TABLE S1. Distribution of enzymes related to *de novo* and salvage pathways of NAD^+ in archaea.

		Salvage		Salvage and *de novo*		*De novo*		
	Organism	Nm deamidase	NaPRT	NMNAT	NAD^+ synthetase	QaPRT	Qa synthase	Asp oxidase
		TK1650 or MtPncA[a]	TK1676	TK0067	TK1798	TK0218	TK0296	TK0297 or TmNadX[b]
Crenarchaeota	*Thermoproteus tenax*		TTX_0495 WP_014126419.1(52%)	TTX_0354 WP_014126286.1(44%)	TTX_1283 WP_014127173.1(42%)	TTX_0292 WP_014126225.1(48%)	TTX_0294 WP_014126227.1(44%)	TTX_0293 WP_014126226.1(40%)
				TTX_1599 WP_014127478.1(37%)	TTX_0625 WP_014126542.1(42%)	TTX_0251 WP_014126184.1(37%)		TTX_0864 WP_014126775.1(32%)
								TTX_1104 WP_014127002.1(31%)
	Pyrobaculum aerophilum	PAE3001 AAL64601.1(30%)*	PAE2990 AAL64593.1(51%)	PAE2259 AAL64068.1(43%)		PAE3114 AAL64682.1(46%)		PAE0716 AAL62973.1(32%)
	Aeropyrum pernix	APE_2350.1 BAA81363.2(30%)*	APE_1273.1 BAA80263.2(50%)					APE_0950.1 BAA79934.2(31%)
	Sulfolobus acidocaldarius	Saci_0543 WP_011277441.1(25%)*	Saci_1470 WP_011278293.1(56%)	Saci_0300 WP_011277218.1(41%)	Saci_0009 WP_011276940.1(39%)	Saci_0547 WP_011277445.1(46%)	Saci_0023[c] WP_011276954.1(48%)	Saci_0549 WP_011277447.1(37%)
								Saci_0982 WP_011277845.1(33%)
	Sulfolobus solfataricus	SSO2455(25%)*[c]	SSO0354 WP_009990652.1(55%)	SSO0255 WP_014511516.1(45%)	SSO2172(38%)[c]	SSO0996 WP_009989246.1(47%)	SSO0998 WP_009989240.1(49%)	SSO0997 WP_009989244.1(39%)
								SSO2356 WP_009989496.1(32%)
	Sulfolobus tokodaii	STK_05820 WP_010978560.1(28%)*	STK_13760 WP_010979420.1(56%)	STK_06480 WP_052846904.1(44%)	STK_21590 WP_052847022.1(40%)	STK_11980 WP_010979217.1(50%)	STK_11970 WP_052846504.1(49%)	STK_11960 WP_010979215.1(36%)
								STK_04970 WP_010978474.1(32%)
	Desulfurococcus amylolyticus	Desfe_0074 WP_014766892.1(34%)	Desfe_0973 WP_014767757.1(54%)	Desfe_1385 WP_014768139.1(44%)	Desfe_0092 WP_014766910.1(40%)			
				Desfe_0747 WP_014767544.1(42%)				
Euryarchaeota	*Thermoplasma acidophilum*	Ta0454 WP_010900881.1(33%)	Ta1145 WP_010901554.1(50%)	Ta0774 WP_048162318.1(47%)	Ta0899 WP_010901309.1(44%)			Ta1001 WP_010901412.1(29%)
	Thermoplasma volcanium	TVG0787823 WP_010917029.1(32%)	TVG1306677 BAB60408.1(49%)	TVG0879099 WP_010917103.1(47%)	TVG1071522 WP_010917275.1(43%)			TVG0761794 WP_010917005.1(29%)
	Archaeoglobus fulgidus DSM 4304	AF_2151 WP_010879640.1(41%)	AF_0821 WP_010878324.1(54%)	AF_2315 WP_010879804.1(51%)	AF_1000 WP_010878500.1(48%)	AF_1839 WP_010879333.1(51%)		AF_0681 WP_010878184.1(31%)
				AF_1488 WP_010878985.1(39%)				AF_1838 WP_010879332.1(39%)†

