			InVyo	mma Plus RG	CC
Patient NAME Mr John Doe	DATE OF BIRTH 1992-Jun-12	DISEASE Prostate	STAGE II	Physician NAME Administrator	
SPECIMEN 20ml Blood	VIAL IDs 1				

# **REPORT SUMMARY**

The result of analysis of the gut microbiome is indicative of a relative balance environment with some evidence of disturbance of the normal flora. Normal level of diversity is observed, which represents a healthy gut environment. Firmicutes/Bacteroidetes ratio is marginally increased, which indicates a partial balance in the gut environment.

Analysis of the detected dominant species:

- Eubacterium\_eligens (Phylum Firmicutes) : gram-negative, anaerobic bacteria, which is one of the most abundant and important bacteria of human gut microbiome. It produces Short Chain Fatty Acids (SCFA), mostly butyric acid, promoting health and balance in general.
- Faecalibacterium prausnitzii (Phylum Firmicutes): gram-positive, anaerobic bacteria, which is one of the most abundant and important bacteria of human gut microbiome. It produces Short Chain Fatty Acids (SCFA), mostly butyric acid, promoting health and balance in general.
- Clostridium spp: (Phylum Firmicutes) : gram-positive, anaerobic bacteria, with great heterogenicity in terms of its function.
- Prevotella\_copri: (phylum Bacteroidetes) : Gram-negative, anaerobic bacteria, which has been associated in the literature with increased incidence rates of rheumatoid arthritis.
- Ruminococcus\_bromii (Phylum Firmicutes) : gram-positive, anaerobic bacteria, which is one of the most abundant and important bacteria of human gut microbiome. It participates directly in the degradation of resistant starch.

Administration of probiotics, enriched in Bifidobacterium and Lactobacillus genus and prebiotics are recommended, for the restoration of the normal flora in the gut environment. Repetition of the analysis is recommended two months after the completion of the treatment

#### Disclaimer

\*This Test Report is issued based on testing the sample / specimen examined by the Laboratory. Modification of data, selective breeding and using portions of this test report is forbidden. The laboratory assumes no liability for improper use or improper interpretation of the result





# **MICROBIOME ANALYSIS**

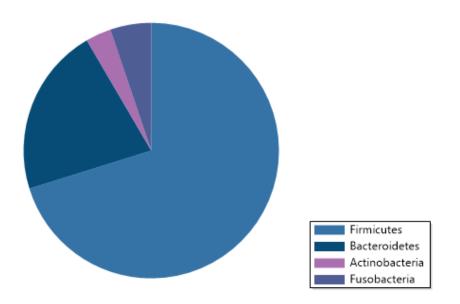
# MICROBIAL DIVERSITY OF GUT MICROBIOME

SHANNON INDEX	NORMAL VALUES
3.43	> 2.0

Shannon Index expresses the degree of diversity of gut microbiome and by extension the stability and health of gut environment. High index values represent a healthy gut environment, while lower gut bacterial diversity has been reproducibly observed in patients with a wide range of conditions, including obesity, cardiovascular disease, autoimmune disease and neurological disorders, along with vaccine responses, and carcinogenesis suggesting a direct relationship between GM diversity and functional outcomes.

# DETECTION RATE OF BACTERIAL PHYLA (%)

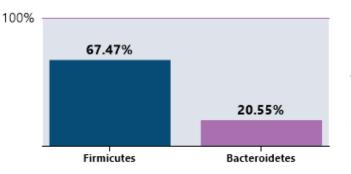
Phylum	%
Firmicutes	67.47%
Bacteroidetes	20.55%
Actinobacteria	3.1%
Fusobacteria	5%





### FIRMICUTES/BACTEROIDETES (RATIO)

Firmicutes/Bacteroidetes Ratio	NORMAL VALUES
3.28	0.7 - 1.1



Firmicutes and Bacteroidetes are the most common representatives of colon bacteria. A high ratio is associated with being overweight, whereas a lack of Firmicutes (low ratio) is associated with poor calorie utilization and being underweight.

