Review paper

# Does the gut microbiome play a role in osteoarthritis?

Submitted by:
MSc Molecular Medicine course student
at the University of East Anglia
Aizhan Uteubayeva

Supervised by:
Prof Ian Clark

# Table of Contents

## Abstract

Osteoarthritis (OA) is a joint degenerative disease of the synovial joint, leading to chronic pain and loss of functionality. Although the pathoetiology of OA is still unknown, this review approaches to link the main OA risk factors such as age, obesity and low-grade inflammation with gut microbiome. It has been identified that joint tissue remodelling due to metalloproteases, could be activated via two potential routes when changes in gut microbiota occur. First, low intestinal permeability may allow LPS to escape into circulation, where TLR4 activation leads to release of cytokines IL-6, IL-8 and MMP-1. Secondly, dendritic cells capture microbial products leading to increased ATM infiltration of adipose tissue and increased synthesis of IL-6 and TNF-α. Both pathways may lead to increased cartilage degradation in OA. Obesity may contribute to OA progression via increased MMP production by senescent cells that increased due to large loading weight. However, more research is needed to establish the proposed mechanism of interaction in OA.

## Introduction

Osteoarthritis (OA) is a debilitating multifactorial joint disease, the pathoetiology of which is still unknown. The major risk factors of OA include age, obesity and mechanical stress, and a common unifying component such as low-grade inflammation has only been recently identified. An implication of gut microbiome in a variety of diseases has also sparked an interest in its effect in osteoarthrosis. The following narrative review is going to examine the evidence for potential direct and indirect links of gut microbiome contribution to osteoarthritis development through the prism of systematic low-grade inflammation present in major risk factors.

## 1. Osteoarthritis, pathoetiology and treatment

Osteoarthritis (OA) is multifactorial degenerative disease, resulting in diarthrodial joint's cartilage, tissue remodelling, subchondral bone alteration and synovitis (Martel-Pelletier et al., 2016a). A current approximation of 250 million patients diagnosed with osteoarthritis worldwide, predicting further an exponential increase in after the age of 66 years old (Murray *et al.*, 2013; Q. Wu *et al.*, 2010). The disease burden undermines patient's daily quality of life, inflates loss in economic productivity and wages, further complicating payment for medical services (Zhao et al., 2019).

Until the 18$^{th}$ century osteoarthritis could not be phenotypically differentiated from rheumatoid arthritis (Garrod, 1859). Now osteoarthritis is categorized into primary (or idiopathic) structural and symptomatic (trauma, injury etc), with the large prevalence of the latter in patients (Martel-Pelletier et al., 2016a). The 'wear and tear' concept of OA has been revolutionized into broader 'whole joint' multifaceted disease, affecting multiple structural and systemic components (Courties *et al.*, 2015; Ishijima *et al.*, 2014). Primary OA is associated with the major risk factors of OA such as adiposity and increasing age; whereas secondary with joint injury. Other risk factors of OA incidences of OA also gender, genetics and lifestyle (*Figure 1*) (Johnson and Hunter, 2014; Perruccio *et al.*, 2017; Punzi *et al.*, 2016; Rogers *et al.*, 2015; Sellam and Berenbaum, 2013).

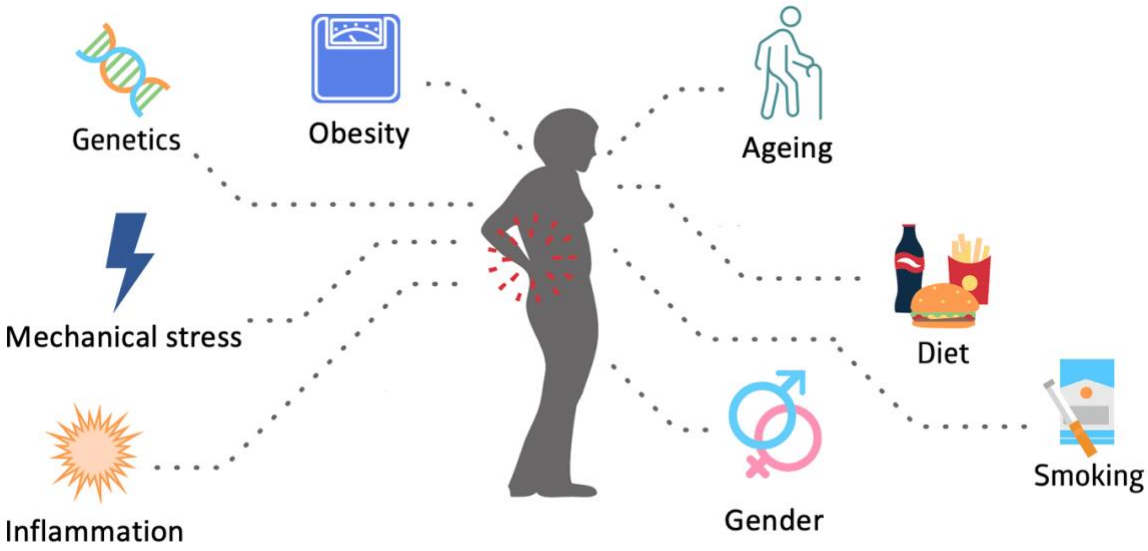


*Figure 1. Schematic representation of risk factors of osteoarthritis: mechanical stress, gender, lifestyle, genetics, ageing and obesity. Adapted from: ("Degenerative Osteoarthritis," n.d.).*

Synovial joints connect two adjoining bones via encapsulating and covering the bone surface by a specialized articular cartilage layer of chondrocytes. The joint structure and composition determine its optimal load-bearing and motile function (*Figure 2*). Adult chondrocytes are normally in quiescent state and maintain cartilaginous extracellular matrix (ECM)(den Hollander *et al.*, 2015; Goldring and Otero, 2011). The articular cartilage ECM consisting of collagen fibres (mainly type II), proteoglycans and non-collagenous proteins provide tensile force and resilience via entrapping water molecules within the extracellular matrix (Andriacchi and Favre, 2014; Quinn *et al.*, 2013). The synovial membrane of the articular capsule is a semipermeable layer that produces synovial fluid for lubrication and low-friction motion. As cartilage has no vasculature or neurons, the synovium plays an important role of cell nurturing, waste removal providing immunomodulating agents of innate and adaptive immune response (Goldring *et al.*, 2011; Scanzello and Goldring, 2012).

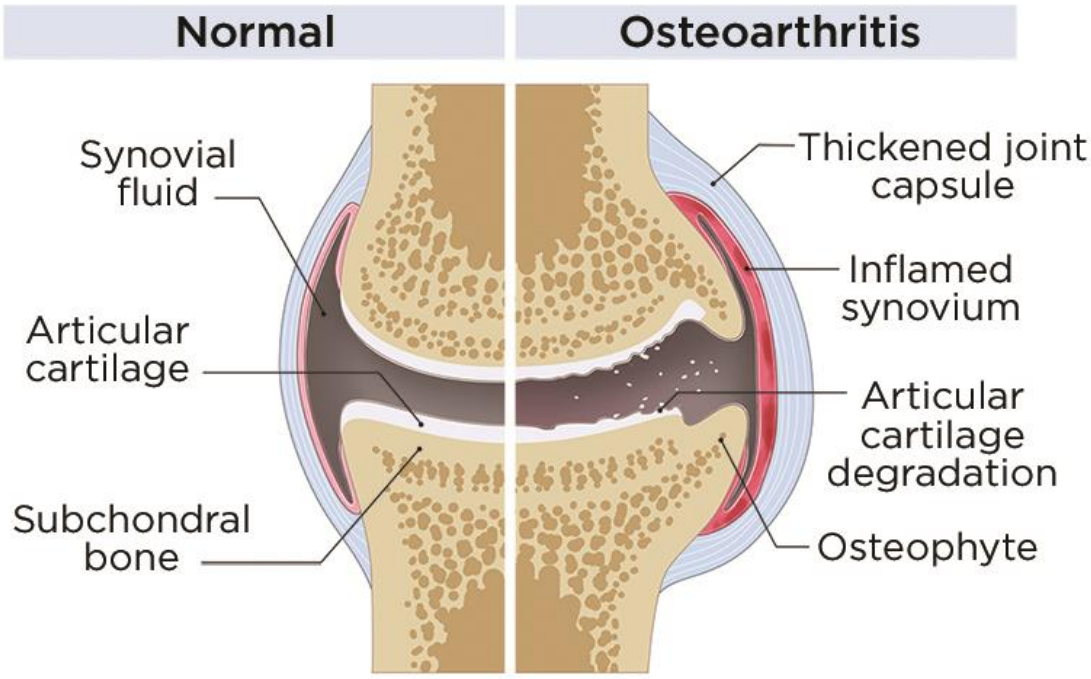


*Figure 2. Schematic representation of normal synovial joint structure versus osteoarthritic joint; (Left) Healthy joint structure featuring articular cartilage, synovial fluid and subchondral bones; (Right)Pathological structure of osteoarthritic joint featuring thickening of joint capsule, inflammation of synovial fluid, articular cartilage degradation and osteophyte formation (Illustration by "Kip Carter," n.d.).*

The primary manifestations of osteoarthritis include joint pain, stiffness and reduced motor function, followed by inflammation, bone and join remodelling (Dell'Isola *et al.*, 2016). The diagnostic tools such as MRI and radiography techniques reveal other pathological changes: degradation of articular cartilage and ligaments, thickening of joint capsule, subchondral bone remodelling, osteophyte formation and synovitis (Loeser *et al.*, 2012a). The disease onset is slow but can evolve gradually or via rapid phases. Therefore, early periodic screenings are recommended for elderly individuals. Investigations into mechanisms of actions of OA are crucial for targeting disease and therefore benefit the public healthcare system.

### 1.1 Pathoetiology:

The pathoetiology of osteoarthritis has long been unclear. It's known to be implicated with both local and systemic immunomodulating factors in clinical manifestations (Martel-Pelletier et al., 2016a). In OA pathological changes involve destruction of articular cartilage, exposure of calcified cartilage and bone, bone remodelling; tidemark duplication which is identified histologically and separates calcified region articular cartilage from non-calcified. As

a result, a new vasculature penetrates from subchondral bone contributing for development of new nerve ends formation possibly contribution for progressive pain (Imhof *et al.*, 2000).

The submolecular layered structure of chondrocytes supports cellular life cycle. The ECM of articular cartilage maintains low turnover of chondrocytes via protecting cells from other non-cartilaginous signalling molecules (Loeser, 2014; Xu *et al.*, 2014). Low-oxygen environment of chondrocytes is maintained via hypoxia-inducible factor-1-α (HIF-1α) (Maes *et al.*, 2012). Primary cilia on the surface of chondrocytes helps to sense mechanical forces and adapt for any changes (Ruhlen and Marberry, 2014). At an early staged of OA, an increased activity of chondrocytes may be due to exposure of cell surface receptors to signals that were previously impenetrable due ECM integrity (Xu et al., 2014). Activated chondrocyte exhibit alterations in tissue order and in cartilage specific gene expression via increased production of proteinases that further deplete collagen and proteoglycan network worsening cartilage erosion (Fosang and Beier, 2011). The collagen *Col2a1* and *aggrecan* genes are downregulated, meanwhile genes coding for metalloproteinases (MMPs), such as *Mmp13*, *Adamts4* and *Adamts5* are upregulated. The family of a disintegrin and metalloproteinase with thrombospondin motifs (ADMATS) include aggrecanases, collagenases and cysteine proteinases, etc (Troeberg and Nagase, 2012). Moreover, activated chondrocytes express vascular endothelial growth factor (VEGF contributing to neoangiogenesis (Walsh *et al.*, 2010). A tight homeostatic balance in cartilage tissue is maintained by regulation of chondrocytes degradation and synthesis. In case of equilibrium failure, catabolism is elevated leading to irreversible tissue destruction.

The main signalling pathways that contributes for upregulation of those genes include nuclear factor kappa B (NF-kB), mitogen-activated protein kinase (MAPK), and wingless-related integration site (Wnt) pathways (Goldring *et al.*, 2011). Among the signalling pathways in OA, Wnt is known to be implicated the most. In the presence of Wnt molecules, it binds to Frizzled and LRP5/6 surface co-receptors, leading to activation of downstream cascade signalling, where β-catenin is not proteosomically degraded but translocated into the nucleus for further gene activation. Wnt pathway related molecules gene expression is altered in OA. The knockout of sFRP3, an antagonist of Wnt, results in increased susceptibility of mice to knee osteoarthritis and the SNPs has been associated with higher risk in osteoarthritis (Thysen *et al.*, 2015). Similarly, the levels of a DKK1, a competitive antagonist of Wnt, are decreased in patients with knee OA (Honsawek *et al.*, 2010). In both spontaneous and induced OA, *Wisp1* gene expression that regulate MMPs and aggrecanases in chondrocytes and macrophages is increased in mice (Blom *et al.*, 2009; Lampropoulou-Adamidou *et al.,* 2014). Activation of β-catenin in *Col2a1* or *Argc1* expressing cells enhances extreme cartilage and

bone remodelling (Wang et al., 2014, 2012). Deletion of both *β-catenin* and *Mmp13* has shown to decrease joint degeneration and pain (Wang *et al.*, 2012). Thus, Wnt signalling holds a potential to be targeted pharmaceutically.

Moreover, bone mineralization and remodelling in OA, include formation of bony outgrowths localized at marginal site called osteophytes (Burr and Gallant, 2012). Osteophytes rather provide bone stability than pathological changes seen in animal models, no correlation between OA progression and osteophytes has been made (Felson *et al.*, 2005; Pottenger *et al.*, 1990; van der Kraan and van den Berg, 2007). An altered distribution of mechanical stress in ligament or menisci injuries may contribute to OA pathology. Chondrocytes cilia or mechanoreceptors sensing change in tensile forces adapt via multiple mechanisms, that yet poorly understood but can be considered as potential therapeutic targets (Quinn *et al.*, 2013).

Recently, new data suggests an implication of extracellular vesicles (ACEV) in osteoarthritis. The vesicles contain biologically active molecules found in the pericellular matrix of articular cartilage and growth plate (Anderson, 1969; Miyaki and Lotz, 2018).These are able to transport regulatory elements like mRNA, microRNA , lipids, DNA and etc (Raposo and Stoorvogel, 2013). It has been identified that chondrocytes internalize ACEV derived mRNA of collagen type 2, aggrecans, transglutaminase in a healthy cartilage (Mitton *et al.*, 2009). Extracellular vesicles may play a bigger role in communication between chondrocytes and tissue cells. These were shown to increase MMP-3 expression levels in synovial fibroblasts, meanwhile decrease expression of ACAN in particular chondrocytes (Kato *et al.*, 2014).

