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Establishment of the first stool bank in an Eastern European country and the first series of successful fecal microbiota transplantations in Bulgaria

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Abstract. – OBJECTIVE: For safe implementation and broader application of fecal microbiota transplantation (FMT), quality controlled stool banking is a must. Establishing a stool bank is a complex, time-consuming, and expensive process, making it a real challenge in an Eastern European country. We aimed to establish the first stool bank in Eastern Europe – in Bulgaria.

SUBJECTS AND METHODS: A multidisciplinary team of gastroenterologists, microbiologists, infectionists, and geneticists was set up. We used a questionnaire based on the First European FMT Consensus in order to recruit possible stool donors. Laboratory blood and stool tests were performed on all potential donors.

RESULTS: Between October 2018 and April 2019, 112 donor volunteers completed a questionnaire; 70 (62.5%) were excluded, mainly because of age above 50, an unhealthy BMI, and risk behavior. Fourty-two (37.5%) donor candidates were invited for laboratory testing of blood and feces, of which 12 (28.6%) passed this screening. Of 12 donors, 4 (33%) failed at the following screening test, which is performed every 3-6 months. Finally, 8 (7.14%) active donors were enrolled. Ten successful FMTs were performed on patients with recurrent Clostridium difficile infection.

CONCLUSIONS: Even though we found many healthy volunteers, only a low percentage (7.14%) of them were suitable to become feces donors. Establishing a stool bank in an Eastern European country is essential for making FMT safe and more popular as a treatment method, finding further implementation and regulation of FMT and supporting physicians offering this treatment to their patients.

Key Words:
Stool bank, Microbiota, Fecal microbiota transplantation, Donors.

Introduction

Clostridioides difficile infection (CDI) is the main cause of antibiotic-associated gastrointestinal disease inducing significant morbidity and mortality[1]. The clinical answer to antibiotic treatment of CDI is 80% in the first episode[2] and rapidly drops to only 30-40% in recurrent disease[3].

Fecal microbiota transplantation (FMT) is an extremely efficient treatment against recurrent *Clostridium difficile* infection (rCDI)[1-5]. Guidelines now recommend FMT as a treatment choice for both mild and severe recurrent CDI and refractory CDI[4]. However, there is still inadequate evidence to recommend FMT as a treatment for the first episode of CDI[4]. FMT is increasingly being used for other disorders, including irritable bowel syndrome (IBS), inflammatory bowel disease (IBD), metabolic syndrome, and critically ill patients[6,7], but none of them emerged an evidence-based recommendation to use FMT except that in the context of research[4,5]. Several case reports have demonstrated positive outcomes of using FMT for treating septic shock and intractable diarrhea in the intensive care unit (ICU)[8,9].

To ensure a safe, disseminated, and cost-effective implementation of FMT, stool banks that produce ready-to-use donor feces suspensions

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are needed. Such stool banks may work at a national, institutional, or international level and are currently being set up in several European countries[10,11]. To date, FMT and stool banking protocols differ significantly within institutions, mostly due to the modernity of this treatment method, and the lack of guidelines addressing FMT and stool banking.

In 2017 the First European Consensus for FMT in clinical practice was published, addressing the following key issues: indications for FMT, donor selection, preparation of fecal material, clinical management and fecal delivery, and basic requirements for implementing an FMT centre[4]. However, the Consensus lacks clear guidelines for the establishment of a stool bank, therefore in 2018, the United European Gastroenterology (UEG) established a multidisciplinary working group, including a participant from Bulgaria, to standardize stool banks for FMT across Europe.

In the same year, a similar multidisciplinary working group was established in Bulgaria that aimed to create the first stool bank in an Eastern European country. The working group aimed to describe a standardized process of stool banking and FMT in Bulgaria, based on available consensus reports, including the new consensus on stool banking for FMT[10], previous experiences[4,11], and lessons learned from blood banks[12]. The establishment of a stool bank may facilitate further implementation and regulation of FMT in Bulgaria and will support physicians who offer this treatment to their patients.

