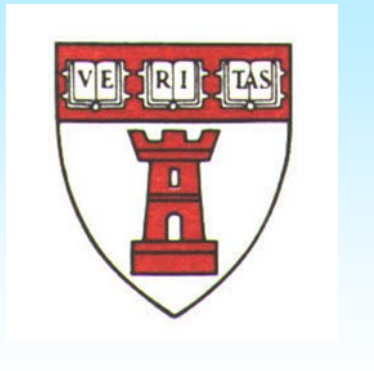# Canine Oral Bacteria Identified in Human Skin Microbiome Libraries

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Abstract

Human skin is constantly exposed to the external environment. It is therefore not surprising that an extremely wide variety of bacteria have been identified from skin samples. Some of the bacteria found are likely endogenous to man, comprising the normal skin microbiome, while others are likely to be exogenous environmental transients. Distinguishing these two groups is difficult, but some genera are known almost exclusively to be mammalian host-associated, while others are known almost exclusively environment-associated such as from soil or water. This approach fails when skin is contaminated with microbes from another mammal. Over 1/3 of U.S. households have dogs. People living with dogs are often exposed to dog saliva either by being licked or touching objects that have been in a dog's mouth. Research on the canine microbiome (2) has identified 353 oral taxa based on 16S rRNA clone library analysis, 295 of which are not shared with the human microbiome. The purpose of this study was to see if canine oral taxa could be found in human skin microbiome studies based on 16S rRNA sequence analysis. BLASTN analysis of 16S rRNA canine reference sequences against GenBank identified 1,185 sequences from human skin with >98.5% similarity to 118 canine oral taxa. The most commonly identified taxa were *Porphyromonas cangingivalis* (101), *Jeotgalicoccus halotolerans* (97), *Conchiformibius* sp. COT-286 (65), and *Porphyromonas gulae* COT-108 (56). When the human subjects were ranked by number of canine taxa recovered, the top 10 subjects had 50, 48, 44, 28, 23, 22, 17, 16, 15, and 9, taxa respectively. In conclusion, it appears human skin sites, such as volar forearm, can be massively contaminated with canine oral bacteria. This cross mammalian contamination may make distinguishing "oral" from "skin" and "endogenous" from "exogenous" bacteria a more complex task. Supported by NIDCR DE016937.

Introduction

In order to fully understand human health and disease, and health and disease in other mammals, it is necessary to know the microorganisms that comprise the host associated microbiomes. Our laboratory has focused on the oral microbiomes of human (1), canine (2) and other mammals. In recent work on the canine microbiome we identified 353 canine oral taxa based on operational taxonomic units with >98.5% full 16S rRNA sequence similarity (2) While examining the similarity of the canine 16S rRNA reference sequences to sequences in GenBank, we noticed that many of the canine sequences were highly similar to sequences from studies of the human skin microbiome. The purpose of this study was to examine the source and frequency of clones from human skin libraries that match reference sequences for the canine oral cavity.

Methods

Four hundred sixteen full length 16S rRNA reference sequences for 353 canine oral taxa (GenBank JN713151–JN713566) were subject to BLASTN analysis against the NCBI non redundant nucleotide database (dated 2012-04-23). The top 200 results for each reference sequence with similarity >90.0% were recorded. A script retrieved header information for each sequence, including host species, subject ID and body site. The header information was combined with BLAST results in an output Excel file. Hits with >98.5% similarity were selected. Hits not from a human source were eliminated. Hits not from human skin were eliminated. Hits for canine taxa with more than one reference sequence were de-replicated. Sequences which did not match over the full length of the alignment were discarded.