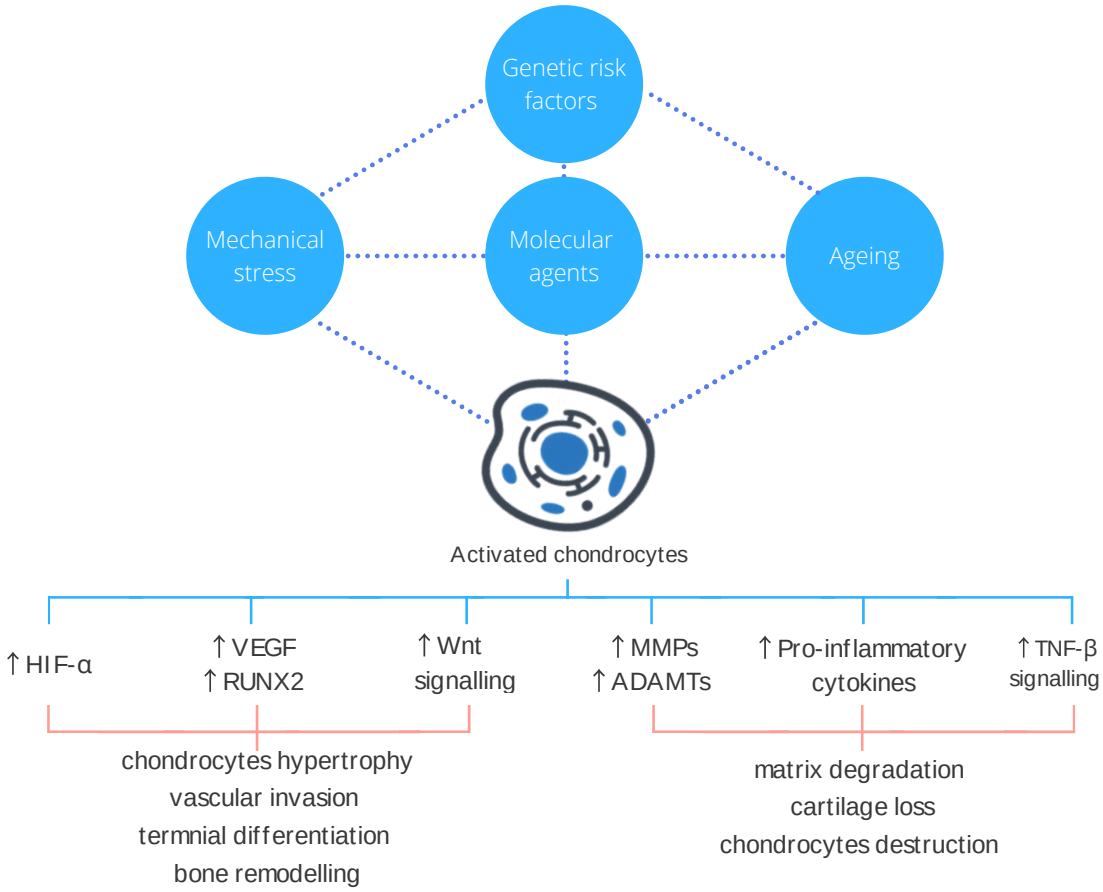


*Figure 3. Schematic representation of pathways and signalling molecules upregulation by activated chondrocytes: hypoxia-induced factor-1-α (HIF-1α), vascular endothelial growth factor (VEGF), Wnt signalling, metalloproteinases (MMPs), ADAMTs, pro-inflammatory cytokines and tumour-necrosis factor β (TNF-β) signalling. The resulting activation leads to chondrocytes hypertrophy, vascular invasion, terminal differentiation of chondrocytes, bone remodelling, matrix degradation, articular cartilage loss and chondrocytes destruction.*

### 1.2 Treatment:

The current treatment of OA mostly includes analgesic or anti-rheumatic medications such as cyclosporin or hydroxychloroquine, that show no improvement but rather symptom palliation (Ghouri and Conaghan, 2019). No osteoarthritis modifying drug has been clinically approved yet (Martel-Pelletier et al., 2016a; Yu and Hunter, 2015). It's a challenge to treat OA with a plethora of risk factors still being recognised and evolving. The current hypothesis as a consequence of evolutionary urbanization in boosting osteoarthritis is due to sedentary lifestyle, high-fat diet and other chronic diseases. Therefore, modifications in lifestyle and nutrition might help with disease alleviation or prevention, as discussed further.

## 2. Gut microbiome in health and disease

Microbiome is a stable-dynamic equilibrium of all commensals, their genes and gene products confined within the human host environment (Hernandez *et al.*, 2016). The morphology of gut microbiota is divided into three large domains of archaea, bacteria and eukaryote. There are an estimated 100 trillion living organisms on the human GI tract. First, the microbiome develops in humans prenatally receiving commensals from mother an then altered according to factors such as mode of delivery, hygiene and age. By the age of three to five years old, a full adult-like microbiome is formed and remains stable until prolonged changes in lifestyle or diet, or an antibiotic treatment (Rodríguez *et al.*, 2015).

A 'halobiont' is a term describing the contributions of the microbiome to its host (Kundu *et al.*, 2017). The total of 3 million protein-coding genes in the microbiome acting as neuronal, immunological and endocrine 'organ' (*Figure 4*)(Clarke *et al.*, 2014; Lazar *et al.*, 2018; Voreades *et al.*, 2014, p. 20). The balance between bacterial niches and the nutritional environment are crucial for disease prevention (Quigley, 2013). The microbiome sensitivity to dietary fat, fibre and food additives has been recently established (Desai *et al.*, 2016; Devkota *et al.*, 2012; J.-Y. Li *et al.*, 2016; Sonnenburg *et al.*, 2016; Thaiss *et al.*, 2014; Wan *et al.*, 2019). Diet can largely influence microbiome due to its quick adaptation to pH and food (David *et al.*, 2014).

A healthy gut microbiome is hard to identify as many factors contribute to the 'normal' state, such as diurnal rhythms, hormonal cycle, travel, immunological profile, diet, genetics and treatment (D'Argenio and Salvatore, 2015; Koren *et al.,* 2012, p. 20; Martin-Belmonte and Perez-Moreno, 2011; Yatsunenko *et al.*, 2012). Although, the alterations of microbiome in disease states suggest its implication and these include: cancer, rheumatoid arthritis, inflammatory bowel disease and Parkinson's disease, type II diabetes, allergic asthma (*Figure 5*) (Cani *et al.*, 2007; J.-Y. Li *et al.*, 2016; Malkki, 2017; Trompette *et al.*, 2014; Wlodarska *et al.*, 2015; Zhang *et al.*, 2015). The concept of microbiome involvement in forementioned conditions is still elusive and active research is needed for its investigation.

Gut microbiome commensals may promote healthy structural, metabolic and signalling functioning. The estimated variation between 1:1 to 10:1 ratio in number of bacterial cells and host cells, suggest the scope of its relative regulatory potential (Sender *et al.*, 2016a, 2016b). The dominant phyla in the healthy gut are *Firmicutes*, *Bacteroidetes* and *Actinobacteria*, the ratio between which varies individually (*Figure 5*) (Qin et al., 2010). However, the specific role that each organism plays within a habitat is difficult to identify. An association of

*Faecalibacterium prausnitzii* with IBD has sceptical views on some evidence (Gerasimidis *et al.*, 2014; Sokol *et al.*, 2009).

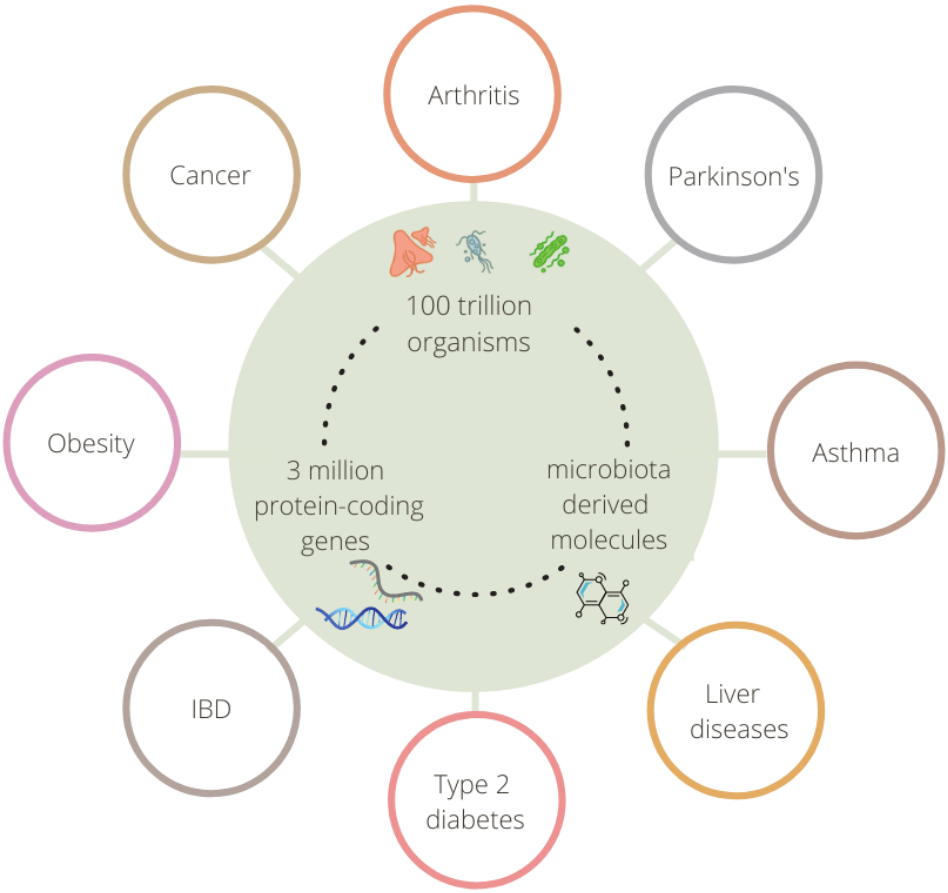


*Figure 4. Schematic representation of links between various diseases and microbiome. Microbiome is estimated to compose of 100 trillion of living organisms, with a total of 3 million protein-coding genes and subsequent microbiota derived molecules. The association of gut microbiome with diseases include irritable bowel disease (IBD), type-2 diabetes, liver diseases, asthma, Parkinson's disease, arthritis, cancer and obesity.*

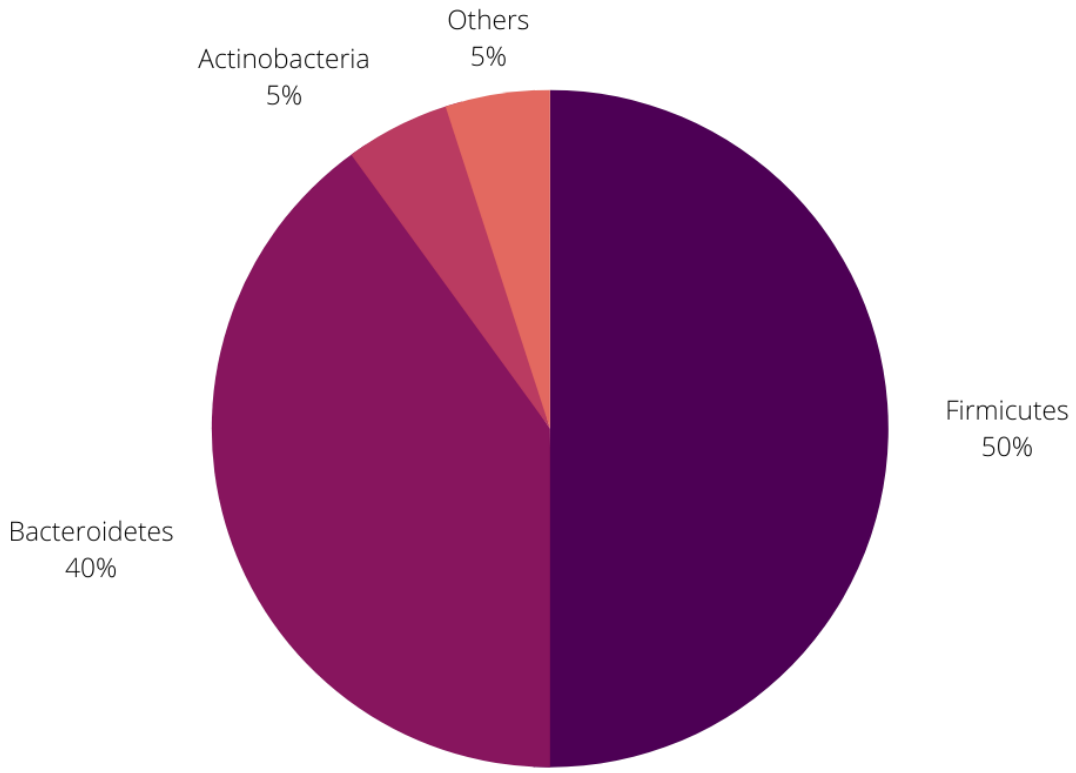


*Figure 5. Schematic pie chart representing approximate percentage population of dominant phyla in normal gut microbiome. From the largest to the smallest enlisted: Firmicutes (50%), Bacteroidetes (40%, Actinobacteria (5%) and others (5%).*

Importantly, that microbiome was found to modulate epithelial development and influence immunological function (Ley *et al.*, 2006; Wang and Li, 2015). First, the suggested mechanisms include: (1) contribution to nutrients and energy harvest (Gill *et al.*, 2006; Turnbaugh *et al.*, 2006); (2) alteration in parasympathetic system promoting insulin, ghrelin levels and hyperphagia due to increased acetate levels derived from dysbiosis (Perry *et al.*, 2016); (3) versatile range of biochemical pathways contributing in fermentation of undigestible carbohydrates and synthesis essential vitamins, such as biotin, cobalamin, vitamin K (Kang *et al.*, 2012; Kau *et al.*, 2011; Roberfroid *et al.*, 1995). For example, Bacteroidetes utilize dietary fibre xyloglucan, meanwhile Lactobacillus and Bifidobacterium use fructo- and oligosaccharides (Goh and Klaenhammer, 2015; Larsbrink *et al.*, 2014). Moreover, short-fatty acids daily produced by commensals can be absorbed by gut epithelium cells regulating inflammatory state, glucose absorption and energy production (Cani *et al.*, 2013; Duncan *et al.*, 2009; Flint *et al.*, 2012).Second, microbiome promotes defence against foreign pathogens via secretion of anti-microbial components (Cash *et al.*, 2006). Finally, microbiota promotes

maturation of intestinal epithelium function. Germ free mice exhibit abnormal mesenteric lymph nodes, spleen, antibody production and T lymphocytes activity (T-helper and T-regulatory cells)(Bouskra *et al.,* 2008; Macpherson and Harris, 2004). Another report showed dual effect of Bacteroidetes and Firmicutes, as these could inhibit anti-inflammatory response of T helper cell (Th-17) via dendritic cells activation, suggesting a tolerogenic effect of gut microbiome on intestinal immune system, and on the other hand promoting inflammation (Magrone and Jirillo, 2013). It is thought that beneficial bacteria Lactobacillus and Bifidobacterium stimulate CD4+ T lymphocyte via polysaccharide A production resulting in increased level of anti-inflammatory IL-10 (Round and Mazmanian, 2009).