Subjects and Methods

In October 2018, a multidisciplinary working group for the establishment of the first stool bank in Bulgaria was set up that aimed at safe and effective FMT. It included gastroenterologists, microbiologist, infectionists and geneticists.

The current study is observational. Between October 2018 and April 2019, we recruited healthy volunteers as potential stool donors. Stool donation is a voluntary action and should not become a subject of commerce. The use of unpaid donors decreases the risk that candidates withhold information during the screening process. We prefer universal donors over patient-selected donors. In individual cases, a patient-selected donor may be accepted; if he or she meets all criteria defined in the screening process for universal donors. We have not used healthcare workers as stool donors to reduce the transmission risk of multiresistant commensals[4]. All donors fulfilled written informed consent. The current study was approved by the Ethics Committee of the Medical University of Sofia. The study protocol conforms to the ethical guidelines of the 1975 Declaration of Helsinki (6th revision, 2008) as reflected in *a priori* approval by the institution's Human Research Committee.

During the first round of the screening process, the medical history and risk behavior of potential donors were evaluated using a dedicated questionnaire based on the First European FMT Consensus (Table I)[4]. A physician evaluated all the results.

Once a potential donor had been found suitable for additional assessment based on the donor questionnaire and the physical examination, he or she experienced blood and feces screening for transmissible pathogens listed in Table II.

When a donor had been approved, he or she performed a second short questionnaire before every donation, evaluating any event that may have occurred between donor approval and donation. Total donor screening based on blood and fecal analyses should be replicated at a minimum every 3-6 months. FMT products made during a donation period were put under quarantine until the repeat donor screening results were ready.

The approved donors deliver their feces within two hours after defecation. After defecation and until further processing, the stool sample was stored at room temperature. If it took more than 30 minutes to deliver the collected feces to the stool bank, temporary storage in a refrigerator was preferred as fecal storage without stabilization buffer significantly changes taxa copiousness from 30 minutes onwards[13,14]. The fecal microbiota is prepared within six hours after defecation.

Storage of FMT Solutions

FMT suspensions were stored in sealed, clean plastic containers, with an individual code guaranteeing the traceability of the sample. Accommodation of FMT suspensions should be supported by the stool bank in a dedicated -80°C freezer with connected alarm notification to ensure constant registration of the storage. Documentation demands biobanking information and management method for registrations, coding, and tracing of the samples.

To guarantee the maximum security and quality of the fecal suspension, it is necessary to specify the best storage time with an expiry date. The

Table I. Screening of potential donors before approval for FMT in the Bulgarian stool bank. The obligatory issues to be evaluated in the questionnaire are listed.

Medical history:
– Previous history of inflammatory, autoimmune, neurodegenerativa or psychiatric disease
– Any current chronic disease
– Any abdominal surgery
– Previous oncological or immunological diagnosis
– Ongoing pregnancy
– Antibiotic, immunosupressive treatment or chemotherapy ongoing, scheduled or received within the last 3 months
– BMI ≥ 18 and ≤ 30 kg/m^2

Current symptoms:
– Diarrhea, obstipation, hematochezia or any other gastrointestinal symptom within the last 3 months
– Fever or rash within the last 3 months
– Any other relevant clinical sign or symptom within the last 3 months

At-risk behavior:
– Current or previous intravenous drug use
– Ongoing high risk sexual behavior
– Travel to high-risk foreign countries within the last 6 months
– Healthcare workers and medical students
– Individual working with animals
– Tattoo, piercing or acupuncture within the last 6 months
– Previous tissue/organ transplantation
– Blood transfusion within the last 6 months
– Recent (< 6 months) needle stick accident

Infectious diseases:
– History or exposure to infectious diseases with chronic activity: particular human immunodeficiency virus (HIV), hepatitis B virus (HBV) or hepatitis C virus (HCV), non-successfully eradicated Helicobacter pylori, syphilis, malaria, trypanosomiasis, tuberculosis, Chagas disease, strongyloidiasis, parasitosis or other gastrointestinal infection.
– Any currently active infection or those of relevance treated within the last 6 months.
– Vaccination with a live attenuated vaccine within the last 8 weeks.

relationship between storage requirements and clinical effectiveness has not been studied. Current data recommend that fecal suspensions may be stored at −20°C for up to one month without loss of efficiency[15]. Long-term accommodation should be at -80°C or lower to limit sample degeneration. High cure rates have been described with frozen FMT suspension collected up to 10 months at -80°C[11,16]; however, this could, in theory, be even longer.

