A Tool for the Treatment of Gastrointestinal Tumors: Micro-Ecological Immunotherapy

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Abstract: It has been proposed that the intestinal microbiota and gastrointestinal tumors are interdependent. Changes in the microbiota can cause dysfunction of the gastrointestinal tract, thereby promoting carcinogenic changes, leading to the occurrence of gastrointestinal tumors. Recent studies on intestinal microbiota have opened up a new area in intestinal micro-ecological immunotherapy. The intestinal microbiota is a double-edged sword. Gut microbes participate in carcinogenesis, but can also be used for immunotherapy. The intestinal microbiota is also regulated by the daily diet. Intestinal micro-ecological immunotherapy combines intestinal immune nutrition and intestinal ecological nutrition to make full use of the intestinal microbiota to strengthen nutritional support. Micro-ecological immunotherapy enhances the body's immune function by providing energy, improving the functional state of tissues and organs, protecting the intestinal mucosal barrier and maintaining normal intestinal microbiota balance. This involves, to some extent, PD-1 and PD-L1. The microbiota is beneficial to improve the clinical efficacy of conventional anti-cancer therapy and to reduce the incidence of complications. At the same time, micro-ecological immunotherapy is itself active and effective in the perioperative treatment of tumors, which is of great significance for the prognosis of the patient. Gastrointestinal tumors are increasingly linked to intestinal microbiota, and various microbiota-related technologies and drugs have been developed. In the future, the intestinal microbiota may represent a screening marker for gastrointestinal tumors. In addition, clinicians may be able to prevent and treat cancers by changing the gene expression levels of certain microbiota, or by regulating the types of microbes present.

Key words: Micro-ecological immunotherapy; Immune nutrition; Ecological nutrition; Gastrointestinal tumors; Microbiota

Introduction

There are abundant microbial communities in the gastrointestinal environment, which are closely related to the physiological and pathological state of the gastrointestinal tract. In recent years, the occurrence of gastrointestinal tumors has been increasingly linked to the activity of the intestinal microbiota. Disorders of the intestinal microbiota can lead to abnormalities in the function of the gastrointestinal tract, thereby promoting the occurrence of gastrointestinal tumors [1]. The intestinal microbiota is also regulated by the daily diet. Importantly, the intestinal microbiota is often a double-edged sword, which can both promote and eradicate gastrointestinal tumors, depending on

the types of microbes present and other conditions present [2]. Recent research on intestinal microbiota has revealed that micro-ecological immunotherapy may represent a novel approach to preventing and/or treating gastrointestinal cancers. Micro-ecological immunotherapy employs a combination of intestinal immune nutrition and intestinal ecological nutrition [3,4]. This article will elaborate on the effects of intestinal micro-ecological immunotherapy, including ways to provide energy, improve the functional states of tissues and organs, improve the body's immune function, and protect the gastrointestinal mucosal barrier by providing an optimal balance in microbes [5]. We also discuss the effects of micro-ecological immunotherapy on perioperative cancer treatment, and the responses to radiotherapy and chemotherapy, as well as the local microenvironment [6]. Gastrointestinal microbes are also expected to be useful as markers to screen for gastrointestinal tumors, and microbes are becoming a new focus for gastrointestinal cancer prevention and treatment.

Epidemiology of Gastrointestinal Tumors

Despite significant progress in the diagnosis and treatment of gastrointestinal tumors, the incidence and mortality rates of gastrointestinal tumors are still high due to

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factors such as heredity, lifestyle, mental state and infections. At present, the incidence and mortality of gastrointestinal tumors ranks at the forefront of all types of tumors in both men and women in China [7]. These tumors reduce the quality of life of patients and cause a serious burden on the social economy. At the same time, the prognosis of gastrointestinal tumors remains a major problem that plagues clinicians and researchers [8]. Among the various pathogenic factors associated with gastrointestinal tumors, disorders of the gastrointestinal microbiota and infectious factors are being increasingly found to play a decisive role in the occurrence, development and progression of tumors.

