Tongue coating microbiome data distinguish patients with chronic heart failure from healthy controls

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Conflicts of interest：
All the funds were from the administration. In the process of research progress, there will be no possibility for me to influence judgment and behavior induced by economic, social relations and academic reputation interests.

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Background:
Clinically, there exists difference in tongue images including tongue coating and tongue color between chronic heart failure (CHF) patients and healthy individuals. Recent reports have suggested alteration in the tongue microbiota, which may play a critical role in diseases. CHF-associated tongue coating microbiome dysbiosis has not yet been clearly defined.

Health controls -associated tongue coating

Description： pale red and somewhat moist with pale white

CHF-associated tongue coating

Description： tongue material redder with pale yellow coating, especially at the rear(root) of tongue

Purpose:

Our aim is to investigate the composition of the tongue microbiome in subjects with and without CHF，which may provide a new impetus to solve the insufficiency of biomarkers in guiding the management of CHF.

Methods:

A prospective case control study were performed. Tongue-coating samples collected from 60 CHF patients and 30 health controls would have been studied by sequencing technology of the 16S rRNA gene of the V3-V4 hyper variable region with an Illumina MiSeq. The sequenced data were analyzed using QIIME, and the sequences obtained were distributed across 7 phyla, 27 genera and 825 operational taxonomic units (OTUs). If the subject had oral, tongue or dental diseases; suffered upper respiratory tract infection in the past week; used antibiotics and immunosuppressants in the past week; and pregnant or in lactation, he/she would have been excluded.

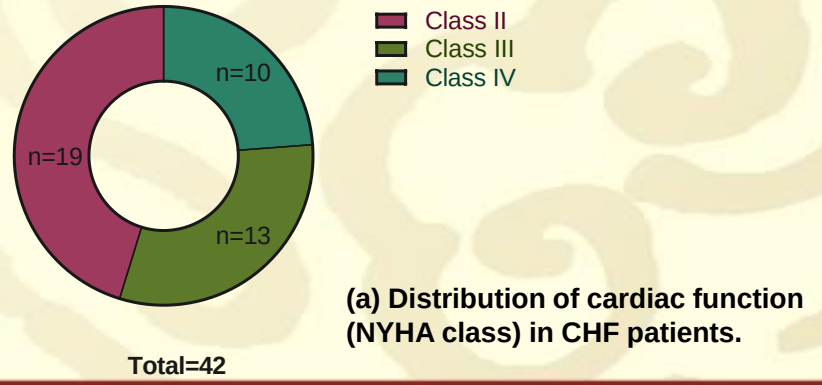
Results:

In total, 42 CHF patients (median age 79.5，range, 30-90 years) with NYHA class II-IV and disease duration from 2 to 5 years, and 28 healthy controls (median age 25, range, 24-45 years) were recruited.

Table 1. Demographic characteristics of healthy controls and CHF patients

	Characteristics	Normal controls	CHF patients
Sample size		28	42
Age	Median (range)	79.5 (30-90)	25 (24-45)
Sex	Male/Female	12/16	18/24

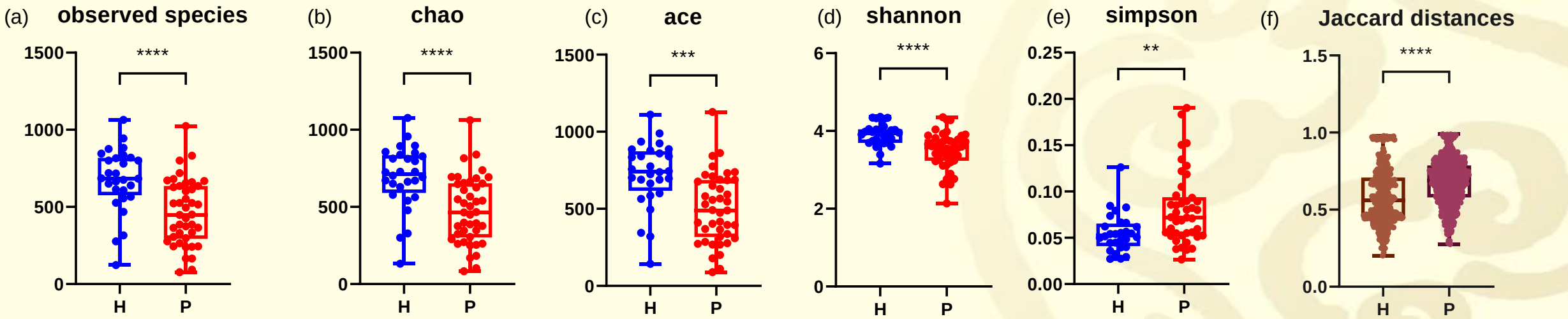
(a)



(a) Distribution of cardiac function (NYHA class) in CHF patients.