Vitamins are essential nutrients that are not host-produced but obtained from the outside sources. Conversely, vitamins exert effect on lowering the population of *Bacteroidetes* and boosting *Lactobacillus*, contributing to overall ratio of *Firmicutes* to *Bacteroidetes* (Conti *et al.*, 2015; Sakaguchi *et al.,* 2011). Systemic lupus erythematosus (SLE), a systemic autoimmune disease symptoms were alleviated by retinoic acid and vitamin A supplementations possible via decreased abundance of *Lachnospiraceae* and increased *Lactobacilli* 14/01/2022 04:29:00. The latter also appeared to boost IL-10 and T-regulatory cells that have anti-inflammatory properties in SLE patients and it's lower abundance is associated with chronic inflammation (Khailova et al., 2013). Other substances, such as omega-3 fatty acid boosts *Lactobacilli* abundance, on the contrary a gluten-free diet alleviates fatigue and myalgias in gluten-sensitive patients with undifferentiated connective tissue disease (Caesar et al., 2015; Conti et al., 2015). Thus suggestion, possible therapeutic implication of diet in reducing inflammation via altering gut microbiome composition.

## 3. Links between major risk factors of osteoarthritis and gut microbiome

Obesity, increasing age and inflammation are major risk factors of osteoarthritis. Both obesity and age have strong correlation with OA, where the approximate risk ratio of being overweight and acquiring OA is 1.9 (Yusuf 2010; Liying Jiang 2016). However, the exact mechanism of OA initiation and progression in still unclear. Recently obesity was observed to affect not only load-bearing joints suggesting a systemic component of the disease (Reyes et al., 2016). A new coined term of increasing inflammation with age called inflammaging, also supports a notion of low-grade inflammation being a unifying component in osteoarthritis. The gut microbiome alterations are also present in aforementioned states, thus it's important to

examine its potential role in implication in age, obesity and low-grade inflammation in causing OA *(Figure 6).* The limitations of studies in this field lack systematic approaches and balanced views, more techniques like preferred reporting items for systematic reviews and meta-analyses (PRISMA) are needed.

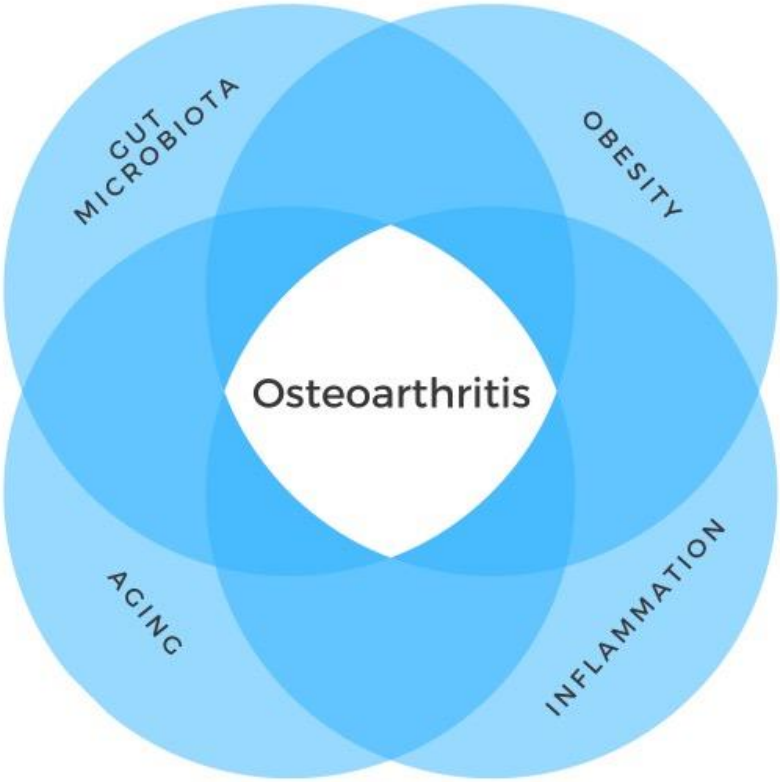


*Figure 6. Schematic representation of a potential cause of osteoarthritis as an overlap between gut microbiota and three main risk factors such as inflammation, obesity and aging.*

### 3.1 Obesity:

Obesity is a worldwide pandemic supposedly caused by reduced overall physical activity and the Western diet (Ogden *et al.*, 2010). Around 66% of osteoarthritis patients are obese or type 2 diabetic (Shi *et al.*, 2006). Interestingly, a systematic review correlates high BMI with osteoarthritis in both non-weight bearing and weight-bearing joints (Yusuf *et al.*, 2010). Germfree mice have developed lower scores of articular cartilage structure degradation than specific pathogen-free mice (Ulici *et al.*, 2018). Thus, suggesting some microbiome-derived systemic players might be involved, even if OA is considered a non-systemic disease.

Gut microbiome alteration has been considered as one of the triggers in adipose tissue associated low-grade systemic inflammation (Winer *et al.*, 2016). Adipokines are pro-inflammatory cytokines released from adipose tissue, the levels of adipokine in vivo has been associated with cartilage degradation and higher frequency of knee replacement in symptomatic OA (Martel-Pelletier *et al.*, 2016b, p. 2015; Stannus *et al.*, 2015).

Liposaccharide (LPS) is a surface-bound endotoxin of Gram-positive bacteria, implicated in a variety of diseases. Obesity can alter gut permeability and immunological profile of intestinal epithelium via gut microbiome alterations (Brun *et al.*, 2007; Cani *et al.*, 2009; Luck *et al.*, 2015). Thus, allowing bacterial LPS to escape into the bloodstream. Indeed, osteoarthritic knee severity is associated with increased levels of serum LPS in patients (Huang *et al.*, 2016). Gram-negative bacterial secrete outer membrane vesicles that can also contain LPS or short-chain fatty acids affecting immune system and permeability accordingly (Brown *et al.*, 2015; Thaiss *et al.*, 2016).

LPS is recognised by toll-like receptors (TLR) via adaptor protein fetuin A, further activating downstream signalling resulting in chemokines secretion (Pal *et al.*, 2012; Shi *et al.*, 2006). The *in vitro* stimulation of dorsal root ganglion cells expressing TLR-4 by LPS showed calcium influx and production of monocyte chemoattractant protein-1 (MCP-1), meanwhile knockout of TLR-4 blocked these responses (Miller *et al.*, 2015). However, a knockout of TLR4 in mice has shown no structural changes in osteoarthritis progression, suggesting that the mechanism of LPS and TLR4 is still unclear (Nasi *et al.*, 2014). Adipose tissue also releases pro-inflammatory markers that promote reactive oxygen species formation and other inflammatory mediators, leading to changes in metabolism and levels of micro-RNAs that regulate NF-kB family (Calçada *et al.*, 2014; Olivieri *et al.,* 2013; Vitale *et al.,* 2013). Thus, suggesting possible activation on chondrocytes and possibly other immune cell via NF-kB pathways in obese patients. Indeed, joint infiltration by macrophages in a rat model of OA with diet-induced-obesity (DIO) correlated with body fat percentage rather than body mass (Collins *et al.*, 2013).

A high-fat diet (HFD), where at least 60% of calories is coming from fat, utilized for experimental purposed in order to mimic Westernized diet, An abnormal bloom of gram-negative bacteria was found in high-fat and low-fibre diet (Dabke *et al*., 2019). The *Lactobacillus Methanobrevibacter* abundance in HFD also correlated with higher Mankin histological scores of osteoarthritic knee and increased serum LPS levels (Collins *et al.*, 2013; Griffin *et al.*, 2012). In a cohort of OA 1444 patients, a *Streptococcus* taxa-specific population was associated with lower hip and knee WOMAC scores, rather than BMI (Boer *et al.*, 2017).

The genetically obese mice exhibited endotoxemia associated with increased gut permeability (Brun *et al.*, 2007). A knockout of β-7 integrin in mice with HFD decreases insulin resistance, the functional regulator of obesity, further showing decrease in intestinal permeability, endotoxemia, adipose tissue inflammation (Luck *et al.*, 2015). Interesting that probiotics supplementation in obese mice improved tight-junction integrity and decreased circulatory LPS (Cani *et al.*, 2009). Thus, obesity-associated gut microbiome alterations may be linked to OA via microbiota contribution to changes in intestinal immune activity and permeability.

Prebiotics are substances that are utilized by microorganisms contributing to beneficial gut functioning exerting a system-wide effect (Gibson *et al.*, 2017). Oligofructose is indigestible by human intestinal tract and is fermented by commensal enzymes (Flint *et al.*, 2012). Schott *et al.*, 2018 in the study showed obesity-associated decreased ratio of Firmicutes to Bacteroidetes, differential expression of colon transcroptome with upregulation genes associated with macrophage activity, increased levels of inflammatory markers (TNF, IL-8, MIP-1B), joint hyperplasia and increased synovial macrophage infiltrations. Moreover, showing oligofructose supplementation of OA mice reversing all of the forementioned conditions via upregulating *Bifidobacterium* and downregulating *Peptococcaceae* species. Prebiotic fibre supplementation, aerobic exercise and combination of both helped to prevent knee joint damage in HFD and high-sucrose rat model (Rios *et al.*, 2019). Indigestible fibre that upregulates the population of *Bifidobacterium* that help fight off advanced glycation end products (AGE) implicated in low-grade chronic inflammation (Kellow *et al.,* 2014). However, some evidence suggests that prebiotics are able to alter intestinal cell signalling directly without modulating gut microflora (Wu *et al.,* 2017).

Although, gut thickness changes with commensal population shifts and microbiota exhibit diurnal oscillations (Thaiss *et al.*, 2014; Zarrinpar *et al.*, 2014). The gene expression of sleep cycle and hepatic clock related proteins are also altered in germ-free free mice during HFD or low-fat diet, suggesting possible mechanism of regulation of natural diurnal rhythms via short-chain fatty-acids (Leone *et al.*, 2015). A new approach of microbiota modulation like faecal transplant has shown to modulate genetic predisposition to obesity (Ussar *et al.,* 2015).

Collectively, the data suggests an ability of obesity-associated gut microbiome changes to modulate immune activity of intestinal epithelium or synovial joint via increased circulatory endotoxins such as LPS; or possibly by immune marker caused by LPS activation in the gut epithelium escaping thought ECM of articular cartilage and activating chondrocytes further promoting inflammation. Moreover, some studies suggesting prebiotic benefits in

maintain balanced gut microflora associated with low-grade inflammation. Therefore, deeper understanding of these mechanisms in warranted.

### 3.2 Inflammation:

Previously osteoarthritis was considered as a 'shear and tear' disease only, but low-grade inflammation was found to be a major contributing factor that unifies OA main risk factors and possibly the gut microbiome. Some research attempted to explain the involvement of microbiota and it's immunomodulating potential, via changing anti-inflammatory and anti-oxidative amino acid profiles or via environmental factors such as smoking, that drive pathologies via microbiome shifts in genetically susceptible patients (Y. Li *et al.*, 2016; Quigley, 2013).

TNF-α, IL-17 are dominant players in regulation of osteoarthritis (Deligne *et al.*, 2015). The Th17m and osteoclastogenic RANKL, IL-17, TNF-α levels failed to rise in germ-free mice with sex steroid deficiency, compared to normal mice gut microbiota with sex steroid deficiency, resulting in no trabecular bone loss (J.-Y. Li *et al.*, 2016). An abundant population of Streptococcus corelated with higher OA knee WOMAC score and knee inflammation in Rotterdam study, further validating results by replication in 867 adults (Boer *et al.*, 2019). One of the OA symptoms, synovitis is characterized by T-cell and B-cell infiltration, increased proteinase activity resulting release of cartilage degradation that may act as damage-associated molecular patterns (DAMPs). DAMPs signalling thought TLR-4 and integrins lead to further pro-inflammatory and catabolic activity (Scanzello *et al.*, 2008; Sohn *et al.*, 2012). In tissue samples of knee OA patients' synovial hypertrophy both in early and late OA exhibited high infiltration of inflammatory markers (CD4+ and CD68+), overexpression of VEGF and IL-1β (Benito *et al*., 2005). Another study has also revealed differential protein composition of the of knee OA patients involved in cartilage turnover, inflammatory and protease inhibition (Sohn *et al*., 2012).

Moreover, lipopolysaccharides present in circulation are also DAMPs that activate macrophages, which both present in cartilage and adipose tissue (Blom *et al*., 2014; Jong *et al*., 2016; Tsuneyoshi *et al.*, 2012). The infiltration of fat cells by adipose tissue macrophages (ATM) expressing tumour necrosis factor α (TNF-α) genes results in release of proinflammatory mediators into the bloodstream. TNF increases macrophages levels in osteoarthritic HFD mice and type 2 diabetic patients (Hamada *et al*., 2016). Thus, suggesting both local and systemic obesity-related inflammation due microbiome components and

alteration. The microbiome can be capable of modulating inflammation via crosstalk between microbiota-derived components and their relative immunoreceptors leading to systemic and local inflammation in osteoarthritis (Portune *et al.*, 2017, Lumeng *et al.*, 2007; Sellam and Berenbaum, 2013).

3.3 Age:

Age is considered as one of the main contributors to osteoarthritis development. The incidences of osteoarthritis symptoms and radiographic images, and in particularly osteophyte formations increase with age (Shane Anderson and Loeser, 2010). Mechanisms such as oxidative stress, muscle mass loss and cartilage thinning were used to bring light onto contribution of age in OA (Musumeci *et al.*, 2015). A few hallmarks of ageing include genome instability, cellular senescence and altered intercellular communication, that can promote chronic low-grade inflammation (O'Toole and Jeffery, 2015)

Differential gene expression of joint tissue between 12-week and 12-month old mice undergoing sham surgery to mimic osteoarthritis compared to uninjured mice has revealed a significant increase in expression of age-related IL-33 and CXCL13, CCL8, and CCL5 chemokines (Loeser *et al.*, 2012b). Older adults have differential composition of microbiome compared to younger population, with the higher abundance of Bacteroides and Clostridium species in former (Claesson *et al.*, 2011; O'Toole and Jeffery, 2015). More specifically, aging reduces bacterial diversity lowering beneficial species such as *Firmicutes*, *Faecalibacterium prausnitzii* and *Bifidobacterium*, meanwhile increasing less beneficial Proteobacteria (Odamaki *et al.*, 2016; Salazar *et al.*, 2013). Alterations in microbiome may affect bioavailability of beneficial components such as SCFAs utilized by gut epithelial cells, and less beneficial LPS. Age-associated reduction in SCFAs may promote abnormal mitochondrial function in skeletal muscle cells leading to irregular oxidation state and insulin resistance (Poggiogalle *et al.*, 2019; Saint-Georges-Chaumet and Edeas, 2016). Decline in physical activity, change in appetite or increased energy consumption due to underlying health conditions has also been associated with physiological GI alterations and commensal variation, all linked with ageing (Lorenzo-López *et al.*, 2017; Morley, 2017). Microbial products may modulate parasympathetic system controlling appetite, such as Escherichia coli associated with increased satiety (Fetissov, 2017; van de Wouw *et al.*, 2017). These imply that age-related malnutrition could possibly give opportunistic microbes to negatively modulate microbiome and brain-related activity further contributing to low-grade inflammation and OA.

