FLUORIDE
Quarterly Journal of The International Society for Fluoride Research Inc.

Fluoride Toxicity and Gut Health: Targeting Gut Microbiome and Barrier Function

Unique digital address (Digital object identifier [DOI] equivalent):
https://www.fluorideresearch.online/epub/files/324.pdf

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Accepted: 2025 Feb 19.
Published as e324: 202% Feb 20.

ABSTRACT

Fluoride exposure, especially at high doses, can disrupt the gut microbiome by promoting the growth of harmful bacteria while diminishing beneficial microbes, potentially leading to gastrointestinal and systemic health issues. While low fluoride levels may have negligible effects on microbial diversity, excessive fluoride intake can significantly alter gut flora composition, triggering metabolic disruptions and impairing gut barrier function. The review aims to understand how these changes can result in increased intestinal permeability, damage to the epithelial lining, and developing conditions such as leaky gut, where toxins and harmful bacteria pass through the compromised gut lining into the bloodstream. Furthermore, fluoride exposure induces inflammation and oxidative stress, which can aggravate gut dysfunction by affecting the integrity of tight junction proteins essential for maintaining the intestinal barrier. Altered calcium ion homeostasis caused by fluoride also destabilizes these tight junctions and increases intestinal permeability. To address this review, extensive search of peer-reviewed articles was conducted. This review explores the diverse effects of fluoride on gastrointestinal health, focusing on its impact on microbial diversity, intestinal permeability, and inflammatory responses. The evidence suggests that excessive fluoride exposure may lead to significant disruptions in gut health, with long-term consequences such as chronic inflammation, metabolic disturbances, and compromised immune function. A knowledge gap exists in the understanding molecular mechanisms behind these changes. Given the growing concern over fluoride's role in health, particularly in communities with high fluoride exposure, further research is needed to understand its impact on the gut microbiome and overall health outcomes.

Key-words: *Fluoride; Gut microbiome; High dose; Intestinal permeability; Leaky gut; Microbial diversity*

INTRODUCTION

Organisms are increasingly exposed to various environmental toxic substances, leading to complex biological responses [1]. Fluoride (F), a common environmental contaminant, is widespread and poses significant risks to organisms. It is primarily ingested through contaminated water, drugs, toothpaste, and other sources. Drinking water, in particular, is the main route of fluoride exposure, with many countries worldwide reporting elevated fluoride levels in their groundwater [2-5]. Research has focused on fluoride's role in dental health, such as cavity prevention, and its potential toxicity at high concentrations, particularly concerning bones, kidneys, liver, and the nervous system [6-8]. Several mechanisms have been proposed to explain fluoride-induced diseases, including oxidative stress, signaling pathway alterations, cell cycle disruptions, apoptosis, and epigenetic changes [9-10]. A brief outline of fluoride toxicity in humans is shown in Figure 1. While the effects of fluoride on gut health are an emerging area of research, studies in this field remain limited. Alterations in gut microbiota have been linked to various health problems affecting key systems, including the digestive, cardiovascular, nervous, reproductive, and renal systems [10-11].

