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## Recipient factors in faecal microbiota transplantation: one stool does not fit all

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1 *NRGH Perspective*

2 **Title: Recipient factors in faecal microbiota transplantation: one stool does not fit all**

3

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17

18 Abstract:

19 Fecal microbiota transplantation is a promising therapy for chronic diseases associated with gut  
20 microbiota alterations. FMT cures 90% of recurrent *C. difficile* infections. However, in complex  
21 diseases, such as inflammatory bowel disease, irritable bowel syndrome and metabolic syndrome, its  
22 efficacy remains variable. It is accepted that donor selection and sample administration are key  
23 determinants of FMT success, and great effort has been made to standardize these procedures.  
24 However, little is known about recipient factors impacting FMT success and long-term clinical  
25 improvement. In this review, we discuss the effects of the recipient parameters on donor microbiota  
26 engraftment and clinical efficacy. Especially, emerging evidence supports that controlling the  
27 inflammation level in the recipient's intestine might facilitate engraftment by reducing host immune  
28 system pressure on the newly transferred microbiota. Deciphering FMT engraftment rules and  
29 developing novel therapeutic strategies are priorities to alleviate the burden of chronic diseases  
30 associated with an altered gut microbiota.

31

32

### 33 **Introduction**

34           The intestinal microbiota plays major roles in the maintenance of human health, including  
35 functions in nutrition, metabolism, immune development, and host defense. In healthy adults, it is  
36 dominated by Firmicutes and Bacteroidetes, with a minor representation of Proteobacteria,  
37 Actinobacteria, Fusobacteria and Verrucomicrobia. Other microorganisms present, such as viruses,  
38 archaea and fungi, are currently essential subjects of exploration. Despite a conserved "core"  
39 microbiota, each intestine harbors a unique assembly of diverse microbial communities. Both  
40 environmental and host factors shape microbiota composition, leading to considerable variability  
41 among individuals<sup>1</sup>.

42           Numerous diseases are associated with alterations in gut microbiota composition and  
43 function, such as recurrent *Clostridioides difficile* infection (rCDI), inflammatory bowel diseases (IBDs),  
44 including Crohn's disease (CD) and ulcerative colitis (UC), irritable bowel syndrome (IBS), hepatic  
45 encephalopathy (HE), metabolic syndrome, and graft-versus-host disease (GvHD)<sup>2</sup>. The common point  
46 of these alterations is a loss of diversity, with enrichment of some microorganisms and loss of others.  
47 Microbiota transfer experiments in mice suggest a causative role in these conditions<sup>3</sup>. Despite available  
48 treatments, there is still a significant unmet medical need for these diseases. In this context, new  
49 therapeutic strategies targeting the microbiota are currently being developed. Fecal microbiota  
50 transplantation (FMT) consists of the administration of fecal material from a healthy donor into the  
51 intestinal tract of a patient to induce therapeutic effects.

52           FMT has been used for centuries before randomized controlled trials (RCTs) proved its efficacy  
53 for treating rCDI in 90% of cases<sup>4,5</sup>. More recently, positive therapeutic effects were also described in  
54 the induction of remission in UC<sup>6-9</sup>, with a modest success rate (24-32 vs 5-9% for placebo) compared  
55 to the results obtained for rCDI<sup>6,8,9</sup>. Similarly, in CD, small case series, uncontrolled studies and a single  
56 pilot RCT suggest a beneficial effect of FMT<sup>10-15</sup>. In IBS, two out of five RCTs revealed significant  
57 symptom relief at three months in patients receiving FMT compared to that in patients receiving  
58 placebo (65 vs 43%<sup>16</sup>; 89.1 vs 23.6%<sup>17</sup>), together with changes in microbiota profiles<sup>16,18-20,17</sup>. Moreover,

59 patients with GvHD positively respond to FMT, with an improvement in gastrointestinal symptoms and  
60 no adverse effects despite their severely immunocompromised status<sup>21-23</sup>. FMT might also be effective  
61 in extraintestinal disorders, such as recurrent HE<sup>24,25</sup>, metabolic disorders<sup>26,27</sup> and autism<sup>28,29</sup>.

62 Thus, FMT is a promising therapeutic strategy in various diseases associated with altered gut  
63 microbiota, and hundreds of RCTs are currently ongoing. However, the differences in the efficacy of  
64 FMT between rCDI and other disorders demonstrate that the gut microbiota is not the only actor in  
65 many complex diseases. The influence of host and environmental factors on the gut microbiota also  
66 takes place after FMT and might limit its efficacy in multifactorial diseases. In this review, we define  
67 the effects of host factors on the intestinal microbiota. We discuss recipient factors that play crucial  
68 roles in FMT success and identify possible avenues for progress in FMT therapies.

69

70

## 71 **1- Microbiota manipulation by the host**

72

### 73 **1-1 Genetics impacts the microbiota**

74 Genetics affect the intestinal microbiota composition (FIG. 1), as suggested by the higher similarity  
75 between the microbiotas of homozygotic twins than dizygotic twins<sup>30</sup>. As the variability in human  
76 genomes and environmental influences makes it challenging to understand polymorphic mechanisms  
77 regulating the intestinal microbiota, mice have been a valuable tool in microbiota research. For  
78 example, defects in the nucleotide-binding oligomerization domain-containing protein 2 (*Nod2*) gene,  
79 which encodes a receptor for bacterial molecules<sup>31,32</sup>, or in the inflammasome NOD-like receptor family  
80 pyrin domain containing 6 (*Nlrp6*) induce alterations in the gut microbiota composition<sup>33</sup>. *Nod2*<sup>-/-</sup> and  
81 *Nlrp6*<sup>-/-</sup> mice exhibit increased susceptibility to colitis-associated cancer<sup>31,33</sup>. This phenotype is  
82 transmissible to wild-type mice upon cohousing, indicating that an altered microbiota plays a direct  
83 role in disease development<sup>31,33</sup>. Deletion of the caspase recruitment domain family member 9 (*Card9*)  
84 gene, encoding an adaptor protein, indicates that it is responsible for the modification of both

85 microbiota composition and function<sup>34</sup>. Indeed, *Card9*<sup>-/-</sup> mice exhibit a decrease in *Lactobacillus reuteri*  
86 abundance and have a reduced capacity to metabolize tryptophan into aryl hydrocarbon receptor  
87 (AhR) ligands, which leads to increased susceptibility to colitis<sup>34</sup>.

88 Moreover, germ-free mice with different defects in innate (antimicrobial peptides, complement,  
89 pentraxins, and enzymes affecting microbial survival) and adaptive (MHC-dependent and MHC-  
90 independent) immunity shape the gut microbiota differently<sup>35</sup>. Mice lacking the myeloid  
91 differentiation primary response 88 (*MyD88*) gene, encoding an essential signaling protein for both  
92 innate and adaptive immunity, or Toll-like receptor 5 (TLR5) also have an altered intestinal microbiota  
93 composition<sup>36,37</sup>. RAG-deficient mice, which lack T and B cells, have a reduced intestinal microbiota  
94 diversity compared to that of controls<sup>38</sup>, with outgrowth of the mucus-associated bacteria  
95 *Akkermansia muciniphila*<sup>39</sup>. Moreover, these mice exhibit an increased interindividual variability of  
96 their microbial communities, suggesting that adaptive immunity contributes to stabilizing the gut  
97 microbiota<sup>40</sup>.

98 Host genetics also influences the mycobiota. Some of the genes mentioned above are involved in  
99 fungal sensing, such as *Card9*, which controls intestinal fungal loads<sup>41,42</sup>. In addition, the multiple  
100 interactions between bacteria and fungi suggest that variation in bacterial communities impacts fungal  
101 populations<sup>43,42,44</sup>.

102 Altogether, these data highlight the role of immune and regulatory genes in shaping intestinal  
103 microbiota composition and function, with a direct impact on disease development risk.

104

105 In humans, the association of the intestinal taxa with several polymorphisms suggests that host  
106 genetics also impacts gut microbiota composition (Table 1). Heritability analyses have identified four  
107 genetic loci statistically associated with microbial taxa (*Rikenellaceae*, *Faecalibacterium*, *Lachnospira*  
108 and *Eubacterium*) in two cohorts from geographically and culturally different areas<sup>45</sup>. Multiple genetic  
109 variants in innate immunity players (particularly C-type lectins) and vitamin D receptor were associated  
110 with intestinal microbiota composition and function, highlighting their role in gut homeostasis<sup>46,47</sup>.

111 These studies provide clues for genetic effects on the microbiota composition in healthy individuals.  
112 However, a comparison of independent studies highlights the complexity of the genetic architecture  
113 underlying microbiota composition, with only little overlap among identified loci<sup>48</sup>. Moreover, the  
114 effect of major disease-causing genetic defects on the human gut microbiota remains elusive, even  
115 though notable advances have been made in the context of IBD.