Although no systematic study of age-microbiome-osteoarthritis axis has been conducted in human yet, combining knowledge that with age intestinal permeability may decline due higher levels of proinflammatory markers such as IL-6 and TNF-α, the smaller diversity and dominance of Bacteroidetes in older people have potential to contribute to inflammation at larger extent. In fact, age-related intestinal integrity and permeability is predisposed by the gut microbiome dysbiosis in *Drosophila* (Srikanth *et al.*, 2005). TGF-β signals via its receptors on surface on chondrocytes, associated with increased expression of MMP-13 in osteoarthritic human cartilage and loss of tissue integrity (Blaney Davidson *et al.*, 2009; van der Kraan *et al.*, 2017). Age-related TGF-β receptor function in mice contributed to chondrocyte activation and destruction (Blaney Davidson *et al.*, 2009). Although, some evidence suggested supportive properties of TGF- β in cartilage homeostasis linked with lower incidences of OA in ageing animal models TGF-β ta can also contribute to osteophytes formation and chondrocyte hypertrophy (Blaney Davidson *et al.*, 2015; van der Kraan, 2017). Therefore, suggesting a tight balancing mechanism of inflammaging in osteoarthritis progression associated with ageing.

Inflammaging is a new term that coins up both systemic and local states of pro-inflammatory mediators increasing with age. Indeed, higher systemic levels of C-reactive protein (CRP), tumour necrosis factor-α (TNF-α) and interleukin-6 (IL-6) has been associated with age and osteoarthritis of the knee (Bruunsgaard, 2002; Ershler, 1993; Livshits *et al.*, 2009; Strandberg and Tilvis, 2000). A prospective study hold over 5 years revealed that serum TNF, IL-6 and CRP sensitivity are key indicators of knee structural abnormalities and pain in older adults (Stannus *et al*., 2013). Conversely, lower IL-6 levels measured at cross-sectional cohort of 90 years old patients corelated with absence of hip, knee and hand pain (Goekoop *et al.*, 2010). The complexity of inflammaging in potential balancing with anti-inflammatory markers has been reported in inCHIANTI study via Morrisette-Thomas *et al.*, 2014 proinflammatrory mediators such as IL-6, hsCRP and macrophage inflammatory protein-1b (MIP-1b) were more abundant with age; meanwhile anti-inflammatory markers namely IL-1 receptor antagonist (IL-1RA), soluble glycoprotein 130 , and some soluble TNF receptors were also increased. Diet and/or exercises with at least 10% of body weight loss shown to decrease IL-6 levels and OA pain in older adults, also suggesting an implication of adiposity in age-related inflammation (Beavers *et al*., 2013; Messier *et al.*, 2013).

The senescent cells were also found to collect in OA cartilage with age (Campisi, 2011). These cells are associated with the production of cytokines, growth factors and MMPs featuring senescence-associated secretory phenotype (SASP)(Campisi, 2005; Coppé *et al*., 2008). These local age-related pro-inflammatory and enzymatic processes may

contribute to OA development via varoid interleukins(IL-6,IL-8) and metalloproteinases(MMP-1, MMP-10, MMP-3) (Clutterbuck *et al*., 2011; Freund *et al.*, 2010; Sohn *et al.*, 2012; Wang *et al.*, 2013). However, previously senescence thought to occur due joint loading or mechanical stress, rather than proliferation of chondrocytes in an adult articular cartilage (Harbo *et al.*, 2012; Rodier and Campisi, 2011; Rose *et al.*, 2012). Thus, cell senescence may occur rather via increased weight derived from obesity than age. A more sensitive assay is required to identify the degree of the limitation and whether sources are chondrocytes or SASP proteins secretions, or both.

## Discussion

There are overlapping components between age, obesity, inflammation and microbiota in osteoarthrosis. In order to put everything into perspective, possible routes of gut microbiome are suggested, but based on presented research, more evidence is still required (*Figure 7*). An expansion of gram-negative bacteria populations such as *Firmicutes* are seen in obese and/or older adults. This alteration in microbiota results in potentially two routes: (1) LPS escapes into circulation via increased intestinal permeability, further binding to TLR4 and via NF-kB pathways increases production of chemokines that stimulate synthesis of IL-6, IL8 and MMP-1 in cartilage; (2a) dendritic cells take up bacterial antigens to adipose tissue resulting in increased infiltration of adipose tissues macrophage, that increases TNF-α secretion; (2b) As adipose tissue secretes pro-inflammatory markers, including mRNA that regulate NF-kB pathways cell, these suggest a multitude synthesis of circulatory IL-6. Meanwhile, TNF-α was found to be implicated in obese to patients to stimulate chondrocytes to express MMP-1, MMP13 and ADAMTS, older OA patients have also increased circulatory TNF-α and IL-6. There is no mechanism established for inflammaging, perhaps gut microbiome fills the missing link via the second route. A large BMI in overweight patients may exert enough of a loading weight on chondrocytes to increase number of senescent cells, further leading to expression of senescent-associated secretory proteins (SASP) that include metalloproteases MMP-1, MMP-3 and IL-8.

A *Streptococcus* abundance was identified in both SLE patients and OA mice. The SLE patients' faecal examples exhibited increased Th17 differentiation, meanwhile germ-free and sex steroid deficient murine models failed to increase Th17 and TNF-α, suggesting a possible link between *Streptococcus* and Th17 cells. Moreover, some evidence of

associations between Bifidobacterium and lower serum TNF-α and LPS, or *Lactobacillus* and increased pro-inflammatory markers, imply some role of gut microflora in OA progression.

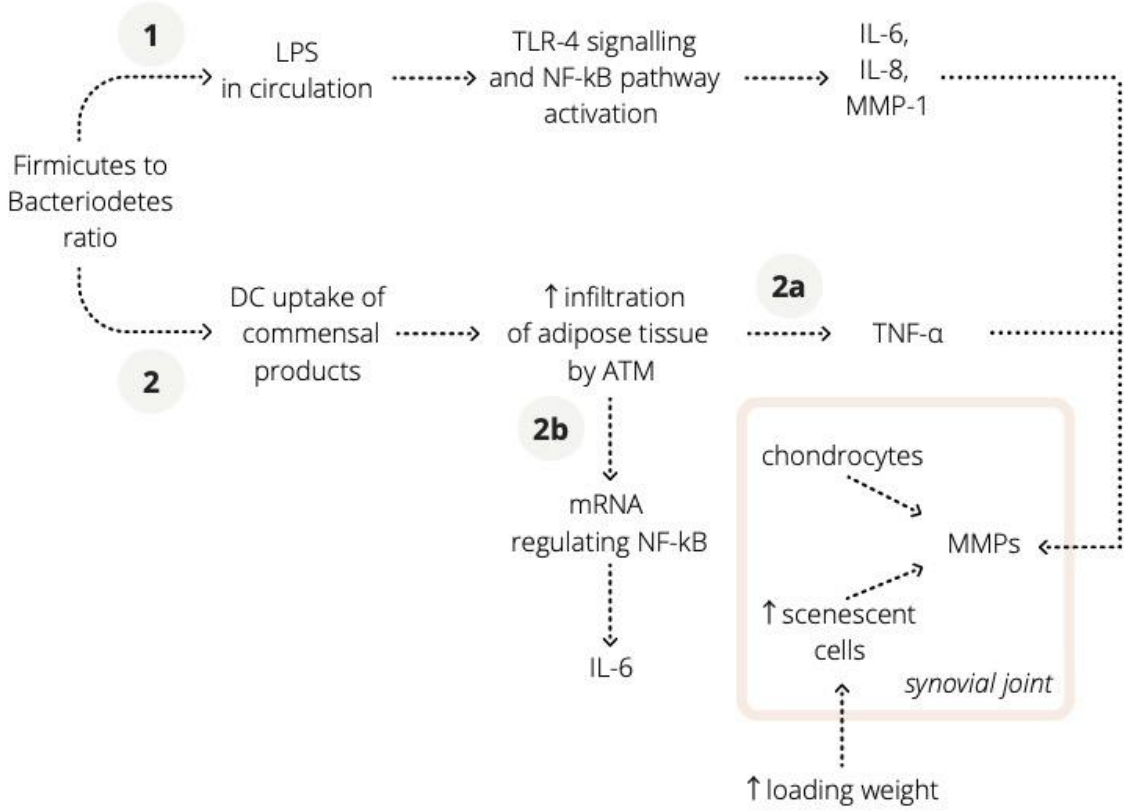


*Figure 7. The microbiota alteration, namely firmicute to Bacteroidetes ration results in two potential routes of action: (1) LSP escapes intro circulation via increased intestinal permeability, further binding to TLR4 and via NF-kB pathways increases production of chemokines that stimulate synthesis of IL-6, IL8 and MMP-1 in cartilage; (2) dendritic cells take up bacterial antigens to adipose tissue resulting in (2a) increased infiltration of adipose tissues macrophage, that increases TNF-α secretion; (2b) As adipose tissue secretes pro-inflammatory markers, including mRNA that regulate NF-kB pathways cell, these suggest a multitude synthesis of circulatory IL-6. Meanwhile, TNF-α was found to be implicated in obese to patients to stimulate chondrocytes to express MMP-1, MMP13 and ADAMTS. Older OA patients have also increased circulatory TNF-α and IL-6. There is no mechanism established for inflammaging, so it could be that gut microbiome fills the missing link via the second route. A large BMI in overweight patients may exerts enough of a loading weight on chondrocytes to increase number of senescent cells, further leading to expression of senescent-associated secretory proteins (SASP) that include metalloproteases MMP-1, MMP-3 and IL-8.*

## Conclusion

Overall, gut microbiome may play role in OA factors obesity, inflammation and ageing via commonly observed agents (*Figure 8*). The general mechanism involves co-interaction between age, inflammation and obesity in osteoarthritis with gut microbiota, that first leads to changes in its composition or metabolites. Secondly, commensal products or components translocate into the circulation where immune system is activated. The bloodstream may bring either metabolites or pro-inflammatory agents into bone marrow or joint synovium, further leading to joint tissue remodelling, synovial inflammation and osteophyte formation.

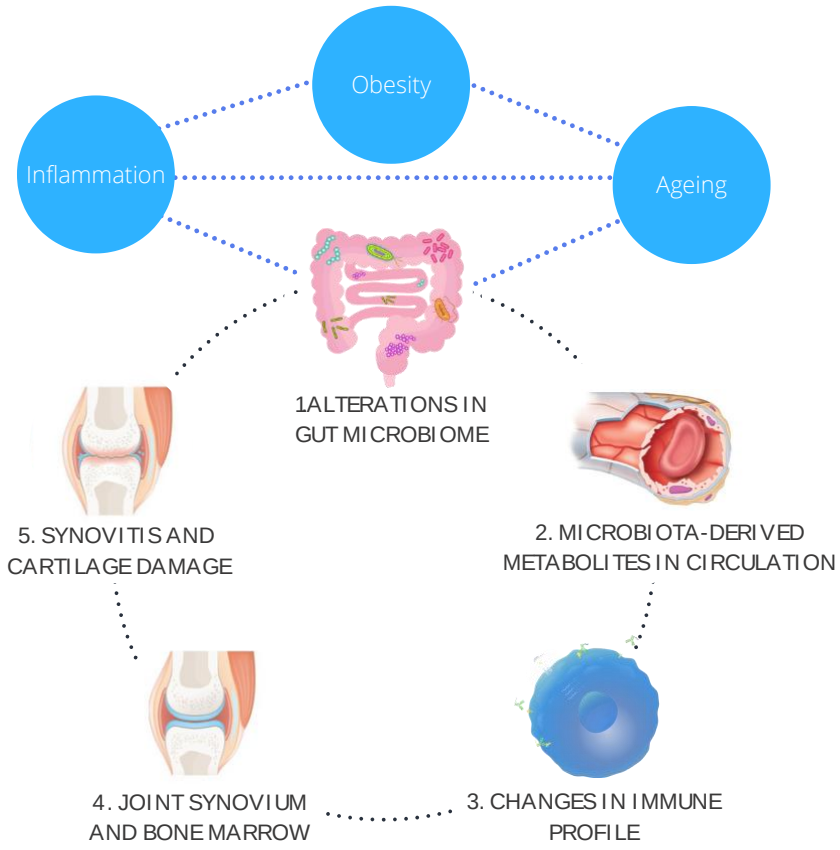


*Figure 8. The schematic diagram of potential mechanism of gut microbiome role in osteoarthritis progression via implication in inflammation, obesity and/or ageing. Three major risk factors of OA (inflammation, obesity and ageing) co-influence each other and eventually alter gut microbiome, leading to changes in immunological profile, that reaches bone marrow and joint synovial fluid, thus promoting synovitis, cartilage damage and other OA pathological features.*

Further research could aim to identify a patient's unique gut microbiota composition and suggest personalised treatments in chronic disease prevention or symptoms alleviation. The link between OA and microbiome requires more patient-based studies. For example, observe groups of non-overweight and overweight OA patients in order to compare their fat mass with serum TNF-α and examine whether there are other sources rather than adipose tissue macrophages that may contribute to TNF-α levels via paired t-test.

