


Washed preparation of faecal microbiota changes the transplantation related safety, quantitative method and delivery

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ulcerative colitis (UC), which made UC a considerable disease model to reflect the quality control of faecal microbiota preparation. We defined one treatment unit as 10 cm³ microbiota precipitation (1.0 × 10¹³ bacteria) based on enriched microbiota instead of rough faecal weight. For delivering microbiota, colonic transendoscopic enteral tube is a promising way especially for multiple WMTs or frequent colonic administration of drugs combined with WMT. This study should help improve the better practice of FMT for helping more patients in the future.

Summary

The safety, quantitative method and delivery of faecal microbiota transplantation (FMT) vary a lot from different countries in practice. Recently, the improved methodology of FMT based on the automatic filtration, washing process and the related delivery was named as washed microbiota transplantation (WMT). First, this study aimed to describe the methodology development of FMT from manual to washing preparation from 2012 to 2021 in China Microbiota Transplantation System (CMTS), a centralized stool bank for providing a national non-profit service. The secondary aim is to describe donor screenings, the correlation between faecal weight and treatment doses, incidence of adverse events and delivery decision. The retrospective analysis on the prospectively recorded data was performed. Results showed that the success rate of donor screening was 3.1% (32/1036). The incidence rate of fever decreased significantly from 19.4% (6/31) in manual FMT to 2.7% (24/902) in WMT in patients with

Introduction

Fecal microbiota transplantation (FMT) has been used in clinical medicine for over one thousand years (Zhang *et al.*, 2012). As an effective method for reconstructing the gut microbiota of recipients, FMT is gaining great attention in the increasing clinical research settings, including gastrointestinal disorders, neurological disorders, cardiovascular disease and even cancer (Bajaj *et al.*, 2017; Kang *et al.*, 2017; Wang *et al.*, 2018; Costello *et al.*, 2019; Ianiro *et al.*, 2020a; Baruch *et al.*, 2021). Stool banks are emerging as high-level facilities to improve the safety and efficacy of FMT.

The recent surveys on doctors, medical students and patients showed that they have a negative perception of FMT (Paramsothy *et al.*, 2015a; Ren *et al.*, 2016; McSweeney *et al.*, 2020), especially due to its manual preparation methods (Zipursky *et al.*, 2012, 2014; Wu *et al.*, 2019). In 2019, two serious adverse events (SAEs) (death and infection) occurred due to drug-resistant *E. coli* bacteraemia transmitted by FMT (DeFilipp *et al.*, 2019), which aroused the public attention to the safety of FMT. Therefore, physicians seemed to be more receptive to but cautious of FMT with consistently increasing evidence of the safety and efficacy of FMT.

The methodology of FMT based on the automatic washing process and the related delivery since 2014 was named as washed microbiota transplantation (WMT) (Zhang *et al.*, 2020a), which was released in Nanjing consensus by the (Fecal Microbiota Transplantation-standardization Study Group, 2020). The integrated clinical findings, animal experiments and *in vitro* tests

Received 14 December, 2021; accepted 30 April, 2022.

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Microbial Biotechnology (2022) 15(9), 2439–2449

doi:10.1111/1751-7915.14074

Funding information

This work was supported by public donated grants from Intestine Initiative and the Nanjing Medical University Fan Daiming Research Funds for Holistic Integrative Medicine; National Natural Science Foundation of China (81670495 and 81873548), and China National Center for Clinical Research of Digestive Diseases (2015BAI13B07).

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demonstrated that the core of WMT is 'washing', which significantly decreased adverse events (AEs) in patients with mucosal barrier injury by removing certain bacterial fragments, pro-inflammatory metabolites, soluble molecules and virus (Zhang *et al.*, 2020a). The traditional manual FMT for determining the treatment dose was mainly based on the faecal weight or the volume of faecal water. Washing process can quantify enriched microbiota for precise treatment. Our recent studies indicated that the improved methodology of the washing process does not decrease the efficacy of FMT while improving the safety (Wang *et al.*, 2018; Ding *et al.*, 2019; Zhang *et al.*, 2020a).

We started the stool bank in Nanjing Medical University for clinical research in 2012 (Zhang *et al.*, 2013; Cui *et al.*, 2015). Then, the hospitalized stool bank was supported by China National Clinical Research Centre for Digestive Diseases in 2015, and was named as fmtBank, a stool bank providing national non-profit FMT service for refractory intestinal infections (Cui *et al.*, 2016; Zhang *et al.*, 2018). In 2017, this system was developed as China Microbiota Transplantation System (CMTS). This article aims to report the methodological development of FMT for improving quantitative method, the safety, delivery and the related clinical decision.

Results

Donor screening and population recruited

From 2014 to 2021, 1,036 (95.1%, 1036/1089) valid questionnaires were collected from 1,089 individuals (aged 20.54 ± 1.51 , BMI 21.04 ± 3.37 , Fig. 1). Four hundred and ninety-five candidates (47.8%, 495/1036) were excluded because of irregular bowel habits and 262 candidates (25.3%, 262/1036) were unqualified due to BMI > 24 or < 18. Among 541 candidates who expressed having regular bowel habits in the questionnaire, there were other reasons for which candidates were excluded. The most common reason for elimination was candidates with abnormal immunity-related diseases (71.9%, 389/541), followed by typical

digestive system diseases (59.3%, 321/541), medication history within 6 months (39.4%, 213/541), family history and genetic diseases (28.1%, 152/541), and infectious diseases (17.2%, 93/541). Thirty-two eligible donors (3.1%, 32/1036) were finally screened for providing faecal microbiota.