116 The role of the intestinal microbiota in IBD pathogenesis is now accepted<sup>49-52</sup>. Beyond mouse  
117 studies on IBD susceptibility genes such as *Nod2* and *Card9*, interaction with genetic factors is being  
118 explored in humans (Table 1). Patients harboring autophagy-related 16-like 1 (ATG16L1) polymorphism  
119 show significant microbiota alterations, with increased abundances of *Fusobacteriaceae*<sup>53</sup> and  
120 *Enterobacteriaceae* and decreased amounts of Clostridium groups IV and XIVa<sup>44</sup>. NOD2 variants are  
121 associated with decreased abundances of *Roseburia* and *Faecalibacterium prausnitzii*<sup>47</sup>. Mathematical  
122 modeling proposes that the effect of *NOD2* risk variants on IBD is partially mediated by decreased *F.*  
123 *prausnitzii* abundance<sup>54</sup>. Moreover, the gut microbiota of IBD patients with *CARD9* but not *NOD2* or  
124 *ATG16L1* risk alleles exhibits an impaired ability to metabolize tryptophan into AhR agonists, as  
125 observed in *Card9*<sup>-/-</sup> mice<sup>34</sup>. Furthermore, analysis of the microbiota composition of patients with rare  
126 monogenic primary immunodeficiency causing an IBD-like phenotype (chronic granulomatous disease,  
127 X-linked inhibitor of apoptosis defect and tetratricopeptide repeat domain 7A defect) showed gene-  
128 specific differences compared to healthy subjects and patients with conventional IBD<sup>55</sup>.

129 Taken together, these data in mice and humans demonstrate that host genetics has an impact on gut  
130 microbiota composition and function. In the FMT context, it suggests that the genetics of the recipients  
131 should be taken into account.

132

### 133 1-2 Immunity affects the microbiota

134 Intestinal inflammation drastically modifies the gut environment and leads to changes in the gut  
135 microbiota through several mechanisms (FIG. 1). The released antimicrobial peptides sequester  
136 specific host-derived nutrients, including iron and zinc, and favor the growth of bacteria that are more

137 efficient at acquiring those nutrients, such as *Enterobacteriaceae*<sup>56–58</sup>. The production of reactive  
138 oxygen and nitrogen species by host cells leads to the luminal generation of exogenous electron  
139 acceptors, which selectively enhances the growth of facultative anaerobic bacteria, especially  
140 *Enterobacteriaceae*<sup>59–61</sup>. Unlike the obligate anaerobic Bacteroidia and Clostridia, *Enterobacteriaceae*  
141 are more likely to encode enzymes for nitrate respiration and thrive in this new metabolic niche. Thus,  
142 inflammation confers a fitness advantage to facultative anaerobes, possibly explaining the expansion  
143 of *Enterobacteriaceae* in the inflammatory setting<sup>59</sup>.

144

145 Intestinal secretory IgA also regulates the composition and function of the gut microbiota in both  
146 humans and mice (FIG. 1). IgA coating affects microbial fitness through various effector functions, such  
147 as modulation of bacterial gene expression and metabolism, immune exclusion, enhanced antigen  
148 uptake, alteration of bacterial motility and niche occupancy, and neutralization of toxins or other  
149 secreted factors<sup>62</sup>.

150 Only a fraction of commensals, such as Proteobacteria and *A. muciniphila*, are coated by IgA *in vivo*,  
151 while most members are not<sup>62</sup>. Increased IgA coating of the intestinal microbiota is observed in  
152 humans with IBD and mouse colitis models<sup>63–65</sup>. In humans, a high IgA coating identifies taxa associated  
153 with disease, such as proinflammatory bacteria in patients with IBD<sup>64,65</sup>. Interestingly, some patients  
154 with IgA deficiency have an IBD-like phenotype<sup>66,67</sup> and exhibit alterations in gut microbiota  
155 composition despite the compensatory secretion of microbiota-targeted IgM<sup>68</sup>.

156 IgA-deficient mice exhibit increased abundances of Proteobacteria<sup>69</sup> and segmented filamentous  
157 bacteria (SFB)<sup>70</sup> and produce abnormal IgG responses to intestinal bacteria, suggesting exposure of  
158 their systemic immune system to microorganisms due to ineffective compartmentalization<sup>71</sup>. Similarly,  
159 mice lacking MyD88 in the T cell compartment fail to control mucosa-associated bacterial communities  
160 due to an abnormal IgA response<sup>72</sup>. This effect induces an increase in the abundances of mucolytic  
161 Proteobacteria and Firmicutes (*Ruminococcus*) and worsens colitis<sup>72</sup>. Naturally occurring  
162 polymorphisms in MHC genes, which control intestinal IgA phenotypes, result in differences in



163 microbiota composition between different lines of MHC-congenic mice and are sufficient to explain  
164 susceptibility to enteric infection<sup>73</sup>.

165 Finally, IgA can modulate gene expression in *Bacteroides thetaiotaomicron*, a prominent  
166 commensal, facilitating functional symbiosis with Firmicutes and promoting homeostasis and  
167 protection from colitis<sup>74</sup>. Thus, diversified and well-selected IgA repertoires might be essential not only  
168 to eliminate potentially harmful microorganisms but also to maintain diverse and balanced commensal  
169 communities.

170 Collectively, these results reveal a unique and nonredundant role for intestinal secretory IgA in  
171 shaping and stabilizing bacterial communities. In parallel with intestinal inflammatory status, the  
172 variability in IgA repertoires and the resulting dynamic immunoselection might play a role in the  
173 evolution of the gut microbiota following FMT.

174

## 175 **2- Factors associated with FMT success**

176

### 177 **2-1 What is a successful FMT?**

178 FMT success is primarily defined by a positive clinical response in the recipient, with the main  
179 objective being a long-term therapeutic effect. However, the shift in gut microbiota composition of the  
180 recipient toward that of the donor, corresponding to the engraftment of the donor microbiota, is also  
181 a useful marker *per se* to assess the success of the procedure. Indeed, engraftment success is often  
182 directly associated with clinical improvement. The first RCT investigating FMT effects in patients with  
183 rCDI revealed a major increase in microbial diversity of the recipient microbiota toward that of the  
184 donor<sup>4</sup>. Similarly, a larger proportion of fungal and bacterial OTUs from the donor microbiota, with  
185 more similar abundances, are transferred in responding rCDI patients than in patients not responding  
186 to treatment<sup>75</sup>. In the same way, patients with IBD who respond to FMT demonstrate a significant shift  
187 in microbiota composition toward their donor's profile, whereas nonresponders do not<sup>6,13</sup>. Typically,

188 changes in microbiota composition following FMT are associated with increased microbial diversity<sup>8,13</sup>,  
189 which is much more pronounced in responders than in nonresponders<sup>13,76,77</sup>.  
190 In addition to composition, metabolic functions of intestinal microbiota might also be crucial to  
191 restore, as metabolites are one of the primary modes by which the gut microbiota interacts with the  
192 host<sup>78,79</sup>. For instance, the presence of bacterial taxa transforming primary to secondary bile acids is  
193 associated with resistance to *C. difficile* infection<sup>80-84</sup>. Moreover, FMT restores the short-chain fatty  
194 acid (SCFA) and polyunsaturated fatty acid composition in patients with rCDI<sup>83,84</sup>. Similarly, in UC,  
195 remission is associated with the restoration of SCFA biosynthesis pathways and secondary bile acid  
196 levels<sup>77</sup>. In a case series, filtrated bacteria-free FMT was also able to alleviate rCDI symptoms,  
197 suggesting that bacterial molecules, metabolites, or bacteriophages mediate some of the FMT effects  
198 in the rCDI context<sup>85</sup>. Bacteriophage transfer during FMT is also associated with rCDI symptom  
199 resolution<sup>86</sup>, supporting that live bacteria are not the only players in FMT therapy. These data highlight  
200 that the transfer of a variety of microorganisms and microbial molecules with diverse engraftment  
201 patterns might be necessary to restore a homeostatic intestinal environment in the patient. In line  
202 with these findings, complete transfer of the donor microbiota is not required to resolve rCDI<sup>82</sup>.

203       Following FMT, some components of the transplanted microbiota engraft and combine with the  
204 remaining recipient microbiota to rebuild a new commensal community, a process that can take  
205 months to years to stabilize<sup>87,88</sup>. Although many studies report long-term FMT impact<sup>6,86,25,89</sup>, the effect  
206 can also be transient. This situation is the case in patients with metabolic syndrome for whom insulin  
207 sensitivity improvement was observed at week 6 post-FMT but was lost at week 18, in line with  
208 transient donor microbiota engraftment<sup>26,27,90</sup>. Thus, long-term clinical improvement relies on the  
209 successful establishment and adaptation of the newly transferred microbiota to the specific rules  
210 governing its novel host habitat.

