

## Supplementary Information

### *Description of Study Sites*

Situated at the western end of the Taylor Glacier, Beacon Valley (BV, 77°48' S, 160°48' E) contains the oldest glacial ice on Earth (Schäfer et al. 2000). The ice layer, primarily localized to central Beacon Valley, rests below a 50 cm thick sublimation till composed of clasts of Ferrar Dolerite, Beacon Sandstone, and granite erratics foreign to Beacon Valley (Kowalewski et al. 2006). There is very limited sublimation from the buried glacial ice even during the austral summer (Kowalewski et al. 2006). The average air relative humidity (RH) content in the austral summer is around 35%, and the air temperature ranges from -16°C to 3.3°C, although ground-surface temperature can be 5-10°C higher (Kowalewski et al. 2006). These factors, in combination with Beacon Valley's high elevations (alt. ~1500 m) and exposure to katabatic winds coming from the nearby Polar Plateau, suggest extremely limited water availability for microorganisms. The sampling site, situated at the center of a very large sublimation polygon, is typical of the soil described above.

Battleship Promontory (BP, 76°54' S, 160°55' E) in the Convoy Range is by far the northern-most study site included in the study. Summer average relative humidity is around 54%, and rock surface temperature ranges from -14.2°C to 9.9°C (Wynn-Williams 2000). Battleship Promontory has long been considered a "hot spot" for endolithic life (Banerjee et al. 2000) and is thought to harbor considerable free-living cyanobacteria populations (Johnston & Vestal 1991; Wynn-Williams & Edwards 2000). Despite the relatively high altitude, transient ponds formed from snow melt water are frequently observed at Battleship Promontory. The sampling site, covering several sublimation polygons, is composed of weathered Beacon Sandstone beneath a layer of Ferrar Dolerite pebbles.

Upper Wright Valley (UW, 77°10' S, 161°50' E) is defined as the areas between

Wright Upper Glacier and Lake Vanda and comprises three distinct areas: the Labyrinth, the Dais, and the North and South forks bifurcated by the Dais (McLeod et al. 2009). The underlying geology of the area is Ferrar Dolerite and Beacon Sandstone, which are clearly visible along the sides of the valley. Air relative humidity and temperature are considered to be too low to produce significant soil moisture, and salt pans are a common occurrence in the area (McLeod et al. 2009). The sampling site is located in an elevated area north of the boundary between Wright Upper Glacier and the Labyrinth, where the soil is a mixture of Ferrar Dolerite and Beacon Sandstone derived from glacial till. The soil is likely to be relatively old due to the presence of locally weathered boulders and finely weathered surface material.

Miers Valley (MV, 78°60' S, 164°00' E) is the southern-most and the only coastal valley (alt. ~150 m) included in the study. Its predominant underlying geology is marble and granite, although most of the valley floor is covered by moraine of glacial and/or marine origin, as is typical of the eastern Dry Valleys (Bockheim & McLeod 2008). Due to its coastal location and low altitude, Miers Valley has higher precipitation than other valleys, and during the austral summer, glacial melt streams from Miers and Adams glaciers become highly active and form a hyporheic zone west of the Miers Lake. The sampling site is west of the Miers Lake and at least 250 m north of the clearly defined hyporheic zone, in an area dominated by moraine with very few large boulders.

#### *Detailed DNA Extraction Protocol*

0.8 g of soil was placed into a screw-cap microcentrifuge tube containing 0.1 mm and 2.5 mm silica-zirconia beads (0.5 g each) (BioSpec Products, Bartlesville, Oklahoma). 300  $\mu$ l of phosphate buffer (100 mM  $\text{NaH}_2\text{PO}_4$ ) and 300  $\mu$ l of SDS lysis buffer (100 mM NaCl, 500 mM Tris pH 8.0, and 10% SDS) were added to the tube, and the tube was shaken at 4.2  $\text{ms}^{-1}$

for 30 seconds on a MiniBeadBeater (Glen Mills Inc, Clifton, New Jersey). After centrifugation at 16,000 x g for 3 minutes, the aqueous phase was transferred to a new tube, and 200 µl of cetyltrimethylammonium bromide (CTAB) extraction buffer (100 mM Tris·HCl pH 8.0, 1.4 M NaCl, 25 mM EDTA, 2% hexadecyltrimethylammonium bromide, 1% polyvinylpyrrolidone, 0.4% v/v beta-mercaptoethanol) was added, followed by incubation at 60°C for 30 minutes. After centrifugation at 16,000 x g for 5 minutes, the aqueous phase (transferred to a new tube) was mixed with equal volume of chloroform:isoamyl alcohol (24:1) and incubated on a platform rocker at room temperature for 20 minutes. After centrifugation at 16,000 x g for 5 minutes, 7 M ammonium acetate was added to the aqueous phase in a new tube to a final concentration of 2.5 M, and the tube was centrifuged again at 16,000 x g for 5 minutes. The supernatant was transferred to a new tube, and 0.54 volumes of ice-cold isopropanol was added. After overnight incubation at -20°C, the precipitated DNA was centrifuged at 16,000 x g for 20 minutes, washed with ice-cold 70% ethanol, and air dried. The recovered DNA was resuspended in 20 µl TE buffer and quantified using a NanoDrop 1000 spectrophotometer (NanoDrop Products, Wilmington, Delaware). Procedural blanks were included at appropriate steps and yielded no DNA detectable by PCR.

#### 70 *PCR Components and Conditions and Quality Control Procedure for Community Fingerprinting Analysis*

Duplicate PCR of 50 µl was performed with 10 to 50 ng of template DNA per tube. The PCR components included: 1X Platinum *Taq* PCR buffer, 0.2 mM dNTPs, 20 µg/ml bovine serum albumin (BSA), 3 mM MgCl<sub>2</sub>, 1 unit of Platinum *Taq* (Invitrogen Inc., Carlsbad, California), 300 nM of each primer, and de-ionized water. All PCR runs were performed on a Bio-Rad DNA Engine (PTC-200, Bio-Rad Laboratories Inc, Hercules,

California), and the thermocycler program was as follows: 94°C for 2 minutes, followed by 30/35 cycles (bacterial/cyanobacterial ARISA) of 94°C for 45 seconds, 55°C for 30 seconds, 72°C for 2 minutes, and a final extension at 72°C for 7 minutes. Duplicate PCR products were  
80 combined and visualized on 1% agarose gel, then cleaned up using a QuickClean PCR Purification kit (GenScript Corporation, Piscataway, New Jersey). PCR amplicons were quantified using a NanoDrop 1000 spectrophotometer (NanoDrop Products, Wilmington, Delaware) and diluted 1:10 using de-ionized water.