## References:


Anderson, H.C., 1969. Vesicles associated with calcification in the matrix of epiphyseal cartilage. J Cell Biol 41, 59–72. https://doi.org/10.1083/jcb.41.1.59

Andriacchi, T.P., Favre, J., 2014. The nature of in vivo mechanical signals that influence cartilage health and progression to knee osteoarthritis. Curr Rheumatol Rep 16, 463. https://doi.org/10.1007/s11926-014-0463-2

Beavers, K.M., Hsu, F.-C., Houston, D.K., Beavers, D.P., Harris, T.B., Hue, T.F., Kim, L.J., Koster, A., Penninx, B.W., Simonsick, E.M., Strotmeyer, E.S., Kritchevsky, S.B., Nicklas, B.J., Health ABC Study, 2013. The role of metabolic syndrome, adiposity, and inflammation in physical performance in the Health ABC Study. J Gerontol A Biol Sci Med Sci 68, 617–623. https://doi.org/10.1093/gerona/gls213

Benito, M.J., Veale, D.J., FitzGerald, O., van den Berg, W.B., Bresnihan, B., 2005. Synovial tissue inflammation in early and late osteoarthritis. Ann Rheum Dis 64, 1263–1267. https://doi.org/10.1136/ard.2004.025270

Blaney Davidson, E.N., Remst, D.F.G., Vitters, E.L., van Beuningen, H.M., Blom, A.B., Goumans, M.-J., van den Berg, W.B., van der Kraan, P.M., 2009. Increase in ALK1/ALK5 ratio as a cause for elevated MMP-13 expression in osteoarthritis in humans and mice. J Immunol 182, 7937–7945. https://doi.org/10.4049/jimmunol.0803991

Blaney Davidson, E.N., van Caam, A.P.M., Vitters, E.L., Bennink, M.B., Thijssen, E., van den Berg, W.B., Koenders, M.I., van Lent, P.L.E.M., van de Loo, F. a. J., van der Kraan, P.M., 2015. TGF-β is a potent inducer of Nerve Growth Factor in articular cartilage via the ALK5-Smad2/3 pathway. Potential role in OA related pain? Osteoarthritis Cartilage 23, 478–486. https://doi.org/10.1016/j.joca.2014.12.005

Blom, A.B., Brockbank, S.M., van Lent, P.L., van Beuningen, H.M., Geurts, J., Takahashi, N., van der Kraan, P.M., van de Loo, F.A., Schreurs, B.W., Clements, K., Newham, P., van den Berg, W.B., 2009. Involvement of the Wnt signaling pathway in experimental and human osteoarthritis: prominent role of Wnt-induced signaling protein 1. Arthritis Rheum 60, 501–512. https://doi.org/10.1002/art.24247

Blom, A.B., Lent, P. van, Bosch, M. van den, Cats, H., Hoogen, F. van den, Lafeber, F., Kraan, P. van der, Berg, W. van den, 2014. Identification of synovial genes and pathways associated with disease progression in a cohort of early osteoarthritis patients (check). Osteoarthritis and Cartilage 22, S23–S24. https://doi.org/10.1016/j.joca.2014.02.067

Boer, C.G., Radjabzadeh, D., Medina-Gomez, C., Garmaeva, S., Schiphof, D., Arp, P., Koet, T., Kurilshikov, A., Fu, J., Ikram, M.A., Bierma-Zeinstra, S., Uitterlinden, A.G., Kraaij, R., Zhernakova, A., van Meurs, J.B.J., 2019. Intestinal microbiome composition and its relation

to joint pain and inflammation. Nat Commun 10, 4881. https://doi.org/10.1038/s41467-019-12873-4

Boer, C.G., Radjabzadeh, D., Uitterlinden, A.G., Kraaij, R., Meurs, J.B. van, 2017. The role of the gut microbiome in osteoarthritis and joint pain. Osteoarthritis and Cartilage 25, S10. https://doi.org/10.1016/j.joca.2017.02.033

Bouskra, D., Brézillon, C., Bérard, M., Werts, C., Varona, R., Boneca, I.G., Eberl, G., 2008. Lymphoid tissue genesis induced by commensals through NOD1 regulates intestinal homeostasis. Nature 456, 507–510. https://doi.org/10.1038/nature07450

Brown, L., Wolf, J.M., Prados-Rosales, R., Casadevall, A., 2015. Through the wall: extracellular vesicles in Gram-positive bacteria, mycobacteria and fungi. Nat Rev Microbiol 13, 620–630. https://doi.org/10.1038/nrmicro3480

Brun, P., Castagliuolo, I., Di Leo, V., Buda, A., Pinzani, M., Palù, G., Martines, D., 2007. Increased intestinal permeability in obese mice: new evidence in the pathogenesis of nonalcoholic steatohepatitis. Am J Physiol Gastrointest Liver Physiol 292, G518-525. https://doi.org/10.1152/ajpgi.00024.2006

Bruunsgaard, H., 2002. Effects of tumor necrosis factor-alpha and interleukin-6 in elderly populations. Eur Cytokine Netw 13, 389–391.

Burr, D.B., Gallant, M.A., 2012. Bone remodelling in osteoarthritis. Nat Rev Rheumatol 8, 665–673. https://doi.org/10.1038/nrrheum.2012.130

Caesar, R., Tremaroli, V., Kovatcheva-Datchary, P., Cani, P.D., Bäckhed, F., 2015. Crosstalk between Gut Microbiota and Dietary Lipids Aggravates WAT Inflammation through TLR Signaling. Cell Metab 22, 658–668. https://doi.org/10.1016/j.cmet.2015.07.026

Calçada, D., Vianello, D., Giampieri, E., Sala, C., Castellani, G., de Graaf, A., Kremer, B., van Ommen, B., Feskens, E., Santoro, A., Franceschi, C., Bouwman, J., 2014. The role of low-grade inflammation and metabolic flexibility in aging and nutritional modulation thereof: a systems biology approach. Mech Ageing Dev 136–137, 138–147. https://doi.org/10.1016/j.mad.2014.01.004

Campisi, J., 2011. Cellular senescence: putting the paradoxes in perspective. Curr Opin Genet Dev 21, 107–112. https://doi.org/10.1016/j.gde.2010.10.005

Campisi, J., 2005. Senescent cells, tumor suppression, and organismal aging: good citizens, bad neighbors. Cell 120, 513–522. https://doi.org/10.1016/j.cell.2005.02.003

Cani, P.D., Amar, J., Iglesias, M.A., Poggi, M., Knauf, C., Bastelica, D., Neyrinck, A.M., Fava, F., Tuohy, K.M., Chabo, C., Waget, A., Delmée, E., Cousin, B., Sulpice, T., Chamontin, B., Ferrières, J., Tanti, J.-F., Gibson, G.R., Casteilla, L., Delzenne, N.M., Alessi, M.C., Burcelin, R., 2007. Metabolic endotoxemia initiates obesity and insulin resistance. Diabetes 56, 1761–1772. https://doi.org/10.2337/db06-1491

Cani, P.D., Everard, A., Duparc, T., 2013. Gut microbiota, enteroendocrine functions and metabolism. Current Opinion in Pharmacology, Gastrointestinal • Endocrine and metabolic diseases 13, 935–940. https://doi.org/10.1016/j.coph.2013.09.008

Cani, P.D., Possemiers, S., Van de Wiele, T., Guiot, Y., Everard, A., Rottier, O., Geurts, L., Naslain, D., Neyrinck, A., Lambert, D.M., Muccioli, G.G., Delzenne, N.M., 2009. Changes in gut microbiota control inflammation in obese mice through a mechanism involving GLP-2-driven improvement of gut permeability. Gut 58, 1091–1103. https://doi.org/10.1136/gut.2008.165886

Cash, H.L., Whitham, C.V., Behrendt, C.L., Hooper, L.V., 2006. Symbiotic Bacteria Direct Expression of an Intestinal Bactericidal Lectin. Science 313, 1126–1130. https://doi.org/10.1126/science.1127119

Claesson, M.J., Cusack, S., O'Sullivan, O., Greene-Diniz, R., de Weerd, H., Flannery, E., Marchesi, J.R., Falush, D., Dinan, T., Fitzgerald, G., Stanton, C., van Sinderen, D., O'Connor, M., Harnedy, N., O'Connor, K., Henry, C., O'Mahony, D., Fitzgerald, A.P., Shanahan, F., Twomey, C., Hill, C., Ross, R.P., O'Toole, P.W., 2011. Composition, variability, and temporal stability of the intestinal microbiota of the elderly. Proc Natl Acad Sci U S A 108 Suppl 1, 4586–4591. https://doi.org/10.1073/pnas.1000097107

Clarke, G., Stilling, R.M., Kennedy, P.J., Stanton, C., Cryan, J.F., Dinan, T.G., 2014. Minireview: Gut microbiota: the neglected endocrine organ. Mol Endocrinol 28, 1221–1238. https://doi.org/10.1210/me.2014-1108

Clutterbuck, A.L., Smith, J.R., Allaway, D., Harris, P., Liddell, S., Mobasheri, A., 2011. High throughput proteomic analysis of the secretome in an explant model of articular cartilage inflammation. J Proteomics 74, 704–715. https://doi.org/10.1016/j.jprot.2011.02.017

Collins, H.M., Abdelghany, M.K., Messmer, M., Yue, B., Deeves, S.E., Kindle, K.B., Mantelingu, K., Aslam, A., Winkler, G.S., Kundu, T.K., Heery, D.M., 2013. Differential effects of garcinol and curcumin on histone and p53 modifications in tumour cells. BMC Cancer 13, 37. https://doi.org/10.1186/1471-2407-13-37

Conti, V., Leone, M.C., Casato, M., Nicoli, M., Granata, G., Carlesimo, M., 2015. High prevalence of gluten sensitivity in a cohort of patients with undifferentiated connective tissue disease. Eur Ann Allergy Clin Immunol 47, 54–57.

Coppé, J.-P., Patil, C.K., Rodier, F., Sun, Y., Muñoz, D.P., Goldstein, J., Nelson, P.S., Desprez, P.-Y., Campisi, J., 2008. Senescence-associated secretory phenotypes reveal cell-nonautonomous functions of oncogenic RAS and the p53 tumor suppressor. PLoS Biol 6, 2853–2868. https://doi.org/10.1371/journal.pbio.0060301

Courties, A., Gualillo, O., Berenbaum, F., Sellam, J., 2015. Metabolic stress-induced joint inflammation and osteoarthritis. Osteoarthritis Cartilage 23, 1955–1965. https://doi.org/10.1016/j.joca.2015.05.016

Dabke, K., Hendrick, G., Devkota, S., 2019. The gut microbiome and metabolic syndrome. J Clin Invest 129, 4050–4057. https://doi.org/10.1172/JCI129194

D'Argenio, V., Salvatore, F., 2015. The role of the gut microbiome in the healthy adult status. Clin Chim Acta 451, 97–102. https://doi.org/10.1016/j.cca.2015.01.003

David, L.A., Maurice, C.F., Carmody, R.N., Gootenberg, D.B., Button, J.E., Wolfe, B.E., Ling, A.V., Devlin, A.S., Varma, Y., Fischbach, M.A., Biddinger, S.B., Dutton, R.J., Turnbaugh, P.J., 2014. Diet rapidly and reproducibly alters the human gut microbiome. Nature 505, 559–563. https://doi.org/10.1038/nature12820

Degenerative Osteoarthritis, n.d. . Singapore Sports and Orthopaedic Clinic. URL https://ssoc.com.sg/degenerative-osteoarthritis/ (accessed 12.14.20).

Deligne, C., Casulli, S., Pigenet, A., Bougault, C., Campillo-Gimenez, L., Nourissat, G., Berenbaum, F., Elbim, C., Houard, X., 2015. Differential expression of interleukin-17 and interleukin-22 in inflamed and non-inflamed synovium from osteoarthritis patients. Osteoarthritis Cartilage 23, 1843–1852. https://doi.org/10.1016/j.joca.2014.12.007

Dell'Isola, A., Allan, R., Smith, S.L., Marreiros, S.S.P., Steultjens, M., 2016. Identification of clinical phenotypes in knee osteoarthritis: a systematic review of the literature. BMC Musculoskelet Disord 17, 425. https://doi.org/10.1186/s12891-016-1286-2

den Hollander, W., Ramos, Y.F.M., Bomer, N., Elzinga, S., van der Breggen, R., Lakenberg, N., de Dijcker, W.J., Suchiman, H.E.D., Duijnisveld, B.J., Houwing-Duistermaat, J.J., Slagboom, P.E., Bos, S.D., Nelissen, R.G.H.H., Meulenbelt, I., 2015. Transcriptional associations of osteoarthritis-mediated loss of epigenetic control in articular cartilage. Arthritis Rheumatol 67, 2108–2116. https://doi.org/10.1002/art.39162

Desai, M.S., Seekatz, A.M., Koropatkin, N.M., Kamada, N., Hickey, C.A., Wolter, M., Pudlo, N.A., Kitamoto, S., Terrapon, N., Muller, A., Young, V.B., Henrissat, B., Wilmes, P., Stappenbeck, T.S., Núñez, G., Martens, E.C., 2016. A Dietary Fiber-Deprived Gut Microbiota Degrades the Colonic Mucus Barrier and Enhances Pathogen Susceptibility. Cell 167, 1339-1353.e21. https://doi.org/10.1016/j.cell.2016.10.043

Devkota, S., Wang, Y., Musch, M.W., Leone, V., Fehlner-Peach, H., Nadimpalli, A., Antonopoulos, D.A., Jabri, B., Chang, E.B., 2012. Dietary-fat-induced taurocholic acid promotes pathobiont expansion and colitis in Il10-/- mice. Nature 487, 104–108. https://doi.org/10.1038/nature11225

Duncan, S.H., Louis, P., Thomson, J.M., Flint, H.J., 2009. The role of pH in determining the species composition of the human colonic microbiota. Environmental Microbiology 11, 2112–2122. https://doi.org/10.1111/j.1462-2920.2009.01931.x

Ershler, W.B., 1993. Interleukin-6: a cytokine for gerontologists. J Am Geriatr Soc 41, 176–181. https://doi.org/10.1111/j.1532-5415.1993.tb02054.x

Felson, D.T., Gale, D.R., Elon Gale, M., Niu, J., Hunter, D.J., Goggins, J., LaValley, M.P., 2005. Osteophytes and progression of knee osteoarthritis. Rheumatology 44, 100–104. https://doi.org/10.1093/rheumatology/keh411

Fetissov, S.O., 2017. Role of the gut microbiota in host appetite control: bacterial growth to animal feeding behaviour. Nat Rev Endocrinol 13, 11–25. https://doi.org/10.1038/nrendo.2016.150

Flint, H.J., Scott, K.P., Duncan, S.H., Louis, P., Forano, E., 2012. Microbial degradation of complex carbohydrates in the gut. Gut Microbes 3, 289–306. https://doi.org/10.4161/gmic.19897

Fosang, A.J., Beier, F., 2011. Emerging Frontiers in cartilage and chondrocyte biology. Best Pract Res Clin Rheumatol 25, 751–766. https://doi.org/10.1016/j.berh.2011.11.010

Freund, A., Orjalo, A.V., Desprez, P.-Y., Campisi, J., 2010. Inflammatory networks during cellular senescence: causes and consequences. Trends Mol Med 16, 238–246. https://doi.org/10.1016/j.molmed.2010.03.003

Garrod, A.B., 1859. The Nature and Treatment of Gout and Rheumatic Gout. Walton and Maberly.

