Gut Microbiota in the Elderly

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Gut microbiota is involved in the maintenance of physiological homeostasis, thus the alteration of its composition and functionality, called dysbiosis, has been associated with many pathologies, and could also be linked with the progressive degenerative process in aging. Specific gut microbiota taxa could be associated to the development of inflammation underlying aging, but also it has been identified some beneficial profiles related to a healthy status in the elderly. Thus, gut microbiota emerges as a therapeutic target with a double impact in the elderly, counteracting both aging itself and associated diseases.

Keywords: Gut Microbiota ; Physical Exercise ; Elderly ; Fatty LIver ; NAFLD ; Nutrition

1. Introduction

As previously stated, even in physiological conditions, the gut microbiota is a dynamic ecosystem. The composition of the gut microbiota is based on permanent and transitory bacterial species of 17 different phyla such as *Firmicutes*, *Bacteroidetes* and *Proteobacteria*, which can reach as much as 70%, 30% and 5% of the total abundance, respectively, between others[1]. This composition changes depending on the anatomic region of the gastrointestinal tract, due to pH, secretions, motility or substrate availability. A gradual increase of bacterial concentration and complexity exists through the stomach and the gut, reaching the maximum in the colon. Moreover, GM experiments with taxonomical and functional changes during the life of an individual since the prenatal period. The microbiota colonization on the gastrointestinal tract may be started in utero with the placenta and amniotic fluid microbial communities of the mother, as recent research has observed when comparing these microbial populations with the meconium ones[2]. Microbiota profile constitution is affected by numerous factors, such as genetic components, type of delivery (vaginal or cesarean), the feeding (breast-feeding or formula-feeding) or antibiotics and/or probiotics consumption during the first days of life. The initial gut microbiota of infants is a quite instable simple structure dominated by bifidobacteria[3] and is in continuous change until the age of three years. At this moment, the microbiota profile is established and acquires an adult pattern that is relatively stable over time. Nevertheless, there are many causes that can modify this adult profile, for instance lifestyle, exercise, dietary patterns, stress or pathophysiology. Microbiota changes drastically in the elderly and these age-related changes are directly correlated with an inflammatory pattern linked with many diseases. Generally, these changes are orientated to a loss of diversity, a reduction of the abundance of beneficial bacteria such as those which produce short-chain fatty acids (SCFAs)[4], a change in the dominant species or an increase of enteropathogens[5]. All of these modifications are associated with physiological changes in the gastrointestinal tract and in dietary patterns, and with a decrease in the immune system function[6], an increase in the inflammatory state and a feasible contribution to the progression of diseases[4] and frailty[6]. Regarding the changes in dietary choices in the elderly, reductions in taste, dentition, chewing ability and intestinal transit time are factors that contribute considerably[7].

2. Latest Research

Some researchers have focused on establishing the specific age-related microbiota profile. At phylum level, *Firmicutes* is predominant in adults, being reduced in the elderly, whereas there is some discrepancy about the increase or decrease of *Bacteroidetes* phylum with age[6][8][9]. Moreover, high levels of *Proteobacteria* phylum especially *Enterobacteriaceae* family[8] and *Clostridia* class[9]), as well as a decrease of *Actinobacteria* (especially *Bifidobacterium*, a genus with intestinal protective capacity) have been reported in old people[6][8][9][10]. In fact, inflammatory markers such as IL-6 or IL-8 have been associated with an enrichment in *Proteobacteria* phylum, which increases with age[11]. Biagi et al.[11] identified in an elder population in Italy a decrease in bacterial diversity, as well as a change in the relative proportion of *Firmicutes* phylum, an increase of *Bacili* and low levels of *Clostridium* cluster XIVa. This reduction in the abundance of *Clostridium* cluster XIVa was corroborated by later studies[3]. Moreover, Rahayu et al.[9] analyzed the gut microbiota composition of 80 volunteers from Bali and Java arranged in two groups depending on the age (25–45 age old and 70 years old), identifying

a reduction of *Bifidobacterium, Prevotella* and *Lactobacillus plantarum* taxa and an increase in *Enterobacteriaceae* and *Lactobacillus reuteri* in the elderly subjects). Furthermore, Bian et al.[12] developed a study with 1095 healthy volunteers from different cities of China and showed a decrease of *Bifidobacterium* and *Bacteroides* genera in older subjects, whereas *Dorea, Clostridium* and *Marvinbryantia* genera were increased in that population. In this research, *Faecalibacterium* genus was identified as a core and stable microorganism among life. Additionally, Claesson et al.[13] observed that low levels of diversity were correlated with inflammatory markers, frailty and impaired health parameters, as well as diet patterns. Frailty has also been associated with a low abundance of butyrate-producers like *Faecalibacterium prausnitzii*[8][14], *Lachnospiraceae* family and *Roseburia* genus, whereas there are some species that associate positively with frailty, such as *Eggerthella dolichum* or *E. lenta*[14].

