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THE ROLE OF MICROBIOME IN IMMUNE-MEDIATED UVEITIS -LITERATURE REVIEW

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Abstract

The microbiome has important physiological functions and is an essential element in the control of the immune response in the body. Its dysbiosis is today associated with the pathogenesis of a number of diseases, including non-infectious uveitis. These are immunologically mediated conditions, where disruption of autotolerance towards intraocular structures and immune mimicry are the basis of the pathophysiological mechanism.

The aim of this paper is to present the current knowledge related to this complex interaction between the microbiome and intraocular homeostasis, as well as directions for possible therapeutic solutions through quantitative and qualitative modification of the microbiota.

During the preparation of this literature review, a meta-analysis of data obtained from several large databases was approached: PubMed, Medline and EMBASE. Keywords such as: non-infectious uveitis, microbiome, intestinal microbiome, probiotics, immune-mediated diseases, human microbiota were used in the search. The obtained results were selected, processed and didactically arranged in order to present relevant current scientific views.

***Keywords*:** non-infectious uveitis, microbiome, microbiota, dysbiosis, microbiome modulation.

Introduction

The human microbiome is the sum of the genes of all the microbiota living on the surface of the body or in physiological body cavities and biofluids. These are all microorganisms (bacteria, archaea, fungi, protists and viruses) that live and are in constant interaction with the host.

The greatest diversity and abundance of microbiota is observed on the skin and intestinal tract, but the microbiome is also present at the level of the ducts of the mammary glands, oral mucosa and saliva, tooth surface, biliary tract, distal segment of genital organs, upper segment of respiratory tract, external ear and of course ocular surface.

The microbiota includes those types of microorganisms that have a symbiotic relationship with the human body, that participate in the maintenance of human homeostasis through interaction with the host's immune system, and that do not cause pathological processes. It is important to note that the microbiome is a dynamic "organ" that changes during life under the influence of various modifying and non-modifying factors. Traditional views claim that it is established immediately after birth, although new studies indicate that some types of microorganisms are still present intrauterine.

Various factors such as: age, gender, diet, geographic origin, malnutrition, race, ethnicity and socioeconomic status have a greater or lesser impact on the quantity and quality of the human microbiome [1].

Special attention is currently being paid to the intestinal microbiome as a large and diverse treasury of microorganisms, most of which are still unknown to science. So in the last decade, a discussion has developed in the scientific community about the role of the microbiome, mostly intestinal, on the development and course of a large number of diseases that are increasingly common in human pathology.

The intestinal microbiome is the sum of the intestinal bacteriome, virome and mycobiome. It is estimated that it is composed of over 1000 species of microorganisms whose total genome is 100 times

larger than that of man. Of all microorganisms, bacteria are the most represented, with a diversity between 300-1000 species. As much as 99% of the bacterial flora belongs to 30-40 species. In general, these are anaerobic bacteria (over 99% of the total population), but some aerobic species are also found in the cecum.

Bacteria that are present in the intestinal tract mainly belong to one of four types: Firmicutes, Bacteroides, Actinobacteria and Proteobacteria [2].

The intestinal microbiome has a large number of physiological functions, most of which are still unknown, on maintaining and improving human health. One of the most important roles is the inhibition of pathogens through competition for space, utilization of nutrients and secretion of microbicidal compounds. A second well-known function is the formation of the mucosal barrier on the surface of the intestines and the interaction with the immune system through GALT (Gut-associated lymphoid tissue) as well as the stimulation of the production of cells from the innate immunity.

A third, no less significant function is related to metabolism and the creation of various substances. That is, fermentation of the so-called indigestible carbohydrates, with the end product of that process being short-chain fatty acids (SCFA), substances with a large number of positive effects on the human body. Also, the intestinal microbiota is involved in the production of some vitamins such as: biotin, folate, vitamin K and a small amount of vitamin B12, but also helps in the absorption of minerals from the diet such as: magnesium, calcium and iron. The bacterial intestinal flora is capable of metabolizing a number of xenobiotics such as drugs, toxins and phytochemicals [3].

As living and dynamic structures, the inhabitants of the intestinal microbiome are susceptible to influence by various stressors. Disturbances of the circadian rhythm, chronic fatigue, diseases, exposure to medications, toxins and infections can significantly disrupt the qualitative and quantitative composition of the microbiota and lead to its imbalance, a condition known as intestinal dysbiosis.

Dysbiosis can create a microenvironment that is suitable for the development of potentially pathogenic microorganisms and cause a disturbance in the interaction between the microbiome and the host's immune system, which will ultimately trigger the onset of a systemic or local disease [4].

