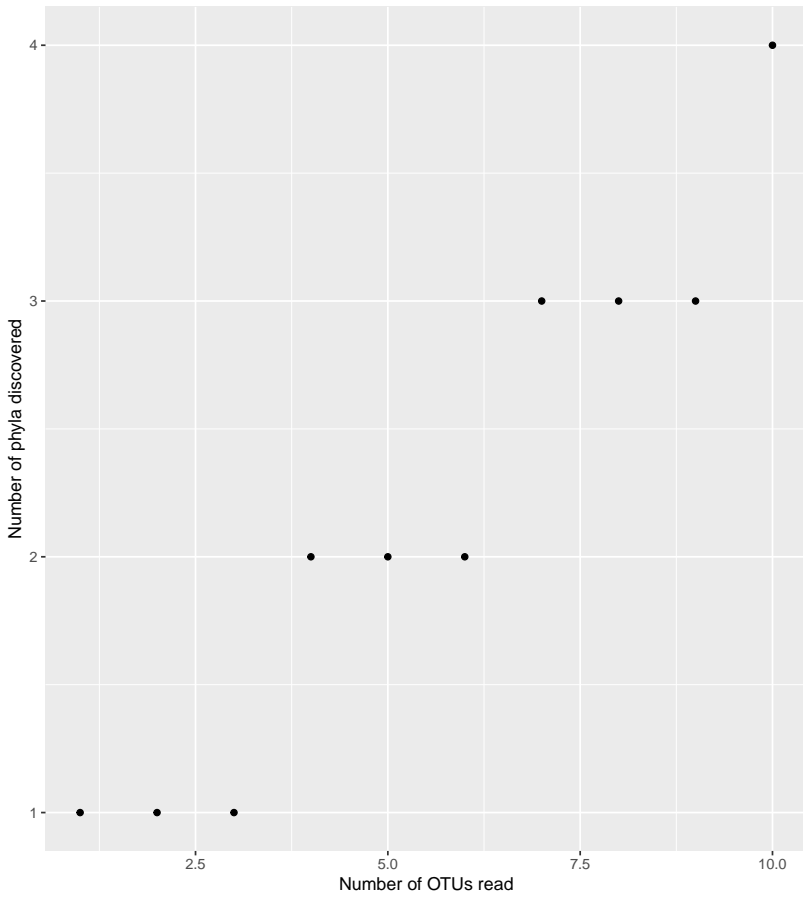
89. 18S.2.11.D.PCM.LT



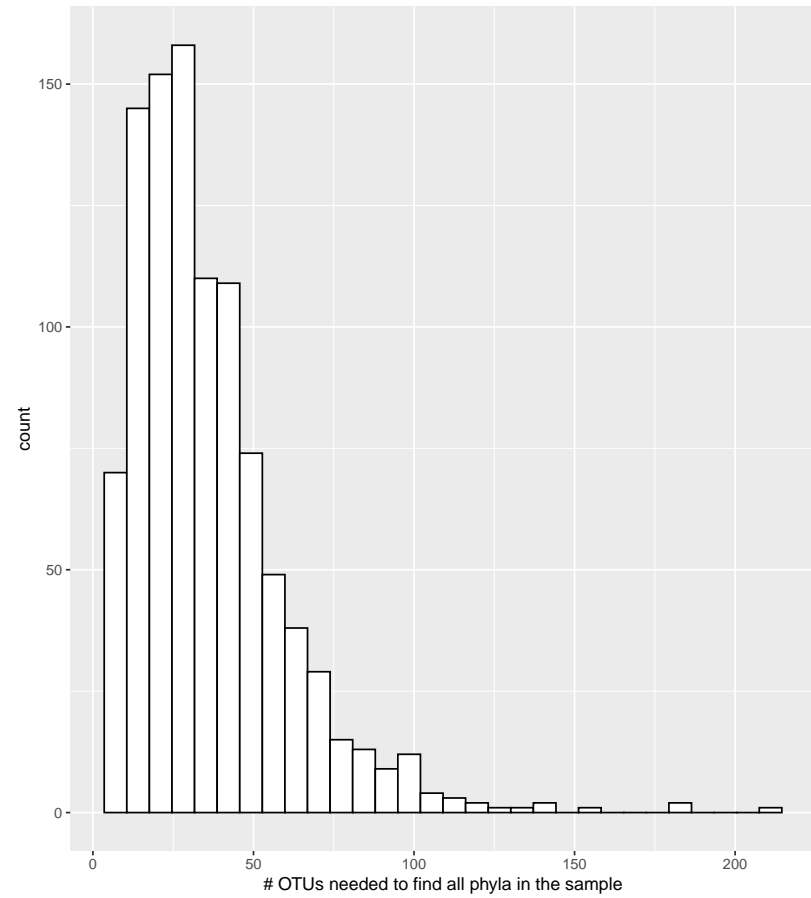
18S.2.11.D.PCM.LT



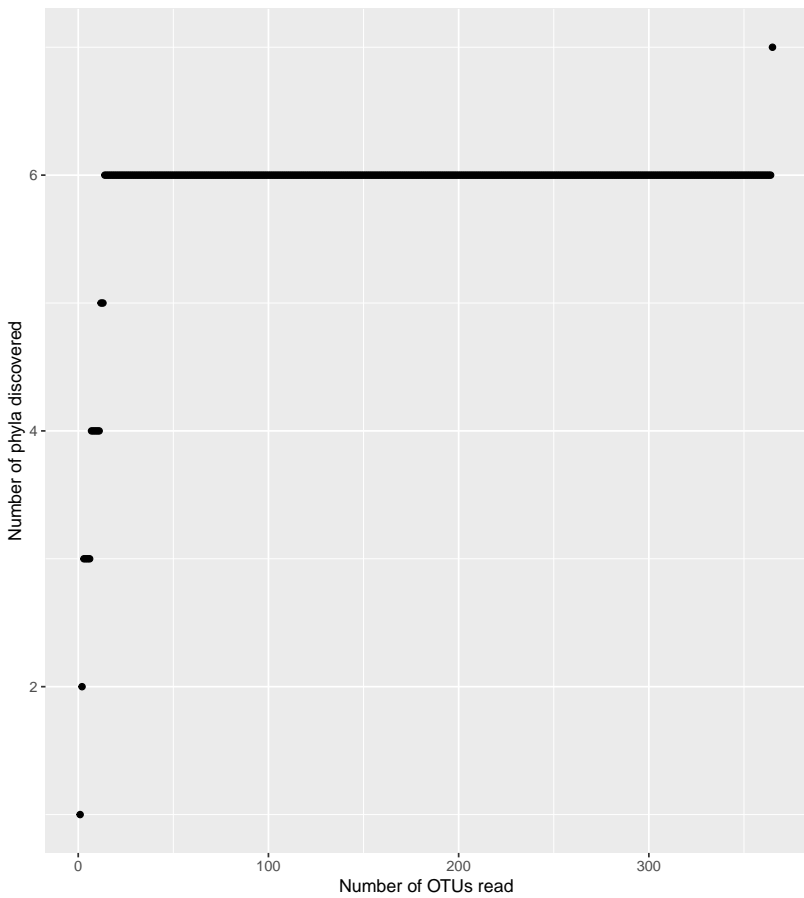
90. 18S.2.11.E.PCM.LT



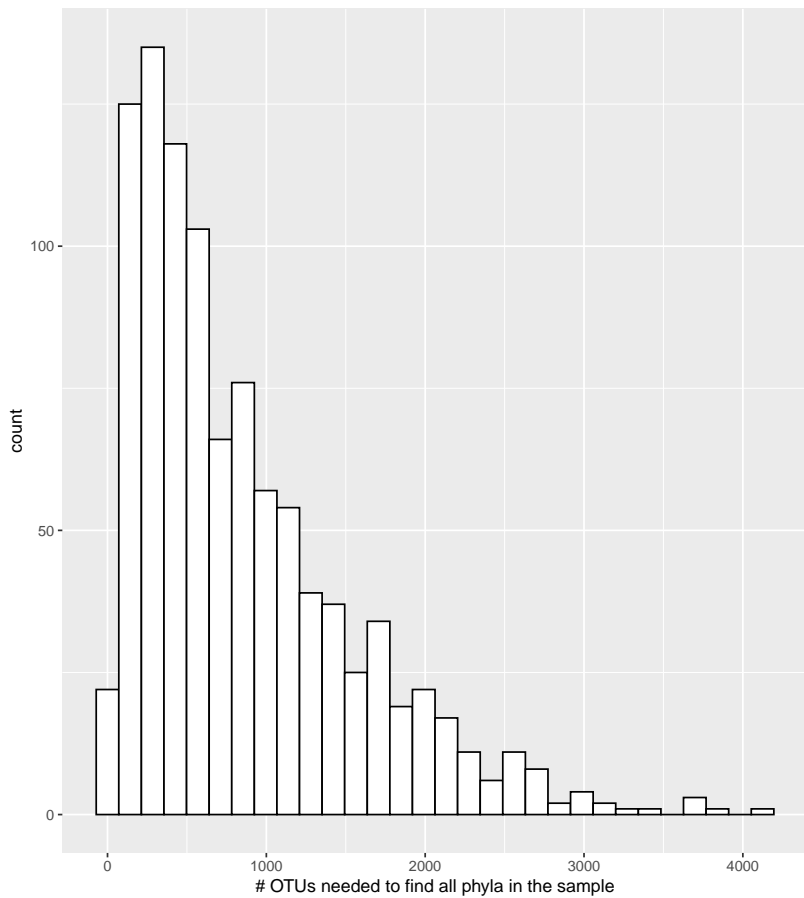
18S.2.11.E.PCM.LT



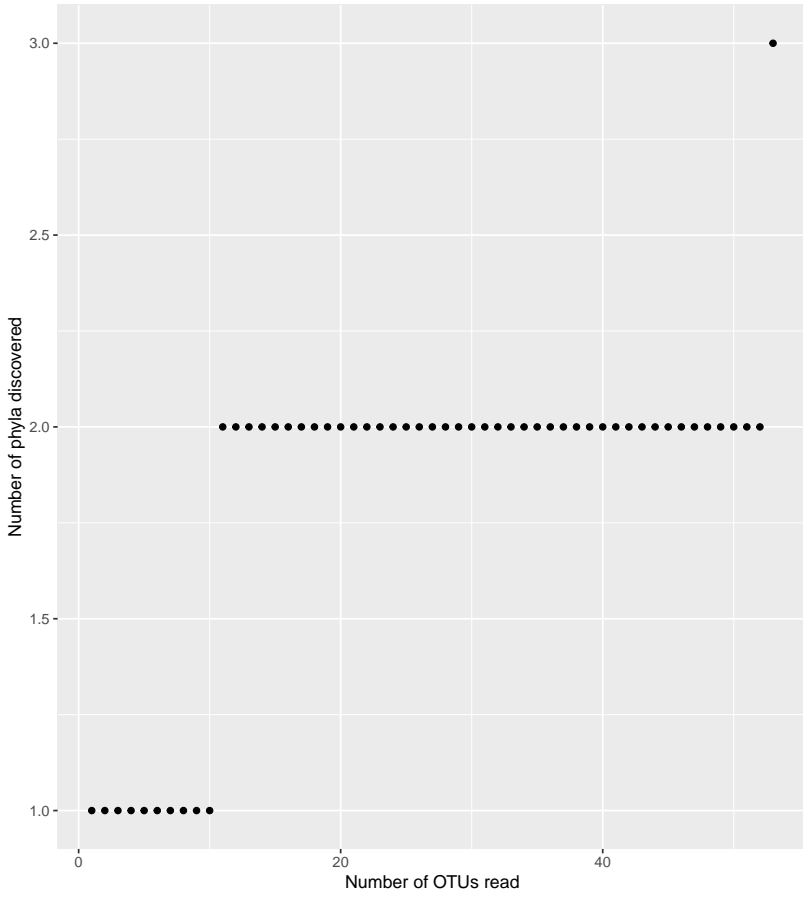
91. 18S.2.11.F.PCM.LT



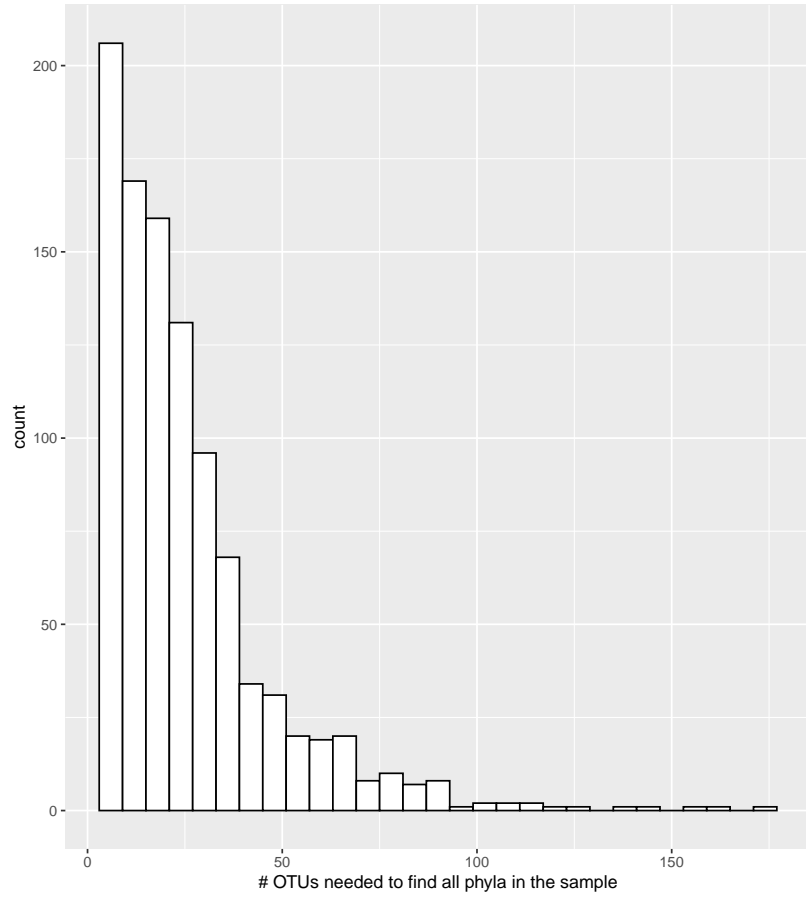
18S.2.11.F.PCM.LT



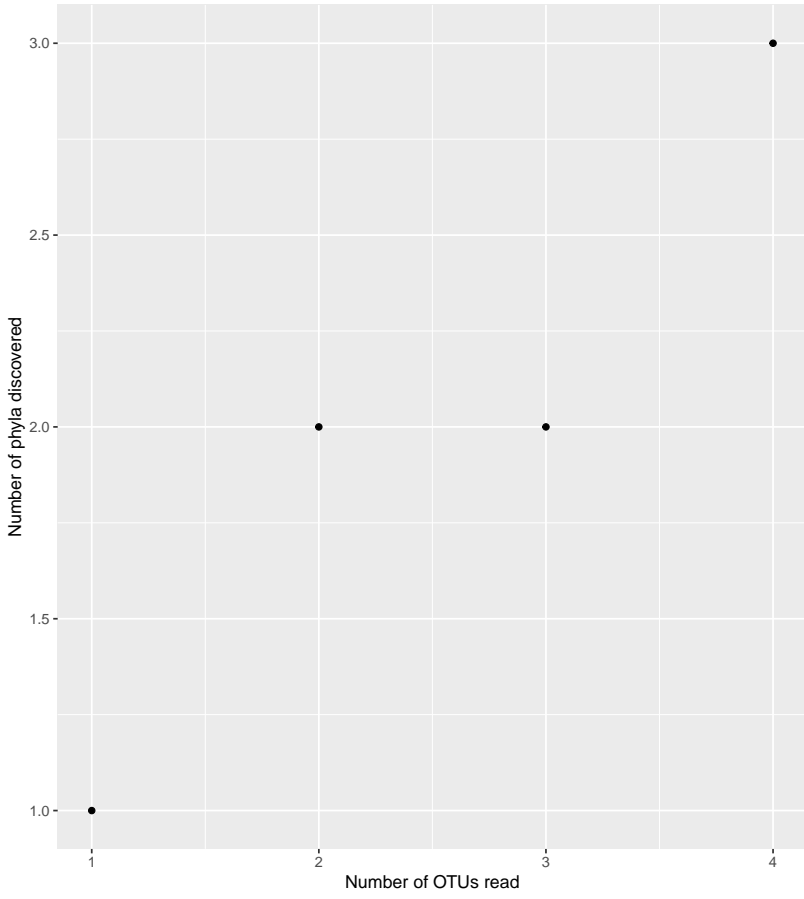
92. 18S.2.2.A.PCM.ME



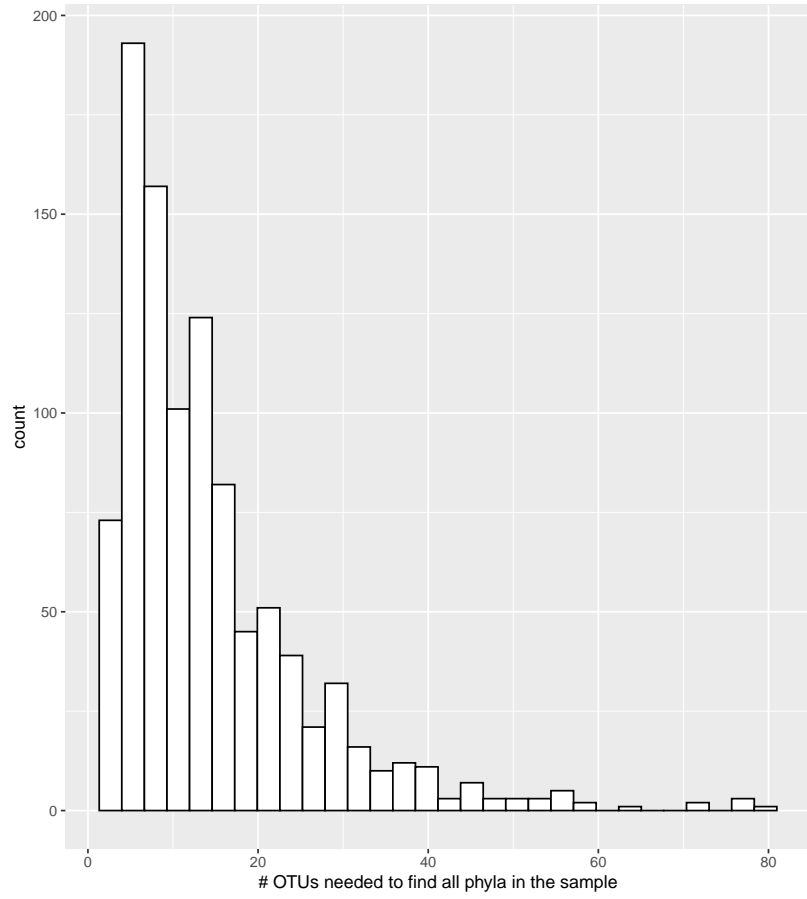
18S.2.2.A.PCM.ME



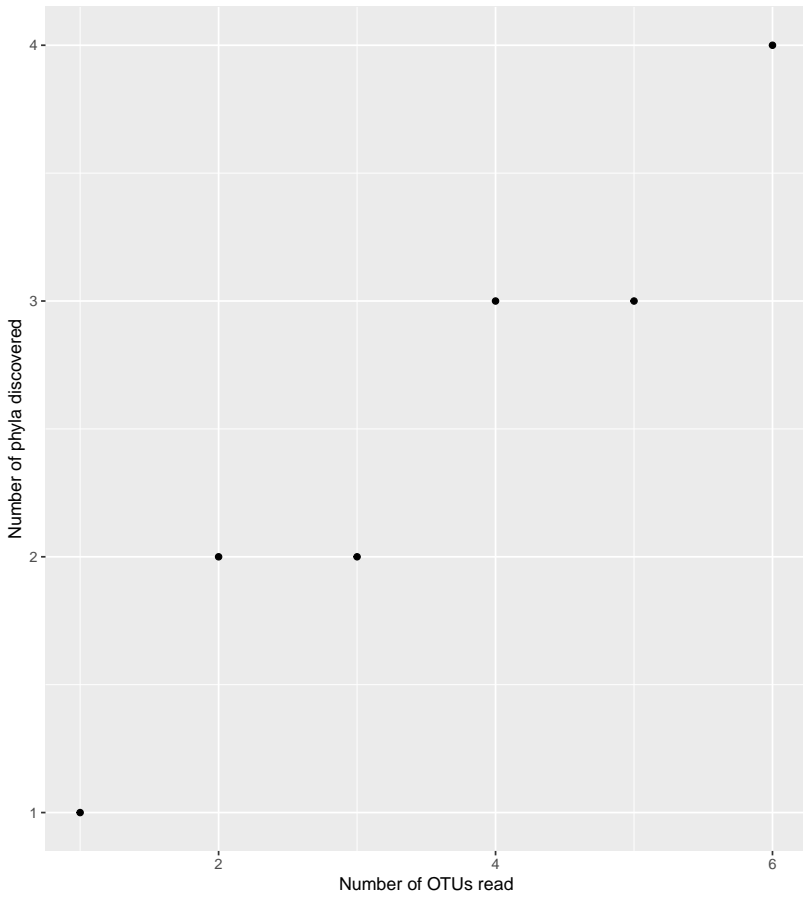
93. 18S.2.2.B.PCM.ME



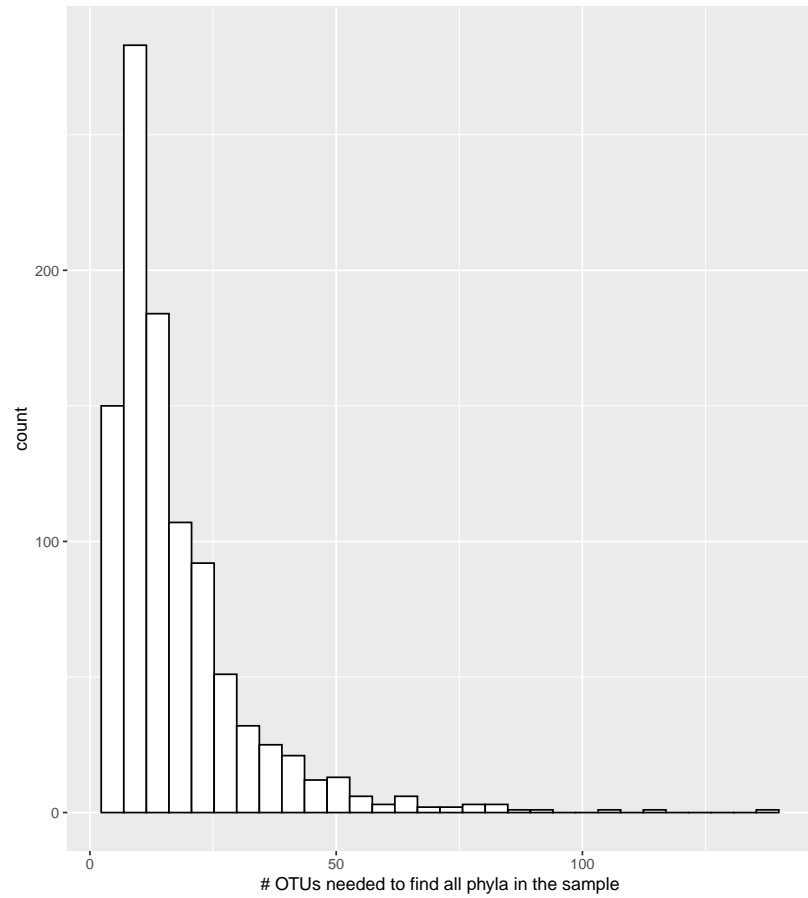
18S.2.2.B.PCM.ME



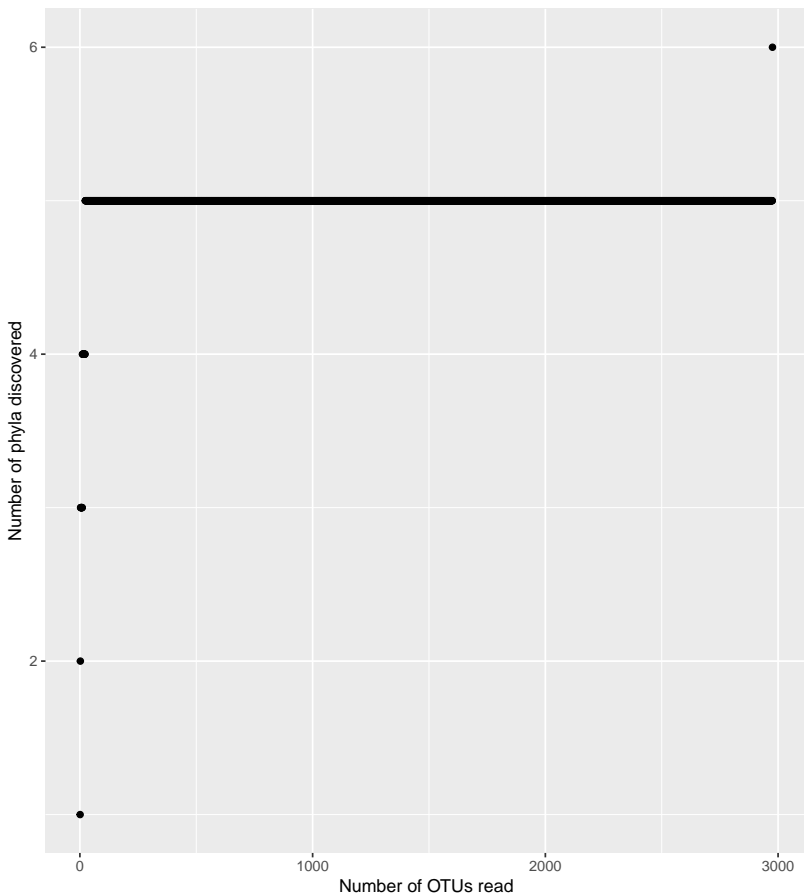
94. 18S.2.2.C.PCM.LT



