

Table S1. General and oral conditions of participants with different total bacterial density on the tongue microbiota.

| | Total bacterial density ^a | | <i>P</i> -value |
|---|--------------------------------------|------------------|--------------------|
| | Low (n= 242) | High (n= 242) | |
| Age (Mean ± SD) | 74.9±2.9 | 75.1±3.0 | 0.426 ^b |
| No. (%) of female | 139 (57.4) | 127 (52.5) | 0.273 ^c |
| Body mass index (Mean ± SD) | 23±3.4 | 23.4±3.2 | 0.202 ^b |
| Smoking intensity (n (%)) | | | 0.582 ^c |
| Nonsmokers | 149 (61.6) | 141 (58.3) | |
| Moderate smokers | 36 (14.9) | 34 (14) | |
| Heavy smokers | 57 (23.6) | 67 (27.7) | |
| Current smokers (n (%)) | 16 (6.6) | 16 (6.6) | 1 ^c |
| Never drinkers (n (%)) | 97 (40.1) | 82 (33.9) | 0.158 ^c |
| Physically active (n (%)) | 135 (55.8) | 149 (61.6) | 0.196 ^c |
| Dental conditions | | | |
| No. of teeth | 20.8±7.6 | 21.4±6.9 | 0.347 ^a |
| No. (%) of subjects with decayed teeth | 45 (18.6) | 50 (20.7) | 0.567 ^c |
| Mean % of caries-experienced teeth | 66.2±26.9 | 66.4±24.7 | 0.905 ^c |
| Mean max plaque index | 1.1±0.8 | 1.2±0.7 | 0.063 ^b |
| Periodontal conditions ^d (n (%)) | | | 0.358 ^c |
| No/Mild | 165 (68.2) | 160 (66.1) | |
| Moderate/Severe | 66 (27.3) | 77 (31.8) | |
| No. (%) of edentulous individuals | 11 (4.5) | 5 (2.1) | 0.203 ^e |
| Presence of airflow limitation | 41 (16.9) | 59 (24.4) | 0.043 ^c |

^aStratified by median value. ^bStudent's t-test. ^cChi-square test. ^dOnly 468 dentulous individuals were included. ^eFisher's exact test.

Table S2 Adjusted odds ratio for airflow limitation in multiple logistic regression analysis.

| | Adjusted odds ratio (95% CI) |
|--|------------------------------|
| Age | 1.12 (1.04–1.21)** |
| Gender (Male) | 0.98 (0.45–2.10) |
| Body mass index | 0.87 (0.80–0.94)*** |
| Smoking intensity | |
| Nonsmokers | 1 |
| Moderate smokers | 2.02 (0.87–4.71) |
| Heavy smokers | 3.68 (1.65–8.20)** |
| Never drinkers | 1.11 (0.63–1.97) |
| Presence of decayed teeth | 1.35 (0.77–2.37) |
| Periodontal condition | |
| No/Mild | 1 |
| Moderate/Severe | 1.07 (0.64–1.78) |
| Edentulous | 1.44 (0.41–5.04) |
| Total bacterial density on the tongue (Log copies) | 1.80 (1.01–3.18)* |

Each odds ratio was adjusted by age, gender, body mass index, smoking intensity, alcohol intake, presence of decayed teeth and periodontal condition with P values < 0.2 in a bivariate analysis. *** $P < 0.001$, ** $P < 0.01$, * $P < 0.05$.

Table S3 Odds ratio for airflow limitation of total bacterial density on the tongue in multiple logistic regression analysis for 452 individuals without any current habit of smoking.

| | Odds ratio | |
|---------------------------------------|-------------------|-------------------|
| | Crude (95% CI) | Adjusted (95% CI) |
| Total bacterial density on the tongue | | |
| Low | 1 | 1 |
| High | 1.81 (1.12–2.97)* | 1.81 (1.09–3.04)* |

Odds ratios were calculated after excluding 32 current smokers. The odds ratio was adjusted by age, sex, body mass index, smoking intensity, alcohol intake, presence of decayed teeth and periodontal condition. * $P < 0.05$.

Table S4 Community types of the tongue microbiota and the presence of airflow limitation.

| | Normal airflow (n= 384) | Airflow limitation (n= 100) | <i>P</i> -value ^a |
|------------------------------|----------------------------|--------------------------------|------------------------------|
| Community types ^b | | | 0.112 |
| Type 1 (n= 87) | 71 (18.5) | 16 (16.0) | |
| Type 2 (n= 110) | 84 (21.9) | 26 (26.0) | |
| Type 3 (n= 152) | 129 (33.6) | 23 (23.0) | |
| Type 4 (n= 135) | 100 (26.0) | 35 (35.0) | |

^aChi-square test. ^bCommunity types of the tongue microbiota were classified by partitioning around medoids (PAM) cluster analysis using the Jensen-Shannon divergence (JSD) and Calinski-Harabasz (CH) index which were used for enterotypes clustering.

