**Supplementary Information**

**Additional file 1**

**Title: Analysis of microbial populations in plastic–soil systems after exposure to high poly(butylene succinateco-adipate) load using high-resolution molecular technique**

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**Appendix 1.** PBSA chemical and physical properties

Total C, H and N concentrations were determined with an Elementar Vario EL III elemental analyzer (Elementar Analysensysteme GmbH, Hanau, Germany). Physical properties are provided in BioPBS™ FD92PM / FD92PB Technical Data Sheet, PTT MCC Biochem Company Limited.

|  |  |  |  |
| --- | --- | --- | --- |
| **Sample type** | **Total C (%)** | **H (%)** | **Total N (%)** |
| PBSA | 56.940 | 7.371 | 0.000 |
|  |  |  |  |
| **Properties Test** | **Test method** | **Unit** | **FD92PM / FD92PB** |
| Density | ISO 1183 | g/cm3 | 1.24 |
| Melt index flow rate (190°C, 2.16 kg) | ISO 1133 | g/ 10 min | 4 |
| Melting point | ISO 3146 | °C | 84 |
| Tensile modulus | MD | ISO 527-3 | MPa | 280 |
| TD | 320 |
| Yield stress | MD | ISO 527-3 | MPa | 18 |
| TD | 17 |
| Stress at break | MD | ISO 527-3 | MPa | 32 |
| TD | 27 |
| Strain at break | MD | ISO 527-3 | % | 600 |
| TD | 580 |
| Elmendorf tear strength | MD | ISO 6383-2 | N/mm | 2 |
| TD | 5 |
| Puncture impact |  | PTTMCCmethod | kJ/m | 6 |

**Figure S1.** Rarefaction curves of archaea (a), bacteria (b), and fungi (c).



**Figure S2**. Links between soil pH and microbial richness: a) soil pH values of initial soil, control S, control SN, initial-N soil, and soils in PS, and PSN treatment on 90 days. Correlations between soil pH and richness of archaea (b), bacteria (c) and fungi (d).



**Figure S3**. Factors explain variations in microbial community composition: percent explainable variations of archaeal (a), bacterial (b), and fungal (c) communities explained by PBSA addition, N fertilization, soil pH, and combinations of these factors. Total variations of archaeal, bacterial, and fungal communities explained by PBSA addition, N fertilization, soil pH, and combinations of these factors are 13, 23 and 28%, respectively.

**Table S1**. Information on relative abundance (%) of archaeal ASVs detected in poly(butylene succinate-co-adipate) (PBSA) and soils of all treatments. Please see in another excel file.

**Table S2.** Information on relative abundance (%) of bacterial ASVs detected in poly(butylene succinate-co-adipate) (PBSA) and soils of all treatments. Please see in another excel file.

**Table S3.** Information on relative abundance (%) of fungal ASVs detected in poly(butylene succinate-co-adipate) (PBSA) and soils of all treatments. Please see in another excel file.

**Table S4.** Comparisons between microbial community compositions of (i) PBSA and soil counterparts and (ii) soils of different treatments and (iii) control soils (Control S and Control SN) and PBSA (PS and PSN treatments) using analysis of similarities (ANOSIM) and non-parametric multivariate analysis of variance (NPMANOVA).

(i)

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| --- | --- | --- | --- |
| **Comparison** | **Archaea** | **Bacteria** | **Fungi** |
| **ANOSIM** | **NPMANOVA** | **ANOSIM** | **NPMANOVA** | **ANOSIM** | **NPMANOVA** |
| PBSA-soil: PBSA *vs*. soil community composition | *R* = 0.91, ***P* = 0.010** | *F* = 12.22, ***P* = 0.008** | *R* = 1.00, ***P* = 0.010** | *F* = 4.26, ***P* = 0.008** | *R* = 0.70, ***P* = 0.010** | *F* = 3.07, ***P* = 0.008** |
| PBSA-soil-N: PBSA *vs*. soil community composition | *R* = 0.37, ***P* = 0.007** | *F* = 5.01, ***P* = 0.008** | *R* = 0.96, ***P* = 0.007** | *F* = 3.74, ***P* = 0.008** | *R* = 0.39, ***P* = 0.040** | *F* = 2.38, ***P* = 0.031** |

(ii)