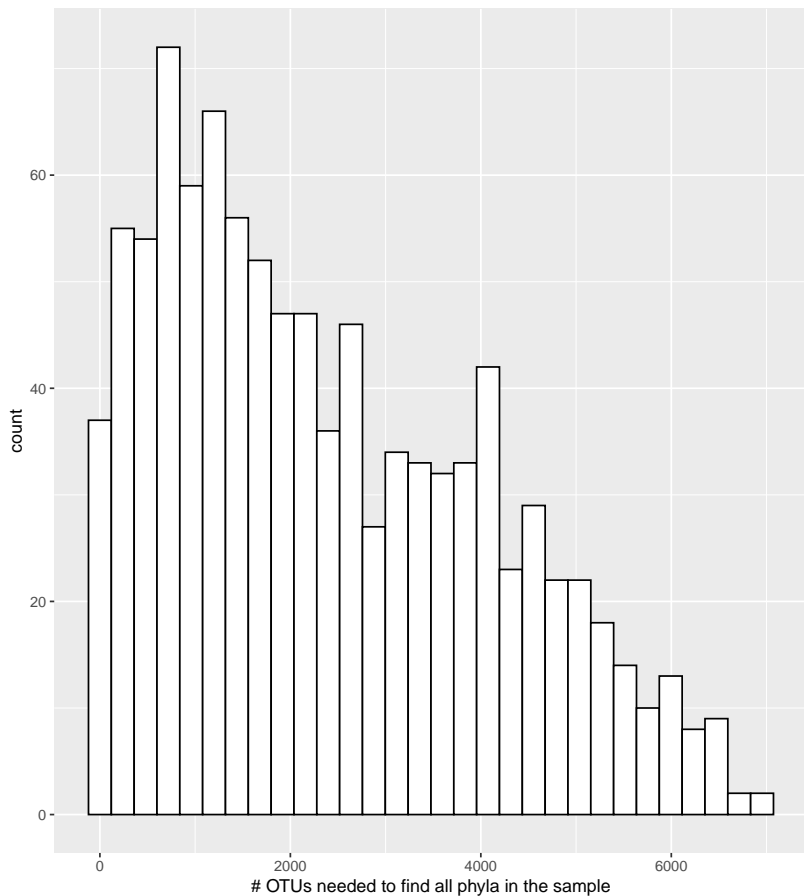
18S.2.2.C.PCM.LT



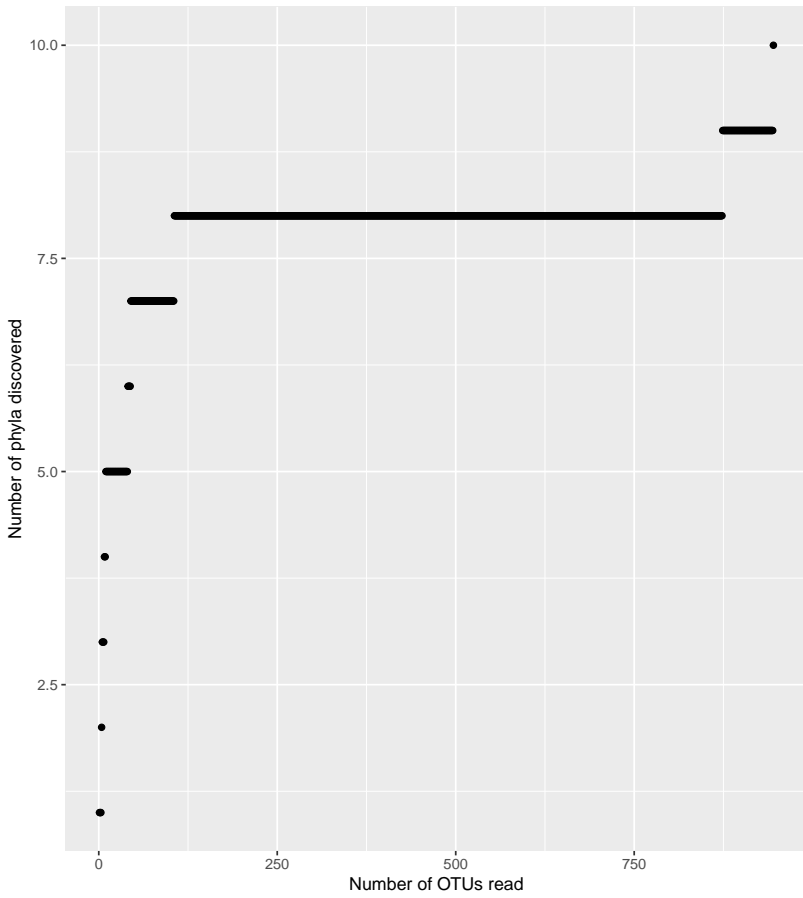
95. 18S.2.2.D.PCM.LT



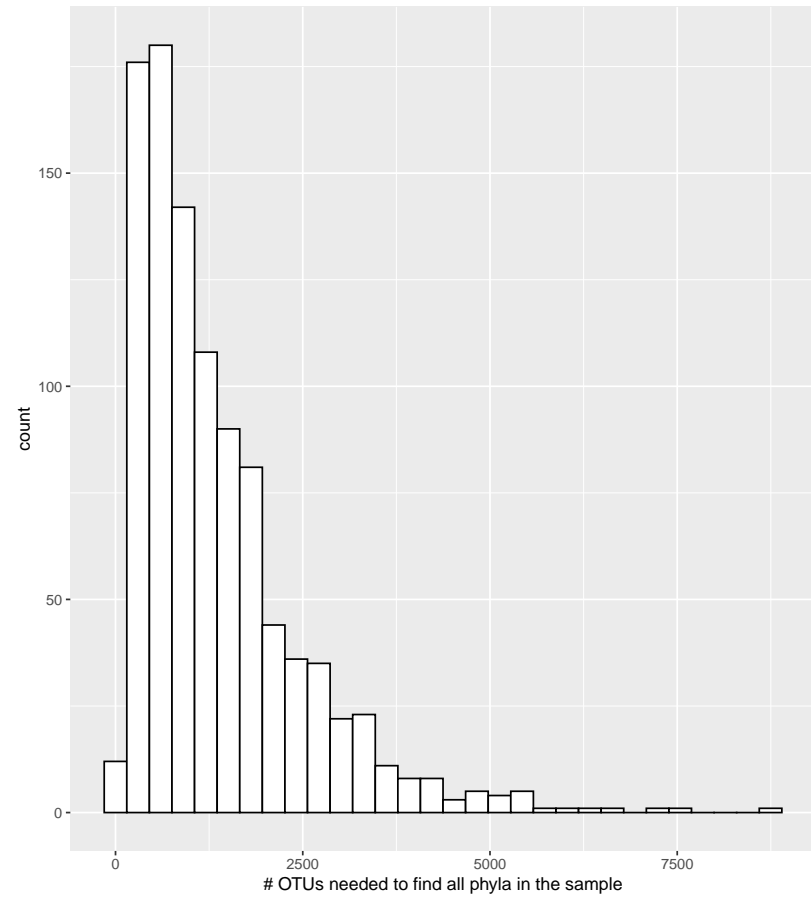
18S.2.2.D.PCM.LT



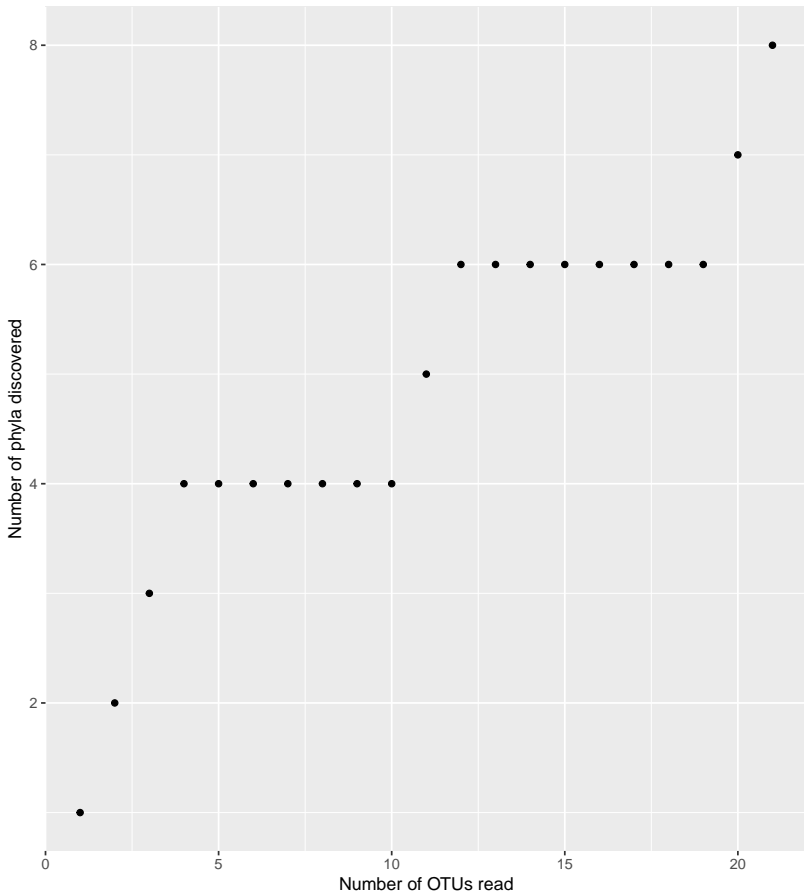
96. 18S.2.2.E.PCM.LT



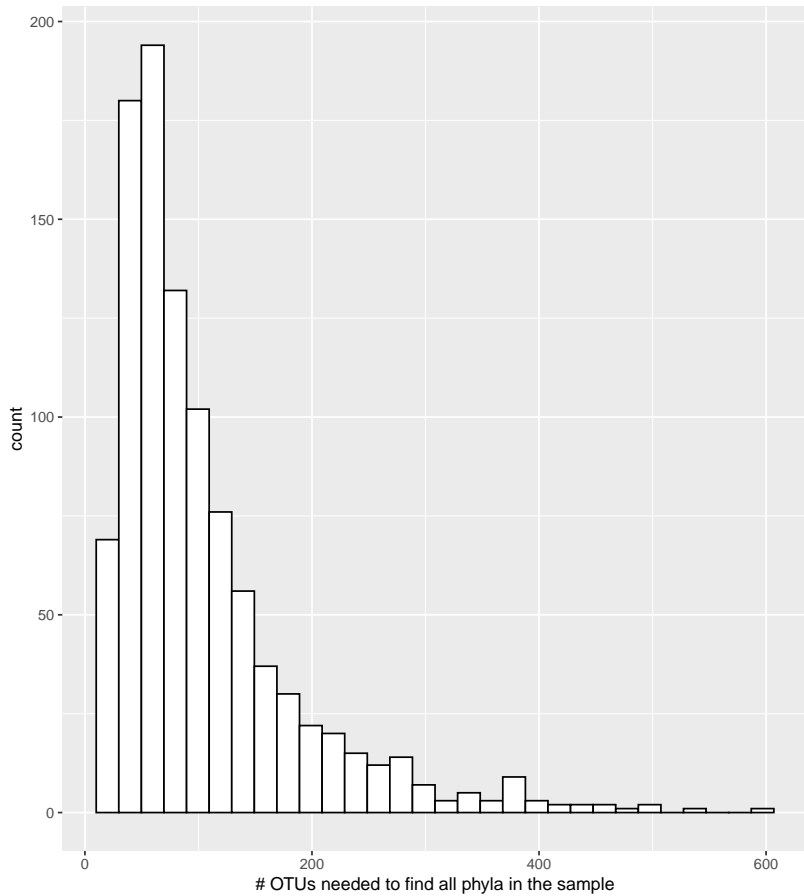
18S.2.2.E.PCM.LT



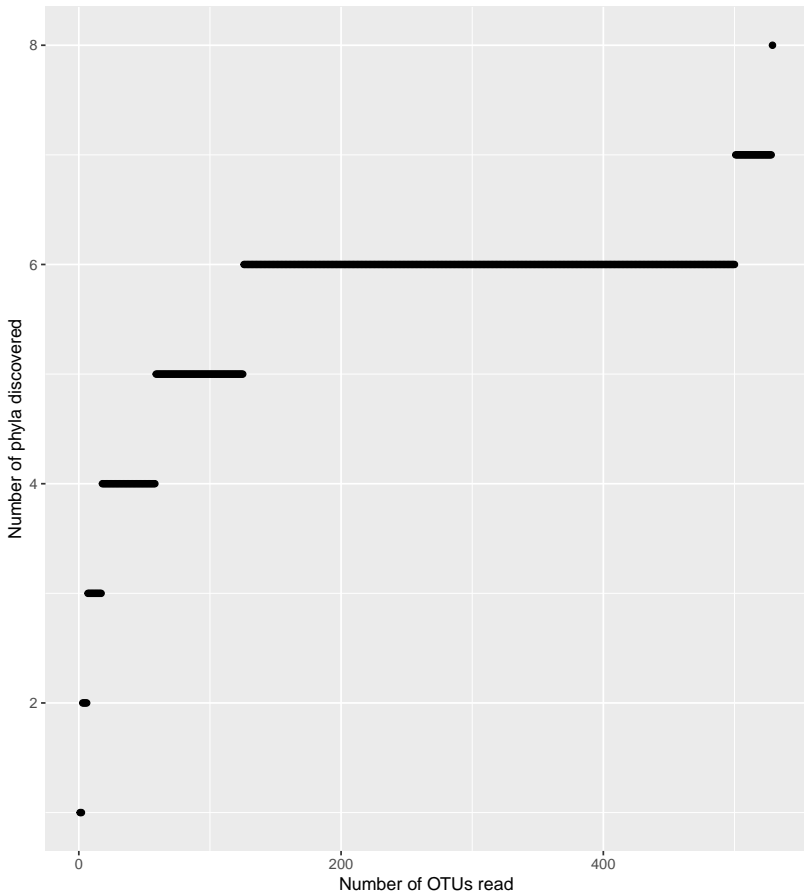
97. 18S.2.2.F.PCM.LT



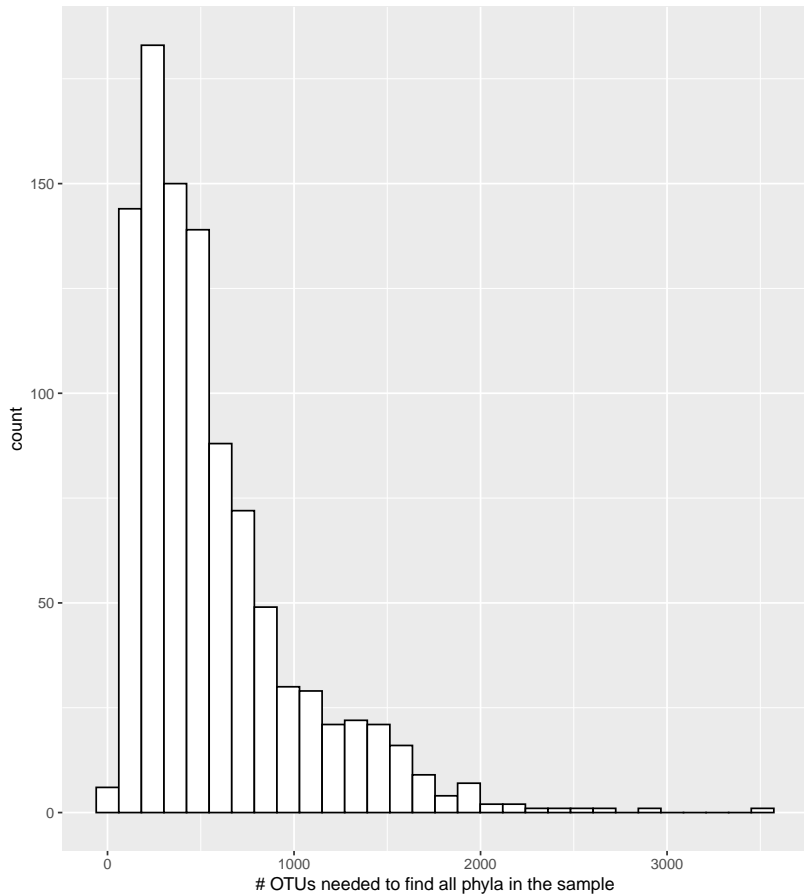
18S.2.2.F.PCM.LT

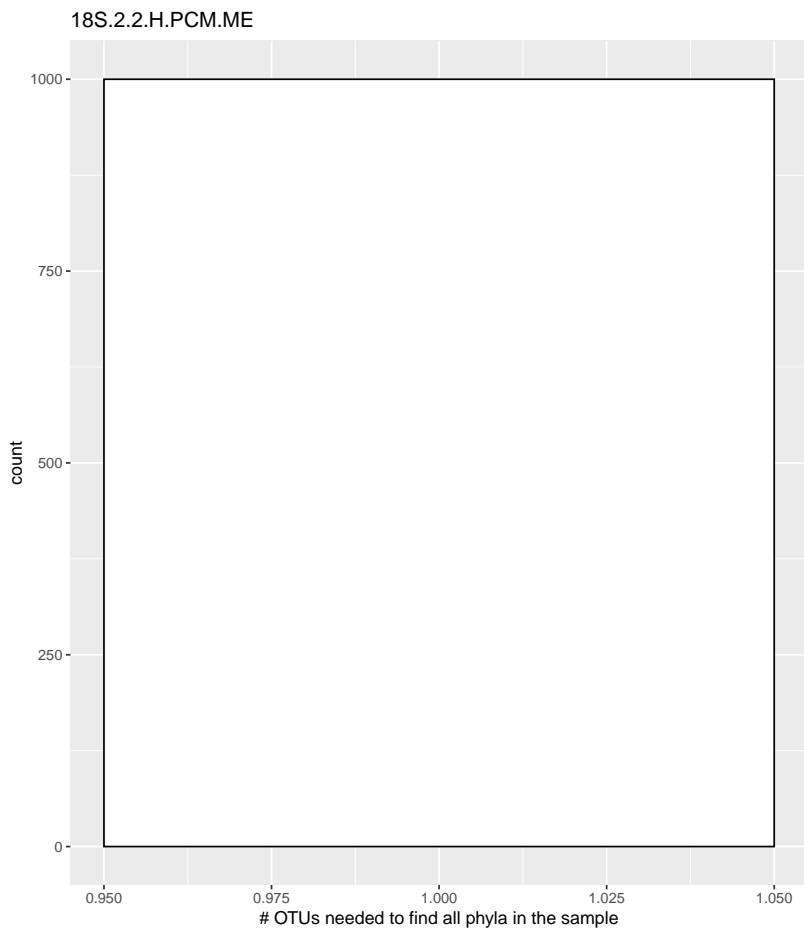
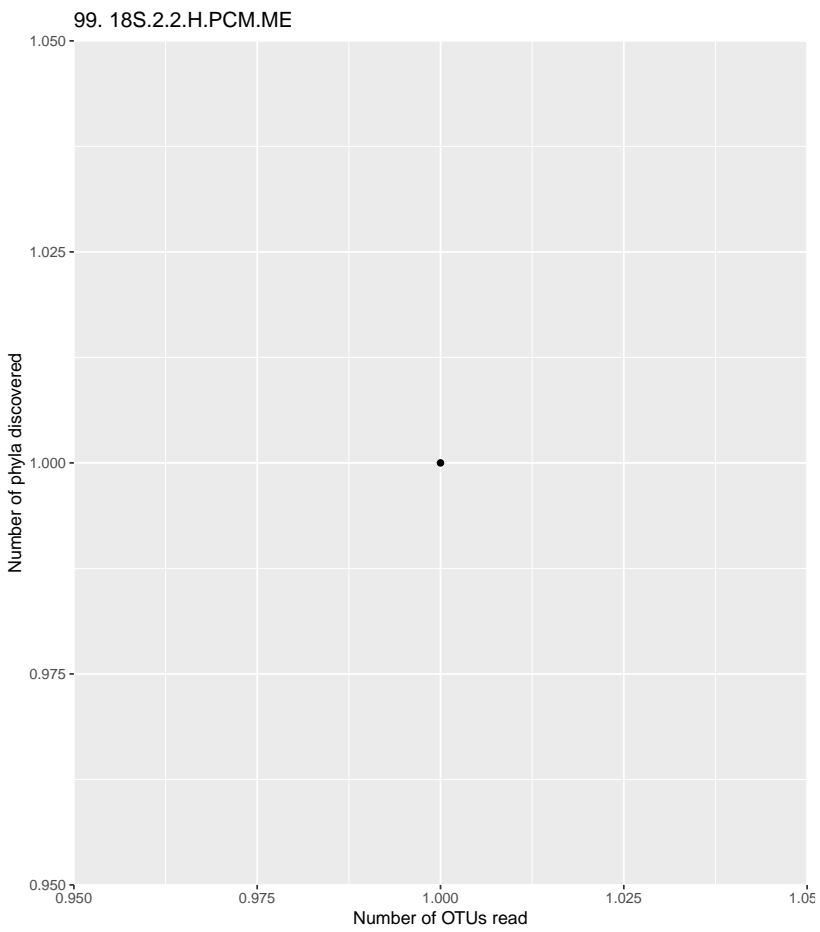


98. 18S.2.2.G.PCM.LT

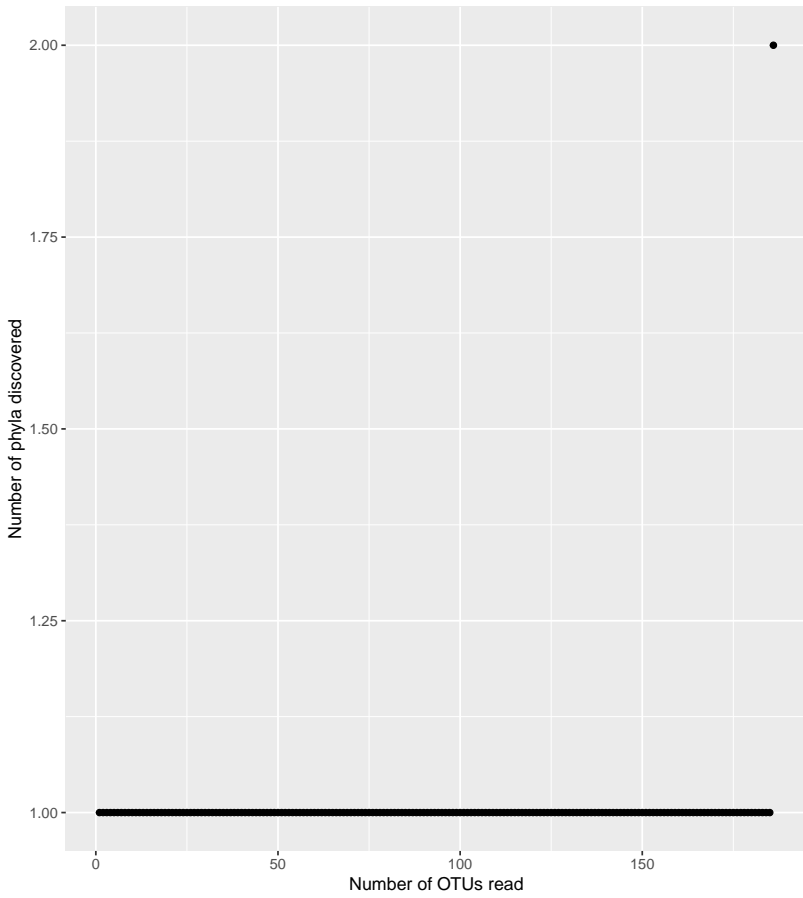


18S.2.2.G.PCM.LT

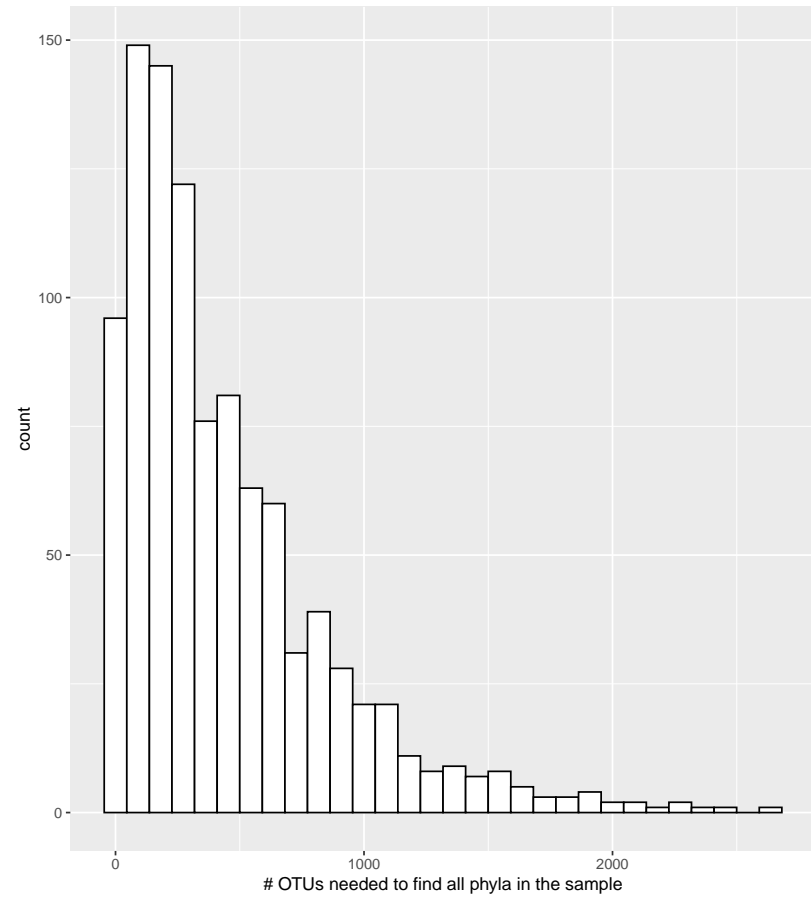


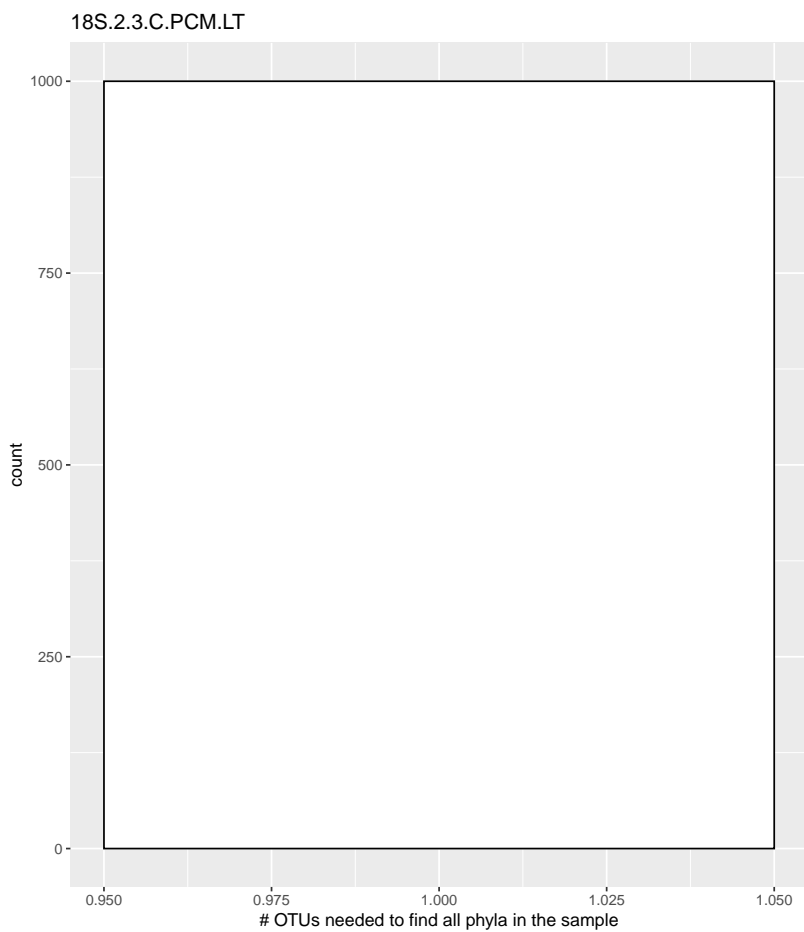
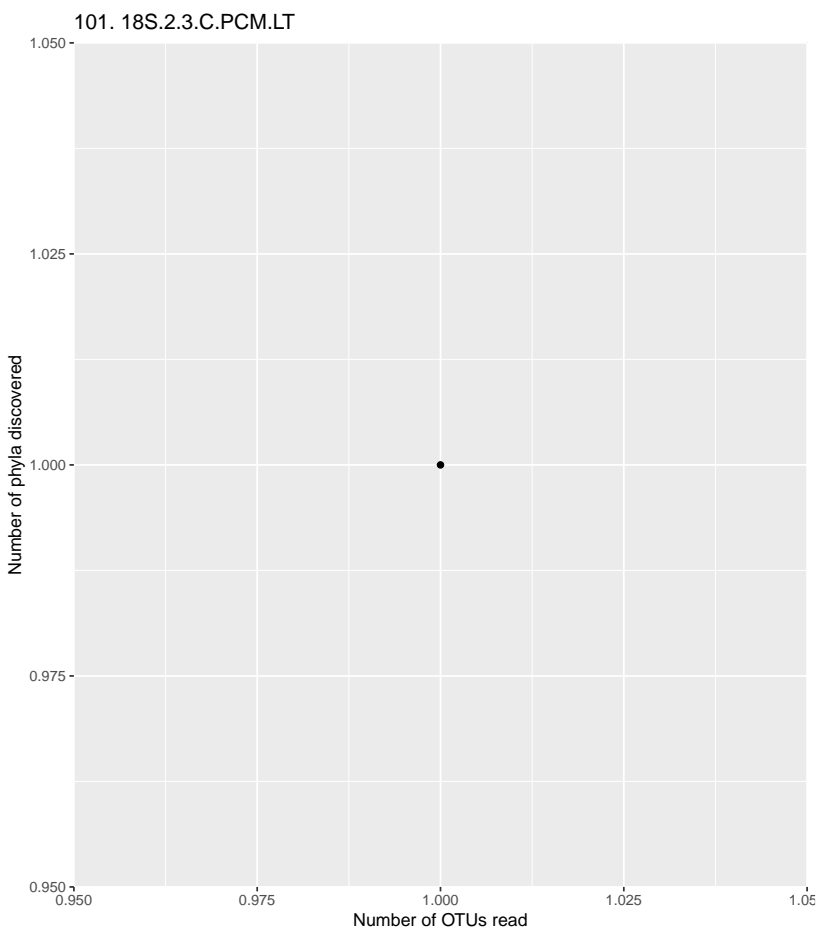