	Archaeoglobus fulgidus DSM 8774	AFULGI_00024210 WP_010879640.1(41%)	AFULGI_00009070 AIG97699.1(54%)	AFULGI_00026150 WP_010879804.1(51%)	AFULGI_00010940 WP_010878500.1(48%)	AFULGI_00020950 AIG98839.1(51%)	AFULGI_00020930 AIG98837.1(48%)	AFULGI_00007610 WP_010878184.1(31%)
				AFULGI_00017400 WP_010878985.1(39%)			AFULGI_00020920 AIG98836.1(62%)	AFULGI_00020940 WP_010879332.1(39%)†
	Halobacterium salinarum	VNG_2537G WP_010903898.1(29%)*	VNG_2541C AAG20598.1(50%)	VNG_0301C WP_010902177.1(46%)	VNG_2031G WP_010903494.1(41%)	VNG_1884G WP_010903380.1(42%)	VNG_1882G WP_049892521.1(32%)	VNG_1883G WP_010903379.1(32%)
		VNG_1100C WP_010902791.1(35%)*						VNG_1306G WP_010902947.1(32%)
	Methanosarcina mazei			MM_0626 AAM30322.1(49%)	MM_0446WP_011032399.1(45%)	MM_2070WP_011034002.1(48%)	MM_2073WP_011034005.1(49%)	MM_2072AAM31768.1(38%)†
					MM_0612 WP_011032563.1(28%)	MM_1577 AAM31273.1(31%)		
	Methanosarcina acetivorans			MA_3731 WP_048065785.1(47%)	MA_3526 WP_011023442.1(47%)	MA_0955 WP_011020993.1(48%)	MA_0959 WP_011020997.1(49%)	MA_0958 WP_011020996.1(38%)†
	Thermococcus onnurineus	TON_1968 WP_012570973.1(82%)	TON_1362 WP_012572324.1(88%)	TON_1164 WP_012572124.1(94%)	TON_1770 WP_012572732.1(78%)	TON_1889 WP_012572852.1(86%)	TON_1887 WP_012572850.1(86%)	TON_1885 WP_012572848.1(75%)
	Thermococcus litoralis	OCC_10179 WP_004066119.1(77%)	OCC_06996 WP_004067753.1(89%)	OCC_07636 WP_004066969.1(83%)	OCC_12951 WP_004070194.1(72%)	OCC_06601 WP_004067606.1(74%)	OCC_06606 WP_004067607.1(79%)	OCC_06611 WP_004067610.1(72%)
	Pyrococcus furiosus	PF1105 WP_011012244.1(86%)	PF1904 WP_011013044.1(83%)	PF0458 WP_011011575.1(86%)	PF0098 WP_011011210.1(69%)	PF1978 WP_011013120.1(78%)	PF1977 WP_011013119.1(83%)	PF1976 WP_011013118.1(68%)
	Pyrococcus abyssi	PAB1720 WP_010868100.1(85%)	PAB1131 WP_010868844.1(83%)	PAB1318 WP_010868691.1(86%)	PAB2244 WP_010867289.1(76%)	PAB2347 WP_010867134.1(84%)	PAB2345 WP_048146454.1(86%)	PAB2343 WP_048147208.1(67%)
	Pyrococcus horikoshii	PH0999 WP_010885086.1(86%)	PH1868 BAA30990.1(81%)	PH0464 WP_010884571.1(85%)	PH0182 WP_010884291.1(73%)	PH0011 WP_048053010.1(82%)	PH0013 WP_010884133.1(84%)	PH0015 WP_048053535.1(67%)
	Methanocaldococcus jannaschii			MJ0541 WP_010870045.1(55%)	MJ1352 WP_064496806.1(50%)	MJ0493 WP_010869994.1(50%)	MJ0407 WP_064496486.1(54%)	MJ0033 WP_010869524.1(31%)
								MJ0915 WP_010870429.1(38%)†
	Methanococcus maripaludis			MMP1578 WP_011171522.1(45%)	MMP1349 WP_011171293.1(47%)	MMP0877 WP_011170821.1(39%)	MMP1242 WP_011171186.1(53%)	MMP1277 WP_011171221.1(31%)
								MMP0737 WP_011170681.1(34%)†