Gerasimidis, K., Bertz, M., Hanske, L., Junick, J., Biskou, O., Aguilera, M., Garrick, V., Russell, R.K., Blaut, M., McGrogan, P., Edwards, C.A., 2014. Decline in presumptively protective gut bacterial species and metabolites are paradoxically associated with disease improvement in pediatric Crohn's disease during enteral nutrition. Inflamm Bowel Dis 20, 861–871. https://doi.org/10.1097/MIB.0000000000000023

Ghouri, A., Conaghan, P.G., 2019. Update on novel pharmacological therapies for osteoarthritis. Ther Adv Musculoskelet Dis 11, 1759720X19864492. https://doi.org/10.1177/1759720X19864492

Gibson, G.R., Hutkins, R., Sanders, M.E., Prescott, S.L., Reimer, R.A., Salminen, S.J., Scott, K., Stanton, C., Swanson, K.S., Cani, P.D., Verbeke, K., Reid, G., 2017. Expert consensus document: The International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of prebiotics. Nat Rev Gastroenterol Hepatol 14, 491–502. https://doi.org/10.1038/nrgastro.2017.75

Gill, S.R., Pop, M., DeBoy, R.T., Eckburg, P.B., Turnbaugh, P.J., Samuel, B.S., Gordon, J.I., Relman, D.A., Fraser-Liggett, C.M., Nelson, K.E., 2006. Metagenomic Analysis of the Human Distal Gut Microbiome. Science 312, 1355–1359. https://doi.org/10.1126/science.1124234

Goekoop, R.J., Kloppenburg, M., Kroon, H.M., Frölich, M., Huizinga, T.W.J., Westendorp, R.G.J., Gussekloo, J., 2010. Low innate production of interleukin-1beta and interleukin-6 is associated with the absence of osteoarthritis in old age. Osteoarthritis Cartilage 18, 942–947. https://doi.org/10.1016/j.joca.2010.03.016

Goh, Y.J., Klaenhammer, T.R., 2015. Genetic Mechanisms of Prebiotic Oligosaccharide Metabolism in Probiotic Microbes. Annual Review of Food Science and Technology 6, 137–156. https://doi.org/10.1146/annurev-food-022814-015706

Goldring, M.B., Otero, M., 2011. Inflammation in osteoarthritis. Curr Opin Rheumatol 23, 471–478. https://doi.org/10.1097/BOR.0b013e328349c2b1

Goldring, M.B., Otero, M., Plumb, D.A., Dragomir, C., Favero, M., El Hachem, K., Hashimoto, K., Roach, H.I., Olivotto, E., Borzì, R.M., Marcu, K.B., 2011. Roles of inflammatory and anabolic cytokines in cartilage metabolism: signals and multiple effectors converge upon MMP-13 regulation in osteoarthritis. Eur Cell Mater 21, 202–220. https://doi.org/10.22203/ecm.v021a16

Griffin, T.M., Huebner, J.L., Kraus, V.B., Yan, Z., Guilak, F., 2012. Induction of osteoarthritis and metabolic inflammation by a very high-fat diet in mice: effects of short-term exercise. Arthritis Rheum 64, 443–453. https://doi.org/10.1002/art.33332

Hamada, D., Maynard, R., Schott, E., Drinkwater, C.J., Ketz, J.P., Kates, S.L., Jonason, J.H., Hilton, M.J., Zuscik, M.J., Mooney, R.A., 2016. Suppressive Effects of Insulin on Tumor Necrosis Factor-Dependent Early Osteoarthritic Changes Associated With Obesity and Type 2 Diabetes Mellitus. Arthritis Rheumatol 68, 1392–1402. https://doi.org/10.1002/art.39561

Harbo, M., Bendix, L., Bay-Jensen, A.-C., Graakjaer, J., Søe, K., Andersen, T.L., Kjaersgaard-Andersen, P., Koelvraa, S., Delaisse, J.-M., 2012. The distribution pattern of critically short telomeres in human osteoarthritic knees. Arthritis Res Ther 14, R12. https://doi.org/10.1186/ar3687

Hernandez, C.J., Guss, J.D., Luna, M., Goldring, S.R., 2016. Links Between the Microbiome and Bone. J Bone Miner Res 31, 1638–1646. https://doi.org/10.1002/jbmr.2887

Honsawek, S., Tanavalee, A., Yuktanandana, P., Ngarmukos, S., Saetan, N., Tantavisut, S., 2010. Dickkopf-1 (Dkk-1) in plasma and synovial fluid is inversely correlated with radiographic severity of knee osteoarthritis patients. BMC Musculoskelet Disord 11, 257. https://doi.org/10.1186/1471-2474-11-257

Huang, Z.Y., Stabler, T., Pei, F.X., Kraus, V.B., 2016. Both systemic and local lipopolysaccharide (LPS) burden are associated with knee OA severity and inflammation. Osteoarthritis Cartilage 24, 1769–1775. https://doi.org/10.1016/j.joca.2016.05.008

Imhof, H., Sulzbacher, I., Grampp, S., Czerny, C., Youssefzadeh, S., Kainberger, F., 2000. Subchondral bone and cartilage disease: a rediscovered functional unit. Invest Radiol 35, 581–588. https://doi.org/10.1097/00004424-200010000-00004

Ishijima, M., Kaneko, H., Kaneko, K., 2014. The evolving role of biomarkers for osteoarthritis. Ther Adv Musculoskelet Dis 6, 144–153. https://doi.org/10.1177/1759720X14541175

Johnson, V.L., Hunter, D.J., 2014. The epidemiology of osteoarthritis. Best Pract Res Clin Rheumatol 28, 5–15. https://doi.org/10.1016/j.berh.2014.01.004

Jong, A.J. de, Lange-Brokaar, B.J. de, Klein-Wieringa, I.R., Heijink, M., Hoekstra, A., Everts, B., Kwekkeboom, J.C., Berkers, C.R., Giera, M., Huizinga, T.W., Kloppenburg, M., Toes, R.E., Ioan-Facsinay, A., 2016. Synovial CD4+ T cells associate with pain in osteoarthritis: is there a role for fatty acids? Osteoarthritis and Cartilage 24, S321. https://doi.org/10.1016/j.joca.2016.01.575

Kang, Z., Zhang, J., Zhou, J., Qi, Q., Du, G., Chen, J., 2012. Recent advances in microbial production of δ-aminolevulinic acid and vitamin B12. Biotechnology Advances, Special issue on ACB 2011 30, 1533–1542. https://doi.org/10.1016/j.biotechadv.2012.04.003

Kato, T., Miyaki, S., Ishitobi, H., Nakamura, Y., Nakasa, T., Lotz, M.K., Ochi, M., 2014. Exosomes from IL-1β stimulated synovial fibroblasts induce osteoarthritic changes in articular chondrocytes. Arthritis Res Ther 16, R163. https://doi.org/10.1186/ar4679

Kau, A.L., Ahern, P.P., Griffin, N.W., Goodman, A.L., Gordon, J.I., 2011. Human nutrition, the gut microbiome and the immune system. Nature 474, 327–336. https://doi.org/10.1038/nature10213

Kellow, N.J., Coughlan, M.T., Savige, G.S., Reid, C.M., 2014. Effect of dietary prebiotic supplementation on advanced glycation, insulin resistance and inflammatory biomarkers in adults with pre-diabetes: a study protocol for a double-blind placebo-controlled randomised crossover clinical trial. BMC Endocr Disord 14, 55. https://doi.org/10.1186/1472-6823-14-55

Khailova, L., Baird, C.H., Rush, A.A., McNamee, E.N., Wischmeyer, P.E., 2013. Lactobacillus rhamnosus GG improves outcome in experimental pseudomonas aeruginosa pneumonia: potential role of regulatory T cells. Shock 40, 496–503. https://doi.org/10.1097/SHK.0000000000000066

Kip [WWW Document], n.d. URL http://vmerc.uga.edu/web/phone/kip.html (accessed 12.14.20).

Koren, O., Goodrich, J.K., Cullender, T.C., Spor, A., Laitinen, K., Bäckhed, H.K., Gonzalez, A., Werner, J.J., Angenent, L.T., Knight, R., Bäckhed, F., Isolauri, E., Salminen, S., Ley, R.E., 2012. Host remodeling of the gut microbiome and metabolic changes during pregnancy. Cell 150, 470–480. https://doi.org/10.1016/j.cell.2012.07.008

Kundu, P., Blacher, E., Elinav, E., Pettersson, S., 2017. Our Gut Microbiome: The Evolving Inner Self. Cell 171, 1481–1493. https://doi.org/10.1016/j.cell.2017.11.024

Lampropoulou-Adamidou, K., Lelovas, P., Karadimas, E.V., Liakou, C., Triantafillopoulos, I.K., Dontas, I., Papaioannou, N.A., 2014. Useful animal models for the research of osteoarthritis. Eur J Orthop Surg Traumatol 24, 263–271. https://doi.org/10.1007/s00590-013-1205-2

Larsbrink, J., Rogers, T.E., Hemsworth, G.R., McKee, L.S., Tauzin, A.S., Spadiut, O., Klinter, S., Pudlo, N.A., Urs, K., Koropatkin, N.M., Creagh, A.L., Haynes, C.A., Kelly, A.G., Cederholm, S.N., Davies, G.J., Martens, E.C., Brumer, H., 2014. A discrete genetic locus confers xyloglucan metabolism in select human gut Bacteroidetes. Nature 506, 498–502. https://doi.org/10.1038/nature12907

Lazar, V., Ditu, L.-M., Pircalabioru, G.G., Gheorghe, I., Curutiu, C., Holban, A.M., Picu, A., Petcu, L., Chifiriuc, M.C., 2018. Aspects of Gut Microbiota and Immune System Interactions in Infectious Diseases, Immunopathology, and Cancer. Front Immunol 9, 1830. https://doi.org/10.3389/fimmu.2018.01830

Leone, V., Gibbons, S.M., Martinez, K., Hutchison, A.L., Huang, E.Y., Cham, C.M., Pierre, J.F., Heneghan, A.F., Nadimpalli, A., Hubert, N., Zale, E., Wang, Y., Huang, Y., Theriault, B., Dinner, A.R., Musch, M.W., Kudsk, K.A., Prendergast, B.J., Gilbert, J.A., Chang, E.B., 2015. Effects of diurnal variation of gut microbes and high-fat feeding on host circadian clock function and metabolism. Cell Host Microbe 17, 681–689. https://doi.org/10.1016/j.chom.2015.03.006

Ley, R.E., Peterson, D.A., Gordon, J.I., 2006. Ecological and Evolutionary Forces Shaping Microbial Diversity in the Human Intestine. Cell 124, 837–848. https://doi.org/10.1016/j.cell.2006.02.017

Li, J.-Y., Chassaing, B., Tyagi, A.M., Vaccaro, C., Luo, T., Adams, J., Darby, T.M., Weitzmann, M.N., Mulle, J.G., Gewirtz, A.T., Jones, R.M., Pacifici, R., 2016. Sex steroid deficiency-associated bone loss is microbiota dependent and prevented by probiotics. J Clin Invest 126, 2049–2063. https://doi.org/10.1172/JCI86062

Li, Y., Xiao, W., Luo, W., Zeng, C., Deng, Z., Ren, W., Wu, G., Lei, G., 2016. Alterations of amino acid metabolism in osteoarthritis: its implications for nutrition and health. Amino Acids 48, 907–914. https://doi.org/10.1007/s00726-015-2168-x

Livshits, G., Zhai, G., Hart, D.J., Kato, B.S., Wang, H., Williams, F.M.K., Spector, T.D., 2009. Interleukin-6 is a significant predictor of radiographic knee osteoarthritis: The Chingford Study. Arthritis Rheum 60, 2037–2045. https://doi.org/10.1002/art.24598

Loeser, R.F., 2014. Integrins and chondrocyte-matrix interactions in articular cartilage. Matrix Biol 39, 11–16. https://doi.org/10.1016/j.matbio.2014.08.007

Loeser, R.F., Goldring, S.R., Scanzello, C.R., Goldring, M.B., 2012a. Osteoarthritis: A Disease of the Joint as an Organ. Arthritis Rheum 64, 1697–1707. https://doi.org/10.1002/art.34453

Loeser, R.F., Olex, A.L., McNulty, M.A., Carlson, C.S., Callahan, M.F., Ferguson, C.M., Chou, J., Leng, X., Fetrow, J.S., 2012b. Microarray analysis reveals age-related differences in gene expression during the development of osteoarthritis in mice. Arthritis Rheum 64, 705–717. https://doi.org/10.1002/art.33388

Lorenzo-López, L., Maseda, A., de Labra, C., Regueiro-Folgueira, L., Rodríguez-Villamil, J.L., Millán-Calenti, J.C., 2017. Nutritional determinants of frailty in older adults: A systematic review. BMC Geriatr 17, 108. https://doi.org/10.1186/s12877-017-0496-2

Luck, H., Tsai, S., Chung, J., Clemente-Casares, X., Ghazarian, M., Revelo, X.S., Lei, H., Luk, C.T., Shi, S.Y., Surendra, A., Copeland, J.K., Ahn, J., Prescott, D., Rasmussen, B.A., Chng, M.H.Y., Engleman, E.G., Girardin, S.E., Lam, T.K.T., Croitoru, K., Dunn, S., Philpott, D.J., Guttman, D.S., Woo, M., Winer, S., Winer, D.A., 2015. Regulation of obesity-related insulin resistance with gut anti-inflammatory agents. Cell Metab 21, 527–542. https://doi.org/10.1016/j.cmet.2015.03.001

Lumeng, C.N., Bodzin, J.L., Saltiel, A.R., 2007. Obesity induces a phenotypic switch in adipose tissue macrophage polarization. J Clin Invest 117, 175–184. https://doi.org/10.1172/JCI29881

Macpherson, A.J., Harris, N.L., 2004. Interactions between commensal intestinal bacteria and the immune system. Nature Reviews Immunology 4, 478–485. https://doi.org/10.1038/nri1373

Maes, C., Araldi, E., Haigh, K., Khatri, R., Van Looveren, R., Giaccia, A.J., Haigh, J.J., Carmeliet, G., Schipani, E., 2012. VEGF-independent cell-autonomous functions of HIF-1α regulating oxygen consumption in fetal cartilage are critical for chondrocyte survival. J Bone Miner Res 27, 596–609. https://doi.org/10.1002/jbmr.1487

Magrone, T., Jirillo, E., 2013. The Interplay between the Gut Immune System and Microbiota in Health and Disease: Nutraceutical Intervention for Restoring Intestinal Homeostasis. Current Pharmaceutical Design 19, 1329–1342. https://doi.org/10.2174/138161213804805793