Excessive fluoride intake from drinking water can trigger immune responses, damage the structure of the caecum and rectum, inhibit the proliferation of intestinal epithelial cells, reduce glycoprotein secretion, decrease the number of goblet cells and hypertrophic cells, and alter the diversity and composition of the gut microbiome [12,13]. Disruptions in gut microbiota can compromise the integrity of the intestinal barrier, potentially leading to various diseases. Recent studies suggest that disturbances in the gut microbiota and their impact on metabolism and physiological functions play a critical role in disease development [14]. Given the known toxicity of fluoride, it is essential to investigate how high levels of fluoride exposure affect the gut microbiota, along with its metabolic and functional changes. The microbiota residing in the gastrointestinal tract plays a crucial role in maintaining overall health, and emerging evidence suggests that fluoride exposure can disrupt the balance of gut bacteria, potentially impairing host health [15]. This indicates a complex interaction between environmental toxicants, gut microbiota, and host health. Excessive fluoride exposure causes significant damage to the rectal structure as well [13]. The structural integrity of the intestine is crucial for various biological processes, including digestion, nutrient absorption, and immune defense [16]. The intestinal mucosa, which consists of epithelial cells lining the mucosal surface, represents the largest interface between the body and the external environment. As such, the intestinal mucosa is highly susceptible to various biological and environmental factors, which can disrupt its function and even lead to the complete loss of its biological activity [17]. Exposure to excessive fluoride notably reduced the thickness of the rectal mucosa and intestinal glands, causing the rectal cavity to either expand or collapse [18]. Disruptions were also observed in the arrangement of rectal epithelial cells and the blurring of nuclear membrane boundaries. Interestingly, the thickness of the rectal muscle layer is significantly increased with fluoride due to edema in the intestinal muscle cells and the collapse of the intestinal cavity due to fluoride toxicity [19]. The damage to the intestinal mucosa could be linked to oxidative stress and lipid peroxidation [20,21]. Fluoride exposure induces cytotoxicity by triggering the generation of reactive oxygen species (ROS) such as superoxide (O_2^-), hydroxyl radicals (OH•), and hydrogen peroxide (H_2O_2), leading to oxidative stress. This oxidative stress causes lipid peroxidation, DNA damage (including strand breaks and mutations), and disruption of DNA repair mechanisms. As a result, cell membranes become destabilized, leading to cellular dysfunction, apoptosis, and the potential development of cancer. Chronic fluoride exposure has been shown to increase ROS levels, damage cellular structures, and promote inflammation, which intensifies its harmful effects on cells and tissues [22-23]. Given the growing concerns over fluoride contamination and its potential impact on gut health, it is essential to elucidate the underlying mechanisms and the broader implications for public health. While much of the existing research on fluoride focuses on its effects on dental health, bones, and neurological development, the impact of fluoride on the gut remains less explored. Thus this review aims to examine the effects of fluoride on the gut and its wider impact on human health.

METHODOLOGY

We conducted an extensive search of research databases such as PubMed, Medline, and Google Scholar to collect peer-reviewed articles for the preparation of this article.

