



GUIDELINES ON MICROBIOME TESTING

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1. Guideline Purpose and Brief

This guideline outlines best practices for gut microbiome testing in clinical settings. It incorporates internationally recognized standards from authoritative bodies such as the American Gastroenterological Association (AGA), Human Microbiome Project (HMP), and the European Society of Clinical Microbiology and Infectious Diseases (ESCMID).

The guidelines are designed to offer recommendations and guidance for healthcare professionals in conducting microbiome testing, including best practices for testing. These guidelines should not be interpreted as comprehensive or exclusive of other valid approaches or methods. They do not guarantee specific outcomes, treatment decisions for individual patients must be made based on the independent judgment of healthcare providers, considering each patient's unique circumstances.

2. Definitions and Abbreviations

No.	Term / Abbreviation	Definition
2.1	AGA (American Gastroenterological Association)	An international authority cited in the guideline for best practices in gastroenterology and microbiome.
2.2	Amplicon Sequencing	A technique targeting specific genes (e.g., 16S rRNA) to identify and profile microbial communities at the genus level.
2.3	Alpha Diversity	A measure of the complexity (richness and evenness) of microbial species in a single sample.
2.4	Beta Diversity	A measure of the similarity or difference in microbial composition between two samples.
2.5	Bristol Stool Chart	A diagnostic tool used to classify stool consistency.
2.6	DNA Extraction	The process of isolating DNA from samples before sequencing.
2.7	Dysbiosis	A change in the diversity of the microbiome, involving loss of beneficial microbes (symbionts) and expansion of harmful ones (pathobionts).
2.8	ESCMID (European Society of Clinical Microbiology and Infectious Diseases)	An international standard-setting organization for clinical microbiology.
2.9	HMP (Human Microbiome Project)	A major research initiative providing reference standards and data for human microbiome studies.
2.10	IBS (Irritable Bowel Syndrome)	A common functional gastrointestinal disorder characterized by abdominal pain, bloating, and altered bowel habits (diarrhea, constipation, or both)
2.11	Metagenomics	The study of genetic material recovered directly from environmental samples, used to analyze microbiomes comprehensively.
2.12	Microbiome	The total collection of microorganisms (bacteria, viruses, fungi, etc.) in or on the human body.
2.13	NIH (National Institutes of Health)	The primary U.S. government agency responsible for biomedical and public health research.
2.14	Pathobionts	Microorganisms that are normally harmless but can cause disease under certain conditions.
2.15	PCR (Polymerase Chain Reaction)	A method to amplify DNA; used in pathogen detection but not sufficient for whole-community microbiome profiling.
2.16	SCFAs (Short-chain Fatty Acids)	Metabolites produced by microbial fermentation, important for maintaining gut health.

2.17	Sexually Transmitted Infections (STIs)	Sexually Transmitted Infections
2.18	Species Plural (spp.)	The term “spp.” Stands for “species plural” and is commonly used in scientific literature to denote more than one species within a particular genus
2.19	Symbionts	Beneficial microorganisms that live in a mutual relationship with the host.
2.20	Taxonomic Profiling	The identification and categorization of microbes in a sample, ideally down to species or strain level.
2.21	16S rRNA Gene	A bacterial gene used in amplicon sequencing to identify and classify microbial taxa.
2.22	Whole Genome Sequencing (WGS)	A method of sequencing the entire DNA content of an organism. In the context of microbiome testing, this entails sequencing the entirety of a microbial community, enabling species/strain identification and functional analysis.

3. Guideline Content

3.1. Clinical Guidelines

- 3.1.1. This guideline outlines best practices for microbiome testing in clinical settings. It incorporates internationally recognized standards from authoritative bodies such as the American Gastroenterological Association (AGA), Human Microbiome Project (HMP), and the European Society of Clinical Microbiology and Infectious Diseases (ESCMID).

3.2. Background

3.2.1. Microbiome Overview

- 3.2.1.1. The microbiome is the collection of all microbes, including bacteria, archaea, fungi, microbial eukaryotes, and viruses/phages, that live naturally on or inside the body.
- 3.2.1.2. The microbiome is responsible for protecting against pathogens, aiding immune system development, and enabling food digestion to produce energy. Such a complex symbiotic interaction is the result of a remarkable metabolic activity driven by a genetic pool whose size is a hundred times larger than the human genome.
- 3.2.1.3. The microbiome serves as a key interface between the body and the environment, and its microbes can significantly impact health in various ways.
- 3.2.1.4. The human microbiome is diverse, with each body site hosting a different community of microbes.
- 3.2.1.5. An individual’s core microbiome is formed within the first few years of life, but can change over time in response to different factors, including diet, medications, and environmental exposures. The composition and rate of change of each person’s microbiota are distinctive from one individual to another, as they are influenced by variables such as age, lifestyle, diet, antibiotics, occupation, and environment.
- 3.2.1.6. Differences in the microbiome may lead to different health effects from environmental exposures and may also help determine an individual’s susceptibility to certain illnesses