Faecal weight and treatment

During medical practice, the centre defined one treatment unit (1 U) as 10 cm^3 microbiota precipitation (1.0×10^{13} bacteria) for convenience to calculate the volume of enriched washed microbiota (Group., 2020; Zhang *et al.*, 2020a). Generally, a full dose of 5 units is required in single treatment for adults and children over than 7 years old. And the dose of enriched microbiota for children from 1–7 year-old ranged from 1 to 5 units (Group., 2020; Zhang *et al.*, 2020a). The frequency of WMTs for most patients in each hospitalization case ranges from one to three times.

As shown in Fig. 2A, among total 2517 donations, the faecal weight was not well correlated with the amount of enriched washed microbiota (which we defined as treatment unit) ($r = 0.65$, 95% CI, 0.63–0.68, $P < 0.0001$), the correlation between faecal weight and the amount of enriched washed microbiota differs from each other in children donor ($r = 0.37$, 95% CI, 0.21–0.52, $P < 0.0001$, Fig. 2B) and adult donor ($r = 0.71$, 95% CI, 0.64–0.76, $P < 0.0001$, Fig. 2B). Even for the same weight of faeces from different defecation samples in one donor, the relationship between the faecal weight and enriched microbiota was not satisfactory (Fig. 2C).

Generally, the amount of enriched microbiota accumulates with the increase of faecal weight. Based on the stratified analysis of faecal weight, donations that meet a minimum weight of 150 g are cost-effective to handle because more than 75% (525/692) of which can be used to treat at least one patient (Fig. 3A). Donations less than 50 g cannot even meet the basic treatment unit for single WMT (Fig. 3B).

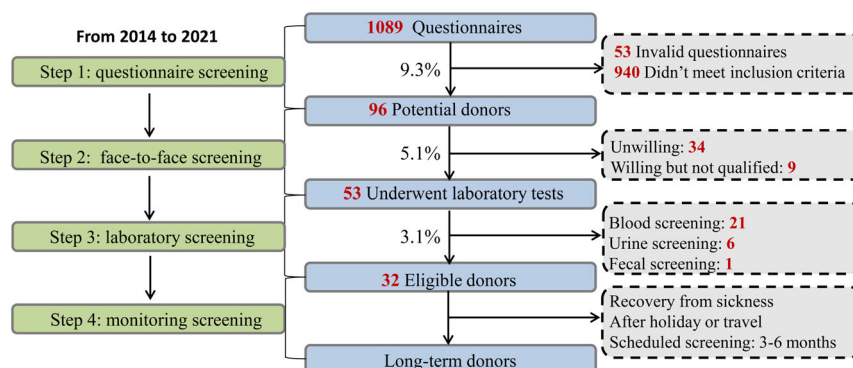


Fig. 1. Donor screening flow.

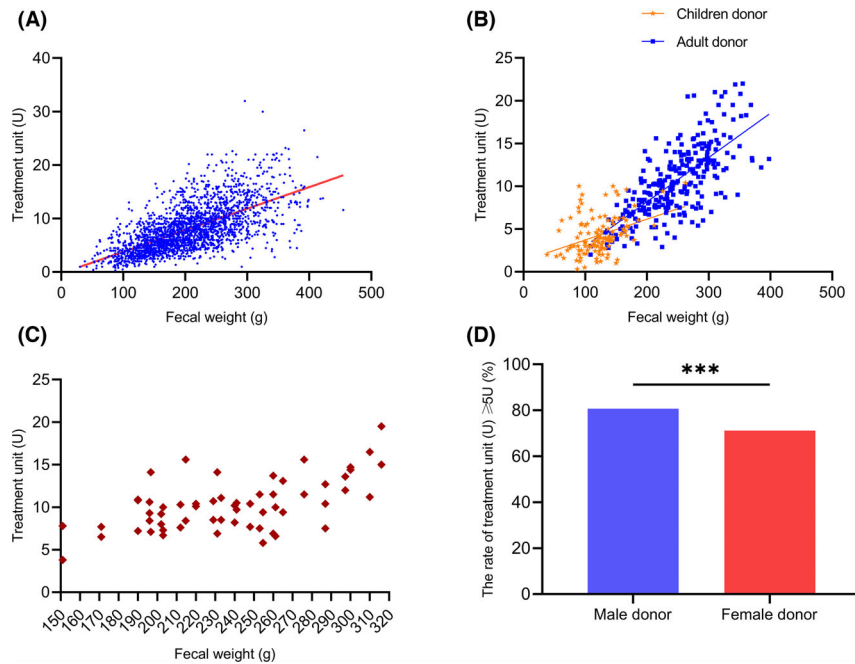


Fig. 2. Correlation between the faecal weight and the amount of enriched washed microbiota.