A number of studies have shown a connection between the occurrence of intestinal dysbosis with various diseases: autoimmune (rheumatoid arthritis, ankylosing spondylitis and multiple sclerosis), inflammatory (ulcerative colitis and Crohn's disease), neurodegenerative (Alzheimer's and Parkinson's disease) and ophthalmological (age-related macular degeneration), glaucoma, dry eye and autoimmune mediated intraocular inflammations-uveitis [4,5].

Immunology and immune privilege of the eye

The surface of the eye (cornea and conjunctiva) comes into contact with the external environment, which is why it is covered with a large number of microorganisms, specific to that location. All types of microorganisms present on the ocular surface make up the so-called ocular microbiome. This microbiome certainly differs both in composition and population density compared to the intestinal microbiome, but with a similar role, only at the local level.

The ocular microbiome, just like the intestinal microbiome, differs from individual to individual depending on: age, gender, external influences and diet. Stressful factors such as application of topical drugs, contact lenses, systemic antibiotic therapy, diabetes and other chronic systemic diseases can lead to its dysbiosis. Since it is in constant contact with the external environment, it represents a passive, but also an immunologically active barrier against the penetration of pathogenic microorganisms. Its dysbiosis is associated with a number of surface inflammatory diseases of the eye such as: chronic blepharitis and conjunctivitis, keratitis, trachoma, dry eye, dysfunction of meibomian glands, etc.

In such diseases, a qualitative and quantitative imbalance between certain types of ocular microorganisms has been observed. However, the occurrence and course of chronic intraocular diseases (glaucoma, senile macular degeneration, diabetic retinopathy and non-infectious uveitis) have not yet been brought into a statistically significant correlation with ocular dysbiosis, as is the case with intestinal dysbiosis [6,7].

The normal ocular microbiota mainly contains representatives of the genera: Corynebacterium, Propionibacterium and Staphylococcus.

The interior of the eye is a sterile environment, and its maintenance is "per se" for the establishment of intraocular homeostasis. A large number of immune cells and inflammatory mediators are present in the intraocular structures, but the immune privilege of the tissues prevents the occurrence of intraocular inflammation through immune tolerance and ignoring [8].

Immune privilege refers to a protective response to immune challenge in tissues with limited capacity for regeneration such as intraocular structures and brain tissue. It is important to note that the structures of the ocular interior are heterogeneous in their composition, that is, they are composed of different types of tissues, so they possess a different level of immune privilege (each tissue carries out a different degree of modulation of the immune response to a certain antigen).

At the apex of this immunological privilege in the eye is the retina with the complex molecular and neuro-immunological mechanisms possessed by this tissue, in contrast to the anterior chamber, whose degree of privilege is probably at a lower level, with the consequence that anterior uveitis is the most common type of uveitis in daily practice [9].

The idea of the existence of an immunological privilege in certain human tissues dates back to the 19th century, but its importance in explaining the complex immunological phenomena that occur in some organs has been established in the last fifty years.

This concept is a kind of balance between the ability of tissues to regenerate and the degree of immune response in the presence of a certain antigen.

Thus, tissues can be arranged "hierarchically" according to the degree of immunological privilege. In other words, tissues that have a great regenerative ability, such as the mucosa of the skin, respiratory and gastrointestinal tract, have little immune privilege, so the inflammatory response and tissue damage in them in the presence of a pathogen (antigen) is quite pronounced.

On the contrary, brain tissue (including the retina) has almost no regenerative ability, so it is considered that the immune response in the presence of a certain antigen is quite silent, based on immune tolerance and ignoring, all in order to prevent tissue damage.

Between these two extremes of immunological privilege, other tissues are spectrally arranged: uveal tract, spleen, meninges, liver, cornea/sclera, tendons/muscles, kidney, glandular tissue, etc. [10]

The immunological privilege of the eye is thought to be due to the presence of its own antigens that are protected by the immune system behind the blood-tissue barrier due to a lack of blood vessels, lymphatic drainage, the absence of MHC II antigenic cells, a high concentration of immunosuppressor tissue immunomodulators, as well as of regulatory T cell activity.

A second phenomenon in the eye that is directly related to the immunological privilege of this organ is the so-called ACAID (anterior chamber-associated immune deviation). It is basically an experimental model, of introducing an antigen into the anterior chamber of the eye, through which the systemic regulatory response is perceived [11].