- 3.2.1.7. Environmental exposures can also disrupt an individual's microbiome, leading to an increased likelihood of developing diabetes, obesity, cardiovascular and neurological diseases, allergies, and inflammatory bowel disease.
- 3.2.1.8. Composition of microbiota is unique, governed bacteria found in the human gut microbiota are primarily from the phyla Bacteroides and Firmicutes, whereas Actinobacteria and Proteobacteria command the skin microbiome, though there is some overlap and it is important to note that there are differences depending on exact location

3.2.2. Gut Microbiome

- 3.2.2.1. The gut microbiome is a key mediator of essential human functions, including metabolism, immune regulation, colonization resistance, and response to drugs
- 3.2.2.2. There is increasing evidence that imbalance of the gut microbiome is associated with broad range of intestinal and extraintestinal disorders and response to treatments
- 3.2.2.3. There is growing interest in the analysis of the gut microbiome in clinical practice, including the diagnosis, prognostics, or risk assessment for particular disorders, as well as the prediction of patient response to certain treatments targeting the gut microbiome.
 - 3.2.2.3.1. Gut microbiota-based testing is currently not endorsed by the AGA or NIH for the diagnosis of IBS, obesity, or neurological disorders.

3.2.3. Other (non-gut) Microbiomes

3.2.3.1. Skin Microbiome

- 3.2.3.1.1. The skin microbiome is established at birth and is influenced by various factors, including delivery mode, the mother's microbiota, antibiotic treatment, hygiene, nutrient deficiency, housing, animal/pet contact, and environmental exposure.
- 3.2.3.1.2. Microbial colonization at birth and associated factors are known to be essential for immune system development.
- 3.2.3.1.3. Factors such as host physiology, biological sex, age, skin site, climate, geographical region, lifestyle (e.g., occupation and hygiene practices), immune system, host genotype, and underlying conditions (e.g., diabetes) can also influence the skin microbiome throughout life.
- 3.2.3.1.4. Common sample types for evaluating skin microbiomes include skin swabs, tape strips, and/or biopsies.

3.2.3.2. Vaginal Microbiome

- 3.2.3.2.1. Vaginal microbiomes are important in protecting from pathogens such as sexually transmitted infections, urinary tract infections, and yeast infections.
- 3.2.3.2.2. Vaginal microbiota arises from various intrinsic and external factors such as genetics, age, diet, medications, hygiene level, and habits. Vaginal microbiome is dominated by certain lactobacilli species, such as *Lactobacillus crispatus*, which protect women from pathogenic microbes.
- 3.2.3.2.3. The development of imbalanced microbiota composition leads to a pathological condition called dysbiosis, a state which has been linked to various disorders and diseases typical for the urogenital tract. Anaerobic bacteria such as *Gardnerella*, *Atopobium*, and *Prevotella* spp. are shown to dominate microbiota in bacterial vaginosis and increase the risk of vaginal and urogenital infections as well as various sexually transmitted infections.
- 3.2.3.2.4. Recent evidence has linked unhealthy, unbalanced vaginal microbiota

even to poor perinatal outcomes such as miscarriage and preterm birth, and also to severe STIs and even increased risk of cervical cancer.

- 3.2.3.2.5. Evaluation of the vaginal microbiome is examined microscopically and stained by Gram staining. Analysis of vaginal microbiome by sequencing is currently not recommended in clinical practice.

