Review

Gut microbiota: A player in aging and a target for anti-aging intervention

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ABSTRACT

Aging-associated alterations in composition, diversity and functional features of intestinal microbiota are well-described in the modern literature. They are suggested to be caused by an age-related decline in immune system functioning (immunosenescence) and a low-grade chronic inflammation (inflammaging), which accompany many aging-associated pathologies. The microbiota-targeted dietary and probiotic interventions have been shown to favorably affect the host health and aging by an enhancement of antioxidant activity, improving immune homeostasis, suppression of chronic inflammation, regulation of fat deposition and metabolism and prevention of insulin resistance. Recently, a high effectiveness and safety of novel therapeutic application such as fecal microbiota transplantation in the prevention and treatment of age-related pathological conditions including atherosclerosis, type 2 diabetes and Parkinson’s disease has been demonstrated. In this review, recent research findings are summarized on the role of gut microbiota in aging processes with emphasis on therapeutic potential of microbiome-targeted interventions in anti-aging medicine.

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1. Introduction

Over the past decades, life expectancy has increased dramatically across the world. This process is, however, not accompanied by a corresponding improvement in health outcomes, thereby causing many socio-economic problems. The noticeable rise in the proportion of older persons in populations of developed countries can likely explain growing interest of the public and professionals in biogerontological research devoted to the development of approaches to healthspan extension.

It is commonly believed that human healthspan is determined by complex interactions between genetic, epigenetic, and environmental (primarily dietary and lifestyle) factors (Govindaraju et al., 2015; Passarino et al., 2016). In recent years, the crucial role of the microbiota inhabiting the gastrointestinal tract in regulating health status and lifespan was demonstrated (Biagi et al., 2016). This bacterial community is collectively referred to as gut microbiota, or microbiome, referring to all the genes these bacteria have. The dependence of health conditions including the protection against pathogens as well as nutritional status and energy expenditure on the composition of intestinal microbial community has been reported in a body of studies (Lakshminarayanan et al., 2014). The microbiota was shown to influence several important physiological and metabolic functions of the host organism, and drives the maturation of the immune response during early development, thereby contributing to life-long homeostasis. A healthy gut microbiome plays a key role in the control of metabolism, resistance to infection and inflammation, preventing against autoimmunity and cancer and also in regulating the brain-gut axis (Konturek et al., 2015). Moreover, gut microbiota was shown to be able to influence the risk of gastrointestinal pathologies such as colorectal cancer, inflammatory bowel disease and irritable bowel syndrome, and some extra-intestinal disorders, including those affecting the liver and the respiratory tract (bronchial asthma, allergy, cystic fibrosis, etc.) (Tojo et al., 2014; Iqbal and Quigley, 2016; Honda and Littman, 2016). The alteration in number and composition of the populations of Bifidobacterium genus, species of which are normal inhabitants of a healthy human intestinal tract, is seemingly the most frequent feature of these diseases, and probiotics containing Bifidobacterium species show the powerful potential to prevent and treat these and other diseases (Tojo et al., 2014; Arboleya et al., 2016). Recent, microbiota-targeted approach was proposed as a promising therapeutic mode in treating age-associated metabolic and neurodegenerative disorders (Bischoff, 2016). In this review, recent evidence that microbiome-targeted interventions can have a therapeutic potential not only for particular age-related diseases, but also for slowing down the aging process per se and for promoting human healthspan and longevity is summarized and discussed.

2. Early-life microbial programming of adult health and disease

There is convincing findings that adverse environmental exposures early in life may ‘program’ the life-long health and disease risk (Hanson, 2015). Over the last years, emerging evidence has been obtained that gut microbiota plays an important role in developmental programming of adult health and disease. The adult-like intestinal microbial community is established over the first 3–5 years of life, and it remains relatively stable during the rest of the life but may be changed depending on lifestyle, diet, bacterial infections, antibiotic or surgical interventions, etc (Rodríguez et al., 2015; Kashtanova et al., 2016). The early microbial colonization of the guts can influence the subsequent symbiotic host-bacterial interactions and affect the occurrence of diseases in later life, such as metabolic disorders including obesity and type 2 diabetes, chronic intestinal inflammation, autoimmune diseases, allergy, irritable bowel syndrome, allergic gastroenteritis and necrotizing enterocolitis (Goulet, 2015; Wallace et al., 2016). This process is referred to as ‘microbial programming’ (Koleva et al., 2015).