100. 18S.2.3.A.PCM.ME

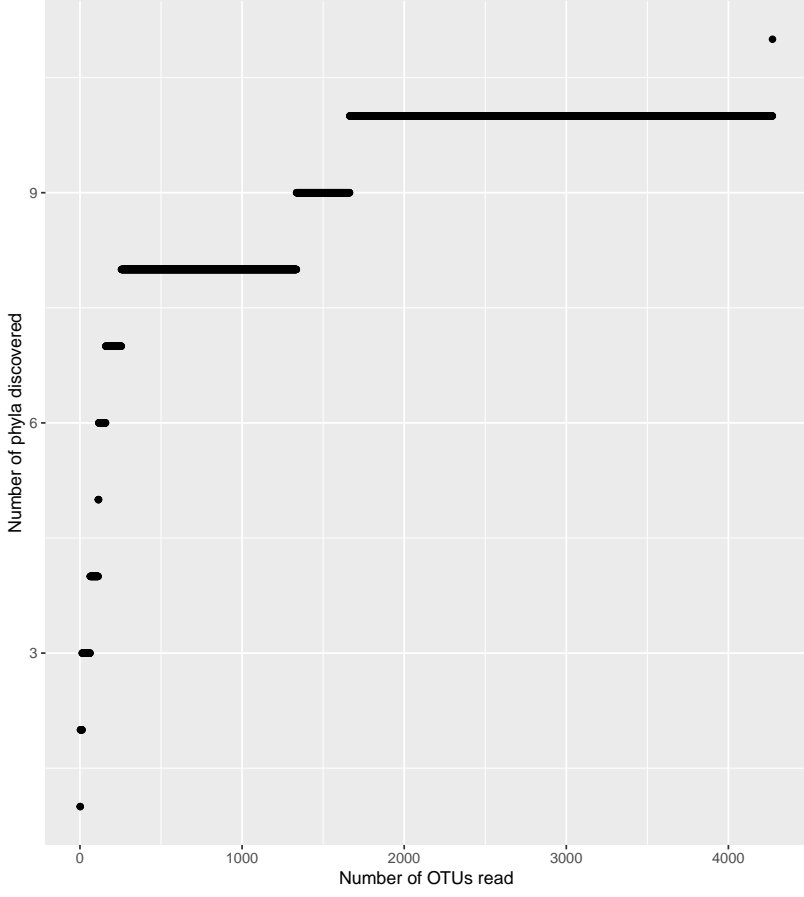


18S.2.3.A.PCM.ME

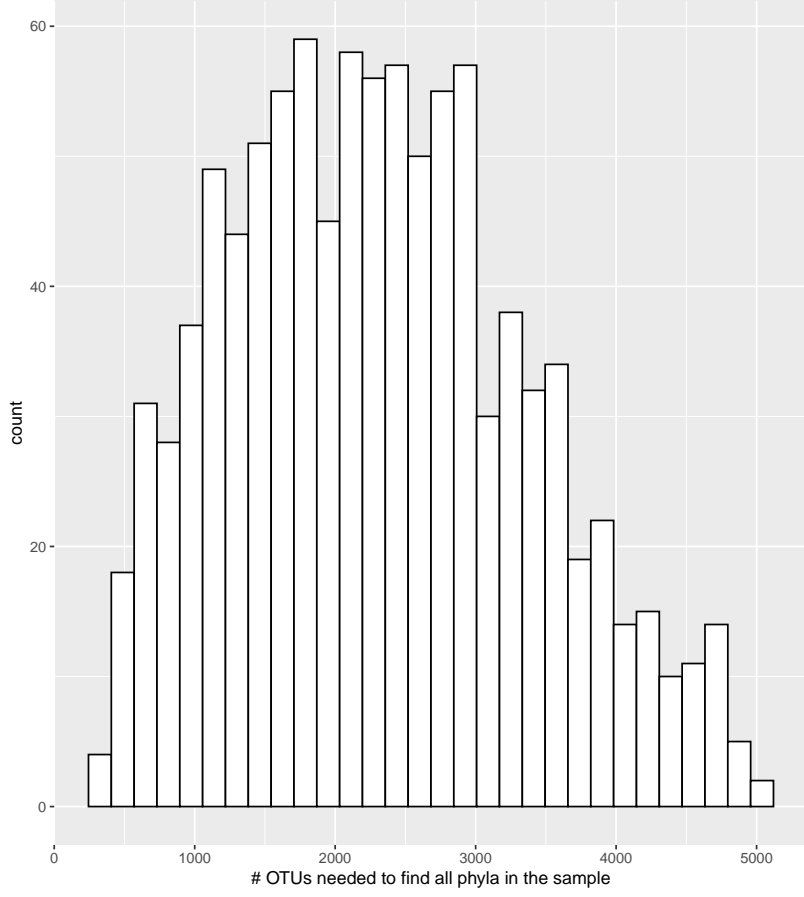




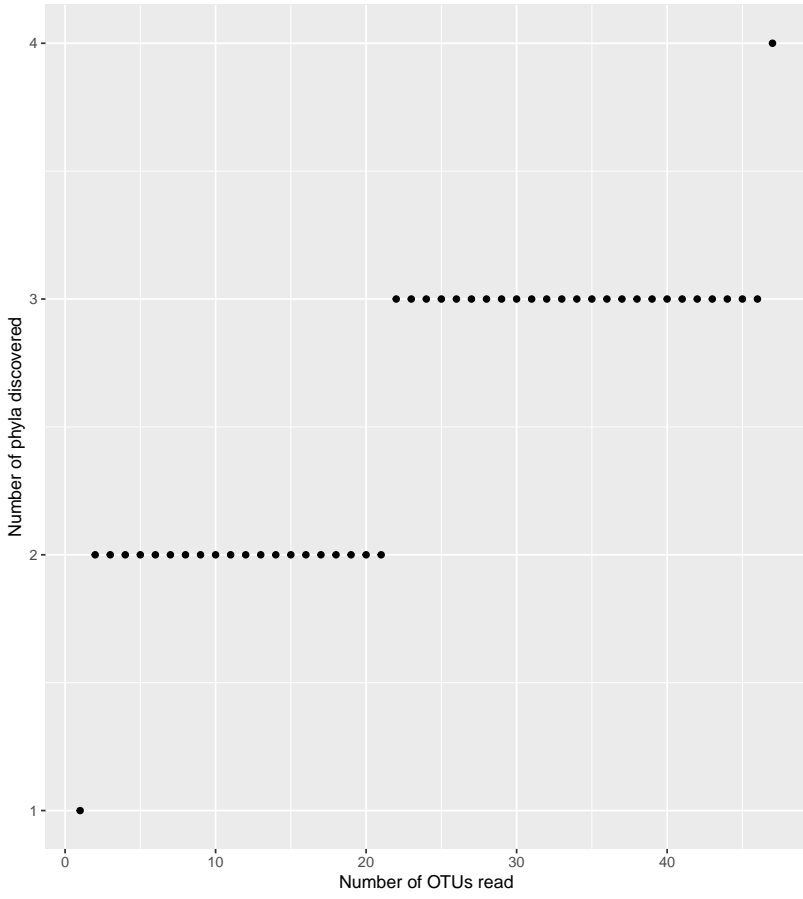
102. 18S.2.3.D.PCM.LT



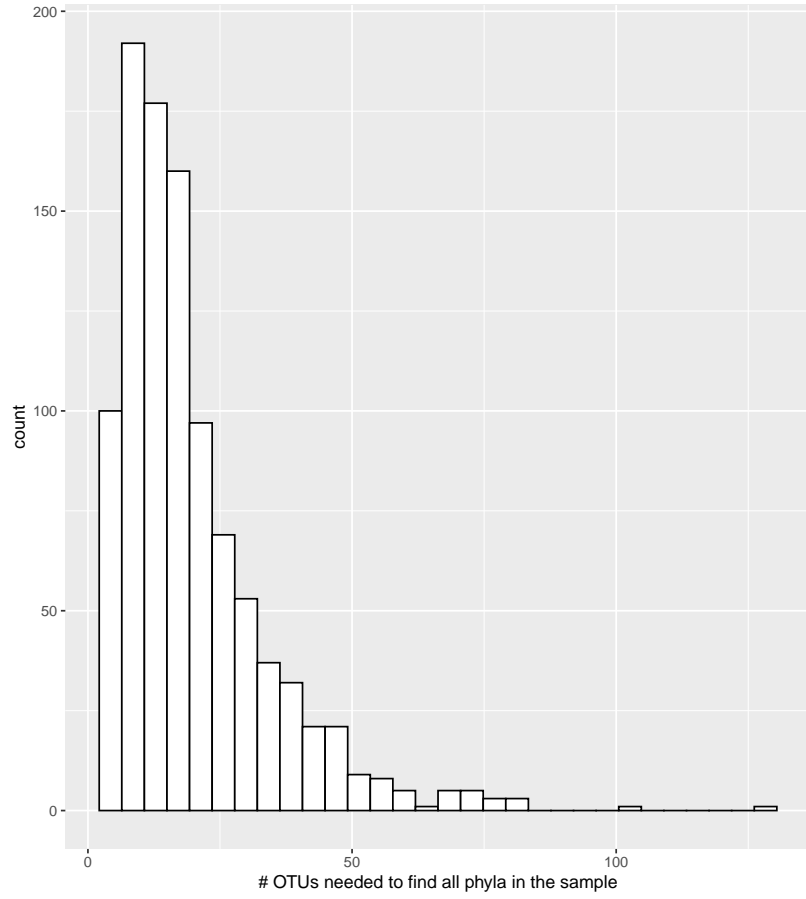
18S.2.3.D.PCM.LT

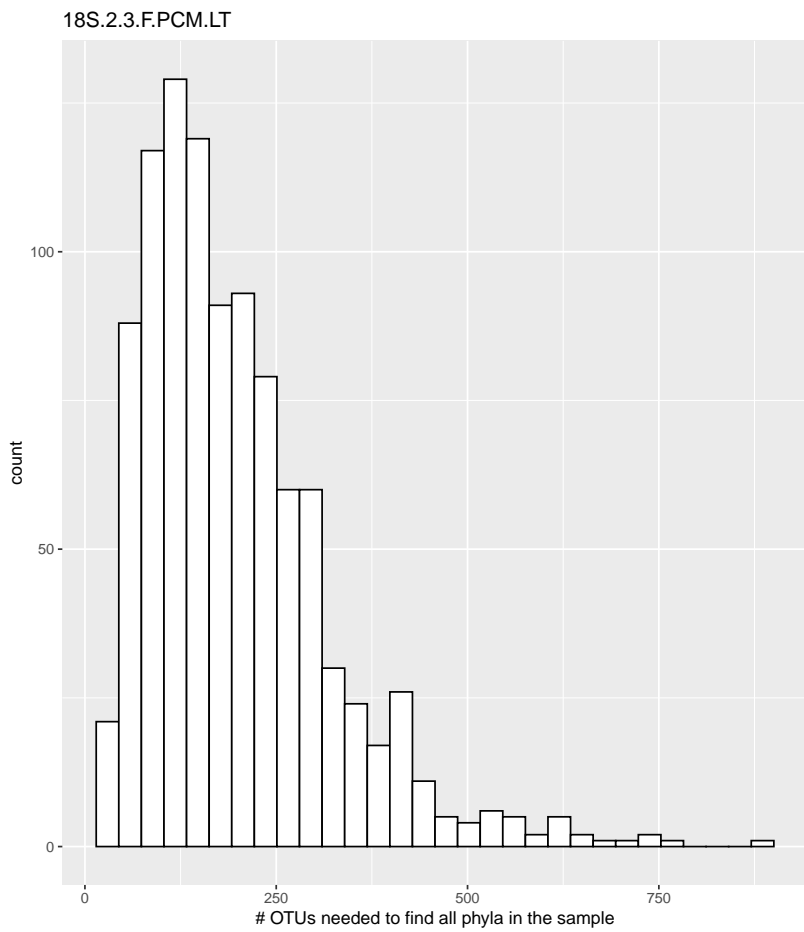
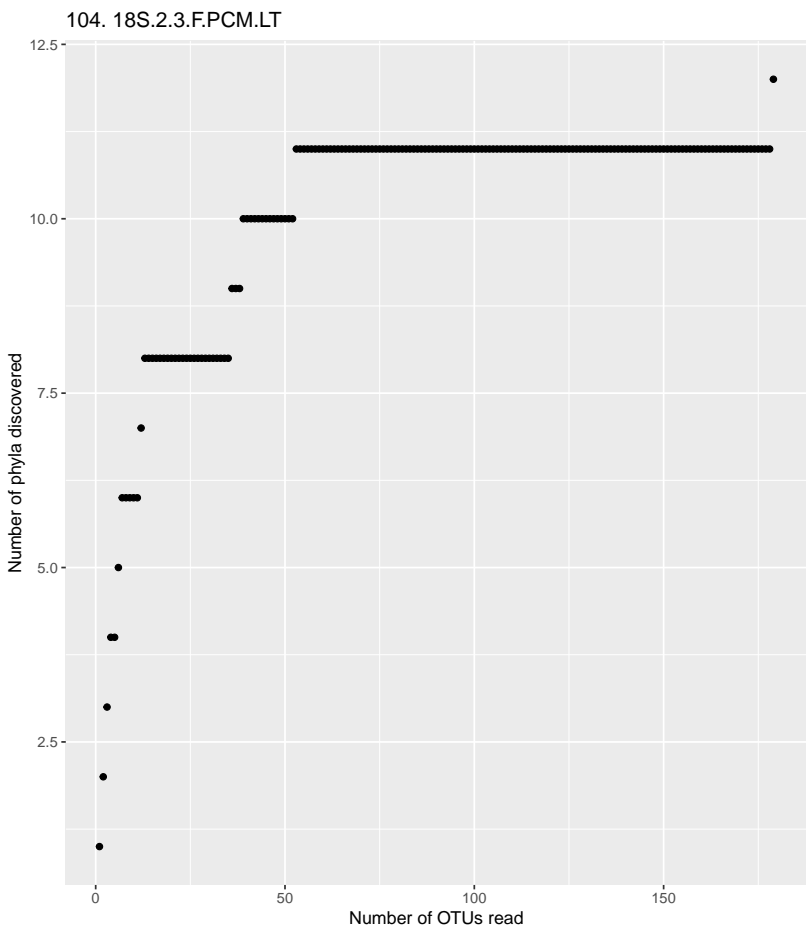


