

SI Figure 1a. Taxa detected in acidic (pH<4.5) high temperature (>73C) sulfidic (>3 μM) springs of western North America. Asterisk indicates that a *merA* sequence most closely affiliated with this taxon was also recovered from one or more sites in which an isolate or the 16S rRNA of this taxon was obtained. Numeral indicates number of sites with Hg data.

Figure 1a References

- Beam, J.P., Jay, Z.J., Kozubal, M.A. and Inskeep, W.P. (2014) Niche specialization of novel Thaumarchaeota to oxic and hypoxic acid geothermal springs of Yellowstone National Park. *ISME J.* 8, 938-951.
- Boyd, E.S., Hamilton, T.L., Wang, J., He, L. and Zhang, C.L. (2013) The role of tetraether lipid composition in the adaptation of thermophilic archaea to acidity. *Front. Microbiol.* 4, doi: 10.3389/fmicb.2013.00062.
- Ellis, D.G., Bizzoco, R.W. and Kelley, S.T. (2008) Halophilic Archaea determined from geothermal steam vent aerosols. *Environ. Microbiol.* 10, 1582-1590.
- Hamamura, N., Macur, R.E., Korf, S., Ackerman, G., Taylor, W.P., Kozubal, M., Reysenbach, A.-L. and Inskeep, W.P. (2009) Linking microbial oxidation of arsenic with detection and phylogenetic analysis of arsenite oxidase genes in diverse geothermal environments. *Environ. Microbiol.* 11, 421-431.
- Inskeep, W.P., Jay, Z.J., Herrgard, M.J., Kozubal, M.A., Rusch, D.B., Tringe, S.G., Macur, R.E., Jennings, R.d., Boyd, E.S., Spear, J.R. and Roberto, F. (2013a) Phylogenetic and functional analysis of metagenome sequence from high-temperature archaeal habitats demonstrate linkages between metabolic potential and geochemistry. *Front. Microbiol.* 4, doi: 10.3389/fmicb.2013.00095.
- Inskeep, W.P., Jay, Z.J., Tringe, S.G., Herrgard, M.J., Rusch, D.B. and Members, Y.M.P.S.C.a.W.G. (2013b) The YNP metagenomic project: environmental parameters responsible for microbial distribution in the Yellowstone geothermal ecosystem. *Front. Microbiol.* 4, 67. doi: 10.3389/fmicb.2013.00067.
- Inskeep, W.P. and McDermott, T.M. (2005) Geomicrobiology of acid-sulfate-chloride springs in Yellowstone National Park, in: Inskeep, W.P., McDermott, T.M. (Eds.), *Geothermal Biology and Geochemistry in Yellowstone National Park*. Montana State University, Bozeman, pp. 143-162.
- Jay, Z.J. (2014) Linking geochemistry with microbial community structure and function in sulfidic geothermal systems of Yellowstone National Park, *Land Resources and Environmental Sciences*. Montana State University, Bozeman, MT, p. 258.
- Jay, Z.J. and Inskeep, W.P. (2015) The distribution, diversity, and importance of 16S rRNA gene introns in the order Thermoproteales. *Biol. Direct* 10, 1-10.
- Jay, Z.J., Rusch, D.B., Tringe, S.G., Bailey, C., Jennings, R.M. and Inskeep, W.P. (2014) Predominant *Acidilobus*-like populations from geothermal environments in Yellowstone National Park exhibit similar metabolic potential in different hypoxic microbial communities. *Appl. Environ. Microbiol.* 80, 294-305.

