



Opinion

Does Intestine Morphology Still Have Secrets to Reveal? A Proposal about the “Ghost” Layer of the Bowel

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Abstract: In this brief Opinion paper, the term “muco-microbiotic layer” is introduced to describe the innermost layer of the intestinal wall. This layer may contribute not only to the overall health of the bowel, but also to that of extraintestinal organs. Its constituents, in terms of soluble molecules and nanovesicles, need to be studied further. Moreover, one can hypothesize the existence of an analogous layer in other organs, such as the airways or some parts of the genital tracts. Further studies on it are needed.

Keywords: microbiota; bowel; muco-microbiotic layer; cell differentiation; tissue homeostasis; organ remodeling; human anatomy; histology; pathophysiology; nanovesicles; exosomes



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1. An Anatomical Introduction

In Anatomy and Histology treatises, within the chapters on the structure of the intestine, little space is left to uncertainty and the readers’ imagination, whether they be young students or expert scientists: the intestinal wall—just like the other organs of the alimentary canal—consists of four overlapping tunicae as if they were four soft cylinders inserted one inside the other, which have been named—from the inner to the outer—“mucosa”, “submucosa”, “muscularis propria” and “adventitia” (the latter incorporated in the submesothelial connective tissue of the serosa, the peritoneum, when present) [1].

Particular attention is given to the mucosa, usually defined as the innermost tunic, which is, in turn, composed of three layers, all of the equal importance: the epithelium (the layer facing the lumen), whose cellular elements morpho-functionally better characterize the two portions into which it is divided the intestine (i.e., small and large); the lamina propria, containing not only the blood vessels for tissue trophism but also lymphatics, immune cells, undifferentiated mesenchymal cells, nerve fibers, and other cells and structures that harbor—in some parts—glandular introflections derived from the lining epithelium; and, last but not least, the muscularis mucosae, a thin bundle of smooth muscle fiber cells, which has the dual purpose of protecting the submucosa (more richly vascularized and innervated than the lamina propria) from chemical or traumatic insults that can affect the more abluminal layers on one hand, and to physically separate the two connective layers of the lamina propria and submucosa, thus actively participating in the creation of two different microenvironments, on the other [1].

The concept of the microenvironment becomes more relevant as our knowledge of the constituents of the human body increases, since it is now clear that cells can differentiate, perform metabolic functions, communicate with each other, and cease (more or less voluntarily) their existence based on the chemical–physical stimuli provided by the environment [2].

2. The Microbiota and Its Microenvironment

In the last few decades, a further constituent of the intestine structure has emerged in the scientific literature: the microbiota, that is, the array of all living elements, bacterial and not, that colonize our intestine, not having been generated together with it—therefore not sharing their genetic heritage with the cells of the individual who hosts them—but having reached a saprophytic balance with it [3]. The constituents of microbiotic flora are presented in Figure 1 and Tables 1–3.