Tables and Figures

Table 1. Rank abundance of canine oral taxa

Rank	Canine oral taxon	Query Organism	Clones / taxon max	Shared human oral cavity
1	109	Porphyromonas cangingivalis	101	
2	347	Jeotgalicoccus halotolerans	97	
3	286	Conchiformibius sp.	78	
4	289	Conchiformibius sp. COT-289	65	
5	052	Porphyromonas gulae COT-052	56	
6	279	Streptococcus sp. COT-279	51	
7	331	Aerococcus viridans	50	
8	271	Pasteurellaceae [G-2] sp. COT-271	41	
9	186	Bergeyella zoohelcum	32	
10	115	Eikenella corrodens	29	yes
11	121	Selenomonas noxia	29	yes
12	254	Capnocytophaga cynodegmi	28	
13	280	Conchiformibius steedae	25	
14	022	Porphyromonas gingivicanis	23	
15	083	Actinomyces sp. COT-083	23	
16	108	Porphyromonas canoris	23	
17	229	Abiotrophia sp. COT-229	23	
18	189	Fusobacterium sp. COT-189	21	
19	343	Mogibacterium diversum	20	yes
20	089	Gemella palaticanis	19	
21	017	Moraxella sp. COT-017	18	
22	080	Pasteurellaceae [G-2] sp. COT-080	15	
23	018	Moraxella sp. COT-018	14	
24	060	Lautropia sp. COT-060	13	
25	111	'Eubacterium' infirmum	13	yes
26	311	Erysipelotrichaceae [G-1] sp. COT-311	12	
27	342	Selenomonas sputigena	10	yes
28	374	Actinomyces sp. COT-374	10	

Table 2. Taxa and Clones per subject

Subject ID	Sites / subject	Taxa / subject	Clones / subject	Pet	Type pet
1033	4	50	150	yes	3 dogs
1049	3	48	224	yes	lizard
1139	3	44	264	yes	2 dogs 1 cat
1027	3	28	49	yes	3 dogs and 2 snakes
1052	4	23	40	no	
1039	3	22	40	yes	dog
1089	3	17	45	yes	2 cats
1088	3	16	56	yes	dog and cat
HV1	10	15	38	yes	dog
1100	3	9	18	yes	dog
1022	4	9	15	no	-
1028	4	8	10	yes	3 dogs and 2 snakes
HV10	4	6	9	yes	2 cats
1104	3	5	8	no	-
1115	4	5	8	yes	hamster
1048	3	5	7	yes	lizard
1019	4	2	22	no	-
1	1	2	2	Unknown	
HV4	2	2	2	yes	2 dogs
HV7	4	1	36	no	-
HV2	1	1	4	no	-
1031	1	1	2	no	-
1036	2	1	1	no	-
1050	2	1	1	yes	lizard
1113	3	1	1	no	-
1121	1	1	1	no	-
1137	3	1	1	yes	fish