3. Age-related changes in the intestinal microbial composition

An important point in this context is that composition of the intestinal microbiota substantially changes with aging and related disease outcomes (Lakshminarayanan et al., 2014). The differences between the gut microbiota in young and old adults are revealed by using both cell culture-dependent and –independent approaches (O'Toole and Jeffery, 2015). Aging-associated alterations in gut physiology (i.e., gastric motility disorders, hypochlorhydria, degenerative changes in enteric nervous system, etc.) have profound effects on the diversity, composition and functional features of intestinal microbiome (Konturek et al., 2015). These age-related differences can appear, in particular, because long-term stimulation of immune system may cause decline in an immune system functioning (“immunosenescence”). With that, a low-grade chronic inflammation (“inflammaging”) occurs, which accompanies many aging-associated diseases, including both gastrointestinal (e.g., Clostridium difficile colitis) and non-gastrointestinal pathological conditions such as atherosclerosis, cachexia, frailty, cancer, fatty liver disease, metabolic syndrome, type 2 diabetes, neurodegenerative diseases etc. (Pérez Martínez et al., 2014; Konturek et al., 2015; Bischoff, 2016). Such inflammatory state might make the host more sensitive to gut bacteria, and vice versa, since alterations in the composition of intestinal microbiota are associated with the progression of various pathological conditions in older adults.

The age-related changes in the gut microbiota composition include a decline in microbiota diversity, a decrease in saccharolytic bacteria and an increase in proteolytic bacteria (Bischoff, 2016), decreased abundance of core (dominant) species and increased abundance of subdominant species (Biagi et al., 2016), an increase of certain Proteobacteria, a reduction of bifidobacterial counts, and also a decrease of the ratio of Firmicutes to Bacteroides (F/B) (Pérez Martínez et al., 2014; Rondanelli et al., 2015). For example, in the vaginally delivered breast-fed infants, the bifidobacteria numbers decrease throughout the life course from 90% of the total colon microbiota following birth to less than 5% in the colon of adult individuals, and these numbers are even smaller in elderly persons or in patients with certain diseases including inflammatory bowel disease, antibiotic-associated diarrhea, irritable bowel syndrome, allergies, obesity, and regressive autism (Rivière et al., 2016).

The pronounced changes in gut microbiota occur through the transition from adulthood to old age. In elderly people (>65 years of age), a reduction in the microbiota diversity and a greater inter-individual variation in microbiota composition have been demonstrated compared to adults (Claesson et al., 2012), with reduced numbers of Bifidobacteria, Firmicutes, Faecalibacterium prausnitzii, Clostridium cluster XIV, Blautia cocoides-Eubacterium rectal and greater presence of Bacteroidetes and Enterobacteriaceae (Rondanelli et al., 2015). It should be, however, noted that data on age-related changes in microbiome composition are different in different populations. A summary of research evidence for aging-related changes in relative composition of the human gut microbiome is presented in Table 1.

The centenarian’s microbiota have shown to be less diverse than in adult persons, and have demonstrated decreased levels of Bifidobacterium, Bacteroides and Enterobacteriaceae, and increased Clostridium spp. levels compared to younger adults (Biagi et al., 2010; Drago et al., 2012). Such aging-associated differences in gut microbiota can not necessarily be caused by aging, but they
might be associated with the general decline in the health status accompanied by malnutrition and rising need for medications, including non-steroidal anti-inflammatory drugs and antibiotics. In discussing recent findings on age-related changes in microbiome diversity, several authors concluded that loss of diversity in the core microbiota groups is rather associated with aging-associated frailty than with chronological age per se (Claesson et al., 2012; Jackson et al., 2016; Jeffery et al., 2016; O’Toole and Jeffery, 2015).

The age-related perturbation in the gut microbiome is shown to be an important determinant of age-associated pathological states, such as chronic inflammation (Rehman, 2012), neurodegeneration (Friedland, 2013), cognitive decline (Magnusson et al., 2015), frailty (Meehan et al., 2015), type 1 and type 2 diabetes (Paun and Danska, 2016), as well as nonalcoholic fatty liver disease and cardiovascular disease (Sanduzi Zamparelli et al., 2016). The intestinal microbiota is also recognized as an important factor modulating the risk of cancer development, supposedly via the action of the specific factors, such as particular toxins, metabolites or microbial structures, as well as induced immune responses, on the process of carcinogenesis (Pope et al., 2017). In addition, the microbial ecology is significantly different between lean and obese subjects (Graham et al., 2015; Saad et al., 2016), e.g., microbiota of obese individuals is characterized by a decrease in Bacteroides and an increase in Firmicutes, and it is more efficient in harvesting food energy than those of normal-weight and lean subjects. Therefore, the gut microbiota composition can be a criterion for the metabolic health (Janssen and Kersten, 2015).