A. Correlation between the faecal weight and enriched microbiota in WMT donors ($n = 2517$).

B. Difference in correlation between the faecal weight and enriched microbiota from one children donor ($n = 124$) and one adult donor ($n = 247$).

C. Different doses of enriched microbiota among the same weight of faeces.

D. The rate of treatment unit differs between male and female donor. Correlation analysis was performed using Spearman correlation analysis. Statistical comparisons were performed using chi-square test; $*P < 0.05$, $**P < 0.01$, $***P < 0.001$.

Even for the same range of faecal weight, the microbiota output differs from each other among different donors. As shown in Fig. 3C, donor A showed more doses of microbiota output than donor B and C among each range of faecal weight. Male donors are more likely to have better microbiota output than female donors ($P < 0.001$) (Fig. 2D).

Different delivering ways

Colonic transendoscopic enteral tube (TET) is recommended for patients who need multiple WMTs or colonic administration of drugs combined with WMT. Figure 4 showed that delivering WMT through colonic TET is a primary method for ulcerative colitis (UC) (67.2%, 627/933). While gastroscope (55.5%, 528/951) and mid-gut tube (32.4%, 308/951) are two predominant mid-gut delivering ways in patients with Crohn's disease (CD). For refractory intestinal infections rescued by WMT, 82.8% (211/255) of patients underwent WMT through mid-gut tube. No tube obstruction was observed through mid-gut or colonic TET for WMT.

Microbiota transplantation -related adverse events

The current data from a total of 574 patients with inflammatory bowel disease (285 UC and 289 CD) who

underwent FMT/WMT were recorded in CMTS from 2012 to 2021. These patients were classified as those who had mucosal barrier injury. When using 1 month cut-off to define short-term or long-term AEs related to FMT/WMT, analysis showed that only two AEs were reported to occur one month after treatment, which was consistent with previous findings (Fig. 5A and B). During short-term follow-up, the rate of undefined AEs in patients with UC significantly decreased from 35.5% (11/31) by manual FMT to 7.2% (65/902) by WMT ($P < 0.001$, Fig. 5C). The rate of AEs in patients with CD decreased from 21.7% (15/69) to 4% (35/882) ($P < 0.001$, Fig. 5E).

The rate of fever after microbiota transplantation significantly decreased from 19.4% (6/31) in manual FMT to 2.7% (24/902) in WMT in patients with UC ($P < 0.001$, Fig. 5D). However, the rate of fever was not related to the fresh or frozen status of microbiota storage in patients with UC (3.2% vs. 2.9%, 28/863 and 2/70 respectively, $P > 0.05$, Fig. 5F).

Discussion

The methodological development from FMT to WMT mainly involves donor screening, laboratory preparation and delivery. In CMTS, the candidate donors are mainly from college students and children and the success rate

