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Review Article

Microbiome-based stratification to guide dietary interventions to improve human health

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ARTICLE INFO

Article history:
Received 29 February 2020
Accepted 10 July 2020

Keywords:
Diet
Health
Gut
Microbiome
Stratification
Personalized nutrition

ABSTRACT

Diverse evidence has suggested that the gut microbiome is closely associated with overall human health. Modulation of the gut microbiome through nutritional intervention is recognized as a robust and attainable strategy to prevent disorders/diseases and improve human health. However, universal dietary recommendations demonstrated to have different, sometimes even opposite, effects due to the considerable inter-individual variability between subjects, especially in the gut microbiome. Hence, implementation of personalized nutrition or other treatment strategies have been suggested to tackle the individuality problem. A first step into this direction includes the stratification of subjects into specific groups based on their gut microbiome. The gut microbiome could serve as a pool of potential biomarkers for distinguishing "responders" and "non-responders" to specific treatments, which subsequently can be used to classify subjects with ambition to increase treatment efficacy. In this review, we explain the need for human gut microbiome stratification, introduce the concepts and show with specific examples potential options of microbiome-based stratifications. Finally, we propose a strategy for how microbiome-based stratification can be introduced to obtain improvements in dietary efficacy that can be implemented in real-life settings.

Article Outline

Abbreviations: BMI, body mass index; CD, Crohn's disease; FMT, fecal microbiota transplantation; GI, gastrointestinal; IBD, inflammatory bowel disease; IBS, irritable bowel syndrome; SCFAs, short-chain fatty acids; UC, ulcerative colitis.

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https://doi.org/10.1016/j.nutres.2020.07.004

1. Introduction

The human gastrointestinal (GI) tract harbors trillions of microbes, commonly referred to as the "gut microbiota" that is dominated by bacteria, but also contains archaea, eukaryotes, and viruses. The term microbiome has been used to refer to the collection of microbes and all the genes and functionalities they encode, and it has been found that the collective microbial metagenome outnumbers our own eukaryote genome by more than 100 times [1]. Several factors including age, dietary habits, genetics, mode of birth delivery, and medication use have been found to affect the composition and functionality of the gut microbiota. Among them, diet has been identified as one of the main drivers in the modulation of the gut microbiome [2]. Moreover, numerous studies have demonstrated that the human gut microbiome plays a vital role in host health and in the onset and progression of a variety of gut-associated as well as more systemic disorders or diseases such as inflammatory bowel disease (IBD), irritable bowel syndrome (IBS), obesity, and diabetes [3,4]. Indeed, there is a plethora of studies describing differences in microbiota composition between healthy and compromised subjects. However, these comparisons, which are mostly cross-sectional, do not provide consensus observations, such as a microbial taxon that is specific for a certain disease or disorder. Even at phylum level, observations from different studies are sometimes contradictory, as has been observed for the Bacteroidetes to Firmicutes ratio in relation to BMI [5,6]. Reasons for these inconsistencies could include technical aspects, such as differing laboratory protocols, recruitment strategies, and cross-sectional study designs, but also biological aspects, such as intrinsic differences in microbiota composition, functional redundancy, and microbial flexibility towards different environmental conditions within an ecosystem (e.g. availability of substrates, pH variation) [7]. Furthermore, changes in activity are not always reflected in corresponding changes in microbiota composition, and, for example, a recent study has indeed shown that switching traditional African and Western diets for 2 weeks had a limited impact on microbiota composition, but large impact on metabolite production, notably short chain fatty acids (SCFAs) [8]. Even with more drastic measures, such as fecal microbiota transplantation (FMT), it has been demonstrated that only a subset of subjects shows positive responsiveness, even in the control group [9]. Although the variation in outcomes between different studies might at least in part be explained by variations in study set-up, recruitment criteria, and laboratory protocols, as indicated previously, it is evident that a significant part of the inconsistency is due to the considerable inter-individual variability in microbiota composition. Therefore, just continuing to randomly select individuals for intervention studies only based on clinical and/or demographic parameters will likely not further improve our understanding of the mechanisms underlying response efficacy of an intervention. Instead, we should consider stratifying participants beforehand based on microbiome characteristics (eg, composition, metabolic capacity, metabolite production) in order to better predict efficacy of an intervention that ultimately will lead, for example, to personalized dietary recommendations. Assuming that the occurrence of a certain disease/disorder is found to be related with a certain microbiota composition profile, patients can be stratified according to this profile prior to the envisaged intervention. Ultimately, personalized stratification-based diagnostics and therapeutics would be employed to improve efficacy.