103. 18S.2.3.E.PCM.LT

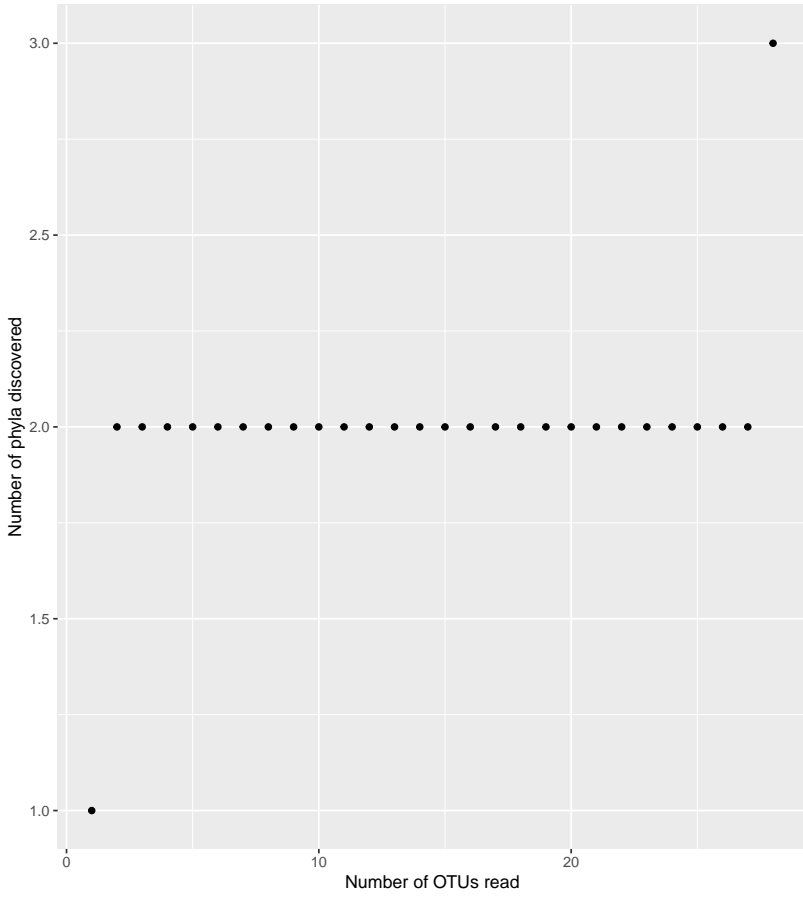


18S.2.3.E.PCM.LT

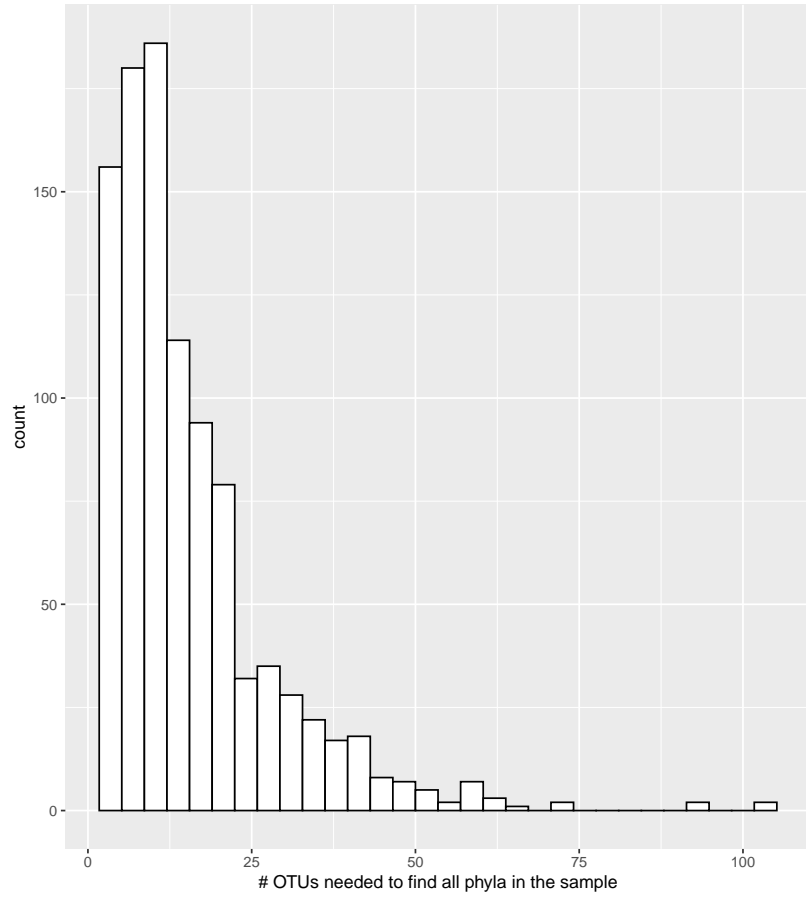




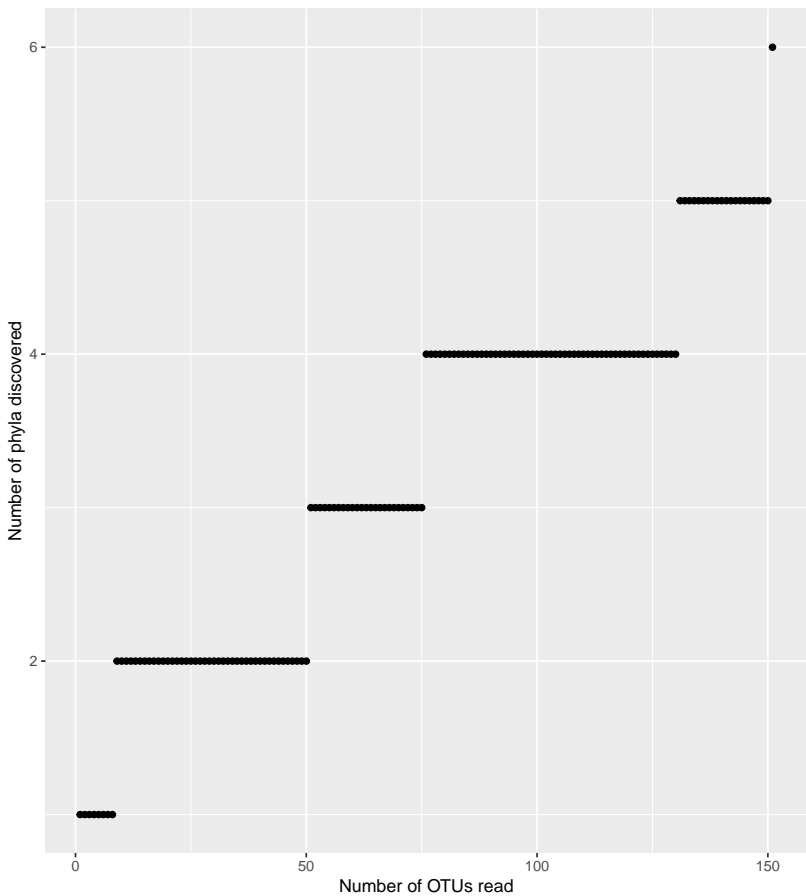
105. 18S.2.3.G.PCM.LT



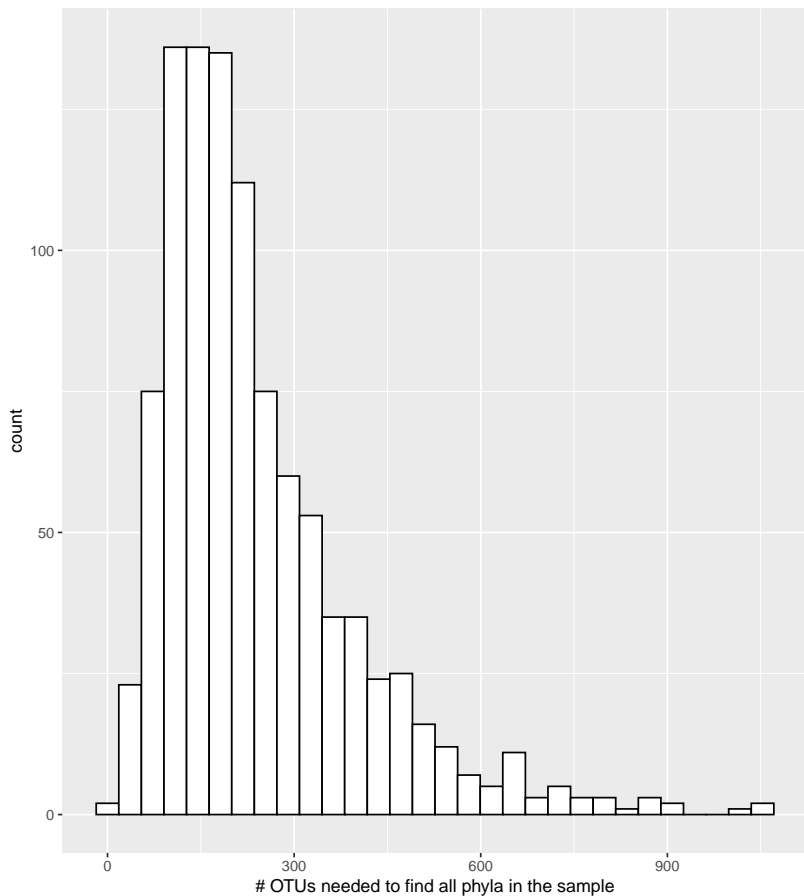
18S.2.3.G.PCM.LT



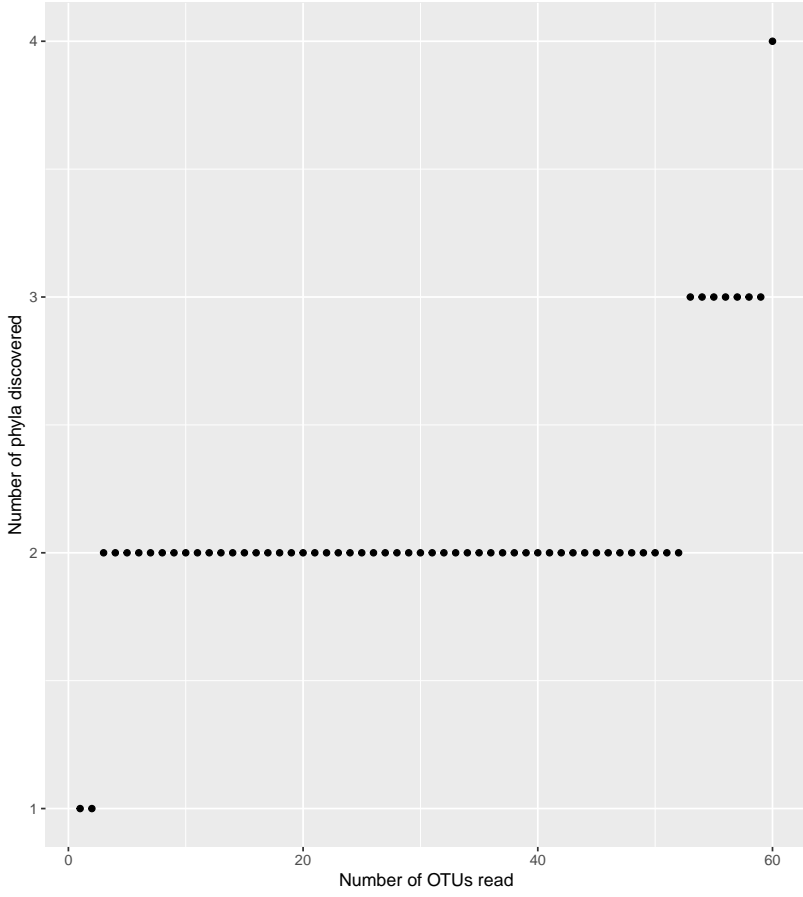
106. 18S.2.4.A.PCM.ME



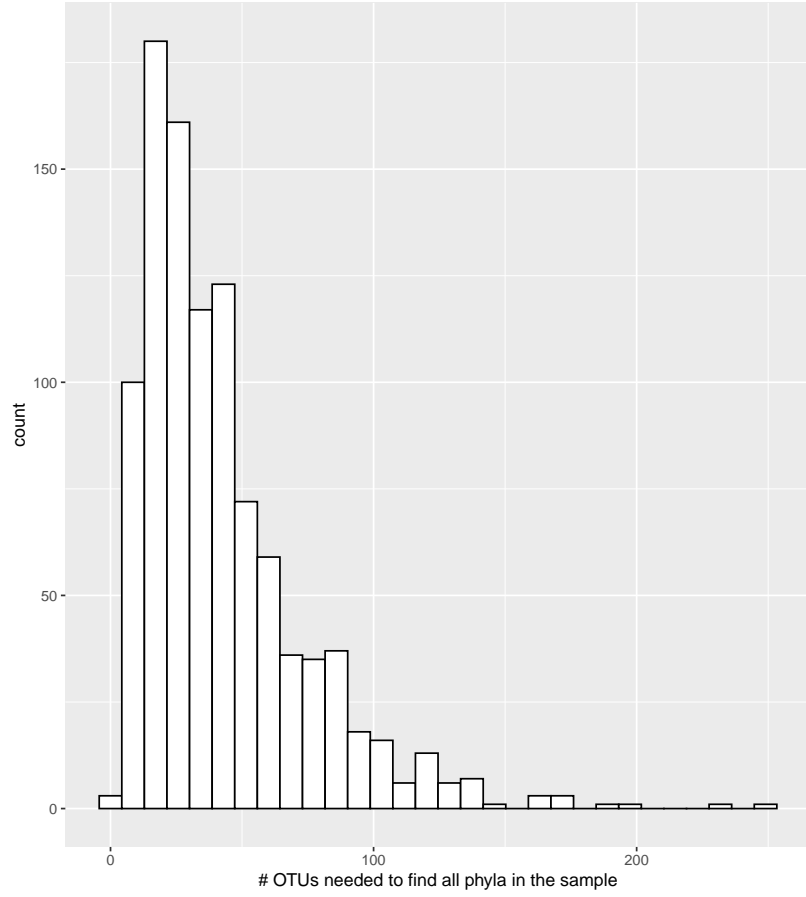
18S.2.4.A.PCM.ME



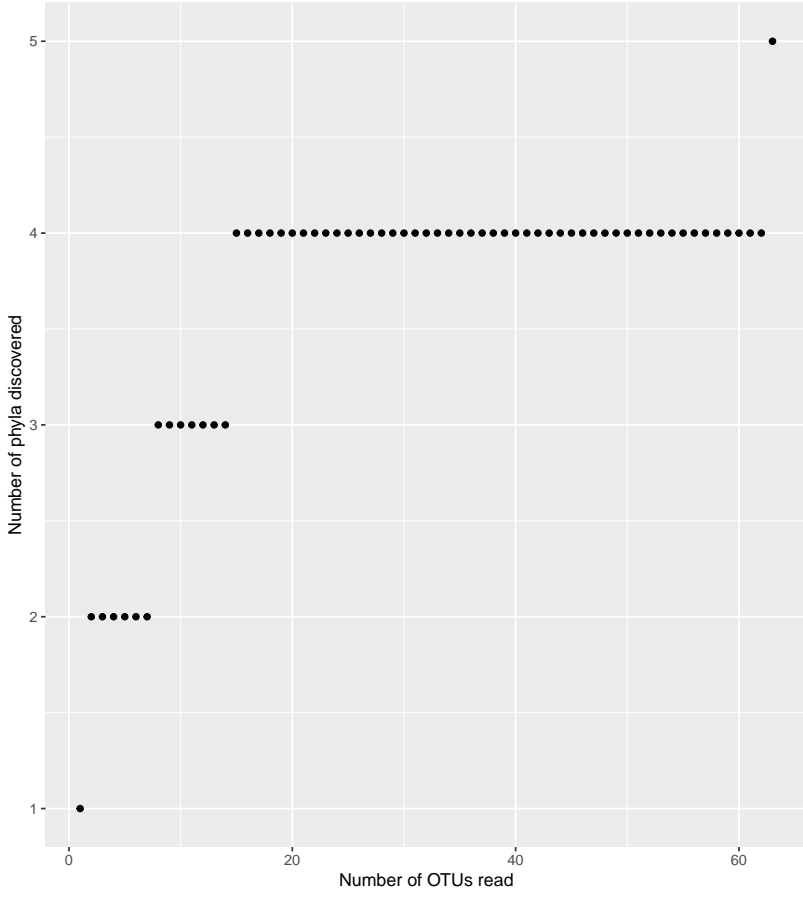
107. 18S.2.4.B.PCM.ME



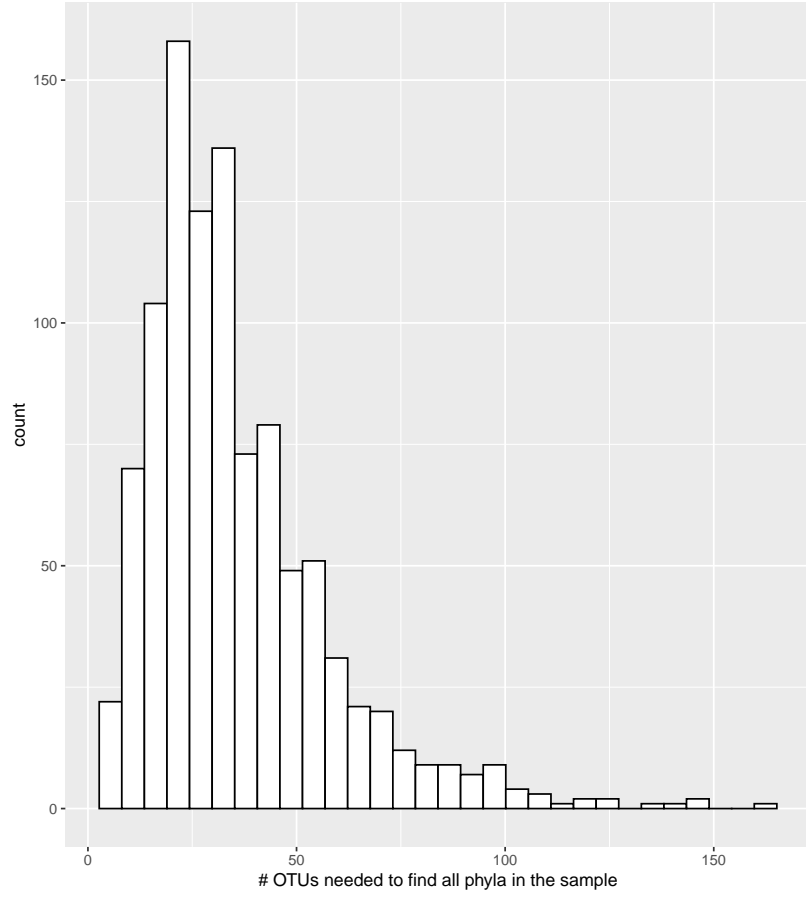
18S.2.4.B.PCM.ME



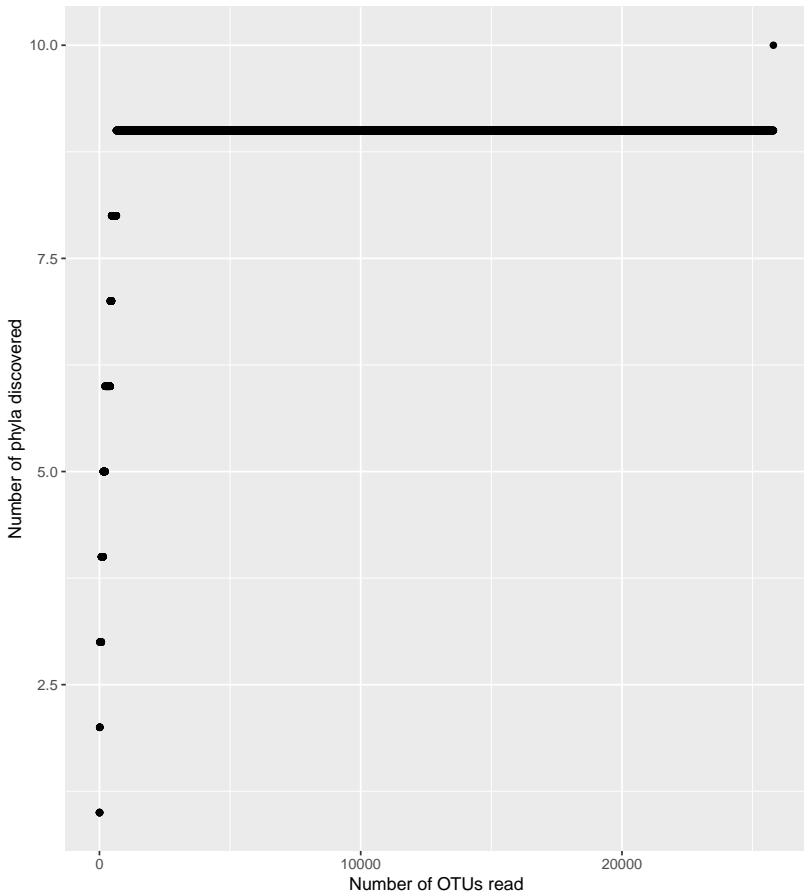
108. 18S.2.4.C.PCM.LT



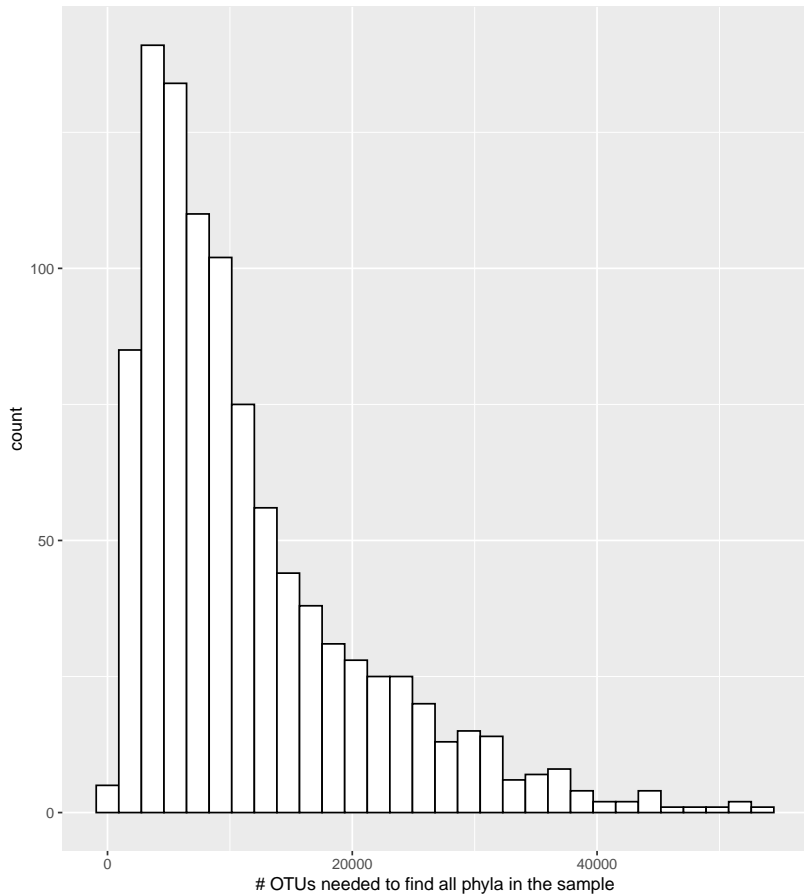
18S.2.4.C.PCM.LT



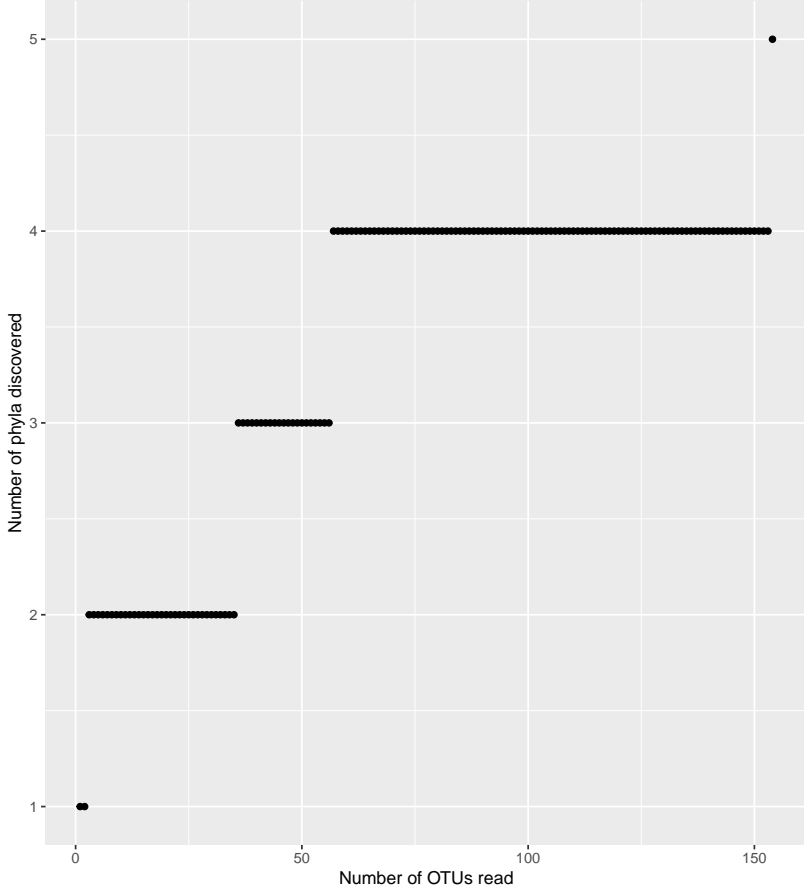
109. 18S.2.4.D.PCM.LT



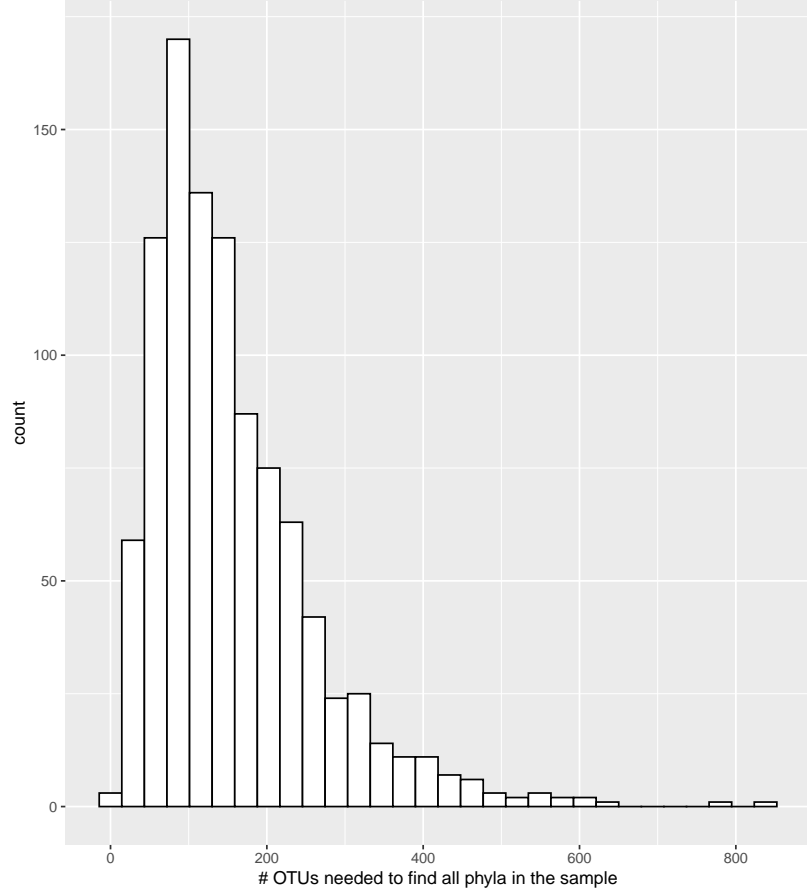
18S.2.4.D.PCM.LT



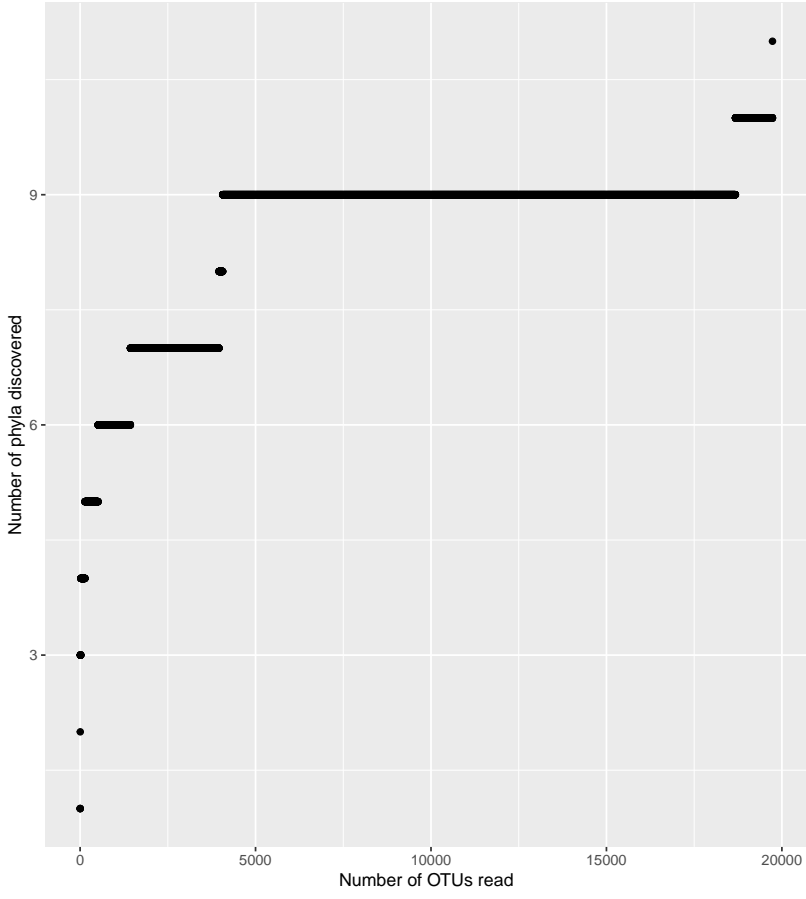
110. 18S.2.4.E.PCM.LT



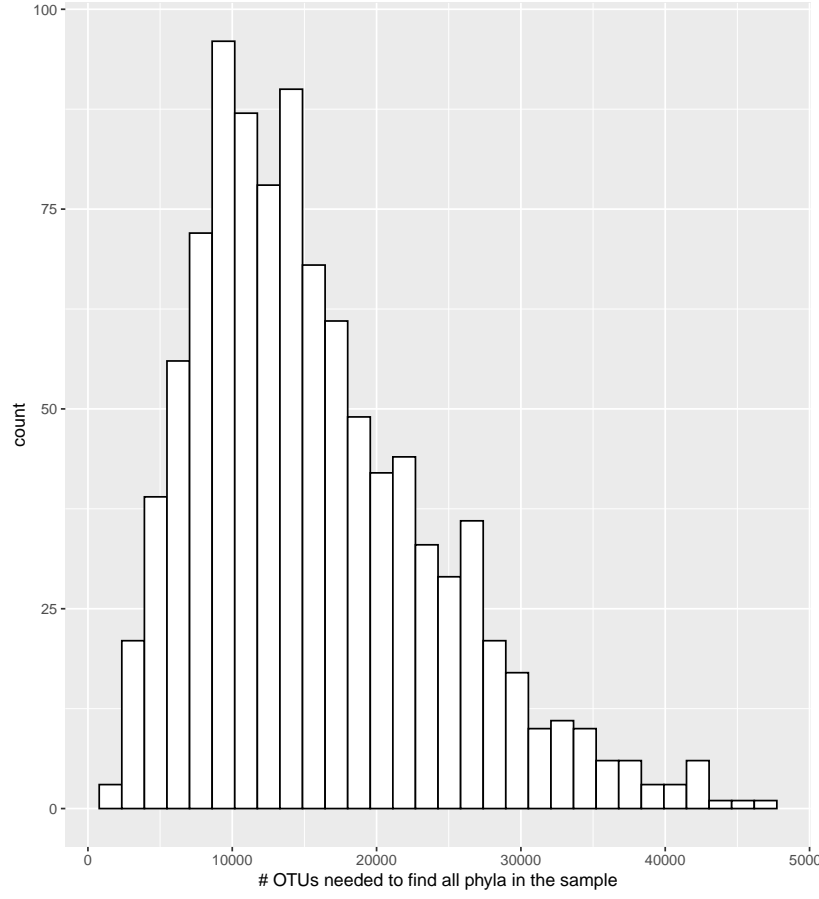
18S.2.4.E.PCM.LT



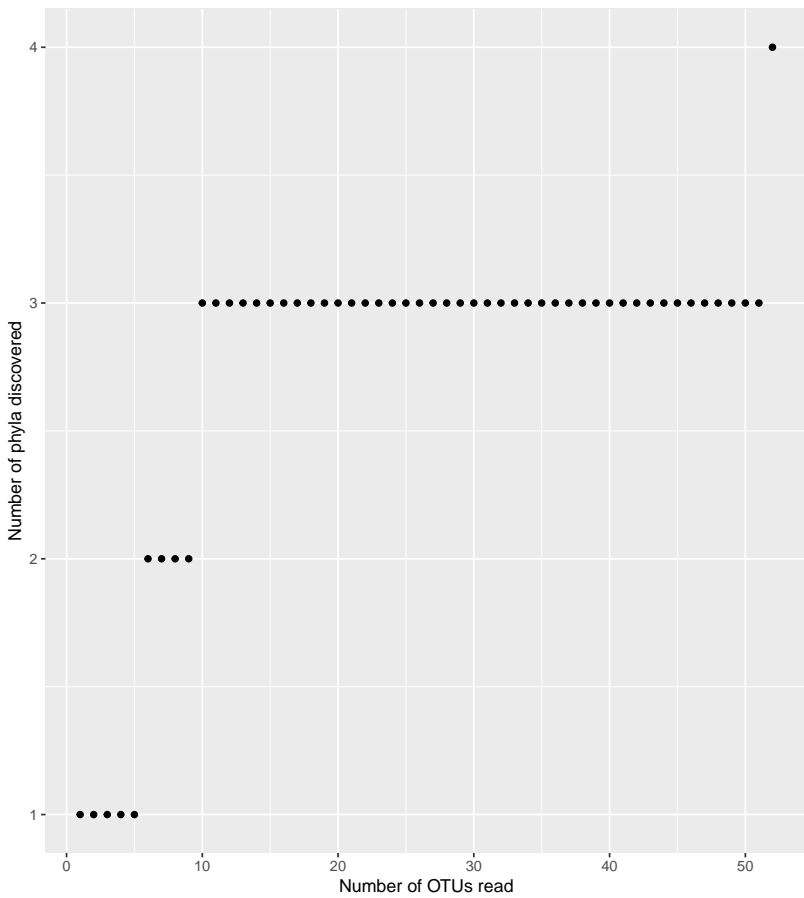
111. 18S.2.4.F.PCM.LT



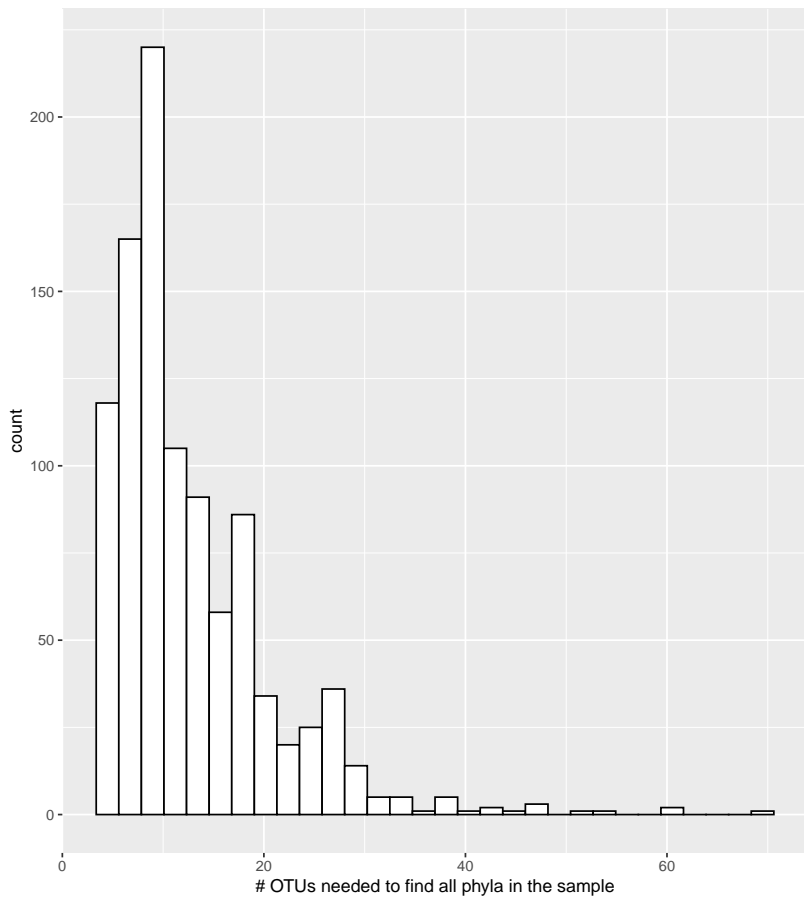