#### 85 *Soil Sample Preparation for Elemental Analysis (ICP-MS)*

4 ml of concentrated HNO<sub>3</sub> (first diluted 1:1 using de-ionized water) and 10 ml of concentrated HCl (first diluted 1:4 using de-ionized water) were added to 1 g of soil in a covered beaker and incubated at room temperature for 30 minutes. The samples were then digested at 95°C for 30 minutes, cooled, and diluted to a final volume of 100 ml. Sub-samples  
90 (approx. 30 ml) were transferred to 50 ml Falcon tubes and centrifuged at 800 x g for 10 minutes. 1.6 ml of concentrated HNO<sub>3</sub> was added to 20 ml of supernatant and consequently diluted to a final volume of 100 ml, an aliquot of which was then filtered through a 0.45 µl filter into a 15 ml Falcon tube. The resulting samples were analyzed using an ELAN DRC II mass spectrometer (Perkin-Elmer Inc., Münster, Germany). A procedural blank containing no  
95 soil returned negligible readings.

#### *PCR Components and Conditions and Quality Control Procedure for Pyrosequencing of PCR Amplicons*

For each study site, triplicates of 30 µl PCR with 15 or 30 ng of pooled genomic DNA  
100 template per tube were used to reduce the effects of stochastic PCR bias (Wintzingerode et al. 1997). The PCR components included: 1X PrimerSTAR Buffer (includes 1 mM Mg<sup>2+</sup>), 0.2

mM dNTPs, 5 units of PrimeSTAR HD DNA Polymerase (Takara Bio Inc., Shiga, Japan), 400 nM of each primer, and de-ionized water. All PCR runs were performed on a Bio-Rad DNA Engine (Bio-Rad Laboratories Inc.), and the thermocycler program was as follows:  
105 94°C for 2 minutes, followed by 30 cycles of 94°C for 20 seconds, 55°C (decrease 0.2 per cycle) for 10 seconds, 72°C for 20 seconds, and a final extension at 72°C for 3 minutes. Triplicate PCR products were combined and run on a 2% agarose gel, which was stained with SYBR Gold Nucleic Acid Gel Stain (Invitrogen Inc.) and visualized on a Safe Imager Blue-Light Transilluminator (Invitrogen Inc.) to avoid UV-related damage to the amplicons. DNA  
110 bands of the correct size were extracted from the agarose gel and purified using a QuickClean Gel Extraction kit (GenScript Corporation). The resulting PCR amplicons were quantified using both a NanoDrop 1000 spectrophotometer and a QuBit Fluorometer (Invitrogen Inc.).

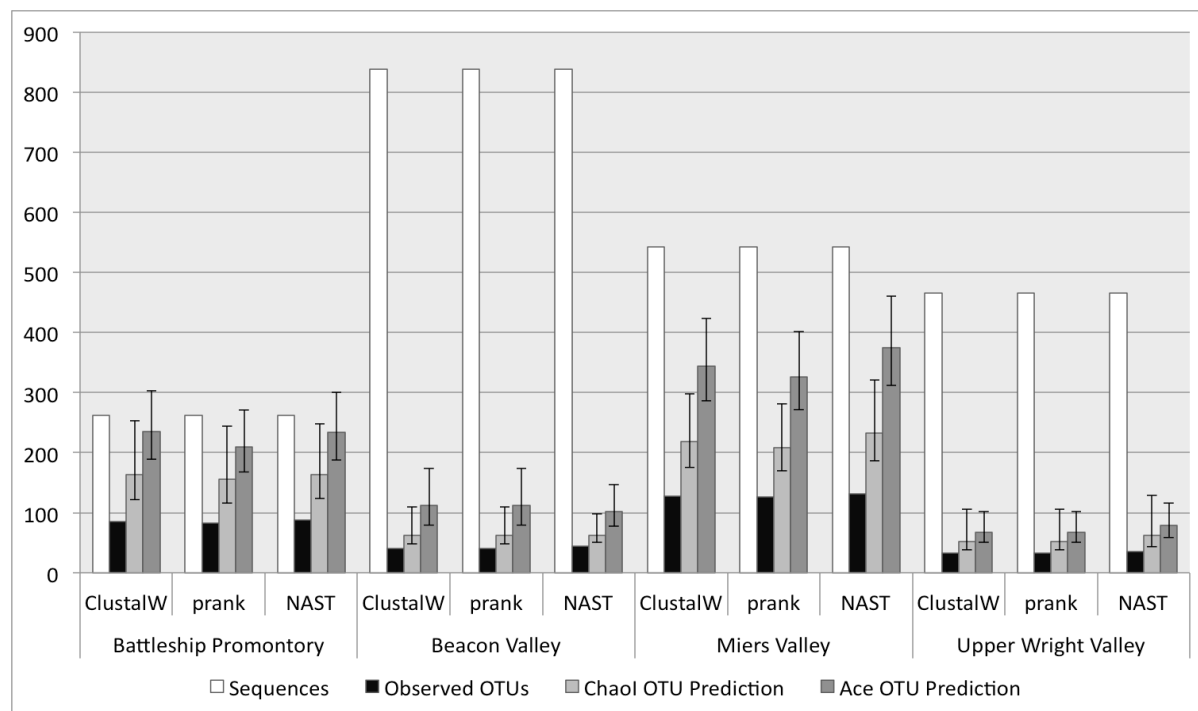
#### *Noise-Filtering and Binning of ARISA Run Data*

115 ARISA run data were processed using Genetic Profiler (Version 2, GE Healthcare) to yield all detectable peaks without applying any arbitrary cutoff. The resulting data was processed using a modified version of a previously described program (Abdo et al. 2006), which defined the background noise baseline through recursive analysis, and peaks within 6 S.D. of the noise baseline were removed. The remaining peaks were binned to counter size variance  
120 between ARISA runs. ARISA results from procedural blanks were used to remove background signals due to contamination or noise. For the FAM-labeled cyanobacterial ARISA samples, potential crosstalk AFLs from the ROX-labeled size standards were manually removed based on known ETR900-R sizes. All AFLs smaller than 120 bp for bacterial ARISA and 180 bp for cyanobacterial ARISA were removed since they were too  
125 short to contain true ITS regions (Wood et al. 2008).

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### Supplementary Figure 1: Comparison of Observed and Predicted Diversities from Multiple Sequence Alignment Algorithms

160 Non-redundant high quality reads were aligned using ClustalW, PRANK<sub>+F<sub>2</sub></sub>, and NAST. The numbers of raw reads are represented by white bars, numbers of observed OTUs<sub>0.03</sub> by black bars, ChaoI indices for OTUs<sub>0.03</sub> by light gray bars, and ACE indices for OTUs<sub>0.03</sub> by dark gray bars. Error bars correspond to standard errors for the alpha diversity indices.