Therefore, the aim of this review is to explore how the current knowledge on the gut microbiome and microbial signatures associated with human health parameters can be used to stratify subjects for intervention studies, with a focus on the bacterial part of the microbiota. In addition, we will highlight some examples of potential gut microbiome targets that can be used for stratification of subjects in order to predict the effect of certain dietary components in an intervention and thereby improving the success rate of the intervention.

2. Overview of gut microbiome-based stratification strategies

To ensure that primary end-points of clinical studies are reached, target group selection is important. Detailed selection of prospective subjects before starting an intervention is very common and starts with specifying inclusion and exclusion criteria. These criteria may include stratification based on age, gender, as well as on specific measurable health biomarkers, such as insulin resistance or inflammation scores. In addition, selection of subjects can also be based on questionnaire-derived criteria, such as the Rome criteria to classify IBS into different subtypes. Sometimes the selection of subjects can be straightforward to obtain high efficacy of a given intervention when it concerns, for example, a disease with known underlying mechanism and mode of action of the respective medication that is evaluated. However, this is not the case when diseases are multifactorial, have an unknown underlying mechanism, and/or include a role of the microbiome. In those cases the efficacy of interventions is

difficult to predict, and as a result, the selection of subjects with respect to the target of the intervention is often random. Typical examples are interventions targeting the microbiome, such as dietary interventions based on fiber or other non-digestible dietary components where subject selections are random from a microbiome point of view (Fig. 1). Although the efficacy of such dietary interventions may sometimes be disappointing, these studies are very relevant as they may provide associations between subject-specific response to the diet and microbiome features that can be used to define or refine microbiome-based stratification of subjects in a subsequent intervention and thereby increasing efficacy with the ultimate goal of reaching personalized dietary recommendations.

The first step to move towards such a microbiome-based personalized recommendation is stratification of subjects in subgroups based on specific microbiome features (Fig. 1). This microbiome-based stratification can, in principle, be based on any microbial feature of which 16S rRNA gene data, metagenome data, and metabolite data are currently most practical (Table 1). The potential merits and drawbacks of these stratifications will be discussed in this section.

2.1. 16S rRNA gene-based stratification strategies

Direct sequencing of the bacterial 16S rRNA gene has become the most widely adopted method to obtain information with respect to microbiota composition of any given ecosystem, including that of the human gut. This type of microbiota profiling has provided a phylogenetic framework of the gut microbiota. Typical ecosystem features that can be obtained from such analyses include microbial community typing, determining microbial diversity, as well as identifying microbes that are differently abundant between groups of subjects, all of which can be used as targets for microbiome-based classification (Table 1). Microbial community typing based on 16S rRNA genes includes identifying the presence of community types and the existence of alternative stable compositional states [10]. Although originally based on metagenome data, the study by Arumugam et al [11] laid the foundation for microbial community typing of the gut microbiome. This study proposed that individuals could be divided into three groups based on their gut microbiota composition. The three groups were defined based on robust clusters, which are each dominated by different genera, namely *Bacteroides*, *Prevotella*, and *Ruminococcus*, and were