Gene numbers, accession numbers and identities based on the results of BLAST searches. Unless indicated otherwise, BLAST searches were performed with protein sequences from *T. kodakarensis* involved in NAD^+ biosynthesis.

[a]Nicotinamide deamidase from the bacterium *Mycobacterium tuberculosis* (MtPncA) was also used in the BLAST search. Proteins identified in the search are indicated with asterisks.

[b]Aspartic acid dehydrogenase from the bacterium *Thermotoga maritima* (TmNadX) was also used in the BLAST search. Proteins identified in the search are indicated with daggers.

[c]Sequences identified with BLAST searches at Kyoto Encyclopedia of Genes and Genomes (KEGG). These sequences were not present in the NCBI database. Identity values were determined manually after alignment.

TABLE S2. Primers used in this study.

No.	Primer name	Sequence (5'-3')
1	eTK1676-F	AAGGAGATATACATATGCGCGACTTCTACATTGC
2	eTK1676-R	GCTCGAATTCGGATCTCAATCAATGCTCAAGTTG
3	dTK0218-F	TCATCTCCCGGAACTTCCTCCGGAGGTCTT
4	dTK0218-R	AACGATAGCCTTCGCCATCTTCGGCGGGTTG
5	dTK1650-F	CCCCTTAGTCTGGACTCTTACTGCTCTCT
6	dTK1650-R	CTGACTAAATAAATCCTCCGGGAAAACATG
7	dTK1676-F	CGGTCTTGCTGCTATTGCCGCGTTGG
8	dTK1676-R	GCGCGTTAAGGCCCCGAGCGTGTGGAGCA
9	invdTK0218-F	TGAACTTTTTCGCTCTTCGCCAATACTTT
10	invdTK0218-R	CACTCTCACCAGTTCTCACTCTAGTTCTCT
11	invdTK1650-F	TACCCTCCACGATGAGCTTCCCAGTCTT
12	invdTK1650-R	TCAGGCTCGTCCCAGTGGTTGACGG
13	invdTK1676-F	GCGTCTTCTTTCTTGTTTTCTTTATCTCTTTTTG
14	invdTK1676-R	GGTTCTCACCTTGGAAAATAGTTAAGCCC
15	idTK0218-F	ATGGTTCCGCTCGAGTATCTGCTCAGGTT
16	idTK0218-R	TCAAAGCCTGCCAATTATCTCAAGGCTTA
17	idTK1650-F	ATGCCGAAGGAGGCCCTCATAGTTGTT
18	idTK1650-R	TGCCCTTAACTGCATCGCTCAGGAGGT
19	idTK1676-F	ATGCGCGACTTCTACATTGCCCATGAGGA
20	idTK1676-R	TCAATCAATGCTCAAGTTGAGCTTCTTCG
21	odTK0218-F	TACCCTCCACGATGAGCTTCCCAGTCTT
22	odTK0218-R	TCAGGCTCGTCCCAGTGGTTGACGG
23	odTK1650-F	AGAAGAATTCCGTAAAAGTGAGAAG
24	odTK1650-R	CATCCCAACCACCGGGAGGTTTTTAT
25	odTK1676-F	TGGCATTCCGCTGGGGGCAGTTATCAAGAG
26	odTK1676-R	GCCAGCACAGACCATCCCCTGGGGAATCGA