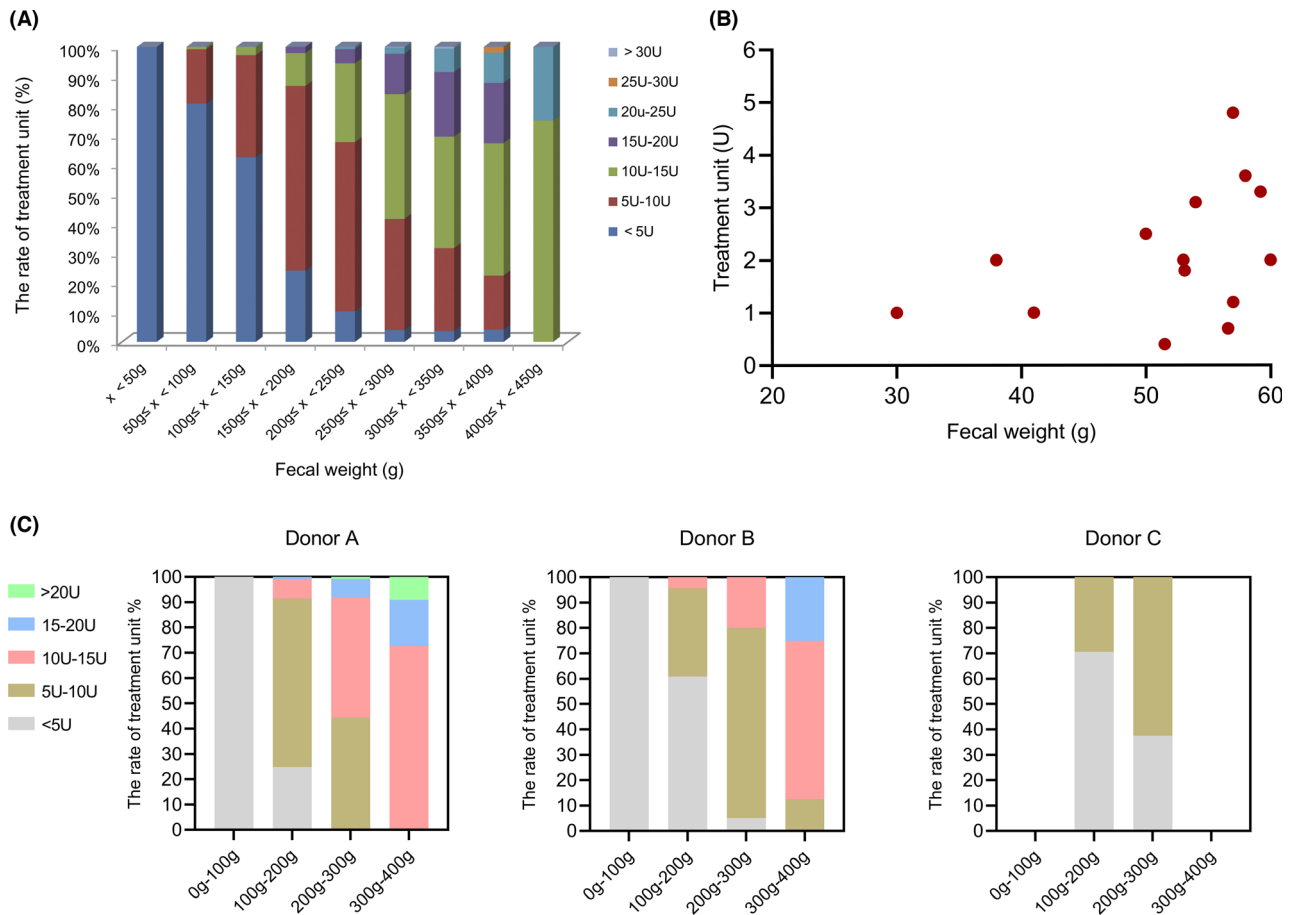


Fig. 3. Specific distribution of faecal weight and the related treatment unit. A. X-axis: The distribution of faecal weight. Y-axis: The rate of treatment unit. B. The detailed distribution of treatment unit among faecal weight less than 60 g. C. Different distribution of treatment unit among three different donors.

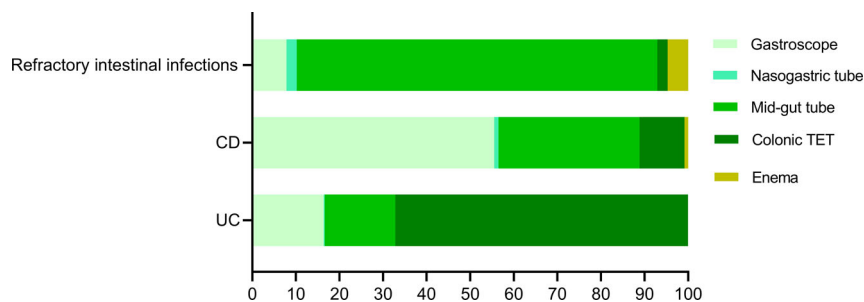


Fig. 4. Different delivering ways for WMT in patients with UC, CD and refractory intestinal infections. The X-axis shows the percentage of different delivering ways. CD, Crohn's disease; TET, transendoscopic enteral tube; UC, ulcerative colitis.

of donor screening was 3.1%. The rate varies from different stool banks, such as OpenBiome (3%) in the United States (Kassam *et al.*, 2019), Canada (2.2%) (Craven *et al.*, 2017), the Netherlands (2.4%) (Termeer *et al.*, 2017), Australian (10%) (Paramsothy *et al.*, 2015b) and Italy (25%) (Ianiro *et al.*, 2021). Importantly,

the methods on donor screening were changing dynamically all over the world (Woodworth *et al.*, 2017; Ianiro *et al.*, 2020b; McSweeney *et al.*, 2020).

We found that the faecal weight was not well correlated with the amount of enriched microbiota. Quantified washed microbiota preparations can provide precise