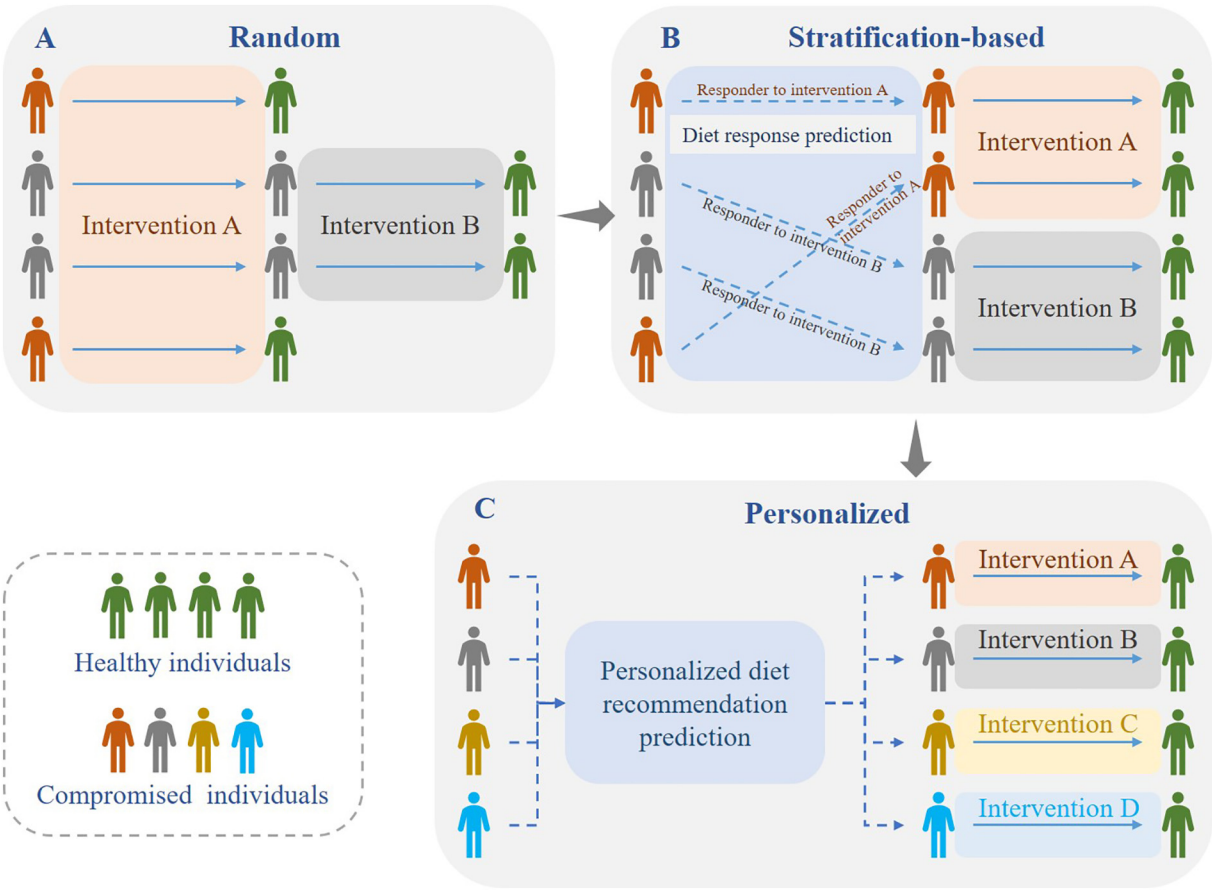

Fig. 1 – Overview of different intervention strategies with microbiome-based stratification as an intermediate step between random and personalized dietary intervention. A, Random selection without prior knowledge on microbiome. B, Stratification of subjects in groups based on microbiome features with predicted efficacy. C, Personalized approach based on individual microbiome-disease characteristics.

Table 1 – Overview of the human microbiome-based stratification possibilities based on different microbial characteristics

	Characteristic	Drawback	Stratification possibility	Clarifying example	Reference
16S rRNA gene profiles	Identification of microbial taxa	No insights in functional capacity or activity	Community typing	*Bacteroides* vs *Prevotella*-dominated profiles, enterotypes	[10,14,15,18,21]
			Diversity differentiation	Diversity differences in elderly	
			Classification on differential taxa	Abundance differences in *Akkermansia muciniphila*	[23]
Metagenome profiles	Identification of microbial taxa and functional capacity	No insight in microbial activity	Community typing	Enterotypes	[11,25,33]
			Diversity differentiation	High and low richness in obese subjects	
			Classification on differential functional capacity	Acetate production capacity differences	[26]
Metabolite profiles	Overall metabolic activity	No phylogenetic information	Untargeted metabonome profiling	Distinct metabonomes between Africans and Americans	[8,29-31]
			Classification based on differential metabolite concentrations	Increased butyrate concentrations in fecal material	[32]

found to be independent of gender, nationality, ethnicity, health status, age, and BMI. However, with more studies focusing on the stratification of the human gut microbiome based on enterotypes, it has been recognized that the number and the category of alternative states vary between studies, with the level of *Prevotella* and *Bacteroides* being the most common drivers for identification of alternative compositional states [12-17]. Although the number of alternative states and the way to identify them is still a point of ongoing discussion, the concept of identifying different alternative states might be used as a target for stratifying people based on their microbiota at the start of an intervention, as has been suggested earlier [18].

