

THE GUT MICROBIOTA IMMUNE AXIS IN IRAQI ADULTS: REVEALING THE INTERACTION BETWEEN MICROBIAL COMPOSITION AND HOST IMMUNE RESPONSE

Eman Sadiq Abd-ali¹, Nawal Barzan yazea²

¹College of Dentistry, University of Mustansiriyah, Baghdad, Iraq

²College of Science, University of Mustansiriyah, Baghdad, Iraq.

ARTICLE INFO

Received: 21 June 2024

Revised: 23 July 2024

Accepted: 24 August 2024

Keywords:

Immune, inflammation, Gut microbiota, Interleukins, T and B cells

Corresponding Author:

Eman Sadiq Abd-ali

Email:

emansadiq1992@uomustansiriyah.edu.iq

Copyright © 2024 by author(s)

This work is licensed under the Creative Commons Attribution International License (CC BY 4.0).
<http://creativecommons.org/licenses/by/4.0/>



ABSTRACT

The gut-immune axis refers to the reciprocal interaction between the gut bacteria and the immune system. The dynamic interaction discussed here is a significant factor in determining one's overall health, since it has an impact on various aspects such as immunological responses, inflammation, and the preservation of gut homeostasis. The objective of this study is to elucidate the intricate relationship between microbial prevalence and important factors, so providing insights into potential consequences for immune responses. A cross-sectional study involved 80 consenting Iraqi people aged 18–65. Feces and blood were carefully collected and processed for microbiota and immune response studies. The poll was pilot-tested for clarity and cultural fit. Safely transported samples by cold chain. Immune markers were measured with ELISA kits. Data analysis was done using SPSS Version 25.0 and chi-square and p-value testing. The study found significant correlations between gut microbiota prevalence and demographics. As people aged, Lactobacillus and Bifidobacterium decreased and Colistriduim and E. coli increased. Lactobacillus and E. coli prevalence differed by gender. All tested individuals' microbiota was influenced by their place of residence, showing urban and rural differences. Microbe incidence was significantly correlated with education and income. Diet and smoking greatly affected microbial profiles. The study also found links between gut microbiota, interleukins, and T and B cells. This study illuminates the intricate interaction between gut microbiota and numerous variables in Iraqi adults. Lifestyle and demographic characteristics may affect microbial composition and immunological responses, as shown by the relationships.

INTRODUCTION

The gut microbiota encompasses a heterogeneous assemblage of microorganisms, comprising bacteria, viruses, and fungi, which inhabit the gastrointestinal system. The intricate ecosystem serves a crucial function in upholding human well-being (Wang et al., 2021). The process of digestion is facilitated by this substance, which also plays a crucial role in the absorption of nutrients and the synthesis of vital vitamins (Chang & Martinez-Guryn, 2019). It also helps the immune system fight pathogens and avoid chronic diseases. The gut flora produces short-chain fatty acids and regulates metabolism and weight (van der Hee & Wells, 2021; Segal et al., 2020).

Gut Microbiota Composition:

The gut microbiota in adult humans has an aggregate weight of approximately 1.5-2 kg. The makeup of the microbiota exhibits inter-individual variability, primarily consisting of two prominent bacterial phyla, namely Firmicutes and Bacteroidetes. However, it also encompasses additional phyla such as Proteobacteria, Actinobacteria, and various others (Banaszak et al., 2023; Bhargava et al., 2022). The makeup of the gut microbiota is influenced by multiple variables. The function of nutrition is of utmost importance, since high-fiber diets have been found to facilitate the promotion of microbial diversity and a well-balanced microbiota, whereas excessive intake of processed foods has been associated with the occurrence of dysbiosis (Martinez et al., 2021). The role of genetics is also influential in shaping an individual's propensity to specific microbial profiles. The makeup of microbiota can be influenced by factors such as the use of antibiotics, adherence to cleanliness measures, and the geographical location. Moreover, it has been demonstrated that lifestyle factors, including stress levels, sleep patterns, and levels of physical activity, exert an influence on the composition and functioning of the gut microbiota (Zhang et al., 2021; Lobionda et al., 2019).

The Immune System and Gut Health:

The immune system assumes a crucial role in the preservation of gut health through its regulation of the intricate equilibrium between the acceptance of helpful gut microorganisms and the protection against detrimental infections. The gut is under constant surveillance by specialized immune cells, including gut-associated lymphoid tissue (GALT) (Croese et al., 2021; Mörbe et al., 2021). The immune system plays a dual function within the gastrointestinal tract, encompassing both protective mechanisms against foreign pathogens and the facilitation of symbiotic relationships with indigenous gut microbiota (Yamamoto & Aizawa, 2021). The maintenance of a dynamic equilibrium serves to protect the gastrointestinal tract from infections, while also providing support for good bacteria. The interaction between various factors is crucial for maintaining a healthy gastrointestinal tract and promoting effective immune system functioning. This interplay serves to protect against chronic inflammation, autoimmune reactions, and the invasion of harmful pathogens, while also fostering a mutually beneficial connection with the microbiota (Nikolenko et al., 2021; Wiertsema et al., 2021).

Immune responses and their impact on microbial composition:

The immune responses within the gastrointestinal tract exert a significant influence on the composition of the gut microbiota, and this reciprocal relationship plays a pivotal role in sustaining a well-balanced and mutually beneficial microbial community. Immunoglobulin A (IgA), an essential constituent of the mucosal immune system, assumes a pivotal function in modulating the microbiota. The substance forms a protective layer on the bacterial surfaces, inhibiting their ability to attach to the lining of the gastrointestinal tract and thereby restricting the proliferation of specific bacterial species (Espirito Santo et al., 2021). When infections are present, the immune system initiates inflammatory responses that are characterized by the release of proinflammatory cytokines and the recruitment of immune cells in order to remove the invading pathogens (Al-Banna et al., 2018). Regulatory T cells, also known as Tregs, play a crucial role in the maintenance of immunological tolerance. These mechanisms serve to inhibit exaggerated immune responses towards commensal microorganisms, so facilitating the proliferation of beneficial germs while preventing detrimental immunological reactions (Okeke & Uzonna, 2019).

Gut Microbiota Dysbiosis:

Gut microbiota dysbiosis refers to the perturbation or disturbance in the structure and operation of the microbial population inhabiting the gastrointestinal system (Das & Nair, 2019). Multiple

causes can contribute to this condition, such as alterations in eating patterns, the utilization of antibiotics, persistent stress, infections, and genetic predispositions. Modified dietary patterns, namely those characterized by reduced fiber content and elevated sugar intake, have the potential to induce a decline in advantageous microbial populations and a concurrent rise in potentially detrimental microorganisms (Pushpanathan et al., 2019; Pulikkan et al., 2019). The administration of antibiotics has the potential to perturb the composition of the gut microbiota by indiscriminately eradicating both beneficial and pathogenic bacteria (Yamamoto & Aizawa, 2021). The gut-brain axis is a bidirectional communication system between the central nervous system and the gastrointestinal tract. Chronic stress has been found to have an impact on the makeup of the gut microbiota (Lobionda et al., 2019). Dysbiosis has the potential to initiate a series of immune-related complications. First and foremost, the disturbance in microbial composition frequently elicits chronic inflammation in the gastrointestinal tract, hence potentially activating the immune system. The chronic nature of this inflammation might give rise to systemic consequences, hence playing a role in the development of several diseases (Vijay & Valdes, 2022; Torun et al., 2021). Moreover, dysbiosis has the potential to undermine immunological tolerance, so eliciting improper immune reactions towards self-tissues and potentially culminating in the development of autoimmune disorders (Mousa et al., 2022). On the other hand, the gut microbiota plays a role in instructing and regulating the immune system. The condition of dysbiosis can disturb the intricate balance within the body, which may result in immunological dysregulation (Zhang et al., 2021).