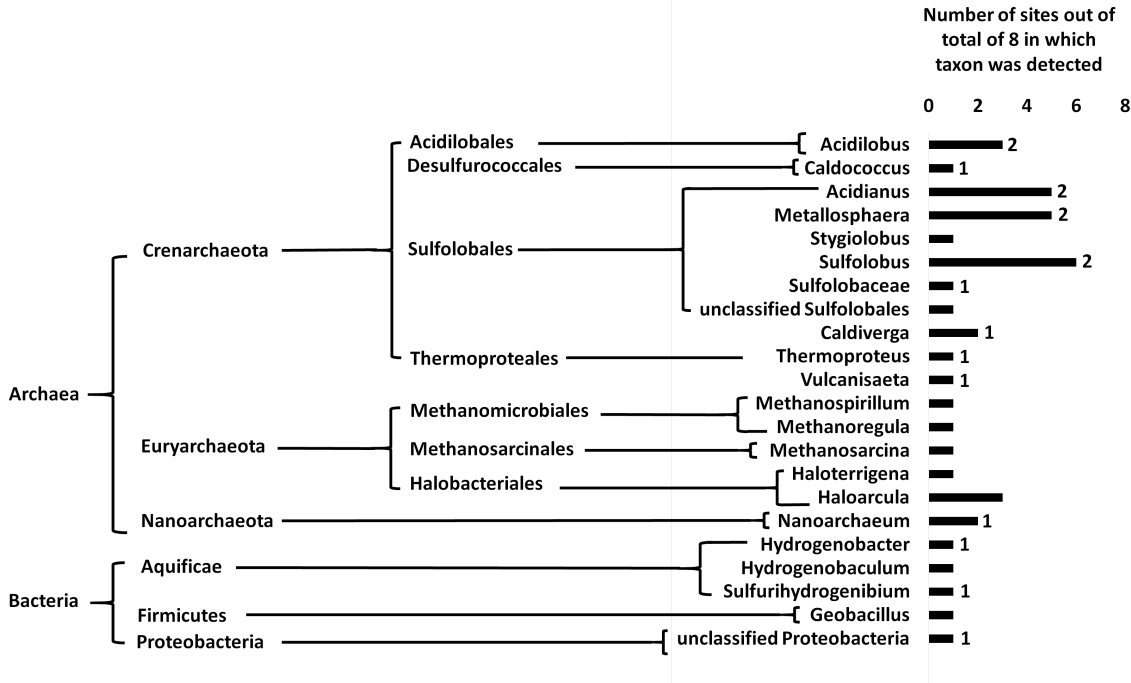
- Kozubal, M., Macur, R.E., Korf, S., Taylor, W.P., Ackerman, G.G., Nagy, A. and Inskeep, W.P. (2008) Isolation and distribution of a novel iron-oxidizing crenarchaeon from acidic geothermal springs in Yellowstone National Park. *Appl. Environ. Microbiol.* 74, 942-949.
- Kozubal, M.A., Macur, R.E., Jay, Z.J., Beam, J.P., Malfatti, S.A., Tringe, S.G., Kocar, B.D., Borch, T. and Inskeep, W.P. (2012) Microbial cycling in acidic geothermal springs of Yellowstone National Park: integrating molecular surveys, geochemical processes, and isolation of novel Fe-active microorganisms. *Front. Microbiol.* 3, doi:10.3389/fmicb.2012.00109.
- Kozubal, M.A., Romine, M., Jennings, R.d., Jay, Z.J., Tringe, S.G., Rusch, D.B., Beam, J.P., McCue, L.A. and Inskeep, W.P. (2013) Geoarchaeota: a new candidate phylum in the Archaea from high-temperature acidic iron mats in Yellowstone National Park. *ISME J* 7, 622-634.
- Macur, R.E., Jay, Z.J., Taylor, W.P., Kozubal, M.A., Kocar, B.D. and Inskeep, W.P. (2013) Microbial community structure and sulfur biogeochemistry in mildly-acidic sulfidic geothermal springs in Yellowstone National Park. *Geobiology* 11, 86-99.
- Macur, R.E., Langner, H.W., Kocar, B.D. and Inskeep, W.P. (2004) Linking geochemical processes with microbial community analysis: Successional dynamics in an arsenic-rich, acid-sulfate-chloride geothermal spring. *Geobiology* 2, 163-177.
- Menzel, P., Gudbergdottir, S.R., Rike, A.G., Lin, L., Zhang, Q., Contursi, P., Moracci, M., Kristajansson, J.K., Bolduc, B., Gavrilov, S., Raviin, N., Mardanov, A., Osmolovskaya, E.B.-., Young, M., Krogh, A. and Peng, X. (2015) Comparative metagenomics of eight geographically remote terrestrial hot springs. *Microb. Ecol.* 70, 411-424.
- Reysenbach, A.-L., Banta, A., Civello, S., Daly, J., Mitchel, K., Lalonde, S., Konhauser, K., Rodman, A., Rusterholtz, K. and Takacs-Vesbach, C. (2005) Aquificales in Yellowstone National Park, in: Inskeep, W.P., McDermott, T.M. (Eds.), *Geothermal Biology and Geochemistry in Yellowstone National Park*. Thermal Biology Institute and Department of Land Resources & Environmental Sciences, Bozeman, pp. 129-142.
- Siering, P.L., Clarke, J.M. and Wilson, M.S. (2006) Geochemical and Biological Diversity of Acidic, Hot Springs in Lassen Volcanic National Park. *Geomicrobiol. J.* 23, 129-141.
- Siering, P.L., Wolfe, G.V., Wilson, M.S., Yip, A.N., Carey, C.M., Wardman, C.D., Shapiro, R.S., Stedman, K.M., Kyle, J., Yuan, T., Van Nostrand, J.D., He, Z. and Zhou, J. (2013) Microbial biogeochemistry of Boiling Springs Lake: a physically dynamic, oligotrophic, low-pH geothermal ecosystem. *Geobiology* 11, 356-376.
- Simbahan, J., Dribjer, R. and Blum, P. (2004) *Alicyclobacillus vulcanalis* sp.nov., a thermophilic, acidophilic bacterium isolated from Coso Hot Springs, California, USA. *Internat. J. Syst. Evol. Microbiol.* 54, 1703-1707.