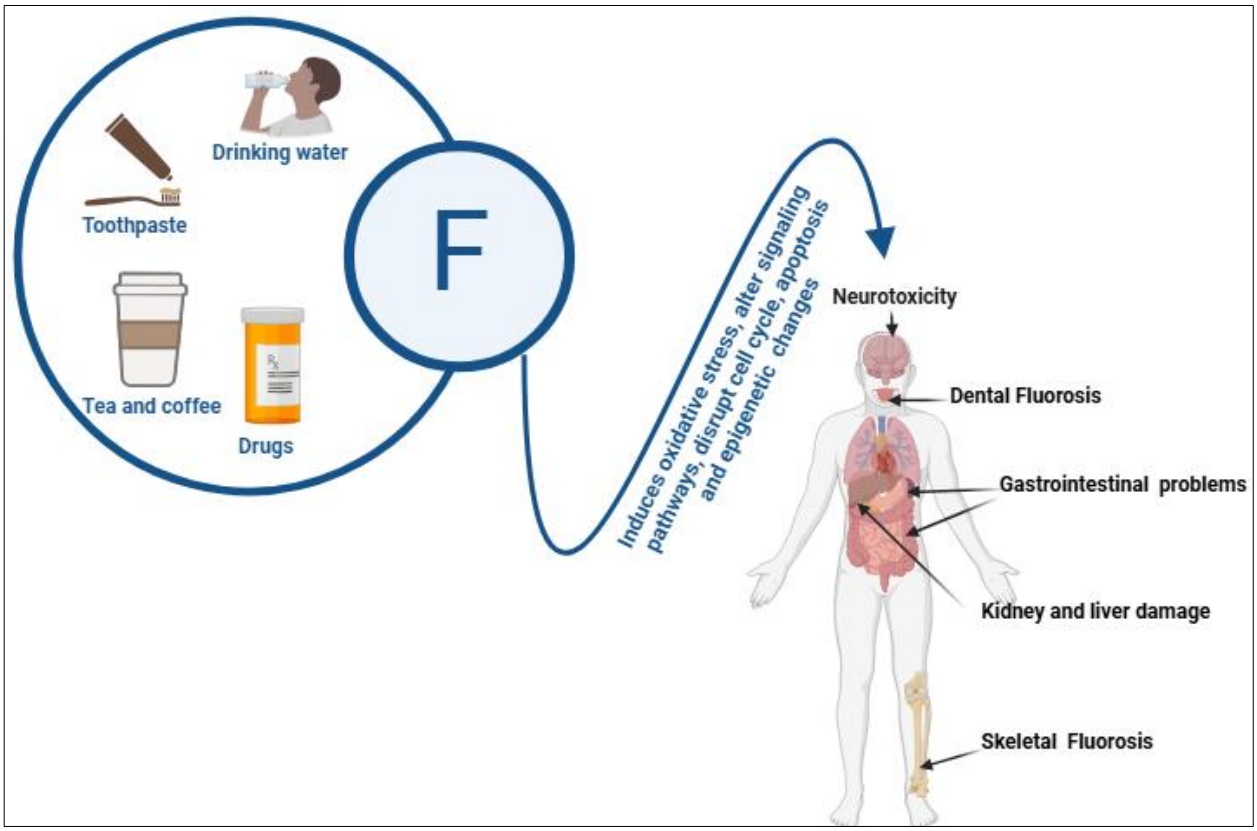

Figure 1. Fluoride toxicity from contaminated water, drugs, toothpaste, and other sources affects bones, kidneys, liver, and the nervous system, with mechanisms involving oxidative stress, signaling pathway alterations, cell cycle disruption, apoptosis, and epigenetic changes.

Fluoride as a Disruptor of the Gut Microbiome

Gut microbiomes and their functions-Our understanding of the microbiome's role in human health has greatly advanced. The gut microbiome, a complex community of bacteria, fungi, and other microorganisms, plays an essential role in digestion, immune function, and overall health [24]. The gastrointestinal (GI) tract hosts around 10^{14} microorganisms, including bacteria, fungi, and viruses, which protect against harmful pathogens, support immune system development, regulate metabolism, and produce vital vitamins [25]. This microbiome begins to form shortly after birth and continues to evolve throughout life [26]. Maintaining a balanced microbiome in the oral cavity is crucial for preventing dental issues such as caries and periodontal disease [27].

Recent research has suggested that fluoride, particularly when consumed through fluoridated water or toothpaste, could disrupt the balance between the microbiome and its host, leading to potential health issues [28]. Fluoride absorption increases as it moves through the GI tract: about 40% is absorbed in the stomach as hydrofluoric acid (HF), while the remainder is absorbed in the small intestine. Approximately 10% of ingested fluoride is excreted in the feces, indicating exposure of the entire GI tract to some degree of fluoride [29].

Chronic fluoride toxicity places physiological stress on the body [30,31], and evidence suggests that it may also affect the gut microbiome. Studies have shown that ingested fluoride can alter intestinal symptoms and microbial communities in animal models, with high fluoride concentrations reducing beneficial bacteria while promoting the growth of harmful bacteria in vitro fecal fermentation model [32,33]. Additionally, acidogenic oral microbiota, which contributes to tooth decay, has been identified as a major factor in dental fluorosis [34]. Other studies have also linked depleted carbohydrate metabolism pathways in fecal microbiota to an increased risk of dental fluorosis in children [35,36].

Intestinal microflora, a key component of the intestinal mucosal barrier, plays a crucial role in regulating nutrient absorption and preventing the growth of harmful microorganisms. Disruption of the intestinal microflora has been linked to intestinal mucosal barrier dysfunction. It is associated with various diseases, including ulcerative colitis, Crohn's disease, obesity, liver diseases, and immune system disorders [37]. The effects of fluoride on gut microbiota have produced mixed results in research. Some studies have reported that exposure to fluoride concentrations as high as 100 mg/L increases the diversity and richness of the gut microbiome in Kunming and Institute of Cancer Research (ICR) mice [12]. However, other research found no significant changes in the composition or function of the microbiota in BALB/c mice exposed to a 4 ppm fluoride dose [38]. Population studies have shown that children with dental fluorosis, particularly in high-fluoride areas, tend to have slightly lower bacterial diversity and richness than children without fluorosis [39]. Although most studies focus on mice, some evidence suggests that rats have a microbiota more similar to humans, making them valuable models for such research [40]. Most studies have primarily examined the impact of fluoride on microbiota diversity, with fewer exploring the dose-response relationship between fluoride exposure and changes in the microbiota. The networks of metabolic, signaling, and immune-inflammatory axes connect the gut to other organs such as the brain and liver [41].

