PROTOCOL

[Analyse of Reporting and Conduct of studies assessing Fecal Microbiota Transplantation: Methodological systematic review]

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1. Introduction

Majority of body are colonized by widely of microbes, almost all them live in gut and constitute "gut microbiota"[1]. Intestinal microbiome plays an important role in the regulation of both host immunity and metabolism[1-4]. Alterations of gut microbiota composition may be implicated in several chronic diseases associated with its disruption[5-8]. Thus, the restoration of a healthy intestinal microbiome became a main clinical therapeutic. Consequently, the interest in the gut microbiota has considerably increased in recent years[9-11]. Most of clinicians and researches are enthusiastic about new therapeutic manipulations of microbiota. Several clinical approaches have been proposed to restore the microbiome, such as the probiotic that is the most widely used treatment[12,13].

However the microorganism used in probiotics are less diverse than those living in healthy persons[14]. Another intervention, fecal microbiota transplantation (FMT) has gained ground since few years in the face of remerging *Clostridium difficile* infections (ref). FMT is considered as promising option for the treatment of diseases involving an alteration of the gut microbiota, although the mechanisms by which the composition of microbiota imply in disease initiative or progression is not understood[15,16]. FMT is the complex intervention needing both the application of cutting-edge technologies such as the identification of gut microbiota composition using the meta-genomics and the development of well-designed, large trials.

However, as with any new therapeutic, the limitations, biases, and methods associated with research on FMT have raised increasing concern[16-22]. Several challenges should be relieved to improve the therapeutic potential of FMT in practice; for example the relationship between donor's microbiota composition and clinical results, standardization of stool preparation[20].

As with any results of research, it is essential that their reports are adequately and clearly reported to enable the reader to judge studies strengths and limitations. Moreover, transparent reporting facilitates decision making by all readers and reproducibility of methods by interested researchers[23]. It is crucial to identify confounding factors in FMT study in order to perform more uniform and effective research in the future.

To our knowledge, the reporting and conduct of studies assessing efficacy or safety of FMT for any condition did not evaluate. Therefore, we will perform a methodological systematic review of published and ongoing reports of studies assessing FMT to examine how they were reported and conducted.

2. Methods

2.1.Eligibility criteria

All studies assessing efficacy or safety of FMT will be eligible. We will exclude systematic reviews or meta-analyses, diagnostic studies, methodological publication, editorial style reviews, reports, abstract and poster, case report of only one case, studies not involving human participant. When we are duplicate publications, i.e. publication of the same study by the same authors without modification of methods or results, we will keep the most recent publication only.

2.2.Search strategy

We will search for eligible studies using the following databases Cochrane Central Register of Controlled Trials, PubMed, Web of Science and clinicaltrial.gov. Search equations will be developed for each database around specific free-text words pertaining to FMT. We also will screen the references of methodological papers, existing reviews. Duplicate records of the same publications were removed.

"Fecal microbiota transplantation" OR "faecal microbiota transplantation" OR **"Fecal microbiome transplantation"** OR

"fecal transplantation" OR "faecal transplantation" OR "feces transplantation" OR "faeces transplantation" OR "stool transplantation" OR "fecal flora transplantation" OR

" fecal transplant" OR "faecal transplant" OR "feces transplant" OR "faeces transplant" OR "stool transplant" OR "microflora transplant" OR "fecal flora transplant" OR "faecal flora transplant" OR "microbiota transplant" OR "microbial transplant" OR "microbiome transplant"

"fecal microbiota transplant" OR "faecal microbiota transplant" OR "Fecal microbiome transplant" OR

"fecal bacteriotherapy" OR "faecal bacteriotherapy" OR "feces bacteriotherapy" OR "faeces bacteriotherapy" OR "rectal bacteriotherapy" OR "fecal flora bacteriotherapy" OR

"donor fecal " OR "donor stool" OR "donor feces"

"fecal transfer" OR "faecal transfer" OR

"fecal reconstitution" OR "microbiome reconstitution" OR "feces reconstitution" OR "fecal flora reconstitution" OR

"duodenal infusion " AND (feces OR faeces OR fecal OR stool OR faecal)

2.3.Selection of relevant paper

One reviewer independently will examine. One reviewer will examine each title and abstract to exclude obviously irrelevant reports. After screening the title and abstract, one reviewer will select full-text articles according to the pre-specified eligibility criteria. A second reviewer will check all included and excluded studies. Disagreements will be discussed by the authors to reach consensus. We will list excluded studies and document the primary reason for exclusion.

2.4.Data extraction

A standardized data collection form will use to collect all data from original reports and supplementary appendices, when available. Two reviewers independently will extract all data of reports. Disagreements will be resolved by discussion.