Table II. Blood and feces tests screening for potential stool donors in the Bulgarian stool bank.

Stool analysis:
– **Bacterial enteral pathogens:** *Shiga-like toxin-producing E. coli* (STEC) stx1/stx2, *Shigella spp*., *Campylobacter jejuni/coli/lari*, *Salmonella spp.*, *Yersinia enterocolitica*, *Yersinia pseudotuberculosis, Clostridioides difficile* (GDH, Toxin A/B) *Clostridioides difficile* (PCR), *Helicobacter pylori* (fecal antigen) – **Multidrug-resistant commensals**: Extended spectrum beta-lactamase producing bacteria (ESBLs), *Vancomycin* resistant *enterococci* (VRE), Carbapenem-resistant bacteria, Methicillin-resistant *Staphylococcus aureus.* – **Viruses:** Norovirus, Rotavirus, Adenovirus, Парechovirus, Astrovirus, Enterovirus. – **Parasites:** *Cryptosporidium spp*., *Giardia lamblia*, *helminths*, *Entamoeba histolytica and dispar*, *Dientamoeba fragilis*, *Cyclospora isospora*, *microsporidia*. – **Others:** Candida, fecal calprotectin.
Blood analysis:
– **General laboratory:** complete blood count, CRP, ESR, creatinine, bilirubin, albumin, electrolytes, ALT, AST, GGT, AP. – **Viruses**: Hepatitis A (IgM), Hepatitis B (HbsAg, HBc-Total), Hepatitis C (anti HCV), Hepatitis E (IgM), HIV 1 и 2, CMV, EBV. – **Bacteria:** *Treponema pallidum* (TPHA)

A sample of the first donor feces and/or of the processed FMT suspension was collected for a minimum of two years for retrospective quality evaluation in case of an unfavorable event.

Distribution of FMT suspensions should be performed on dry-ice shipment through a certified courier service.

Preparation of FMT Solution and Fecal Microbiota Transplantation

We used just fresh fecal material from approved donors for FMT in our patients. We utilized a minimum amount of 50 g of feces, which was suspended in saline in a 1:5 ratio using a blender or manual effort and sieved in order to avoid the clogging of infusion. The fresh stool was used within 6 hours after defecation. In order to protect anaerobic bacteria, the storage and preparation were as brief as possible. We prepared everything in a dedicated disinfected space, and we used protective gloves and facial masks.

We froze fecal material from each approved donor for further FMT. We used at least 30 g of donor feces and 150 mL of saline solution. Before freezing, the cryoprotectant glycerol was added up to a final concentration of 10%. The final suspension was clearly labelled and traceable and stored at –80°C[4].

In all the patients, we inserted fresh fecal suspension by colonoscopy. After the FMT, the patients were followed-up every 1-3 months to assess the efficacy.

Statistical Analysis

The statistical analysis was performed using SPSS for Windows, Version 25.0. (SPSS Inc., Chicago, USA). Descriptive statistic for tabular and graphical presentation of results was used.

Results

Between October 2018 and April 2019, 112 donor volunteers completed a questionnaire; 70 (62.5%) were excluded, mainly because of age above 50, an unhealthy BMI, and risk behavior. Fourty-two (37.5%) donor candidates were invited for laboratory testing of blood and feces, of which 12 (28.6%) passed this screening.

The presence of *Helicobacter pylori* fecal antigen and Multi-Drug Resistant Organisms were the most observed exclusion criteria. Of 12 donors, 4 (33%) failed at the following screening test, which is performed every 3-6 months. Finally, 8 (7.14%) active donors were enrolled (Figure 1). The mean age of donors was 30.43 (23-42, SD- 5.74) years.