Besides the stratification of individuals based on different alternative steady states of microbiota composition, differentiating microbiota based on 16S rRNA gene diversity opens another avenue for microbiome-based stratification. Diversity of a microbiota includes the number of different taxa (richness) as well as their (relative) abundance distribution (evenness) within an ecosystem. A high microbial diversity is considered to be beneficial as it is suggested to contribute to resilience after disturbance of the microbiome [19]. Indeed, the microbiota diversity is generally higher in healthy subjects than compromised subjects [20]. It is interesting to note that the average diversity of the microbiota declines during aging, with high subject-to-subject variation [21]. Since this reduced diversity is hypothesized to be associated with a decline in health status, stratification of the elderly based on diversity could be an interesting approach for intervention studies.

Apart from the above mentioned approaches, the presence or absence of specific microbes or their differential abundance between groups of subjects could also be regarded as a characteristic for stratification. For example, individuals who had a higher abundance of combinations of taxa, i.e. the genera *Lactobacillus*, *Leuconostoc*, and *Pediococcus*, at baseline lost less weight and rapidly regained weight [22]. Other examples include higher baseline levels of *Akkermansia muciniphila* that were associated with a greater improvement in insulin sensitivity markers after a low-calorie diet [23], and low initial numbers of *Bifidobacterium* that were associated with the biggest increase in the abundance of this genus after a bifidogenic (partially hydrolyzed guar gum and fructo-oligosaccharides in the biscuit) intervention [24].

2.2. *Metagenome-based stratification strategies*

Besides stratification based on 16S rRNA genes, the collection of microbial genomes (commonly referred to as the metagenome) can also be used as targets for microbiome-based stratification (Table 1). Metagenomics enhances resolution of identifying and characterizing microbial strains as compared to 16S rRNA gene sequencing and provides potentially important information about the capabilities of the organisms in the community. Besides 16S rRNA gene- data, metagenomic data can also be used for stratification. As indicated before, the discovery of enterotypes is based on metagenome data [11]. Similarly, microbial diversity can also be determined based on metagenomic datasets. A hallmark example is that obese subjects with a low diversity (richness) microbiome were found to be more prone to weight gain and developing insulin resistance as compared to obese subjects with a high diversity microbiome [25].

The most promising feature of metagenome-based microbiome stratification is the fact that differences in functional capacities between groups of subjects can be identified. For example, a comprehensive meta-analysis by Armor and colleagues [26] revealed a variety of functional signatures in the human gut microbiome associated to Crohn's disease and obesity, such as increased abundance of modules for lipopolysaccharide biosynthesis, iron transport, and acetate production. With studies establishing correlations between features and responsiveness, stratification of subjects based on these microbiome features in an intervention could be a promising approach to selecting for increased response efficacy.

2.3. *Metabolite-based stratification strategies*

Although 16S rRNA gene and metagenome data provide insight into which microbes are present and into their functional capacity, these approaches do not reflect actual microbial activity. Excreted metabolites are the end result of metabolic activity and those produced by the microbiota can directly impact numerous processes in the body. Also, metabolic diversity influences nutrient requirements and responses to diet between individuals [27]. Hence, stratification based on metabolic profiles provides another way of stratifying populations based on microbiome features. Metabonomics, i.e. profiling of all produced metabolites (metabonome), has demonstrated to be powerful in discriminating subjects with differences in their health status as well as human populations from different geographic locations with their traditional long term dietary habits [28].

Untargeted metabonomics has demonstrated to be successful for identification of disease biomarkers in human blood, urine, and fecal samples [29-31], which subsequently aids in disease diagnosis and proposed guidelines for potential therapeutics. In addition, Africans and African Americans demonstrated completely different metabonome profiles in urine and feces, which are in line with their microbiota compositional differences [8]. Such distinct features between subjects can be used for microbiome-based stratification.

In addition to metabonomics, monitoring of specific metabolite levels in a targeted approach is another way to distinguish subjects with potential responsiveness to a given intervention. For example, fecal propionate and butyrate levels, which are considered beneficial and suggested as biomarkers for IBS diagnosis [32], could be used as targets for stratification. The drawback of this approach is that it provides limited to no insight in which phylogenetic groups are involved in the microbial activities displayed, and for some metabolites, it is difficult to separate microbial from host activities.