211

212 2-2 Donor selection and FMT protocol

213 To date, the exceptionally high success rate of FMT therapy in rCDI has not been observed for any  
214 other diseases. The heterogeneity of FMT efficacy from one disease to the other likely reflects  
215 differences regarding the importance of the gut microbiota in their respective pathogenesis. While  
216 rCDI is almost purely microbiota related, other diseases, such as IBD or metabolic syndrome, are much  
217 more complex and involve dysfunctional host-microbe interactions and immune and genetic factors<sup>91</sup>.  
218 In this context, discrepancies in FMT procedures make it even more challenging to compare  
219 independent studies. Various approaches have been chosen in terms of donor selection (related or  
220 unrelated donors), preparation of fecal material (fresh or frozen, aerobic or anaerobic), route of  
221 delivery (colonoscopy, upper endoscopy, nasointestinal tube, enema or capsule), intestinal  
222 preparation (antibiotics or colon cleansing), and dose and frequency of administration<sup>92,93</sup>. Even with  
223 the same procedure, response to FMT is variable from one recipient to the next. Specificities of the  
224 donor material can partly explain these differential responses, with the emergence of the  
225 "superdonor" concept<sup>94</sup>. The first study revealing a superdonor effect was an RCT evaluating FMT  
226 efficacy in UC, in which seven of the nine FMT responders received material from the same donor<sup>6</sup>.

227 Since then, several studies have confirmed that high donor bacterial diversity is crucial to restoring  
228 a stable commensal community in recipients and is one of the most significant factors influencing FMT  
229 efficacy in IBD<sup>76,95,77</sup>. A high diversity of bacteriophage communities in donors is also associated with  
230 FMT success in rCDI<sup>96</sup>. Donor microbiota composition also matters. Members of *Clostridium* clusters  
231 IV and XIVa in the *Ruminococcaceae* and *Lachnospiraceae* families were found to be enriched in the  
232 microbiota of a superdonor for UC<sup>6</sup>, and their enrichment was associated with a positive FMT outcome  
233 in several IBD studies<sup>7,8,97</sup>. Similarly, high abundances of *A. muciniphila*, *Ruminococcaceae* members  
234 and *Bacteroides* species were more likely to induce remission in patients with UC, whereas a high  
235 amount of *Streptococcus* was associated with no response<sup>77,95</sup>.

236

237 2-3 Recipient genetics and lifestyle

238 For rCDI, the long-term stability of the donor microbiota might not be crucial. FMT rapidly induces  
239 clearance of the pathogen and restoration of a commensal community, and a gradual drift away from  
240 the donor's microbiota profile due to the recipient selection pressures is unlikely to lead to disease  
241 recurrence if there are no further insults to the gut microbiota. By contrast, the sustainability of the  
242 donor microbiota in patients with chronic diseases, such as IBD or metabolic syndrome, may be much  
243 more determinant. As microbiota alterations contribute to disease progression, FMT is essential to  
244 provide the initial relief, with a likely need for subsequent FMT to maintain the effect.

245 Strong evidence supports the theory of donor-recipient compatibility, which depends not only on  
246 donor material specificities but also on recipient genetics and environment. Recipient genes and  
247 immunity, as well as diet and antibiotic exposures, impact FMT engraftment and long-term  
248 maintenance. In metabolic syndrome, engraftment failure observed 18 weeks after FMT could be due  
249 to particular immune responses and dietary habits<sup>27</sup>. Along with variable FMT outcomes, colonization  
250 levels differ largely among recipients sharing the same donor<sup>98</sup>, suggesting the existence of individual  
251 permissivity patterns regarding donor microbiota. In hematopoietic stem-cell transplantation  
252 recipients undergoing FMT for rCDI, the functional and taxonomic concordance between donor  
253 material and their own post-FMT microbiota diminished after one year, supporting that environmental  
254 and host factors are essential determinants in the long-term shaping of the transferred microbiota<sup>90</sup>.  
255 Thus, aspects such as strain and species fitness and colonization resistance should be investigated in  
256 the context of microbiota resilience, genetic traits, immune responses and lifestyle of the recipient.  
257 Indeed, it is likely that complex ecological interactions and donor-recipient compatibilities underlie the  
258 success of FMT.

259 As genetic traits are inherent to the recipient and cannot be changed, FMT might be combined with  
260 modulation of immune responses and dietary intervention (see section 3). Furthermore, donor-  
261 recipient matching approaches could rely on screening of functional perturbations in recipient  
262 microbiota to identify a specific donor material enriched in the metabolic pathways that need to be

263 restored. Thus, FMT is not a "one stool fits all" strategy, especially in the context of complex chronic  
264 diseases involving genetic, immune and environmental factors.

265

#### 266 2-4 Recipient clinical status

267 Although rarely discussed, the clinical status of the recipient before FMT is also a key element to  
268 consider (Table 2). Severe and complicated indications, inpatient status during FMT, and the number  
269 of previous CDI-related hospitalizations are strongly associated with early FMT failure<sup>99</sup>. Patients who  
270 need repeated FMT for rCDI have higher intestinal inflammation than patients cured with one single  
271 FMT<sup>100</sup>. rCDI patients with an active IBD requiring medication escalation have reduced FMT success,  
272 with no impact of other parameters, such as CDI recurrence<sup>101</sup>. In the context of IBD, it is also crucial  
273 to question the timing and extent of the disease before performing FMT. Is the recipient in flare or  
274 remission? Is the inflammation in a maintenance phase after successful immunosuppressive  
275 treatment? No published human trial is powered to assess the role of these factors in FMT success,  
276 but several elements converge to support this idea<sup>10,91</sup>.

277 Whereas a randomized FMT study in UC patients demonstrated no effect of concomitant steroid  
278 therapy on remission<sup>8</sup>, two others suggest the opposite. One study shows that patients taking  
279 immunosuppressants tend to have a greater benefit from FMT<sup>6</sup>. In another RCT, the reduction in  
280 disease score following FMT was greatly increased in patients taking oral steroids at baseline<sup>9</sup>.  
281 Recently, we published the first RCT in CD, evaluating the role of FMT in patients who achieved clinical  
282 remission using corticosteroids. Failure to maintain remission was associated with microbiota factors  
283 at baseline and weaker control of clinical activity<sup>15</sup>. In multivariate analysis, FMT and Crohn's disease  
284 activity index (CDAI) score were the only independent parameters associated with maintenance of  
285 remission, with FMT dividing the risk of a flare by 8 and higher CDAI score (although below the  
286 remission threshold) increasing the risk by more than 20-fold (Table 3).

287 Donor-recipient incompatibilities leading to FMT failure may partly be due to an active immune  
288 response toward the transplanted microbiota. An immune screening approach was examined in an

289 FMT case study for UC to minimize FMT failure<sup>102</sup>. Patient-derived lymphoid cells isolated from a rectal  
290 biopsy were incubated with the intestinal microbiota of different donors. FMT, performed with the  
291 donor microbiota inducing the lowest production of proinflammatory cytokines, induced a  
292 remission<sup>102</sup>. In addition to the noncontrolled nature of this study, the time and costs involved by  
293 personalized approaches limit their implementation. Controlling the inflammation level in the  
294 recipient's intestine using immunosuppressants might be a simpler strategy.

295

## 296 2-5 Recipient microbiota composition

297 Following FMT, donor bacterial strains cohabit with those of the recipient. In metabolic syndrome,  
298 colonization success for a given strain is greater if another representative of the same species is also  
299 present in the recipient before FMT<sup>98</sup>. In both rCDI and metabolic syndrome, bacterial abundance and  
300 taxonomic identity in both the donor and recipient before FMT are strong predictors of engraftment<sup>103</sup>.  
301 Following FMT, the recipient microbiota contains a combination of recipient- and donor-derived as  
302 well as newly acquired species<sup>103</sup>, suggesting that complex microbial interactions contribute to overall  
303 FMT engraftment. Thus, two recipients receiving the same donor microbiota will not show the same  
304 clinical outcome, as both recipient genetics and microbiota are idiosyncratic and determinant in FMT  
305 success.

306 In numerous human studies, both microbial diversity and the presence of specific species in the  
307 recipient microbiota impact FMT engraftment (Table 3). In UC, higher fecal and mucosal microbiota  
308 richness before FMT is associated with positive therapeutic outcomes<sup>8,77</sup>, while *Fusobacterium* and  
309 *Sutterella* species are associated with FMT failure<sup>8</sup>. In CD, FMT failure is associated with enrichment in  
310 different members of Gammaproteobacteria, such as *Klebsiella*, *Actinobacillus*, and *Haemophilus*, in  
311 the recipient microbiota at baseline<sup>15</sup>. In metabolic syndrome, responders have lower fecal microbial  
312 diversity before FMT, with higher relative abundances of *Subdoligranulum variabile* and *Dorea* and  
313 lower relative abundances of *Eubacterium ventriosum* and *Ruminococcus torques* than

314 nonresponders<sup>27</sup>. Patients with IBS responding to FMT have a higher baseline abundance of  
315 *Streptococcus* species and higher microbiota diversity than nonresponders<sup>104</sup>.

