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Hot Topic

Fifty shades of graft: How to improve the efficacy of faecal microbiota transplantation for decolonization of antibiotic-resistant bacteria

Benjamin Davido[a,*], Rui Batista[b], Aurélien Dinh[a], Pierre de Truchis[a], E.M. Terveer[c,d], Bruce Roberts[e], Ed J. Kuijper[c,d], Silvia Caballero[e]

[a] Maladies Infectieuses, Hôpital Universitaire Raymond-Poincaré, Groupe Hospitalier Paris Ile de France Ouest, AP-HP, Garches, France
[b] Pharmacie Hospitalière, Hôpital Universitaire Cochin, AP-HP, Paris, France
[c] Department of Medical Microbiology, Leiden, The Netherlands
[d] Netherlands Donor Faeces Bank at Leiden University Medical Centre, Leiden, The Netherlands
[e] Vedanta Biosciences Inc., Cambridge, MA, USA

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ABSTRACT

Background: Spontaneous decolonization of antibiotic-resistant bacteria (ARB) takes time: approximately 25% after 30 days for carbapenem-producing Enterobacteriaceae or extended-spectrum beta-lactamase-producing Enterobacteriaceae. Faecal microbiota transplantation (FMT) has been proposed as a new strategy to promote decolonization in order to reduce the risk of superinfection due to these ARB. This paper discusses the literature on the use of FMT for this indication, and the improvement levers available to promote its efficacy.

Methods: Literature available to date concerning the use of FMT to eradicate ARB was reviewed, and the different factors that may have influenced the efficacy of decolonization were evaluated.

Results: Four axes that could have played major roles in the efficacy of FMT were identified: bowel preparation before FMT; donor; dose; and thermal conditioning of faeces. The positive or negative impact of each on the outcome of FMT is discussed.

Conclusion: Although FMT is very efficient for the eradication of *Clostridium difficile*, the same 'recipe' cannot be used for the eradication of ARB. Working together with expert centres may help to improve the efficacy of FMT for this indication, and enable the reduction of in-hospital isolation precautions.

1. Introduction

Over the past century, bacterial resistance to antibiotics has proven to be ineluctable, resulting in the need to develop new antimicrobial strategies. The European Centre for Disease Prevention and Control reported that antibiotic-resistant bacteria (ARB) accounted for approximately 33,000 deaths in 2015 in Europe [1]. These data particularly concern multi-drug-resistant and CTX-M extended-spectrum beta-lactamase (ESBL)-producing Enterobacteriaceae, which have spread to endemic levels worldwide and constitute a real threat. To prevent spread of ARB in hospitals, patients colonized with drug-resistant bacteria are nursed in isolation with specific infection control measures. Additionally, digestive tract colonization due to ESBL-producing Enterobacteriaceae is a risk factor for individual patients to develop infection with ARB [2]. Unfortunately, spontaneous decolonization takes time: approximately 25% after 30 days for carbapenem-producing Enterobacteriaceae (CPE) or ESBL-producing Enterobacteriaceae [3], and up to 50% after 50 days for CPE or vancomycin-resistant enterococci (VRE) [4]. Moreover, oral decontamination with antibiotics such as colistin or aminoglycosides is potentially associated with further development of antimicrobial resistance [5,6]. As these drugs, especially colistin, are deemed to be 'last-resort antibiotics', extra care should be taken when using them for a non-systemic infection.

There is an urgent need to identify new decolonization strategies to decrease the rate of ARB-colonized patients. In 2013, Van Nood et al. reported the high efficacy (approximately 90%) of faecal microbiota transplantation (FMT) against *Clostridium difficile* infection (CDI) [7]. This publication led to newfound interest in FMT in humans as a new weapon to fight ARB, but with heterogeneous results. For instance, Dubberke et al. studied a population of dysbiotic patients suffering from CDI and colonized with VRE ($n = 8$), and reported that FMT resulted in eradication

* Corresponding author. Address: Service de Maladies Infectieuses et Tropicales, Hôpital Raymond-Poincaré, Garches 92380, France. Tel.: +33 1 47107758; fax: +33 1 47107790.
E-mail address: benjamin.davido@aphp.fr (B. Davido).