Previous studies have demonstrated a decline in the richness of the gut microbiota, which is frequently associated with a drop in advantageous bacterial strains such as *Bifidobacterium* and *Lactobacillus* species. The alterations mentioned above have been linked to dietary modifications in Iraq, which involve a rise in the consumption of processed foods and a decrease in fiber intake. These changes have been found to contribute to dysbiosis (Bhargava et al., 2022; Martinez et al., 2021).

Host immune response:

Pathogens are fought by the immune system's vast network of cells, tissues, and chemicals. It is innate and adaptable. Innate immune systems defend against a wide spectrum of invaders quickly and nonspecifically (Banaszak et al., 2023), while adaptive immune systems produce particular antibodies and immunological memory for long-term protection (Bhargava et al., 2022). Genetic, environmental, and age-related immune system variations exist. Due to genetic variances in immune responses, certain people are more susceptible to particular diseases. Environmental factors including food and infections affect immune system function. Immune responses vary in children, adults, and the elderly due to age-related immune system alterations (Martinez et al., 2021; Zhang et al., 2021).

Variations in immune responses affect gut health. Genetic and environmental variables affecting immune function can affect gut microbial balance. These variants can affect infection risk, pathogen control, and gut microbiota responses to diet and environment (Lobionda et al., 2019).

Interaction between Gut Microbiota and Host Immune Response:

The dynamic and complicated link between microbial makeup and immune response is of crucial importance to overall health. The gut microbiota plays a significant role in the modulation of the immune system, and conversely, the immune system influences the composition and function of the gut microbiota (Segal et al., 2020; Vijay & Valdes, 2022). The microbiota plays a crucial role in providing an antigenic stimulus that serves to educate and train the immune system in differentiating between benign commensal microorganisms and dangerous infections (Torun et al., 2021). The immune cells consistently surveil the gastrointestinal tract in

order to sustain immunological tolerance towards the indigenous microorganisms, while also being alert to potential intruders. The immune system, in a reciprocal manner, exerts control over the composition of the gut microbiota through the secretion of immunoglobulins such as IgA. This immunoglobulin has the capacity to impact the prevalence of particular bacterial species (Mousa et al., 2022).

Gut microbiota composition and Immune Dysregulation link to diseases:

The correlation between the composition of the gut microbiota and many diseases has been shown, highlighting its significant influence on human health. Dysbiosis has been linked to gastrointestinal illnesses such as irritable bowel syndrome (IBS) and inflammatory bowel disease (IBD) (Zhang et al., 2021). Moreover, there is evidence suggesting that changes in the composition of the gastrointestinal microbiota are associated with metabolic disorders such as obesity and type 2 diabetes, as it exerts an impact on energy metabolism and the assimilation of nutrients (Mousa et al., 2022; Zhang et al., 2021). The control of the immune system by the gut microbiota signifies that dysbiosis has the potential to contribute to the development of autoimmune disorders. In several pathological states such as rheumatoid arthritis, multiple sclerosis, and lupus, the presence of immunological dysregulation serves as a catalyst for persistent inflammation and subsequent tissue damage (Torun et al., 2021). On the other hand, a compromised immune response can facilitate the uncontrolled progression of diseases, such as HIV and specific types of malignancies. Furthermore, immunological dysregulation has been identified as a potential factor in the pathogenesis of various conditions, including allergies, asthma, and metabolic disorders such as diabetes (Vijay & Valdes, 2022; Zhang et al., 2021).

This study examines the microbial makeup of adult Iraqis to determine if their gut microbiota patterns differ. Second, we want to study the host immunological response in a consistent population to understand this group's immune processes. Finally, we aim to understand the complicated links between gut microbiota diversity and immunological responses in adult Iraqis.

METHODOLOGY

The present work utilizes a meticulous observational cross-sectional study design to thoroughly examine the complex association between gut microbiota and the immune system in a varied group of Iraqi adults. The study involved a sample of 80 Iraqi adults for the purpose of assessing this correlation.

Inclusion and Exclusion criteria:

Individuals must be 18–65, which includes Iraqi adults, to be eligible. The applicants must be Iraqi residents and represent the country's diverse geography and ethnicity. People must give informed consent to participate in the research, understanding its goals and methods. Physical health and cognitive aptitude are essential for understanding and answering survey questions. Dietary patterns, lifestyles, and medical backgrounds are purposely broad to include many aspects that may affect gut microbiota and immune response. Exclusion of individuals from participation will be implemented in cases where their age falls outside the designated range of 18 to 65 years. Since the study's major goal is to study gut microbiota-immune system interactions in Iraqi adults, non-Iraqis will be excluded. In addition, people with cognitive disorders or other barriers to informed consent will be excluded. People with medical issues that might prevent them from participating will be removed from the study to improve data collection and integrity. This study will also exclude participants with physical limitations that prevent them from following sample collection protocols. This study uses a carefully designed survey instrument to collect data on various parameters that may affect gut flora and immune response. Demographic factors including age, gender, and location are included. Lifestyle issues include

smoking and supplement use. The survey will be pilot tested with a small number of participants to ensure clarity, relevance, and cultural appropriateness. Based on valuable feedback, adjustments will be made. Fecal samples will be collected and stored at -80°C to study the microbiota. A cold chain system will convey these samples to the lab safely, where microbiota will be examined.

In parallel, trained phlebotomists will collect blood samples, which will then undergo processing to extract serum and plasma. Subsequently, these samples will be subjected to analysis utilizing enzyme-linked immunosorbent assay (ELISA) kits to measure essential immune response markers (IL6, IL8, IL10) and identify immune cell types (T cells, B cells). The correctness and reliability of the immune response data can be ensured through the rigorous implementation of standardized laboratory processes and the implementation of strong quality control measures. The collected data, encompassing demographics, food patterns, lifestyle characteristics, and medical history, will be subjected to thorough statistical analysis in order to investigate potential correlations between gut microbiota composition and immune response markers. The suitable statistical tests such as chi-square, t-tests, and regression will be applied. To assess the data, we employed Student's Chi-square test, taking into account statistical significance at $P \leq 0.05$. The data analysis was performed with the SPSS Version 25.0 program.

RESULTS AND DISCUSSION

Correlation between age and different gut-microbiota prevalence:

The findings in table 1 reveal intricate trends in microbial makeup across different age groups. There is a greater occurrence of *Lactobacillus* and *Bifidobacterium* in individuals aged 18-30, with a steady decline observed as age increases. In contrast, it is observed that *Colistridium* has a divergent trend, characterized by a greater incidence among individuals in the older adult population (specifically, the 51-60 age group). Of particular significance is the observed prevalence of *Escherichia coli* (*E. coli*), which exhibits a compelling upward trend in association with advancing age. The statistical studies done demonstrate significant relationships, as evidenced by p-values ranging from 0.00009 to less than 0.00001.

Table 1: Gut microbiota correlation to age.

		18-30	31-50	51-60	P-value
<i>Lactobacillus</i>	High	13	7	3	0.00009
	Low	8	16	33	
<i>Bifidobacterium</i>	High	14	10	7	0.001691
	Low	7	13	29	
<i>Colistridium</i>	High	9	15	30	0.00068
	Low	12	8	6	
<i>E.Coli</i>	High	7	20	34	<0.00001
	Low	14	3	2	

Different gut-microbiota prevalence correlation to gender:

The study found a gender gap in *Lactobacillus* prevalence, with females having higher levels for high prevalence and males for low prevalence. The exceptionally low p-value (<0.00001) suggests a strong link between gender and *Lactobacillus* prevalence. *Bifidobacterium* is more common to be low in men and its high prevalence is observed in females, with a statistically significant p-value of 0.022992. No gender-related differences are seen in *Colistridium* prevalence (p-value = 0.775246). *E. coli* has a gender-associated pattern, with high levels being more common in men and low levels in women. The low p-value of 0.000836 shows a gender-*E. Coli* prevalence association as demonstrated in table 2.