Gastrointestinal Microbiota and Tumorigenesis

The stomach has long been considered sterile, but it actually contains a variety of microbes. One such microbe is *Helicobacter pylori* (*H. pylori*). In many parts of the world, the infection rate of *H. pylori* correlates with the rate of gastric cancer. *H. pylori* can cause pathological changes in the host stomach, including chronic gastritis, atrophy, intestinal metaplasia and gastric cancer [9]. The bacterium secretes vacuolar cytotoxins such as CagL and VacA, which interact with mucosal epithelial cells, leading to changes in signaling pathways and cell polarity, which ultimately lead to the development of gastric cancer [10]. These oncoproteins activate different pathways, such as Csk/Crk/SHP-2 (through a phosphorylation-dependent pathway), leading to cytoskeletal reorganization and cell death [11]. CagA also activates Crb2/c-Met via a phosphorylation-independent pathway, resulting in pro-inflammatory and mitogenic gene expression, which also leads to cancer formation [12]. VacA has immunomodulatory and inhibitory effects on various mammalian immune cells, ensuring the persistence of *H. pylori* in the host. VacA can inhibit the activation and proliferation of both T cells and B cells and interferes with the antigen expression of B cells [13]. VacA prevents normal antigen processing, phagocytic expression, intracellular killing and cytokine production. Animal experiments also revealed that VacA can cause damage to the gastric mucosa, increase the recruitment of

inflammatory cells, and induce cancer [14].

There are thousands of microbes in the human intestine, which form a symbiotic system with the intestinal cells to maintain the intestinal balance. However, some intestinal microbes are pathogenic or carcinogenic, with some exerting these effects when the intestinal environment is disturbed. For example, *Enterococcus faecalis* infection causes colitis and the expression of TGF- β , which activates the Smad4 signaling pathway. It also produces extracellular superoxide, participates in bystander effects, causes DNA damage, and thus induces cancer [15]. The B2 strain of *E. coli* produces cell necrosis factor, which inhibits mismatch repair. When these bacteria also produce pathogenic pks, the chromosome can become unstable and cancers can occur [16]. *Bacteroides fragilis* releases an enterotoxin (BFT), activates the STAT3 signaling pathway, induces the production of Th17 and interleukin-17, and promotes colon cancer formation [17]. The BFT can rapidly induce the expression of SMO, promoting the production of ROS, and destroying the DNA of intestinal epithelial cells [18]. At the same time, BFT is able to amplify signaling via the Wnt and NF- κ B signaling pathways, and leads to the release of pro-inflammatory substances. *Fusobacterium nucleatum* stimulates FadA, activates wnt/ β -catenin/TLR4 signaling and upregulates oncogene expression [17], and *Fusobacterium nucleatum* can inhibit the activity of natural killer cells in the tumor microenvironment, leading to the occurrence of colorectal cancer [19].

Relationship between Changes in Intestinal Microbiota and Intestinal Nutrition

Undoubtedly, changes in the intestinal microbiota are also closely related to intestinal nutrients. The most direct source of intestinal nutrition is the daily diet. Carbohydrates are divided into digestible carbohydrates and non-digestible carbohydrates. Digestible carbohydrates are enzymatically degraded in the small intestine, including starches and sugars such as glucose, fructose, sucrose and lactose. The consumption of high levels of glucose, fructose and sucrose increases the relative abundance of bifidobacteria,

Table 1 Relationship between changes in the intestinal microbiota and intestinal nutrition.