Table S5 Bacterial taxa corresponding to differentially abundant OTUs in tongue microbiota of the individuals with airflow limitation and those with normal airflow.

| OTU No. | Bacterial taxa corresponding to OTU ^a | Normal airflow (n= 384) | Airflow limitation (n= 100) | LDA score |
|--|--|-------------------------|-----------------------------|-----------|
| Differentially abundant in the individuals with airflow limitation | | | | |
| #6 | <i>Neisseria flavescens</i> (610) | 5.5±7.8 | 7.3±10.5 | 4.00 |
| #4 | <i>Prevotella melaninogenica</i> (469) | 8.9±6.5 | 10.1±6.1 | 3.71 |
| #421 | Genus <i>Neisseria</i> ^b | 2.1±4.8 | 3.0±5.7 | 3.70 |
| Differentially abundant in the individuals with normal airflow | | | | |
| #19 | <i>Streptococcus parasanguinis</i> (411) | 4.5±3.9 | 3.7±3.8 | 3.53 |

Linear discriminant analysis effect size (LEfSe) analysis was conducted and OTUs with a high LDA score (>3.5) were shown.

^aOral taxon IDs in HOMD are given in parentheses following bacterial names. ^bNo BLAST hit with ≥98.5% identity was found in the HOMD.

Table S6 Bacterial amounts of 21 predominant (with mean relative abundance >1%) operational taxonomic units (OTUs) in the tongue coating samples of participants with normal airflow and those with airflow limitation

| OTU No. | Median bacterial density per sample | | Bacterial taxa corresponding to each OTU ^a |
|---------|-------------------------------------|-----------------------------|---|
| | Normal airflow (n= 384) | Airflow limitation (n= 100) | |
| #4 | 16730119 | 25656687** | <i>Prevotella melaninogenica</i> (469) |
| #2 | 14446642 | 15183719 | <i>Streptococcus salivarius</i> (755) |
| #1 | 12662001 | 14488616* | <i>Rothia mucilaginosa</i> (681) |
| #9 | 12629204 | 15106277 | <i>Veillonella atypica</i> (524) |
| #19 | 6851811 | 6427302 | <i>Streptococcus parasanguinis_II</i> (411) |
| #10 | 5923537 | 9753585** | <i>Actinomyces odontolyticus</i> (701) |
| #7 | 5627024 | 6546995* | <i>Granulicatella adiacens</i> (534) |
| #6 | 4699346 | 9303785** | <i>Neisseria flavescens</i> (610) |
| #3 | 4589601 | 5660723 | <i>Prevotella histicola</i> (298) |
| #5 | 3766301 | 4181986 | <i>Haemophilus parainfluenzae</i> (718) |
| #25 | 2738309 | 3023472 | <i>Actinomyces</i> sp. (172) |
| #15 | 2299137 | 3407161 | <i>Prevotella pallens</i> (714) |
| #11 | 1840369 | 1707299 | <i>Gemella sanguinis</i> (757) |
| #8 | 1644017 | 2326732 | <i>Fusobacterium periodonticum</i> (201) |
| #28 | 1490319 | 2405718* | Genus <i>Streptococcus</i> ^b |
| #22 | 1441301 | 2117044* | <i>Streptococcus</i> sp. (074) |
| #23 | 1183495 | 1860080* | <i>Actinomyces graevenitzii</i> (866) |
| #17 | 1142547 | 1164849 | <i>Alloprevotella</i> sp. (308) |
| #12 | 380751 | 698806* | <i>Leptotrichia</i> sp. (417) |
| #14 | 337964 | 635212 | <i>Porphyromonas pasteri</i> (279) |
| #421 | 27296 | 147826* | Genus <i>Neisseria</i> ^b |

We calculated the bacterial amounts of 21 OTUs by multiplying total bacterial density and the relative abundance of each OTU. Bacterial amount of each OTU was compared between participants with normal airflow and those with airflow limitation using Wilcoxon Rank Sum test. ** $P < 0.01$, * $P < 0.05$; ^aOral taxon IDs in HOMD are given in parentheses following bacterial names. ^bNo BLAST hit with $\geq 98.5\%$ identity was found in the HOMD.

Supplemental figure legends

Figure S1 Total bacterial density (Log 10 transformed) of the tongue microbiota of the elderly adults with various oral conditions. Dots indicate the mean; the error bars indicate 95% confidence intervals. No significant difference was observed between the groups by Kruskal-Wallis analysis.