Table 3. Taxa and clones per site

| Study	Subject Isolation Sites	Clones / study +	Clones / study -	Taxa / site +	Taxa / site -	Clones / site +	Clones / site -	Subjects / site +	Subjects / site -
	Includes shared human taxa & clones								
1	antecubital fossa (inner elbow) skin	2	2	2	2	2	2	1	1
2	skin, volar forearm	1067	963	91	82	655	605	27	15
2	skin, antecubital fossa	1067	963	60	54	216	197	23	16
2	skin, popliteal fossa	1067	963	45	39	173	157	20	18
2	skin, nare	1067	963	7	4	23	4	9	3
3	skin, interdigital web space	116	91	10	9	35	34	3	3
3	skin, antecubital fossa	116	91	9	6	13	10	4	2
3	skin, volar forearm	116	91	5	5	18	12	5	4
3	skin, toe web space	116	91	2	2	2	2	2	2
3	skin, occiput	116	91	2	1	2	1	2	1
3	skin, hypothenar palm	116	91	2	1	24	23	3	2
3	skin, buttock	116	91	1	1	1	1	1	1
3	skin, elbow	116	91	1	1	2	2	2	2
3	skin, plantar heel	116	91	1	1	2	2	1	1
3	skin, glabella	116	91	1	1	1	1	1	1
3	skin, alar crease	116	91	1	1	1	1	1	1
3	skin, nare	116	91	1	1	1	1	1	1
3	skin, back	116	91	1	1	1	1	1	1
3	skin, axillary vault	116	91	2	0	10	0	1	0
3	skin, external auditory canal	116	91	1	0	1	0	1	0
3	skin, umbilicus	116	91	1	0	2	0	1	0
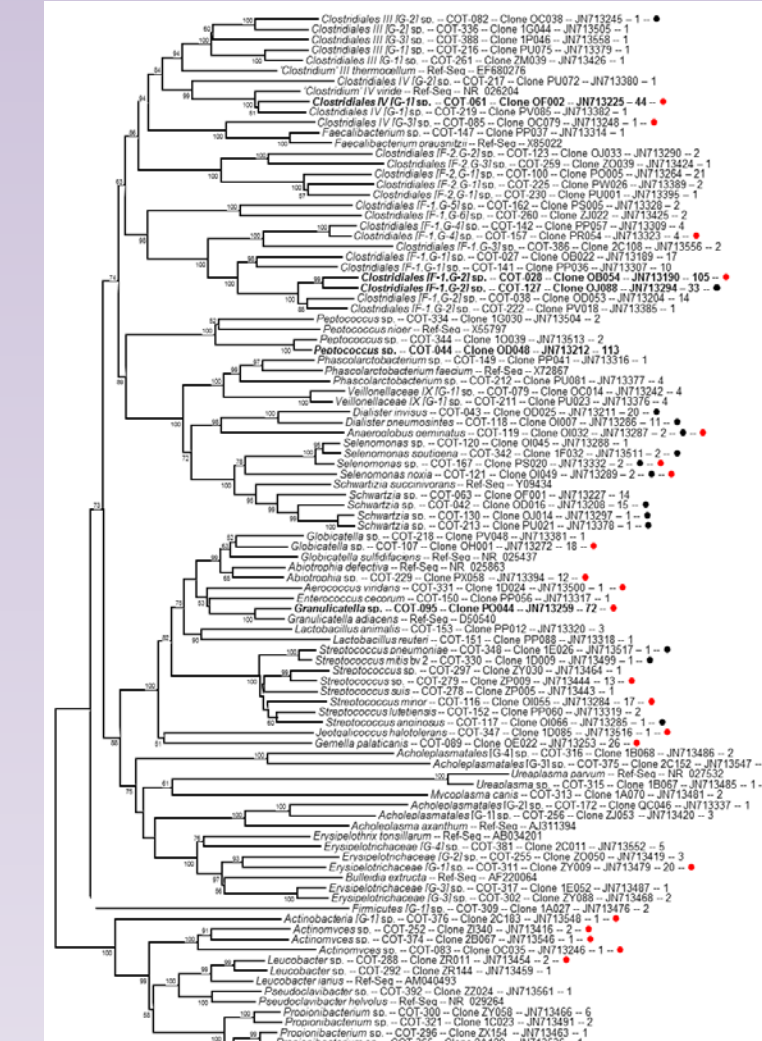
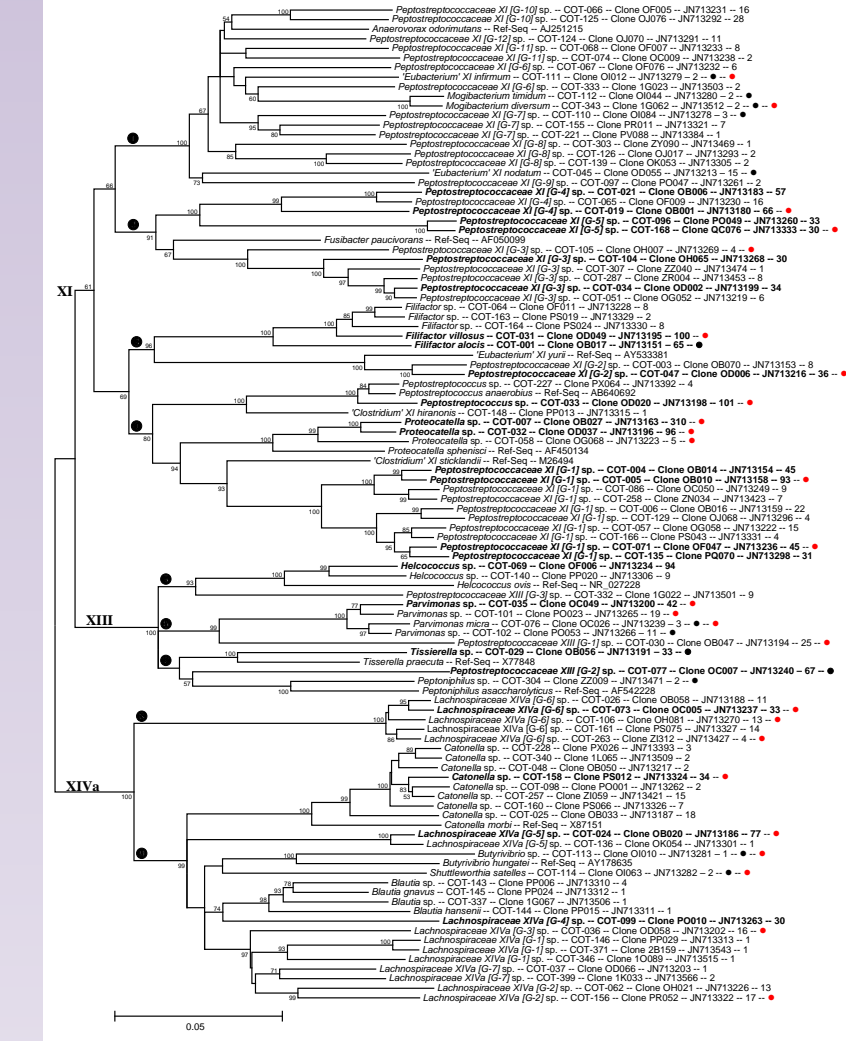**Fig. 1.** *Firmicutes, Actinobacteria & Tenericutes.*

