Role and progress of arginine and its rate-limiting enzyme in the treatment of colorectal cancer

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Abstract

The treatment for colorectal cancer presents a major challenge worldwide, ranking second to lung cancer in terms of yearly fatalities. Early detection and surgery are the mainstays of current treatment; additionally, adjuvant anticancer agents are imperative. There is controversy surrounding the use of arginine in treating colorectal cancer, with two main approaches: arginine deprivation and arginine supplementation. Arginine is essential for the development and progression of colorectal cancer. As a result, arginine deprivation therapy suggests that pegylated arginine deaminase (ADI-PEG) and pegylated human recombinant Arg-1 (rhArg1-PEG) supplements should be taken in the absence of argininosuccinate synthetase-1 (ASS1) and argininosuccinate lyase (ASL). This would further reduce the supply of exogenous arginine, which can effectively inhibit tumor development. Nevertheless, studies have revealed that low L-arginine levels in tumors restrict T cell activation and proliferation, which, in turn, reduces the immune response and negatively impacts tumor therapy—furthermore, colonizing MC38 tumors with ECN that produce.

High levels of L-Arg combined with anti-PD-L1 antibody significantly improved the effectiveness of PD-L1-mediated immunotherapy. This paper reviews arginine's theoretical and preclinical findings and its rate-limiting enzyme-related adjuvant therapy for colorectal cancer.

Keywords: colorectal cancer, arginine, rate-limiting enzyme, metabolic, immunity

According to the World Health Organization's latest cancer statistics

for 2020, colorectal cancer (CRC) ranked third among all cancers with 1,931,590 new cases per year and second among all cancers with 935,173 new deaths per year [1]. Currently, available treatment options for CRC include surgery, chemotherapy, radiotherapy, targeted therapy, and so on. Surgery is the main curative treatment for colorectal cancer [2]. However, the overall therapeutic effect of colorectal cancer still depends on early detection and surgery. The arginine pathway is overactive in colorectal cancer, and several molecules involved in this pathway are potential targets for chemoprevention and targeted therapy of colorectal cancer [3]. Arginine is a semi-essential amino acid closely associated with tumor metabolism [3-9]. Many enzymes are involved in the arginine pathway, including synthetases and rate-limiting enzymes, and the actions of these enzymes affect the intracellular concentration of arginine, which in turn affects the rate of tumor metabolism [3]. This paper will review the role of arginine and its rate-limiting enzymes in treating and progressing colorectal cancer.

1. Arginine metabolic pathways

There are several intracellular pathways for arginine metabolism. Three are the most common:

(1)Citrulline and NO can be generated from arginine through the Action of arginine deiminase (ADI) or the rate-limiting enzyme nitric oxide synthase (NOS). Argininosuccinate synthetase-1 (ASS1) can use citrulline to generate the intermediary product arginine succinate, which can then be converted back into arginine through argininosuccinate lyase (ASL).

(2)Arginine is catalyzed by arginase-1 (Arg-1) to produce ornithine and urea, and the product, ornithine, is then subjected to ornithine decarboxylase (ODC) to participate in polyamine synthesis;

(3)Arginine generates Agmatine via arginine decarboxylase (ADC), which is involved in cell signaling pathways. [4-5].

2. Advances in the study of arginine in the treatment of CRC

2.1 Role and progress of arginine deprivation in CRC therapy:

arginine is required to initiate and develop CRC, and almost all CRC cells cannot grow in arginine-free conditions [9]. Arginine deprivation leads to multiple response mechanisms in cancer cells that prepare cancer cells to tolerate the arginine-deficient state and activate the apoptotic pathway to promote cancer cell apoptosis and inhibit CRC progression [10]. Studies have shown that in the process of some CRC tumor development, the

role of metabolic is to meet the energy needs of the rapid growth of tumor cells.

reprogramming, the expression of key enzymes of the ornithine cycle is abnormal, mainly the deletion of the expression of ASS1. This reduces the tumor cells' arginine synthesis, which must be achieved by increasing exogenous arginine intake and then relying more on exogenous supplementation, a phenomenon known as arginine nutritional deficiency [7-8]. Therefore, new therapeutic ideas have emerged on how to reduce exogenous arginine supplementation to achieve tumor suppression. Currently, pegylated arginine deaminase (ADI-PEG) and pegylated human recombinant Arg-1 (rhArg1-PEG) are the most widely used and studied for the treatment of arginine-deficient tumors [4]. ADI can metabolize extracellular arginine to citrulline to reduce serum arginine concentration. However, after extracellular citrulline is taken up into the cell, citrulline can be synthesized directly into arginine intracellularly under ASS1 and ASL, so the effectiveness of ADI treatment must be based on the absence of ASL or ASS1 expression; Arg-1 metabolizes arginine to ornithine extracellularly, and its therapeutic efficacy is dependent on the absence of ornithine transcarbamylase (OTC), ASL, or ASS [11]. Both aim to interfere with the exogenous replenishment of arginine, showing an inhibitory effect on some, but not all, CRC cells [9].