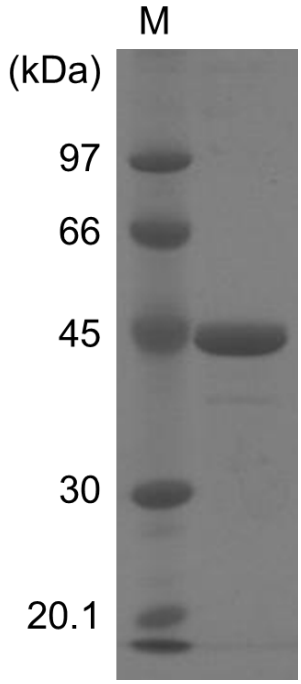

FIG S1. Purification of the recombinant TK1676 protein. Purified TK1676 protein (4 μg) was applied and separated by SDS-PAGE and stained with Coomassie Brilliant Blue. M: molecular mass marker.

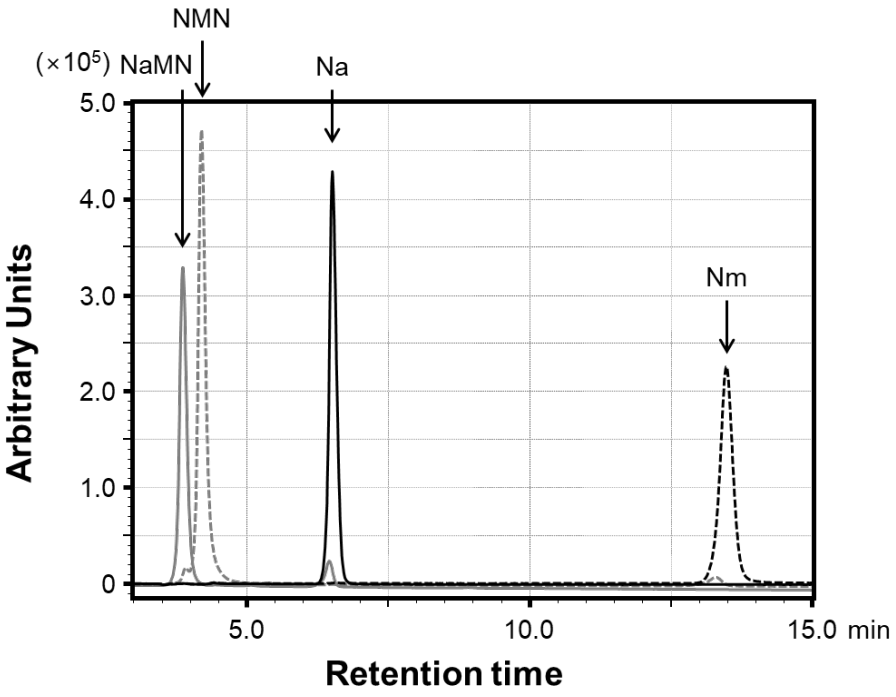

FIG S2. HPLC chromatograms of standard compounds. Standard compounds (2 mM) were analyzed by HPLC. Black solid, black dashed, gray solid and gray dashed lines correspond to Na, Nm, NaMN and NMN standards, respectively.