Intestinal	Nutrition composition	Example dietary sources	Changes in the intestinal microbiota	
			Increase	Decrease
Carbohydrates	Digestible carbohydrate	Starch, sugar (glucose, fructose, lactose)	<i>Bifidobacteria</i>	<i>Bacteroides spp.</i> , <i>Clostridium spp.</i>
	Non-digestible carbohydrate	Fiber (whole wheat, wheat bran, resistant starch)	<i>Bifidobacteria</i> , <i>Lactobacilli</i> , <i>Rumenococci</i> , <i>E. rectale</i> , <i>Roseburia spp.</i>	
Fats	Saturated fat	Fat, butter	<i>Clostridium spp.</i> , <i>Bacteroides spp.</i> , <i>Faecalibacteria</i> ,	<i>Lactobacilli</i>
	Unsaturated fat	Fish	<i>Bifidobacteria</i> , <i>Streptococci</i>	
Proteins	Animal protein	Dairy products	<i>Bacteroides spp.</i> , <i>Alistipes spp.</i> , <i>Clostridium spp.</i>	<i>Bifidobacteria</i>
	Plant protein	Bean products	<i>Bifidobacteria</i> , <i>Lactobacilli</i>	<i>Bacteroides spp.</i> , <i>Clostridium spp.</i>

while the abundance of *Bacteroides* species is relatively reduced [20]. The addition of lactose to the diet replicates the same bacterial changes and also reduces the abundance *Clostridium* species [21] (Table 1). Many studies have shown that diets rich in non-digestible carbohydrates steadily increase the number of bifidobacteria and lactobacilli [22]. For example, a diet rich in whole wheat or wheat bran is associated with an increase in *bifidobacteria* and *lactobacilli* [23]. Other non-digestible carbohydrates, such as resistant starch and whole-wheat barley, also appear to increase the number of intestinal microbiota such as *Rumenococci*, *E. rectale*, and *Roseburia* spp. [24,25].

It is well-known that the risk of cardiovascular disease is closely related to the daily lipid intake. Several studies have shown that a diet high in saturated fat increases the number of anaerobic microbiota and *Bacteroides* spp. [26]. The number of *Clostridium* organisms also increases in the presence of a high saturated fat diet, but the number of lactobacilli decreases [27]. In contrast, an unsaturated fat diet results in an increase in the number of *Bifidobacterium* in the feces compared to baseline, while also leading to a decrease in fasting blood glucose [28].

Protein intake is divided into plant proteins and animal proteins. A large number of studies have shown that protein consumption is positively correlated with the overall microbial diversity. Excessive intake of dairy products can cause an increase in *Bacteroides* and *Clostridium* spp., while the number of bifidobacteria is relatively reduced [29,30]. It has been reported that the intake of whey and pea protein extracts can increase the number of bifidobacteria and lactobacilli in the intestine, while whey can also reduce the pathogenic *Bacteroides fragilis* and *Clostridium perfringens* [31].

Therefore, the daily diet determines the physiological state and abundance of the intestinal microbiota. Thus, clinicians can provide dietary guidance for patients with gastrointestinal cancer that will help reduce their disease burden or may improve the response to treatment.

Nutritional Support for Patients with Gastrointestinal Tumors

Patients with gastrointestinal tumors often develop malnutrition during the perioperative period. The causes of malnutrition are two-fold, involving both inadequate intake of nutrients and increased consumption of nutrients and energy. Tumor-induced pain or localized compression can lead to eating or emptying disorders. Inadequate intake can lead to cachexia and early satiety, which stimulates the hypothalamus to produce factors that further worsen anorexia [32]. In addition, malnutrition in cancer patients often leads to complications such as nausea, vomiting, and coagulopathy [33]. In patients with cancer, the local cellular metabolism is hyperactive, with increased glucose consumption as a result of the tumor-associated Warburg effect (anaerobic glycolysis) [34]. The host's proteins are

also consumed to increase the formation of the tumor cells' proteins, resulting in a negative nitrogen balance in the body.