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Table 1
Summary of case series and studies performed regarding faecal microbiota transplantation and gut decolonization in patients harbouring antibiotic-resistant bacteria.

Authors	Year	Country	*n*	Study design	Targeted micro-organism(s)	Stool quantity	Stool type	Use of antibiotics
Huttner et al. [11]	2019	France, Switzerland, Netherlands and Israel	39	Randomized open-label trial	Mainly ESBL, some CRE	15–40 g	Frozen	Oral colistin + neomycin
Saïdani et al. [12]	2018	France	10	Case–control	CRE and *Acinetobacter* spp.	50 g	Fresh	Oral colistin + aminoglycoside or sulfadiazine + fusidic acid
Davido et al. [9]	2018	France	8	Uncontrolled trial	VRE	50 g	Frozen	No
Singh et al. [14]	2018	Netherlands	15	Uncontrolled trial	ESBL *E. coli* or *K. pneumoniae*	50 g	Fresh	No
Dinh et al. [10]	2018	France	17	Uncontrolled trial	CRE versus VRE	50 g	Frozen	No
Davido et al. [9]	2017	France	8	Uncontrolled trial	Mainly CRE, some VRE	50 g	Frozen	No
Innes et al. [20]	2017	UK	1	Case report	ESBL *E. coli* + *C. difficile*	100 mL	Frozen	Oral vancomycin
Stalenhoef et al. [16]	2017	Netherlands	1	Case report	ESBL *E. coli* and *Pseudomonas aeruginosa*	75 g	Fresh	Intravenous colistin
Bilinski et al. [13]	2017	Poland	20	Uncontrolled trial	Mainly ESBL/CRE, rare VRE	100 g	Fresh	No
Biliński et al. [21]	2016	Poland	1	Case report	*K. pneumoniae* CRE with ESBL *E. coli*	100 g	Fresh	No
Dubberke et al. [8]	2016	USA	8	Uncontrolled trial	VRE + *C. difficile*	50 g	Frozen	Oral vancomycin
García-Fernández et al. [22]	2016	Spain	1	Case report	*K. oxytoca* CRE + *C. difficile*	100 g	Fresh	Oral vancomycin
Sohn et al. [23]	2016	Korea	3	Case series	VRE + *C. difficile*	100 g	Fresh	Oral vancomycin
Crum-Cianflone et al. [24]	2015	USA	1	Case report	CRE + MRSA + *C. difficile, Acinetobacter* spp. and *Pseudomonas aeruginosa*	480 mL	Fresh	Oral vancomycin
Jang et al. [25]	2015	Korea	1	Case report	VRE + *C. difficile*	300 g	Fresh	Oral vancomycin + metronidazole
Lagier et al. [26]	2015	France	1	Case report	*K. pneumoniae* CRE	50 g	Fresh	Oral colisitin + gentamicin
Stripling et al. [27]	2015	UK	1	Case report	VRE + *C. difficile*	25–30 g	Fresh	Oral vancomycin
Wei et al. [28]	2015	China	5	Case series	MRSA + *C. difficile*	60 g	Fresh	Oral vancomycin or intravenous ceftriaxone-tazobactam
Singh et al. [19]	2014	Netherlands	1	Case report	ESBL *E. coli* + *C. difficile*	50 g	Fresh	Oral vancomycin

ESBL, extended-spectrum beta-lactamase; CRE, carbapenem-resistant Enterobacteriaceae; VRE, vancomycin-resistant enterococci; MRSA, meticillin-resistant *Staphylococcus aureus; E. coli, Escherichia coli; K. pneumoniae, Klebsiella pneumoniae; C. difficile, Clostridium difficile; K. oxytoca, Klebsiella oxytoca*.

of VRE in six patients (75%) after 30 days [8]. In addition, Davido et al. showed that FMT had moderate decolonization efficacy (50%) after 3 weeks in a non-CDI population, including patients with chronic kidney failure colonized with VRE ($n = 8$) [9] or CPE ($n = 9$) [10]. Currently, nine clinical trials have been reported on clinicaltrial.gov to eradicate ARB with FMT (NCT03367910, NCT03029078, NCT03167398, NCT03479710, NCT02543866, NCT03391674, NCT02906774, NCT02472600, NCT02312986). The only randomized controlled trial to date failed to demonstrate non-inferiority of FMT to decolonize ARB (composed of ESBL carriers including 27% co-colonized with CRE) [11]. However, this trial had two main limitations: it did not include the targeted number of patients ($n = 64$) because of delay (only recruited 39 patients), and oral antimicrobial therapy was used before FMT.

