**Supplementary Materials:**

**Isolation and Biochemical Characterization of Long-Tailed Macaques Aerobic Gut Bacteria from Tinjil Island, Indonesia**

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**A diagram of a cell structure

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Figure S1. The flowchart of the study methods includes two types of Bergey's Manual of Determinative Bacteriology.

Table S1 Total viable bacteria in four fecal samples of LTM from Tinjil Island, Banten

|  |  |  |
| --- | --- | --- |
| **No.** | **Sample** | **Total viable bacteria (CFU/g)** |
| 1 | C72 | 2.81 x 109 |
| 2 | LB2 | 1.37 x 109 |
| 3 | LB3 | 2.18 x 109 |
| 4 | RB1 | 1.11 x 109 |
|  | Average | 1.86 x 109 |

Note: Fecal samples were collected from semi-wild long-tailed macaques onTinjil Island. Samples were labeled as follows: C72 (Cage 7 Sample 2), LB2 (Left Base camp Sample 2), LB3 (Left Base camp Sample 3), RB1 (Right Base camp Sample 1). CFU/g indicates colony-forming units per Gram.

Table S2 Morphological characteristics of bacterial colonies on NA medium

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **No.** | **Isolate**  **Code** | **Colony Morphology** | | | | |  |
| **Shape** | **Margin** | **Colour** | **Elevation** | **Transparency** | **Texture** |
| 1 | C72.1 | Irregular | Undulate | White | Flat | Opaque | Viscid |
| 2 | C72.2 | Irregular | Undulate | White | Flat | Opaque | Viscid |
| 3 | C72.3 | Irregular | Undulate | Yellow | Flat | Opaque | Viscid |
| 4 | LB2.1 | Irregular | Entire | White | Flat | Opaque | Mucoid |
| 5 | LB2.2 | Circular | Entire | Yellow | Flat | Opaque | Mucoid |
| 6 | LB2.3 | Circular | Entire | White | Raised | Translucent | Viscid |
| 7 | LB2.4 | Circular | Entire | White | Flat | Opaque | Viscid |
| 8 | LB2.5 | Irregular | Undulate | White | Flat | Opaque | Viscid |
| 9 | LB3.1 | Irregular | Undulate | Light yellow | Flat | Translucent | Viscid |
| 10 | LB3.2 | Circular | Entire | Yellow | Flat | Opaque | Viscid |
| 11 | LB3.3 | Circular | Entire | Light pink | Flat | Translucent | Viscid |
| 12 | LB3.4 | Irregular | Undulate | Light yellow | Flat | Translucent | Viscid |
| 13 | LB3.5 | Circular | Entire | White | Raised | Opaque | Viscid |
| 14 | LB3.6 | Circular | Entire | Light yellow | Raised | Opaque | Viscid |
| 15 | RB1.1 | Circular | Entire | White | Raised | Translucent | Viscid |
| 16 | RB1.2 | Circular | Entire | Yellow | Convex | Opaque | Viscid |
| 17 | RB1.3 | Circular | Entire | White | Raised | Opaque | Viscid |
| 18 | RB1.4 | Circular | Entire | Light pink | Flat | Translucent | Viscid |
| 19 | RB1.5 | Irregular | Undulate | White | Flat | Opaque | Mucoid |

Note : Isolate codes represent bacterial isolates obtained from fecal samples of semi-wild long-tailed macaques from Tinjil Island. Isolates include C72.1 (Cage 7 Sample 2 Isolate 1), C72.2 (Cage 7 Sample 2 Isolate 2), C72.3 (Cage 7 Sample 2 Isolate 3), LB2.1 (Left Base camp Sample 2 Isolate 1), LB2.2 (Left Base camp Sample 2 Isolate 2), LB2.3 (Left Base camp Sample 2 Isolate 3), LB2.4 (Left Base camp Sample 2 Isolate 4), LB2.5 (Left Base camp Sample 2 Isolate 5), LB3.1 (Left Base camp Sample 3 Isolate 1), LB3.2 (Left Base camp Sample 3 Isolate 2), LB3.3 (Left Base camp Sample 3 Isolate 3), LB3.4 (Left Base camp Sample 3 Isolate 4), LB3.5 (Left Base camp Sample 3 Isolate 5), LB3.6 (Left Base camp Sample 3 Isolate 6), RB1.1 (Right Base camp Sample 1 Isolate 1), RB1.2 (Right Base camp Sample 1 Isolate 2), RB1.3 (Right Base camp Sample 1 Isolate 3), RB1.4 (Right Base camp Sample 1 Isolate 4), RB1.5 (Right Base camp Sample 1 Isolate 5).