316 In rCDI, a low abundance of fecal *Candida albicans*, both in recipients at baseline and in donors,  
317 correlates with a positive FMT outcome<sup>75</sup>. In contrast, a high initial abundance of *C. albicans* was  
318 associated with increased FMT success in patients with UC, suggesting that high fungal abundance  
319 allows better bacterial engraftment in this context<sup>105</sup>. In rCDI, a higher richness of the bacteriophage  
320 Caudovirales in the donor than in the recipient microbiota predicts a positive therapeutic outcome<sup>86</sup>.  
321 Moreover, in UC, responders to FMT have a lower baseline relative abundance of Caudovirales  
322 bacteriophages than nonresponders<sup>106</sup>. Similarly, low eukaryotic virus richness before FMT in patients  
323 with UC is associated with a clinical response<sup>107</sup>. These data suggest that in addition to bacteria, fungi  
324 and both eukaryotic and prokaryotic viruses in the recipient microbiota play a role in the establishment  
325 of a novel commensal community after FMT and in therapeutic effects.

326 Altogether, these data demonstrate that the baseline recipient microbiota affects FMT clinical  
327 outcome, probably by influencing the success of donor microbiota engraftment and maintenance but  
328 also by directly contributing to the new microbial ecosystem. Thus, extensive analysis of bacterial,  
329 fungal and viral species in patients before transfer might provide a valuable biomarker for FMT success  
330 prediction.

331

### 332 **3- Avenues for progress**

333

#### 334 3-1 Gut preparation for engraftment

335 Inadequate bowel preparation is associated with FMT failure in rCDI<sup>108</sup>. Although it seems  
336 logical to clear ecological niches for the new microbiota before administrating it, procedures for  
337 preparing the recipient (broad-spectrum antibiotics, colon cleansing with polyethylene glycol or no  
338 preparation) vary between studies, and the best approach remains to be identified (FIG. 2). Antibiotic  
339 usage is questionable, as they may induce deleterious side effects (particularly in IBD) and/or enrich

340 for antibiotic-resistant bacteria. Few clinical studies suggest that antibiotic pretreatment improves the  
341 clinical response rate of FMT in UC, but control arms are missing<sup>109,110</sup>. Mouse study results are  
342 divergent, with one study demonstrating a positive impact of bowel cleansing prior to FMT<sup>111</sup>, while  
343 another found no effect of lavage but a positive effect of antibiotics<sup>112</sup>. Conversely, antibiotic  
344 pretreatment does not improve the overall engraftment of donor microbiota compared to that with  
345 lavage alone but does increase the engraftment of specific taxa, such as *Bifidobacterium*<sup>113</sup>. Thus,  
346 further well-designed RCTs are needed to determine the best approach to prepare the gut  
347 environment for engraftment.

348

### 349 3-2 Targeting recipient immunity

350 Modulation of immune responses within the new gut habitat could improve engraftment (FIG.  
351 2). In IBD, control of disease activity using corticosteroids is associated with improved FMT efficacy<sup>6,9,15</sup>.  
352 Indeed, a healthy microbiota can rapidly be altered by existing intestinal inflammation, thus limiting  
353 its potential therapeutic effect. Moreover, transferring a large number of microbes into an inflamed  
354 intestine with a disrupted epithelial barrier could be detrimental and both exacerbate inflammation  
355 and promote bacterial translocation. Thus, treatment with immunosuppressants in addition to FMT  
356 protocols might be useful to alleviate immune pressure on the newly transferred microbiota and avoid  
357 additional damage to the intestinal tissue.

358 Furthermore, human leukocyte antigen (HLA) and other genetic compatibility factors between donors  
359 and recipients might play a role in FMT efficacy. Indeed, FMT engraftment failure can be related to  
360 graft rejection, which corresponds to the immunologic destruction of transplanted tissues between  
361 two individuals of the same species differing at the MHC for that species (i.e., HLA in humans). HLAs  
362 are extremely polymorphic in different populations and determine the specificity of T cell responses  
363 against the intestinal microbiota. MHC-dependent mechanisms impact the microbiota composition in  
364 mice<sup>73,114</sup>. In humans, specific HLA polymorphisms are associated with the microbiota composition and



365 with several common infections<sup>46,115,116</sup>. Considering HLA as a matching factor for FMT should be  
366 investigated.

367

### 368 3-3 Dietary intervention

369 Environmental factors impacting the microbiota, particularly dietary habits, might play a significant  
370 role in FMT success (FIG. 2). Dietary interventions show promising results in CD<sup>117-119</sup> and could help  
371 maintain an optimal ecosystem after transplantation in IBD and non-IBD contexts. Adopting a  
372 “microbiota-protecting” diet enriched in fibers to promote SCFA-producing bacteria and reduced in  
373 ultraprocessed food might be beneficial in the post-FMT setting. Further basic and clinical research  
374 studies are required to validate the integration of such approaches to FMT protocols.

375

### 376 **Conclusions**

377 FMT efficacy could be improved by carefully selecting donors, but the specific parameters for  
378 implementing selection criteria are still mostly unknown. Similarly, no consensus exists on the best  
379 procedure for each condition. Standardization of methods and development of stool banks will  
380 improve follow-up over the years and help to optimize donor-to-recipient microbiota transfer.  
381 However, the role of recipient factors in FMT success remains underestimated. The main drivers of  
382 donor material engraftment into the recipient's intestine include microbiota composition, genetics,  
383 inflammation status and environmental factors, such as diet. Additional interventions, alone or in  
384 combination, could potentiate FMT. Optimization of donor/recipient matching at both the genetic and  
385 microbiota levels could help minimize engraftment failure due to incompatibilities. Deciphering the  
386 best bowel preparation method is also crucial to promote remission. Furthermore, strategies to limit  
387 immune pressure toward the newly transferred microbiota could be powerful, such as the  
388 administration of immunosuppressants at least during the first stages after FMT in chronic  
389 inflammatory diseases. Finally, improved control of the recipient environment, especially via dietary

390 intervention, might improve FMT engraftment and maintenance and support long-term clinical  
391 remission.

392

393 **Figure legends:**

394 - **Figure 1: Demonstrated effects of the host immune system on the intestinal microbiota**  
395 **composition and functions**

396 Multiple host factors, including intestinal immunity, shape the bacterial, fungal and viral populations  
397 of the microbiota. **A.** Intestinal secretory immunoglobulin A (SIgA) eliminates potentially harmful  
398 microorganisms and stabilizes bacterial communities. **B.** Production of antimicrobial peptides (AMPs)  
399 favors the growth of *Enterobacteriaceae* by sequestering specific host-derived nutrients, including iron  
400 and zinc. **C.** Production of reactive oxygen and nitrogen species (ROS and RNS) by host cells leads to  
401 the luminal generation of exogenous electron acceptors, which selectively enhance the growth of  
402 facultative anaerobic bacteria, including *Enterobacteriaceae*. **D.** Finally, immune and regulatory  
403 proteins are essential to maintain a balanced composition and function of the microbiota, especially  
404 the ratio between Bacteroidetes and Firmicutes. Mutation of these genes is associated with alteration  
405 of the microbiota composition. NOD2, nucleotide-binding oligomerization domain-containing protein  
406 2; ATG16L1, autophagy-related 16-like 1; NLRP6, NOD-like receptor family pyrin domain containing 6;  
407 VDR, vitamin D receptor; MyD88, myeloid differentiation primary response 88; TLR5, Toll-like receptor  
408 5; CARD9, caspase recruitment domain family member 9; TTC7A, tetratricopeptide repeat domain 7A;  
409 XIAP, X-linked inhibitor of apoptosis; CGD, chronic granulomatous disease.

410

411 - **Figure 2: Potential targets and strategies on the recipient side to improve FMT efficacy**

412 The main drivers of donor material engraftment into the recipient's intestine include microbiota  
413 composition, genetics, inflammation status and environmental factors, such as dietary habits.  
414 Additional interventions before or after transplantation, alone or in combination, could help to  
415 improve FMT success. **A.** Optimization of donor/recipient matching at the genetic, immune and

416 microbiota levels could help minimize engraftment failure due to incompatibilities. **B.** Identifying the  
 417 best bowel preparation method between antibiotics and bowel cleansing is also crucial to promote  
 418 engraftment. **C.** Furthermore, strategies to limit immune pressure toward the newly transferred  
 419 microbiota, such as the administration of immunosuppressants at least before and during the first  
 420 stages after FMT in chronic inflammatory diseases could be a powerful method. **D.** Finally, improved  
 421 control of the recipient environment, especially via dietary intervention, might improve FMT  
 422 engraftment and maintenance and support long-term clinical remission.