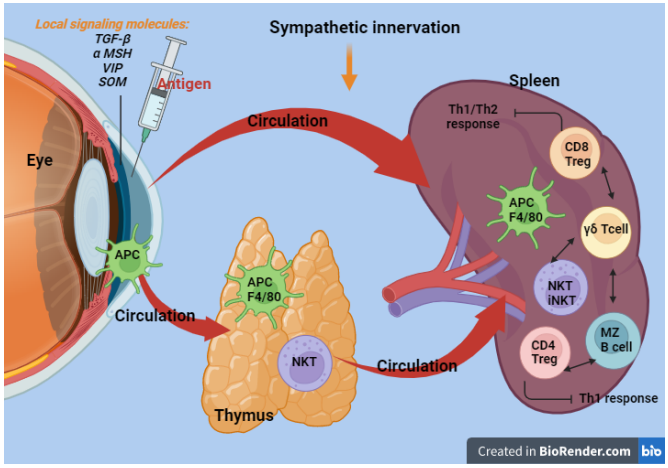


Figure 1. ACAID-anterior chamber-associated immune deviation. (Created in: BioRender.com

In this phenomenon, suppression of a delayed-type cell-mediated hypersensitivity reaction occurs and IgG2 antibodies are generated as a result of humoral immunity activity.

Antigen entering the anterior chamber is phagocytosed by antigen-presenting cells whose origin remains controversial (blood monocytes or resident iris antigen-presenting cells). After being phagocytized by these F4/80+ CD11b+ ocular APCs, they migrate to the thymus and there stimulate the generation of natural killer cells (NKT), thus generated NKT through the blood network reach the spleen where they play an important role in the creation of splenic suppressor cells.

The process takes place in the marginal zone of the spleen. There APC from the eye and NKT from the thymus participate in the generation of immunomodulatory cells: T-regulatory cells (CD8+ and CD4+), B-cells, MZ-B-cells, NKT-regulatory and iNKT- regulatory cells that through the systemic blood network induce an immune deviation (Figure 1). The result is an inhibitory effect on the progression of the inflammatory response, with the aim of reduced destruction of the ocular structures and preservation of their function- the vision of the affected individual [12].

External and internal tissue barrier

The immunological privilege of the eye is certainly ensured through the existence of the so-called external and internal tissue barrier, which protect against the penetration of a potential pathogen into the intraocular structures (Figure 2). The external tissue barrier consists of the skin and mucosa of the intestinal tract, which are in constant contact with the external environment.

They are the first lines of defense of the organism against pathogenic agents. It is important to note that they do not represent "simple" physical barriers, but are made up of numerous immune cells (myeloid cells, dendritic cells, intraepithelial and stromal T effector and T regulatory lymphocytes, NKT cells and many others) as well as immunomodulatory molecules that are of exceptional importance for ensuring the overall homeostasis in the organism (defensives, neuropeptides, TGF-β, thrombospondin), cytokines, chemokines, etc. The intestinal microbiome, which is in direct contact with the intestinal mucosa, obviously plays a very important role in maintaining this so-called external barrier.

The microbiome does this both through competition with pathogenic microorganisms and through the creation of a large number of anti-inflammatory molecules, with which it maintains mucosal integrity, but also regulates the local immune response [13].

Internal barriers are at the level between blood vessels and tissues in a particular organ. They differ from tissue to tissue, both in terms of the type of endothelial cells that build the blood vessels, and in terms of the surrounding accompanying elements that are involved in the formation of their integrity. At the level of the liver, spleen and uveal tract, these barriers have a moderate degree of selectivity, in contrast to the CNS and retinal tissue, where they provide a complex physical, molecular and immunological restriction on the entry of a potential antigen. It is assumed that the blood-retinal barrier develops gradually in the perinatal period, and the microbiome itself plays a major role in that process.

Evidence for this thesis is presented by several preclinical studies that showed the importance of the microbiome in mice on the development of the blood-brain barrier (BBB), through the creation of more competent protein molecules (occludin and claudin 5) that participate in the creation of endothelial tight junctions. Blood-retinal barier (BRB), as the counterpart of BBB, is considered to undergo the same creation processes. According to this, it can be concluded that the creation of a healthy microbiome in the neonatal period has a key role on the formation and integrity of these life-important tissue barriers [9,14].

Previous views on the BRB were that it represents a physical barrier built by endothelial cells that are connected to each other by tight junctions that prevent the penetration of any antigen into the retinal tissue.

However, new research has pointed to the fact that BRB is an immunologically active barrier and not just a passive endothelial barrier. In addition to endothelial cells, pericytes with immunomodulatory activity, perivascular macrophages, extensions of surrounding glia and Müller cells, as well as the dendrites of retinal neurons participate in its composition, which together constitute the so-called neurovascular unit. It is believed that T-regulatory cells play a major role in the functioning of the BRB, which perform immunomodulation and regulation of this specific barrier, thereby enabling a high immunoprivilege of the

retinal tissue. An additional barrier that protects the neurosensory retina from antigenic attack is the retinal pigment epithelium with Bruch's membrane. They represent a highly selective barrier between the choriocapillary blood vessels of the choroid and the neurosensory retina [15].