#### **PROTEOBACTERIA INDEX**

Proteobacteria index	NORMAL VALUES
0	< 2.0

A healthy intestinal flora/gut microbiome should contain only a small proportion of proteobacteria, as many potential pathogens belong to this bacterial phylum. Having a small number of these bacteria is perfectly normal.

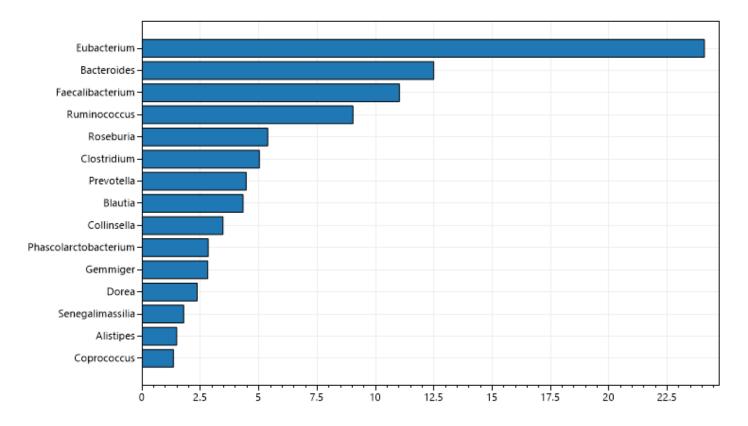
### SPECIES RICHNESS

SPECIES RICHNESS	NORMAL VALUES
80	100 - 350

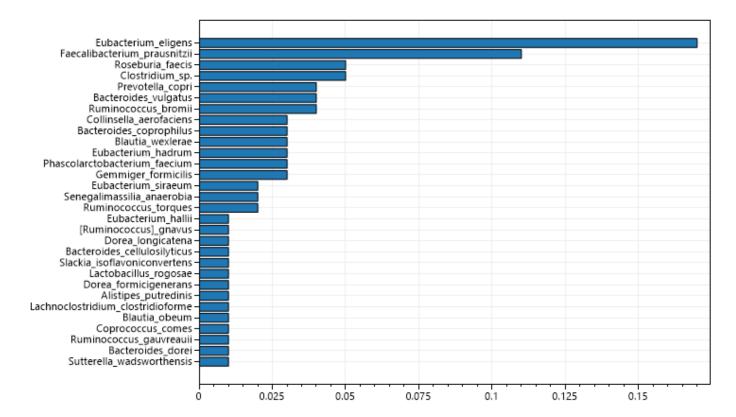
Species richness describes the number of different species / types of bacteria in the gut system. In a more diverse microbiome, the large number of different bacterial species can contribute to many different functions being carried out by bacteria. As a consequence, the body can utilize food and nutrients better, as well as handle stress and malnutrition more easily.



#### 15 DOMINANT BACTERIA IN GENUS LEVEL (%)



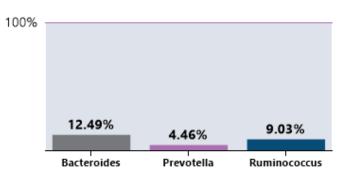
### 30 DOMINANT BACTERIA IN SPECIES LEVEL (%)





Patient NAME Mr John Doe	DATE OF BIRTH 1992-Jun-12	InVyomma Plus RGCC
ENTEROTYPE		

ENTEROTYPE		
Bacteroides (%)	12.49	
Prevotella (%)	4.46	
Ruminococcus (%)	9.03	



The enterotype develops during the first years of life. It is independent of gender, age or geographical origin and it is mainly related to genetics and eating habits. Each enterotype is dominated by a different bacterial strain/grenus. It affects energy production from food, as well as the production of vitamins.