3. Stratification-focused interventions to promote human health

Gut microbiome homeostasis is extremely important for maintaining overall human health, and its dysfunction or changes to its composition/activity have been associated with not only intestinal diseases (IBS, IBD, and colorectal cancer), but also extra-intestinal disorders (e.g. obesity, type 2 diabetes mellitus, and metabolic syndrome) [34,35]. Hence, understanding underlying mechanisms that govern these associations as well as predicting the efficacy of diets or other treatments in order to cure the respective disease or improve its symptoms remains a major challenge. An additional challenge lies in the fact that individuals often respond differently to similar or identical diets [33] or other treatments, and thus, individuals can be differentiated into responders and non-responders. In this section, we summarize the associations between specific gut microbial features and a selected number of well-studied disorders including IBD, IBS, and metabolic syndrome; how these associations are linked to response variation; and how these findings could lead to microbiome-based stratification of subjects with improved prediction of the efficacy of interventions.

3.1. *Inflammatory bowel disease (IBD)*

IBD is a group of chronic inflammatory conditions of the gut of which ulcerative colitis (UC) and Crohn's disease (CD) are the most common. Both conditions share a lot of similarities with regard to disease symptoms, but location of inflammation is one of the key differences between the two. In CD patients, inflammation could be found in any part of the GI tract, while in UC patients it is exclusively restricted to the inner lining of the colonic and rectal mucosa [36]. Although the onset and maintenance of IBD is largely unknown, mounting evidence supports the notion that the microbiome plays a crucial role. There are a variety of studies that have shown marked differences in microbiota composition between IBD and healthy controls, which generally show reduced diversity and butyrate production capacity in IBD [36-38]. Similarly, the fecal metabonome of IBD patients has been found to be very distinct from that of healthy subjects and even enables discrimination between CD and UC [31], showing a clear association between disease and microbiota activity. FMT interventions in subjects suffering from IBD offer a promising approach to treat IBD, but studies performed so far have shown varying success. Although this is often disappointing from a medical point of view, such studies are needed to identify retrospectively whether there are specific microbial features that are associated to relapse and remission. Taking UC as an example, one study demonstrated that patients that have relatively high relative abundance of *Clostridium* clusters IV and XIVa and butyrate production capacity in their microbiota, are more likely to enter sustained remission after FMT [39]. Moreover, Zhu et al [40] recently found that in mice, colitis could be ameliorated by tungstate treatment, which prevented the expansion of *Enterobacteriaceae* via inhibiting molybdenum-cofactor-dependent microbial respiratory pathways, whilst influencing the microbiota composition minimally. These examples of intervention studies revealed different microbial characteristics that could be used to select subjects based on specific microbial features prior to a next intervention study.

3.2. *Irritable bowel syndrome (IBS)*

IBS is characterized by increased intestinal sensation to triggers reflected by chronic or recurrent intestinal symptoms in the intestine in the absence of other pathological disorders [41]. Although there is no cure for IBS, in general the low FODMAP (fermentable oligosaccharides, disaccharides, monosaccharides, and polyols) diet had been proven to ameliorate gut symptoms in adult IBS patients [42]. However, it has to be noted that despite the evidence supporting the high efficacy of a low FODMAP diet, nearly a quarter of adult IBS patients does not show a positive response to the diet [43]. Among all the taxonomic changes in those non-responders, a lower relative or absolute abundance of *Bifidobacterium* was found to be consistent in most studies [44,45]. Similarly, the

baseline gut microbiota composition in children with IBS was found to be related to the efficacy of low FODMAP diet intervention as well. For example, in an IBS study in children, Chumpitazi et al [46] demonstrated that the fecal microbiota of responders was enriched at baseline in i) taxa with known greater saccharolytic metabolic capacity (eg, *Bacteroides*, *Ruminococcaceae*, *Faecalibacterium prausnitzii*) and ii) two Kyoto Encyclopedia of Genes and Genomes (KEGG) orthologues that are related to carbohydrate metabolism. However, other studies have found that baseline microbiota composition data currently cannot accurately predict response to a low FODMAP diet [47]. In contrast, Rossi and co-authors [48] were able to predict responses of patients with IBS to a low FODMAP diet with a mean accuracy of 97% based on 15 features in fecal volatile organic compounds profiles before the intervention, suggesting that the metabolic function of bacteria may be a suitable biomarker in determining response. This is a promising observation, as the efficacy of the low-FODMAP dietary intervention can be improved by metabolite-based stratification of subjects. Therefore, the efficacy of low FODMAP diet in IBS patients could be improved if we take the microbial features into account for stratification of subjects.