Table 2: Gender and gut microbiota correlation.

		Male	Female	P-value
<i>lactobacillus</i>	High	5	18	<0.00001
	Low	46	11	
<i>Bifidobacterium</i>	High	15	16	0.022992
	Low	36	13	
<i>Colistriduim</i>	High	35	19	0.775246
	Low	16	10	
<i>E.Coli</i>	High	45	16	0.000836
	Low	6	13	

Different gut-microbiota prevalence correlation to residence:

Table 3 shows that lactobacillus levels are higher in rural people for high prevalence and urban residents for low prevalence, also significance is found (p-value 0.000105). Bifidobacterium is in the same vein like lactobacillus (P value 0.000326). Colistriduim and E. coli prevalence differ between rural and urban inhabitants, with greater urban prevalence and lower rural prevalence. The low p-value of (0.000363, 0.0000148) shows a strong prevalence and residence connection.

Table 3: Gut microbiota and residence association.

		Rural	Urban	P-value
<i>lactobacillus</i>	High	14	9	0.000105
	Low	9	45	
<i>Bifidobacterium</i>	High	16	15	0.000326
	Low	7	42	
<i>Colistriduim</i>	High	5	49	0.000363
	Low	18	8	
<i>E.Coli</i>	High	11	50	0.0000148
	Low	12	7	

Different gut-microbiota prevalence correlation to education level:

Education level and Lactobacillus prevalence are substantially correlated (p=0.001347). Education level is significantly correlated with Bifidobacterium prevalence (p-value 0.000642). Colistriduim prevalence also varies by education level, with high prevalence in primary school graduates and low prevalence in secondary and university graduates. The low p-value of 0.000166 indicates a substantial link between education and Colistriduim prevalence. High levels of E. coli are more common in people with primary education, while low levels are the opposite. The extremely low p-value of 0.000011 shows a strong association between education and E. coli prevalence as table 4 illustrates.

Table 4: Education and gut microbiota correlation.

		Primary	Secondary	University	P-value
<i>lactobacillus</i>	High	6	13	5	0.001347
	Low	40	12	4	
<i>Bifidobacterium</i>	High	11	12	8	0.000642
	Low	35	13	1	
<i>Colistriduim</i>	High	39	13	2	0.000166
	Low	7	12	7	
<i>E.Coli</i>	High	43	14	4	0.000011
	Low	3	11	5	

Different gut-microbiota prevalence correlation to income level:

Table 5 show that increased income correlates with increased Lactobacillus and Bifidobacterium prevalence. The 0.000753 and 0.000023 p-values show a strong link between income and their prevalence. Income also affects Colistriduim and E.Coli prevalence. Income is significantly correlated with Colistriduim prevalence (p=0.000135). The 0.000013 p-value shows a strong association between income and E. Coli prevalence.

Table 5: correlation between income level and gut microbiota prevalence

		Poor	Moderate	High	P-value
<i>lactobacillus</i>	High	3	8	12	0.000753
	Low	33	13	11	
<i>Bifidobacterium</i>	High	4	12	15	0.000023
	Low	32	9	8	
<i>Colistriduim</i>	High	33	11	10	0.000135
	Low	3	10	13	
<i>E.Coli</i>	High	36	15	11	0.000013
	Low	0	7	12	

Different gut-microbiota prevalence correlation to daily diet:

The incidence of Lactobacillus is lowest in meat eaters and highest in vegetarians (p-value o0.00335). Vegetarians have the highest Bifidobacterium prevalence, while meat-eaters have the lowest (p-value of 0.000016). Colistriduim prevalence is higher in meat-eaters and lowest in vegetarians (p=0.0002419). E. coli prevalence is highest in meat eaters and lowest in vegetarians. An exceptionally low p-value of <0.00001 indicates a strong association between daily diet and E. coli prevalence as table 6 elucidate.

Table 6: Daily diet association with gut microbiota.

		Vegetarian	Omnivorous	Meateraian	P-value
<i>lactobacillus</i>	High	10	8	5	0.00335
	Low	7	19	31	
<i>Bifidobacterium</i>	High	12	15	4	0.000016
	Low	5	12	32	
<i>Colistriduim</i>	High	6	19	29	0.0002419
	Low	11	8	7	
<i>E.Coli</i>	High	8	20	33	<0.00001
	Low	19	7	3	

Different gut-microbiota prevalence correlation to life style habits:

Table 7 shows that Lactobacillus prevalence is statistically different between supplement users and non-users, with a p-value of 0.005221. Similarly, Bifidobacterium prevalence is significantly affected by dietary supplement use (p-value 0.01402). Colistriduim prevalence is correlated with dietary supplement use (p=0.00392). E. Coli prevalence differ by dietary supplement use, with a p-value of 0.000348. Smokers have a much lower prevalence of Lactobacillus and Bifidoacteruim than non-smokers, as shown by the highly significant p-value of less than 0.00001. Smokers have higher levels of colonitriduim and E. coli than non-smokers. The p-value of 0.000119 and <0.00001 implies that smoking increases Colistriduim and E. coli prevalence.

Table 7: dietary supplements and smoking relation to gut microbiota prevalence.

		Dietary supplements		Smoking	
		Yes	No	Yes	No
<i>Lactobacillus</i>	High	18	5	5	18
	Low	15	32	49	8
		P-Value 0.005221		P-Value <0.00001	
<i>Bifidobacterium</i>	High	22	9	11	20
	Low	21	28	43	6
		P-Value 0.01402		P-Value <0.00001	
<i>Clostridium</i>	High	23	31	44	10
	Low	20	6	10	16
		P-Value 0.00392		P-Value 0.000119	
<i>E.Coli</i>	High	26	35	50	11
	Low	17	2	4	15
		P-Value 0.000348		P-Value <0.00001	

Correlation with different gut-microbiota prevalence and interleukins levels:

Table 8 shows how gut microbiota prevalence affects interleukin levels (IL6, IL8, and IL10) in subjects. High Lactobacillus concentrations affect interleukin levels.

Results show a substantial connection between Lactobacillus prevalence and higher IL6 and lower IL10 levels ($P < 0.00001$). IL8 levels are significantly associated with normal Lactobacillus presence ($P = 0.000326$). In Bifidobacterium, greater abundance is linked to different interleukin levels. High Bifidobacterium levels raise normal IL6 ($P = 0.000088$), IL8 ($P = 0.000023$), and decreased levels of IL10 ($P = 0.000129$). An association exists between Clostridium levels and significant changes in IL6 ($P = 0.000554$) and IL10 ($P < 0.00001$) levels. Escherichia coli levels correlate with interleukin levels. High levels of E. coli are strongly correlated with changed levels of IL6, IL8, and IL10, all with p-values below 0.00001.

Table 8: Interleukins association with gut microbiota prevalence.