Bacterial infections can easily lead to microbial disorders in patients with gastrointestinal tumors, reducing postoperative recovery and the tolerance of radiotherapy and chemotherapy [35]. Immune function is impaired by malnutrition, which leads to abnormal lymphocyte development, decreased macrophage function, and the abnormal secretion of cytokines [36]. Damage to the immune function leads to an increase in infection and a decrease in the patients' prognosis [37]. Immune dysfunction also promotes tumorigenesis. For example, tumor-associated macrophages stimulate the proliferation of cancer cells and the epithelial-mesenchymal transition by accelerating the formation of blood vessels and lymphatic vessels, promoting the metastasis of tumor cells [38]. Neutrophil elastic hydrolase also promotes tumor cell invasion [39]. Of note, T cells play a dual role in tumorigenesis and metastasis. In the early stages of tumorigenesis, T cells can control tumor progression [40]. Due to the sustained selective pressure of the effector response, tumor variants are selected and evade immune recognition. The immune system is then unable to eradicate these selected tumors, which can proliferate and metastasize [41].

Clinicians have found that malnutrition is common in patients with gastrointestinal cancer. In most patients with severe malnutrition, immediate surgery is not the best option. There is also currently no clear way to significantly change an individual's body composition in a short period of time before surgery. However, perioperative nutritional support can significantly improve the postoperative outcomes of patients with severe malnutrition. When early intestinal nutrition does not meet the requirements to alleviate malnutrition, adding microecological agents and immune nutrition has been advocated [42]. Nutritional support is especially important after surgery. Postoperative malnutrition in patients with cancer is associated with increased infection rates, decreased immune function, increased postoperative complications, and a reduced tolerance of radiotherapy and chemotherapy [33]. This not only increases the financial burden on patients, but also increases their risk of mortality. In order to more directly regulate post-operative inflammation and other complications, the implementation of intestinal microecological immunotherapy can provide not only energy and nitrogen, but also a replacement population of the microbiota normally in the human body [43]. This concept is expected to open up a new field of research.

A New Weapon against Cancer: the Emergence of Micro-ecological Immunotherapy

The intestinal microbes have rapidly become a focus of extensive research in a variety of fields. The gut microbiota

is considered to be the hidden immune organ of the human body, regulating the function and homeostasis of many organs throughout the body [44]. Nutritional support plays a crucial role in the immune and postoperative recovery of cancer patients. Intestinal microbes are being used by researchers as a new type of ecological nutrient for the treatment of tumors [45]. Intestinal micro-ecological immunotherapy integrates intestinal immune nutrition with intestinal ecological nutrition. Micro-ecological preparations are added to strengthen the clinical efficacy of nutritional support, reduce the occurrence of complications, and exert therapeutic effects either directly or by increasing the tolerance to conventional therapies [46]. Special nutrients such as glutamine and arginine are added to traditional intestinal nutrition to stimulate the body to produce an immune response, reduce excessive inflammation, and protect the integrity of the intestinal mucosa [47]. In a study of the effects of glutamine-based nutritional support on the perioperative period of patients with advanced gastric cancer, Wang J et al. found that the supplement protected the intestinal mucosal barrier function, reduced the adverse reaction rate and increased the patient quality of life [48].

Omega-3 polyunsaturated fatty acids have anti-inflammatory effects and are also used to reduce postoperative infections and complications [49]. Xie H et al. found that perioperative intestinal nutritional support is superior to preoperative and postoperative nutritional support in pre-

venting postoperative infections and complications. Moreover, Wilhelm SM et al. used a combination of arginine and omega-3 polyunsaturated fatty acids to effectively reduce infection rates and improve patient survival of colorectal cancer [49,50].

While intestinal nutritional support enhances the immunity of cancer patients and improves the prognosis of patients, the emergence of microbial preparations means that intestinal ecological nutrition can also be provided. Microbiota, especially probiotics, can be incorporated in the patient's daily nutrition, where the microbes will regulate the intestinal micro-ecological environment and reconstitutes the bacterial homeostasis, preventing the invasion of pathogenic bacteria [51]. The innovation of intestinal ecological immune nutrition lies in the combination of immune nutrition and microbial preparations (Figure 1). In support of this concept, Tan CK et al. gave patients microbial preparations before surgery, and found that this significantly promoted the recovery of postoperative intestinal function in patients with colorectal cancer and shortened their hospital stay [52]. Microbiota can also stimulate toll-like receptors (TLRs), trigger complex molecular cascades, produce cytokines and activate T cells [53]. Studies have shown that the regeneration of T cells can also be induced by metabolites produced by the microbiota.

