**SUPPLEMENTARY MATERIAL**

**­­­Altered patterns of compositional and functional disruption of the gut microbiota in typhoid fever and non-typhoidal febrile illness**

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**Supplementary Table 1:** **Overview of infectious diagnoses, microbiological data and antibiotic exposure prior to faecal sampling**

| **Characteristic** | **Fever, no typhoid (n=46)** | **Typhoid (n=17)** | **P** |
| --- | --- | --- | --- |
| **INFECTION TYPE** |  |  |  |
| Typhoid fever, n (%) | 0 (0.0) | 14 (100) |  |
| Lower respiratory tract infection, n (%) | 17 (36.9) | 0 (0.0) |  |
| Urinary tract infection, n (%) | 13 (28.2) | 0 (0.0) |  |
| Acute febrile illness, n (%) | 10 (21.7) | 0 (0.0) |  |
| Cholecystitis, n (%) | 2 (4.3) | 0 (0.0) |  |
| Tuberculosis, n (%) | 2 (4.3) | 0 (0.0) |  |
| Malaria, n (%) | 2 (4.3) | 0 (0.0) |  |
| **MICROBIOLOGICAL DATA** |  |  |  |
| *S.* Typhi PCR, positive/taken (%) | 0/46 (0.0) | 6/14 (42.8) |  |
| Blood cultures, positive/taken (%) | 7/46 (15.2) | 8/14 (57.1) |  |
| *S. Typhi* | 0 | 8 |  |
| *Escherichia coli* | 1 | 0 |  |
| *Serratia liquefaciens* | 1 | 0 |  |
| Other (suspected contaminants) | 5 | 0 |  |
| **HISTORY OF PRIOR ANTIBIOTIC EXPOSURE** |  |  |  |
| Antibiotics prior to faecal sampling, n (%) | 40 (87.0) | 14 (100.0) | 0.360 |
| Cephalosporin, n (%) | 31 (67.4) | 11 (82.4) | 0.641 |
| Cephalosporin exposure in days, median [IQR] | 2.00 [1.00, 3.00] | 2.00 [1.00, 5.50] | 0.578 |
| Penicillin, n (%) | 9 (19.6) | 1 (7.1) | 0.118 |
| Penicillin exposure in days, median [IQR] | 2.00 [1.00, 2.00] | 1.00 [1.00, 1.00] | 0.287 |
| Fluoroquinolone, n (%) | 9 (19.6) | 8 (57.1) | 0.017 |
| Fluoroquinolone exposure in days, median [IQR] | 6.00 [4.00, 7.00] | 2.00 [1.00, 5.50] | 0.262 |
| Metronidazole, n (%) | 5 (10.9) | 3 (21.4) | 0.570 |
| Metronidazole exposure in days, median [IQR] | 2.00 [1.00, 2.00] | 2.00 [1.50, 2.50] | 0.752 |
| Macrolide , n (%) | 9 (19.6) | 3 (21.4) | 1.000 |
| Macrolide exposure in days, median [IQR] | 2.00 [1.00, 2.00] | 2.00 [1.50, 3.50] | 0.695 |

Abbreviations: IQR, interquartile range; PCR, polymerase chain reaction

**Supplementary Table 2: Differences in beta diversity among groups**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Group** | **UniFrac (unweighted)** | | **UniFrac (weighted)** | |
| Control vs Fever, no typhoid | *P* =0.001 | R2 = 0.046 | *P* = 0.001 | R2 = 0.140 |
| Control vs Typhoid | *P* = 0.001 | R2 = 0.080 | *P* = 0.037 | R2 = 0.063 |
| Typhoid vs Fever, no typhoid | *P* = 0.014 | R2 = 0.027 | *P* = 0.504 | R2 = 0.016 |

Differences in microbiota composition among groups were tested for using permutational multivariate analysis of variance (PERMANOVA) on beta diversity matrices. False discovery rate was adjusted for with Benjamini-Hochberg.