423

424

425 **Tables:**

426 **Table 1: Associations between host genetics and intestinal microbiota in healthy individuals and**

427 **IBD patients**

Study (year)	Gene	Association with microbiota composition or function
<b>Healthy individuals (GWAS)</b>		
Turpin et al. (2016) <sup>45</sup>	rs62171178 (near UBR3) rs1394174 (CNTN6) rs59846192 (DMRTB1) rs28473221 (SALL3)	Presence of <i>Rikenellaceae</i> Presence of <i>Faecalibacterium</i> Presence of <i>Lachnospira</i> Presence of <i>Eubacterium</i>
Bonder et al. (2016) <sup>46</sup>	<b>CLEC4F–CD207</b> and <b>CLEC4A–FAM90A1</b> (C-type lectin) <b>HLA-B</b> (human leukocyte antigen)	Intestinal homeostasis Microbiota composition
Wang et al. (2016) <sup>47</sup>	<b>VDR</b> (vitamin D receptor)	Overall microbial variation and individual taxa
<b>Patients with IBD</b>		
Frank et al. (2011) <sup>120</sup>	<b>NOD2</b> (nucleotide-binding oligomerization domain-containing protein 2) in IBD <b>ATG16L1</b> (inflammasome NOD-like receptor family pyrin domain containing 6) in IBD	Increased abundance of <i>Enterobacteriaceae</i> (Proteobacteria) and decreased abundance of Clostridium groups IV and XIVa (Firmicutes)
Knights et al. (2014) <sup>121</sup>	<b>NOD2</b> in IBD	Increased abundance of <i>Enterobacteriaceae</i>
Sadaghian Sadabad et al. (2015) <sup>53</sup>	<b>ATG16L1</b> in CD	Increased abundance of <i>Fusobacteriaceae</i>
Lamas et al., (2016) <sup>34</sup>	<b>CARD9</b> (caspase recruitment domain family member 9) in IBD	Impaired ability to metabolize tryptophan into AhR agonists
Aschard et al. (2019) <sup>54</sup>	<b>NOD2</b> in IBD	Decreased abundance of the <i>Roseburia</i> genus and <i>Faecalibacterium prausnitzii</i>

IBD-like phenotype in Primary immunodeficiency		
Sokol et al. (2019) <sup>55</sup>	<p><b>CGD</b> (Chronic granulomatous disease) <b>XIAP</b> (X-linked inhibitor of apoptosis)</p> <p><b>TTC7A</b> (Tetratricopeptide Repeat Domain 7A)</p>	<p>Increased abundance of <i>Ruminococcus gnavus</i></p> <p>Increased abundance of disease-associated taxa from Proteobacteria, Firmicutes, Actinobacteria, and Fusobacteria</p> <p>Presence of the pathogen <i>Lactococcus garvieae</i></p> <p>Increased abundance of Proteobacteria, decreased abundance of the <i>Ruminococcaceae</i> family (notably the <i>Oscillospira</i> genera)</p>

428

429

430 **Table 2: Donor and recipient factors at baseline associated with FMT success**

Condition	Donor microbiota composition	Recipient microbiota composition	Recipient clinical status
rCDI	<ul style="list-style-type: none"> <li>- Low abundance of <i>Candida albicans</i><sup>75</sup></li> <li>- High bacteriophage diversity<sup>86,96</sup></li> </ul>	<ul style="list-style-type: none"> <li>- Low abundance of <i>Candida albicans</i><sup>75</sup></li> <li>- Low relative abundance of Caudovirales bacteriophages<sup>86</sup></li> </ul>	<ul style="list-style-type: none"> <li>- Low disease severity<sup>99,108</sup></li> <li>- Low intestinal inflammation level (calprotectin)<sup>100</sup></li> <li>- Low number of disease recurrence<sup>99</sup></li> <li>- Adequate bowel preparation<sup>108</sup></li> <li>- No active associated IBD requiring medication escalation<sup>101</sup></li> </ul>
UC	<ul style="list-style-type: none"> <li>- High microbiota diversity<sup>77,95</sup></li> <li>- Enrichment in members of Clostridium clusters IV and XIVa in the <i>Ruminococcaceae</i> and <i>Lachnospiraceae</i> families<sup>6-8,97</sup></li> <li>- High relative abundance of <i>A. muciniphila</i>, <i>Ruminococcaceae</i> members and <i>Bacteroides</i> species<sup>77,95</sup></li> <li>- Low amount of <i>Streptococcus</i><sup>77,95</sup></li> </ul>	<ul style="list-style-type: none"> <li>- High fecal and mucosal microbiota richness<sup>8,77</sup></li> <li>- Absence of <i>Fusobacterium</i> and <i>Sutterella</i><sup>8</sup></li> <li>- High abundance of <i>C. albicans</i><sup>105</sup></li> <li>- Low abundance of Caudovirales bacteriophages<sup>106</sup></li> <li>- Low eukaryotic virus richness<sup>107</sup></li> </ul>	<ul style="list-style-type: none"> <li>- Immunosuppressant therapy, such as oral steroids<sup>6,9</sup></li> </ul>
CD	<ul style="list-style-type: none"> <li>- High microbiota diversity<sup>76</sup></li> </ul>	<ul style="list-style-type: none"> <li>- Low abundance in members of Gammaproteobacteria such as <i>Klebsiella</i>, <i>Actinobacillus</i> and <i>Haemophilus</i><sup>15</sup></li> </ul>	<ul style="list-style-type: none"> <li>- Low disease activity<sup>15</sup></li> </ul>
Metabolic syndrome	<ul style="list-style-type: none"> <li>- High abundance of <i>Bifidobacterium</i><sup>122</sup></li> </ul>	<ul style="list-style-type: none"> <li>- Low relative microbial diversity<sup>27</sup></li> <li>- High abundance of <i>Subdoligranulum variabile</i> and <i>Dorea</i><sup>27</sup></li> <li>- Low abundance of <i>Eubacterium ventriosum</i> and <i>Ruminococcus torques</i><sup>27</sup></li> </ul>	N/A
IBS	N/A	<ul style="list-style-type: none"> <li>- High relative microbiota diversity<sup>104</sup></li> </ul>	N/A

		- High relative abundance of <i>Streptococcus</i> <sup>104</sup>	
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432

433 **Table 3: Factors associated with remission maintenance in the whole population of the study in**  
 434 **univariate and multivariate analyses (from <sup>15</sup>)**

Factors	P value univariate	P value multivariate	OR (Odds Ratio)	95% CI (confidence interval)
FMT vs Sham	0.23	0.0295*	0.13	[0.009 – 0.82]
Age	0.60			
Gender	0.70			
Active smoking	0.53			
Montreal : L2 / L3	0.91			
Disease duration	0.26			
Hemoglobin	0.46			
Platelet count	0.55			
CRP	0.21	0.42		
Fecal Calprotectin	0.06	0.72		
Previous azathioprine treatment	0.28			
Previous anti-TNF therapy	0.39			
CDAI Score	0.03	0.018*	26.9	[1.8 – 645.7]
CDEIS Score	0.31			

435 CRP: C reactive protein; CDAI: Crohn's disease activity index; CDEIS: Crohn's disease endoscopic index  
 436 of severity; TNF: Tumor Necrosis Factor; L2: colonic involvement only ; L3: ileo-colonic involvement.

437

438

439 **References:**

440 1. Durack, J. & Lynch, S. V. The gut microbiome: Relationships with disease and opportunities for  
 441 therapy. *J. Exp. Med.* **216**, 20–40 (2019).

442 2. D’Haens, G. R. & Jobin, C. Fecal Microbial Transplantation for Diseases Beyond Recurrent  
 443 *Clostridium Difficile* Infection. *Gastroenterology* **157**, 624–636 (2019).

444 3. Round, J. L. & Palm, N. W. Causal effects of the microbiota on immune-mediated diseases. *Sci.*  
 445 *Immunol.* **3**, eaao1603 (2018).

446 4. van Nood, E. *et al.* Duodenal Infusion of Donor Feces for Recurrent *Clostridium difficile*. *N. Engl. J.*  
 447 *Med.* **368**, 407–415 (2013).