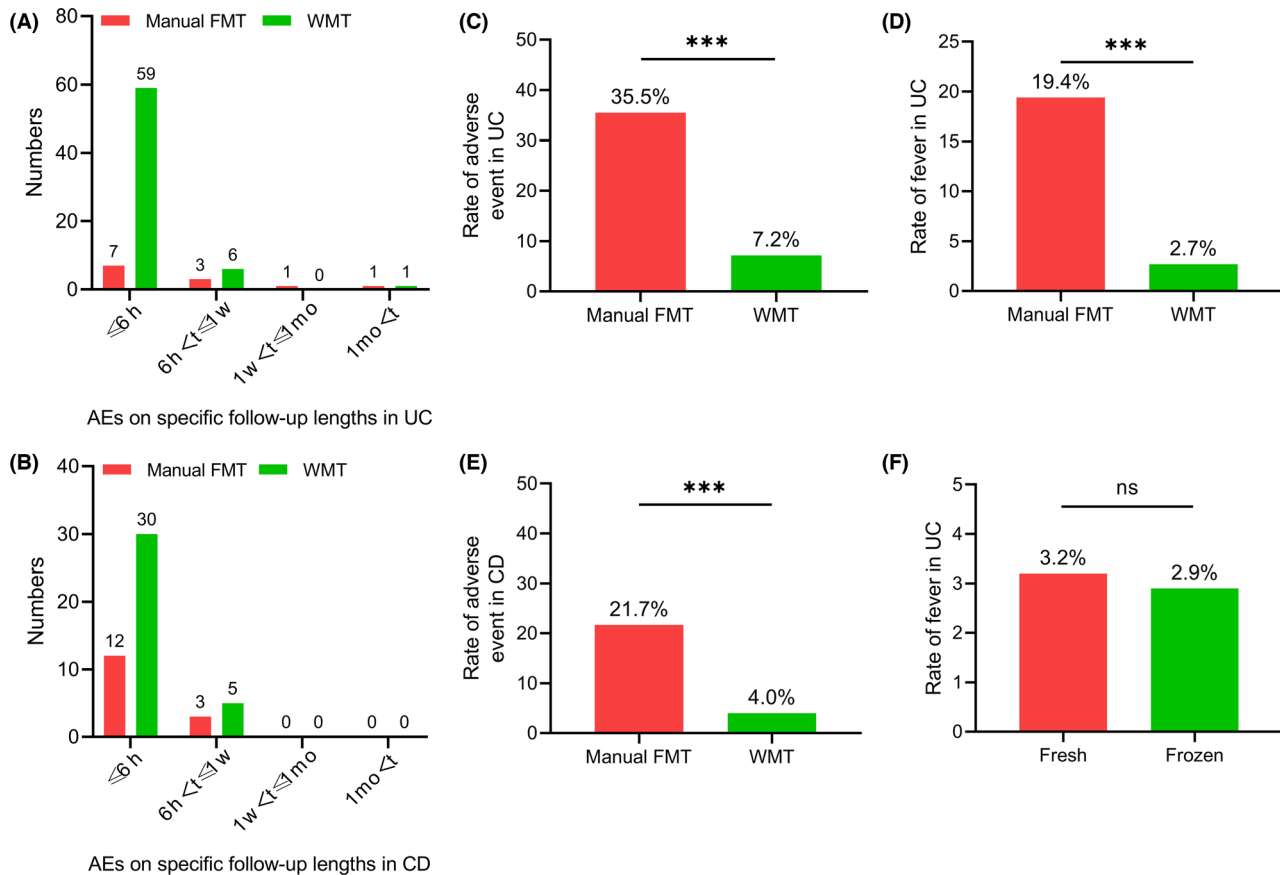


Fig. 5. The incidence rate of microbiota transplantation related adverse events.

A–B. The frequency numbers of AEs on specific follow-up lengths related to manual FMT and WMT in patients with UC and CD respectively.

C. The rate of short-term AEs related to manual FMT and WMT in patients with UC.

D. The rate of fever related to manual FMT and WMT in short-term follow-up in patients with UC.

E. The rate of short-term AEs related to manual FMT and WMT in patients with CD.

F. The rate of fever related to fresh and frozen microbiota in short-term follow-up in patients with UC. AE, adverse events; CD, Crohn's disease; FMT, fecal microbiota transplantation; t, time; UC, ulcerative colitis; WMT, washed microbiota transplantation. Statistical comparisons were performed using chi-square test; ns: no significance, * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.

doses of microbiota. The analysis on the precise dose of enriched faecal microbiota showed that the faecal weight less than 50 g was not cost-effective and provided very few doses for WMT treatment, thus faeces from donors less than 50 g were not recommended in Nanjing consensus. We defined 10 cm^3 microbiota precipitation (1.0×10^{13} microbial cells) as one basic treatment unit (Group., 2020; Zhang *et al.*, 2020b). Our treatment strategy is 1–5 units for each delivery in children and adults. The present findings highlight the significance of metrological faecal microbiota and a favourable cost-effectiveness during the washing process.

The delivery decision is made based on the clinician's judgement of patients' condition. For example, patients who have difficulty in swallowing capsules may choose mid-gut tubes (nasojunal tube, gastrostomy tube or jejunostomy tube). Mid-gut tube is useful for repeated infusion of microbiota and the combination of enteral

nutrition. As for patients who are not suitable for the upper gastrointestinal delivery, lower gastrointestinal delivery such as colonic TET, colonoscopy and enema can be taken into consideration. Since the treatment dose is based on washed microbial cells, different delivering ways have no effect on it. However, different studies which use manual faecal microbiota suspension recommend different treatment volume of suspension according to upper or lower gastrointestinal delivery because of physiological structure (different parts of the digestive tract can hold different volumes of fluid). The data from this real-world study indicated that colonic TET is the predominant delivery for WMT. The safety of colonic TET has already been proved in adults (≥ 18 years old) and children aged over 3 years old (Zhang *et al.*, 2020b; Zhong *et al.*, 2021a,b). Colonic TET is helpful to increase the frequency of WMT for increasing the efficacy, which is the aim of step-up WMT

strategy (Ding *et al.*, 2020; Xiang *et al.*, 2020). A prospective study including 224 patients reported 97.8% of satisfaction with the colonic TET, and the success rate of colon TET was 100% (Zhang *et al.*, 2020b). Recently, the colonic TET was first time used to rescue endoscopy-associated perforation, increasing our confidence in the different usage of colonic TET (Zhang *et al.*, 2021a,b). Colonic TET is also a convenient way for local microbiota analysis because researchers could directly acquire microbiota through this tube (Liu *et al.*, 2021). However, the colonic TET is not recommended for delivering the manual preparation of FMT because of the possible obstruction in tube (Wang *et al.*, 2019).

