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REVIEW

Immunotherapy Targeting Heparanase-1 May Be the Dawn of Cancer Sufferers

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ABSTRACT

Although there have been great advances in preventive and therapeutic approaches, malignant tumor is still one of the major causes of death in the world. The control of invasion and metastasis of advanced-stage malignancies remains the pain in the neck. Recently, cancer immunotherapy has emerged as a new supplement for cancer treatment that has weak side effects and favorable applicability. Antigen presentation is critical for triggering such immune response, and Dendritic cells and Cytotoxic T Lymphocytes are very important in this process. Although cancer immunotherapy has shown encouraging results in some human clinical trials, there have been some momentous setbacks: (1) Most Tumor Associated Antigens (TAAs) that have been described are either tissue specific or nonsignificant for cell survival; (2) the poor reactivity of T cells against TAAs; (3) immunosuppressive features of tumor cells; and (4) the expansion of Treg cells. Therefore, it is critical to identify promising TAAs for tumor immunotherapy, and a class of TAA termed 'Universal Tumor Antigens' has been proposed that is supposed not only to induce antitumor immunity against most tumor types, but also to have crucial functional roles in tumor growth and development. Heparanase-1 can degrade the heparan sulfate proteoglycans in the extracellular matrix and basement membrane. Unlike most other TAA, heparanase-1 is found highly expressed in most mammalian malignant tumors, and many hurdles could be overcome when cancer immunotherapy is targeting heparanase-1. It was demonstrated many years ago that the metastatic potential of tumor cells is correlated with heparanase-1 expression. Thereby, it can be hypothesized that heparanase-1 is a key enzyme involved in the metastasis of malignant tumors, and immunotherapy aiming at

heparanase-1 may be the dawn of advanced-stage cancer sufferers.

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Key words: Immunotherapy; Heparanase; Cancer

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IMMUNOTHERAPY OFFERS A NEW HOPE FOR MALIGNANCIES VICTIMS

Although there have been enormous advances in preventive and therapeutic approaches, malignant tumor is still one of the major causes of death in the world. Invasion and metastasis remain the major bewilderments in curing cancer patients, thus effective therapy to combat malignancies is still not established. Until the present, surgical resection, radiotherapy and chemotherapy have been the most common forms of management used in treating cancers. Although surgical intervention may play a vital role in patients with resectable lesions, the undesirable progresses of local or even distant metastasis that often occur in many advanced-stage tumors make surgical therapy unsuitable. And it is quite common for patients to stand unpleasant side effects when receiving radiotherapy or chemotherapy. In order to establish a complementary therapeutic approach in cancer therapeutic area, immunotherapy is under intent investigation during the past few years. It has emerged as a new modality for cancer treatment for its weak side effects and applicability. It utilizes the immune system to recognize and specifically eradicate tumor cells and has shown encouraging results in certain human clinical trials^[1]. The quintessential goals of such therapy are manufacturing vaccines that could elicit potent anti-tumor immune responses without side effects, and subsequently cause recognition of tumors with succedent elimination.

Antigen presentation is critical for triggering an immune response, and Dendritic cells (DCs) are important in this process because they are the most potent professional antigen-presenting cells^[2]. To accomplish this mission, DCs present TAA to the immune system and initiate a TAA-specific immune response. TAA-loaded DC presentation is the major mechanism of tumor immunotherapy, and DC-based cancer immunotherapy has become a hotspot in the study

of cancer immunotherapy. It has the advantage of strong immunogenicity and minimal side effects^[3].

Cytotoxic T Lymphocytes (CTLs) are also essential in anti-cancer immuno-response. They are chief mediators of tumor immunosurveillance through the recognition of TAAs as cognate peptides bound to MHC molecules expressed on the surface of tumor cells. A major advance in tumor immunology in the last two decades was the verification that CTL epitopes bind to MHC rather than to integral TAAs to induce CTL activation. These epitope peptides are often 8 to 10 amino acids long, with 2 to 3 primary anchor residues that interact with MHC class I molecules and 2 to 3 amino acid residues that bind to the T-cell receptor^[4].