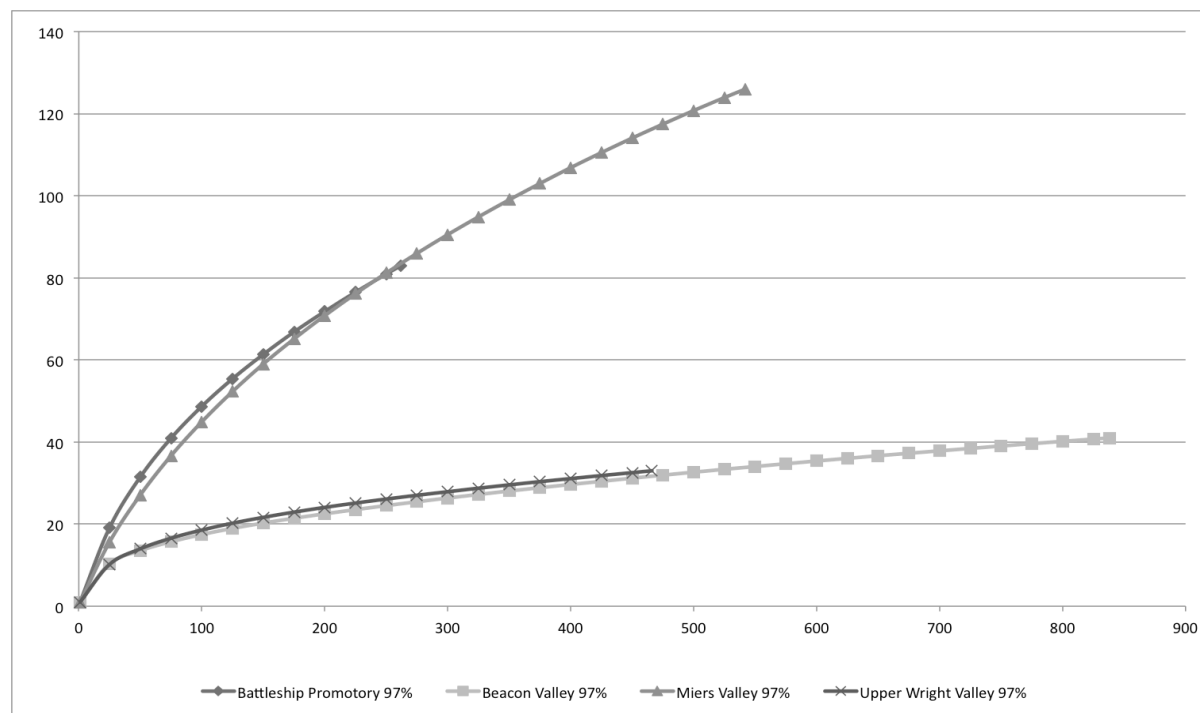


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Supplementary Figure 2: Rarefaction Curves for OTU<sub>S0.03</sub>

The rarefaction curves for OTU<sub>S0.03</sub> observed in the four study sites are as follows: Miers Valley, light gray triangles; Battleship Promontory, dark gray diamonds; Beacon Valley, light gray squares; Upper Wright Valley, light gray crosses.

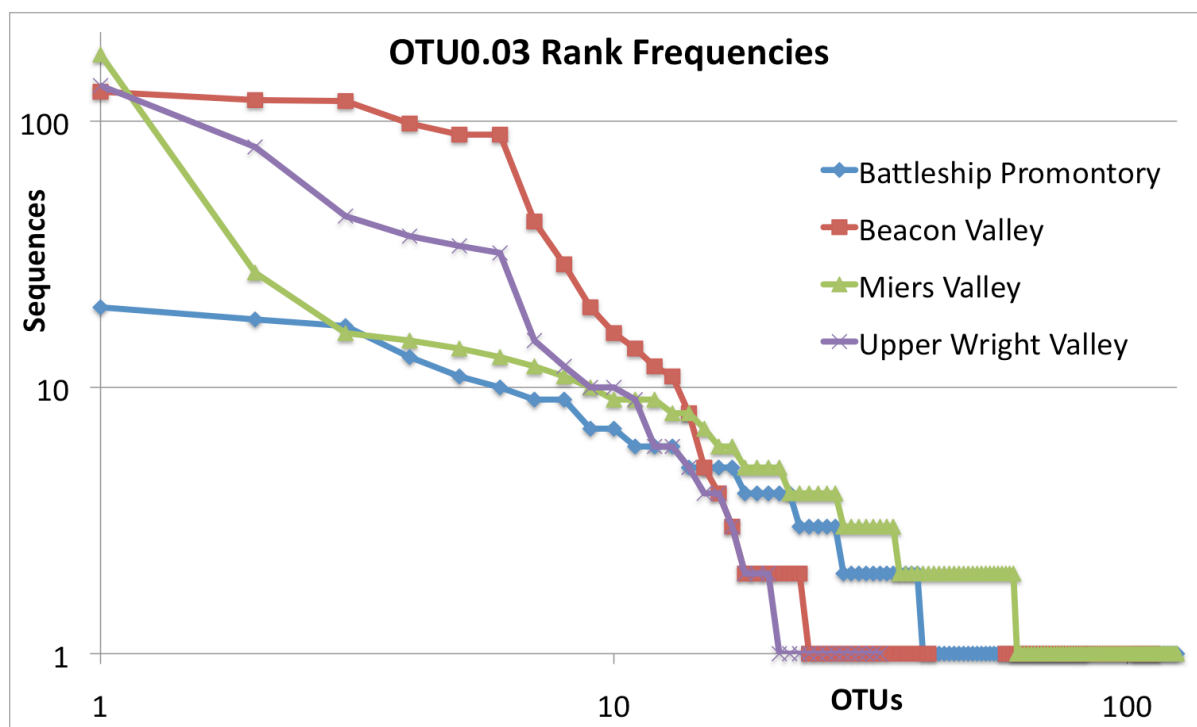
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Supplementary Figure 3: Rank-Abundance Distributions of OTU<sub>S0.03</sub>

175 The X axis ranks all OTU<sub>S0.03</sub> in order of abundance, and the Y axis represents the number of sequences in each OTU<sub>S0.03</sub>. Both axes are in logarithmic scale. The study sites are represented using the following symbols: Miers Valley, green triangles; Battleship Promontory, blue diamonds; Beacon Valley, red squares; Upper Wright Valley, purple crosses.