3.2.3.3. Oral Microbiome

- 3.2.3.3.1. The oral microbiota is the second most abundant and diverse microbial community, after the gut microbiota.
- 3.2.3.3.2. Dysbiosis in the oral microbiome is characterized by the involvement of bacterial metabolites in periodontal disease, as well as a notable difference in the metabolic profiles of the oral microbiota compared to those of healthy microbiota.
- 3.2.3.3.3. An imbalance between the microbiota and the host, caused by certain factors, can lead to diversity within the microbial community. This can cause oral and dental diseases in which beneficial bacteria are destroyed, and pathogens proliferate. Under these conditions, the immune system is unable to fight the pathogens, exposing the host to tissue and dental damage such as dental caries, periodontal disease, and carcinogenic metabolites that can cause cancer and other systemic diseases
- 3.2.3.3.4. In addition, imbalances in oral microbiota have been correlated to various types of malignancies, including gastric, colorectal, liver, lung, esophageal, and breast malignancies

3.2.3.4. Respiratory Microbiome

- 3.2.3.4.1. Asthma, chronic obstructive pulmonary disease (COPD), cystic fibrosis (CF), and idiopathic pulmonary fibrosis (IPF) are common chronic respiratory diseases characterized by disrupted and distinct respiratory microbiomes.
- 3.2.3.4.2. There is evidence suggesting that viral infections exacerbate asthma, the lung microbiome plays a significant role in COPD severity, and CF and IPF. Lungs possess unique microbiomes that impact disease transmission and progression.

3.2.3.5. Urinary Tract Microbiome

- 3.2.3.5.1. Urinary microbiota may consist of species residing within the bladder, urinary tract, or urogenital tract
- 3.2.3.5.2. Role of this microbial community in urogenital health and disease, such as urinary tract infection (UTI), is not fully understood.

3.2.3.6. Placental Microbiome

- 3.2.3.6.1. Microbial invasion of the amniotic cavity is associated with significant neonatal and maternal morbidity and mortality
- 3.2.3.6.2. Cultivation-dependent and independent studies have linked placental and amniotic fluid microorganisms with miscarriage, chorioamnionitis, preterm rupture of membranes, preterm delivery and stillbirth

3.2.3.7. Blood Microbiome

- 3.2.3.7.1. Recent reports have revealed the presence of genetic materials of microbes or pathogens in the blood circulation, leading to the conceptualization of a blood microbiome that is vital for physical wellbeing.

- 3.2.3.7.2. Dysbiosis of the blood microbial profile has been implicated in a wide range of health conditions.
- 3.2.3.8. Testing of microbiomes from anatomical sites other than the gut (oral, vaginal, skin, respiratory, urinary, placental, or blood) should be restricted to trained healthcare professionals and ethically approved research settings due to insufficient evidence for routine clinical use.
- 3.2.4. General Principles and Minimal Requirements for Testing of Gut Microbiome**
 - 3.2.4.1. Currently, microbiome testing is only recommended for testing the gut microbiome, bearing in mind the best practices and limitations noted below.
 - 3.2.4.2. General Principles**
 - 3.2.4.2.1. Informed Consent: Patients must be made aware of the scarce evidence for testing and be fully informed on the return of results
 - 3.2.4.2.2. Multidisciplinary team: The team providing the care for the patient must include healthcare professionals who are experts in diagnosing and managing gastrointestinal diseases and conditions
 - 3.2.4.2.3. Adherence to high-quality standards: Laboratories must guarantee high-quality standards and be accredited in accordance with DoH regulations.
 - 3.2.4.2.4. Microbiome testing results should be taken into consideration with all other clinical findings, to ensure holistic view prior to making any recommendations or dietary changes.
 - 3.2.4.2.5. Laboratories must ensure they are using up-to-date software
 - 3.2.4.2.6. Treatment and Implications: any potential change in patients' treatment based on microbiome testing results should be made or supervised by the referring clinician
 - 3.2.4.3. Pre-testing Analytics
 - 3.2.4.3.1. Requirements for testing
 - 3.2.4.3.1.1. A concerned clinician should initiate request for testing
 - 3.2.4.3.1.2. Suspension of chronic treatment or change in diet prior to testing is discouraged, unless recommended by the referring physician to address specific clinical questions and under clinical supervision, as diet and individuals drugs can change the gut microbiome composition.
 - 3.2.4.3.2. Clinical Metadata Collection
 - 3.2.4.3.2.1. Important metadata should be gathered from the patient including:
 - 3.2.4.3.2.1.1. Personal patient demographic and clinical data: age, gender, BMI, smoking status, dietary habits, gut transit time (e.g. Bristol stool scale)
 - 3.2.4.3.2.1.2. Current medical history: current comorbidities, current medications
 - 3.2.4.3.2.1.3. Past medical history: previous diseases, previous relevant surgical interventions, previous drugs (within 3 months of testing)
 - 3.2.4.3.3. Sample Collection and Storage:
 - 3.2.4.3.3.1. Preferred sample type for gut microbiome analysis is stool samples.
 - 3.2.4.3.3.2. Stool samples should be collected utilizing a stool catcher or validated stool collection kit, with available preservative media