Table S3 Bacterial cell morphology through Gram staining

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **No** | **Isolate** | **Characteristics of bacteria** | | |
| **Form** | **Dominant Arrangement** | **Gram** |
| 1 | C72.1 | Bacilli | Streptobacilli | Positive |
| 2 | C72.2 | Cocci | Staphylococci | Positive |
| 3 | C72.3 | Cocci | Staphylococci | Positive |
| 4 | LM2.1 | Bacilli | Streptobacilli | Positive |
| 5 | LM2.2 | Cocci | Monococci | Positive |
| 6 | LM2.3 | Cocci | Staphylocci | Positive |
| 7 | LM2.4 | Bacilli | Diplobacilli | Positive |
| 8 | LM2.5 | Bacilli | Palisade | Positive |
| 9 | LM3.1 | Bacilli | Monobacilli | Positive |
| 10 | LM3.2 | Cocci | Staphylococci | Positive |
| 11 | LM3.3 | Coccobacilli | Coccobacilli | Positive |
| 12 | LM3.4 | Bacilli | Palisade | Positive |
| 13 | LM3.5 | Cocci | Staphylococci | Positive |
| 14 | LM3.6 | Cocci | Tetracocci | Positive |
| 15 | RM1.1 | Cocci | Monococci | Positive |
| 16 | RM1.2 | Cocci | Staphylococci | Positive |
| 17 | RM1.3 | Cocci | Staphylococci | Positive |
| 18 | RM1.4 | Coccobacilli | Coccobacilli | Positive |
| 19 | RM1.5 | Bacilli | Streptobacilli | Positive |

Note : Isolate codes represent bacterial isolates obtained from fecal samples of semi-wild long-tailed macaques from Tinjil Island. Isolates include C72.1 (Cage 7 Sample 2 Isolate 1), C72.2 (Cage 7 Sample 2 Isolate 2), C72.3 (Cage 7 Sample 2 Isolate 3), LB2.1 (Left Base camp Sample 2 Isolate 1), LB2.2 (Left Base camp Sample 2 Isolate 2), LB2.3 (Left Base camp Sample 2 Isolate 3), LB2.4 (Left Base camp Sample 2 Isolate 4), LB2.5 (Left Base camp Sample 2 Isolate 5), LB3.1 (Left Base camp Sample 3 Isolate 1), LB3.2 (Left Base camp Sample 3 Isolate 2), LB3.3 (Left Base camp Sample 3 Isolate 3), LB3.4 (Left Base camp Sample 3 Isolate 4), LB3.5 (Left Base camp Sample 3 Isolate 5), LB3.6 (Left Base camp Sample 3 Isolate 6), RB1.1 (Right Base camp Sample 1 Isolate 1), RB1.2 (Right Base camp Sample 1 Isolate 2), RB1.3 (Right Base camp Sample 1 Isolate 3), RB1.4 (Right Base camp Sample 1 Isolate 4), RB1.5 (Right Base camp Sample 1 Isolate 5).