The approved donors donated fresh fecal material for ten FMTs, performed at Tsaritsa Yoanna University Hospital, Sofia. The FMTs were done by colonoscopy, and the fecal suspension was inserted into the cecum of each patient through the working channel. The mean age of the patients was 61.1 (55-71, SD – 5.22) years. They were fulfilling ESCMID guidelines[17] for recurrent CDI. All the patients were positive for C. difficile glutamate dehydrogenase and toxins A and/or B. All patients had a history of antibiotic treatment, a history of C. difficile infection treated with metronidazole and/or vancomycin, and all sought medical attention due to diarrhea syndrome (average ten bowel movements per day). Four of the patients (40%) reported fever, 8 (80%) had abdominal pain, and 5 (50%) had tachycardia. Four patients (40%) had blood in their stools, all of whom had concomitant IBD – two with Crohn's disease and two with ulcerative colitis.

After FMT, all ten patients responded positively - the disappearance of diarrhea and normal-

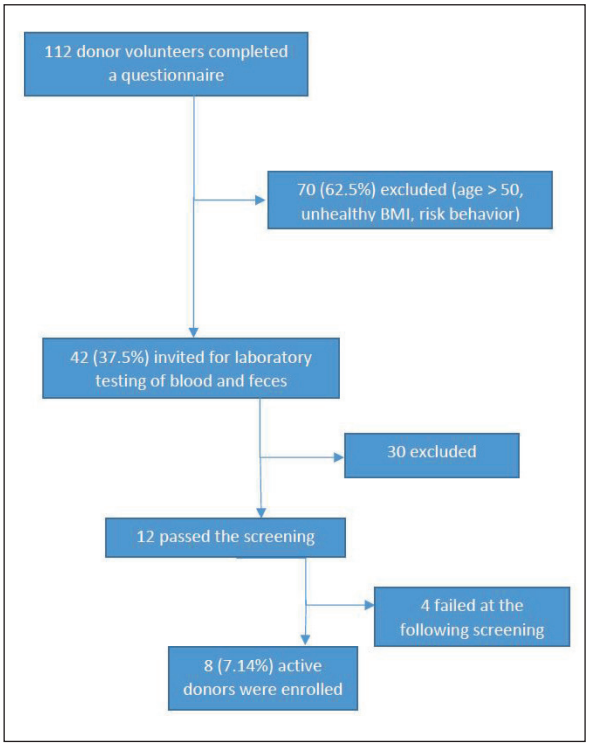

Figure 1. Stool donors enrollment.

ization of accompanying symptoms. *C. difficile* toxins normalized in 4 (40%) patients up to one month after FMT and in all of them up to 6 months after FMT.

Discussion

In the current study, we present the creation of the first stool bank for Bulgaria and Eastern Europe and the first series of effective FMTs in Bulgaria. FMT has entered mainstream medicine for the treatment of rCDI in the last few years, with FMT being widely available in many locations, either using locally processed samples or samples obtained from commercial stool banks. However, appropriate donor selection remains a challenge, especially with few studies describing donor selection and retention processes, the rationale for exclusion, and results from donor screening[18].

Several studies describe physicians' knowledge and attitude toward FMT showing increased acceptance of FMT for rCDI among physicians[19-21]. Furthermore, a recent study demonstrates[22] that stool donation is well accepted by healthy individuals; however, a significant proportion of healthy individuals (57%) who volunteer for stool donation failed prescreening with a history questionnaire. Previous studies[18,20] show that many donors who sign up for stool donation fail prescreening and stool testing due to asymptomatic colonization by potential pathogens, with very few ultimately serving as donors. Overall, data from a broad USA stool bank[23] suggest that nearly 90% of applicants are excluded after medical evaluation. Similar data have been performed by an Australian stool bank, where approximately 50% of donors are excluded after fulfillment of the clinical questionnaire[21]. We obtained similar results in our study, where 62.5% of the volunteers failed after the questionnaire. Even though we found many healthy volunteers, only a low percentage (7.14%) of them were suitable to become feces donors.

