COMPOSITION AND DIVERSITY OF MICROBIOME IN CERVICAL TUMORS INFECTED WITH HPV16, HPV18, HPV45 AND IN MULTIPLE INFETIONS

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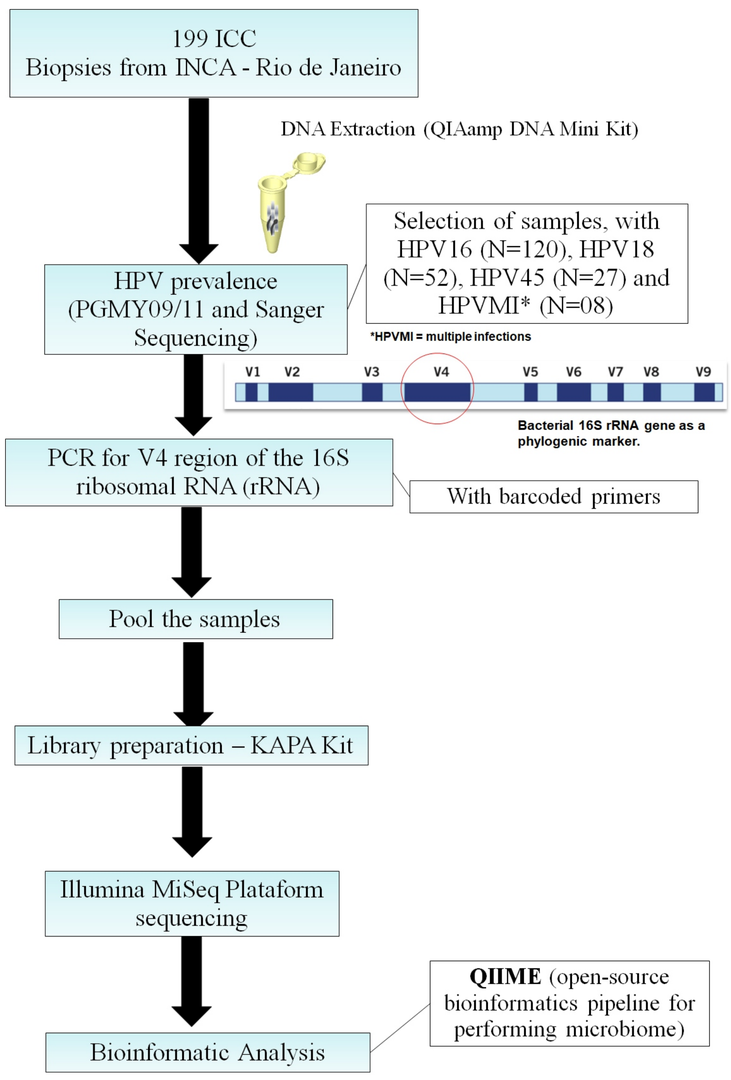
INTRODUCTION

Despite all progress regarding HPV-induced cervical carcinogenesis, risk factors for HPV-infection persistence and clearance are still unclear. Recent data suggests cervico-vaginal microbiota as an important element influencing HPV acquisition, persistence and cervical lesion progressions. Characterize microbiota diversity in cervical specimens is one of the initial steps to understand the mechanisms involved in the cervical cancer pathogenesis.

GOALS

Thus, our goal was to describe the microbiome present in invasive cervical cancer (ICC) biopsies of patients from Rio de Janeiro, Brazil, associating with clinical and biological characteristics.

METTHODS



RESULTS

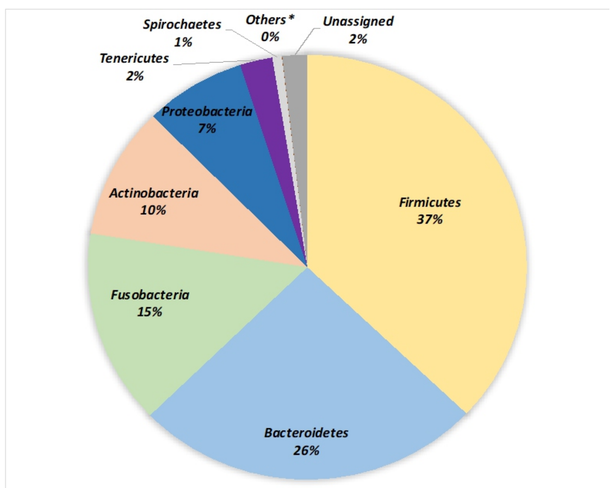


Figure 1. Composition of the most abundant phyla of bacteria present in the DNA extracted from cervical cancer biopsies.