THERE ARE STILL MANY SETBACKS IN CANCER IMMUNOTHERAPY

TAAs presented by DCs trigger a TAA-specific immune response, thus they are the crux of anti-tumor immunotherapy. Unfortunately, most TAAs that have been described are either possess tissue specific or unessential for cell survival, such as in the case of carcinoembryonic antigen (CEA) in colon and lung carcinoma, prostate-specific antigen (PSA) in prostatic carcinoma, and fetoprotein (AFP) in hepatocellular carcinoma. Immunotherapy aiming at these TAAs has shown a narrow therapeutic window and an unsatisfied efficiency. On the other hand, the reactivity of T cells against TAAs remains poor in spite of the assistant of immunologic adjuvants. Perhaps the greatest hurdle in identifying an approving immunogenic cancer antigen is that the majority of TAAs are selfantigens, which either are over-expressed or vary only in small mutations or altered post-translational modifications^[5]. Accordingly, these antigens tend to be unreliable in eliciting efficient immunity owing to pre-existing tolerance. As a result, T cell receptors (TCRs) often exhibit low affinity for these TAAs, due to prior deletion of T cells with high-affinity TCRs during negative selection in the thymus.

Immunosuppressive feature of tumor cells is a crucial obstacle for cancer immunotherapy. And some cancer cells can make alterations in TAA presentation and even secrete immunosuppressive factors, evading T cell responses mainly by avoiding immune “recognition”^[6], and such alterations may eventually accelerating the so-called “immunoediting” phenomenon, which pictures an early vigorous immune response that initially recognizes and arrests tumor growth, but may eventually induce the development of highly aggressive phenotypes due to the principle of “survival of the fittest”^[7].

And another fundamental problem in anti-tumor (vaccination) immunotherapy is the expansion of Treg cells (Tregs). Tregs have been verified to possess the undesirable ability to inhibit specific CTL responses against tumor antigens, and it evokes tumor-related immune tolerance instead of eliminating tumor cells^[8]. Generally speaking, the capacity of Tregs to suppress effector cell expansion has been attributed to the intracellularly expressed cytotoxic T lymphocyte antigen 4 (CTLA-4, CD152) molecule. In many cases, TAAs which could otherwise be appropriate targets for anti-cancer immunotherapy, expanding TAA specific Treg cells rather than stimulate effector T cells. Accordingly, in the course of immunotherapy it is important to address this issue^[9].

IT IS CRITICAL TO IDENTIFY PROMISING TAAS FOR TUMOR IMMUNOTHERAPY

The central issue in the development of cancer immunotherapeutic strategies is the identification of relevant TAAs capable of medi-

ating anti-tumor elimination by competent immune system. Immunotherapeutic protocols based on vaccination with autologous antigen-presenting cells (APCs) sensitized with TAA have been used for almost a decade in the treatment of metastatic human melanoma^[10], although a large proportion of patients showed a detectable immune response, clinical trials demonstrated only limited evidence of tumor regression. Recently, some researchers advocated that the effective immunotherapy requires cancer cells to have ideal TAAs on their surface that should be: (1) unique, distinctly different than on normal cells; (2) constitutively expressed during the cell cycle; and (3) their constitutive expression is essential for cell survival^[11]. Currently, dozens of TAAs have been identified and described^[12]. The expression of most TAAs is restricted to a few tumor types and to a fraction of patients with these types of tumor. Nevertheless, as mentioned above, the appearance of antigen-loss or antigen-alteration mutations in tumor cells in response to immune pressure is highlighted and well described by many researchers. To circumvent this issue, a class of TAA termed ‘Universal Tumor Antigens’ (UTAs) has been proposed that is hypothesized not only to induce antitumor immunity against a vast range of tumor types, but also to have crucial functional roles in tumor growth and development. For years, enormous scientific works have been done to find out promising UTAs before the identification of heparanas-1 (HPA-1).