Recent studies have demonstrated that high fluoride doses, ranging from 45 to 100 mg/L, can significantly alter the gut microbiota in a dose-dependent manner [12]. Fecal samples from healthy individuals aged 20–30 years demonstrated an increased relative abundance of *Proteobacteria*, a phylum containing several pathogens such as Escherichia, Yersinia, Rickettsia, Shigella, Brucella, and Salmonella, when exposed to high levels of fluoride in the fermentation model. These pathogens are linked to a range of diseases [33]. An overabundance of *Proteobacteria* is increasingly seen as an indicator of dysbiosis, which can lead to disease [42]. Fluoride exposure also significantly impacts gut microbiota at the genus level. *Faecalibacterium*, a key bacterium in the healthy human gut, plays an important role in overall health, with *F. prausnitzii* showing potential as a probiotic [43]. Similarly, *Phascolarctobacterium* has been linked to successful weight loss [44]. However, high fluoride exposure resulted in a marked decrease in the relative abundances of both *Faecalibacterium* and *Phascolarctobacterium*, which may contribute to health risks [33]. A similar decrease in *Faecalibacterium* was observed in mice treated with high fluoride doses [45]. *Firmicutes, Bacteroidetes,* and *Proteobacteria* dominant microbial phyla in rodent intestines are decreased flouride exposure [46]. Within this phylum, *Lactobacillaceae* help lower intestinal pH by producing organic acids and inhibit harmful bacterial growth by altering the permeability of the cytomembrane [47]. The decline in *Lactobacillaceae* following fluoride exposure likely increased the risk of pathogenic infections. Additionally, the relative abundance of *Bacteroidetes* and *Proteobacteria* increased, further exacerbating the imbalance in the intestinal microflora [48]. Furthermore, fluoride treatment led to a significant decrease in the proportions of *Actinobacteria* and *Verrucomicrobia. Bifidobacterium,* a key member of *Actinobacteria*, plays a beneficial role in regulating the intestinal microbiome, and its metabolites can inhibit the growth of harmful bacteria such as *Clostridium perfringens* and *Pseudomonas aeruginosa* [48,49] *Akkermansia*, a member of *Verrucomicrobia*, helps maintain the integrity of the intestinal mucosa by releasing signaling molecules that prevent the passage of lipopolysaccharides [50]. This microbial change could contribute to the damage of the intestinal barrier following fluoride exposure. *Akkermansia,* which interacts with mucin, is decreased with excessive fluoride [51].

Conversely, low fluoride doses have been shown to increase the relative abundance of beneficial bacteria, such as *Lactobacillus* and *Faecalibacterium. Lactobacillus* species, widely recognized as beneficial probiotics, play an important role in preventing and

treating various diseases. Many species of *Lactobacillus* are commercially used as probiotics, according to the U.S. Food and Drug Administration [52]. Thus, low doses of fluoride may support the growth of beneficial gut bacteria, potentially enhancing host health. In contrast, high fluoride exposure primarily increased the abundance of *Enterobacteriaceae,* a family of intestinal and systemic pathogens, including *Escherichia coli, Shigella, Klebsiella, and Salmonella* [33]. These pathogens are associated with chronic inflammation and colorectal cancer. Miao et al. [53] found that high sodium fluoride intake altered the cecal microbial community in laying hens by increasing the relative abundance of *Gammaproteobacteria, Escherichia-Shigella, Streptococcaceae,* and *Enterobacteriaceae*, while decreasing *Lactobacillus.* Moreover, studies utilizing high-throughput 16S rRNA gene sequencing have indicated that fungi play an important role in intestinal health and that excessive fluoride exposure can disrupt the species richness of intestinal fungi, potentially leading to adverse gastrointestinal symptoms by causing imbalances in the fungal microbiota. While high fluoride concentrations have been shown to disrupt microbial communities and potentially contribute to disease risk, low fluoride doses may have a more beneficial effect by promoting the growth of beneficial bacteria. Fluoride exposure not only disturbs the balance of the gut microbiome but also triggers metabolic disruptions. Alterations in the intestinal microbiota and metabolome are key factors in regulating disease susceptibility and multi-organ damage following excessive fluoride exposure. Research has increasingly shown significant changes in the abundance of gut bacteria after excessive fluoride intake, highlighting the role of fluoride in altering the composition of intestinal microbes in animals. Future research is needed to understand better the mechanisms behind these changes and their implications for human health.