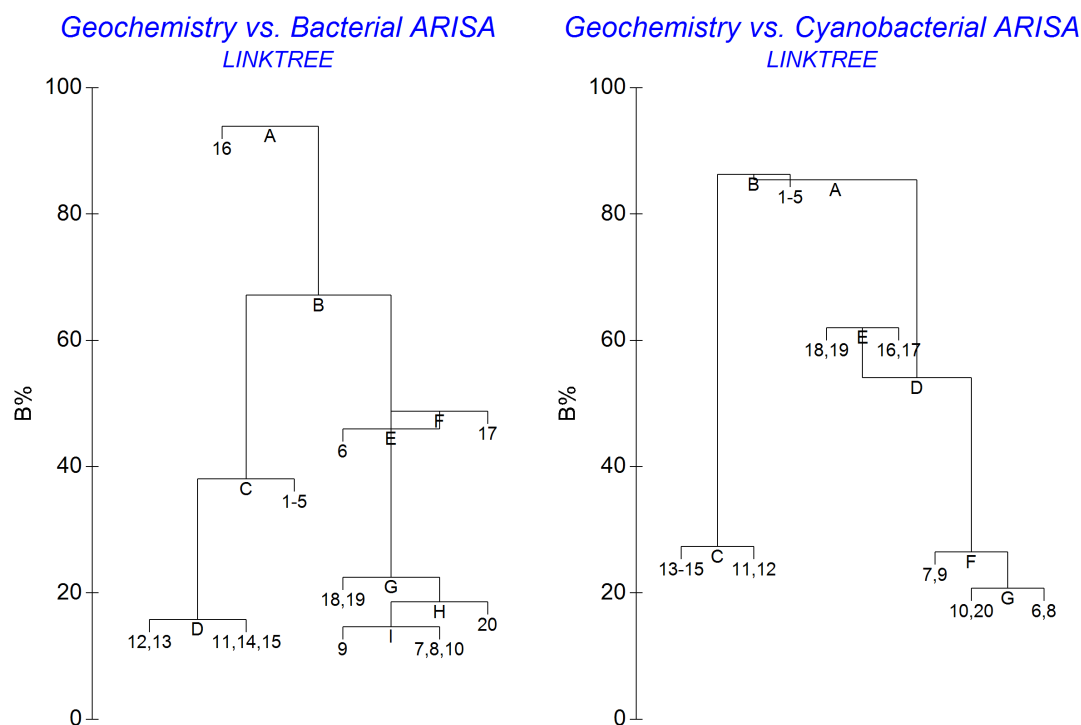


Supplementary Figure 4: LINKTREE Analysis of Bacteria (A) and Cyanobacterial (B) ARISA Patterns in Conjunction with Physicochemical Parameters

185 The LINKTREE analysis binary-splits the samples successively using a single most divisive environmental variable (although multiple variables may be equally divisive), which are listed in Supplementary Table 6. Samples within each group that cannot be split further (i.e.,  $p$  value greater than 0.05) are thus biologically and physicochemically similar. ARISA samples are presented as numbers in the trees: samples #1 through #5, MV\_A through MV-X; samples #6 through #10, BV\_A through BV\_X; samples #11 through #15: BP\_A through BP\_X; samples #16 through #20: UW\_A through UW-X.

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| A | B |
|---|---|
|---|---|



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## Supplementary Table 1: ANOSIM Analysis of Soil Geochemical Properties

Pairwise R-values for all geochemical properties shown in Table 1 are listed in the lower triangular, and pairwise R-values for ICP/MS values are listed in the upper triangular and in parentheses. p-values for all pairs are 0.008.

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|                       | Beacon Valley | Upper Wright Valley | Battleship Promontory | Miers Valley |
|-----------------------|---------------|---------------------|-----------------------|--------------|
| Beacon Valley         | X             | (0.524)             | (0.524)               | (0.812)      |
| Upper Wright Valley   | 0.564         | X                   | (0.712)               | (0.94)       |
| Battleship Promontory | 0.672         | 0.856               | X                     | (0.96)       |
| Miers Valley          | 0.956         | 0.996               | 0.98                  | X            |