3.3. *Metabolic syndrome*

Metabolic syndrome is a collection of conditions associated with metabolic disorder and increased risk of developing cardiovascular diseases. It has not only been shown to associate with an aberrant gut microbiota, but is also highly influenced by long term dietary patterns [49]. In a recent barley beta glucans intervention study, it was observed that three participants with higher relative abundance of *Bifidobacterium* spp. and *Akkermansia muciniphila* before the intervention experienced total cholesterol level reduction, while the others who had lower amounts or even none of these microbial groups did not [50]. Similarly, higher relative abundance of *A. muciniphila* at baseline was associated with a healthier metabolic status in overweight/obese humans, characterized by the improvement in glucose homoeostasis, blood lipids, and body composition [23]. Recently the first intervention study with *A. muciniphila* as a probiotic in obese and overweight subjects has been published and demonstrated improvement of some metabolic parameters, supporting a causal role of this microorganism [51].

The relative contribution of diet and gut microbiome to fat accumulation has been characterized by a recent study, which indicated that certain nutrients alone were hardly able to affect fat accumulation, whereas specific gut bacteria contributed to host adiposity, irrespective of dietary intake [52]. This indicated that modulation of gut microbiota composition might be a target for losing visceral fat mass. Moreover, a 6-month intervention study described that subjects with high *Prevotella/Bacteroides* ratio appeared to more easily lose body fat on a high fiber and whole grain diet than subjects with a low *Prevotella/Bacteroides* ratio [53]. Other dietary components involved in weight loss have also been reported with microbiota associations. Capsaicin, a compound obtained from chili peppers, has a potential in controlling obesity, and one study has shown that the Firmicutes/Bacteroidetes ratio as well as the relative abundance of the genus *Faecalibacterium* was increased by capsaicin intervention, accompanied with higher plasma level of glucagon-like peptide 1 [14]. Meanwhile, these beneficial effects were mainly found among subjects that were clustered into the *Bacteroides* enterotype rather than the *Prevotella* enterotype. Similarly, in the case of diabetes, a recent acarbose intervention study showed that patients with higher baseline relative abundance of *Bacteroides* in their microbiota responded better, characterized by plasma secondary bile acids, reduced BMI, and improved insulin resistance, compared with those with a higher relative abundance of *Prevotella* [54]. This highlights the potential of microbiota-based pre-treatment selection for better predicting antidiabetic metabolic benefits. Collectively, all the above-mentioned examples provided microbial hints that could be used for stratifying subjects based on microbial features in order to optimally select the right target group for a given intervention.

4. Potential strategies to increase the success rate of intervention

An ideal scenario for personalized nutrition would be that a personal microbiome-based analysis for each individual would be done based on which health-care practitioners (eg, dieticians, clinicians) could provide the individual with personalized dietary advices and/or medication. A hallmark study by Zeevi et al [55] described a first concept towards such a personalized nutritional strategy. A high variability in the response to identical dietary components was observed between subjects, which could be accurately predicted by making use of a device that integrated blood parameters, dietary habits, physical activity, body measurements, and the gut microbiota. This study showed that personalized dietary recommendations may modify elevated postprandial blood glucose levels, possibly leading to diminished disease symptoms in type 2 diabetes mellitus. Although the same study demonstrated a significant increase in bacterial species that are generally considered beneficial when following a healthy diet (with low postprandial glycemic responses level) compared to a control group, the intervention only lasted for 1 week. It would be very insightful to continue this study for months or even years.

Although very promising, such a personalized dietary recommendation approach is not immediately applicable yet for disorders with unknown biomarkers or biomarkers that cannot be monitored continuously. Hence, a good intermediate step between this complete personal approach and universal recommendations, are stratification patterns as mentioned earlier. To reach this, we propose a five-step approach for a microbiome-based stratification, as shown in Fig. 2, to ultimately bridge the gap between general recommendations and more personalized nutrition.