Simbahan, J., Kurth, E., Schelert, J., Dillman, A., Moriyama, E., Jovanovich, S. and Blum, P. (2005) Community analysis of a mercury hot spring supports occurrence of domain-specific forms of mercuric reductase. *Appl Environ Microbiol* 71, 8836-8845.

Spear, J.R., Walker, J.J., McCollom, T.M. and Pace, N.R. (2005) Hydrogen and bioenergetics in the Yellowstone geothermal ecosystem. *PNAS (USA)* 102, 2555-2560.

Spear, J.R., Walker, J.J. and Pace, N.R. (2006) Microbial ecology and energetics in Yellowstone hot springs. *Yellowstone Science* 14, 17-24.

Whitaker, R.J., Grogan, D.W. and Taylor, J.W. (2003) Geographic Barriers Isolate Endemic Populations of Hyperthermophilic Archaea. *Science* 301, 976-978.



SI Figure 1b. Taxa detected in acidic (pH<4.5) high temperature (>73C) nonsulfidic (<3μM) hot springs in western North America. Numeral indicates number of sites with Hg data.

Figure 1b References

Boyd, E.S., Hamilton, T.L., Wang, J., He, L. and Zhang, C.L. (2013) The role of tetraether lipid composition in the adaptation of thermophilic archaea to acidity. *Front. Microbiol.* 4, doi: 10.3389/fmicb.2013.00062.

Inskeep, W.P., Jay, Z.J., Herrgard, M.J., Kozubal, M.A., Rusch, D.B., Tringe, S.G., Macur, R.E., Jennings, R.d., Boyd, E.S., Spear, J.R. and Roberto, F. (2013a) Phylogenetic and functional analysis of metagenome sequence from high-temperature archaeal habitats demonstrate linkages between metabolic potential and geochemistry. *Front. Microbiol.* 4, doi: 10.3389/fmicb.2013.00095.

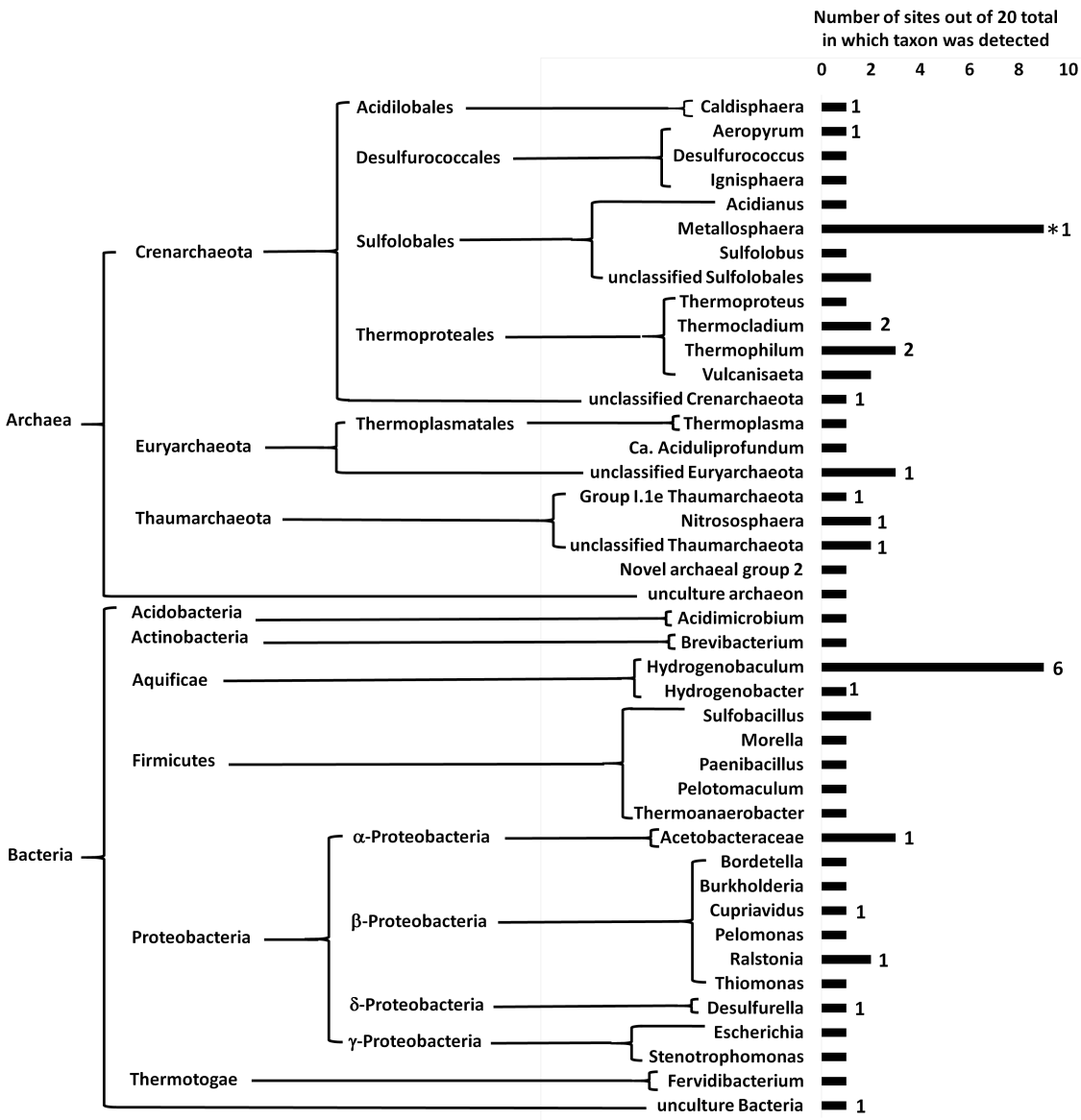
Inskeep, W.P., Jay, Z.J., Tringe, S.G., Herrgard, M.J., Rusch, D.B. and Members, Y.M.P.S.C.a.W.G. (2013b) The YNP metagenomic project: environmental parameters responsible for microbial distribution in the Yellowstone geothermal ecosystem. *Front. Microbiol.* 4, 67. doi: 10.3389/fmicb.2013.00067.