		IL6		IL8		IL10	
		Normal	> Normal	Normal	> Normal	Normal	< Normal
<i>Lactobacillus</i>	High	18	5	16	7	20	3
	Low	10	47	15	42	13	44
		P – Value <0.00001		P – Value 0.000326		P – Value <0.00001	
<i>Bifidobacterium</i>	High	19	12	21	10	21	10
	Low	9	40	10	39	12	37
		P – Value 0.000088		P – Value 0.000023		P – Value 0.000129	
<i>Clostridium</i>	High	12	42	17	37	12	42
	Low	16	10	18	12	21	5
		P – Value 0.000554		P – Value 0.011074		P – Value <0.00001	
<i>E.Coli</i>	High	13	48	14	47	15	46
	Low	15	4	17	2	18	1
		P – Value < 0.00001		P – Value < 0.00001		P – Value < 0.00001	

Correlation of different gut-microbiota prevalence and antibodies levels:

High levels of Lactobacillus are linked to normal Treg cells and B lymphocytes cell populations ($P < 0.00001$). A substantial link ($P < 0.00001$) exists between high Bifidobacterium levels and normal T and B cells ($P = 0.001245$). The results found strong correlations between Clostridium

levels and T and B cell changes ($P = 0.000265$ for T cells, and $P < 0.00001$ for B cells). *E. coli* significantly affects T and B cell levels ($P = 0.001906$ for T cells, $P = 0.000836$ for B cells). As table 9 illustrates.

Table 9: antibodies levels and gut microbiota prevalence.

		Treg Cells		B lymphocytes Cells	
		\geq Normal	< Normal	Normal	> Normal
<i>Lactobacillus</i>	High	17	6	21	2
	Low	10	47	8	49
		P – Value < 0.00001		P – Value < 0.00001	
<i>Bifidobacterium</i>	High	22	9	18	13
	Low	5	44	11	38
		P – Value < 0.00001		P – Value 0.001245	
<i>Colistridium</i>	High	11	43	10	44
	low	16	10	19	7
		P – Value 0.000265		P – Value < 0.00001	
<i>E.Coli</i>	High	15	46	16	45
	Low	12	7	13	6
		P – Value 0.001906		P – Value 0.000836	

Health self-assessment of participants:

Tale 10 demonstrates that few people self-reported "excellent" (8.75%) or "very good" (12.5%) health. A percentage of respondents rated their health as "good" (13.75%), "fair" 27.5%), or "poor" (37.5%). Subjective health evaluations can reveal individuals' health views and match them to gut microbiota composition and immunological responses. Some participants (12.5%) reported no sick days, while others reported 1-10 (22.5%), 11-20 (37.5%), and more than 20 (27.5%) sick days in the past year.

Table 10: Self-assessment of health of the participants.

		No.	%
Overall health	Excellent	7	8.75%
	Very good	10	12.5%
	Good	11	13.75%
	Fair	22	27.5%
	Poor	30	37.5%
Sick days last year	0 days	10	12.5%
	1-10	18	22.5%
	11-20	30	37.5%
	More than 20	22	27.5%

Discussion:

The intricate relationship between gut microbiota and the immune system in Iraqi adults is elucidated in this discussion. The findings suggest links between demographics, lifestyle, and immune responses. Age-related microbial composition variations, gender differences, and urbanization are significant. This discussion also analyzes how education and poverty affect gut microbiome. The data show how the gut and immune systems interact in Iraq.

This study found a substantial correlation between age and gut microbiome. *Lactobacillus* and *Bifidobacterium* are more common in 18-30-year-olds and decrease with age. *Clostridium* and *E. coli* have different trends, with older adults having a higher prevalence. This investigation

confirms prior findings that age increases *Clostridium* and *E. coli* prevalence (Tuomisto et al., 2019). A comprehensive review paper supports age-related gut microbiome alterations, and our study supports this. The study found that *Bifidobacterium* and *Lactobacillus* are common in young adults but decrease with age (Chandra et al., 2023).

This study found a gender-related connection between *Lactobacillus*, *Bifidobacterium*, and *E. coli*. No similar association was found with *Clostridium*. This supports previous research showing that sex strongly affects the human microbiome. Thus, "microgenderome" describes the complicated interplay between microbiota, sex hormones, and the immune system (Levy & Solt, 2018). A 2017 study found that females have more *Bifidobacterium* ($P = 0.046$) than males (Suzuki et al., 2017). Two studies found that *Lactobacillus*, a prevalent microbiota genus in women, is susceptible to female estrogens (Pelzer et al., 2012; Martin et al., 2016).

Our analysis found no significant gender correlation with *Clostridium* prevalence, which is consistent with a prior study (Zhang et al., 2021). A separate study found that male mice have more *Proteobacteria*, which includes *E. coli*, than female mice (Valeri & Endres, 2021).

The current investigation found a connection between participant residence and gut microbiota prevalence. Rural residents had more *Lactobacillus* and *Bifidobacterium*, while urban individuals had more *Clostridium* and *E. coli*. Microbial composition and urbanization are linked, as shown by previous research (Naito et al., 2019). Urbanization decreased *Actinobacteria* (*Bifidobacterium*) but boosted *Proteobacteria* (*E. coli*), according to a study. *Lactobacillus* is more abundant in rural areas than *Clostridium* in urban areas, according to another study (Lu et al., 2021). An analysis particularly found a higher percentage abundance of *Lactobacillus* in rural communities, supporting our findings (Aslam, 2022).

In this investigation, we found a strong connection between gut microbiota species and education levels, with differing prevalence patterns across people of different income and education levels. These findings support research linking gut microbiome to socioeconomic factors. One study found differences in gut microbiota prevalence between low-to-middle-income and high-income countries (Ecklu-Mensah et al., 2023). Another study showed *Clostridium* is common in middle- and low-income countries (Tayyib et al., 2023). Among 2022 studies, low *Bifidobacterium* levels were found among low-income and low-educated people, emphasizing the link between gut microbiota and socioeconomic position (Lapidot et al., 2022). A UK study discovered reduced *Lactobacilli* levels among low-income and low-educated people (Bowyer et al., 2019).

Our investigation found a link between dietary preferences and gut microbiota prevalence. The vegetarian group had more *Lactobacillus* and *Bifidobacterium*, while the meat-eating group had more *Clostridium* and *E. coli*. These findings support previous research linking food to gut microbial makeup. Vegan diets enhanced *Bifidobacterium* and *Lactobacillus* and decreased *Clostridium*, according to a study. The study found that a vegetarian/vegan diet promotes a rich ecology of beneficial bacteria that maintain the gut microbiome and wellness (Tomova et al., 2019). Another study found that a high-fiber diet, like vegetarian and vegan diets, lowers colonic pH, limiting *E. coli* growth. This suggests that diet, especially fiber-rich foods, may shape gut microbiota and improve microbial conditions (Scott et al., 2008).

In our study, smoking was linked to elevated gut *Clostridium* and *E. coli* levels. Conversely, *Lactobacillus* and *Bifidobacterium* are more prevalent in people who take probiotics. This supports other studies showing that probiotics and other dietary supplements help improve gut flora (Li et al., 2019). Probiotic supplements boost intestinal microbiota variety and good bacteria like *Lactobacillus* and *Bifidobacterium*, according to an investigation. This study also found a decline in opportunistic pathogenic bacteria, supporting our findings of high prevalence in *Clostridium* and *E. coli* (Yang et al., 2020). By raising *Bifidobacterium* levels, a 2020 study validated the gut microbiota benefits of dietary supplements. Another study linked dietary

supplement use to high *Bifidobacterium* and *Lactobacillus* levels (Yu et al., 2019). Research also shows that dietary supplements help prevent *Clostridium* and *E. coli* (Sun et al., 2020).

In 2022, Kumar linked smoking to gut microbiota dysbiosis. Smoking causes dysbiosis, according to studies (Kumar & Kumar, 2022). In mice not exposed to cigarettes, *Lactobacillus* and *Bifidobacterium* were abundant (Zhong et al., 2022). This supports 2022 human research that found lower *Bifidobacterium* abundances in smokers than non-smokers (Yang et al., 2022).