A schematic representation of major age-associated changes in human intestinal microbiota composition is given in Fig. 1.

### 4. The role of gut microbiota in the host aging

The evidence was obtained that microbiome composition may affect the rate of aging (Candela et al., 2014; Saraswati and Sitaraman, 2015). No chronological threshold or age at which the microbiota composition harshly changes has been identified; rather, these changes proceed gradually with time (O’Toole and Jeffery, 2015). The rate of age-related deterioration of a mutualistic interaction between the host and intestinal microbiota is strongly influenced by particular factors such as age-associated alterations in lifestyle, nutrition, frailty and inflammation (Candela et al., 2014).

Certain variants of gut microbiome were shown to be predictors of exceptional human longevity. Biagi et al. (2010) have demonstrated that the microbial composition is highly similar in young adults and seventy-year-old people, but differs significantly from that in the centenarians. In individuals living for over 100 years, the gut microbiota was characterized by a rearrangement in the Firmicutes population and enrichment in Proteobacteria, including the opportunistic pro-inflammatory bacteria (“pathobionts”). Such a compromised microbiota-host homeostasis is associated with an increased inflammatory status in centenarians, as determined by a range of peripheral blood markers of inflammation. The remodeling of the centenarians’ microbiome is also accompanied by a substantial decrease in Faecalibacterium prausnitzii and relatives, which are symbiotic species with pronounced anti-inflammatory activity. Furthermore, Eubacterium limosum and relatives were specifically identified as signature bacteria of the long life, as they were more than 10-fold increased in centenarians. In a more recent phylogenetic microbial analysis of semi-supercenarians (105–109 years of age), in comparison to adults, elderly, and centenarians, a higher prevalence of particular health-associated groups (e.g., Akkermansia, Bifidobacterium, and Christensenellaceae) in semi-supercenarians was observed (Biagi et al., 2016). In another centenarian study, the age-associated trajectory of the human intestinal microbiome was shown to be characterized by loss of genes involved in the short-chain fatty acid synthesis and by a decrease in the saccharolytic potential, while proteolytic functions of intestinal metagenome of elderly people were more abundant than those of younger adults (Rampelli et al., 2013). Such changed functional profile was associated with enrichment in “pathobionts”, i.e. the opportunistic pro-inflammatory bacteria commonly present in the adult gut ecosystem in low numbers. In addition, 116 microbial genes significantly correlating with aging were identified as a signature for long life. In the Park et al. (2015a,b) study, maintaining the gut microbiota, including Faecalibacterium spp. EF402172_s and EF404388_s, and also low lipopolysaccharides levels was shown

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<th>Taxon</th>
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<td><strong>Phyla</strong></td>
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<td>Firmicutes</td>
<td>Increase</td>
<td>Biagi et al. (2010); Claesson et al. (2011); Hayashi et al. (2003); Mueller et al. (2006); Makivuokko et al. (2010)</td>
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<td>Bacteroidetes</td>
<td>Decrease</td>
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<td><strong>Minor groups</strong></td>
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<td>Bacteroides species diversity</td>
<td>Decrease</td>
<td>Claesson et al. (2011); Hopkins et al. (2002); Hopkins and MacFarlane (2002)</td>
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<td>Clostridium abundance</td>
<td>Decrease</td>
<td>Hopkins and MacFarlane (2002)</td>
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<td>Bifidobacterial species diversity</td>
<td>Decrease</td>
<td>Gavini et al. (2001); Hébuterne (2003); Hopkins and MacFarlane (2002); Mueller et al. (2006); Woodmansey et al. (2004)</td>
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<tr>
<td>Bifidobacteria</td>
<td>Decrease</td>
<td>Gavini et al. (2001)</td>
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<td><strong>Enterobacter and Klebsiella species</strong></td>
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<td>Providencia and Proteus species</td>
<td>Increase</td>
<td>Gavini et al. (2001)</td>
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<tr>
<td>Enterobacter and Klebsiella species</td>
<td>Decrease</td>
<td>Gavini et al. (2001)</td>
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<tr>
<td>Faecalibacterium prausnitzii</td>
<td>Increase</td>
<td>Biagi et al. (2010); Mueller et al. (2006)</td>
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<td>Facultative anaerobes: streptococci, staphylococci, enterococci, enterobacteria</td>
<td>Decrease</td>
<td>Gavini et al. (2001); Hébuterne, 2003; Makivuokko et al. (2010); Mariat et al. (2009); Mueller et al. (2006); Rajilic-Stojanovic et al. (2009); Woodmansey et al. (2004)</td>
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Table 1: Summary of findings on aging-related changes in relative composition of the human gut microbial composition at genus and species levels.
to preserve residents' health in village communities with a large number of centenarians.