Table 1 provides details of all studies on the use of FMT for decolonization of ARB published to date.

1.1. Impact of bowel preparation

Huttner et al. used a 5-day course of oral antimicrobial therapy prior to FMT ($n = 21$), and observed a paradoxal regrowth of ARB from 23.8% before FMT to 57.1% after 30 days. The real impact of FMT alone cannot be studied in this trial as the control group ($n = 16$) did not receive oral antibiotic therapy. Saïdani et al. [12] also used oral antimicrobial therapy prior to FMT, but without a control group.

Of note, Huttner et al. did not perform bowel lavage before FMT, as recommended by Van Nood et al. [7]. The potential impact of bowel lavage on decolonization cannot be assessed as no controlled study on this point is available to date.

1.2. Possible dose effect

Bilinski et al. reported 60% efficacy of FMT after 30 days in patients with blood disorders ($n = 20$), mainly co-colonized with ESBL/CPE or VRE ($n = 2$) [13]. However, it should be noted that FMT was repeated in some patients (25 FMT for 20 patients), which enhanced the decolonization outcome.

Most studies used a single FMT dose containing 50 g of fresh faeces, as recommended in the original publication by Van Nood et al. [7]. However, this dose has only been evaluated for CDI. The present authors used a single dose of FMT in their studies on ARB decolonization [9,10].

More recently, Singh et al. [14] attempted to eradicate ESBL colonization in 15 patients, but failed to achieve acceptable findings using a single dose of FMT (efficacy of 20% after 30 days). Interestingly, when two doses of FMT were given ($n = 7$), patients had a two-fold increased chance of being decolonized (40%). Likewise, in a single-centre study ($n = 10$), Saïdani et al. [12] observed increased efficacy of FMT against CPE from 40% to 80% after a second dose of FMT (total 100 g).

Conversely, Huttner et al. [11] used two different doses of faeces in the same intervention group. Sixteen patients received capsulized faeces for 2 days (derived from a total of 15–30 g of faecal material), and six patients received 40 g (lower dose than recommended) via a nasogastric approach. This may have played a role in the unexpected findings of the clinical trial, but this cannot be examined further as the data were pooled.

1.3. Possible donor impact

Importantly, although Singh et al. [14] found that FMT had low efficacy to eradicate ESBL-producing Enterobacteriaceae from the

intestinal tract, one of two donors seemed to be associated with better ARB decolonization efficacy ($n = 3/6$ vs n = 3/16; not significant). The idea of a donor-dependent response makes sense when one considers the findings of Leung et al. [15]. Out of eight FMT donor–recipient pairs, they identified 37 and 95 antimicrobial resistance genes that were acquired by or removed from FMT recipients, respectively [15]. This finding supports the suggestion that some donors may be more suitable to eradicate antimicrobial resistance genes in the recipient's microbiota, such as ARB. Likewise, Bilinski et al. [13] observed better eradication of ARB with donors showing higher microbial richness.

The present authors are currently investigating the impact of the donor using datasets from their FMT clinical study [10] [FEDeX study registered at clinicaltrial.gov (NCT03029078)]. Preliminary data suggest that there was no difference between the 'best' donor (success rate 6/9) and the other four donors (success rate 4/12) ($P = 0.19$) in the eradication of CPE and VRE. This observation can be explained by the lack of power due to the small sample size of these uncontrolled trials using FMT to decolonize ARB.

In addition, microbiological failures to eradicate ARB colonization might be explained by the absence of significant changes in microbiota diversity between donor and recipient, before and after FMT, as reported by Stalenhoef et al. [16].