Gut microbiota and interleukin levels—both pro-inflammatory and anti-inflammatory—are complexly linked. Our study found that distinct gut microbiota species correspond with varied interleukin levels, demonstrating a key relationship between microbial makeup and immune responses. IL-6 and IL-8 concentrations increased significantly in low *Lactobacillus* prevalence, elucidated in a study in 2015 (Carasi et al., 2015). This matches another study that related lower *Lactobacillus* spp. levels to higher pro-inflammatory cytokines (IL-6 and IL-8) and higher anti-inflammatory IL-10 cytokine levels. These findings suggest *Lactobacillus* may modulate immune response through anti-inflammatory effects (Rastogi & Singh, 2022). A study found that a microflora deficit, characterized by low levels of lactobacilli and bifidobacteria, led to elevated levels of pro-inflammatory cytokines such as IL-1 β , IL-6, and IL-8. This shows that fewer helpful bacteria may cause intestinal inflammation (Kondratiuk et al., 2020).

Another notable finding in our study was that a high prevalence of dangerous *E. coli* was related to a considerable rise in pro-inflammatory IL-6 and a significant decrease in anti-inflammatory IL-10 (He et al., 2022). A study found that low *Clostridium*, mainly *Clostridium*, levels caused pro-inflammatory effects. This shows that *Clostridium* abundance may reduce pro-inflammatory responses (Yusufu et al., 2021). Positive bacteria like *Lactobacillus* and *Bifidobacterium* may control interleukin levels, promoting a balanced immune response. Conversely, reducing dangerous bacteria like *E. coli* may lessen inflammation.

Our investigation found that beneficial and detrimental gut microbiota species affect B lymphocytes and regulatory T cells. Gut health and immunological modulation depend on bacterial strains and immune cell populations, especially Tregs (Sun et al., 2020). Studies have shown that beneficial gut microbiota species like *Bifidobacterium* and *Lactobacillus* regulate the immune system, notably Tregs. *Bifidobacterium* and *Lactobacillus* change the gut microbiome, encouraging regulatory T cell responses. Probiotic *Lactobacillus* increases Treg cells (Kazmierczak-Siedlecka et al., 2021). Our study and a 2023 study found a positive correlation between *Bifidobacterium* and Treg cell responses (Oh et al., 2023). Studies in germ-free mice have illuminated how gut microbiome affects Treg cell populations. In germ-free mice, beneficial gut microbiome colonization increases Treg cell populations. However, exposure to damaging and pathogenic germs had the opposite effect, demonstrating the importance of microbial colonization in Treg cell dynamics (Atarashi et al., 2011). Treg cell inhibition of microbial-induced intestinal inflammation is essential in research (Ning et al., 2022). Treg cells control microbial-induced inflammation and reduce overactive immune responses. This highlights Treg cells' role in intestinal homeostasis and inflammation prevention.

The 2022 study found a drop in B lymphocytes with a high prevalence of probiotic gut microbiota, raising issues regarding the complicated relationships between individual microbial strains and immune cell populations (Tiwari, 2022). An investigation reported that gut microbiota activates T cells to cause immunological diseases highlights the intestinal immune system's delicate balance between commensal flora tolerance and pathogenic organism response. Dysbiosis may cause immunological dysregulation and inflammatory disorders by increasing T cells in response to pathogenic and toxic bacteria (Ma et al., 2019).

The current study found substantial relationships between lifestyle factors, microbial makeup, immunological markers, and health status. Many individuals smoked, ate meat, had low socioeconomic position, primary education, and had high levels of dangerous bacteria such

Colistridium and E. coli. Beneficial bacteria like Lactobacillus and Bifidobacterium were less common.

The immune system reflected this microbial imbalance, with most subjects having high pro-inflammatory cytokine levels and below-normal anti-inflammatory levels. Most subjects had low Tregs and high B lymphocytes.

These findings led to a health status assessment that showed 37.5% of individuals were in poor health and 8.75% were in great health. This result is significant in relation to interleukins and antibodies because it suggests that gut microbiota makeup may affect immunological responses and health.

One indicator was the number of sick days recorded, with 27.5% of individuals having more than 20 and 12.5% having none. This gap shows a link between immunological indicators, microbial composition, and health status, highlighting the complex interaction between lifestyle, gut microbiota, and well-being.

CONCLUSION

This study shows complex relationships between gut microbiota prevalence and demographic, lifestyle, and health characteristics in Iraqi people. Age, gender, residence, education, income, food, and smoking practices correlated with microbiological profiles. The study ties gut microbiota to immunological responses by examining interleukin levels and T and B cell antibody populations. These data highlight the complex relationship between gut microbiota, lifestyle, and immunological parameters.

REFERENCES

- Al-Banna, N. A., Cyprian, F., & Albert, M. J. (2018). Cytokine responses in campylobacteriosis: Linking pathogenesis to immunity. *Cytokine & Growth Factor Reviews*, 41, 75-87. <https://doi.org/10.1016/j.cytogfr.2018.03.002>
- Ameen, A., Amir, A., Aziz, M. N., & Irshad, A. (2023). Dysfunction of the gut microbiome and its onset progression of chronic and mental health disorders. *Pakistan Journal of Medical & Health Sciences*, 17(05), 425. <https://doi.org/10.53350/pjmhs2023175425>
- Andersson, O. P. (2023). Is the microbiome the cause of irritable bowel syndrome and inflammatory bowel disease? Lessons to consider from odontology. *International Journal of Colorectal Disease*, 38(1), 117. <https://doi.org/10.1007/s00384-022-04300-3>
- Aron-Wisnewsky, J., Warmbrunn, M. V., Nieuwdorp, M., & Clément, K. (2021). Metabolism and metabolic disorders and the microbiome: The intestinal microbiota associated with obesity, lipid metabolism, and metabolic health—Pathophysiology and therapeutic strategies. *Gastroenterology*, 160(2), 573-599. <https://doi.org/10.1053/j.gastro.2020.10.050>
- Aslam, M. (2022). The demographic and geographic variations mediate fecal and oral microbial dynamics alteration in human population of District Faisalabad. *Journal of Basic Microbiology*. <https://doi.org/10.1002/jobm.202200148>
- Atarashi, K., Tanoue, T., Shima, T., Imaoka, A., Kuwahara, T., Momose, Y., Cheng, G., Yamasaki, S., Saito, T., Ohba, Y., & Taniguchi, T. (2011). Induction of colonic regulatory T cells by indigenous *Clostridium* species. *Science*, 331(6015), 337-341. <https://doi.org/10.1126/science.1198469>
- Aziz, T., Khan, A. A., Tzora, A., Voidarou, C., & Skoufos, I. (2023). Dietary implications of the bidirectional relationship between the gut microflora and inflammatory diseases with special emphasis on irritable bowel disease: Current and future perspective. *Nutrients*, 15(13), 2956. <https://doi.org/10.3390/nu15132956>