The molecular mechanisms by which microbiota may favorably affect the host health and aging processes are now intensively investigated in controlled clinical trials using prebiotics and probiotics that were shown to be efficient to prevent particular pathological conditions in elderly populations. Among these mechanisms, the most important is believed to be the suppression of inappropriate chronic inflammation. It includes the decrease of synthesis of proinflammatory cytokines known to be up-regulated in elderly subjects, such as interleukins (ILs)-6, -8 and -10, as well as tumor necrosis factor (TNF), and thereby the increase of levels of activated lymphocytes, natural killer cells, and phagocytic activity (Pérez-Martínez et al., 2014; Lowry et al., 2016). It assumes that direct modulation of intestinal microbial community can reduce inflammatory responses and improve adaptive immune responses, thereby counteracting immunosenescence. Other mechanisms supposedly mediating these effects include degradation of nondigestible carbohydrates, enhancement of antioxidant activity, production of vitamin B and conjugated linoleic acids (Riviére et al., 2016), regulation of host fat deposition and metabolism (Nehra et al., 2016), prevention of insulin resistance (Zhang et al., 2016), and improved maintenance of mucosal barrier integrity and immune homeostasis (Patel and Dupont, 2015). Moreover, these mechanisms include improved gut barrier function, elevated production of short-chain fatty acids, increasing gut peptides important for lipid metabolism and glucose homeostasis, and mimicking some effects of calorie restriction (CR), i.e., a dietary regimen low in calories without malnutrition, and also up-regulation of particular genes involved in xenobiotic metabolism (Keenan et al., 2015). Moreover, gut microbiota could affect the host's aging by modulating its general gene expression profile, and microRNAs may play a crucial role in these processes (Patrignani et al., 2014).

An important point in terms of anti-aging medicine is that cross-feeding interactions take place between the gut Bifidobacteria and butyrate-producing colon bacteria, such as Faecalibacterium prausnitzii and Roseburia (Riviére et al., 2016). The short-chain fatty acid butyrate is an essential metabolite produced in the colon, since it is a preferred energy source for colon epithelial cells, contributing to the maintenance of the gut barrier functions, and demonstrating immunomodulatory, anti-inflammatory and anti-cancer capabilities (Keenan et al., 2015; Riviére et al., 2016). It can also exert its beneficial metabolic effects via prevention of metabolic endotoxemia, enhanced mitochondrial activity, and activation of gut gluconeogenesis (Hartstra et al., 2015). Moreover, butyrate is suggested to play an important role in aging and age-related diseases due to its role in modulating epigenetic processes by inhibition of histone deacetylase activity (Vaiserman and Pasyukova, 2012). Recently, evidence has been obtained on the therapeutic potential of butyrate in immune diseases, cancer and neurological disorders (Falkenberg and Johnstone, 2014), as well as in type 2 diabetes (Khan and Jena, 2015).

Fig. 1. Age-associated changes in human intestinal microbiota composition.
5. Gut microbiota in healthspan-promoting interventions

5.1. Dietary interventions

Alterations in the composition of the gut microbiota are revealed in various healthspan-promoting interventions. In a number of studies, an association has been found between the intestinal microbiota composition and weight loss caused by CR, a most reproducible life-extending strategy now. The F/B ratio, in particular, was consistently found to be increased in obesity and reduced with weight loss–producing CR-based interventions (Mathur and Barlow, 2015). For example, in the study of the surgical and dietary weight loss therapy for obesity, the energy-reabsorbing potential of the gut microbiota, indicated by the F/B ratio, was decreased by CR and increased following laparoscopic sleeve gastrectomy (Damm-Machado et al., 2015). Remarkably, the Firmicutes changes were accompanied by alterations in butyrate–producing bacterial species in both groups. The F/B ratio was also significantly decreased in obese individuals receiving a weight-loss dietary intervention (Remely et al., 2015). The weight gain–causing bacteria can, in turn, induce the expression of genes linked to carbohydrate and lipid metabolism thereby influencing dietary energy harvest (John and Mullin, 2016). In animal models, the CR-induced life extension was accompanied by structural modulation of gut microbiota. For instance, life-long low-fat diet significantly altered the overall structure of the intestinal microbiota in C57BL/6j mice. CR was shown to enrich phylotypes positively correlating with longevity, e.g., the genus Lactobacillus, and to reduce phylotypes negatively associating with lifespan (Zhang et al., 2013). Such CR-induced changes were accompanied by significantly reduced levels of serum lipopolysaccharide-binding protein, suggesting that a structurally balanced architecture of gut microbiota may be established via CR. The authors suggest that CR can cause health benefits for the host through reduction of antigen load from the gut.

