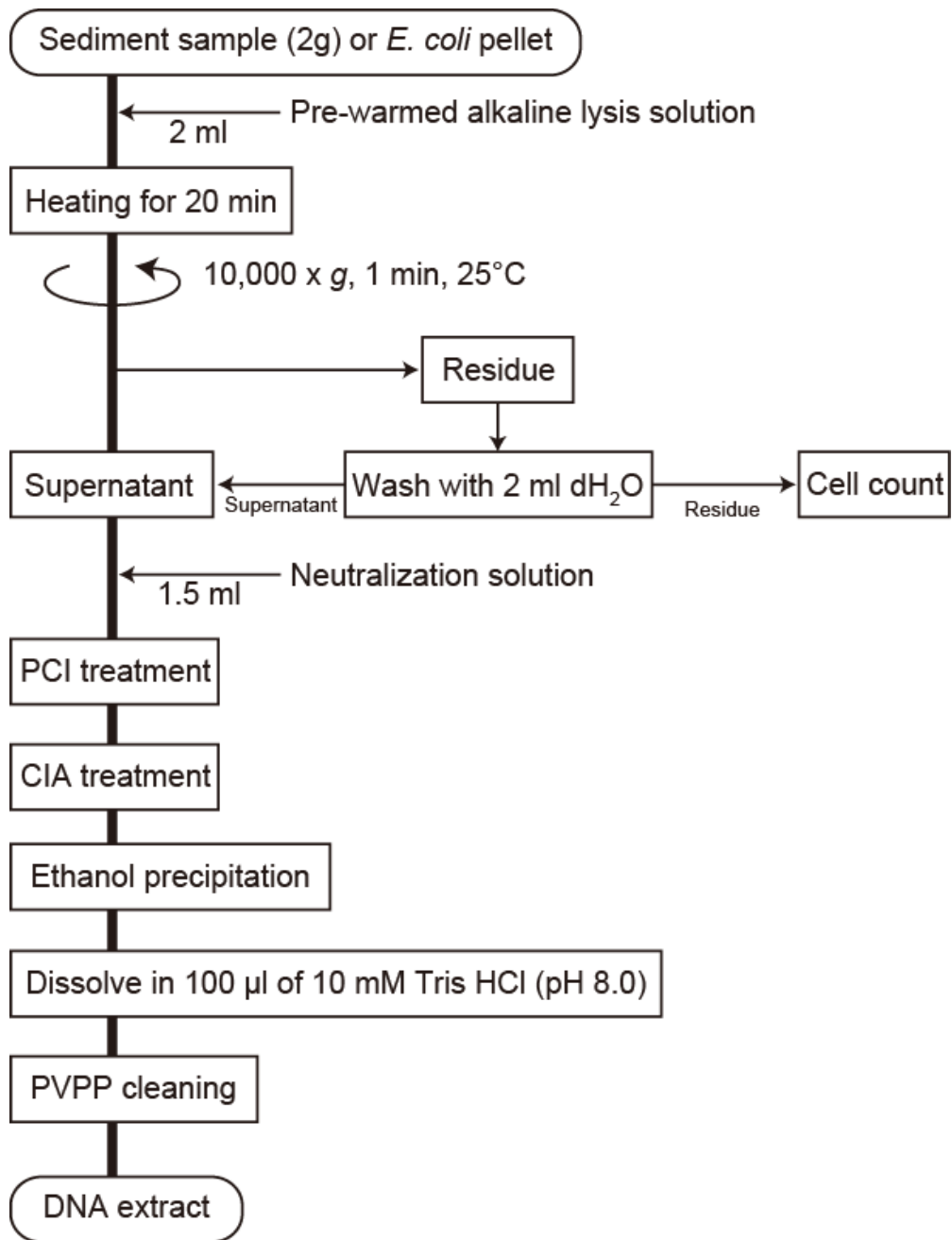
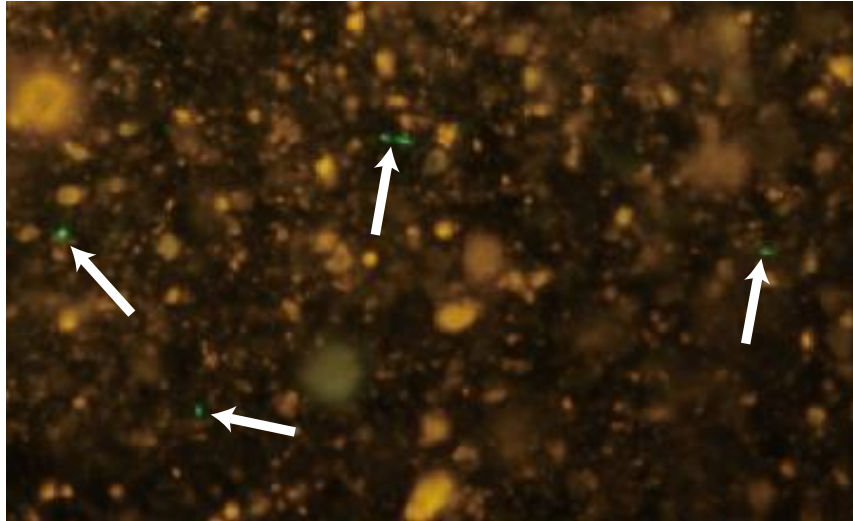