Inskeep, W.P., Rusch, D.B., Jay, Z., Herrgard, M.J., Kozubal, M.A., Richardson, T.H., Macur, R.E., Hamamura, N., Jennings, R.d., Fouke, B.W., Reysenbach, A.-L., Roberto, F., Young, M., Schwartz, A., Boyd, E.S., Badger, J., Mathur, E.J., Ortmann, A.C., Bateson, M., Geesey, G. and Frazier, M. (2010) Metagenomes from high-temperature chemotrophic systems reveal geochemical controls on microbial community structure and function. *PLoS One* 5, e9773.

Kozubal, M., Macur, R.E., Korf, S., Taylor, W.P., Ackerman, G.G., Nagy, A. and Inskeep, W.P. (2008) Isolation and distribution of a novel iron-oxidizing crenarchaeon from acidic geothermal springs in Yellowstone National Park. *Appl. Environ. Microbiol.* 74, 942-949.

Kozubal, M.A., Macur, R.E., Jay, Z.J., Beam, J.P., Malfatti, S.A., Tringe, S.G., Kocar, B.D., Borch, T. and Inskeep, W.P. (2012) Microbial cycling in acidic geothermal springs of Yellowstone National Park: integrating molecular surveys, geochemical processes, and isolation of novel Fe-active microorganisms. *Front. Microbiol.* 3, doi:10.3389/fmicb.2012.00109.

Menzel, P., Gudbergsdottir, S.R., Rike, A.G., Lin, L., Zhang, Q., Contursi, P., Moracci, M., Kristajansson, J.K., Bolduc, B., Gavrillov, S., Raviin, N., Mardanov, A., Osmolovskaya, E.B.-., Young, M., Krogh, A. and Peng, X. (2015) Comparative metagenomics of eight geographically remote terrestrial hot springs. *Microb. Ecol.* 70, 411-424.



SI Figure 1c. Taxa detected in acidic moderate temperature (55-73°C) sulfidic (>3μM) springs of western North America. Asterisk indicates that a *merA* sequence most closely affiliated with this taxon was also recovered from one or more sites in which an isolate or the 16S rRNA of this taxon was obtained. Numeral indicates number of sites with Hg data.