Since the supply and conversion of nutrients is highly dependent on the composition of gut microbiota and vice versa (Jonkers, 2016), it can be assumed that certain anti-aging interventions may cause specific variations of gut microbial communities causing chronic CR conditions and by this promoting both healthspan and lifespan.

5.2. Probiotics

The WHO defines probiotics as “live microorganisms which, when administered in adequate amounts, confer a health benefit on the host.” The main mechanisms underlying their favorable effects on the host's health status include improvement of barrier function, immunomodulation and production of neurotransmitters; furthermore, they can influence the host's microbiota and cellular components of the gut–brain axis (Sánchez et al., 2016). Considering that major age-related changes in microbiota composition can be attenuated by probiotics, it is not surprising that probiotics can be successfully applied in elderly individuals for treating variety of gastrointestinal and respiratory diseases (Bischoff, 2016).

Among the microbiota-targeted interventions to improve health status in elderly populations, effects of prebiotics and probiotics, particularly those containing *Bifidobacterium* and *Lactobacillus*, are most investigated and documented in clinical trials (Pérez Martínez et al., 2014; Rondanelli et al., 2015; Saraswati and Sitaraman, 2015). The physiological benefits have been repeatedly found to be associated with consumption of probiotics by healthy people. For example, the probiotic *Lactobacillus rhamnosus* GG ATCC 53103 was shown to be able to promote anti-inflammatory pathways of the resident microorganisms (Eloe-Fadrosh et al., 2015). These benefits are suggested by several authors to be mediated to the promotion of the microbiota homeostasis, rather than to change in its composition (Sanders, 2016).

Oral supplementation with probiotics containing *Bifidobacterium breve* B-3 and *Lactobacillus plantarum* HY7714 has been shown to be effective in preventing skin photo-aging induced by chronic ultraviolet irradiation in a mouse models (Kim et al., 2014; Ra et al., 2014; Satoh et al., 2015), and administration of *Lactobacillus plantarum* HY7714 resulted in a reduction of symptoms of UV-induced skin photo-aging in humans (Lee et al., 2015). Oral administration of *Lactobacillus brevis* OW28 to aged mice resulted in amelioration of aging–related colitis and memory impairments by inhibiting gut microbiota lipopolysaccharide production, p16 expression and activation of NF-κB (Jeong et al., 2016). Moreover, there are strong experimental and clinical evidence that probiotic supplementation may improve metabolic and cardiovascular health status by modulating parameters playing crucial roles in aging processes. Probiotic therapy resulted in lowering of low-density lipoprotein cholesterol levels and improving the low-density/high density lipoprotein ratio, and also in reducing inflammatory mediators, blood glucose levels, blood pressure and body mass index (Thushara et al., 2016).

In experimental studies, probiotics have been also shown to be able to promote longevity. Significant life-extending effects of probiotic supplementation were repeatedly found in nematode *Caenorhabditis elegans*. These pro-longevity effects were accompanied by stimulating the innate immune response signaling (Nakagawa et al., 2016; Kwon et al., 2016), improved resistance to oxidative stress (Grompone et al., 2012), decreased lipofuscin accumulation (Komura et al., 2013) and modulated serotonin signaling (Park et al., 2015a,b). There is also evidence that probiotic treatment can promote longevity in mice, possibly through suppression of chronic low-grade inflammatory processes in the colon (Matsumoto et al., 2011).

Most existing experimental evidence as well as data from clinical trials supports the general view that probiotic use is safe for most populations. However, they should be applied with caution in particular patient groups. Potential risks of probiotic treatment include gastrointestinal side effects, unfavorable metabolic profile, excessive immune stimulation and systemic infections in susceptible individuals, as well as horizontal transfer of genes (Doron and Snyderman, 2015). Moreover, since properties of different probiotic species may be strain-specific, the effects of certain strains of probiotics should not be propagated to others without strong confirmation in separate studies (Boyle et al., 2006). Therefore, further investigation is needed to comprehensive evaluation of the incidence and severity of disadvantageous events linked to probiotic consumption.