Table S4 Biochemical test on bacterial isolates of Gram-positive cocci

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **No** | **Isolate**  **Code** | |  | | **Biochemical Tests** | | | | | | |
| **Ca** | | **M** | | **Y** | | **G** | | |
| 1 | C72.2 | | Bubbles  **+** | | Control  **+** | | **+** | | Not Tested | | |
| 2 | C72.3 | | Bubbles  **+** | | Control  **+** | | **+** | | Not Tested | | |
| 3 | LB2.2 | | Bubbles  **+** | | Control  **-** | | **+** | | Control  **-** | | |
| 4 | LB2.3 | | Bubbles  **+** | | Control  **-** | | **-** | | Not Tested | |
| 5 | LB3.2 | | Bubbles  **+** | | Control  **-** | | **+** | | Control  **-** | | |
| 6 | | LB3.5 | | Bubbles  **+** | | A close-up of two vials with green liquid  Description automatically generated  Control  **-** | | A white oval object with a light reflecting off it  Description automatically generated  **-** | | Not Tested |
| 7 | | LB3.6 | | A close-up of a black object  Description automatically generated  Bubbles  **+** | | A close-up of two vials with green liquid  Description automatically generated  Control  **-** | | A group of white round objects  Description automatically generated  + | | A couple of test tubes with green liquid  Description automatically generated  Control  **-** |
| 8 | | RB1.1 | | A bubble on a surface  Description automatically generated  Bubble  + | | A test tubes with green liquid in them  Description automatically generated  Control  - | | A white sphere in the sky  Description automatically generated  - | | Not Tested |
| 9 | | RB1.2 | | A close-up of a rectangular object  Description automatically generated  Bubble  + | | **A couple of test tubes with green liquid  Description automatically generated**  Control  - | | A white bubble with a light reflection  Description automatically generated with medium confidence  + | | A close-up of a test tube  Description automatically generated  Control  - |
| 10 | | RB1.3 | | A close-up of a white object  Description automatically generated  Bubble  + | | A close-up of test tubes  Description automatically generated  Control  - | | A white ball in the sky  Description automatically generated  - | | Not Tested |

Note : Isolate codes represent bacterial isolates obtained from fecal samples of semi-wild long-tailed macaques from Tinjil Island. Isolates include C72.2 (Cage 7 Sample 2 Isolate 2), C72.3 (Cage 7 Sample 2 Isolate 3), LB2.2 (Left Base camp Sample 2 Isolate 2), LB2.3 (Left Base camp Sample 2 Isolate 3), LB3.2 (Left Base camp Sample 3 Isolate 2), LB3.5 (Left Base camp Sample 3 Isolate 5), LB3.6 (Left Base camp Sample 3 Isolate 6), RB1.1 (Right Base camp Sample 1 Isolate 1), RB1.2 (Right Base camp Sample 1 Isolate 2), RB1.3 (Right Base camp Sample 1 Isolate 3). Type of biochemical tests: Ca(Catalase: (+) bubbles, (-) no bubbles); M(Mannitol: (+) yellow+bubbles in durham tube, (-) green); Y(Yellow pigment ); G(Glucose: (+)yellow+bubbles in durham tube, (-) green).

Table S5 Biochemical tests (Endospore stain, acid fast stain, and Catalase test) on bacterial isolates of Gram-positive bacilli

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **No** | **Isolate code** | **Biochemical Tests** | | | |
| **E** | **Af** | **Ca** | |
| 1 | C72.1 | A screenshot of a computer  Description automatically generated  E  FS  V  Swollen cell  (E >V; FS>V) | Not Tested | A white powder on a black surface  Description automatically generated  Bubble  + | |
| 2 | LB2.1 | A close-up of a microscope  Description automatically generated  V  FS  + | Not Tested | A close-up of a white substance  Description automatically generated  Bubble  + | |
| 3 | LB2.4 | A close-up of a microscope  Description automatically generated  - | Blue speckled specks on a white background  Description automatically generated  - | A close-up of a fingerprint  Description automatically generated  Bubble  + | |
| 4 | LB2.5 | A close-up of a white surface  Description automatically generated  - | A close-up of a blue and white cell  Description automatically generated  - | A white substance on a black surface  Description automatically generated  Bubble  + | |
| 5 | LB3.1 | A close-up of a microscope  Description automatically generated  - | A close-up of a microscope  Description automatically generated  - | A close-up of a microscope slide  Description automatically generated  Bubble  + | |
| 6 | LB3.3 | A close-up of a white surface  Description automatically generated  - | A close-up of a microscope  Description automatically generated  - | A close-up of a plastic  Description automatically generated  Bubble  + |
| 7 | LB3.4 | A close-up of a microscope  Description automatically generated  - | A close-up of a microscope  Description automatically generated  - | A close-up of a glass  Description automatically generated  Bubble  + |
| 8 | RB1.4 | A close-up of a microscope  Description automatically generated  - | A close-up of a microscope  Description automatically generated  - | A close-up of a white object  Description automatically generated  Bubble  + |
| 9 | RB1.5 | A close-up of a microscope  Description automatically generated  ES  V  FSE  + | Not Tested | A close-up of a white substance  Description automatically generated  Bubble  + |