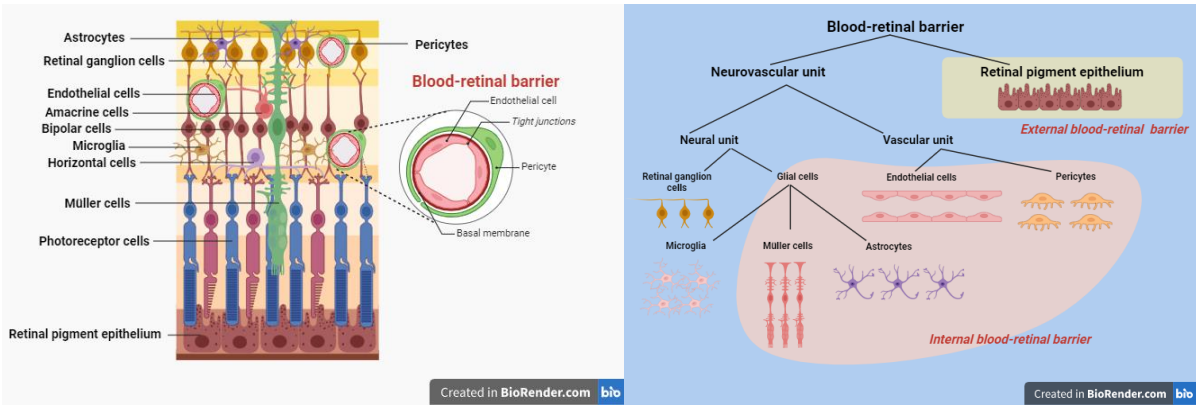


Figure 2 Cytoarchitecture of the retina and cells involved in the construction of the blood-retinal barrier and the neurovascular unit. (Created in: BioRender.com)

Current views are that the disruption of this retinal neurovascular unit is the basis for a number of retinal diseases such as: diabetic retinopathy, age-related macular degeneration and retinal vasculitis [15].

Due to the immune privilege of the retina, inflammations in the anterior segment of the eye are not subject to an accompanying reaction by the retina. On the contrary, retinal affections in BRB disorders (for example, retinal vasculitis) are very often accompanied by an inflammatory response from the structures of the anterior part of the uvea.

It is precisely through this phenomenon of uveitis progression that the special immune privilege of the retinal tissue is perceived, most likely to protect it from potentially irreversible damage that would seriously impair vision and lead to blindness. In one of these findings, it can be concluded about the immunologically privileged hierarchy of intraocular structures, where the retina is at the top of it, in contrast to the anterior chamber and the uveal tract, which have a lower degree of immunological privilege (according to the direction of the inflammatory process).

As mentioned above, the development of BRB occurs in the perinatal period and early childhood. Therefore, it is considered that the majority of retinal tissue infections occur during this period, when BRB is not yet sufficiently developed. However, they remain latent for many years under the influence of local T regulatory lymphocytes that do not allow immune activation and the initiation of an inflammatory response.

Evidence for this thesis is also AIDS patients, in whom, due to the decline of the T-cell population, the reactivation of latent infections, including those at the level of the retina (HSV, CMV, VZV, Toxoplasma gondii and Mycobacterium tuberculosis), begins. In this regulation, between T regulatory cells and controlled activation of T effector cells, a major role is considered to be played by the intestinal microbiome, which, in addition to participating in the development of BRB, also participates through immunomodulatory molecules in the creation and maintenance of the balance between these two cell populations. When the BRB is damaged, it is often accompanied by intestinal dysbiosis, the immune privilege is lost and intraocular inflammation begins with a serious threat to vision [9, 16].

Gut-brain axis and GUT-ocular axis

In recent years, the existence of the so-called Gut-brain axis, communication between the gastrointestinal system and intestinal microbiota with the enteric nervous system and the central nervous system, has been discussed. Several preclinical studies have demonstrated this association, as well as the importance of the microbiota in this complex interaction. Namely, the intestinal microbiota and the Gut-

brain axis are connected through a bidirectional communication link that is mediated under the influence of the immune system. Components of this, for now insufficiently known interaction are: intestinal microbiota, products of its metabolism, enteric, sympathetic and parasympathetic nervous system, neuro-immune, neuro-endocrine and central nervous system. Several routes of communication between these two instances have been established: neural network, neuroendocrine axis (hypothalamic-pituitary-adrenal axis), Gut immune system, some neurotransmitters and neural regulators synthesized by intestinal microbiota and of course, external and internal (BBB) tissue barrier.

There are theories that explain the possible etiopathogenesis of some neurodegenerative diseases such as: Alzheimer's dementia, Parkinson's disease and multiple sclerosis through the disruption of this communication [17].

The appearance of ophthalmic manifestations (uveitis, episcleritis and scleritis) in some inflammatory bowel diseases (ulcerative colitis and Crohn's disease) may be the first sign of the existence of a connection between these two organ systems. N.opticus and retinal tissue are an extension of the central nervous system (brain tissue), so the Gut-brain axis also functions at the level of the retina, or in conjunction with the entire nervous system, but also probably as a separate axis gut-retinal (ocular) axis .