HPA-1 MAYBE THE DAWN IN TUMOR IMMUNOTHERAPY

The heparanase mRNA encodes a 61.2 kDa protein with 543 amino acids. This pro-enzyme is post translationally cleaved into 8 and 50 kDa subunits that non-covalently associate to form the active heparanase^[13]. Heparanase protein is an endo- β -glucuronidase that cleaves HS side chains presumably at sites of low sulfation, releasing saccharide products with appreciable size (4–7 kDa) that can still associate with protein ligands and facilitate their biological potency. It is the only endogenous endoglycosidase, as yet, that can degrade the heparan sulfate proteoglycans (HSPGs) in the extracellular matrix (ECM) and basement membrane (BM)^[14]. Mammalian cells express primarily a single dominant functional heparanase enzyme (HPA-1)^[15]. A second heparanase (HPA-2) has been cloned and sequenced but has not been shown to have HS degrading activity^[16].

Unlike most other TAAs, HPA-1 is found highly expressed in most mammalian malignant tumors, and its expression has been linked to tumor metastasis and angiogenesis^[17]. HPA-1 has been detected at a relatively low levels at mammalian lymphoid organs, placenta and platelets, and it is either not expressed or expressed at very low levels in other normal tissue^[18]. Activation of HPA-1 is a determining factor in metastasis that enables tumor cells to break through the ECM and BE barriers, releasing multiple types of cytokine and cause the formation of new vessels and local permanent planting^[19], thus it can be hypothesized that HPA-1 is a key enzyme involved in the metastasis of malignant tumors. In fact, it was demonstrated many years ago that the metastatic potential of tumor cells is correlated with HPA-1 expression. Evidence for a direct role of HPA-1 in tumor metastasis was provided by the conversion of tumor cells, such as T-lymphoma and melanoma cells, from a non-metastatic to metastatic phenotype following stable transfection and overexpression of HPA-1^[20].

Some tumor cells can downregulate or mutate TAA expression to evade immune surveillance^[21]. However, because of the crucial role of HPA-1 in tumor progression, down-regulation or mutation of

its expression as a means of immune escape might itself have deleterious effects on the proliferation and metastasis of tumor cells. Tregs usually accumulate at the tumor site, where they suppress the function of effector T cells, resulting in tumor growth despite the presence of TAA-specific CTLs^[22]. Interestingly, Tregs against HPA-1 were not found in patients with colorectal cancer^[23]. HPA-1-targeted immunotherapy is thus expected to be prolonged and more efficient owing to the absence of T-suppressor cells. More attractively, HPA-1-specific Tregs are absent in some patients with cancer^[23], suggesting that the CTL response induced by HPA-1 might result in more efficient tumor suppression than that induced by other TAAs.

Based on the above features, HPA-1 is therefore considered to be an ideal universal immunotherapeutic target in human cancers. To date, Several HLA-A2-restricted CTL epitopes in the human HPA-1 protein have been shown to elicit specific antitumor immunity in vitro and in vivo against various tumor cells. In addition, there have been no obvious toxicity or autoimmune reactions observed in the evaluation of this strategy in any experimental models so far^[24,25,26,19]. Except for CTL epitopes, B-cell epitopes of HPA-1 also showed an encouraging prospect: Yang and colleagues designed 3 multiple antigen peptide vaccines of human heparanase based on B-cell epitopes. They found that antibodies induced by these MAP vaccines could inhibit the heparanase activity of liver cancer cells and, therefore, reduce the invasiveness of tumor cells in vitro^[27]. All these studies provide evidence that HPA-1 is an ideal universal target immuno-antigen, and immunotherapy aiming at HPA-1 may be the dawn of advanced-stage cancer patients.

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