Supplementary Material

Temporal Dynamics of Air Bacterial Communities in a University Health Centre using Illumina MiSeq Sequencing

Maneet Kumar Chakrawarti, Madhuri Singh, Vijay Pal Yadav, Kasturi Mukhopadhyay

*Correspondence author:
Prof. Kasturi Mukhopadhyay

School of Environmental Sciences, Jawaharlal Nehru University, New Delhi, India-110067

Phone: 91-11-26704307

E-mail: kasturim@mail.jnu.ac.in
**Figure S1:** Map of JNU Health Centre (28.54 N, 77.16 E).

**Figure S2:** Rarefaction analysis of bacterial communities in air samples collected from indoor and outdoor area of JNU Health Centre in each season: Monsoon Indoor (MI), Monsoon Outdoor (MO), Spring Indoor (SPI), Spring Outdoor (SPO), Summer Indoor (SUI), Summer Outdoor (SUO), Winter Indoor (WI), and Winter Outdoor (WO).

**Table S1:** Meteorological conditions, PM$_{2.5}$ and PM$_{10}$ concentration during air sampling at JNU Health Centre in each season: Spring Indoor (SPI), Spring Outdoor (SPO), Monsoon Indoor (MI), Monsoon Outdoor (MO), Winter Indoor (WI), Winter Outdoor (WO), Summer Indoor (SUI), Summer Outdoor (SUO).

**Table S2:** SRCCs between dominant phyla of bacterial community and Temperature (T), Relative Humidity (RH), PM$_{2.5}$ and PM$_{10}$.

**Table S3:** Diversity and richness estimation of 16S rRNA gene libraries of air samples collected from indoor and outdoor area of JNU Health Centre in each season: Spring Indoor (SPI), Spring Outdoor (SPO), Monsoon Indoor (MI), Monsoon Outdoor (MO), Winter Indoor (WI), Winter Outdoor (WO), Summer Indoor (SUI), Summer Outdoor (SUO) using Illumina MiSeq PE300 sequencing.

**Table S4:** Numbers of taxa classified at different taxonomic levels in air samples collected from indoor and outdoor area of JNU Health Centre in each season: Spring Indoor (SPI), Spring Outdoor (SPO), Monsoon Indoor (MI), Monsoon Outdoor (MO), Winter Indoor (WI), Winter Outdoor (WO), Summer Indoor (SUI), Summer Outdoor (SUO).

**Table S5:** The proportions of unclassified taxa reported in air samples collected from indoor and outdoor area of JNU Health Centre in each season: Spring Indoor (SPI), Spring Outdoor (SPO), Monsoon Indoor (MI), Monsoon Outdoor (MO), Winter Indoor (WI), Winter Outdoor (WO), Summer Indoor (SUI), Summer Outdoor (SUO) at different taxonomic levels.

**Table S6:** Raw data of ESKAPE pathogens determined in the total sequences acquired in each season: Monsoon Indoor (MI), Monsoon Outdoor (MO), Spring Indoor (SPI), Spring Outdoor (SPO), Summer Indoor (SUI), Summer Outdoor (SUO), Winter Indoor (WI), Winter Outdoor (WO).

**Table S7:** Inter-phyla correlations using Spearman’s Rank Correlation Coefficient (SRCC)
Figure S1: Map of JNU Health Centre (28.54 N, 77.16 E).
Figure S2: Rarefaction analysis of bacterial communities in air samples collected from indoor and outdoor areas of JNU Health Centre in each season: Monsoon Indoor (MI), Monsoon Outdoor (MO), Spring Indoor (SPI), Spring Outdoor (SPO), Summer Indoor (SUI), Summer Outdoor (SUO), Winter Indoor (WI), and Winter Outdoor (WO).
Table S1: Meteorological conditions, PM$_{2.5}$ and PM$_{10}$ concentration during air sampling at JNU Health Centre in each season: Spring Indoor (SPI), Spring Outdoor (SPO), Monsoon Indoor (MI), Monsoon Outdoor (MO), Winter Indoor (WI), Winter Outdoor (WO), Summer Indoor (SUI), Summer Outdoor (SUO).