18S.2.4.F.PCM.LT



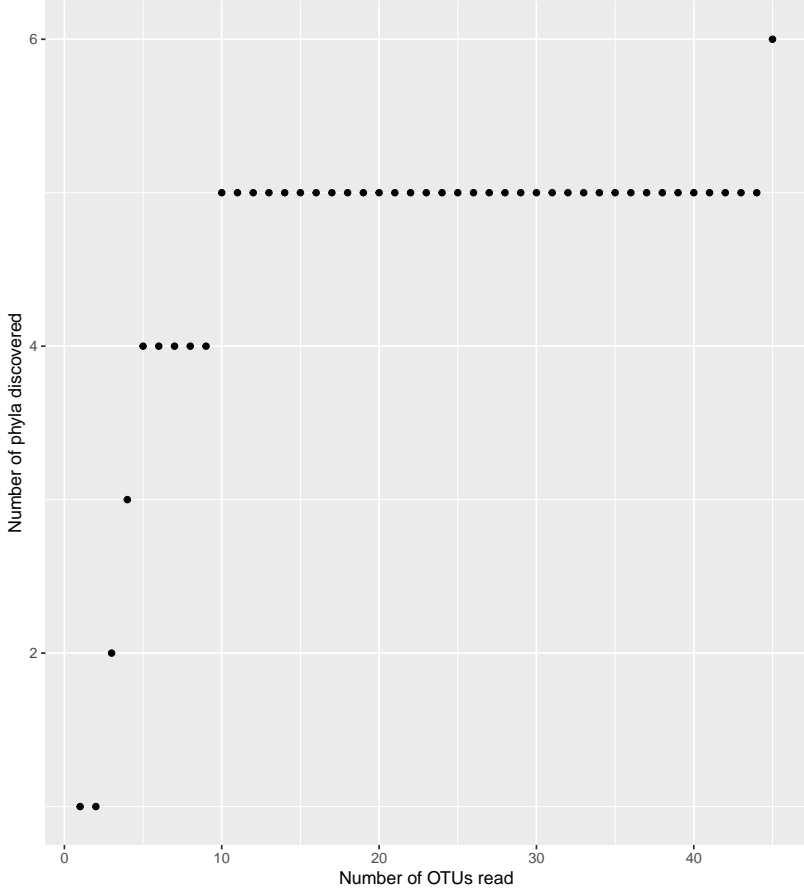
112. 18S.2.4.G.PCM.LT



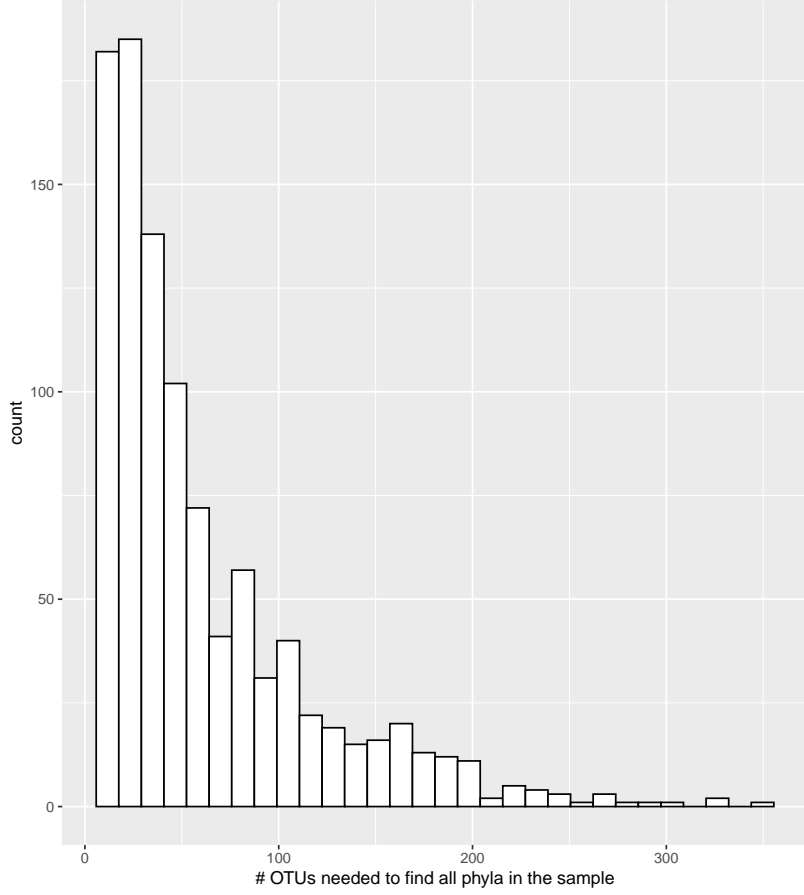
18S.2.4.G.PCM.LT



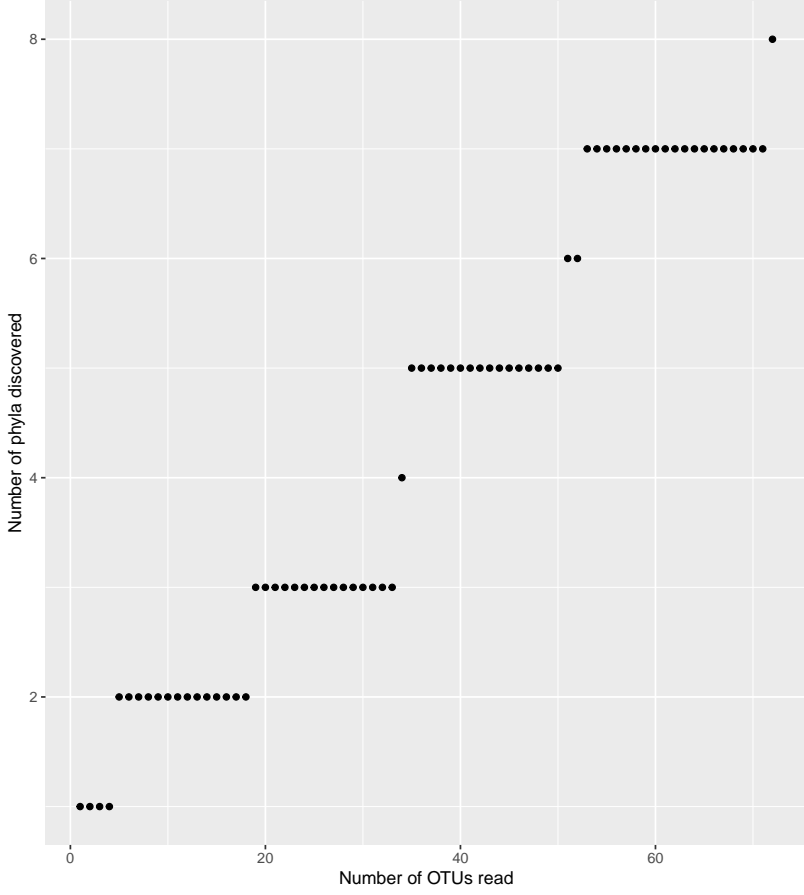
113. 18S.2.5.A.PCM.ME



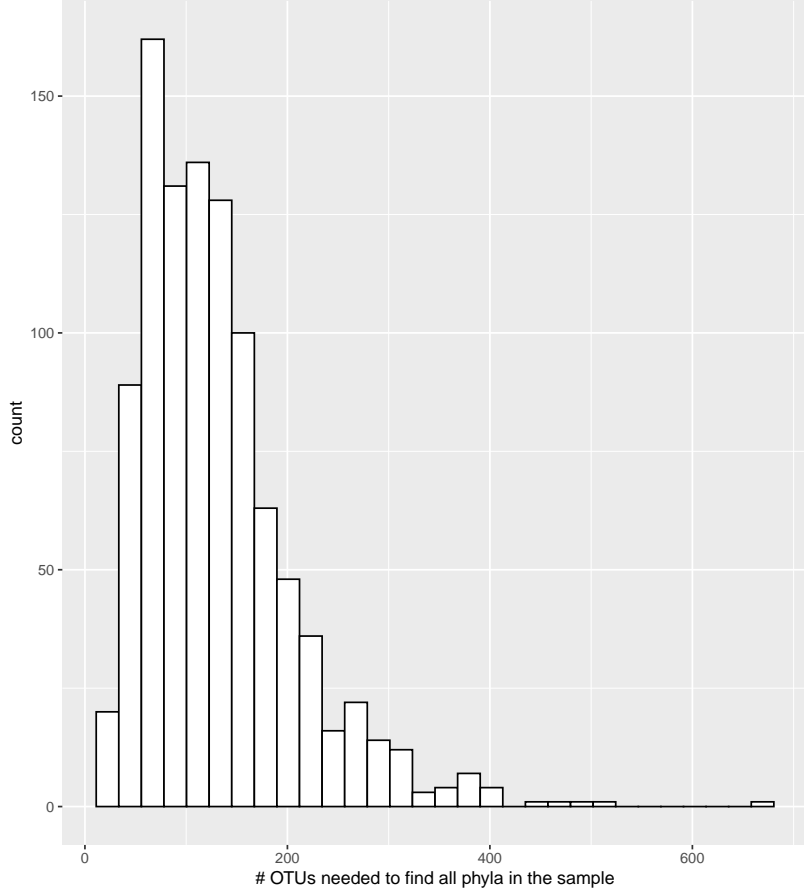
18S.2.5.A.PCM.ME



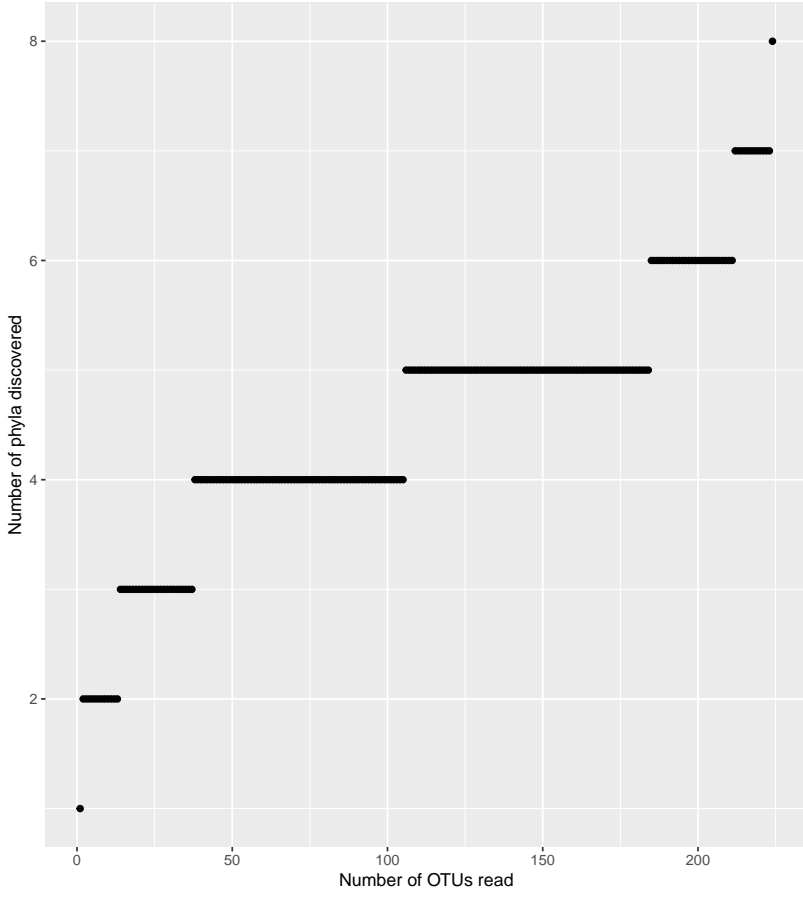
114. 18S.2.5.B.PCM.ME



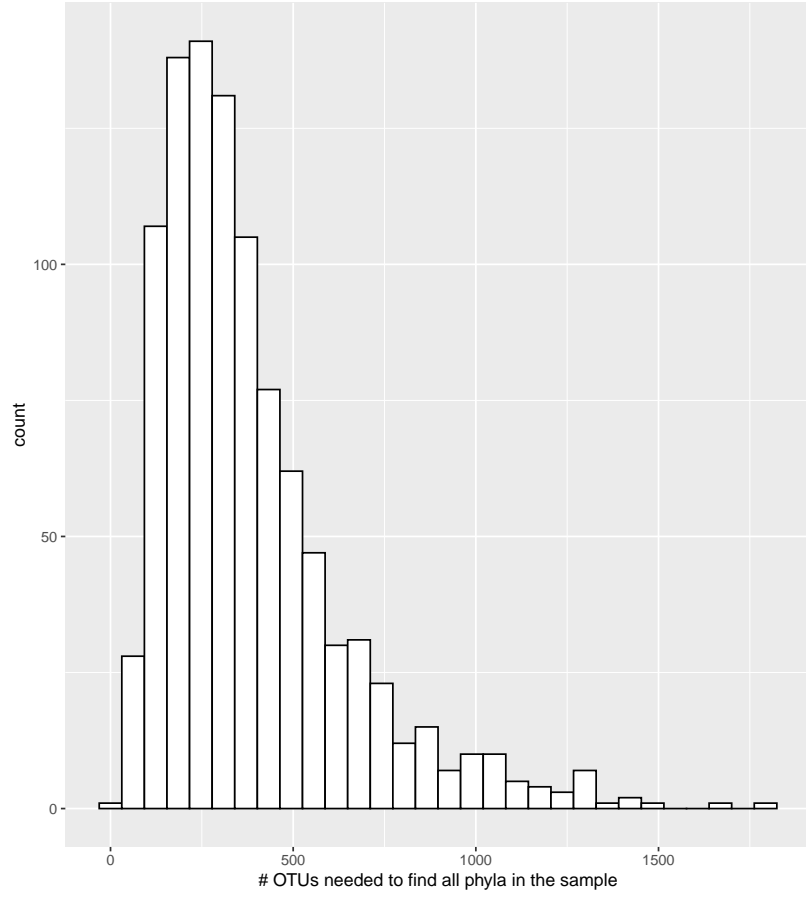
18S.2.5.B.PCM.ME



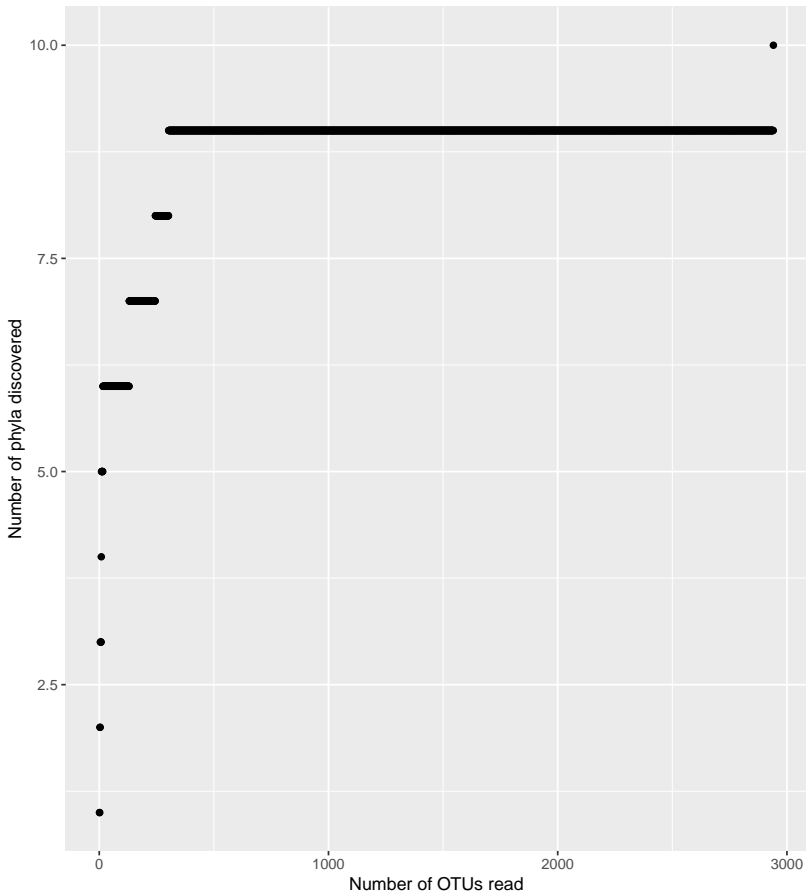
115. 18S.2.5.C.PCM.LT



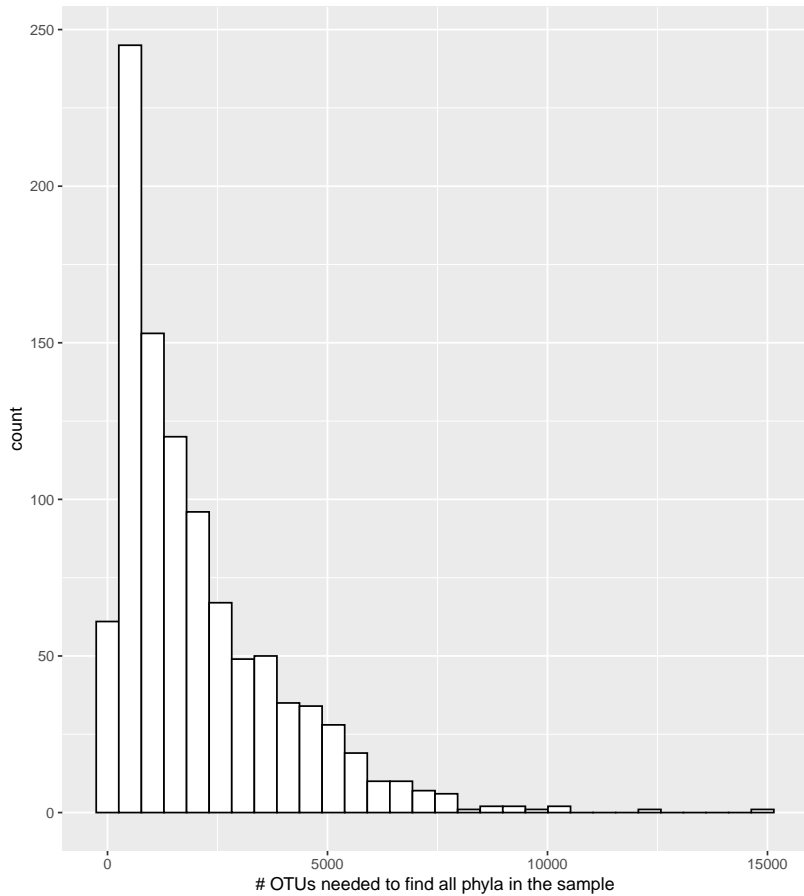
18S.2.5.C.PCM.LT



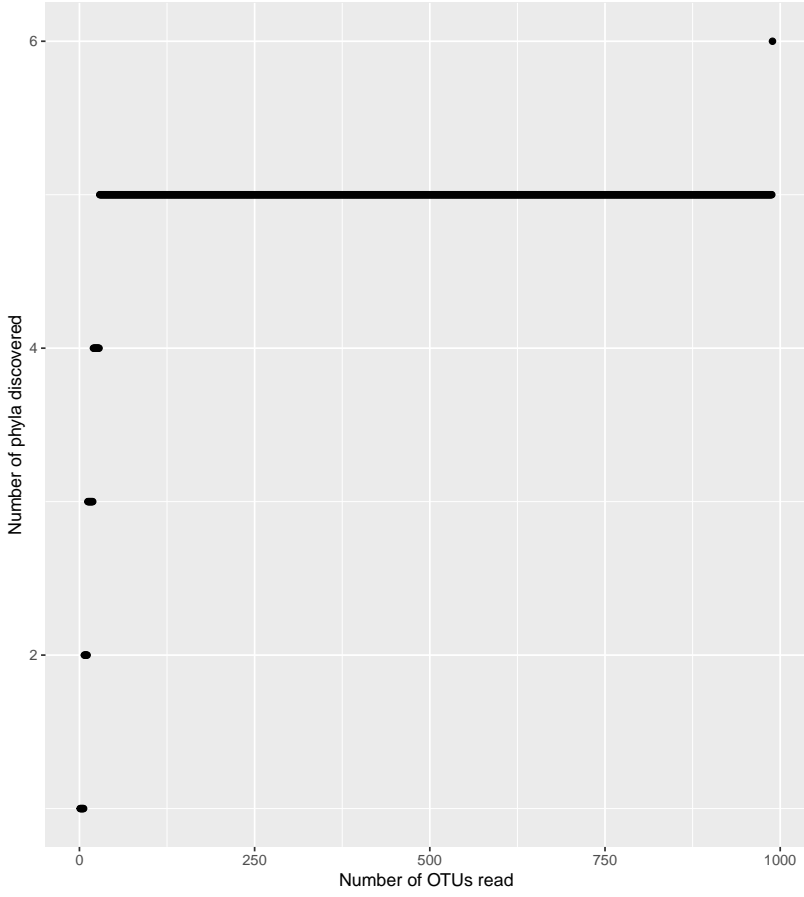
116. 18S.2.5.D.PCM.LT



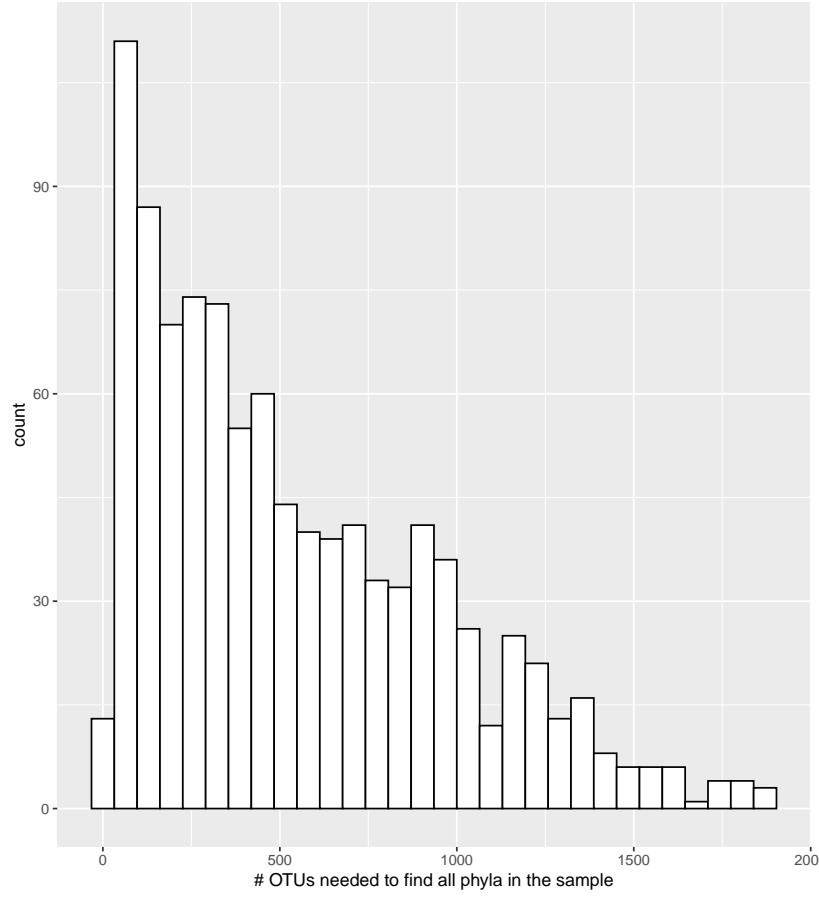
18S.2.5.D.PCM.LT



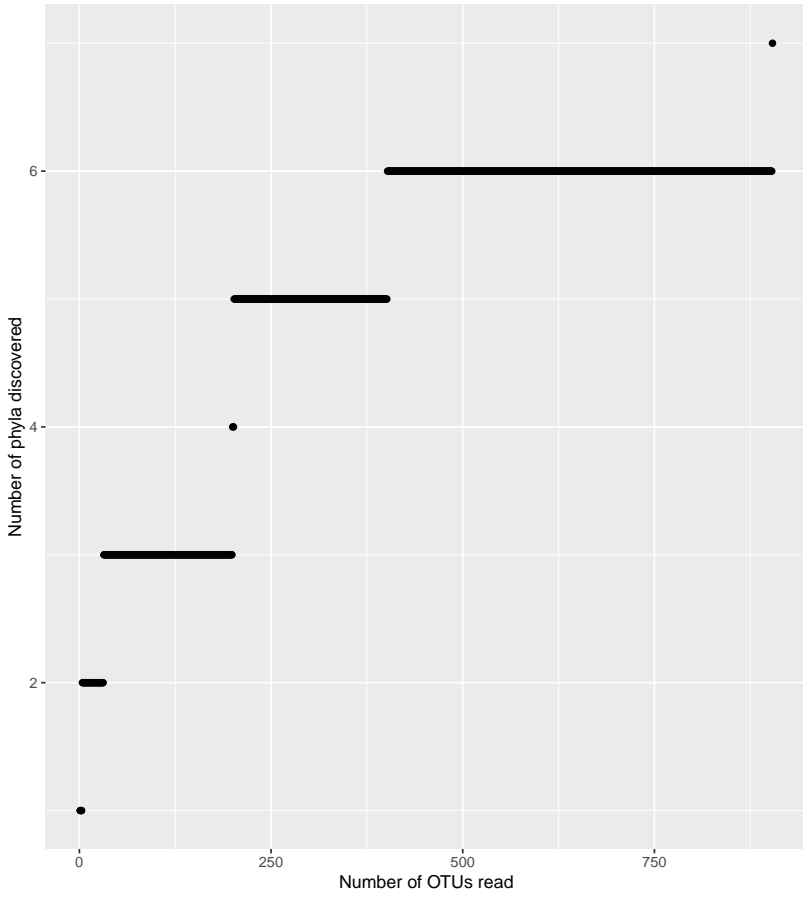
117. 18S.2.5.F.PCM.LT



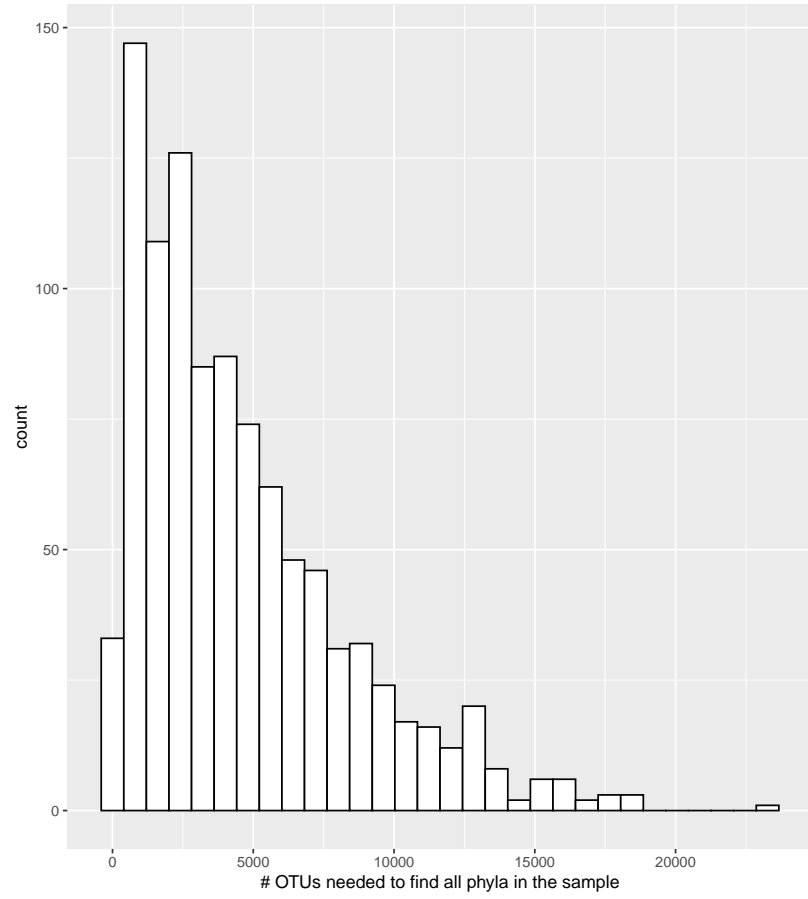
18S.2.5.F.PCM.LT



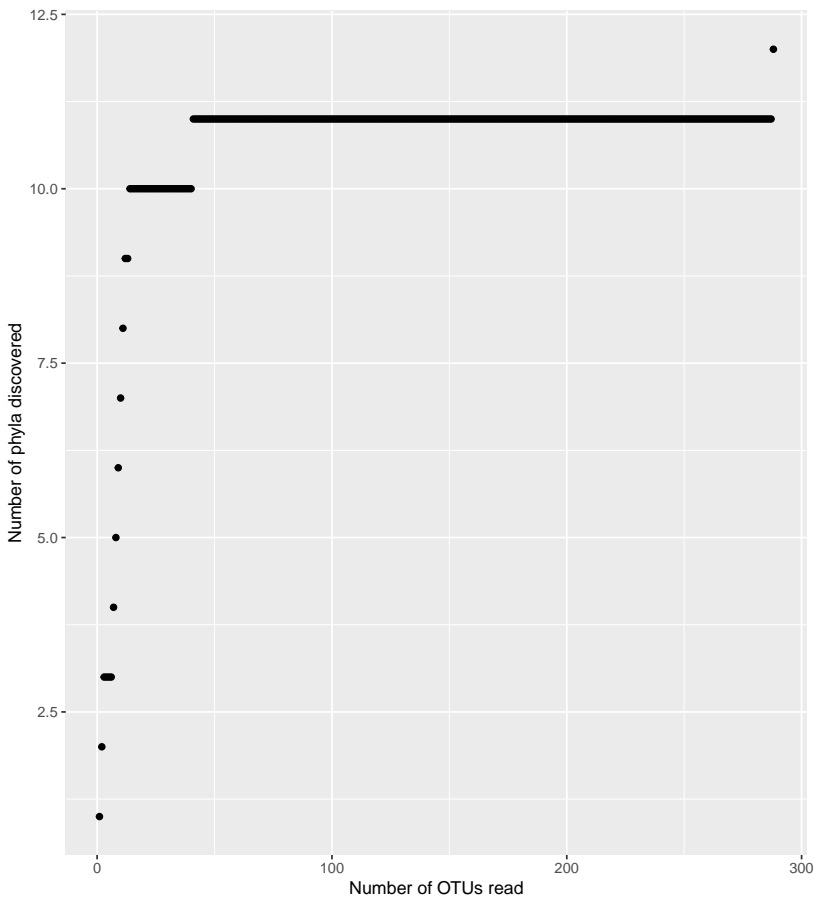
118. 18S.2.5.G.PCM.IT



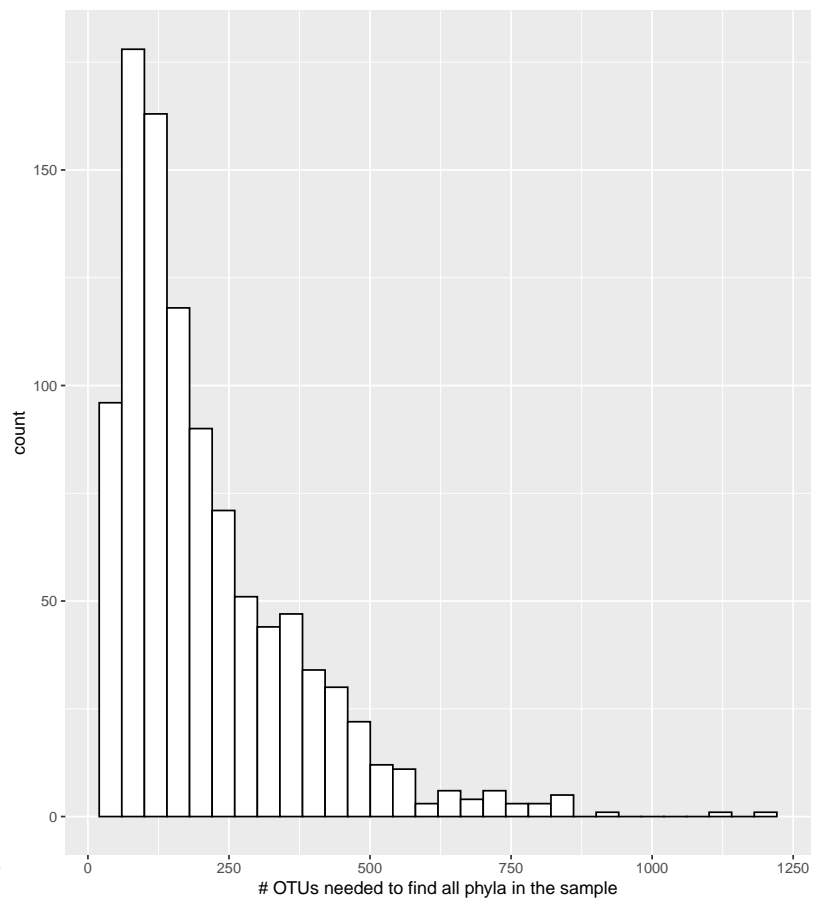
18S.2.5.G.PCM.IT



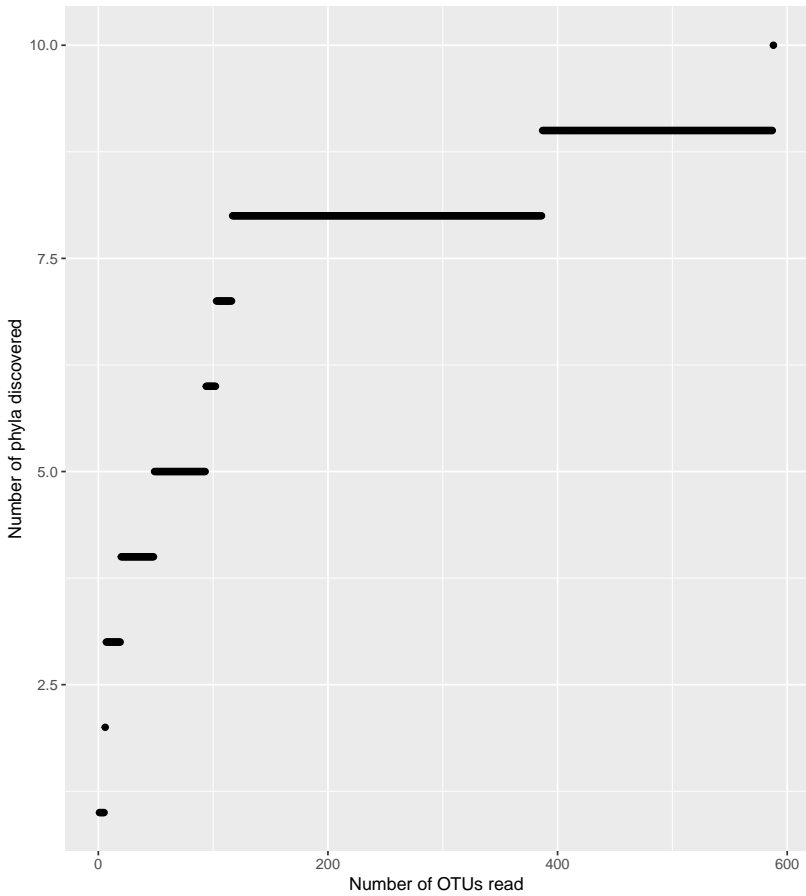
119. 18S.2.6.A.PCM.ME