Effect of Fluoride on intestinal mucosa

The intestine is a crucial organ in the human body, playing vital roles in food digestion and absorption, maintaining water and electrolyte balance, and regulating immune responses. It absorbs essential nutrients, helps maintain plasma osmotic pressure, and uses specific and non-specific immunity to detect and eliminate harmful pathogens, thereby protecting the body's internal environment. Individuals residing in areas with high fluoride concentrations often experience gastrointestinal symptoms such as loss of appetite, nausea, bloating, constipation, and intermittent diarrhea. A prospective case-controlled study by Das et al. [54] investigated the long-term effects of fluoride ingestion on the gastrointestinal system, revealing significant gastrointestinal disturbances and structural abnormalities. Fluoride-exposed individuals reported symptoms including abdominal pain, vomiting, and nausea. Additionally, petechiae, erosions, and erythema of the mucous membranes were observed in these subjects. Epithelial damage, characterized by a cracked-clay appearance, diminished microvilli, and desquamation, was noted. A report by Spak et al [55] highlighted that dental treatment with 0.42%-fluoride gel led to gastric irritation due to accidental swallowing of the gel. This irritation was attributed to the direct toxic effects of fluoride on the gastric mucosa. Petechiae and erosions were observed in the mucosa and surface epithelium was identified as the most affected area of the mucosa. Patients undergoing prolonged fluoride therapy may experience gastric damage unless luminal acidity is reduced with inhibitors or neutralized with antacids. Fluoride acts as a barrier-disrupting agent, promoting ultrafiltration of fluid from the interstitial space into the gastric lumen, which is accompanied by increased acid back-diffusion, a release of glycoproteins, and a reduction in adherent mucus [56]. The rapid penetration of fluoride into the mucosa as hydrofluoric acid may aggravate this process, as it induces local vascular stasis, leading to increased intramucosal acidity and, ultimately, significant mucosal damage. The gastrointestinal impact of fluoride is mainly influenced by the concentration of aqueous fluoride in the stomach, rather than the overall fluoride dose from the ingested liquid or solid, emphasizing the importance of fluoride intake concentration in determining health effects.

The fluoride concentration that the gut epithelium is exposed to can be altered by gastric fluids when fluoride is ingested. The population is expected to experience GI symptoms in regions with endemic fluorosis. Therefore, understanding the biological and physiological mechanisms behind fluoride's impact on the GI system is essential. Ingesting high doses of fluoride [57-60], such as from fluoride-rich oral care products, contaminated drinking water during fluoridation accidents, or fluoride drugs used for

osteoporosis treatment, can worsen GI symptoms. Fluoride has been shown to stimulate stomach acid secretion, reduce blood flow from the stomach lining, dilate blood vessels, increase redness of the stomach lining, and cause cell death and shedding of the GI tract epithelium. Since fluoride inhibits several key intracellular enzymes, it is not surprising that at high exposures, it causes cell death and desquamation of the GI epithelium. The mechanisms by which fluoride affects secretion are not entirely understood, but they are likely linked to fluoride's ability to activate guanine nucleotide regulatory proteins (G proteins) [61]. Whether fluoride activates these G proteins in the gut epithelium at chronic exposure of fluoridated water at 4.0 mg/L, and has significant effects on gut cell function, remains unclear [62].

Fluoride-Related Inflammation in Gut

The small intestine is one potential site affected by fluoride. Inflammation is known to increase epithelial permeability and inflammatory cytokines such as Interleukin 1 beta (IL-1β), Tumor necrosis factor alpha (TNF-α), and Interferon gamma (IFN-γ) are associated with enhanced gut permeability Conversely, IL-10, an anti-inflammatory cytokine, plays a crucial role in maintaining gut homeostasis [63]. Excessive fluoride intake leads to intestinal inflammation by elevating pro-inflammatory cytokines (TNF-α, IL-1β, and IFN-γ) and reducing the anti-inflammatory cytokine IL-10 [30]. High concentrations of fluoride have been linked to inflammation. In the digestive system, inflammation could play a role in conditions like Irritable Bowel Syndrome (IBS), Crohn's disease, and ulcerative colitis [64] Chronic excessive fluoride harms the intestine by inhibiting epithelial cell proliferation, increasing intestinal permeability, disrupting intestinal mechanical and immune barrier functions, and promoting inflammation in the gut lining [64]. Furthermore, fluoride-induced oxidative damage can lead to a reduction in the percentage of specific T cell subgroups in the cecal tonsil and decrease the levels of Immunoglobulin A (IgA), Immunoglobulin G (IgG), and Immunoglobulin M (IgM) in this region, which compromises local mucosal immune function [65]. Excess fluoride is absorbed through the stomach and intestinal epithelium, where it can inhibit the proliferation of intestinal epithelial cells and mast cells, stimulate inflammatory cell activity, activate lymphocytes, reduce immunoglobulin concentrations, and impair the ability to combat pathogens, ultimately affecting the immune function of the intestinal mucosa [66-69]. The intestinal barrier is a critical defense against microbial pathogens that enter the host via the gut.