Results:

Finding 1: The overall tongue microbial community (both alpha and beta diversity) differed between patients with CHF and healthy control (P<0.05). In terms of alpha diversity, the community richness indexes including the observed species, the chao1 estimator and the ace estimator, showed significantly decreased in heart failure patients when compared with those of healthy controls (Fig.a-c), indicating a lower number of species in heart failure patients. Besides, the Shannon index was significantly lower and the Simpson index significantly higher in heart failure patients than those of healthy controls, which revealed a less uniform species distribution in heart failure patients (Fig.d-e). In terms of beta diversity, the Jaccard distances between CHF patients were higher than that between normal controls (P<0.05), reflecting that the tongue coating microbiota in CHF patients were higher dissimilar (Fig.f).

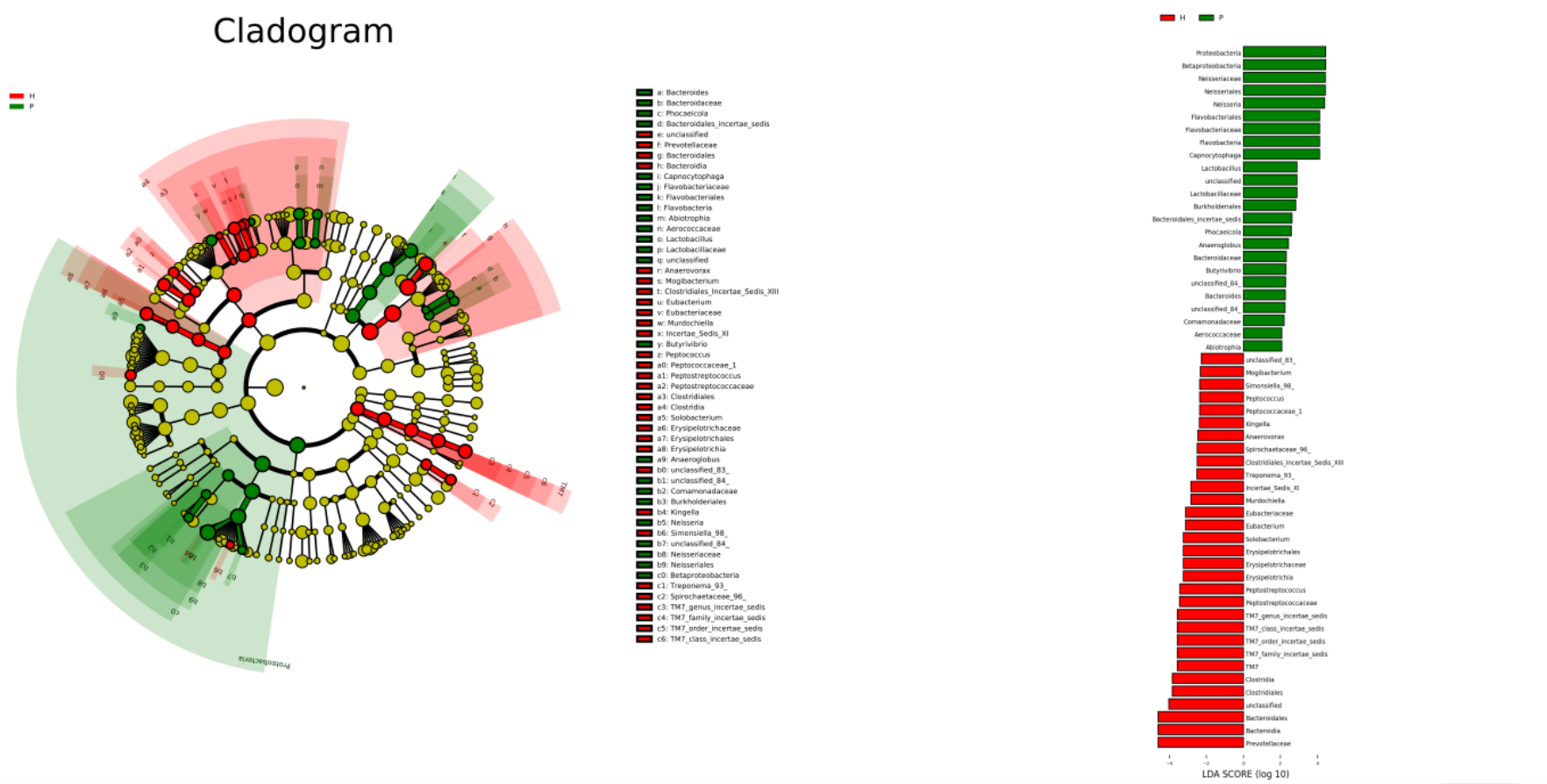


(a) Number of observed species in normal controls and CHF patients. (b) Chao ace index in normal controls and CHF patients.
(c) Ace index in normal controls and CHF patients. (d) Shannon index in normal controls and CHF patients.
(e) Simpson index in normal controls and CHF patients. (f) Jaccard distances between normal controls and between CHF patients.

Finding 2:

Heart failure patients group was characterized by a dominant of Neisseria, Capnocytophaga, Lactobacillus, Phocaeicola, Anaeroglobus, Butyrivibrio, Bacteroides and Abiotrophia (P<0.05, P<0.001) , while TM7_genus_incertae_sedis, Solobacterium, eubacterium, Murdochiella, Treponema_93, Anaerovorax, kingella, Peptococcus and Mogibacterium were preponderant in the healthy control group(P<0.05, P<0.001).