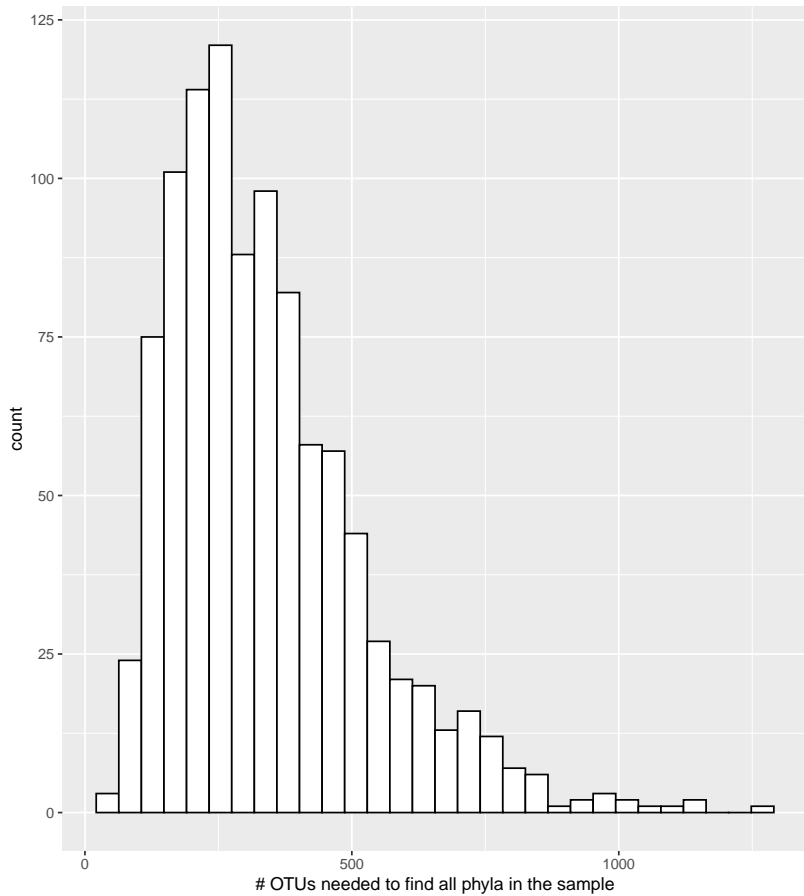
18S.2.6.A.PCM.ME



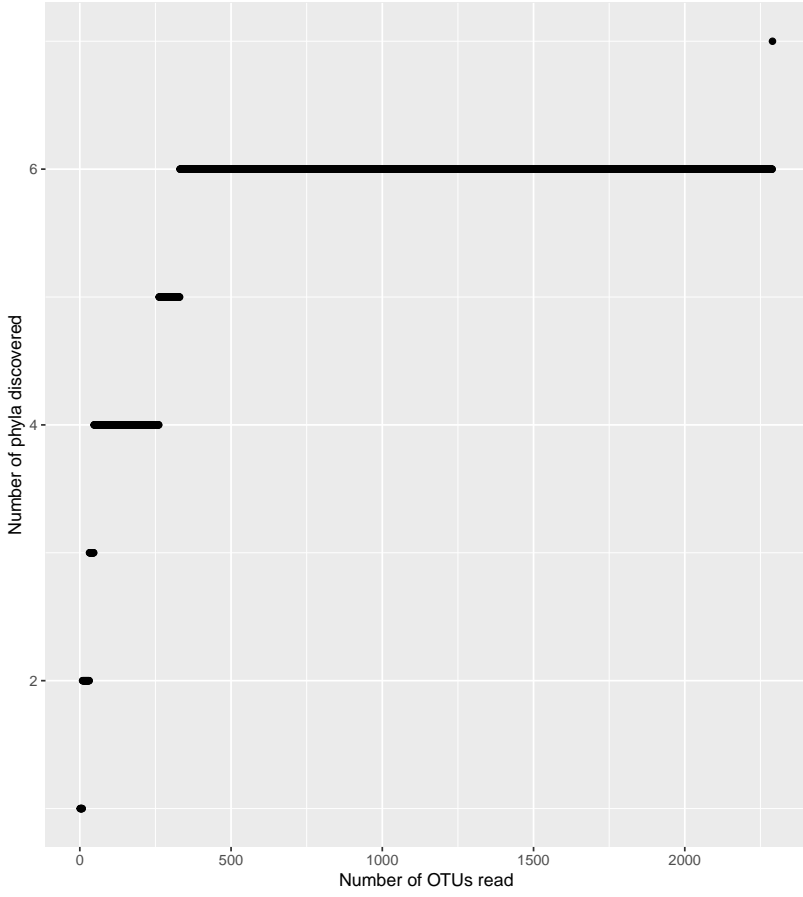
120. 18S.2.6.B.PCM.ME



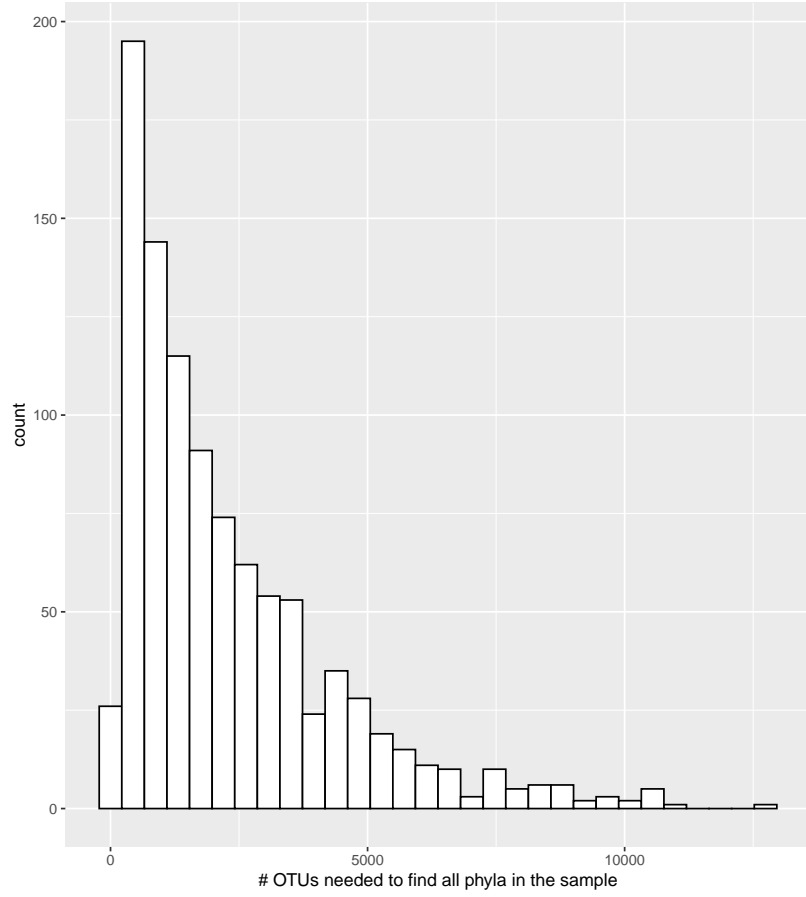
18S.2.6.B.PCM.ME



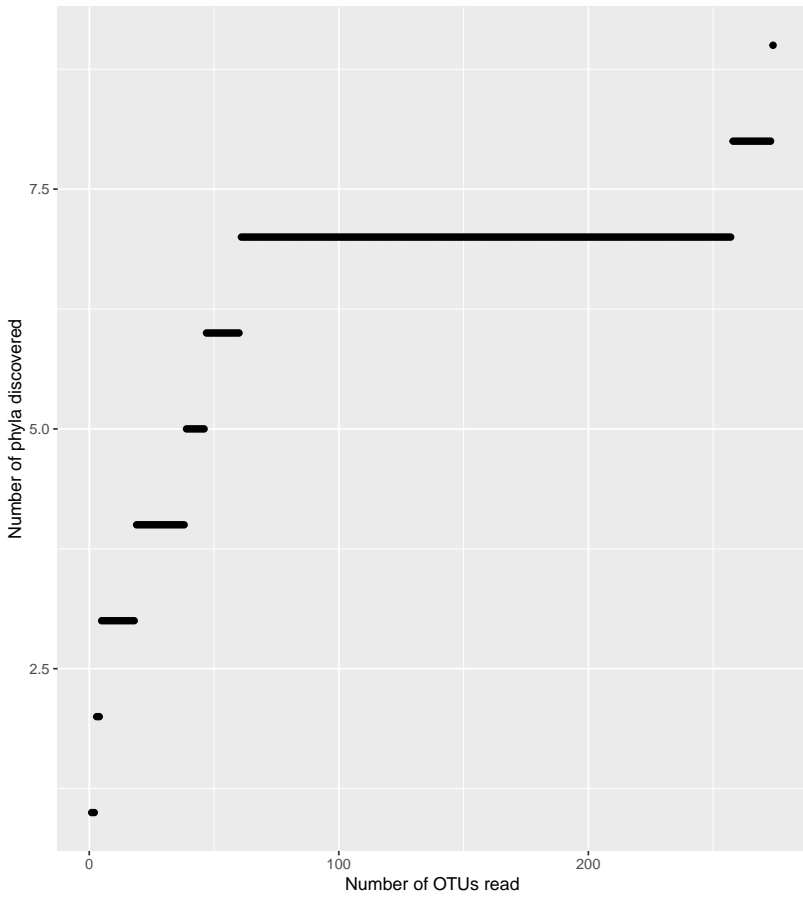
121. 18S.2.6.C.PCM.LT



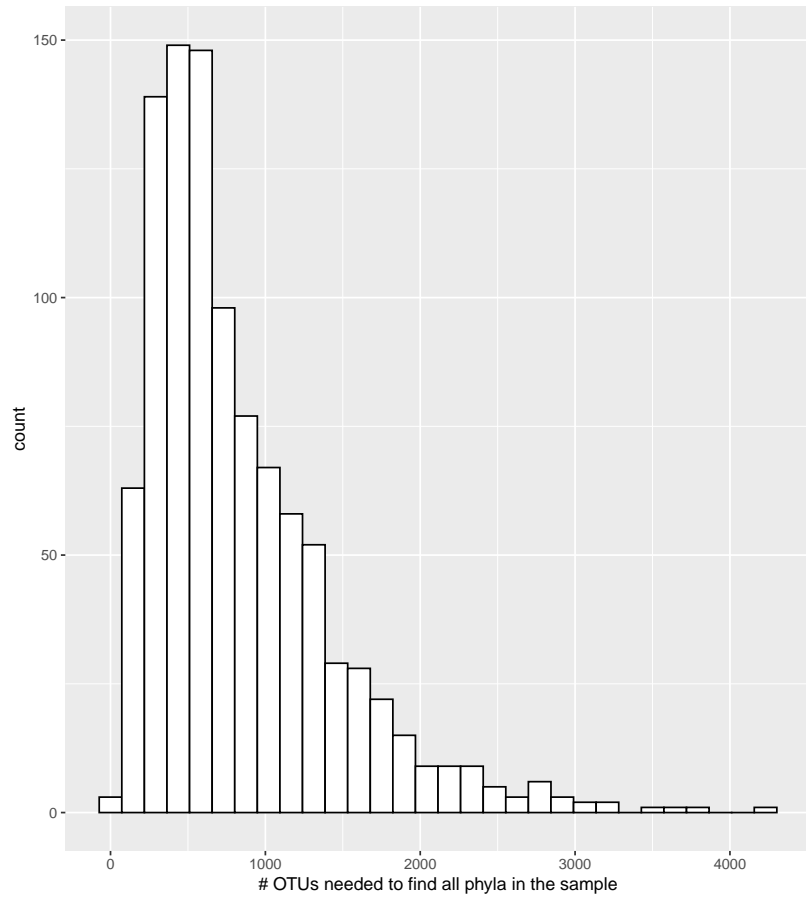
18S.2.6.C.PCM.LT



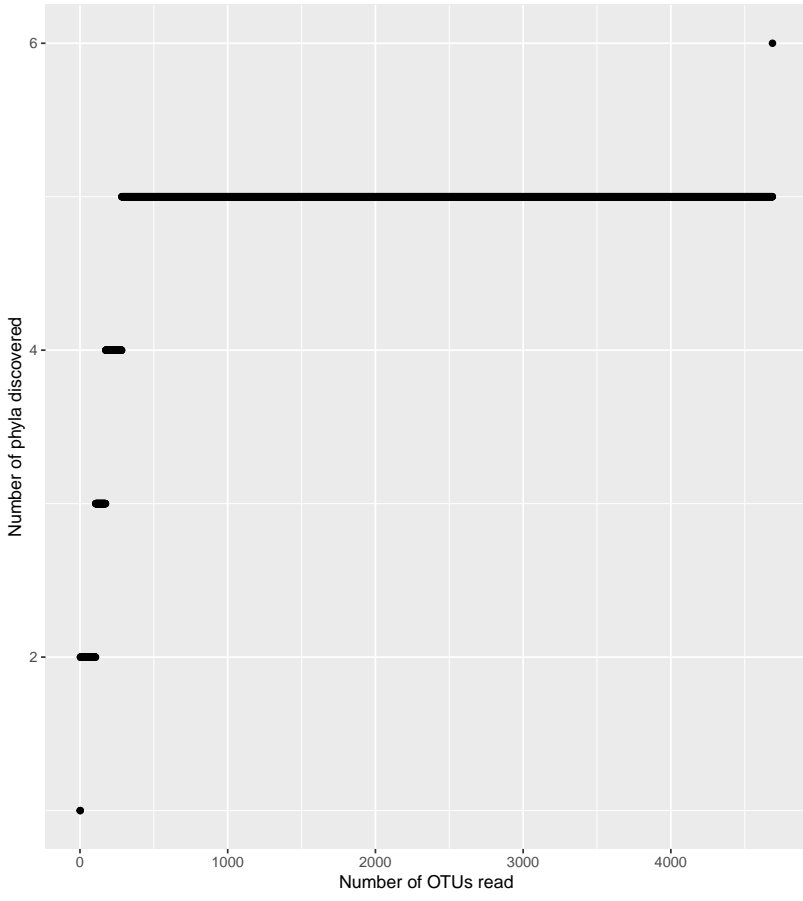
122. 18S.2.6.D.PCM.LT



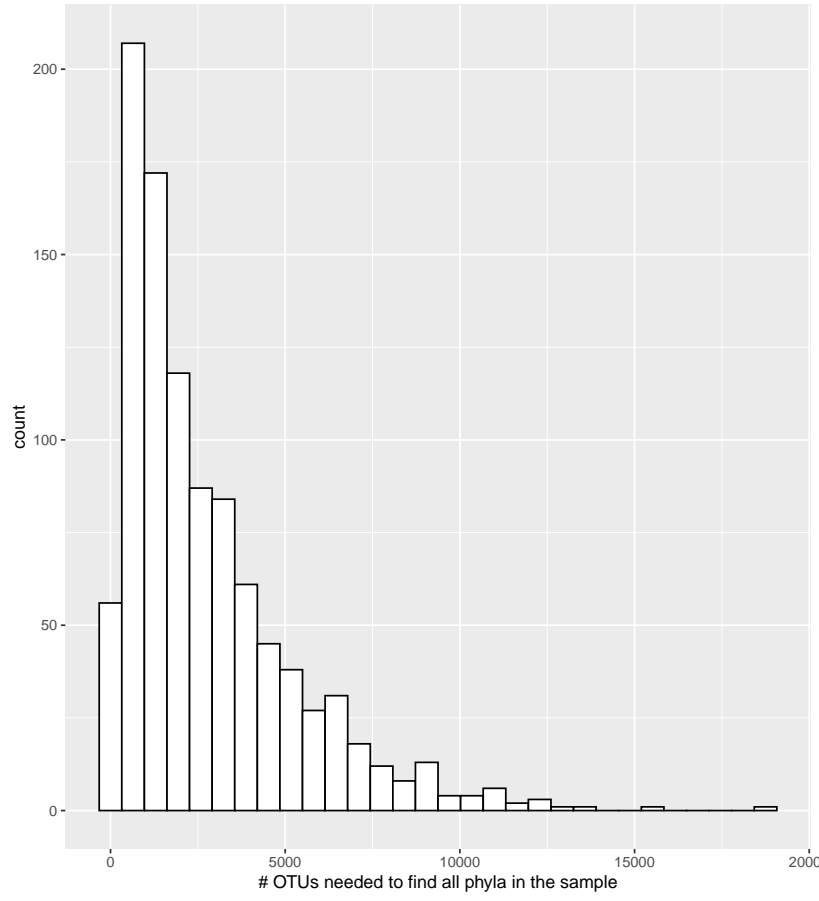
18S.2.6.D.PCM.LT



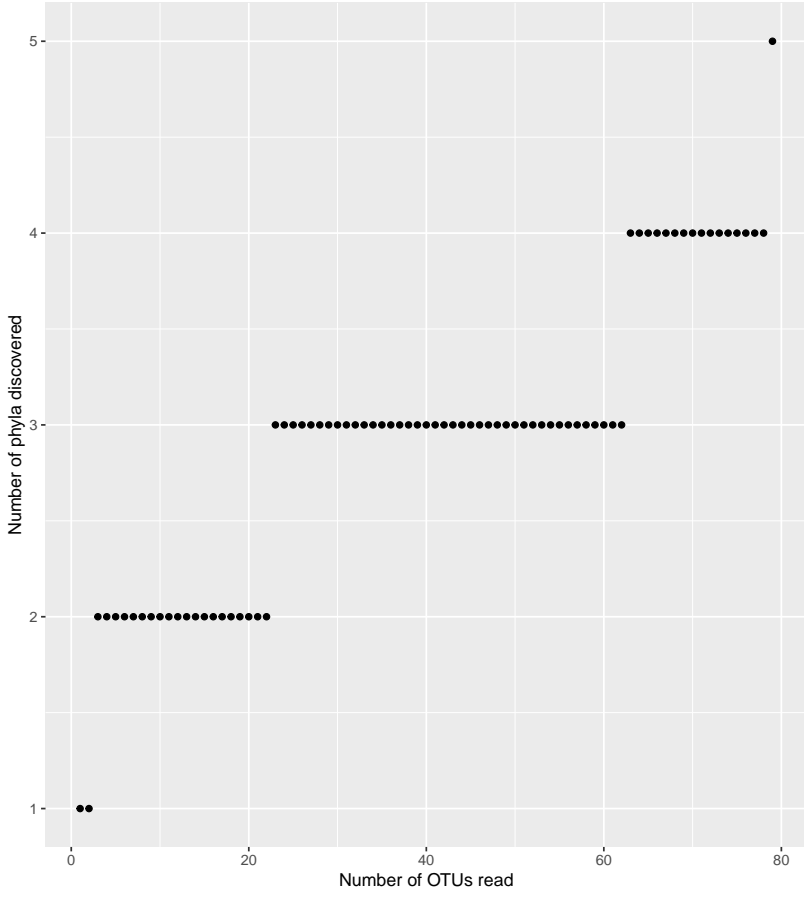
123. 18S.2.6.E.PCM.LT



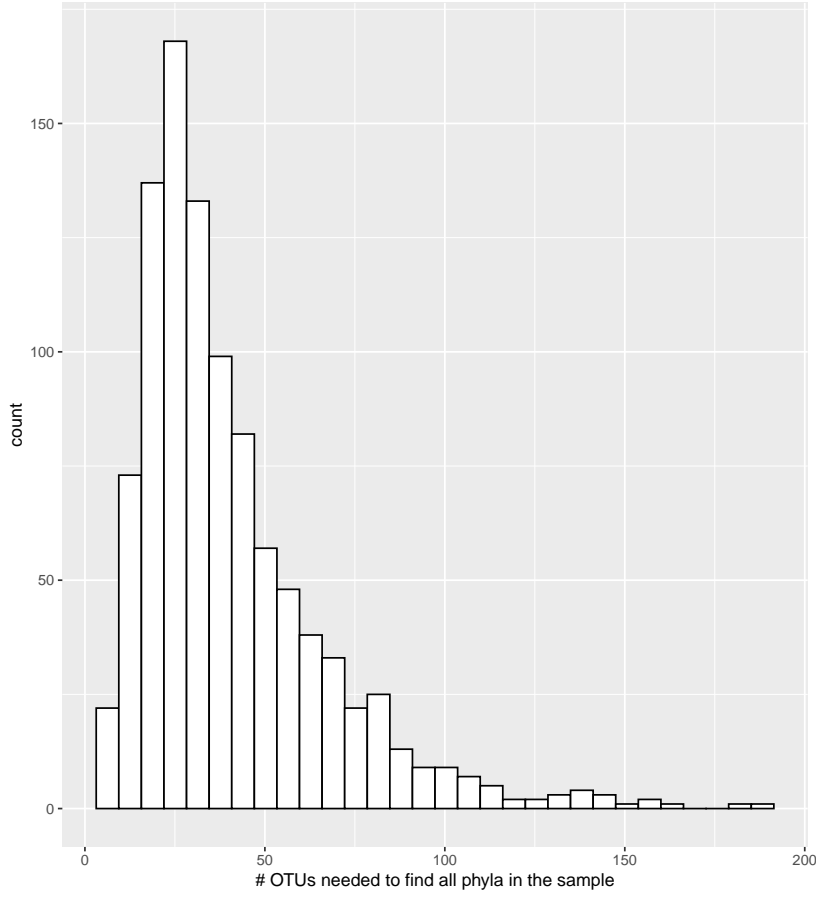
18S.2.6.E.PCM.LT



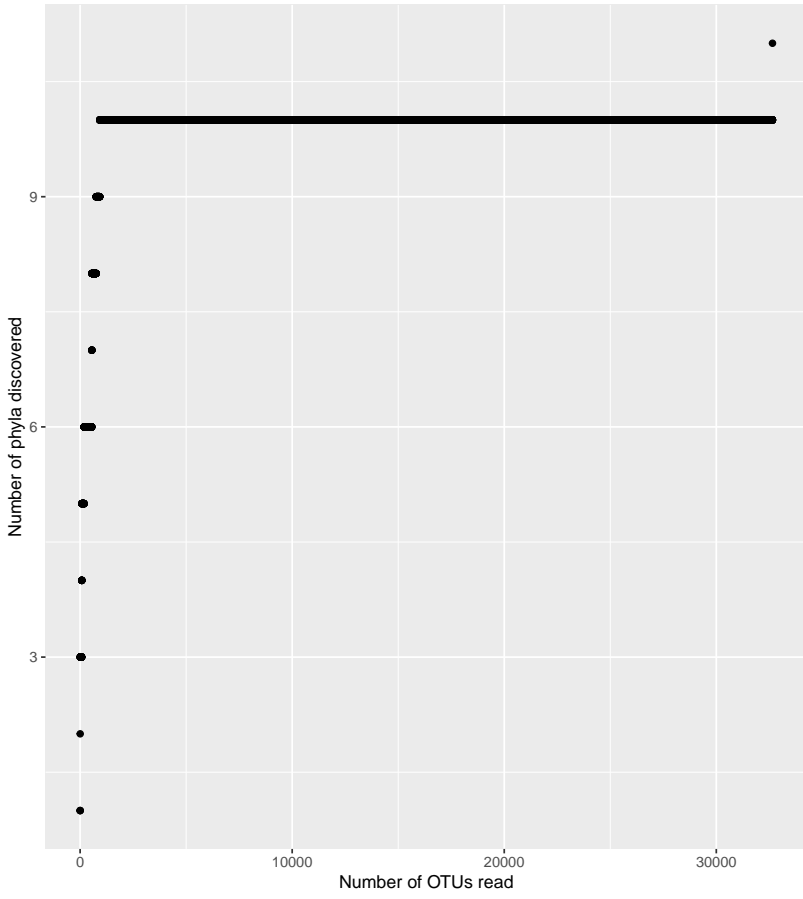
124. 18S.2.6.F.PCM.LT



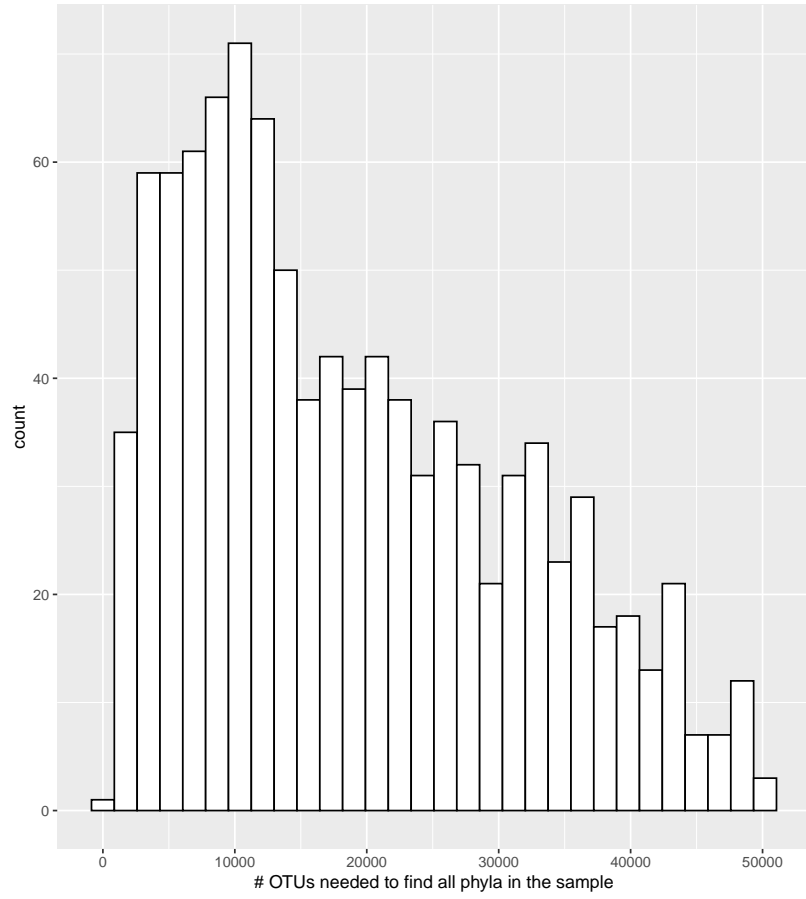
18S.2.6.F.PCM.LT



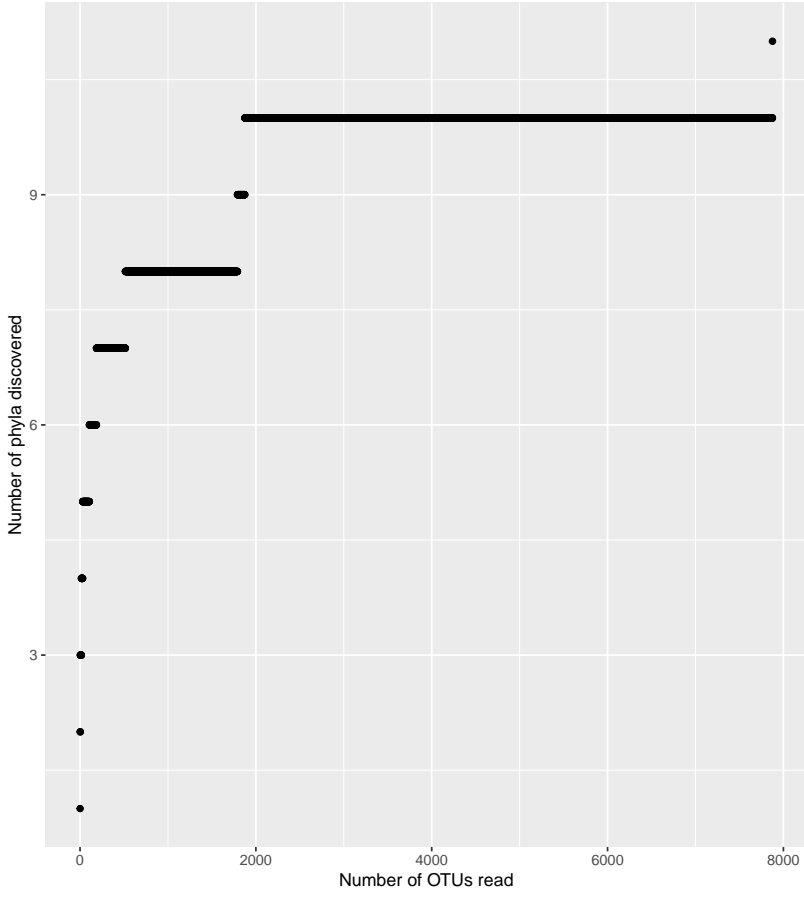
125. 18S.2.7.A.PCM.ME



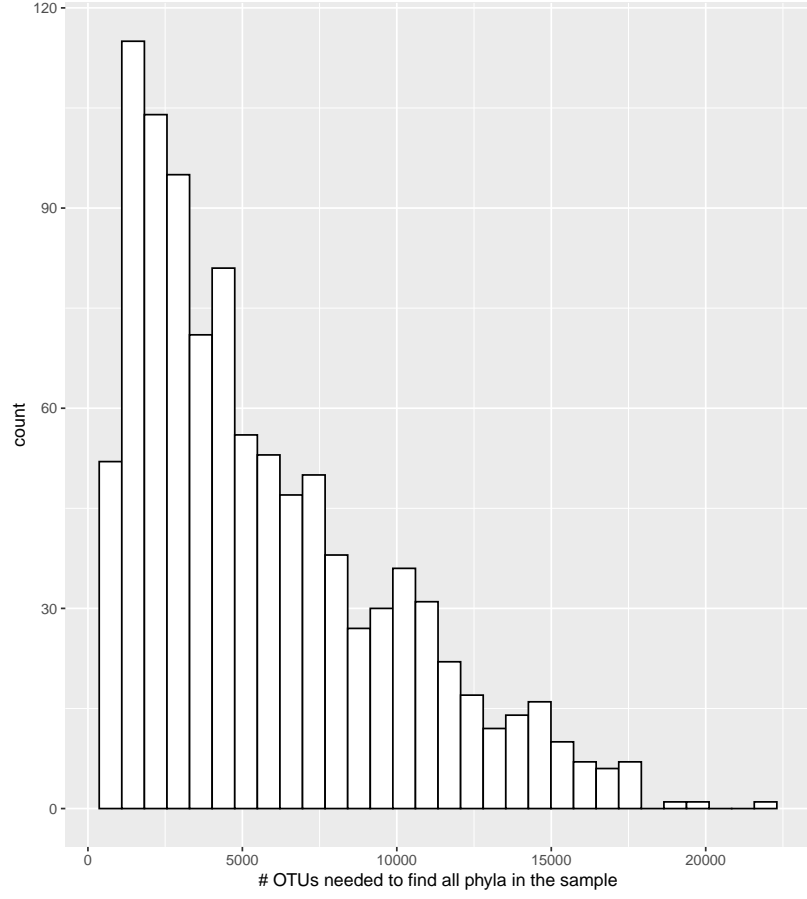
18S.2.7.A.PCM.ME



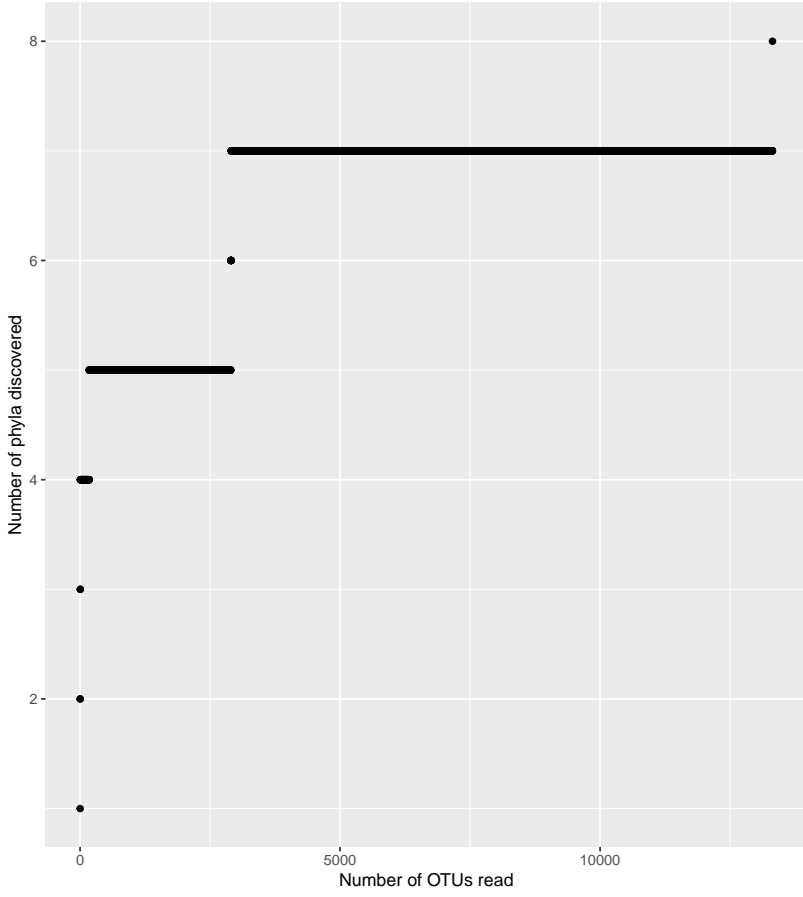
126. 18S.2.7.C.PCM.LT



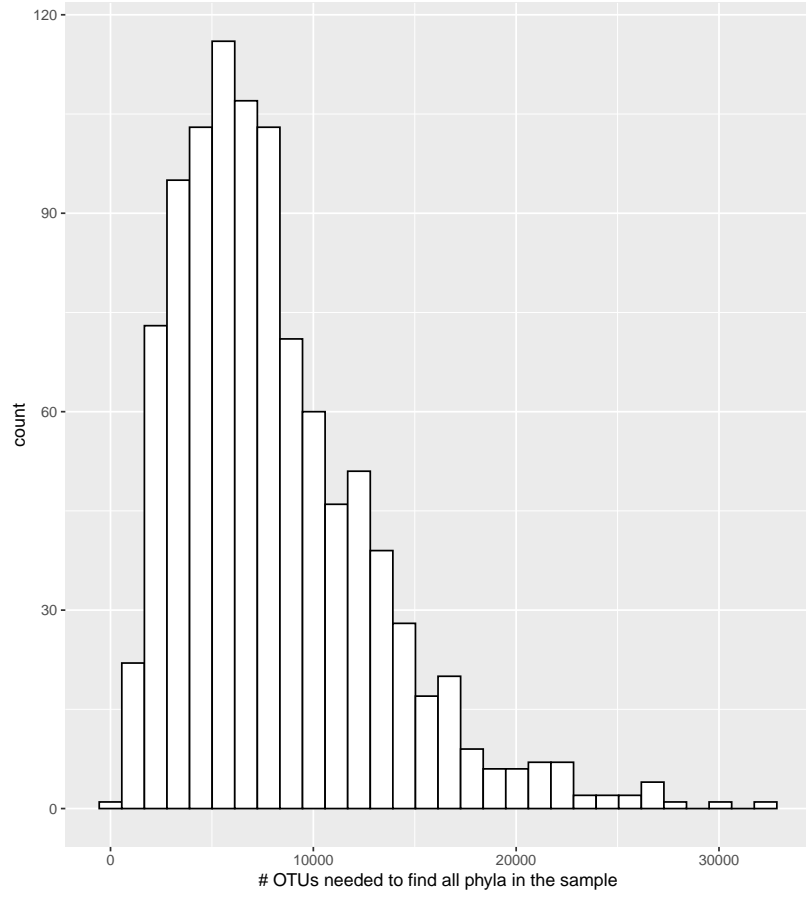
18S.2.7.C.PCM.LT



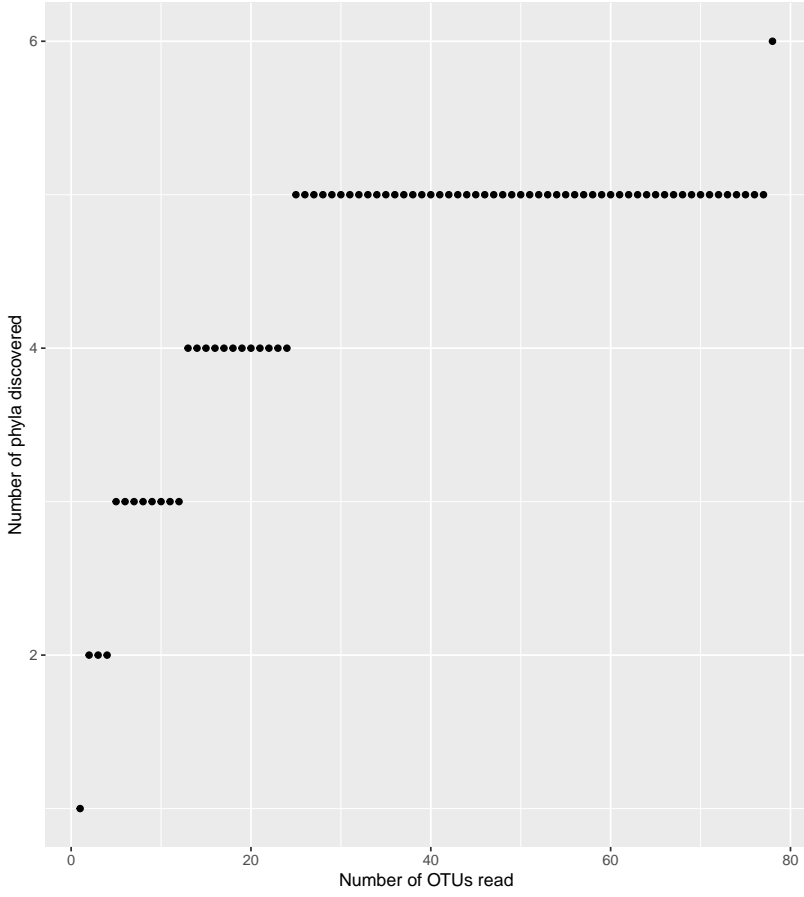
