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Modulatory effect of faecal microbiota transplantation on procarcinogenic colibactin-producing bacteria in patients with recurrent *Clostridioides difficile* infection

Introduction

Faecal microbiota transplantation (FMT) is an established treatment for recurrent *C. difficile* infections (rCDI). Recently, it was shown that *Escherichia coli* may produce genotoxic colibactin, which is encoded on the pks gene island that holds 19 *clb* genes. Also, pks$^+$ *E. coli* may be transmitted or cleared after FMT. We studied deep sequenced faecal microbiomes of healthy faeces donors and rCDI patients for pks genes and the possibility of transmission to patients and clearance by negative donor faeces after FMT.

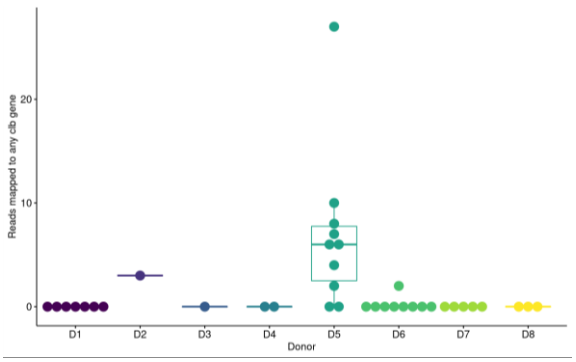

Figure 1, pks is present in faecal samples of three out of eight donors. Five donors never have pks genes in their faeces, three donors did have at least one pks-positive faecal sample. Pks values are measured as absolute number of reads matched to the reference.

Materials and methods

CDI patient faeces

× **49**

Healthy donor faeces

× **38** (from 9 donors)

Patient faeces after FMT

× **49**

49 sample triplets → **136** deep sequenced metagenomes

Method details

Thirty-eight donor faecal suspensions were used to treat 49 patients (some suspensions were used for multiple patients). Metagenomes were deep sequenced by Vedanta Biosciences, Inc. (Cambridge, MA, USA) to 20M+ Illumina 150-PE reads. Metagenomes were analysed with Jovian (https://doi.org/10.5281/zenodo.3666156) and *clb* reads were detected by mapping cleaned reads to a reference sequence of the pks island: accession ID AM229678 from GenBank.

Results

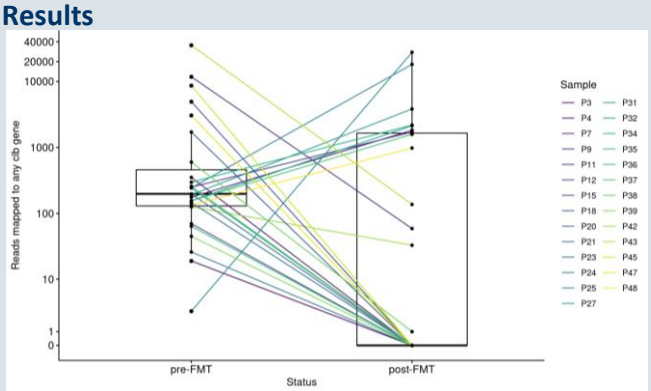

Figure 2a, patients with pks before FMT may lose pks after FMT. Twenty-seven patients were pks-positive before FMT, of which 13 are still pks-positive after FMT and 14 patients lose pks after FMT. All but one of the patients who lose pks were treated with pks-negative donor material. Patients whose pks levels decrease were mostly treated with pks-negative donor material and patients whose pks levels rise were mostly treated with pks-positive donor samples.

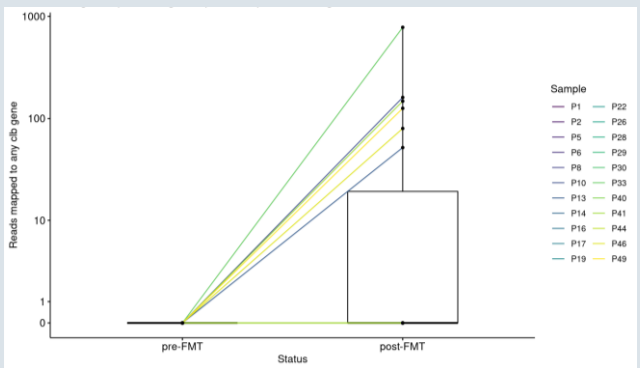

Figure 2b, patients with no pks before FMT may obtain pks after FMT, although they often remain pks-negative. Twenty-two patients had no pks prior to FMT. Sixteen of them remain negative after FMT, whereas six patients have detectable pks after FMT. All six were treated by negative donor samples. Pks values are measured as absolute number of reads matched to the reference – scales are non-linear.

Conclusions

- Pks is often absent in healthy donors (74% of samples)
- Pks is prevalent in rCDI patients (55% of patients)
- In rCDI patients, pks generally decreases after FMT
- Donor pks status is not always predictive for pks status in recipients after FMT

NDFB Nederlandse Donor Feces Bank

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