**Supplemental Figure 1: Overview of microbiota composition and alpha diversity of each patient, combined with a timeline of hospitalization stay**

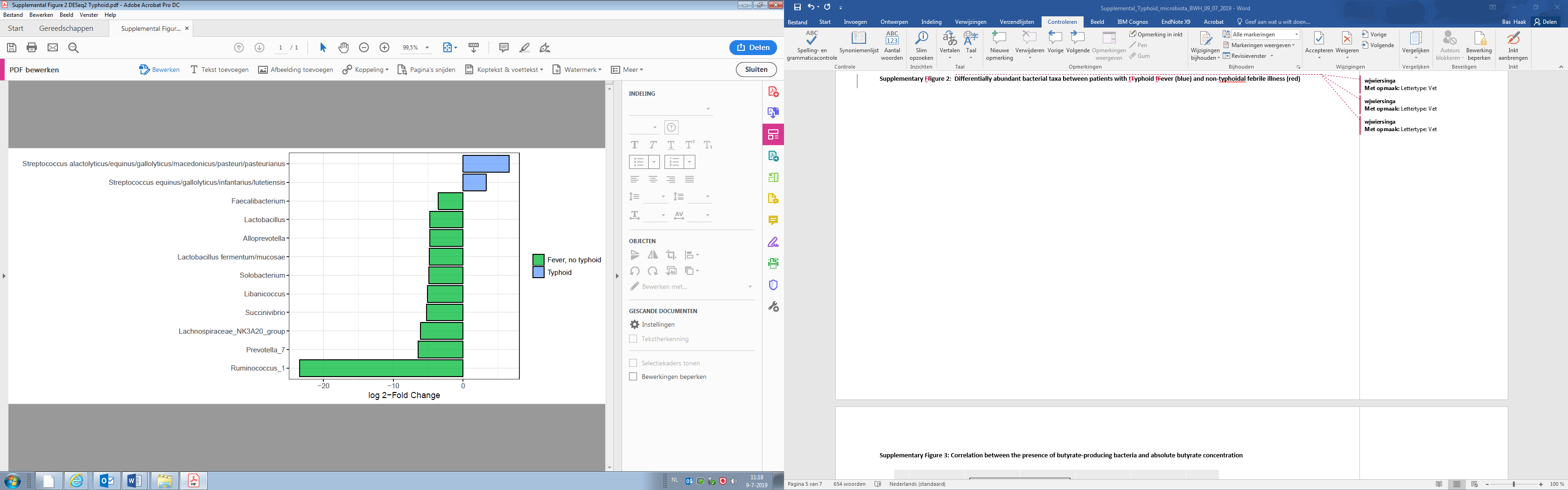


**Figure legend**

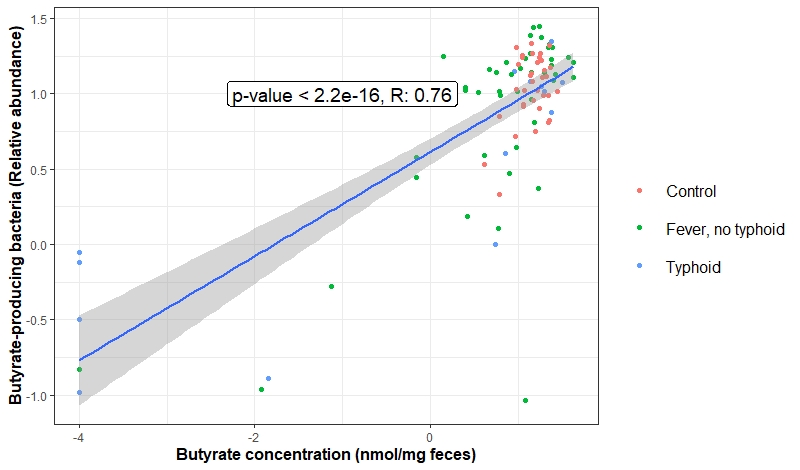
Overview of the microbiota composition (left panel), alpha diversity (Shannon index), combined with a timeline of hospitalization stay and antibiotic exposure of each patient. Non-typhoidal febrile illness patients are displayed at the top. Typhoid fever patients are displayed at the bottom.