In spite of the great inter-variability among studies, there are some taxa which may be less susceptible to be modified by external factors and may constitute the core of gut microbiota composition in the elderly. The reduction in the abundance of *Ruminococcus*, *Blautia* or *Clostridium* cluster XIVa and *Clostridium* cluster IV and the major prevalence of facultative anaerobes like *Escherichia coli* are changes that have been observed in the elderly population across many studies[15]. Even so, it is difficult to establish a unique aging gut microbiota composition profile, due to many factors that modulate this internal ecosystem such as the ethnicity, lifestyle, dietary patterns, host genetics, the presence of comorbidities or even methodological tools. Those reasons reveal the importance to do more studies to reach a homogeneous aging gut microbiota signature.

In the elderly, the presence of comorbidities is a very common situation that requires polypharmacy in order to improve the health status. Thus, not only the comorbidities but also the polypharmacy are factors that modify drastically the composition and diversity of microbiota. In fact, Ticinesi et al.[16] studied the differences between a cohort of 76 elderly hospitalized and multimorbid patients and a group of 25 healthy active elderly volunteers. The β-diversity index showed that the microbiota profile of hospitalized patients was significantly different in comparison with non-hospitalized group. The number of drugs was negatively correlated with Chao-index α-diversity and with the taxa *Massilia* and *Lachnospiraceae*. Furthermore, *Coprobacter*, *Helicobacter* and *Prevotella* were positively correlated with polypharmacy. The hospitalization is another factor that modifies gut microbiota composition, characterized by a substantial decrease in *Faecalibacterium prausnitzii*, *Desulfovibrio* spp. or *Bifidobacterium*, between others, and a major increase of the abundance of enterobacteria[8]. Moreover, Claesson et al. [13] and Jeffery et al.[7]identified in a study which compared the gut microbiota of Irish elderly by their type of residence (community-dwelling, one day at hospital, short-term rehabilitation and residential care) a relationship between the institutionalization of elderly people and an increase of *Firmicutes* phylum and *Parabacteroides*, *Eubacterium*, *Anaerotruncus* and *Coprobacillus* genus, as well as a reduction in bacterial diversity and the abundance of short-chain fatty acid (SCFAs) producers. These results are in agreement with previous reports, indicating that the microbiota profile related to age is aggravated by polypharmacy, reducing the number of SCFAs producers such as *Lachnospiraceae* family and increasing the abundance of some enteropathogens like *Helicobacter*.

It is important to consider the importance and the difference between biological and chronological age, being first physiological age, which takes into consideration many issues such as lifestyle or environmental and genetic factors, and the second one the number of years a person has been alive. Bacterial diversity has been negatively correlated with biological age, but not with chronological age. Moreover, *Ruminococcus*, *Coprobacillus* and *Eggerthella* genera have been associated positively with biological age, independently of the chronological one[4]. Related to that, many studies have identified the microbiota profile of centenarians—those people close to 100 years old—showing a specific healthy composition closer to adults' pattern and remarkably different to that commonly observed in elders over 65 years. Wang et al.[17] described the composition of gut microbiota of centenarians in East China and observed that the α-diversity was significantly increased, as well as the genus *Escherichia* and *Roseburia* in centenarians. Moreover, volunteers more than 100 years old also showed a decrease in the abundance of *Lactobacillus*, *Butyricimonas*, *Coprococcus*, *Parabacteroides*, *Akkermansia*, *Sutterella* and *Faecalibacterium* genera compared with the groups between 80 and 99 years old. Afterwards, Wang et al.[18] conducted a similar study in a larger population and described that community richness and α-diversity was significantly lower in the 65–70 years age group compared with the 90–99 and the 100+ year age groups. An increase in the relative abundance of *Synergistetes* phylum (with special mention of *Prevotellaceae*, *Lachnospiraceae* and *Porphyromonadaceae*) was observed in the longevity group compared with the younger elderly group. Similar results related to the increase of microbial diversity and *Porphyromonas* genus in centenarians were observed in a study of 367 Japanese volunteers[10] . Furthermore, Kim et al.[19] identified a minor relative abundance of *Faecalibacterium* and *Prevotella*, as well as an increase of *Escherichia* and *Proteobacteria* in centenarians of South Korea. Additionally, Kong et al.[20] , considering the results of their own Chinese cohort and the results previously reported by Biagi et al.[21], identified an enrichment of *Clostridium* cluster XIVa, *Akkermansia*, *Ruminococcaceae* and *Christensenellaceae* in long-living groups. While many genera of *Clostridium* cluster XIVa are producers of SCFAs, *Akkermansia* and *Chistensenellaceae* have been identified as good metabolic health-related bacteria, associated with healthy homeostasis and