Other evidence for the association and bidirectional communication between the gut microbiome and the retina are preclinical studies of inducing retinal diseases in murine models through dietary manipulation of macro- and micronutrients.

The consequence of such nutritional modifications is the appearance of intestinal dysbiosis, accompanied by the appearance and progression of various retinal diseases: diabetic retinopathy, senile macular degeneration and retinitis pigmentosa. In accordance with these findings is the previous presentation of the connection between the microbiome, its metabolites (eg SCFAs), T regulatory cells and the appearance of uveitis, which supports the existence of such a connection. Further studies and evidence are expected in the future that will more precisely define this bidirectional biochemical-immunological communication between the intestinal microbiome and the eye [17,18].

Animal models for autoimmune uveitis

Most of the association between the gut microbiome and uveitis has been obtained through preclinical studies with experimental autoimmune uveitis (EAU) models. The most commonly used model is the EAU mouse model through active immunization with IRBP (inter-photoreceptor retinoid-binding protein) and the spontaneous EAU model of transgenic mice expressing auto-reactive T-cell receptors [19].

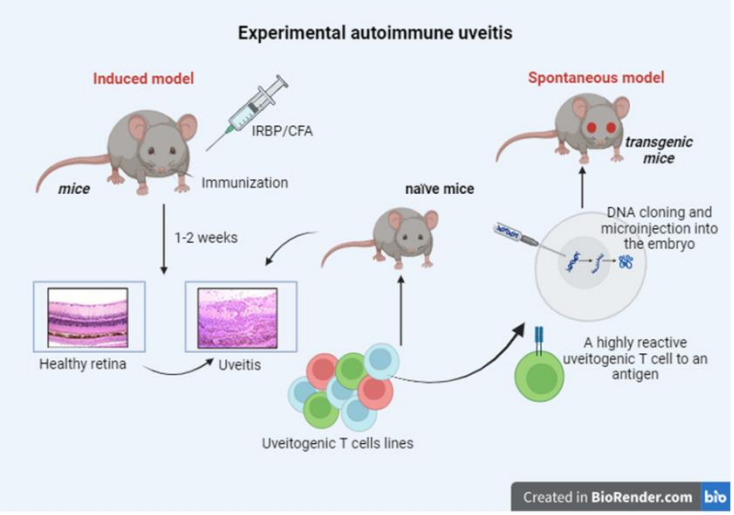
Basically, experimental autoimmune uveitis is an animal model of the disease, which is the counterpart of human endogenous uveitis. It occurs by immunization of retinal antigens in animals that are sensitive to this kind of inflammation.

The resulting intraocular inflammation targets the neurosensory retina, via a T-cell-mediated mechanism, similar to what occurs in human uveitis. Uveitogenic retinal antigens used in the preparation of such models are: arrestin (retinal soluble antigen), internal photoreceptor protein that binds retinoid, rhodopsin, opsin, recoverin, phosducin, etc.

EAU can be induced in different species of animals such as: rats, mice, monkeys and rabbits. None of these animal models are ideal representatives of the full spectrum of human uveitis. Each of them has specific characteristics, which make it suitable for the examination of a certain disease or a characteristic pathophysiological mechanism.

The classic EAU model has been known since 1988, and has been modified several times in the past decades. Induction is carried out by an internal photoreceptor protein that binds rhodopsin, which is in emulsion with a suitable adjuvant (CFA-complete Freund's adjuvant), consisting of a suspension of tubercle bacilli in mineral oil. CFA is critical for disease induction. It has been applied to mice and rats, some strains requiring additional costimulation with pertussis toxin. After being stimulated by intradermal immunization, animals develop panuveitis in 1-2 weeks. The adjuvant induces a type 1 T-helper cell-mediated response. Such a model is characterized by a clinical picture of retinitis or chorioretinitis, retinal vasculitis with subsequent impairment of visual function in the affected individual. A modification of this model is the method where the disease is induced by injecting lymphocytes that are specific for retinal antigens, obtained from donors who have been immunized for the induction of EAU (Figure 3). In this way, the clinical picture is much more similar to that of clinical practice [20].