Note: Isolate codes represent bacterial isolates obtained from fecal samples of semi-wild long-tailed macaques from Tinjil Island. Isolates include C72.1 (Cage 7 Sample 2 Isolate 1), LB2.1 (Left Base camp Sample 2 Isolate 1), LB2.4 (Left Base camp Sample 2 Isolate 4), LB2.5 (Left Base camp Sample 2 Isolate 5), LB3.1 (Left Base camp Sample 3 Isolate 1), LB3.3 (Left Base camp Sample 3 Isolate 3), LB3.4 (Left Base camp Sample 3 Isolate 4), RB1.4 (Right Base camp Sample 1 Isolate 4), RB1.5 (Right Base camp Sample 1 Isolate 5). Type of biochemical tests E(Endospore stain: (+) endospore-forming, (-) non-endospore forming), Af(Acid-fast: (+) red cells, (-) blue cell, Ca(Catalase (+) bubbles, (-) no bubbles). (E) Endospore, (FS) Free spore, (V) Vegetative.

Table S6 Biochemical tests (Voges-Proskauer, Cell diameter, Motility, NaCl) on bacterial isolates of Gram-positive bacilli

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **No** |  | **Isolate Code** | **Biochemical Tests** | | | |
|  |  |  | **VP** | **Cd** | **Mo** | **Na** |
| 1 |  | C72.1 | A test tube with a liquid in it  Description automatically generated  - | Not Tested | Not Tested  Control - | A couple of test tubes with liquid in them  Description automatically generated  - |
| 2 |  | LB2.1 | A test tube with a red liquid  Description automatically generated  + | A screenshot of a computer  Description automatically generated  +  Cell diameter > 1 um | (Observation under the microscope)  + | Not Tested |
| 3 |  | LB2.4 | Not Tested | Not Tested | Not Tested | Not Tested |
| 4 |  | LB2.5 | Not Tested | Not Tested | Not Tested | Not Tested |
| 5 |  | LB3.1 | Not Tested | Not Tested | Not Tested | Not Tested |
| 6 |  | LB3.3 | Not Tested | Not Tested | Not Tested | Not Tested |
| 7 |  | LB3.4 | Not Tested | Not Tested | Not Tested | Not Tested |
| 8 |  | RB1.4 | Not Tested | Not Tested | Not Tested | Not Tested |
| 9 |  | RB1.5 | A test tube with a red liquid  Description automatically generated+ | A screenshot of a computer  Description automatically generated  +  Cell diameter > 1 um | (Observation under the microscope)  + | Not Tested |

Note : Isolate codes represent bacterial isolates obtained from fecal samples of semi-wild long-tailed macaques from Tinjil Island. Isolates include C72.1 (Cage 7 Sample 2 Isolate 1), LB2.1 (Left Base camp Sample 2 Isolate 1), LB2.4 (Left Base camp Sample 2 Isolate 4), LB2.5 (Left Base camp Sample 2 Isolate 5), LB3.1 (Left Base camp Sample 3 Isolate 1), LB3.3 (Left Base camp Sample 3 Isolate 3), LB3.4 (Left Base camp Sample 3 Isolate 4), RB1.4 (Right Base camp Sample 1 Isolate 4), RB1.5 (Right Base camp Sample 1 Isolate 5). Type of biochemical: VP(Voges-Proskauer: (+) red, (-) oranye/brown), Cd (Cell diameter: (+) Cd > 1 um, (-) Cd < 1 um), M(Motility test: (+) motile, (-) nonmotile), Na(NaCl 6.5%: (+) cloudy, (-) almost clear/same with negative control).