

Supplementary File 1:

Full Details, Sequencing and Read Cleaning:

Our transcriptomic sequencing was of reasonably good quality, with *B. bacillifera* (A2) and *L. abietina* (A10) of good average quality (Phred median >30) through to the final base in both read directions. The *L. baikalensis* (A8) sample was of less robust quality, possessing reads which declined in base call confidence score towards the end of the read. The use of Trimmomatic fixed these issues, with few reads removed from samples A2 and A10, while sample A8 had 40.84% of read pairs removed, either due to poor quality or short read length after truncation. Where possible, single unpaired reads were retained from these filtered read pairs. Unpaired reads were used in assembly, but not in read mapping.

Our genomic sequencing required attention before use in genome assembly, due to a large number of apparent singleton *k*-mers (Supplementary File 2), which could be indicative of read errors. Trimmomatic was applied to these reads first, removing and truncating large quantities of reads, leaving just over half of the original read pairs and bases. The 99,040,746 (53.68%) of pairs remaining after Trimmomatic were fed into rCorrector, which corrected 67,842,587 bases in 34,392,948 read pairs. rCorrector also discarded 17,998,543 pairs of reads, which were assessed as being unfixable. This left a total of 81,042,203 read pairs for further analyses. This process is conservative and highly stringent. In a more complete genome assembly, low complexity sequences could be spanned using long reads and a variety of library sizes, but for our purposes, this ensured the quality of sequence data used in assembly, and thus the reliability of assembled data.

Additional Details, Assembly:

SPAdes was chosen as the best-performing assembler, as noted in the main text. For our purposes, the higher median contig size, larger number of long (>1kb) contigs and higher number of bases in these 1kb+ contigs (155,282,664 bp) meant that an increased number of long contigs was available for our work. That SPAdes outperformed other assemblers was not in itself a surprise, as its more recent publication and paired assembly graph approach meant that it incorporates methods not always found in other assemblers. However, other assemblers were better performers by some metrics (for example, ABySS generated the largest single contig). The addition of long read data or multiple library sizes would allow us to further scaffold this data in the future.

The number of bases recovered by our SPAdes assembly (209,989,122 at 500 bp minimum contig size) falls short of the estimated genome size of this sponge by a considerable margin, but would comfortably contain the coding sequence of the average

sponge genome. It is likely that repetitive sequences are poorly represented in our dataset, due to the single small fragment size used in sequencing.

Additional Details, Annotation:

For all three species present in our sampling, the most commonly top-hit species by BLAST searches was the sponge *Amphimedon queenslandica* with 11,328, 12,236 and 12,358 hits respectively. The affinity of this species, the first sequenced poriferan, with our samples is obvious. As *A. queenslandica* is also a demosponge, and has its genome on the *nr* database, it is unsurprising that it is the species with the highest number of BLAST top-hits. The top five most hit species for all three samples also included *Orbicella faveolata*, *Stylophora pistillata*, *Exaiptasia pallida* and *Branchiostoma belcheri*, although the order in which these occurred slightly changes from sample to sample. The first three of these species are cnidarians, and *B. belcheri* is a cephalochordate. These species have a relatively slow rate of molecular evolution, likely resulting in their similarity to our sequences under BLAST search.

Tables

<i>Prior to Cleaning</i>	<i>Baikalospongia bacillifera (A2)</i>	<i>Lubomirskia abietina (A10)</i>	<i>Lubomirskia baikalensis (A8)</i>	<i>Lubomirskia baikalensis</i> gDNA Reads
Number of Read Pairs	50,694,150	52,345,337	54,439,423	184,491,682
Read Length	101	101	101	151
GC%	49	49	49	43.5
Average Quality	34.4	34.39	33.74	37.5
Total bases	10,240,218,300	10,573,758,074	10,996,763,446	55,716,487,964
<i>After Cleaning</i>	A2	A10	A8	gDNA Reads
Number of Read Pairs	48,445,101	50,007,487	32,204,065	81,042,203
GC% (Paired)	49	49	49	41
Average Quality (Paired)	36.25	36.25	36.15	39.75
Total Bases (Paired)	9,518,960,084	9,822,304,994	6,321,981,078	19,426,404,098
Unpaired Reads	2129241	2214127	1381971	n/a
GC% (Unpaired)	50	50	50	n/a
Average Quality (Unpaired)	33	33	32.9	n/a
Total bases (Unpaired)	183279397	190482063	117844996	n/a

Table 1: Metrics relating to reads, before and after read cleaning.