- 448 5. Kelly, C. R. *et al.* Effect of Fecal Microbiota Transplantation on Recurrence in Multiply Recurrent  
449 *Clostridium difficile* Infection: A Randomized Trial. *Ann. Intern. Med.* **165**, 609 (2016).
- 450 6. Moayyedi, P. *et al.* Fecal Microbiota Transplantation Induces Remission in Patients With Active  
451 Ulcerative Colitis in a Randomized Controlled Trial. *Gastroenterology* **149**, 102-109.e6 (2015).
- 452 7. Rossen, N. G. *et al.* Findings From a Randomized Controlled Trial of Fecal Transplantation for  
453 Patients With Ulcerative Colitis. *Gastroenterology* **149**, 110-118.e4 (2015).
- 454 8. Paramsothy, S. *et al.* Multidonor intensive faecal microbiota transplantation for active ulcerative  
455 colitis: a randomised placebo-controlled trial. *The Lancet* **389**, 1218–1228 (2017).
- 456 9. Costello, S. P. *et al.* Effect of Fecal Microbiota Transplantation on 8-Week Remission in Patients  
457 With Ulcerative Colitis: A Randomized Clinical Trial. *JAMA* **321**, 156 (2019).
- 458 10. Colman, R. J. & Rubin, D. T. Fecal microbiota transplantation as therapy for inflammatory bowel  
459 disease: A systematic review and meta-analysis. *J. Crohns Colitis* **8**, 1569–1581 (2014).
- 460 11. Cui, B. *et al.* Fecal microbiota transplantation through mid-gut for refractory Crohn’s disease:  
461 Safety, feasibility, and efficacy trial results: Fecal microbiota transplantation. *J. Gastroenterol.*  
462 *Hepatol.* **30**, 51–58 (2015).
- 463 12. Suskind, D. L. *et al.* Fecal Microbial Transplant Effect on Clinical Outcomes and Fecal Microbiome  
464 in Active Crohn’s Disease: *Inflamm. Bowel Dis.* **21**, 556–563 (2015).
- 465 13. Vaughn, B. P. *et al.* Increased Intestinal Microbial Diversity Following Fecal Microbiota Transplant  
466 for Active Crohn’s Disease: *Inflamm. Bowel Dis.* **22**, 2182–2190 (2016).
- 467 14. He, Z. *et al.* Multiple fresh fecal microbiota transplants induces and maintains clinical remission  
468 in Crohn’s disease complicated with inflammatory mass. *Sci. Rep.* **7**, 4753 (2017).
- 469 15. Sokol *et al.* Fecal microbiota transplantation to maintain remission in Crohn’s disease: a pilot  
470 randomized controlled study. *Microbiome* **8**, 12 (2020).
- 471 16. Johnsen, P. H. *et al.* Faecal microbiota transplantation versus placebo for moderate-to-severe  
472 irritable bowel syndrome: a double-blind, randomised, placebo-controlled, parallel-group, single-  
473 centre trial. *Lancet Gastroenterol. Hepatol.* **3**, 17–24 (2018).

- 474 17. El-Salhy, M., Hatlebakk, J. G., Gilja, O. H., Bråthen Kristoffersen, A. & Hausken, T. Efficacy of  
475 faecal microbiota transplantation for patients with irritable bowel syndrome in a randomised,  
476 double-blind, placebo-controlled study. *Gut* **69**, 859–867 (2020).
- 477 18. Halkjær, S. I. *et al.* Faecal microbiota transplantation alters gut microbiota in patients with  
478 irritable bowel syndrome: results from a randomised, double-blind placebo-controlled study. *Gut*  
479 **67**, 2107–2115 (2018).
- 480 19. Holster, S. *et al.* The Effect of Allogenic Versus Autologous Fecal Microbiota Transfer on  
481 Symptoms, Visceral Perception and Fecal and Mucosal Microbiota in Irritable Bowel Syndrome: A  
482 Randomized Controlled Study. *Clin. Transl. Gastroenterol.* **10**, e00034 (2019).
- 483 20. Aroniadis, O. C. *et al.* Faecal microbiota transplantation for diarrhoea-predominant irritable  
484 bowel syndrome: a double-blind, randomised, placebo-controlled trial. *Lancet Gastroenterol.*  
485 *Hepatol.* **4**, 675–685 (2019).
- 486 21. Kakihana, K. *et al.* Fecal microbiota transplantation for patients with steroid-resistant acute  
487 graft-versus-host disease of the gut. *Blood* **128**, 2083–2088 (2016).
- 488 22. Spindelboeck, W. *et al.* Repeated fecal microbiota transplantations attenuate diarrhea and lead  
489 to sustained changes in the fecal microbiota in acute, refractory gastrointestinal graft- versus -  
490 host-disease. *Haematologica* **102**, e210–e213 (2017).
- 491 23. Qi, X. *et al.* Treating Steroid Refractory Intestinal Acute Graft-vs.-Host Disease With Fecal  
492 Microbiota Transplantation: A Pilot Study. *Front. Immunol.* **9**, 2195 (2018).
- 493 24. Bajaj, J. S. *et al.* Fecal microbiota transplant from a rational stool donor improves hepatic  
494 encephalopathy: A randomized clinical trial: Bajaj et al. *Hepatology* **66**, 1727–1738 (2017).
- 495 25. Bajaj, J. S. *et al.* Long-term Outcomes of Fecal Microbiota Transplantation in Patients With  
496 Cirrhosis. *Gastroenterology* **156**, 1921-1923.e3 (2019).
- 497 26. Vrieze, A. *et al.* Transfer of Intestinal Microbiota From Lean Donors Increases Insulin Sensitivity in  
498 Individuals With Metabolic Syndrome. *Gastroenterology* **143**, 913-916.e7 (2012).

- 499 27. Kootte, R. S. *et al.* Improvement of Insulin Sensitivity after Lean Donor Feces in Metabolic  
500 Syndrome Is Driven by Baseline Intestinal Microbiota Composition. *Cell Metab.* **26**, 611-619.e6  
501 (2017).
- 502 28. Kang, D.-W. *et al.* Microbiota Transfer Therapy alters gut ecosystem and improves  
503 gastrointestinal and autism symptoms: an open-label study. *Microbiome* **5**, 10 (2017).
- 504 29. Kang, D.-W. *et al.* Long-term benefit of Microbiota Transfer Therapy on autism symptoms and  
505 gut microbiota. *Sci. Rep.* **9**, 5821 (2019).
- 506 30. Goodrich, J. K. *et al.* Human Genetics Shape the Gut Microbiome. *Cell* **159**, 789–799 (2014).
- 507 31. Couturier-Maillard, A. *et al.* NOD2-mediated dysbiosis predisposes mice to transmissible colitis  
508 and colorectal cancer. *J. Clin. Invest.* JCI62236 (2013) doi:10.1172/JCI62236.
- 509 32. Petnicki-Ocwieja, T. *et al.* Nod2 is required for the regulation of commensal microbiota in the  
510 intestine. *Proc. Natl. Acad. Sci.* **106**, 15813–15818 (2009).
- 511 33. Hu, B. *et al.* Microbiota-induced activation of epithelial IL-6 signaling links inflammasome-driven  
512 inflammation with transmissible cancer. *Proc. Natl. Acad. Sci.* **110**, 9862–9867 (2013).
- 513 34. Lamas, B. *et al.* CARD9 impacts colitis by altering gut microbiota metabolism of tryptophan into  
514 aryl hydrocarbon receptor ligands. *Nat. Med.* **22**, 598–605 (2016).
- 515 35. Khan, A. A. *et al.* Polymorphic Immune Mechanisms Regulate Commensal Repertoire. *Cell Rep.*  
516 **29**, 541-550.e4 (2019).
- 517 36. Wen, L. *et al.* Innate immunity and intestinal microbiota in the development of Type 1 diabetes.  
518 *Nature* **455**, 1109–1113 (2008).
- 519 37. Vijay-Kumar, M. *et al.* Metabolic Syndrome and Altered Gut Microbiota in Mice Lacking Toll-Like  
520 Receptor 5. *Science* **328**, 228–231 (2010).
- 521 38. Kawamoto, S. *et al.* Foxp3+ T Cells Regulate Immunoglobulin A Selection and Facilitate  
522 Diversification of Bacterial Species Responsible for Immune Homeostasis. *Immunity* **41**, 152–165  
523 (2014).



- 524 39. Zhang, H., Sparks, J. B., Karyala, S. V., Settlage, R. & Luo, X. M. Host adaptive immunity alters gut  
525 microbiota. *ISME J.* **9**, 770–781 (2015).
- 526 40. Dimitriu, P. A. *et al.* Temporal stability of the mouse gut microbiota in relation to innate and  
527 adaptive immunity: Mouse gut microbiota dynamics. *Environ. Microbiol. Rep.* **5**, 200–210 (2013).
- 528 41. Sokol, H. *et al.* Card9 Mediates Intestinal Epithelial Cell Restitution, T-Helper 17 Responses, and  
529 Control of Bacterial Infection in Mice. *Gastroenterology* **145**, 591-601.e3 (2013).
- 530 42. Richard, M. L. & Sokol, H. The gut mycobiota: insights into analysis, environmental interactions  
531 and role in gastrointestinal diseases. *Nat. Rev. Gastroenterol. Hepatol.* (2019)  
532 doi:10.1038/s41575-019-0121-2.
- 533 43. Sovran, B. *et al.* Enterobacteriaceae are essential for the modulation of colitis severity by fungi.  
534 *Microbiome* **6**, 152 (2018).
- 535 44. van Tilburg Bernardes, E. *et al.* Intestinal fungi are causally implicated in microbiome assembly  
536 and immune development in mice. *Nat. Commun.* **11**, 2577 (2020).
- 537 45. Turpin *et al.* Association of host genome with intestinal microbial composition in a large healthy  
538 cohort. *Nat. Genet.* **48**, 1413–1417 (2016).
- 539 46. Bonder, M. J. *et al.* The effect of host genetics on the gut microbiome. *Nat. Genet.* **48**, 1407–  
540 1412 (2016).
- 541 47. Wang, J. *et al.* Genome-wide association analysis identifies variation in vitamin D receptor and  
542 other host factors influencing the gut microbiota. *Nat. Genet.* **48**, 1396–1406 (2016).
- 543 48. Benson, A. K. The gut microbiome-an emerging complex trait. *Nat. Genet.* **48**, 1301–1302 (2016).
- 544 49. Hansen, J. J. Immune Responses to Intestinal Microbes in Inflammatory Bowel Diseases. *Curr.*  
545 *Allergy Asthma Rep.* **15**, 61 (2015).
- 546 50. Gevers, D. *et al.* The treatment-naive microbiome in new-onset Crohn’s disease. *Cell Host*  
547 *Microbe* **15**, 382–392 (2014).
- 548 51. Morgan, X. C. *et al.* Dysfunction of the intestinal microbiome in inflammatory bowel disease and  
549 treatment. *Genome Biol.* **13**, R79 (2012).

