Review of: "Analysis of colonic mucosa-associated microbiota using endoscopically collected lavage"

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Miyauchi et al., presents a comprehensive analysis to assess the use f colon lavage as a representative sample to study mucosal microbiota. Human studies are mainly based on fecal sample, which is a challenge to capture true host-microbial interactions. This is also reason behind that recent up take in large animal model studies to uncover true host-microbial interactions affecting host health. While it has its own merits, I think the way that the authors approached it has diluted the importance and significances to the microbiome research. I believe the comparison between smokers and non-smokers could have been used to support you speculations but should not have been a main focus.

In addition, I think it is a good idea to acknowledge that colon is not necessarily the suitable region to study host-microbial interactions and its impact on health. How would we able to develop and use this knowledge to study the small intestine? It is probably better to define lavage microbiota as a proxy to study mucosal microbiota.

Did the authors collected 3 different samples at the same time

I think the higher abundance of 16S rRNA DNA in lavage samples could be simply due to the less host DNA in your samples

Generally, when preparing PCR products for sequencing from biopsy and brush samples (which has high host DNA) need a nested PCR to enrich bacterial DNA. So, it is no wonder that authors seeing higher % of low quality and chimeric reads from those samples. It is questionable whether you are successful getting enough bacterial PCR products to go forward with sequencing. So, given the approach used in the current study to profile microbiota from biopsy samples, I do not think it is reasonable to conclude that biopsy samples generate unwanted sequences or mitochondrial sequences. In large animal models, with the use of nested-PCR they have generated consistent results on mucosal microbiota.