Figure 1c References

- Beam, J.P., Jay, Z.J., Kozubal, M.A. and Inskeep, W.P. (2014) Niche specialization of novel Thaumarchaeota to oxic and hypoxic acid geothermal springs of Yellowstone National Park. *ISME J.* 8, 938-951.
- Boyd, E.S., Hamilton, T.L., Wang, J., He, L. and Zhang, C.L. (2013) The role of tetraether lipid composition in the adaptation of thermophilic archaea to acidity. *Front. Microbiol.* 4, doi: 10.3389/fmicb.2013.00062.
- Boyd, E.S., Leavitt, W.D. and Geesey, G.G. (2009) CO₂ uptake and fixation by a thermoacidophilic microbial community attached to precipitated sulfur in a geothermal spring. *Appl. Environ. Microbiol.* 75, 4289-4296.
- Inskeep, W.P., Jay, Z.J., Tringe, S.G., Herrgard, M.J., Rusch, D.B. and Members, Y.M.P.S.C.a.W.G. (2013) The YNP metagenomic project: environmental parameters responsible for microbial distribution in the Yellowstone geothermal ecosystem. *Front. Microbiol.* 4, 67. doi: 10.3389/fmicb.2013.00067.
- Inskeep, W.P., Macur, R.E., Harrison, G., Bostick, B.C. and Fendorf, S. (2004) Biomineralization of As(V)-hydrous ferric oxyhydroxide in microbial mats of an acid-sulfate-chloride geothermal spring, Yellowstone National Park. *Geochim. Cosmochim. Acta* 68, 3141-3155.
- Inskeep, W.P. and McDermott, T.M. (2005) Geomicrobiology of acid-sulfate-chloride springs in Yellowstone National Park, in: Inskeep, W.P., McDermott, T.M. (Eds.), *Geothermal Biology and Geochemistry in Yellowstone National Park*. Montana State University, Bozeman, pp. 143-162.
- Jackson, C.R., Langner, H.W., Donahoe-Christiansen, J., Inskeep, W.P. and McDermott, T.R. (2001) Molecular analysis of microbial community structure in an arsenite-oxidizing acidic thermal spring. *Environ. Microbiol.* 3, 532-542.
- Kozubal, M., Macur, R.E., Korf, S., Taylor, W.P., Ackerman, G.G., Nagy, A. and Inskeep, W.P. (2008) Isolation and distribution of a novel iron-oxidizing crenarchaeon from acidic geothermal springs in Yellowstone National Park. *Appl. Environ. Microbiol.* 74, 942-949.
- Kozubal, M.A., Macur, R.E., Jay, Z.J., Beam, J.P., Malfatti, S.A., Tringe, S.G., Kocar, B.D., Borch, T. and Inskeep, W.P. (2012) Microbial cycling in acidic geothermal springs of Yellowstone National Park: integrating molecular surveys, geochemical processes, and isolation of novel Fe-active microorganisms. *Front. Microbiol.* 3, doi:10.3389/fmicb.2012.00109.
- Macur, R.E., Langner, H.W., Kocar, B.D. and Inskeep, W.P. (2004) Linking geochemical processes with microbial community analysis: Successional dynamics in an arsenic-rich, acid-sulfate-chloride geothermal spring. *Geobiology* 2, 163-177.

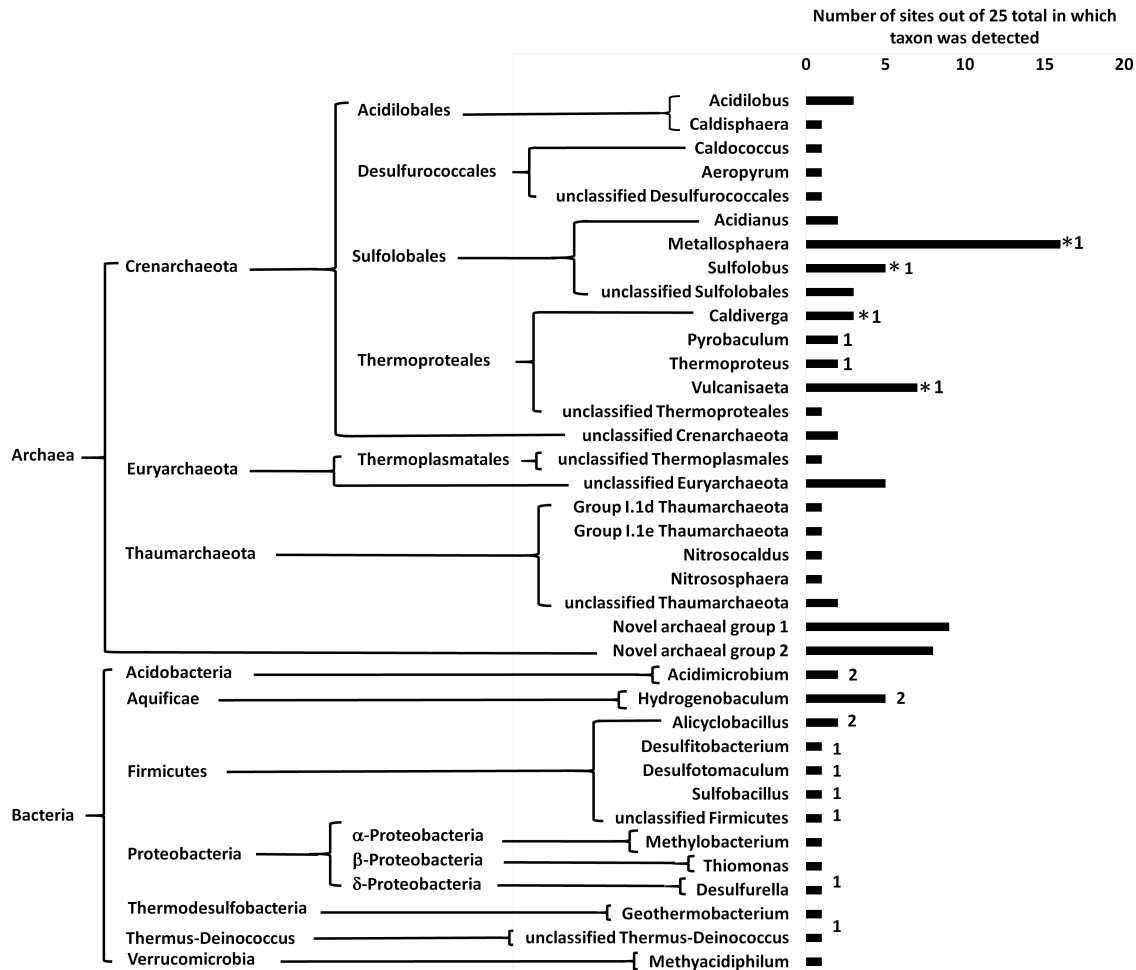
Mathur, J., Bizzoco, R.W., Ellis, D.G., Lipson, D.A., Poole, A.W., Levine, R. and Kelley, S.T. (2007) Effects of abiotic factors on the phylogenetic diversity of bacterial communities in acidic thermal springs. *Appl. Environ. Microbiol.* 73, 2612-2623.