Excess fluoride exposure has been shown to trigger pro-inflammatory factor expression, decrease the levels of tight junction-related genes and proteins, activate inflammatory responses, promote cell fever, and hinder intestinal development, leading to inflammation and diarrhea [70]. Increased levels of inflammatory cytokines such as TNF-α and IL-1β, and elevated Nuclear factor 'kappa-light-chain-enhancer' of activated B-cells (NF-κB) protein expression in the small intestine are reported in mice having fluorinated drinking water. NF-kB plays a crucial role in mediating the inflammatory response [71]. White blood cells (WBCs) are recruited by inflammatory cytokines in acute and chronic inflammation, making them a reliable marker of inflammation. The positive association between WBC counts and plasma fluoride concentrations suggests a link between fluoride exposure and heightened inflammation. Recently Den et al [72] reported the positive associations between plasma fluoride and neutrophils and monocytes suggesting that fluoride may influence tissue-specific inflammatory responses. Neutrophils, monocytes, and lymphocytes respond to peripheral inflammation, Lymphocytes are involved in antibody production while neutrophils, are recruited to sites of tissue damage in response to inflammation and are later cleared by monocytes. Fluoride ingestion through drinking water leads to inflammatory lesions in the small intestine resembling those found in Crohn's disease in rodents [19, 73]. Increased neutrophils and monocytes are key components in the chronic inflammation seen in Crohn's disease.

Macrophages are the primary source of reactive oxygen species (ROS) in the human body and play a crucial role in the immune response to various pathogens, as well as in inflammation. When exposed to high fluoride concentrations, macrophages experience an increase in ROS production, leading to oxidative stress, which in turn affects the expression of inflammatory factors [74]. Similarly, fluoride exposure in bovine neutrophils results in the formation of neutrophil extracellular traps (NETs), along with elevated ROS levels and reduced antioxidant enzyme activity [75]. This oxidative stress and NET formation may

contribute to immunotoxicity in neutrophils through activation of the p38/MAPK and ERK pathways. T lymphocytes, including CD4+ T helper (Th) cells and CD8+ cytotoxic T lymphocytes (CTL), are involved in the immune response by regulating the functions of other immune cells. Excessive fluoride exposure has been shown to reduce the number of lymphocytes, particularly decreasing the percentages of CD4+ and CD8+ T cells in the thymus, which can lead to T cell dysfunction. Fluoride also impacts the expression of transcription factors like T-bet and GATA3. These factors are key regulators of Th1/Th2 cell development, differentiation, and memory formation. Disruption of the balance between Th1 and Th2 cells can cause immune dysregulation. Furthermore, excessive fluoride intake has been linked to changes in the number of Th17 cells and alterations in the expression of related cytokines, ultimately affecting the immune system's overall function [76].