Figure 3 Induced and spontaneous model of experimental autoimmune uveitis. The resulting uveitogenic T-cell line after induction with IRBP/CFA induces uveitis in a naïve mouse. Obtaining a



highly reactive uveitogenic T-cell line, DNA cloning and microinjection into a mouse embryo, leads to the formation of a transgenic mouse that spontaneously develops uveitis at a young age. (Created in: BioRender.com)

Pathophysiological mechanisms of microbiome dysbiosis and uveitis

Unlike infectious uveitis, where the microorganism with its pathogenicity acts directly through invasion and intoxication on the ocular structures, in non-infectious uveitis, microorganisms can affect the inflammatory process through immune mechanisms. As the main microbiome, the gut is a major source for such mechanisms in a number of autoimmune diseases, including autoimmune uveitis. So far, four main pro-inflammatory mechanisms have been discussed (Figure 4). Their beginning is at the level of the relationship between the intestinal microbiome and the immune components in its mucosa. It is important to note that these mechanisms are not separate in their action in autoimmune uveitis. They overlap and take place successively during one uveitic process. In the future, it is expected that they will be understood in more detail, as well as the discovery of new mechanisms through which the intestinal microbiota maintains uveal homeostasis, and how it triggers the occurrence and reactivation of an autoimmune cascade during its dysbiosis.

1. Antigenic or microbial mimicry

The mechanism of microbial mimicry is well known in the pathogenesis of a number of autoimmune diseases. Basically, it represents cross-reactivity in which microbial peptides and autoantigens have similar protein domains, so when generating T-cells against the microbial agent, cross-reactivity to

autoantigens occurs, and they are attacked by the immune system. Examples of this mechanism of pathogenesis are found in HLA-B27-associated diseases, including uveitis associated with: ankylosing spondylitis, reactive arthritis, psoriatic arthritis, and inflammatory bowel disease.

This pathophysiological mechanism is considered the basis through which intestinal dysbiosis is involved in the generation of autoimmunity [21]. Under investigation are several peptides and bacteria that are considered to have a high percentage of homology in amino acid sequences, and these are related to the pathogenesis of HLA-B27 associated uveitis. Such peptides that are involved in microbial mimicry have so far been associated with some species of bacteria such as Chlamydia trachomatis, Campylobacter jejuni as well as representatives of: Klebsella, Salmonella, Yersinia and Shigella [21,22].

Apart from preclinical studies, potential peptides associated with microbial mimicry have been demonstrated in some autoimmune diseases from clinical practice. An example of this is the disease neuromyelitis optica, an autoimmune demyelinating disease in which the optic nerve and spinal cord are the target of attack. A humoral and cellular autoimmune process attacks the protein water channels AQP4-aquaporin 4 underlying the pathophysiological mechanism.

A peptide molecule (ABC-TP: adenosine triphosphate-binding cassette transporter permease) from the bacterium Clostridium perfringens has been shown to have over 90% homology with the T-cell epitope of the amino acid chain of AQP4, which explains the microbial mimicry in the pathogenesis of this neuro-ophthalmic disease. A similar process has been demonstrated in patients with Systemic Lupus, where cross-reactivity is thought to occur between the RNA-binding autoantigen Ro60 and a peptide molecule from Propionibacterium propionicum and Bacteroides thetaiotaomicron [22].

2. Increased intestinal permeability

Intestinal integrity is an important barrier to overall internal homeostasis in humans. It represents both passive and immunologically active protection with a high degree of control over the entry of various substances into the body's blood network. Undoubtedly, the microbiota found on its mucosal surface plays an important role in the permeability and integrity of this important immune protection.

On the other hand, dysbiosis in the intestinal microbiome can cause inflammation at the level of the mucosa that will lead to damage of this barrier, disruption of permeability and penetration of intestinal substances into the blood or lymphatic network through the rich vascular systems of the lamina propria [23].

Microorganisms and their products such as lipopolysaccharides and β-glucans through the intestinal circulation can reach peripheral tissues such as joints and the uveal tract, depositing in those places as antigens causing the initiation of an inflammatory cascade, with the clinical manifestation of arthritis or uveitis, respectively. This mechanism of pathogenesis has been observed in patients with ankylosing spondylitis, in which the dysregulation of tight intestinal cell connections (tight junction) and lipopolysaccharide translocation through the blood network are considered to be two main factors for the exacerbation of the inflammatory process, and they are related to intestinal dysbiosis. Experimental studies with EAU mice confirmed this relationship between intestinal dysbiosis and impaired intestinal integrity with the occurrence and degree of uveal inflammation [23,24].

3. Loss of intestinal immune homeostasis

As is known, T cells play a significant role in the development of uveitis, as well as in autoimmune diseases in general. In intestinal dysbiosis, there is a disturbance of intestinal homeostasis, and thus a disturbance of the balance between T helper 17 (Th17) and Tregs, in the direction of increasing the activation of Th17, which is considered a key moment in the pathogenesis of uveitis.