6. Fecal microbiota transplantation: gerontological aspects

Dietary and probiotic interventions are currently most useful in modulating the gut microbiota. Fecal microbiota transplantation (FMT), or bacteriotherapy, a transfer of liquid filtrate feces from a healthy donor into the recipient's gastrointestinal tract to treat a particular disease or condition (Young and Hayden, 2016), is a more radical approach to restore the intestinal ecosystem. This procedure is usually performed by colonoscopy and less commonly by nasogastric or nasoduodenal tube, enema, or capsule (Gupta et al., 2016). In 2012, the United States Food and Drug Administration (FDA) classified FMT as a new investigational therapeutic option in treatment of particular diseases (Vyas et al., 2015). Initially, bacteriotherapy was developed as an effective method of treating *Clostridium difficile* infection (CDI), which is a major cause of healthcare-associated diarrhea through perturbation of the normal gastrointestinal microbiome (Rao and Salfdar, 2016). More recently, however, its potential effectiveness and safety has been demonstrated in the prevention and treatment of non-gastrointestinal
pathological conditions including those commonly associated with aging, e.g., atherosclerosis, metabolic syndrome, type 2 diabetes, neurodegenerative disease, etc (Konturek et al., 2015; Choi and Cho, 2016). Major research findings on this topic are described in the subsections below.

6.1. Clostridium difficile infection

CDI is the most common cause of nosocomial diarrhea, disproportionately affecting elderly individuals. Among the risk factors of this infection, the most important are antibiotic exposure and age 65+ (Jump, 2013). The pathogenesis of CDI substantially depends on innate immune system, responsible for the early defense against the tissue inflammation. Therefore, the age-related functional loss in the innate immune system can cause CDI development. In healthy people, gut microbiota can prevent colonization of CDI, thus conferring resistance to CDI. Age-related alterations in microbiota composition may impair this resistance and thereby increase risk of CDI (Shin et al., 2016).

The applicability of FMT in treatment of CDI has been extensively studied in randomized controlled trials. For example, in the study by van Nood et al. (2013), the recurrent CDI patients receiving vancomycin were compared with those patients who received vancomycin followed by bowel lavage and with those received vancomycin followed by both bowel lavage and FMT procedure. Within three months of investigation, the symptoms of CDI resolved in 31% of patients receiving vancomycin alone, in 23% of patients receiving vancomycin and bowel lavage, and in 81% of patients receiving FMT. The research was stopped prematurely since FMT proved to be more than twice as effective in resolving the CDI symptoms as antibiotic therapy alone. In the pilot study by Youngster et al. (2014a), after the single FMT in CDI patients, an overall cure rate achieved was 70%, and, after retreatment, it reached 90%. However, this study had several weaknesses such as absence of the control group non-treated with FMT; moreover, it was unblinded. In a very recent systematic review by Li et al. (2016) on the long-term outcomes of FMT for CDI patients which included 18 observational studies with total of 611 participants, the primary cure rate was estimated at 91.2%, and the overall recurrence rate was 5.5%. Most unfavorable events were shown to be expected, short-term, self-limited and manageable.

6.2. Metabolic disorders

Recent preclinical data indicate a great therapeutic potential of FMT in treatment of metabolic disorders. In the Kulecka et al. (2016) study, long-term high-fat diet-fed obese mice supplemented with feces from lean mice through the fecal-oral route demonstrated altered diversity and richness of bacterial species, as well as improving the symptoms of their metabolic syndrome compared to obese mice, non-supplemented with feces from lean mice. In the elegant research by Ridaura et al. (2013), the diet of germ-free mice was supplemented with feces from human female twin pairs discordant for obesity. Supplementation with fecal microbiota from the obese twins led to 20% higher adiposity rate than supplementation with bacteria from the lean twins. These differences in the composition of the body were negatively correlated with levels of fermentation of short-chain fatty acids and positively correlated with metabolism of branched chain amino acids. Since mice are coprophagic, the authors placed animals from different experimental groups in the same cages, and it prevented the development of obesity-associated metabolic phenotypes in mice supplemented with fecal samples from obese-twin donors.

In the preliminary study of effectiveness of FMT in humans, the transfer of gut microbiota from lean donors to persons with metabolic syndrome was implemented (Vrieze et al., 2012). FMT beneficially affected the microbiota composition in recipients, as manifested by increased amounts of butyrate-producing bacteria along with improved insulin sensitivity six weeks after the FMT procedure. Interestingly, not all lean donors exerted similar effects on insulin sensitivity. Several donors demonstrated very significant effects, while others showed no effect. The authors suggest that “super-fecal donor” effects are most likely to occur through high amounts of butyrate-producing bacteria in the donors’ fecal matter (Udayappan et al., 2014).