(a)



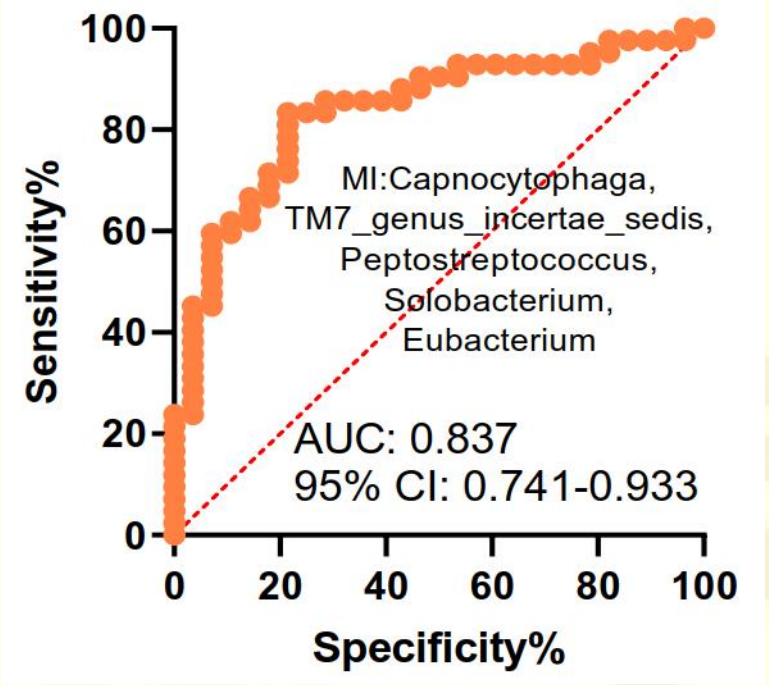
(b)

(a) Cladogram using the LEfSe method indicating the phylogenetic distributionof tongue coat microbes associated with patients with CHF (green indicates phylotypes statistically overrepresented in CHF) and healthy subjects (red indicates phylotypes overrepresented in healthy controls). Each filled circle represents one phylotype, and phylum and class are indicated in their names on the cladogram and the order, family, or genera are given on the right panel.

(b) A histogram of the linear discriminant analysis (LDA) scores was calculated for the selected taxa which showed the significant bacterial difference between the CHF and HC. LDA score at the log 10 scale is indicated at the bottom. The greater the LDA score is, the more significant the microbial biomarker is in the comparison.

Finding 3:
Strikingly, in genus level, Capnocytophaga, TM7_genus_incertae_sedis, Peptostreptococcus, Solobacterium and Eubacterium could distinguish CHF patients from healthy control (AUC: 83.7%, 95% confidential interval 74.1%–93.3%).

(a)



(a) Prediction of microbial index (MI, the key genera which can distinguish CHF patients from healthy controls). The area under the ROC curve (AUC = 0.837).

Conclusion:
Our findings identified the microbiota dysbiosis of the tongue coat in CHF patients, and provide insight into the association between the human microbiome and CHF. It is worth to explore the underlying mechanisms between tongue coating microbiome and cardiac function.

Thanks for your attention