Based on the current understanding, several major bacterial metabolites play key roles in the pathogenesis of

Figure 1 Introduction to micro-ecological immunotherapy. Micro-ecological immunotherapy is a new type of nutritional therapy that combines intestinal ecological nutrition with intestinal immune nutrition. It is based on the immunological factors included in intestinal immune nutrition, such as arginine, glutamic acid and omega-3 polyunsaturated fatty acids. This is combined with microbial preparations, such as probiotics, to provide nutrition and anti-tumor effects. Micro-ecological immunotherapy provides energy, improves the tissue and organ functional status, improves immunity, protects the intestinal mucosa, and maintains the intestinal microbiota. Micro-ecological immunotherapy can reduce the risk of postoperative infections, reduce the rates of complications and adverse reactions, and promote the regeneration of immune cells, so that patients can improve their survival and quality of life and reduce their hospitalization time.

colorectal cancer and the immune microenvironment that permits the growth of these cancers. These metabolites include SCFAs, polyamines, secondary bile acids, and phytochemicals. Their roles in colorectal cancer have been widely documented [54,55]. Several mechanisms have been proposed to underlie the effects of probiotics in the prevention and treatment of gastrointestinal tumors. First, some probiotics are capable of inducing anti-proliferative and apoptotic processes in colorectal cancer cells. For example, *Propionibacterium freudenreichii* induces apoptosis in colorectal and gastric cancers by producing SCFAs. This is because colorectal cancer cells can use the glycolysis pathway to create an acidic environment in the gastrointestinal tract, which results in the production of high SCFAs by *Propionibacterium freudenreichii* [53-56]. SCFAs block the proliferation of colorectal cancer cells primarily in the G1 phase of cell division [57]. *Bifidobacteria* can also inhibit the growth of colorectal cancer cells by stopping the cell cycle and increasing alkaline phosphatase activity during G0/G1. Alkaline phosphatase is associated with a reduced degree of malignancy in cancer cells [58,59]. This finding suggests that the addition of specific probiotic bacteria to lactic acid products can provide patients with both good postoperative nutrition and some therapeutic benefits. Microbes have also been shown to exert apoptosis via multiple mechanisms in HT29 and HCT116 cell lines; exerting cytotoxic effects on cancer cells, secreting specific cytotoxic metabolites, and positively regulating the C-fos gene [60].

Second, probiotics can enhance the function of the intestinal epithelial barrier. SCFAs can activate adenosine monophosphate-activated protein kinase to enhance the tight junctions of the intestinal epithelium and form a strong barrier. This protein kinase is a key factor that regulates the energy metabolism of human colon cells. *In vitro* and *in vivo* experiments have demonstrated that supplementation of SCFAs enhances this intestinal barrier [54].

Third, probiotics can inhibit the enzymatic activity of pathogenic bacteria. The intestinal purification of toxins and absorption of nutrients largely relies on enzymes produced by various microbiota. Disordered microbiota was result in the release of a large number of oncogenic enzymes, such as β -glucuronidase, β -glucosidase and nitroreductase [61,62]. These enzymes are susceptible to toxic carcinogenic metabolites such as hydrogen sulfide, acetaldehyde aromatic amines, and secondary cholates, which can inhabit these products [63]. Therefore, probiotics can reduce the production of carcinogens by reducing the activity of these enzymes. For example, lactic acid bacteria reduce the dehydroxylation of primary bile acids [64]. The combination of various probiotics and vegetables can reduce the level of carcinogenic products such as β -glucuronidase, which are increased by the consumption of protein and fat. Probiotics can also participate in regulating the immune function. Studies have shown that probiotics interact with TLRs to

produce anti-inflammatory cytokines [65]. Certain types of lactic acid bacteria increase the activity of regulatory T cells (Tregs), enhance the antibacterial phagocytic activity of peripheral blood neutrophils, and increase the activity of natural killer cells [66]. Soluble substances produced by probiotics may also induce a systemic immune response through secondary gastrointestinal antigen-presenting cells.