Romano, C., D'Imperio, S., Woyke, T., Mavromatis, K., Lasken, R., Shock, E.L. and McDermott, T.R. (2013) Comparative genomic analysis of phylogenetically closely related *Hydrogenobaculum* sp. isolates from Yellowstone National Park. *Appl. Environ. Microbiol.* 79, 2932-2943.

Takacs-Vesbach, C., Inskeep, W.P., Jay, Z.J., Herrgard, M.J., Rusch, D.B., Tringe, S.G., Kozubal, M.A., Hamamura, N., Macur, R.E., Fouke, B.W., Reysenbach, A.-L., McDermott, T.R., Jennings, R.d., Hengartner, N.W. and Xie, G. (2013) Metagenome sequence analysis of filamentous microbial communities obtained from geochemically distinct geothermal channels reveals specialization of three aquificales lineages. *Front. Microbiol.* 4, doi:10.3389/fmicb.2013.00084.

Whitaker, R.J., Grogan, D.W. and Taylor, J.W. (2003) Geographic barriers isolate endemic populations of hyperthermophilic Archaea. *Science* 301, 976-978.

Wilson, M., Siering, P., White, C., Hauser, M. and Bartles, A. (2008) Novel Archaea and Bacteria Dominate Stable Microbial Communities in North America's Largest Hot Spring. *Microb. Ecol.* 56, 292-305.



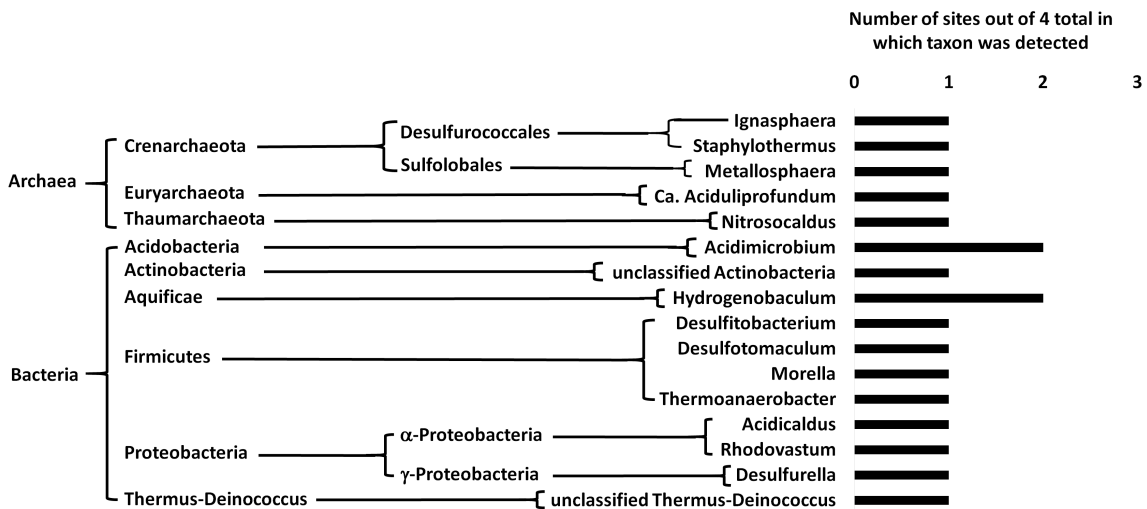
SI Figure 1d. Taxa detected in acidic (pH<4.5) moderate temperature (55-73C) nonsulfidic (<3 μM) springs of western North America. Asterix indicates that a *merA* sequence most closely affiliated with this taxon was also recovered from one or more sites in which an isolate or the 16S rRNA of this taxon was obtained. Numeral indicates number of sites with Hg data.