There are variations in the donor screening protocols adapted by different institutions with standardized recommendations on blood and stool tests for donors. Most institutions including ours screen for chronic hepatitis, HIV and syphilis, which are pathogens not known to be transmitted by stool. It is unclear if *Helicobacter pylori*, symptomatic Cytomegalovirus, and Epstein-Barr can be transmitted by FMT, but we suggest that they must be added to donor screening.

Since progressing age has been associated with modified gut microbiota composition, young individuals (aged <50 years) are preferred as possible donors[10]. Although some stool banks like ours exclude healthcare workers with exposure to patients, available data recommend a low predominance of antibiotic-resistant bacteria colonization in this population[24]. On the other hand, we prefer not to risk and exclude all medical professionals and medical students from the list of potential donors. Long-term cost-effectiveness analyses are necessarily needed for donor screening for FMT.

Thanks to the establishment of a stool bank in Bulgaria, ten successful and safe FMTs were performed in patients with rCDI. The presence of verified donors enabled safety, high efficiency, and centralization of the process and created the potential for this methodology to be performed routinely throughout the country by trained specialists.

FMT is regarded as a medical therapy that is performed by registered specialists[4,10]. FMT is not yet consistently regulated within Europe; however, it seems evident that the voluntarily donated fecal material should be collected, handled, and used according to the standards determined by the EU Commission in the EU Tissue and Cells Directive (2004/23/ec)[25,26]. Faeces should be considered a substance of human origin (SoHo) and may be considered equal to a tissue[27]. Any alteration of donor feces other than those required for conservation and/or administration of the fecal microbiota indicates that the changed products from this step should be considered a drug and classified according to this legislation (Directive 2004/23/ec §6 and Directive 2001/83/ec). Regrettably, some countries within the EU have currently listed donor feces suspensions as a drug. Currently, the FMT in Bulgaria is considered neither as medicine nor as a tissue or organ.

In addition to the classification of the fecal microbiota as tissue or drug, there are many unanswered questions about FMT. There needs to be awareness among healthcare professionals about FMT and other microbiome-based therapies. Moreover, it will be interesting to know how gut microbiota interacts with drugs, herbal supplements, dairy products, pollutants, and other xenobiotics[28]. More than physician awareness, there needs to be public awareness so that potential patients can make informed decisions regarding procedure methodology and donor selection.

Several limitations should be noted in this study. First of all, we performed FMT on a small

sample of patients. However, this method was done for the first time in Bulgaria; therefore, this is a good start. Secondly, the composition of fecal microbiota was not assessed in the donors and the patients due to a lack of resources.

Conclusions

FMT is a safe and effective method for the treatment of rCDI. An essential process for the implementation of a successful FMT is the process of donor selection and screening, which is why stool banks are significant for the centralization of the process. For the first time, a stool bank was established in Bulgaria, which helped ten successful and safe FMTs to be performed. This will allow the implementation of this methodology in the country by quality assurance and guaranteeing the availability of fecal donor suspensions for patients in need.

Conflict of Interest

The Authors declare that they have no conflict of interests.