Supplementary Table 2: Complete Soil Geochemical Properties

| (Element concentrations in ppb) | MV A     | MV B     | MV C     | MV D     | MV X     | BV A     | BV B     | BV C     | BV D     | BV X     |
|---------------------------------|----------|----------|----------|----------|----------|----------|----------|----------|----------|----------|
| pH                              | 8.9      | 8.8      | 8.1      | 8.6      | 8.7      | 6.9      | 7        | 7        | 7        | 7.6      |
| Conductivity (µS)               | 300      | 300      | 300      | 300      | 300      | 3920     | 3920     | 3920     | 3920     | 3920     |
| Gravimetric Water Content (%)   | 0.53%    |          | 0.52%    |          |          | 2.25%    |          | 2.47%    |          |          |
| % C                             | 0.27     | 0.46     | 0.47     | 0.41     | 0.70     | 0.11     | 0.20     | 0.17     | 0.11     | 0.11     |
| % N                             | 0.01     | 0.05     | 0.05     | 0.06     | 0.06     | 0.06     | 0.08     | 0.09     | 0.08     | 0.08     |
| C/N                             | 54.00    | 9.20     | 9.40     | 6.83     | 11.67    | 1.83     | 2.50     | 1.89     | 1.38     | 1.38     |
| Li                              | 18.92    | 11.95    | 15.63    | 20.04    | 18.96    | 12.55    | 10.59    | 20.31    | 12.67    | 14.47    |
| B                               | 1029.13  | 1080.90  | 1012.14  | 974.69   | 956.64   | 944.42   | 1010.81  | 1075.94  | 975.18   | 1062.91  |
| Na                              | 7615.44  | 5886.48  | 7362.35  | 9681.16  | 8291.59  | 3316.58  | 3219.94  | 6827.89  | 4354.56  | 4414.27  |
| Mg                              | 42724.53 | 29478.33 | 41811.32 | 48057.06 | 41376.36 | 9136.12  | 9283.76  | 14285.56 | 9670.57  | 10429.54 |
| Al                              | 24602.74 | 16071.96 | 22145.90 | 29281.29 | 25104.35 | 24230.63 | 18844.32 | 38396.40 | 25558.47 | 29333.16 |
| Si                              | 13739.24 | 9046.16  | 12929.20 | 17651.65 | 8573.35  | 2002.72  | 1913.98  | 1857.00  | 1858.11  | 1799.48  |
| P                               | 1314.75  | 1573.20  | 1724.62  | 2169.05  | 1971.41  | 1008.13  | 1237.78  | 1000.76  | 782.91   | 842.45   |
| K                               | 5375.36  | 3539.77  | 4525.19  | 6395.32  | 5950.87  | 1291.35  | 1349.68  | 3466.62  | 2095.38  | 2290.63  |
| Ca                              | 24663.46 | 16804.91 | 23436.45 | 30856.43 | 27604.81 | 17984.44 | 9099.23  | 21134.72 | 16395.50 | 16322.57 |
| Ti                              | 5674.47  | 3418.16  | 4754.81  | 6794.96  | 4918.48  | 842.92   | 356.82   | 1242.88  | 718.32   | 1008.47  |
| V                               | 78.49    | 54.80    | 76.50    | 98.62    | 83.52    | 122.37   | 82.77    | 215.15   | 148.25   | 210.56   |
| Cr                              | 53.31    | 36.87    | 55.93    | 64.10    | 48.99    | 8.66     | 6.94     | 16.36    | 11.02    | 11.85    |
| Fe                              | 45971.43 | 30961.85 | 43262.72 | 54330.99 | 47124.06 | 41521.27 | 40086.20 | 66271.29 | 43673.53 | 52303.92 |
| Mn                              | 742.19   | 518.72   | 727.17   | 892.11   | 787.33   | 417.19   | 323.34   | 584.29   | 379.91   | 421.56   |
| Co                              | 31.90    | 20.95    | 32.17    | 37.47    | 31.91    | 20.82    | 20.71    | 27.73    | 20.39    | 22.04    |
| Ni                              | 174.81   | 117.41   | 185.82   | 200.45   | 169.66   | 24.43    | 24.86    | 38.26    | 27.67    | 29.87    |
| Cu                              | 24.78    | 15.22    | 23.47    | 29.92    | 26.06    | 123.12   | 149.88   | 188.97   | 134.87   | 139.79   |
| Zn                              | 114.51   | 96.34    | 103.57   | 100.23   | 107.56   | 92.69    | 79.44    | 137.74   | 99.19    | 111.59   |
| As                              | 0.87     | 1.20     | 1.30     | 4.38     | 1.54     | 2.19     | 1.70     | 3.00     | 1.91     | 2.19     |
| Hf                              | 0.87     | 0.34     | 0.58     | 0.73     | 0.52     | 0.41     | 0.25     | 0.63     | 0.31     | 0.40     |
| Se                              | 0.62     | 0.91     | 1.06     | 1.33     | 0.88     | 1.20     | 1.31     | 1.95     | 1.41     | 2.03     |
| Sr                              | 328.09   | 196.06   | 299.73   | 415.31   | 366.36   | 60.07    | 41.93    | 94.54    | 74.76    | 80.94    |
| Zr                              | 35.65    | 16.32    | 29.68    | 41.96    | 30.11    | 14.68    | 8.16     | 22.60    | 12.43    | 14.40    |
| Ag                              | 0.03     | 0.00     | 0.00     | 0.10     | 0.00     | 0.00     | 0.00     | 0.14     | 0.03     | 0.05     |
| Cd                              | 0.29     | 0.18     | 0.23     | 0.29     | 0.27     | 0.13     | 0.14     | 0.31     | 0.25     | 0.20     |
| In                              | 0.04     | 0.02     | 0.02     | 0.04     | 0.03     | 0.02     | 0.01     | 0.04     | 0.02     | 0.03     |
| Ba                              | 150.30   | 95.38    | 133.86   | 200.44   | 172.51   | 26.18    | 19.65    | 115.24   | 73.71    | 40.06    |
| Hg                              | 0.06     | 0.04     | 0.03     | 0.05     | 0.03     | 0.03     | 0.03     | 0.10     | 0.04     | 0.04     |
| Tl                              | 0.10     | 0.04     | 0.07     | 0.11     | 0.11     | 0.04     | 0.05     | 0.11     | 0.09     | 0.08     |

|    |      |      |      |      |      |      |      |       |      |      |
|----|------|------|------|------|------|------|------|-------|------|------|
| Pb | 3.80 | 2.60 | 3.77 | 5.84 | 5.07 | 7.27 | 7.12 | 11.99 | 8.69 | 9.16 |
| Bi | 0.14 | 0.02 | 0.05 | 0.11 | 4.42 | 0.06 | 0.05 | 0.10  | 0.10 | 0.08 |
| U  | 1.01 | 0.85 | 1.04 | 1.56 | 1.37 | 1.01 | 0.96 | 1.42  | 0.96 | 1.12 |