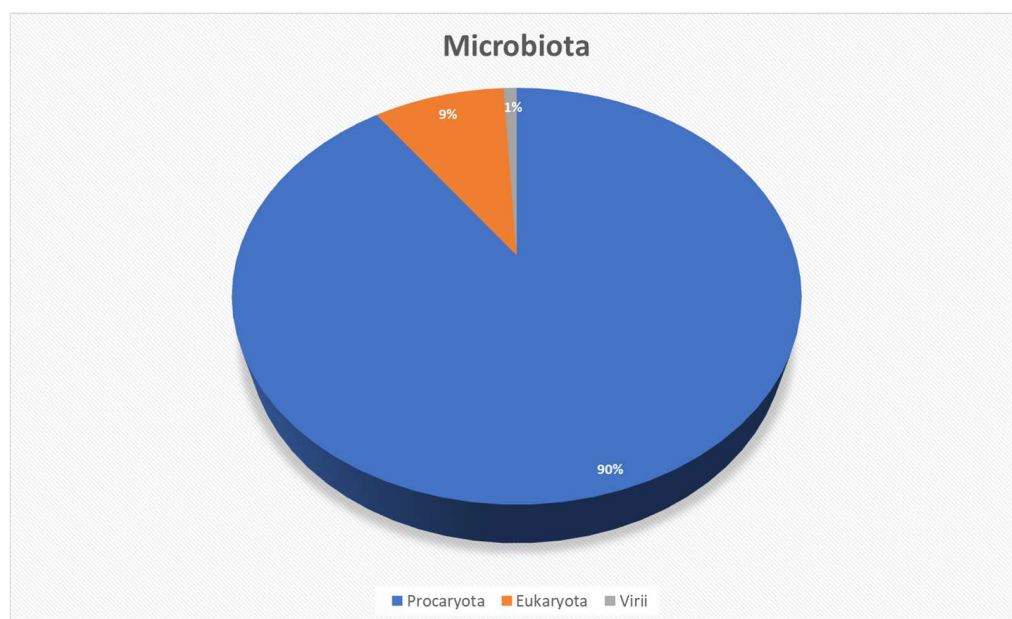


Figure 1. The microbiotic flora. The microbiota is mainly made up of prokaryotic elements (90%) and, in a lesser extent, of eukaryotic (9%) and virii (1%) species. See Tables 1–3 for more information about its composition.

The microbiota is defined by some—improperly, from a strictly morphological point of view—as “the organ within the organ”; instead, we would like to redefine and reframe it in a morpho-functionally more appropriate way in the short text that follows. To do this, we have to start by correlating the “morphology” with the “pathophysiology”. Thus, please forgive us for the slightly pedantic nature of the following paragraphs.

The human body can be subdivided into apparatuses (or systems), which can be split into organs, each formed of tissues, in turn composed of cells, each specialized for their function (just remember how different they are, morpho-functionally, not just a neuron from a muscle cell, but also an absorbing epithelial cell from a secreting one) [4].

The alteration of cellular differentiation triggers the disorganization of the tissue, which, in turn, causes organ malfunction, causing the organism to display signs of illness. Physicians, through the knowledge of topographical anatomy, are called to identify the organ and/or the anatomical area affected by the disease, using semiotic tools (physical and instrumental); the knowledge of the structures of the human body permits them to identify where those natural mechanisms regulating cell differentiation, tissue homeostasis and organ remodeling (all phenomena that continue throughout life, both pre- and postnatally, including not only infancy and adolescence but also tissue senescence and organ meiorpragia later in life) are malfunctioning [5].

Table 1. The Prokaryotic kingdom and their phyla in the human microbiota. The percentages derive from literature data.

Prokaryota				
Kingdoms	Phylum	Genus	(%)	References
Bacteria	Firmicutes	Lactobacillus	5%	[6–12]
		Faecalibacterium	5%	[12]
		Halomonas	3%	[12]
		Clostridium	3%	[12]
		Ruminococcus	7%	[12]
		Roseburia	4%	[12]
		Streptococcus	2%	[12]
		Others	21%	[12]
	Bacteroidetes	Bacteroides	19%	[6–13]
		Others	2%	[12]
	Actinobacteria	Bifidobacterium	4%	[9,12,14]
		Others	1%	[12]
	Proteobacteria	Escherichia	5%	[10,13]
		Shigella	2%	[10]
		Others	9%	[13]
	Spirochaetes	Brachyspira	3%	[12]
		Others	2%	[12]
	Others	Others	2%	[11]
	Archea	Euryarchaeota	Halobacteria	2%

Table 2. The Eukaryotic kingdom and their phyla in the human microbiota. The percentages derive from literature data.

Eukaryota			
Phylum	Genus	(%)	References
Ascomycota	Debaryomycetaceae	13%	[16–18]
	Dipodascaceae	14%	[17]
	Saccharomycetaceae	13%	[17]
Basidiomycota	Tremellomycetes	20%	[16]
	Malasseziaceae	20%	[19]
Mucorimycota	Fusicatenibacter	5%	[17]
	Aspergiullus	5%	[19]

Table 3. The Virii kingdom in the human microbiota. The percentages derive from literature data.

Virii			
Phylum	Genus	(%)	References
Caudovirales	Siphoviridae	20%	[11,20–23]
	Myoviridae	20%	[17,22,23]
	Podoviridae	20%	[17,22,23]
	Others	40%	[11,22,23]

The identification of “breakdown” at the cellular level can lead to targeted therapy (sometimes just lifestyle changes are enough; other times, drug therapy may be necessary; and, in extremis, surgical intervention could be inevitable for the demolition and eventual reconstruction of the irretrievably sick part), and the efforts of the biomedical and bioengineering community are concentrated above all on developing such therapy protocols.

It now seems certain that the microbiota is involved in many intestinal pathologies, therefore implying that it can transform itself from a combination of commensal germs to a mix of pathogens under certain conditions and for causes that are not yet fully understood [24–27]. What mainly affects the health of the microbiota is certainly the microenvironment in which we live and, in the first place, our diet: what passes through the intestinal lumen throughout our existence. However, to better understand how and why the microbiota is altered and, above all, how and why this alteration affects not only intestinal homeostasis but also that of other tissues physically distant from it, the context of its microenvironment, i.e., the mucus that lines the epithelial surface of the intestine and in which the microbiota is contained, must be contemplated [28].