127. 18S.2.7.D.PCM.LT



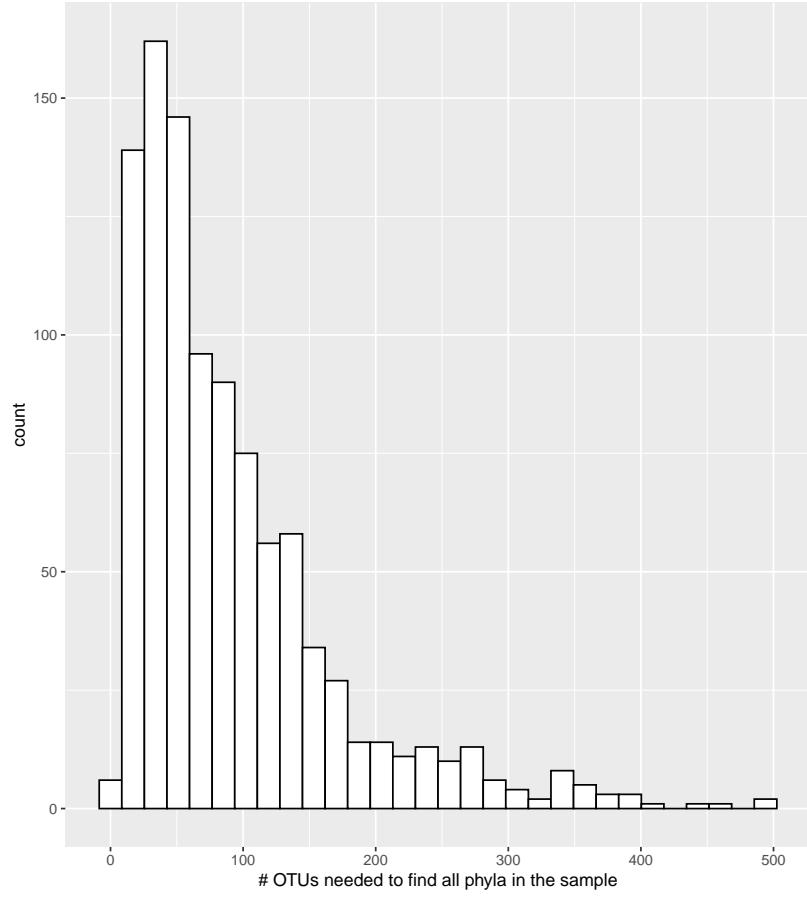
18S.2.7.D.PCM.LT



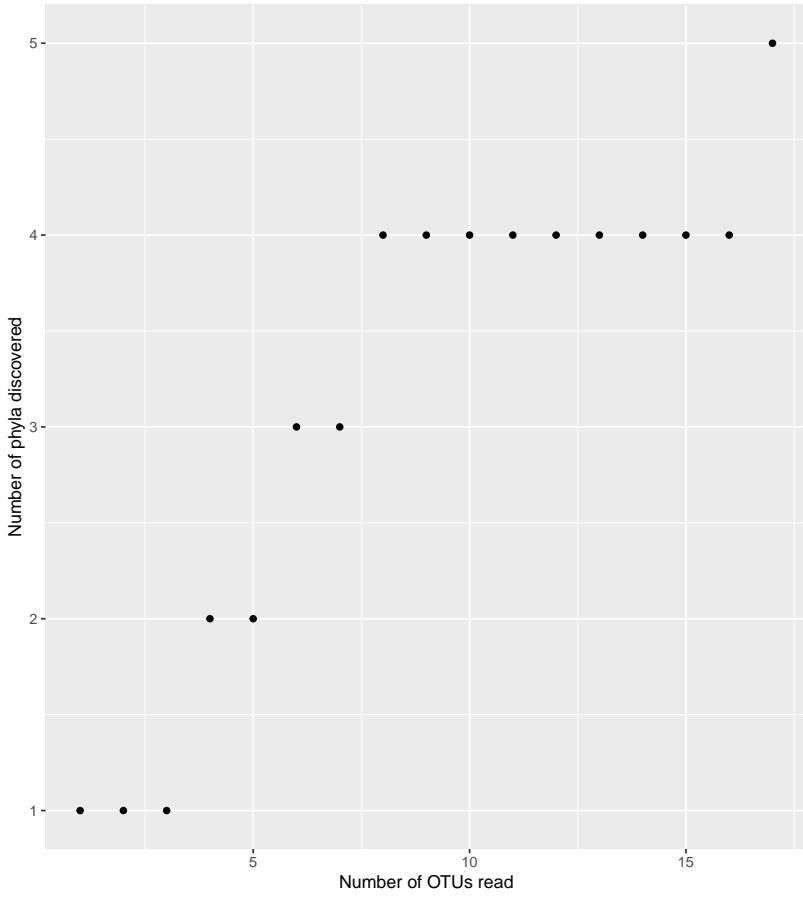
128. 18S.2.7.E.PCM.LT



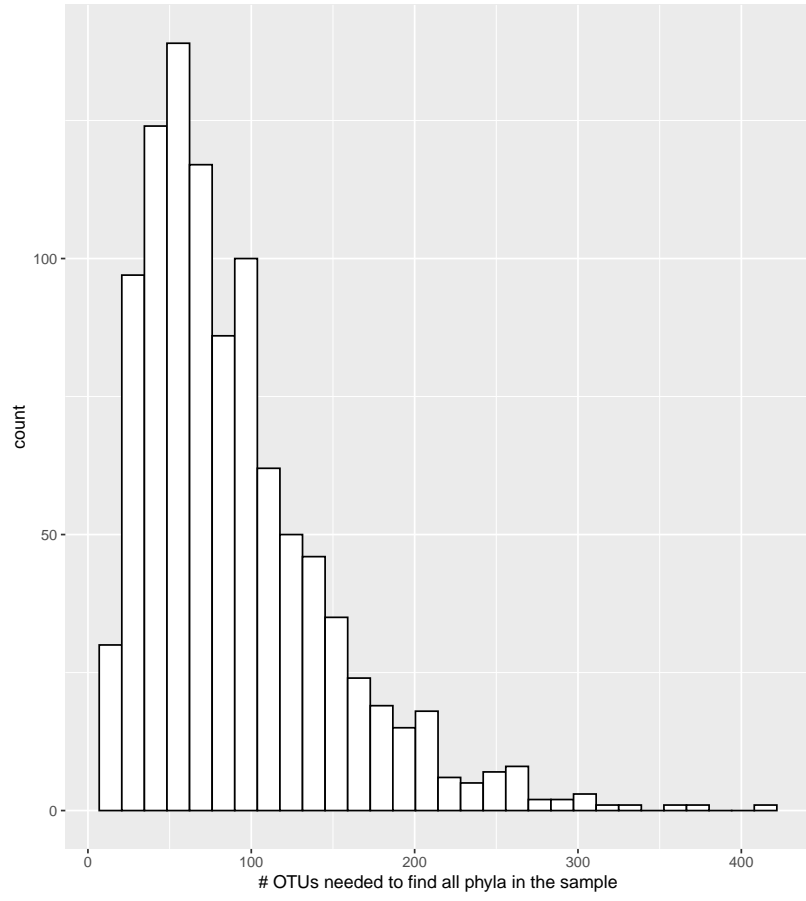
18S.2.7.E.PCM.LT



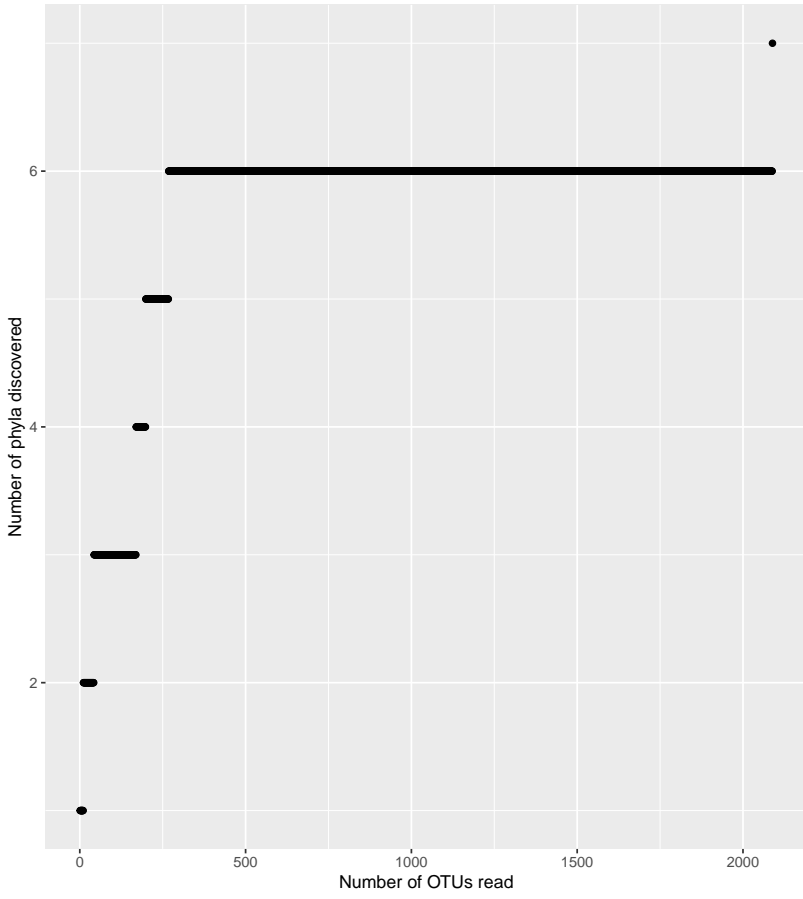
129. 18S.2.7.F.PCM.LT



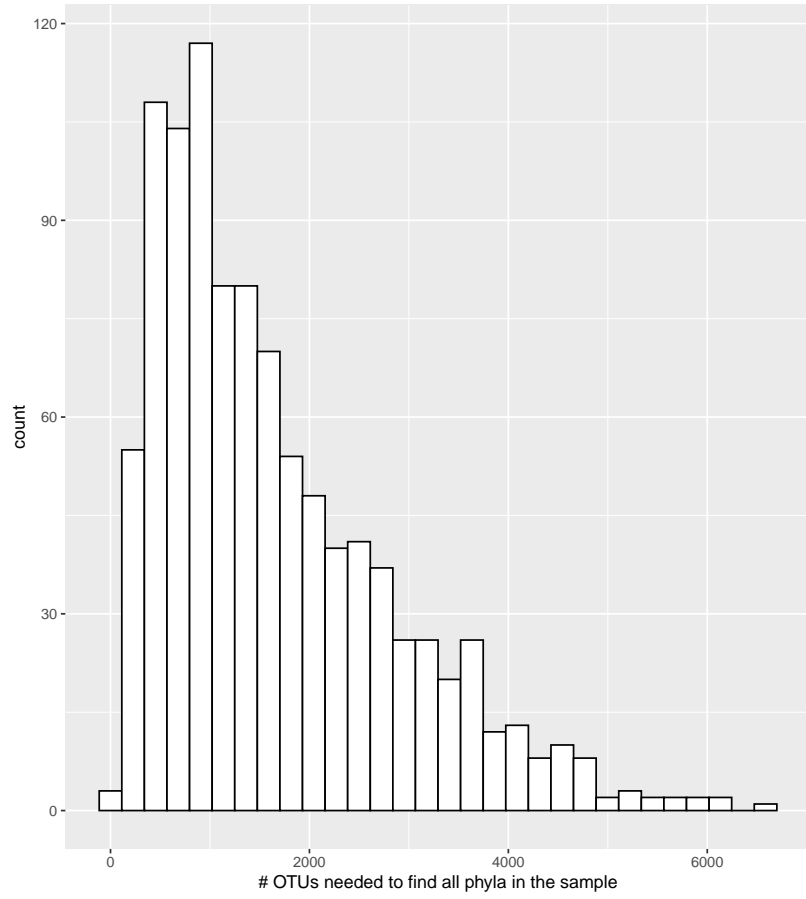
18S.2.7.F.PCM.LT



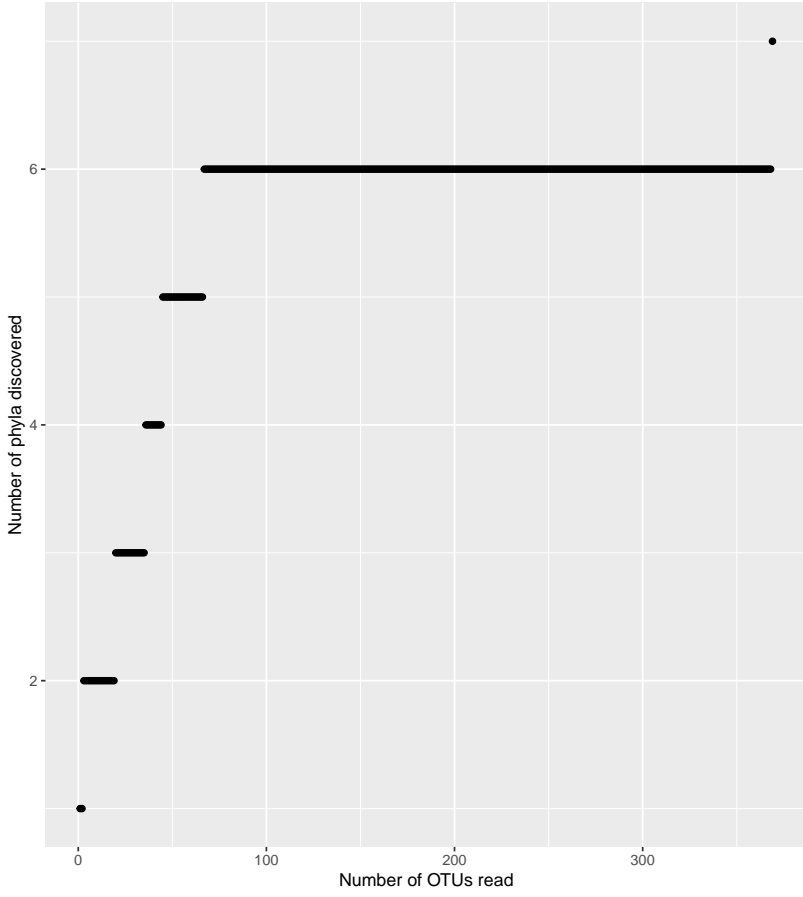
130. 18S.2.8.A.PCM.ME



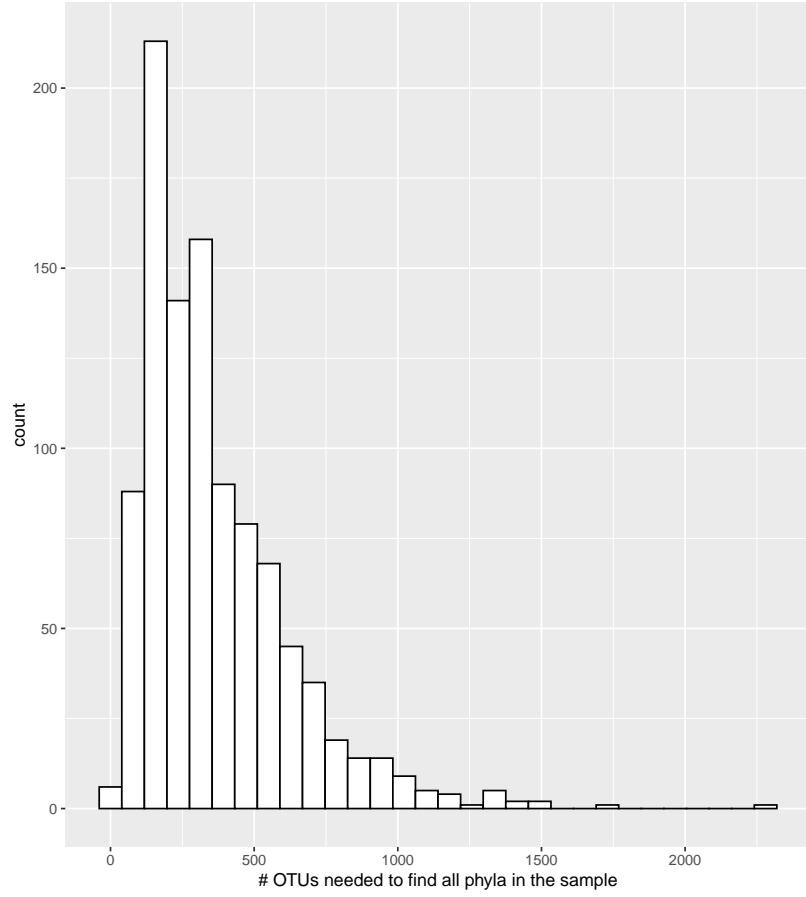
18S.2.8.A.PCM.ME



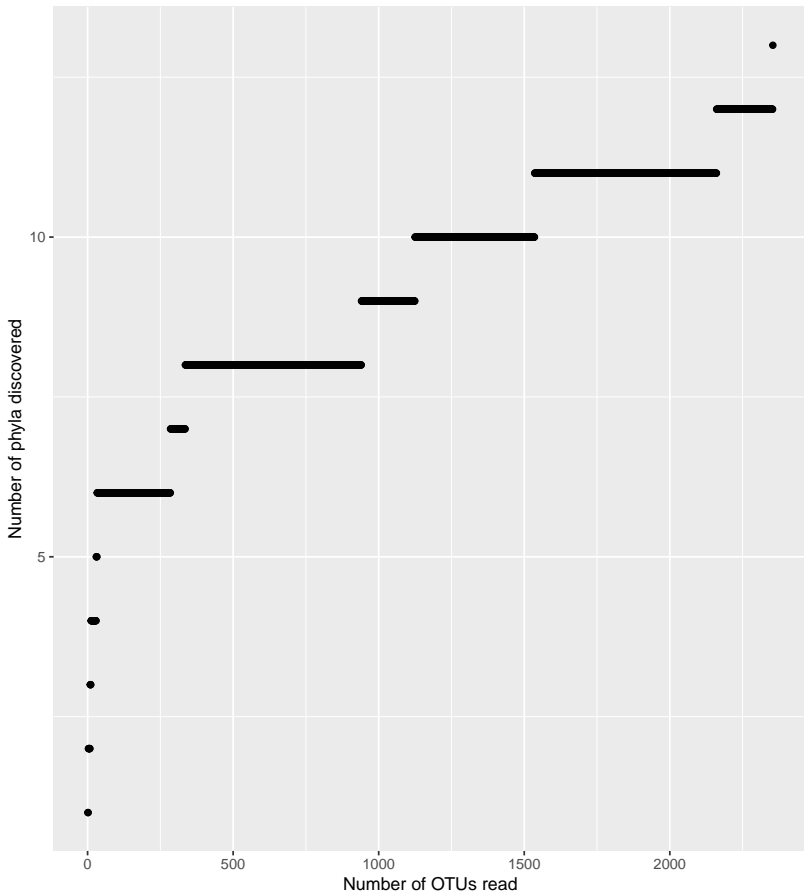
131. 18S.2.8.B.PCM.LT



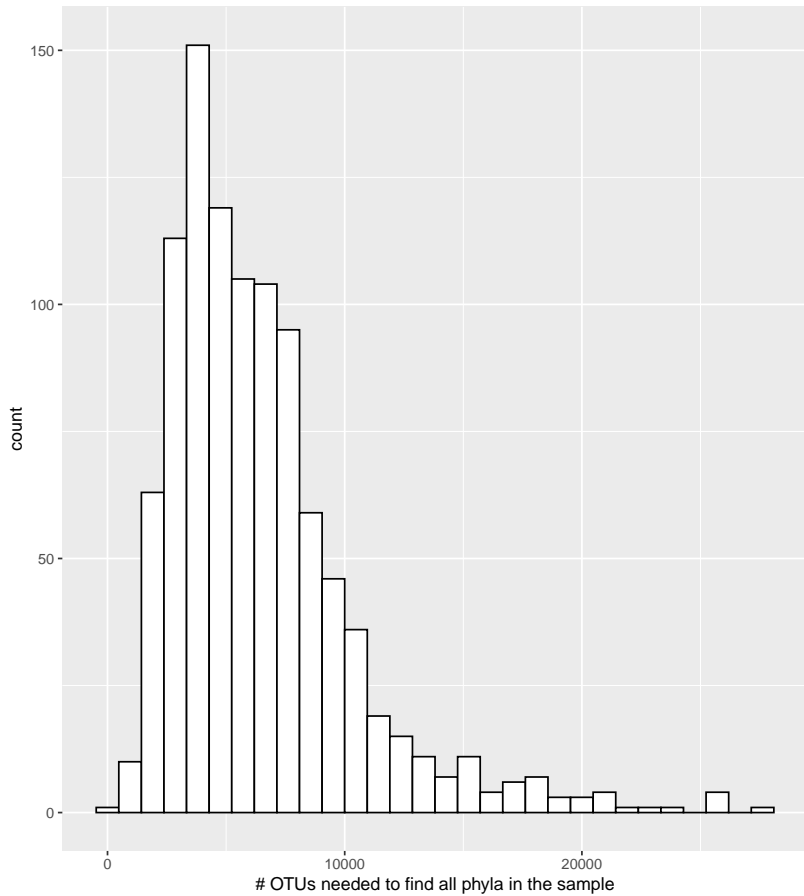
18S.2.8.B.PCM.LT



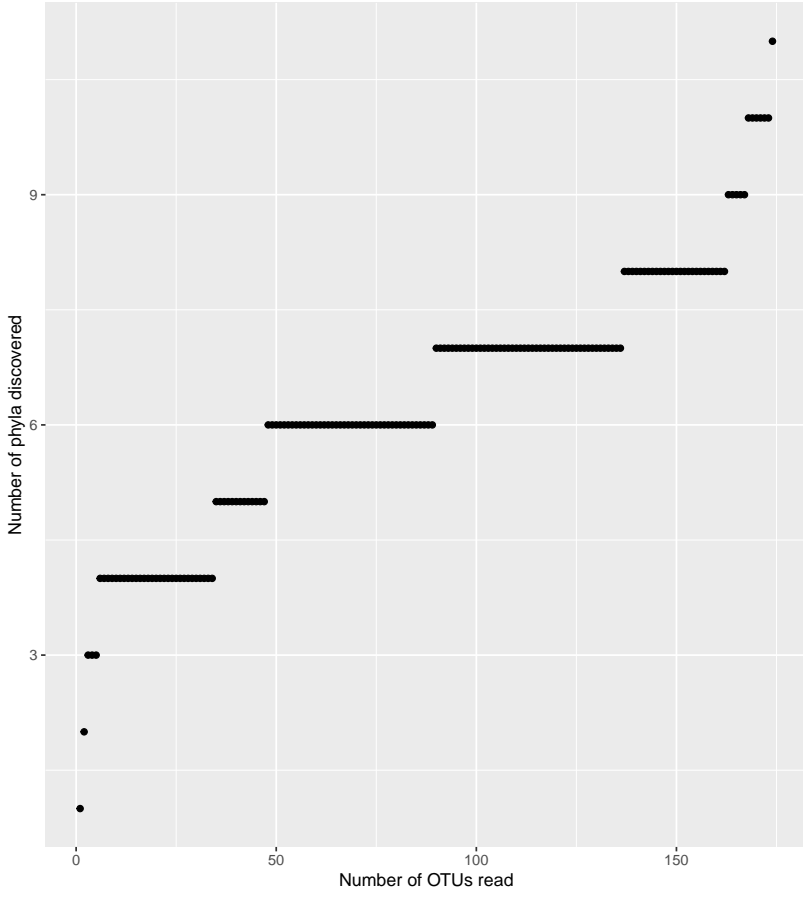
132. 18S.2.8.C.PCM.LT



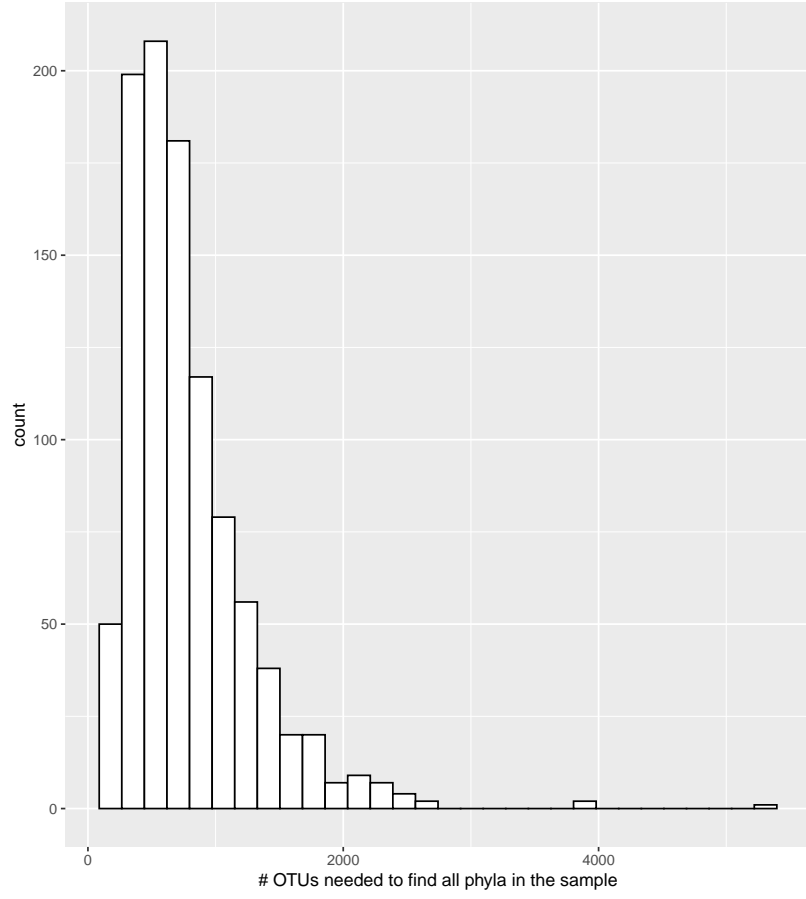
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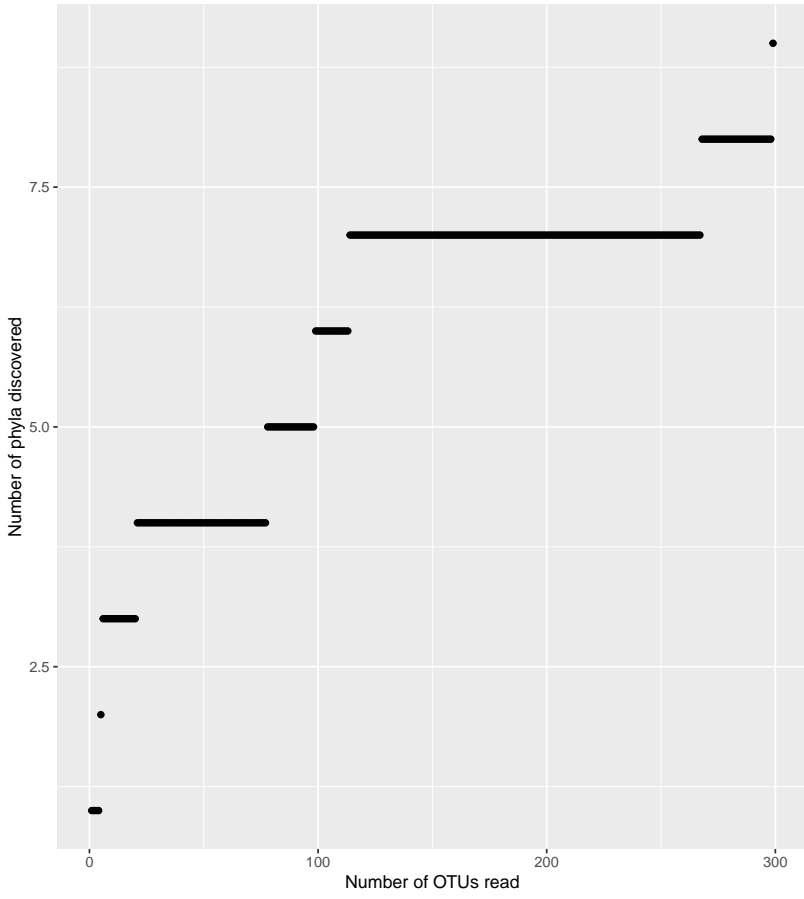
133. 18S.2.8.D.PCM.LT



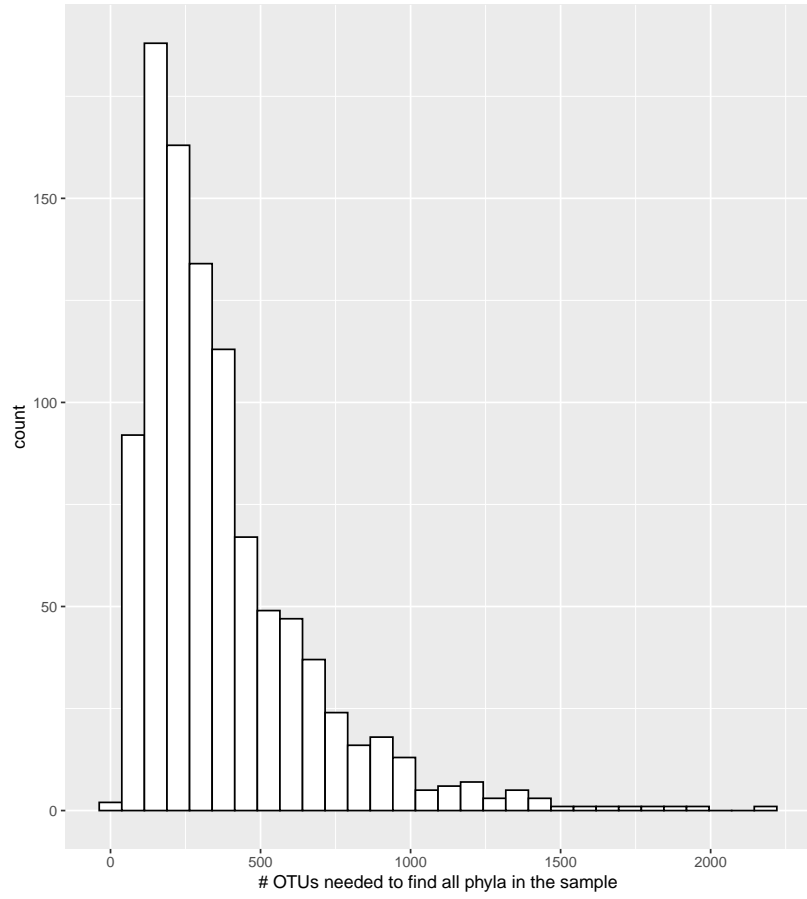
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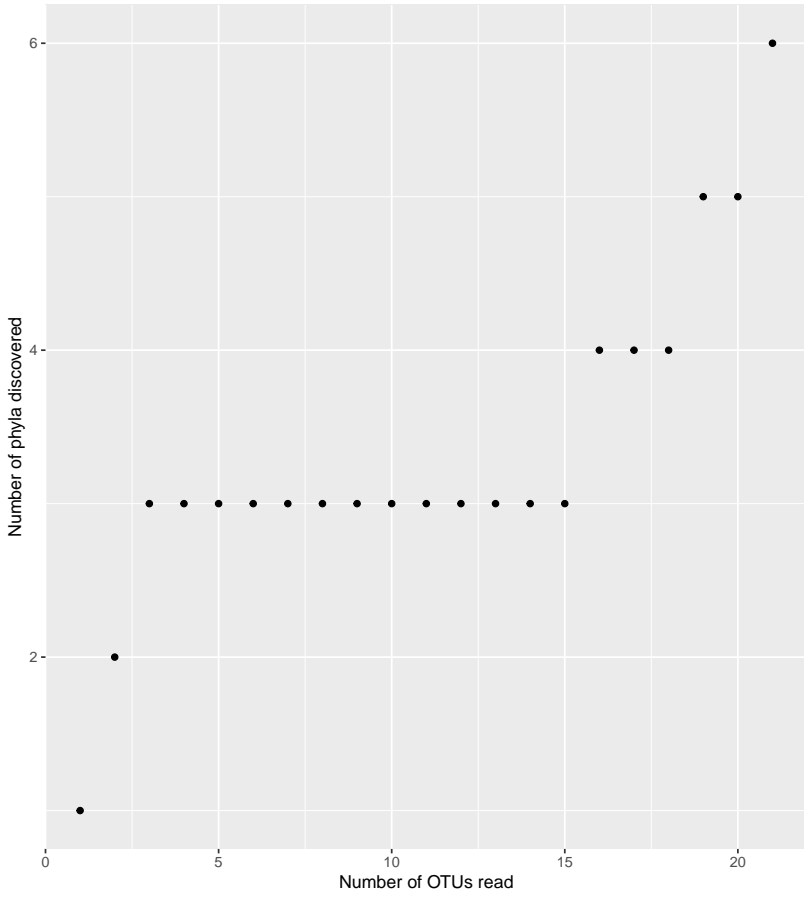
134. 18S.2.8.E.PCM.LT