Therefore, to circumvent this donor-dependent effect, a substudy is being conducted of Patients 20–60 using a universal donor from the Netherlands Donor Faeces Bank. Donor selection was based on experimental testing using animal models of CRE and VRE decolonization, whereby antibiotic-treated mice densely colonized with either ARB were administered with faecal material from various FMT donors. Consistent with clinical findings, variability in FMT-mediated decolonization was observed, and a donor enriched in both CRE and VRE decolonization activity, as determined by complete CRE/VRE clearance in 80% of treated animals, was selected. To date, it is unclear if these data can be extrapolated to humans. However, the authors are confident that these preclinical models will shed new light on the donor–recipient impact in FMT by allowing the selection of donors with the highest anti-ARB activity which are most likely to provide a strong clinical benefit.

1.4. Fresh or frozen faeces

The authors used frozen faeces in their previous studies [9,10], as did Huttner et al. [11], in order to simplify the faeces donation process and to speed up FMT administration. From the authors' experience, frozen faeces maintained at -80°C is viable for up to 12 months without the quality of the graft being affected (data not shown). However, it is unclear whether the FMT conditions for CDI also apply to ARB decolonization, as shown by Lee et al. in a randomized clinical trial ($n = 219$) [17].

1.5. Use of antibiotic course before FMT

Oral decontamination with antibiotics such as colistin or aminoglycosides has been proposed to enhance the efficacy of FMT, as suggested by Saïdani et al. [12] and Huttner et al. [11]. However, data are lacking to support that such antibiotic induction before FMT plays a major role in the observed findings. Incidentally, the latter trial failed to support the benefit of FMT compared with placebo. Moreover, to prove the value of these regimens, including prior use of last-resort intravenous antibiotics such as colistin [16], studies with two groups comparing FMT with FMT plus a course of antibiotics are needed. The authors' position would be to minimize the use of such treatment to avoid regrowth with more highly resistant pathogens. Nevertheless, it could be worth proposing an induction when alternatives to antibiotics are well documented. Indeed, as suggested *in vitro* and in a mice model, repurposing niclosamide could be insightful before performing FMT [18].

2. Perspectives

Another interesting approach is to prevent colonization by performing autologous transplantation with faeces preserved before the occurrence of colonization in high-risk patients, such as haematological patients. This would reduce the risk of superinfection in such patients [2], and would prevent in-hospital epidemics, as described previously with a well-contained outbreak of VRE [9]. Of note, caution should be exercised concerning potential unintended consequences of FMT, especially transmission of infections and long-term consequences of microbiota alteration. In January 2019, the French Group of Fecal Microbiota Transplantation started to evaluate the relevance of FMT for the decolonization of ARB as a compassionate treatment. Regarding ethical queries, a committee composed of three physicians and one pharmacist decides whether or not FMT is indicated based on the literature and the targeted condition.

Finally, FMT raises one very important question: can isolation precautions be reduced for decolonized patients in hospital wards, thus avoiding the detrimental effects of quarantine? The answer clearly depends on the overall efficacy of FMT as an infection control strategy, and demonstration that patients are not subject to recolonization over time, especially if they will undergo antimicrobial therapy.

3. Conclusion

Overall, studies concerning ARB decolonization using FMT require more harmonization for consistent efficacy. It seems that the same 'recipe' used for the eradication of *C. difficile* cannot be applied for the decolonization of ARB. Working together with expert centres may help to improve the efficacy of FMT for this indication. Randomized controlled trials without previous oral decontamination are needed to answer these questions, with long-term patient follow-up.

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Competing interests

Bruce Roberts and Silvia Cabellero are employees of Vendanta Biosciences. The remaining authors declare no competing interests.

Ethical approval

Not required.

Authors' contributions

BD, AD and EK designed the paper. BD and PDT prepared the 1st draft of the manuscript. SC, RB, PDT and ET revised the 2nd version of the manuscript. BR and SC were responsible of editing and proofreading the entire manuscript. All the authors participated in manuscript preparation and approved the final manuscript for publications.

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