We have recently explored the roles of PD-1 and PD-L1 in regulating the effects of microbial metabolites, with a focus on colorectal cancer and its microenvironment. Tumor immunotherapy is currently a hot topic of research. The PD-1 receptor is an inhibitory receptor expressed by activated T lymphocytes, B cells, natural killer T cells (NKT) and Treg cells, making it an important target for immunotherapy. PD-1 plays a key role in regulating T cell function in peripheral tissues, where it recognizes PD-L1 and PD-L2. These two ligands are expressed not only on antigen-presenting cells (APCs), but also on the surface of lymphoid tissues and non-lymphoid tissues (including tumor cells). PD-1 contains an immunoreceptor tyrosine-based inhibition motif (ITIM) and an immunoreceptor tyrosine-stimulated motif (ITSM). When PD-1 recognizes its ligand, this interaction induces the phosphorylation of tyrosine residues in the ITSM, leads to the recruitment of tyrosine phosphatase SHP2, induces Zap70 dephosphorylation and inactivation in T cells, and down-regulates TCR signaling [67]. Therefore, PD-1 down-regulates the activity of T cells and inhibits the normal immune response. When PD-1 binds to PD-L1 in the tumor microenvironment, the function of T cells is impaired, allowing tumor cells to evade immune surveillance. Many studies have shown that blocking PD-1 or PD-L1 restores T cell function [68].

As noted above, micro-ecological immunotherapy is an excellent method for providing tumor control (Figure 2). Some intestinal microbiota can affect the interaction between PD-1 and PD-L1. For example, Sivan A et al. [71] compared melanoma cells from two different mice. These mice had different microbiota but were genetically similar. They found that bifidobacteria had a positive effect on activated APCs, which can effectively control tumors and IFN- γ secretion [69]. The researchers also found that PD-1 expression is involved in experimental colitis. Wild-type (WT) mice had severe wasting disease caused by experimentally-induced colitis, whereas PD-1 deficient mice did not develop colonic inflammation [70]. Sivan A et al. compared the growth of mouse melanoma in mice with different commensal microbiota and observed differences in spontaneous anti-tumor immunity that were eliminated after co-administration or fecal metastasis [71]. Subsequent 16S ribosomal RNA sequencing revealed that Bifidobacteria have anti-tumor effects. The effects of oral administration of Bifidobacteria alone on tumors were the same as those of PD-L1-specific antibody treatment, and combination therapy almost eliminated tumor growth [71]. When Wang T et al. administered *Lactobacillus reuteri* to

regulate intestinal microbiota, it alleviated PD-1 antibody-induced autoimmunity, thereby enabling immunotherapy to produce the desired immune response without significant immunopathology [72]. This suggests that the resistance of PD-1-deficient mice to colitis depends on their intestinal microbiota. These experiments provided new ideas about the potential mechanisms underlying micro-ecological immunotherapy. The addition of specific microbiota to micro-ecological immunotherapy can thus assist tumor immunotherapy, reduce complications, and provide greater opportunities for tumor elimination.

Figure 2 The intestinal microbiota is involved in regulating the relationship between PD-1, PD-L1 and tumorigenesis. PD-1 is produced by a variety of cells, including T cells, B cells, and NK cells. It recognizes PD-L1 on the surface of tumor cells and APCs, and induces an immunoreceptor tyrosine stimulating motif (ITSM). Tyrosine is dephosphorylated and inactivated via the ITSM, inhibiting T cell activity. Microbiota may be involved in the process of immune recognition, preventing the binding of PD-1 to PD-L1, thereby inhibiting tumor growth and progression.