6.3. Neurodegenerative disorders

Several lines of evidence suggest that gut microbiota is an important part of the microbiota-gut-brain axis including multiple bidirectional systems through which the intestinal microbiota and brain communicate, encompassing hormonal (HPA axis), neural (vagus nerve) and immune pathways (Dinan and Cryan, 2016; Prendergill et al., 2015). Aging-associated alterations in the composition of the gut microbiota are likely contributed to immunosenescence and to the development of a pro-inflammatory phenotype (inflamm-aging). Inflamm-aging, in turn, can significantly impair brain function by an increase in inflammatory cytokine expression and elevated oxidative stress, blood-brain barrier breakdown, peripheral immune cell infiltration, and glial cell activation. These processes may likely contribute to aging-related cognitive impairments and behavioral changes, such as anxiety, depression and reduced sociability.

The age-related changes in microbiota composition have been shown to play a role in development of neurodegenerative disorders, including Parkinson’s disease (PD), affecting 1–2% of the population over age 65+. This disease is characterized by the accumulation of misfolded α-synuclein (αSyn) affecting all aspects of the brain-gut axis functioning including the central, autonomic, and enteric nervous systems (Mulak and Bonaz, 2015). Since brain-gut axis interactions can be significantly affected by the intestinal microbiota through neuroendocrine, immunological and direct neural mechanisms, dysregulation of the brain-gut-microbiota axis in PD can cause gastrointestinal manifestations often preceding motor symptoms, and also may be involved in the pathogenesis of this disease itself. Remarkably, αSyn aggregates, known to be substantially implicated in the pathogenesis of PD, are found in both myenteric and submucosal plexuses of the enteric nervous system before they appear in the brain, indicating that these proteinopathies may spread from gut to brain in a ‘prion-like’ manner (Felice et al., 2016). Since complaints of constipation in patients with PD begin approximately ten years before the start of the impairment of motor functions and since these constipation complaints can be treated with antibiotics, it is assumed that this disease originates in the gut (Xu et al., 2015). In a recent research with mice overexpressing αSyn, convincing evidence was obtained that gut microbiota is key player in motor deficits, microglia activation, and αSyn pathology (Sampson et al., 2016). In this study, antibiotic supplementation ameliorated, whereas microbial re-colonization promoted pathological conditions in adult animals, and oral administration of specific microbial metabolites to germ-free mice promoted the motor symptoms and neuroinflammation. Remarkably, when αSyn-overexpressing mice were mated with microbiota from patients with PD, it resulted in enhanced physical impairment in comparison with those caused by microbiota transplants from healthy donors. These findings suggest that change in the microbiome represents an important risk factor for PD. Improvements in symptoms of PD in patients receiving FMT has been described in one case report only until now (Ananthaswamy, 2011). Therefore, further research is needed to assess the effectiveness of FMT in patients with this pathology.
In numerous studies, a key role of gut microbiota in the development of another neurodegenerative disorder, Alzheimer’s disease (AD), has been demonstrated. AD is a most common neurodegenerative disorder associates with cerebral accumulation of amyloid-β fibrils driving neuroinflammation and neurodegeneration and resulting in gradual memory loss and severe impairment of mental and behavioral functioning (for recent reviews, see Hu et al., 2016; Pistollato et al., 2016). The bacterial species residing in the intestine have been shown to release substantial amounts of amyloids and lipopolysaccharides, thereby promoting the production of proinflammatory cytokines and modulating signaling pathways involved in pathogenesis of AD (Cattaneo et al., 2017; Pistollato et al., 2016). Numerous research findings showed that AD can begin in the gut, and thereby is closely associated with the microbiota imbalance. This suggests that modulation of the intestine microbiome and, consequently, amyloidogenesis via specific dietary interventions and/or FMT can provide a promising therapeutic strategy to prevent or delay the onset of dementia. Such approaches, however, have not been thoroughly investigated until now and could be the subject of future research.

7. Conclusions and perspectives

In the last few years, great progress has been made in understanding the role of the gut microbiota in human healthspan, aging and longevity. Numerious limitations and challenges, however, still exist within this newly emerging field, which require further investigation and improvement. Most of these limitations are related to research methodology generally applied in these studies. Indeed, although there is growing evidence for the relationship between gut microbiota and age-related pathological conditions, one must be cautious in making causal inferences, taking into account that cross-sectional studies commonly used in this field can provide correlative but not causative evidence (Devkota, 2016). Since microbiota composition can be profoundly affected by several confounding factors, the interpretation of these data should be conducted with caution. In particular, the microbiome–drug interactions must definitely be taken into consideration. Indeed, there is increasing evidence that treatment with various drugs including host-targeted medicines and antibiotics may significantly influence the expression levels of microbial genes (Maurice et al., 2013) and, on the contrary, data are obtained that microbiota can affect the bioavailability of medications (Haiser et al., 2013). Other potentially confounding factors are diet (Sandhu et al., 2017), probiotic supplementation (Kristensen et al., 2016), level of physical activity (Cerdá et al., 2016), exposure to psychological stressors (Gur and Bailey, 2016) and host genetics (Dąbrowska and Witkiewicz, 2016). In most cases, however, a proper control of confounding factors and minimization of bias is a very difficult task. For example, it is very difficult to find enough patients who have not undergone medicamentous treatment to provide sufficient statistical power for investigation. In further research, it would be very important to identify the confounding factors and try to control for them.