	<i>Baikalospongia bacillifera</i> (A2)	<i>Lubomirskia abietina</i> (A10)	<i>Lubomirskia baikalensis</i> (A8)
Number of Trinity Transcripts	80,925	93,404	81,951
Number of Trinity 'Genes'	54,606	62,809	54,913
Min contig length:	201	201	201
Max contig length:	16,639	30,430	11,157
Mean contig length:	850.38	849.74	854.37
N50 contig length:	1,595	1,628	1,572
Number of contigs >=1kb:	20,558	22,946	21,595
Number of contigs in N50:	12,300	13,341	12,943
Number of bases in all contigs:	68,817,041	79,368,987	70,016,550
Number of bases in contigs >=1kb:	44,913,440	52,015,444	45,916,970
GC Content of contigs: (%)	46.75	46.62	46.78

Table 2: Statistics relating to transcriptome assemblies

	SPAdes, 500 min	SPAdes, 200 min	ABYSS, 61mer	SOAPdenovo, 61mer	Velvet, 61mer
Min contig length:	500	200	200	200	200
Max contig length:	124,926	124,926	216,201	99,521	21,493
Mean contig length:	1553.28	681.89	729.9	544.51	464.06
Median contig length:	845	351	294	324	292
N50 contig length:	2,213	1,019	1,931	661	544
Number of contigs:	135,191	451,479	235,631	579,486	357,804
Number of contigs >=1kb:	54,728	54,728	22,065	49,540	28,260
Number of contigs in N50:	19,573	53,387	14,759	95,954	72,346
Number of bases in all contigs:	209,989,122	307,857,163	171,988,112	315,536,346	166,041,199
Number of bases in contigs >=1kb:	155,282,664	155,282,664	96,787,949	120,598,815	51,363,918
GC Content of contigs: (%)	43.68	42.64	43.85	40.94	44.39

Table 3: Genome assembly using a variety of programmes. SPAdes (with a minimum size cutoff of 500 bp) used for further analyses, but other assemblies also available for download.

property	min	max
Heterozygosity	1.75375%	1.80378%
Genome Haploid Length	558,344,824 bp	565,078,853 bp
Genome Repeat Length	350,461,528 bp	354,688,339 bp
Genome Unique Length	207,883,296 bp	210,390,514 bp
Model Fit	85.7458%	98.2203%
Read Error Rate	0.574939%	0.574939%

Table 4: Genome metrics for *L. baikalensis*, computed using Genoscope, using Jellyfish-derived *k* mer counts (size = 21 bp).

<i>Eukaryote Cassette:</i>	<i>Baikalospongia bacillifera (A2)</i>	<i>Lubomirskia abietina (A10)</i>	<i>Lubomirskia baikalensis (A8)</i>	SPAdes Assembly
Complete BUSCOs	299	297	291	235
-Complete (Single Copy)	192	196	178	204
-Complete (Duplicated)	107	101	113	31
Fragmentary BUSCOs	2	2	9	51
Missing BUSCOs	2	4	3	17
Total BUSCO genes	303	303	303	303
<i>Metazoan Cassette:</i>	<i>A2</i>	<i>A10</i>	<i>A8</i>	SPAdes Assembly
Complete BUSCOs	918	914	904	562
-Complete (Single Copy)	555	556	515	493
-Complete (Duplicated)	363	358	389	69
Fragmentary BUSCOs	19	18	31	247
Missing BUSCOs	41	46	43	169
Total BUSCO genes	978	978	978	978

Table 5: BUSCO results for genome and transcriptome assemblies.