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| --- | --- | --- | --- |
| **Comparison** | **Archaea** | **Bacteria** | **Fungi** |
| **ANOSIM** | **NPMANOVA** | **ANOSIM** | **NPMANOVA** | **ANOSIM** | **NPMANOVA** |
| Control S *vs*. Control SN | *R* = 0.03, *P* = 0.31 | *F* = 1.04, *P* = 0.374 | *R* = 1.00, ***P* = 0.007** | *F* = 4.67, ***P* = 0.008** | *R* = 1.00, ***P* = 0.008** | *F* = 2.99, ***P* = 0.009** |
| Control S *vs*. Soil-PS  | *R* = 0.12, *P* = 0.24 | *F* = 1.39, *P* = 0.275 | *R* = 0.64, ***P* = 0.008** | *F* = 2.09, ***P* = 0.007** | *R* = 0.88, ***P* = 0.008** | *F* = 3.44, ***P* = 0.011** |
| Control S *vs*. Soil-PSN | *R* = 0.67, ***P* = 0.008** | *F* = 4.67, ***P* = 0.008** | *R* = 1.00, ***P* = 0.007** | *F* = 4.97, ***P* = 0.006** | *R* = 0.59, ***P* = 0.008** | *F* = 4.62, ***P* = 0.007** |
| Control SN *vs*. Soil-PS | *R* = 0, *P* = 0.696 | *F* = 0.80, *P* = 0.679 | *R* = 0.87, ***P* = 0.009** | *F* = 3.91, ***P* = 0.006** | *R* = 0.94, ***P* = 0.008** | *F* = 3.92, ***P* = 0.010** |
| Control SN *vs*. Soil-PSN | *R* = 0.53, ***P* = 0.008** | *F* = 4.60, ***P* = 0.006** | *R* = 0.94, ***P* = 0.008** | *F* = 4.88, ***P* = 0.009** | *R* = 0.59, ***P* = 0.007** | *F* = 4.43, ***P* = 0.008** |
| Soil-PS *vs*. Soil-PSN | *R* = 0.47, ***P* = 0.015** | *F* = 3.47, ***P* = 0.024** | *R* = 0.98, ***P* = 0.009** | *F* = 3.84, ***P* = 0.007** | *R* = 0.48, ***P* = 0.008** | *F* = 3.14, ***P* = 0.007** |

(iii)

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| **Comparison** | **Archaea** | **Bacteria** | **Fungi** |
| **ANOSIM** | **NPMANOVA** | **ANOSIM** | **NPMANOVA** | **ANOSIM** | **NPMANOVA** |
| Control S *vs*. PBSA-PS  | *R* = 0.92, ***P* = 0.008** | *F* = 12.16, ***P* = 0.008** | *R* = 1.00, ***P* = 0.008** | *F* = 5.82, ***P* = 0.009** | *R* = 0.96, ***P* = 0.008** | *F* = 5.20, ***P* = 0.009** |
| Control S *vs*. PBSA-PSN | *R* = 0.37, ***P* = 0.007** | *F* = 5.41, ***P* = 0.009** | *R* = 1.00, ***P* = 0.009** | *F* = 6.59, ***P* = 0.007** | *R* = 1.00, ***P* = 0.008** | *F* = 9.12, ***P* = 0.007** |
| Control SN *vs*. PBSA-PS | *R* = 0.92, ***P* = 0.008** | *F* = 12.98, ***P* = 0.008** | *R* = 1.00, ***P* = 0.008** | *F* = 6.36, ***P* = 0.007** | *R* = 0.97, ***P* = 0.009** | *F* = 5.16, ***P* = 0.008** |
| Control SN *vs*. PBSA-PSN | *R* = 0.37, ***P* = 0.008** | *F* = 5.71, ***P* = 0.009** | *R* = 1.00, ***P* = 0.009** | *F* = 7.15, ***P* = 0.008** | *R* = 1.00, ***P* = 0.008** | *F* = 8.87, ***P* = 0.008** |
| PBSA-PS *vs*. PBSA-PSN | *R* = 0.22, ***P* = 0.033** | *F* = 2.18, ***P* = 0.031** | *R* = 1.00, ***P* = 0.008** | *F* = 3.99, ***P* = 0.008** | *R* = 0.96, ***P* = 0.007** | *F* = 4.03, ***P* = 0.007** |