3. The Muco-Microbiotic Layer

Mucus and microbiota form a morpho-functional layer, the biological elements of the microbiota being comparable to a “cellular component”, and the mucus to the “matrix” in which not only these cells reside, but also in which their products—both soluble molecules (proteins, lipids, etc.) and nanovesicles (for example, the “outer membrane vesicles”, OMV, of prokaryotic origin, equivalent to the exosomes produced by eukaryotic cells)—are released [28,29].

Figure 2 shows, schematically, the composition of the muco-microbiotic (MuMi) layer with its main constituents. Why has the MuMi layer been ignored until now, and why has it not yet been described in anatomy and histology books? This is probably due to the fact that widely used histochemical stains contain alcohol, which solubilizes the mucus by sweeping it away along with the microbiota. Therefore, in histological sections, neither of these two components are visible, if not in minute traces, and this has led scientists to dismiss it as a physiological constituent of our bodies.

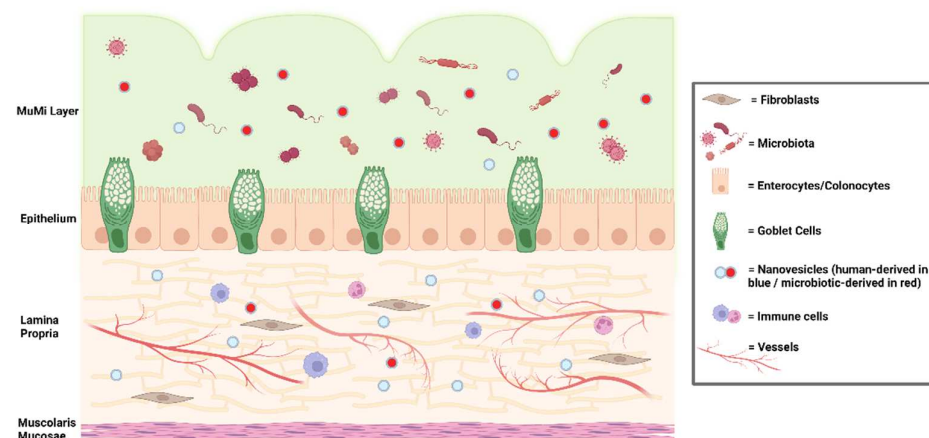


Figure 2. The muco-microbiotic (MuMi) layer. This figure shows, schematically, the MuMi layer, its main constituents (in terms of microbiotic elements and nanovesicles) and its relationships with the underlying mucosal components, i.e., epithelial cells and lamina propria. Whereas nanovesicles, either human- and microbiota-derived ones, can easily reach the lamina propria (in turn contributing to determine its microenvironment), the muscularis mucosae represents a wall between the connective tissue of the lamina propria and that of the submucosa (not shown).

The MuMi layer is therefore located in the bowel internally to the mucosa, representing the innermost “fifth layer” (or the first one) of the intestinal wall; the alterations of the mucous matrix can cause a qualitative and quantitative modification of the populations of

germs that housed in it, and vice versa [28,29]. In addition, the intense traffic of soluble products and nano-vesicles mutually influence human and microbiotic cells; the increase in pathogenic bacteria generates, for example, a disturbance in the microenvironment of the lamina propria, triggering or perpetuating inflammatory processes; and since the nanovesicles (both human and bacterial) can easily reach both the bloodstream and the lymphatic one, the well-being of our intestinal MuMi layer can affect the homeostasis of extra-intestinal tissues, thus compromising the function of other organs, including the liver, lung, heart, and brain, just to mention the most important ones considering the anatomical progression of the blood mass originating from the abdominal splanchnic district [28].

4. Conclusions

Based on the consideration reported above, the existence of the MuMi layer, in our modest opinion, should not only no longer be ignored, but must also be investigated (also in other anatomical districts, e.g., the airways or some genital tract organs) in order to characterize, from a morpho-molecular point of view, its constituents and the mechanisms that fine-tune all the related physiological and pathophysiological events.

Limiting our conclusion to the bowel, if the Hippocratic axiom “we are what we eat” is still true, and if it is correct to think that the health of our body depends, in the first instance, on the health of our intestine, omitting the observations of molecular phenomena that occur in this layer from scientific reasoning—as well as from scientific protocols—would undermine the achievement of better knowledge on human health.

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