This study further confirms our previous finding that washing preparation is an independent contributor to reduce the incidence rate of FMT-related AEs by improving intestinal mucosal permeability and decreasing pro-inflammatory metabolites (Zhang *et al.*, 2020a). So far, water, sterile saline, phosphate-buffered saline have been used as a vector solvent of faecal material (Liao and Shollenberger, 2003; Mattila *et al.*, 2012; Cammarota *et al.*, 2017, 2019). Normal saline is used to prepare most faecal microbiota suspensions, as this solvent enables better preservation of microbes (Mattila *et al.*, 2012). Metabolism analysis has proved a significant decrease in pro-inflammatory metabolites during the washing process such as prostaglandin G2, leukotriene B4, TRPV1 and the related differentially enriched metabolic pathways, which play important roles in fever and inflammation. Metagenomic next-generation sequencing (NGS) indicated the increasing types and amount of viruses could be washed out during the washing process. Further animal experiments and *in vitro* screening also supported the evidence for linking the clinical findings to the safety of WMT (Zhang *et al.*, 2020a). Published data from CMTS have shown the reliable safety of rescue WMT in critically ill patients (Dai *et al.*, 2019) and patients with graft-versus-host disease (Qi *et al.*, 2018). The recent studies in other centres showed the similar findings on the safety and efficacy of WMT (Chen *et al.*, 2020; Zheng *et al.*, 2021; Zhong *et al.*, 2021a,b).

AEs were generally calculated in two ways: the frequency of AEs post-FMT during follow-up, or the proportion of patients with symptoms. Recently, Kelly *et al.* reported the initial results from the American Gastroenterological Association FMT National Registry programme using the proportion of patients with symptoms to evaluate the safety of FMT in CDI (Kelly *et al.*, 2021). Mayo Clinic reported the incidence rate of AEs calculated by the proportion of patients with symptoms: 13% gastrointestinal symptoms, 10% weight gain and 11.8% new infections (Saha *et al.*, 2021). In our recent systematic review, the global data calculated by using the frequency of AEs post-FMT during follow-up showed that the incidence rate of

FMT-related AEs, FMT-related SAEs and death was observed in 19%, 1.39% and 0.12% of total FMT courses, respectively (Marcella *et al.*, 2021), which shows a higher trend than the online data of CMTS (<http://fmtbank.org/>).

Which item should be selected as a quality control model of WMT is another practical issue. According to this study, since one month post-FMT was regarded as a cut-off between short-term and long-term follow-up, more attention needs to be paid to the short-term evaluation. Our recent systematic review on the global incidence of FMT-related AEs from 2000 to 2020 demonstrated that FMT-related SAEs, including serious infections and deaths, had been reported in 1.4% of patients who underwent FMT (0.99% microbiota-related SAEs) (Marcella *et al.*, 2021). All reported FMT-related SAEs were happened in patients with mucosal barrier injury, which was defined as endoscopically confirmed mucosa broken (e.g. ulcerative or erosive lesions) or clinically confirmed acute intestinal infections with determined or undetermined pathogens (See *et al.*, 2013; Marcella *et al.*, 2021). Based on our previous study, Wang *et al.* reported a 1 month cut-off could be suggested to define short-term and long-term AEs of FMT/WMT because no AE beyond 1 month was observed in a total of 184 frequencies of FMT/WMT in CD patients. During a follow-up of up to eight years, only two patients reported myasthenia gravis and rash/pruritus, respectively 1 month after FMT/WMT. The remaining AEs all happened within 1 month, especially within 6 hours after FMT/WMT. Therefore, the incidence rate of fever as a short-term FMT/WMT-related AE in patients with UC should be considered as a valuable indicator to evaluate the quality of laboratory processes for WMT. In another words, the safety of microbiota transplantation in patients with light intestinal barrier injury or even without mucosa damage, such as constipation, irritable bowel syndrome and autism, cannot really reflect the quality control of delivered microbiota.

The development of WMT from FMT is similar to the history of blood transfusion. The revolutionary development of blood transfusion significantly improves the safety (Goodnough *et al.*, 2013). This public platform works hard to promote the transformation of WMT from clinical benefits to breakthrough in disease cognition. The latest pilot studies indicate the role of WMT on radiation colitis (Ding *et al.*, 2020), complex infections (Dai *et al.*, 2019), recurrent invasive fungal infection (Wu *et al.*, 2021), refractory *Helicobacter pylori* infection (Ye *et al.*, 2020).

However, there are limitations in this study. We did not integrate the microbiome analysis and efficacy evaluation on WMT, although we have reported these in our previous studies. The policy on the use of FMT in China is the permitted medical therapy for CDI and many other diseases. This is the reason why we focused on the studies in populations beyond CDI, which has already

been well studied in Europe and North America (Allegretti *et al.*, 2019; Baunwall *et al.*, 2021).

In conclusion, donor screening, washing process, dose, time, frequency and delivery were all closely related to the safety and efficacy of WMT. The experience of washed preparation of faecal microbiota should be helpful to improve the better practice of FMT for helping more patients in the future.