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| (Element concentrations in ppb) | BP A     | BP B     | BP C     | BP D     | BP X     | UW A     | UW B     | UW C     | UW D     | UW X     |
|---------------------------------|----------|----------|----------|----------|----------|----------|----------|----------|----------|----------|
| pH                              | 7.9      | 7.7      | 7.2      | 7.2      | 8.4      | 6.9      | 7.1      | 7        | 6.9      | 6.9      |
| Conductivity (µS)               | 107      | 107      | 107      | 107      | 107      | 6130     | 6130     | 6130     | 6130     | 6130     |
| Gravimetric Water Content (%)   |          | 1.17%    |          | 1.10%    |          |          | 1.05%    |          | 1.10%    |          |
| % C                             | 0.10     | 0.10     | 0.09     | 0.10     | 0.10     | 0.10     | 0.11     | 0.13     | 0.11     | 0.10     |
| % N                             | 0.04     | 0.05     | 0.04     | 0.05     | 0.04     | 0.15     | 0.09     | 0.11     | 0.13     | 0.10     |
| C/N                             | 2.50     | 2.00     | 2.25     | 2.00     | 2.50     | 0.67     | 1.22     | 1.18     | 0.85     | 1.00     |
| Li                              | 10.28    | 7.37     | 7.56     | 7.87     | 8.62     | 12.70    | 13.13    | 9.82     | 15.23    | 16.01    |
| B                               | 1011.43  | 1066.09  | 1096.96  | 1014.18  | 1000.37  | 976.82   | 1060.32  | 1032.47  | 1043.78  | 1028.77  |
| Na                              | 3513.85  | 2709.95  | 2446.86  | 2831.78  | 2494.66  | 6920.16  | 4318.82  | 3800.44  | 7395.78  | 12718.43 |
| Mg                              | 6629.99  | 5155.96  | 4618.98  | 5494.16  | 5721.86  | 9631.34  | 7680.43  | 6344.37  | 8952.06  | 6800.97  |
| Al                              | 29820.62 | 23813.09 | 23162.74 | 24991.45 | 21661.85 | 19038.46 | 18678.92 | 13653.22 | 21619.86 | 27174.52 |
| Si                              | 1895.41  | 1797.64  | 1855.81  | 1610.42  | 1654.85  | 1534.66  | 2075.08  | 1851.26  | 2044.63  | 1938.68  |
| P                               | 892.97   | 744.50   | 616.47   | 620.59   | 722.81   | 636.18   | 593.10   | 370.90   | 503.58   | 483.16   |
| K                               | 1234.61  | 922.62   | 915.68   | 979.78   | 1077.94  | 2472.14  | 2700.96  | 2252.89  | 4000.77  | 7336.59  |
| Ca                              | 11552.11 | 10078.24 | 8940.83  | 9862.03  | 8631.41  | 5504.80  | 5041.14  | 2944.20  | 5521.00  | 8458.98  |
| Ti                              | 921.21   | 808.58   | 764.65   | 953.06   | 861.24   | 599.48   | 508.61   | 360.81   | 439.76   | 396.18   |
| V                               | 88.56    | 73.98    | 72.46    | 92.64    | 75.95    | 71.44    | 82.30    | 53.30    | 58.99    | 56.94    |
| Cr                              | 8.63     | 7.38     | 7.15     | 7.13     | 6.99     | 8.61     | 9.59     | 6.91     | 10.36    | 7.60     |
| Fe                              | 37354.94 | 30446.37 | 27730.78 | 30070.54 | 34114.71 | 29648.20 | 31110.01 | 21054.85 | 29985.89 | 26991.41 |
| Mn                              | 358.64   | 291.03   | 257.07   | 315.41   | 364.96   | 271.77   | 283.09   | 191.07   | 314.40   | 442.49   |
| Co                              | 19.59    | 14.95    | 13.05    | 15.60    | 17.39    | 14.79    | 14.31    | 9.51     | 13.61    | 10.41    |
| Ni                              | 27.75    | 22.05    | 19.16    | 22.34    | 21.27    | 19.82    | 19.75    | 14.50    | 20.30    | 16.39    |
| Cu                              | 121.75   | 96.28    | 77.57    | 98.08    | 104.83   | 78.02    | 73.48    | 52.64    | 70.78    | 55.87    |
| Zn                              | 68.52    | 57.30    | 49.62    | 68.56    | 72.41    | 65.35    | 73.76    | 48.12    | 69.05    | 66.79    |
| As                              | 1.40     | 1.28     | 0.69     | 1.13     | 1.33     | 1.95     | 1.77     | 2.16     | 2.18     | 1.71     |
| Hf                              | 0.33     | 0.27     | 0.20     | 0.28     | 0.31     | 0.31     | 0.38     | 0.29     | 0.45     | 0.51     |
| Se                              | 1.12     | 1.06     | 0.97     | 1.56     | 1.08     | 1.38     | 1.01     | 1.02     | 1.08     | 1.15     |
| Sr                              | 44.54    | 36.53    | 32.90    | 38.96    | 33.14    | 34.68    | 28.93    | 20.79    | 36.80    | 40.93    |
| Zr                              | 10.99    | 9.94     | 6.67     | 11.60    | 11.47    | 13.36    | 13.47    | 12.02    | 24.38    | 26.85    |
| Ag                              | 0.00     | 0.00     | 0.00     | 0.00     | 0.00     | 0.01     | 0.00     | 0.00     | 0.00     | 0.00     |
| Cd                              | 0.15     | 0.08     | 0.11     | 0.17     | 0.16     | 0.30     | 0.12     | 0.10     | 0.13     | 0.20     |
| In                              | 0.02     | 0.01     | 0.02     | 0.02     | 0.02     | 0.01     | 0.02     | 0.01     | 0.02     | 0.02     |
| Ba                              | 28.27    | 21.91    | 19.31    | 27.99    | 25.76    | 42.41    | 36.89    | 28.75    | 42.73    | 49.54    |
| Hg                              | 0.02     | 0.01     | 0.01     | 0.02     | 0.02     | 0.03     | 0.01     | 0.01     | 0.02     | 0.04     |
| Tl                              | 0.05     | 0.01     | 0.03     | 0.03     | 0.04     | 0.09     | 0.07     | 0.04     | 0.09     | 0.12     |
| Pb                              | 4.31     | 3.24     | 3.03     | 3.64     | 3.88     | 9.07     | 8.78     | 6.69     | 11.45    | 9.70     |

|    |      |      |      |      |      |      |      |      |      |      |
|----|------|------|------|------|------|------|------|------|------|------|
| Bi | 0.03 | 0.01 | 0.01 | 0.02 | 0.02 | 0.04 | 0.06 | 0.04 | 0.06 | 0.07 |
| U  | 1.10 | 0.85 | 0.78 | 0.87 | 0.87 | 0.83 | 0.91 | 0.60 | 1.12 | 1.79 |

Supplementary Table 3: Numbers of ARISA Fragment Length Groups (AFLs) Identified in Bacterial and Cyanobacterial Profiles

|                       | Total Bacterial AFL | Average Bacterial AFL±S.D. | Total Cyanobacterial AFL | Average Cyanobacterial AFL±S.D. |
|-----------------------|---------------------|----------------------------|--------------------------|---------------------------------|
| Beacon Valley         | 83                  | 16.6±4.0                   | 43                       | 8.6±2.9                         |
| Upper Wright Valley   | 98                  | 19.6±8.7                   | 24                       | 4.8±1.3                         |
| Battleship Promontory | 177                 | 35.4±9.4                   | 31                       | 6.2±4.0                         |
| Miers Valley          | 174                 | 34.8±5.1                   | 56                       | 11.2±4.2                        |

Supplementary Table 4: ANOSIM Analysis of Bacterial and Cyanobacterial ARISA Profiles

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Pairwise R-values for bacterial ARISA profiles are listed in the lower triangular, and pairwise R-values for cyanobacterial ARISA profiles are listed in the upper triangular and in parentheses. p-values for all pairs are 0.008, except for the BV-UW pair (0.024 for bacterial ARISA and 0.103 for cyanobacterial ARISA)

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|                       | Beacon Valley | Upper Wright Valley | Battleship Promontory | Miers Valley |
|-----------------------|---------------|---------------------|-----------------------|--------------|
| Beacon Valley         | X             | (0.216)             | (1)                   | (0.998)      |
| Upper Wright Valley   | 0.214         | X                   | (0.594)               | (0.752)      |
| Battleship Promontory | 0.958         | 0.596               | X                     | (0.99)       |
| Miers Valley          | 0.974         | 0.616               | 0.952                 | X            |