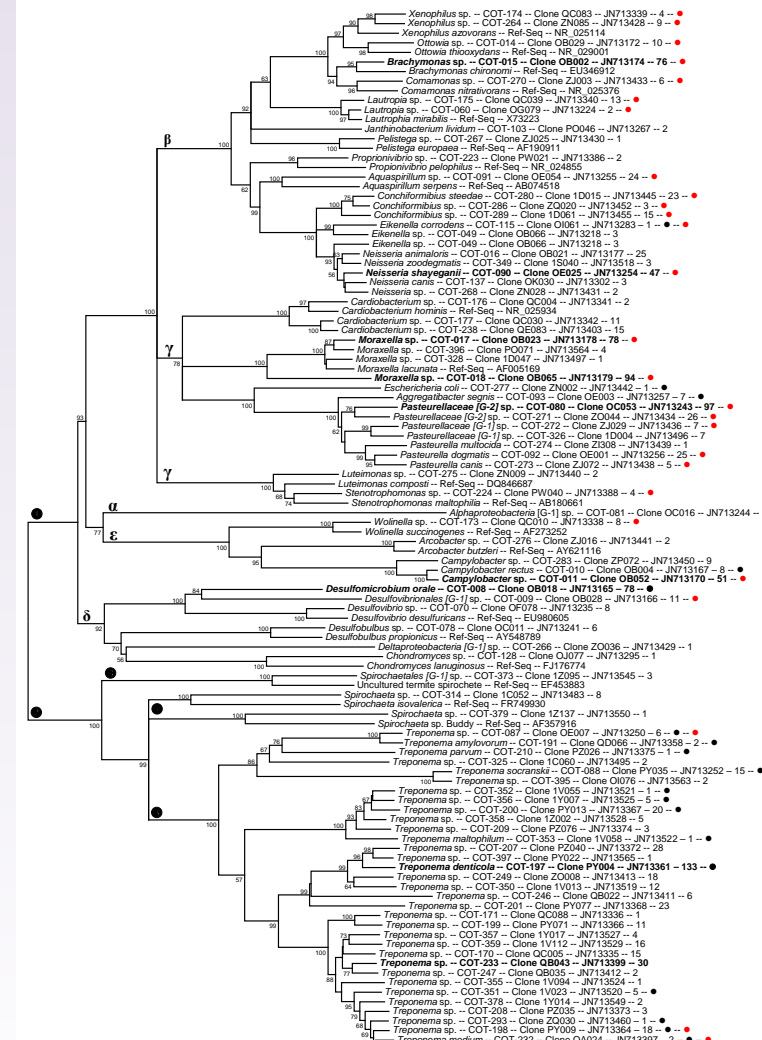**Fig. 2.** *Clostridia: Peptostreptococceae & Lachnoslpiraceae.*