Figure 1d References

- Beam, J.P., Jay, Z.J., Kozubal, M.A. and Inskeep, W.P. (2014) Niche specialization of novel Thaumarchaeota to oxic and hypoxic acid geothermal springs of Yellowstone National Park. *ISME J.* 8, 938-951.
- Boyd, E.S., Hamilton, T.L., Wang, J., He, L. and Zhang, C.L. (2013) The role of tetraether lipid composition in the adaptation of thermophilic archaea to acidity. *Front. Microbiol.* 4, doi: 10.3389/fmicb.2013.00062.
- Inskeep, W.P., Jay, Z.J., Herrgard, M.J., Kozubal, M.A., Rusch, D.B., Tringe, S.G., Macur, R.E., Jennings, R.d., Boyd, E.S., Spear, J.R. and Roberto, F. (2013a) Phylogenetic and functional analysis of metagenome sequence from high-temperature archaeal habitats demonstrate linkages between metabolic potential and geochemistry. *Front. Microbiol.* 4, doi: 10.3389/fmicb.2013.00095.
- Inskeep, W.P., Jay, Z.J., Tringe, S.G., Herrgard, M.J., Rusch, D.B. and Members, Y.M.P.S.C.a.W.G. (2013b) The YNP metagenomic project: environmental parameters responsible for microbial distribution in the Yellowstone geothermal ecosystem. *Front. Microbiol.* 4, 67. doi: 10.3389/fmicb.2013.00067.
- Inskeep, W.P., Macur, R.E., Harrison, G., Bostick, B.C. and Fendorf, S. (2004) Biomineralization of As(V)-hydrous ferric oxyhydroxide in microbial mats of an acid-sulfate-chloride geothermal spring, Yellowstone National Park. *Geochim. Cosmochim. Acta* 68, 3141-3155.
- Inskeep, W.P., Rusch, D.B., Jay, Z., Herrgard, M.J., Kozubal, M.A., Richardson, T.H., Macur, R.E., Hamamura, N., Jennings, R.d., Fouke, B.W., Reysenbach, A.-L., Roberto, F., Young, M., Schwartz, A., Boyd, E.S., Badger, J., Mathur, E.J., Ortmann, A.C., Bateson, M., Geesey, G. and Frazier, M. (2010) Metagenomes from high-temperature chemotrophic systems reveal geochemical controls on microbial community structure and function. *PLoS One* 5, e9773.
- Jay, Z.J. (2014) Linking geochemistry with microbial community structure and function in sulfidic geothermal systems of Yellowstone National Park, *Land Resources and Environmental Sciences*. Montana State University, Bozeman, MT, p. 258.
- Johnson, D.B., Okibe, N. and Roberto, F.F. (2003) Novel thermo-acidophilic bacteria isolated from geothermal sites in Yellowstone National Park: physiological and phylogenetic characteristics. *Arch. Microbiol.* 180, 60-68.
- Kozubal, M.A., Macur, R.E., Jay, Z.J., Beam, J.P., Malfatti, S.A., Tringe, S.G., Kocar, B.D., Borch, T. and Inskeep, W.P. (2012) Microbial cycling in acidic geothermal springs of Yellowstone National Park: integrating molecular surveys, geochemical processes, and isolation of novel Fe-active microorganisms. *Front. Microbiol.* 3, doi:10.3389/fmicb.2012.00109.

Kozubal, M.A., Romine, M., Jennings, R.d., Jay, Z.J., Tringe, S.G., Rusch, D.B., Beam, J.P., McCue, L.A. and Inskeep, W.P. (2013) Geoarchaeota: a new candidate phylum in the Archaea from high-temperature acidic iron mats in Yellowstone National Park. *ISME J.* 7, 622-634.

Mathur, J., Bizzoco, R.W., Ellis, D.G., Lipson, D.A., Poole, A.W., Levine, R. and Kelley, S.T. (2007) Effects of abiotic factors on the phylogenetic diversity of bacterial communities in acidic thermal springs. *Appl. Environ. Microbiol.* 73, 2612-2623.



SI Figure 1e. Taxa detected in acidic (pH<4.5) low temperature (<55C) sulfidic (>3 μ M) springs of western North America.