- Banaszak, M., Górna, I., Woźniak, D., Przysławski, J., & Drzymała-Czyż, S. (2023). Association between gut dysbiosis and the occurrence of SIBO, LIBO, SIFO, and IMO. *Microorganisms*, 11(3), 573. <https://doi.org/10.3390/microorganisms11030573>
- Bhargava, S., Merckelbach, E., Noels, H., Vohra, A., & Jankowski, J. (2022). Homeostasis in the gut microbiota in chronic kidney disease. *Toxins*, 14(10), 648. <https://doi.org/10.3390/toxins14100648>
- Bowyer, R. C., Jackson, M. A., Le Roy, C. I., Ni Lochlainn, M., Spector, T. D., Dowd, J. B., & Steves, C. J. (2019). Socioeconomic status and the gut microbiome: A TwinsUK cohort study. *Microorganisms*, 7(1), 17. <https://doi.org/10.3390/microorganisms7010017>
- Carasi, P., Racedo, S. M., Jacquot, C., Romanin, D. E., Serradell, M. A., & Urdaci, M. C. (2015). Impact of kefir-derived *Lactobacillus kefir* on the mucosal immune response and gut microbiota. *Journal of Immunology Research*, 2015, 1-11. <https://doi.org/10.1155/2015/361604>
- Chandra, S., Sisodia, S. S., & Vassar, R. J. (2023). The gut microbiome in Alzheimer's disease: What we know and what remains to be explored. *Molecular Neurodegeneration*, 18(1), 1-21. <https://doi.org/10.1186/s13024-023-00603-3>
- Chang, E. B., & Martinez-Guryn, K. (2019). Small intestinal microbiota: The neglected stepchild needed for fat digestion and absorption. *Gut Microbes*, 10(2), 235-240. <https://doi.org/10.1080/19490976.2018.1470006>
- Croese, T., Castellani, G., & Schwartz, M. (2021). Immune cell compartmentalization for brain surveillance and protection. *Nature Immunology*, 22(9), 1083-1092. <https://doi.org/10.1038/s41590-021-00971-4>
- Das, B., & Nair, G. B. (2019). Homeostasis and dysbiosis of the gut microbiome in health and disease. *Journal of Biosciences*, 44(1), 1-8. <https://doi.org/10.1007/s12038-019-9902-5>
- Ecklu-Mensah, G., Choo-Kang, C., Maseng, M. G., Donato, S., Bovet, P., Viswanathan, B., Bedu-Addo, K., Plange-Rhule, J., Oti Boateng, P., Forrester, T. E., & Williams, M. (2023). Gut microbiota and fecal short chain fatty acids differ with adiposity and country of origin: The METS-microbiome study. *Nature Communications*, 14(1), 5160. <https://doi.org/10.1038/s41467-023-41091-8>
- Espirito Santo, C., Caseiro, C., Martins, M. J., Monteiro, R., & Brandão, I. (2021). Gut microbiota, in the halfway between nutrition and lung function. *Nutrients*, 13(5), 1716. <https://doi.org/10.3390/nu13051716>
- Han, Y., Jia, Q., Jahani, P. S., Hurrell, B. P., Pan, C., Huang, P., Gukasyan, J., Woodward, N. C., Eskin, E., Gilliland, F. D., & Akbari, O. (2020). Genome-wide analysis highlights contribution of immune system pathways to the genetic architecture of asthma. *Nature Communications*, 11(1), 1776. <https://doi.org/10.1038/s41467-020-15592-1>
- He, L., Wang, C., Simujide, H., Aricha, H., Zhang, J., Liu, B., Zhang, C., Cui, Y., & Aorigele, C. (2022). Effect of early pathogenic *Escherichia coli* infection on the intestinal barrier and immune function in newborn calves. *Frontiers in Cellular and Infection Microbiology*, 12, 173. <https://doi.org/10.3389/fcimb.2022.826898>
- Hill, D. L., Carr, E. J., Rutishauser, T., Moncunill, G., Campo, J. J., Innocentin, S., Mpina, M., Nhabomba, A., Tumbo, A., Jairoce, C., & Moll, H. A. (2020). Immune system development varies according to age, location, and anemia in African children. *Science Translational Medicine*, 12(529), eaaw9522. <https://doi.org/10.1126/scitranslmed.aaw9522>
- Hoden, B., DeRubeis, D., Martinez-Moczygemba, M., Ramos, K. S., & Zhang, D. (2022). Understanding the role of toll-like receptors in lung cancer immunity and immunotherapy. *Frontiers in Immunology*, 13, 1033483. <https://doi.org/10.3389/fimmu.2022.1033483>
- Hou, K., Wu, Z. X., Chen, X. Y., Wang, J. Q., Zhang, D., Xiao, C., Zhu, D., Koya, J. B., Wei, L., Li, J., & Chen, Z. S. (2022). Microbiota in health and diseases. *Signal Transduction and Targeted Therapy*, 7(1), 135. <https://doi.org/10.1038/s41392-022-00997-0>

- Kazmierczak-Siedlecka, K., Roviello, G., Catalano, M., & Polom, K. (2021). Gut microbiota modulation in the context of immune-related aspects of *Lactobacillus* spp. and *Bifidobacterium* spp. in gastrointestinal cancers. *Nutrients*, 13(8). <https://doi.org/10.3390/nu13082686>
- Kita, H. (2022). How are airborne allergens remembered by the immune system? *Journal of Allergy and Clinical Immunology*, 149(6), 1940-1942. <https://doi.org/10.1016/j.jaci.2021.11.028>
- Kogut, M. H., Lee, A., & Santin, E. (2020). Microbiome and pathogen interaction with the immune system. *Poultry Science*, 99(4), 1906-1913. <https://doi.org/10.1016/j.psj.2019.12.046>
- Kondratiuk, V. E., Tarasenko, O. M., Karmazina, O. M., & Taranchuk, V. V. (2020). Impact of the synbiotics and urate-lowering therapy on gut microbiota and cytokine profile in patients with chronic gouty arthritis. *Journal of Medicine and Life*, 13(4), 490-498. <https://doi.org/10.25122/jml-2020-0044>
- Kumar, S., & Kumar, A. (2022). Microbial pathogenesis in inflammatory bowel diseases. *Microbial Pathogenesis*, 163, 105383. <https://doi.org/10.1016/j.micpath.2021.105383>
- Lapidot, Y., Reshef, L., Maya, M., Cohen, D., Gophna, U., & Muhsen, K. (2022). Socioeconomic disparities and household crowding in association with the fecal microbiome of school-age children. *NPJ Biofilms and Microbiomes*, 8(1), 10. <https://doi.org/10.1038/s41522-022-00295-y>
- Levy, G., & Solt, I. (2018). The human microbiome and gender medicine. *Gender and the Genome*, 2(4), 123-127. <https://doi.org/10.1177/2470289718810965>
- Li, H., Zhou, Y., Ling, H., Luo, L., Qi, D., & Feng, L. (2019). The effect of dietary supplementation with *Clostridium butyricum* on the growth performance, immunity, intestinal microbiota and disease resistance of tilapia (*Oreochromis niloticus*). *PLoS One*, 14(12), e0223428. <https://doi.org/10.1371/journal.pone.0223428>
- Lobionda, S., Sittipo, P., Kwon, H. Y., & Lee, Y. K. (2019). The role of gut microbiota in intestinal inflammation with respect to diet and extrinsic stressors. *Microorganisms*, 7(8), 271. <https://doi.org/10.3390/microorganisms7080271>
- Lobionda, S., Sittipo, P., Kwon, H. Y., & Lee, Y. K. (2019). The role of gut microbiota in intestinal inflammation with respect to diet and extrinsic stressors. *Microorganisms*, 7(8), 271. <https://doi.org/10.3390/microorganisms7080271>
- Lu, J., Zhang, L., Zhai, Q., Zhao, J., Zhang, H., Lee, Y. K., Lu, W., Li, M., & Chen, W. (2021). Chinese gut microbiota and its associations with staple food type, ethnicity, and urbanization. *NPJ Biofilms and Microbiomes*, 7(1), 71. <https://doi.org/10.1038/s41522-021-00233-y>
- Ma, H., Tao, W., & Zhu, S. (2019). T lymphocytes in the intestinal mucosa: Defense and tolerance. *Cellular & Molecular Immunology*, 16(3), 216-224. <https://doi.org/10.1038/s41423-018-0021-6>
- Mahmud, M. A. (2022). Assessing factors that shape neonatal gut microbiota in Erbil Province, Iraq. *Jordan Journal of Biological Sciences*, 15(3), 383-390. <https://doi.org/10.54319/jjbs/150311>
- Martin, R., Makino, H., Cetinyurek Yavuz, A., Ben-Amor, K., Roelofs, M., Ishikawa, E., Kubota, H., Swinkels, S., Sakai, T., Oishi, K., & Kushiro, A. (2016). Early-life events, including mode of delivery and type of feeding, siblings and gender, shape the developing gut microbiota. *PLoS One*, 11(6), e0158498. <https://doi.org/10.1371/journal.pone.0158498>
- Martinez, J. E., Kahana, D. D., Ghuman, S., Wilson, H. P., Wilson, J., Kim, S. C., Lagishetty, V., Jacobs, J. P., Sinha-Hikim, A. P., & Friedman, T. C. (2021). Unhealthy lifestyle and gut dysbiosis: A better understanding of the effects of poor diet and nicotine on the intestinal microbiome. *Frontiers in Endocrinology*, 12, 667066. <https://doi.org/10.3389/fendo.2021.667066>
- Mörbe, U. M., Jørgensen, P. B., Fenton, T. M., von Burg, N., Riis, L. B., Spencer, J., & Agace, W. W. (2021). Human gut-associated lymphoid tissues (GALT): Diversity, structure, and function. *Mucosal Immunology*, 14(4), 793-802. <https://doi.org/10.1038/s41385-021-00370-3>
- Mousa, W. K., Chehadeh, F., & Husband, S. (2022). Microbial dysbiosis in the gut drives systemic autoimmune diseases. *Frontiers in Immunology*, 13, 906258. <https://doi.org/10.3389/fimmu.2022.906258>