In the perioperative period, intestinal ecological immune nutrition can improve the intestinal barrier, thus effectively reducing the stress response following the operation. In the surgical treatment of enteropathy, the use of this nutritional method can increase the diversity of the intestinal microbiota and reduce the symptoms of the digestive tract. At present, intestinal eco-immune nutrition mainly involves the addition of probiotics for nutritional intervention [65]. Sharaf LK et al. found that the administration of a combination of probiotics and celecoxib during the week before the induction of colorectal cancer in mice decreased the expression of proto-oncogene K-ras and increased the expression of *p53* [73]. These results implied that the addition of probiotics seem to help activate a tumor suppressor gene, maintain the normal cell activity and cell cycle, and effectively control the development of colorectal cancer. The researchers subsequently treated 140 patients with colorectal cancer receiving chemotherapy with supplemental probiotics and intestinal nutrition, while

the control group received a placebo for eight weeks. The results showed that the level of inflammatory markers related to the side effects of chemotherapy were reduced in the probiotic and intestinal nutrition group. Moreover, this reduction was more significant than that associated with traditional therapies [74].

Intestinal ecological immune nutrition shows obvious advantages in the promotion of postoperative recovery. In a study by Consoli ML et al., oral probiotics reduced the time to the first major complication, the duration of the first infection and the time to the first bowel movement [75]. Intestinal immune nutrition improves the patient's survival and discharge rate, as well as the quality of life. The interaction of probiotics with immature dendritic cells results in activation of the complement cascade and increased proliferation of NK cells [76]. The combination of various probiotics also promotes the apoptosis of colonic cells with DNA damage caused by carcinogens, down-regulates oncogenic enzymes, and enhances immune regulation [77].

In summary, the main mechanisms underlying the effects of micro-ecological immunotherapy in the intestine are the provision of energy, improvement of the functional state of tissues and organs, improvement of the immune function, protection of the intestinal mucosa and maintenance of the balance of intestinal microbiota.

Evaluation and Future Prospects of Micro-ecological Immunotherapy

The development of intestinal immune nutrition has been rapid. Researchers have discovered a powerful anti-cancer probiotic with ancient origins: Kefir [78]. Bioactive peptides in kefir induce macrophage activation and phagocytosis [79]. The active peptides also induce ROS-mediated apoptosis and activate $\text{Ca}^{2+}/\text{Mg}^{2+}$ -dependent endonuclease for DNA cleavage [80]. The intake of kefir reduces TGF- α and Bcl2 by increasing the secretion of Bax and inducing apoptosis in cancer cell [78]. Kefir has anti-tumor, anti-proliferative and apoptosis-inducing effects *in vitro* and *in vivo*, and can be used for the treatment of colorectal cancer, breast cancer and lung cancer [79].

The intestinal microbiota are not only useful in the treatment or prevention of tumors, but can also be used for tumor screening. By using sequencing technology to comprehensively analyze the types and quantities of intestinal microbes in a population, clinicians can successfully distinguish between colorectal adenomas, cancers, and non-colorectal adenomas [81]. This may be related to an increase in the abundance of proteobacteria in the stool of patients with colorectal adenomas and cancers. Moreover, it has recently been found that intestinal microbial-based predictive models are more accurate in the diagnosis of colorectal cancer than standard fecal occult blood tests [82]. This suggests that changes in the microbiota in the feces can be used as a non-invasive

sensitive screening indicator for detecting the health status of high-risk populations, permitting the early diagnosis of colorectal cancer, thereby permitting the early treatment of colorectal cancer, reducing mortality and prolonging the patient's life-span. By analyzing the shared functions between different microbiota, we can further clarify the microbial markers of gastrointestinal tumors. Establishing a causal relationship between microbiota and cancer is expected to help lock in which microbes are associated with various effects, making it possible to alter the genes of the microbes, and prevent and inhibit tumor growth associated with the microbes and their environment.

Conflict of Interests

There is no conflict of interest to declare.

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