| Sample ID | Date of Sampling | Temperature (°C) | Relative Humidity (%) | PM$_{2.5}$ (µg m$^{-3}$) | PM$_{10}$ (µg m$^{-3}$) |
|-----------|------------------|------------------|-----------------------|--------------------------|-------------------------|
| SPI       | 23/Feb/2018      | 25.1             | 51.8                  | 156.76                   | 560.10                  |
| SPO       | 23/Feb/2018      | 27.9             | 39.7                  | NA*                     | NA*                     |
| MI        | 12/Sept/2018     | 29.8             | 67.5                  | 13.53                   | 80.17                   |
| MO        | 12/Sept/2018     | 30.5             | 70.0                  | 46.82                   | 201.06                  |
| WI        | 03/Jan/2019      | 17.6             | 58.7                  | 477.6                   | 739.02                  |
| WO        | 03/Jan/2019      | 17.2             | 54.8                  | 326.38                  | 604.30                  |
| SUI       | 08/Jun/2019      | 35.4             | 33.6                  | 46.32                   | 254.92                  |
| SUO       | 08/Jun/2019      | 40.9             | 19.9                  | 43.99                   | 377.15                  |

*NA= Not Available
Table S2: SRCCs between dominant phyla of bacterial community and Temperature (T), Relative Humidity (RH), PM$_{2.5}$ and PM$_{10}$.

| Variables | Proteobacteria | Actinobacteria | Firmicutes | Bacteroidetes | Planctomycetes | Verrucomicrobia | Themi | Acidobacteria | TM7 | Others |
|-----------|----------------|----------------|------------|---------------|----------------|----------------|-------|---------------|-----|--------|
| **T**     |                |                |            |               |                |                |       |               |     |        |
| SRCC      | -0.548         | .762*          | 0.643      | -0.143        | 0.571          | 0.571          | 0.619 | 0.214         | 0.571| 0.357   |
| p values  | 0.160          | 0.028          | 0.086      | 0.736         | 0.139          | 0.139          | 0.102 | 0.610         | 0.139| 0.385   |
| **RH**    |                |                |            |               |                |                |       |               |     |        |
| SRCC      | 0.000          | 0.143          | 0.024      | 0.000         | -0.238         | -0.238         | -0.286| -0.619        | -0.310| -0.548  |
| p values  | 1.000          | 0.736          | 0.955      | 1.000         | 0.570          | 0.570          | 0.493 | 0.102         | 0.456| 0.160   |
| **PM2.5** |                |                |            |               |                |                |       |               |     |        |
| SRCC      | .857*          | -.893**        | -.893**    | 0.357         | -0.250         | -0.250         | -0.286| 0.107         | -0.107| 0.071   |
| p values  | 0.014          | 0.007          | 0.007      | 0.432         | 0.589          | 0.589          | 0.535 | 0.819         | 0.819| 0.879   |
| **PM10**  |                |                |            |               |                |                |       |               |     |        |
| SRCC      | 0.714          | -.964**        | -0.750     | 0.143         | -0.179         | -0.179         | -0.143| 0.393         | 0.036| 0.357   |
| p values  | 0.071          | 0.000          | 0.052      | 0.760         | 0.702          | 0.702          | 0.760 | 0.383         | 0.939| 0.432   |

* Correlation is significant at the 0.05 level (2-tailed).