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References

1) Freeman J, Bauer MP, Baines SD, Corver J, Fawley WN, Goorhuis B, Kuijper EJ, Wilcox MH. The changing epidemiology of Clostridium difficile infections. Clin Microbiol Rev 2010; 23: 529-549.
2) Wenisch C, Parschalk B, Hasenhundl M, Hirschl A, Graninger W. Comparison of vancomycin, teicoplanin, metronidazole, and fusidicacid for the treatment of Clostridium difficile-associated diarrhea.Clin Infect Dis 1996; 22: 813-818.
3) McFarland LV, Surawicz CM, Rubin M, Fekety R, Elmer GW, Greenberg RN. Recurrent Clostridium difficile disease: epidemiology and clinical characteristics. Infect Control Hosp Epidemiol 1999; 20: 43-50.
4) Cammarota G, Ianiro G, Tilg H, Rajilic-Stojanovic M, Kump P, Satokari R, Sokol H, Arkkila P, Pintus C, Hart A, Segal J, Aloi M, Masucci L, Molinaro A, Scaldaferri F, Gasbarrini G, Lopez-Sanroman A, Link A, de Groot P, de Vos WM, Högenauer C, Malfertheiner P, Mattila E, Milosavljević T, Nieuwdorp M, Sanguinetti M, Simren M, Gasbarrini A. European consensus conference on fecal microbiota transplantation in clinical practice. Gut 2017; 66: 569-580.
5) Cammarota G, Ianiro G, Bibbo S, Gasbarrini A. Fecal microbiota transplantation: a new old kid on the block for the management of gut microbiota-related disease. J Clin Gastroenterol 2014; 48: S80-S84.
6) Nakov R, Segal JP, Settanni CR, Bibbò S, Gasbarrini A, Cammarota G, Ianiro G. Microbiome: what intensivists should know. Minerva Anestesiol 2020; 86: 777-785.
7) Zatorski H, Nakov R. Faecal microbiota transplantation in Inflammatory Bowel Disease: current concepts and future challenges [published online ahead of print, 2020 Jun 2]. Curr Drug Targets.
8) Li Q, Wang C, Tang C, He Q, Zhao X, Li N, Li J. Successful treatment of severe sepsis and diarrhea after vagotomy utilizing fecal microbiota transplantation: a case report. Crit Care 2015; 19: 37.
9) Wurm P, Spindelboeck W, Krause R, Plank J, Fuchs G, Bashir M, Petritsch W, Halwachs B, Langner C, Högenauer C, Gorkiewicz G. Antibiotic-associated apoptotic enterocolitis in the absence of a defined pathogen: the role of intestinal microbiota depletion. Crit Care Med 2017; 45: e600-e606.
10) Cammarota G, Ianiro G, Kelly CR, Mullish BH, Allegretti JR, Kassam Z, Putignani L, Fischer M, Keller JJ, Costello SP, Sokol H, Kump P, Satokari R, Kahn SA, Kao D, Arkkila P, Kuijper EJ, Vehreschild MJG, Pintus C, Lopetuso L, Masucci L, Scaldaferri F, Terveer EM, Nieuwdorp M, López-Sanromán A, Kupcinskas J, Hart A, Tilg H, Gasbarrini A. International consensus conference on stool banking for fecal microbiota transplantation in clinical practice. Gut 2019; 68: 2111-2121.
11) Terveer EM, van Beurden YH, Goorhuis A, Seegers J, Bauer MP, van Nood E, Dijkgraaf MGW, Mulder CJJ, Vandenbroucke-Grauls CMJE, Verspaget HW, Keller JJ, Kuijper EJ. How to: establish and run a stool bank. Clin Microbiol Infect 2017; 23: 924-930.
12) Jorgensen SMD, Hvas CL, Dahlerup JF, Mikkelsen S, Ehlers L, Hammeken LH, Licht TR, Bahl MI, Erikstrup C. Banking feces: a new frontier for public blood banks? Transfusion 2019; 59: 2776–2782.
13) Guo Y, Li SH, Kuang YS, He JR, Lu JH, Luo BJ, Jiang FJ, Liu YZ, Papasian CJ, Xia HM, Deng HW, Qiu X. Effect of short-term room temperature storage on the microbial community in infant fecal samples. Sci Rep 2016; 6: 26648.
14) Shaw AG, Sim K, Powell E, Cornwell E, Cramer T, McClure ZE, Li MS, Kroll JS. Latitude in sample handling and storage for infant fecal microbiota studies: the elephant in the room? Microbiome 2016; 4: 40.
15) Lee CH, Steiner T, Petrof EO, Smieja M, Roscoe D, Nematallah A, Weese JS, Collins S, Moayyedi P, Crowther M, Ropeleski MJ, Jayaratne P, Hig-

gins D, Li Y, Rau NV, Kim PT. Frozen vs fresh fecal microbiota transplantation and clinical resolution of diarrhea in patients with recurrent clostridium difficile infection: a randomized clinical trial. JAMA 2016; 315: 142-149.