- 550 52. Chu, H. *et al.* Gene-microbiota interactions contribute to the pathogenesis of inflammatory  
551 bowel disease. *Science* **352**, 1116–1120 (2016).
- 552 53. Sadaghian Sadabad, M. *et al.* The *ATG16L1-T300A* allele impairs clearance of pathosymbionts in  
553 the inflamed ileal mucosa of Crohn’s disease patients. *Gut* **64**, 1546–1552 (2015).
- 554 54. Aschard, H. *et al.* Genetic effects on the commensal microbiota in inflammatory bowel disease  
555 patients. *PLOS Genet.* **15**, e1008018 (2019).
- 556 55. Sokol, H. *et al.* Intestinal dysbiosis in inflammatory bowel disease associated with primary  
557 immunodeficiency. *J. Allergy Clin. Immunol.* **143**, 775-778.e6 (2019).
- 558 56. Raffatellu, M. *et al.* Lipocalin-2 resistance confers an advantage to *Salmonella enterica* serotype  
559 Typhimurium for growth and survival in the inflamed intestine. *Cell Host Microbe* **5**, 476–486  
560 (2009).
- 561 57. Liu, J. Z. *et al.* Zinc sequestration by the neutrophil protein calprotectin enhances *Salmonella*  
562 growth in the inflamed gut. *Cell Host Microbe* **11**, 227–239 (2012).
- 563 58. Deriu, E. *et al.* Probiotic bacteria reduce *salmonella typhimurium* intestinal colonization by  
564 competing for iron. *Cell Host Microbe* **14**, 26–37 (2013).
- 565 59. Winter, S. E. *et al.* Host-Derived Nitrate Boosts Growth of *E. coli* in the Inflamed Gut. *Science* **339**,  
566 708–711 (2013).
- 567 60. Winter, S. E. & Bäumler, A. J. Dysbiosis in the inflamed intestine: Chance favors the prepared  
568 microbe. *Gut Microbes* **5**, 71–73 (2014).
- 569 61. Faber, F. & Bäumler, A. J. The impact of intestinal inflammation on the nutritional environment  
570 of the gut microbiota. *Immunol. Lett.* **162**, 48–53 (2014).
- 571 62. Bunker, J. J. & Bendelac, A. IgA Responses to Microbiota. *Immunity* **49**, 211–224 (2018).
- 572 63. van der Waaij, L. A. *et al.* Immunoglobulin coating of faecal bacteria in inflammatory bowel  
573 disease. *Eur. J. Gastroenterol. Hepatol.* **16**, 669–674 (2004).
- 574 64. Palm, N. W. *et al.* Immunoglobulin A Coating Identifies Colitogenic Bacteria in Inflammatory  
575 Bowel Disease. *Cell* **158**, 1000–1010 (2014).

- 576 65. Viladomiu, M. *et al.* IgA-coated *E. coli* enriched in Crohn's disease spondyloarthritis promote T<sub>H</sub>  
577 17-dependent inflammation. *Sci. Transl. Med.* **9**, eaaf9655 (2017).
- 578 66. Aghamohammadi, A. *et al.* IgA Deficiency: Correlation Between Clinical and Immunological  
579 Phenotypes. *J. Clin. Immunol.* **29**, 130–136 (2009).
- 580 67. Ludvigsson, J. F., Neovius, M. & Hammarström, L. Association Between IgA Deficiency & Other  
581 Autoimmune Conditions: A Population-Based Matched Cohort Study. *J. Clin. Immunol.* **34**, 444–  
582 451 (2014).
- 583 68. Catanzaro, J. R. *et al.* IgA-deficient humans exhibit gut microbiota dysbiosis despite secretion of  
584 compensatory IgM. *Sci. Rep.* **9**, 13574 (2019).
- 585 69. Mirpuri, J. *et al.* Proteobacteria-specific IgA regulates maturation of the intestinal microbiota.  
586 *Gut Microbes* **5**, 28–39 (2014).
- 587 70. Suzuki, K. *et al.* Aberrant expansion of segmented filamentous bacteria in IgA-deficient gut. *Proc.*  
588 *Natl. Acad. Sci.* **101**, 1981–1986 (2004).
- 589 71. Macpherson, A. J. A Primitive T Cell-Independent Mechanism of Intestinal Mucosal IgA  
590 Responses to Commensal Bacteria. *Science* **288**, 2222–2226 (2000).
- 591 72. Kubinak, J. L. *et al.* MyD88 Signaling in T Cells Directs IgA-Mediated Control of the Microbiota to  
592 Promote Health. *Cell Host Microbe* **17**, 153–163 (2015).
- 593 73. Kubinak, J. L. *et al.* MHC variation sculpts individualized microbial communities that control  
594 susceptibility to enteric infection. *Nat. Commun.* **6**, 8642 (2015).
- 595 74. Nakajima, A. *et al.* IgA regulates the composition and metabolic function of gut microbiota by  
596 promoting symbiosis between bacteria. *J. Exp. Med.* **215**, 2019–2034 (2018).
- 597 75. Zuo, T. *et al.* Gut fungal dysbiosis correlates with reduced efficacy of fecal microbiota  
598 transplantation in *Clostridium difficile* infection. *Nat. Commun.* **9**, 3663 (2018).
- 599 76. Vermeire, S. *et al.* Donor Species Richness Determines Faecal Microbiota Transplantation Success  
600 in Inflammatory Bowel Disease. *J. Crohns Colitis* **10**, 387–394 (2016).

- 601 77. Paramsothy, S. *et al.* Specific Bacteria and Metabolites Associated With Response to Fecal  
602 Microbiota Transplantation in Patients With Ulcerative Colitis. *Gastroenterology* **156**, 1440-  
603 1454.e2 (2019).
- 604 78. Lavelle, A. & Sokol, H. Gut microbiota-derived metabolites as key actors in inflammatory bowel  
605 disease. *Nat. Rev. Gastroenterol. Hepatol.* **17**, 223–237 (2020).
- 606 79. McCarville, J. L., Chen, G. Y., Cuevas, V. D., Troha, K. & Ayres, J. S. Microbiota Metabolites in  
607 Health and Disease. *Annu. Rev. Immunol.* **38**, 147–170 (2020).
- 608 80. Weingarden, A. R. *et al.* Microbiota transplantation restores normal fecal bile acid composition in  
609 recurrent *Clostridium difficile* infection. *Am. J. Physiol.-Gastrointest. Liver Physiol.* **306**, G310–  
610 G319 (2014).
- 611 81. Buffie, C. G. *et al.* Precision microbiome reconstitution restores bile acid mediated resistance to  
612 *Clostridium difficile*. *Nature* **517**, 205–208 (2015).
- 613 82. Staley, C., Kelly, C. R., Brandt, L. J., Khoruts, A. & Sadowsky, M. J. Complete Microbiota  
614 Engraftment Is Not Essential for Recovery from Recurrent *Clostridium difficile* Infection following  
615 Fecal Microbiota Transplantation. *mBio* **7**, e01965-16, /mbio/7/6/e01965-16.atom (2016).
- 616 83. Seekatz, A. M. *et al.* Restoration of short chain fatty acid and bile acid metabolism following fecal  
617 microbiota transplantation in patients with recurrent *Clostridium difficile* infection. *Anaerobe* **53**,  
618 64–73 (2018).
- 619 84. Brown, J. R.-M. *et al.* Changes in microbiota composition, bile and fatty acid metabolism, in  
620 successful faecal microbiota transplantation for *Clostridioides difficile* infection. *BMC*  
621 *Gastroenterol.* **18**, 131 (2018).
- 622 85. Ott, S. J. *et al.* Efficacy of Sterile Fecal Filtrate Transfer for Treating Patients With *Clostridium*  
623 *difficile* Infection. *Gastroenterology* **152**, 799-811.e7 (2017).
- 624 86. Zuo, T. *et al.* Bacteriophage transfer during faecal microbiota transplantation in *Clostridium*  
625 *difficile* infection is associated with treatment outcome. *Gut* gutjnl-2017-313952 (2017)  
626 doi:10.1136/gutjnl-2017-313952.