Supplementary Table 5: Biogeochemical Analysis

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The Biota-Environmental Stepwise (BEST) analysis was carried out to investigate potential links between physicochemical parameters (up to five) and ARISA patterns (both bacterial and cyanobacterial ARISA). p-values for both BEST analyses are 0.01. A Rho value (aka Spearman's rank correlation coefficient) of 1 indicates perfect correlation, and 0 no correlation.

| Geochem vs. Bacterial ARISA |                                      | Geochem vs. Cyanobacterial ARISA |  |
|-----------------------------|--------------------------------------|----------------------------------|--|
| Rho                         | Variables                            | Rho                              | Variables  |
| 0.567                       | Altitude, $\mu\text{S}$ <sup>1</sup> | 0.622                            | Altitude, $\mu\text{S}$ , Cu                                 |
| 0.560                       | Altitude, $\mu\text{S}$ , %N, Pb     | 0.623                            | Altitude, $\mu\text{S}$ , Cu, Ni                             |
| 0.560                       | $\mu\text{S}$ , pH, Ca, Pb           | 0.623                            | Altitude, $\mu\text{S}$ , $\text{W}_G$ <sup>2</sup> , Cu     |
| 0.563                       | Altitude, $\mu\text{S}$ , pH, Pb, B  | 0.623                            | Altitude, $\mu\text{S}$ , $\text{W}_G$ <sup>2</sup> , Cu, Ni |
| 0.560                       | Altitude, $\mu\text{S}$ , pH, Pb, Ca | 0.621                            | Altitude, $\mu\text{S}$ , Cu, Ni, Si                         |

<sup>1</sup>conductivity; <sup>2</sup>gravimetric water content.

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Supplementary Table 6: Environmental Variables Corresponding to LINKTREE Splits

230 Environmental variables that facilitate successful binary splits (i.e., p-value <0.05) in LINKTREE analysis (Supplementary Figure 4) are listed here. The soil physicochemical values shown here have been transformed and normalized as described in Material and methods and can only be interpreted in relative terms. Nodes and samples are represented using the same scheme as Supplementary Figure 4.

| Node->Split                                      | Variable   | LHS <sup>1</sup> (RHS) <sup>2</sup> Split   | $\pi^3$ | p value | R <sup>4</sup> | B% <sup>5</sup> |
|--|--|---|---------|---------|----------------|-----------------|
| <b>Bacterial ARISA (Supplementary Figure 4A)</b> |  |   |         |         |                |                 |
| A->(16), B                                       | %N<br>C/N<br>Si<br>W <sub>G</sub>  | >1.77 (<1.43)<br><-1.08 (>-0.93)<br><-0.765 (>-0.709)<br><-0.662 (>-0.64)   | 1.92    | 0.001   | 0.87           | 93.9%           |
| B->C, E  | $\mu$ S<br>Pb  | <-0.664 (>0.818)<br><-2.39E-2 (>0.265)  | 1.9     | 0.001   | 0.83           | 67.1%           |
| C->D, (1-5)                                      | W <sub>G</sub><br>Altitude<br>Mg<br>Si<br>Cr<br>Ni<br>Ba<br>Sr<br>K<br>%C<br>Ti<br>Cu<br>Na<br>Zn<br>C/N<br>Mn<br>P<br>Zr<br>Ca<br>$\mu$ S<br>Li<br>Hg<br>Co<br>Cd<br>Hf | <-0.623 (>1.68)<br>>0.421 (<-1.65)<br><-0.704 (>1.19)<br><-0.518 (>1.26)<br><-0.558 (>1.2)<br><-0.336 (>1.24)<br><-0.738 (>0.821)<br><-0.489 (>1.06)<br><-0.97 (>0.549)<br><-0.675 (>0.68)<br><-0.16 (>1.17)<br>>0.157 (<-1.15)<br><-0.681 (>0.37)<br><-0.387 (>0.6)<br><-0.128 (>0.784)<br><-0.264 (>0.573)<br><2.46E-2 (>0.815)<br><-0.658 (>-2.87E-3)<br><-3.03E-2 (>0.558)<br><-1.23 (>-0.664)<br><-0.644 (>-0.184)<br><-0.253 (>0.186)<br><5.2E-2 (>0.227)<br><-0.228 (>-9.61E-2)<br><-0.469 (>-0.375) | 2.58    | 0.001   | 0.95           | 38%             |
| D->(12,13), (11,14,15)                           | B<br>Cd<br>Zn<br>Hg<br>Pb<br>Ba<br>Mn<br>Zr<br>Li<br>Bi<br>U<br>Co<br>Se<br>K<br>Mg  | >1 (<-0.178)<br><-1.27 (>-0.441)<br><-1.19 (>-0.577)<br><-1.3 (>-0.787)<br><-1.21 (>-0.978)<br><-1.05 (>-0.853)<br><-0.799 (>-0.61)<br><-0.946 (>-0.758)<br><-1.56 (>-1.44)<br><-0.653 (>-0.541)<br><-0.721 (>-0.61)<br><-0.644 (>-0.536)<br><-0.388 (>-0.301)<br><-1.39 (>-1.3)<br><-1.02 (>-0.941)  | 0.31    | 0.005   | 0.58           | 15.8%           |