Malkki, H., 2017. Parkinson disease: Could gut microbiota influence severity of Parkinson disease? Nat Rev Neurol 13, 66–67. https://doi.org/10.1038/nrneurol.2016.195

Martel-Pelletier, J., Barr, A.J., Cicuttini, F.M., Conaghan, P.G., Cooper, C., Goldring, M.B., Goldring, S.R., Jones, G., Teichtahl, A.J., Pelletier, J.-P., 2016a. Osteoarthritis. Nat Rev Dis Primers 2, 16072. https://doi.org/10.1038/nrdp.2016.72

Martel-Pelletier, J., Raynauld, J.-P., Dorais, M., Abram, F., Pelletier, J.-P., 2016b. The levels of the adipokines adipsin and leptin are associated with knee osteoarthritis progression as assessed by MRI and incidence of total knee replacement in symptomatic osteoarthritis patients: a post hoc analysis. Rheumatology 55, 680–688. https://doi.org/10.1093/rheumatology/kev408

Martin-Belmonte, F., Perez-Moreno, M., 2011. Epithelial cell polarity, stem cells and cancer. Nat. Rev. Cancer 12, 23–38. https://doi.org/10.1038/nrc3169

Messier, S.P., Mihalko, S.L., Legault, C., Miller, G.D., Nicklas, B.J., DeVita, P., Beavers, D.P., Hunter, D.J., Lyles, M.F., Eckstein, F., Williamson, J.D., Carr, J.J., Guermazi,

A., Loeser, R.F., 2013. Effects of intensive diet and exercise on knee joint loads, inflammation, and clinical outcomes among overweight and obese adults with knee osteoarthritis: the IDEA randomized clinical trial. JAMA 310, 1263–1273. https://doi.org/10.1001/jama.2013.277669

Miller, R.E., Belmadani, A., Ishihara, S., Tran, P.B., Ren, D., Miller, R.J., Malfait, A.-M., 2015. Damage-associated molecular patterns generated in osteoarthritis directly excite murine nociceptive neurons through Toll-like receptor 4. Arthritis Rheumatol 67, 2933–2943. https://doi.org/10.1002/art.39291

Mitton, E., Gohr, C.M., McNally, M.T., Rosenthal, A.K., 2009. Articular cartilage vesicles contain RNA. Biochem Biophys Res Commun 388, 533–538. https://doi.org/10.1016/j.bbrc.2009.08.038

Miyaki, S., Lotz, M.K., 2018. Extracellular vesicles in cartilage homeostasis and osteoarthritis. Curr Opin Rheumatol 30, 129–135. https://doi.org/10.1097/BOR.0000000000000454

Morley, J.E., 2017. Anorexia of ageing: a key component in the pathogenesis of both sarcopenia and cachexia. J Cachexia Sarcopenia Muscle 8, 523–526. https://doi.org/10.1002/jcsm.12192

Morrisette-Thomas, V., Cohen, A.A., Fülöp, T., Riesco, É., Legault, V., Li, Q., Milot, E., Dusseault-Bélanger, F., Ferrucci, L., 2014. Inflamm-aging does not simply reflect increases in pro-inflammatory markers. Mech Ageing Dev 139, 49–57. https://doi.org/10.1016/j.mad.2014.06.005

Murray, C.J.L., Richards, M.A., Newton, J.N., Fenton, K.A., Anderson, H.R., Atkinson, C., Bennett, D., Bernabé, E., Blencowe, H., Bourne, R., Braithwaite, T., Brayne, C., Bruce, N.G., Brugha, T.S., Burney, P., Dherani, M., Dolk, H., Edmond, K., Ezzati, M., Flaxman, A.D., Fleming, T.D., Freedman, G., Gunnell, D., Hay, R.J., Hutchings, S.J., Ohno, S.L., Lozano, R., Lyons, R.A., Marcenes, W., Naghavi, M., Newton, C.R., Pearce, N., Pope, D., Rushton, L., Salomon, J.A., Shibuya, K., Vos, T., Wang, H., Williams, H.C., Woolf, A.D., Lopez, A.D., Davis, A., 2013. UK health performance: findings of the Global Burden of Disease Study 2010. Lancet 381, 997–1020. https://doi.org/10.1016/S0140-6736(13)60355-4

Musumeci, G., Aiello, F.C., Szychlinska, M.A., Di Rosa, M., Castrogiovanni, P., Mobasheri, A., 2015. Osteoarthritis in the XXIst century: risk factors and behaviours that influence disease onset and progression. Int J Mol Sci 16, 6093–6112. https://doi.org/10.3390/ijms16036093

Nasi, S., Ea, H.-K., Chobaz, V., van Lent, P., Lioté, F., So, A., Busso, N., 2014. Dispensable role of myeloid differentiation primary response gene 88 (MyD88) and MyD88-dependent toll-like receptors (TLRs) in a murine model of osteoarthritis. Joint Bone Spine 81, 320–324. https://doi.org/10.1016/j.jbspin.2014.01.018

Odamaki, T., Kato, K., Sugahara, H., Hashikura, N., Takahashi, S., Xiao, J.-Z., Abe, F., Osawa, R., 2016. Age-related changes in gut microbiota composition from newborn to centenarian: a cross-sectional study. BMC Microbiol 16, 90. https://doi.org/10.1186/s12866-016-0708-5

Ogden, C.L., Carroll, M.D., Curtin, L.R., Lamb, M.M., Flegal, K.M., 2010. Prevalence of high body mass index in US children and adolescents, 2007-2008. JAMA 303, 242–249. https://doi.org/10.1001/jama.2009.2012

Olivieri, F., Rippo, M.R., Monsurrò, V., Salvioli, S., Capri, M., Procopio, A.D., Franceschi, C., 2013. MicroRNAs linking inflamm-aging, cellular senescence and cancer. Ageing Res Rev 12, 1056–1068. https://doi.org/10.1016/j.arr.2013.05.001

O'Toole, P.W., Jeffery, I.B., 2015. Gut microbiota and aging. Science 350, 1214–1215. https://doi.org/10.1126/science.aac8469

Pal, D., Dasgupta, S., Kundu, R., Maitra, S., Das, G., Mukhopadhyay, S., Ray, S., Majumdar, S.S., Bhattacharya, S., 2012. Fetuin-A acts as an endogenous ligand of TLR4 to promote lipid-induced insulin resistance. Nat Med 18, 1279–1285. https://doi.org/10.1038/nm.2851

Perruccio, A.V., Chandran, V., Power, J.D., Kapoor, M., Mahomed, N.N., Gandhi, R., 2017. Systemic inflammation and painful joint burden in osteoarthritis: a matter of sex? Osteoarthritis Cartilage 25, 53–59. https://doi.org/10.1016/j.joca.2016.08.001

Perry, R.J., Peng, L., Barry, N.A., Cline, G.W., Zhang, D., Cardone, R.L., Petersen, K.F., Kibbey, R.G., Goodman, A.L., Shulman, G.I., 2016. Acetate mediates a microbiome–brain–β-cell axis to promote metabolic syndrome. Nature 534, 213–217. https://doi.org/10.1038/nature18309

Poggiogalle, E., Lubrano, C., Gnessi, L., Mariani, S., Di Martino, M., Catalano, C., Lenzi, A., Donini, L.M., 2019. The decline in muscle strength and muscle quality in relation to metabolic derangements in adult women with obesity. Clin Nutr 38, 2430–2435. https://doi.org/10.1016/j.clnu.2019.01.028

Portune, K.J., Benítez-Páez, A., Del Pulgar, E.M.G., Cerrudo, V., Sanz, Y., 2017. Gut microbiota, diet, and obesity-related disorders-The good, the bad, and the future challenges. Mol Nutr Food Res 61. https://doi.org/10.1002/mnfr.201600252

Pottenger, L.A., Phillips, F.M., Draganich, L.F., 1990. The effect of marginal osteophytes on reduction of varus-valgus instability in osteoarthritic knees. Arthritis Rheum 33, 853–858. https://doi.org/10.1002/art.1780330612

Punzi, L., Galozzi, P., Luisetto, R., Favero, M., Ramonda, R., Oliviero, F., Scanu, A., 2016. Post-traumatic arthritis: overview on pathogenic mechanisms and role of inflammation. RMD Open 2, e000279. https://doi.org/10.1136/rmdopen-2016-000279

Qin, J., Li, R., Raes, J., Arumugam, M., Burgdorf, K.S., Manichanh, C., Nielsen, T., Pons, N., Levenez, F., Yamada, T., Mende, D.R., Li, J., Xu, J., Li, Shaochuan, Li, D., Cao, J., Wang, B., Liang, H., Zheng, H., Xie, Y., Tap, J., Lepage, P., Bertalan, M., Batto, J.-M., Hansen, T., Le Paslier, D., Linneberg, A., Nielsen, H.B., Pelletier, E., Renault, P., Sicheritz-Ponten, T., Turner, K., Zhu, H., Yu, C., Li, Shengting, Jian, M., Zhou, Y., Li, Y., Zhang, X., Li, Songgang, Qin, N., Yang, H., Wang, Jian, Brunak, S., Doré, J., Guarner, F., Kristiansen, K., Pedersen, O., Parkhill, J., Weissenbach, J., MetaHIT Consortium, Bork, P., Ehrlich, S.D., Wang, Jun, 2010. A human gut microbial gene catalogue established by metagenomic sequencing. Nature 464, 59–65. https://doi.org/10.1038/nature08821

Quigley, E.M.M., 2013. Gut bacteria in health and disease. Gastroenterol Hepatol (N Y) 9, 560–569.

Quinn, T.M., Häuselmann, H.-J., Shintani, N., Hunziker, E.B., 2013. Cell and matrix morphology in articular cartilage from adult human knee and ankle joints suggests depth-associated adaptations to biomechanical and anatomical roles. Osteoarthritis Cartilage 21, 1904–1912.

Raposo, G., Stoorvogel, W., 2013. Extracellular vesicles: exosomes, microvesicles, and friends. J Cell Biol 200, 373–383. https://doi.org/10.1083/jcb.201211138

Reyes, C., Leyland, K.M., Peat, G., Cooper, C., Arden, N.K., Prieto-Alhambra, D., 2016. Association Between Overweight and Obesity and Risk of Clinically Diagnosed Knee, Hip, and Hand Osteoarthritis: A Population-Based Cohort Study. Arthritis Rheumatol 68, 1869–1875. https://doi.org/10.1002/art.39707

Rios, J.L., Bomhof, M.R., Reimer, R.A., Hart, D.A., Collins, K.H., Herzog, W., 2019. Protective effect of prebiotic and exercise intervention on knee health in a rat model of diet-induced obesity. Sci Rep 9, 3893. https://doi.org/10.1038/s41598-019-40601-x

Roberfroid, M.B., Bornet, F., Bouley, C., Cummings, J.H., 1995. Colonic Microflora: Nutrition and Health. Summary and Conclusions of an International Life Sciences Institute (ILSI) [Europe] Workshop held in Barcelona, Spain. Nutrition Reviews 53, 127–130. https://doi.org/10.1111/j.1753-4887.1995.tb01535.x

Rodier, F., Campisi, J., 2011. Four faces of cellular senescence. J Cell Biol 192, 547–556. https://doi.org/10.1083/jcb.201009094

Rodríguez, J.M., Murphy, K., Stanton, C., Ross, R.P., Kober, O.I., Juge, N., Avershina, E., Rudi, K., Narbad, A., Jenmalm, M.C., Marchesi, J.R., Collado, M.C., 2015. The composition of the gut microbiota throughout life, with an emphasis on early life. Microb Ecol Health Dis 26, 26050. https://doi.org/10.3402/mehd.v26.26050

Rogers, E.L., Reynard, L.N., Loughlin, J., 2015. The role of inflammation-related genes in osteoarthritis. Osteoarthritis Cartilage 23, 1933–1938. https://doi.org/10.1016/j.joca.2015.01.003

Rose, J., Söder, S., Skhirtladze, C., Schmitz, N., Gebhard, P.M., Sesselmann, S., Aigner, T., 2012. DNA damage, discoordinated gene expression and cellular senescence in osteoarthritic chondrocytes. Osteoarthritis Cartilage 20, 1020–1028. https://doi.org/10.1016/j.joca.2012.05.009

Round, J.L., Mazmanian, S.K., 2009. The gut microbiota shapes intestinal immune responses during health and disease. Nature Reviews Immunology 9, 313–323. https://doi.org/10.1038/nri2515

Ruhlen, R., Marberry, K., 2014. The chondrocyte primary cilium. Osteoarthritis Cartilage 22, 1071–1076. https://doi.org/10.1016/j.joca.2014.05.011

Saint-Georges-Chaumet, Y., Edeas, M., 2016. Microbiota-mitochondria inter-talk: consequence for microbiota-host interaction. Pathog Dis 74, ftv096. https://doi.org/10.1093/femspd/ftv096

Sakaguchi, S., Tanaka, S., Tanaka, A., Ito, Y., Maeda, S., Sakaguchi, N., Hashimoto, M., 2011. Thymus, innate immunity and autoimmune arthritis: interplay of gene and environment. FEBS Lett 585, 3633–3639. https://doi.org/10.1016/j.febslet.2011.10.026

Salazar, N., López, P., Valdés, L., Margolles, A., Suárez, A., Patterson, A.M., Cuervo, A., de los Reyes-Gavilán, C.G., Ruas-Madiedo, P., Gonzalez, S., Gueimonde, M., 2013. Microbial targets for the development of functional foods accordingly with nutritional and immune parameters altered in the elderly. J Am Coll Nutr 32, 399–406. https://doi.org/10.1080/07315724.2013.827047

Scanzello, C.R., Goldring, S.R., 2012. The role of synovitis in osteoarthritis pathogenesis. Bone 51, 249–257. https://doi.org/10.1016/j.bone.2012.02.012

Scanzello, C.R., Plaas, A., Crow, M.K., 2008. Innate immune system activation in osteoarthritis: is osteoarthritis a chronic wound? Curr Opin Rheumatol 20, 565–572. https://doi.org/10.1097/BOR.0b013e32830aba34

Schott, E.M., Farnsworth, C.W., Grier, A., Lillis, J.A., Soniwala, S., Dadourian, G.H., Bell, R.D., Doolittle, M.L., Villani, D.A., Awad, H., Ketz, J.P., Kamal, F., Ackert-Bicknell, C., Ashton, J.M., Gill, S.R., Mooney, R.A., Zuscik, M.J., 2018. Targeting the gut microbiome to treat the osteoarthritis of obesity. JCI Insight 3. https://doi.org/10.1172/jci.insight.95997

Sellam, J., Berenbaum, F., 2013. Is osteoarthritis a metabolic disease? Joint Bone Spine 80, 568–573. https://doi.org/10.1016/j.jbspin.2013.09.007