2.2 Role and progress of arginine supplementation in the treatment of CRC

However, early studies found that the expression of ASS1, ASL, and OTC, key enzymes of arginine metabolism, were abnormally up-regulated in colorectal tumors, resulting in a low dependence on exogenous arginine, which limited research into the application of pre-arginine deprivation in the treatment of colorectal cancer [9]. Another limiting reason is that low levels of L-arginine in the tumor limit the activation and proliferation of T-cells, which suppresses the immune response and is not conducive to tumor therapy. In contrast, additional supplementation with arginine improves the immune capacity of the T-cells [12-13]. Therefore, increasing arginine levels within the tumor can potentially enhance the anti-tumor effects of immunotherapy.

In mice bearing subcutaneous MC38 tumors, daily oral administration of L-arginine (2 g/kg body weight) has been shown to enhance the therapeutic effect of anti-PD-L1 therapy and improve the survival rate of the mice [14]. Therefore, increasing arginine levels in tumors may enhance the anti-tumor effects of immunotherapy. However, direct arginine injection at the tumor site did not reduce tumor size, with or without combined PD-L1 blockade therapy. This may be because L-arginine diffuses very rapidly beyond the tumor site, resulting in a tumor microenvironment that is not one with a very high concentration of arginine [14]. Some studies have found the Escherichia coli (E. coli Nissle, ECN) strain deregulated the feedback inhibitory effect of arginine by knocking out the arginine inhibitory protein encoded by ArgR. Also, the activity of N-acetylglutamate synthase (ArgA), the first enzyme of the arginine synthesis pathway, would be limited by high endogenous arginine levels. By introducing Argfbr, a dominant mutant that deregulates feedback inhibition was obtained so that the ECN would not be affected by high levels of L-arginine expression, and ultimately, arginine accumulated up to 400 μM within three h of pre-induction with five mM NH4Cl [14]. When cultured in vitro in a medium with only ammonium chloride as the sole nitrogen source, the modified L-Arg ECNs had higher L- arginine expression levels than wild-type ECNs. The researchers colonized MC38 tumors with L-Arg bacteria to create a tumor microenvironment with a high concentration of arginine to see if this could support anti-PD-L1 immunotherapy. The results showed that combining the L-Arg strain with anti-PD-L1 antibodies reduced tumor growth and significantly enhanced the therapeutic effect of PD-L1- mediated immunotherapy. The researchers then investigated whether this immunotherapeutic effect was T-cell-based. By repeating the experiment in T-cell-deficient cd3e-/- mice, it was found that the L- Arg strain in combination with anti-PD-L1 treatment did not have an inhibitory effect on tumor growth, suggesting that the anti-tumor effect of the L-Arg strain is highly T-cell dependent. In conclusion, the L-Arg strain combined with anti-PD-L1 immunotherapy enhanced the immune memory of tumor-specific T cells, but this may only apply to MC38 tumors [14].

2.3 Effect of NOS on CRC tumor tissue

NOS is the rate-limiting enzyme induced by NO and produces changes in activity that directly affect NO production in the arginine pathway and arginine levels. NOS is currently classified into three main types: (1) endothelial NOS (eNOS), (2) neural NOS (nNOS), and (3) inducible NOS (iNOS). Among these, iNOS plays an important role in intracellular arginine levels and is mainly found in the cytoplasm of hepatocytes, neutrophils, and tumor cells. Studies have shown that iNOS promotes the expression of vascular endothelial growth factor (VEGF) during tumor angiogenesis [15-16]. In all aspects of solid tumor growth and metastasis, angiogenesis significantly promotes tumorigenesis [15]. Therefore, tumor angiogenesis can be inhibited by inhibiting or blocking the action of VEGF and iNOS,

which can be used to treat tumors [16]. In addition, studies have shown that there is a correlation between the expression of eNOS and VEGF-C in tumor tissue in CRC treatment, but the specific mechanism remains to be seen [17].

3. Summary and Outlook

Colorectal cancer is a highly prevalent tumor worldwide, and its main cause of death is postoperative metastasis. Therefore, elucidating the pathogenesis of CRC and the mechanism of metastasis is crucial for treating CRC and reducing mortality. Despite the successes in treating colorectal cancer in recent years, the enzymes in the pathway of arginine metabolism and whether the combination of arginine with other drugs can produce more benefit are still the next questions to be explored.

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