Experimental procedures

Microbiota transplantation system and data management

CMTS aims to support non-profit WMT practice and research in China for long-term evaluation of the decision, treatment, efficacy and safety of microbiota transplant (Fig. 6A). The data were recorded by research teams and doctors.

Donor recruitment and evaluation

Faeces donations were approved by the institutional ethics committee of the Second Affiliated Hospital of

Nanjing Medical University. Healthy adults and adolescents (preferably aged 6–24 years old) are potential donors in the clinical practice according to the consensus (Group., 2020). Candidates were informed of the potential risks and benefits of WMT for recipients (Wu *et al.*, 2019; McSweeney *et al.*, 2020) and should provide written informed consent. Children donors should be screened after obtaining parental consent. Questionnaire screening, face-to-face screening and laboratory screening are taken step-by-step to exclude candidates based on the criteria including age, physiology, pathology, psychology, integrity, time, environment and recipient status (Ding *et al.*, 2019; Zhang *et al.*, 2019).

Washed microbiota preparation and quality control

The method for preparation of microbiota is based on the automatic microbiota purification system (GenFMter, FMT Medical, Nanjing, China) followed with centrifugation plus suspension three times in a specially designed exclusive laboratory at good manufacture practice level (Cui *et al.*, 2016; Zhang *et al.*, 2018) (Fig. 6B). The time

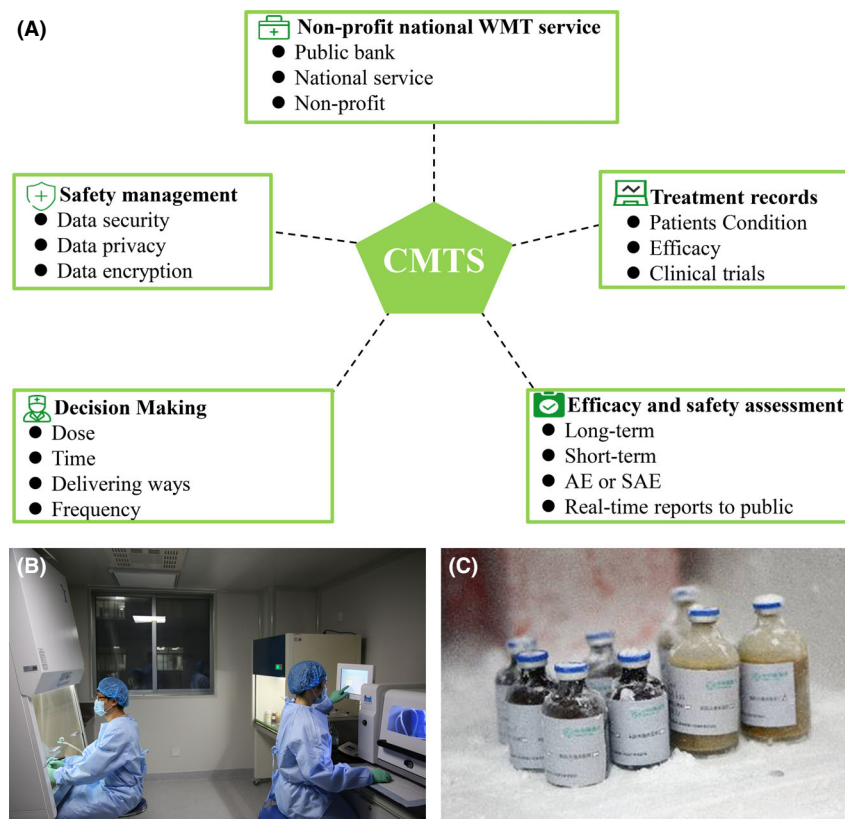


Fig. 6. The framework and laboratory of CMTS.

A. The function framework of CMTS.

B. The laboratory for washed microbiota preparation.

C. The frozen washed microbiota. AE, adverse event; CMTS, China Microbiota Transplantation System; SAE, serious adverse event; WMT, washed microbiota transplantation.

from defecation of a donor, laboratory preparation for enriching microbiota to the time of microbiota delivering or microbiota storing was limited within one hour (He *et al.*, 2017; Zhang *et al.*, 2018; Ding *et al.*, 2019), which was called 'one-hour protocol' (Group., 2020).

Faeces are collected on-site by a disposable faeces container in a dedicated room only for donors. The donated faeces (generally ≥ 50 g) and sterile saline were put together into the GenFMTer at the ratio of 500 ml of saline per 100 g faeces to become faecal microbiota suspension. The faecal suspension is transferred to centrifugation tubes for centrifugation with 700 *g* for 3 min and the supernatant is discarded. The above centrifugation plus washing and suspension using sterile saline should be repeated three times (Zhang *et al.*, 2020a). Washed microbiota suspension consists of final precipitation and vector solution with the volume ratio of 1:2 for fresh use. Frozen samples can be safely stored by mixed with 10% glycerol at -80°C for frozen use. The frozen samples should be thawed in a warm water bath (37°C water temperature, 30–45 min from -80°C to 37°C). Retention of stool samples is recommended for 'look-back' testing.