Sender, R., Fuchs, S., Milo, R., 2016a. Are We Really Vastly Outnumbered? Revisiting the Ratio of Bacterial to Host Cells in Humans. Cell 164, 337–340. https://doi.org/10.1016/j.cell.2016.01.013

Sender, R., Fuchs, S., Milo, R., 2016b. Revised Estimates for the Number of Human and Bacteria Cells in the Body. PLoS Biol 14, e1002533. https://doi.org/10.1371/journal.pbio.1002533

Shane Anderson, A., Loeser, R.F., 2010. Why is osteoarthritis an age-related disease? Best Pract Res Clin Rheumatol 24, 15–26. https://doi.org/10.1016/j.berh.2009.08.006

Shi, H., Kokoeva, M.V., Inouye, K., Tzameli, I., Yin, H., Flier, J.S., 2006. TLR4 links innate immunity and fatty acid-induced insulin resistance. J Clin Invest 116, 3015–3025. https://doi.org/10.1172/JCI28898

Sohn, D.H., Sokolove, J., Sharpe, O., Erhart, J.C., Chandra, P.E., Lahey, L.J., Lindstrom, T.M., Hwang, I., Boyer, K.A., Andriacchi, T.P., Robinson, W.H., 2012. Plasma proteins present in osteoarthritic synovial fluid can stimulate cytokine production via Toll-like receptor 4. Arthritis Res Ther 14, R7. https://doi.org/10.1186/ar3555

Sokol, H., Seksik, P., Furet, J.P., Firmesse, O., Nion-Larmurier, I., Beaugerie, L., Cosnes, J., Corthier, G., Marteau, P., Doré, J., 2009. Low counts of Faecalibacterium prausnitzii in colitis microbiota. Inflamm Bowel Dis 15, 1183–1189. https://doi.org/10.1002/ibd.20903

Sonnenburg, E.D., Smits, S.A., Tikhonov, M., Higginbottom, S.K., Wingreen, N.S., Sonnenburg, J.L., 2016. Diet-induced extinctions in the gut microbiota compound over generations. Nature 529, 212–215. https://doi.org/10.1038/nature16504

Srikanth, V.K., Fryer, J.L., Zhai, G., Winzenberg, T.M., Hosmer, D., Jones, G., 2005. A meta-analysis of sex differences prevalence, incidence and severity of osteoarthritis. Osteoarthritis Cartilage 13, 769–781. https://doi.org/10.1016/j.joca.2005.04.014

Stannus, O.P., Cao, Y., Antony, B., Blizzard, L., Cicuttini, F., Jones, G., Ding, C., 2015. Cross-sectional and longitudinal associations between circulating leptin and knee cartilage thickness in older adults. Ann Rheum Dis 74, 82–88. https://doi.org/10.1136/annrheumdis-2013-203308

Stannus, O.P., Jones, G., Blizzard, L., Cicuttini, F.M., Ding, C., 2013. Associations between serum levels of inflammatory markers and change in knee pain over 5 years in older adults: a prospective cohort study. Ann Rheum Dis 72, 535–540. https://doi.org/10.1136/annrheumdis-2011-201047

Strandberg, T.E., Tilvis, R.S., 2000. C-reactive protein, cardiovascular risk factors, and mortality in a prospective study in the elderly. Arterioscler Thromb Vasc Biol 20, 1057–1060. https://doi.org/10.1161/01.atv.20.4.1057

Thaiss, C.A., Levy, M., Korem, T., Dohnalová, L., Shapiro, H., Jaitin, D.A., David, E., Winter, D.R., Gury-BenAri, M., Tatirovsky, E., Tuganbaev, T., Federici, S., Zmora, N., Zeevi, D., Dori-Bachash, M., Pevsner-Fischer, M., Kartvelishvily, E., Brandis, A., Harmelin, A., Shibolet, O., Halpern, Z., Honda, K., Amit, I., Segal, E., Elinav, E., 2016. Microbiota Diurnal Rhythmicity Programs Host Transcriptome Oscillations. Cell 167, 1495-1510.e12. https://doi.org/10.1016/j.cell.2016.11.003

Thaiss, C.A., Zeevi, D., Levy, M., Zilberman-Schapira, G., Suez, J., Tengeler, A.C., Abramson, L., Katz, M.N., Korem, T., Zmora, N., Kuperman, Y., Biton, I., Gilad, S., Harmelin, A., Shapiro, H., Halpern, Z., Segal, E., Elinav, E., 2014. Transkingdom control of microbiota diurnal oscillations promotes metabolic homeostasis. Cell 159, 514–529. https://doi.org/10.1016/j.cell.2014.09.048

Thysen, S., Luyten, F.P., Lories, R.J., 2015. Loss of Frzb and Sfrp1 differentially affects joint homeostasis in instability-induced osteoarthritis. Osteoarthritis Cartilage 23, 275–279. https://doi.org/10.1016/j.joca.2014.10.010

Troeberg, L., Nagase, H., 2012. Proteases involved in cartilage matrix degradation in osteoarthritis. Biochim Biophys Acta 1824, 133–145. https://doi.org/10.1016/j.bbapap.2011.06.020

Trompette, A., Gollwitzer, E.S., Yadava, K., Sichelstiel, A.K., Sprenger, N., Ngom-Bru, C., Blanchard, C., Junt, T., Nicod, L.P., Harris, N.L., Marsland, B.J., 2014. Gut microbiota metabolism of dietary fiber influences allergic airway disease and hematopoiesis. Nat Med 20, 159–166. https://doi.org/10.1038/nm.3444

Tsuneyoshi, Y., Tanaka, M., Nagai, T., Sunahara, N., Matsuda, T., Sonoda, T., Ijiri, K., Komiya, S., Matsuyama, T., 2012. Functional folate receptor beta-expressing macrophages in osteoarthritis synovium and their M1/M2 expression profiles. Scand J Rheumatol 41, 132–140. https://doi.org/10.3109/03009742.2011.605391

Turnbaugh, P.J., Ley, R.E., Mahowald, M.A., Magrini, V., Mardis, E.R., Gordon, J.I., 2006. An obesity-associated gut microbiome with increased capacity for energy harvest. Nature 444, 1027–1031. https://doi.org/10.1038/nature05414

Ulici, V., Kelley, K.L., Azcarate-Peril, M.A., Cleveland, R.J., Sartor, R.B., Schwartz, T.A., Loeser, R.F., 2018. Osteoarthritis induced by destabilization of the medial meniscus is reduced in germ-free mice. Osteoarthritis Cartilage 26, 1098–1109. https://doi.org/10.1016/j.joca.2018.05.016

Ussar, S., Griffin, N.W., Bezy, O., Fujisaka, S., Vienberg, S., Softic, S., Deng, L., Bry, L., Gordon, J.I., Kahn, C.R., 2015. Interactions between Gut Microbiota, Host Genetics and Diet Modulate the Predisposition to Obesity and Metabolic Syndrome. Cell Metab 22, 516–530. https://doi.org/10.1016/j.cmet.2015.07.007

van de Wouw, M., Schellekens, H., Dinan, T.G., Cryan, J.F., 2017. Microbiota-Gut-Brain Axis: Modulator of Host Metabolism and Appetite. J Nutr 147, 727–745. https://doi.org/10.3945/jn.116.240481

van der Kraan, P., Matta, C., Mobasheri, A., 2017. Age-Related Alterations in Signaling Pathways in Articular Chondrocytes: Implications for the Pathogenesis and Progression of Osteoarthritis - A Mini-Review. Gerontology 63, 29–35. https://doi.org/10.1159/000448711

van der Kraan, P.M., 2017. The changing role of TGFβ in healthy, ageing and osteoarthritic joints. Nature Reviews Rheumatology 13, 155–163. https://doi.org/10.1038/nrrheum.2016.219

van der Kraan, P.M., van den Berg, W.B., 2007. Osteophytes: relevance and biology. Osteoarthritis Cartilage 15, 237–244. https://doi.org/10.1016/j.joca.2006.11.006

Vitale, G., Salvioli, S., Franceschi, C., 2013. Oxidative stress and the ageing endocrine system. Nat Rev Endocrinol 9, 228–240. https://doi.org/10.1038/nrendo.2013.29

Voreades, N., Kozil, A., Weir, T.L., 2014. Diet and the development of the human intestinal microbiome. Front Microbiol 5, 494. https://doi.org/10.3389/fmicb.2014.00494

Walsh, D.A., McWilliams, D.F., Turley, M.J., Dixon, M.R., Fransès, R.E., Mapp, P.I., Wilson, D., 2010. Angiogenesis and nerve growth factor at the osteochondral junction in rheumatoid arthritis and osteoarthritis. Rheumatology 49, 1852–1861. https://doi.org/10.1093/rheumatology/keq188

Wan, Y., Wang, F., Yuan, J., Li, J., Jiang, D., Zhang, J., Li, H., Wang, R., Tang, J., Huang, T., Zheng, J., Sinclair, A.J., Mann, J., Li, D., 2019. Effects of dietary fat on gut microbiota and faecal metabolites, and their relationship with cardiometabolic risk factors: a 6-month randomised controlled-feeding trial. Gut 68, 1417–1429. https://doi.org/10.1136/gutjnl-2018-317609

Wang, B., Li, L., 2015. Who determines the outcomes of HBV exposure? Trends in Microbiology 23, 328–329. https://doi.org/10.1016/j.tim.2015.04.001

Wang, M., Li, S., Xie, W., Shen, J., Im, H.J., Holz, J.D., Wang, M., Diekwisch, T.G., Chen, D., 2014. Activation of β-catenin signalling leads to temporomandibular joint defects. Eur Cell Mater 28, 223–235. https://doi.org/10.22203/ecm.v028a15

Wang, M., Liu, C., Zhang, Y., Hao, Y., Zhang, X., Zhang, Y.M., 2013. Protein interaction and microRNA network analysis in osteoarthritis meniscal cells. Genet Mol Res 12, 738–746. https://doi.org/10.4238/2013.March.13.2

Wang, M., Tang, D., Shu, B., Wang, B., Jin, H., Hao, S., Dresser, K.A., Shen, J., Im, H.-J., Sampson, E.R., Rubery, P.T., Zuscik, M.J., Schwarz, E.M., O'Keefe, R.J., Wang, Y., Chen, D., 2012. Conditional activation of β-catenin signaling in mice leads to severe defects in intervertebral disc tissue. Arthritis Rheum 64, 2611–2623. https://doi.org/10.1002/art.34469

Winer, D.A., Luck, H., Tsai, S., Winer, S., 2016. The Intestinal Immune System in Obesity and Insulin Resistance. Cell Metab 23, 413–426. https://doi.org/10.1016/j.cmet.2016.01.003

Wlodarska, M., Kostic, A.D., Xavier, R.J., 2015. An integrative view of microbiome-host interactions in inflammatory bowel diseases. Cell Host Microbe 17, 577–591. https://doi.org/10.1016/j.chom.2015.04.008

Wu, Q., Zhu, M., Rosier, R.N., Zuscik, M.J., O'Keefe, R.J., Chen, D., 2010. Beta-catenin, cartilage, and osteoarthritis. Ann N Y Acad Sci 1192, 344–350. https://doi.org/10.1111/j.1749-6632.2009.05212.x

Wu, R.Y., Määttänen, P., Napper, S., Scruten, E., Li, B., Koike, Y., Johnson-Henry, K.C., Pierro, A., Rossi, L., Botts, S.R., Surette, M.G., Sherman, P.M., 2017. Non-digestible oligosaccharides directly regulate host kinome to modulate host inflammatory responses without alterations in the gut microbiota. Microbiome 5, 135. https://doi.org/10.1186/s40168-017-0357-4

Xu, L., Golshirazian, I., Asbury, B.J., Li, Y., 2014. Induction of high temperature requirement A1, a serine protease, by TGF-beta1 in articular chondrocytes of mouse models of OA. Histol Histopathol 29, 609–618. https://doi.org/10.14670/HH-29.10.609

Yatsunenko, T., Rey, F.E., Manary, M.J., Trehan, I., Dominguez-Bello, M.G., Contreras, M., Magris, M., Hidalgo, G., Baldassano, R.N., Anokhin, A.P., Heath, A.C., Warner, B., Reeder, J., Kuczynski, J., Caporaso, J.G., Lozupone, C.A., Lauber, C., Clemente, J.C., Knights, D., Knight, R., Gordon, J.I., 2012. Human gut microbiome viewed across age and geography. Nature 486, 222–227. https://doi.org/10.1038/nature11053

Yu, S.P.-C., Hunter, D.J., 2015. Emerging drugs for the treatment of knee osteoarthritis. Expert Opin Emerg Drugs 20, 361–378. https://doi.org/10.1517/14728214.2015.1037275

Yusuf, E., Nelissen, R.G., Ioan-Facsinay, A., Stojanovic-Susulic, V., DeGroot, J., van Osch, G., Middeldorp, S., Huizinga, T.W.J., Kloppenburg, M., 2010. Association between weight or body mass index and hand osteoarthritis: a systematic review. Ann Rheum Dis 69, 761–765. https://doi.org/10.1136/ard.2008.106930

Zarrinpar, A., Chaix, A., Yooseph, S., Panda, S., 2014. Diet and feeding pattern affect the diurnal dynamics of the gut microbiome. Cell Metab 20, 1006–1017. https://doi.org/10.1016/j.cmet.2014.11.008

Zhang, X., Zhang, D., Jia, H., Feng, Q., Wang, D., Liang, D., Wu, X., Li, Junhua, Tang, L., Li, Yin, Lan, Z., Chen, B., Li, Yanli, Zhong, H., Xie, H., Jie, Z., Chen, Weineng, Tang, S., Xu, Xiaoqiang, Wang, X., Cai, X., Liu, S., Xia, Y., Li, Jiyang, Qiao, X., Al-Aama, J.Y., Chen, H., Wang, L., Wu, Q.-J., Zhang, F., Zheng, W., Li, Yongzhe, Zhang, M., Luo, G., Xue, W., Xiao, L., Li, Jun, Chen, Wanting, Xu, Xun, Yin, Y., Yang, H., Wang, Jian, Kristiansen, K., Liu, L., Li, T., Huang, Q., Li, Yingrui, Wang, Jun, 2015. The oral and gut microbiomes are perturbed in rheumatoid arthritis and partly normalized after treatment. Nat Med 21, 895–905. https://doi.org/10.1038/nm.3914

Zhao, X., Shah, D., Gandhi, K., Wei, W., Dwibedi, N., Webster, L., Sambamoorthi, U., 2019. Clinical, humanistic, and economic burden of osteoarthritis among noninstitutionalized

adults in the United States. Osteoarthritis Cartilage 27, 1618–1626. https://doi.org/10.1016/j.joca.2019.07.002
f.