Each bar represents one patient; microbiota taxa are indicated with colours and expressed in percentage of the total DNA reads. Only genera that made up ≥5 of the total microbiota in at least one sample are included; other genera are pooled within their respective phyla, or within the Ruminococcaceae and Lachnospiraceae families.

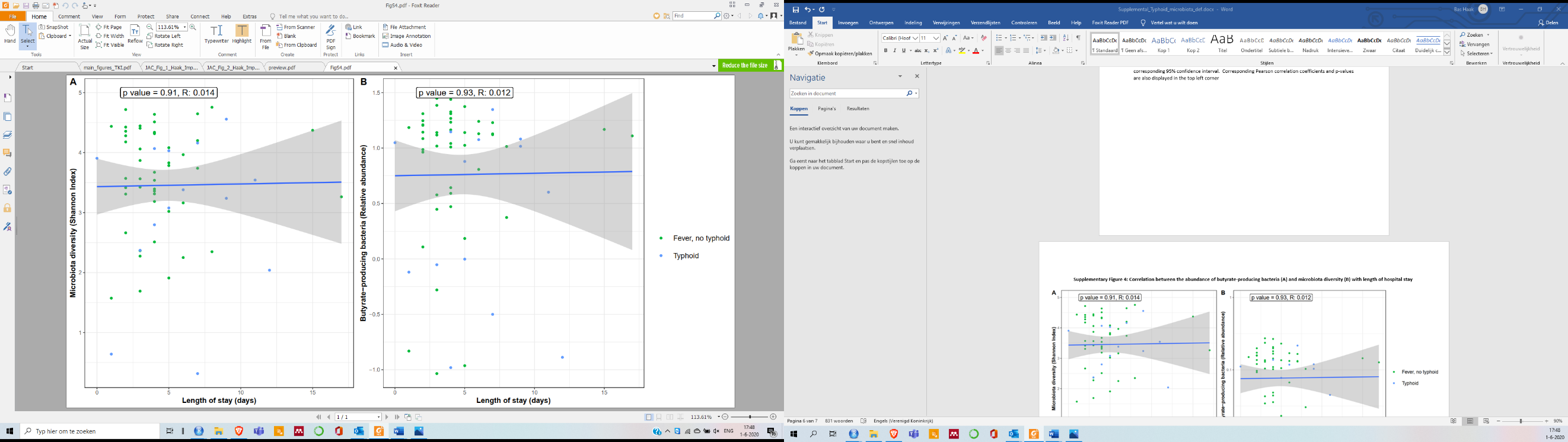
**Supplementary Figure 2**:  **Differentially abundant bacterial taxa between patients with typhoid fever (green) and non-typhoidal febrile illness (blue)**



**­­­­­Supplementary Figure 3: Correlation between the presence of butyrate-producing bacteria and absolute butyrate concentration**



Scatter plot of butyrate concentrations (nmol/mg feces after log10 transformation, x-axis) versus relative abundance of butyrate-producing bacteria (percentage after log10 transformation, y-axis). The line represents the linear regression fit (tested for linearity with Wald tests) and the shade the corresponding 95% confidence interval. Corresponding Pearson correlation coefficients and p-values are also displayed in the figure.

**­­Supplementary Figure 4: Correlation between the abundance of butyrate-producing bacteria (A) and microbiota diversity (B) with length of hospital stay**

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Scatter plot of hospital length of stay (days, x-axis) versus **(A)** microbiota diversity (Shannon Index, y-axis) or **(B)** relative abundance of butyrate-producing bacteria (% after log10 transformation, y-axis). The line represents the linear regression fit (tested for linearity by Wald tests) and the shade the corresponding 95% confidence interval. Corresponding Pearson correlation coefficients and p-values are displayed in the figure.