During medical practice, we defined one treatment unit (1 U) as 10 cm^3 microbiota precipitation (1.0×10^{13} bacteria) to calculate the volume of enriched washed microbiota. We used fluorescent staining for total bacteria counts (Magge *et al.*, 2009; Emerson *et al.*, 2017).

Delivering ways for washed microbiota transplantation

The delivery of microbiota transplantation includes the upper gut, mid-gut and lower gut (Peng *et al.*, 2016). Oral intake (capsules or drinking) and nasogastric tube are two means of upper gut delivery way (Lee *et al.*, 2016; Zhang *et al.*, 2021a,b). Mid-gut delivery includes infusing the microbiota suspension into the small intestine beyond the second duodenal segment through endoscopy and mid-gut tube (nasojejun tube or mid-gut TET) (Cui *et al.*, 2015; Long *et al.*, 2018). Faecal microbiota can also be delivered to the lower gut through colonoscopy, enema, stoma in ileocolon and colonic TET in adults and paediatrics (Peng *et al.*, 2016; Wen *et al.*, 2020; Zhong *et al.*, 2021a,b). The endoscopic delivery and TET technique can be performed by endoscopist and gastroenterologist in the CMTS.

Clinical trials and non-profit national washed microbiota transplantation service

The clinical data of this study was based on our pooled registered trials for UC, CD and refractory intestinal infections (ClinicalTrials.gov, NCT01790061, NCT01793831, and NCT03895593). The system also records the

information about the population for attending clinical trials such as autism, epilepsy and radiation enteritis. However, data from these trials were not included in this study. CMTS provides rescue WMT service for patients nationally (Fig. 6C) (Dai *et al.*, 2019). The rescue WMT is a teamwork in which there are at least two intensive care specialists in charge from destination hospitals, professional WMT clinicians, laboratory managers and clinical research coordinators from the CMTS. The rescue team members communicated closely throughout the whole process to ensure the safety fluent work flow of WMT. Conditions, contraindications and the potential risk factors of SAEs were evaluated by the team. Rescue WMT has been successfully applied in treating GVHD and antibiotic-associated diarrhoea (Qi *et al.*, 2018; Huang *et al.*, 2020; Wang *et al.*, 2021).

Safety evaluation for washed microbiota transplantation

In this study, any new onset of symptom and the exacerbation of previous symptoms were recorded and evaluated according to the Code of Federal Regulations and researchers' clinical experience. SAEs include: an AE that is disease spreading, fatal, or life threatening, or requires professional intervention that requires hospitalization or prolonged hospital stays, or results in infertility, congenital anomaly, permanent disability or disfigurement. Daily records were used for AEs within one week after treatment. AEs beyond hospital were recorded via telephone follow-up or hospital visits. AEs were evaluated at 6 h, 1 week, 1 month, 3 months, 6 months, 1 year and every year after FMT/WMT. A one month cut-off could be suggested to define short-term and long-term AEs of FMT/WMT according to previous study (Wang *et al.*, 2018). Causality between AEs and FMT/WMT was categorized as definitely related, probably related, possibly related and unrelated (Kelly *et al.*, 2014). Only FMT/WMT-related (including definitely, probably and possibly related) AEs were reported in this study. A group discussion from two or more physicians is needed to form a common opinion once the relativity of AEs could not be identified. The recorded incidence rate of AEs/SAEs was calculated using the frequency of AEs to the total FMT courses ratio.

Statistical analysis

Descriptive analysis was used in this article. Patient characteristics were evaluated using proportions for categorical variables. Categorical variables were analysed by chi-square test. Pearson correlation coefficient is used to describe the degree of correlation between two variables. *P* values < 0.05 were considered significant. Data were analysed by IBM SPSS 24.0 or GRAPHPAD 7.0.

Ethic approval

All procedures followed were in accordance with the ethical standards of the institutional review board of the Second Affiliated Hospital of Nanjing Medical University and with the Helsinki Declaration of 1975, as revised in 2000. The clinical data of this study was based on our pooled registered trials for UC, CD and refractory intestinal infections (ClinicalTrials.gov, NCT01790061, NCT01793831, and NCT03895593). Informed consent prior to participate was obtained from all patients for being included in the study. For the participants aged below 18, informed consent was obtained from their parents/legally authorized representatives.

Acknowledgements

The authors appreciate the kindly help from Jie Zhang for providing data from China Microbiota Transplantation System, the technical support by Yaoyao Chen and Miao Wang, and language polishing by Ms. Thea Wooten. This work was supported by public donated grants from Intestine Initiative and the Nanjing Medical University Fan Daiming Research Funds for Holistic Integrative Medicine; National Natural Science Foundation of China (81670495 and 81873548), and China National Center for Clinical Research of Digestive Diseases (2015BAI13B07).

Conflicts of interest

FZ conceived the concept of GenFMTer and transendoscopic enteral tubing and devices related to them. The remaining authors declare that they have no competing interests.

Author contributions

GL and WW designed the study, analysed data and wrote the manuscript. PL, QW and BC participated in the data collection. FZ had the study conception, manuscript revision and grants support.

Data availability statement

The data are not publicly available due to the containing information that could compromise research participants' privacy/consent. The basic data that support the findings of this study are available on request from the corresponding author's e-mail.

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