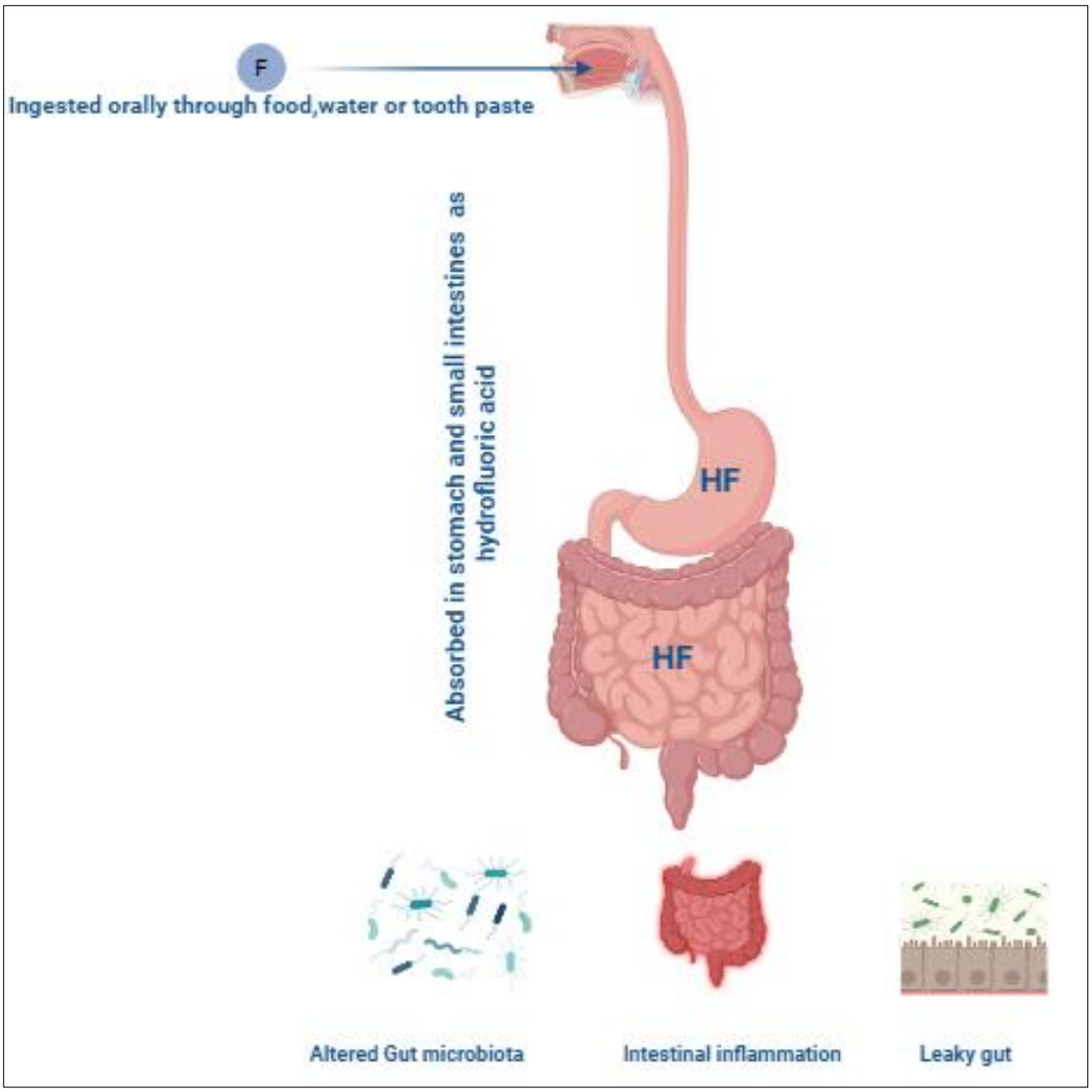

Figure 2 - Chronic fluoride toxicity can disrupt the gut microbiota, increase gut inflammation, and lead to a leaky gut.

Fluoride and Gut Barrier Function

The intestinal tract is a protective layer of cells that lines the intestines and prevents harmful substances from entering the bloodstream [77]. The gastrointestinal tract is the earliest and most vulnerable site to toxin exposure. Its health is an indispensable factor for the maintenance of host homeostasis. The physical barrier, the key line of defense, is formed by epithelial cells and the Tight Junctions.TJ acts as paracellular gates that impose size- and charge-dependent restrictions on diffusion, constructed by a network of protein interactions in the apical region of the lateral membrane [78]. Existing research has demonstrated that alterations in the continuity and/or number of tight junction proteins control epithelial permeability. Intestinal permeability, often referred to as "intestinal leakage," can allow the translocation of bacteria and their products, which has been implicated in the development of various health disorders. Tight junction (TJ) proteins, such as ZO-1 and occludin, are essential in maintaining the intestinal barrier by facilitating cell-to-cell adhesion and preventing harmful molecules from crossing the epithelial sheet into the bloodstream [79]. Fluoride induces intestinal inflammation and disrupts mucosal integrity, increasing intestinal permeability [80,81]. The mRNA expression of TJ proteins is significantly reduced in fluoride-exposed rodents contributing to gut inflammation and resulting in increased levels of serum diamine oxidase (DAO) activity and D-lactate. DAO activity is primarily associated with the small intestine, and serum DAO is largely derived from this region in mammals [82]. Additionally, D-lactate, which cannot be produced by mammals but originates from gut bacteria, accumulates in the bloodstream when the intestinal barrier is compromised [83]. Both DAO activity and D-lactate levels are slow to metabolize and serve as valuable markers of mucosal integrity [84]. Long-term fluoride intake directly affects the proliferation of intestinal epithelial cells and destroys the intestinal barrier structure. Intestinal integrity is associated with the construction of tight junctions. Fluoride is absorbed by the stomach and intestines in the form of weakly acidic un-dissociated hydrogen fluoride with the intestine absorbing up to 60% or more. Furthermore, epidemiological investigations showed that among patients with long-term fluoride intake, more than 70% exhibited symptoms such as nausea, vomiting, diarrhea and abdominal pain. The results of duodenal biopsy from patients with otosclerosis who had consumed 30 mg of sodium fluoride daily showed inflammation of the intestinal mucosa [54]. In addition, some animal studies have suggested that fluoride exposure might alter gut barrier function, potentially leading to "leaky gut," a condition in which the gut lining becomes more permeable.