Table 1. HPV prevalence in ICC in a Brazilian cohort from Rio de Janeiro. Samples were randomly selected from the three most prevalent HPV infections.

HPV type	N	%
16	**370**	**62.3**
18	**77**	**13.0**
45	**33**	**5.6**
35	12	2.0
58	11	1.9
52	8	1.3
73	8	1.3
31	7	1.2
33	7	1.2
39	6	1.0
59	6	1.0
26	2	0.3
51	2	0.3
56	2	0.3
68	2	0.3
83	1	0.2
Co-infection	**20**	**3.4**
Undetermined	20	3.4
Total	594	100.0

Table 2. Average of relative abundance measures of the most dominant bacteria genera in all selected samples (N=144) grouped by the seven most frequent phyla.

Phyla	Genera	Abundance (%)*
Bacteroidetes	*Prevotella*	16.38%
	Porphyromonas	4.48%
	Bacteroides	2.92%
Firmicutes	*Lactobacillus*	10.37%
	Streptococcus	3.44%
	Peptoniphilus	2.85%
	Dialister	1.92%
	Parvimonas	1.74%
	Megasphaera	1.58%
	Anaerococcus	1.36%
	Clostridium	1.29%
	Anaerococcus	1.16%
	Veillonella	1.05%
	Peptostreptococcus	0.93%
	Finegoldia	0.83%
	WAL_1855D	0.80%
	Staphylococcus	0.70%
	Moryella	0.60%
Fusobacteria	*Fusobacterium*	6.67%
	Sneathia	5.19%
	Fusobacterium	2.70%
Actinobacteria	*Gardnerella*	5.85%
	Atopobium	1.54%
	Corynebacterium	0.52%
Proteobacteria	*Campylobacter*	2.27%
	Acinetobacter	0.93%
	Haemophilus	0.84%
Tenericutes	*Ureaplasma*	1.23%
	Mycoplasma	0.89%
Spirochaetes	*Treponema*	0.60%
Unassigned/Others	-	16.00%
Total		**100.00%**

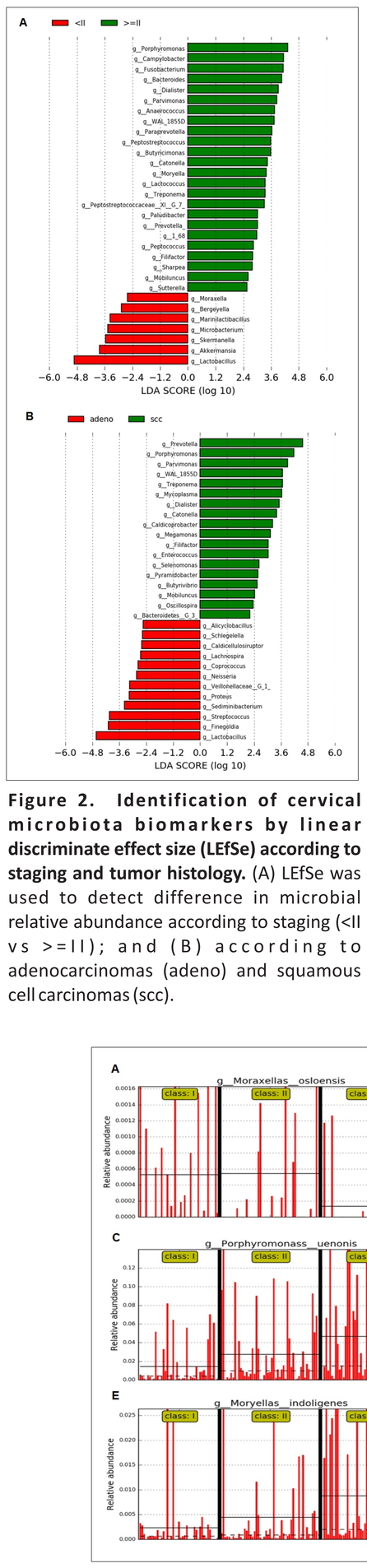


Figure 2. Identification of cervical microbiota biomarkers by linear discriminate effect size (LEfSe) according to staging and tumor histology. (A) LEfSe was used to detect difference in microbial relative abundance according to staging (<II vs >=II); and (B) according to adenocarcinomas (adeno) and squamous cell carcinomas (scc).