| Node->Split   | Variable   | LHS <sup>1</sup> (RHS) <sup>2</sup> Split  | $\pi^3$ | <i>p</i> value | R <sup>4</sup> | B% <sup>5</sup> |
|---|--|--|---------|----------------|----------------|-----------------|
|   | Hf<br>V<br>Ti<br>Cu  | <-0.938 (>-0.863)<br><-0.421 (>-0.355)<br><-0.329 (>-0.264)<br><0.464 (>0.491)   |         |                |                |                 |
| E->(6), F   | B<br>%N<br>Tl<br>K   | <-1.86 (>-1.1)<br><-0.148 (>0.385)<br><-0.791 (>-0.607)<br><-0.906 (>-0.842)   | 1.23    | 0.001          | 0.68           | 46%             |
| F->G, (17)  | Se<br>Si   | >-0.502 (<-0.533)<br><-0.429 (>-0.412)   | 1.34    | 0.001          | 0.82           | 48.8%           |
| G->(18,19), H   | Ca<br>Hg<br>Se<br>% N<br>Cd<br>Sr<br>Mn  | <-1.19 (>-0.519)<br><-0.572 (>-0.137)<br><-0.294 (>-8.2E-2)<br>>1.05 (<0.841)<br><-0.77 (>-0.569)<br><-0.679 (>-0.573)<br><-0.617 (>-0.551)  | 1.25    | 0.001          | 0.59           | 22.5%           |
| H->I, (20)  | Co<br>Fe<br>Na<br>Cu<br>K<br>U<br>P<br>V<br>Zn<br>Altitude<br>Se<br>Ni<br>Mg<br>Zr<br>$\mu$ S<br>C/N<br>%N<br>W <sub>G</sub><br>Tl<br>pH<br>Ca<br>%C<br>Sr<br>Si | >0.157 (<-1.55)<br>>0.306 (<-1.09)<br><0.672 (>1.94)<br>>0.95 (<-0.304)<br><0.519 (>1.61)<br><1.3 (>2.29)<br>>-0.243 (<-1.22)<br>>-0.14 (<-1.07)<br>>-6.83E-2 (<-0.664)<br>>0.879 (<0.351)<br>>0.397 (<-8.2E-2)<br>>-0.45 (<-0.872)<br>>-0.279 (<-0.672)<br><0.643 (>0.992)<br><0.818 (>1.08)<br>>-0.595 (<-0.819)<br><0.621 (>0.841)<br>>-0.431 (<-0.623)<br><1.18 (>1.36)<br>>-0.809 (<-0.959)<br>>-0.404 (<-0.519)<br>>-0.565 (<-0.675)<br>>-0.549 (<-0.573)<br><-0.507 (>-0.492) | 0.99    | 0.001          | 0.75           | 18.6%           |
| I->(9), (7,8,10)                                      | B<br>P<br>Cu<br>Co   | <-1.1 (>-0.256)<br><-0.243 (>-9.39E-2)<br><0.95 (>1)<br><0.157 (>0.197)  | 1.22    | 0.001          | 1              | 14.6%           |
| <b>Cyanobacterial ARISA (Supplementary Figure 4B)</b> |  |  |         |                |                |                 |
| A->B, D   | $\mu$ S<br>Pb  | <-0.664 (>0.818)<br><-2.39E-2 (>0.265)   | 1.91    | 0.001          | 0.54           | 85.4%           |
| B->C, (1-5)   | W <sub>G</sub><br>Altitude<br>Mg<br>Si<br>Cr<br>Ni<br>Ba<br>Sr<br>K<br>%C<br>Ti  | <-0.623 (>1.68)<br>>0.421 (<-1.65)<br><-0.704 (>1.19)<br><-0.518 (>1.26)<br><-0.558 (>1.2)<br><-0.336 (>1.24)<br><-0.738 (>0.821)<br><-0.489 (>1.06)<br><-0.97 (>0.549)<br><-0.675 (>0.68)<br><-0.16 (>1.17)   | 2.58    | 0.001          | 0.99           | 86.3%           |

| Node->Split         | Variable | LHS <sup>1</sup> (RHS) <sup>2</sup> Split | $\pi^3$ | <i>p</i> value | R <sup>4</sup> | B% <sup>5</sup> |
|---------------------|----------|---|---------|----------------|----------------|-----------------|
|                     | Cu       | >0.157 (<-1.15)                           |         |                |                |                 |
|                     | Na       | <-0.681 (>0.37)                           |         |                |                |                 |
|                     | Zn       | <-0.387 (>0.6)                            |         |                |                |                 |
|                     | C/N      | <-0.128 (>0.784)                          |         |                |                |                 |
|                     | Mn       | <-0.264 (>0.573)                          |         |                |                |                 |
|                     | P        | <2.46E-2 (>0.815)                         |         |                |                |                 |
|                     | Zr       | <-0.658 (>-2.87E-3)                       |         |                |                |                 |
|                     | Ca       | <-3.03E-2 (>0.558)                        |         |                |                |                 |
|                     | $\mu$ S  | <-1.23 (>-0.664)                          |         |                |                |                 |
|                     | Li       | <-0.644 (>-0.184)                         |         |                |                |                 |
|                     | Hg       | <-0.253 (>0.186)                          |         |                |                |                 |
|                     | Co       | <5.2E-2 (>0.227)                          |         |                |                |                 |
|                     | Cd       | <-0.228 (>-9.61E-2)                       |         |                |                |                 |
|                     | Hf       | <-0.469 (>-0.375)                         |         |                |                |                 |
| C->(13-15), (11,12) | P        | <-0.405 (>-0.345)                         | 0.32    | 0.004          | 0.83           | 27.3%           |
|                     | Cr       | <-0.767 (>-0.732)                         |         |                |                |                 |
|                     | Ca       | <-0.278 (>-0.244)                         |         |                |                |                 |
| D->E, F             | Ca       | <-1.19 (>-0.519)                          | 1.2     | 0.001          | 0.49           | 54.1%           |
|                     | Sr       | <-0.679 (>-0.573)                         |         |                |                |                 |
|                     | Mn       | <-0.617 (>-0.551)                         |         |                |                |                 |
| E->(18,19), (16,17) | V        | <-0.981 (>-0.507)                         | 0.44    | 0.012          | 1              | 62%             |
|                     | P        | <-1.14 (>-0.806)                          |         |                |                |                 |
|                     | As       | >0.626 (<0.367)                           |         |                |                |                 |
|                     | Ti       | <-0.952 (>-0.804)                         |         |                |                |                 |
|                     | Co       | <-0.881 (>-0.754)                         |         |                |                |                 |
|                     | Cu       | <2.76E-2 (>8.03E-2)                       |         |                |                |                 |
| F->(7,9), G         | Hf       | <-0.588 (>1.66E-2)                        | 1.06    | 0.001          | 0.57           | 26.5%           |
|                     | Zr       | <-0.527 (>-0.245)                         |         |                |                |                 |
|                     | Mn       | <-0.169 (>5.33E-2)                        |         |                |                |                 |
|                     | U        | <-0.235 (>-5.75E-2)                       |         |                |                |                 |
| G->(10,20),(6,8)    | P        | <-9.39E-2 (>0.257)                        | 0.8     | 0.001          | 1              | 20.7%           |
|                     | C/N      | <-0.595 (>-0.378)                         |         |                |                |                 |
|                     | Ca       | <0.512 (>0.665)                           |         |                |                |                 |

235 <sup>1</sup>LHS: Left-Half Split, properties of the group to the left of the node; <sup>2</sup>RHS: Right-Half Split, properties of the group to the right of the node; <sup>3</sup> $\pi$ : the statistic  $\pi$ , which is defined as the absolute deviation of the real profile from the mean of all permuted profiles and constitutes the metric for formal hypothesis testing; <sup>4</sup>R: Spearman Rho value; <sup>5</sup>B%: an absolute measure of group differences. A lower value indicates similar samples.

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