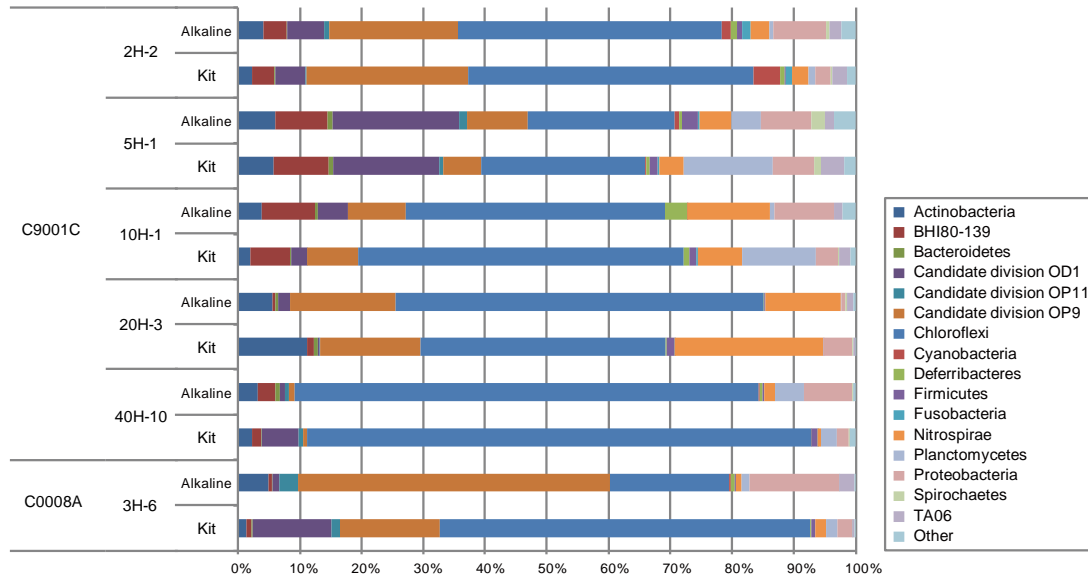
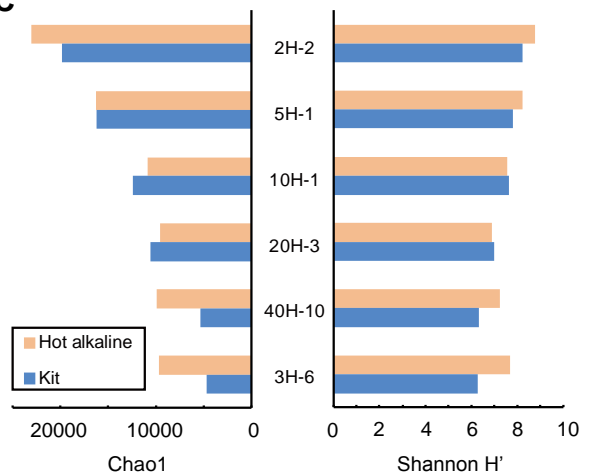
## Supplementary materials



Supplementary Figure 1. Summarized scheme for hot alkaline DNA extraction



**Supplementary Figure 2. Microbial cells found in residue of sediment sample of 2H-2 after DNA extraction with a commercial kit. Microbial cells are indicated by white arrows.**

**A****B****C**

### Supplementary Fig. 3. Characterization of extracted bacterial DNA

(A) Phylogenetic composition of bacterial 16S rRNA genes as determined by pyrosequencing of extracted DNAs.

(B) Bacterial diversity estimated by Chao 1 species richness and Shannon-Wiener ( $H'$ ) indices calculated using

pyrosequence data with a similarity cut-off of 97%, and (C) 99%.

**Supplementary Table 1. Effect of extracellular DNA washing procedure on microbial cell number.** To evaluate the potential effect of the extracellular DNA washing procedure on intact microbial cells, we determined the number of intact cells before and after washing and found that there was no significant difference in cell number even after washing for 1 h at room temperature.

Microbial cell abundance (cells cm <sup>-3</sup> )	
Directly suspended	$4.43 \pm 1.72 \times 10^7$
ASW wash 1h	$4.24 \pm 1.79 \times 10^7$

Supplementary Table 2. Summary of pyrosequence data

Expedition	Drilling site	Core	Depth (mbsf)	DNA extraction method	Number of sequence reads for bacteria	Chao 1 estimates		Shannon index	
						97%	99%	97%	99%
Chikyu shakedown cruise CK06-06	2H-2		9.5	kit	17051	5900	19805	6.87	8.21
				hot-alkaline	20899	5390	23036	7.19	8.78
	5H-1		35.4	kit	10368	861	4353	4.97	6.90
				hot-alkaline	10345	1112	11063	5.79	8.03
	10H-1	C9001C	84.0	kit	16359	3951	16200	6.39	7.81
				hot-alkaline	18223	3608	16289	6.62	8.23
	20H-3		177.4	kit	14171	703	4114	5.10	6.95
				hot-alkaline	8728	819	8224	5.48	7.65
	40H-10	C0008A	364.0	kit	15126	2860	12403	6.11	7.62
				hot-alkaline	16904	1889	10867	5.72	7.55
3H-6	Integrated Ocean Drilling Program Expedition 316	22.0	kit	12724	280	2286	3.89	5.80	
			hot-alkaline	11215	187	3507	3.16	5.96	
			kit	18587	1883	10580	5.27	6.99	
			hot-alkaline	21086	1624	9592	5.12	6.89	
			kit	10439	225	1458	3.40	5.21	
			hot-alkaline	8789	535	4552	4.10	6.59	
			kit	29414	1401	5309	5.28	6.31	
			hot-alkaline	19750	1795	9895	5.52	7.25	
			kit	16350	168	1331	3.32	4.78	
			hot-alkaline	6997	417	4284	4.23	6.79	
			kit	27270	1209	4677	5.14	6.27	
			hot-alkaline	15985	1625	9702	5.70	7.67	
			kit	13919	187	1182	3.05	4.88	
			hot-alkaline	11731	259	4029	3.85	6.55	