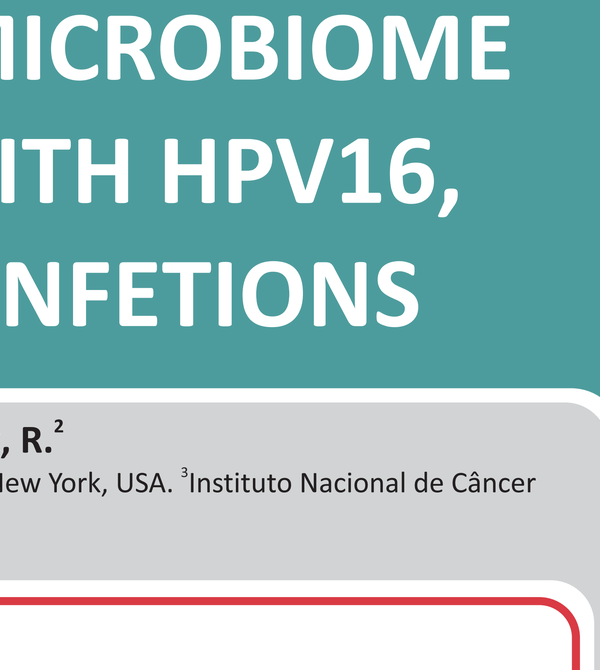


Figure 3. Apha diversity by HPV type and staging. HPV45 positive samples had statistically higher level of diversity (Observed) than HPV16 and HPV18 positive cancers (p=0.0059 and 0.0001). Advanced FIGO stages (>=II) displayed higher Observed and Shannon diversity (p=0.0403 and 0.0009, respectively). Statistical significance was assessed by ANOVA test.

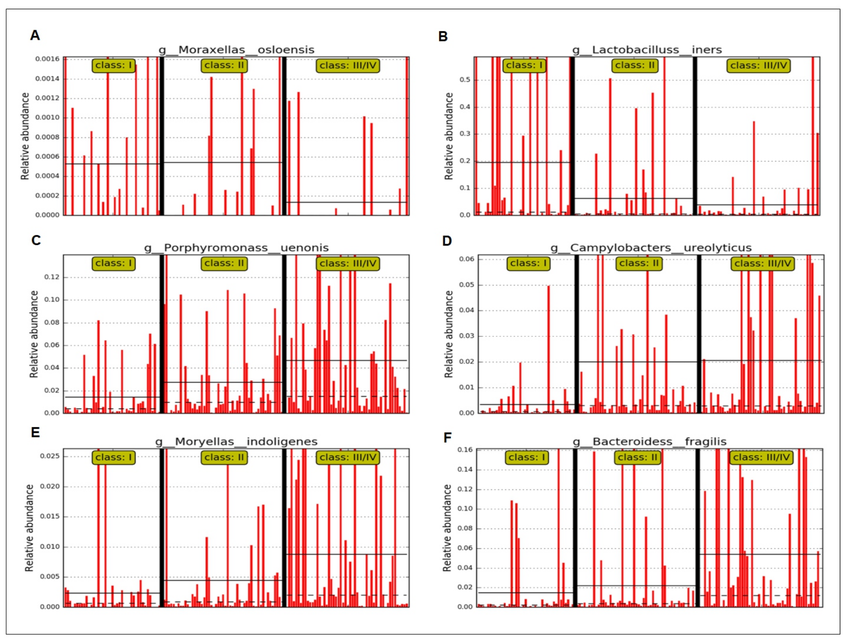


Figure 4. Histogram showing distribution of relative abundance counts of bacteria species identified by LEfSe as biomarkers of early staging (A and B) and advanced staging (C, D, E and F). Higher abundance of Moraxellas osloensis (A) and Lactobacillus iners (B) in stages I and II in comparison with III/IV was observed. On the other hand, *Porphyromonas uenonis* (C), *Campylobacter ureolyticus* (D), *Moryellas indoligenes* (E) and *Bacteroides fragilis* were most abundant in stages II and III/IV in comparison with stage

CONCLUSION

Preliminary results shows:

- We are able to identify microbiome in DNA from tissues,
- Point out differences of diversity by HPV types,
- and by HPV staging,
- Suggest that specific microbiota can be used for discrimination of tumor histology and staging.

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