2.4.1. General characteristics

We will record the following general characteristic:

For published study, the type of journal (i.e. general or specialized journal), the year of publication will be noted. For ongoing study, we will record the primary date of registration year. For all study, we will assess the location such as the countries (single or international study) and the centres involved (number of centres). We will evaluate the context clinic such as medical area and disease (disorder, syndrome, illness, or injury). Also we will examine study characteristic such as the study design (randomized controlled trial [RCT] or observational study); the type of comparator (i.e., placebo or active) and when the comparator was active, we will classify the comparator as pharmacological intervention, non-pharmacological intervention or both. We will record the number of arms and number of

participants. Finally, we will note trial number registration, funding source and a statement of conflict of interest.

2.4.2. Reporting of key methodological components of study

We will interest on reporting of key methodological components that allow assessing or judging the quality and conduct of study such as objectives, characteristics of patients, description of intervention, location, and outcome. Thus, we will assess whether key methodological components were reported or not according to Weil and colleagues[22], Nieuwdorp and colleagues[18], Smith and colleagues[19], Spector and colleagues, Vyas and colleagues[21].

- Objectives: We will evaluate whether the authors provided specific objectives, any rationale or justification for their study question. When the rational was reported, we will determine whether the authors stated animal studies, human studies or both.
- Description of patients: We will assess whether the authors reported a description of patients giving: the eligibility criteria, age of participants (child, adult and senior), disease status, disease status and severity of patients, mode of recruitment. Also, we will evaluate whether the authors reported the medications taken by patients
- Description of intervention: We will assess whether the authors reported description of FMT:
    - ***Donor***: First, we will evaluate whether the authors reported any information about eligible criteria of donor, a methods of selection of donor and the periods of recruitment. We will determine whether the authors reported any preparation of donors such as advice for modification of diet or pre-treatment with antibiotics or laxative. We will examine whether the authors gave a definition of healthy donor and we will record the definition. We will assess whether the authors gave donor characteristics (for example, age, and sex) and described a

methods for infectious diseases screening (type of test [blood and stool], number and period of analysis), metabolic diseases, family history of autoimmune and malignancies. We will examine whether they reported number of donor, number of donation by donor, the relationship between the donors and participants. We will assess whether the author reported a follow up of donor. Also, we will evaluate whether the authors reported the medications taken by donor

- ***Procedures for stool preparation***: We will assess whether the authors described how and when the stool was collected. We will determine whether they gave a method for preparation of stool infusion (methods for dilution and homogenization). We will note type of stool (fresh or frozen). We will evaluate whether the volume, frequency and number of infusion were reported. We will define if the volume of infusion was large when patient were given > 500 ml (ref). Also, we will examine whether specific material for stool preparation were reported. , for example determination of stool microbiota composition by DNA microarray. Finally we will evaluate whether the authors proposed or described a methods to examine the stability of anaerobic conditions.
- ***Procedures for patient preparation***: We will determine whether the authors reported any methods for preparation of patients for example any pre-treatment such as antibiotics.
- ***Duration of follow up***: We will evaluate whether the duration of follow-up. We will classify the duration of follow up as short (0-3 months) or medium (3-6 months) or long (>6months).

- ***Method of transplantation***: We will assess whether they gave the route of administration. Also we will evaluate whether study reported the skill of care provider.

- Description of co-intervention: If any co-intervention was given in control or experimental group or both, we will assess whether the author reported a description.
- Description of setting, locations: We will evaluate whether the authors reported a description of setting location. We will record the type of structure.
- Description of outcomes: We will assess whether the authors gave a clear definition of outcome and we will record the definition. We will determine whether the outcomes were pre-specified and whether the main outcomes were clearly mentioned. We will note whether the report mentioned type of measure and time point for each outcome. We will assess the number of outcome and type of outcome: Binary, continuous outcomes and time to event. Lastly, we will classify the main outcomes in Patient-Important Outcomes or surrogate outcomes according to previous works on this topic[24-26]. We will define patient important outcome as measure that are directly impact on quality of life such as major morbid events (stroke, myocardial infarction, amputation) and minor morbid events (pain and functional status); Surrogate Outcomes will be define as measure that may indicate disease progression and increased risk for patient-important outcomes (e.g., glycated hemoglobin, cholesterol level); or assessed response to physiological or laboratory manoeuvres without direct tangible effects on patients (e.g., insulin, C-peptide levels) (ref). For example, high blood pressure does not reflect how a patient feels, functions, or survives but is known to be associated with increased risk of stroke. Other surrogate outcomes were for example treatment adherence, patient knowledge, satisfaction and acceptability of experimental interventions.

- Reporting of adverse events: we will assess whether the authors reported definition and a strategy to record the adverse event.

2.4.3. Composite outcome assessing inadequate reporting or inadequate conduct quality

Firstly, we will plan to contact specialists of the FMT to identify the most relevant of methodological components of study that allow evaluate quality study and ability to be applied in clinical practice. Secondly, we will build composite outcome assessing inadequate reporting or inadequate conduct quality (when the reporting was adequate). The composite outcome will be based on expect recommendation and methodological paper.

2.5.Statistical analysis

The analyse will be descriptive. We will summarize the quantitative data by medians and interquartile ranges, and categorical data by numbers and percentages. Statistical analyses will involve use of SAS version 9.3 (SAS Institute).

3. Reference

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