A leaky gut can allow toxins and undigested food particles to pass into the bloodstream, triggering immune responses and inflammation. Sun et al [85] reported the decreased expression of tight junction protein zonula occludens-1 (ZO-1) in human vascular endothelial cells through the PI3K/AKT signaling pathway through excessive fluoride exposure. Fluoride affects cellular calcium ion homeostasis, with an enhancement in intracellular calcium levels observed in multiple model systems [86]. Fluoride inhibited calcium transporter and channel activity, promoting calcium release from intracellular stores [87] Xu et al. [88] proved that high fluoride exposure leads to oxidative stress and apoptosis by elevating intracellular calcium concentrations in cultured neuroblastoma cells. Calcium ion is a highly versatile messenger in the modulation of TJs and actin filaments. It is well established that extracellular Ca^{2+} interacts with tight junctions to promote stability. Cytoplasmic calcium is conducive to the assembly of TJs with cytoskeleton. Additionally, calcium ions have been widely reported as mediators of cellular signaling. $Ca2^{+}$ activates MLCK to drive actomyosin reorganization, resulting in the disconnection of tight junctions. Additionally, calcium ion oscillations have been proven to modulate the status of the RhoA/ROCK pathway, and forestalling its overload may rescue barrier hyperpermeability caused by ethanol. The permeability of the intestinal barrier primarily depends on the integrity of TJs between adjacent cells. In contrast to adherens junctions, which initiate cell-cell contacts and control tensile force, TJs act as paracellular gates to confine the trafficking of molecules based on their sizes and charges.TJ disruption mirrored epithelial integrity loss, which triggers multiple diseases that endanger health. NaF administration significantly decreased the expression of the scaffolding protein ZO-1. Consistently, NaF, administered at 100 ppm in drinking water, remarkably reduced the mRNA and/or protein levels of ZO-1 in the ileum of mice [89] Additionally, 1 mM fluoride impeded TJ formation in rat ameloblast HAT-7 cells [90]. TJ

function is also critically governed by subcellular localization. The rearrangement of TJs has been reported to favor barrier hyperpermeability [91]. However, most studies on the correlation between fluoride and TJs have been limited to protein quantification. As confocal microscopy can capture subtle changes in target proteins, NaF noticeably interfered with the continuity of ZO-1 distribution. Ochratoxin A-treated IPEC-J2 monolayers exhibited irregular ZO-1 staining patterns and barrier dysfunction. Another study showed that the bradykinin-mediated blood-tumor barrier (BTB) resulted from the redistribution of ZO-1.

CONCLUSIONS

Fluorine is a highly abundant element that poses toxicity to various organisms, ranging from bacteria to humans. However, the precise mechanisms by which eukaryotes protect themselves from fluoride toxicity are still not well understood. Fluoride disrupts numerous cellular processes, but here, we focus on its impact on the gut.

Excessive fluoride exposure may negatively affect gut health by impairing the intestinal mucosal barrier, disrupting the gut microbiome, and promoting inflammation. Specifically, fluoride exposure decreased beneficial bacteria such as *Lactobacillaceae* and *Bifidobacterium*, while increasing harmful bacteria like *Bacteroidetes* and *Proteobacteria.* This microbial imbalance, coupled with reduced levels of tight junction proteins (ZO-1 and occludin), contributed to increased intestinal permeability. Furthermore, fluoride-induced inflammation was marked by higher levels of pro-inflammatory cytokines and a decrease in the anti-inflammatory cytokine IL-10. Although most health guidelines indicate that typical fluoride levels found in drinking water and dental products are generally safe, maintaining a balanced diet rich in fiber, probiotics, and prebiotics can support a healthy gut microbiome and potentially offset the negative effects of environmental toxins like fluoride. Reducing fluoride exposure should be prioritized in areas with high natural fluoride levels in the water. Individuals concerned about fluoride exposure might consider limiting fluoride toothpaste usage (particularly swallowing large amounts) and avoiding fluoride supplements to lower overall exposure. Future research is needed to understand better the mechanisms behind these changes and their implications for human health.

FUNDING

Not applicable

CONFLICT OF INTERESTS

None

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