** Correlation is significant at the 0.01 level (2-tailed).
Table S3: Diversity and richness estimation of 16S rRNA gene libraries of air samples collected from indoor and outdoor area of JNU Health Centre in each season: Spring Indoor (SPI), Spring Outdoor (SPO), Monsoon Indoor (MI), Monsoon Outdoor (MO), Winter Indoor (WI), Winter Outdoor (WO), Summer Indoor (SUI), Summer Outdoor (SUO) using Illumina MiSeq PE300 sequencing.

| Sample ID | OTUs  | Shannon | Simpson | Chao1  | Equitability |
|-----------|-------|---------|---------|--------|--------------|
| SPI       | 1239  | 4.7     | 0.90    | 1934.2 | 0.46         |
| SPO       | 1075  | 3.2     | 0.66    | 1671.4 | 0.31         |
| MI        | 822   | 3.7     | 0.83    | 1198.9 | 0.39         |
| MO        | 1185  | 6.0     | 0.96    | 1694.0 | 0.58         |
| WI        | 1702  | 5.0     | 0.89    | 2413.0 | 0.46         |
| WO        | 1818  | 4.6     | 0.83    | 2643.5 | 0.42         |
| SUI       | 3253  | 7.7     | 0.94    | 3835.9 | 0.66         |
| SUO       | 3606  | 9.1     | 0.99    | 3879.2 | 0.77         |
Table S4: Numbers of taxa classified at different taxonomic levels in air samples collected from indoor and outdoor area of JNU Health Centre in each season: Spring Indoor (SPI), Spring Outdoor (SPO), Monsoon Indoor (MI), Monsoon Outdoor (MO), Winter Indoor (WI), Winter Outdoor (WO), Summer Indoor (SUI), Summer Outdoor (SUO).

| Taxonomic level | Total | SPI | SPO | MI | MO | WI | WO | SUI | SUO |
|-----------------|-------|-----|-----|----|----|----|----|-----|-----|
| Phylum          | 34    | 17  | 19  | 16 | 22 | 21 | 21 | 26  | 27  |
| Class           | 96    | 47  | 50  | 39 | 49 | 58 | 55 | 74  | 78  |
| Order           | 182   | 77  | 82  | 56 | 85 | 101| 106| 144 | 146 |
| Family          | 339   | 170 | 165 | 115| 178| 209| 211| 281 | 283 |
| Genus           | 749   | 315 | 302 | 220| 340| 404| 412| 578 | 604 |
Table S5: The proportions of unclassified taxa reported in air samples collected from indoor and outdoor area of JNU Health Centre in each season: Spring Indoor (SPI), Spring Outdoor (SPO), Monsoon Indoor (MI), Monsoon Outdoor (MO), Winter Indoor (WI), Winter Outdoor (WO), Summer Indoor (SUI), Summer Outdoor (SUO) at different taxonomic levels.

| Taxonomic level | SPI   | SPO   | MI    | MO    | WI    | WO    | SUI   | SUO   |
|-----------------|-------|-------|-------|-------|-------|-------|-------|-------|
| Phylum          | 0     | 0     | 0     | 0     | 0     | 0     | 0     | 0     |
| Class           | 0.003 | 0.007 | 0.008 | 0.063 | 0.007 | 0.018 | 0.070 | 0.162 |
| Order           | 0.055 | 0.045 | 0.047 | 0.598 | 0.247 | 0.237 | 1.269 | 1.559 |
| Family          | 0.847 | 0.795 | 0.986 | 3.727 | 1.990 | 2.363 | 8.813 | 9.353 |
| Genus           | 18.581| 10.789| 21.404| 35.373| 43.634| 18.675| 29.055| 30.677|
| Species         | 79.779| 35.240| 96.849| 77.278| 61.462| 52.551| 65.149| 90.191|

*Sequences that could not be assigned to any known group were assigned as unclassified.
Table S6: Raw data of ESKAPE pathogens determined in the total sequences acquired in each season: Monsoon Indoor (MI), Monsoon Outdoor (MO), Spring Indoor (SPI), Spring Outdoor (SPO), Summer Indoor (SUI), Summer Outdoor (SUO), Winter Indoor (WI), Winter Outdoor (WO).