The first step is to determine potential associations between microbial characteristics within the microbiome and certain diseases or conditions that could be used as targets for a dietary intervention and allow easy stratification of subjects. As mentioned in section 3, there are a variety of examples that could be used as a start for microbiome-based stratification options. Evidently, this step requires a focus on a specific target group of subjects with a potentially matching intervention, such as the low-FODMAP diet for IBS patients as

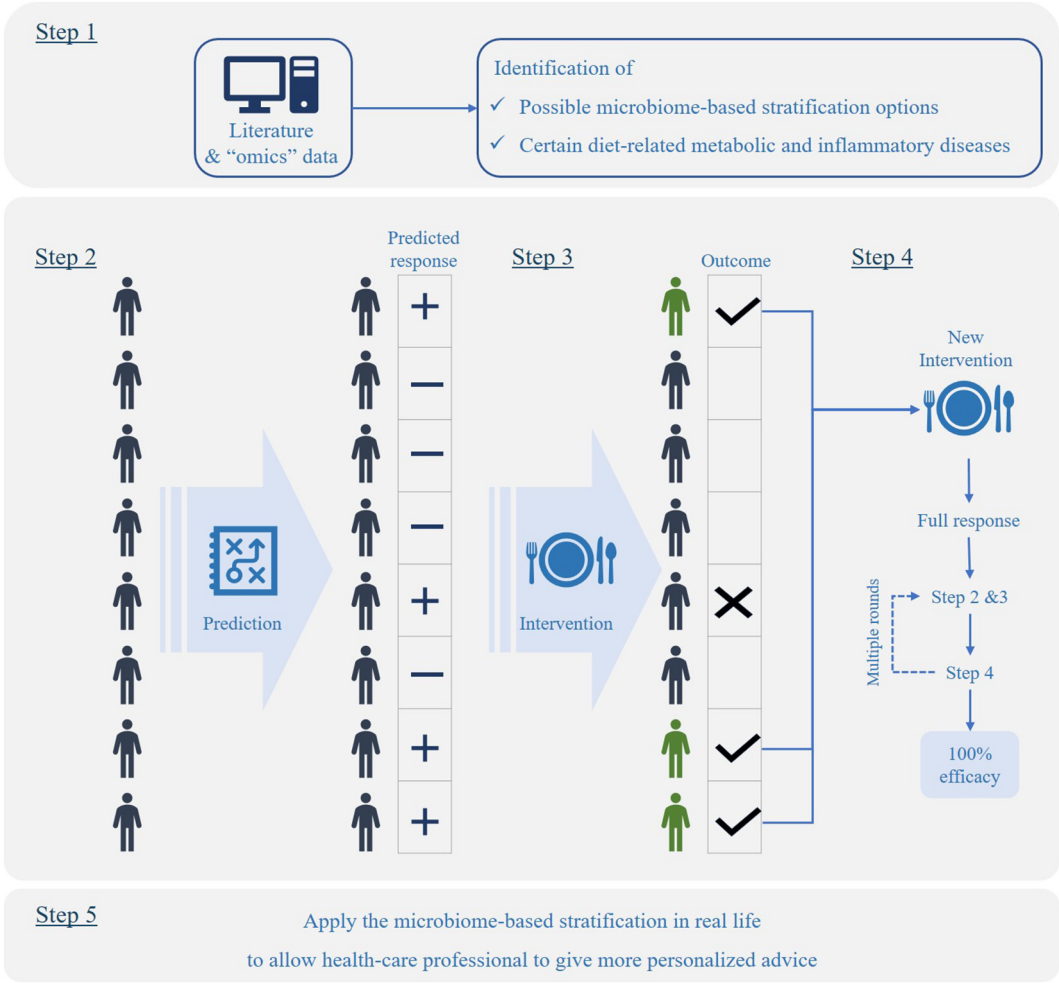

Fig. 2 – Schematic representation of a five-step approach to apply the microbiome-based stratification in personalized nutrition. Step 1: identify associations between microbiome and diseases that could be used as treatment targets. Step 2: **screen subjects and predict response to the planned intervention.** Step 3: **perform dietary intervention, check the outcome, and alter the prediction algorithm accordingly.** Step 4: **perform a new dietary intervention with the responders in step 3, and ideally achieve 100% success rate after several rounds from step 2 to step 4.** Step 5: **incorporate the microbiome-based stratification set-up in real-life settings.**

mentioned previously. Ideally, targets for screening and subsequent stratification should be present in feces, blood, or urine as these are relatively easy and cheap to obtain, and their sampling is already integrated in the current health-care system. However, for this to be feasible, it might be that specific attention should to be paid to storage methods, as metabolites are often not very stable. In addition, targets should preferably be amenable to screening using relatively easy-to-apply assays, such as PCR-based approaches to identify and/or quantify a specific microbial taxon or functional gene, or metabolites that can easily been detected by HPLC (high-performance liquid chromatography) or GC (gas chromatography).