Figure 1e References

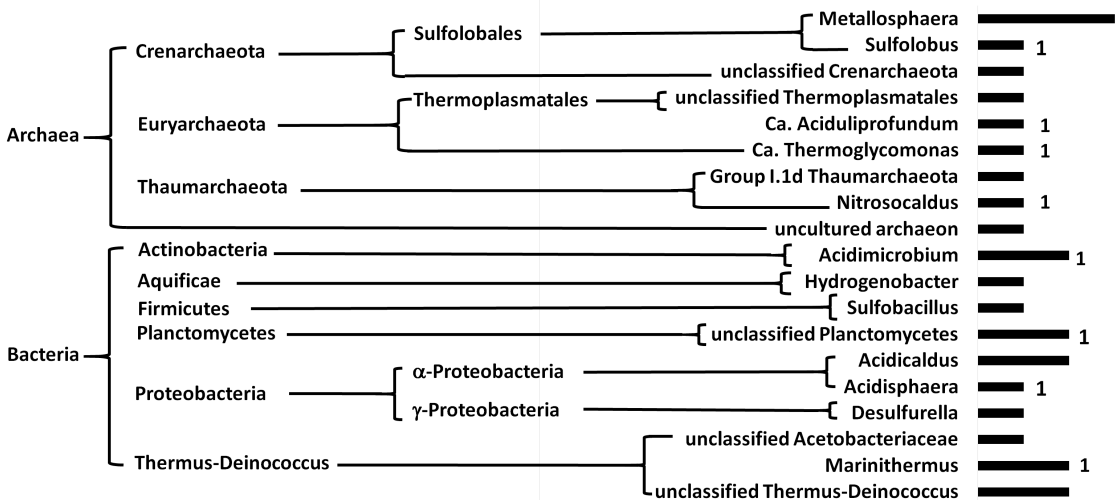
Brown, P.B. and Wolfe, G.V. (2006) Protist genetic diversity in the acidic hydrothermal environments of Lassen Volcanic National Park, USA. *J. Eukaryot. Microbiol.* 53, 420-431.

Ferris, M.J., Magnuson, T.S., Fagg, J.A., Thar, R., Kuhl, M., Sheehan, K.B. and Henson, J.M. (2003) Microbially mediated sulfide production in a thermal, acidic algal mat community in Yellowstone National Park. *Environ. Microbiol.* 5, 954-960.

Siering, P.L., Wolfe, G.V., Wilson, M.S., Yip, A.N., Carey, C.M., Wardman, C.D., Shapiro, R.S., Stedman, K.M., Kyle, J., Yuan, T., Van Nostrand, J.D., He, Z. and Zhou, J. (2013) Microbial biogeochemistry of Boiling Springs Lake: a physically dynamic, oligotrophic, low-pH geothermal ecosystem. *Geobiology* 11, 356-376.

Number of sites out of 8 total in which taxon was detected

0 1 2 3



SI Figure 1f. Taxa detected in acidic (pH<4.5) low temperature (<55C) nonsulfidic (<3 μM) springs of western North America. Numeral indicates number of sites with Hg data.

Figure 1f References

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Boyd, E.S., Hamilton, T.L., Wang, J., He, L. and Zhang, C.L. (2013) The role of tetraether lipid composition in the adaptation of thermophilic archaea to acidity. *Front. Microbiol.* 4, doi: 10.3389/fmicb.2013.00062.

Inskeep, W.P., Macur, R.E., Harrison, G., Bostick, B.C. and Fendorf, S. (2004) Biomineralization of As(V)-hydrous ferric oxyhydroxide in microbial mats of an acid-sulfate-chloride geothermal spring, Yellowstone National Park. *Geochim. Cosmochim. Acta* 68, 3141-3155.

Jackson, C.R., Langner, H.W., Donahoe-Christiansen, J., Inskeep, W.P. and McDermott, T.R. (2001) Molecular analysis of microbial community structure in an arsenite-oxidizing acidic thermal spring. *Environ. Microbiol.* 3, 532-542.

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Kozubal, M.A., Macur, R.E., Jay, Z.J., Beam, J.P., Malfatti, S.A., Tringe, S.G., Kocar, B.D., Borch, T. and Inskeep, W.P. (2012) Microbial cycling in acidic geothermal springs of Yellowstone National Park: integrating molecular surveys, geochemical processes, and isolation of novel Fe-active microorganisms. *Front. Microbiol.* 3, doi:10.3389/fmicb.2012.00109.

Macur, R.E., Langner, H.W., Kocar, B.D. and Inskeep, W.P. (2004) Linking geochemical processes with microbial community analysis: Successional dynamics in an arsenic-rich, acid-sulfate-chloride geothermal spring. *Geobiology* 2, 163-177.