- 3.2.4.3.3.3. Collection kits should contain proper instructions for the recommended amount of stool to be collected and proper instructions for labelling, packaging, short-term storage, and waste disposal
- 3.2.4.3.3.4. Stool samples should be collected either at home or in dedicated areas within healthcare facilities by patients
- 3.2.4.3.3.5. Bristol stool chart can be used to record the consistency of stool samples.
- 3.2.4.3.3.6. Collected samples should be shipped to testing laboratories with assurance standards for microbiome sequencing within recommended timeframes and following the conditions described by the collection kit.
- 3.2.4.3.3.7. Samples should be stored at -80 C in the laboratory until further processing.
- 3.2.4.3.3.8. Samples should be stored according to specimen type within the laboratory to ensure integrity and quality of the sample prior to testing.
- 3.2.4.3.3.9. The processing center should have a clear criteria for acceptance and rejection of the samples.
- 3.2.4.3.3.10. Analysis of the microbiome from extraintestinal body sites is a promising field in research, but requires further development before being applied in clinical practice. This includes analysis of samples from the microbiome involving specimen other than fecal samples, including vaginal, skin, oral swabs, saliva, and breastmilk.
- 3.2.4.3.3.11. Samples should be stored at -80 °C in the laboratory until further processing.
- 3.2.4.3.3.12. Analysis of the microbiome from extraintestinal body sites is a promising field in research but requires further development before being applied in clinical practice. This includes analysis of samples from the microbiome involving specimen other than fecal samples, including vaginal, skin, oral swabs, saliva, and breastmilk.
- 3.2.4.3.3.13. Stool samples should be collected utilizing a stool catcher or validated stool collection kit, with available preservative media
- 3.2.4.3.3.14. Collection kits should contain proper instructions for the recommended amount of stool to be collected and proper instructions for labelling, packaging, short-term storage, and waste disposal
- 3.2.4.3.3.15. Stool samples should be collected either at home or in dedicated areas within healthcare facilities by patients
- 3.2.4.3.3.16. Bristol stool chart can be used to record the consistency of stool samples

3.2.4.4. Sequencing Technology

- 3.2.4.4.1. For microbiome testing, amplicon or whole genome sequencing (WGS) are reliable methods for community-based profiling of microbiomes.
- 3.2.4.4.2. Advantages and disadvantages of amplicon vs WGS are noted in Table 1.

Table 1: Comparison of Amplicon vs Whole Genome Sequencing

Function	Amplicon sequencing (e.g. 16S rRNA)	WGS
Sample requirement	Lower amount of biological sample required	Higher amount of biological sample required
Risk of contamination by host DNA	Hardly affected by host DNA	Can be affected by host DNA
Target of sequencing	Specific gene (e.g. 16S rRNA) or portion (e.g. Specific 16S rRNA variable region)	Whole DNA analysis of the sample
Sequencing costs	Lower cost per sample	Higher cost per sample
Taxonomic resolution	Up to the genus taxonomic level	Strain-level resolution
Functional analysis	Not available	Identified genes and functions of microbial communities

3.2.4.4.3. Conventional microbial cultures or molecular techniques (e.g., multiplex PCR) are useful in several clinical contexts, primarily for identifying specific pathogens. However, they are not suitable for evaluating the composition of microbial communities and therefore cannot be considered microbiome testing or used as a proxy for microbiome profiling.

3.2.4.4.4. Quality indicators should be part of the processing and analysis steps.

3.2.4.5. Pre-processing

3.2.4.5.1. Standard operating protocols for processing raw sequenced data must be described in detail, following adherence to high-quality standards.

3.2.4.6. Community and Taxonomic profiling

3.2.4.6.1. Microbiome analysis should include alpha diversity metrics, including richness and evenness, and beta diversity measures should be included in the microbiome analysis.

3.2.4.6.2. Alpha diversity, an ecological measure of the complexity and variety of an ecosystem that might associate with clinical response, should always be calculated within testing.

3.2.4.6.3. Beta diversity, an ecological measure of the similarity between the compositions of two microbial communities, should be calculated within the testing when longitudinal sample or multiple samples from different sites are compared or when they are contextualized with other normal or pathological result.

3.2.4.6.4. Complete taxonomic profiling: Complete taxonomic profiling is an essential component of microbiome testing

3.2.4.6.5. Taxa should be identified at all possible levels

3.2.4.6.6. Amplicon sequencing provides details from phylum to genus level.

3.2.4.6.7. WGS provides details from phylum to species or strain with their relative contribution to the whole microbiota community.