The second step is to screen subjects based on the selected target for the dietary intervention and predict who will respond positively to the intervention. During this step, in principle the same rules apply compared to stratification of subjects based on other inclusion or exclusion criteria, including optimal intervention set-up and power calculations to determine the number of subjects. The third step will be the implementation of dietary intervention based on the prediction in the second step, collect the data, and evaluate the accuracy of the prediction. Based on the outcome of the prediction, fine-tuning or altering the prediction algorithm might be needed, which will subsequently lead to the fourth step, that performs a new intervention study with the responders that were predicted to respond based on microbiome-based stratification pattern. This will not only lead to determine the improvement of the prediction, but also indicate whether the response in the same individual is reproducible or coincidental. Ideally, a 100% success rate of the new intervention will be achieved. However, it is likely that multiple rounds of validation of the accuracy of specific microbiome-based stratification pattern will be needed.

The fifth and final step will be incorporating the microbiome-based stratification set-up in real-life settings. This step will certainly need the involvement of health professionals to implement it in such a way that it allows

more personalized advice to individual subjects as well as to make its implementation accessible for larger populations. For the latter part, educating people will be an important aspect. We also foresee this real-life implementation will drive innovation in the private sector. This could include the development of devices to measure the target molecules which are used for the stratification in a home setting. Typical examples of such devices include e-noses to detect specific odorous metabolites or chips that measure blood glucose levels continuously in diabetic individuals. Another private sector that may benefit from this implementation concerns those involved in development of specific food supplements or probiotics. Not only may novel products be produced, but microbiome-based stratification could also lead to better definition of target populations that will benefit from these products.

It is evident that this proposed five-step approach will result in non-responders for which the respective intervention will not offer a solution. However, the benefit of predicting accurately which subjects will not respond to the respective intervention is that these non-responders can focus on finding alternative interventions using the same five-step approach.

5. Future research

Although there are increasing numbers of studies investigating the gut microbiome, mechanistic insights with respect to health and how the microbiome can be affected by a dietary intervention remains largely unknown. As we indicated in section 4, identifying a robust and specific response between a given intervention and disease is the next key step to apply existing microbiome knowledge into real life nutritional recommendations. To achieve this, mining of massive omics and metabolite data sets complemented with mechanistic studies, as well as standardizing methodologies between laboratories will lay a solid foundation for the further clinical studies. This should be followed by setting inclusion and exclusion criteria of target diseases/disorders and stratification strategies based on the predicted efficacy of the intervention on the microbiome. In addition, more information on the effectiveness of prediction algorithms will be needed. Since inconsistencies are often encountered in gut microbiome studies, large-scale clinical studies should also be employed to confirm the reproducibility of predictions and findings.

6. Conclusion

The gut microbiome is vastly influenced by diet and ultimately affects human health. However, it is evident that gut microbiomes vary greatly among individuals, and as a result, traditional stratifiers for grouping subjects, such as age groups, gender, disease, and respective subtypes, might not be sufficient to obtain high efficacy of dietary interventions that target the gut microbiome. Hence, this inter-individual variability of gut microbiomes should be taken into account. Therefore, individual microbiome-based stratification, as an intermediate step towards personalized recommendations, may be a promising strategy to improve the success rate of certain dietary treatments. This is still a challenging approach as studies often show inconsistent, even contrasting associations between health parameters and specific characteristics of the microbiome. Nevertheless, the first steps moving forward into this field have been taken and are promising [18]. We are convinced that implementing characteristics of the microbiome, such as differences in composition, functional capacity, and/or its activity, as stratifiers for targeted dietary interventions will not only lead to improved understanding of the microbiome in health and disease, but also lead to innovations that will ultimately lead to personalized dietary recommendations in a real-life setting.

Acknowledgment

This work is funded by Winclove Probiotics B.V. (Amsterdam, The Netherlands). Zhuang Liu is also financially supported by the China Scholarship Council (File No. 201806850091).

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