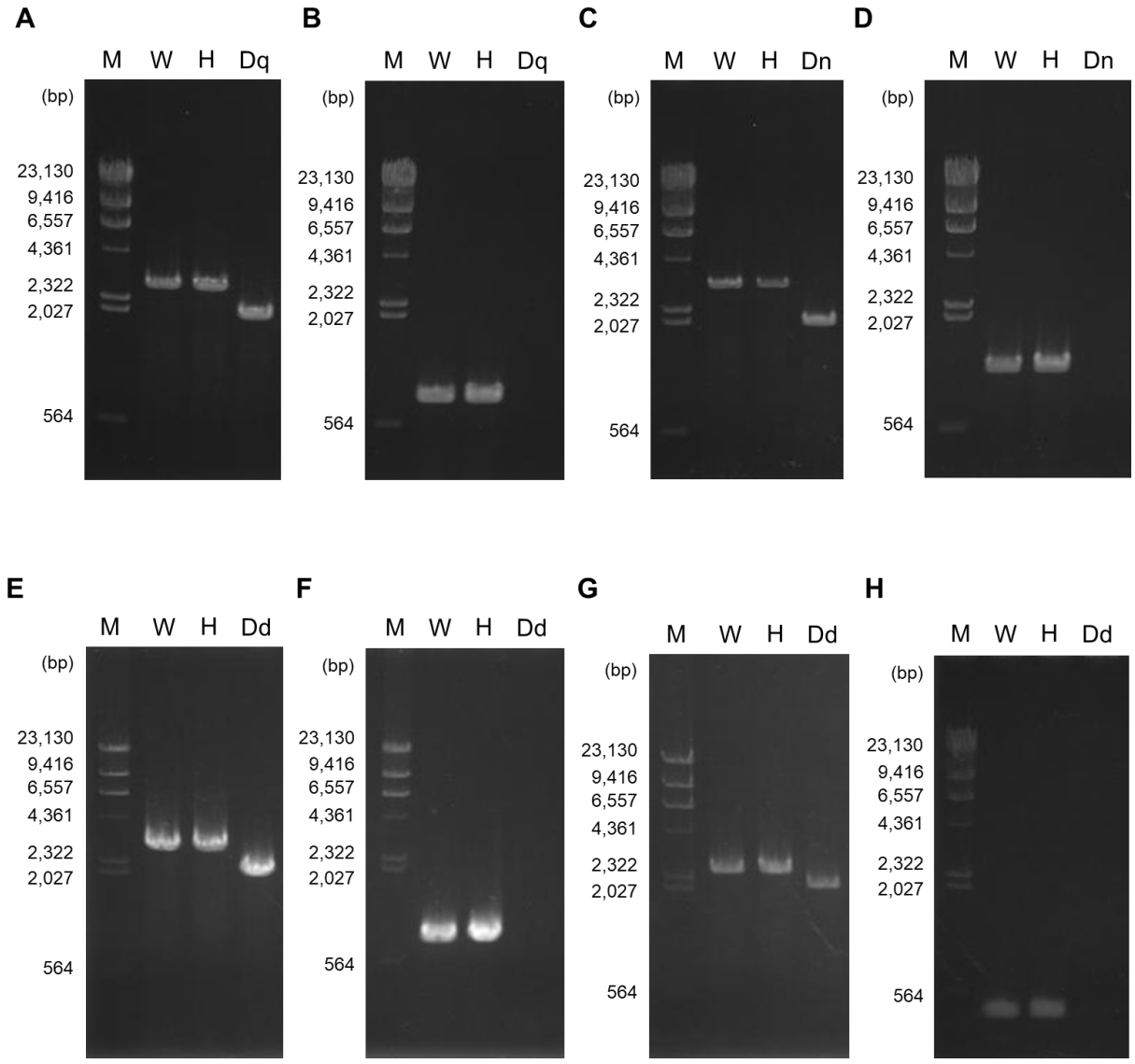

FIG S3. Genotypic analyses of the gene disruption strains by PCR. PCR analyses of the TK0218 and TK1676 gene disruption strains were performed with primer sets that anneal outside of the TK0218-flanking regions (A) and TK1676-flanking regions (C) for homologous recombination and within the TK0218 (B) and TK1676 (D) genes. PCR analyses of the ΔTK0218ΔTK1650 double gene disruption strain were performed with primer sets that anneal outside of the TK0218- and TK1650-flanking regions for homologous recombination (E and G) and within the TK0218 and TK1650 genes (F and H). Abbreviations: M; marker, W; *T. kodakarensis* wild-type KOD1, H; the host strain KU216, Dq; the ΔTK0218 strain, Dn; the ΔTK1676 strain, Dd; the ΔTK0218ΔTK1650 strain.