- Naito, Y., Takagi, T., Inoue, R., Kashiwagi, S., Mizushima, K., Tsuchiya, S., Itoh, Y., Okuda, K., Tsujimoto, Y., Adachi, A., & Maruyama, N. (2019). Gut microbiota differences in elderly subjects between rural city Kyotango and urban city Kyoto: An age-gender-matched study. *Journal of Clinical Biochemistry and Nutrition*, 65(2), 125-131. <https://doi.org/10.3164/jcbrn.18-124>
- Nikolenko, V. N., Oganessian, M. V., Sankova, M. V., Bulygin, K. V., Vovkogan, A. D., Rizaeva, N. A., & Sinelnikov, M. Y. (2021). Paneth cells: Maintaining dynamic microbiome-host homeostasis, protecting against inflammation and cancer. *BioEssays*, 43(3), 2000180. <https://doi.org/10.1002/bies.202000180>
- Ning, X., Lei, Z., Rui, B., Li, Y., & Li, M. (2022). Gut microbiota promotes immune tolerance by regulating ROR γ t⁺ Treg cells in food allergy. *Advanced Gut & Microbiome Research*, 2022. <https://doi.org/10.1002/agm2.12118>
- Oh, S. H., Kim, I. S., Kim, G. I., Kim, J. A., Moon, Y. S., Jang, J. C., Lee, S. S., Jung, J. H., Park, H. C., & Cho, K. K. (2023). Effects of lactic acid bacteria fermented feed and three types of lactic acid bacteria (*L. plantarum*, *L. acidophilus*, *B. animalis*) on intestinal microbiota and T cell polarization (Th1, Th2, Th17, Treg) in the intestinal lymph nodes and spleens of rats. *Animal Bioscience*, 36(1), 156-164. <https://doi.org/10.5713/ab.22.0301>
- Okeke, E. B., & Uzonna, J. E. (2019). The pivotal role of regulatory T cells in the regulation of innate immune cells. *Frontiers in Immunology*, 10, 680. <https://doi.org/10.3389/fimmu.2019.00680>
- Pascale, A., Marchesi, N., Govoni, S., Coppola, A., & Gazzaruso, C. (2019). The role of gut microbiota in obesity, diabetes mellitus, and effect of metformin: New insights into old diseases. *Current Opinion in Pharmacology*, 49, 1-5. <https://doi.org/10.1016/j.coph.2019.03.011>
- Pelzer, E. S., Allan, J. A., Theodoropoulos, C., Ross, T., Beagley, K. W., & Knox, C. L. (2012). Hormone-dependent bacterial growth, persistence and biofilm formation—A pilot study investigating human follicular fluid collected during IVF cycles. *PLoS One*, 7(12), e49965. <https://doi.org/10.1371/journal.pone.0049965>
- Pengpid, S., & Peltzer, K. (2021). Overweight and obesity among adults in Iraq: Prevalence and correlates from a national survey in 2015. *International Journal of Environmental Research and Public Health*, 18(8), 4198. <https://doi.org/10.3390/ijerph18084198>
- Pulikkan, J., Mazumder, A., & Grace, T. (2019). Role of the gut microbiome in autism spectrum disorders. In *Reviews on Biomarker Studies in Psychiatric and Neurodegenerative Disorders* (pp. 253-269). https://doi.org/10.1007/978-3-030-18230-3_11
- Pushpanathan, P., Mathew, G. S., Selvarajan, S., Seshadri, K. G., & Srikanth, P. (2019). Gut microbiota and its mysteries. *Indian Journal of Medical Microbiology*, 37(2), 268-277. https://doi.org/10.4103/ijmm.IJMM_19_387
- Radek, K., & Gallo, R. (2007). Antimicrobial peptides: Natural effectors of the innate immune system. In *Seminars in Immunopathology* (Vol. 29, pp. 27-43). Springer. <https://doi.org/10.1007/s00281-007-0064-3>
- Rastogi, S., & Singh, A. (2022). Gut microbiome and human health: Exploring how the probiotic genus *Lactobacillus* modulates immune responses. *Frontiers in Pharmacology*, 13, 1042189. <https://doi.org/10.3389/fphar.2022.1042189>
- Ruff, W. E., Greiling, T. M., & Kriegel, M. A. (2020). Host–microbiota interactions in immune-mediated diseases. *Nature Reviews Microbiology*, 18(9), 521-538. <https://doi.org/10.1038/s41579-020-0367-1>
- Scott, K. P., Duncan, S. H., & Flint, H. J. (2008). Dietary fibre and the gut microbiota. *Nutrition Bulletin*, 33(3), 201-211. <https://doi.org/10.1111/j.1467-3010.2008.00706.x>
- Segal, J. P., Mak, J. W., Mullish, B. H., Alexander, J. L., Ng, S. C., & Marchesi, J. R. (2020). The gut microbiome: An under-recognised contributor to the COVID-19 pandemic? *Therapeutic Advances in Gastroenterology*, 13, 1756284820974914. <https://doi.org/10.1177/1756284820974914>