The mechanism of inflammation is triggered by intestinal dysbiosis and stimulation of Th17 cells by antigen presenting cells in the GALT of the gut. This data has been confirmed through experimental models of EAU-mice where the application of broad-spectrum antibiotics caused a modification of the intestinal microbiota. Reduction of representatives from Firmicutes, Bacteroidetes and Alphaproteobacteia, and increase of Gamaproteobacteria. This led to an increase in Tregs cells in the lymphatic circulation and the eye, thereby reducing the severity of uveal inflammation. Through these mechanisms, the movement of

T cells and other immune cells originating from the gut towards the ocular structures has been proven. In other words, it can be concluded that immune homeostasis is of exceptional importance on the gut-ocular axis, to maintain a constant balance between pro-inflammatory and anti-inflammatory signals in the uveal tissues. That balance is maintained under the influence of various microorganisms from the intestinal microbiota. Dysbiosis can trigger a disturbance in the ratio between Th17 and Tregs, with subsequent generation of a pro-inflammatory cascade [25].

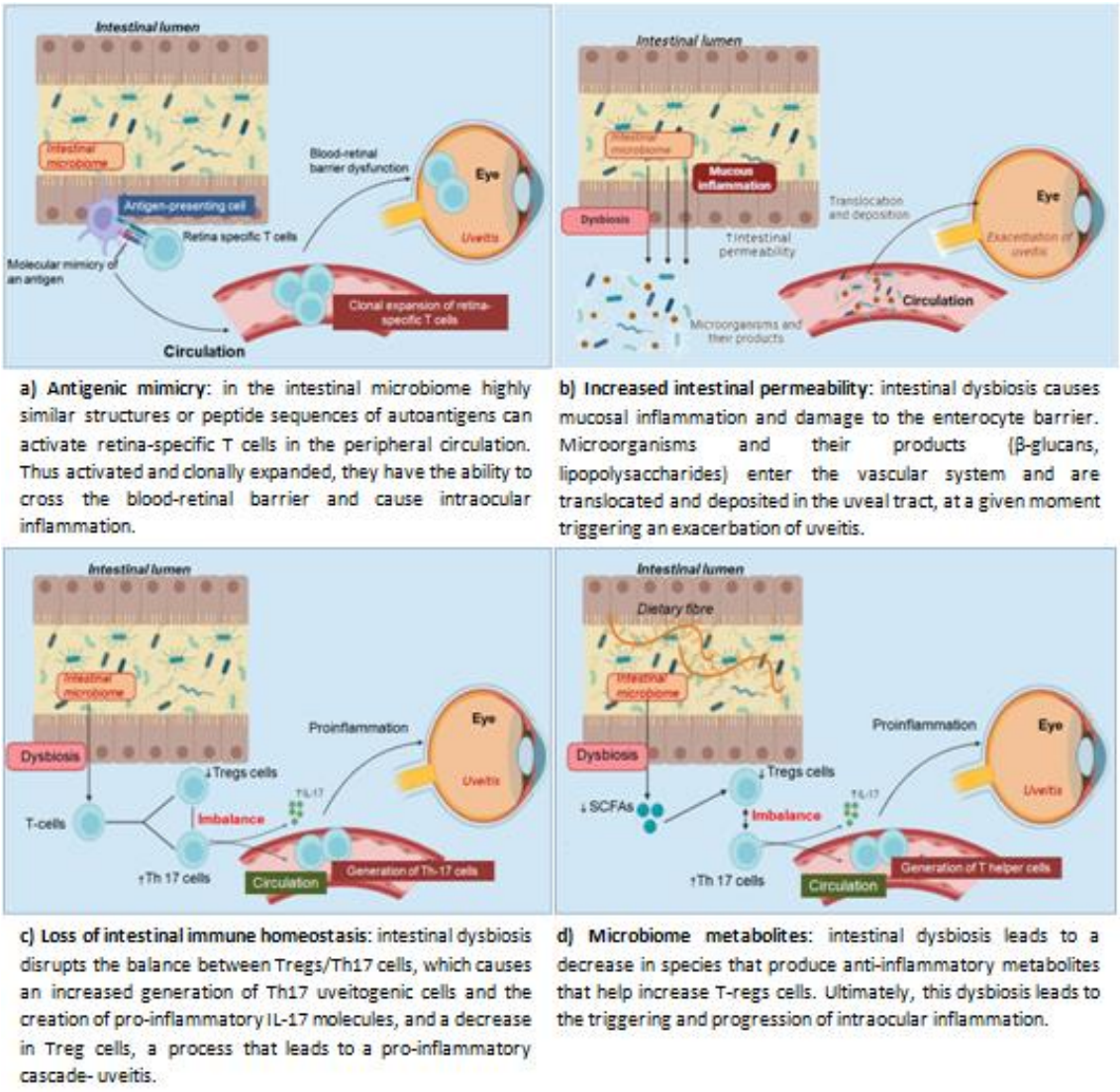
4. Reduction of anti-inflammatory microbial metabolites

Intestinal microorganisms exert their positive influence on the body through the final metabolites from their metabolism. In other words, the microorganisms in the colon have the ability to metabolize indigestible vegetable fibers to short-chain fatty acids (SCFAs): butyric, propionic, and acetic acid. The role of these end products is seen both in protection from pathogenic microorganisms and in improving the condition of a large number of inflammatory diseases (eg, ankylosing spondylitis). Similar to other immune-mediated diseases, SCFAs in uveitis play a role in reducing inflammation through two main processes:

1. increasing the number and efficiency of Tregs cells in intestinal tissues and lymph nodes;
2. suppression of effector T cells and reduction of their communication between the intestinal system and the spleen.