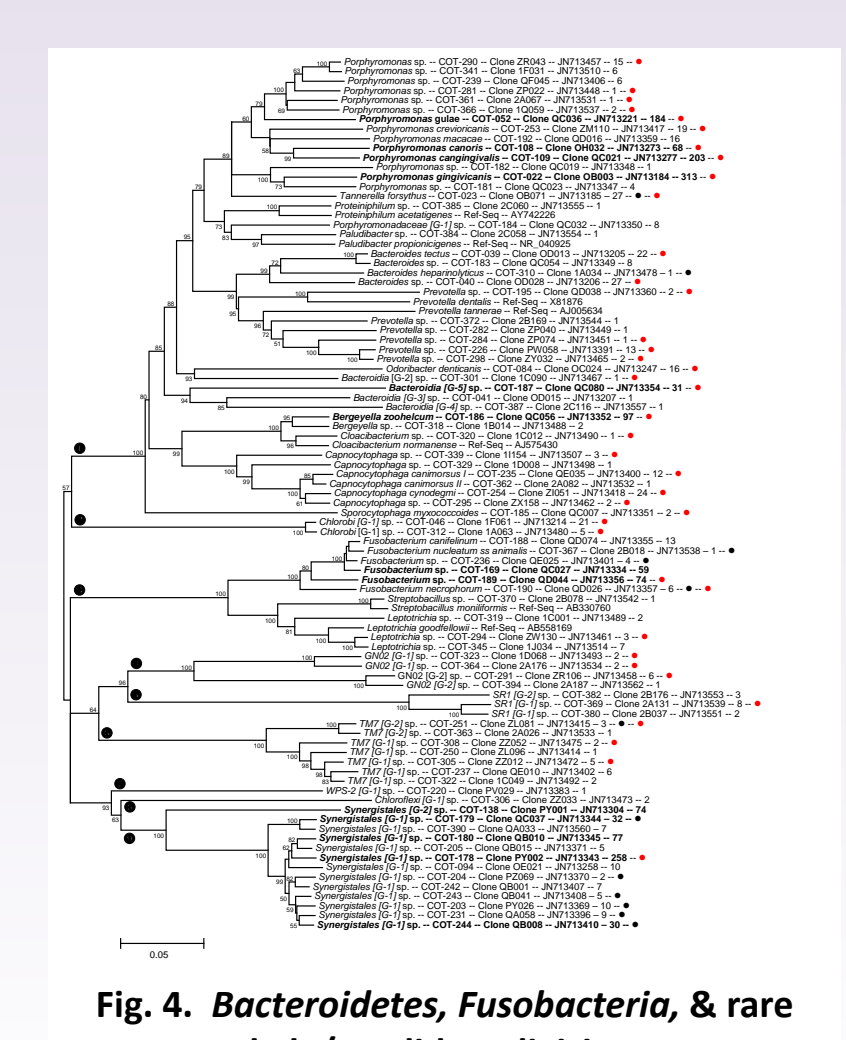**Fig. 3.** *Proteobacteria & Spirochaetes.*

Fig. 4. *Bacteroidetes, Fusobacteria,* & rare phyla/candidate divisions.

Results

BLASTN analysis of 417 full length 16S rRNA references canine sequences representing 353 canine oral taxa produce 7,272 hits >98.5% identity. Limiting to human source produced 3,858 hits. Limiting to skin sites reduce hits to 1,592. Forty-seven sequences that did not align over their full lengths were eliminated. Finally, de-replicating hits to different reference sequences of the same canine oral taxon produce a final total of 1,185 sequence hits. The 118 canine oral taxa were seen in 41 subjects. The rank abundance of the 28 most common canine oral taxa on skin sites is shown in Table 1. *Porphyromonas cangingivalis* was the most common with 101 clones. Fifteen canine oral taxa with a total of 128 clones are shared with the human oral cavity (98.5% sequence similarity) leaving 103 unique canine taxa with 1,057 clones. Shown in Table 2 are the canine unique taxa and clones for each subject . Twelve of the 41 subjects had only taxa shared with the human oral cavity, leaving 29 subject in Table 2. Subject 1033 has 3 dogs and had 150 clones from 50 canine taxa. Shown in Table 3 are the number of canine taxa and clones seen by human sampling site. The majority of canine taxa and clones are on exposed arm and hand sites The phylogenetic distribution of the canine oral taxa found in the human skin libraries is shown in Figures 1-4 by red dots. Taxa found in humans have black dots. These figures are modified from reference (2).

Discussion

With over 1/3 of U.S. households having one or more dogs, it is not surprising that skin swabs from human microbiome studies contain dog oral bacteria. The subjects with the highest number of canine taxa and clones reported dog ownership. While possession of canine oral bacteria on human skin generally correlates with dog ownership, the metadata does not contain information of whether subjects were licked by dogs, regardless of ownership. Thus, while it seems likely that the subjects with 10 to 50 canine oral taxa were likely licked by a dog, that specific conclusion will have to await future studies designed to address transmission through licking. The human health impact of being licked by a dog, while generally thought to benign, deserves further study. Dog bites, which transfer canine oral bacteria across the epithelial barrier are known to cause serious infections. An important question in human microbiome studies is determining which organisms are endogenous to a particular body site, and which organisms are transients from the environment. This distinction is particularly difficult with the skin sites which are directly exposed to the environment. The potential exposure of human skin to the endogenous microbiomes of other mammals further complicates the task of identifying endogenous human skin bacteria.

Acknowledgements

References

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