| Species                    | MI  | MO  | SPI | SPO | SUI | SUO | WI  | WO  |
|----------------------------|-----|-----|-----|-----|-----|-----|-----|-----|
| Enterococcus cecorum       | 0   | 0   | 0   | 0   | 0   | 3   | 3   | 0   |
| Enterococcus casseliflavus | 0   | 0   | 0   | 0   | 0   | 3   | 1   | 0   |
| Enterococcus asini         | 0   | 2   | 0   | 0   | 0   | 2   | 5   | 7   |
| Enterococcus unclassified  | 0   | 9   | 1   | 0   | 7   | 169 | 52  | 26  |
| Staphylococcus unclassified| 17  | 60  | 44  | 54  | 35  | 2814| 88  | 52  |
| Staphylococcus sciuri      | 5   | 49  | 1   | 0   | 1   | 212 | 1   | 1   |
| Staphylococcus epidermidis | 2   | 3   | 0   | 0   | 2   | 44  | 1   | 0   |
| Staphylococcus pettenkoferi| 0   | 0   | 0   | 0   | 0   | 7   | 0   | 0   |
| Staphylococcus equorum     | 0   | 0   | 1   | 1   | 0   | 25  | 18  | 22  |
| Staphylococcus aureus      | 0   | 0   | 1   | 1   | 0   | 6   | 1   | 2   |
| Staphylococcus saprophyticus| 0   | 0   | 0   | 0   | 0   | 8   | 0   | 0   |
| Klebsiella unclassified    | 3   | 0   | 1   | 1   | 9   | 288 | 0   | 0   |
| Acinetobacter unclassified | 164 | 51  | 21  | 8   | 359 | 3359| 18  | 42  |
| Acinetobacter schindleri   | 21  | 1   | 5   | 0   | 47  | 581 | 2   | 0   |
| Acinetobacter johnsonii    | 3   | 0   | 1   | 4   | 8   | 85  | 4   | 4   |
| Acinetobacter gillouiae    | 2   | 3   | 0   | 0   | 3   | 0   | 1   | 1   |
| Acinetobacter lwoffii      | 1   | 10  | 33  | 7   | 63  | 102 | 3   | 5   |
| Acinetobacter rhizosphaerae| 0   | 0   | 1   | 0   | 2   | 13  | 0   | 0   |
| Pseudomonas unclassified   | 10065| 1953| 148 | 13 | 58  | 361 | 26  | 7   |
| Pseudomonas pseudoalcaligenes| 23   | 56  | 0   | 0   | 1   | 2   | 0   | 0   |
| Pseudomonas stutzeri       | 9   | 10  | 3   | 0   | 1   | 4   | 0   | 0   |
| Pseudomonas viridiflava    | 4   | 2   | 0   | 0   | 0   | 0   | 0   | 0   |
| Pseudomonas alcaligenes    | 4   | 2   | 0   | 0   | 2   | 48  | 0   | 0   |
| Pseudomonas veronii        | 0   | 0   | 0   | 0   | 0   | 0   | 0   | 2   |
| Pseudomonas nitroreducens  | 0   | 0   | 0   | 0   | 4   | 6   | 0   | 0   |
| Pseudomonas fragi          | 0   | 0   | 0   | 0   | 0   | 0   | 1   | 1   |
| Enterobacter turicensis    | 0   | 0   | 0   | 0   | 0   | 4   | 0   | 0   |
| Enterobacter gergoviae     | 0   | 0   | 0   | 0   | 2   | 6   | 0   | 0   |
| Enterobacter cowanii       | 0   | 0   | 0   | 1   | 0   | 3   | 0   | 0   |
| Enterobacter cloacae       | 0   | 0   | 0   | 0   | 1   | 5   | 0   | 0   |
| Enterobacter unclassified  | 0   | 0   | 0   | 0   | 4   | 6   | 0   | 0   |
| Total Pathogenic Sequences | 10323| 2211| 261 | 90 | 606 | 8169| 224 | 172 |
Table S7: Inter-phyla correlations using Spearman’s Rank Correlation Coefficient (SRCC)