Enterotype 1 = Especially for people who often eat meat: Bacteriodes

Enterotype 2 = Especially for people who eat vegan or vegetarian food: Prevotella

**Enterotype 3** = Especially for people who prefer a balanced diet: Ruminococcus

### DETECTION RATE OF BIFIDOBACTERIUM GENUS (%)

Bifidobacterium (%)	NORMAL VALUES
0.21	0.5 - 12

Bacteria belonging to Bifidobacterium genus are considered as healthpromoting bacteria. They have a significant impact on infant health. Despite decreasing abundance in adult microbiomes, these bacteria play an important role in restoring intestinal/gut health, prevention of inflammation and allergies and dispersal of pathogenic microorganisms. Therefore, they are added to different probiotic dairy products, such as kefir and yoghurt.

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#### DETECTION RATE OF LACTOBACILLUS GENUS (%)

Lactobacillus (%)	NORMAL VALUES
1.09	0.03 - 19

Lactobacillus is a genus of Gram-positive, aerotolerant anaerobes or microaerophilic, rod-shaped, non-spore-forming bacteria. Their role in the maintenance of the stability and health of gut is important as they participate in the formation of biofilms and by extension in the creation of barriers though their adhesion to the mucus membrane and their production of antimicrobial peptides and H202. Lactobacillus directly metabolize glycogen into lactic acid, which is essential for gut health.

# **METABOLIC INDICATORS**

METABOLIC INDICATORS	VALUE	NORMALI VALUES
Total SCFA	28 µmol/g	>= 23 µmol/g
Butyrate	4.5 µmol/g	>= 3.6 µmol/g

% OF TOTAL SCFA	VALUE	NORMAL VALUES
Acetate	65%	50% - 72%
Butyrate	18%	11% - 33%
Propionate	17%	<= 29%

Sincerely,

Dr. Ioannis Papasotiriou MD, PhD, SCym

# InVyomma Plus RGCC

#### Human Gut Microbiome In Health And Disease

The gut microbiome (GM) refers to the genetic makeup of all microbes that exist within the human gastrointestinal tract, including bacteria, viruses, yeast, protozoa, fungi and archaea. The GM contains -100 trillion micro-organisms, which encode over three million genes producing thousands of metabolites, which replace or modulate many of the functions of the human host. Constituents of the GM have been shown to interact with one another and the host immune system in ways that influence physiological homeostasis and the development of disease. Disruption of the delicate balance of commensal bacteria is seen in the setting of what is often referred to as dysbiosis, a somewhat ill-defined state characterised by a less diverse and less stable microbiota, with potential enrichment of opportunistic pathogenic bacteria. Such an imbalance can lead to impaired local, locoregional and systemic immune responses through the breakdown of mucosal barriers, alterations in cytokine signalling, inhibition of probiotic commensal bacterial colonisation and proliferation of enteropathogens.

It has been discovered that the microbiome is closely related to the occurrence and development of a variety of cancer types in the epithelial barrier and sterile tissues. Symbiotic ecosystems that inhabit the gut or other mucosa perform a role in both local and distal carcinogenesis. On the one hand, microbiome can provide toxic metabolites or carcinogenic products directly as cancer-transforming agents. On the other hand, it can indirectly play a role in promoting cancer by the induction of inflammation or immunosuppression.

#### Laboratory Process

Information

DNA extraction was performed from the clinical sample, followed by amplification with Polymerase Chain Reaction – PCR. Sequencing of 16S ribosomal RNA bacterial gene was performed by NGS-next generation sequencing, using the platform Ion Torrent PGM<sup>™</sup> System (life technologies/ThermoFisher SCIENTIFIC). The hypervariable regions, V2, V3, V4,V6, V7, V8 and V9 of 16S rRNA gene were sequenced and data analysis was performed via advanced bioinformatic programs. 16S rRNA gene is conserved in all bacteria and its sequence enables the taxonomic assignment and relative quantification of each bacteria detected in the sample.

According to literature, the sensitivity of the method is 99%.

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