In discussing the most promising avenues for development and clinical implementation of microbiota-targeted treatment modalities, it may be assumed, based on the data reviewed above, that

Fig. 2. Schematic representation of potential pathways to extend human healthspan by gut microbiota modulation.
FMT, among other microbiome-targeted interventions, have great therapeutic potential in preventing and treating age-related disorders, including metabolic and neurodegenerative pathological conditions. However, despite the high success rates and safety of FMT procedure, its implementation in clinical practice is complicated by several concerns. Among them, there are problems with donor screening, limited viability of fresh stool samples, as well as in their preparation and administration, fears about the pathogen transmission, lack of standardized treatment regimen, and also patients consenting to be treated (Choi and Cho, 2016). To address these issues, novel approaches are being developed, including the use of frozen transplants, which were shown to be nearly as efficient and safe as the fresh ones (Youngster et al., 2014b; Satokari et al., 2015). Another reasonable treatment alternative is the application of stool substitutes, i.e., preparations of multi-species communities of bacteria isolated in pure culture from single donors. For example, in the proof-of-principle study by Petrof et al. (2013), a stool substitute mixture comprising a multi-species bacterial community derived from a single healthy donor was shown to be effective in treating antibiotic-resistant D. difficile colitis. These favorable therapeutic effects were accompanied by pronounced alterations in gut microbial composition and these changes reflected the isolates from the synthetic mixture.

Importantly, several research findings suggest that direct modulation of gut microbiome may be applied not only in treating particular age-related disorders, but also can be promising therapeutic option to combat the aging process per se. For example, in the Xu et al. (2011) study, oral administration of mice with fractions of the purified exopolysaccharides from Bifidobacterium animalis RH that were isolated from the fecal samples of centenarians residing in Bama longevity villages (Guangxi, China) resulted in significantly increased activity of superoxide dismutase, catalase and total antioxidant capability in serum, as well as reduced level of lipofuscin accumulation in mouse brain. Supplementation with intact cells, cell culture supernatants, and intracellular cell-free extracts of Bifidobacterium animalis 01 isolated from centenarians significantly improved several parameters related to aging, such as activity of antioxidative enzymes and scavenging free radicals, and also significantly reduced the lipofuscin level in mice (Shen et al., 2011). Moreover, oral supplementation with live Bifidobacterium strains isolated from healthy Bama centenarians resulted in improvement of parameters indicative of cellular, humoral and nonspecific immune functions (Yang et al., 2009a), and also the immune barrier function in the intestine of mice (Yang et al., 2009b). Thus, isolation of pure cultures of intestinal bacteria associated with human exceptional longevity and development of standardized procedures for FMT treatment with such cultures seems promising to promote healthspan and lifespan in elderly. A schematic overview of potential pathways to extend human healthspan by gut microbiota modulation is given in Fig. 2.

In addition, use of genetically modified microorganisms seems to be very promising because they can produce substances with potential health-promoting and anti-aging properties. For example, supplementation with E. coli Nissle 1917 bacteria therapeutically modified for the synthesis of N-acylphosphatidylethanolamine (a precursor of the N-acylthanolamines belonging to a family of lipid-related signaling molecules synthesized in the small intestine to reduce food intake), was shown to have potential to prevent the metabolic abnormalities, including insulin resistance, adiposity and hepatosteatosis, in mice fed a high-fat diet (Chen et al., 2014). Through bioengineered, it may be possible not only improve beneficial properties of existing bacterial strains, but also create probiotics with fundamentally new properties and functions. Such bioengineering constructs could be composed not only of bacterial components but could also include particular enzymes or elements of regulatory systems derived from foreign-human-sources (Steidler, 2003). Since engineered bacteria can deliver particular proteins, drugs, or gene therapy vectors more precisely than conventional therapeutic agents, using of bioengineered probiotics seems an attractive treatment option, especially if these novel therapeutic strategies will be comprehensively tested and optimized to meet current clinical standards (Paton et al., 2012; Kumar et al., 2016). In such a case, they will have the potential to revolutionize the healthcare system.

Conflict of interests

The authors have no conflict of interests.

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