immunomodulation. This suggests a tendency in the microbiota profile of centenarians towards a healthy and anti-inflammatory status[19][20]. Moreover, related to SCFAs producers, the microbiota profile of centenarians showed an increase in some butyrate producers (*Anaerotruncus colihominis* and *Eubacterium limosum*) and a decrease in others (*Ruminococcus obeum*, *Roseburia intestinalis*, *E. ventriosum*, *E. rectale*, *E. hallii*, *Papillibacter cinnamovorans* and *Faecalibacterium prausnitzii*), suggesting with these differences the presence of bacteria characteristics of longevity[22]. The association of longevity with *Ruminococcus*, a genus known as a SCFAs producer and with an important role in gut protection, is still contradictory[18]. All of these findings related to gut microbiota composition in elderly and in centenarians are summarized in Table 1.

Table 1. Gut microbiota composition of the elderly (≥60 years old) and centenarians (≥99 years old).

Reference	Subjects	Methodological Approach	Main Findings in Gut Microbiota Composition	
			Elderly	**Centenarians**
[67] Biagi et al. 2010	84 subjects from Northern Italy (50F, 34M). Young adults (20–40 years old) **(Y)**, elderly (60–80 years old) **(E)**, centenarians group (99–104 years old) **(C)** and offspring of the centenarians (59–78 years old) **(F)**	HITChip analysis and qPCR (16S rRNA)	. ↑ *Akkermansia muciniphila*	. ↓ α-diversity index . ↑ facultative anaerobes from *Proteobacteria* phylum (*E. coli, Haemophilus, K. pneumoniae* or *Pseudomonas*) and *Bacilli* class (*Bacillus* or *Staphylococcus*) . ↓ *Clostridium* cluster XIVa . ↓ bifidobacteria . Rearrangement of *Clostridium* cluster IV (↓ *Faecalibacterium prausnitzii* and ↑ *Clostridium leptum*)
[58] Claesson et al. 2011	161 **elderly** Irish subjects (82F, 79M) (>65 years old) and a **control** group (5F, 4M) (28–46 years old)	Pyrosequencing with 454 system (16S rRNA V4 region)	. ↓ *Firmicutes* proportion . ↑ *Clostridium* cluster IV (specially, *Faecalibacterium* spp.) . ↓ *Clostridium* cluster XIVa	
[72] Wang et al. 2015	24 volunteers from China (14F, 10M) classified in **Group RC** (100–108 years old), **Group RE** (85–99 years old) and **Group CE** (80–92 years old)	Illumina MiSeq and qPCR (16S rRNA V4 region)	. ↑ α-diversity in centenarians . ↑ *Escherichia, Roseburia* in centenarians . ↓ *Lactobacillus, Butyricimonas, Coprococcus, Parabacteroides, Akkermansia, Sutterella, Faecalibacterium* in centenarians	

			Elderly	**Centenarians**
[76] Biagi et al. 2016	24 semi-supercentenarians (>105 years old, **group S**) (18F, 6M) vs. 15 young adults (22–48 years old, **group Y**) (8F, 7M) from Northern Italy. Results of **C and E groups** from Biagi et al. 2010 study were incorporated.	Illumina MiSeq and qPCR (16S rRNA V3-V4 region)	. ↓ *Bifidobacterium* . ↓ *Bacteroidaceae, Lachnospiraceae, Ruminococcacea* with age . ↑ *Eggerthella, Bilophila, Akkermansia, Anaerotruncus, Christensenellaceae* and *Synergistaceae* with age	. ↓ *Bifidobacterium* in C . ↑ *Bifidobacterium* in S
[75] Kong et al. 2016	168 Chinese individuals (85F, 83M) grouped into **long-living group** (≥90 years old), **elderly group** (65–83 years old) and a **younger age group** (24–64 years old)	Illumina MiSeq (16S rRNA V3-V4 region)	. ↑ *Clostridium* cluster XIVa, *Ruminococcaceae, Akkermansia* and *Christensenellaceae* in long-living group . ↑ microbial diversity in long-living group	
			Elderly	**Centenarians**
[64] Odamaki et al. 2016	367 Japanese volunteers between 0 and 104 years old (210F, 157M)	Illumina MiSeq and qPCR (16S rRNA V3-V4 region)	-	. ↑ microbial diversity in centenarians
			. ↑ *Bacteroidetes* (*Bacteroides* and *Clostridiaceae*) *and Proteobacteria* (*Betaproteobacteria* and *Deltaproteobacteria*) with age . ↓ *Actinobacteria* with age . ↑ *Porphyromonas, Treponema, Fusobacterium* and *Pseudoramibacter* with age	
			Elderly	**Centenarians**
[68] Bian et al. 2017	A total of 1095 healthy Chinese volunteers (533F, 562M) classified into eight groups according to their age (**children, adults, elderly, centenarians**)	Illumina MiSeq (16S rRNA V4 region)	. ↓ *Blautia* after 60 years old . ↑ *Prevotella* and *Bacteroides* in 60–79 years old group	. ↓ *Bacteroides* and *Bifidobacterium* genera in the oldest groups vs. the youngest groups . ↓ *Prevotella* and *Bacteroides* in centenarians
			. ↑ *Dorea, Clostridium insertae sedis, Clostridium sensu strictu 1, Marvinbryantia* and members of *Prevotella* in older subjects vs. young groups	