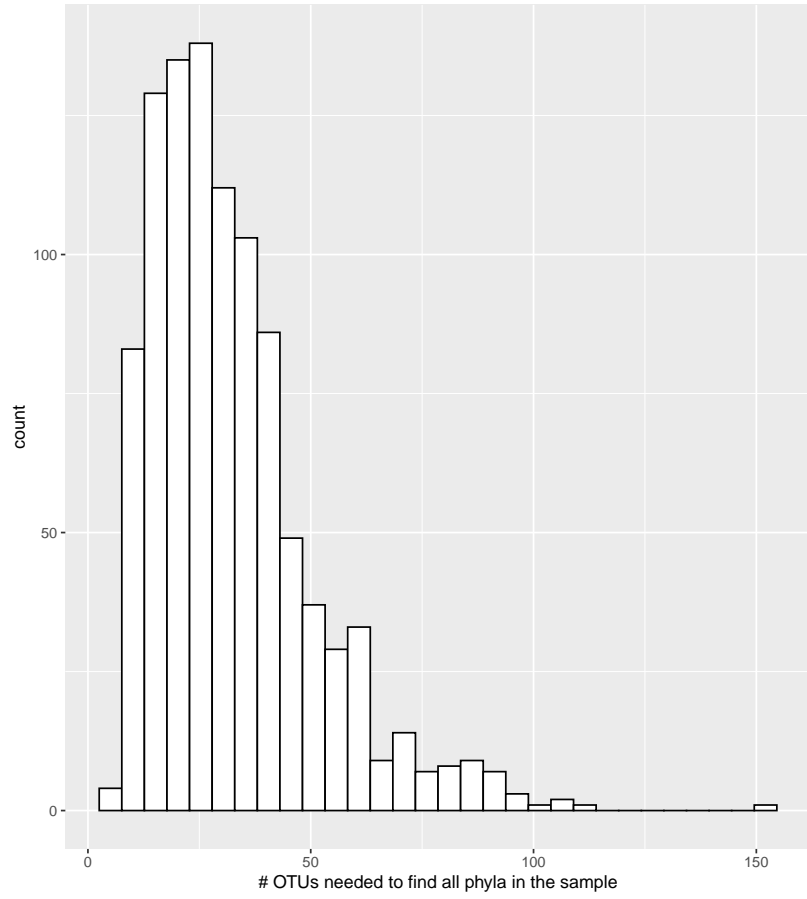
18S.2.8.E.PCM.LT



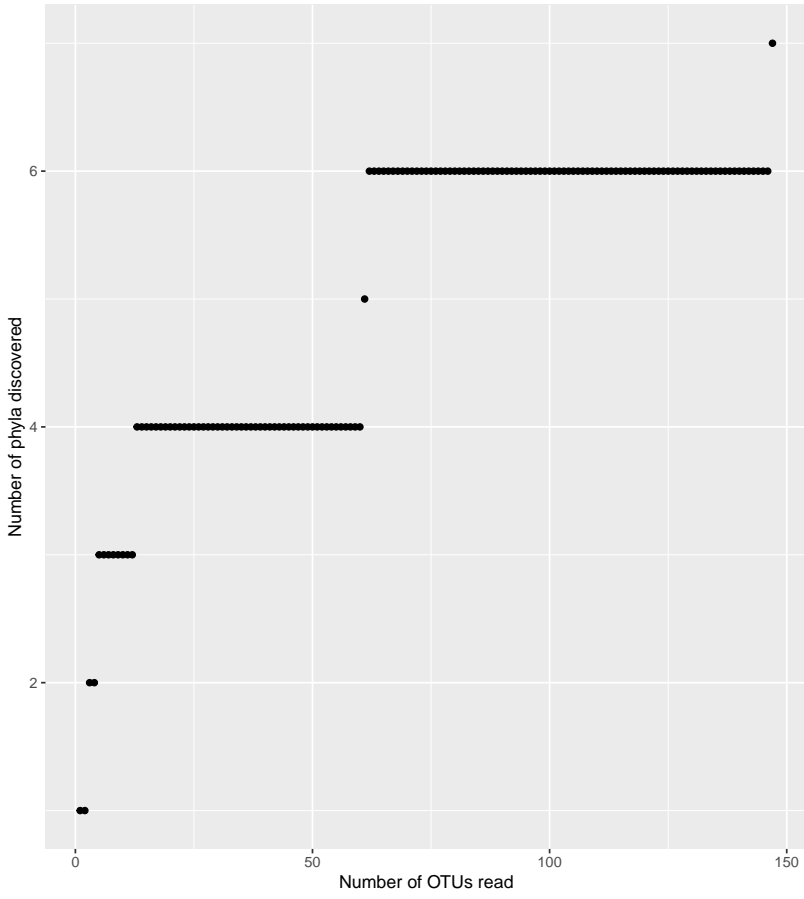
135. 18S.2.8.F.PCM.LT



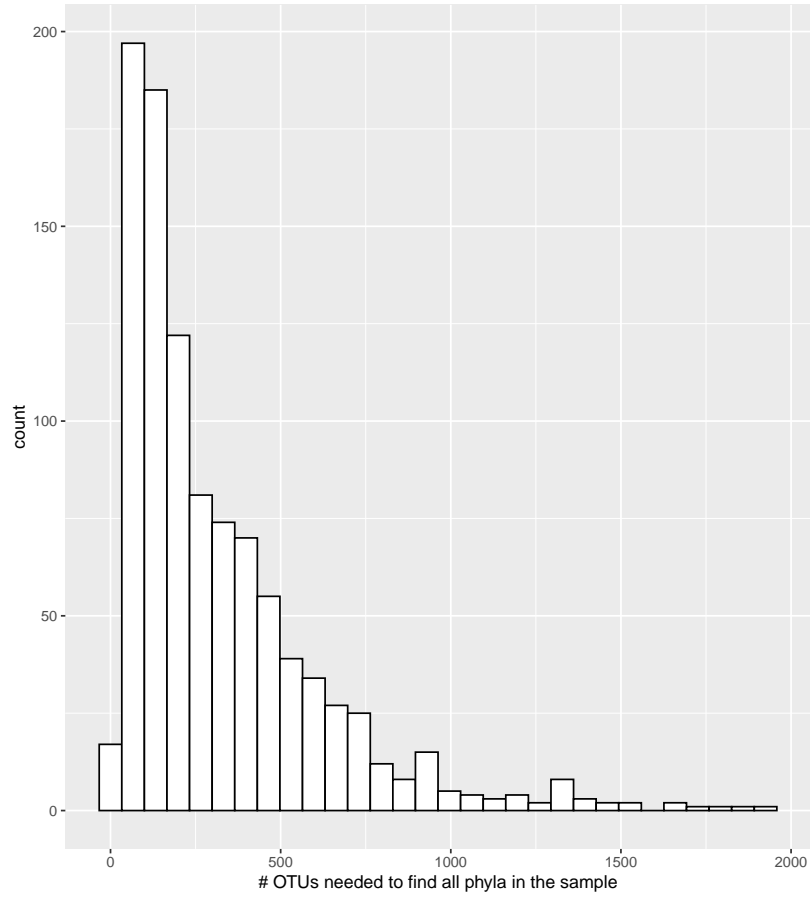
18S.2.8.F.PCM.LT



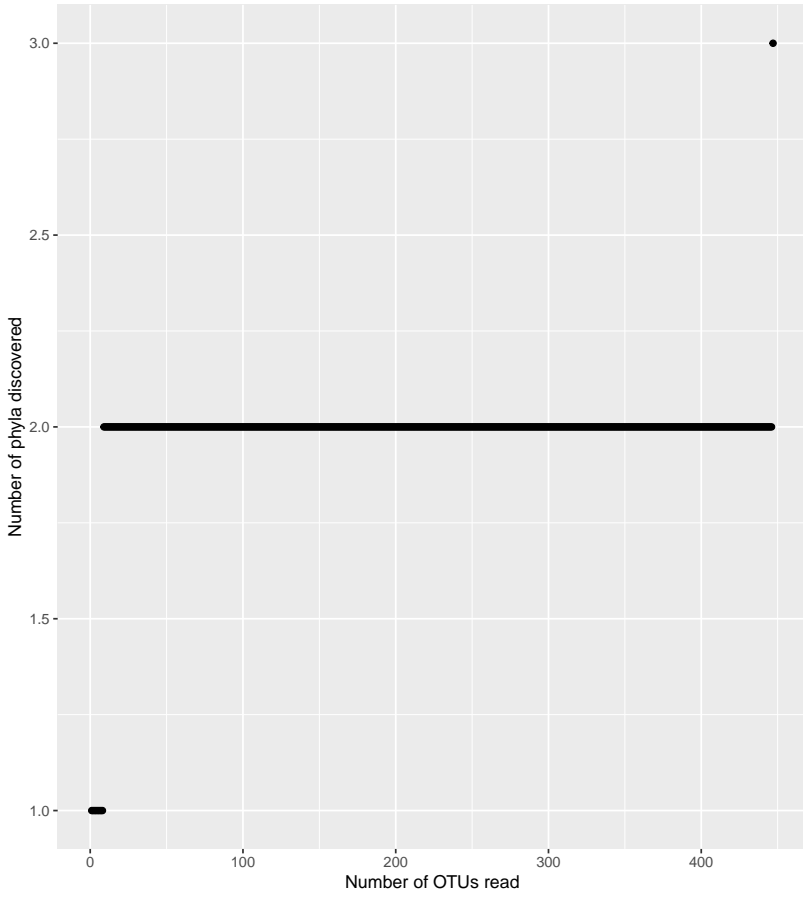
136. 18S.2.8.G.PCM.LT



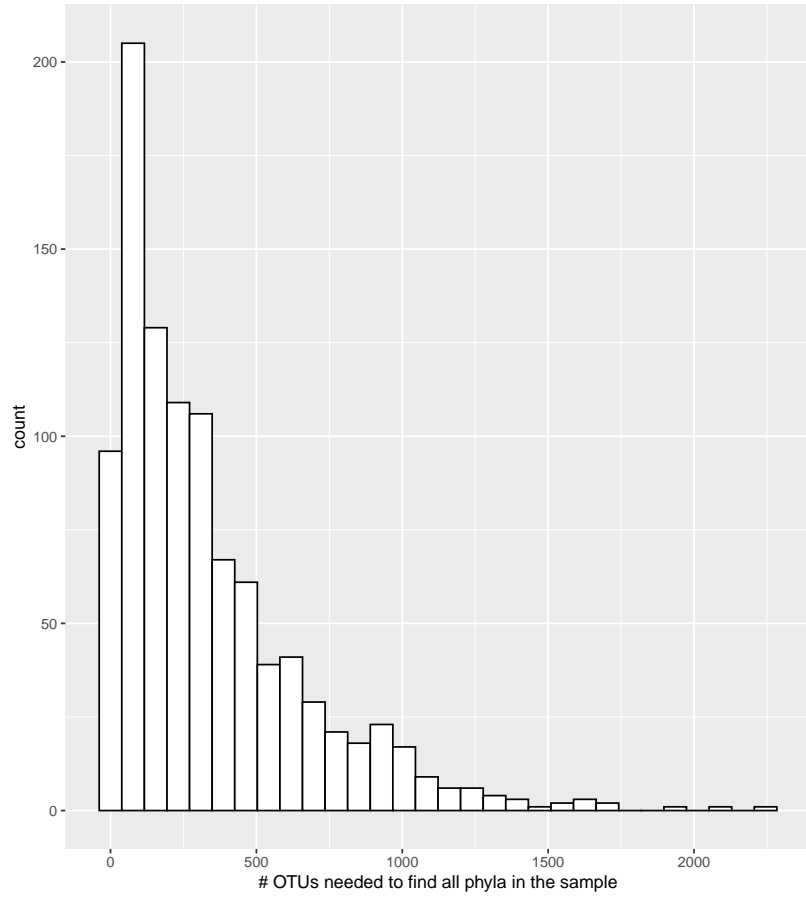
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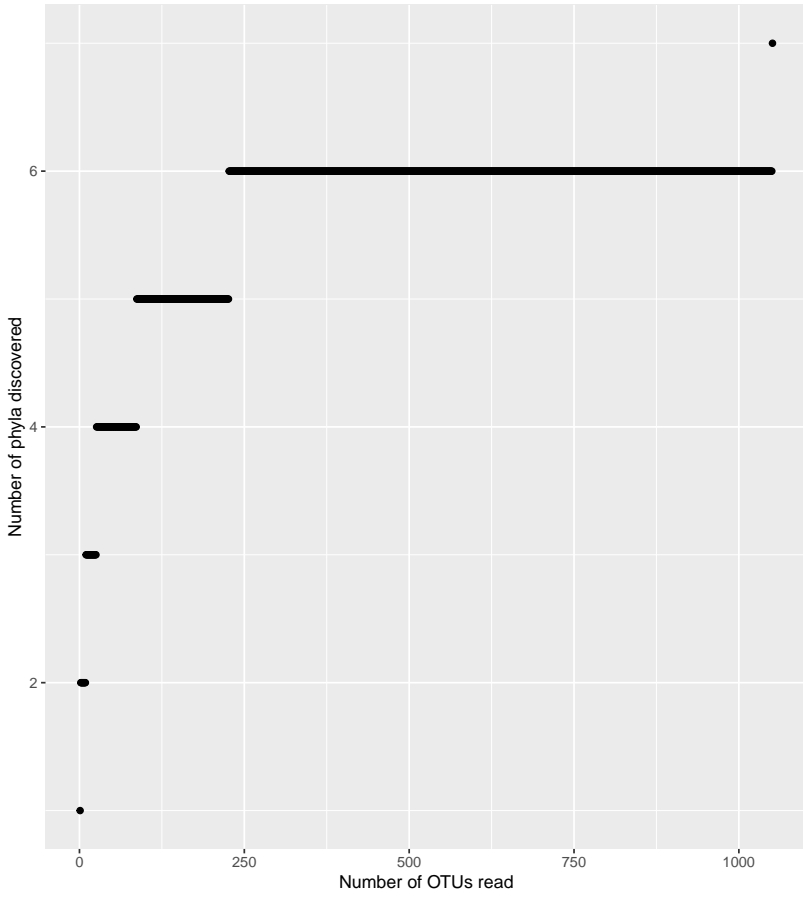
137. 18S.2.9.A.PCM.ME



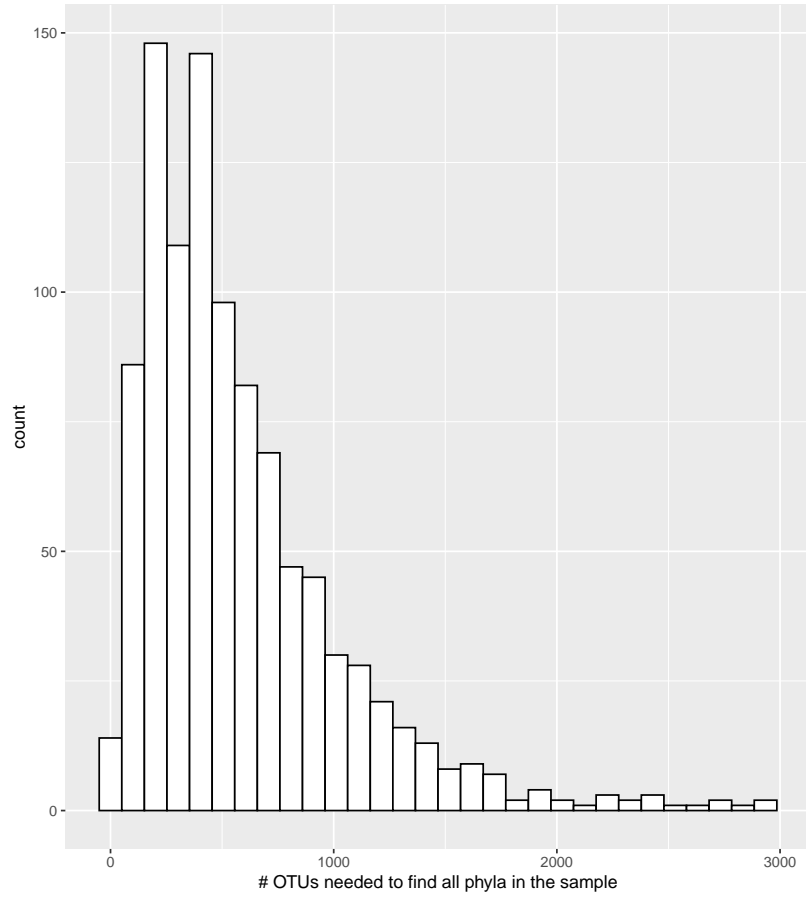
18S.2.9.A.PCM.ME



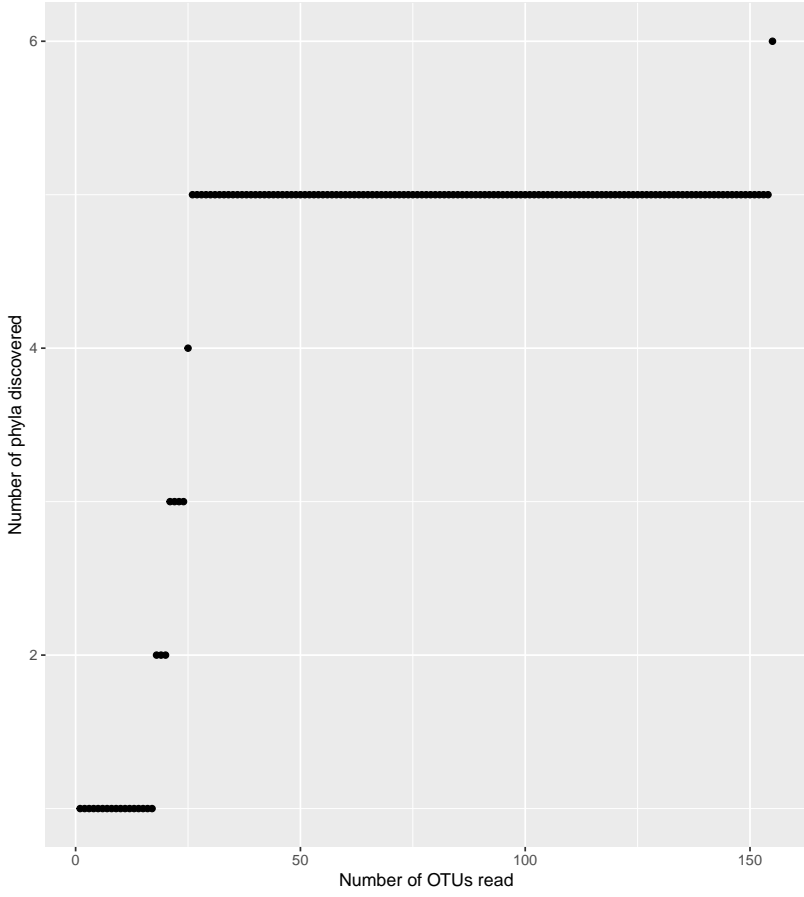
138. 18S.2.9.C.PCM.LT



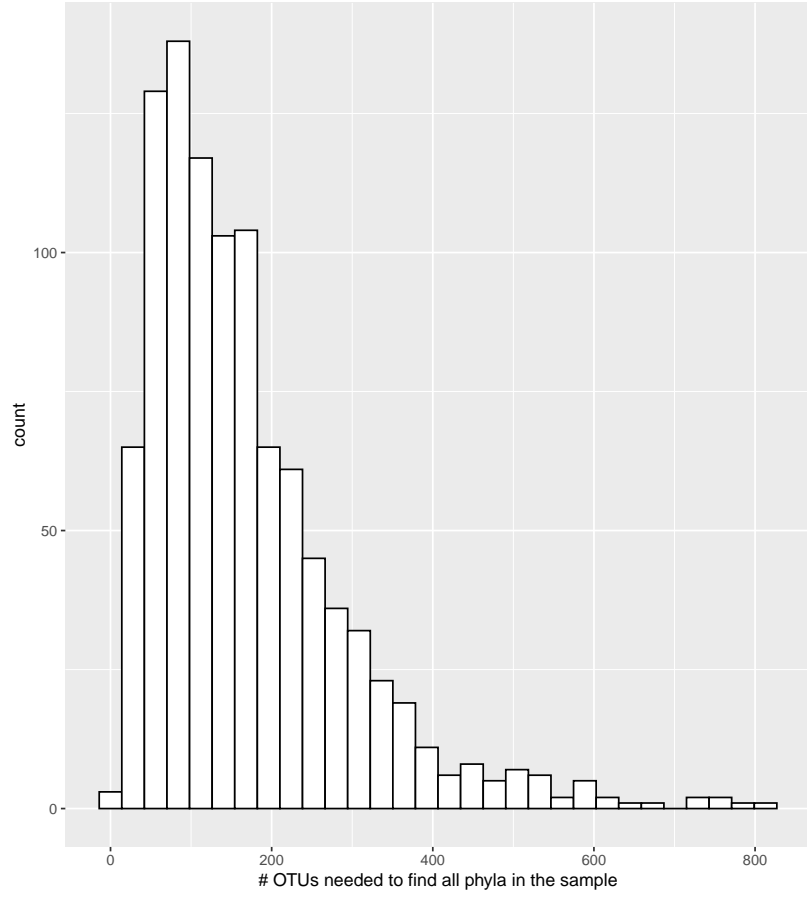
18S.2.9.C.PCM.LT



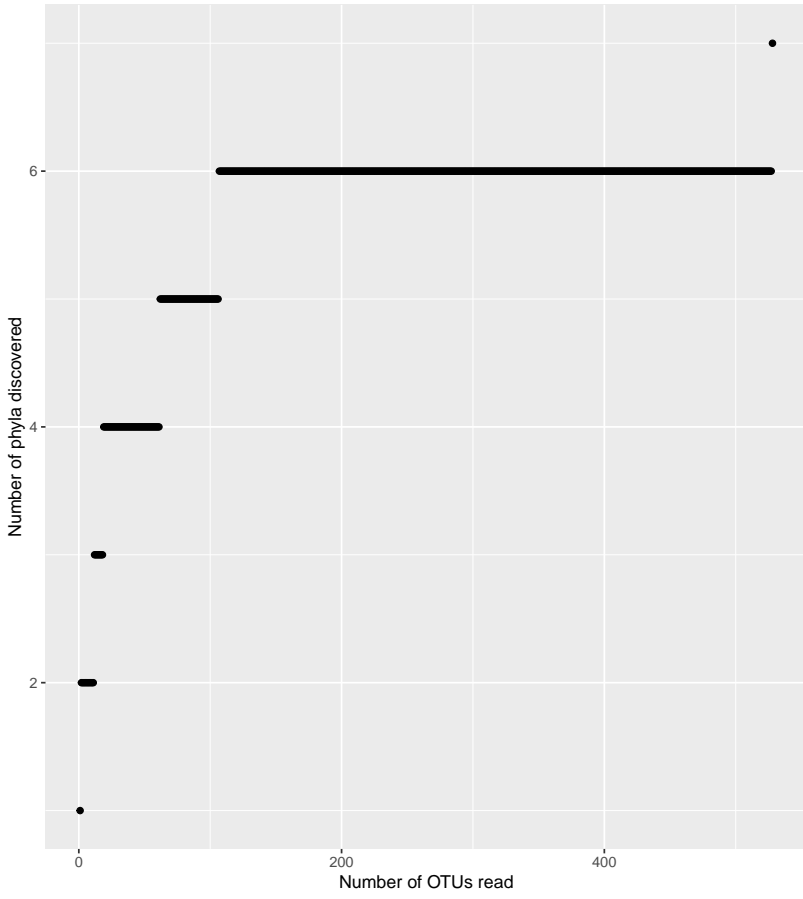
139. 18S.2.9.D.PCM.LT



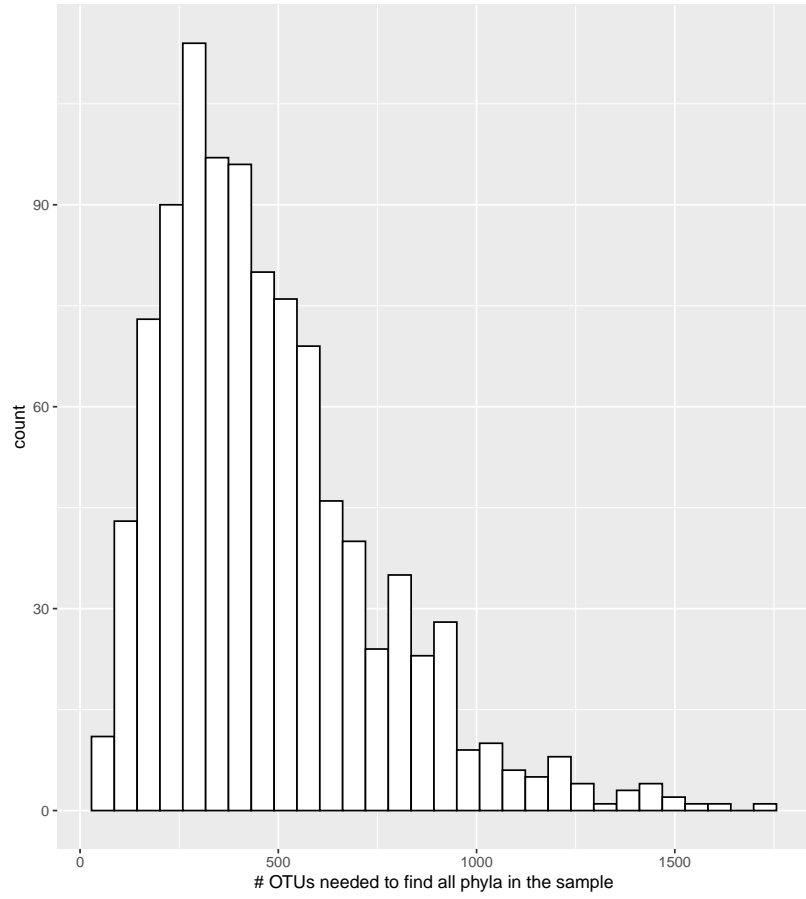
18S.2.9.D.PCM.LT



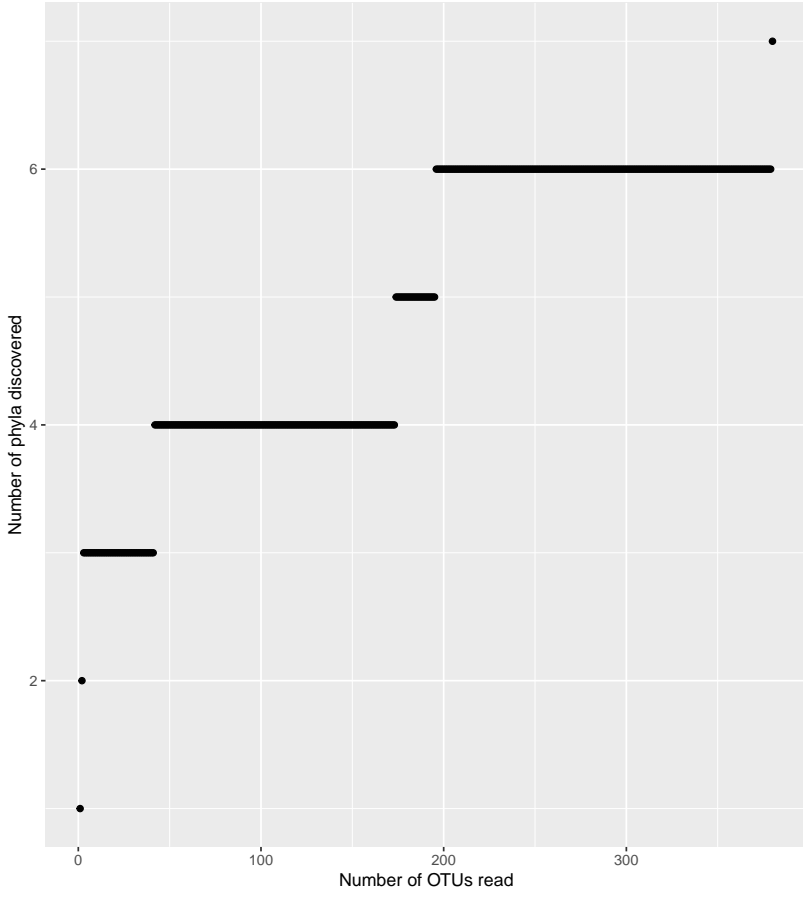
140. 18S.2.9.E.PCM.LT



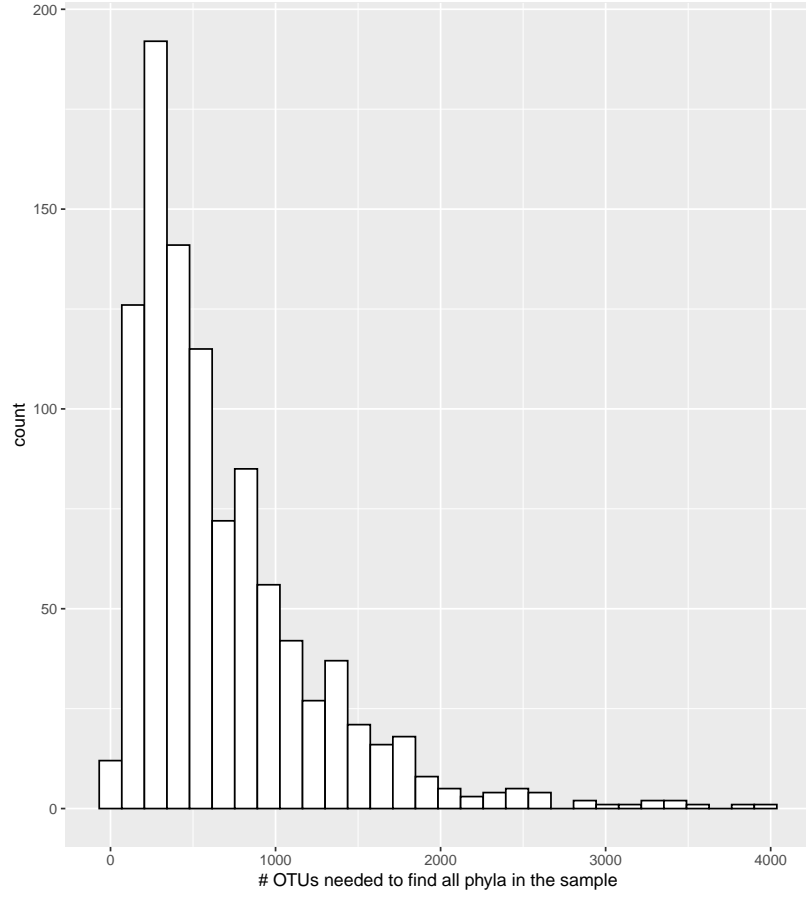
18S.2.9.E.PCM.LT



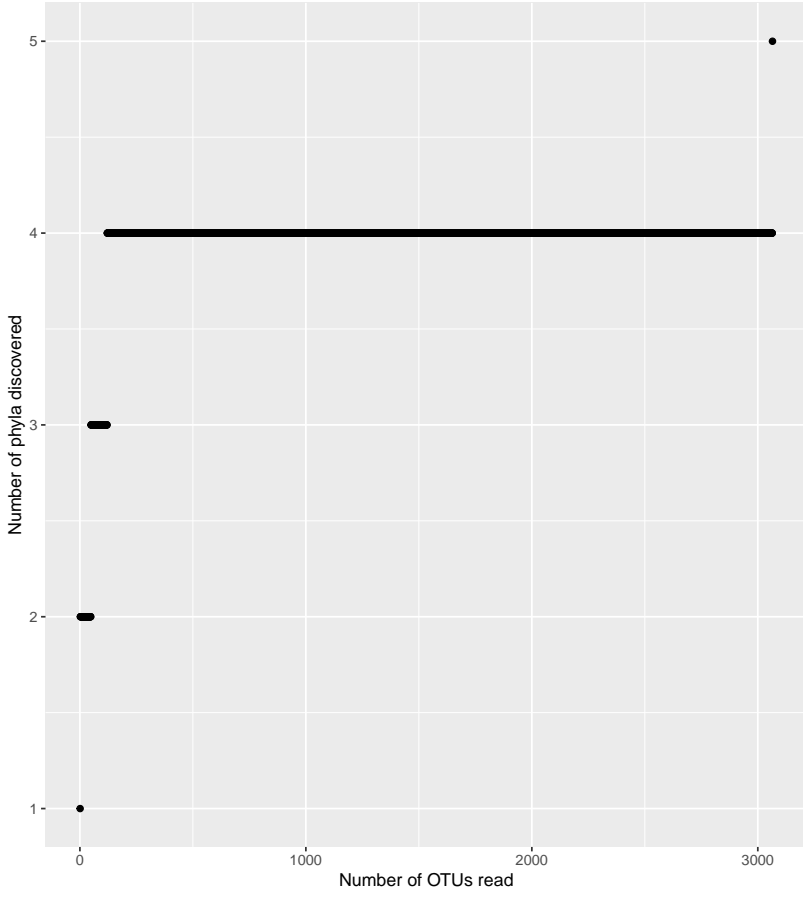
141. 18S.2.9.F.PCM.LT



18S.2.9.F.PCM.LT



142. 18S.2.9.G.PCM.LT



18S.2.9.G.PCM.LT