- Sun, Q., Zhang, S., Liu, X., Huo, Y., Su, B., & Li, X. (2020). Effects of a probiotic intervention on *Escherichia coli* and high-fat diet-induced intestinal microbiota imbalance. *Applied Microbiology and Biotechnology*, 104(3), 1243-1257. <https://doi.org/10.1007/s00253-019-10300-4>
- Sun, S., Luo, L., Liang, W., Yin, Q., Guo, J., Rush, A. M., Lv, Z., Liang, Q., Fischbach, M. A., Sonnenburg, J. L., & Dodd, D. (2020). *Bifidobacterium* alters the gut microbiota and modulates the functional metabolism of T regulatory cells in the context of immune checkpoint blockade. *Proceedings of the National Academy of Sciences*, 117(44), 27509-27515. <https://doi.org/10.1073/pnas.2015622117>
- Suzuki, Y., Ikeda, K., Sakuma, K., Kawai, S., Sawaki, K., Asahara, T., Takahashi, T., Tsuji, H., Nomoto, K., Nagpal, R., & Wang, C. (2017). Association between yogurt consumption and intestinal microbiota in healthy young adults differs by host gender. *Frontiers in Microbiology*, 8, 847. <https://doi.org/10.3389/fmicb.2017.00847>
- Tang, T. W., Chen, H. C., Chen, C. Y., Yen, C. Y., Lin, C. J., Prajnamitra, R. P., Chen, L. L., Ruan, S. C., Lin, J. H., Lin, P. J., & Lu, H. H. (2019). Loss of gut microbiota alters immune system composition and cripples postinfarction cardiac repair. *Circulation*, 139(5), 647-659. <https://doi.org/10.1161/CIRCULATIONAHA.118.035235>
- Tayyib, H. M., Ali, A., Jabeen, S., Kamran, H., Bajaber, M. A., Usman, M., & Zhang, X. (2023). Restoration of gut dysbiosis through *Clostridium butyricum* and magnesium possibly balances blood glucose levels: An experimental study. *Nutrients*, 15(7), 1635. <https://doi.org/10.3390/nu15071635>
- Tiwari, S. K. (2022). Bacteriocin-producing probiotic lactic acid bacteria in controlling dysbiosis of the gut microbiota. *Frontiers in Cellular and Infection Microbiology*, 12, 851140. <https://doi.org/10.3389/fcimb.2022.851140>
- Tomova, A., Bukovsky, I., Rembert, E., Yonas, W., Alwarith, J., Barnard, N. D., & Kahleova, H. (2019). The effects of vegetarian and vegan diets on gut microbiota. *Frontiers in Nutrition*, 6, 47. <https://doi.org/10.3389/fnut.2019.00047>
- Torun, A., Hupalowska, A., Trzonkowski, P., Kierkus, J., & Pyrzynska, B. (2021). Intestinal microbiota in common chronic inflammatory disorders affecting children. *Frontiers in Immunology*, 12, 642166. <https://doi.org/10.3389/fimmu.2021.642166>
- Tuomisto, S., Huhtala, H., Martiskainen, M., Goebeler, S., Lehtimäki, T., & Karhunen, P. J. (2019). Age-dependent association of gut bacteria with coronary atherosclerosis: Tampere Sudden Death Study. *PLoS One*, 14(8), e0221345. <https://doi.org/10.1371/journal.pone.0221345>
- Valeri, F., & Endres, K. (2021). How biological sex of the host shapes its gut microbiota. *Frontiers in Neuroendocrinology*, 61, 100912. <https://doi.org/10.1016/j.yfrne.2020.100912>
- van der Hee, B., & Wells, J. M. (2021). Microbial regulation of host physiology by short-chain fatty acids. *Trends in Microbiology*, 29(8), 700-712. <https://doi.org/10.1016/j.tim.2021.03.001>
- Vijay, A., & Valdes, A. M. (2022). Role of the gut microbiome in chronic diseases: A narrative review. *European Journal of Clinical Nutrition*, 76(4), 489-501. <https://doi.org/10.1038/s41430-021-00991-7>
- Wang, R., Tang, R., Li, B., Ma, X., Schnabl, B., & Tilg, H. (2021). Gut microbiome, liver immunology, and liver diseases. *Cellular & Molecular Immunology*, 18(1), 4-17. <https://doi.org/10.1038/s41423-020-00569-1>
- Wiertsema, S. P., van Bergenhenegouwen, J., Garssen, J., & Knippels, L. M. (2021). The interplay between the gut microbiome and the immune system in the context of infectious diseases throughout life and the role of nutrition in optimizing treatment strategies. *Nutrients*, 13(3), 886. <https://doi.org/10.3390/nu13030886>
- Yamamoto, M., & Aizawa, R. (2021). Maintaining a protective state for human periodontal tissue. *Periodontology 2000*, 86(1), 142-156. <https://doi.org/10.1111/prd.12382>

- Yang, H. T., Xiu, W. J., Liu, J. K., Yang, Y., Zhang, Y. J., Zheng, Y. Y., Wu, T. T., Hou, X. G., Wu, C. X., Ma, Y. T., & Xie, X. (2022). Characteristics of the intestinal microorganisms in middle-aged and elderly patients: Effects of smoking. *ACS Omega*, 7(2), 1628-1638. <https://doi.org/10.1021/acsomega.1c06318>
- Yang, Q., Liang, Q., Balakrishnan, B., Belobrajdic, D. P., Feng, Q. J., & Zhang, W. (2020). Role of dietary nutrients in the modulation of gut microbiota: A narrative review. *Nutrients*, 12(2), 381. <https://doi.org/10.3390/nu12020381>
- Yu, M., Li, Z., Chen, W., Wang, G., Cui, Y., & Ma, X. (2019). Dietary supplementation with citrus extract altered the intestinal microbiota and microbial metabolite profiles and enhanced the mucosal immune homeostasis in yellow-feathered broilers. *Frontiers in Microbiology*, 10, 2662. <https://doi.org/10.3389/fmicb.2019.02662>
- Yusufu, I., Ding, K., Smith, K., Wankhade, U. D., Sahay, B., Patterson, G. T., Pacholczyk, R., Adusumilli, S., Hamrick, M. W., Hill, W. D., & Isales, C. M. (2021). A tryptophan-deficient diet induces gut microbiota dysbiosis and increases systemic inflammation in aged mice. *International Journal of Molecular Sciences*, 22(9), 5005. <https://doi.org/10.3390/ijms22095005>
- Zhang, L., Cao, H., Li, L., Zhao, W., & Zhang, F. (2022). Oral and external intervention on the crosstalk between microbial barrier and skin via foodborne functional component. *Journal of Functional Foods*, 92, 105075. <https://doi.org/10.1016/j.jff.2022.105075>
- Zhang, X., Zhong, H., Li, Y., Shi, Z., Ren, H., Zhang, Z., Zhou, X., Tang, S., Han, X., Lin, Y., & Yang, F. (2021). Sex- and age-related trajectories of the adult human gut microbiota shared across populations of different ethnicities. *Nature Aging*, 1(1), 87-100. <https://doi.org/10.1038/s43587-020-00015-2>
- Zhang, Z. J., Huang, M. F., Qiu, L. F., Song, R. H., Zhang, Z. X., Ding, Y. W., Zhou, X., & Zhang, X. (2021). Diversity and functional analysis of Chinese bumblebee gut microbiota reveal the metabolic niche and antibiotic resistance variation of *Gilliamella*. *Insect Science*, 28(2), 302-314. <https://doi.org/10.1111/1744-7917.12789>
- Zhong, L., Qin, L., Ding, X., Ma, L., Wang, Y., Liu, M., Chen, H., Yan, H., & Song, L. (2022). The regulatory effect of fermented black barley on the gut microbiota and metabolic dysbiosis in mice exposed to cigarette smoke. *Food Research International*, 157, 111465. <https://doi.org/10.1016/j.foodres.2022.111465>
- Ahmed, A., & Sonnenberg, G. F. (2023). Immunologic regulation of health and inflammation in the intestine. In *Pediatric Inflammatory Bowel Disease* (pp. 15-32). Cham: Springer International Publishing. https://doi.org/10.1007/978-3-031-08244-4_2