| Variables   | Proteobacteria | Actinobacteria | Firmicutes | Bacteroidetes | Planctomycetes | Verrucomicrobia | Thermi | Acidobacteria | TM7 | Others |
|-------------|----------------|----------------|------------|---------------|----------------|----------------|--------|---------------|-----|--------|
| Proteobacteria | SRCC 1.000 | -0.762* | -0.595 | 0.048 | -0.143 | -0.143 | -0.119 | 0.167 | -0.119 | 0.262 |
| p values     | 0.028 | 0.120 | 0.911 | 0.736 | 0.736 | 0.779 | 0.693 | 0.779 | 0.531 |
| Actinobacteria | SRCC -0.762* | 1.000 | 0.738* | -0.190 | 0.238 | 0.238 | 0.286 | -0.333 | 0.119 | -0.214 |
| p values     | 0.028 | 0.037 | 0.651 | 0.570 | 0.570 | 0.493 | 0.420 | 0.779 | 0.610 |
| Firmicutes   | SRCC -0.595 | 0.738* | 1.000 | -0.524 | 0.595 | 0.595 | 0.643 | 0.119 | 0.429 | 0.238 |
| p values     | 0.120 | 0.037 | 0.183 | 0.120 | 0.120 | 0.086 | 0.779 | 0.289 | 0.570 |
| Bacteroidetes | SRCC 0.048 | -0.190 | -0.524 | 1.000 | 0.048 | 0.048 | -0.095 | 0.167 | 0.071 | -0.095 |
| p values     | 0.911 | 0.651 | 0.183 | 0.911 | 0.911 | 0.823 | 0.693 | 0.867 | 0.823 |
| Planctomycetes | SRCC -0.143 | 0.238 | 0.595 | 0.048 | 1.000 | 1.000** | .952** | .762* | .929** | .786* |
| p values     | 0.736 | 0.570 | 0.120 | 0.911 | 0.000 | 0.028 | 0.001 | 0.021 |
| Verrucomicrobia | SRCC -0.143 | 0.238 | 0.595 | 0.048 | 1.000** | 1.000 | .952** | .762* | .929** | .786* |
| p values     | 0.736 | 0.570 | 0.120 | 0.911 | 0.000 | 0.028 | 0.001 | 0.021 |
| Thermi       | SRCC -0.119 | 0.286 | 0.643 | -0.095 | .952** | .952** | 1.000 | 0.690 | .905** | .833* |
| p values     | 0.779 | 0.493 | 0.086 | 0.823 | 0.000 | 0.000 | 0.058 | 0.002 | 0.010 |
| Acidobacteria | SRCC 0.167 | -0.333 | 0.119 | 0.167 | .762* | .762* | 0.690 | 1.000 | .786* | .881** |
| p values     | 0.693 | 0.420 | 0.779 | 0.693 | 0.028 | 0.028 | 0.058 | 0.021 | 0.004 |
| TM7          | SRCC -0.119 | 0.119 | 0.429 | 0.071 | .929** | .929** | .905** | .786* | 1.000 | .857** |
| p values     | 0.779 | 0.779 | 0.289 | 0.867 | 0.001 | 0.001 | 0.002 | 0.021 | 0.007 |
| Others       | SRCC 0.262 | -0.214 | 0.238 | -0.095 | .786* | .786* | .833* | .881** | .857** | 1.000 |
| p values     | 0.531 | 0.610 | 0.570 | 0.823 | 0.021 | 0.021 | 0.010 | 0.004 | 0.007 |

*. Correlation is significant at the 0.05 level (2-tailed).
**. Correlation is significant at the 0.01 level (2-tailed).