16) Goldenberg SD, Batra R, Beales I, Digby-Bell JL, Irving PM, Kellingray L, Narbad A, Franslem-Elumogo N. Comparison of different strategies for providing fecal microbiota transplantation to treat patients with recurrent clostridium difficile infection in two English hospitals: a review. Infect Dis Ther 2018; 7: 71-86.

17) Debast SB, Bauer MP, Kuijper EJ. European Society of Clinical Microbiology and Infectious Diseases. European Society of Clinical Microbiology and Infectious Diseases: update of the treatment guidance document for Clostridium difficile infection. Clin Microbiol Infect 2014; 20: 1-26.

18) Burns LJ, Dubois N, Smith MB, Mendolia GM, Burgess J, Edelstein C, Noh A, Alm E, Kassam Z. Donor recruitment and eligibility for fecal microbiota transplantation: results from an International Public Stool Bank. Gastroenterology 2015; 148: 96-97.

19) Moossavi S, Salimzadeh H, Katoonizadeh A, Mojarrad A, Merat D, Ansari R, Vahedi H, Merat S, Malekzadeh R. Physicians' knowledge and attitude towards fecal microbiota transplant in Iran. Middle East J Digestive Diseases 2015; 7: 155-160.

20) Zipursky JS, Sidorsky TI, Freedman CA, Sidorsky MN, Kirkland KB. Physician attitudes toward the use of fecal microbiota transplantation for the treatment of recurrent Clostridium difficile infection. Canadian J Gastroenterol Hepatol 2014; 28: 319-324.

21) Paramsothy S, Borody TJ, Lin E, Finlayson S, Walsh AJ, Samuel D, van den Bogaerde J, Leong RWL, Connor S, Ng W, Mitchell HM, Kaakoush N, Kamm MA. Donor recruitment for fecal microbiota transplantation. Inflamm Bowel Dis 2015; 21: 1600-1606.

22) Tariq R, Weatherly R, Kammer P, Pardi DS, Khanna S. Donor screening experience for fecal microbiota transplantation in patients with recurrent C. difficile Infection. J Clin Gastroenterol 2018; 52: 146-150.

23) Kassam Z, Dubois NE, Ling K, Qazi T, Smith M, Kelly CR, Fischer M, Allegretti JR, Budree S, Panchal P, Kelly CP, Osman M. 512 – donor health screening for fecal microbiota transplantation: prospective evaluation of 15,317 candidate donors. Gastroenterology 2019;156: 100-101.

24) Decker BK, Lau AF, Dekker JP, Spalding CD, Sinaii N, Conlan S, Henderson DK, Segre JA, Frank KM, Palmore TN. Healthcare personnel intestinal colonization with multidrug-resistant organisms. Clin Microbiol Infect 2018; 24: 82e1-82e4.

25) EU. Directive 2004/23/ec of the European Parliament and of the Council of 31 March 2004 on setting standards of quality and safety for the donation, procurement, testing, processing, preservation, storage and distribution of human tissues and cells: European Union; 2004

26) Keller JJ, Vehreschild MJGT, Hvas CL, Jorgensen SMD, Kupcinskas J, Link A, Mulder CJJ, Goldenberg SD, Arasaradnam R, Sokol H, Gasbarrini A, Hoegenauer C, Terveer EM, Kuijper EJ, Arkkila P. Stool for Fecal Microbiota Transplantation should be classified as a transplant product and should not become a pharma cash cow. United European Gastroenterol J 2019; 7: 1408-1410.

27) EU. Competent Authorities on Substances of Human Origin Expert Group (CASoHO E01718), Meeting of the Competent Authorities for Tissues and Cells, 3-4 December 2014. Summary Report: European Commission, Directorate-general for health and food safety; 2015

28) Nakov R, Velikova T. Chemical metabolism of xenobiotics by gut microbiota. Curr Drug Metab 2020; 21: 260-269.