[66] Rahayu et al. 2019	80 Indonesian subjects (50F, 30M): **young group** (25–45 years old) and **elderly group** (≥70 years old)	Yakult intestinal flora-scan (YIF-SCAN) (qPCR method)	· ↓ microbiota concentration · ↑ *Lactobacillus reuteri* and *Enterobacteriaceae* · ↓ *Clostridium cocoides, Bacteroides fragilis, Clostridium leptum, Bifidobacterium, Prevotella* and *Lactobacillus plantarum*	
			Elderly	**Centenarians**
[73] Wang et al. 2019	187 elderly subjects from three groups of age (**65–70 years old**), (**90–99 years old**) and (**100+ years old**) from East China (120F, 67M)	Illumina MiSeq (16S rRNA V3, V4 and V5 regions)	· ↑ *Clostridium, Parabacteroides* and *Streptococcus* in 90–99 years old group vs. 65–70 years old group · ↓ *Megamonas, Blautia* and *Coprococcus* in 90–99 years old group vs. 65–70 years old group · ↑ *Bacteroides fragilis, Parabacteroides merdae, Ruminococcus gnavus, Coprococcus* and *Clostridium perfringens* in 90–99 years old group	· ↑ community richness (Ace and Chao1 index) in centenarians (90–99 years old and 100+ years old groups) · ↑ *Ruminococaccaeae, Alistipes and Barnesiella* in 100+ years old group vs. 65–70 years old group · ↓ *Lachnospira and Prevotella* in 100+ years old group vs. 65–70 years old group · ↑ *Synergistetes, Verrucomicrobia* and *Proteobacteria* in longevity group vs. younger elderly group

			Elderly	Centenarians
[74] Kim et al. 2019	56 South Korea subjects classified in **centenarians** (95–108 years old) (27F, 3M), **elderly** (67–79 years old) (7F, 10M) and **adults** (26–43 years old) (3F, 6M)	Pyrosequencing with 454 system (16S rRNA V1–V3 regions)	. ↑ *Proteobacteria* in elderly vs. adults . ↓ *Bacteroidetes* in elderly vs. adults	. ↑ *Verrucomicrobia* in centenarians vs. elderly . ↑ *Proteobacteria, Actinobacteria* and *Verrucomicrobia* in centenarians vs. adults . ↑ *Akkermansia, Clostridium, Collinsella, Escherichia, Streptococcus* and *Christensenellaceae* in centenarians vs. elderly and adults . ↓ *Faecalibacterium* and *Prevotella* in centenarians vs elderly and adults

F: female; M: male. Changes (↑: increase; ↓: decrease) in the relative abundance of selected microbial taxa and in bacterial diversity with age. Names in bold denote each group for the corresponding study and are defined in the table.

In these terms, not only the composition but also the metabolic pathways of microbiota change with age. Collino et al.[23] identified some alterations in a Northern Italian population linked to age, such as low concentrations of tryptophan and lysophospatidylcholines and increased levels of sphingomyelins and phospatidylcholine 32:0. On the other hand, some plasma metabolomic patterns such as lipids and amino acids have been related to health span markers in elderly[24]. Nevertheless, it has been observed that phosphatidylinositol, glycosphingolipid and N-glycan biosynthesis signaling pathways are increased in centenarians, all of them being associated with anti-inflammation and healthy status of gut microbiota[19]. Low levels of markers of lipid peroxidation, as 9-hydroxy-octadecadienoic acid (9-HODE) and 9-oxo-octadecadienoic acid (9-oxoODE), have been identified in longevity phenotype in a population of Italy[23], while centenarians in China showed high levels of SCFAs and total bile acids[25]. These results seem to reinforce the existence of a specific altered microbiota pattern in the elderly with the particularity of a healthy microbiota composition and functionality in centenarians, with more research being necessary to elucidate such patterns.

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