Experimental models have demonstrated the influence of SCFAs on reducing uveal inflammation through exogenous administration of these substances in animal models.

Conversely, with intestinal dysbiosis and reduction of the species that have the ability to create these anti-inflammatory metabolites, there is an increase in the frequency and severity of ocular autoimmune inflammations. Additional studies have shown that not only are these substances reduced in the intestines of patients with anterior autoimmune uveitis, but such patients also have a large number of pro-inflammatory molecules in their feces. Some of these molecules have so far been associated with triggering inflammatory cascades in a number of inflammatory diseases. Currently known: linolenic acid, azelaic acid, palmitoleic acid, isomaltose-1, shikimic acid, N-acetyl-beta-D-mannosamine and 6-deoxy-D-glucose-1. The mechanisms through which these substances are involved in uveitis are currently unknown, but their pro-inflammatory nature is known - generation of pro-inflammatory cascades, oxidative damage and carcinogenesis [26].



a) Antigenic mimicry: in the intestinal microbiome highly similar structures or peptide sequences of autoantigens can activate retina-specific T cells in the peripheral circulation. Thus activated and clonally expanded, they have the ability to cross the blood-retinal barrier and cause intraocular inflammation.

b) Increased intestinal permeability: intestinal dysbiosis causes mucosal inflammation and damage to the enterocyte barrier. Microorganisms and their products (β-glucans, lipopolysaccharides) enter the vascular system and are translocated and deposited in the uveal tract, at a given moment triggering an exacerbation of uveitis.

c) Loss of intestinal immune homeostasis: intestinal dysbiosis disrupts the balance between Tregs/Th17 cells, which causes an increased generation of Th17 uveitogenic cells and the creation of pro-inflammatory IL-17 molecules, and a decrease in Treg cells, a process that leads to a pro-inflammatory cascade- uveitis.

d) Microbiome metabolites: intestinal dysbiosis leads to a decrease in species that produce anti-inflammatory metabolites that help increase T-regs cells. Ultimately, this dysbiosis leads to the triggering and progression of intraocular inflammation.

Figure 4 Mechanisms of microbiome dysbiosis and uveitis (Created in: BioRender.com)

Therapeutic approaches aimed at modifying the gut microbiome to achieve uveitis remission

From the previous preclinical and clinical studies, certain directions have been obtained for possible therapeutic approaches that would perform some kind of modification of the microbiome, and through it would help in reducing intraocular inflammation. These include: application of probiotics and prebiotics, antibiotic therapy, fecal transplantation, as well as application of immunomodulatory drugs and biological preparations. Some of these therapeutic modalities directly and others indirectly influence both the quantitative and the qualitative composition of the intestinal microbiota. Probiotics and prebiotics do this directly, by modulating the intestinal microbiome and improving the anti-inflammatory response in the intestinal immune system, thereby reducing pro-inflammatory cytokines to intraocular structures and cervical lymph nodes. Prebiotics and SCFAs from probiotics also influence Th17 cell regulation, by reducing their number and increasing Tregs cells, which is currently considered a key driver of ocular immune homeostasis. Antibiotics and fecal transplants cause the effect by changing the populations of microorganisms in the gut. Immunomodulatory and biological agents achieve their effect through modulation and modification of the pathophysiological immune mechanism and determining which patients

are responders and which are not to this therapeutic regimen by following changes in the intestinal microbiota (they play the role of biomarkers) [22,27].

Although such knowledge is currently quite scarce about the impact of these therapeutic modalities in patients with non-infectious uveitis, they are a significant harbinger of a new era of treatment known as personalized medicine in the treatment of autoimmune diseases, including uveitis.

Conclusion

According to everything previously mentioned, it can be concluded that there is a strong causal relationship between the intestinal microbiome, its dysbiosis and immune-mediated uveitis. The existence of a gut-ocular axis is clearly functional in each individual, and the balance between pro-inflammatory and anti-inflammatory mediators is a key link in maintaining intraocular homeostasis.

Finally: the diet, the application of probiotics and prebiotics, antibiotic therapy and fecal transplantation, as well as monitoring the uveitic process in patients treated with biological therapy through the qualitative-quantitative changes in the microbiome; they may be a "first step" towards a personalized medicine as well as possible "game-changers" in the treatment and follow-up of patients with autoimmune mediated uveitis.

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