- 627 87. Broecker, F. *et al.* Long-term changes of bacterial and viral compositions in the intestine of a  
628 recovered *Clostridium difficile* patient after fecal microbiota transplantation. *Mol. Case Stud.* **2**,  
629 a000448 (2016).
- 630 88. Staley, C. *et al.* Durable Long-Term Bacterial Engraftment following Encapsulated Fecal  
631 Microbiota Transplantation To Treat *Clostridium difficile* Infection. *mBio* **10**, e01586-19,  
632 /mbio/10/4/mBio.01586-19.atom (2019).
- 633 89. Goloshchapov, O. V. *et al.* Long-term impact of fecal transplantation in healthy volunteers. *BMC*  
634 *Microbiol.* **19**, 312 (2019).
- 635 90. Moss, E. L. *et al.* Long-term taxonomic and functional divergence from donor bacterial strains  
636 following fecal microbiota transplantation in immunocompromised patients. *PLOS ONE* **12**,  
637 e0182585 (2017).
- 638 91. Pigneur, B. & Sokol, H. Fecal microbiota transplantation in inflammatory bowel disease: the  
639 quest for the holy grail. *Mucosal Immunol.* **9**, 1360–1365 (2016).
- 640 92. Kim, K. O. & Gluck, M. Fecal Microbiota Transplantation: An Update on Clinical Practice. *Clin.*  
641 *Endosc.* **52**, 137–143 (2019).
- 642 93. Allegretti, J. R., Mullish, B. H., Kelly, C. & Fischer, M. The evolution of the use of faecal microbiota  
643 transplantation and emerging therapeutic indications. *The Lancet* **394**, 420–431 (2019).
- 644 94. Wilson, B. C., Vatanen, T., Cutfield, W. S. & O’Sullivan, J. M. The Super-Donor Phenomenon in  
645 Fecal Microbiota Transplantation. *Front. Cell. Infect. Microbiol.* **9**, 2 (2019).
- 646 95. Kump, P. *et al.* The taxonomic composition of the donor intestinal microbiota is a major factor  
647 influencing the efficacy of faecal microbiota transplantation in therapy refractory ulcerative  
648 colitis. *Aliment. Pharmacol. Ther.* **47**, 67–77 (2018).
- 649 96. Park, H. *et al.* The success of fecal microbial transplantation in *Clostridium difficile* infection  
650 correlates with bacteriophage relative abundance in the donor: a retrospective cohort study. *Gut*  
651 *Microbes* **10**, 676–687 (2019).

- 652 97. Fuentes, S. *et al.* Microbial shifts and signatures of long-term remission in ulcerative colitis after  
653 faecal microbiota transplantation. *ISME J.* **11**, 1877–1889 (2017).
- 654 98. Li, S. S. *et al.* Durable coexistence of donor and recipient strains after fecal microbiota  
655 transplantation. *Science* **352**, 586–589 (2016).
- 656 99. Fischer, M. *et al.* Predictors of Early Failure After Fecal Microbiota Transplantation for the  
657 Therapy of Clostridium Difficile Infection: A Multicenter Study: *Am. J. Gastroenterol.* **111**, 1024–  
658 1031 (2016).
- 659 100. Gallo, A. *et al.* Fecal calprotectin and need of multiple microbiota transplantation infusions in  
660 *Clostridium difficile* infection. *J. Gastroenterol. Hepatol.* jgh.15072 (2020) doi:10.1111/jgh.15072.
- 661 101. Hirten, R. P. *et al.* Microbial Engraftment and Efficacy of Fecal Microbiota Transplant for  
662 Clostridium Difficile in Patients With and Without Inflammatory Bowel Disease. *Inflamm. Bowel*  
663 *Dis.* **25**, 969–979 (2019).
- 664 102. Ponce-Alonso, M. *et al.* P782 A new compatibility test for donor selection for faecal microbiota  
665 transplantation in ulcerative colitis. *J. Crohns Colitis* **11**, S480–S481 (2017).
- 666 103. Smillie, C. S. *et al.* Strain Tracking Reveals the Determinants of Bacterial Engraftment in the  
667 Human Gut Following Fecal Microbiota Transplantation. *Cell Host Microbe* **23**, 229-240.e5  
668 (2018).
- 669 104. Holvoet, T. *et al.* Assessment of faecal microbial transfer in irritable bowel syndrome with  
670 severe bloating. *Gut* **66**, 980–982 (2017).
- 671 105. Leonardi, I. *et al.* Fungal Trans-kingdom Dynamics Linked to Responsiveness to Fecal Microbiota  
672 Transplantation (FMT) Therapy in Ulcerative Colitis. *Cell Host Microbe* **27**, 823-829.e3 (2020).
- 673 106. Gogokhia, L. *et al.* Expansion of Bacteriophages Is Linked to Aggravated Intestinal Inflammation  
674 and Colitis. *Cell Host Microbe* **25**, 285-299.e8 (2019).
- 675 107. Conceição-Neto, N. *et al.* Low eukaryotic viral richness is associated with faecal microbiota  
676 transplantation success in patients with UC. *Gut* **67**, 1558–1559 (2018).

- 677 108. Ianiro, G. *et al.* Predictors of failure after single faecal microbiota transplantation in patients  
678 with recurrent *Clostridium difficile* infection: results from a 3-year, single-centre cohort study.  
679 *Clin. Microbiol. Infect.* **23**, 337.e1-337.e3 (2017).
- 680 109. Keshteli, A. H., Millan, B. & Madsen, K. L. Pretreatment with antibiotics may enhance the  
681 efficacy of fecal microbiota transplantation in ulcerative colitis: a meta-analysis. *Mucosal*  
682 *Immunol.* **10**, 565–566 (2017).
- 683 110. Ishikawa, D. *et al.* Changes in Intestinal Microbiota Following Combination Therapy with Fecal  
684 Microbial Transplantation and Antibiotics for Ulcerative Colitis: *Inflamm. Bowel Dis.* **23**, 116–125  
685 (2017).
- 686 111. Le Roy, T. *et al.* Comparative Evaluation of Microbiota Engraftment Following Fecal Microbiota  
687 Transfer in Mice Models: Age, Kinetic and Microbial Status Matter. *Front. Microbiol.* **9**, 3289  
688 (2019).
- 689 112. Ji, S. K. *et al.* Preparing the Gut with Antibiotics Enhances Gut Microbiota Reprogramming  
690 Efficiency by Promoting Xenomicrobiota Colonization. *Front. Microbiol.* **8**, 1208 (2017).
- 691 113. Freitag, T. L. *et al.* Minor Effect of Antibiotic Pre-treatment on the Engraftment of Donor  
692 Microbiota in Fecal Transplantation in Mice. *Front. Microbiol.* **10**, 2685 (2019).
- 693 114. Khan, A. A. *et al.* Polymorphic Immune Mechanisms Regulate Commensal Repertoire. *Cell Rep.*  
694 **29**, 541-550.e4 (2019).
- 695 115. Tian, C. *et al.* Genome-wide association and HLA region fine-mapping studies identify  
696 susceptibility loci for multiple common infections. *Nat. Commun.* **8**, 599 (2017).
- 697 116. Andeweg, S. P., Keşmir, C. & Dutilh, B. E. *Quantifying the impact of Human Leukocyte Antigen on*  
698 *the human gut microbiome.* <http://biorxiv.org/lookup/doi/10.1101/2020.01.14.907196> (2020)  
699 doi:10.1101/2020.01.14.907196.
- 700 117. Levine, A. *et al.* Crohn's Disease Exclusion Diet Plus Partial Enteral Nutrition Induces Sustained  
701 Remission in a Randomized Controlled Trial. *Gastroenterology* **157**, 440-450.e8 (2019).

- 702 118. Svolos, V. *et al.* Treatment of Active Crohn’s Disease With an Ordinary Food-based Diet That  
703 Replicates Exclusive Enteral Nutrition. *Gastroenterology* **156**, 1354-1367.e6 (2019).
- 704 119. Sabino, J., Lewis, J. D. & Colombel, J.-F. Treating Inflammatory Bowel Disease With Diet: A Taste  
705 Test. *Gastroenterology* **157**, 295–297 (2019).
- 706 120. Frank, D. N. *et al.* Disease phenotype and genotype are associated with shifts in intestinal-  
707 associated microbiota in inflammatory bowel diseases: *Inflamm. Bowel Dis.* **17**, 179–184 (2011).
- 708 121. Knights, D. *et al.* Complex host genetics influence the microbiome in inflammatory bowel  
709 disease. *Genome Med.* **6**, 107 (2014).
- 710 122. Mizuno, S. *et al.* Bifidobacterium-Rich Fecal Donor May Be a Positive Predictor for Successful  
711 Fecal Microbiota Transplantation in Patients with Irritable Bowel Syndrome. *Digestion* **96**, 29–38  
712 (2017).

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715 CD, NR, and HS designed the systematic review, did the literature search and assessed data quality.  
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