3.2.4.6.8. Comparison with a matched control group: Appropriate comparison to a healthy control group should be included to aid in the

- interpretation of patient taxonomic and diversity profiles.
- 3.2.4.6.9. Publicly accessible resources should be used to ensure sufficient control group size and potential confounding factors.
 - 3.2.4.6.10. Longitudinal assessment of the patient microbiome at different points in time can be useful in specific clinical scenarios, such as assessing the effect of a treatment or diet or to evaluate the microbiome composition after a stressful event.
- 3.2.4.7. Reporting Requirements
- 3.2.4.7.1. Standard operating procedures for sequencing, post-processing, and analysis should be detailed and readily available.
 - 3.2.4.7.2. The reporting of demographic and clinical metadata should ease the interpretation of test results by the referring physician.
 - 3.2.4.7.3. Stool collection protocols should be reported in addition to details on DNA extraction, as these variables could influence the outcome of the analysis.
 - 3.2.4.7.4. Main features of sequencing methods (e.g., amplicon-based vs. WGS), depth of sequencing, the technology and amplicon region (if available), should be reported.
 - 3.2.4.7.5. Alpha and beta diversity measures assessed should be included in the final report, and the microbiome composition should be described with the deepest possible taxonomic resolution.
 - 3.2.4.7.6. Reported taxonomic profile should provide at least a degree of reference to the percentage of sequencing data that could not be assigned to a particular taxonomy.
 - 3.2.4.7.7. The report should include all taxa shift from significantly healthy matched controls, as well as known microbial pathogens.
 - 3.2.4.7.8. All taxa that diverge significantly from matched health ranges tailored to the patient population, such as *C. difficile*, *Salmonella* spp, *Shigella* spp, or pathogenic *Escherichia coli* strains.
 - 3.2.4.7.9. It is recommended to report other health-relevant taxa and clusters (e.g., *Akkermansia* spp, *Bifidobacterium* spp, *Enterobacteriaceae*, *Fusobacterium* spp, *Lactobacillus* spp, and short-chain fatty acid-producers), regardless of their abundance, could help the clinical management of patients.
 - 3.2.4.7.10. It is recommended to report microbial taxa abundance as percentages, as there is a lack of evidence to report strict healthy reference ranges for the relative abundances of bacterial taxa.
 - 3.2.4.7.11. Post-testing therapeutic advice strongly discouraged, and should be charged to the referring physician who requested the testing.
 - 3.2.4.7.12. Reporting *Firmicutes: Bacteroidetes* ratio and dysbiosis indices are discouraged.
 - 3.2.4.7.13. User-friendly infographics within the report to ease interpretation are strongly recommended.
 - 3.2.4.7.14. Taxa and microbial clusters relevant for human health should be reported.

4.Relevant References Documents

No.	Reference Date	Reference Name	Relation Explanation / Coding / Publication Links
1	2025	National Institute of Environmental Health Sciences	https://www.niehs.nih.gov/health/topics/science/microbiome
2	2019	What is the Healthy Gut Microbiota Composition? A	https://pmc.ncbi.nlm.nih.gov/articles/PMC6351938/pdf/microorganisms-07-00014.pdf
3	2017	European Journal of Nutrition	https://link.springer.com/article/10.1007/s00394-017-1445-8
4	2023	Microorganisms	https://www.mdpi.com/2076-2607/11/5/1222
5	2023	Microorganisms	https://www.mdpi.com/2076-2607/11/2/298
6	2024	Nature Reviews Microbiology	https://www.nature.com/articles/s41579-024-01075-5
7	2024	Microbiome	https://www.mdpi.com/2076-2607/12/9/1797
8	2025	Microbial Biotechnology	https://link.springer.com/article/10.1007/s00394-017-1445-8
9	2024	Science Direct European Urology Focus	https://www.sciencedirect.com/science/article/abs/pii/S2405456924002669
10	2005	De Gruyter Brill	https://www.degruyterbrill.com/document/doi/10.1515/JPM.2003.016/html
11	2023	International Journal of Molecular Sciences	https://www.mdpi.com/1422-0067/24/6/5633
12	2025	The Lancet Gastroenterology and Hepatology	https://www.thelancet.com/journals/langas/article/PIIS2468-1253(24)00311-X/abstract
13	2024	Guidelines for Clinical & Translational Research in Genomics	https://www.doh.gov.ae/en/resources/guidelines
14	2023	Standard for Laboratory Accreditation for Genomic-Related Services and Products	https://www.doh.gov.ae/en/resources/standards